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Import Risk Analysis Eviscerated or Trunked Fish for Human Consumption

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Version 3.2

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New Zealand Government

Ministry for Primary Industries
Manatū Ahu Matua

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Science and Risk Assessment
Ministry for Primary Industries

Import Risk Analysis: Eviscerated or Trunked Fish for Human Consumption

Version 3.2

June 2019

Approved for IHS Development

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Ministry for Primary Industries

Version Information

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1.0	First peer-reviewed version	22 March 2015
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New Zealand is a member of the World Trade Organisation and a signatory to the Agreement on the Application of Sanitary and Phytosanitary Measures ("The Agreement"). Under the Agreement, countries must base their measures on an International Standard or an assessment of the biological risks to plant, animal or human health.

This document provides a scientific analysis of the risks associated with eviscerated or trunked fish, that may be fresh, chilled or frozen. It assesses the likelihood of entry, exposure, establishment and spread of various diseases and pests in relation to this commodity and assesses the potential impacts of those organisms should they enter and establish in New Zealand. The document has been internally and externally peer reviewed.

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Contents

1. Index of tables and figures

2. Glossary of abbreviations

Biosecurity New Zealand	Previous New Zealand government biosecurity agency, now a business unit of the Ministry for Primary Industries, New Zealand
Competent Authority	Government veterinary authority or other government authority having the responsibility and competence for ensuring or supervising the implementation of animal health and welfare measures, international veterinary certification and other standards and recommendations in the OIE Code and Aquatic Code
EFSA	European Food Safety Authority
ESR	Environmental Science and Research Ltd, Crown Research Institute, New Zealand
FAO	Food and Agriculture Organisation of the United Nations, Rome, Italy
MPI	Ministry for Primary Industries, New Zealand
NIWA	National Institute for Water and Atmospheric Research, Crown Research Institute, New Zealand
OIE	World Organisation for Animal Health, Paris, France
IHS	Import Health Standard
RMP	Risk management proposal
SPS Agreement	Agreement on the Application of Sanitary and Phytosanitary Measures
WTO	World Trade Organisation, Geneva, Switzerland

3. Executive summary

This document is a qualitative biosecurity import risk analysis for non-viable fresh, chilled or frozen fish. This includes eviscerated bony fishes (class Actinopterygii) and trunked cartilaginous fishes (class Elasmobranchii) and their products (including minced, salted, smoked or mechanically recovered fish), imported for human consumption. These fish may be sourced from fresh, brackish, or marine waters. This analysis was requested by the Animal Imports team of the Ministry for Primary Industries (MPI).

This risk analysis is consistent with the guidelines described in *Biosecurity New Zealand Risk Analysis Procedures – Version 1* and in Chapter 2 of the *Aquatic Animal Code* 2016 of the World Organisation for Animal Health (hereafter referred to as the *Code*).

The consequences to human health arising from the introduction of a zoonotic pathogen are noted in this report. However, all imported food for human consumption must meet the legislative requirements of the Food Act 2014 and the Animal Products Act 1999, which manage the risks to public health associated with food in New Zealand. These requirements are in addition to the requirements of the Biosecurity Act 1999.

From a preliminary list of hazards comprising of 569 organisms of concern, a total of 20 viruses or viral species complexes, 12 bacterial pathogens or species complexes, 4 fungal or microsporidian pathogens and 5 groups of metazoan pathogens were identified as representing a non-negligible risk. These include the following risk organisms, where those with zoonotic potential are indicated by *:

Virus	• Epizootic haematopoietic necrosis virus/European catfish virus/ European sheatfish virus • European eel virus • European eel herpesvirus • Grass carp haemorrhagic virus • Grouper iridovirus • Infectious haematopoietic necrosis virus • Infectious pancreatic necrosis virus /Halibut birnavirus/Viral deformity of yellowtail virus • Infectious salmon anaemia virus • Koi herpesvirus • Marine Hirame rhabdovirus • New Japan virus of salmonids • Nodaviruses, including viral nervous necrosis virus • *Oncorhynchus masou* virus • Piscine aquareovirus/Salmon aquareovirus/Tasmanian salmon reovirus/Grass carp reovirus/Turbot reovirus/Heart & skeletal muscle inflammation syndrome virus • Red sea bream iridovirus/Infectious spleen and kidney necrosis virus/Gourami iridovirus • Salmon alphavirus/Salmon pancreatic disease virus • Salmon gill poxvirus/Carp oedema virus/Koi sleepy disease virus • Spring viraemia of carp virus/Pike fry rhabdovirus • Viral erythrocytic necrosis/ Piscine erythrocytic necrosis virus • Viral haemorrhagic septicaemia virus
Bacteria	• *Aeromonas hydrophila* (exotic strains) * • "Atypical" *Aeromonas salmonicida* var. *salmonicida* • "Typical" *Aeromonas salmonicida* var. *salmonicida*

	• *Edwardsiella* spp. * • *Flavobacterium columnare* (exotic strains) • *Francisella* spp. • *Moritella viscosa* • *Piscirickettsia salmonis* and related Rickettsia-like organisms • *Pseudomonas anguilliseptica* • *Renibacterium salmoninarum* • *Streptococcus agalactiae* (serotype III: 283), *Streptococcus iniae* * • *Yersinia ruckeri* (Hagerman and other exotic strains) *
Fungi and Microsporidia	• *Aphanomyces invadans* • *Ichthyophonus hoferi* • *Sphaerothecum destruens* • Microsporidian pathogens (*Glugea plecoglossi, Pleistophora anguillarum, Microsporidium seriolae, Nucleospora salmonis*) *
Metazoa	• Myxosporidian pathogens (*Enteromyxum, Henneguya, Kudoa, Myxobolus, Sphaerospora, Unicapsula*) * • *Anguillicola crassus** • *Gyrodactylus salaris* and related monogenean parasites • Digenean larvae * • Cestode larvae *

Potential pathways within New Zealand for introduction of biosecurity risk organisms (Figure 1) include trade wastes and landfill, as well as fish product wastes from households (Appendix 1). Other potential pathways not included in this risk analysis include food imported but diverted from the human consumption pathway (including use as fish bait, animal/fish food or industrial processing), as they are considered less significant or unlikely pathways.

Risk management options are provided for each identified risk organism in the appropriate chapter of this report. These actions may be completed offshore, or in a transitional facility (as defined in the Biosecurity Act 1999). The disposal of all liquid and solid wastes generated during transport, storage and processing is assumed to be in accordance with the transitional facility requirements of the Biosecurity Act 1999[A]. Risk management options may include one or a combination of the following measures:

[A] Transitional Facilities for Animal Products (2016). MPI-STD-ANIPRODS. Facility Standards, Ministry for Primary Industries, Wellington, New Zealand, 18p.

- Identification of family, genus, species and common name of fish
- Acceptance of declaration of country or zone freedom, through MPI Country Approval Procedures[B]
- Restriction of eviscerated fish to wild-caught only (excluding fish from aquaculture)
- Processed state consistent with requirements of the OIE Aquatic *Code*
- Heat treatment or frozen storage (the time/temperature combination that has been demonstrated to inactivate the pathogen of concern)
- Additional processing beyond evisceration/trunking:
    - removal of the gills
    - removal of head and gills
    - processing to skin-off fillets.

These are summarised by fish host family (Appendix 2), and by processing, heat treatment and frozen storage options (Appendix 3). A summary evaluation (Appendix 4) is provided as a guide for selection of appropriate risk management options. Where uncertainty around life cycle details and infection pathways exists, details are provided in the appropriate disease chapters.

From the summary evaluation of risk management options (see below), overall biosecurity risks are negligible where fish of families associated with an OIE-listed disease are imported under the conditions specific to the appropriate OIE Aquatic Code disease chapter.

Where fish of families associated with a non-OIE listed disease are imported in a processed state consistent with the recommended treatments for OIE-listed diseases in the OIE Aquatic Code (OIE 2018a) (see Appendix 3), the biosecurity risks are assessed as negligible.

Where general OIE treatments for OIE-listed diseases (Appendix 3) have not been chosen by an exporting country and the commodity includes fish of families identified as susceptible to one or more of the risk organisms identified in this report, then the risk is non-negligible. Risk management options can be selected as provided in the appropriate chapter and summarised in Appendix 3 and 4 of this report.

[B] Export controls and certification systems for animals and animal products. Guidance for Competent Authorities of exporting countries. Ministry for Primary Industries, Wellington, New Zealand [Online] Available from: http://www.mpi.govt.nz/news-and-resources/publications/ [Accessed 15 March 2019].

Effect of Risk Management Options on Pathogen (see Appendix 3 and 4)

Category	Risk Management Option	Impact on Pathogen type[3]		
		Viruses	**Bacteria**	**Other**
OIE-listed diseases	Country freedom[1]	Substantial	N/A	Substantial
	Species declaration	Substantial	N/A	Substantial
	Processed following specific OIE *Code* guidelines	Eliminate	N/A	Eliminate
Non-OIE listed diseases	Country freedom[1]	Substantial	Substantial	Substantial
	Species declaration	Substantial	Substantial	Substantial
	Restricted to wild-sourced fish, not aquaculture	No effect	Moderate	Moderate
	Removal of gills (GGU)	Slight	Slight	Slight
	Removal of head and gills (HGU)	Moderate	Moderate	Moderate
	Skin-off fillet (SKF)	Moderate	Moderate	Moderate
	Heat treatment	Eliminate	Eliminate	Eliminate
	Frozen storage	No effect	No effect	Eliminate[2]
	Follow OIE *Aquatic Code* treatments for OIE-listed diseases	Eliminate	Eliminate	Eliminate

1 Country freedom may be accepted following the MPI Country Approval Procedures

2 Myxosporean pathogens are unaffected by frozen storage

Levels of pathogen load[3]

Eliminate > 95% reduction in pathogen load[4]
Substantial 71-95% reduction in pathogen load
Moderate 51-70% reduction in pathogen load
Slight < 50% reduction in pathogen load

3 Where pathogen load is a qualitative assessment of the presence of the pathogen in the commodity

In preparing these options, the following assumptions have been made:

- Country Freedom
  Declaration of country freedom may be accepted for countries approved under the MPI Country Approval Procedures.

- Identification of Fish Host
  A wide variety of fish species may potentially be imported into New Zealand. It is assumed that if an identified risk agent is reported from a species in a family, then the whole family may be susceptible to that disease. Accordingly, the risk management options proposed apply to all members of the listed fish families. The families of fish associated with these identified risk organisms are provided in Appendix 2.

4. Introduction

The Animal Imports team of MPI (Ministry for Primary Industries) has requested the development of a qualitative risk analysis to examine the biosecurity risks associated with the importation of fresh, chilled or frozen non-viable fish for human consumption. This includes eviscerated bony fishes (class Actinopterygii) and trunked cartilaginous fish (class Elasmobranchii) and their products (including frozen, chilled, minced, salted, smoked or mechanically recovered fish). These commodities may be derived from wild fisheries or aquaculture stock, from fresh, brackish or marine waters.

The risks associated with fish imports into New Zealand were previously examined for salmonids (MacDiarmid 1994; Stone *et al.* 1997; MAF 1999), *Pangasius* spp. (Johnston 2008a), *Oreochromis* spp. (Johnston 2008b; Melville & Johnston 2010), and yellowtail kingfish (*Seriola lalandi*) (Diggles 2003), while diseases associated with salmonid aquaculture were reviewed by Diggles (2011, 2016). No previous analysis of the risks associated with importation of other fish species into New Zealand has been completed.

Risk analyses prior to 2016 considered that simple evisceration provided sufficient mitigation to ensure a pathogen-free commodity for international trade in fish for human consumption (B. Oidtmann *pers. comm.* 2017). Eviscerated teleost fish are now recognised as representing a significant risk of pathogen introduction (Oidtmann *et al.* 2013a, 2017; Pearce *et al.* 2014; C. Rodgers *pers. comm.* 2017). It has subsequently been acknowledged (OIE 2016a) that further risk management measures may be necessary, where supported by an import risk analysis.

5. New Zealand fish fauna

The continental shelf and slope of New Zealand is relatively large and ranges over 30 degrees of latitude, but the coastal shelf and reef areas are relatively small (Paul 2000; Webber *et al.* 2010). The New Zealand fish fauna which consists of approximately 200 families and 1000 species of marine and freshwater fish (Table 1). While this includes both tropical/subtropical and sub-polar fish species, New Zealand is situated too far south to support a true sub-tropical fauna in its northern waters. It is also too remote from other significant land masses to support a significant sub-polar fauna in its southern waters (Paul 2000).

5.1. New Zealand marine fisheries

New Zealand has a diversity of marine fish, as shown in Table 1. Marine deep-water fish species support major export fisheries for hoki (*Macruronus novaezelandiae*), Southern hake (*Merluccius australis*), ling (*Genypterus blacodes*), southern blue whiting (*Micromesistius australis*), jack mackerel (*Trachurus* spp.), oreo (*Allocyttus* spp.) and orange roughy (*Hoplostethus atlanticus*).

Significant inshore fisheries exist for snapper (*Pagrus auratus*), blue cod (*Parapercis colias*), bluenose (*Hyperoglyphe antarctica*), tarakihi (*Nemadactylus macropterus*), warehou (*Seriolella brama*), gurnard (*Chelidonichthys kumu*), rig (*Mustelus lenticulatus*), blue moki (*Latridopsis cilliaris*), flounder (*Rhombosolea* spp.), hapuku (*Polyprion oxygeneios*), groper), trevally (*Pseudocarynz dentex*), school shark (*Galeorhinus galeus*) and John dory (*Zeus faber*). Major pelagic fisheries include tuna (*Scomber* spp., *Thunnus* spp.), kawahai (*Arripis trutta*)

and mackerels (*Trachurus* spp.). Over 200,000 tonnes of fish are caught each year, with an estimated export value of $660 million in 2016 (MPI 2018).

Table 1. Orders and Families of New Zealand Marine fish (Family and Common Names After Paulin *et al.* 2001, Fishbase 2018)

Actinopterygii: Teleostei (Bony Fishes)	
Order	**Family and Common Name**
Anguilliformes	Anguillidae (freshwater eels), (Chlopsidae (false moray eels), Congridae (conger eels), , Derichthyidae (longneck eels), Muraenidae (moray eels), Nemichthyidae (snipe eels), Netastomatidae (duckbill eels), Ophichthidae (snake eels), Serrivomeridae (sawtooth eels) , Synaphobranchidae (Cut-throat eels)
Atheriniformes	Atherinidae (hardyheads), Isonidae (surf silversides)
Aulopiformes	Alepisauridae (lancetfishes), Anotopteridae (daggertooths), Aulopidae (flagfins), Bathysauridae (deepsea lizardfish), Bathysauropsidae (black deepsea lizardfishes), Chlorophthalmidae (greeneyes), Evermannellidae (sabretooth fishes), Ipnopidae (tripodfishes), Notosudidae (waryfishes), Omosudidae (hammerjaw), Paralepididae (barracoudinas), Paraulopidae (Cucumberfish), Scopelarchidae (pearleyes), Synodontidae (lizardfishes)
Beloniformes	Belonidae (neddlefish), Exocoetidae (flyingfishes), Hemiramphidae (halfbeaks), Scomberesocidae (sauries)
Beryciformes	Anoplogastridae (fangtooths), Berycidae (alfonsinos), Diretmidae (Discfishes), Holocentridae (squirrelfishes), Monocentridae (pineapplefishes), Trachichthyidae (roughies)
Bythitidae	Dinematichthyidae (fleshfishes)
Cetomimiformes	Barbourisiidae (red whalefishes), Cetomimidae (flabby whalefish), Rondeletiidae (redmouth whalefishes)
Clupeiformes	Clupeidae (sardines), Engraulidae (anchovies)
Gadiformes	Bregmacerotidae (codlets), Euclichthyidae (eucla cods), Gadidae (true cods), Lotidae (rocklings), Macrouridae (rattails), Melanonidae (pelagic cods), Merlucciidae (hakes), Moridae (morid cods), Muraenolepididae (moray cods), Phycidae (Phycid hakes)
Gobiesociformes	Gobiidae (gobies), Gobiesocidae (clingfish, lumpfishes)
Gonorynchiformes	Gonorynchidae (sandfishes)
Lampriformes	Lampridae (opahs), Lophotidae (crestfishes), Regalecidae (oarfishes),Trachipteridae (dealfishes), Veliferidae (Velifers)
Lophiiformes	Antennariidae (frogfishes), Caulophrynidae (fanfin anglerfishes), Ceratiidae (seadevils), Chaunacidae (seatoads), Diceratiidae (doublespine anglerfishes), Gigantactinidae (slender anglerfishes), Himantolophidae (prickly anglerfishes), Linophrynidae (leftvent seadevils), Lophiidae (goosefishes), Melanocetidae (humpback anglerfishes), Neoceratiidae (toothed seadevils), Ogcocephalidae (batfishes), Oneirodidae (dreamers)
Macrouridae	Bathygadidae (codhead rattails), Macrouroididae (balloonhead rattails), Trachyrincidae (rough rattails)
Mugiliformes	Mugilidae (mullets)
Myctophiformes	Myctophidae (lanternfishes), Neoscopelidae (blackchins)
Notacanthiformes	Halosauridae (halosaurs), Notacanthidae (spiny eels)
Ophidiiformes	Aphyonidae (blind cusk eels), Bythitidae (brotulas), Carapidae (pearlfishes), Ophidiidae (cusk eels)
Osmeriformes	Alepocephalidae (slickheads), Argentinidae (silversides), Bathylagidae (deepsea smelts), Galaxiidae (Galaxiids, whitebaits, smelts), Microstomatidae (white smelts), Opisthoproctidae (spookfishes), Platytroctidae (tubeshoulders), Retropinnidae (Southern smelts)
Perciformes	Acanthuridae (surgeonfishes), Acropomatidae (ocean basses), Aplodactylidae (marblefishes), Apogonidae (cardinalfishes), Ariommatidae (Ariommids), Arripidae (kahawai), Blenniidae (blennies), Bovichtidae (thornfishes), Bramidae (pomfrets), Callanthiidae (splendid perches), Callionymidae (dragonets), Carangidae (trevallies), Caristiidae (manefishes), Centrolophidae (rudderfish), Cepolidae (bandfishes), Chaetodontidae (butterflyfishes), Champsodontidae (gapers), Cheilodactylidae (moki and terakihi), Cheimarrichthyidae (torrentfish), Chiasmodontidae (swallowers), Chironemidae (kelpfishes), Cirrhitidae (hawkfishes), Clinidae (weedfishes), Coryphaenidae (dolphinfishes), Creediidae (tommyfishes), Draconettidae (deepsea dragonets), Echeneidae (remoras), Eleotridae (bullies), Emmelichthyidae (bonnetmouths), Ephippidae (spadefishes), Epigonidae (deepsea cardinalfishes), Gempylidae (gemfishes), Girellidae (nibblers), Grammatidae (soapfishes), Howellidae (pelagic cardinalfishes), Istiophoridae (billfishes), Kuhliidae (flagtails), Kyphosidae (drummers), Labridae (wrasses), Latridae (trumpeters), Leptoscopidae (stargazers), Lutjanidae (tropical snappers), Luvaridae (luvar), Malacanthidae (tilefishes),

	Microcanthidae (stripeys), Microdesmidae (dart gobies), Mullidae (goatfishes), Nomeidae (cubeheads), Nototheniidae (ice cods), Oplegnathidae (knifejaws), Pempheridae (bulleyes), Pentacerotidae (boarfishes), Percophidae (Opalfishes), Pinquipedidae (Sandperches, weevers), Plesiopidae (rockfishes), Polyprionidae (wreckfishes), Pomacanthidae (Angelfishes), Pomacentridae (damselfishes), Priacanthidae (bigeyes), Scorpididae (sweeps), Scombridae (mackerels and tunas), Scombrolabracidae (black mackerel), Serranidae (gropers and perches), Sparidae (seabreams), Sphyraenidae (barracudas), Tetragonuridae (Squaretails), Thalasseleotridae (marine sleepers), Trichiuridae (scabbardfishes), Tripterygiidae (triplefins), Uranoscopidae (armourhead stargazers), Xiphiidae (swordfish), Zanclidae (moorish idol), Zoarcidae (eelpout)
Pleuronectiformes	Achiropsettidae (finless flounders), Bothidae (left eye flounders), Cynoglossidae (tongue soles), Pleuronectidae (southern righteye flounders), Soleidae (true soles)
Polymixiiformes	Polymixiidae (bearfishes)
Saccopharyngiformes	Cyematidae (bobtail snipe eels), Eurypharyngidae (pelican eels/gulpers), Saccopharyngidae (whiptail gulpers)
Salmoniformes	Salmonidae (Trouts, salmon, chars)
Scorpaeniformes	Congiopodidae (pigfishes), Cottidae (sculpins), Dactylopteridae (flying gurnards), Hoplichthyidae (ghost flathead), Liparidae (snailfishes), Neosebastidae (gurnard perches), Plectrogeniidae (small-mouth scorpionfishes), Psychrolutidae (toadfishes), Scorpaenidae (scorpionfishes and lionfishes), Sebastidae (rockfish, seaperches), Setarchidae (deepwater scorpionfishes), Tetrarogidae (waspfishes), Triglidae (gurnards)
Stephanoberyciformes	Melamphaidae (bigscales), Stephanoberycidae (pricklefishes)
Stomiiformes	Diplophidae (portholefishes), Gonostomatidae (bristelmouths), Phosichthyidae (lighthousefishes), Sternoptychidae (marine hatchetfishes), Stomiidae (dragonfishes)
Syngnathiformes	Aulostomidae (trumpetfishes), Centriscidae (bellowsfish, pipefish, seahorses, snipefish), Fistulariidae (cornetfishes)
Tetraodontiformes	Balistidae (triggerfishes), Diodontidae (porcupinefishes), Molidae (sunfishes), Monacanthidae (leatherjackets), Ostraciidae (boxfishes), Tetraodontidae (puffers), Triacanthodidae (spikefishes)
Zeiformes	Cyttidae (Cyttid dories), Grammicolepididae (tinselfishes), Macrurocyttidae (elongate dories), Oreosomatidae (oreo dories), Parazenidae (slender dories), Zeidae (dories), Zeniontidae (armoureye dories)
Elasmobranchii (Sharks, Skates, Rays, Chimaeras)	
Order	**Family and Common Name**
Carcharhiniformes	Carcharhinidae (requiem sharks), Proscyllidae (finback cat sharks), Pseudotriakidae (false cat sharks), Scyliorhinidae (cat sharks), Sphyrnidae (hammerhead sharks), Triakidae (smoothhounds)
Chimaeriformes	Callorhinchidae (elephant fishes), Chimaeridae (chimaeras, ghost sharks), Rhinochimaeridae (longnosed chimaeras)
Heterodontiformes	Heterodontidae (horn sharks)
Hexanchiformes	Chlamydoselachidae (frill sharks), Hexanchidae (cow sharks)
Lamniformes	Alopiidae (thresher sharks), Cetorhinidae (basking sharks), Lamnidae (mackerel sharks), Mitsukurinidae (goblin sharks), Odontaspididae (sandtiger sharks)
Myliobatiformes	Dasystidae (stingrays), Mobulidae (manta rays), Myliobatidae (eagle rays)
Orectolobiformes	Rhincodontidae (whale shark)
Rajiformes	Arhynchobatidae (kongtailed skate), Rajidae (skates)
Squaliformes	Centrophoridae (gulper sharks), Dalatiidae (kitefin sharks), Echinorhinidae (bramble sharks), Oxynotidae (rough sharks), Squalidae (dogfishes)
Torpediniformes	Narcinidae (slender electric rays), Narkidae (blind electric rays), Torpedinidae (electric rays)

Family Lutjanidae are restricted to northern tropical waters including the Kermadec Islands. These tropical snappers are considered rare and of no commercial significance (Paulin *et al.* 2001). Their specific pathogens will not be considered further.

5.2. New Zealand freshwater fish fauna

The New Zealand freshwater fish fauna (Table 2) is relatively depauparate (NIWA 2018). It was supplemented by the introduction of salmonids late in the 1800s, while other freshwater fish have been subsequently introduced or become established (Table 3).

Table 2. Orders and Families of New Zealand Freshwater Fish (Family and Common Names After Paulin *et al.* 2001), NIWA 2018)

Order	Family and Common Name
Anguilliformes	Anguillidae (eels)
Mugiliformes	Mugilidae (mullets)
Osmeriformes	Retropinnidae (smelts)
Perciformes	Eleotridae (bullies), Pinguipedidae (torrentfish), Tripterygiidae (triplefin)
Pleuronectiformes	Pleuronectidae (black flounder *Rhombosolea retiaria*)
Salmoniformes	Galaxiidae (galaxiids, mudfish)

No salmonid broodstock or ova have been imported since the early 1900s (NIWA 2018). While commercial trout farming is illegal in New Zealand, trout support significant recreational fisheries that are maintained or supplemented by non-commercial hatchery operations. There are no significant commercial fisheries for wild salmonid stocks and the recreational fisheries are largely confined to fresh water (NIWA 2018).

Table 3. Order, Family and Genera of Freshwater Fish Introduced into New Zealand (After Paulin *et al*. 2001, NIWA 2018)

Order	Family	Species and Common Name
Cypriniformes	Cyprinidae	*Carassius auratus* (goldfish), *Cyprinus carpio* (common carp, ornamental varieties including koi (*C. Carpio koi*)), *Gobio gobio* (gugeon), *Ctenopharyngodon idella* (grass carp), *Leuciscus idus* (orfe), *Scardinius erythrophthalmus (rudd), Hypophthalmichthys molitrix* (silver carp), *Tinca tinca* (tench)
Cyprinodontiformes	Poeciliidae	*Phallocerus caudimaculatus* (dusky millions fish), *Poecilia reticulata* (guppy), *gambusia affinis* (mosquitofish), *Poecilia latipinna*, (sailfin molly), *Xiphophorus helleri* (green swordtail)
Perciformes	Percidae	*Perca fluviatilis* (European perch)
Salmoniformes	Salmonidae	*Salmo salar* (Atlantic salmon), *Salmo trutta* (brown trout), *Salvelinus fontinalis* (brook char), *Oncorhynchus mykiss* (rainbow trout), *O. nerka* (sockeye salmon), *O tshawytscha* (Chinook or king salmon), *Salvelinus* namaycush (Mackinaw)
Siluriformes	Ictaluridae (Ameiuridae)	*Ameiurus nebulosus* (brown bullhead)

Brown trout (*Salmo trutta*) are widely distributed in central and southern New Zealand fresh waters, but may also spend time at sea. Both brown and rainbow trout (*Oncorhynchus mykiss*) populations are supplemented by hatchery release, but rainbow trout are now self-sustaining in most fresh water catchments throughout New Zealand fresh waters. No sea-run populations of rainbow trout exist in New Zealand (NIWA 2018). Brook char (*Salvelinus fontinalis*) are widely distributed in fresh waters of the North and South Island and are maintained by hatchery release (NIWA 2018).

Small populations of Atlantic, Mackinaw (*Salvelinus namaycush*) and sockeye salmon (*Oncorhynchus nerka*) are restricted to the South Island. Atlantic salmon are confined to the lakes and raceways of the upper Waiau River catchment by the dams of the central Otago

hydro-electric power system. A small broodstock is maintained at a Wanaka hatchery, but Atlantic salmon are essentially extinct in New Zealand fresh waters (NIWA 2018). A small population of Macinaw is confined to Lake Pearson in the Waimakariri River catchment, but a small broodstock population is held in several hatcheries. Sockeye salmon are confined to the Waitaki River catchment and also considered essentially extinct in New Zealand (NIWA 2018). Atlantic salmon, Mackinaw and sockeye salmon are uncommon, considered extinct or partially extinct in New Zealand. Accordingly, Atlantic salmon, Mackinaw and sockeye salmon are not regarded as potential hosts for exotic diseases in New Zealand.

Chinook (king) salmon (*Oncorhynchus tshawytscha*) occur throughout the South Island. Adults mature at sea, returning to fresh water to spawn. Brown trout, brook char and chinook (king) salmon are widespread throughout New Zealand (NIWA 2018) and represent potential hosts for fish pathogens. The introduction of exotic diseases would indirectly affect the farming of salmonids for recreational fishing, worth in excess of $80 million per annum (Fish & Game 2014), as well as affecting the tourism industries associated with salmon and trout fishing.

Non-salmonid fish have also been introduced into New Zealand freshwaters (Table 3). These include cyprinids: common and ornamental carp (*Cyprinus carpio*), grass carp (*Ctenopharyngodon idella*), orfe (*Leuciscus idus*), European perch (*Perca fluviatilis*), silver carp (*Hypophthalmichthys molitrix*), rudd (*Scardinius erythrophthalmus*), tench (*Tinca tinca*), mainly in North Island lakes and rivers. Other introduced species (including brown bullhead (*Ameiurus nebulosus*), mosquitofish (*Gambusia affinis*) and other poeciliids (*Phalloceros caudimaculatus Poecilia reticulata, P. latipinna, Xiphophorus helleri*) and goldfish (*Carassius auratus*) have little, if any value and are considered pests because they either reduce native biodiversity, degrade other species' habitats, or contribute to the decline of water quality in lakes (NIWA 2018).

5.3. Aquaculture in New Zealand

New Zealand aquaculture depends heavily on the provision of clean, disease-free water supplies. As the water supplies of New Zealand aquaculture are commonly untreated, hatchery facilities are at risk from aquatic pathogens (Sim-Smith *et al.* 2014).

Commercial aquaculture is concentrated on king salmon (*O. tshawytscha*). These are produced in freshwater hatcheries and on grown in 57 locations in freshwater or coastal marine waters of central and southern New Zealand. Exports of king salmon represented over $63 million in 2011 (Aquaculture New Zealand 2014).

Brown (*S. trutta*) and rainbow trout (*O. mykiss*) populations are maintained by significant hatchery operations that restock lakes and rivers throughout New Zealand for recreational fishing (Fish & Game 2014). Recreational trout fishing supports economically important local and associated tourist industries (Fish & Game 2014). While the dollar value of New Zealand's freshwater fisheries is not fully known, the Lake Taupo trout fishery alone is valued at $70 to 80 million per annum (Marsh & Mkwra 2013).

Other freshwater aquaculture species include grass carp (*C. idella*) and silver carp (*H. molitrix*) that are farmed for weed management in waterways (Mitchell 1980, 2008; Hofstra 2014; NIWA 2014b). While their economic value is unknown, these species play an important role in New Zealand waterway management. Eels (*Anguilla* spp.) (Anguillidae) have previously been

farmed commercially in New Zealand. While no commercial farming occurs at present, eel farming is currently under development (NIWA 2018).

Marine non-salmonid aquaculture under development in New Zealand include snapper (*Pagrus auratus*) (Sparidae), hapuku (*Polyprion oxygeneios* (Polyprionidae) and yellowtail kingfish (*Seriola lalandi*) (Carangidae) (LBAAF 2018; NIWA 2018).

5.4. Imports of fish into New Zealand

Imported fish for human consumption include marine, brackish and freshwater bony fish and elasmobranch species. The total amounts increased between 2013 and 2015 (Table 4), with considerable inter-annual variation in the most important species imported. As 27% of the commodity is not classified to family, these data are approximations and should be treated with caution.

Bony fish are imported in several processed states (Table 5), including whole fish (viscera, gills and head-on), headed and gutted (eviscerated, with head and gills removed), filleted, or further processed (other).

A relatively small amount of elasmobranch product is imported, mainly from families Callorhinchidae and Triakidae. These may be imported either as trunks, or as fillets.

In 2015, 32% of total fish were imported as whole fish. These fish, which include the viscera, contain a substantially higher pathogen load and represent an increased biosecurity risk (Oidtmann *et al*. 2017).

Table 4: Imports of Fish (T) by Family into New Zealand, 2013 – 2015 (Source: Statistics NewZealand, 2017)

Family	Year		
	2013	2014	2015
Anguillidae	<1	<1	0
Berycidae	<1	<1	6
Callorhinchidae	0	0	6
Carangidae	0	18	63
Centrolophidae	2	17	0
Cheilodactylidae	27	48	47
Clupeidae	1,053	993	734
Engraulidae	3	3	6
Epigonidae	0	<1	<1
Gadidae	785	307	469
Gempylidae	0	<1	<1
Latidae	0	0	0
Lophiidae	3	2	4
Merlucciidae	1,456	734	1,612
Monacanthidae	<1	<1	<1
Mugilidae	50	20	53
Nototheniidae	38	62	80
Ophidiidae	<1	0	7
Oreosomatidae	7	49	29
Pleuronectidae	<1	284	7
Salmonidae	177	1,064	579
Scombridae	422	1,191	1,697
Serranidae	1	<1	0
Soleidae	<1	0	0
Sparidae	1	18	36
Trachichthyidae	0	6	197
Triakidae	<1	0	0
Triglidae	0	21	0
Xiphiidae	7	12	44
Zeidae	93	116	49
Fish unspecified	1,787	2,161	2,096
Total imports (T)	7,321	8,316	7,820

Table 5: Imports of Fish (T) by Family and by Processed State (Whole, Headed & Gutted, Fillet, Other) During 2015 (Source: Statistics New Zealand, 2017)

Family	Whole	H&G	Fillet	Other	All
Anguillidae	0	0	0	0	0
Berycidae	6	0	0	0	6
Centrolophidae	<1	0	0	0	0
Callorhinchidae	0	6	0	0	6
Carangidae	63	0	0	0	63
Cheilodactylidae	0	0	47	0	47
Clupeidae	723	0	10	1	734
Engraulidae	0	0	6	0	6
Epigonidae	<1	0	0	0	0
Gadidae	0	<1	467	2	469
Gempylidae	0	0	0	0	0
Latidae	0	0	0	<1	0
Lophiidae	0	4	0	0	4

Table 5 (Continued)

Family	Whole	H&G	Fillet	Other	All
Merlucciidae	0	1	1,611	0	1,612
Monacanthidae	0	0	<1	0	0
Mugilidae	41	11	0	<1	53
Nototheniidae	0	0	1	79	80
Ophidiidae	<1	7	0	0	7
Oreosomatidae	1	0	28	0	29
Pleuronectidae	<1	<1	7	<1	7
Salmonidae	0	0	188	391	579
Scombridae	1,420	9	114	155	1,697
Soleidae	0	0	0	0	0
Sparidae	<1	0	0	36	36
Trachichthyidae	0	90	106	0	197
Triakidae	0	<1	0	0	0
Triglidae	0	0	0	0	0
Xiphiidae	0	6	38	0	44
Zeidae	0	0	49	0	49
Various	287	19	625	1,166	2,096
Total	2,541	153	3,295	1,830	7,820

6. Scope

This qualitative risk analysis assesses the biosecurity risks to animal health (including economic, environmental, social and cultural values) and to human health, that may be associated with the importation of eviscerated teleost fish, trunked elasmobranch fish and their products (including shark fins) imported for human consumption under the Biosecurity Act 1995[C]. These products may be from both farmed and wild stocks and be derived from marine, brackish or fresh waters.

6.1. Distribution pathways

The potential distribution pathways for non-viable fish products imported for human consumption are shown in Figure 1. The commodity may also re-directed post-border into other pathways (such as fish bait, stock feed, shark oil, sandpaper, leather, fish food, or pet food) (Skall *et al.* 2005; Cobb 2008; Oidtmann *et al.* 2011; Borzycem *et al*. 2013; Blackwell 2013). These pathways are not within the scope of this import risk analysis.

The quantity of fish product wastes derived from domestic food processing is so low as to be negligible (Appendix 1). This pathway is considered insignificant.

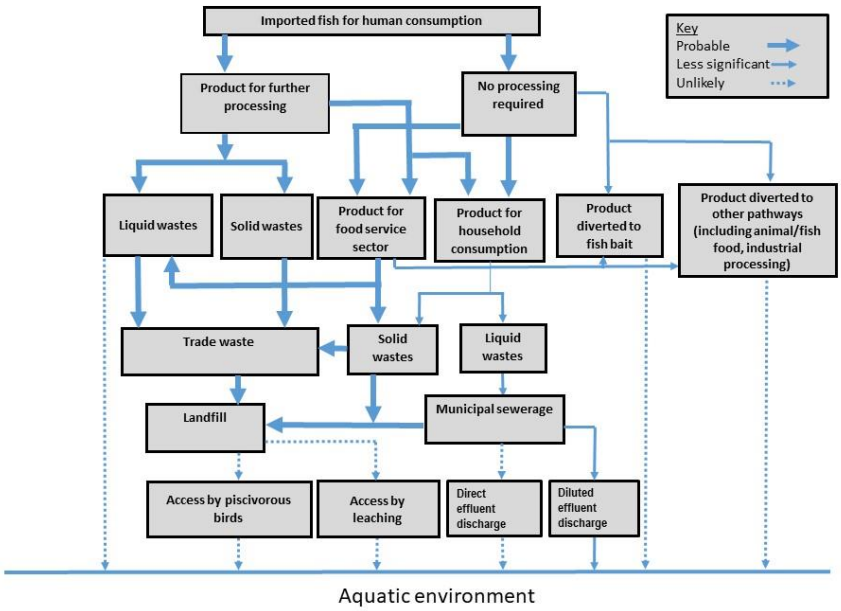
Minor pathways include liquid wastes from processing or transport, leaching from landfill, the activity of piscivorous and scavenging birds, or sewerage effluent discharges. These pathways associated with commercial processing are managed by the Transitional Facility regulations[D] (MPI 2016). They are considered where relevant for each identified pathogen.

Other pathways for pathogen introduction exist. These include the importation of live ornamental fish for aquarium display, for zoos or for biological research purposes (Hine & Diggles 2005; MPI 2010, 2017a, 2017b). These pathways are not within the scope of this import risk analysis, but diseases reported from ornamental fish are considered to be present in New Zealand, where evidence exists that the host species may have become established in the New Zealand environment.

[C] Biosecurity Act 1993, The Animal Products Act 1999, The Food Act 2014. The Parliamentary Counsel Office provides access to New Zealand legislation [Online] Available from: http://www.legislation.govt.nz/act

[D] Transitional Facilities for Animal Products, MPI-STD-ANIPRODS, Ministry for Primary Industries, Wellington, New Zealand, 18 p [Online] Available from: https://www.mpi.govt.nz/dmsdocument/12774/send [Accessed 14 April 2019].

Figure 1. Potential Distribution Pathways for the Commodity



6.2. Human health consequences

Fish and fish products may contain pathogenic organisms, which may also release chemicals into the fish that can cause allergic reactions in some people (Novoslavskij *et al.* 2015). In New Zealand, the Food Act 2014[C] and the Animal Products Act 1999[C] manage risks to public health associated with food. The human health consequences to New Zealand consumers from the consumption of imported fish and fish products (food safety) has previously been evaluated by MPI (2016) and more recently alongside this IRA. Parasites (nematodes, cestodes and trematodes), *Clostridium botulinum* and antimicrobial resistant variants of common pathogens were identified as hazards of concern. The US-FDA additionally identified histamine, scombroid toxin and ciguatoxin as significant hazards in finfish (FDA 2018). These are outside the scope of this IRA.

While human health considerations are noted in the consequence assessment of the relevant risk analysis chapters, the food safety risks and additional risk management measures related to imported fish and fish products have not been further considered in this IRA. Consignments of product imported into New Zealand for human consumption must meet the food safety and suitability requirements of this legislation, in addition to the requirements of the Biosecurity Act 1999. These requirements are independent of the IHS.

7. Commodity definition

Fish is defined in the Fisheries Act (1986)[E] as "*all species of finfish of the classes Agnatha, Chondrichthyes, and Osteichthyes, at any stage of their life history, whether living or dead*".

For the purposes of this risk analysis, fish is defined as "*limbless cold blooded vertebrate animals with gills and fins, living wholly in water; or the flesh of fish as food*" (Stevenson & Waite 2011). This includes bony or "teleost" fish (Class Actinopterygii) as well as cartilaginous sharks, skates, rays and chimaeras (Class Elasmobranchii), but excludes primitive vertebrates (Agnatha), as well as all other aquatic organisms, such as crustaceans and shellfish, mammals and amphibians (Fishbase 2018; WoRMS 2018).

7.1. Eviscerated fish of class Actinopterygii

The World Organisation for Animal Health (OIE) defines "eviscerated fish" as "*fish from which internal organs, excluding the head, brain and gills, have been removed*". This definition, which includes frozen fish, chilled fish, minced fish and mechanically recovered fish tissue[F] (OIE 2016a) has been used in this risk analysis.

7.2. Trunked cartilagenous fish of class Elasmobranchii

The OIE Aquatic *Code* does not specifically define appropriate processing of elasmobranchs, which differs from "evisceration" of teleosts (OIE 2016a). The blood and tissues of elasmobranchs contain high concentrations of urea (Wood *et al.* 1995; Musick 2005) which quickly breaks down post-mortem, releasing ammonia. This imparts a strong smell and tainting to the flesh and may be toxic at high concentrations (Musick 2005). It makes the product unpalatable and unfit for human consumption (Clarke *et al.* 2013), so elasmobranchs are routinely processed immediately after capture, to remove the blood from the tissues (Musick 2005). Processed elasmobranch tissues may also be salted, smoked or dried (Musick 2005). It is assumed in this risk analysis that all elasmobranch tissue product for human consumption has been processed as described below, before entering the human consumption pathway.

[E] The Fisheries Act (1986) and amendments. The Parliamentary Counsel Office provides access to New Zealand legislation [Online] Available from: http://www.legislation.govt.nz/act

[F] This assumes that mechanically recovered fish tissue for human consumption is derived from eviscerated fish product.

7.3. Chimaera, guitarfish, sharks and sawfish

For chimaera (Chimaeridae), guitarfish (Rhinobatidae), sharks (Selachii) and sawfish (Pristidae), the freshly caught carcass is routinely beheaded (removing the gills, brain and heart), eviscerated (removing the major body organs), trimmed (removing the lower body flaps, fins and tail), then thoroughly washed in seawater to remove the blood from the tissues. The remaining "trunk" is stored as chilled or frozen product (Musick 2005; Clarke *et al.* 2013).

7.3.1. Shark fins

Shark fins are processed to harvest the fine collagenous fibres or "needles" which support the margin of the shark fin. The cartilaginous fin base and all attached musculature are discarded as they cause spoilage of the shark fin product (Musick 2005). The "needles" are most numerous in the first dorsal fin, pectoral fins and the lower lobe of the caudal fin and when cooked and dried, these fins are sold as a complete set for each shark. The remaining fins (second dorsal fin, pelvic fins and the upper lobe of the caudal fin) are of much lower value. These are commonly sold in either processed or unprocessed form, as a bulk commodity (Musick 2005).

7.3.2. Skates and Rays

For skates (Rajidae), stingrays (Dasyatidae), eagle and manta rays (Myliobatidae) only the large muscular wings are removed from the carcass. These are thoroughly washed in seawater to remove the blood from the tissues. The "wings" are then stored as chilled or frozen product (Musick 2005; Clarke *et al.* 2013, 2014; FAO 2015).

8. Risk analysis methodology

8.1. General procedures

The methodology used in this risk analysis is guided by the *Biosecurity New Zealand Risk Analysis Procedures – Version 1* (Biosecurity New Zealand 2006), the Handbook on Import Risk Analysis for animals and animal products (OIE 2010) and in Chapter 2 of the *Aquatic Animal Health Code* (OIE 2016a). The risk analysis process comprises two main steps: Hazard identification, and risk assessment.

8.1.1. Hazard identification

A list of organisms of potential concern is compiled from the OIE list of aquatic animal diseases (OIE 2016a), published MPI risk analyses, and from the relevant published scientific literature, to represent the preliminary potential hazard list.

For each organism on the preliminary potential hazard list, several steps are completed (OIE 2016a). These include formal identification of the organism, its status as agent of an OIE listed disease, its New Zealand status, (present, under a control programme, or exotic), together with an assessment of the relevant aspects of the epidemiology and characteristics of the organism (Figure 2).

Hazard identification concludes with an assessment of whether or not the organism is identified as a hazard in the commodity. The results of the hazard identification are commonly summarised as a table. All hazards are subjected to risk assessment. The likelihood of each criterion being met will be different for each pathogen, host and country of origin (MacDiarmid 1994).

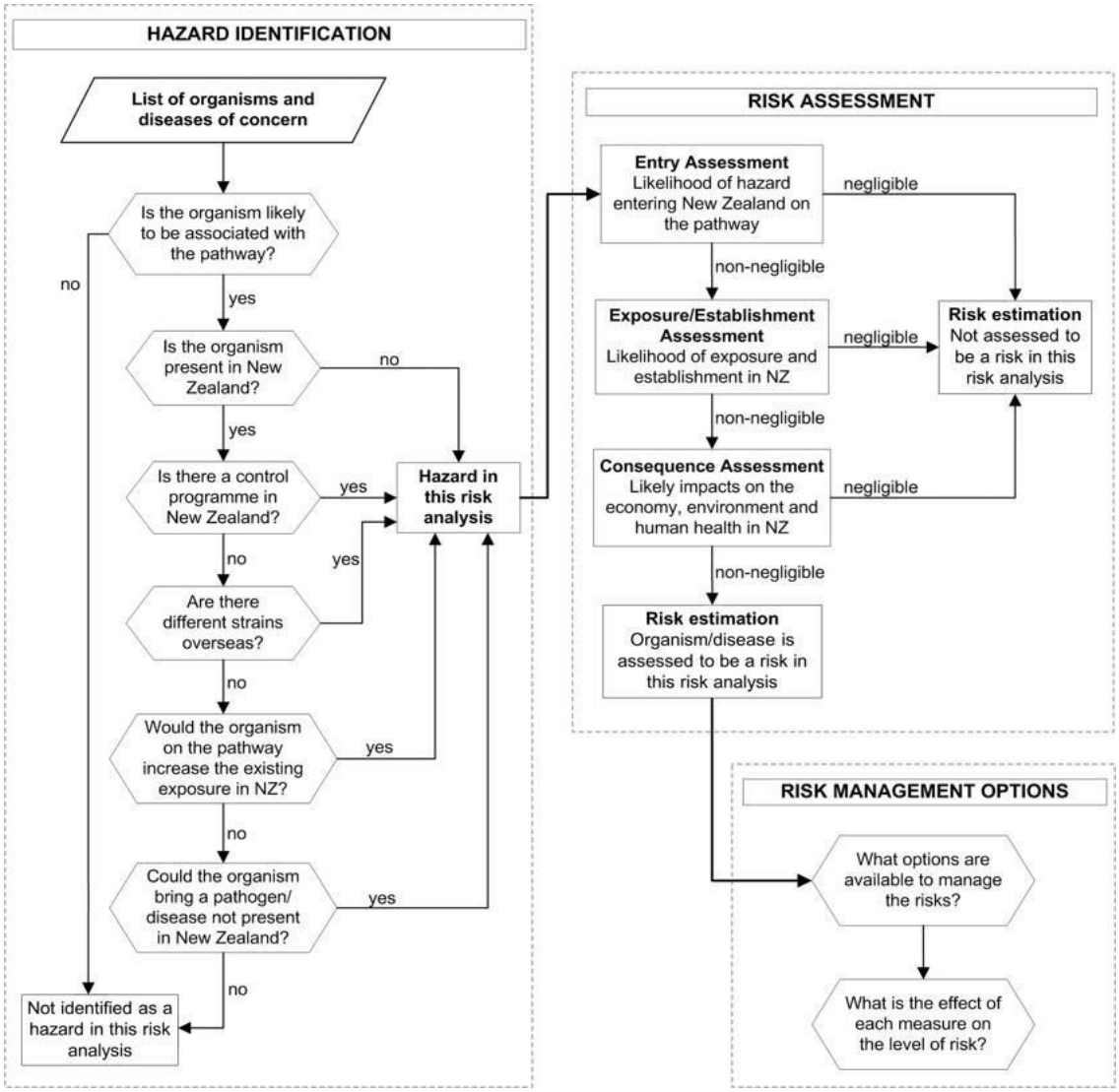
8.1.2. Risk assessment

Risk assessment (Figure 2) consists of four steps: entry assessment, exposure assessment, consequence assessment and risk estimation. At each of the first two steps, a qualitative assessment is made on the likelihood, based on the epidemiological information. Risk assessment concludes with an assessment of the risk associated with each identified hazard organism, based on the likelihood of entry and exposure, and the consequence assessments.

- *If the risk is assessed as negligible*, then the risk estimate is automatically negligible and the remaining steps of the risk assessment are redundant. The same situation arises when the likelihood of entry is non-negligible but the exposure assessment concludes that the likelihood of susceptible species being exposed is negligible, or when both entry and exposure are non-negligible but the consequences of introduction are assessed as negligible.

- *If the risk is non-negligible*, then the hazard is assessed as a risk. Risk management measures are considered to reduce the level of risk to an acceptable level.

Figure 2. The Risk Analysis Process



8.1.3. Risk management options

Risk management identifies the options available for managing that risk, based on the epidemiology of the risk organism. Where the OIE *Code* (OIE 2016a) lists recommendations for the management of a risk, these are described alongside options of similar, lesser or greater stringency, where available from the scientific literature. In addition to the options presented, unrestricted entry or prohibition may also be considered. Recommendations for the appropriate sanitary measures to achieve the effective management of risks are not made in this document. These will be determined when an IHS is drafted.

As obliged under Article 3.1 of the World Trade Organisation (WTO) SPS Agreement (WTO 2017), the measures adopted in IHSs will be based on international standards, guidelines and recommendations where they exist, except as otherwise provided for under Article 3.3. That is, measures providing a higher level of protection than international standards can be applied if there is scientific justification, or if there is a level of protection that the member country considers is more appropriate, that is based on a scientific risk analysis.

8.1.4. Risk communication

After a draft import risk analysis has been written, MPI analyses the options available and proposes draft measures for the effective management of the identified risks. These are then presented in a draft Import Health Standard (IHS) that is released for public comment, together with a risk management proposal (RMP) that summarises the options analysis, the rationale for the proposed measures and provides a link to the draft risk analysis.

Not every risk organism identified in the risk analysis may be associated with a particular imported fish species and require risk management in an IHS. The RMP will take into account specific information that would affect the need for risk management measures. For instance, factors considered in an RMP would include (but not be limited to) the country of origin of the commodity and the presence or absence of risk organisms in that country, from what species the commodity was derived, and any manufacturing processes that inactivate risk organisms.

The document package (draft IHS, RMP and draft risk analysis) is released for a pre-defined period of stakeholder consultation. Stakeholder submissions in relation to these documents are reviewed and published, including any supplementary risk analyses that may be required, before a final IHS is issued.

8.2. Risk analysis methods specific to this risk analysis

8.2.1. Hazard identification

For this risk analysis, organisms of potential concern were those identified from the OIE list of aquatic animal diseases (OIE 2016a), the peer-reviewed scientific literature, health databases, previous MPI risk analyses and information freely provided by the governments of other countries. In addition, diseases or disease agents suggested by MPI, experts and interested parties that were consulted or involved in reviewing this risk analysis were included.

Not all the organisms in the previous risk analyses are relevant to the commodities considered here. For this reason, organisms on the potential hazard list were then subjected to the following specific criteria, to rule out those organisms that were clearly not identified as hazards in the commodity.

OIE Status

Is the disease associated with the organism listed in the OIE Aquatic Code (OIE 2016a).

New Zealand Status

This reviews the organisms presence in New Zealand and whether it is under an Official Control programme. For the purposes of this risk analysis, if an organism has not been formally identified as being present in New Zealand, by notification in the New Zealand Organism Register (NZOR 2017), or in the scientific literature, then has been assumed to be exotic.

Presence in the Commodity

For the purposes of this risk analysis, pathogens reported to target the visceral organs and not otherwise reported from head, muscle or skin tissues, have been assumed to be present in negligible or insignificant quantities in the eviscerated or trunked commodity.

Detection by Inspection

Fish with obvious infections should not be present in the commodity (OIE 2016a), but pathogens may occur at sub-clinical levels of infection, disease may be in a latent or incubation phase, or disease may progress with few, or no external clinical signs. These carrier fish may be present in the commodity.

Opportunistic Organisms and Organisms that are not Associated with Significant Disease

Opportunistic pathogens may be present in the commodity, but only cause disease where the host is weakened or stressed. Infection may only occur following physical injury, or the organism may only be pathogenic following co-infection by another pathogen. For other organisms, infection may not cause significant disease. Opportunistic organisms and those of low pathogenicity are not considered further.

Presence of Virulent Exotic Strains

Whilst a pathogen may be present in New Zealand, more virulent exotic strains may have been identified that could be present in the commodity. These exotic strains are considered further.

Is a Vector or Intermediate Host Necessary to Complete the Life Cycle

Organisms with indirect life cycles require one or more intermediate hosts or vectors to complete their life cycle. Such organisms may be less likely to establish in New Zealand.

Potential Hosts in New Zealand

An organism may be present in the commodity, but the primary or intermediate host/vector necessary to complete the life cycle may not be present in New Zealand. Where no alternative intermediate host/vector exists, these organisms are unlikely to establish. These organisms are not considered further.

Zoonotic Potential

Some pathogenic organisms are also zoonotic (capable of causing human disease). These organisms are considered further.

8.2.2. Risk assessment

The methodology used herein assesses risk to be negligible, or non-negligible.

Entry Assessment

While fish showing signs of clinical disease are unlikely to pass visual inspection, larval stages of aquatic pathogens may not be visible in the commodity. In addition, sub-clinically infected hosts with little or no external signs of infection may pass visual inspection and be present in the commodity.

Exposure Assessment

In general, uncertainty exists on the infectious doses for marine/aquatic pathogens, as most available data have been determined in experimental studies. Where experimental methods

reasonably follow known exposure pathways, the host species are assumed to be susceptible to disease (OIE 2016a).

Consequence Assessment

Little published information is available to determine the social and environmental consequences of pathogen establishment. Where potential hosts are considered as having significant economic, social or environmental importance, the risk associated with the consequence assessment is assessed to be non-negligible.

8.2.3. Determination of Organisms of Potential Concern

The organisms of potential concern identified from the sources listed above, were collated to determine the preliminary hazard list of all pathogenic organisms reported to be associated with fish from wild and farmed eviscerated bony fish and trunked cartilaginous fish, sourced from marine, freshwater or brackish waters.

This preliminary hazard list was then subject to criteria as defined in Chapter 8, to rule out those organisms that were clearly not identified as hazards in the commodity, in the hazard identification table (Table 6). All organisms identified as hazards were subjected to risk assessment.

8.2.4. Summary Analysis of Risk Management Options

For each identified risk organism, a list of identified hosts are presented and the relevant risk management options are identified. Risk organisms are summarised by host fish family (Appendix 2 a-c) and by risk management option (Appendix 3). A qualitative evaluation of the risk management options is provided for each risk organism (Appendix 4). This summarises the expected levels of pathogen reduction for each risk management option, based on the epidemiology of the risk organism within the host.

The levels of pathogen reduction are approximated **as**:

Eliminate:	> 95% reduction in pathogen load
Substantial:	71-95% reduction in pathogen load
Moderate:	51-70% reduction in pathogen load
Slight:	< 50% reduction in pathogen load

No effect: Risk management option has no effect on pathogen load.

This evaluation is intended as a guide for selection of appropriate risk management options. When evaluating these options, consideration was given to the uncertainty around life cycle details and infection pathways, as outlined in the appropriate disease chapters.

9. Determination of preliminary hazards in the commodity

From the organisms of potential concern, a preliminary potential hazard list was completed for all pathogenic organisms reported to be associated with fish from wild and farmed eviscerated bony fish and trunked cartilaginous fish, sourced from marine, freshwater or brackish waters (Table 6). Following the criteria defined in Chapter 8, all organisms that were not hazards in the commodity were identified and excluded from further analysis. All organisms identified as hazards were subjected to risk assessment.

Table 6. Hazard Identification Table

Pathogen	Fish Host Family	Host Distribution	OIE Listed Disease	Pathogen Reported from New Zealand	Present in the Commodity	Likely to be Detected by Inspection	Opportunistic/ not Cause Significant Disease	Virulent Exotic Strains	Vector/ Intermediate Host Required	Potential Hosts in New Zealand	Zoonotic Potential	Retained for Risk Analysis	Reference
Adenovirus of dogfish, guitarfish	Rhinobatidae, Triakidae	Widespread	N	N	N (Not reported from wild populations)	Y (dermal lesions)	Y	N	N	Y	N	N	Paulin *et al.* 2001, Bowman *et al.* 2008, Garner 2013, Borocinska 2016, Camus *et al.* 2016
Adenovirus 2 (viral papilliomatosis)	Rhinidae	Africa, IndoPacific	N	N	Y	N	Y	N/A	N	N	N	N	Camus *et al.* 2016a
Atlantic salmon papillomatosis agent	Salmonidae	Europe	N	N	Y	N	Y	N/A	N	Y	N	N	Stone *et al.* 1997
American grass carp reovirus (AGCRV)	Cyprinidae	North America	N	N	Y	N	Y	N/A	N	Y	N	N	Plumb & Hanson 2011, Yan *et al.* 2014
Bohle iridovirus (Ranavirus) (BIV)	Latidae, Cichlidae	Asia, Australia	N	N	Y	N	Y	N/A	N	N	N	N	Whittington *et al.* 2010
Carp pox herpesvirus (CyHV-1)	Cyprinidae	Widespread	N	N	Y	N	Y	N/A	N	Y	N	N	Cobb 2008, Lepa & Siwicki 2012
Channel catfish viral disease (CCVD)	Ictaluridae (does not affect *Ameiurus nebulosus*)	Widespread	N	N	Y	N	N	N/A	N	N	N	N	Plumb 1989, Hanson *et al.* 2011
Chinook salmon paramyxovirus (CSPV)	Salmonidae	Pacific North America, Norway	N	N	Y	N	Y	N/A	N	Y	N	N	Cobb 2008, Lepa & Siwicki 2012
Dab ascites birnavirus	Pleuronectidae	Europe	N	N	N (viscera)	N/A	Y	N/A	N	Y	N	N	Munro & Midtyling 2011
Eel stomatopapilloma virus (EV-2)	Anguillidae	Widespread	N	N	Y	Y (dermal lesions)	Y	N/A	N	Y	N	N	Cobb 2008, Marino *et al.* 2010, Nagabayashi & Wolf 1979; Plumb & Hanson 2011
Epizootic haemopoietic necrosis virus (EHNV), including European catfish virus (ECV) and European sheatfish virus (ESV)	Salmonidae, Percidae, Galaxiidae, Ictaluridae	Australia	Y	N	Y	N	N	N/A	N	Y	N	Y	Whittington *et al.* 2010
Erythrocytic inclusion body virus (EIBS)/ Pacific salmon anaemia virus (PSAV), Heart and skeletal muscle inflammation (HSMI)	Wide teleost host range	Widespread	N	N	Y	N	Y	N/A	N	Y	N	N	Rodger 2007
European eel virus (EVE)	Anguillidae	Widespread	N	N	Y	N	N	N/A	N	Y	N	Y	Van Beurden *et al.* 2012

Pathogen	Fish Host Family	Host Distribution	OIE Listed Disease	Pathogen Reported from New Zealand	Present in the Commodity	Likely to be Detected by Inspection	Opportunistic/ not Cause Significant Disease	Virulent Exotic Strains	Vector/ Intermediate Host Required	Potential Hosts in New Zealand	Zoonotic Potential	Retained for Risk Analysis	Reference
European eel herpesvirus (EEHV)/ Eel herpesvirus (HVA)	Anguillidae	Europe	N	N	Y (latent infection)	Y (dermal lesions	N	N/A	N	Y	N	Y	Van Nieuwstadt *et al.* 2001, Van Ginnekin *et al.* 2004, Threader *et al.* 2010, Hanson *et al.* 2011
Eel virus European-X (EVEX)	Anguillidae	Europe	N	Y	Y	Y	Y	N/A	N	Y	N	N	Van Ginnekin *et al.* 2004
Goldfish herpesvirus (Cy-HV-2)	Cyprinidae:	Widespread	N	Y	Y	N	Y	N	N	Y	N	N	Stone *et al.* 1997, Hine *et al.* 2006
Graceful catshark herpacivirus (Flaviviridae)	Proscylliidae	Asia	N	N	N	N/A	N	N/A	N	N	N	N	Shi *et al.* 2015
Grass carp haemorrhagic virus (GCHV)	Cyprinidae	Widespread	N	N	Y	N	N	N/A	N	Y	N	Y	Jiang 2009, Cudmore & Mandrak 2011, Hofstra 2014
Grouper iridovirus	Serranidae	Asia, Australia	N	N	Y	N	N	N/A	N	Y	N	Y	Tubbs *et al.* 2007
Infectious haematopoietic necrosis virus (IHNV)	Wide teleost host range	Widespread	Y	N	Y	N	N	N/A	N	Y	N	Y	OIE 2016a
Infectious pancreatic necrosis virus (IPNV), including Halibut birnavirus (HBV), Tasmanian salmon aquabirnavirus (TaBV) and Viral deformity of yellowtail virus (VDV)	Wide teleost host range	Widespread	N	N	Y	N	N	N/A	N	Y	N	Y	Nakajima *et al.* 1998, Crane *et al.* 2000, McDowall 2000, McColl *et al.* 2009,Crane & Hyatt 2011, Diggles 2011, NIWA 2014
Infectious salmon anaemia virus (ISAV)	Wide teleost host range	Widespread	Y	N	Y	N	N	N/A	N	Y	N	Y	OIE 2016a
Koi herpesvirus (CyHV-3)	Cyprinidae	Widespread	Y	N	Y	N	N	N/A	N	Y	N	Y	OIE 2016a
Lymphocystis iridovirus	Carangidae, Zeidae	Widespread	N	Y	Y	N	Y	N/A	N	Y	N	N	Hine & Diggles 2005, Tubbs *et al.* 2007, Diggles 2011
Hirame rhabdovirus (HIRRV)	Wide teleost host range	Widespread	N	N	Y	N	N	N/A	N	Y	N	Y	Skall *et al.* 2005
Milkfish ulcer disease virus (Aquabirnaviridae)	Chanidae:	Asia	N	N	Y	N	Y	N	N	N	N	N	FAO 2012
New Japan virus of salmonids	Salmonidae	Japan	N	N	Y	N	N	N/A	N	Y	N	Y	Kahn *et al.* 1999

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Nodaviruses including viral nervous necrosis virus (VNN), viral encephalopathy and retinopathy (VER)	Wide teleost host range	Widespread	N	Y	N	N	N	N	N	Y	N	Y	Nguyen *et al*. 1996, Tubbs *et al.* 2007
Oncorhynchus masou virus (Herpesvirus type 2)	Salmonidae	Northern Pacific, Kuwait	N	N	Y	N	N	N/A	N	Y	N	Y	Yoshimizu 2012
Picornavirus and picorna-like (PKL) viruses	Anguillidae, Centrarchidae, Cyprinidae, Ictaluridae, Salmonidae	Widespread	N	Y (eels)	N (viscera)	N/A	Y (low mortality)	N	N	Y	N	N	Stone *et al.* 1997, Van Ginneken *et al.* 2004, Roberts 2012, Barbknecht *et al.* 2014
Pilchard herpesvirus	Clupeidae	Australia, New Zealand	N	Y	Y	N	Y	N	N	Y	N	N	Whittington *et al.* 1997
Pilchard orthomyxovirus (POMV), Tasmanian salmon orthomyxovirus (SOMV)	Clupeidae, Gadidae, Salmonidae	Australia	N	N	Y	N	N	N/A	N	Y	N	Y	Crane & Williams 2008, McDowall 2000, TASAL 2014, Huon 2014, Diggles 2016, B. Jones *pers.comm* 2017, PROMED 2017
Tilapia orthomyxovirus	Cichlidae	United States	N	N	Y	N/A	N	N/A	N	N	N	N	Bacharach *et al.* 2016
Piscine aquareovirus (PRV), including salmon, Tasmanian salmon (TasSRV), grass carp (GCRV) and turbot aquareoviruses (SMReV, TRV) / Heart and skeletal muscle inflammation syndrome (HSMI)	Cyprinidae, Retropinnidae, Salmonidae, Scophthalmidae,	Widespread	N	N	Y	N	N	N/A	N	Y	N	Y	Cobb 2008, King *et al.* 2011, Garseth *et al.* 2012, Roberts 2012, Carlile *et al.* 2014
Piscine myocarditis virus (cardiomyopathy syndrome), (Totiviridae)	Salmondiae, Argentinidae	Europe, Canada	N	N	N (viscera, very low titre in muscle)	N	N	N	N	N	N	N	Mikalsen *et al.* 2016
Plasmatoid leukaemia virus (PL)	Salmonidae	Canada, Australia	N	N	N (viscera, very low titre in muscle)	N	Y	N	N	Y	N	N	Eaton & Kent 1992, Eaton *et al.* 1994, MPI 1999, B. Jones *pers. comm* 2015
Rainbow trout intraerythrocytic virus	Salmonidae	Widespread	N	N	N (juvenile salmon)	N	Y	N	N	Y	N	N	Stone *et al*. 1997

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Red sea bream iridovirus (RSIV), Infectious spleen and kidney necrosis virus (ISKNV), dwarf gourami iridovirus (DGIV)	Wide teleost host range	Widespread	Y	N	Y (some)	N	N	N/A	N	Y	N	Y	OIE 2016a, Rimmer *et al.* 2015, 2017
Salmon alphavirus (SAV), Salmon pancreatic disease virus (SPDV)	Wide teleost host range	Widespread	Y	N	Y	N	N	N/A	N	Y	N	Y	Diggles 2011, OIE 2016a
Salmon gill poxvirus (carp oedema virus, koi sleepy disease)	Cyprinidae, Plecoglossidae, Salmonidae	Europe, Japan	N	N	Y	N	N	N/A	N	Y	N	Y	Cobb 2008, Lewish *et al.* 2015
Salmonid herpesvirus (HSV-(Type 1)	Salmonidae	United States, Japan	N	N	N (juveniles)	N/A	Y	N/A	N	Y	N	N	Stone *et al.* 1997, Sano *et al.* 1988, Hetrick & Sano 1989
Scale drop disease of barramundi (Megalocytiviridae)	Latidae	Southeast Asia (Malaysia, Singapore)	N	N	Y	N	N	N/A	N	N	N	N	Gibson-Kueh *et al.* 2012, de Groof *et al.* 2015
Smooth dogfish herpesvirus	Triakidae	North America	N	N	Y (skin)	Y (dermal lesions)	Y	N/A	N	Y	N	N	Leibovitz & Lebouitz 1985a, Bowman *et al.* 2008, Paulin *et al.* 2001, Garner 2013, Borocinsca 2016, Camus *et al.* 2016a, 2016b
Spring viraemia of carp virus (SVCV) including pike fry rhabdovirus (PFRV)	Wide teleost host range	Widespread	Y	N	Y	N	N	N/A	N	Y	N	Y	OIE 2016a
Tilapia lake virus (TiLV)	Cichlidae	Israel, South America, Egypt, Thailand	N	N	Y	N	N	N/A	N	N	N	N	Eynegor *et al.* 2014, FAO 2017, OIE 2017
Undiagnosed virus Y	Salmonidae	Norway	N	N	Y	N	Y	N/A	N	Y	N	N	Hjeltnes 2015
Viral erythrocytic necrosis virus (VEN), Piscine erythrocytic necrosis (PEN) (Iridoviridae)	Wide teleost host range, Triakidae	Widespread	N	N	Y (blood, gills, viscera, muscle)	N/A	Y	N/A	N	Y	N	Y	Reno *et al.* 1985, Meyers *et al.* 1986, Stone *et al.* 1997, Kahn *et al.* 1999, Cobb 2008, Glen *et al.* 2012, Winton & Hershberger 2014, Hick *et al.* 2016

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Viral haemorrhagic septicaemia virus (VHSV)	Wide teleost host range	Widespread	Y	N	Y	N	N	N/A	N	Y	N	Y	OIE 2016a
White sturgeon iridovirus (WSIVD)	Acipenseridae	Widespread	N	N	N	N	N	N/A	N	N	N	N	Hedrick *et al.* 1990, OIE 2016b
Yellowtail ascites virus (YAV)	Carangidae	Japan, Europe, Australia	N	N	Y (skin)	Y (skin lesions)	N	N/A	N	Y	N	N	Diggles 2003, Tubbs *et al.* 2007, Munro & Midtyling 2011
Bacterial pathogens													
Acinetobacter spp.	Wide teleost host range	Widespread	Y	Y	Y	N	Y	N/A	N	Y	N	N	Melville & Johnston 2010
Acinetobacter baumannii	Cyprinidae, Ictaluridae	China, Myanmar	N	N	N (viscera)	N/A	N	N/A	N	Y	N	N	Johnston 2008a, Melville & Johnston 2010, Abel *et al.* 2012
Actinomyces sp.	Cichlidae	Widespread	N	Y	N	N/A	Y	N	N	Y	N	N	Johnston 2008b, Gutierrez *et al.* 2013
Aeromonas hydrophila	Wide teleost host range; Lamnidae	Ubiquitous	N	Y	N (viscera)	N/A	Y	Y	N	Y	Y	Y	Boustead 1982, Tubbs *et al.* 2007, Johnston 2008a, Interaminense *et al.* 2010, Melville & Johnston 2010, DermNet NZ 2014, Kluzik & Woodford 2016,
Aeromonas sp.	Wide elasmoranch and teleost host range	Widespread	N	Y	Y	N	N	N	N	Y	Y	N	Youngren-Grimes 1990, Praveen *et al.* 2016, DermNet 2014
Aeromonas salmonicida var. *salmonicida* (atypical strains)	Wide teleost host range; Lamnidae	Widespread	N	N	Y	N	N	N/A	N	Y	N	Y	Briones *et al.* 1998, Lowry & Smith 2007, Tubbs *et al.* 2007, Keeling *et al.* 2013, Borucinska 2016
Aeromonas salmonicida var. *salmonicida* (typical strains)	Wide teleost host range	Widespread	N	N	Y	N	N	N/A	N	Y	N	Y	Lowry & Smith 2007, Tubbs *et al.* 2007, Keeling *et al.* 2013
Aeromonas sobria	Wide teleost host range	Widespread	N	Y	Y	N	Y	N	N	Y	Y	N	Diggles *et al.* 2002, Kluzik & Woodford 2016
Aliivibrio salmonicida var *salmonicida* (Hitra disease)	Gadidae (*Gadus morhua*), Salmonidae (*Salmo salar, Oncorhynchus mykiss*)	Widespread in marine waters	N	N	Y	N	Y	N/A	N	Y	N	N	McDowell 1976, Hine & Diggles 2005, Tubbs *et al.* 2007, Morris 2012, Haenen 2017
Alteromonas spp.	Lamnidae, Triakidae	Widespread	N	N	N (viscera)	N	N	N/A	N	Y	N	N	Borocinska & Frasca 2002a, Howgate 2010, Buller 2014

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Campylobacter spp.	Wide teleost host range	Widespread	N	Y	Y	N	Y	N	N	Y	Y	N	Kluzik & Woodford 2016
Carnobacterium sp.	Wide teleost host range	Widespread	N	Y	Y	N	Y	N	N	Y	N	N	Johnston 2008a
Carnobacterium maltaromaticum (=*Carnobacterium piscicola*)	Wide teleost host range, Lamnidae	Widespread	N	Y	Y (Teleosts) N (Lamnidae)	N	Y	N	N	Y	N	N	Stone *et al.* 1997, DermNet 2014
Chlamydia-like organisms (Uncultured fish Chlamydia-1 UFC1) (Bacterial epitheliocystosis)	Wide teleost host range, Triakidae	Widespread	N	Y	N (viscera)	N	Y	N	N	Y	N	N	Whittington *et al.* 1997, Nowak & Clark 2001, Nowak & LaPatra 2006, Polkingorne *et al.* 2010, Arkush & Bartholomew 2011, Stride & Nowak 2014, Haenen 2017
Chromobacterium violaceum	Cyprinidae	Widespread	N	N	Y	N	Y	N/A	N	Y	N	N	Midani & Rathore 1998, Melville & Johnston 2010, Abel *et al.* 2012
Chryseobacterium spp.	Wide fish host range	Widespread	N	Y	Y	N	N	N/A	N	Y	Y	N	Loch & Faisal 2015, NZOR 2018
Citrobacter freundii, Citrobacter sp.	Wide teleost host range, Lamnidae	Widespread	N	Y	N (viscera)	N	Y	N/A	N	Y	N	N	Wolf & Smith 2000, Cobb 2008, Toranzo *et al.* 1994, Interaminensce *et al.* 2010, DermNet 2014
Clostridium botulinum	Wide teleost host range	Widespread	N	Y (Not zoonotic types A & E)	Y	N	Y	N	N	Y	Y	N	Stone *et al.* 1997, Sobel 2005, Smyth *et al.* 2015
Cyanobacterium spp.	Carcharhinidae, Ginglymostomatidae, Sphyrnidae	Widespread	N	Y	N	N	Y	No	No	Y	N	N	*Paulin et al.* 2001, Wall *et al.* 2014, Hammerschlag *et al.* 2016
Cytophaga spp. (bacterial gill disease)	Wide teleost host range	Widespread	N	Y	Y	N	Y	N	N	Y	N	N	Stone *et al.* 1997, Tubbs *et al.* 2007, Starlipper & Schill 2011
Edwardsiella species complex (*E. angillulara, E. hoshinae, E. piscicida, E. tarda* and isolates)	Wide teleost host range, Dasyatidae	Widespread	N	N	Y	N	N	N/A	N	Y	Y	Y	Munday 2002, Johnston 2008a, Evans *et al.* 2011, Kluzik & Woodford 2016, Haenen 2017, Reichley *et al.* 2018
Enterobacter spp.	Wide teleost host range	Ubiquitous	N	Y	N (viscera)	N	Y	N	N	Y	Y	N	Heffernan *et al.* 2005, Cobb 2008, Sekar *et al.* 2008, Melville & Johnston 2010, Ikman 2013

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Epitheliocystis agent (*Chlamydia*-like organisms)	Wide teleost host range	Widespread	N	Y	Y	N	Y	N	N	Y	N	N	Whittington *et al.* 1997, Nowak & Clark 2001, Nowak & LaPatra 2006, Arkush & Bartholomew, Stride & Nowak 2014
Erysipelothrix rhusiopathiae	Wide teleost host range	Widespread	N	Y	Y	N	Y	N	N	Y	Y	N	DermNet 2014, NZOR 2017
Escherichia coli	Wide teleost host range	Widespread	N	Y	Y	N	Y	N	N	Y	Y	N	DermNet NZ 2014, NZOR 2017
Flavobacterium spp.	Wide teleost host range	Widespread	N	Y	Y	N	Y	N	N	Y	N	N	Stone *et al.* 1997, Starlipper & Schill 2011, Loch & Faisal 2015, Anon. 2016, Haenen 2017, NZOR 2017
Flavobacterium columnare (=*Cytophaga columnaris*)	Wide teleost host range	Widespread	N	Y	Y	N	N	Y	N	Y	N	Y	Stone *et al.* 1997, Michel *et al.* 2002, Johnston 2008a, Melville & Johnston 2010, Haenen 2017, NZOR 2017
Flavobacterium psychrophilum (=*Cytophaga psychrophila*)	Wide teleost host range	Widespread	N	Y	Y	N	Y	N	N	Y	N	N	Stone *et al.* 1997, Cobb 2008, Starliper & Schill 2011, MPI 2013, Haenen 2017, NZOR 2017
Francisella spp.	Wide teleost host range	Widespread	N	N	Y	N	N	N/A	N	Y	N	Y	Johnston 2008a, Colquhoun & Doudo 2011
Hafnia alvei	Wide teleost host range	Ubiquitous	N	Y	Y	N	Y	N	N	Y	Y	N	Austin 1983, Klapholz *et al.* 1994, Stone *et al.* 1997, Heffernan *et al.* 2005, NZOR 2017
Janthinobacterium lividum	Salmonidae	Widespread	N	N	Y	Y (skin lesions)	Y	N/A	N	Y	N	N	Austin 1983, Stone *et al.* 1997
Klebsiella spp.	Wide teleost host range	Widespread	N	Y	Y	N	Y	N	N	Y	Y	N	Anon. 2016, Kluzik & Woodford 2016, NZOR 2017
Lactococcus garvieae/ Lactococcus spp.	Wide teleost host range	Widespread	N	Y	Y	N	Y	N/A	N	Y	N	N	Diggles 2011, Gibello *et al.* 2016, Haenen 2017, NZOR 2017
Listeria monocytogenes	Wide teleost host range	Widespread	N	Y	Y	N	Y	N	N	N	Y	N	Boustead 1981, Rozman *et al.* 2016, NZOR 2017
Micrococcus spp.	Wide teleost host range, Lamnidae	Widespread	N	Y	Y	N	Y	N	N	Y	N	N	Johnston 2008a, Interaminensce *et al.* 2010, DermNet 2014
Moraxella spp.	Carcharhinidae, Ginglymostomatidae	Widespread	N	N	N (viscera)	N/A	N	N/A	N	Y	N	N	Austin & Austin 2007, Buller 2014

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Moritella (*Vibrio*) *viscosa*	Cyclopteridae, Gadidae, Mugilidae, Pleuronectidae, Salmonidae, Scophthalmidae	Widespread	N	N	Y	Y (skin lesions)	N	N/A	N	Y	N	Y	Austin & Austin 2007, Tubbs *et al.* 2007; Björnsdóttir 2011, Georgiades *et al.* 2016
Mycobacterium spp.	Wide elasmobranch and teleost host range	Widespread	N	Y	Y (skin, viscera)	Y (lesions on skin, mouth and gills, granuloma)	Y	N	N	Y	Y	N	Sinderman 1990, Shaperclaus 1992; Anderson 1997, Stone *et al.* 1997, Bruno *et al.* 1998, Montgomery 1998, Decostere 1999, Hine & Diggles 2005, Cobb 2008, Borucinska 2016, DermNet 2017, Haenen 2017
Plesiomonas shigelloides	Wide teleost host range	Widespread	N	Y	Y	N	Y	N	N	Y	Y	N	Staples 2000, Gonzalez-Rey *et al.* 2011, Griffiths 2015
Neisseria spp. (Meningococcal disease)	Carcharhinidae, Ginglymostomidae	Widespread	N	Y	N (viscera)	N/A	N	N	N	Y	Y	N	Frank & Jeffery 2000; Buller 2014, MOH 2018
Nocardia spp., *N. asteroides*	Wide teleost host range	Widespread	N	Y	Y	Y (skin lesions, ascites)	Y	N	N	N	Y	N	Boustead 1981, Stone *et al.* 1997, MAF 1999, Achla & Szyfres 2001, Labrie *et al.* 2008, Lewis & Chinabut 2011, Griffiths 2015
Photobacterium damselae, P. piscicida (=*Vibrio damsela*)	Wide elasmobranch and teleost host range	Widespread	N	Y	Y	N	Y	N	N	Y	Y	N	Paulin *et al.* 2001, Asato & Kayana 2004, Johnston 2008a, Buller 2014, NZ Fungi 2017
Piscirickettsia salmonis species complex, *Piscirickettsia*-like organisms,(PLOs), (*Rickettsia*-like organisms (RLOs), Tasmanian rickettsia-like organism,	Wide teleost host range	Widespread	N	Y (Official control)	Y	N	N	N/A	N (Alternative vectors present in New Zealand)	Y	N	Y	Kahn *et al.* 1999, Johnston 2008b, Corbeil & Crane 2009, Haenen 2017, Diggles 2011
Planococcus spp.	Salmonidae	Widespread	N	N	Y	Y (skin lesions, spots)	N	N/A	N	Y	N	N	Austin *et al.* 1988, Stone *et al.* 1997
Plesiomonas shigelloides	Wide teleost host range, Triakidae	Widespread	N	Y	Y	N	Y	N	N	Y	Y	N	Staples 2000, Paulin *et al.* 2001
Proteus spp. (= *Providencia rettgeri*)	Cyprinidae, Salmonidae	Ubiquitous	N	N	Y	Y (skin lesions)	Y	N/A	N	Y	Y	N	Austin & Austin 2007, Cobb 2008, Interaminensce *et al.*

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													2010, Daly & Akoi 2011, Chandler et al. 2006
Pseudomonas aeruginosa	Wide teleost host range	Widespread	N	Y	Y	N	Y	N	N	Y	Y	N	Johnston 2008a, 2008b, Haenen *et al.* 2013
Pseudomonas anguilliseptica	Wide teleost host range	Widespread	N	N	Y	N	N	N/A	N	Y	Y	Y	Tubbs *et al.* 2007, Haenen *et al.* 2013
Pseudomonas fluorescens	Wide teleost host range	Widespread	N	Y	Y	N	Y	N	N	Y	Y	N	Johnston 2008a, 2008b, Haenen *et al.* 2013
Pseudomonas sp.	Wide teleost host range, Lamnidae	Widespread	N	Y	Y	N	Y	N	N	Y	N	N	Interaminensce *et al.* 2010, Abrahaminian & Goldstein 2011, Buller 2014, Wall *et al.* 2014, DermNet 2014, Haenen 2017
Renibacterium salmoninarum	Wide teleost host range	Widespread	N	N	Y	N	N	N/A	N	Y	N	Y	Tubbs *et al.* 2007, Diggles 2011
Rhodococcus spp.	Salmonidae	Widespread	N	N	Y (ocular oedema)	N	Y	N/A	N	Y	N	N	Speare *et al.* 1992, Stone *et al.* 1997, Avendano-Herrera *et al.* 2011
Salmonella spp.	Wide teleost host range	Widespread	N	Y	Y	N	Y	N	N	Y	Y	N	Kluzik & Woodford 2016
Serratia marcescens	Moronidae, Sphyrnidae	Ubiquitous	N	Y	Y	Y (skin lesions)	Y	N	N	Y	N	N	Baya *et al.* 1992, Borucinska 2016, DermNet 2017
Serratia spp.	Cyprinidae, Salmonidae	Ubiquitous	N	Y	Y	Y (skin lesions)	Y	N	N	Y	N	N	Melville & Johnston 2010
Shewanella putrefaciens	Wide teleost host range, Triakidae	Widespread	N	Y	Y	N	Y	N	N	Y	Y	N	Kozinska & Pekala 2004, NZOR 2017
Staphylococcus spp.	Wide teleost host range, Lamnidae	Widespread	N	Y	Y	N	Y	N	N	Y	Y	N	Johnston 2008a, Interaminensce *et al.* 2010, ,Abrahaminian & Goldstein 2011
Stenotrophomonas maltophilia	Ictaluridae	Widespread	N	Y	Y	N	Y	N	N	Y	Y	N	Melville & Johnston 2010, Brooke 2012, NZ Fungi 2017
Strawberry disease of trout/ red mark syndrome (possible Rickettsia-like organism RLO)	Salmonidae	United Kingdom, North, South America	N	N	Y	N	Y	N/A	N	Y	N	N	Oidtmann *et al.* 2013b, Sandoval *et al.* 2016
Streptococcus (Enterococcus) spp., *S. agalactiae, S.*	Wide teleost host range	Widespread	N	Y	Y	N	Y	N	N	Y	Y	N	Sinton & Donnison 1994, MAF 2000, Paulin *et al.* 2001, Johnston 2008a,

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dysgalactiae, S. fascium, S. fascialis													McDougall *et al.* 2014, DermNet 2014
Streptococcus agalactiae (Lancefield Group B)	Anabatidae, Clariidae, Ginglymostomatidae	Widespread	N	Y	N (viscera)	N	Y	N	N	Y	Y	N	Carr *et al.* 2003, Kingklib & Suanyuk 2017, NZOR 2017
Streptococcus agalactiae (Group B type 283)	Channidae, Cyprinidae	Singapore	N	Y	Y (muscle)	N	N	Y	N	Y	Y	Y	Diggles 2011, Rajendran *et al.* 2016, NZOR 2017
Streptococcus iniae	Wide teleost host range	Widespread	N	N	Y	N	N	N/A	N	Y	Y	Y	Johnston 2008a, Kluzik & Woodford 2016
Tenacibaculum maritimus (=Cytophaga marina)	Wide teleost host range	Widespread	N	Y	Y	N	Y	N	N	Y	N	N	Anderson 1996, Diggles 2011, MPI 2013
Vibrio alginolyticus, V. harveyi (=V. carchariae)	Wide elasmobranch and teleost host range	Widespread	N	Y	Y	N	Y	N	N	Y	Y	N	Austin & Austin 2007, Noga 2014, Brosnahan *et al.* 2015, NZOR 2017
Vibrio (= *Listonella*) *anguillarum*	Wide teleost host range	Widespread	N	Y	Y	N	Y	Y	N	Y	Y	N	Powell & Loutit 1990, Stone *et al.* 1997, Kahn *et al.* 1999, Tubbs *et al.* 2007, Kluzik & Woodford 2016
Vibrio spp. *(V. cholera, V. ordalii, V. fluvialis, V. parahaemolyticus, V. vulnificus* (biotype 2)	Wide elasmobranch and teleost host range	Widespread	N	Y (including virulent Vp S03:tdh strain)	Y	N	Y	Y (but virulent Vp strains rare in fish)	N	Y	Y	N	Boustead 1981, Buck *et al.* 1984, Powell & Loutit 1990, Stone *et al.* 1997, Chisnall 2000, ESR 2001, Diggles *et al.* 2002, Upton & Taylor 2002, Huang & Wei-Hsu 2005, Abrahaminian & Goldstein 2011, Kirs et al. 2011; Whittaker 2013, Cruz 2015, Klluzik & Woodford 2016, Borucinska 2016, NZOR 2017
Yersinia ruckeri	Salmonidae	Widespread	N	Y	Y	N	N	Y	N	Y	N	Y	Stone *et al.* 1997, Diggles 2011, Barnes *et al.* 2016, Haenen 2017
Fungal and Pseudofungal pathogens													
Chromista (water moulds)													
Aphanomyces invadans (Epizootic ulcerative syndrome)	Wide teleost host range	Widespread	Y	N	Y	N	N	N/A	N	Y	N	Y	Johnston 2008a, OIE 2016a
Aphanomyces spp.	Wide teleost host range	Widespread	N	Y	N	N/A	N	N	N	Y	N	N	Johnston 2008a, 2008b

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Branchiomyces spp.	Wide teleost host range	Widespread	N	N (sub-tropical waters)	Y (gills)	N	Y	N	N	Y	N	N	Stone *et al.* 1997, *Tubbs et al.* 2007, Melville & Johnston 2010. Goodwin *et al.* 2012
Dermocystidium spp.	Wide teleost host range	Widespread	N	Y	Y	N	Y	N	N	Y	N	N	MacDiarmid 1994, Cobb 2008
Ichthyophonus hoferi	Wide teleost host range	Widespread	N	N	Y	N	N	N/A	N	Y	N	Y	Johnston 2008a, Diggles 2011
Sphaerothecum destruens (Rosette agent)	Wide teleost host range	Widespread	N	N	Y	N	N	N/A	N	Y	N	Y	Diggles 2011, Spikmans *et al.* 2013
Thraustochytriaceae													
Schizochytrium – like sp.	Salmonidae	United Kingdom	N	Y	Y	Y (epidermal lesions)	Y	N	N	Y	N	N	Humphrey 1995, Stone *et al.* 1997, FSANZ 2017
Saprolegniales													
Achlya spp.	Wide teleost host range	Widespread	N	Y	Y	N	Y	N	N	Y	N	N	Stone *et al.* 1997, Kumari & Kumar 2015
Leptolegnia spp.	Wide teleost host range	Widespread	N	Y	Y	N	Y	N	N	Y	N	N	Stone *et al.* 1997
Saprolegnia spp.	Wide teleost host range	Widespread	N	Y	Y	N	Y	N	N	Y	N	N	Boustead 1982, Stone *et al.* 1997, Diggles 2011
Leptomitales													
Leptomitus lacteus	Wide teleost host range (affects eggs/larvae only)	Widespread	N	N	N	N/A	Y	N/A	N	N	N	N	Humphrey 1995, Stone *et al.* 1997
Ascomycota incerta sedis													
Botrytis spp.	Wide teleost host range	Widespread	N	Y	N	N//A	Y	N	N	Y	N	N	Boustead 1982, Stone *et al.* 1997, NZOR 2017
Cladosporiaceae													
Ochroconis spp. (Phaeohyphomycosis)	Salmonidae, Sparidae, Sebastidae	Widespread	N	Y	Y	N	Y	N/A	N	Y	N	N	Stone *et al.* 1997, Wong *et al.* 2010, Samerpitak *et al.* 2013, NZOR 2017
Didymellaceae													
Peyronella (=Phoma) herbarum, Phoma spp.	Salmonidae (juveniles only)	United States United Kingdom	N	Y	N (viscera)	N/A	Y	N/A	N	Y	N	N	Boustead 1982, Stone *et al.* 1997, Humphrey 1995, Aveskamp *et al.* 2010, NZOR 2017

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Herpotrichiellaceae													
Exophiala pisciphila	Wide teleost host range, Triakidae	Widespread	N	Y	Y	N	Y	N	N	Y	N	N	Gaskins & Cheung 1986, Stone *et al.* 1997, MAF 2009, Seyedmousavi *et al.* 2011, NZOR 2017
Exophiala salmonis (Cerebral mycetoma)	Salmonidae, Ictaluridae, Paralichthyidae, Sillaginidae	Canada, Norway, Scotland, Faero Islands	N	N	N (viscera, rarely in muscle)	N	Y (farmed fish)	N/A	N	Y	N	N	Carmichael 1966, Fijan 1969, Stone *et al.* 1997, Reuter *et al.* 2003, Kurata *et al.* 2008, MAF 2009, de Hoog *et al.* 2011, Bruno 2016
Hyphomyceta													
Aspergillus spp.	Wide teleost host range	Widespread	N	Y	N	N/A	Y	N	N	Y	N	N	Boustead 1982, Stone *et al.* 1997
Isaria farinosa (=Paecilomyces farinosus)	Salmonidae	Widespread	N	N	N (viscera)	N/A	Y	N/A	N	N	N	N	Stone *et al.* 1997, Zimmermann 2010
Hypocreaceae													
Trichoderma spp.	Wide teleost host range	Widespread	N	Y	Y	N	Y	N	N	Y	N	N	Boustead 1982, Stone *et al.* 1997, Diggles 2011
Mucoraceae													
Rhizomucor spp. (Mucormycosis)	Wide teleost host range	Widespread	N	Y	Y	Y	Y	N	N	N	N	N	Johnston 2008a , Gomes *et al.* 2011
Nectriaceae													
Fusarium merismoides	Wide teleost host range	Widespread	N	Y	Y	N	Y	N	N	Y	N	N	Boustead 1982, Stone *et al.* 1997
Fusarium solani	Dasyatidae, Sphyrnidae	Widespread	N	Y	Y	Y (epidermis, gills)	Y	Y	N	Y	N	N	Pirat *et al.* 2016, DermNet 2014
Saccharomycetales incertae sedis													
Candida spp. (Canidiasis)	Salmonidae	Widespread	N	Y	N	N	N	N	N	Y	N	N	Stone *et al.* 1997, Cobb 2008, NZOR 2017
Microsporidia													
Glugea plecoglossi	Plecoglossidae, Salmonidae	Widespread	N	N	Y	N	N	N	N	Y	N	Y	Johnston 2008a
Glugea stephani	Pleuronectidae	Widespread	N	N	N (viscera)	N/A	N	N/A	N	Y	N	N	Dykova 2006
Glugea spp.	Wide teleost host range	Widespread	N	N	N (viscera)	N/A	Y	N	N	Y	N	N	Dykova 2006, Johnston 2008a
Kabatana arthuri	Pangasiidae	Widespread	N	N	Y	N	N	N	N	Y	N	Y	Johnston 2008b
Kabatana tadekai	Salmonidae	Japan	N	N	N (viscera)	N/A	Y	N	N	Y	N	N	Stone *et al.* 1997

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Loma salmonae/Loma fontinalis	Salmonidae	Widespread	N	N	N (viscera)	N/A	N	N	N	Y	N	N	Dykova 2006
Microsporidium rhabdophilia	Cichlidae, Salmonidae	Africa	N	N	N (viscera)	N/A	N	N	N	Y	N	N	FAO 2012
Microsporidium seriolae	Carangidae	Widespread	N	N	Y	N	N	N	N	Y	N	Y	Dykova 2006, Johnston 2008a
Microsporidium sp.	Triakidae	Widespread	N	Y	N (viscera)	N/A	N	N	N	N	N	N	Garner *et al.* 1998
Nucleospora (*Enterocytozoon*) *salmonis*	Salmonidae	Widespread	N	N	Y	N	N	N	N	Y	N	Y	Dykova 2006, Johnston 2008a
Pleistophora (*Heterosporis*) *anguillarum*	Anguillidae	Japan	N	N	Y	N	N	N	N	Y	N	Y	Hine & Diggles 2005, Johnston 2008a
Tetramicra brevifilum	Scophthalmidae	Widespread	N	N	Y	N	N	N	N	Y	N	Y	Dykova 2006, Johnston 2008a
Algae													
Blue green algae	Hexanchidae	Widespread	N	Y	N	N	Y	N	N	Y	N	N	Blasiola & Turnier 1979, Noga 2011
Protozoan pathogens													
Ciliates (Ciliophora)													
Prorodontida: Holophyridae													
Cryptocaryon irritans	Wide teleost host range, Triakidae	Widespread	N	Y	Y	N	Y	N	N	Y	N	N	Burgess & Matthews 1995, Colorni & Burgess 1997,Hine & Diggles 2005, Johnston 2008b
Dysteriidae: Hartmannulidae													
Brooklynella hostilis	Wide teleost host range	Ubiquitous	N	N	Y	N	Y	N	N	Y	N	N	Hine & Diggles 2005, Basson & Van As 2006, Pavlidis & Mylonas 2011
Brooklynella sp.	Heterodontidae	Widespread	N	Y	Y	N	Y	N	N	Y	N	N	Lom & Dykova 1992, Hine *et al*. 2000, Goertz 2004, Pavlidis & Mylonas 2011
Peritrichia: Trichodinidae													
Trichodina oviducti	Dasyatidae, Rajidae	Widespread	N	Y	N	N/A	N	N	N	Y	N	N	Kahn 1972, Hine *et al.* 2000
Trichodina rajae	Rajidae	Western Atlantic	N	Y	N	N/A	N	N	N	Y	N	N	Evdokimova *et al*. 1969, Hine *et al*. 2000, Goertz 2004
Trichodina spp.	Wide teleost host range	Widespread	N	Y	Y (skin)	N	Y	N	N	Y	N	N	Hine *et al.* 2000, Xu *et al.* 2002, Diggles 2011

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Tripartiella spp.	Wide teleost host range	Widespread	N	Y	Y (skin)	N	Y	N	N	Y	N	N	Johnston 2008b, Mohilal & Hemananda 2012
Peritrichia: Ellobiophryidae													
Caliperia brevipipes	Rajidae	Atlantic	N	N	N	N/A	N	N	N	Y	N	N	Laird 1959, Goertz 2004
Sessilida: Epistylidae													
Apiosoma spp.	Pangasiidae	Widespread in freshwater	N	N	Y (gills)	N	Y	N	N	N	N	N	Johnston 2008b
Epistylus spp.	Wide teleost host range	Widespread	N	Y	Y (skin)	N	Y	N	N	Y	N	N	Maskell 1886, Johnston 2008a, Valladao *et al.* 2014
Ciliophora: Scuticociliata													
Uronema marinum, Uronema spp.	Dasyatidae	Widespread	N	N	N (gills)	N/A	N	N	N	Y	N	N	Cheung 1993, Goertz 2004
Philasterides dicentrarchi	Heterodontidae, Stegosomatidae, Triakidae	Widespread	N	Y	Y	N	N	N	N	Y	N	N	Jung *et al.* 2007, Song *et al.* 2009, Stidworthy *et al.* 2017
Ciliophora: Chlamydodontidae													
Chilodonella spp.	Pangasiidae, Cyprinidae	Widespread	N	Y	Y (gills)	N	Y	N	N	N	N	N	Johnston 2008a, 2008b, Digges 2011
Ciliophora: Vestibuliferida: Balantiidae													
Balantidium spp.	Wide teleost host range	Widespread	N	N	N (viscera)	N/A	Y	N/A	N	N	Y	N	Stone *et al.* 1997, Johnston 2008b, Boylan 2011
Ciliophora: Hymenostomatida: Ichthyopthiuridae													
Ichthyophthirius multifiliis	Wide teleost host range	Widespread	N	Y	Y (skin)	N	Y	N	N	Y	N	N	Anderson 1996, Stone *et al.* 1997, Diggles 2011
Tetrahymena spp.(host of *Edwardsiella tarda, Listeria monocytogenes, Legionella pneumophila*)	Cyprinidae	Widespread	N	Y	Y (skin)	N	Y	N	N	Y	Y	N	Barker & Brown 1994, Dopheide *et al.* 2011
Amoebozoa													
Lobosa: Euamoebida: Hartmannellidae													
Hartmannella vermiformis (host to *Legionella pneumophila*)	Wide teleost host range	Widespread	N	Y	N (viscera)	N/A	Y	N	N	Y	Y	N	Barker & Brown 1994, Diggles *et al.* 2002, Johnston 2008a, MOH 2017
Mayorella-like spp.	Cichlidae	Europe	N	N	N (viscera)	N/A	Y	N	N	Y	N	N	Dykova *et al.* 2008, Johnston 2008a

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Platyamoeba-like spp.	Cichlidae	Europe	N	N	N (viscera)	N/A	Y	N	N	Y	N	N	Dykova *et al.* 20047, Johnston 2008a
Heterolobosea													
Rosculus ithacus	Cichlidae	Widespread	N	N	N (viscera)	N/A	Y	N	N	*N*	N	N	Johnston 2008a
Protozoa incertae sedis													
Amoeba spp.	Carcharhinidae, Stegostomatidae	Widespread	N	Y	Y	N	Y	N	N	Y	N	N	Goertz 2004
Neoparamoeba perurans / *Cochliopodida* sp. (amoebic gill disease)	Salmonidae	Widespread	N	Y	Y	N	Y	N	N	Y	N	N	Anderson 1996, Stone *et al.* 1997; Munday *et al.* 2001, Young *et al.* 2008, Diggles 2011, 2016
Paramoeba spp.	Wide teleost host range	Widespread	N	Y	Y	N	Y	N	N	*Y*	N	N	Stone *et al.* 1997
Dermamoebida: Dermamoebida incertae sedis													
Mayorella spp.	Cichlidae	Widespread	N	N	N (viscera)	N/A	Y	N/A	N	Y	N	N	Johnston 2008a
Discosea: Thecamoebidae													
Thecamoeba hoffmani	Salmonidae	Widespread in freshwater	n	N	N	N (juveniles)	N/A	N	N	N	N	N	Stone *et al.* 1997
Dinoflagellates													
Blastodiniales: Oodiniaceae													
Amyloodinium ocellatum	All elasmobranch and teleost fish	Widespread	N	N	Y (epidermis)	Y	Y	N	N	Y	N	N	Paperna 1980, Lom & Dykova 1992, Alvarez-Pellitero 2004, Hine & Diggles 2005
Dinoflagellata incertae sedis													
Piscinoodinium spp.	All fish	Widespread in freshwater	N	Y	Y (slimy skin)	Y	Y	N	N	Y	N	N	Austin *et al.* 1988, Hine & Diggles 2005, Stone *et al.* 1997
Protozoa: Trichozoa													
Distomatida: Hexamitidae													
Hexamita (*Spironucleus*) *salmonis*	Salmonidae	Widespread	N	N	N (viscera)	N/A	N	N/A	N	Y	N	N	Kent & Poppee 1998, Diggles 2011, Williams et al. 2011
Chromista: Apicomplexa: Myzozoa													
Eucoccidiorida: Cryptosporiidae													
Cryptosporidium spp.	Wide teleost host range	Widespread	N	N	N (viscera)	N/A	Y	N/A	N	Y	Y	N	Stone *et al.* 1997, Kahn *et al.* 1999, Certad *et al.* 2015

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Eucoccidiorida: Eimeriidae													
Eimeria anguillae	Anguillidae	Widespread	N	Y	N (viscera)	N/A	Y	N/A	N	Y	N	N	Stone *et al*. 1997, Kahn *et al*. 1999, Hine *et al*. 2000
Eimeria atherinae	Atherinidae	Widespread	N	N	N (viscera)	N/A	Y	N/A	N	N	N	N	Daoudi *et al.* 1987
Eimeria chollanensis	Urolophidae	Central America	N	N	N (viscera)	N/A	N	N/A	N	N	N	N	Upton *et al.* 1988
Eimeria eizeti	Myliobatidae	Europe	N	N	N (viscera)	N/A	N	N/A	N	Y	N	N	Upton *et al.* 1988
Eimeria gigantea	Lamnidae	Widespread	N	N	N (viscera)	N/A	N	N/A	N	Y	N	N	Levine 1985, Paulin *et al.* 2001
Eimeria lucida (=*E. scylli)*	Scyliorhinidae, Triakidae	Widespread	N	N	N (viscera)	N/A	N	N/A	N	Y	N	N	Boulard 1977, Lom & Dykova 1992, Goertz 2004
Eimeria ottojiroveci	Rajidae	Europe	N	N	N (viscera)	N/A	N	N/A	N	Y	N	N	Dykova & Lom 1983
Eimeria raiarum	Rajidae	Europe	N	N	N (viscera)	N/A	N	N/A	N	Y	N	N	Boulard 1977, Levine 1985
Eimeria southwelli (=*E. quentini*)	Myliobatidae	Widespread	N	N	N (viscera)	N/A	N	N/A	N	Y	N	N	Halawani 1930, Stamper *et al.* 1988, Cheung 1993, Dove *et al.* 2017
Eimeria squali	Triakidae	Widespread	N	N	N (viscera)	N/A	N	N/A	N	Y	N	N	Fitzgerald 1975
Eimeria vanasi	Cichlidae	Widespread	N	N	N (viscera)	N/A	N	N/A	N	Y	N	N	Johnston 2008b, Kahn *et al*. 1999, Hine & Diggles 2000
Eimeria zygaenae	Sphyrnidae	Widespread	N	N	N (viscera)	N/A	N	N/A	N	Y	N	N	Mandal & Chakravarty 1965
Eimeria sp.	Carcharhinidae	Widespread	N	N	N (viscera)	N/A	N	N/A	N	Y	N	N	Goertz 2004, Stidworthy *et al.* 2017
Goussia cichlidarum	Cichlidae	Widespread	N	N	N (viscera)	N/A	Y	N/A	N	N	N	N	Johnston 2008b, Kahn *et al.* 1999
Goussia gadi	Gadidae	Widespread	N	N	Y	N	Y	N/A	N	N	N	N	Molnar 2006
Eucoccidiorida: Haemogregarinidae													
Haemogregarina dasyatis	Rajidae, Triakidae	Widespread	N	N	N (blood)	N/A	Y	N/A	N	Y	N	N	So 1972, 1979, Kahn *et al.* 1980
Hemogregarina hemiscylli	Hemiscylliidae	Southeast Asia, Australia	N	N	N (blood)	N/A	Y	N/A	N	Y	N	N	Mackerras & Mackerras 1961
Haemogregarina lobianci, H. torpedinis	Wide elasmobranch host range	Europe	N	N	N (blood)	N/A	Y	N/A	N	Y	N	N	Neuman 1909, Kohl-Yakimoff & Yakimoff 1915, Goertz 2004
Haemogregarina sachai	Scophthalmidae	Widespread	N	N	N (blood)	N	Y	N	N	Y	N	N	Stone *et al*. 1997, Bricknell *et al*. 2005, Molnar 2006

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Apicomplexa: Haemohormiidae													
Haemohormidium sp.	Etmopteridae, Triakidae	Widespread	N	N	N (blood)	N/A	N	N	N	Y	N	N	Kahn *et al.* 1980, Clewley *et al.* 2002
Chromista: Bigyra: Opalinata													
Protoopalina spp.	Wide teleost host range	Widespread	N	Y	N (viscera)	N/A	Y	N	N	N	N	N	Stone *et al.* 1997, McKenna 2001, Johnston 2008b
Protozoa: Euglenozoa													
Eubodonida: Cryptobiaceae													
Cryptobia spp.	Anguillidae, Pangasiidae	Widespread	N	Y	Y (gills)	N	Y	N	N	N	N	N	Johnston 2008b
Kinetoplastidea: Ichthyobodonidae													
Ichthyobodo (=*Costia*) species complex	Wide teleost host range, Triakidae	Widespread	N	N	Y	N	Y	N/A	N	Y	N	N	Leibovitz & Lebovitz 1985b, Amlacher 1970, Hine *et al.* 2000, Paulin *et al.* 2001, Johnston 2008b, Isaksen 2011
Kinetoplastea: Trypanosomatidae													
Trypanosoma sp.	Wide elasmobranch & teleost host range	Widespread	N	Y	N (blood)	N/A	N	N	N	Y	N	N	Laird 1952, Paulin *et al.* 2001, Johnston 2008b,
Trypanosoma carchariasi	Carcharhinidae	Widespread	N	Y	N (blood)	N/A	N	N	N	Y	N	N	Hine *et al.* 2000, Paulin *et al.* 2001
Trypanosoma gargantua	Hemiscylliidae, Rajidae	Australia	N	Y	N (blood)	N/A	N	N	N	Y	N	N	Laird 1951, Mackerras & Mackerras 1961, Paulin *et al.* 2001
Trypanosoma giganteum	Rajidae	Widespread	N	N	N (blood)	N/A	N	N/A	N	Y	N	N	Neuman 1909
Trypanosoma humboldti	Scyliorhinidae	South America	N	N	N (blood)	N/A	N	N/A	N	Y	N	N	Morillas *et al.* 1987
Trypanosoma marplantensis	Dasyatidae	Indo-Pacific, Australia	N	N	N (blood)	N/A	N	N/A	N	Y	N	N	Bacigalupo & de la Plaza 1948
Trypanosoma rajae	Etmopteridae, Rajidae	Widespread	N	N	N (blood)	N/A	N	N/A	N	Y	N	N	So 1972, Kahn *et al.* 1980
Trypanosoma scylii	Scyliorhinidae	Europe, Africa	N	N	N (blood)	N/A	N	N/A	N	Y	N	N	Pulsford 1984
Cnidaria: Myxozoa, Myxosporea													
Ceratomyza abbreviata, C. attenuata, C. sphairophora	Carcharhinidae	Widespread	N	N	N (viscera)	N/A	N	N/A	N	Y	N	N	Gleeson & Adlard 2011
Ceratomyxa auerbachi	Clupeidae	Widespread	N	N	N (viscera)	N/A	Y	N/A	Y	Y	N	N	Koie *et al.* 2008

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Ceratomyxa buri	Carangidae	Japan	N	N	N (viscera)	N/A	N	N/A	N	Y	N	N	Yokoyama & Fukuda 2001, Woo & Bruno 2014
Ceratomyxa carcharhini, C. curvata, C. flagellifera, C. lunata, C. melanopteri, C. negaprioni	Carcharhinidae	Widespread	N	N	N (viscera)	N/A	N	N/A	N	Y	N	N	Gleeson & Adlard 2011
Ceratomyxa filiformis	Chimaeridae	Northeast Pacific	N	N	N (viscera)	N/A	N	N/A	N	Y	N	N	Gleeson & Adlard 2011
Ceratomyxa jamesoni	Triakiae	North America	N	N	N (viscera)	N/A	N	N/A	N	Y	N	N	Gleeson & Adlard 2011
Ceratomyxa mesospora, C. recurvata	Sphyrnidae	Widespread	N	N	N (viscera)	N/A	N	N/A	N	Y	N	N	Eiras 2006
Ceratomyxa riolae	Carangidae	Japan	N	N	N (viscera)	N/A	N	N/A	N	Y	N	N	Lom & Dykova 2006
Ceratomyxa seriolae	Carangidae	Widespread	N	N	N (viscera)	N/A	N	N/A	N	Y	N	N	Yokoyama & Fukuda 2001
Ceratomyxa shasta	Salmonidae	Widespread	N	N	Y (gills)	N	N	N/A	N	Y	N	Y	Lom & Dykova 2006
Ceratomyxa sphaerulosa (=*C. orientalis*)	Triakidae	Widespread	N	N	N (viscera)	N	N	N/A	N	Y	N	N	Tolonen & Karlsbakk 2003, Eiras 2006, Koie *et al.* 2008
Ceratomyxa sp.	Pangasiidae	Southeast Asia	N	N	N (viscera)	N	N	N/A	N	N	N	N	Johnston 2008a
Chloromyxum clavatum, C. dogieli	Rajidae	Europe	N	N	N (viscera)	N	N	N/A	Y	Y	N	N	Kovaljova 1988, Gleeson & Adlard 2011, Rocha *et al*. 2016
Chloromyxum hemiscyllii	Hemiscylliidae	Western Pacific, Australia	N	N	N (viscera)	N	N	N/A	Y	N	N	N	Gleeson & Adlard 2011
Chloromyxum kuhlii	Dasyatidae	Southwest Pacific/ Australia	N	N	N (viscera)	N	N	N/A	Y	N	N	N	Gleeson & Adlard 2011
Chloromyxum levigatum	Squatinidae	Eastern Pacific	N	N	N (viscera)	N	N	N/A	Y	N	N	N	Jameson 1931
Chloromyxum lesteri	Scylliorhinidae	Australia	N	N	N (viscera)	N	N	N/A	Y	N	N	N	Gleeson & Adlard 2011
Chloromyxum leydigi	Somniosidae, wide elasmobranch host range	Widespread	N	N	N (viscera)	N	N	N/A	Y	Y	N	N	Gleeson & Adlard 2011
Chloromyxum liae	Carcharhinidae	Widespread	N	N	N (viscera)	N	N	N/A	Y	Y	N	N	Kuznetsova 1977
Chloromyxum lissosporum, C. multicostatum	Squatinidae	Eastern Atlantic	N	N	N (viscera)	N	N	N/A	Y	N	N	N	Kovaljova 1988

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Chloromyxum mingazzinii	Pristiophoridae	Southern Australia	N	N	N (viscera)	N	N	N/A	Y	N	N	N	Gleeson & Adlard 2011
Chloromyxum sp., *C. myliobati*	Myliobatidae, Triakidae	Southern Australia	N	Y	N (viscera)	N	N	N/A	Y	Y	N	N	Gleeson & Adlard 2011
Chloromyxum nobelei	Dasyatidae, Hemiscyllidae	Northern Australia	N	N	N (viscera)	N	N	N/A	Y	N	N	N	Gleeson & Adlard 2011
Chloromyxum ovatum	Triakidae	Widespread	N	N	N (viscera)	N	N	N/A	Y	Y	N	N	Gleeson & Adlard 2011
Chloromyxum parvicostatum	Pristiophoridae	Southern Australia	N	N	N (viscera)	N	N	N/A	Y	Y	N	N	Kuznetsova 1977
Chloromyxum riorajum	Arhynchobatidae	South America	N	N	N (viscera)	N	N	N/A	Y	N	N	N	Azevado *et al.* 2009
Chloromyxum scyliorhinum	Scyliorhynidae	South Korea, Japan, Taiwan	N	N	N (viscera)	N	N	N/A	Y	N	N	N	Noble 1948
Chloromyxum schulmani	Rajidae	Africa	N	N	N (viscera)	N	N	N/A	Y	Y	M	N	Kovaljova 1988
Chloromyxum sphyrnae	Sphyrnidae	Widespread	N	N	N (viscera)	N	N	N/A	Y	Y	N	N	Gioia & Corderio 1996
Chloromyxum striatellus	Scyliorhynidae	Northeast Atlantic	N	N	N (viscera)	N	N	N/A	Y	N	N	N	Kovaljova 1988
Chloromyxum testeri	Scyliorhinidae	Australia	N	N	N (viscera)	N	Y	N/A	Y	Y	N	N	Gleeson & Adlard 2011
Chloromyxum transversocostatum	Triakidae	South America	N	N	N (viscera)	N	Y	N/A	Y	Y	N	N	Kuznetsova 1977
Enteromyxum fugu	Tetraodontidae	Asia	N	N	Y	N	Y	N/A	N	Y	N	Y	Paulin *et al.* 2001
Enteromyxum leei	Sparidae	Widespread	N	N	Y (gills, muscle)	N	N	N/A	N	Y	N	Y	Feist & Longshaw 2006, Johnston 2008b, Woo & Bruno 2014
Enteromyxum scophthalmi	Scophthalmidae	Widespread	N	N	Y	N	N	N/A	N	Y	N	Y	Redondo *et al.* 2002
Hennegoides malayensis	Pangasiidae	Southeast Asia	N	N	Y (gills)	N	N	N/A	N	N	N	N	Molnar *et al*. 2006, Melville & Johnston 2010
Hennegoides berlandi	Pangasiidae	Southeast Asia	N	N	Y (gills)	N	N	N/A	N	N	N	N	Molnar *et al*. 2006, Melville & Johnston 2010
Hennegoides pangasii	Pangasiidae	Southeast Asia	N	N	Y (gills)	N	N	N/A	N	N	N	N	Molnar *et al*. 2006, Melville & Johnston 2010
Henneguya creplini	Cyprinidae	Widespread	N	N	Y (gills)	N	N	N/A	N	Y	N	Y	Feist & Longshaw 2006
Henneguya excilis	Ictaluridae	Widespread	N	N	Y (gills)	N	N	N/A	N	Y	N	Y	Eiras 2002
Henneguya ghaffari	Percidae	Widespread	N	N	Y (gills)	N	N	N/A	N	Y	N	Y	Eiras 2002
Henneguya ictaluri	Ictaluridae	Widespread	N	N	Y (gills)	N	N	N/A	N	Y	N	Y	Wise *et al.* 2008
Hennegya nuesslini	Salmonidae	Widespread	N	N	Y	N	N	N/A	N	Y	N	Y	Feist & Longshaw 2006
Henneguya piaractus	Cichlidae	Brazil	N	N	Y (gills)	N	N	N/A	N	Y	N	Y	Johnston 2008b

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Henneguya salminicola (=H. zschokkei)	Salmonidae	Widespread	N	N	Y	N	N	N/A	N	Y	N	Y	Feist & Longshaw 2006
Henneguya shariffi	Pangasiidae	Southeast Asia	N	N	Y (gills)	N	N	N/A	N	N	N	N	Johnston 2008b
Henneguya waltairensis	Channidae	India	N	N	Y (gills)	N	N	N/A	N	N	N	N	Kalavati & Narasimhamurti 1985
Henneguya sp.1	Anguillidae	Widespread	N	Y	Y	N	N	N/A	N	Y	N	N	Hine & Diggles 2000, Johnston 2008b
Henneguya sp.2	Siluridae	Widespread	N	N	Y	N	N	N/A	N	N	N	N	Martins *et al.* 2004
Henneguya sp. 3	Characidae	South America	N	N	Y (gills)	N	N	N/A	N	N	N	N	Barassa *et al.* 2003
Kudoa clupeidae	Clupeidae	Widespread	N	N	Y	N	N	N/A	N	Y	N	Y	Reimschuessel *et al.* 2003
Kudoa lemniscati	Lutjanidae	Asia (Absent from mainland New Zealand)	N	N	Y	N	N	N/A	N	N	N	N	Paulin *et al.* 2001
Kudoa neothunni	Carangidae	Western Pacific	N	N	Y	N	N	N/A	N	Y	N	Y	Kasai *et al.* 2017
Kudoa paniformis	Merlucciidae	Widespread	N	N	Y	N	N	N/A	N	Y	N	Y	Stehr & Whittaker 1986
Kudoa spp.	Wide elasmobranch and teleost host range	Widespread	N	N	Y	N	N	N/A	N	Y	N	Y	Feist & Longshaw 2006, Gleeson &Adlard 2012, Stideworthy *et al.* 2017
Kudoa thyrsites	Wide teleost host range	Widespread	N	N	Y	N	N	N/A	N	Y	N	Y	Stehr & Whittaker 1986
Myxidium spp.	Wide teleost host range	Widespread	N	N	N (viscera)	N/A	N	N/A	N	Y	N	N	Feist & Longshaw 2006, Woo & Bruno 2014
Myxidium giardi (= *M. zealandicum*), *M. rhodei*	Wide teleost host range	Widespread	N	Y	N (viscera)	N/A	N	N	N	Y	N	N	Hine *et al.* 2000
Myxobolus aeglefini	Wide teleost host range	Widespread	N	N	Y	N	N	N/A	N	Y	N	Y	Feist & Longshaw 2006
Myxobolus arcticus	Salmonidae	Japan	N	N	Y	N/A	N	N/A	N	Y	N	Y	Feist & Longshaw 2006
Myxobolus baskai	Pangasiidae	Southeast Asia	N	N	Y (gills)	N	N	N/A	N	N	N	N	Melville & Johnston 2010
Myxobolus bramae	Cyprinidae	Widespread	N	N	Y	N	N	N/A	N	N	N	Y	Eszterbauer *et al.* 2000
Myxobolus buckei	Cyprinidae	Widespread	N	N	Y	N	N	N/A	N	Y	N	Y	Feist & Longshaw 2006
Myxobolus buri	Carangidae	Widespread	N	N	N (viscera)	N/A	N	N/A	N	Y	N	N	Woo & Bruno 2014

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Myxobolus artus, M. carassii. M. cultus, M. drjagini, M. ergensi, M. ginbuna, M. hungaricus, M. intimus, M. koi, M. macrocapsularis, M.paratoyami, M. parviformis, M. pavlovski, M. pfeifferi, portucalensis, M. pseudodispar (=*M. cyprini*), M. tanakai,	Cyprinidae	Widespread	N	N	Y	N	N	N/A	N	Y	N	Y	Ogawa *et al.* 1992, Eiras *et al.* 2005, Feist & Longshaw 2006, Kato 2017
Myxobolus cerebralis	Salmonidae	Widespread	N	Y	Y	N	N	N	N	Y	N	N	Stone *et al.* 1997, Tubbs *et al.* 2007; Kahn *et al.* 1999
Myxobolus (*Ceratomyxa*) *dogieli*	Cyprinidae, Rajidae	Widespread	N	N	N (viscera)	N/A	N	N/A	Y	Y	N	N	Bedell 1971, Molnar *et al.* 2008, Bauer *et al.* 1991, Rocha *et al.* 2016
Myxobolus ergensi	Cyprinidae	Widespread	N	N	N (viscera)	N/A	N	N/A	Y	Y	N	N	Eszterbauer *et al.* 2001
Myxobolus omari	Pangasiidae	Malaysia	N	N	Y	N	Y	N/A	N	N	N	N	Melville & Johnston 2010
Myxobolus pangasii	Pangasiidae	Southeast Asia	N	N	Y (eye, muscle, skin)	N	Y	N/A	Y	N	N	N	Feist & Longshaw 2006, Johnston 2008b, Melville & Johnston 2010
Myxobolus spp.	Wide teleost host range	Worldwide	N	Y (some)	Y	N	N	N	Y	Y	N	Y	Feist & Lonshaw 2006, Melville & Johnston 2010
Myxobolus spirosulcatus	Carangidae	Widespread	N	N	N (viscera)	N/A	N	N/A	N	N	N	N	Woo & Bruno 2014
Parvicapsula spp.	Wide teleost host range	Widespread	N	N	N (viscera)	N/A	N	N/A	N	Y	N	N	Woo & Bruno 2014
Parvicapsula pseudobranchicola,	Salmonidae	Widespread	N	N	Y (gills)	N	N	N	N	Y	N	Y	Woo & Bruno 2014
Sphaerospora spp.	Wide host range	Widespread	N	N	N (viscera)	N/A	N	N/A	N	Y	N	N	Johnston 2008a, Woo & Bruno 2014
Sphaerospora chinensis, S. molnari, S. renicola	Cyprinidae	Widespread	N	N	Y (gills)	N	N	N/A	N	Y	N	Y	Woo & Bruno 2014
Sphaerospora truttae	Salmonidae	Widespread	N	N	N (viscera)	N	N	N	N	Y	N	N	Woo & Bruno 2014
Tetracapsula bryosalmonae	Salmonidae	Widespread	N	N	N (viscera)	N/A	N	N/A	N	Y	N	N	Woo & Bruno 2014
Thelohanellus hovorkai, T. nikolskii, Thelohanellus spp.	Cyprinidae	Widespread	N	N	Y	N	N	N/A	N	Y	N	Y	Yokoyama *et al.* 1998, Woo & Bruno 2014

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Unicapsula spp.., *andersenae, U. chirocentrusi, U. galatea, U. marquesi, U. muscularis, U. pacifica, U. pflugfelderi. U. pyrimidata, U. seriolae, U. schulmani, U. stenolepis*	Wide elasmobranch and teleost host range	Widespread	N	N	Y	N	N	N/A	N	Y	N	Y	Borokinska & Fraska 2002, Feist & Longshaw 2006; Gleeson & Adlard 2012, Miller & Adlard 2013, Woo & Bruno 2014, Yokoyama *et al.* 2014
Zschokkella spp.	Wide teleost host range, Triakidae	Widespread	N	Y	N (viscera)	N/A	N	N/A	N	Y	N	N	Lom & Dykova 1995, Hine & Diggles 2000, Benz & Bullard 2004, Woo & Bruno 2014
Annelid pathogens													
Cancellariidae													
Cancellaria cooperii	Squatinidae, Torpedinidae	Widespread	N	N	Y (skin)	N	Y	N/A	N	Y	N	N	Benz & Bullard. 2004
Glossosiphoniidae													
Batrachobdelloides tricarinata	Bagridae, Clariidae, Cichlidae, Cyprinidae, Mormyridae, Synodontidae	Widespread	N	N	Y (skin)	N	Y	N/A	N	Y	N	N	Oosthuizen 1989, Paperna 1996
Hemiclepsis lucida, H. marginata, H. quadrata	Esocidae	Europe	N	N	Y (skin)	N	Y	N/A	N	Y	N	N	Moore 1939, Khalifa 1985, Oosthuizen 1987,Paperna 1996
Piscicolidae													
Branchellion torpedinis, B. ravenelii, Branchellion sp.	Scophthalmidae, Labridae, wide elasmobranch host range	Widespread	N	Y	Y (skin)	N	Y	N	N	Y	N	N	Hine *et al.* 2000, Benz & Bullard. 2004, Goveditch 2017, NZOR 2017
Myzobdella sp.	Cichlidae, Mugilidae	Widespread	N	N	Y (skin)	N	Y	N/A	N	Y	N	N	Khalifa 1985
Piscicola geometra	Wide teleost host range	Ubiquitous	N	N	Y (skin)	N	Y	N/A	N	Y	N	N	Burrelson 2006, Johnston 2008b
Phyllobdella sp.	Cyprinidae, Mormyridae	Widespread	N	N	Y (skin)	N	Y	N/A	N	Y	N	N	Moore 1939, Khalifa 1985, Paperna 1996
Undescribed piscicolids	Mugilidae	Widespread	N	N	Y (skin)	N	Y	N/A	N	Y	N	N	Moore 1939, Kabata 1985

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Nematode pathogens													
Acanthocheilus rotundatus, Terranova galeocerdonis (=*Porocaecum galeocerdonis, P. rochalimai*)	Carcharhinidae, Sphyrnidae	Widespread	N	Y	N (viscera)	N/A	Y	N/A	N	Y	Y	N	Hine *et al.* 2000, Moravec & Justine 2006, Samshi *et al.* 2018
Adenocephalus pacificus	Triakidae	Widespread	N	N	N (viscera)	N	Y	N/A	Y	Y	Y	N	Kuchta *et al.* 2015, Scholz & Kuchta 2016
Anguillicola crassus	Wide teleost host range	Widespread	N	N	Y	N	N	N/A	Y	Y	N	Y	Lefebre *et al*. 2004; Yanong 2011
Anisakis spp.	Wide elasmobranch & teleost host range	Widespread	N	Y	Y	N	N	N/A	Y	Y	Y	N	Hine *et al.* 2000, Borocinska & Frasca 2002b,Pascual *et al.* 2018
Bothriocephalus acheilognathi	Cyprinidae	Widespread	N	Y	N (viscera)	N/A	N	N/A	Y	Y	Y	N	Hine *et al.* 2000,
Camallanus spp.	Wide teleost host range	Widespread	N	Y	Y	N	N	N/A	Y	Y	N	N	Hine *et al.* 2000, D'Silva *et al.* 2012
Capillaria spp.	Clupeidae	Bangladesh, India	N	Y	N (viscera)	N	N	N/A	Y	Y	Y	N	Hine *et al.* 2000, D'Silva *et al.* 2012, Lindquist & Cross 2017
Contracaecum spp.	Wide teleost host range	Widespread	N	N	Y	N	N	N/A	Y	Y	Y	N	Johnston 2008b, Shamsi *et al.* 2018
Cucullanus spp.	Wide teleost host range	Widespread	N	Y	Y	N	N	N/A	Y	Y	N	N	Hine *et al.* 2000
Diphyllobothrium spp.	Wide teleost host range	Widespread	N	N	Y	N	N	N/A	Y	Y	Y	N	Scholz & Kuchta 2016
Diplogonoporus spp.	Clupeidae, Scombridae	Japan	N	N	Y	N	Y	N/A	Y	Y	Y	N	Scholz & Kuchta 2016
Echinocephalus overstreeti	Dasyatidae	Asia-Pacific	N	N	N (viscera)	N/A	N	N	Y	Y	N	N	Beveridge 1991, Moravec & Justine 2006
Echinocephalus sinensis, E. uncinatus	Myliobatidae	Widespread	N	N	N (viscera)	N/A	N	N	Y	Y	N	N	Beveridge 1985, Moravec & Justine 2006
Goezia bangladeshi	Clupeidae	Bangladesh, India	N	N	N (viscera)	N/A	N	N/A	Y	Y	Y	N	D'Silva *et al.* 2012, Bhuiyan 2013
Gnathostoma spp.	Wide teleost host range	Widespread	N	N	Y	N	N	N/A	Y	Y	Y	N	Lette & Temesgen 2007, Lindquist & Cross 2017
Huffmanela carcharhinae, H. lata	Carcharhinidae	Widespread	N	N	Y (skin)	Y (dermal lesions)	Y	N/A	Y	N	N	N	Huffman & Moravec 1988, Moravec 2001, Paulin *et al.* 2001, Justine 2005
Hysterothylacium spp. (=Maricostula spp.)	Wide teleost host range	Widespread	N	Y	Y	N	N	N/A	Y	Y	Y	N	Hine *et al.* 2000, Smith *et al*. 2007, D'Silva *et al.* 2012, Shamsi *et al.* 2018

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Ligula spp.	Cyprinidae	Widespread	N	Y	Y	N	N	N/A	Y	Y	Y	N	Hine *et al.* 2000, Scholz & Kuchta 2016
Pancreatonema americanum	Rajidae, Triakidae	Widespread	N	N	N (viscera)	N/A	N	N/A	Y	Y	N	N	Moravec *et al.* 2001, Borucinska & Frasca 2002b, McVicar & Gibson 1975, Benz & Bullard 2004
Parascarophis sphyrnae	Sphyrnidae	Widespread	N	N	N (viscera)	N/A	N	N	Y	Y	N	N	Knoff *et al.* 2001a, Millar 2016
Phlyctainophora lamnae, P. squali	Triakidae	Widespread	N	Y	Y (skin)	N/A	N	N	Y	Y	N	N	Adamson *et al.* 1987, Jones & Delahunt 1995
Philometra spp.	Wide teleost host range	Widespread	N	Y	Y	N	N	N	Y	Y	Y	N	Hine *et al.* 2000, Dadar *et al.* 2016
Porrocaecum galeocerdonis, P. rochalimi	Carcharhinidae, Sphyrnidae	Widespread	N	Y	Y	N	N	N	Y	Y	Y	N	Hine *et al.* 2000, Youn 2009
Porrocaecum spp.	Clupeidae	Bangladesh, India	N	Y	N (viscera)	N	N	N/A	Y	Y	Y	N	Hine *et al.* 2000, D'Silva *et al.* 2012
Procamellanus spp.	Wide teleost host range	Widespread	N	Y	N (viscera)	N	N	N	Y	Y	N	N	Hine *et al.* 2000. Arthur & Ahmed 2002
Procamellanus pereirai	Arhynchobatidae	South America	N	N	N (viscera)	N/A	Y	N/A	Y	N	N	N	Knoff *et al.* 2001a
Pseudoterranova spp.	Wide teleost host range	Widespread	N	Y	Y	N	N	N	Y	Y	Y	N	Hine *et al.* 2000, Shamsi & Suthar 2016, Hine *et al.* 2000,
Pulchrascaris chiloscyllae	Sphyrnidae, Squatinidae	Europe, Africa	N	Y	N (viscera)	N/A	Y	N	Y	Y	Y	N	Deardorf 1987, Hine *et al.* 2000, Tanzolla & Sardella 2006, Shamsi & Suthar 2016
Terranova gingylmostomae, T. pristus, T. scoliodontis, Terranova sp.	Wide elasmobranch host range	Widespread	N	Y	N (viscera)	N/A	Y	N	Y	N	Y	N	Hine *et al.* 2000, Borocinska & Fraska 2002b, Moravec & Justine 2006, Shamsi *et al.* 2018
Unidentified larval nematodes	Wide elasmobranch and teleost host range	Widespread	N	N	Y	N	N	N/A	Y	Y	Y	N	Benz *et al.* 1987, Hine *et al.* 2000, Molnar 2006, Johnston 2008a, 2008b, Borocinska & Adams 2013, Shamsi & Suthar 2016
Platyhelminth pathogens													
Turbellaria													
Micropharynx parasitica	Rajidae	Eastern Atlantic	N	N	Y (skin)	N	Y	N/A	N	Y	N	N	Ball & Khan 1975, Benz & Bullard 2004

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Monogenea													
Actinocleidus fusiformis, A. gracilus, A. leiognathidae	Centrarchidae, Leiognathidae	India	N	N	Y	N	Y	N/A	N	N	N	N	Chaudry *et al.* 2013
Alcopenteron ureteroecetes	Wide host range	Widespread	N	N	N (viscera)	N/A	Y	N/A	N	Y	N	N	Buchmann & Bresciani 2006, Petrie-Hanson 2011.
Allencotyla mcintoshi	Carangidae	Widespread	N	N	Y	N	N	N/A	N	Y	N	Y	Yamaguti 1963, Montero *et al.* 2003
Allobivagina sp.	Siganidae	Europe	N	N	Y	N	N	N/A	N	N	N	N	Buchmann & Bresciani 2006
Allopyragraphorus hippos	Carangadae	Americas	N	N	Y	N	N	N/A	N	Y	N	Y	Kritsky *et al.* 2011
Ancyrocephalus chakrabartii	Cyprinidae	India, Asia	N	N	Y	N	N	N/A	N	Y	N	Y	Chaudry *et al.* 2013
Ancylodiscoides vistulensis	Siluridae	Europe	N	N	Y	N	N	N/A	N	N	N	N	Buchmann & Bresciani 2006
Anoplodiscus australis	Sparidae	Australia	N	Y	Y	Y	Y	N/A	N	Y	N	N	Roubal & Whittington 1990, Tubbs *et al.* 2007
Anoplodiscus cirrusspiralis	Sparidae	New Zealand	N	Y	Y	N	Y	N	N	N	N	N	Hine *et al.* 2000, Tubbs *et al.* 2007
Anoplodiscus richardii	Sparidae	Australasia	N	Y	Y	N	Y	N	N	N	N	N	Roubal & Whittington 1990
Anoplodiscus spari	Sparidae	Japan	N	Y	Y	N	Y	N	N	N	N	N	Ogawa & Egusa 1981
Anoplodiscus tai	Sparidae	Japan	N	N	Y	N	N	N/A	N	Y	N	Y	Ogawa & Egusa 1981
Anthocotyle merlucci	Wide teleost host range	Widespread	N	N	Y	N	Y	N/A	N	Y	N	N	Baldwin *et al.* 2011
Benedenia monticelli	Mugilidae	Widespread	N	N	Y	Y (skin infection)	Y	N/A	N	Y	N	N	Buchmann & Bresciani 2006. Ogawa 2012
Benedenia sekii	Sparidae	Widespread	N	Y	Y	N	Y	N	N	Y	N	N	Hine *et al.* 2000
Benedenia seriolae	Carangidae	Widespread	N	Y	Y	N	Y	Y	N	Y	N	N	Hine *et al.* 2000
Benedeniella posterocolpa	Myliobatidae, Rajidae	Widespread, New Zealand	N	N	Y (skin)	N	Y	N/A	N	Y	N	N	Lawler 1981 (Not reported from New Zealand)
Bifurcohaptor spp.	Cichlidae	Widespread	N	N	Y (skin)	N	N	N/A	N	Y	N	Y	Johnston 2008b
Bivagina spp.	Sparidae	Australia, New Zealand	N	Y	Y	N	Y	N	N	Y	N	N	Hine *et al.* 2000
Bychowslyella spp.	Clariidae	Southeast Asia	N	N	Y	N	Y	N/A	N	N	N	N	Woo & Bruno 2014
Calicobenedenia polyprioni	Percichthyidae	Widespread	N	N	Y	N	N	N/A	N	Y	N	Y	Kritsky & Fennessy 1999
Cemocotyle noveboracensis	Carangidae	North America	N	N	Y	N	N	N/A	N	Y	N	Y	Kritsky *et al.* 2011
Chauhanellus alatus	Ariidae	India	N	N	Y	N	Y	N/A	N	N	N	N	Chaudhary *et al.* 2013a

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Choricotyle chrysophri	Sparidae	Widespread	N	N	Y	N	N	N/A	N	Y	N	Y	Sanchez-Garcia *et al.* 2014
Cichlidogyrus spp., *C. tilapiae*	Cichlidae	Widespread	N	N	Y	N	N	N/A	N	Y	N	Y	Buchmann & Bresciani 2006
Clavunculus bursatus	Centrarchidae	United States	N	N	Y	N	Y	N/A	N	N	N	N	Collins & Janovy 2003
Cleidodiscus pricei	Cyprinidae	United States	N	N	Y	N	N	N/A	N	Y	N	Y	Bauer 1991
Dactylogyrus sp., *D. anchoratus, D. aristichthys, D. batae, D. bifurcatus, D. brevitubus, D. calbasi, D. chagunionis , D. chauhani, D. ctenopharyngodonis, D. extensus, D. fotedari, D. glossogobii, D. hypophthalmichthys, D. inexpectatus, D. kalyanensis, D. labei, D. lamellatus, D. lampam, D. leptobarbus, D. lohanii, D. minutus, D. mrigali, D. nobilis, D. scriabini, D. speciosus, D. suchengtaii, D. vastator, D. vicinus, D. yogendrai*	Wide teleost host range	Widespread	N	Y (some)	Y	N	N	Y	N	Y	N	Y	Hine *et al.* 2000, Buchmann & Bresciani 2006
Dawestrema cycloancistrium	Arapaimidae	Widespread	N	N	Y	N	Y	N/A	N	N	N	N	Santos *et al.* 2017
Dermophthirius nigrelli, D. penneri	Carcharhinidae	Widespread	N	N	Y (skin)	N	Y	N/A	N	Y	N	N	Grimes *et al.* 1985, Bullard *et al.* 2000a, Benz 1987
Dermophthirioides pristidis, Neoheterocotyle inpristi	Pristidae	Widespread	N	N	Y (skin)	N	Y	N/A	N	N	N	N	Cheung & Nigrella 1983
Dionchus postoncomiracidia	Carcharhinidae	Widespread	N	N	Y (skin)	N	Y	N/A	N	Y	N	N	Bullard et al. 2000b
Diplectanum sp.	Wide teleost host range	Widespread	N	N	Y	N	N	N/A	N	Y	N	Y	Buchmann & Bresciani 2006
Diplozoon indicum	Cyprinidae	India	N	N	Y	N	N	N/A	N	Y	N	Y	Prakash 2015
Discocotyle sagitata	Salmonidae	Widespread	N	N	Y	N	N	N/A	N	Y	N	Y	Buchmann & Bresciani 2006

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Dogielius catalius	Cyprinidae	India	N	N	Y	N	N	N/A	N	Y	N	Y	Chaudhary *et al.* 2013
Encotyllable spari	Sparidae	Pacific	N	N	Y	N	N	N/A	N	Y	N	Y	Robinson 1961
Engraulicola forcepopenis	Cyprinidae	India	N	N	Y	N	Y	N/A	N	Y	N	N	Chaudhary 2013
Enterogyrus cichlidarum	Cichlidae	Widespread	N	N	N (viscera)	N/A	N	N/A	N	Y	N	N	Buchmann & Bresciani 2006, Petrie-Hanson 2011
Entobdella soleae	Soleidae	Europe	N	N	Y	N	Y	N/A	N	Y	N	N	Kearn *et al.* 1993
Gyrodactylus anguillae	Anguillidae	Widespread	N	N	Y	N	N	N/A	N	Y	N	Y	Buchmann & Bresciani 2006
Gyrodactylus bychowskii	Salmonidae	Europe	N	N	Y	N	N	N/A	N	Y	N	Y	Buchmann & Bresciani 2006
Gyrodactylus cichlidarum	Cichlidae	Widespread	N	N	Y	N	N	N/A	N	Y	N	Y	Mousavi *et al.* 2012, Garcia-Vasquez *et al.* 2011
Gyrodactylus colemanensis	Salmondiae	North America	N	N	Y	N	N	N/A	N	Y	N	Y	Buchmann & Bresciani 2006
Gyrodactylus derjavini	Salmondae	Europe	N	N	Y	N	N	N/A	N	Y	N	Y	Buchmann & Bresciani 2006
Gyrodactylus elegans	Salmonidiae	Widespread	N	N	Y	N	N	N/A	N	Y	N	Y	Buchmann & Bresciani 2006
Gyrodactylus groschafti	Clariidae	Africa	N	N	Y	N	Y	N/A	N	N	N	N	Buchman & Bresciani 2006
Gyrodactylus katharineri	Cyprinidae	Europe	N	N	Y	N	N	N/A	N	Y	N	Y	Buchmann & Bresciani 2006
Gyrodactylus longipipes	Sparidae	Europe	N	N	Y	N	N	N/A	N	Y	N	Y	Paladini *et al.* 2011
Gyrodactylus orecchiae	Sparidae	Europe	N	N	Y	N	N	N/A	N	Y	N	Y	Paladini *et al.* 2011
Gyrodactylus salaris	Wide host range	Widespread	Y	N	Y	N	N	N/A	N	Y	N	Y	Buchmann & Bresciani 2006
Gyrodactylus salmonis	Salmonidae	North America	N	N	Y	N	N	N/A	N	Y	N	Y	Buchmann & Bresciani 2006
Gyrodactylus sp.	Wide teleost host range	Widespread	N	Y	Y	N	N	N/A	N	Y	N	Y	Diggles *et al.* 2002
Haplocleidus furcatus	Centrarchidae	North America	N	N	Y	N	Y	N/A	N	N	N	N	Hoffman 1967
Haplocleidus vachi	Cyprinidae	Widespread	N	N	Y	N	N	N/A	N	Y	N	Y	Hoffman 1967
Heteraxine heterocerca	Carangidae	Japan	N	N	Y	N	N	N/A	N	Y	N	Y	Buchmann & Bresciani 2006
Heterobothrium okamotoi	Tetraodontidae	Japan	N	N	Y	Y (dermal lesions)	N	N/A	N	Y	N	N	Buchmann & Bresciani 2006; Ogawa 2012
Lamellodiscus (*Furnestia*) *echeneis*	Sparidae	Europe	N	N	Y	N	N	N/A	N	Y	N	Y	Buchmann & Bresciani 2006
Laticola latesi	Latidae	Asia	N	N	Y	N	N	N/A	N	N	N	N	Tingbao *et al.* 2006

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Leptocotyle minor	Scyliorhinidae	Europe, North Africa	N	N	Y (skin)	N	Y	N/A	N	N	N	N	Kearn 1965
Mazocraes mamaevi, M. singhi	Cyprinidae	India	N	N	Y	N	N	N/A	N	Y	N	Y	Chaudhary *et al.* 2013
Metamicrocotyla macraecantha	Mugilidae	North America	N	N	Y	N	Y	N/A	N	Y	N	N	Baker *et al.* 2008
Microcotyle sebastis	Sebastidae	North America	N	N	Y	N	N	N/A	N	Y	N	Y	Buchmann & Bresciani 2006
Neobenedenia hirame	Paralichthyidae	Asia	N	N	Y	N	Y	N/A	N	Y	N	N	Buchmann & Bresciani 2006
Neobenedenia melleni	Wide teleost host range	Widespread	N	N	Y	N	N	N/A	N	Y	N	Y	Buchmann & Bresciani 2006; Ogawa 2012
Neoheterobothrium hirame	Paralichthyidae	Asia	N	N	Y	N	Y	N/A	N	Y	N	N	Buchmann & Bresciani 2006
Neomazocraes anadontosomae, N. sardinopsi	Clupeidae	Widespread	N	N	Y	N	Y	N/A	N	Y	N	N	Reed *et al.* 2012
Onchocleidus ferox, O. principalis	Centrarchidae	Widespread	N	N	Y	N	N	N/A	N	N	N	N	Hoffman 1967
Pangasitrema spp.	Pangasiidae	Southeast Asia	N	N	Y	N	N	N/A	N	Y	N	Y	Johnston 2008b
Paradactylogyrus catalius	Cyprinidae	India	N	N	Y	N	N	N/A	N	Y	N	Y	Chaudhary *et al.* 2013
Paramazocraes gorakhanati	Cyprinidae	India	N	N	Y	N	N	N/A	N	Y	N	Y	Chaudhary *et al.* 2013
Paramicrocotyloides reticularis	Carangidae	Widespread	N	Y	Y	N	Y	N/A	N	Y	N	N	Diggles & Hutson 2005, Smith *et al.* 2007
Protomicrocotyle mirabilis	Carangidae	Widespread	N	N	Y	N	N	N/A	N	Y	N	Y	Wahl 1971
Pseudodactylogyrus anguillae, P. bini,	Anguillidae	Widespread	N	N	Y	N	N	N/A	N	Y	N	Y	Umeda *et al.* 2006
Pseudo-rhabdosynochus spp.	Serranidae	Widespread	N	N	Y	N	N	N/A	N	Y	N	Y	Tingbao *et al.* 2006
Quadriacanthus spp.	Clariidae	Asia	N	N	Y	N	Y	N/A	N	N	N	N	Woo & Bruno 2014
Rhabdosynochus rhabdosynochus	Centropomidae	Widespread	N	N	Y	N	Y	N/A	N	N	N	N	Kritsky 2010
Scutogyrus longicornis	Cichlidae	Africa	N	N	Y	N	N	N/A	N	Y	N	Y	Parisielle & Euzet 1995
Silurodiscoides spp.	Pangasiidae	Southeast Asia	N	N	Y (skin)	Y	N	N/A	N	Y	N	Y	Johnston 2008b
Singhiogyrus exotica	Cyprinidae	India	N	N	Y	N	N	N/A	N	Y	N	Y	Chaudhary *et al.* 2013
Sparicotyle chrysophrii	Sparidae	Mediterranean	N	N	Y	N	N	N/A	N	Y	N	Y	Antonelli *et al.* 2010, Sitjà-Bobadilla & Alvarez-Pellitero (2009).

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Tetraonchus awakurai	Salmonidae	Japan	N	N	Y	N	N	N/A	N	Y	N	Y	Buchmann & Bresciani 2006
Tetraonchus oncorhynchi	Salmonidae	Japan	N	N	Y	N	N	N/A	N	Y	N	Y	Buchmann & Bresciani 2006
Thaparocleidus spp. *T. caecus, T. pangasi, T. siamensis*	Pangasiidae.	Southeast Asia	N	N	Y	N	N	N/A	N	Y	N	Y	Melville & Johnston 2010
Thaparogyrus lucknowius	Cyprinidae	India	N	N	Y	N	N	N/A	N	Y	N	Y	Chaudhary *et al.* 2013
Trimusculotrema schwartzi	Dasyatidae	Asia, China	N	N	Y (skin)	N	Y	N/A	N	Y	N	N	Anon. 2018
Trianchoratus agrawali, T. keami	Anabantidae	India	N	N	Y	N	Y	N/A	N	N	N	N	Chaudhary *et al.* 2013
Tristoma adintegrum	Xiphiidae	Pacific	N	Y	Y	N	Y	N/A	N	Y	N	N	Hine *et al.* 2000, Smith *et al.* 2007
Tristoma adcoccineum	Xiphiidae	Pacific	N	Y	Y	N	Y	N/A	N	Y	N	N	Hine *et al.* 2000, Smith *et al.* 2007
Unidentified monogeneans	Wide elasmobranch & teleost host range	Widespread	N	Y (some)	Y (skin)	N	Y	N/A	N	Y	N	N	Benz & Bullard 2004, Caira *et al.* 2012
Zeuxapta japonica	Carangidae	Pacific	N	Y	Y	N	Y	N/A	N	Y	N	N	Tubbs *et al.* 2007
Zeuxapta seriolae	Carangidae	Pacific	N	Y	Y	N	Y	N/A	N	Y	N	N	Hine *et al.* 2000
Digenea													
Acipersericola petersoni	Polyodontidae	United States	N	N	N (viscera)	N/A	N	N/A	Y	N	Y	N	Bullard *et al.* 2008, Durburrow *et al.* 2015
Aliptrema tilapiae	Cichlidae	Egypt	N	N	Y (muscle)	N/A	N	N/A	Y	Y	Y	N	Gupta *et al.* 2008
Allocreadium fossilis	Heteropneustidae	India, Mynamar	N	N	N (viscera)	N/A	N	N/A	Y	N	Y	N	Arthur & Ahmed 2002
Amirthalingamia sp.	Cichlidae	China, Brazil	N	N	N (viscera)	N/A	N	N/A	N	Y	N	N	Johnston 2008b
Ankistromeces dunwichensis, A. olsoni	Monacanthidae, Siganidae	IndoPacific	N	N	N (blood)	N/A	N	N/A	Y	N	Y	N	Nolan & Cribb 2006
Aphanurus stossichii	Clupeidae	Bangladesh, India	N	N	N (viscera)	N/A	N	N/A	Y	N	Y	N	Arthur & Lumanian-Mayo 1997, D'Silva *et al.* 2012
Aporocotyle spp.	Merlucciidae	Widespread	N	Y	N (blood)	N	Y	N	Y	Y	N	N	Fernandez 1985, Hernandez-Orts *et al.* 2017
Ascocotyle ascolonga	Cichlidae	China, Brazil	N	N	Y	N	N	N/A	N	N	Y	Y	Johnston 2008b, Lobna *et al.* 2010
Ascocotyle (*Phagicola*) *longa*	Mugilidae	Widespread	N	N	Y	N	N	N/A	Y	Y	Y	Y	Rodriguez *et al.* 2015
Aspidogastrea (undescribed)	Wide elasmobranch host range	Widespread	N	Y	Y	N	N	N	Y	Y	N	N	Hine *et al.* 2000

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Bolbophorus levantinus	Cichlidae	China, Brazil	N	N	Y	N	N	N/A	Y	N	Y	N	Johnston 2008b
Bolbophorus spp.	Ictaluridae, Percidae	Widespread	N	N	Y	N	N	N/A	Y	Y	Y	Y	Venable *et al.* 2000, Paperna & Dzikowski 2006
Bucephalus anguillae	Anguillidae	Widespread	N	Y	N (viscera)	N/A	Y	N/A	Y	Y	Y	N	Paperna & Dzikowski 2006
Carassotrema tilapiae	Cichlidae	China, Brazil	N	N	Y (skin, eyes, viscera)	N/A	N	N/A	N	N	N	N	Johnston 2008b
Centrocestus spp.	Cichlidae	China, Brazil	N	N	Y	N	N	N/A	N	N	Y	N	Chai & Lee 2002, Johnston 2008b
Centrocestus formosanus	Wide teleost host range	Widespread	N	N	Y	N	N	N/A	Y	Y	Y	Y	Paperna & Dzikowski 2006
Chimaerohemecus trondheimensis	Chimaeridae, Rajidae	Widespread	N	N	N (blood)	N/A	Y	N/A	Y	Y	N	N	Cribb *et al.* 2017
Clinostomum spp.	Cichlidae	China, Brazil	N	N	Y	N	N	N/A	N	N	Y	Y	Johnston 2008b, Sutili et al. 2014
Clinostomum tilapiae	Cichlidae	Widespread	N	N	Y	N/A	N	N/A	Y	N	Y	Y	Johnston 2008b, Sutili et al. 2014
Clonorchis sinensis	Cichlidae	China, Brazil	N	N	Y	N	N	N/A	N	N	Y	N	Muller 2002, Johnston 2008b
Clonorchis sp.	Wide teleost host range	Widespread	N	N	Y	N	N	N/A	Y	Y	Y	Y	Muller 2002, Paperna & Dzikowski 2006, Youn 2009
Diplostomum spp.	Wide teleost host range	Widespread	N	N	Y	N	N	N/A	Y	Y	N	Y	Johnston 2008b
Echinochasmus spp.	Cichlidae , Cyprinidae	Widespread	N	N	Y	N	N	N/A	Y	Y	Y	Y	Chai & Lee 1991, Murrel & Fried 2007, Johnston 2008b
Echinostomes spp.	Teleosts	India, southeast Asia	N	Y	Y	N	N	N/A	Y	Y	Y	N	Hine *et al.* 2000, Muller 2002
Erilepturus hamate	Wide teleost host range	Widespread	N	N	N (viscera)	N/A	N	N/A	Y	N	Y	N	Wang 1982, Shen 1990,Vo *et al.* 2008, Liu *et al.* 2010
Euclinostomum heterostomum	Atherinopsidae, Channidae, Cichlidae, Siluridae	Widespread	N	N	Y (muscle)	N	N	N/A	Y	Y	Y	Y	Johnston 2008b, Athokpam & Tandon 2015
Exorchis spp.	Cichlidae	China, Brazil	N	N	Y (muscle)	N	N	N	Y	Y	N	N	Johnston 2008a, Krailis *et al.* 2014
Faustula brevichrus, F. gangetica, F. ilishii	Clupeidae	Bangladesh, India	N	N	N (viscera)	N/A	N	N/A	Y	Y	N	N	D'Silva *et al.* 2012
Haplorchis spp.	Cichlidae, Salmonidae	China, Brazil	N	N	Y	N	N	N/A	Y	Y	Y	Y	Johnston 2008b

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Heterophyes spp.	Cichlidae, Mugilidae, Soleidae	Widespread	N	N	Y	N	N	N/A	Y	Y	Y	Y	Wells & Randall 1955, Muller 2002, Johnston 2008b, Youn 2009
Heterophyopsis continua	Serranidae	Asia	N	N	Y	N	N	N/A	Y	Y	Y	Y	Vo *et al.* 2008
Hirudinella spp.	Scombridae, Xiphiidae	Widespread	N	Y	N (viscera)	N	N	N	Y	Y	Y	N	Hine *et al.* 2000, Smith *et al.* 2007, Dias *et al.* 2011
Hyperandrotrema cetorhini, H. walterboergeri	Cetorhinidae, Lamnidae	Widespread	N	N	N (blood)	N/A	Y	N/A	Y	Y	N	N	Maillard & Ktari 1978, Oerlis-Ribeiro *et al.* 2013
Lecithochirium pisoodonophis	Ophichthidae	China, Australia, New Zealand	N	Y	N (viscera)	N/A	N	N	Y	Y	N	N	Hine *et al*. 2000, Liu *et al*. 2010
Lecithaster indicus	Clupeidae., Muglidae	Bangladesh, India	N	N	N (viscera)	N/A	N	N/A	Y	Y	N	N	D'Silva *et al.* 2012
Lophosicya-diplostomum sp.	Channidae, Cichlidae, Siluridae	Bangladesh, India	N	N	Y	N/A	N	N/A	Y	N	Y	N	Arhokpam & Tandon 2015
Macvicaria sp.	Sparidae	Bangladesh, India	N	N	N (viscera)	N/A	N	N/A	Y	N	N	N	Bartoli 1989
Mesostephanus spp.	Cichlidae	Widespread	N	N	Y (muscle)	N	N	N	Y	Y	Y	Y	Abou-Eisha *et al.* 2008, Johnston 2008a
Metagonimus spp.	Wide teleost host range	Widespread	N	N	Y	N	N	N/A	Y	Y	Y	Y	Muller 2002, Ko 2006, Abou-Eisha *et al.* 2008, Yeon 2009
Metorchis bilis, M. conjunctus	Wide teleost host range	Widespread	N	N	Y	N	N	N/A	Y	Y	Y	Y	Kennedy 2004, Chai *et al.* 2005, Sitko *et al.* 2016
Moedlingeria amphoraeformis	Cichlidae	China, Brazil	N	N	Y (muscle)	N	N	N	Y	N	Y	Y	Shalaby *et al.* 1993, Johnston 2008a
Monorchis fusiformis	Ophichthidae	Widespread	N	N	N (viscera)	N/A	N	N/A	Y	N	N	N	Wang 1982. Liu *et al*. 2010
Myliobaticola richardheardi	Dasyatidae	Eastern United States	N	N	N (blood)	N/A	N	N/A	Y	Y	N	N	Bullard & Jensen 2008
Neopecoelina spp.	Cnannidae, Heteropneustidae	Bangladesh, India	N	N	N (viscera)	N/A	N	N/A	Y	N	N	N	Gupta 1955, Froese & Pauly 2018
Opegaster sp.	Heteropneustidae	Bangladesh, India	N	N	N (viscera)	N/A	N	N/A	Y	N	N	N	Bray & Justine 2013, Froese & Pauly 2016
Opisthorchis spp.	Cyprinidae	Widespread	N	N	Y	N	N	N/A	Y	Y	Y	Y	Muller 2002, Doanh & Nawa 2016
Orchispirium heterovitellatum	Dasyatidae	Indo West Pacific	N	N	N (blood)	N/A	N	N/A	Y	Y	N	N	Bullard & Jensen 2008
Otodistomium veliporum, O. cestoides	Rajidae, Squatinidae	South America	N	Y	N (viscera)	N/A	N	N	Y	Y	N	N	Hine *et al.* 2000, Knoff *et al.* 2001b

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Paradeontacylix grandispinus	Scombridae	Japan	N	N	N (blood)	N	N	N/A	Unknown	Y	Y	N	Ogawa & Egusa 1986, 1994
Paradeontacylix kampachi	Scombridae	Japan	N	N	N (blood)	N	N	N/A	Unknown	Y	Y	N	Ogawa & Egusa 1986, 1994
Phagicola ornata, P. ornamentata	Cichlidae	Brazil, China, Egypt	N	N	N (viscera)	N/A	N	N	Y	Y	Y	N	Shalaby *et al.* 1993, Johnston 2008b
Pharyngostomum spp.	Cichlidae	China, Brazil	N	N	Y (muscle)	N	N	N/A	Y	Y	Y	Y	Shalaby *et al.* 1996, Johnston 2008b
Philopinna spp.	Channidae, Cyprinidae, Heteropneustidae	Bangladesh, Egypt, India, Japan	N	N	Y (fins, eye, muscle)	N/A	N	N/A	Y	Y	N	N	Arthur & Ahmed 2002, Froese & Pauly 2016
Phyllotrema bicaudatum, P. quadricaudatum	Congridae, Ophichthidae	Australia, China, Southease Asia,	N	N	N (viscera)	N/A	N	N/A	Y	N	N	N	Liu *et al.* 2010
Polylekithum sp.	Channidae, Siluridae	India	N	N	Y (muscle)	NA	N	N/A	Y	Y	Y	Y	Athokpam & Tandon 2015
Posthodiplostomum spp.	Channidae, Cichlidae, Cyprinidae, Siluridae	Widespread	N	N	Y (skin, muscle)	N	N	N/A	Y	Y	Y	Y	Johnston 2008b, Arhokpam & Tandon 2015, Nareaho *et al.* 2017
Procerovum sp.	Cichlidae, Serranidae	China, Asia, Brazil	N	N	Y (muscle)	N	N	N/A	Y	Y	Y	Y	Shalaby *et al.* 1993, Johnston 2008b, Vo *et al.* 2008
Prohemistomum vivax	Cichlidae	China, Brazil	N	N	Y (muscle)	N	N	N	Y	N	Y	Y	Abou-Eisha *et al.* 2008, Johnston 2008b
Prosostephanus industrius	Cichlidae	Brazil, China, Egyptl	N	N	Y (muscle)	N	N	N/A	Y	N	Y	Y	Shalaby *et al.* 1993, Johnston 2008b
Protocladorchis pangasii	Pangasiidae	Southeast Asia	N	N	N (viscera)	N/A	N	N/A	Y	Y	N	N	Melville & Johnston 2010
Pygidiopsis spp.	Cichlidae, Mugilidae	Widespread	N	N	Y (muscle)	N	N	N/A	Y	Y	Y	Y	Johnston 2008b, Lobna *et al.* 2010. Hegazi *et al.* 2014
Selachohemecus benzi, S. olseni	Carcharhinidae	Widespread	N	N	N (blood)	N/A	N	N/A	Y	Y	N	N	Short 1954, Bullard *et al.* 2006
Stellantchasmus falcatus	Cyprinidae, Hemiramphidae, Mugilidae	Southeast Asia, China, Japan, Hawaii	N	N	N (viscera)	N/A	N	N/A	Y	Y	Y	N	Youn 2009, Chai *et al.* 2016
Tylodelphys spp.	Wide teleost host range	Widespread	N	Y	Y	N	N	N/A	Y	Y	N	N	Blasco-Costa *et al.* 2017
Transversotrema spp.	Wide teleost host range	Widespread in marine and fresh waters	N	N	Y (skin)	N	N	N/A	Y	Y	N	Y	Cribb *et al.* 1992, 2011, 2014
Unidentified digenean metacercaria	Wide teleost host range	Ubiquitous	N	N	Y (muscle)	N	N	N	Y	Y	Y	Y	Johnston 2008b

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Cestoda													
Adenocephalus pacificus (=*Diphyllobothrium pacificum*) (Pacific broad tapeworm)	Wide teleost host range, Triakidae	Widespread	N	Y	Y	N	N	N/A	N	Y	Y	N	Felix 2015, Kuchta *et al.* 2015, Scholtz & Kuchta 2016
Amphilina foliacea, A. bipunctata	Acipenseridae	Widespread	N	N	Y	N	N	N/A	N	N	N	N	Paulin *et al.* 2001, Dick *et al.* 2006
Bialovarium spp.	Heteropneustidae	Bangladesh, India	N	N	N (viscera)	N/A	N	N/A	Y	N	N	N	Fischthal 1953, Froese & Pauly 2016
Bothriocephalus acheilognathi	Cyprinidae	Widespread	N	N	N (Viscera)	N/A	Y	N/A	N	Y	Y	N	Dick *et al.* 2006, Yera *et al.* 2013
Bothriocephalus claviceps	Anguillidae, Percidae, Poeciliidae	Widespread	N	N	N (viscera)	N	N	N/A	N	Y	N	Y	Scholz 1997
Bothriomonas sturionis	Salmonidae	Widespread	N	N	N (Viscera)	N/A	Y	N/A	N	Y	N	N	Dick *et al.* 2006
Callitetrarhynchus spp.	Wide elasmobranch and teleost host range	Widespread	N	Y	Y (muscle)	N	N	N/A	N	Y	N	N	Hine *et al.* 2000, Felizardo *et al.* 2010, Morsey *et al.* 2013, Beveridge *et al.* 2014
Caryophyllaeus spp.	Cyprinidae, Siluridae	Widespread	N	N	N (viscera)	N/A	Y	N/A	N	Y	N	N	Dick *et al.* 2006
Cetorhinicola acanthocapax	Cetorhinidae	Widespread	N	Y	N (viscera)	N/A	Y	N/A	N	Y	N	N	Beveridge & Campbell 1988, Beveridge & Duffy 2005
Corallobothrium sp.	Ictaluridae	Widespread	N	N	N (viscera)	N/A	Y	N/A	N	Y	Y	N	Piasecki *et al.* 2004, Dick *et al.* 2006
Cyathocephalus truncatus	Salmonidae	Widespread	N	N	N (viscera)	N/A	Y	N/A	N	Y	N	N	Dick *et al.* 2006
Dasyrhynchus basipunctatus	Carcharhinidae	IndoPacific, Australia	N	Y	N (viscera)	N/A	Y	N/A	N	Y	N	N	Hine & Diggles 2000
Dibothriocephalus alaskensis (*Diphyllobothrium alascense*)	Wide teleost host range	Widespread	N	N	Y (muscle)	N	N	N/A	N	Y	N	N	Scholz & Kutcha 2016
Dibothriocephalus dalliae (*Diphyllobothrium dalliae*)	Lotidae, Salmonidae	Widespread	N	N	Y (muscle)	N	N	N/A	N	Y	Y	Y	Scholz & Kutcha 2016

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Dibothriocephalus dendriticus (*Diphyllobothrium dendriticum, Bothriocephalus dedriticus*)	Wide teleost host range	Widespread	N	N	Y (muscle)	N	N	N/A	N	Y	Y	Y	Felix 2013, Kutcha 2013, Scholz & Kutcha 2016
Dibothriocephalus ditremus (*Diphyllobothrium ditremum*)	Osmeridae, Salmonidae	Widespread	N	N	Y (muscle)	N	N	N/A	N	N	N	N	Paulin *et al.* 2001, Scholz *et al.* 2009, Felix 2013
Dibothriocephalus latus (*Diphyllobothrium latum)*	Wide teleost host range	Widespread	N	N	Y (muscle)	N	N	N/A	N	Y	Y	Y	Paulin *et al.* 2001, Scholz *et al.* 2009, Felix 2015
Dibothriocephalus (*Diphyllobothrium*) *ursi*	Ariidae, Carangidae, Sciaenidae	Widespread	N	N	Y (muscle)	N	N	N/A	N	Y	Y	Y	Paulin *et al.* 2001, Waeschenbach 2017
Digramma interrupta	Cyprinidae	Widespread	N	N	Y (muscle)	N	N	N/A	N	Y	N	Y	Reshnetikova 1965
Diphyllobothrium balaenopterae Diplogonoporus balaenopterae, D. grandis)	Clupeidae, Engraulidae	Widespread	N	N	Y (muscle)	N	N	N/A	N	Y	Y	Y	Sinderman 1990, Chung *et al.* 1995, Kino *et al.* 2002
Diphyllobothrium nihonkaiensis (Asian tapeworm)	Wide teleost host range	Widespread	N	Y	Y (muscle)	N	N	N/A	N	N	Y	N	Felix 2013, Kuchta *et al.* 2015, Scholtz & Kuchta 2016
Diphyllobothrium sp.	Anguillidae	Widespread	N	N	Y (muscle)	N	N	N/A	N	Y	Y	Y	Paulin *et al.* 2001
Floriceps saccatus	Carangidae, Coryphaenidae	Widespread	N	N	N (viscera)	N	N	N/A	N	Y	N	N	Sinderman 1990
Floriceps minacanthus	Carcharhinidae	Widespread	N	N	N (viscera)	N/A	Y	N/A	N	Y	N	N	Capmbell & Beveridge 1987a, Beveridge *et al.* 2014
Gilquinia sp., *G. minor, G. robertsoni, G. squali*	Centrophoridae, Triakidae	Widespread	N	Y	N (viscera)	N/A	Y	N	N	Y	N	N	Beveridge & Jones 2002, Beveridge & Justine 2006
Gymnorhynchus sp.	Wide teleost host range	Widespread	N	Y	Y (muscle)	N	N	N/A	N	Y	Y	N	Hine *et al.* 2000. Paulin *et al.* 2001, Pelayo *et al.* 2009
Hepatoxylon sp.	Wide elasmobranch and teleost host range	Widespread	N	Y	Y	Y	N	N/A	Y	Y	N	N	Smith *et al.* 2007
Heteronybelinia nipponica	Paralichthyidae	South America	N	N	Y (muscle)	N	N	N/A	Y	Y	Y	Y	Felizardo *et al.* 2010
Homelliella annandalei	Hemiscylliidae, Stegosomatidae	IndoPacific, Australia	N	N	N (viscera)	N/A	Y	N/A	Y	N	N	N	Campbell & Beveridge 1987b, Waterman & Sin 1991

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Ilisha parthenogenetica	Clupeidae	Bangladesh, India	N	N	N (viscera)	N/A	Y	N/A	Y	Y	N	N	D'Silva *et al.* 2012
Kawhia spp.	Cyprinidae, Siluridae	Widespread	N	N	N (Viscera)	N/A	Y	N/A	N	Y	N	N	Dick *et al.* 2006
Kotorella pronosoma	Sparidae	Europe	N	N	Y (muscle)	N	N	N/A	Y	Y	N	Y	Morsey *et al.* 2013
Lacistorhynchus dollfusi, Dolfusiella martini, Trimacranthus aetobatidis	Wide elasmobranch and teleost host range	Widespread	N	Y	Y (muscle)	N/A	Y	N/A	N	Y	N	N	Beveridge & Sakanar 1987, Hine *et al.* 2000, Felizardo *et al.*.2010
Lacistorhynchus tenuis	Moronidae	Widespread	N	N	Y	N	N	N/A	Y	N	N	N	Paulin *et al.* 2001, Felizardo *et al.*.2010
Ligula intestinalis	Galaxiidae	Widespread	N	Y	Y	Y	N	N	Y	Y	Y	N	Weeks & Penlington 1986, Hine *et al.* 2000, Scholz & Kuchta 2016
Lucknowia fossilisi	Heteropneustidae	Bangladesh, India	N	N	N (viscera)	N/A	N	N/A	Y	N	N	N	Ash *et al.* 2011
Neogryporhynchus cheilancristrotus	Cyprinidae	Widespread	N	N	N (viscera)	N/A	Y	N/A	Y	Y	N	N	Scholz *et al.* 2004
Nybelinia goreensi, N. indica, N. queenslandii,	Nemipteridae	Australia	N	Y	Y (muscle)	N	N	N/A	Y	N	Y	N	Hine *et al.* 2000, Palm *et al.* 1997, Paulin *et al.* 2001
Nybelinia sp.	Carangidae	Australia	N	Y	Y (muscle)	N	N	N	Y	N	Y	N	Hine *et al.* 2000, Felizardo *et al.*.2010
Nybelina bisulcata, N. narinari	Sparidae	Europe	N	N	Y (muscle)	N	N	N	Y	Y	Y	Y	Hine *et al.* 2000, Morsey *et al.* 2010
Nybelinia lingularis	Paralithyidae	South America	N	N	Y (muscle)	N	N	N/A	Y	Y	Y	Y	Felizardo *et al.*.2010
Nybelinia surmenicola	Carangidae, Gadidae, Lamnidae, Merlucciidae	Widespread	N	Y	Y (muscle)	N	N	N/A	Y	Y	Y	N	Sinderman 1990. Felizardo *et al.*.2010; Bryan *et al.* 2012; Lee *et al.* 2016
Paradilepis scolecina	Cyprinidae	Widespread	N	N	Y (muscle)	N	N	N/A	Y	Y	N	N	Forrest & Cone 2011
Pseudogrillotia spp.	Sparidae	Europe	N	N	Y (muscle)	N	N	N/A	Y	Y	N	Y	Morsey *et al.* 2013
Otobothrium spp.	Wide teleost host range	Widespread	N	N	Y (muscle)	N/A	N	N/A	Y	Y	Y	Y	Felizardo *et al.*2010, D'Silva *et al.* 2012, Bhuiyan 2014,
Poecilancistrium caryophyllum (robustum), P. dipsacum	Carangidae, Sciaenidae	Widespread	N	N	Y (muscle)	N	N	N/A	N	Y	N	Y	Overstreet 1977, Sinderman 1990
Proteocephalus ambloptis	Centrarchidae	Widespread	N	N	Y (muscle)	N	N	N/A	N	N	N	N	Paulin *et al.* 2001
Proteocephalus exiguuis	Salmonidae	Widespread	N	N	N (viscera)	N/A	Y	N/A	N	Y	N	N	Dick *et al.* 2006, Johnston 2008a

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Protocladorchis spp.	Wide teleost host range	Widespread	N	N	N (viscera)	N/A	Y	N/A	N	Y	N	N	Johnston 2008a
Pseudobothrium sp.	Xiphiidae	Widespread	N	Y	Y	Y	N	N	Y	Y	N	N	Smith *et al.* 2007; Alves *et al.* 2018
Pseudocaryophyllaeus heteropneustus	Heteropneustidae	Bangladesh, India	N	N	N (viscera)	N/A	N	N/A	Y	N	N	N	Arthur & Ahmed 2002
Pterobothrium acanthotruncatum, P. australiense. P. Kingstoni	Dasyatidae, Pristidae	Asia, Australia	N	N	N (viscera)	N/A	Y	N/A	N	Y	N	N	Palm *et al.* 1997, Beveridge *et al.* 2014
Pterobothrium kingstoni, P. crassicole	Paraliththyidae	South America	N	N	Y (muscle)	N/A	N	N/A	N	Y	N	Y	Palm *et al.* 1997, Palm & Overstreet 2000, Beveridge *et al.* 2014
Pyramicocephalus phocarum	Wide teleost host range	Widespread	N	N	Y (muscle)	Y	N	N/A	Y	Y	Y	Y	Rausch & Adams 2000, da Fonseca *et al.* 2012, Scholz & Kuchta 2016
Shirleyrhynchus butlerae	Orectolobidae	Australia	N	N	N (viscera)	N/A	Y	N/A	Y	Y	N	N	Beveridge & Campbell 1988
Stragulorhynchus orectologi	Orectolobidae	Australia	N	N	N (viscera)	N/A	Y	N/A	Y	Y	N	N	Beveridge & Campbell 1988
Tentacularia coryphaenae	Wide teleost host range, Carcharhinidae	Widespread	N	Y	N (viscera)	N/A	Y	N/A	Y	Y	N	N	Hine *et al.* 2000, Beveridge & Jones 2002, Palm *et al.* 2007
Triaenophorus nodulosus, T. crassus	Cichlidae, Cyrinidae, Esocidae, Salmonidae	Widespread	N	N	Y (muscle)	N	N	N/A	Y	Y	N	Y	Valtonen *et al.* 1989
Unidentified plerocercoid larvae (*Scolex pleronectes*)	Wide elasmobranch and teleost host range	Widespread	N	N	Y (muscle)	N	N	N/A	Y	Y	Y	N	Beveridge & Campbell 1988, Sinderman 1990
Valipora campylancristrota	Cyprinidae	Widespread	N	N	N (viscera)	N/A	Y	N/A	Y	Y	N	N	Scholz *et al.* 2004
Vittirhynchus sp., V. squali, Sagittirhynchus aculeatus	Centrophoridae, Triakidae	Australia	N	N	N (viscera)	N/A	Y	N/A	Y	Y	N	N	Beveridge & Campbell 1988
Acanthocephala													
Acanthocephalans	Wide elasmobranch & teleost host range	Widespread	N	Y	N (viscera)	N/A	Y	N	N	Y	N	N	Adams *et al.* 1997; Stone *et al.* 1997, Benz *et al.* 2004, Dick *et al.* 2006, Johnston 2008b
Acanthocentis indica, A. hilsai	Clupeidae	Bangladesh, India	N	N	N (viscera)	N/A	Y	N/A	N	Y	N	N	D'Silva *et al.* 2012, Bhuiyan 2013
Corynosoma sp.	Pomatomidae, Squatinidae, Triakidae	Widespread	N	Y	N (viscera)	N/A	Y	N	N	Y	N	N	Hine *et al.* 2000, Knoff *et al.* 2001b, *Benz et al. 2004*

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Dollfusentis chandleri	Dasyatidae	Widespread	N	N	N (viscera)	N/A	Y	N/A	N	Y	N	N	Benz *et al.* 2004
Echinorhynchus gadi	Triakidae	Widespread	N	Y	N (viscera)	N/A	Y	N	N	Y	N	N	Hine *et al.* 2000
Gorgorhynchus trachinotus	Sphyrnidae	South America	N	N	N (viscera)	N/A	Y	N/A	N	Y	N	N	Knoff *et al.* 2001b
Megapriapus ungriai	Potamotrygonidae	South America	N	N	N (viscera)	N/A	Y	N/A	N	Y	N	N	Benz *et al.* 2004
Tegorhynhcus furcatus	Dasyatidae	Widespread	N	N	N (viscera)	N/A	Y	N/A	N	Y	N	N	Benz *et al.* 2004
Crustacean pathogens													
Amphipoda													
Lafystius morhuanus	Rajidae, Sebastidae	Canada, North America	N	N	Y (skin)	N/A	Y	N/A	N	Y	N	N	Kabata 1970, Paulin *et al.* 2001, Benz *et al.* 2004, Moran *et al.* 2011
Lafystius sturionis	Gadidae, Rajidae	Canada, North America	N	N	Y (skin)	N/A	Y	N/A	N	Y	N	N	Kabata 1970, Paulin *et al.* 2001, Benz *et al.* 2004, Montero *et al.* 2008
Opisa tridentata	Triakidae	Widespread	N	N	Y (skin)	N	Y	N/A	N	Y	N	N	Bousefield & Kabata 1988, Paulin *et al.* 2001, Benz *et al.* 2004
Trischizostoma raschi	Etmopteridae	Europe, North Atlantic	N	N	Y (skin)	N	Y	N/A	N	Y	N	N	Benz *et al.* 2004
Cirripedia													
Anelasma squalicola	Etmopteridae	Widespread	N	Y	Y (skin)	N	Y	N	N	Y	N	N	Hine *et al.* 2000, Yano & Musick 2000
Conchoderma auritum, C. virgatum (rabbit eared barnacle, goose barnacle)	Wide elasmobranch and teleost host range	Widespread	N	N	Y(skin)	N	Y	N/A	N	Y	N	N	Benz *et al.* 2004
Copepoda													
Anthosoma crassum	Carcharhinidae, Lamnidae, Molidae	Widespread	N	Y	Y (skin)	N	Y	N	N	Y	N	N	Hine *et al.* 2000, Paulin *et al.* 2001, Benz *et al.* 2004, Benz & Bullard 2004
Brachiella spp.	Wide teleost host range	Widespread	N	N	Y (skin)	N/A	N	N/A	N	Y	N	N	Lester & Heyward 2006
Caligus spp.	Wide teleost host range	Widespread	N	Y	Y (skin)	N/A	N	N/A	N	Y	N	N	Jones 1988, Lester & Heyward 2006
Colobomatus lamnae	Lamnidae	Widespread	N	N	Y (skin)	N	Y	N/A	N	Y	N	N	Grabda & Linowski 1978, Jones 1998, Caira *et al.* 2012
Driocephalus cerebrinoxius	Leptochariidae	Eastern Atlantic	N	N	N (olfactory lobes)	N	Y	N/A	N	N	N	N	Diebakate *et al.* 1997, Benz *et al.* 2004

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Prosaetes rhinodontis	Rhincodontidae	Widespread	N	N	Y (skin)	N	Y	N/A	N	N	N	N	Cressy 1970, Hine *et al.* 2000, Norman *et al.* 2000
Kroyeria lineata, K. longicauda	Carcharhinidae, Triakidae	Widespread	N	Y	Y (skin)	N	Y	N	N	Y	N	N	Cressy 1970, Deets 1994, Henderson *et al.* 2003
Lernaea spp.	Wide teleost host range	Widespread	N	Y	Y (skin)	N/A	N	N/A	N	Y	N	N	Hine *et al.* 2000, Lester & Heyward 2006
Lernaeocera spp.	Heteropneustidae	Bangladesh, India	N	N	N (viscera)	N/A	N	N/A	Y	N	N	N	Froese & Pauly 2016
Lernanthropus kroyeri	Moronidae	Europe	N	Y	Y (skin)	N	Y	N	N	Y	N	N	Hine *et al.* 2000, Khidr *et al.* 2014
Nesippus vespa (=*N. orientalis*)	Lamnidae, Rhinobatidae	Widespread	N	Y	Y (skin)	N	Y	N	N	Y	N	N	Kurtisinghe 1964, Hine *et al.* 2000, Dippenaar *et al.* 2010
Pandarus cranchii, P. satyrus	Carcharhinidae, Lamnidae	Widespread	N	Y	Y (skin)	N	Y	N	N	Y	N	N	Hewitt 1967, Hine *et al.* 2000, Benz *et al.* 2004
Perissopus dentatus	Wide elasmobranch host range	Widespread	N	Y	Y (skin)	N	Y	N	N	Y	N	N	Kurtisinghe 1964, Hine *et al.* 2000
Tripaphylus vaissierei	Sphyraenidae	Widespread	N	N	N (gills)	N/A	Y	N/A	N	Y	N	N	Lewis 1966
Isopoda													
Alitropus spp.	Wide teleost host range	Widespread	N	N	Y (skin)	N/A	Y	N/A	N	Y	N	N	Nair & Nair 1983, Lester & Heyward 2006
Gnathia sp.	Rhinobatidae, Serranidae	Australia, Europe	N	Y	Y (skin)	N	Y	N/A	N	Y	N	N	Monod 1926; Poore 1981, McKierman *et al.* 1995, Genc 2007
Brachuira													
Argulus spp.	Wide teleost host range	Widespread	N	N	Y (skin)	N/A	Y	N/A	N	Y	N	N	Lester & Heyward 2006
Ostracoda													
Photeros parasitica	Sphyraenidae	Widespread	N	N	Y (skin)	N	Y	N/A	N	Y	N	N	Benz *et al.* 2004
Sheina orri	Dasyatidae, Hemiscylliidae	Australia	N	N	Y (skin, gills)	N	Y	N/A	N	Y	N	N	Bennett *et al.* 1997, Benz *et al.* 2004
Other Metazoan pathogens													
Ulcerative dermal necrosis agent	Salmonidae	Widespread	N	N	Y	Y (Skin ulcers)	N	N/A	N	Y	N	N (Unknown pathogen)	Anon. 2015
Simenchelys parasitica (pugnosed eel)	Gadidae, Lamnidae, Pleuronectidae	Widespread	N	Y	Y (muscle, viscera)	N/A	Y	N/A	N	Y	N	N	Paulin *et al.* 2001, Benz *et al.* 2004

9.1. Hazard list for risk assessment

The following hazard list for imported eviscerated bony fishes (class Actinopterygii) and trunked cartilagenous fishes (class Elasmobranchii) and their products (including minced, salted, smoked or mechanically recovered fish) has been compiled from the preliminary hazard list as identified in Table 6. All of these hazards are subject to a risk assessment:

9.1.1. Viruses

- Epizootic haematopoietic necrosis virus (EHNV), including European catfish virus (ECV), European sheatfish virus (ESV)
- European eel virus
- European eel herpesvirus (HVA/EEHV)
- Grass carp haemorrhagic virus
- Grouper iridovirus
- Infectious haematopoietic necrosis virus
- Infectious pancreatic necrosis virus (IPNV), including halibut birnavirus (HBV), Tasmanian salmon birnavirus (TaBV), viral deformity of yellowtail virus VDV)
- Infectious salmon anaemia virus
- Koi herpesvirus (CyHV-3)
- Hirame rhabdovirus
- New Japan virus of salmonids
- Nodaviruses including viral necrosis virus (VNN), viral encephalopathy and retinopathy (VER)
- *Oncorhynchus masou* virus (OMV)
- Pilchard orthomyxovirus (POMV)/Tasmanian salmon orthomyxovirus (SOMV)
- Piscine aquareovirus (PRV), including heart and skeletal muscle inflammation syndrome (HSMI), salmon aquareovirus (SAV), Tasmanian salmon reovirus (TasSRV), grass carp reovirus (GCRV) and turbot (TRV) aquaeroviruses
- Red sea bream iridovirus (RSIV), including infectious spleen and kidney necrosis virus (ISKNV), dwarf gourami iridovirus (DGIV) and other megalocytiviruses
- Salmon alphavirus (SAV)
- Salmon gill poxvirus, including carp oedema virus and koi sleepy disease
- Spring viraemia of carp virus (SVCV), including pike fry rhabdovirus (PFRV)
- Viral erythrocytic necrosis virus (VEN)/piscine erythrocytic necrosis virus (PEN) (Iridoviridae)
- Viral haemorrhagic septicaemia virus (VHSV)

9.1.2. Bacteria

- *Aeromonas hydrophila* (exotic strains)
- *Aeromonas salmonicida* var. *salmonicida* (atypical strains)
- *Aeromonas salmonicida* var *salmonicida* (typical strains)
- *Edwardsiella* spp.
- *Flavobacterium columnare* (exotic strains)
- *Francisella* spp.
- *Moritella viscosa*

- *Piscirickettsia salmonis* species complex
- *Pseudomonas anguilliseptica*
- *Renibacterium salmoninarum*
- *Streptococcus* agalactiae (serovar III: 283)
- *Streptococcus iniae*
- *Yersinia ruckeri* (exotic strains)

9.1.3. Fungi and Mesomycetozoea

- *Aphanomyces invadans*
- *Ichthyophonus hoferi*
- *Sphaerothecum destruens* (Rosette agent)

9.1.4. Microsporidia

- *Glugea plecoglossi*
- *Kabatana arthuri*
- *Microsporidium seriolae*
- *Nucleospora (Enterocytozoon) salmonis*
- *Pleistophora (Heterosporis) anguillarum*
- *Tetramicra brevifilum*

9.1.5. Myxosporea

- *Ceratomyxa shasta.*
- *Enteromyxum fugu, E. leei, E. scophthalmi*
- *Henneguya creplini, H. excilis, H. ghaffari, H. ictaluri, H. nuesslini, H. piaractus, H. salmonicola* (*=H. zschokkei*)
- *Kudoa* spp., *K. clupeidae, K. neothunni, K. paniformis, K. thyrsites*
- *Myxobolus* spp., *M. aeglefini, M. articus, M. artus, M. bramae, M. buckei, M. carassii, M. cultus, M. drjagini, M. ergensi, M, hungaricus, M. intimus, M. koi, M. macrocapsularis, P. parviformis, M. pavlovski, M. portucalensis, M. pseudodispar* (*=M. cyprini*)
- *Parvicapsula pseudobranchicola*
- *Sphaerospora chinensis, S. molnari, S. renicola*
- *Thelohanellus spp., T. hovorkai, T. nikolskii*
- *Unicapsula spp., U. andersenae, U. chirocentrus, U. galatea, U. marqesi, U. muscularis, M. pacifica, U. pflugfelderi, U. pyrimidata, U. schulmani, U. seriolae, U. stenolepis*

9.1.6. Nematoda

- *Anguillicola crassus*

9.1.7. Monogenea

- *Allencotyla mcintoshi*
- *Allopyragraphorus hippos*
- *Ancyrocephalus chakrabartii,*
- *Anoplodiscus tai*
- *Bifurcohaptor* spp.

- *Calicobenedenia polyprioni*
- *Cemocotyle noveboracensis*
- *Choriocotyle chrysophryi*
- *Cichlidogyrus* spp., *C. tilapiae*
- *Cleidodiscus pricei*
- *Dactylogyrus* spp., *D. anchoratus, D. aristichthys, D. batae, D. bifurcatus, D. brevitubus, D. calbasi, D. chagunionis , D. chauhani, D. ctenopharyngodonis, D. extensus, D. fotedari, D. glossogobii, D. hypophthalmichthys, D. inexpectatus, D. kalyanensis, D. labei, D. lamellatus, D. lampam, D. leptobarbus, D. lohanii, D. minutus, D. mrigali, D. nobilis, D. scriabini, D. speciosus, D. suchengtaii, D. vastator, D. vicinus, D. yogendrai*
- *Diplectanum* sp.
- *Diplozoon indicum*
- *Discocotyle sagittata*
- *Dogielius catalius*
- *Encotyllable spari*
- *Gyrodactylus* spp., *G. anguillae, G. bychowskii, G. cichlidarum, G. colemanensis, G. derjavini, G. elegans, G. katharineri, G. longipipes, G. orecchiae, G. salaris, G. salmonis*
- *Haplocleidus vachi*
- *Heteraxine heterocerca*
- *Lamellodiscus echeneis*
- *Mazocraes mamaevi, M. singhi*
- *Microcotyle sebastis*
- *Neobenedenia melleni*
- *Pangasitrema* spp.
- *Paradactylogyrus catalius*
- *Paramazocraes gorakhanati*
- *Protomicrocotyle mirabilis*
- *Pseudodactylogyrus anguillae, P. bini,*
- *Pseudorhabdosynochus* spp.
- *Scutogyrus longicornis*
- *Silurodiscoides* spp.
- *Singhiogyrus exotica*
- *Sparicotyle chrysophrii*
- *Tetraonchus awakurai, T. oncorhynchi*
- *Thaparocleidus* spp., *T. caecus, T. pangasi, T. siamensis*
- *Thaparogyrus lucknowius,*

9.1.8. Digenea

- *Ascocotyle ascolonga, A. longa*
- *Bolbophorus* spp.
- *Centrocestus formosanus*
- *Clinostomum spp. C. tilapiae*
- *Clonorchis* sp.
- *Diplostomum* spp.
- *Echinochasmus* spp.
- *Euclinostomum heterostomum*
- *Haplorchis* spp.

- *Heterophyes* spp.
- *Heterophysopsis continua*
- *Lophosicyadiplostomum* sp.
- *Mesostephanus* spp.
- *Metagonimus* spp.
- *Metorchis bilis, Metorchis conjunctus*
- *Moedlingeria amphoraeformis*
- *Opisthorchis* spp.
- *Pharyngostomum* spp.
- *Polylekithum* sp.
- *Posthodiplostomum* spp.
- *Procerovum* sp.
- *Prohemistomum vivax*
- *Prosostephanus industrius*
- *Pygidiopsis* spp.
- *Transversotrema* spp.
- Unidentified digenean metacercaria

9.1.9. Cestoda

- *Bothriocephalus claviceps*
- *Dibothriocephalus dalliae, D. dendriticus, D. latus, D. ursi*
- *Digramma interrupta*
- *Diphyllobothrium* sp., *D. balaenopterae*
- *Heteronybelinia nipponica*
- *Kotorella pronosoma*
- *Nybelinia bisulcata, N. lingularis, N. narinari*
- *Poecilancistrium caryophyllum, P. dipsacum*
- *Otobothrium* spp.
- *Pseudogrillotia* sp.
- *Pterobothrium crassicole, P. kingstoni*
- *Pyramicocephalus phocarum*
- *Triaenophorus nodulosus, T. crassus*

9.2. References

Abel EA, Pakingking RV, Pagador G, Wint MT, Huyop F (2012). Characteristics of dehalogenase from bacteria isolated from the gut of pond-reared rohu (*Labeo rohita*) juveniles in Myanmar. *Advances in Bioscience and Biotechnology* 3: 353–361.

Abou-Eisha AM, Saleh RE, Fadel HM, Youseff EM, Helmy YA (2008). Role of freshwater fishes in the epidemiology of some zoonotic trematodes in Ismailia Province (Egypt). *Suez Canal Veterinary Medicine Journal* 13(2): 653–676.

Abrahamian FM, Goldstein EJC (2011). Microbiology of animal bite wound infections. Clinical Microbiology Reviews 24(2): 231–246 [Online] Available from: http://www.doi: 10.1128/CMR.00041-10 [Accessed 20 October 2017].

Achla PN, Szyfres B (2001). Zoonoses and communicable diseases common to man and animals, Volume 1: Bacterioses and mycoses. Pan American Health Organisation, WHO (World Health Organisation of the United Nations), Wasington D.C., United States of America. 384 p.

Adams AM, Murrell KD, Cross JH (1997). Parasites of fish and risks to public health. *OIE Reviews in Science and Technology* 16(2): 652–660.

Adamson ML, Deets GB, Benz GW (1987). Description of male and redescription of female *Phlyctainophora squali* Mudry and Dailey, 1969 (Nematoda; Dracunculoidea) from elasmobranchs. *Canadian Journal of Zoology* 65(12): 3006–3010**.**

Alvarez-Pellitero P (2004). Report about fish parasitic diseases. pp 103-130. *In:* Alvarez-Pellitero P, Barja JL, Basurco B, Berthe F, Toranzo AE (*Eds*.). Mediterranean aquaculture diagnostic laboratories. Zaragoza: CIHEAM, 2004. *Options Méditerranéennes: Série B. Etudes et Recherches*: n. 49, 130 p.

Alve AM, Rangel e Sousa GT, Takemoto RM, Tavares LER, de Melo CM, Madi RR, Jeraldo VL (2018). Occurrence of larvae of trypanorhynch cestodes in snappers (Lutjanidae) from northeast Brazil. *Brazilian Journal of Veterinary Parasitology* Online] Available from : http://dx.doi.org/10.1590/S1984-296120180019 [Accessed 27 July 2018].

Amlacher E (1970). Protozoan diseases. pp 179-182. *In:* Textbook of fish diseases (translated), Conroy DA, Herman RL (*Eds*.). T.F.H. Publications, New York, Unites States of America, 302 p.

Anderson CD (1996). Distribution of salmonid diseases in New Zealand. *Surveillance* 23(4): 23–24.

Anderson CD (1997). Fish facts from New Zealand. *Microbiology Australia* (*May 1997*): 20–21.

Andreou D, Gozlan RE, Paley R (2009). Temperature influence on production and longevity of *Sphaerothecum destruens* zoospores. *Journal of Parasitology* 95(6): 1539–1531.

Anonymous (2015). UDN – Ulcerative dermal necrosis [Online]. Available from: http://www.gov.scot/resource/0042/00425990.pdf [Accessed 9 April 2015].

Anonymous (2018). Sharks and rays in New Zealand. National Museum of New Zealand [Online] Available from: https://collections.tepapa.govt.nz/topic/2775 [Accessed 26 March 2018].

Antonelli L, Quilichini Y, Marchland B (2010). *Sparicotyle chrysophrii* (Van Beneden and Hesse 1863) (Monogenea: Polyopisthocotylea) parasite of cultured Gilthead sea bream *Sparus aurata* (Linnaeus 1758) (Pisces: Teleostei) from Corsica: ecological and morphological study. *Parasitological Research* 107: 389–398.

Aquaculture New Zealand (2014). Salmon farming: An overview [Online]. Available from: http://aquaculture.org.nz/industry/overview.pdf [Accessed 15 July 2014].

Arkush KD, Bartholomew JL (2011). *Piscirickettsia, Francisella* and epitheliocystis. pp 302–337. *In*: Woo PTK, Bruno DW (*Eds*.). Fish Diseases and Disorders: Volume 3 - Viral, bacterial and fungal infections. 2nd edition. CABI International, Wallingford, United Kingdom. 930 p.

Arthur JR, Lumanlan-Mayo S (1997). Checklist of the parasites of fishes of the Philippines. *FAO Fisheries Technical Paper 369.* Food and Agriculture Organisation of the United Nations, Rome, Italy. 102 p.

Arthur JR, Ahmed ATA (2002). Checklist of the parasites of fishes of Bangladesh. *FAO Fisheries Technical Paper* T369/1), 77 p. Food and Agriculture Organisation of the United Nations, Rome, Italy [Online] Available from: http://www.fao.org/fishery/publications/technical-papers/en. [Accessed 19 April 2018].

Asato J, Kayana F (2004). Fatal infection of the hand due to *Photobacterium damsela*: A case report] Available from https://doi.org/10.1086/383468 [Accessed 25 June 2018].

Ash A, Oros M, Scholz T, Kar PK (2011). Cestodes (Caryophyllidea) of the stinging catfish *Heteropneustes fossilis* (Siluriformes: Heteropneustidae) from Asia. *Journal of Parasitology* 97(5):899–907.

Athokpam VD, Tandon V (2015). A survey of metacercarial infections in commonly edible fish and crab hosts prevailing in Manipur, Northeast India. *Journal of Parasitic Diseases* 39(3): 429–440. [Online] Available from: http://www. 10.1007/s12639-013-0360-z [Accessed 9 July 2018].

Austin B (1983). Bacterial microflora associated with a coastal marine fish-rearing unit. *Journal of the Marine Biological Association of the United Kingdom* 63(3): 585–592.

Austin B, Austin DA (2007). Bacterial fish pathogens: Diseases of wild and farmed fish. Springer Science, 580 p.

Austin B, McIntosh D, Murray KR (1988). Infection of Atlantic salmon (*Salmo salar*) with *Planococcus* sp. *Bulletin of the European Association of Fish Pathologists* 8(2): 21–22.

Avendano-Herrera R, Balboa S, Doce A, Ilardi, P, Lovera P, Toranzo AE, Romalde JL (2011). Pseudo-membranes on internal organs associated with *Rhodococcus qingshengii* infection in Atlantic salmon (*Salmo salar*). *Veterinary Microbiology* 147(1–2): 200–204.

Aveskamp MM, de Gruyter J, Verkley GJM, Crous PW (2010). Highlights of the *Didymellaceae*: A polyphasic approach to characterise *Phoma* and related pleosporalean genera. Studies in Mycology 65: 1–60.

Azevedo C, Casal G, Garcia P, Matos P, Teles-Grilo L, Matos E (2009). Ultrastructural and phylogenetic data of *Chloromyxum riorajum sp. nov*. (Myxozoa), a parasite of the stingray *Rioraja agassizii* in Southern Brazil. *Diseases of Aquatic Organisms* 85: 41–51.

Bacharach E, Mishra N, Briese T (2016). Characterization of a novel orthomyxo-like virus causing mass die-offs of tilapia. *M Bio* (American Society for Microbiology) 7(2): [Online] Available from: www. doi: 10.1128/mBio.00431-16 [Accessed 13 June 2017].

Bacigalupo J, De la Plaza N (1948). Presencia de tripanosomas en las rays de Mar del Plata. *Revista de la Sociedad Argentina de Biologia* 24: 269-274.

Baker TG, Pante E, Levesque EM, Roumillat WA, de Buron I (2008). *Metamicrocotyla macracantha*, a polyopisthocotylid gill parasite of the striped mullet, *Mugil cephalus*: population dynamics in South Carolina estuaries. *Parasitological Research* 102(5): 1085–1088.

Baldwin RE, Banks MA, Jacobsson KC (2011). Integrating fish and parasite data as a holistic solution for identifying the elusive stock structure of Pacific sardines (*Sardinops sagax*). *Reviews in Fish Biology and Fisheries* 22(1): 137–156.

Ball IR, Khan RA (1975). On *Micropharynx parasitica* Jagerskikild, a marine planarian ectoparasitic on thorny skate, *Raja radiata* Donovan, from the North Atlantic Ocean. J*ournal of Fish Biology* 8: 419–426.

Barbknecht M, Sepsenwol S, Leis E, Tuttle-Lau M, Gaikowski M, Knowles NJ, Lasee B, Hoffman MA (2014). Characterisation of a new picornavirus isolated from the freshwater fish *Lepomis macrochirus. Journal of General Virology* 95: 601–613.

Barker J, Brown MRW (1994). Trojan Horses of the microbial world : protozoa and the survival of bacterial pathogens in the environment. *Microbiology* 140: 1253–1259.

Barnes AC, Deboutteville JD, Gudkovs N, Brosnahan C, Morrison R, Carson J (2016). Whole genome analysis of *Yersinia ruckeri* isolated over 27 years in Australia and New Zealand reveals geographical endemism over multiple lineages and recent evolution under host selection. *Microbial Genomics* 2(11): e000095 [Online] Available from www.doi: 10.1099/mgen.0.000095 [Accessed 23 August 2017].

Bartoli P. Bray RA, Gibson DI (1989). The Opecoelidae (Digenea) of sparid fishes of the western Mediterranean. III *Macvicaria* Gibson & Bray, 1982. *Systematic Parasitology* 13(3): 167–192.

Basson L, Van As J (2006). Trichodinidae and other Ciliophorans (Phylum Ciliophora). pp 154–182. *In:* Woo PTK (*Ed*.) Fish Diseases and Disorders, Volume 1: Protozoan and metazoan infections, Second Edition. CABI International, Wallingford, United Kingdom, 791 p.

Bauer ON (1991). Spread of parasites and diseases of aquatic organisms by acclimatization: a short review. *Journal of Fish Biology* 39(5): 697–686.

Bauer ON, Voronin VN, Yunchis ON (1991). Infection of the heart in carp caused by *Myxobolus dogidi* Myxosporea, Myxobolidae) *Angewandte Parasitologie* 32: 42–44.

Baya AM, Toranzo AE, Lupiani B, Santos Y, Hetrick FM (1992). *Serratia marcescens*: a potential pathogen for fish. *Journal of Fish Diseases* 15(1): 15–26.

Bedell GW (1971). Infection of the heart in carp caused by *Myxobolus dogieli* (Myzosporea, Myxobolidae Angew. *Parasitology* 32: 42–44.

Bennett MB, Heupel MR, Bennett SM, Parker AR (1997). *Sheina orri* (Myodocopa: Cypridinidae), an ostracod parasitic on the gills of the epaulette shark, *Hemiscyllium ocellatum* (Elasmobranchii: Hemiscyllidae). *Parasitology* 27(3): 275–281.

Benz GW, Pratt HL, Adamson ML (1987). Larval philometrid nematodes (Philometridae) from the uterus of a sandbar shark, *Carcharhinus plumbeus*. *Proceedings of the Helminthological Society of Washington* 54(1): 154–155.

Benz GW, Bullard SA (2004). Metazoan parasites and associates of chondrichthyans with emphasis on taxa harmful to captive hosts. pp 325–346 *In:* The Elasmobranch husbandry manual: Captive Care of Sharks, Rays and their relatives. Ohio Biological Survey, Inc. Columbus, Ohio, United States of America, 481 p.

Beveridge I (1985). A redescription of *Echinocephalus uncinarus* Molin, 1858 (Nematoda, Gnathostomatoidea) from European rays, *Dusyatis pastinaca* (Linnaeus, 1758). *Bulletin du Museum National d'Hisfoire NaturelIe* (Paris, *4 series)* 7: 78 l–790.

Beveridge I (1991). The distribution of *Echinocephalus overstreeti* Deardorff and Ko (Nematoda), a parasite of Elasmobranch fishes in Australian waters. *Transactions of the Royal Society of South Australia* 115: 107.

Beveridge I, Sakanari JA (1987). *Lacistorhynchus dollfusi* sp. nov. (Cestoda: Trypanorhyncha) in elasmobranch fishes from Australian and North American coastal waters. *Transactions of the Royal Socieiy of South Australia* 111: 147–154.

Beveridge I, Campbell RA (1988). *Ceforhinicolu* n. g., *Shirleyrhynchus* n. g. and *Stragulorhynchus* n. g., three new genera of trypanorhynch cestodes from elasmobranchs in Australian waters. *Systematic Parasitology* 12: 47–60.

Beveridge I, Jones MK (2002). Diversity and biogeographical relationships of the Australian cestode fauna. *International Journal for Parasitology* 32: 343–351**.**

Beveridge I, Duffy C (2005). Redescription of *Cetorhinicola acanthocapax* Beveridge & Campbell, 1988 (Cestoda: Trypanorhyncha) from the basking shark *Cetorhinus maximus* (Gunnerus). *Systematic Parasitology* 62(3): 191–198 [Online] Available from: https://doi.org/10.1007/s11230-005-5492-9 [Accessed 25 July 2018].

Beveridge I, Justine JL (2006). Gilquiniid cestodes (Trypanorhyncha) from elasmobranch fishes off New Caledonia with descriptions of two new genera and a new species. *Systematic Parasitology* 65(3): 235–249.

Beveridge I, Bray RA, Cribb TH, Justine JL (2014). Diversity of trypanorhynch metacestodes in teleost fishes from coral reefs off eastern Australia and New Caledonia. *Parasite* 21:60 [Online] Available from: www. doi: 10.1051/parasite/2014060 [Accessed 9 October 2017].

Bhuiyan AI (2013). Endoparasitic helminths of *Tenualosa ilisha* in Bangladesh in relation to sex, seasons and habitat type. *Bangladesh Journal of Zoology* 41(2): 153–163.

Biosecurity New Zealand (2006). Risk analysis procedures. Version 1. Biosecurity New Zealand, Wellington [Online]. Available from: http://www.biosecurity.govt.nz/files/pests/surv-mgmt/surv/review/risk-analysis-procedures.pdf. [Accessed 24th January 2013].

Björnsdóttir B (2011). *Moritella viscosa* Virulence: Extracellular products and host-pathogen interaction. Ph.D thesis, Faculty of Medicine, University of Iceland, 184 p.

Blackwell RG (2013). Baitfish use in New Zealand: recent trends. Memorandum prepared for the Animal Imports team by Animals and Aquatics team, Ministry for Primary Industries, Wellington, New Zealand. 22 p.

Blasco-Costa I, Poulin R, Presswell B (2017). Morphological description and molecular analyses of *Tylodelphys* sp. (Trematoda: Diplostomidae) newly recorded from the freshwater fish *Gobiomorphus cotidianus* (common bully) in New Zealand. *Journal of Helminthology* 91: 332–345 [Online] Available from http://www.doi:10.1017/S0022149X16000298 [Accessed 29 January 2018].

Blasiola GC, Turnier JC (1979). Algal infection of the sevengill shark, *Notorynchus macula* Ayers. *Journal of Fish Diseases* 2: 161–163.

Borucinska D, Frasca S (2002a). Naturally occurring lesions and micro-organisms in two species of free-living sharks: the spiny dogfish, *Squalus acanthias* L., and the smooth dogfish, *Mustelus canis* (Mitchill), from the north-western Atlantic. *Journal of Fish Diseases* 25: 287–298.

Borucinska D, Frasca S (2002b). Pancreatic fibrosis and ductal ectasia associated with the nematode *Pancreatonema americanum* sp. nov., in spiny dogfish, *Squalus acanthias* L., from the north-western Atlantic. *Journal of Fish Diseases* 25(6): 367–370.

Borucinska D, Adams DH (2013). Parasitic metritis and oophoritis associated with nematode larvae in an Atlantic sharpnose shark, *Rhizoprionodon terraenovae* (Richardson), collected in the western Atlantic off Florida. *Journal of Fish Diseases* 36(9): 811–817.

Borucinska J (2016). Opportunistic infections in elasmobranchs, pp 67–80 *In:* Hurst CJ (*Ed*.). The Rasputin Effect: When Commensals and Symbionts Become Parasitic. Elsevier Science, New York, United States of America, 357 p.

Borzycm E, Matras M, Maj J, Samdomiersks A, Olesen NJ, Eliassen M, Baud M, Talbi C, Bigarre L (2013). First detection of hirame rhabdovirus (HIRRV) in Europe. Conference abstracts from the 16th Annual Meeting of the European Union National Reference Laboratories for Fish Diseases. Aarhus, Denmark, May 30–31, 2012, 78 p.

Bousfield EL, Kabata Z (1988). Amphipoda, pp 149–163 *In:* Margolis L, Kabata Z [*Eds*.] Guide to the parasites of fishes of Canada. Part II - Crustacea. *Canadian Special Publications of Fisheries and Aquatic Sciences* 101: 184p [Online] Available from: http://www.dfo-mpo.gc.ca/Library/181449.pdf [Accessed 16 April 2018].

Boustead NC (1981). Diseases in salmon hatcheries. pp 59–61. *In:* Taylor JL, Ogilvie RM, Todd, PR (*Eds.*). Proceedings of the Salmon Farming Conference, Ministry of Fisheries, New Zealand, August 1980. Occasional Publications No. 47. Fisheries Research Division Ministry of Fisheries, New Zealand.

Boustead NC (1982). Fish diseases recorded in New Zealand with a discussion on potential sources and certification procedures. Occasional Publications No. 34, Fisheries Research Division. Ministry of Fisheries, New Zealand, 18 p.

Boylan SM (2011). Zoonoses associated with fish. *Veterinary Clinics of North America Exotic Animal Practice* 14(3): 427–438.

Bowman M, Ramer J, Proudfoot J, Stringer E, Garner M, Trupkiewicz J, Giray C (2008). A novel adenovirus in a collection of wild-caught dusky smooth-hounds (*Mustelus canis*), pp 208−209 *In:* Proceedings of the 2008 AAZV (American Association of Zoo Veterinarians) Conference, Los Angeles, California, United States of America.

Bray RA1, Justine JL. A digenean parasite in a mudskipper: *Opegaster ouemoensis* sp. n. (Digenea: Opecoelidae) in *Periophthalmus argentilineatus* Valenciennes (Perciformes: Gobiidae) in the mangroves of New Caledonia. *Folia Parasitologica*; 60(1): 7–16.

Bricknell IR, Bron J, Bowden TJ (2005). Diseases of gadoid fish in cultivation: A review. *ICES Journal of Marine Science* 63(2): 253–266.

Bricknell I (2017). Types of Pathogens in Fish, Waterborne Diseases, pp. 261– 264 In: Fish diseases: Prevention and control strategies. Academic Press, New York, United States of America.

Briones V, Fernandez A, Blanco M, Ramiro F, de Vicente ML, Garcia J, Mendez JL, Goyache J (1998). Haemorrhagic septicaemia by *Aeromonas salmonicida* subsp. *salmonicida* in a blacktip reef shark (*Carcharhinus melanopterus*). *Zentralblatt fur Veterinarmedizin Reihe* B 45:443–445.

Brooke JS (2012). *Stenotrophomonas maltophilia*: an emerging global opportunistic pathogen. *Clinical Microbiology Reviews* 25(1): 2–41.

Brosnahan C, Pande A, Jones B (2015). Snapper mortalities: finding a cause. *Surveillance* 42(14): 23–26.

Bruno DW, Griffiths J, Mitchell CG, Wood BP, Fletcher ZI, Drobniewski FA, Hastings GS (1998). Pathology attributed to *Mycobacterium chelonae* among farmed and laboratory-infected Atlantic salmon (*Salmo salar*). *Diseases of Aquatic Organisms* 33: 101–109.

Bruno DW (2016). Infection with *Exophiala salmonis.* ICES identification leaflets for diseases and parasites of fish and shellfish No. 42. ICES Working Group on Pathology and Diseases of Marine Organisms. International Council for the Exploration of the Sea, Copenhagen, Denmark, 5 p.

Bryan DR, Jacobson KC, Buchanan JC (2012). Recent Increase in *Nybelinia surmenicola* Prevalence and Intensity in Pacific Hake (*Merluccius productus*) Off the United States West Coast. *Journal of Parasitology* 98(1): 85–92.

Buchmann K, Bresciani J (2006). Monogenea (Phylum Platyhelminthes), pp 297–344. *In:* Woo PTK (*Ed.*). Fish diseases and disorders, Volume 1. Protozoan and metazoan infections, 2nd edition. CABI International, Wallingford, United Kingdom, 791 p.

Buck JD (1990). Potentially pathogenic marine *Vibrio* species in seawater and marine animals in the Sarasota, Florida area. *Journal of Coastal Research* 6: 943–948.

Buck JD, Spotte S, Gadbaw JJ (1984). Bacteriology of the teeth from a great white shark: potential medical implications for shark bite victims. *Journal of Clinical Microbiology* 20(5): 849–851.

Burgess PJ, Mathews RA (1995). Fish host range of seven isolates of *Cryptocaryon irritans* (Ciliophora). *Journal of Fish Biology* 46: 727–729.

Bullard SA, Frasca S, Benz GW (2000a). Skin lesions caused by *Dermophthirius penneri* (Monogenea: Microbothriidae) on wild-caught blacktip sharks (*Carcharhinus limbatus). Journal of Parasitology* 86(3): 618–622

Bullard SA, Benz GW, Braswell JS (2000b). *Dionchus postoncomiracidia* (Monogenea: Dionchidae) from the skin of blacktip sharks, *Carcharhinus limbatus* (Carcharhinidae). *Journal of Parasitology* 86: 245–250.

Bullard SA, Overstreet RM, Carlson JK (2006). *Selachohemecus benzi* n. sp. (Digenea: Sanguinicolidae) from the blacktip shark *Carcharhinus limbatus* (Carcharhinidae) in the northern Gulf of Mexico. *Systematic Parasitology* 63: 143–154.

Bullard SA, Jensen K. (2008). Blood flukes (Digenea: Aporocotylidae) of stingrays (Myliobatiformes: Dasyatidae): *Orchispirium heterovitellatum* from *Himantura imbricata* in the Bay of Bengal and a new genus and species of Aporocotylidae from *Dasyatis sabina* in the Northern Gulf of Mexico. *Journal of Parasitology* 94: 1311–1321.

Buller NB (2014). Bacteria and fungi from fish and other aquatic animals, 2nd edition, CABI International, Wallingford, United Kingdom, 919 p.

Burreson E (2006). Phylum Annelida: Hirudinea as vectors and disease agents, pp 566–591. *In:* Woo PTK, Bruno DW, Lin LH (*Eds.*). Diseases and disorders of finfish in cage culture. CABI International, Wallingford, United Kingdom, 791 p.

Caira JN, Healy CJ, Jensen K (2012). An updated look at elasmobranchs as hosts of metazoan parasites, pp 548–650 *In:* Carrier JC, Musick JA, Heithaus MR (*Eds.*) Biology of sharks and their relative, 2nd Edition. CRC Press, 666 p.

Camus A, Dill J, McDermott A, Camus M, Ng TFF (2016a). Virus-associated papillomatous skin lesions in a giant guitarfish *Rhynchobatus djiddensis*: a case report. *Diseases of Aquatic Organisms* 117: 253–258.

Camus A, Dill J, McDermott A, Hatcher N, Griffin M (2016b). *Edwardsiella piscicida*-associated septicaemia in a blotched fantail stingray *Taeniura meyeni* (Müeller & Henle). *Journal of Fish Diseases* 39(9) 1125–1131.

Campbell RA, Beveridge I (1987a). *Florieceps minacanthus* sp. nov. (Cestoda: Trypanorhyncha) from Australian fishes. *Transactions of the Royal Society of South Australia* 111: 189–194.

Campbell RA, Beveridge I (1987b). *Hornelliella macropora* (Shipley & Hornell. 1906) comb. nov. (Cestoda: Trypanorhyncha) from Australian elasmobranch fishes and a re-assessment of the family Hornelliellidae. *Transactions of the Royal Society of South Australia* 111: 195–200.

Carlile G, East IJ, McColl KA, Ellard K, Browning GF, Crane M St J. (2014). The spatial and temporal variation of the distribution and prevalence of Atlantic salmon reovirus (TSRV) infection in Tasmania, Australia. *Preventive Veterinary Medicine* 116: 214–219.

Carr WH, Khoo L, Stoskopf MK (2003). Acute streptococcal septicemia in a nurse shark (*Ginglymostoma cirratum*). Session 5: Infectious diseases. Proceedings of the 34th International Association for Aquatic Animal Medicine **(**IAAAM) Conference, Kohala, Hawaii, May 10–14, 2003 [Online] Available from: http://www.vin.com/doc/?id=3863983 [Accessed 3 October 2017].

Carmichael JW (1966). Cerebral mycetoma of trout due to a phialophora-like fungus. *Sabouraudia* 5(2):120–123.

Certad G, Dupouy-Camet J, Gantois N, Hammouma-Ghelboun O, Pottier M, Guyot K, Benamrouz S, Osman M, Delaire B, Creusy C, Viscogliosi E, Dei-Cas E, Aliouat-Denis CM, Follet J (2015). Identification of *Cryptosporidium* species in fish from Lake Geneva (Lac Léman) in France. *PLoS One* 10(7) [Online] Available from: http://www.doi: 10.1371/journal.pone.0133047 : e0133047 [Accessed 26 June 2018].

Chacko A J (1993). Fungal and algal diseases of salmonids. *In*: Stoskopf MK (Ed.) Fish Medicine. Saunders, Pennsylvania, United States of America.

Chai JY, Lee SH (1991). Intestinal trematodes infecting humans in Korea. *Southeast Asian Journal of Tropical Medicine and Public Health* 22: 163–170.

Chai JL, Murrell KD, Lymbery AJ (2005). Fish-borne parasitic zoonoses: Status and issues. *International Journal for Parasitology* 35(11-12): 1233–1254 [Online] Available from: https://doi.org/10.1016/j.ijpara.2005.07.013 [Accessed 9 July 2018].

Chai JL, Sohn WM, Na BK, Jeong HG, Sinon M, Socheat D (2016). *Stellantchasmus falcatus* (Digenea: Heterophyidae) in Cambodia: Discovery of metacercariae in mullets and recovery of adult flukes in an experimental hamster. *Korean Journal of Parasitology* 54(4): 537–541.

Chandler Y, Goyal SM, Gupta SC (2006). Antimicrobial resistance of Providencia spp. Isolated from animal manure. *Veterinary Journal* 172(1): 188-191.

Chaudhary A, Verma C, Manu-Varma S, Singh HS (2013). A review of Monogenean diversity in India: Pathogens of fish diseases. *Journal of Coastal Life Medicine* 1(2): 151–168.

Chaudhary A, Verma C, Singh HS (2013). Morphological description of two new species of *Mastacembelocleidus* and *Trianchoratus* (Monogenea: Dactylogyridae) from India. *Science and Parasitology* 14(2): 69–76.

Cheung P (1993). Parasitic diseases of elasmobranchs, 782–808 *In:* Stoskopf MK (*Ed.*) Fish Medicine, Saunders, Philadelphia, United States of America.

Cheung PJ, Nigrelli RF (1983). *Dermophthirioides pristidis* n. gen., n. sp. (Microbothriidae) from the skin and *Neoheterocotyle ruggierii* n. sp. (Monocotylidae) from the gills of the smalltooth sawfish, *Pristis pectinata. Transactions of the American Microscopical Society* 102: 366–370.

Chisnall BL (2000). The Australian long finned eel *Anguilla reinhardtii* in New Zealand. Conservation Advisory Notes No. 302. Department of Conservation, Wellington, 16 p.

Chung DI1, Kong HH, Moon CH, Choi DW, Kim TH, Lee DW, Park JJ (1995). The first human case of *Diplogonoporus balaenopterae* (Cestoda: Diphyllobothriidae) infection in Korea. *Korean Journal of Parasitology* 33(3): 225–230.

Clarke SC, Francis MP, Griggs LH (2013). Review of shark meat markets, discard mortality and pelagic shark data availability, and a proposal for a shark indicator analysis. *New Zealand Fisheries Assessment Report* 2013/65, 74 p.

Clarke S, Sato M, Small C, Sullivan B, Inoe Y, Ochi D (2014). Bycatch in longline fisheries for tuna and tuna-like species: a global review of status and mitigation measures, pp 1-99 *In: FAO Fisheries and Aquaculture Technical Paper* 588. Food and Agriculture Organization of the United Nations, Rome, Italy, 226 p.

Clewley A, Kocan RM, Kocan AA (2002). An intraerythrocytic parasite from the spiny dogfish, *Squalus acanthias* L., from the Pacific Northwest. *Journal of Fish Diseases* 25: 693–696.

Cobb S (2008). Import risk analysis: Fish food. Biosecurity New Zealand, Ministry of Agriculture and Forestry, Wellington, New Zealand, 57 p.

Collins MR, Janovy J (2003). Host specificity among Ancyrocephalinae (Monogenoidea) of Nebraska sunfish. *Journal of Parasitology* 89(1): 80– 83**.**

Colorni A, Burgess P (1997). *Cryptocaryon irritans* Brown 1951, the cause of `white spot disease' in marine fish: an update. *Aquarium Sciences and Conservation* 1: 217–238.

Colquhoun DJ1, Duodu S (2011). *Francisella* infections in farmed and wild aquatic organisms. *Veterinary Research* 42(1): 47 [Online] Available from http://www/. doi: 10.1186/1297-9716-42-47 [Accessed 22 June 2018].

Corbeil S, Crane MS (2009). *Piscirickettsia salmonis. Australia New Zealand Standard Operating Procedure*. SCAHLS (Sub-committee on Aquatic Animal Health laboratory standards). Government of Australia, Canberra, 20 p.

Crane MStJ, Smith PH, Williams LM, Hyatt AD, Eaton LM, Gould A, Handlinger J, Kattenbelt J, Gudkovs N (2000). First isolation of an aquatic birnavirus from farmed and wild fish species in Australia. *Diseases of Aquatic Organisms* 43: 1–14.

Crane M StJ, Williams LM (2008). Viruses of salmonids: Virus isolation in cell lines. Australia and New Zealand Standard Diagnostic Procedure. 36 p. [Online] Available from http://www.agriculture.gov.au/ SiteCollectionDocuments/animal/ahl/ANZSDP-Virus-isolation.pdf) [Accessed 22 June 2018].

Crane M, Hyatt A (2011). Viruses of fish: An overview of significant pathogens. *Viruse*s 3: 2025–2046.

Cressy RF (1970). Copepods parasites on sharks from the west coast of Florida. *Smithsonian Contributions to Zoology* 38: 1–30, figs. 1–110.

Cribb TH, Bray RA, Barker SC (1992). A review of the Family Transversotrematidae (Trematoda: Digenea) with the description of a new Genus, *Crusziella. Invertebrate Taxonomy* 6: 909–935.

Cribb TH, Bray RA, Diaz PE, Huston DC, Kudlai GK, Martin SB, Yong RQ, Cutmore SC (2011). Trematodes of fishes of the Indo-west Pacific: told and untold richness. *Systematic Parasitology* 93: 237–247.

Cribb TH, Bott NJ, Bray RA, McNamara MKA, Miller TL, Nolan MJ, Cutmore SC (2014). Trematodes of the Great Barrier Reef, Australia: emerging patterns of diversity and richness in coral reef fishes. *International Journal for Parasitology* 44: 929–939.

Cribb TH, Chick RC, O'Connor W, O'Connor S, Johnson DJ, Sewell KB, Cutmore SC (2017). Evidence that blood flukes (Trematoda: Aporocotylidae) of chondrichthyans infect bivalves as intermediate hosts: indications of an ancient diversification of the Schistosomatoidea. *International Journal of Parasitology* (In press 2017), [Online] Available from: http://dx.doi.org/10.1016/j.ijpara.2017.05.008 [Accessed 9 October 2017].

Cruz CD (2015). Long-term study of *Vibrio parahaemolyticus* prevalence and distribution in New Zealand shellfish. *Applied and Environmental Biology* 81(7): 2320–2327.

Cudmore B, Mandrak NE (2011). Biological synopsis of tench (*Tinca tinca*). *Canadian Manuscript Report of Fisheries and Aquatic Sciences* 2948, 28 p.

Dadar M, Alborzi A, Peyghan R, Adel M (2016). Occurrence and intensity of Anisakid nematode larvae in some commercially important fish species in the Persian Gulf. *Iranian Journal of Parasitology* 11(2): 239–246.

da Fonseca MCG, de São Clemente SC, Felizardo NN, Gomes DC, Knoff M (2012). Trypanorhynch cestodes of hygienic-sanitary importance infecting flounders *Paralichthys patagonicus* Jordan, 1889 and *Xystreurys rasile* (Jordan, 1891) of the Neotropical region, Brazil. *Parasitological Research* 111(2): 865–874 [Online] Available from: https://doi.org/10.1007/s00436-012-2912-z [Accessed 2 August 2018].

Daoudi F, Radujkovi ČB, Marqués A, Bouix G (1987). Nouvelles espéces de coccidies (Apicomplexa, Eimeriidae) des genres *Eimeria* Schneider, 1875, et *Epieimeria* Dyková et Lom, 1981 parasites de poissons marins de la baie de Kotor (Yougoslavia). *Bulletin du Museum National d'Histoire Naturelle*, Paris, 4(9): 321–332.

Daly JG, Akoi T (2011). Pasturellosis and other bacterial diseases, pp 632–668 *In:* Woo PTK, Bruno DW (*Eds*.). Fish diseases and disorders. Volume 3: Viral, bacterial and fungal infections. CABI International, Wallingford, United Kingdom, 930 p.

Deardoff TL (1987). Redescription of *Pulchrascaris chiloscyllii* (Johnston and Mawson, 1951) (Nematoda: Anisakidae), with Comments on Species in *Pulchrascaris* and *Terranova. Proceedings of the Helminthological Society of Washington* 54(1): 28–39.

Decostere A, Hermans K, Haesebrouck F (1999). Piscine mycobacteriosis: A literature review covering the agent and the disease it causes in fish and humans. *Veterinary Microbiology* 99(3-4): 159–166.

Deets GB (1994). Copepod-Chondrichthyan coevolution: A cladistic consideration. Ph.D thesis, University of British Columbia. Canada.

de Groof A, Guelen L, Deijs M, van der Wal Y, Miyata M, Ng KS, van Grinsven L, Simmelink B, Biermann Y, Grisez L, van Lent J, de Ronde A, Chang SF, Schrier C, van der Hoek L (2015). A novel virus causes scale drop disease in *Lates calcarifer*. *PLoS Pathogen* 2015 Aug 7;11(8):e1005074 [Online]. Available from: doi: 10.1371/journal.ppat.1005074. eCollection 2015 [Accessed 12 August 2015].

De Hoog DS, Vicente VA, Najafzadeh MJ, Harrak MJ, Badeli H, Seyedmousavi S (2011). *Exophiala* species causing disease in cold-blooded animals. *Persoonia* 27: 46-72.

DermNet NZ (2014). Online medical dermatology database. DermNet New Zealand Trust [Online] Available from: https://www.dermnetnz.org/topics/erysipeloid/ [Accessed 21 June 2018].

Dias FJE, São Clemente SC, Pinto RM, Knoff M (2011). Anisakidae nematodes and Trypanorhyncha cestodes of hygienic importance infecting the king mackerel *Scomberomorus cavalla* (Osteichthyes: Scombridae) in Brazil. *Veterinary Parasitology* 175: 351–355.

Dick TA, Chambers C, Isinguzo I (2006). Cestoidea (Phylum Platyhelminthes), pp 391–416 *In:* Woo PTK (*Ed.*). Fish Diseases and Disorders, Volume 1: Protozoan and metazoan infections. CABI International, Wallingford, United Kingdom. Second Edition, 791 p.

Diebakate C, Raibaut A, Kabata Z (1997). *Thamnocephalus cerebrinoxius* n. g., n. sp. (Copepoda: Sphyriidae), a parasite in the nasal capsules of *Leptocharias smithii* (Müller & Henle, 1839) (Pisces: Leptochariidae) off the coast of Senegal. *Systematic Parasitology* 38(3):231–235.

Diggles BK (2003). Import risk assessment: Juvenile yellowtail kingfish (*Seriola Lalandi*) from Spencer Gulf aquaculture, South Australia. NIWA Client Report: WLG 2002/03. National Institute of Water and Atmospheric Research, Wellington, New Zealand, 54 p.

Diggles BK (2011). Environmental assessment report: Diseases of cultured salmon in New Zealand. DigsFish Services Client Report DF 11-02. August 2011, 68 p.

Diggles BK (2016). Updated disease risk assessment report – relocation of salmon farms in Marlborough Sounds, New Zealand. Digsfish Services Report: DF 16-01 prepared for Ministry of Fisheries, Nelson, New Zealand by Digsfish Services, Banks Beach, Queensland, Australia. 77 p.

Diggles BK, Hine PM, Handley S, Boustead NC (2002). A handbook of diseases of importance to aquaculture in New Zealand. NIWA Science and Technology Series, No. 49, 200 p.

Diggles BK, Hutson K (2005). Diseases of kingfish (*Seriola lalandi*) in Australasia. *Aquaculture Health International* 3: 12-14.

Doanh PN, Nawa & (2016). *Clonorchis sinensis* and *Opisthorchis* spp. in Vietnam: current status and prospects. *Transactions of the Royal Society for Tropical Medicine and Hygiene* 110(1):13–20.

Dopheide A, Lear G, Stott R, Lewis G (2011). Preferential feeding by the ciliates *Chilodonella* and *Tetrahymena* spp. and effects of these protozoa on bacterial biofilm structure and composition. *Applied and Environmental Microbiology* 77(13): 4564–4572. {Online] Available from Http://www.doi.10.1128/AEM.02421-10 [Accessed 26 June 2018].

Dove AM, Clauss M, Marancik DP, Camus AC (2017). Emerging diseases of elasmobranchs in aquaria, pp 263–276. Chapter 27 *In:* Smith M, Warmolts D, Thoney D, Hueter R, Murray M, Ezcurra J (*Eds.*) *The Elasmobranch Husbandry Manual II: Recent Advances in the Care of Sharks, Rays and their Relatives*. Special Publication of the Ohio Biological Survey, Columbus, Ohio, United States of America, 504 p.

Dippenaar SM, Mathibela RB, Bloomer P (2010). Cytochrome oxidase I sequences reveal possible cryptic diversity in the cosmopolitan symbiotic copepod *Nesippus orientalis* Heller, 1868 (Pandaridae: Siphonostomatoida) on elasmobranch hosts from the KwaZulu-Natal coast of South Africa. *Experimental Parasitology* 125(1): 42–50 [Online] Available from: www.doi:10.1016/j.exppara.2009.08.017 [Accessed 10 October 2017].

D'Silva J, Bhuiyan AI, Bristow GA (2012). Distribution of helminth parasites in size groups and organs of hilsa shad, *Tenualosa ilisha. Dhaka Iniversity Journal of Biological Science* 21(1): 55–65.

Durburrow RM, Kuchta R, Scholz T (2015). Paddlefish diseases, pp 227–265 (Chapter 8) *In:* Mims SD, Shelton WL (*Eds.*). Paddlefish Aquaculture (First edition), John Wiley, New York, United States of America, 320 p.

Dykova I (2006). Phylum Microspora, pp 205–229. *In:* Woo PTK (*Ed.*). Fish Diseases and Disorders, Volume 1: Protozoan and Metazoan Infections. CABI International, Wallingford, United Kingdom. Second edition, 791 p.

Dykova I, Lom J (1983). Fish coccidia: an annotated list of described species. *Folia Parasitologica* (Praha) 30: 193–208.

Dykova I, Peckova H, Kostka M (2008). Introduction of *Mayorella gemmifera* Schaeffer, 1926 into phylogenetic studies of Amoebozoa. *Acta Protozoologica* 47: 205–210.

Eaton WD, Kent ML (1992). A retrovirus in Chinook salmon (*Oncorhynchus tshawytscha*) with plasmacytoid leukaemia and evidence for the etiology of the disease. *Cancer Research* 52(23): 6496–6500.

Eaton WD, Folkin, B, Kent ML (1994). Biochemical and histological evidence of plasmacytoid leukaemia and salmon leukaemia virus (SLV) in wild-caught chinook salmon *Oncorhynchus tshawytscha* from British Columbia expressing plasmacytoid leukaemia. *Diseases of Aquatic Organisms* 19: 147-151.

Eiras JC (2002). Synopsis of the species of the genus *Henneguya* Thélohan, 1892 (Myxozoa: Myxosporea: Myxobolidae. *Systematic Parasitology* 52(1): 43–54.

Eiras J (2006). Synopsis of the species of *Ceratomyxa* Thélohan, 1892 (Myxozoa: Myxosporea: Ceratomyxidae). *Systematic Parasitology* 65: 49–71.

Eiras JC, Molnar K, Lu YS (2005). Synopsis of the species of *Myxobolus* Butschli, 1882 (Myxozoa: Myxosporea: Myxobolidae). *Systematic Parasitology* 61(1): 1–46.

ESR (2001). *Vibrio vulnificus*. Report prepared by Environmental Science and Research Ltd, Porirua, Wellington, for the Ministry of Health, New Zealand, 2 p.

Eszterbauer E, Szekely C, Molnar K, Ferenc B (2000). Development of *Myxobolus bramae* (Myxosporea: Myxobolidae) in an oligochaete alternate host, *Tubifex. Journal of Fish Diseases* 23(1):19–25.

Esterbauer E, Benko M, Dan A, Molnar K (2001). Identification of fish-parasitic *Myxobolus* (Myxosporea) species using a combined PCR-RFLP method. *Diseases of Aquatic Organisms* 44: 35–39.

EFSA (European Food Safety Authority) (2007). Possible vector species and live stages of susceptible species not transmitting disease as regards certain fish diseases. *EFSA Journal* 584: 1–163.

Evans JJ, Klesius PH, Plumb JA, Shoemaker CA (2011). *Edwardsiella* septicaemias, pp 512–569 *In:* Woo PTK, Bruno DW (*Eds.*). Fish Diseases and Disorders: Volume 3 - Viral, Bacterial and Fungal Infections, 2nd edition. CABI International, Wallingford, United Kingdom, 930 p.

Evdokimova EB, Kuznetzora IG, Stein GA (1969). Parasitic ciliates of the family Urceolariidae (Peritricha, Mobilia) from some fishes of the southwest Atlantic. *Zoologicheskii Zhurnal* 48: 1451–1455.

Eyngor OM, Zamostiano R, Kembou Tsofack JE, Berkowitz A, Bercovier H, Tinman S, Lev M, Hurvitz A, Galeotti M, Bacharach E, Eldar A (2014). Identification of a novel RNA virus lethal to tilapia. *Journal of Clinical Microbiology* 52: 4137– 4146 [Online] Available from: https://doi.org/10.1128/JCM.00827-14 [Accessed 13 June 2017].

FAO (2012). The state of world fisheries and aquaculture in 2012. FAO Fisheries and Aquaculture Department, Food and Agriculture Organisation of the United Nations, Rome, 209 p.

FAO (2015). State of the global market for shark products. Globefish – Analysis and information on world fish trade. FAO Fisheries and Aquaculture Department, Food and Agriculture Organisation of the United Nations, Rome, 188 p.

FAO (2016). The state of world fisheries and aquaculture (SOFIA) in 2016. FAO Fisheries and Aquaculture Department, Food and Agriculture Organisation of the United Nations, Rome, 209 p.

FAO (2017). Outbreaks of Tilapia Lake Virus (TiLV) threatens the livelihoods and food security of millions of people dependent on tilapia farming. FAO/GIEWS Special Alert No. 338. (Global Information and Early Warning System on Food and Agriculture, Food and Agriculture Organisation of the United Nations, Rome, Italy. 6 p. [Online] Available from: http://www.fao.org/3/a-i7326e.pdf [Accessed 14 June 2017].

FDA (Food and Drug Administration) (2018). Import alert for industry fishery/seafood products. Food and Drug Administration (FDA), United States Department of Health and Human Services, Silver Springs, Maryland, United States of America [Online] Available from: https://www.accessdata.fda.gov/cms_ia/industry_16.html [Accessed 16 August 2018].

Feist SW (2013). Differential characterization of emerging skin diseases of Rainbow Trout—a standardized approach to capturing disease characteristics and development of case definitions. *Journal of Fish Diseases* 36:921–937.

Feist SW, Longshaw M (2006). Phylum Myxozoa, pp 230–296 *In:* Woo PTK (*Ed.*) Fish diseases and disorders, Volume 1. Protozoan and Metazoan Infections, 2nd edition. CABI International, Wallingford, United Kingdom, 791 p.

Felix J (2013). Reported incidences of parasitic infections in marine mammals from 1892 to 1978. Zea Books, University of Nebraska, Lincoln, Nebraska, United States of America. [Online] Available from: HTTp:/www/digitalcommons.unl.edu/zeabook/] Accessed 20 July 2017].

Felizardo NN, Torres EJL, Fonseca GMC, Pinto RM, Gomes CD, Knoff M (2010). Cestodes of the flounder *Paralichthys isosceles* Jordan, 1890 (Osteichthyes - Paralichthyidae) from the state of Rio de Janeiro, Brazil. *Neotropical Helminthology* 4(2): 113–125.

Fernandez JB (1985). Estudo parasitilógici de *Merluccius australis* (Hutton 1872) (Pisces, Merlucciidae): aspectos sistemáticos, estadisticos y zoogeográficos. *Buletin de la Sociedad de Biologia de Concepción* 56: 31–41 (In Spanish).

Fijan N (1969). Systemic mycosis in channel catfish. *Wildlife Diseases* 5(2): 109 –10.

Fischthal JH (1953). *Bialovarium nocomis* n.g., n.sp. (Cestoda: Caryophyllaeidae) from the hornyhead chub, *Nocomis biguttatus* (Kirtland). *Journal of Parasitology* 39(4-2): 1–24.

Fish and Game (2014). New Zealand Fish and Game Council [Online]. Available from: http://www.fishandgame.org.nz/ [Accessed 24 September 2014].

Fishbase (2018). Fishbase: A global information system on Fishes, Fishbase. Froese R, Pauly D (Eds.). Metro Manilla, The Philippines [Online] Available from: https://www.Fishbase.de/home.htm# [Accessed 16August 2018].

Fitzgerald PR (1975). New coccidia from the spiny dogfish shark (*Squalus acanthias*) and great sculpin (*Myoxocephalus polyacanthocephalus*). *Journal of the Fisheries Research Board of Canada* 32: 649–651.

Forrest JJH, Cone DK (2011). Helminth parasites of golden shiner *Notemigonus crysoleucas* (Pisces: Cyprinidae) from Ontario and Nova Scotia, Canada. *Comparative Parasitology* 78(1): 220–222.

Frank SA, Jeffrey JS (2000). The probability of severe disease in zoonotic and commensal infections. *Journal of the Royal Society B* [Online] Available from http://www. doi.10.1098rsp.2000.1329 [Acessed 22 June 2018].

Froese R, Pauly D (2018). Fishbase. World Wide Web electronic publication version (02/2018) [Online] Available from: www.Fishbase.org [Accessed 26 March 2018].

FSANZ (2017). Australia New Zealand food standards code — Amendment No. 170. *New Zealand Gazette* 54: 1–4.

Garner MM (2013). A retrospective study of disease in elasmobranchs. *Veterinary Pathology* 50(3): 377–389.

Garner MM, Miller-Morgan TJ, Brown SR, Reimschuessel R, Poynton SL, Baldwin TI, Olson RE (1998). Microsporidiosis in Leopard Sharks. *In:* Abstracts of Proceedings of the Third International Symposium on Aquatic Animal Health. August 30-September 3, 1998. University of Maryland Aquatic Pathobiology Center, Baltimore, Maryland, United States of America, 145 p.

Garseth ÅH, Fritsvold C, Opheim M, Skjerve E, Biering E (2012). Piscine reovirus (PRV) in wild Atlantic salmon, *Salmo salar* L and seatrout, *Salmo trutta* L. in Norway. *Journal of Fish Diseases*, 36: 483–493.

Gaskins JE, Cheung PJ (1986). *Exophialia pisciphila*: a study on its development. *Mycopathologia* 93: 173– 184.

Genc E (2007). Infestation status of gnathiid isopod juveniles parasitic on Dusky grouper (*Epinephelus marginatus*) from the northeast Mediterranean Sea. *Parasitology Research* 101(3): 761-766.

Georgiades E, Fraser R, Joned B (2016). Options to strengthen on-farm biosecurity management for commercial and non-commercial aquaculture. *MPI Technical Paper* 2016/47. Ministry for Primary Industries, Wellington, New Zealand, 360 p.

Gibello A, Galan-Sanchez F, Bianco MM, Rodruiguez-Iglesias M, Dominiquez L, Fernandez-Garayzabal JF (2016). The zoonotic potential of *Lactococcus garvieae:* An overview on microbiology, epidemiology, virulence factors and relationship with its presence in foods. *Research in Veterinary Science* 109:59–70 [ON;ine] Available from http://www.doi: 10.1016/j.rvsc.2016.09.010 [Accessed 22 June 2018 [Accessed 22 June 2018].

Gibson-Kueh S, Chee D, Chen J, Wang YH, Tay S, Leong LN, Ng ML, Jones JB, Nicholls PK, Ferguson HW (2012). The pathology of 'scale drop syndrome' in Asian seabass, *Lates calcarifer* Bloch, a first description. *Journal of Fish Diseases* 35(1): 19–27 [Online]. Available from: http://www.doi.10.1111/j.1365-2761.2011.01319.x. Epub 2011 Nov 21. [Accessed 12 August 2015].

Gioia I, Cordeiro NS (1996). Brazilian Myxosporidians' check-list (Myxozoa). *Acta Protozoologica* 35: 137–49.

Gleeson RJ, Adlard RD (2011). Morphological and genetic analysis of three new species of *Ceratomyxa* Thélohan, 1892 (Myxozoa: Myxosporea) from carcharhinid sharks off Australia. *Systematic Parasitology* 80 (2): 117–124. {Online] Available from: www.DOI: 10.1007/s11230-011-9316-9 [Accessed 13 October 2017].

Gleeson RJ, Adlard RD (2012). Phylogenetic relationships amongst *Chloromyxum* Mingazzini, 1890 (Myxozoa: Myxosporea), and the description of six novel species from Australian elasmobranchs. *Parasitology International* 61: 267–274.

Glenn JA, Emmenegger EJ, Grady CA, Roon SR, Gregg JL, Conway CM, Winton JR, Hershberger PK (2012). Kinetics of viral load and erythrocytic inclusion body formation in Pacific herring artificially infected with erythrocytic necrosis virus. *Journal of Aquatic Animal Health* 24:195–200.

Goertz CEC (2004). Protozoan diseases of elasmobranchs, pp 417–427 *In:* Smith M, Warmolts D, Thoney D, Heuter R (*Eds*.) The Elasmobranch husbandry manual: captive care of sharks, rays and their relatives. Ohio Biological Survey, Inc., Colombus, Ohio, United States of America, 588 p.

Gomes MZR, Lewis RE, Kontoyiannis DP (2011). Mucormycosis caused by unusual *Mucormycetes*, non-*Rhizopus*, -*Mucor*, and -*Lichtheimia* species. *Clinical Microbiology Reviews* 24(2): 411–445.

Gonzales-Rey C, Stiltonen A, Pavlova A, Ciznar I, Svenson SB, Krovacek K (2011). Molecular evidence of *Plesiomonas shigelloides* as a possible zoonotic agent. *Folia Microbiologica* 56: 178. [Online] Available from https://www.doi.org/10.1007/s12223-011-0032-2 [Accssed 22 June 2018].

Goodwin AE (2012). Branchiomycosis. American Fisheries Service, United States Department of Agriculture [Online]. Available from: http://www.afsfhs.org/perch/resources/ 14069250504.2.1branchiomycosis2014.pdf [Accessed 12 November 2014].

Govedich FR (2017). Alphabetical list of Australian and New Zealand leech species. School of Biological Sciences, Monash University, Victoria, Australia [Online] Available from: https://www.invertebrate.us/leech/splist.htm [Accessed 6 October 2017].

Griffiths L (2015). Potential for and prevention of zoonoses associated with zebrafish. Environmental health and Safety, Princeton University, Princeton, New Jersey, United States of America [Online] Available from: https://www.ehs.princeton.edu/sites/ehs/files/media_files/Zebrafish_Zoonoses_EHS.pdf [Accessed 26 June 2018].

Gupta SP (1955). Two new trematodes belonging to the subfamily Opecoelinae Stunkard, 1931, with a key to the known genera. *Indian Journal of Helminthology* 5: 87–100.

Gupta N, Gupta DK, Shalaby SI (2008). Parasitic zoonotic infections in Egypt and India: an overview. *Journal of Parasitic Diseases* 32(1): 1–9.

Gutierrez R, Whangchai N, Sompong U, Prarom W, Iwami N, Itayama T, Nomura N, Sugira N (2013). Off-flavour in Nile tilapia (*Oreochromis niloticus*) cultured in an integrated pond-cage culture system. *Majeo International Journal of Science and Technology* 7: 1–13.

Haenen O (2017). Major bacterial diseases affecting aquaculture: Responsible management of bacterial diseases in Aquaculture. Aquatic AMR Workshop 1: 10-11 April 2017, Mangalore, India, FAO (Food and Agriculture Organisation of the United Nations, Rome, Italy [Online] Available from: http://www.fao.org/fi/static-media/MeetingDocuments/WorkshopAMR/presentations/07_Haenen.pdf [Accessed 25 June 2018].

Haenan OLM, Evans JJ, Berthe F (2013). Bacterial infections from aquatic species: potential for and prevention of contact zoonoses. *OIE Reviews in Science and Technology* 32(2): 497–507.

Halawani A (1930). On a new species of *Eimeria* (*E. southwelli*) from *Aetobatus narinari*. *Annals of Tropical Medicine and Parasitology* 24: 1–3.

Hammerschlag N, Davis DA, Mondo K, Seely MS, Murch SJ, Glover WB, Divoll T, Evers DC, Mash DC (2016). Cyanobacterial neurotoxin BMAA and Mercury. *Toxins* 8: 238. [Online] Available from: http://www.doi.10.3390/toxins8080238 [Accessed 19 October 2016].

Hanson L, Dishon A, Kotler M (2011). Herpesviruses that affect fish. *Viruses* 3: 2160–2191.

Heffernan H, Woodhouse R, Blackmore T (2005). Prevalence of extended-spectrum β-lactamases among urinary *Escherichia coli* and *Klebsiella* in New Zealand in 2006. Report FW0598 prepared by Environmental Science and Research Ltd, Porirua, Wellington, for the Ministry of Health, New Zealand, 29 p.

Hedrick RP, Groff JM, McDowell T, Wingfield WH (1990). An iridovirus infection of the integument of the white sturgeon *Acipenser transmontanus*. *Diseases of Aquatic Organisms* 8: 39–44.

Heffernan H, Woodhouse R, Blackmore T (2005). Report FW0598 prepared by Environmental Science and Research Ltd, Porirua, Wellington, for the Ministry of Health, New Zealand, 29 p.

Hegazi MA, Hassan AT, Al-Nashar TM, Abo-Elkheir OI, El-Lessi FM (2014). Encysted metacercariae of family Heterophyidae in infected fish in Dakahlia Governorate, an endemic focus in Egypt. *Journal of the Egyptian Society for Parasitology* 44(3): 547–558.

Henderson AC, Flannery K (2003). An investigation into the metazoan parasites of the spiny dogfish (*Squalus acanthias* L.), off the west coast of Ireland. *Journal of Natural History* 36(14): 1747–1760.

Hernández-Orts JS, Hernández-Mena DI2, Alama-Bermejo G, Kuchta R, Jacobson KC (2017). Morphological and molecular characterisation of *Aporocotyle margolisi* Smith, 1967 (Digenea: Aporocotylidae) from the North Pacific hake *Merluccius productus* (Ayres) (Gadiformes: Merlucciidae) off Oregon, USA. *Systematic Parasitology* 94(7): 819–829 [Online] Available from: http://www.doi: 10.1007/s11230-017-9740-6. Epub 2017 Jul 6 [Accessed 2 July 2018].

Hetrick RP, Sano T (1989). Herpesviruses of fish, *p* 161 *In:* Ahne W, Kurstak E (*Eds*.) Viruses of Lower Vertebrates, Springer-Verlag, Berlin, Germany, 496 p.

Hewitt GC (1967). Some New Zealand parasitic copepoda of the family Pandaridae, *New Zealand Journal of Marine and Freshwater Research* 1(2): 180–264 [Online] Available from www.doi:10.1080/00288330.1967.9515202 [Accessed 10 October 2017].

Hick P, Becker J, Whittington R (2016). Iridoviruses of fish, pp 127–152 *In:* Kibenge FSB, Godoy MG (*Eds*.). Aquaculture Virology, Academic Press, London, United Kingdom, 549 p.

Hine PM, Jones JB, Diggles BK (2000). A checklist of the parasites of New Zealand fishes, including previously unpublished records. *NIWA Technical Report* 75. National Institute for Water and Atmospheric Research, Wellington, New Zealand, 95 p.

Hine PM, Diggles BK (2005). Import risk analysis: Ornamental fish. Biosecurity New Zealand, Ministry of Agriculture and Forestry, Wellington, 262 p.

Hine PM, **Tham KM, Morrison R** (**2006**)**.** Cyprinid herpesvirus 2 in New Zealand goldfish. *Surveillance* 33: 3–5.

Hjeltnes B (2015). Virus associated disease in rainbow trout (virus Y), Fact sheet. Reitehaug E, Haukaas A (*Eds*.) Norwegian Veterinary Institute, 2 p, [Online] Available from: http://www.oestuecask.com/eng/Factsheets/Virus-associated-disease-in-rainbow-trout-virus-Y.html [Accessed 16 August 2018].

Hjeltnes B, Walde C, Bang-Jensen B, Haukaas A (2016). The Fish Health Report: 2015. The Norwegian Veterinary Institute's report series, no. 3: 2016. 76 p.

Hoffman GL (1967). Parasites of North American freshwater fishes. University of California Press, California, United States of America, 486 p.

Hofstra DE (2014). Grass carp effectiveness and effects. Stage 2: Knowledge review. *NIWA Client Report* HAM2014-060, prepared for the Department of Conservation, Wellington, New Zealand, 140 p.

Howgate P (2010). A critical review of total volatile bases and trimethylamine as indices of freshness of fish: Part 1. Determination. *Electronic Journal of Environmental, Agricultural and Food Chemistry* 9(1): 29–57.

Huang KC, Wei-Hsu RD (2005). *Vibrio fluvialis* haemorrhagic cellulitis and cerebritis. *Clinical and Infectious Diseases* 40(9): 75–77.

Huffman DG, Moravec F (1988). First description of adult *Huffmanela huffmani* Moravec, 1987 (Nematoda: Trichosomoididae) from the swimbladder of centrarchid fishes of the upper San Marcos River, central Texas. *Folia Parasitologica* 35: 227–234.

Humphrey J D (1995). Australian quarantine policies and practices for aquatic animals and their products: a review for the scientific working party on aquatic animal quarantine. Bureau of Resource Sciences, Department of Primary Industries and Energy, Canberra, Australia.

Huon (2014). Huon Aquaculture: Prospectus. Huon Aquaculture, Sydney, New South Wales, Australia, 155 p.

Ikmam R (2013). Antimicrobial resistance in New Zealand: What is my role in primary care? *Best Practice Journal* 54: 3–7.

Interaminense JA, Nascimento DCO, Ventura RF, Batista JEC, Souza MMC, Hazin FHV, Pontes-Filho NT, Lima-Filho JV (2010). Recovery and screening for antibiotic susceptibility of potential bacterial pathogens from the oral cavity of shark species involved in attacks on humans in Recife, Brazil. *Journal of Medical Microbiology* 59: 941–947 [Online] Available from: http://www.doi.10.1099/jmm.0.020453-0 1 [Accessed 19 October 2017].

Isaksen TE, Karlsbakk E, Watanabe K, Nylund A (2011). *Ichthyobodo salmonis* sp. n. (Ichthyobodonidae, Kinetoplastida), an euryhaline ectoparasite infecting Atlantic salmon (*Salmo salar* L.). Parasitology 138(9): 1164–1175 [Online] Available from: http://www.doi.10.1017/S0031182011000916 [Accessed 14 April 2018].

Jameson AP (1931). Notes on Californian Myxosporidia. *Journal of Parasitology* 18: 59–68.

Jiang Y (2009). Haemorrhagic disease of grass carp: Status of outbreaks, diagnosis, surveillance and research. *Israeli Journal of Aquaculture* 61(3): 188–197.

Johnston C (2008a). Import risk analysis: Frozen, skinless and boneless fillet meat of *Oreochromis* spp. from China and Brazil for human consumption. MAF Biosecurity New Zealand, 96 p.

Johnston C (2008b). Import risk analysis: Frozen skinless and boneless fillet meat of *Pangasius* spp. fish from Vietnam for human consumption. MAF Biosecurity New Zealand, 60 p.

Jones JB (1988). New Zealand parasitic Copepoda; genus *Caligus* Müller, 1785 (Siphonostomatoida: Caligidae). *New Zealand Journal of Zoology* 15(3): 397–413.

Jones JB, Delahunt B (1995). *Phlyctainophora lamnae* (Nematoda; Philometridae) from dogfish *Squalus acanthias* off southern New Zealand. *International Journal for Parasitology* 25(3): 395–397.

Jung S, Kitamura SI, Song JY, Oh MJ (2007). *Miamiensis avidus* (Ciliophora : Scuticociliatida) causes systemic infection of olive flounder *Paralichthys olivaceus* and is a senior synonym of *Philasterides dicentrarchi. Diseases of Aquatic Organisms* 73: 227–234.

Justine JL (2005). *Huffmanela lata n. sp.* (Nematoda: Trichosomoididae: Huffmanelinae) from the shark *Carcharhinus amblyrhynchos* (Elasmobranchii: Carcharhinidae) off New Caledonia. *Systematic Parasitology* 61: 181–184 [Online] Available from http://www.DOI 10.1007/s11230-005-3160-8 [Accessed 19 October 2017].

Kabata Z (1970). Crustaceans as enemies of fishes, pp 1–171 *In:*. Snieszko SF, Axelrod HR (*Eds.*). Diseases of fishes. Tropical Fish Hobbyist Publications, Jersey City, New Jersey, United States of America.

Khan RA, Barrett M, Murphy J (1980). Blood parasites of fish from the north-western Atlantic Ocean. *Canadian Journal of Zoology* 58: 770–781.

Kahn SA, Beers PT, Findlay VL, Peebles IR, Durham PJ, Wilson DW, Gerrity SE (1999). Import risk analysis on non-viable salmonids and non-salmonid marine finfish. Australian Quarantine and Inspection Service, Department of Agriculture, Fisheries and Forestry, Canberra, Australia, 409 p.

Kalavati C, Narasimhamurti CC (1985). Histopathological changes in the gills of *Channa punctatus* BL. infected with *Henneguya waltairensis. Archiv für Protistenkunde* 129:199–202 [Online] Available from: http://www. doi: 10.1016/S0003-9365(85)80023-3 [Accessed 15 August 2018].

Kasai A, Tsuduki H, Jimenez LA, Li YC, Tanaka S, Sato H (2017). Incidence of three *Kudoa* spp., K. *neothunni, K. hexapunctata*, and *K. thunni* (Myxosporea: Multivalvulida), in *Thunnus* tunas distributed in the western Pacific Ocean. *Parasitological Research* 116: 1137–1150 [Online] Available from: http://www.DOI 10.1007/s00436-016-5369-7 [Accessed 21 February 2018].

Kato E, Kasai A, Tomochi H, Li YC, Sato H (2017). Four *Myxobolus* spp. (Myxosporea: Bivalvulida) from the gill lamellae of common carp (*Cyprinus carpio*) and Japanese silver crucian carp (*Carassius langsdorfii*) in the western part of Japan, with the description of three new species (*M. tanakai* n. sp., *M. paratoyamai* n. sp., and *M. ginbuna* n. sp.). *Parasitological Research* 2017:5 [Online] Available from: http://www. doi:10.1007/s00436-017-5545-4 [Accessed 21 February 2018].

Kearn GC (1965). The biology of *Leptocotyle minor*, a skin parasite of the dogfish, *Scyliorhinus canicula. Parasitology* 55:473–480.

Kearn GC, James R, Evans-Gowing R (1993). Insemination and population density in *Entobdella soleae*, a monogenean skin parasite of the common sole, *Solea solea. International Journal of Parasitol*ogy 23: 891–899.

Keeling SE, Brosnahan CL, Johnston C, Wallis R, Gudkovs N, McDonald WL (2013). Development and validation of a real-time PCR assay for the detection of *Aeromonas salmonicida. Journal of Fish Diseases* 36: 495–503.

Kennedy M (2004). Flatworm Cysts in Fish: the North American Liver Fluke Alberta Agriculture and Forestry, Government of Alberta, Canada [Online] Available from: https://www.agriculture.alberta.ca/app21/ministrypage [Accessed 10 August 2018].

Khalifa KA (1985). Leeches on freshwater farmed fish in Iraq. *Journal of Wildlife Diseases* 21: 312–313.

Khan RA (1972). Taxonomy, prevalence, and experimental transmission of a protozoan parasite, *Trichodina oviducti* Polyanskii (ciliata: Peritrichida) of the thorny skate, *Raja radiata* Donovan. *Journal of Parasitology* 58: 680–685.

Khidr AE, Samak OA, Said AE, Ghoneim AM, Fahmy SA (2014). Structural and functional observations on the appendages of gill parasite, *Lernanthropus kroyeri* (Copepoda: Lernanthropidae) infesting the sea bass *Dicentrarchus Labrax. Nature and Science* 12(2): 101–107.

King AMQ, Lefkowitz E, Adams M, Carstens EB (2011). Virus taxonomy: Ninth report of the International Committee on the Taxonomy of Viruses. Elsevier Science, New York, United States of America, 1338 p.

Kingklib C, Suanyuk N (2017). *Streptococcus agalactiae* serotype Ib, an emerging pathogen affecting climbing perch (*Anabas testudineus*) and Günther's walking catfish (*Clarias macrocephalus*) polycultured in southern Thailand. *The Thai Journal of Veterinary Medicine* 47(2), 183–197 [Online] Available from: https://www.tci-thaijo.org/index.php/tjvm/article/view/90276 [Accessed 13 April 2018].

Kino H, Hori W, Kobayashi H, Nakamura N, Nagasawa K (2002). A mass occurrence of human infection with *Diplogonoporus grandis* (Cestoda: Diphyllobothriidae) in Shizuoka Prefecture, central Japan. *Parasitology International* 51(1): 73–79 [Online] Available from: https://doi.org/10.1016/S1383-5769(01)00106-4 [Accessed 27 July 2018].

Kirs M, Dpanola A, Fyfe R, Krantz J, Van Laanen A, Cotton D, Castle M (2011). A survey of oysters (*Crassostrea gigas*) in New Zealand for *Vibrio parahaemolyticus* and *Vibrio vulnificus. International Journal of Food Microbiology* 147(2): 149–153.

Klapholz A, Lessnau KD, Huang B, Talavera W, Boyle JF (1994). *Hafnia alvei:* Respiratory tract isolates in a community hospital over a three-year period and a literature review. *Chest* (Journal of the American College of Chest Physicians)105(4): 1098–100.

Kluzik M, Woodford N (2016). Zoonoses associated with fish (including aquarium fish). Washington State University Institutional Animal Care and Use Committee. Washington State University, Spokane, Washington, United States of Amwrica [Online] Available from: https://iacuc.wsu.edu/zoonoses-associated-with-fish/ [Accessed 21 June 2018].

Knoff M, Carmona-de-Sao Clemente S, Pinto RMP, Gomes DC (2001a). Nematodes of elasmobranch fishes from the southern coast of Brazil. *Memoirs of the Institute of Oswaldo Cruz* 96(1): 81–87.

Knoff M, Carmona-de-Sao Clemente S, Pinto RMP, Gomes DC (2001b). Digenea and Acanthocephala of elasmobranch fishes from the southern coast of Brazil. *Memoirs of the Institute of Oswaldo Cruz* 96(8): 1095–1101.

Ko RC (2006). Fish-bourne parasitic zoonoses, pp 592–677 *In:* Woo PTK (*Ed*.). Fish Diseases and Disorders, Volume 1: Protozoan and Metazoan Infections. CABI International, Wallingford, United Kingdom. Second edition, 791 p.

Kohl-Yakimoff N, Yakimoff WL (1915). Hamogregarinen der Seefische. *Centrablatt für Bakteriologie, Parasitenkunde und Infektionskrankheiten* 76: 134–146.

Koie M, Karlsbakk E, Nylund A (2008). The marine herring myxozoan *Ceratomyxa auerbachi* (Myxozoa: Ceratomyxidae) uses *Chone infundibuliformis* (Annelida: Polychaeta: Sabellidae) as invertebrate host. *Folia Parasitologica* 5: 100–104.

Kovaljova AA (1988). Myxosporidia of the genus *Chloromyxum* (Cnidospora, Myxosporea) of cartilaginous fishes from the Atlantic Coast of Africa. *Parazitologiia* 22: 384–388 (In Russian with English abstract).

Kozinska A, Pekala A (2004). First isolation of *Shewanella putrefaciens* from freshwater fish – a potential new pathogen of the fish. *Bulletin of the European Association of Fish Pathologists* 24(4): 199–203.

Krailas D, Namchote S. Koonchornboon T, Dechruksa W, Boonmekam D (2014). Trematodes obtained from the thiarid freshwater snail *Melanoides tuberculata* (Müller, 1774) as vector of human infections in Thailand. *Zoosystematics and Evolution* 90(1): 57–86.

Kritsky DC, Fennessey CJ (1999). *Calicobenedenia Polyprioni* n. gen., n. sp. (Monogenoidea: Capsalidae) from the external surfaces of wreckfish, *Polyprion americanus* (Teleostei: Polyprionidae), in the north Atlantic. *Journal of Parasitology* 85(2): 192–195.

Kritsky DC (2010). *Rhabdosynochus* Spp. (Monogenoidea: Diplectanidae) infecting the gill lamellae of snooks, *Centropomus* spp. (Perciformes: Centropomidae), in Florida, and redescription of the type species, *R. rhabdosynochus. Journal of Parasitology* 96(5):879–868.

Kritsky DC, McAleese WJ, Bakenhaster MD (2011). Heteronchoineans (Monogenoidea) from the gills of Creville jack, *Caranx hippos* (Perciformes, Carangidae), from Everglades National Park, Florida, with a Redescription of *Protomicrocotyle mirabilis* (Gastrocotylinea, Protomicrocotylidae). *Comparative Parasitology* 78(2): 265–274 [Online] Available from https://doi.org/10.1654/4505.1 [Accessed 22 February 2018].

Kumari R, Kuman C (2015). Fungal infection in some economically important fresh water fishes in Gandak River near Muzaffarpur region of Bihar. *International Journal of Life Science and Pharma Research* 5(1): 1–11.

Kurata O, Munchan C, Wada S, Hatai K, Miyoshi Y, Fukuda Y (2008). Novel *Exophiala* infection involving ulcerative skin lesions in Japanese Flounder *Paralichthys olivaceus. Fish Pathology* 43: 35–44.

Kirtisinghe P (1964). A review of the parasitic copepods of fish recorded from Ceylon with description of additional forms. *Bulletin of the Fisheries Research Station, Ceylon* 17: 45–132.

Kuchta R (2013). Tapeworm *Diphyllobothrium dendriticum* (Cestoda)—Neglected or emerging human parasite? *PLoS Neglected Tropical Diseases* 7(12): e2535 [Online] Available from: http://www.doi. 10.1371/journal.pntd.0002535 [Accessed 14 April 2018].

Kutcha R, Serrano-Martinez ME, Scholz T (2015). Pacific Broad Tapeworm *Adenocephalus pacificus* as a Causative Agent of Globally Reemerging Diphyllobothriosis. *Emerging Infectious Diseases* 21(10): 1697–1703 [Online] Available from: https://dx.doi.org/10.3201/eid2110.150516 [Accessed 4 October 2017].

Kuznetsova IG (1977). Myxosporidians of Chondrostei from the Patagonian shelf. *Parazitologiia* 11: 74–7 (In Russian with English abstract).

Labrie L, Tan JN, Komar C, Ho E, Grizel L (2008). Nocardial infections in fish: An emerging problem in both freshwater and marine aquaculture systems in Asia, pp 297–312 *In*: Bondad-Reantaso MG, Mohan CV, Crumlish M, Subasinghe RP (*Eds*.). Diseases in Asian Aquaculture VI. Fish Health Section, Asian Fisheries Society, Manila, Philippines, 505 p.

Laird M (1951). Studies on the trypanosomes of New Zealand fish. *Proceedings of the Zoological Society of London* 1221: 285–309.

Laird M (1952). The protozoa of New Zealand intertidal fishes. *Transactions and Proceedings of the Royal Society of New Zealand* 81(1): 79–143.

Laird M (1959). *Caliperia brevipes* n. sp. (Ciliata: Peritrichia) epizoic on *Raja erinacea* Mitchell at Saint Andres, New Brunswick. *Canadian Journal of Zoology* 37: 283–288.

Landcare (2014). *Stenotrophomonas maltophilia.* New Zealand Fungi, Landcare New Zealand [Online]. Available from: http://nzfungi2.landcareresearch.co.nz/default.aspx?selected=NameDetails&Action=Display& CancelScript=

1&TabNum= 1&NameId=3E858298-7E2E-4F29-90B5-D9DDD283F9FF&StateId=&Sort=0 [Accessed 10 August 2015].

Land based aquaculture assessment framework (LBAAF) (2018). Kingfish [Online] Available from http://www.lbaaf.co.nz/nz-aquaculture-species/modelled-species/kingfish/ [Accessed 2 March 2018].

Lawler AR (1981). Zoogeography and host specificity of the Super-Family Capsaloidea Price 1936 (Monogenea: Monopisthocotylea) *Special Papers in Marine Science* 6. Virginia Institute of Marine Science, Gloucester Point,Virginia, United States of America, 650 p.

LBAAF (2018). Land based aquaculture assessment framework (LBAAF), Ngati Porou Fisheries Ltd, Gisborne, New Zealand [Online] Available from: http://www.lbaaf.co.nz/nz-aquaculture-species/ [Accessed 16August 2018].

Lee JY, Kim JW, Park GM (2016). Plerocercoids of *Nybelinia surmenicola* (Cestoda: Tentacularidae) in Squids, *Todarodes pacificus*, from East Sea, the Republic of Korea. *The Korean Journal of* Parasitology 54(2): 221-224.

Leibovitz L, Lebouitz SS (1985a). A viral dermatitis of the smooth dogfish, *Mustelus canis*. *Journal of Fish Diseases* 8: 273–279.

Leibovitz L, Lebouitz SS (1985b). Skin diseases of sharks. *In:* Sharks: Recent advances in captive biology. A Symposium on the Captive Maintenance of Elasmobranchs, March 31-April 3, 1985. Organized by the National Aquarium in Baltimore and Department of Pathology, University of Maryland School of Medicine, Baltimore, Maryland, USA. Abstract. 6 p.

Lefebvre F, Schuster T, Münderle M, Hine M, Poulin R (2004). Anguillicolosis in the short-finned eel *Anguilla australis*: Epidemiology and pathogenicity. *New Zealand Journal of Marine and Freshwater Research* 38(4): 577–583 [Online] Available from: http://www. Doi.10.1080/00288330.2004.9517263 [Accessed 14 February 2018].

Lepa A, Siwicki AJ (2012). Fish herpesvirus diseases: a short review of current knowledge. *Acta Veterinaria BRNO* 81: 383–389.

Lester RJ, Hayward CJ (2006). Phylum Arthropoda, pp 466–565 *In*: Woo PTK (*Ed.*). Fish diseases and disorders, Volume 1. Protozoan and Metazoan Infections, 2nd edition. CABI International, Wallingford, United Kingdom, 791 p.

Lette V, Temesgen Z (2007). Parasitic and fungal infections, pp 1273–1295 (Chapter 94) *In:* Schapira SHV (*Ed*.). Neurology and Clinical Neuroscience. Mosby, Elsevier, New York, United States of America, 1664 p. [Online] Available from https://doi.org/10.1016/B978-0-323-03354-1.50098-5 [Accessed 26 June 2018].

Levine ND (1985). *Erhardorina* n. g., *Ascogregarina polynesiensis* n. sp., *Eimeria golemanskii* n. sp., *Isospora tamariscini* n. sp., *Gregarina kazumii* n. nom., new combinations and emendations in the names of apicomplexan protozoa. *Journal of Protozoology* 32(2): 359–363.

Lewis AG (1966). Copepod crustaceans parasitic on elasmobranch fishes of the Hawaiian Islands. *Proceedings of the United States National Museum: Smithsonian Institute* 118(3524): 57–154.

Lewis S, Chinabut S (2011). Mycobacteriosis and Nocardiosis, pp 397– 423 *In:* Woo PTK, Bruno DW (*Eds*.). Fish Diseases and Disorders, Volume 3. CABI International, Wallingford, United Kingdom, 930 p.

Lewish E, Gorgoglione B, Way K, El-Matbouli M (2015). Carp edema virus/Koi sleepy sickness: an emerging disease in Central-East Europe. *Transboundary and Emerging Diseases* 62(1): 6–12.

Lindquist HLA, Cross JH (2017). *Capillaria philippinensis*, pp 1763–1799 *In:* Cohen J, Powderly WG, Opal SM (*Eds*.) Infectious Diseases, volume 2 (Fourth Edition). Elsevier, New York, United States of America, 2016 p.

Liu SF, Peng WF, Gao P, Fu MJ, Wu HZ, Lu MK, Gao JQ, Xiao J (2010). Digenean parasites of Chinese marine fishes: a list of species, hosts and geographical distribution. *Systematic Parasitology* 75:1–52.

Lobna SM, Metawea YF, Elsheikha HM (2010). Prevalence of heterophyiosis in Tilapia fish and humans in Northern Egypt. *Parasitology Research* 107(4): 1029–1034.

Loch TP, Faisal M (2015). Emerging flavobacterial infections in fish: A review. *Journal of Advanced Research* 6(3): 283–300.

Lom J, Dykova I (1992). Protozoan Parasites of Fishes, pp 26–28 *In:* Volume 26 (Developments in Aquaculture and Fisheries Science), 1st edition. Elsevier, Amsterdam, the Netherlands. 315 p.

Lom, J. Dyková I (2006). Myxozoan genera: definition and notes on taxonomy, life-cycle terminology and pathogenic species. *Folia Parasitologica* 53: 1–36.

Lowry T, Smith SA (2007). Vet Med Today: Zoonosis Update. *Journal of the Aquatic Veterinary Medical Association* 6: 876–880.

MacDiarmid SC (1994). The risk of introducing exotic diseases of fish into New Zealand through the importation of ocean-caught Pacific salmon from Canada. Ministry of Agriculture Regulatory Authority, Wellington, New Zealand, 170 p.

MacKerras MJ, MacKerras IM (1961). The haematozoa of Australian frogs and fish. *Australian Journal of Zoology* 9: 123–139.

MAF (Ministry of Agriculture and Forestry) (2009). Import risk analysis: Tropical, subtropical and temperate freshwater and marine ornamental fish and marine molluscs and crustaceans. MAF Biosecurity New Zealand, Wellington, New Zealand, 157 p.

MAF (Ministry of Agriculture and Forestry) (1999). Import risk analysis: Head-on, gill-in Australian salmonids for human consumption. Biosecurity Authority, Ministry of Agriculture and Forestry, Wellington, New Zealand, 24 p.

MAF (Ministry of Agriculture and Forestry) (2000). Supplementary import risk analysis: Head-on, gill-in Australian salmonids for human consumption. Review of submissions. MAF Biosecurity Authority, Ministry of Agriculture and Forestry. Wellington, New Zealand, 39 p.

Maillard C, Ktari MH (1978). *Hyperandrotrema cetorhini* n. g., n. sp. (Trematoda, Sanguinicolidae) parasite du système circulatoire de *Cetorhinus maximus* (Selacii). *Annales de Parasitologie Humane et Comparée* 53: 359–365.

Marino F, Lanteri G, Manganaro M, Macri F (2010). European eel stomatopapillomatosis in the south Tyrrhenian Sea: surgical excision and post-surgical recovery. *Bulletin of the European Association of Fish Pathology* 30(1): 30–34.

Marsh D, Mkwra L (2013). Review of freshwater non-market value studies. Department of Economics, University of Waikato, Hamilton, New Zealand, 144 p.

Martins ML, Onaka EM, Bozzo FR, Fenerick J (2004). *Henneguya* sp. (Myxozoa: Myxobolidae) in *Pimelodus maculatus* (Osteichthyes: Siluridae) from Volta Grande reservoir, Minas Gerais, Brazil, *Bulletin Instituta Pescatoria* (Brazil) 30(1): 1–7.

Maskell WM (1886). On freshwater infusoria of the Wellington district. *Transactions and proceedings of the Royal Society of New Zealand* 20(1): 49–61.

McColl KA, Sunarto A, Williams LM, Crane M (2007). Koi herpesvirus: Dreaded pathogen or white knight? *Aquaculture Health International* 9: 4-6.

McColl KA, Davies KR, Young JG, Crane, MStJ (2009). Aquatic birnavirus infections of finfish. Australia and New Zealand Standard Diagnostic Procedure, Department of Agriculture and Water Resources, Canberra, Australia, 20 p. [Online] Available from: http://www.agriculture.gov.au/SiteCollectionDocuments/animal/ahl/ANZSDP-Aquatic-birnaviruses.pdf [Accessed 15 August 2018].

McDougall S, Hussein H, Petrovski K (2014). Antimicrobial resistance in *Staphylococcus aureus*, *Streptococcus uberis* and *Streptococcus dysgalactiae* from dairy cows with mastitis. *New Zealand Veterinary Journal* 62(2): 68–76.

McDowall RM (1976). The role of estuaries in the life cycles of fishes in New Zealand. *Proceedings of the New Zealand Ecological Society* 23: 27–37.

McDowall RM (2000). The Reed field guide to New Zealand freshwater fishes. Reed, Auckland, 256 p.

McKenna PB (2001). An annotated checklist of helminth and protozoan parasites of frogs in New Zealand. *Surveillance* 28(2): 11–12.

McKenna P (2010). An updated checklist of helminth and protozoan parasites of birds in New Zealand. *Parasitology* 1(9): WCM00705 [Online]. Available from: http://www.webmedcentral.com/article_view/705 [Accessed 18 December 2014].

McKiernan JP, Alexandra S, Grutter AS, Davies AJ (2005). Reproductive and feeding ecology of parasitic gnathiid isopods of epaulette sharks (*Hemiscyllium ocellatum*) with consideration of their role in the transmission of a haemogregarine. *International Journal of Parasitology* 35: 19–27.

McVicar AH, Gibson DI (1975). *Pancreatonema torriensis* gen. nov., sp. nov. (Nematoda: Rhabdochonidae) from the pancreatic duct of *Raja naevus. International Journal of Parasitology* 5: 529–535.

Melo FT, e Silva JP, Costa-Goncalves E, Furtado AP, Giese EG, Santos CP, Santos JN (2015). Taxonomic status and redescription of the genus *Brasicystis* Thatcher, 1979 (Digenea: Didymozoidae). *Parasitology International* 62: 201–214.

Melville V, Johnston C (2010). Supplementary risk analysis: extension of countries eligible to export frozen, skinless, boneless fillet meat of *Pangasiu*s spp., under the import health standard fisfilic.spe. MAF Biosecurity New Zealand, Wellington, 9 p.

Meyers TR, Hauck AK, Blankenbeckler WB, Minicucci T (1986). First report of viral erythrocytic necrosis in Alaska, USA, associated with epizootic mortality in Pacific herring, *Clupea harengus pallasii* (Valenciennes). *Journal of Fish Diseases* 9: 479-491.

Michel C, Messiaen S, Bernardet JF (2002). Muscle infections in imported neon tetra *Paracheirodon innesi* Myers: Limited occurrence of microsporidia and predominance of severe forms of columnaris disease caused by an Asian genomovar of *Flavobacterium columnare. Journal of Fish Diseases* 25: 253–263.

Midani S, Rathore M (1998). *Chromobacterium violaceum* infection. *Southern Medical Journal* 91(5): 464–466.

Mikalsen, A. B., Kim, S. H., & Evensen, Ø. (2017). Piscine myocarditis virus ORF3 encodes a cytotoxic protein, which increase virulence and viral titer of an attenuated heterologous model virus. 18th International Conference on Diseases of Fish and Shellfish – EAFP 2017, Belfast, Northern Ireland (Poster) (Quoted in Garseth AH, Frissvold C, Svendsen JC, Bang-Jensen B, Mikalsen AB (2017). Cardiomyopathy syndrome in Atlantic salmon *Salmo salar* L., A review of the current state of knowledge. *Journal of Fish Diseases* 41(1): 11–26 [Online] Available from: http://www. https://doi.org/10.1111/jfd.12735 [Accessed 16 August 2018].

Miller MH (2016). Endangered Species Act status review report: Smooth hammerhead shark (*Sphyrna zygaena*). Report to the National Marine Fisheries Service, Office of Protected Resources, Silver Spring, Maryland, United States of America, 167 p.

Miller TL, Adlard RD (2013). *Unicapsula* species (Myxosporea: Trilosporidae) of Australian marine fishes, including the description of *Unicapsula andersenae* n. sp. in five teleost families off Queensland, Australia. *Parasitology Research* 112(8): 2945–2957.

Mitchell CP (1980). Control of water weed by grass carp in two small lakes. *New Zealand Journal of Marine and Freshwater Research* 14(4): 381–390.

MOH (Ministry of Health) (2017). *Hartmanella vermiformis.* Datasheet 1.2: Guidelines for drinking water quality management for New Zealand. Ministry of Health, Wellington, New Zealand, 51 p.

MOH (Ministry of Health) (2018). Meningococcal disease (including meningitis). Know the symptoms. Health resources: Health Ed, Ministry of Health, Wellington, New Zealand [Online] Available from https://www.healthed.govt.nz/ resource/ meningococcal-disease-know-symptoms [Accessed 27 March 2018].

Mohilal N, Hemananda T (2012). Record of the species of *Tripartiella* (Lom, 1959) from fishes of Manipur. *Journal of Parasitic Diseases* 36(1): 87–93.

Molnar K (2006). Phylum Apicomplexa, pp 183–229 *In:* Woo PTK (*Ed.*). Fish diseases and disorders, Volume 1. Protozoan and Metazoan Infections, 2nd edition. CABI International, Wallingford, United Kingdom, 791 p.

Molnár K, Székely C, Mohamed K, Shaharom-Harrison F (2006). Myxozoan pathogens in cultured Malaysian fishes. I. Myxozoan infections of the sutchi catfish *Pangasius hypophthalmus* in freshwater cage cultures. *Diseases of Aquatic Organisms* 68: 209–218 [Online] Available from: https://doi.org/10.3354/dao068209 [Accessed 17 January 2018].

Molnar K, Cech G, Szekely C (2008). Infection of the heart of the common bream, *Abramis brama* (L.), with *Myxobolus* s.l. *dogieli* (Myxozoa, Myxobolidae). *Journal of Fish Diseases* 31(8): 613–620.

Montero FE, Aznar FJ, Fernández M, Raga JA (2003). *Pseudoallencotyla* gen. n., a new genus for *Allencotyla pricei* (Kritsky, Noble et Moser 1978) (Monogenea: Heteraxinidae), with a key to genera of Heteraxininae. *Folia Parasitologica* 50: 43–47.

Monod T (1926). Les Gnathidae. Essai monographique (morphologie, biologie, systematique). *Memoires de la Societe des Sciences Naturelles du Maroc* 13, 667 p.

Montero FE, Raga JA, Kostadinova A (2008). Composition and structure of the parasite faunas of cod, *Gadus morhua* L. (Teleostei: Gadidae), in the North East Atlantic. *Parasites & Vectors* 1: 23. [Online] Available from http://www.doi:10.1186/1756-3305-1-23 [Accessed 10 April 2018].

Moran JDW, Arthur JR, Burt MDB (2011). Parasits of sharp-beaked redfishes (*Sebastes fasciatus* and Sebastes *mentella*) collected from the Gulf of St Lawrence, Canada. *Canadian Journal of Fisheries and Aquatic Sciences* 53(8): 1821–1826.

Moravec F, Borucinska JD, Frasca S (2001). *Pancreatonema americanum* sp. nov. (Nematoda, Habdochonidae) from the pancreatic duct of the dogfish shark, *Squalus acanthias*, from the coast of Massachusetts, USA. *Acta Parasitologica* 46: 293–298.

Moravec F, Justine JL (2006). Three nematode species from elasmobranchs off New Caledonia. *Systematic Parasitology* 64: 131–145.

Morillas J, George-Nascimento M, Valeria H, Khan RA (1987). *Trypanosoma humboldti* n. sp. from the Chilean catshark, *Schroederichthys chilensis* (Guichenot, 1848). *Journal of Protozoology* 34: 342–344.

Morris T (2012). Investment opportunities in the New Zealand Salmon industry. Report from the Food and Beverage Information Project. Coriolis Ltd, Wellington, New Zealand, 46 p.

Morsey K, Bashtar AR, Ghaffar FA, Al Quraishy S, Al Ghamdi A, Mostafa N (2013). First identification of four trypanorhynchid cestodes: *Callitetrarhynchus speciosus*, *Pseudogrillotia* sp. (Lacistorhynchidae), *Kotorella pronosoma* and *Nybelinia bisulcata* (Tentaculariidae) from Sparidae and Mullidae fish. *Parasitology Research* 112(7): 2523–2532.

Mousavi HAE, Omidzahir S, Soltani M, Shayan P, Ebrahimzadeh E, Mousavi S, Hoseini H (2012). Morphometrical and molecular characterization of *Gyrodactylus cichlidarum* (Gyrodactylidae) from *Astronotus ocellatus* (Cichlidae) in Iran. *Comparative Clinical Pathology* 5 pp. [Online] Available from http://www.DOI 10.1007/s00580-012-1534-2 [Accessed 22 February 2018].

Montgomery H (1998). Mycobacteria in New Zealand. *Surveillance* 26(1): 6–8.

Moore JP (1939). Additions to our knowledge of African leeches (Hirudinea*). Proceedings of the Academy of Natural Sciences, Philadelphia* 90: 297–360.

MPI (Ministry for Primary Industries) (2010). Import health standard for sand tiger sharks (*Carcharias taurus*) from all countries FISSHARIC.ALL, Ministry for Primary Industries, Wellington, New Zealand, 11 p.

MPI (Ministry for Primary Industries) (2013). Animal health laboratory. *Surveillance* 40(3): 9–13.

MPI (Ministry for Primary Industries) (2016). Animal Products Notice: Specifications for products intended for human consumption. Ministry for Primary Industries, Wellington, New Zealand, 126 p.

MPI (Ministry for Primary Industries) (2017a). Import Health Standard: Ornamental fish and marine invertebrates. Ministry for Primary Industries, Wellington, New Zealand, 77 p.

MPI (Ministry for Primary Industries) (2017b). Live animals [Online] Available from http://mpi.govt.nz/importing/live-animals/ [Accessed 14 June 2017].

MPI (Ministry for Primary Industries) (2018). Annual Review report for deepwater fisheries 2016/17. Ministry for Primary Industries, Wellington, New Zealand [Online] Available from: https://www.mpi.govt.nz/dmsdocument/29741/ [Accessed 16August 2018].

Muller R (2002). Worms and human disease (2nd edition). CABI International, Wallingford, United Kingdom, 310 p.

Munchan C, Kurata O, Wada S, Hatai K, Sano A, Kamei K, Nakaoka N (2009). *Exophiala xenobiotica* infection in cultured striped jack, *Pseudocaranx dentex* (Bloch & Schneider), in Japan. *Journal of Fish Diseases* 32: 893–900.

Munday BL (2002). Scientific review of significant disease agents carried by non-viable, non-salmonid freshwater and estuarine finfish and their products. Biosecurity Australia, Department of Agriculture, Fisheries and Forestry, Canberra, Australia, 117 p.

Munro ES, Midtlyng PJ (2011). Infectious pancreatic necrosis virus and associated birnaviruses. pp 1–65. *In:* Woo PTK, Bruno DW (*Eds.*). Fish Diseases and Disorders: Volume 3 - Viral, Bacterial and Fungal Infections, 2nd edition. CABI International, Wallingford, United Kingdom, 930 p.

Murrell KD, Fried B (2007). Food-borne parasitic zoonoses: fish and plant-borne parasites. Springer Science, New York, United States of America, 429 p.

Musick JA (2005). Shark utilisation, pp 243–251 *In:* Musick JA, Bonfil R (*Eds*.). Management techniques for elasmobranch fisheries. *FAO Fisheries Technical Paper* 474. Food and Agriculture Organisation of the United Nations (FAO). Rome, Italy, 251 p.

Nair GA, Nair NB (1983). Effect of infestation with the isopod, *Alitropus typus* M. Edwards (Crustacea: Flabellifera: Aegidae) on the haematological parameters of the host fish, *Channa striatus* (Bloch). *Aquaculture* 30(1–4): 11–19.

Nagabayashi T, Wolf K (1979). Characterisation of EV-2, a virus isolated from European eels (*Anguilla anguilla*) with stomatopapilloma. *Journal of Virology* 30(1): 358–364.

Nakajima K, Inouye K, Sorimachi M (1998). Viral diseases in cultured marine fish in Japan. *Fish Pathology* 33: 181–188.

Nareaho A, Eriksson-Kallio AM, Heikken P, Snellman A, Sukura A, Koski P (2017). High prevalence of zoonotic trematodes in roach (*Rutilus rutilus*) in the Gulf of Finland. *Acta Veterinaria Scandanavica* 59: 75 [Online] Available from: http://www.doi: 10.1186/s13028-017-0343-7 [Accessed 20 July 2018].

Neumann RO (1909). Studien uber protozoische parasiten im blut von meeresfischen. *Zeitschrift fur Hygiene und Infektionskrankheiten* 64: 1–112 (English abstract).

Nickol BB (2006). Phylum Acanthocephala, *pp* 444–465 *In:* Woo PTK, Buchmann K (*Eds*.) Fish parasites: Pathobiology and Protection. CABI International, Wallingford, United Kingdom, 383 p.

NIWA (National Institute of Water and Atmospheric Research) (2014a). Atlantic salmon. NIWA Fish Atlas. National Institute of Water and Atmospheric Research, Auckland, New Zealand. (Website https://www.niwa.co.nz/freshwater-and-estuaries/nzffd/NIWA-fish-atlas/fish-species/atlantic_salmon, downloaded 15 April 2015).

NIWA (National Institute of Water and Atmospheric Research) (2014b). Grass carp *Ctenopharyngodon idella* (Valenciennes, 1844) [Online] Available from: https://www.niwa.co.nz/freshwater-and-estuaries/nzffd/NIWA-fish-atlas/fish-species/grass_carp [Accessed 12 February 2015].

NIWA (National Institute of Water and Atmospheric Research) (2017). Yellowtail Kingfish. National Institute for Water and Atmospheric Research (NIWA), Auckland, New Zealand [Online] Avalable from: https://www.niwa.co.nz/aquaculture/aquaculture-species/yellowtail-kingfish [Accessed 14 August 2017].

NIWA (National Institute of Water and Atmospheric Research) (2018). Freshwater fish species, NIWA Atlas of freshwater fishes. National Institute for Marine and Freshwater Research Ltd, Auckland, New Zealand [Online] Available from: https://www.niwa.co.nz/freshwater-and-estuaries/nzffd/NIWA-fish-atlas/fish-species/ [Accessed 12 January 2018].

Noble ER (1948). A new myxosporidian (Protozoan) parasite from *Scyliorhinus torazame. Transactions of the American Microscopical Society* 67(3): 254–256 [Online] Available from: http://www.doi:10.2307/3223188 [Accessed 23 October 2017].

Noga EJ (2014). Fish disease: Diagnosis and treatment. John Wiley, New York, United States of America. 536 p.

Nolan MJ, Cribb TH (2006). An exceptionally rich complex of Sanguinicolidae von Graff, 1907 (Platyhelminthes: Trematoda) from Siganidae, Labridae and Mullidae (Teleosti: Percifiormes) from the Indo-West Pacific Region. *Zootaxa* 1218: 3–80.

Norman BM, Newbound DR, Knott B (2000). A new species of Pandaridae (Copepoda), from the whale shark *Rhincodon typus* (Smith). *Journal of Natural History*, 34(3): 355–366.

Novoslavskij A, Terentjeva M, Eizenberga I, Valcinja O, artkevics V, Berzins A (2015). Major foodborne pathogens in fish and fish products: a review. *Annals of Microbiology* 2015 [Online] Available from: http://www.doi.10.1007/s13213-015-1102-5 [Accessed 30 July 2018].

Nowak BF, Clark A (2001). Prevalence of epitheliocystis in Atlantic salmon, *Salmo salar* L., farmed in Tasmania, Australia. *Journal of Fish Diseases* 22(1): 73–78.

Nowak BF, LaPatra SE (2006). Epitheliocystis in fish. *Journal of Fish Diseases* 29: 573– 588.

Nguyen HD, Nakai T, Muroga K (1996). Progression of striped jack nervous necrosis disease (SJNNV) infection in naturally and experimentally infected striped jack *Pseudocaranx dentex* larvae. *Diseases of Aquatic Organisms* 24: 99–105.

NZFUNGI (2017). NZFUNGI2 (New Zealand Fungi and Bacteria): Online database. Manaaki Whenua Landcare Research Ltd, Lincoln, New Zealand [Online] Available from: https://www.landcareresearch.co.nz/ resources/data/ nzfungi [Accessed 30 July 2018].

NZOR (New Zealand Organisms Register) (2017). New Zealand Organisms Register [Online]. Available from: http://www.nzor.org.nz/008f7dlb_eb47_4d68_a956_6b9cc0b98202 [Accessed 7 April 2015].

Oerlis-Ribeiro R, Ruiz CF, Curran SS, Bullard SA (2013). Blood Flukes (Digenea: Aporocotylidae) of Epipelagic Lamniforms: Redescription of *Hyperandrotrema cetorhini* from Basking Shark (*Cetorhinus maximus*) and Description of a New Congener from Shortfin Mako Shark (*Isurus oxyrinchus*) off Alabama. *Journal of Parasitology* 99(5): 835–846.

Ogawa K, Fukudome M (1994). Mass mortality caused by blood fluke (*Paradeontacylix*) among amberjack (*Seriola dumerili*) imported to Japan. *Fish Pathology* 29(4):265–269.

Ogawa K (2012). *Heterobothrium okamotoi* and *Neoheterobothrium hirame*, pp 245-259 *In*: Woo PTK, Buchmann K (*Eds.*) Fish parasites: Pathobiology and Protection. CABI International, Wallingford, United Kingdom, 383 p.

Ogawa K, Egusa S (1981). The systematic position of the genus *Anoplodiscus* (Monogenea: Anoplodiscidae). *Systematic Parasitology* 2(4): 253–260.

Ogawa K, Egusa E (1986). Two new species of *Paradeontacylix* McIntosh, 1934 (Trematoda: Sanguinicolidae) from the vascular system of a cultured marine fish *Seriola purpurascens. Fish Pathology* 21: 15–19.

Ogawa K, Delgahapitiya KP, Furuta T, Wakabayayashi H (1992). Histological studies on the host response to *Myxobolus artus* Akhmerov, 1960 (Myxozoa: Myxobolidae) infection in the skeletal muscle of carp, *Cyprinus carpio* L.. *Journal of Fish Biology* 41: 363–371 [Online] Available from: http://www.doi:10.1111/j.1095-8649.1992.tb02665.x [Accessed 21 February 2018].

Oidtmann B, Joiner C, Reese RA, Stone D, Dodge M, Dixon P (2011). Risks associated with commodity trade: Transmission of viral haemorrhagic septicaemia virus (VHSV) to rainbow trout fry from VHSV-carrying tissue homogenates. *Transboundary and Emerging Diseases* 58: 224–231.

Oidtmann B, Johnston C, Klotins K, Mylrea G, Van PT, Cabot S, Rosado-Martin P, Ababouch L, Berthe F (2013a). Assessment of the safety of aquatic animal commodities for international trade: The OIE Aquatic Animal Health Code. *Transboundary and Emerging Diseases* 60(1): 27–38.

Oidtmann B, LaPatra SE, Verner-Jeffreys D, Pond M, Peeler EJ, Noguera PA, Bruno DW, St-Hilaire S, Schubiger CB, Snekvik K, Crumlish M, Green DM, Metselaar M, Rodger H, Schmidt-Posthaus H, Galeotti M, Feist SW (2013b). Differential characterization of emerging skin diseases of rainbow trout--a standardized approach to capturing disease characteristics and development of case definitions. *Journal of Fish Diseases 36*(11): 921–937.

Oidtmann B, Dixon P, Way K, Joiner C, Bayley AE (2017). Risk of waterborne virus spread – review of survival of relevant fish and crustacean viruses in the aquatic environment and implications for control measures. *Reviews in Aquaculture* 0: 1–29 [Online]. Available from: www. doi: 10.111/raq.12192 [Accessed 15 March 2017].

OIE (2010). Handbook on import risk analysis for animals and animal product – Volume 1: Introduction and qualitative risk analysis, 2nd Edition. World Organization for Animal Health. Paris, France, 88 p.

OIE (2014). New disease in rainbow trout from Norway [Online] Available from: http://www.oie.int/wahis_2/public/wahid.php/Reviewreport/Review?page_refer=MapFullEventReportandreportid=15460 [Accessed 21 June 2014].

OIE (2016a). Aquatic Animal Health Code, 19th Edition. World Organization for Animal Health. Paris, France [Online]. Available from: http://www.oie.int/international-standard-setting/aquatic-code/access-online/ [Accessed 15 March 2017].

OIE (2016b). Manual of diagnostic tests for aquatic animal. World Organization for Animal Health. Paris, France [Online] Available from: http://www.oie.int/international-standard-setting/aquatic-manual/access-online/ [Accessed 16 May 2017].

OIE (2017). Tilapia lake virus (TiLV) – a novel orthomyxo-like virus:: Disease card. World Organisation for Animal Health, Paris, France [Online] Available from: http://www.oie.int/fileadmin/Home/eng/Internationa_Standard_Setting/docs/pdf/Aquatic_Commission/A_TiLV_disease_card.pdf [Accessed 14 June 2017].

Oosthuizen JH (1987). Redescription of *Hemiclepsis quadrata* (Moore, 1939) n. comb. (Hirudinea: Glossiphoniidae). Systematic Parasitology 10: 73–78.

Oosthuizen JH (1989). Redescription of the African leech *Batrachobdelloides tricarinata* (Blanchard, 1897). *Hydrobiologia* 184: 153–164.

Overstreet RM (1977). *Poecilancistrium caryophyllum* and other trypanophynch cestode plerocercoids from the musculature of *Cynoscion nebulosus* and other sciaenid fishes in the Gulf of Mexico. Journal of Parasitology 63(5): 780–789.

Paladini G, Hansen H, Fiorvanti ML, Shinn AP (2011). *Gyrodactylus longipes* n. sp. (Monogenea: Gyrodactylidae) from farmed gilthead seabream (*Sparus aurata* L.) from the Mediterranean. *Parasitology International* 60(4): 410–418 [Online] Available from http://www.doi: 10.1016/j.parint.2011.06.022 [Accessed 26 February 2018].

Palm HW, Walter T, Schwerdtfeger G, Reimer LW (1997). *Nybelinia* Poche, 1926 (Cestoda: Trypanorhyncha) from the Moçambique coast, with description of *N. beveridgei* sp. nov. and systematic consideration of the genus, *South African Journal of Marine Science*, 18(1): 273–285 [Online] Available from http://www/doi.10.2989/025776197784161018 [Accessed 20 August 2018].

Palm HW, Overstreet RM (2000). New records of trypanorhynch cestodes from the Gulf of Mexico, including *Kotorella pronosoma* (Stossich, 1901) and *Heteronybelinia palliata* (Linton, 1924) comb. n. *Folia Parasitica* 47: 293–302.

Palm HW, Waeschenbach A, Littlewood DT (2007). Genetic diversity in the trypanorhynch cestode *Tentacularia coryphaenae* Bosc, 1797: evidence for a cosmopolitan distribution and low host specificity in the teleost intermediate host. *Parasitological Research* 101(1): 153–159.

Paperna I (1980). *Amyloodinium ocellatum* (Brown, 1931) (Dinoflagellida) infestations in cultured marine fish at Eilat, Red Sea: Epizootiology and pathology. *Journal of Fish Diseases* 3: 363–372.

Paperna I (1996). Parasites, infections and diseases of fishes in Africa - An update. *FAO/CIFA Technical Paper* 31. Food and Agriculture Organisation of the United Nations, Rome, Italy 220p.

Paperna I, Zwerner DE (1974). Massive leech infection on a white catfish (*Ictalurus catus*): a histological consideration. *Proceedings of the Helminthological Society of Washington* 41: 64–67.

Paperna I, Dzikowski R (2006). Digenea (Phylum Platyhelminthes), pp 345-390 *In*: Woo PTK (*Ed.*), Fish Diseases and Disorders. Volume 1: Protozoan and Metazoan Infections, CABI International, Wallingford, United Kingdom, 791 p.

Parisielle A, Euzet L (1995). *Scufogyrus* gen. n. (Monogenea: Ancyrocephalidae) for *Cichlidogyrus longicornis* minus DOSSOU, 1982, C. *1. longicornis*, and C. *1. gravivaginus* Paperna and Thurston, 1969, with Description of Three New Species Parasitic on African Cichlids. *Journal of the Helminthological Society of Washington* 62(2): 157– 173.

Pascual S, Rodriguez H, Pierce GJ, Hastie LC, Gonzalez AF (1017). The NE Atlantic European hake: A neglected high exposure risk for zoonotic parasites in European fish markets. *Fisheries Research* 202: 69–78 [Online] Available from https://doi.org/10.1016/j.fishres.2017.12.008 [Accessed 26 June 2018].

Paul L (2000). New Zealand fishes. Reed Penguin, London, United Kingdom, 253 p.

Paulin C, Stewart A, Roberts C, McMillan P (2001). New Zealand fish: A complete guide. Te Papa Press, Museum of New Zealand Te Papa Tongarewa, Wellington, New Zealand, 279 p.

Pavlidis M, Mylonas C (2011). Sparidae: Biology and aquaculture of gilthead sea bream and other species. John Wiley, New York, 416 p.

Pearce FM, Oidtmann BC, Thrush MA, Dixon PF, Peeler EJ (2014). Do imports of rainbow trout carcasses risk introducing viral haemorrhagic septicaemia virus into England and Wales? *Transboundary and Emerging Diseases* 61: 247–257.

Pelayo V, Garcia-Hernandez P, Puente P, Rodero M, Cuebilar C (2009). Seroprevalence of anti-*Gymnorhynchus gigas* (Trypanorhyncha, Gymnorhynchidae) antibodies in a Spanish population. *Journal of Parasitology* 95: 778–780.

Petrie-Hanson L (2011). First reported mortality and associated pathology attributed to *Acolpenteron ureteroecetes* in largemouth bass. *Journal of Aquatic Animal Health* 13(4): 364–367.

Piasecki W, Goodwin AE, Eiras JC, Nowak BF (2004). Importance of copepoda in freshwater aquaculture. *Zoological Studies* 43(2): 193–205.

Pirarat N, Sahatrakul K, Lacharoje S, Lombardini E, Chansue N, Techangamsuwan S (2016). Molecular and pathological characterization of *Fusarium solani* species complex infection in the head and lateral line system of *Sphyrna lewini. Diseases of Aquatic Organisms* 120: 195–204.

Plumb JA, 1989. Channel catfish herpesvirus. Viruses of lower vertebrates, pp 198–216 In: Proceedings of the 1st International Symposium on Viruses of Lower Vertebrates, Munich, August 1988.

Plumb JA, Hanson LA (2011). Health maintenance and principal microbial diseases of cultured fishes. John Wiley, New York, 400 p.

Polkinghorne A, Schmidt-Posthaus H, Meijer A, Lehner A, Vaughan L (2010). Novel Chlamydiales associated with epitheliocystis in a leopard *shark Triakis semifasciata. Diseases of Aquatic Organisms* 91: 75–81.

Powell JL, Loutit MW (1990). Isolation and characterisation of *Vibrio anguillarum* from selected marine sites around New Zealand. *New Zealand Journal of Marine and Freshwater Research* 24: 267–273.

Prakash MS (2015). On a new species of Monogenea *Diplozoon chauhani* n.sp. (Diplozooidae) from Indian fresh water food fish *Cirrhinus mrigala. International Journal of Fisheries and Aquatic Studies* 2015; 2(4): 140–141.

Praveen P, Debnath C, Shekhar S, Dalai N, Ganguly S (2016). Incidence of *Aeromonas* spp. infection in fish and chicken meat and its related public health hazards: A review. *Veterinary World* 9(1): 6–11 [Online] Available from: http://www. Doi.10.14202/vetworld.2016.6-11 [Accessed 16August 2018].

PROMED (2017). Pilchard orthomyxovirus – Australia (Tasmania) Farmed salmon [Online] Available from: http://www.promedmail.org/direct.php?id=20171221.5517876 [Accessed 9 July 2018].

Pulsford A (1984). Preliminary studies on trypanosomes from the dogfish, *Scyliorhinus canicula* L. *Journal of Fish Biology* 24: 671–682.

Quinn TP, Kinnison MT, Unwin MT (2001). Evolution of Chinook salmon (*Oncorhynchus tshawytscha*) populations in New Zealand: pattern, rate and process. *Genetica* 112–113: 493–515.

Rajendram P, Kyaw WM, Leo YS, Ho H, Chen WK, Lin R, Pratim P, Badaruddin H, Ang B, Barkham T, Chow A (2016). Group B *Streptococcus* sequence type 283 disease linked to consumption of raw fish, Singapore. *Emerging Infectious Diseases* 22(11): 1974–1977 [Online] Available from: http://dx.doi.org/10.3201/eid2211.160252 [Accessed 24 July 2018].

Rausch RL, Adams AM (2000). Natural transfer of helminths of marine origin to freshwater fishes, with observations on the development of *Diphyllobothrium alascense. The Journal of Parasitology* 86(2): 319–327.

Redondo MJ, Palanzula O, Riaza A, Macias A, Alvarez-Pelletiro P (2002). Experimental transmission of *Enteromyxum scophthalmi* (Myxozoa), an enteric parasite of turbot *Scophthalmus maximus. Journal of Parasitology* 88(3): 482–488.

Reed C, MacKenzie K, van der Lingen CD (2012). Parasites of South African sardines, *Sardinops sagax*, and an assessment of their potential as biological tags. *Bulletin of the European Association of Fish Pathologists* 32(2): 41–48.

Reimschuessel R1, Gieseker CM, Driscoll C, Baya A, Kane AS, Blazer VS, Evans JJ, Kent ML, Moran JD, Poynton SL (2003). Myxosporean plasmodial infection associated with ulcerative lesions in young-of-the-year Atlantic menhaden in a tributary of the Chesapeake Bay, and possible links to *Kudoa clupeidae.* Diseases of Aquatic Organisms 53(2):143–166.

Reichley SR, Ware C, Steadman J, Gaunt PS, Garcia JC, LaFrentz BR, Thachil A, Waldbieser JC, Stine CB, Arias CR, Loch T, Welch TJ, Cipriano RC, Greenway TE, Khoo1 LH, Wise DJ, Lawrence ML, Griffin MJ (2018). Comparative phenotypic and genotypic analysis of *Edwardsiella* spp. isolates from different hosts and geographic origins, with an emphasis on isolates formerly classified as *E. tarda* and an evaluation of diagnostic methods. *Journal of Clinical Microbiology* [Online] Available from: http://www. doi:10.1128/JCM.00970-17 [Accessed 25 June 2018].

Reno PW, Serreze DV, Hellyer SK, Nicholson BL (1985). Hematological and physiological effects of viral erythrocytic necrosis (VEN) in Atlantic cod and herring. *Fish Pathology* 20: 353–360.

Reshnetikova AV (1965). Effect of *Digramma interrupta* plerocercoids on the growth of bream. *Zoological Journal* 44(5): 734–739 (In Russian, English abstract).

Reuter RE, Hutchinson W, Ham J, Davis S. 2003. *Exophiala* sp. infection in captured King George whiting (*Sillaginodes punctata*). *Bulletin of the European Association of Fish Pathologists* 23: 128– 134.

Rimmer AE, Becker JA, Tweedie A, Lintermans M, Landos M, Stephens F, Whittington RJ (2015). Detection of dwarf gourami iridovirus (Infectious spleen and kidney necrosis virus) in populations of ornamental fish prior to and after importation into Australia, with the first evidence of infection in domestically farmed Platy (*Xiphophorus maculatus*). *Preventative Veterinary Medicine* 122(1-2): 181–194. [Online] Available from: http://www.doi: 10.1016/j.prevetmed.2015.09.008. Epub 2015 Sep 21 [Accessed 20 Februay 2018].

Rimmer AE, Whittington RJ, Tweedie A, Becker AJ (2017). Susceptibility of a number of Australian freshwater fishes to dwarf gourami iridovirus (Infectious spleen and kidney necrosis virus). *Journal of Fish Diseases* 40: 293–310.

Roberts RJ (2012). Fish Pathology, Fourth Edition, 592 p.

Robinson ES (1961). Some monogenetic trematodes from marine fishes of the Pacific. *Transactions of the American Microscopical Society* 53(3): 1–3.

Rocha S, Casal G, Al-uiraishy S, Azevado C (2016). Morphological and molecular characterization of a new myxozoan species (Myxosporea) infecting the gall bladder of *Raja clavata* (Chondrichthyes), from the Portuguese Atlantic coast. *The Journal of Parasitology* 99(2): 307– 317.

Rodger HD (2007). Erythrocytic inclusion body virus in Atlantic salmon *Salmo salar* L. *Journal of Fish Diseases* 30(7): 411–418.

Rodrigues MV, de Pérez ACA, Machado TM, Orisaka FM, Kurissio JK, Lafisca A (2015). Research of *Ascocotyle* (*Phagicola*) *longa* in heat treated fillets of mullet (*Mugil platanus*). *Fisheries and Aquaculture Journal* 6: 115 [Online] Available from: http://www.doi:10.4172/2150-3508.10000115 [Accessed 27 July 2018].

Roubal FR, Whittington ID (1990). Observations on the attachment by the monogenean, *Anoplodiscus australis*, to the caudal fin of *Acanthopagrus australis*. *International Journal for Parasitology* 20: 307–314. [Online] Available from: http://www.doi:10.1016/0020-7519(90)90144-C [Accessed 17 January 2018].

Rozman J, Njari B, Kozacinski L (2016). Occurrence of *Listeria monocytogenes* in fish and fishery products. *Meso* 2 (18): 72–76.

Samerpitak K, van der Linde E, Choi JH, van den Ende GB, Machouart M, Gueidan C, Hoog S (2013). Taxonomy of *Ochroconis*, genus including opportunistic pathogens on humans and animals. *Fungal Diversity* 65 [Online] Available from: http://www.doi.10.1007/s13225-013-0253-6 [Accessed 3 April 2018].

Sánchez-García N, Raga J, Francisco EM (2014). Risk assessment for parasites in cultures of *Diplodus puntazzo* (Sparidae) in the Western Mediterranean: Prospects of cross infection with *Sparus aurata*. *Veterinary Parasitology* 204(3-4) [Online] Available from: http://www.doi.10.1016/j.vetpar.2014.05.013. [Accessed 23 February 2018].

Sandoval C, Infante J, Abad J, Ferguson HW, Paredes E, Valdebenito S, Yanez AJ, Ilardi P, Avendano-Herrera R (2016). Case Report: Strawberry disease in farmed Chilean rainbow trout. *Journal of Aquatic Animal Health* 28(1): 1–10 [Online] Available from: https://www.doi.org/10.1080/08997659.2015.1114534 [Accessed 11 January 2018].

Santos CP, DaSilva MT, Moravek F (2017). *Dawestrema cycloancistrium* from the head pores of *Arapaimas. Diseases of Aquatic Organisms* 125(2): 93–100.

Sano T, Sano M, Fukuda H (1988). Characterization of newly isolated herpesvirus from eel *In:* Abstract of Third International Colloquium on Pathology in Marine Aquaculture. Gloucester Point, Virginia, 1.p.

Sekar VT, Santiago TC, Vijayan KK, Alvavandi SV, Raj VS, Rajan JJ, Sanjuthka M, Kalaimani N (2008). Involvement of *Enterobacter cloacae* in the mortality of the fish, *Mugil cephalus. Letters in Applied Microbiology* 46(6): 667–672 [Online] Available from http://www. doi: 10.1111/j.1472-765X.2008.02365.x. Epub 2008 Apr 12. [Accessed 25 June 2018].

Shalaby SI (1993). Communicable parasites from Nile fishes. I- Pathological and parasitological studies on *Aliptrema tilapi* n.sp. *Egyptian Journal of Comparative Pathology and Clinical Pathology* 6: 199–205.

Shalaby SI, Esposito P, Riegler G, Di- Carlo V, Carratu R (1993). Trematode parasites transmitted to man and fish-eating mammals through *Tilapia nilotica*. II-New trematode species. *Acta Mediterranea Di Patologia Infective Tropicale* 12: 111–114.

Shalaby SI, Easa M, El S, Afify MH (1996). Lake Quarun fishes as intermediate hosts for transmitting some trematodes. *Egyptian Journal of Comparative Pathology and Clinical Pathology* 9: 201– 213.

Shamsi S, Suthar J (2016). Occurrence of *Terranova* larval types (Nematoda: Anisakidae) in Australian marine fish with comments on their specific identities [Online] Available from https://doi.org/10.7717/peerj.1722 [Accessed 26 June 2018].

Shamsi S, Steller E, Chen Y (2018). New and known zoonotic nematode larvae within selected fish species from Queensland waters in Australia. *International Journal of Food Microbiology* 272: 73–82.

Shaperclaus W (1992). Fish diseases, Volume 1, CRC Press, Rotterdam, The Netherlands, 748 p.

Scholz T (1997). Life-cycle of *Bothriocephalus claviceps*, a specific parasite of eels. *Journal of Helminthology* 71(3): 241–248.

Scholz T, Kuchta R, Bray RA, Repova R (2004). Larvae of gryporhynchid cestodes (Cyclophyllidea) from fish: A review. *Folia Parasitologica* 51: 131–152.

Scholz T, Garcia HH, Kuchta R, Wicht B (2009). Update on the human broad tapeworm (genus *Diphyllobothrium*), including clinical relevance. *Clinical Microbiology Reviews* 22(1): 146–160.

Scholz T, Kutcha R (2016). Fish-borne, zoonotic cestodes (*Diphyllobothrium* and relatives) in cold climates: A never-ending story of neglected and (re)-emergent parasites. *Food and Waterbourne Parasitology* 4: 23–38.

Seyedmousavi S, Badali H, Chlebicki A, Zhao J, Prenafeta-Boldú FX, de Hoog GS (2011). *Exophiala sideris*, a novel black yeast isolated from environments polluted with toxic alkyl benzenes and arsenic. *Fungal Biology* 115:1030–1037.

Shen JW (1990). Digenetic trematodes of marine fishes from Hainan Island. Science Publications, Beijing, China, 228 p (In Chinese, English abstract).

Shi M, Lin XD, Vasilakis N, Tian JH, Li CX, Chen LJ, Eastwood G, Diao XN, Chen MH, Chen X, Qin XC, Widen SG, Wood TG, Tesh RBm Xu J, Holmes EC, Zhang YZ (2015). Divergent viruses discovered in arthropods and vertebrates revise the evolutionary history of the Flaviviridae and related viruses. *Journal of Virology* 90(2): 659–669.

Short RB (1954). A new blood fluke, *Selachohemecus olsoni*, n. g., n. sp. (Aporocotylidae) from the sharp-nosed shark, *Scoliodon terranovae. Proceedings of the Helminthological Society of Washington* 21:78–82.

Sim-Smith C, Faire S, Lees A (2014). Managing biosecurity risk for business benefit. Report prepared for the Ministry for Primary Industries by Coast and Catchment Ltd, Auckland, New Zealand, 214 p.

Sindermann CJ (1990). Principal diseases of marine fish and shellfish: Diseases of marine fish. Gulf Professional Publishing, New York. 521 p.

Sinton LW, Donnison AM (1994). Characterisation of faecal streptococci from some New Zealand effluents and receiving waters. *New Zealand Journal of Marine and Freshwater Research* 28(2): 145–158.

Sitjà-Bobadilla A, Alvarez-Pellitero P (2009). Experimental transmission of *Sparicotyle chrysophrii* (Monogenea: Polyopisthocotylea) to gilthead seabream (*Sparus aurata*) and histopathology of the infection. *Folia Parasitologica* 56(2): 143–151.

Sitko J, Sherrand-Smith E, Stanton DWC, Komorova P, Heneberg P (2016). Integrative taxonomy of European parasitic flatworms of the genus *Metorchis* Looss, 1899 (Trematoda: Opisthorchiidae). *Parasitology International* 65: 258–267.

Skall HF, Olesen NJ, Mellergaard S (2005). Viral haemorrhagic septicaemia virus in marine fish and its implications for fish farming – A review. *Journal of Fish Diseases* 28(9): 509–529.

Smith PJ, Diggles B, Kim S (2007). Evaluation of parasite markers to access swordfish stock structure. *Scientific Committee, Western and Central Pacific Fisheries Commission Paper* WCPFC-SC3-B1 SWG/1P – 1, 13 p.

Smyth D, Deverell E, Balm M, Nesdale A, Rosmergy I (2015). A case of botulism in New Zealand. *New Zealand Medical Journal* 128 (1425): 97–100.

So BKF (1972). Marine fish haematozoa from Newfoundland waters. *Canadian Journal of Zoology* 50: 543–554.

Sobel J (2005). Botulism. *Clinical and Infectious Diseases* 41:1167–1173.

Sommerville C (1982). The life history of *Haplorchis pumilio* (Looss, 1896) from cultured tilapias. *Journal of Fish Diseases* 5: 233–241 [Online] Available from: http://www.doi,10.1111/j.1365-2761.1982.tb00478.x [Accessed 22 February 2018].

Song JY, Kitamura S, Oh MJ, Kang HS, Lee JH, Tanaka SJ, Jung SJ (2009). Pathogenicity of *Miamiensis avidus* (syn. *Philasterides dicentrarchi*), *Pseudocohnilembus persalinus, Pseudocohnilembus hargisi* and *Uronema marinum* (Ciliophora, Scuticociliatida). *Diseases of Aquatic Organisms* 83(2):133–143 [online] Available from: http://www.doi: 10.3354/dao02017 [Accessed 6 October 2017].

Speare DJ, Brocklebank J, MacNair N (1992). Granulomatous nephritis associated with *Rhodococcus* sp. infection in Atlantic salmon (*Salmo salar*). Cross-Canada Disease Report: Eastern Canada. *Canadian Veterinary Journal* 33(3): 192.

Spikmans F, van Tongeren T, Van Alen TA, Van de Velde G, den Camp HJ (2013). High prevalence of the parasite *Sphaerothecum destruens* in the invasive topmouth gudgeon *Pseudorasbora parva* in the Netherlands, a potential threat to native freshwater fish. *Aquatic Invasions* 8: 1–6.

Stamper MA, Lewbart GA, Barrington PR, Harms CA, Geoly F, Stoskopf MK (1998). *Eimeria southwelli* infection associated with high mortality of cownose rays. *Journal of Aquatic Animal Health* 10: 264–270.

Staples P (2000). *Aeromonas, Plesiomonas* and *Vibrio* bacteria isolated from animals in New Zealand. *Surveillance* 27(1): 3–4.

Starliper CE, Schill WB (2011). Flavobacterial diseases: Columnaris disease, coldwater disease and bacterial gill disease. pp 606–63, *In:* Woo PTK, Bruno DW (*Eds.*). Fish Diseases and Disorders: Volume 3 - Viral, Bacterial and Fungal Infections, 2nd edition. CABI International, Wallingford, United Kingdom, 930 p.

Statistics New Zealand (2017). Import and export trade statistics, Statistics New Zealand, Wellington, New Zealand [Online] Available from: http://www.archive.stats.govt.nz/browse_for_stats/industry_sectors/imports_and_exports.aspx [Accessed 16 August 2018].

Stevenson A, Waite M (2011). Concise Oxford English Dictionary, Luxury edition. Oxford University Press, Oxford, United Kingdom, 1682 p.

Steyr C, Whittaker DJ (1986). Host-parasite interaction of the myxosporeans *Kudoa paniformis* Kabata & Whitaker, 1981 and *Kudoa thyrsites* (Gilchrist, 1924) in the muscle of Pacific whiting, *Merluccius productus* (Ayres): an ultrastructural study. *Journal of Fish Diseases* 9(6): 505–517.

Stidworthy MF, Thornton SM, James R (2017). A review of pathologic findings in elasmobranchs: a retrospective case series, pp 277–288, *In:* Warmolts MD, Thoney D, Hueter R, Murray M, Ezcurra J (*Eds.*). The Elasmobranch

Husbandry Manual II: Recent Advances in the Care of Sharks, Rays and their Relatives. Ohio Biological Survey, Inc. Columbus, Ohio, United States of America. 481 p.

Stone, MAB, MacDiarmid SC, Pharo HJ (1997). Import health risk analysis: salmonids for human consumption. Ministry of Agriculture Regulatory Authority, New Zealand, 269 p.

Stride MC, Nowak BF (2014). Epitheliocystis in three wild fish species in Tasmanian waters. *Journal of Fish Diseases* 37: 157–162.

Tanzolla RD, Sardella NH (2006). *Terranova galeocerdonis* (Thwaite, 1927) (Nematoda: Anisakidae) from *Carcharias taurus* (Chondrichthyes: Odontaspididae) off Argentina, with comments on some related species. *Systematic Parasitology* 64: 27–36.

Tassal (2014). Environmental impact assessment to accompany draft 4 to D'Entrecasteaux channel MFPD 2002: A request to amend zone 8B. Report prepared by Tassal Ltd, Tasmania, 187 p.

Threader R, Blimke L, Groman D (2010). Taking stock in stock. Stocking of American eel (*Anguilla rostrata*) in the Upper St Lawrence River and Lake Ontario, 2006–2009. Atlantic Veterinary College, Prince Edward Island. Report OSPG – 07015.01.EEL, to Ontario Power Generation, 62 p.

Tingbao Y, Kritsky DC, Yuan S, Jianying Z, Suha S, Agrawi N (2006). Diplectanids infesting the gills of the barramundi *Lates calcarifer* (Bloch) (Perciformes: Centropomidae), with the proposal of *Laticola* n. g. (Monogenoidea: Diplectanidae). *Systematic Parasitology* 63(2): 125 139.

Toranzo AE, Cutrin JM, Roberson BS, Nunez S, Abell JM, Hetrick FM, Baya AM (1994). Comparison of the taxonomy, serology, drug resistance transfer and virulence of *Citrobacter freundii* strains from mammals and poikilotherm hosts. *Applied and Environmental Microbiology* 60(6): 1789–1797.

Tubbs L, Lee P, Diggles B, Jones JB, Sheppard M, Sim-Smith C (2007). A review of aquatic diseases of significance to New Zealand. Final research report for project ZBS 2005-17. National Institute of Water and Atmospheric Research, Wellington, New Zealand, 452 p. (unpublished document commissioned by MPI).

Umeda N, Nibe H, Hara T, Hirazawa N (2006). Effects of various treatments on hatching of eggs and viability of oncomiracidia of the monogeneans *Pseudodactylogyrus anguillae* and *Pseudodactylogyrus bini. Aquaculture* 253(1–4): 148–153.

Upton SJ, Gardner SL, Duszynski DW (1988). The round stingray, *Urolophus halleri* (Rajiformes: Dasyatidae), as a host for *Eimeria chollaensis* sp. nov. (Apicomplexa: Eimeriidae). *Canadian Journal of Zoology* 66: 2049–2052.

Upton A, Taylor S (2002). *Vibrio vulnificus* necrotising fasciitis and septicaemia. *New Zealand Medical Journal* 115: 108–109.

Valladao GMR, Levy-Pereira N, Viadanna PHO, Gallani SU, Farias THV, Pilarski F (2014). Haematology and histopathology of Nile tilapia parasitized by *Epistylus* sp., an emerging pathogen in South America. Bulletin of the European Association of Fish Pathologists 35(1): 14–20.

Valtonen FT, Rintamaki P, Lappalainen M (1989). *Triaenophorus nodulosus* and *T. crassus* in fish from northern Finland. *Folia Parasitica* 36(4): 351–370.

Van Beurden SJ, Engelsma MY, Roozenberg I, Voorbergen-Laarman MA, van Tulden PW, Kerkhoff S, van Nieuwstadt AP, Davidse A. Haenen OLM (2012). Viral diseases of wild and farmed European eel *Anguilla anguilla* with particular reference to the Netherlands. *Diseases of Aquatic Organisms* 101: 69–86.

Van Ginnekin V, Haenen O, Coldenhoff K, van den Thillart G (2004). Presence of eel viruses in eel species from various geographic regions. *Bulletin of the European Association of Fish Pathologists* 24(5): 268–272.

Van Nieuwstadt AP, Dijkstra SG, Haenen OLM (2001). Persistence of herpesvirus of eel *Herpesvirus anguillae* in farmed European eel *Anguilla anguilla. Diseases of Aquatic Organisms* 45: 103–107.

Venable DL, Gaudé AP, Klerks PL (2000). Control of the trematode *Bolbophorus confusus* in channel catfish *Ameiurus punctatus* in ponds using salinity manipulation and polyculture with black carp *Mylopharyngodon piceus*. *Journal of the World Aquaculture Society* 31: 158-166.

Vo, DV, Murrell D, Dalsgaard A, Bristow G, Nguyen D, Bui T, (2008). Prevalence of zoonotic metacercariae in two species of grouper, *Epinephelus coioides* and *Epinephelus bleekeri*, and flathead mullet, *Mugil cephalus*, in Vietnam.

The Korean Journal of Parasitology 46: 77–82. [Online] Available from: http://www.doi.10.3347/kjp.2008.46.2.77. [Accessed 10 July 2018].

Waeschenbach A, Brabec J, Scholz T, Littlewood DTJ, Kuchta R. (2017). The catholic taste of broad tapeworms – multiple routes to human infection. *International Journal for Parasitology*. 47(13): 831–843 [Online] Available from: https://www.doi.org/10.1016/j.ijpara.2017.06.004 [Accessed 10 April 2018].

Wahl E (1971). *Protomicrocotyle mirabilis* (Mac Callum 1918) Johnston et Tiegs 1922 et *P. ivoriensis* n. spec., Monogènes parasites de *Caranx hippos* dans la lagune Ebrié (Côte d'Ivoire). *Zeitschrift für Parasitenkunde* 38(4): 319 332 {online] Available from https://doi.org/10.1007/BF00329279 [Accessed 22 February 2018].

Wall JM, Wood SA, Orlovich DA, Rhodes L, Summerfield TC (2014). Characterisation of freshwater and marine cyanobacteria in the Hokianga region, Northland, New Zealand. *New Zealand Journal of Marine and Freshwater Research* 48(2): 177-193.

Wang PQ (1982). Some digenetic trematodes of marine fishes from Fujian Province, China. *Oceanologia et Limnologia Sinica* 13(2): 179–194.

Waterman PB, Sin FYT (11). occurrence of the marine tapeworms, *Hepatoxylon trichiuri* and *Hepatoxylon megacephalum*, in fishes from Kaikoura, New Zealand. *New Zealand Natural Sciences* 18: 71–73.

Webber W, Fenwick G, Bradford-Grieve J, Eagar S, Buckeridge J, Poore G, Dawson E, Watling L, Jones J, Wells J, Bruce N, Ahyong S, Larsen K, Chapman M, Olesen J, Ho J, Green J, Shiel R, Rocha C, Lorz A, Bird G, Charleston W (2010). Phylum Arthropoda subphylum Crustacea: shrimps, crabs, lobsters, barnacles, slaters, and kin, pp 98-232 *In:* Gordon DP (*Ed.*) *New Zealand Inventory of Biodiversity:* Volume Two: Kingdom Animalia - Chaetognatha, Ecdysozoa, Ichnofossils, Canterbury University Press, New Zealand [Online]. Available from: https://www.researchbank.rmit.edu.au/view/rmit:16047 [Accessed 1 March 2017].

Weeks PJ, Penlington B (1986). First records of *Ligula intestinalis* (Cestoda) in rainbow trout *Salmo gairdneri* and common bully *Gobiomorphus cotidianus* in New Zealand. *Journal of Fish Biology* 28: 183–190.

Wells WH, Randall BH (1955). New hosts of the genus *Heterophyes* in Egypt. *The Journal of Parasitology* 42(3): 287–292.

Whittaker SJ (2013). Shellfish acquired *Vibrio cholera* cellulitis and sepsis from a vulnerable leg. *The New Zealand Medical Journal* 126: 1379.

Whittington RG, Jones JB, Hine PM, Hyatt AD (1997). Epizootic mortality in the pilchard *Sardinops sagax neopilchardus* in Australia and New Zealand in 1995, Part 1: Pathology and Epizootiology. *Diseases of Aquatic Organisms* 28: 1–16.

Whittington RJ, Becker JA, Dennis MM (2010). Iridoviruses in finfish. *Journal of Fish Diseases* 33: 95–122.

Williams CF, Lloyd D, Poynton SL, Jorgensen A, Millet COM, Cable J (2011). *Spironucleus* species: Economically important fish pathogens and enigmatic single-celled eukaryotes. *Journal of Aquaculture Research and Development* S2: 002, 13 p. [Online] Available from: http://www.doi: 10.4172/2155-9546.S2-002 [Accssed 22 August 2018].

Winton JR, Hershberger PK (2014). Viral Erythrocytic Necrosis, Chapter 2.2.7 *In:* The Blue Book 2014. Fish Health Section, American Fisheries Society [Online] Available from: http://www.afs-fhs.org/perch/resources/ 14069231372.2.7ven2014.pdf [Accessed 6 October 2017].

Wise DJ, Griffin MJ, Terhune JS, Pote LM, Khoo LH (2008). Induction and evaluation of proliferative gill disease in channel catfish fingerlings. *Journal of Aquatic Animal Health* 20(4): 236–244.

Wolf JC, Smith SA (2000). Seafood safety. pp 697–716. *In*: Martin RE, Carter EP, Flick GJ, Davis LM (*Eds.*) *Marine and Freshwater Products Handbook*. CRC Press, 963 p.

Wong JSJ, Schousboe MI, Metcalf SSL, Endre ZH, Hegarty JM, Maze MJ, Keith ER, Seaward LM, Podmore RG (2010). *Ochroconis gallopava* peritonitis in a cardiac transplant patient on continuous ambulatory peritoneal dialysis. *Transplant Infectious Disease* 12: 455–458.

Woo PTK, Bruno DW (2014). Diseases and disorders of finfish in cage culture. Second edition. CABI Publishers, Wallingford, United Kingdom, 354 p.

Wood (1995) Wood CM, Pärt P, Wright PA (1995). Ammonia and urea metabolism in relation to gill function and acid-base balance in a marine elasmobranch, the spiny dogfish (*Squalus acanthias*). *Journal of Experimental Biology* 198: 1545–1558.

WoRMS (2018). World register of marine species, Flanders Marine Institute, Belgium [Online] Available from: http://www.marinespecies.org at VLIZ [Accessed 26 March 2018].

WTO (World Trade Organisation) (2017). Agreement on trade facilitation. World Trade Organisation, Geneva, Switzerland [Online] Available from: https://www.wto.org/english/docs_e/legal_e/tfa-nov14_e.htm [Accessed 16 August 2018].

Xu K, Song W, Warren A (2002). Taxonomy of trichodinids from the gills of marine fishes in coastal regions of the Yellow Sea, with descriptions of two new species of *Trichodina* Ehrenberg, 1830 (Protozoa: Ciliophora: Peritrichia). *Systematic Parasitology* 51: 107–120.

Yamaguti S (1963). Systema Helminthum, Volume IV. Monogenea and Aspidocotylea, pp 699–699. Wiley Interscience, London, United Kingdom, 699 p.

Yan X, Wang Y, Xiong LF, Jian LC, Wu Zh (2014). Phylogenetic analysis of newly isolated grass carp reovirus. Springer Plus 3: 190–200. Springer Biomedical and Life Sciences.

Yano K, Musick JA (2000). The effect of the mesoparasitic barnacle *Anelasma* on the development of reproductive organs of deep-sea squaloid sharks, *Centroscyllium* and *Etmopterus. Environmental Biology of Fishes* 59(3): 329–339.

Yanong RPE (2011). Nematode (roundworm) infections in fish. Circular 91, Fisheries and Aquatic Services Department, UF/IFAS Extension. Institute of Food and Aquatic Sciences, Gainesville, Florida, United States of America, 8 pp [Online] Available from: https://edis.ifas.ufl.edu/fa091 [Accessed 12 February 2015].

Yera H, Kuchta R, Brabec J, Peyron F, Dupouy-Camet J (2013). First identification of eggs of the Asian fish tapeworm *Bothriocephalus acheilognathi* (Cestoda: Bothriocephalidea) in human stool. *Parasitology international*. 62. [Online] Available from: http://www.doi.10.1016/j.parint.2013.02.001 [Accessed 23 July 2018].

Yokoyama H, Liyanage YS, Sugai A, Wakabayashi H (1998). Hemorrhagic thelohanellosis of color carp caused by *Thelohanellus hovorkai* (Myxozoa:Myxosporea). *Fish Pathology* 33: 85-89.

Yoshimizu M (2012). *Oncorhynchus masou* virus disease. Chapter 2.2.13 *In:* Fish diseases. American Fisheries Society [Online] Available from: http://www.afs-fhs.org/perch/resources/14069233132.2.13omvdisease2014.pdf [Accessed 22 March 2017].

Youn H (2009). Review of zoonotic parasites in medical and veterinary fields in the Republic of Korea. *Korean Journal of Parasitology* 47(Suppl): 133–141 [Online] Available from http://www. Doi.10.3347/kjp.2009.47.S.S133 [Accessed 26 June 2018].

Yokoyama H, Liyanage YS, Sugai A, Wakabayashi H (1998). Hemorrhagic thelohanellosis of color carp caused by *Thelohanellus hovorkai* (Myxozoa:Myxosporea). *Fish Pathology* 33: 85–89.

Yokoyama H, Fukuda Y (2001). *Ceratomyxa seriolae* n. sp. and *C. buri* n. sp. (Myxozoa: Myxosporea) from the gall-bladder of cultured yellowtail *Seriola quinqueradiata. Systematic Parasitology* 48(2): 125–130.

Young ND, Dykova I, Snekvik K, Morrison RN (2008). *Neoparamoeba perurans* is a cosmopolitan aetiological agent of amoebic gill disease. *Diseases of Aquatic Organisms* 78(3): 217–223.

Youngren-Grimes B (1990). Ecology of bacteria in shark tissue, pp 55–63 *In:* Pratt HL. Gruber SH, Taniuchi T (*Eds*.). Elasmobranchs as living resources: Advances in the biology, ecology, systematics, and the status of the fisheries. Proceedings of the Second United States-Japan Workshop East- West Center, Honolulu, Hawaii, 9-14 December 1987. *NOAA Technical Report* NMFS 90. National Oceanic and Atmospheric Administration, United States Department of Commerce, Washington D.C., United States of America, 528 p.

Zimmermann G (2010). The entomopathogenic fungi *Isaria farinosa (*formerly *Paecilomyces farinosus*) and the *Isaria fumosorosea* species complex (formerly *Paecilomyces fumosoroseus*): biology, ecology and use in biological control. *Biocontrol Science and Technology* 18(9): 865– 901.

10. Epizootic haematopoietic necrosis virus, European catfish virus and European sheatfish virus

10.1. Hazard identification

10.1.1. Aetiological agent

Epizootic haematopoietic necrosis virus (EHNV) is a double-stranded DNA non-enveloped virus, classified in the Genus *Ranavirus*, within the Family Iridoviridae (OIE 2016a). However, this classification is under review (Bondad-Reantaso *et al.* 2012). While members of the *Ranavirus* genus primarily infect amphibians, the fish pathogens European sheatfish iridovirus (ESV), which affects the sheatfish (*Silurus glanis*), and the European catfish iridovirus (ECV), which affects catfish (*Ameiurus melas*), as well as epizootic haematopoietic necrosis virus (EHNV), are also included in this genus. EHNV was originally described in 1985 from European perch (*Perca fluviatilis*) and rainbow trout (*Oncorhynchus mykiss*) (Whittington *et al.* 2010).

10.1.2. OIE status

Epizootic haematopoietic necrosis is listed by the OIE (OIE 2016a).

10.1.3. New Zealand status

EHNV, ESV and ECV are considered exotic (MAF 1999). They have not been reported from New Zealand but EHNV is not a notifiable organism in New Zealand (Anon. 2016).

10.1.4. Epidemiology

Until EHNV was identified in Australian salmonid aquaculture, iridoviruses were not associated with economically significant disease in fish (Whittington *et al*. 2010). EHNV has only been reported from New South Wales and South Australia where it naturally infects wild and farmed stocks of European perch and rainbow trout. It has a poor rate of infectivity, but has a high fatality rate (OIE 2016b). During outbreaks, it may be detected in 60 to 80% of moribund fish, but at low rates (0 to 4%) in clinically healthy fish. Between outbreaks, it remains dormant and undetectable in its host populations (OIE 2016b). Differences in susceptibility occur between the originally introduced and latterly introduced stocks of host species (Whittington *et al.* 1996). Infectivity rates are also influenced by factors such as temperature, water quality and fish husbandry.

The host range of EHNV is listed in Table 7, based on bath inoculation data (Bucke *et al.* 1979; Langdon 1989; Whittington *et al.* 2010; Sano *et al.* 2011; Becker *et al.* 2013; OIE 2016a, 2016b).

The patterns of infection remain unknown (OIE 2016b). Disease transmission is horizontal, by direct contact, through transport water, or by the presence of sub-clinically infected fish (OIE 2016b). The viral particles are shed into the water from infected tissue or following death and decomposition of the carcass (Reddacliff & Whittington 1996). Sequential infection of farmed stocks from adjacent wild fish has been proposed to explain some re-infection patterns, but the presence of disease in distinct catchments and patterns of upstream disease progression in Australian catchments indicates EHNV is likely to be spread by factors other than simple water catchment flow (OIE 2016b).

Table 7. Families and Species of Fish Susceptible to Epizootic Haematopoietic Necrosis Virus (EHNV) and Associated Iridoviruses

Epizootic Haematopoietic Necrosis Virus (EHNV)	
Family	**Host Species**
Esocidae	Pike (*Esox lucius*)
Galaxiidae	Mountain galaxiid (*Galaxias olidus*)
Ictaluridae	Black bullhead (*Ameirus melas*)
Melotaenidiidae	Australian rainbowfish (*Melotaenia fluviatilis*)
Percidae	European perch (*Perca fluviatilis*)
Percyithidae	Macquarie perch (*Macquaria australasica*), Murray cod (*Maccullochella peelii)*
Plotosidae	Freshwater catfish (*Tandanus tandanus*)
Salmonidae	Atlantic salmon (*Salmo salar*), rainbow trout (*Oncorhynchus mykiss*)
Terapontidae	Silver perch (*Bidyanus bidyanus*)
European Catfish Virus (ECV)	
Ictaluridae	Black bullhead (*Ameiurus melas*), brown bullhead (*Ameiurus nebulosus*)
European Sheatfish Virus (ESV)	
Siluridae	European sheatfish (*Silurus glanis*)

EHNV is extremely resistant to drying, and may be carried on feathers, bill and feet of scavenging birds. It remains viable in the avian digestive tract and may be shed in the faeces of piscivorous birds, such as *Chroicocephalus novaehollandiae* (silver gull), for 3–9 days after feeding (Whittington *et al.* 1996; Spickler *et al.* 2010).

Dried viral material is resistant to heat, remaining viable after heating to 60°C for up to 15 minutes (Whittington *et al*. 2010). It remains viable in mucous and frozen fish tissues for up to 2 years (Langdon 1989; Whittington *et al*. 1996) and persists for years in water and sediments (OIE 2016b).

Upon entry, EHNV invades hepatic, haematopoietic and endothelial cells including the spleen, kidney, liver, heart and gill tissues (Reddacliff & Whittington 1996; Becker *et al.* 2013). Fish infected with EHNV may show no apparent external clinical signs (CFIA 2014; OIE 2016a).

The two related viruses ECV and ESV are considered causative agents of EHN (Bondad-Reantaso *et al*. 2012) and are sufficiently similar to be essentially considered to be one species complex (Holopainen *et al.* 2009; Bondad-Reantaso *et al.* 2012).

ECV was first isolated in France from the black bullhead (*Ameiurus melas*) (Pozet *et al.* 1992) and is a significant disease of juvenile fish. Infections have been experimentally induced in brown bullhead (*Ameiurus nebulosus*) (Sano *et al.* 2011). Clinical signs include oedema, with petechiae around the pectoral girdle and pale gills, with high mortality. Infection is restricted to spleen and kidney tissues (Plumb & Hanson 2011).

ESV was first isolated in Germany from European sheatfish (*Silurus glanis*) (Ahne *et al.* 1989). It causes high mortality (up to 100%) in sheatfish fry in European aquaculture (Plumb & Hanson 2011). The focus of infection is the spleen and kidney, with hyperplasia and oedema of gill, skin, heart, eye, liver, pancreas, brain and digestive tract tissue (Plumb & Hanson 2011).

While adult and juvenile European sheatfish can be experimentally infected with ESV and ECV, older sheatfish were refractory to ECV, indicating fish age to be an important factor in determining rates of infection (Leimbach *et al.* 2014). No vaccines are available (OIE 2016b).

Susceptible species in New Zealand to the EHNV/ESV/ECV species complex include Atlantic salmon (*Salmo salar*), Chinook salmon and rainbow trout (*Oncorhynchus* spp.) and native

Galaxias sp. (Stone *et al.* 1997; Tubbs *et al.* 2007), as well as the introduced brown bullhead (*Ameiurus nebulosus*) (Johnston 2008) and European perch (*Perca fluviatilis*) (Closs *et al.* 2005).

10.2. Risk assessment

10.2.1. Entry assessment

Clinically diseased fish with no external clinical signs of infection (Plumb & Hanson 2011) are unlikely to be detected by visual inspection. EHNV, ECV and ESV are commonly associated with brain, eye and gill tissues of the head, which are retained on evisceration. These viruses are extremely resistant to drying and freezing (Spikler *et al.* 2010) and are likely to remain viable in the commodity even after extended frozen storage (Whittington *et al*. 1996). The likelihood of entry is assessed as non-negligible.

10.2.2. Exposure assessment

To establish infection through the commodity, infected eviscerated product would have to become available for consumption by a susceptible freshwater fish host, in sufficient quantity and duration (Kahn *et al.* 1999). EHNV, ECV and ESV may remain viable in the offal discarded after commercial processing (Plumb & Hanson 2011). They are extremely resistant to freezing and drying (Langdon 1989, Whittington e*t al*. 1996, 2010) and can remain viable in dried organic material (OIE 2016b).

EHNV, ECV and ESV remain viable in the water column and can be distributed horizontally, through discarded offal. Further distribution may occur between aquatic waterways by scavenging birds, either directly (on feathers, bills and feet), or indirectly (by passage through the digestive system). Transfer may also occur through contaminated transfer and processing machinery, or poor hygiene. Transfer may occur through disposal of organic material into waterways, or through the inadequate treatment of wash-down water after processing (Whittington *et al.* 1996; Spikler *et al.* 2010). These viruses may remain viable for an extended period (months to years) in water or sediment. They are extremely hardy in the environment and multiple potential exposure vectors and pathways exist. The likelihood of exposure to EHNV, ECV and ESV through the commodity is assessed as non-negligible.

10.2.3. Consequence assessment

The establishment of EHNV/ESV/ECV in New Zealand may have a direct economic effect upon the hatchery stages of the Chinook salmon (*O. tshawytscha*) aquaculture industry, which was worth $63.4 million in 2011 (Aquaculture New Zealand 2014). It may indirectly affect the farming of salmonids for recreational fishing, worth in excess of $80 million (Fish & Game 2014), as well as the tourism industries associated with salmon and trout fishing. It may also affect New Zealand native galaxiid fish species, most of which are classified as endangered (Allibone 2010). It may also affect the introduced brown bullhead (*Ameiurus nebulosus*) which is established in New Zealand. The consequence of establishment is assessed as non-negligible.

10.2.4. Risk estimation

Since the entry, exposure and consequence assessments for EHNV/ESV/ECV are non-negligible, the risk estimation is non-negligible. Therefore, these viruses are assessed to be a risk in the commodity and risk management measures may be justified.

10.3. Risk management

EHNV/ESV/ECV have been assessed to be a risk in the commodity. As infection with EHNV is a notifiable disease, the OIE *Aquatic Code* (OIE 2016a) provides specific guidance on importing eviscerated fish from infected countries and the specific processing requirements that would ensure the destruction of the virus.

Article 10.1.3 of the OIE *Aquatic Code* states:

Competent Authorities should not require any conditions related to EHN, regardless of the EHN status of the exporting country, zone or compartment, when authorising the importation or transit of the following aquatic animals and aquatic animal products from the species referred to in Article 10.1.2 which are intended for any purpose and which comply with Article 5.4.1:

a. *heat sterilised, hermetically sealed fish products (i.e. a heat treatment at 121°C for at least 3.6 minutes or any time/temperature equivalent);*
b. *pasteurised fish products that have been subjected to a heat treatment at 90°C for at least ten minutes (or any time/temperature equivalent which has been demonstrated to inactivate IHNV);*
c. *mechanically dried, eviscerated fish (i.e. a heat treatment at 100°C for at least 30 minutes or any time/temperature equivalent which has been demonstrated to inactivate IHNV)*

Article 10.1.12 of the OIE *Aquatic Code* states:

For importation of aquatic animals and aquatic animal products for retail trade for human consumption from a country, zone or compartment not declared free from epizootic haematopoietic necrosis: Competent Authorities should not require any conditions related to EHN, regardless of the EHN status of the exporting country, zone or compartment, when authorizing the importation or transit of fish fillets or steaks (chilled or frozen) which have been prepared and packaged for retail trade and which comply with Article 5.4.2.

Certain assumptions have been made in assessing the safety of the aquatic animal products mentioned above. Member Countries should refer to these assumptions at Article 5.4.2 and consider whether the assumptions apply to their conditions.

For these commodities Member Countries may wish to consider introducing internal measures to address the risks associated with the commodity being used for any purpose other than for human consumption.

Compliance with Articles 10.1.2 and 10.1.12 should eliminate EHNV from the commodity and is a viable risk management option. Acceptance of country/zone freedom from EHNV does not necessarily include freedom from related viruses ECV and ESV. Where declaration of country/zone freedom is approved under the MPI Country Approval Procedures[G], this option should substantially reduce the occurrence of the EHNV/ECV/ESV species complex. Acceptance of a declaration of country/zone freedom is a viable risk management option.

The EHNV/ECV/ESV species complex affects 8 fish families (Table 7), which may be present in the commodity. Species declaration would substantially reduce the occurrence of EHNV/ECV/ESV from the commodity and be a viable risk management option. These viruses are reported from wild and farmed fish (OIE 2016a). Restriction of the commodity to wild-caught fish (not from aquaculture) would have little, or no effect on pathogen load of EHNV/ESV/ECV and is not a viable risk management option.

EHNV/ECV/ESV may be present in the brain and gill tissues of infected fish, so pathogen load is likely to be slightly reduced by removal of the gills and moderately reduced by removal of the head and gills. These viruses are moderately heat labile (Whittington *et al.* 1996), so the OIE-recommended heat treatments and processing guidelines would eliminate these pathogens from the commodity. EHNV/ECV/ESV are resistant to cold temperatures so frozen storage is likely to have no effect on pathogen load.

EHNV/ECV/ESV may be transmitted through the waste products associated with transport, storage and processing of the commodity. The requirement that all wash and wastewater discharges be appropriately chemically treated (e.g., with iodophors) before discharge, and that all solid wastes, tissue scraps and offal be disposed of through a recognised trade waste disposal procedure would be viable management options.

10.3.1. Risk management options

The EHNV/ECV/ESV viruses are reported from fish in families Esocidae, Galaxiidae, Ictaluridae, Percicthyidae, Percidae, Plotosidae, Salmonidae and Terapontidae (Table 7), which are considered likely to be present in the commodity. Other families have not been associated with the EHNV/ECV/ESV species complex. Therefore species declaration indicating the commodity is not originated from any of the above families should substantially reduce the occurrence of EHV/ECV/ESV in the commodity.

For the commodities originated from families associated with EHNV/ECV/ESV, one or a combination of the following additional options could also be considered to effectively manage the risk.

[G] Country freedom may be accepted by MPI for this, and other OIE-listed and non-listed diseases through the MPI Country Approval Procedures (Available from http://www.mpi.govt.nz/news-and-resources/publications/).

Where country/zone freedom from EHNV/ECV/ESV is accepted by MPI:

Option 1

Acceptance of country/zone freedom should substantially reduce pathogen occurrence, so the commodity may be imported without any further restrictions.

Where country/zone freedom from EHNV/ECV/ESV is not accepted by MPI or not available:

Option 2

Processing consistent with the conditions of Article 10.1.3 or 10.1.12 of the OIE *Aquatic Code* (OIE 2016a) should eliminate these pathogens. When these provisions are met, the commodity could be imported without any further restrictions.

Where the imported commodity is not consistent with Article 10.1.3 or 10.1.12 of the OIE *Aquatic Code* (OIE 2016a*)* and further processing is necessary:

Option 3

Heat treatment (by cooking for at least 15 minutes at a temperature exceeding 60°C) should eliminate EHNV/ECV/ESV. When this provision is met, the commodity could be imported without any further restrictions; or,

Option 4

Further processing (by removal of the head and gills) should moderately reduce the occurrence of EHNV/ECV/ESV. When this provision is met, the commodity could be imported without any further restrictions.

10.4. References

ACT (2000). Fish stocking plan for the Australian Capital Territory 2001–2005. Environment ACT, ACT Government, Canberra, Australia.

Ahne W, Schlotfeldt HJ, Thomsen I (1989). Fish viruses: isolation of an icosahedral cytoplasmic deoxyribovirus from sheatfish (*Silurus glanis*). *Journal of Veterinary Medicine (Part B)* 36*:* 333–336.

Allibone R (2010). Conservation status of New Zealand's native fish. Golder Associates Newsletter 20 (Third and fourth quarters 2010) [Online]. Available from: http://www.golder.co.nz/en/ modules.php?name=Newslettersandop= viewarticleandsp_id= 151andpage_id=709andarticle_id=409Options.pdf [Accessed 12 March 2015].

Anonymous (2016). Biosecurity (Notifiable Organisms) Order 2016 SR 2016/73. New Zealand Government, Wellington, New Zealand, 13 p.

Aquaculture New Zealand (2014). Salmon farming: An overview [Online]. Available from: http://aquaculture.org.nz/industry/overview.pdf [Accessed 15 July 2014].

Becker JA, Tweedie A, Gilligan D, Asmus M, Whittington RJ (2013). Experimental infection of Australian freshwater fish with epizootic haematopoietic necrosis virus (EHNV). *Journal of Aquatic Animal Health* 25(1): 66–76.

Bondad-Reantaso MG, McGladdery SE, East I, Subasinghe R (2012). Asia diagnostic guide to aquatic animal diseases. FAO Fisheries Technical Paper 402, Supplement 2. FAO (Food and Agriculture Organisation/Network of Aquaculture Centres in Asia-Pacific, United Nations, Rome, 240 p.

Bucke D, Cawley GD, Craig JF, Pickering AD, Willoughby LG (1979). Further studies of an epizootic of perch, *Perca fluviatili*s L., of uncertain aetiology. *Journal of Fish Diseases* 2: 297–311.

Closs GP, Ludgate B, Goldsmith RJ (2005). Controlling European perch (*Perca fluviatilis*): lessons from an experimental removal. *Science and Technical Report* PF-05, Department of Conservation, Wellington, New Zealand. 108 p.

CFIA (2014). Canadian Food Inspection Agency. Fact sheet for reportable diseases: Epizootic haematopoietic necrosis virus, 3 p [Online] Available from: http://www.inspection.gc.ca/animals/aquatic-animals/offices/eng/1300462382369/1365216058692.html [Accessed 21 May 2014].

Fish and Game (2014). New Zealand Fish and Game Council [Online]. Available from: http://www.fishandgame.org.nz/ [Accessed 24 September 2014].

Holopainen R, Ohlemeyer S, Schutze H, Bergmann SM, Tapiovaara H (2009). Ranavirus phylogeny and differentiation based on capsid protein, DNA polymerase and neurofilament triplet H1-like protein genes. *Diseases of Aquatic Organisms* 85: 81–91.

Johnston C (2008). Import risk analysis: Frozen skinless and boneless fillet meat of *Pangasius* spp. fish from Vietnam for human consumption. MAF Biosecurity New Zealand, 60 p.

Langdon JS (1989). Experimental transmission and pathogenicity of epizootic haematopoietic necrosis virus (HNV) in redfin perch *Perca fluviatilis* L., and 11 other teleosts. *Journal of Fish Diseases* 12(4): 295–310.

Leimbach S, Schutze H, Bergmann SM (2014). Susceptibility of European sheatfish *Silurus glanis* to a panel of ranaviruses. *Journal of Applied Ichthyology* 30(1): 93–101.

MAF (Ministry of Agriculture and Forestry) (1999). Supplementary import risk analysis: Head-on, gill-in Australian salmonids for human consumption. MAF Regulatory Authority. Ministry of Agriculture and Forestry, Wellington, New Zealand, 24 p.

OIE (2016a). Aquatic Animal Health Code, 19th Edition. World Organization for Animal Health. Paris, France [Online] Available from: http://www.oie.int/international-standard-setting/aquatic-code/access-online/ [Accessed 15 March 2017].

OIE (2016b). Manual of diagnostic tests for aquatic animals. World Organization for Animal Health. Paris, France [Online] Available from: http://www.oie.int/international-standard-setting/aquatic-manual/access-online/ [Accessed 17 March 2017].

Plumb JA, Hanson LA. (2011). Health maintenance and principal microbial diseases of cultured fishes. Wiley, New York, United States of America, 400 p.

Pozet F, Morand M, Moussa A, Torhy C, De Kinkelin P (1992). Isolation and preliminary characterisation of pathogenic icosahedral deoxyribovirus from the catfish (*Ameiurus melas*). *Diseases of Aquatic Organisms* 14: 35–42.

Reddacliff LA, Whittington RJ (1996). Pathology of epizootic haematopoietic necrosis virus (EHNV) infection in rainbow trout (*Oncorhynchus mykiss* Walbaum) and redfin perch (*Perca fluviatilis* L.). *Journal of Comparative Pathology* 115(2): 103–115.

Sano M, Nakai T, Fijan N (2011). Viral diseases and agents of warm water fish. *pp.* 166–244. *In:* Woo PTK, Bruno DW (*Eds.*) Fish Diseases and Disorders: Volume 3: Viral, Bacterial and Fungal Infections, 2nd edition. CABI Publishers, Wallingford, UK. 930 p.

Spikler AR, Roth JA, Gaylon J, Lofstedt JL (2010). Emerging and exotic diseases of animals. Fourth Edition. Centre for Food Safety and Public Health, College of Veterinary Medicine, Iowa State University, 383 p.

Stone, MAB, MacDiarmid SC, Pharo HJ (1997). Import health risk analysis: salmonids for human consumption. Ministry of Agriculture Regulatory Authority, New Zealand, 269 p.

Tubbs L, Lee P, Diggles B, Jones JB, Sheppard M, Sim-Smith C (2007). A review of aquatic diseases of significance to New Zealand. Final research report for project ZBS 2005-17. National Institute of Water and Atmospheric Research, Wellington, New Zealand, 452 p.

Whittington RJ, Kearns C, Hyatt AD, Hengstberger S, Rutzou T (1996). Spread of epizootic haematopoietic necrosis virus (EHNV) in European perch (*Perca fluviatilis*) in Southern Australia. *Australian Veterinary Journal* 73: 112–114.

Whittington RJ, Becker JA, Dennis MM (2010). Iridovirus infections in finfish - critical review with emphasis on ranaviruses. *Journal of Fish Diseases* 33(2): 95–122.

11. European eel virus

11.1. Hazard identification

11.1.1. Aetiological agent

European eel virus (EVE) is the agent of gill necrosis of eels (Plumb & Hanson 2011; Van Beurden *et al*. 2012). It is a bi-segmented double stranded RNA virus classified as an aquabirnavirus within the Family Birnaviridae (Munro & Midtlyng 2011). The taxonomy of this group is unclear (Kusuda *et al.* 1993; Crane & Hyatt 2011). The birnaviruses infecting non-salmonids are collectively referred to as aquabirnaviruses, or IPNV-like viruses (Kusuda *et al.* 1993; Castric 1997; Crane & Hyatt 2011; Van Beurden *et al.* 2012). They affect invertebrates as well as marine and freshwater fish, often with low pathogenicity, but do not cross-infect salmonids (Noga 2010; Munro & Midtlyng 2011). The four identified serogroups (A–D) of aquabirnaviruses relate to geographical locations, rather than to host species (Crane & Hyatt 2011). Most fish aquabirnaviruses, including European eel virus are grouped within serogroup A. EVE is considered to group with the IPNV serotype Ab (Van Beurden *et al*. 2012).

11.1.2. OIE status

Infection with EVE is not an OIE listed disease (OIE 2016a).

11.1.3. New Zealand status

EVE has not been reported from New Zealand eels to date (Van Ginneken *et al.* 2004; Tubbs *et al.* 2007; Plumb & Hansen 2011). It is assumed exotic but is not a notifiable organism in New Zealand (Anon. 2016).

11.1.4. Epidemiology

Eels are farmed by on-growing stocks of captured wild glass eels. While eel migration is poorly understood, adults from several countries migrate to a central site in oceanic waters to spawn, providing a conduit for the transfer of viruses among wild fish stocks (Van Beurden *et al.* 2012). The spawning grounds for New Zealand eel species are currently unknown, but are thought to be located off Tonga or New Caledonia to spawn (NIWA 2017).

EVE was originally described in 1970 from European eels (*Anguilla anguilla*) farmed in Japan (Sano *et al.* 1981), where it causes gill necrosis disease in juveniles. It has subsequently spread to the Japanese eel (*Anguilla japonica*) populations in Japan and Taiwan, as well as in wild and farmed European eels in Europe (Denmark, France, Holland, Italy, Germany, Greece, Sweden) and the United Kingdom. It has also spread to the United States, where it infects the American eel *Anguilla rostrata* (Van Ginneken *et al*. 2004; Crane & Hyatt 2011; Van Beurden *et al.* 2012).

It is endemic in wild European eel populations at rates of 44–57%, generally with no external signs of infection (Sano *et al.* 1981; Bandin *et al.* 2014). It has been reported in farmed *A. anguilla* from Italy, with no clinical signs of disease (Van Ginneken *et al.* 2004).

Outbreaks may occur in farmed eels, with mortalities of up to 100% at high stocking densities (Haenen *et al*. 2001). Mortalities are rare in older fish and surviving fish become lifelong carriers of infection (Tubbs *et al*. 2007; Munro & Midtlyng 2011; Van Beurden *et al.* 2012). The

introduction of EVE into Japanese and Taiwanese aquaculture of the native *A. japonica* stocks resulted in mass mortalities of up to 50% (Haenen 2008; Plumb & Hansen 2011). In addition, it has been implicated as a major agent responsible for the decline in European eel populations (Van Ginneken *et al*. 2005). Families and host species susceptible to EVE are given in Table 8.

Table 8. Families and Species of Fish Susceptible to European Eel Virus (EVE)

European Eel Virus (EVE)	
Family	**Host Species**
Anguillidae	American eel (*Anguilla rostrata*), European eel (*A. anguilla*), Japanese eel (*A. japonica*)
Cichlidae	*Tilapia mossambica* (= *Oreochromis mossambicus*)
Salmonidae	Rainbow trout (*Oncorhynchus mykiss*)

EVE has also been isolated from *Tilapia mossambica* (= *Oreochromis mossambicus*) in Taiwanese aquaculture (Kou *et al.* 1982). Infection may be experimentally induced by bath immersion of rainbow trout fry (*Oncorhynchus mykiss*) with 82% mortality (Van Beurden *et al.* 2012).

External signs of infection include external ulcerative lesions in the fins and hyperplasia of the gill filaments, together with swelling of the body (Van Bearden *et al*. 2012). Gross clinical internal signs include hypertrophy and necrosis of the kidney and spleen, petechial haemorrhages of the liver, as well as haematoma and necrosis of gill lamellae cells (Sano *et al*. 1981; Noga *et al.* 2010; Plumb & Hansen 2011; Van Beurden *et al.* 2012). Viral titres are highest in the visceral organs (kidney, liver, pancreas and spleen), with lower levels occurring in the gill lamellae (Munro & Midtlyng 2011). EVE is commonly isolated from both farmed fingerlings and adult eels with no external signs of infection (Van Beurden *et al.* 2012).

The life cycle is direct and disease transmission is horizontal, through infected fish urine or faeces (Munro & Midtlyng 2011). The minimum infective dose for the related IPNV is very low. Experimental data indicates dosage by the oral route of 10^{-1} $TCID_{50}$ ml^{-1}, (defined as the quantity necessary to produce a cytopathic effect in 50% of the inoculated cell culture) with viral shedding peaking at 10^{2} $TCID_{50}$ ml^{-1} (Jarp *et al.* 1994). Infected wild stocks may transfer disease to farmed fish (Raynard *et al*. 2007).

EVE has high environmental tolerance, surviving in fresh and marine coastal waters for at least 15 days (Mortensen *et al*. 1990). It can be spread in transport water, contaminated nets, shipping containers and other equipment used in eel aquaculture. The optimum temperature range is 15 to 23°C (Haenen 2008), although the virus replicates at temperatures from 5 to 30°C and salinities ranging from 0 to 40% (Tubbs *et al.* 2007). Infections persist outside this temperature range without external signs of disease (Haenen 2008). EVE remains viable for several months in water at 4°C and is unaffected by UV light treatments (Tubbs *et al.* 2007).

EVE remains viable in moist decaying fish offal, is resistant to mild acid (1 hour at pH 2.5) (Tubbs *et al*. 2007) and survives in the avian digestive system. It can be transmitted between waterways where scavenging birds feed on infected offal (Raynard *et al.* 2007; Diggles 2011).

Aquabirnaviruses including EVE are unaffected by freezing (to -70°C) (Taksdal *et al*. 2004). Inactivation by heat treatment requires high temperatures (56°C for 2 hours), with moist heat being more effective than dry heat (Munro & Midtlyng 2011). No commercially available vaccines have been developed (Haenen 2012).

Susceptible species in New Zealand include the endemic long finned eel (*Anguilla dieffenbachii*) as well as the shortfin eel (*Anguilla australis*) and the spotted eel (*A. reinhardtii*) (Jellyman 1996).

Aquabirnaviruses have also been recovered from a range of marine invertebrates including blue mussel (*Mytilus galloprovincialis*) (Diggles 2011).

11.2. Risk assessment

11.2.1. Entry assessment

EVE has been suggested as an isolate of IPNV. Aquabirnaviruses have a wide host range with potential to cause new diseases in emerging fish farming species. Infected carrier fish may show no external signs of infection (Munro & Midtlyng 2011). These fish may pass visual inspection and be retained in the gill tissues present in the commodity. EVE is resistant to freezing and resistant to the environmental conditions likely to be encountered in the processing and importation of the commodity. The likelihood of entry is assessed as non-negligible.

11.2.2. Exposure assessment

For an infection to be established in a host population, sufficient infected product would have to become available for consumption by a susceptible marine or freshwater fish host, in sufficient quantity and duration (Kahn *et al.* 1999). EVE may remain viable in moist decaying fish offal and fish heads and can be transferred between waterways through the digestive system of scavenging birds (Raynard *et al.* 2007; Diggles 2011; Van Beurden *et al*. 2012). It may also be transferred horizontally through contaminated wash water and after disposal of contaminated organic material into waterways, through contamination of fish processing utensils and equipment, or through poor hygiene practices (Sano *et al*. 2011). The likelihood of exposure to EVE is assessed as non-negligible.

11.2.3. Consequence assessment

The introduction of EVE into New Zealand may hasten the decline of an already stressed endemic population (Jellyman 2012). Eels are an iconic and highly valued New Zealand species, with a commercial eel fishery valued at $4.9 million (Jellyman 2012). Eels are also important for non-commercial subsistence eel fishers and are considered a taonga by Maori (Parliamentary Commissioner for the Environment 2013). The introduction of EVE would have significant social and economic consequences. The eel fishery is considered stressed at current catch levels (MPI 2014).

The consequences of the introduction and establishment of EVE are assessed as non-negligible.

11.2.4. Risk estimation

Since the entry, exposure and consequence assessments are non-negligible, the risk estimate is non-negligible. Therefore, EVE is assessed to be a risk in the commodity and risk management measures may be justified.

11.3. Risk management

EVE has been assessed as a risk in the commodity. Infection with EVE is a non-notifiable disease so the *Aquatic Code* (OIE 2016) provides no specific guidance on mitigation measures for importing eviscerated fish from infected countries, or specific processing requirements that would ensure the destruction of the pathogen. It is assumed that compliance with the approved processed

states for OIE-listed diseases (Appendix 3) would also eliminate EVE from the commodity and be a viable risk management option.

EVE is reported from three fish families (Table 8) which are considered likely to be present in the commodity. Species declaration would substantially reduce pathogen load and be a viable risk management option. The disease is widely reported at low levels of infection in wild and farmed Japanese, Taiwanese and European eels, but no requirements for dedicated monitoring exist. Restriction of the commodity to wild-caught fish (not from aquaculture) would have little, or no effect on pathogen load and is not a viable risk management option, as the eel populations of the major eel exporting countries in Europe or Asia are linked through migration pathways.

Acceptance of a declaration of country freedom through the MPI Country Approval Procedures should substantially reduce the pathogen load of EVE in the commodity and be a viable risk management option.

EVE is restricted to the gill tissues in the eviscerated product, so removal of the gills would substantially reduce pathogen load and be a viable risk management option.

EVE is highly resistant to high and low temperatures, so extended heat treatment (to 56° C for 2 hours) should eliminate the pathogen and be a viable risk management option. EVE is unaffected by cold temperatures, so frozen storage is unlikely to be a viable management option.

EVE may be transmitted through the waste products associated with transport, storage and processing of the commodity. The requirement that all wash and wastewater discharges be appropriately chemically treated (e.g., with iodophors) before discharge, and that all solid wastes, tissue scraps and offal be disposed of through a recognised trade waste disposal procedure, would be viable management options.

11.3.1. Risk management options

EVE is reported from fish in families Anguillidae, Cichlidae and Salmonidae (Table 8), which are likely to be present in the commodity. Other families have not been associated with EVE. Therefore species declaration indicating the commodity is not originated from any of the above families should substantially reduce the occurrence of EVE in the commodity.

For the commodities originated from families associated with EVE, one or a combination of the following additional options could also be considered to effectively manage the risk.

Where country/zone freedom from EVE is accepted by MPI:

Option 1

Acceptance of country/zone freedom declaration should substantially reduce the occurrence of EVE, so the commodity may be imported without any further restrictions.

Where country/zone freedom from EVE is not accepted by MPI or not available:

Option 2

Processing to a state consistent with the approved states for OIE-listed diseases (Appendix 3) should eliminate EVE. When these provisions are met, the commodity could be imported without any further restrictions.

Where the imported commodity is not in a state consistent with the approved states for OIE-listed diseases (Appendix 3):

Option 3

Heat treatment (by cooking to at least 56° C for at least 120 minutes) should eliminate EVE. When this provision is met, the commodity could be imported without any further restrictions; or,

Option 4

Further processing (by removal of the gills) should substantially reduce the occurrence of EVE. When this provision is met, the commodity could be imported without any further restrictions.

11.4. References

Anonymous (2016). Biosecurity (Notifiable Organisms) Order 2016 SR 2016/73. New Zealand Government, Wellington, New Zealand, 13 p.

Bandin I, Souto S, Cutrin JM, López-Vazquez C, Olivera JG, Esteve C, Alcaide E, Dopazo CP (2014). Presence of viruses in wild eels *Anguilla anguilla* L, from the Albufera Lake (Spain). *Journal of Fish Diseases* 37: 597–607.

Castric J (1997). Viral diseases in fish mariculture. *Bulletin of the European Association of Fish Pathologists* 17(6): 220–228.

Crane M, Hyatt A (2011). Viruses of fish: An overview of significant pathogens. *Viruse*s 3: 2025–2046.

Diggles BK (2011). Risk analysis - aquatic animal diseases associated with domestic bait translocation. DigsFish Services Report 2009/072. Report prepared for the Fisheries Research and Development Corporation, Australia, 297 p.

Haenen OLM (2008). Impact of eel viruses on recruitment of European eel. pp 387–399. *In*: van den Thillart G, Dufour S, Rankin JC (*Eds*.). Spawning migration of the European eel; reproduction index, a useful tool for management. Springer Science, 490 p.

Haenen OLM (2012). Diseases of eels in an international perspective: Workshop on eel diseases at the 15th International Conference on Diseases of Fish and Shellfish, Split, Croatia. *Bulletin of the European Association of Fish Pathologists* 32(3): 109–115.

Haenen OLM, van Tulden PW, Dijkstra SG, Davidse A, Wagenaar F, Botter A, Basavarajappa HN, van Nieuwstadt AP (2001). Viruses isolated from European eel, *Anguilla anguilla* L. in The Netherlands from 1986-2001. Abstract of poster, European Association of Fish Pathologists Conference, Dublin, Ireland, 2001.

Jellyman D (2012). The status of longfin eels in New Zealand – an overview of stocks and harvest. NIWA client report CHC2012-006. Report prepared for the Parliamentary Commissioner for the Environment by the National Institute of Water and Atmospheric Research. Christchurch, New Zealand, 78 p.

Jellyman DJ, Chisnall BL, Dijkstra LH, Boubee JAT (1966). The first record of the Australian long finned eel, *Anguilla reinhardtii*, in New Zealand. *Australian Journal of Marine and Freshwater Research* 47:1037–1040.

Jarp J, Gjevre AG, Olesen AB, Bruheim T (1994). Risk factors for furunculosis, infectious pancreatic necrosis virus and mortality in post-smolt of Atlantic salmon, *Salmo salar*. *Journal of Fish Diseases* 18: 67–78.

Kahn SA, Beers PT, Findlay VL, Peebles IR, Durham PJ, Wilson DW, Gerrity SE (1999). Import risk analysis on non-viable salmonids and non-salmonid marine finfish. Australian Quarantine and Inspection Service, Department of Agriculture, Fisheries and Forestry, Canberra, Australia, 409 p.

Kou GH, Chen SN, Yeno Y, Hedrick RP, Fryer JL (1982). Investigation of viral infections in cultured fishes in Taiwan, pp 42-43 *In*: Report from the Republic of Taiwan-Japan Symposium on Fish Diseases. Tungkang, Taiwan, Republic of China.

Kusuda R, Nishi Y, Hosono N, Suzuki S (1993). Serological comparison of birnaviruses isolated from several species of marine fish in southwest Japan. *Fish Pathology* 28: 91–92.

Mortensen SH, Hjeltnes B, Rodseth O, Krogsrud J, Christie KE (1990). Infectious pancreatic necrosis virus, serotype N1, isolated from Norwegian halibut (*Hippoglossus hippoglossus*), turbot (*Scophthalmus maximus*), and scallops (*Pecten maximus*). *Bulletin of the European Association of Fish Pathology* 10: 42–43.

Ministry for Primary Industries (MPI) (2014). Freshwater eels (SFE, LFE), pp 276–301. *In:* Fisheries Assessment Plenary, May 2014: stock assessments and stock status. Compiled by the Fisheries Science Group, Ministry for Primary Industries, Wellington, New Zealand, 1381 p.

Munro ES, Midtlyng PJ (2011). Infectious pancreatic necrosis virus and associated aquatic birnaviruses. pp 1–65. *In:* Woo PTK, Bruno DW (*Eds*.). Fish Diseases and Disorders: Volume 3 - Viral, Bacterial and Fungal Infections, 2nd edition. CABI Publishers, Wallingford, United Kingdom, 930 p.

NIWA (2017). Tuna-spawning grounds. National Institute for Water and Atmospheric Research, Auckland, New Zealand. [Online] Available from: https://www.niwa.co.nz/te-k%C5%ABwaha/tuna-information-resource/biology-and-ecology/spawning-grounds [Accessed 8 August 2017].

Noga EJ (2010). Fish Disease: Diagnosis and treatment, Second edition. Wiley, New York, 536 p.

OIE (2016). Aquatic Animal Health Code, 19th Edition. World Organization for Animal Health. Paris, France [Online]. Available from: http://www.oie.int/international-standard-setting/aquatic-code/access-online/ [Accessed 15 March 2017].

Parliamentary Commissioner for the Environment (2013). On a pathway to extinction? An investigation into the status and management of the longfin eel. Parliamentary Commission for the Environment, Wellington, New Zealand, 95 p.

Plumb JA, Hanson LA. (2011). Health maintenance and principal microbial diseases of cultured fishes. Wiley, New York, 400 p.

Raynard R, Wahli T, Vatsos I, Mortensen S (2007). Review of disease interactions and pathogen exchange between farmed and wild finfish and shellfish in Europe (DIPNET). VESO Project 1655 report for the European Union. IFREMER, Paris, France, 459 p.

Sano T, Okamoto N, Nishimura T (1981). A new viral epizootic of *Anguilla japonica* Temminck & Schlegel. *Journal of Fish Diseases* 4: 127–139.

Sano M, Nakai T, Fijan N (2011). Viral diseases and agents of warm water fish. *pp.* 166–244. *In:* Woo PTK, Bruno DW (*Eds.*). Fish Diseases and Disorders: Volume 3: Viral, Bacterial and Fungal Infections, 2nd edition. CABI Publishers, Wallingford, United Kingdom, 930 p.

Taksdal T, Ramstad A, Stangeland K, Dannevig BH (2004). Induction of infectious pancreatic necrosis (IPN) in covertly infected Atlantic salmon, *Salmo salar* L., post-smolts by stress exposure, in injection of IPN virus (IPNV) and by cohabitation. *Journal of Fish Diseases* 21(3): 193–204.

Tubbs L, Lee P, Diggles B, Jones JB, Sheppard M, Sim-Smith C (2007). A review of aquatic diseases of significance to New Zealand. Final research report for project ZBS 2005-17. National Institute of Water and Atmospheric Research, Wellington, New Zealand, 452 p.

Van Beurden SJ, Engelsma MY, Roozenberg I, Voorbergen-Laarman MA, van Tulden PW, Kerkhoff S, van Nieuwstadt AP, Davidse A. Haenen OLM (2012). Viral diseases of wild and farmed European eel *Anguilla anguilla* with particular reference to the Netherlands. *Diseases of Aquatic Organisms* 101: 69–86.

Van Ginneken V, Ballieux B, Willemze R, Coldenhoff K, Lentjes E, Antonissen E, Haenen O, Van den Thillart GEE (2005). Hematology patterns of migrating European eels and the role of EVEX virus. *Comparative Biochemistry and Physiology Part C: Toxicology and Pharmacology* 140(1): 97-102.

Van Ginneken V, Haenen O, Coldenhoff, K, Willemze R, Antonissen E, Van Tulden P, Dijkstra S, Wagenaar F, van den Thillart GEE (2004). Presence of eel viruses in eel species from various geographic regions. *Bulletin of the European Association of Fish Pathologists* 24(5): 268-272.

12. European eel herpesvirus

12.1. Hazard identification

12.1.1. Aetiological agent

European eel herpesvirus (EEHV) is also known as herpesvirus anguillae (HVA), or anguillid herpesvirus 1 (AngHV-1) (Van Beurden *et al*. 2012). It is the agent of gill herpesvirus of eels, which was originally described in 1985 from farmed European eels (*Anguilla anguilla*) in Hungary (Bekesi *et al*. 1986).

EEHV is an icosahedral double stranded DNA virus classified as a *Cyprinivirus* within the Family Alloherpesviridae (Van Beurden *et al.* 2012). Herpesviruses in fish may represent a single virus species (Waltzek *et al*. 2009), but recent data suggests two separate clades occur within *Cyprinivirus*; one which includes EEHV and the cyprinid herpesviruses; and another which includes ictalurid and other herpesviruses (Van Beurden *et al.* 2010).

12.1.2. OIE status

Infection with EEHV is not an OIE-notifiable disease (OIE 2016a).

12.1.3. New Zealand status

EEHV is not reported from New Zealand eels and has been assumed to be exotic. Infection with EEHV is not a notifiable disease in New Zealand (Anon. 2016).

12.1.4. Epidemiology

EEHV is considered one of the most significant viral threats to European eels (*A. anguilla*) because of its high pathogenicity (Jakob *et al.* 2009). It is only reported from eels (Family Anguillidae). It occurs in farmed *A. anguilla* in Denmark, France, Germany, Greece, the Netherlands, Poland and the United Kingdom (Armitage *et al.* 2013), as well as in wild *A. anguilla* in Europe (Van Beurden *et al.* 2012). It is also reported from *A. japonica* farmed in Japan and from *A. anguilla* and *A. rostrata* farmed in China and Taiwan (Sano *et al.* 1990; Van Beurden *et al*. 2012; Kempter *et al.* 2014). Eels of all ages appear susceptible (Kibenge & Godoy 2016).

Eels are catadromous, spending most time in estuaries and rivers, but returning to the sea to spawn. The European eels (*A. anguilla*) and the American eel (*A. rostrata*) share the same spawning grounds in the Sargasso Sea. While Australian and New Zealand stocks of *A. australis* are thought to share spawning grounds in the central Pacific Ocean (Bandin *et al*. 2014). Little is known about contacts with other eel fish stocks.

Eels sold for human consumption are sourced from aquaculture, but based on the collection and on-growing of juvenile glass eels, collected from the wild. European and Asian eel aquaculture of *A. anguilla* is largely based on seed stock derived from European sources. *A. rostrata* is extensively farmed in the United States, while *A. australis* is cultured in Australia and Asia (from Australian seed stock). *A. japonica* is farmed in Japan, Taiwan and South Korea from wild-sourced juvenile stocks (McKinnon & Godley 1998).

The occurrence of EEHV may exceed 50% in natural populations (Hanson *et al.* 2016) and natural mortality rates of 10–48% have been implicated in the decline of European *A. anguilla* (Sano *et al.* 1990; Haenen *et al.* 2012). Mortalities of up to 30% are reported from aquaculture, commonly associated with stress factors such as poor water quality, poor husbandry or overstocking (Chang *et al*. 2002; Hanson *et al.* 2016).

External signs of infection are non-specific and similar to infection with other eel viruses (EVE and EVEX). These include fin and skin haemorrhage and necrosis, as well as gill hyperplasia and necrosis (Haenen *et al*. 2010, 2011; Lepa & Siwicki 2912; Van Beurden *et al*. 2012). Internal signs include liver inflammation and necrosis of the liver, kidney and spleen (Armitage *et al.* 2013), with viral presence initially detected in gill and stomach tissues 4 days post-bath challenge (Hagalpura *et al*. 2007; Hanson *et al.* 2011).

Latent infection may occur without clinical signs with apparent low infection prevalence of 2% in both farmed and wild eel stocks (Rijsewijk *et al.* 2005), but this may change rapidly to an acute aetiopathology, caused in part by immune suppression, and to stress-reactivation due to external stress factors (Van Nieuwstadt *et al*. 2001; Jakob *et al.* 2009). EEHV infection appears temperature related, within a range from 15–30°C, with little viral replication occurring below 10°C (Jakob *et al*. 2009).

EEHV is highly resistant to high and low temperatures, requiring heat treatment to 56° C for 2 hours for inactivation. It is resistant to cold temperatures and unaffected by freezing (OIE 2016b).

Viral shedding occurs across the gills and through the digestive system and infection occurs horizontally through the water column (Haenen *et al*. 2001; Hanson *et al*. 2016).

No vaccines are available for EEHV (Hanson *et al.* 2016).

Susceptible species in New Zealand include *A. australis*, *A. dieffenbachii* and *A. reinhardtii* (Jellyman 1996).

12.2. Risk assessment

12.2.1. Entry assessment

EEHV is a virulent and commonly occurring viral infection of farmed and wild eels. Eels with obvious lesions should not be present in the commodity and viral tire would be expected to be lower in the eviscerated commodity. However, latent or sub-clinically infected eels may pass visual inspection and be present in the commodity. Herpesviruses are unaffected by freezing and EEHV is also likely to remain viable in the commodity, or in transport water.

The likelihood of entry of EEHV is assessed as non-negligible.

12.2.2. Exposure assessment

For an infection to be established in a host population, sufficient infected product would have to become available for consumption by a susceptible marine or freshwater fish host, in sufficient quantity and duration (Kahn *et al.* 1999). EEHV may remain viable in the gills and skin tissue of infected eviscerated fish (Van Beurden *et al.* 2012). It may also be transferred through contaminated wash water and disposal of contaminated organic material into waterways, through

contamination of fish processing utensils and equipment, or through poor hygiene practices (Sano *et al.* 2011).

The likelihood of exposure to EEHV is assessed as non-negligible.

12.2.3. Consequence assessment

Eels are a valuable commercial product, supporting a commercial fishery valued at $4.9 million Eels are exported as live fish, or frozen, chilled or smoked product, mainly to Europe (Belgium, Germany and the United Kingdom), South Korea, Taiwan and Hong Kong (Jellyman 2012). Eels support small domestic and recreational fisheries and represent a significant taonga for Maori (Parliamentary Commissioner for the Environment 2013). The eel fishery resource is considered to be stressed at current commercial catch levels (MPI 2014).

The consequences of the introduction of EEHV are assessed as non-negligible.

12.2.4. Risk estimation

Since the entry, exposure and consequence assessments are non-negligible, under the methodology of this risk assessment, the risk associated with EEHV is assessed as non-negligible and further management measures may be established.

12.3. Risk management

EEHV has been assessed as a risk in the commodity. Infection with EEHV is a non-notifiable disease, therefore, the *Aquatic Code* (OIE 2016a) provides no specific guidance on mitigation measures for importing eviscerated fish from infected countries, or specific processing requirements that would ensure the destruction of the pathogen. It is assumed that compliance with the approved processed states for OIE-listed diseases (Appendix 3) would also eliminate EEHV from the commodity and be a viable risk management option.

EEHV only occurs in eels (Anguillidae), so species declaration should substantially reduce the pathogen load of EEHV and is a viable risk management option. EEHV is reported at low levels of infection in wild and farmed European (Europe and the United Kingdom) and Asian (Japanese, South Korean and Taiwanese) eels, where these eel populations are linked through migration and recruitment pathways (Van Beurden *et al.* 2012). Restriction of the commodity to wild-caught fish (not from aquaculture) should have little, or no effect on pathogen load and is not a viable risk management option.

Where declaration of country/zone freedom is approved by MPI, country freedom is likely to substantially reduce the pathogen load of EEHV and be a viable risk management option.

Latent infection is restricted to the head, fins and gill tissues. Pathogen load should be slightly reduced by the removal of the gills and be moderately reduced by removal of the head and gills. Removal of the head and gills is a viable risk management option.

EEHV is resistant to high temperatures, requiring heat treatment to 56° C for 2 hours for inactivation. This would eliminate the pathogen from the commodity and be a viable risk management option. EEHV is unaffected by cold temperatures, so frozen storage would not be a viable risk management option.

EEHV may be transmitted through the waste products associated with transport, storage and processing of the commodity. The requirement that all wash and wastewater discharges be appropriately chemically treated (e.g., with iodophors) before discharge, and that all solid wastes, tissue scraps and offal be disposed of through a recognised trade waste disposal procedure, would be viable management options.

12.3.1. Risk management options

EEHV is restricted to fish of family Anguillidae, which is likely to be present in the commodity. Other families have not been associated with EEHV. Therefore species declaration indicating the commodity is not originated from the above family should substantially reduce the occurrence of EEHV in the commodity.

For the commodities originated from family Anguillidae, one or a combination of the following additional options could also be considered to effectively manage the risk.

Where country/zone freedom from EEHV is accepted by MPI:

Option 1

Acceptance of country/zone freedom should substantially reduce pathogen occurrence, so the commodity may be imported without any further restrictions.

Where country/zone freedom from EEHV is not accepted by MPI, or not available:

Option 2

Processing to a state consistent with the approved states for OIE-listed diseases (Appendix 3) should eliminate EEHV. Where these provisions are met, the commodity could be imported without any further restrictions.

Where the imported commodity is not in a state consistent with the approved states for OIE-listed diseases (Appendix 3):

Option 3

Heat treatment (by cooking to at least 56°C, for at least 120 minutes) should eliminate EEHV. Where this provision is met, the commodity could be imported without any further restrictions; or,

Option 4

Further processing (by removal of the head and gills) should moderately reduce occurrence of EEHV. Where this provision is met, the commodity could be imported without any further restrictions.

12.4. References

Anonymous (2016). Biosecurity (Notifiable Organisms) Order 2016 SR 2016/73. New Zealand Government, Wellington, New Zealand, 13 p.

Armitage J, Hewlett NR, Twigg M, Lewin NC, Reading AJ, Williams CF, Aprehamian M, Way K, Feist SW, Peeler EJ (2013). Detection of *Herpesvirus anguillae* during two mortality investigations of wild European eel in England: implications for fishery management. *Fisheries Management and Ecology* 21(1): 1–12.

Bandin I, Souto S, Cutrin JM, López-Vazquez C, Olivera JG, Esteve C, Alcaide E, Dopazo CP (2014). Presence of viruses in wild eels *Anguilla anguilla* L, from the Albufera Lake (Spain). *Journal of Fish Diseases* 37: 597–607.

Bekesi L, Horvath I, Kovacs-Gayer E, Csaba G (1986). Demonstration of herpesvirus-like particles in skin lesions of European eel (*Anguilla anguilla*). *Journal of Applied Ichthyology* 2(4): 190–192.

Chang PH, Pan YH, Wu CM, Kuo ST, Chung HY (2002). Isolation and molecular characterization of herpesvirus from cultured European eels *Anguilla anguilla* in Taiwan. *Diseases of Aquatic Organisms* 50: 111–118.

Haenen OLM, Lehmann J, Engelsma MY, Stürenberg FJ, Roozenburg I, Kerkhoff S, Klein Breteler J (2010). The health status of European silver eels, *Anguilla anguilla*, in the Dutch River Rhine Watershed and Lake Ijsselmeer. *Aquaculture* 309: 15–24.

Haenen OLM (2012). Diseases of eels in an international perspective: Workshop on eel diseases at the 15th International Conference on Diseases of Fish and Shellfish, Split, Croatia. *Bulletin of the European Association of Fish Pathologists* 32(3): 109–115.

Hangalapura BN, Zwart R, Engelsma MY, Haenen OL (2007). Pathogenesis of Herpesvirus anguillae (HVA) in juvenile European eel *Anguilla anguilla* after infection by bath immersion. *Diseases of Aquatic Organisms* 78(1): 13–22.

Hanson L, Dishon A, Kotler M (2011). Herpesviruses that infect fish. *Viruses* 3(11): 2160–2191.

Hanson L, Doszpoly A, van Beurden SJ, de Oliveira Viadanna PH, Waltzek (2016). Chapter 9, Alloherpesviruses of fish. uses of fish, pp 153-172 *In:* Kibenge FSB, Godoy M (*Eds*.). Aquaculture Virology, Academic Press, New York, United States of America, 568 p.

Jakob E, Neuhaus H, Steinhagen D, Lukhardt B, Hanel R (2009). Monitoring of *Herpesvirus anguillae* (HVA) infections in European eel, *Anguilla anguilla* (L.), in northern Germany. *Journal of Fish Diseases* 32: 557–561.

Kahn SA, Beers PT, Findlay VL, Peebles IR, Durham PJ, Wilson DW, Gerrity SE (1999). Import risk analysis on non-viable salmonids and non-salmonid marine finfish. Australian Quarantine and Inspection Service, Department of Agriculture, Fisheries and Forestry, Canberra, Australia, 409 p.

Kempter J, Hofsoe P, Panicz R, Bergmann SM (2014). First detection of anguillid herpesvirus 1 (AngHV1) in European eel (*Anguilla anguilla*) and imported American eel (*Anguilla rostrata*) in Poland. *Bulletin of the European Association of Fish Pathologists* 34(3): 87-94.

Lepa A, Siwicki AJ (2012). Fish herpesvirus diseases: a short review of current knowledge. *Acta Veterinaria* BRNO 81: 383–389.**McKinnon LJ, Godley GJ (1998).** Key environmental criteria associated with the invasion of *Anguilla australis* glass eels into estuaries of South-Eastern Australia. *Bulletin Français de la Pêche et de la Pisciculture* 349: 117–128.

Ministry for Primary Industries (MPI) (2014). Freshwater eels (SFE, LFE), pp 276–301. *In:* Fisheries Assessment Plenary, May 2014: stock assessments and stock status. Compiled by the Fisheries Science Group, Ministry for Primary Industries, Wellington, New Zealand, 1381 p.

OIE (2016a). Aquatic Animal Health Code, 19th Edition. World Organization for Animal Health. Paris, France [Online]. Available from: http://www.oie.int/international-standard-setting/aquatic-code/access-online/ [Accessed 15 March 2017].

OIE (2016b). Manual of Diagnostic Tests for Aquatic Animals. World Organisaton for Animal Health, Paris, France [Online]. Available from: http://www.oie.int/international-standard-setting/aquatic-manual/access-online/ [Accessed 13 March 2017].

Parliamentary Commissioner for the Environment (2013). On a pathway to extinction? An investigation into the status and management of the longfin eel. Parliamentary Commission for the Environment, Wellington, New Zealand, 95 p.

Rijsewijk F, Pritz-Vershuren S, Kerkhoff S, Botter A, Willemsen M, van Nieuwstadt T, Haenen O (2005). Development of a polymerase chain reaction for the detection of anguillid herpesvirus zDNA in eels based on the herpesvirus DNA polymerase gene. *Journal of Virological Methods* 124: 87–94.

Sano M, Fukuda H, Sano T (1990). Isolation and characterization of a new herpesvirus from eel. pp 15–31. *In*: Perkins TO, Cheng TC (*Eds*.) Pathology in Marine Science, Vol 3. Academic Press, New York, United States of America, 538 p.

Sano M, Nakai T, Fijan N (2011). Viral diseases and agents of warm water fish. *pp.* 166–244. *In:* Woo PTK, Bruno DW (*Eds*.). Fish Diseases and Disorders: Volume 3: Viral, Bacterial and Fungal Infections, 2nd edition. CABI Publishers, Wallingford, UK. , 930 p.

Van Beurden SJ, Bossers A, Voorberg-Laarman MH, Haenen OL, Peters S, Abma-Henkens MH, Peeters BP, Rottier PJ, Engelsma MY (2010). Complete genome sequence and taxonomic position of anguillid herpesvirus 1. *Journal of General Virology* 91(4): 880–887.

Van Beurden SJ, Engelsma MY, Roozenberg I, Voorbergen-Laarman MA, van Tulden PW, Kerkhoff S, van Nieuwstadt AP, Davidse A, Haenen OL (2012). Viral diseases of wild and farmed European eel *Anguilla anguilla* with particular reference to the Netherlands. *Diseases of Aquatic Organisms* 101: 69–86.

Van Nieuwstadt AP, Dijkstra SG, Haenen OLM (2001). Persistence of herpesvirus of eel *Herpesvirus anguillae* in farmed European eel *Anguilla anguilla*. *Diseases of Aquatic Organisms* 45: 103−107.

Waltzek TB, Kelley GO, Alfaro ME, Kurobe T, Davison AJ, Hedrick RP (2009). Phylogenetic relationships in the family Alloherpesviridae. *Diseases of Aquatic Organisms* 84: 179−194.

13. Grass carp haemorrhagic virus

13.1. Hazard identification

13.1.1. Aetiological agent

Grass carp haemorrhagic virus (GCHV) is a double stranded icosahedral RNA virus classified within the Family Reoviridae. It was first reported from grass carp (*Ctenopharyngodon idella*) in China in 1972 (Nie & Pan 1985) and was later identified as an aquareovirus (Chen & Jiang 1984). It primarily affects fry and yearling grass carp, causing haemorrhagic disease and red spot disease (RSD). It is highly contagious and considered one of the most serious viral diseases of grass carp in China and Southeast Asia (Plumb & Hanson 2011).

Four genotypes have been identified, with 25 separate isolates that vary in their pathogenicity and antigenicity (Yan *et al*. 2014). GCHV is also closely related to golden shiner reovirus (GSV) and these viruses are now considered as isolates of Aquareovirus C (Plumb & Hanson 2011).

13.1.2. OIE status

Infection with GCHV is not notifiable to the OIE (OIE 2016).

13.1.3. New Zealand status

The original and each subsequent imported brood stock of grass carp have been certified as free from GCHV. There have been no reports of GCHV in over 10 years of monitoring (B. Jones, *pers. comm*. 2014), so GCHV is considered exotic. However, GCHV is not a notifiable organism in New Zealand (Anon. 2016).

13.1.4. Epidemiology

GCHV has been reported from grass carp aquaculture in Asia (China and Vietnam) and the United States. It is primarily a disease of fry and yearling fish, but fish up to three years old may be affected (Jiang 2009; Plumb & Hanson 2011)

Major host species (Table 9) in Asian aquaculture include grass carp (*Ctenopharyngodon idella*), Asian black carp (*Mylopharyngodon piceus*), and topmouth gudgeon (*Pseudorasbora parva*) (Jiang 2009). Also affected are tench (*Tinca tinca*) in the United States (Cudmore & Mandrak 2011), while viral replication is reported from other cyprinids including bighead carp (*Hypophthalmichthys* (*Aristichthys*) *nobilis*), golden carp (*Carassius auratus*), rare minnow (*Gobiocypris rarus*) and silver carp (*Hypophthalmichthys molitrix*). Infection usually occurs with no clinical signs of disease. These species may also act as a reservoir for infection (Jiang 2009).

Table 9. Families and Species of Fish Susceptible to Grass Carp Haemorrhagic Virus (GCHV)

Grass Carp Haemorrhagic Virus (GCHV)	
Family	**Host Species**
Cyprinidae	Asian black carp (*Mylopharyngodon piceus*), bighead carp (*Hypophthalmichthys (nobilis*), golden carp (*Carassius auratus*), grass carp (*Ctenopharyngodon idella*), rare minnow (*Gobiocypris rarus*), silver carp (*Hypophthalmichthys molitrix*), tench (*Tinca tinca*) topmouth gudgeon (*Pseudorasbora parva*)

The distribution of GCHV may be wider than presently reported, as infection may also occur in areas where water temperatures are too low for infected fish to show clinical signs. Such fish may also function as carriers of disease and spread GCHV both nationally and internationally (Jiang 2009).

Epidemics are generally associated with warm summer water temperatures of 25 to 30°C, when mortalities may reach 80% for grass carp fry and up to 65% for yearling fish (Plumb & Hanson 2011). When water temperatures are lower, fish may not show external signs of infection (Plumb & Hanson 2011). Mortality rates of 30% occur in vaccinated fish (Jiang 2009) and sub-clinically infected fish may represent carriers of infection (Jun *et al.* 1997).

External signs of infection include skin haemorrhages and exophthalmia. Internally, infection is focused on liver, spleen and kidneys, with extensive haemorrhage and necrosis. Infection may spread to gill, brain and muscle tissues in clinical infection (Plumb & Hanson 2011).

GCHV is highly contagious and is transmitted horizontally through the water column, from either clinically infected hosts or surviving adult grass carp and other carrier fish with no external signs of infection. It may also be spread by ectoparasites such as *Argulus* spp. (Plumb & Hanson 2011). The site of virus entry and shedding is across the gill membranes (Wang *et al.* 1993).

Aquareoviruses are extremely environmentally stable, surviving in low-moisture processed food for more than 14 days at room temperature and for more than 2 months at 5°C (Pirtle & Beran 1991). They can remain viable for over a year (630 days) in municipal wastewater at 8°C and for almost two weeks (13.3 days) at 26°C before T_{90} occurs (90% loss of virus activity) (Pirtle & Beran 1991). GCHV remained viable after long-term frozen storage at -80°C and after three freeze-thaw cycles (Fan *et al*. 2013).

Aquareoviruses survived for almost six months at 8°C and almost a week at 26°C before T_{90} was achieved in distilled water (McDaniels *et al.* 1983). They are heat stable, requiring temperatures exceeding 56°C for up to 2 hours for inactivation. GCHV is stable over a wide range of pH, although infectivity is reduced at pH < 3.0 (John *et al.* 2001).

Reoviruses are resistant to freezing at -80°C (Rudd & Lemay 2005) as well as to chemical and physical disinfectants (Thomas *et al.* 2008). GCHV has been controlled in China by extensive use of a simple attenuated live virus prepared from infected fish (Jiang 2009), but more effective inactivated whole-virus vaccines are being developed (Lu *et al.* 2011). GCHV remains an important disease of grass carp aquaculture in other Southeast Asian countries where attenuated vaccines are not used (Jiang 2009).

Susceptible species in New Zealand include introduced carp, goldfish and tench species (Mitchell 2009). However, little is known about the potential hosts of GCHV among native New Zealand fish (Hofstra *et al.* 2014).

13.2. Risk assessment

13.2.1. Entry assessment

GCHV is a significant disease of carp in Southeast Asian and Chinese aquaculture. It is relatively non-host specific (Jiang 2009), infecting several farmed and wild freshwater carp species. Freshwater fish such as carp are currently prohibited imports, as import regulations permit only frozen skinless and boneless fillets of *Pangasius* spp., *Oreochromis* spp. and Nile perch *Lates niloticus* to enter New Zealand for human consumption (MPI 2000, 2011).

While evisceration would reduce viral titre, GCHV present in the gill and muscle tissue (Plumb & Hanson 2011) of infected adult fish would be retained in the commodity. In addition, clinically infected carrier fish with no external clinical signs of infection may pass visual inspection and be present in the commodity. GCHV may be present in offal and blood-water discharges discarded during fish processing.

The likelihood of entry through the commodity is assessed as non-negligible.

13.2.2. Exposure assessment

To establish infection in New Zealand, infected eviscerated product would have to become available for consumption by a susceptible freshwater fish host, in sufficient quantity and duration (Kahn *et al.* 1999). GCHV transferred into waterways through wastewater discharges may remain viable for up to a year (McDaniels *et al.* 1983), and suitable freshwater fish hosts are present in New Zealand (Mitchell 2009).

Reoviruses show high environmental stability. GCHV may remain viable after passage through municipal sewage treatment (McDaniels *et al.* 1983) and remain infective for up to 36 days in landfill (Ware 1980). Reoviruses are not heat labile and are considered unlikely to survive passage through the digestive system of scavenging birds (John *et al*. 2001).

The likelihood of exposure to GCHV is assessed as non-negligible.

13.2.3. Consequence assessment

The consequence of introduction of GCHV into New Zealand indigenous freshwater fish is essentially unknown (Hofstra *et al.* 2014). GCHV is recognised as a potential limiting factor in the use of introduced grass carp and silver carp for the management of waterways (Mitchell 2009) and this weed control activity may potentially act to spread GCHV in New Zealand (Jiang 2009; Hofstra *et al*. 2014). While GCHV is likely to also affect other non-indigenous "pest" species including goldfish (*Carassius auratus*) (Hofstra *et al.* 2014), other more likely pathways for introduction including the importation and release of ornamental fish (Hine & Diggles 1995) also exist.

The consequence of establishment of GCHV is assessed as non-negligible.

13.2.4. Risk estimation

Since the entry, exposure and consequence assessments for GCHV are non-negligible, the risk estimation is non-negligible. Therefore, grass carp haemorrhagic virus is assessed to be a risk in the commodity and risk management measures may be justified.

13.3. Risk management

GCHV has been assessed to be a risk in the commodity. Infection with GCHV is a non-notifiable disease, so the OIE *Aquatic Code* (OIE 2016) provides no specific guidance on mitigation measures for importing eviscerated fish from infected countries, or specific processing requirements that would ensure the destruction of the pathogen. It is assumed that compliance with the approved processed states for OIE-listed diseases (Appendix 3) would also eliminate GCHV from the commodity and be a viable risk management option.

GCHV is restricted to wild and farmed fish of family Cyprinidae (Table 9). Cyprinids present in the commodity may function as carriers with no clinical signs of disease (Jun *et al.* 1997; Jiang 2009). Species declaration should substantially reduce the pathogen load of GCHV and be a viable risk management option. As restriction of the commodity to wild-caught fish (not from aquaculture) is likely to have little, or no effect on pathogen load, this is not considered a viable risk management option.

GCHV is reported extensively throughout South East Asia and China, as well as in the United States (Jian 2009; Cudmore & Mandrak 2011), but is not reported from European or African cyprinids, although no requirements for dedicated monitoring exist.

Where declaration of country/zone freedom is approved through the MPI Country Approvals Procedures, this option should substantially reduce the occurrence of GCHV in the commodity. Approval of a declaration of country/zone freedom by MPI is a viable risk management option.

GCHV may be present in the gills, brain and muscle of eviscerated fish (Plumb & Hanson 2011), so removal of the gills should slightly reduce GCHV occurrence in the commodity. Removal of the head and gills should moderately reduce pathogen load and be a viable risk management option. GCHV is unaffected by freezing, so frozen storage is not a viable risk management option.

GCHV is inactivated by heat treatment (cooking to at least 56°C for up to 2 hours (John *et al.* 2001), so this should eliminate the pathogen from the commodity. Heat treatment is a viable risk management option.

While GCHV is controlled in China by vaccination (Jiang 2009), available vaccines are of limited effectiveness. They can promote sub-clinical infection, or the development of a carrier state (Jun *et al.* 1997; Jiang 2009), with an unknown effect on pathogen load. Vaccination is not considered to be a viable risk management option and is not considered further.

GCHV may be transmitted through the waste products associated with transport, storage and processing of the commodity. The requirement that all wash and wastewater discharges be appropriately chemically treated (e.g., with iodophors) before discharge, and that all solid wastes, tissue scraps and offal be disposed of through a recognised trade waste disposal procedure, would be viable management options.

13.3.1. Risk management options

GCHV is reported from fish in family Cyprinidae (Table 9), which are considered likely to be present in the commodity. Other families have not been associated with GCHV. Therefore, species declaration indicating the commodity is not originated from family Cyprinidae should substantially reduce the occurrence of GCHV in the commodity.

For commodities originating from family Cyprinidae, one or a combination of the following additional options could also be considered to effectively manage the risk.

Where country/zone freedom from GCHV is accepted by MPI:

Option 1

Acceptance of country/zone freedom should substantially reduce the occurrence of GCHV, so the commodity could be imported without any further restrictions.

Where country/zone freedom from GCHV is not accepted by MPI, or not available:

Option 2

Processing to a state consistent with the approved states for OIE-listed diseases (Appendix 3) should eliminate GCHV. When these provisions are met, the commodity could be imported without any further restrictions.

Where the imported commodity is not processed to a state consistent with the approved states for OIE-listed diseases (Appendix 3):

Option 3

Heat treatment (by cooking to at least 56°C for at least 120 minutes) should eliminate GCHV. When this provision is met, the commodity could be imported without any further restrictions; or,

Option 4

Further processing by removal of the head and gills should moderately reduce the occurrence of GCHV. When this provision is met, the commodity could be imported without any further restrictions.

13.4. References

Anonymous (2016). Biosecurity (Notifiable Organisms) Order 2016 SR 2016/73. New Zealand Government, Wellington, New Zealand, 13 p.

Chen Y, Jiang Y (1984). Morphological and physio-chemical characterization of the haemorrhagic virus of grass carp. *Kexue Tongbao* 29: 832–835.

Cudmore B, Mandrak NE (2011). Biological synopsis of tench (*Tinca tinca*). *Canadian Manuscript Report of Fisheries and Aquatic Sciences* 2948, 28 p.

Fan Y, Rao S, Zeng L, Ma J, Zhou Y, Xu J, Zhang H (2013). Identification and genomic analysis of a novel fish reovirus, Hubei grass carp disease reovirus, isolated in 2009 in China. *Journal of General Virology* 94: 2266–2277.

Hine PM, Diggles BK (2005). Import risk analysis: Ornamental fish. Biosecurity New Zealand, Ministry of Agriculture and Forestry, Wellington, New Zealand, 262 p.

Hofstra DE, Rowe DK, Clayton JS (2014). Assessment of grass carp use for aquatic weed control: environmental impacts, management constraints and biosecurity risks in New Zealand waters. NIWA Client Report HAM2014-061, prepared for Ministry of Primary Industries. National Institute of Water and Atmospheric Research, Hamilton, New Zealand, 160 p.

Jiang Y (2009). Haemorrhagic disease of grass carp: Status of outbreaks, diagnosis, surveillance and research. *Israeli Journal of Aquaculture* 61(3): 188–197.

John KR, George MR, Richards RH, Frerichs GN (2001). Characteristics of a new reovirus isolated from epizootic ulcerative syndrome infected snakehead fish. *Diseases of Aquatic Organisms* 46: 83–92.

Kahn SA, Beers PT, Findlay VL, Peebles IR, Durham PJ, Wilson DW, Gerrity SE (1999). Import risk analysis on non-viable salmonids and non-salmonid marine finfish. Australian Quarantine and Inspection Service, Department of Agriculture, Fisheries and Forestry, Canberra, Australia, 409 p.

Lu L, Xu H, He Y, Li J (2011). Protection of grass carp *Ctenopharyngodon idella* (Valenciennes) through the oral administration of a sub unit virus against reovirus. *Journal of Fish Diseases* 34(12): 939–942.

McDaniels AE, Cochran KW, Gannon JJ, Williams GW (1983). Rotavirus and reovirus stability in microorganism free distilled and wastewaters. *Water Research* 17(10): 1349–1353.

Mitchell C (2009). Profile of white amur (grass carp) and silver amur (silver carp) from the results of releases in New Zealand waterways. Charles Mitchell and Associates, Raglan, New Zealand, 15 p.

Ministry for Primary Industries (MPI) (2000). Import health standard for the importation into New Zealand of frozen Nile perch (*Lates niloticus*) skinless, boneless fillets for human consumption from Kenya, Tanzania or Uganda (Lake Victoria) (Fisnpeic.all). Ministry for Primary Industries, Wellington, New Zealand, 2 p.

Ministry for Primary Industries (MPI) (2011). Import health standard for processed tilapia and catfish for human consumption from specified countries. Ministry for Primary Industries, Wellington, New Zealand, 2 p.

Nie DS, Pan JP (1985). Diseases of grass carp (*Ctenopharyngodon idella* Valenciennes, 1844) in China, a review from 1953 to 1983. *Fish Pathology* 20: 323–330.

OIE (2016). Aquatic Animal Health Code, 19th Edition. World Organization for Animal Health. Paris, France [Online]. Available from: http://www.oie.int/international-standard-setting/aquatic-code/access-online/ [Accessed 15 March 2017].

Pirtle EC, Beran GW (1991). Virus survival in the environment. *OIE Review of Science and Technology* 10(3): 733–748.

Plumb JA, Hanson LA (2011). Health maintenance and principal microbial diseases of cultured fishes. John Wiley, New York, United States of America, 400 p.

Rudd P, Lemay G (2005). Correlation between interferon sensitivity of reovirus isolates and ability to discriminate between normal and Ras-transformed cells. *Journal of General Virology* 86(5): 1489–1497.

Thomas NJ, Hunter TD, Atkinson CT (2008). Infectious diseases of wild birds. John Wiley, New York, United states of America, 496 p.

Wang T, Chen H, Liu P, Liu H, Guo W, Yi Y (1993). Observations on the ultra-thin sections of the main organs and tissues of haemorrhagic *Gobiocypris rarus* artificially infected by grass carp haemorrhagic virus (GCHV). *Acta Hydrobiologica Sinatica* 17: 343-346 (In Chinese, English abstract).

Ware SA (1980). A survey of pathogen survival during municipal solid waste and manure treatment processes. Municipal and Environmental Research Laboratory, Environmental Protection Agency, United States, 105 p.

Yan X, Wang Y, Xiong L, Jian J, Wu Z (2014). Phylogenetic analysis of newly isolated grass carp reovirus. *Springer Plus* 3: 190. (Published April 15, 2014) [Online] Available from http://www. doi: 10.1186/2193-1801-3-190 [Accessed 22 August 2018].

14. Grouper iridovirus

14.1. Hazard identification

14.1.1. Aetiological agent

Grouper iridovirus (GIV) is a double stranded DNA virus currently classified within the Genus *Ranavirus* (Xia *et al.* 2010) and grouped within the Family Iridoviridae (Piegu *et al*. 2014). It affects marine groupers of the Genus *Epinephelus*, causing grouper iridoviral disease, which is also known as sleepy grouper disease (OIE 2016a). It was first isolated from the greasy grouper (*Epinephelus tauvina*) in Chinese aquaculture (Xia *et al.* 2010).

While several GIV isolates have been identified, GIV is closely related to other ranaviruses infecting fish, including Singapore grouper iridovirus, orange spotted grouper iridovirus and Taiwan grouper iridovirus (Huang *et al*. 2011; OIE 2016a; DAFF 2014). Genomic studies indicate 100% compatibility with the megalocytoid red sea bream iridovirus (RSIV) (Chen *et al.* 2003; Razak *et al.* 2014). The status of this group is under review to determine whether these iridoviruses should be regarded as clades of RSIV (OIE 2016a).

14.1.2. OIE status

Grouper iridoviral disease is not notifiable to the OIE (OIE 2016a).

14.1.3. New Zealand status

GIV is considered exotic (Gias *et al*. 2011) and is not a notifiable organism in New Zealand (Anon. 2016). It is also related to dwarf gourami iridovirus, gourami iridovirus and infectious spleen and kidney necrosis virus commonly found in exotic ornamental fish (Hine & Diggles 2005; Jeong *et al.* 2008) but these have not been reported from New Zealand (Tubbs *et al.* 2007; Gias *et al.* 2011).

14.1.4. Epidemiology

GIV is an economically significant pathogen mainly affecting serranid groupers (*Epinephelus* spp.) in Asian aquaculture, including China, Singapore, Taiwan, Thailand, Vietnam, Indonesia and Malaysia (Nakajima 2003; Xia *et al.* 2010). Hosts include the greasy grouper (*Epinephelus coioides*) cultured in Chinese and Southeast Asian aquaculture (Huang *et al.* 2011), estuarine rock cod (*E. tauvina*), Malabar grouper (*E. malabaricus*) and yellow grouper (*E. awoara*). Iridoviruses are relatively non-host specific and other species may be experimentally infected (Chua *et al*. 1994; Chen *et al.* 2003; DAFF 2014). GIV is also reported from the Murray cod (*Maccullochella peelii*) in Australia (Go *et al.* 2005; DAFF 2014). Susceptible host families and species are summarised in Table 10.

Table 10. Families and Species of Fish Susceptible to Grouper Iridovirus (GIV)

Grouper Iridovirus (GIV)	
Family	**Host Species**
Percichthyidae	Murray cod (*Maccullochella peelii)*
Serranidae	Serranid grouper (*Epinephelus* sp.), greasy grouper (*E. tauvina*), Malabar grouper (*E. malabaricus*) orange spotted grouper (*E. coioides*), yellow grouper (*E. awoara*).

Iridoviruses of freshwater fish may infect marine fish hosts (Chen *et al*. 2003), while experimental cohabitation trials indicate the reverse infection pathway may also occur (Jeong *et al.* 2008).

Iridovirus transmission is horizontal and direct through the water, by cohabitation (Jeong *et al.* 2008), consumption of infected fish or tissues, or through cross-contamination of infected equipment (Yanong & Waltzek 2010). Vertical transmission has been suggested (Tubbs *et al.* 2007) but this is unproven (Yanong & Waltzek 2010). Juvenile fish are most susceptible, with mortalities in aquaculture of up to 100% occurring 3-4 months after placement in sea cages (DAFF 2014). Older fish are less susceptible, with mortality rates of up to 30% (Huang *et al.* 2011; Lio-Po & de la Pena 2012), while surviving fish are carriers of disease (Crane & Hyatt 2011; Yanong & Waltzek 2010; Nolan *et al.* 2015).

Infected fish may have external lesions, darkened skin and pale gills, but may show no external signs of infection (Tubbs *et al.* 2007; DAFF 2014). Necrosis and lesions of the spleen, kidney and myocardium, together with degeneration of the epithelial layer of the gill filaments are reported (Hanson *et al.* 2011; DAFF 2014).

Iridoviruses are resistant to the range of temperatures likely to be encountered in the human food consumption pathway, remaining viable after freezing at -10°C for 155 days (Plumb & Zilberg 1999), at -80°C for several years (OIE 2016b) and after long-term storage in water at 4°C (Tubbs *et al*. 2007). They are temperature labile, remaining viable for up to 30 minutes at 56°C (OIE 2016b).

Iridoviruses are inactivated by acid conditions (pH 3.0 and lower) and are sensitive to ether or chloroform treatments. They are inactivated by formalin (1% formalin at 37°C for 48 hours (Fan *et al.* 2011). GIV is susceptible to UV irradiation, and treatment with iodophor (7.5 ppm free iodine for 10 minutes) (Tendencia 2003) and chlorine (200 ppm for 2 hours) treatments (Langdon 1989). While formalin-inactivated vaccines are available (Kurita & Nakajima 2012), they are of limited duration and effectiveness due to the widespread practice of feeding whole baitfish as food in Asian aquaculture (Wang *et al.* 2007; Razak *et al.* 2014).

Species at risk in New Zealand include groupers and sea perches (Serranidae) found in northern New Zealand waters (Paul 2000). If GIV is considered as a megalocytivirus then potential host species include snapper[H] (*Pagrus auratus*), kingfish (*Seriola lalandi*), trevally (*Caranx georgianus*), as well as many other marine and freshwater species (Tubbs *et al.* 2007).

[H] The New Zealand snapper is classified in Family Sparidae, as *Pagrus* (=*Chrysophrys auratus*) (MPI 2013). For consistency it will subsequently be referred to as *Pagrus auratus* in this document.

14.2. Risk assessment

14.2.1. Entry assessment

Infection is mainly found in the spleen and kidney, so evisceration would substantially reduce viral titre. As GIV also occurs in gill tissues, which are retained after evisceration (Hanson *et al*. 2011), it may be present in the commodity. Iridoviruses are generally resistant to drying and freezing (Spikler *et al.* 2010) and are likely to remain viable even after extended frozen storage. The likelihood of entry is assessed as non-negligible.

14.2.2. Exposure assessment

To establish infection in New Zealand, infected eviscerated product would have to be consumed by a susceptible fish host, in sufficient quantity and duration (Kahn *et al.* 1999). Iridoviruses are resistant to freezing and drying and may remain viable in fish offal discarded after commercial fish processing (OIE 2016b). GIV may remain viable after dumping in landfill, although its potential transfer by scavenging birds is unknown. The likelihood of exposure to GIV is assessed as non-negligible.

14.2.3. Consequence assessment

Grouper iridovirus affects groupers (Lutjanidae), which occur rarely in northern New Zealand waters. However, GIV is a potential synonym for RSIV. The establishment of GIV/RSIV in New Zealand would have a major effect upon the inshore fisheries for several major New Zealand fish, including snapper (*Pagrus auratus*). This fishery alone was valued at $62 million in 2009 (Statistics New Zealand 2014). The introduction of GIV/RSIV could have significant impacts on the major recreational fisheries for these species, as well as significant economic effects upon a wide range of associated industries including retail sales and tourism. The consequence of the establishment of GIV in New Zealand through the live ornamental fish pathway was considered high to catastrophic (Hine & Diggles 2005). The consequence of exposure through the human food consumption pathway is assessed as non-negligible.

14.2.4. Risk estimation

Since the entry, exposure and consequence assessments for GIV/RSIV are non-negligible, the risk estimation is non-negligible. Therefore, GIV/RSIV is assessed to be a risk in the commodity and risk management measures may be justified.

14.3. Risk management

GIV has been assessed to be a risk in the commodity. Infection with GIV is a non-notifiable disease, but its status is uncertain, as it may be a clade of Red sea bream iridovirus, an OIE-listed disease. The OIE *Aquatic Code* (OIE 2016a) provides no specific guidance on mitigation measures for importing eviscerated fish from infected countries, or specific processing requirements that would ensure the destruction of the pathogen. It is assumed that compliance with the approved processed states for OIE-listed diseases (Appendix 3) would also eliminate GIV from the commodity and be a viable risk management option.

GIV is reported from two fish families (Table 10), which may be present in the commodity. While iridoviruses have low host specificity, species declaration should substantially reduce the pathogen load of GIV in the commodity and be a viable risk management option.

Grouper iridovirus is reported throughout Asia (China, Singapore, Taiwan, Thailand, Vietnam, Indonesia and Malaysia) and Australia, but no requirements for dedicated monitoring exist. Where country/zone freedom from GIV is approved through the MPI Country Approval Procedures, this option should substantially reduce the occurrence of grouper iridovirus in the commodity. Approval of a declaration of country/zone freedom by MPI is a viable risk management option.

GIV is found in wild Murray cod (*Maccullochella peelii*) (Percichthyidae) in Australia (DAFF 2014), but is predominantly associated with farmed serranid groupers (*Epinephelus* spp.) in Chinese and Southeast Asian aquaculture (Nakajima 2003; Xia *et al.* 2010). Restriction of the commodity to wild-caught fish (not from aquaculture) would moderately reduce pathogen load and be a viable risk management option.

Grouper iridovirus may be present in the head and gills, so removal of the gills should slightly reduce pathogen load. Removal of the head and gills should moderately reduce the pathogen load of GIV and be a viable risk management option.

As GIV is unaffected by freezing, frozen storage is not a viable risk management option. GIV is denatured by high-temperature treatment (to 56° C for at least 30 minutes). This would eliminate GIV from the commodity and be a viable risk management option.

Currently available vaccines are of limited effectiveness and application. Vaccination is not considered a viable option and is not considered further.

14.3.1. Risk management options

Grouper iridovirus (GIV) is reported from fish in families Percichthyidae and Serranidae (Table 10), which are considered likely to be present in the commodity. Other families have not been associated with GIV. Therefore species declaration indicating the commodity is not originated from either of the above families should substantially reduce the occurrence of GIV in the commodity.

For commodities originated from families associated with GIV, one or a combination of the following additional options could also be considered to effectively manage the risk.

Where country/zone freedom from GIV is accepted by MPI:

Option 1

Acceptance of country/zone freedom should substantially reduce the occurrence of GIV, so the commodity may be imported without any further restrictions.

Where country/zone freedom from GIV is not accepted by MPI, or not available:

Option 2

Processing to a state consistent with the approved states for OIE-listed diseases (Appendix 3) should eliminate GIV. When these provisions are met, the commodity could be imported without any further restrictions.

Where the imported commodity is not in a state consistent with the approved states for OIE-listed diseases (Appendix 3):

Option 3

Heat treatment (by cooking to at least 56°C for at least 30 minutes) should eliminate GIV. When this provision is met, the commodity could be imported without any further restrictions; or,

Option 4

Further processing (by removal of the head and gills) should moderately reduce the occurrence of GIV. When this provision is met, the commodity could be imported without any further restrictions; or,

Option 5

Restriction of the commodity to wild-caught fish (not from aquaculture) should moderately reduce the occurance of GIV. When this provision is met, the commodity could be imported without any further restrictions.

14.4. References

Anonymous (2016). Biosecurity (Notifiable Organisms) Order 2016 SR 2016/73. New Zealand Government, Wellington, New Zealand, 13 p.

Chen XH, Lin KB, Wang XW (2003). Outbreaks of an iridovirus disease in maricultured large yellow croaker, *Larimichthys crocea* (Richardson) in China. *Journal of Fish Diseases* 26(10): 615–619.

Chua FHC, Ng ML, Ng KL, Loo JJ, Wee JY (1994). Investigation of outbreaks of a novel disease, "Sleepy Grouper Disease" affecting the brown spotted grouper, *Epinephelus tauvina* Forskal. *Journal of Fish Diseases* 17: 417–427.

Crane M, Hyatt A (2011). Viruses of fish: An overview of significant pathogens. *Viruses* 3(11): 2025–2046

DAFF (2014). Grouper iridoviral disease. Aquatic animal diseases significant to Australia: Identification Field Guide, Fourth Edition. Department of Agriculture, Fisheries and Forestry, Canberra, Australia, 29 p.

Fan T, Xiuzhong H, Wang L, Geng X, Guojian J, Yang X, Yu M (2011). Development of an inactivated iridovirus vaccine against turbot reddish body syndrome. *Journal of the Ocean University of China* [Online]. Available from: http://link.springer.com/article/10.1007%2Fs11802-012-1852-7#page-2 [Accessed 28 August 2015].

Gias E, Johnston C, Keeling S, Spence RP, McDonald WL (2011). Development of real-time PCR assays for detection of megalocytiviruses in imported ornamental fish. *Journal of Fish Diseases* 34(8): 609–618.

Go J, Lancaster M, Deece K, Dhungyel O, Whittington R (2005). The molecular epidemiology of iridovirus in Murray cod (*Maccullochella peelii peelii*) and dwarf gourami (*Colisa lalia*) from distant biogeographical regions suggests a link between trade in ornamental fish and emerging iridoviral diseases. *Molecular and Cellular Probes* 20: 212–222.

Hanson L, Dishon A, Kotler M (2011). Herpesviruses that infect fish. *Viruses* 3(11): 2160–2191.

Hine PM, Diggles BK (2005). Import risk analysis: Ornamental fish. Biosecurity New Zealand, Ministry of Agriculture and Forestry, Wellington , New Zealand, 262 p.

Huang Y, Huang X, Yan Y, Cai J, Ouyang Z, Cui H, Wang P, Quin Q (2011). Transcriptome analysis of orange-spotted grouper (*Epinephelus coioides*) spleen in response to Singapore grouper iridovirus. *BMC Genomics* 12: 556.

Jeong JB, Cho HJ, Jun JL, Hong SH, Chung JK, Jeong HD (2008). Transmission of iridovirus from freshwater ornamental fish (pearl gourami) to marine fish (rock bream). *Diseases of Aquatic Organisms* 82: 27–36.

Kahn SA, Beers PT, Findlay VL, Peebles IR, Durham PJ, Wilson DW, Gerrity SE (1999). Import risk analysis on non-viable salmonids and non-salmonid marine finfish. Australian Quarantine and Inspection Service, Department of Agriculture, Fisheries and Forestry, Canberra, Australia, 409 p.

Kurita J, Nakajima K (2012). Megalocytiviruses. *Viruses* 4: 521–538.

Langdon JS (1989). Experimental transmission and pathogenicity of epizootic haematopoietic necrosis virus (EHNV) in redfin perch, *Perca fluviatilis* L., and 11 other teleosts. *Journal of Fish Diseases* 12: 295–310.

Lio-Po GD, de la Pena LD (2012). Viral diseases. pp 3–18. *In:* Nagasawa K, Cruz-Lacierda ER (*Eds.*). Diseases of Cultured Groupers. Southeast Asian Fisheries Development Centre, Aquaculture Department, Iloilo, Philippines, 81 p.

Nakajima K (2003). Grouper iridoviral disease – disease card developed to support the NACA/FAO/OIE regional quarterly aquatic animal disease (QAAD) reporting system in the Asia Pacific. NACA, Bangkok, Thailand, 4 p.

Nolan D, Stephens F, Crockford M, Jones JB, Snow M (2015). Detection and characterisation of viruses of the genus *Megalocytivirus* in ornamental fish imported into an Australian border quarantine premises: an emerging risk to national biosecurity. *Journal of Fish Diseases* 38: 187–195.

OIE (2016a). Aquatic Animal Health Code, 19th Edition. World Organization for Animal Health. Paris, France [Online]. Available from: http://www.oie.int/international-standard-setting/aquatic-code/access-online/ [Accessed 15 March 2017].

OIE (2016b). Manual of diagnostic tests for aquatic animals. World Organization for Animal Health. Paris, France [Online]. Available from: http://www.oie.int/international-standard-setting/aquatic-manual/access-online/ [Accessed 17 March 2017].

MPI (2013). NZ list of scientific names of fish. Animal Product (Specifications for products intended for human consumption) Notice 2004. Ministry for Primary Industries (MPI), Wellington, New Zealand. [Online] Available from: https://mpi.govt.nz/document-vault/963 [Accessed 11 August 2017].

Paul LJ (2000). New Zealand Fishes. Reed Publishing, Wellington, New Zealand, 253 p.

Piegu B, Asgari S, Bideshi D, Federici BA, Bigot Y (2014). Evolutionary relationships of iridoviruses and divergence of ascoviruses from invertebrate iridoviruses in the superfamily Megavirales. *Molecular Phylogenetics and Evolution* 84: 44–52.

Plumb JA, Zilberg D (1999). Survival of largemouth bass iridovirus in frozen fish. *Journal of Aquatic Animal Health* 11: 94–96.

Razak AA, Ransanagan J, Sade A (2014). First report of megalocytivirus (*Iridoviridae*) in grouper culture in Sabah, Malaysia. *International Journal of Current Microbiology and Applied Sciences* 3(3): 896–909.

Spikler AR, Roth JA, Gaylon J, Lofstedt JL (2010). Emerging and exotic diseases of animals. Fourth Edition. Centre for Food Safety and Public Health, College of Veterinary Medicine, Iowa State University, 383 p.

Statistics New Zealand (2014). New Zealand's commercial fish resource, 2009: snapper [Online] Available from: http://www.stats.govt.nz/browse_for_stats/environment/environmental-economic-accounts/fish-monetary-stock-account-1996-2009/results.aspx [Accessed 12 September 2014].

Tendencia EA (2003). Iodine disinfection of grouper *Epinephelus coioides* eggs. *Bulletin of the European Association of Fish Pathologists* 23(4): 191–196.

Tubbs L, Lee P, Diggles B, Jones JB, Sheppard M, Sim-Smith C (2007). A review of aquatic diseases of significance to New Zealand. Final research report for project ZBS 2005-17. National Institute of Water and Atmospheric Research, Wellington, New Zealand, 452 p.

Wang YQ, Lu L, Weng SP, Huang JN, Chan SM, He JG (2007). Molecular epidemiology and phylogenetic analysis of a marine fish infectious spleen and kidney necrosis virus like (ISKNV-like) virus. *Archives of Virology* 152: 763–773.

Xia L, Liang, H, Huang Y, Ou-Yang Z, Quin Q (2010). Identification and characterization of Singapore grouper iridovirus (SGIV) ORF162L, an immediate-early gene involved in cell growth control and viral replication. *Virus Research* 147(1): 30–39.

Yanong RP, Waltzek TB (2010). Megalocytivirus Infections in Fish, with Emphasis on Ornamental Species. Program in Fisheries and Aquatic Sciences FA182, 7 p. School of Forest Resources and Conservation, University of Florida, Gainseville, Florida, United States of America. [Online] Available from: http://edis.ifas.ufl.edu. [Accessed 11 August 2017].

15. Hirame rhabdovirus

15.1. Hazard identification

15.1.1. Aetiological agent

Hirame rhabdovirus (HIRRV) is a disease of cultured bastard halibut (*Paralichthys olivaceus*) and ayu (*Plecoglossus altivelis*) reported from Japan (Sindermann 1990). It was first isolated from bastard halibut in 1984 (Kimura *et al*. 1986) and is classified in the Genus *Novirhabdovirus*, within the Family Rhabdoviridae, which includes the OIE-listed diseases infectious haematopoietic necrosis (IHN), spring viraemia of carp (SVC) and viral haemorrhagic septicaemia (VHS) (OIE 2016b). Three isolates of HIRRV have been identified (Kurath 2012).

15.1.2. OIE status

Hirame rhabdovirus disease is not notifiable to the OIE (OIE 2016).

15.1.3. New Zealand status

HIRRV is considered exotic (Tubbs *et al.* 2007), since it has not been reported from New Zealand.

15.1.4. Epidemiology

HIRRV causes economically significant disease in bastard halibut (*P. olivaceus*) and ayu (*P. altivelis*) in marine aquaculture in Japan, Taiwan and China (Borzym *et al*. 2013). It is highly pathogenic, causing mortalities exceeding 90% in Japanese flounders (Sindermann 1990).

A wide range of fish families and species of fish (Table 11) are naturally susceptible to HIRRV (Kimura *et al*. 1989; Sun *et al.* 2010; *Borzym et al*. 2013). Clinical infection occurs in black sea bream (*Mylio macrocephalus=Acanthopagrus schlegelii*), red sea bream (*Chrysophrys major*), black rockfish (*Sebastes inermis*) (Sparidae), and stone flounder (*Kareius bicoloratus*) (Pleuronectidae) in Chinese marine waters (Kimura *et al.* 1986; Sun *et al.* 2010). The clouded blenny (*Opisthocentrus ocellatus*), alewife (bigeye herring) (*Alosa=Clupea pseudoharengus*) (Skall *et al.* 2005) and spotted seabass (*Lateolabrax maculatus*) are infected in Korean marine aquaculture with 10-12% cumulative mortality (Seo *et al.* 2016).

Table 11. Families and Species of Fish Susceptible to Hirame Rhabdovirus (HIRRV)

Family	Host Species
Clupeidae	Alewife (*Alosa* (*Clupea) pseudoharengus*)
Lateolabracidae	Japanese (spotted) seabass (*Lateolabrax japonicus*)
Paralichthyidae	Bastard halibut (*Paralichthys olivaceus*)
Plecoglossidae	Ayu (*Plecoglossus altivelis*)
Pleuronectidae	Stone flounder (*Kareius bicoloratus*)
Sebastidae	Darkbanded rockfish (*Sebastes inermis*)
Salmonidae	Brown trout (*Salmo trutta*), grayling (*Thymallus thymallus*)
Sparidae	Blackhead sea bream (*Acanthopagrus schlegelii*), red sea bream (*Chrysophrys major*)
Stichaeidae	Ocellated blenny (*Opisthocentrus ocellatus*)

HIRRV causes clinical disease in grayling (*Thymallus thymallus*) in Polish freshwater aquaculture, while brown trout (*Salmo trutta*) derived from the same farm were positive for HIRRV without clinical signs. Genomic analysis indicated the disease had been introduced with frozen food

imported from China originally intended for human consumption, but had been diverted for use as fish food in aquaculture (Borzym *et al.* 2014).

HIRRV causes high mortality in rainbow trout (*Oncorhynchus mykiss*), by experimental injection, but Coho salmon (*O. kisutch*), masu salmon (*O. masou*) and rainbow trout were not able to be infected by bath immersion (Kimura *et al*. 1989). These are not included as susceptible species.

External clinical signs include extensive haemorrhages of skin and prominent ascites (Sun *et al.* 2010), as well as extensive necrosis of the skeletal musculature, haematopoietic tissues and visceral organs (Daniels & Watanabe 2010; Sun *et al.* 2010). While broadly similar to infection with viral haemorrhagic septicaemia virus (VHSV), HIRRV infection results in more extensive haemorrhages of the skin, skeletal musculature and visceral organs (Daniels & Watanabe 2010). VHSV infection also occurs in brain tissue (OIE 2016b) and it is likely that HIRRV also occurs in brain tissue, but sampling protocols for PCR analysis generally examine pooled tissue samples (Sun *et al.* 2010). Disease is most acute in juvenile fish, while surviving older and larger fish may develop resistance and become carriers (Kurath 2012). Infection of *Lateolabrax maculatus* followed a similar internal pathology, but external signs were limited to a darkening of skin colour (Seo *et al.* 2016)

The optimal temperature for HIRRV is between 15 and 20°C (Oseko *et al.* 1988) with infection generally associated with temperatures below 18°C. In response, flounders are now routinely cultured at temperatures above 20°C in Japanese aquaculture (Leber *et al.* 2008; Daniels & Watanabe 2010).

HIRRV remains viable in sea water for up to 96 days (Kimura *et al*. 1986) and is resistant to heat, ether and acid (to pH 3) (Kimura *et al.* 1989). HIRRV survives for up to 2 years at -20°C (Hine & MacDiarmid 1997). Rhabdoviruses remain viable in frozen fish tissues after freezing, as well as in aquatic sediments for several months (Yoshimizu *et al*. 2005; Munro & Midtlyng 2011). The infective dose (LD_{50}), determined by injection for *L. maculatus* was $10^{4.95}$ $TCID_{50}$/mL (Seo *et al.* 2016).

HIRRV may be inactivated (99% reduction in infectivity) by chlorine treatment at 0.34 mg L^{-1} for 1 minute, by total residual oxidant concentration of 0.5 mg L^{-1} for 15 seconds, or 100 ppm iodophor at 15°C for 30 seconds (Yoshimizu *et al*. 2005). Rhabdoviruses are generally resistant to high temperatures, so it is likely that HIRRV would require a similar temperature range to IHNV (at least 65°C for 15 minutes) for inactivation (Stone *et al.* 1997).

Potential hosts in New Zealand include all salmonids (*Oncorhynchus* spp., *Salmo* spp, *Salvelinus* spp., as well as flatfish (Families Pleuronectidae and Bothidae), Clupeidae (Pacific herring, *Clupea* spp.) and Sparidae (snapper, *Sparus aurata*).

15.2. Risk assessment

15.2.1. Entry assessment

HIRRV has a wide host range (Table 11) including marine and freshwater fish, from Asian and European waters. Clinically infected fish generally exhibit extensive darkening and haemorrhaging of the skin, with prominent ascites and would be unlikely to pass inspection. However, surviving adults develop immunity and become carriers of disease (Daniels & Watanabe 2010; Sun *et al*. 2010; Kurath 2012). These fish may pass visual inspection and it is likely that HIRRV present in

the skin and skeletal musculature would be retained in the commodity, or be present in the blood-water discharge from factory processing (Daniels & Watanabe 2010). HIRRV is resistant to freezing and chilled storage and likely to remain viable in the commodity (Hine & MacDiarmid 1997; Yoshimura *et al*. 2005).

The likelihood of entry of HIRRV through the commodity is assessed as non-negligible.

15.2.2. Exposure assessment

To establish infection through the commodity, infected eviscerated product would have to become available for consumption by a susceptible fish host, in sufficient quantity and duration (Kahn *et al*. 1999). HIRRV remains viable in frozen fish tissues and sufficient infected product may be present in the human consumption pathway to initiate and maintain an infection through infected wastewater and offal discharged through factory wastewater and offal.

Infection of Bastard halibut (*P. olivaceus*) (Oseko *et al.* 1998) occurs at a similar range of water temperatures (5-15°C) to that commonly found in New Zealand waters and farmed fish can be infected when fed frozen fish tissues directly as fish feed (Borzym *et al.* 2013).

HIRRV remains viable in seawater for long periods (up to 96 days) Kimura *et al.* 1986) and survives in marine sediments for several months (Yoshimizu *et al*. 2005; Munro & Midtlyng 2011). Infective dose (LD_{50}) for *L. maculatus* was $10^{4.95}$ $TCID_{50}$/mL (Seo *et al* 2016). A range of coastal inshore marine fish, including sea cage salmonids, snapper and flatfish are susceptible to HIRRV.

The likelihood of exposure to HIRRV through the commodity is assessed as non-negligible.

15.2.3. Consequence assessment

HIRRV is an exotic rhabdovirus with a wide host range including several fish of economic importance in marine and freshwater. These include sea-cage salmonids, so introduction into New Zealand would have a significant effect on the New Zealand salmonid aquaculture industry, worth in excess of $60 million in direct exports (Aquaculture New Zealand 2014). HIRRV also affects rainbow trout (*O. mykiss*) in freshwater, so an outbreak may affect recreational and tourist trout and salmon fishing, and incur significant social and environmental costs associated with other domestic fisheries (Fish & Game 2014). While the dollar value of New Zealand's freshwater fisheries is not fully known, the Lake Taupo trout fishery alone is valued at $70 to 80 million (Marsh & Mkwra 2013).

Susceptible marine fish include the developing aquaculture industry for grouper (*Polyprion oxygenios*) and snapper (*Sparus aurata*) (Smith & Taylor 1982; NIWA 2014; Plant & Food 2017). These species also support significant coastal inshore fisheries, where the snapper export fishery was valued at $62 million in 2009 (Statistics New Zealand 2014). The reduction in key inshore recreational fishery target species such as flounder (Families Pleuronectidae and Bothidae), grouper and snapper would incur significant social effects (MPI 2014).

The consequence of establishment of HIRRV is assessed as non-negligible.

15.2.4. Risk estimation

Since the entry, exposure and consequence assessments for HIRRV are non-negligible, the risk estimate is non-negligible. Under the procedures followed in this risk assessment, HIRRV is assessed to be a risk in the commodity and risk management measures may be developed.

15.3. Risk management

HIRRV is assessed to be a risk in the commodity. Infection with HIRRV is not notifiable to the OIE, so the OIE Aquatic Code (OIE 2016a) does not provide guidance on measures that would ensure the destruction of the virus. It is assumed that compliance with the approved processed states for OIE-listed diseases (Appendix 3) would also eliminate HIRRV from the commodity and be a viable risk management option.

HIRRV occurs in several families of wild and farmed fish (Table 11), which may be present in the commodity. Species declaration should substantially reduce the pathogen load of HIRRV and be a viable risk management option. Restriction of the commodity to wild-caught fish (not from aquaculture) should have little, or no effect on pathogen load and is not a viable risk management option.

HIRRV is reported from China, Japan, Poland, South Korea and Taiwan (Skall *et al.* 2015; Seo *et al.* 2016), but no requirements for dedicated monitoring exist. Where declaration of country/zone freedom from HIRRV is approved through the MPI Country Approval Procedures, this option should substantially reduce the occurrence of HIRRV in the commodity. Approval of country/zone freedom by MPI is a viable risk management option.

Infection with HIRRV is commonly associated with the head, skin and skeletal musculature in acute and chronic infection and may be concentrated in the brain and neural tissues (Daniels & Watanabe 2010; Sun *et al.* 2010). Removal of the gills, or the head and gills, would slightly reduce the pathogen load of HIRRV. Further processing to the skin-off fillet state should moderately reduce pathogen load in the commodity and be a viable risk management option.

Rhabdoviruses are unaffected by freezing, so frozen storage is not a viable risk management option. If it is assumed that HIRRV is likely to require a similar temperature range for inactivation as for IHNV (at least 65°C for 15 minutes) (Stone *et al.* 1997), this heat treatment should eliminate HIRRV from the commodity. Heat treatment may be a viable risk management option.

15.3.1. Risk management options

Hirame rhabdovirus (HIRRV) is reported from fish in families Clupeidae, Lateolabracidae, Paralichthyidae, Plecoglossidae, Pleuronectidae, Salmonidae, Sebastidae, Sparidae and Stichaeidae (Table 11), which are considered likely to be present in the commodity. Other families have not been associated with HIRRV. Therefore species declaration indicating the commodity is not originated from any of the above families should substantially reduce the occurrence of HIRRV in the commodity.

For the commodities originated from families associated with HIRRV, one or a combination of the following additional options could also be considered to effectively manage the risk.

Where country/zone freedom from HIRRV is accepted by MPI:

Option 1

Acceptance of country/zone freedom should substantially reduce the occurrence of HIRRV, so the commodity may be imported without any further restrictions.

Where country/zone freedom from HIRRV is not accepted by MPI or not available:

Option 2

Processing to a state consistent with the approved states for OIE-listed diseases (Appendix 3) should eliminate HIRRV. When these provisions are met, the commodity could be imported without any further restrictions.

Where the imported commodity is not in a state consistent with the approved states for OIE-listed diseases (Appendix 3):

Option 3

Heat treatment (by cooking to at least 65°C for at least 15 minutes) should eliminate HIRRV. When this provision is met, the commodity could be imported without any further restrictions; or,

Option 4

Further processing (to the skin-off fillet state) should moderately reduce the occurrence of HIRRV. When this provision is met, the commodity could be imported without any further restrictions.

15.4. References

Anonymous (2016). Biosecurity (Notifiable Organisms) Order 2016. NZLR 2016/73. New Zealand Government, Wellington, New Zealand, 13 p.

Aquaculture New Zealand (2014). New Zealand's aquaculture industry key facts 2013 [Online] Available from: http://www.seafoodnewzealand.org.nz/our-industry/key-facts/ [Accessed 26 September 2014].

Borzym E, Matras M, Maj-Paluch J, Baud M, De Boisséson C, Talbi C, Olesen NJ, Bigarré L (2014). First isolation of hirame rhabdovirus from freshwater fish in Europe. *Journal of Fish Diseases* 37: 423–430.

Daniels HV, Watanabe WO (2010). Practical flatfish culture and stock enhancement. John Wiley, New York, United States of America, 388 p.

Fish and Game (2014). New Zealand Fish and Game Council [Online]. Available from: http://www.fishandgame.org.nz/ [Accessed 24 September 2014].

Hine M, MacDiarmid SC (1997). Contamination of fish products: Risks and prevention. *OIE Reviews in Science and Technology* 16(1): 135–145.

Kahn SA, Beers PT, Findlay VL, Peebles IR, Durham PJ, Wilson DW, Gerrity SE (1999). Import risk analysis on non-viable salmonids and non-salmonid marine finfish. Australian Quarantine and Inspection Service, Department of Agriculture, Fisheries and Forestry, Canberra, Australia, 409 p.

Kimura T, Yoshimizu M, Gorie S (1986). A new rhabdovirus isolated in Japan from cultured hirame (Japanese flounder, *Paralichthys olivaceus*) and ayu (*Plecoglossus altivelis*). *Diseases of Aquatic Organisms* 1: 209–217.

Kimura T, Yohsimizu M, Oseko N, Nishizawa T (1989). Rhabdovirus olivaceus (*Hirame rhabdovirus*), pp 388–390 *In*: Ahne HW, Kurstak E (*Eds.*). Viruses of Lower Vertebrates. Springer-Verlag, Berlin, 518 p.

Kurath G (2012). Fish norhabdoviruses, pp 89–116 *In*: Dietzgen RG, Kuzmin IV (*Eds.*). Rhabdoviruses: Molecular taxonomy, evolution, genomics, ecology, host-vector interactions, cytopathology and control. Horizon Scientific Press, Linton, United Kingdom, 276 p.

Leber LM, Kitada S, Blankenship HL, Svansard T (2008). Stock enhancement and sea ranching: Developments, pitfalls and opportunities. John Wiley, New York, United States of America, 584 p.

Marsh D, Mkwra L (2013). Review of freshwater non-market value studies. Department of Economics, University of Waikato, Hamilton, New Zealand, 144 p.

Ministry for Primary Industries (MPI) (2014). Fisheries Assessment Plenary, May 2014: Stock assessments and stock status. Compiled by the Fisheries Science Group, Ministry for Primary Industries, Wellington, New Zealand, 1381 p.

Munro ES, Midtyling PJ (2011). Infectious pancreatic necrosis virus and associated aquatic birnaviruses, pp 1–65 *In:* Woo PTK, Bruno DW (*Eds.*). Fish diseases and disorders, Volume 3: Viral Bacterial and fungal infections.CABI International, Wallingford, United Kingdom. 930 p.

NIWA (2017). Aquaculture species: Hapuku. Aquaculture. National Institute for Water and Atmospheric Research Ltd (NIWA), Auckland, New Zealand. [Online] Available from: https://www.niwa.co.nz/aquaculture [Accessed 15 August 2017].

OIE (2016). Aquatic Animal Health Code, 19th Edition. World Organization for Animal Health. Paris, France [Online] Available from: http://www.oie.int/international-standard-setting/aquatic-code/access-online/ [Accessed 15 March 2017].

Oseko N, Yoshimizu M, Kimura T (1988). Effect of water temperature on artificial infection of *Rhabdovirus olivaceus* (*Hirame rhabdovirus*: HRV) to Hirame (Japanese flounder, *Paralichthys olivaceus*). *Fish Pathology* 23: 125–132.

Plant and Food (2017). Snapper aquaculture. Rural Delivery, June 2017. Plant and Food Research, Nelson, New Zealand [Online] Available from: https://www.ruraldelivery.net.nz/stories/Snapper-Aquaculture [Accessed 15 August 2017].

Sindermann CJ (1990). Principal diseases of marine fish and shellfish: Chapter 1: Diseases of marine fish caused by microbial pathogens. Gulf Professional, Elsevier, New York, 521 p.

Smith PJ, Taylor JL (1982). Prospects for snapper farming and reseeding in New Zealand. *Fisheries Research Division Occasional Publication* No. 37, 56 p.

Seo HG, Do JW, Jung SH, Han HJ (2016). Outbreak of hirame rhabdovirus infection in cultured spotted sea bass *Lateolabrax maculatus* on the western coast of Korea. *Journal of Fish Diseases* 39: 1239–1246.

Skall HF, Olesen NJ. Mellergaard S. (2005) Prevalence of viral haemorrhagic septicaemia virus in Danish marine fishes and its occurrence in new host species. *Diseases of Aquatic Organisms* 66, 145–151.

Statistics New Zealand (2014). Fish monetary stock account: snapper (*Pagrus auratus*) [Online] Available from: http://www.stats.govt.nz/browse_for_stats/environment/environmental-economic-accounts/fish-monetary-stock-account-1996-2009/results.aspx#snapper [Accessed 10 September 2014].

Stone, MAB, MacDiarmid SC, Pharo HJ (1997). Import health risk analysis: salmonids for human consumption. Ministry of Agriculture Regulatory Authority, Wellington, New Zealand, 269 p.

Sun Y, Yue Z, Liu H, Zhao Y, Liang C, Shi X, Wu B, Xu B, Deng M, Zhu L, Wang Z (2010). Development and evaluation of a sensitive and quantitative assay for hirame rhabdovirus based on quantitative real-time RT-PCR. *Journal of Virological Methods* 169: 391–396.

Tubbs L, Lee P, Diggles B, Jones JB, Sheppard M, Sim-Smith C (2007). A review of aquatic diseases of significance to New Zealand. Final research report for project ZBS 2005-17. National Institute of Water and Atmospheric Research, Wellington, New Zealand, 452 p.

Yoshimizu M, Yoshinaka T, Hatori S, Kasai H (2005). Survivability of fish pathogenic viruses in environmental water and the inactivation of fish viruses. *Bulletin of the Fisheries Research Agency of Japan*, Supplement 2: 47–54.

16. Infectious haematopoietic necrosis virus

16.1. Hazard identification

16.1.1. Aetiological agent

Infectious haematopoietic necrosis virus (IHNV) was first identified from sockeye salmon (*Oncorhynchus nerka*) on the west coast of the United States (Rucker *et al.* 1953) and isolated by Wingfield *et al.* (1969). It is a single stranded RNA virus established as the type species of the Genus *Novirhabdovirus*, classified within the Family Rhabdoviridae (Bootland & Leong 2011; OIE 2016a). It is considered distinct from other fish rhabdoviruses, such as viral haemorrhagic septicaemia virus (VHSV) (OIE 2016b).

Although only one serotype of IHNV has been identified (Bootland & Leong 2011), several genogroups have been described that vary widely in pathogenicity. These appear to be geographically based rather than being determined by host specificity (Bootland & Leong 2011; OIE 2016b).

16.1.2. OIE status

Infection with IHNV is listed by the OIE as a notifiable disease (OIE 2016a).

16.1.3. New Zealand status

IHNV has not been reported from New Zealand (Boustead *et al.* 1993; Stone *et al*. 1997). It is considered an exotic notifiable disease (Anon. 2016).

16.1.4. Epidemiology

IHNV is a major disease of economic importance primarily affecting juvenile salmonids, although fish of all ages may be infected. It affects farmed and wild salmonids in marine and fresh waters of the Pacific and Northwest United States, Asia (China, Japan, South Korea and Taiwan) and South America (Bolivia, Chile), as well as from Europe (Austria, Croatia, Czech Republic, France, Germany, Italy, Netherlands, Poland, Slovenia, Spain), Iran, Kuwait, Russia and Pakistan (Tubbs *et al.* 2007: OIE 2016b).

Susceptible fish (Table 12) include the Salmonidae, including the mountain whitefish (*Prosopium williamsoni*) as well as marine and freshwater fish in the families Acipenseridae, Anguillidae, Clupeidae, Gasterosteidae, Embiotocidae, Esocidae, Gadidae, Moronidae, Salmonidae, Scophthalmidae, and Sparidae (Kahn *et al.* 1999; Bootland & Leong 2011; OIE 2016a; Dhar *et al.* 2016, Dixon *et al.* 2016).

It is also reported from invertebrates including mayflies (*Callibaetis* spp.) salmon leeches (*Piscicola* spp.) and parasitic copepods of salmon (*Salmonicola* spp.) (Bootland & Leong 2011; OIE 2016b).

Table 12. Families and Species of Fish Susceptible to Infectious Haematopoietic Necrosis Virus (IHNV)

Infectious Haematopoietic Necrosis Virus (IHNV)	
Family	**Host Species**
Acipenseridae	White sturgeon (*Acipenser transmontanus*)
Anguillidae	European eel (*Anguilla anguilla*)
Clupeidae	Pacific herring (*Clupea pallasii*),
Gasterosteidae	Tube-snout stickleback (*Aulorhynchus flavidus*)
Embiotocidae	Pile perch (*Rhacochilus* (*Damalichthys*) *vacca*), shiner perch (*Cymatogaster aggregata*)
Esocidae	Northern pike (*Esox lucius*)
Gadidae	Pacific cod (*Gadus macrocephalus*)
Moronidae	Sea bass (*Morone labrax*)
Salmonidae	Amago salmon (*Oncorhynchus masou rhodurus*), Atlantic salmon (*Salmo salar*), brown trout (*Salmo trutta*), char (*Salvelinus namaycush*, *S. alpinus*, *S. fontinalis*, *S. leucomaensis*), Chinook salmon (*O. tshawytscha*), Coho salmon (*O. kisutch*), chum salmon (*O. keta*), cutthroat trout (*O. clarkii*), rainbow trout (*O. mykiss*), masu salmon (*O. masou*), mountain whitefish (*Prosopium williamsoni*), sockeye salmon (*O. nerka*), yamame salmon (*O. masou ishikawae*),
Scophthalmidae	Turbot (*Scophthalmus maximus*)
Sparidae	Gilthead sea bream (*Sparus aurata*)

Clinically infected fish usually show visible external lesions and darkening of the skin, although mortality in salmonids may occur in the absence of clinical signs. Infected fish commonly have petechial haemorrhages at the base of the fins, vent, gills and mouth. The major sites of infection are the haematopoietic tissues of the liver, spleen and kidney, which appear pale and anaemic (Munro & Midtlyng 2011), although viral particles may also be present in brain tissue (Samuelsen *et al*. 2006).

The infectiveness of IHNV isolates varies widely among species (Bootland & Leong 2011). While IHNV isolated from eels (*Anguilla* spp.) remains infective to salmonids (Bergmann *et al.* 2002), isolates from other species, such as herring, are not infective to salmon under laboratory conditions (Hart *et al.* 2011).

Mortality also varies widely between viral strains, host species and environmental factors. While mortality rates in hatcheries may reach 90% (Munro & Midtlyng 2011), mortality in older fish is usually less than 50%. Surviving fish acquire immunity to further infection when transferred to sea cages, but this immunity is temperature related. While these fish may act as carriers, IHNV may also enter a latent state in infected fish, to emerge later if the host is compromised by stress (Bootland & Leong 2011).

Transmission may be vertical (OIE 2016b), but is generally horizontal from fish to fish through the water column. Infection occurs through epidermal cells in the gills, skin, fin bases, oral region, or through the digestive system (Stone *et al*. 1997; Bootland & Leong 2011). Initial viral replication occurs in the fin bases, gills and skin, before progressing to the characteristic widespread degenerative necrosis of the haematopoietic tissues, spleen, liver, kidney and brain tissues (Munro & Midtlyng 2011).

IHNV is extremely environmentally stable and unaffected by salinity, remaining viable for at least 15 days in fresh water at 5°C (Traxler *et al.* 1993), although natural infections usually occur in spring, when water temperatures are lower than 15°C (Stone *et al.* 1997). IHNV does not remain viable in freshwater streams during winter, but may survive for several months in river sediments (Stone *et al.* 1997). It remains viable after passage through the avian digestive system and may be transported between waterways by piscivorous birds (Wang *et al.* 2007).

While IHNV survives for up to 14 days in sea or estuarine water (Stone *et al.* 1997), little is known about the transmission of IHNV in sea water. The existence of a yet-unidentified marine reservoir of infection for sockeye salmon between Russia and the USA has been suggested (OIE 2016b), which may include parasitic copepods (Bootland & Leong 2011) or other fish (Kent *et al.* 1998).

IHNV is highly infective at relatively low viral concentrations of 10^3 pfu/ml (plaque forming units) (Traxler *et al.* 1993; Bootland & Leong 2011). IHNV remains viable at high titres in fish blood and salmonid gill issues retained in the eviscerated product (Stone *et al.* 1997; Fraser *et al.* 2006).

IHNV may be present in wash water discharged into waterways, and in offal discarded after fish processing. It is resistant to the environmental conditions normally encountered in fish processing and storage. It requires high temperatures for thermal inactivation (65°C for 15 minutes, or 82°C for 5 minutes) and is resistant to freezing when associated with serum or high levels of dissolved protein, such as in fish tissues. It has high resistance to freeze-thaw cycles and may persist for several years at -20°C (Stone *et al*. 1997; Tubbs *et al*. 2007).

IHNV may be inactivated by fish ensilage processing at pH 4 (Whipple & Rohovec 1994), ozone treatment or UV treatment at 3.84 mJ cm^{-2} (Afonso *et al*. 2012). Iodophor (100 mg L^{-1}) or chlorine (50 mg L^{-1}) for at least 15 sec has been shown effective for disinfection of fish eggs (Bovo *et al*. 2005; OIE 2016a), but treatments may be ineffective when high levels of organic material are present in the effluent discharge (Fraser *et al.* 2006). Contaminated organic material should be disposed to landfill as a high-risk waste product (Fraser *et al.* 2006), as IHNV may be transferred by other vectors including birds, animals and possibly insects (Stone *et al.* 1997).

Inactivated virus vaccines have been developed for IHNV, but they are of short-term duration and are inconsistent in operation (Anderson *et al*. 2008; Alonso & Leong 2013).). DNA vaccines have been licenced for use in China (Xu *et al.* 2017), the EU (EMA 2016; Houston *et al.* 2017), the United States and Canada (OIE 2016b; CFIA 2017, Dadar *et al.* 2017), where they can be administered by infection. Vaccines have not been licenced in other countries where their application to smaller fish requires further research (Alonso & Leong 2013).

Potential fish hosts in New Zealand include all salmonids and a wide range of other marine, estuarine and freshwater fish (Tubbs *et al.* 2007).

16.2. Risk assessment

16.2.1. Entry assessment

IHNV has not been previously associated with non-viable fresh or frozen product through the human food consumption pathway (Bootland & Leong 2011). Fish clinically infected with IHNV usually show external skin lesions (Samuelsen *et al.* 2006) and would not pass visual inspection. However, surviving adult carrier fish may pass visual inspection and be present in the commodity.

Evisceration would be expected to significantly reduce viral titre, but IHNV is infective even at low concentrations (Bootland & Leong 2011). IHNV may be associated with the brain and gill tissues of the head, which is retained, in the eviscerated commodity, as well as in the blood-water discharge from factory processing (Stone *et al*. 1997; Fraser *et al*. 2006).

The likelihood of entry is assessed as non-negligible.

16.2.2. Exposure assessment

To establish infection in New Zealand, infected eviscerated product would have to be consumed by a susceptible fish or invertebrate host, in sufficient quantity and duration. IHNV is resistant to freezing and drying and may remain viable for several months in river sediments (Kahn *et al.* 1999). It is infective at low concentrations (Bootland & Leong 2011) and infects a wide range of fish and invertebrate hosts. It is likely to survive for long periods in fish offal discarded in commercial food processing (Bootland & Leong 2011) and may be transferred by scavenging birds (Fraser *et al*. 2006).

The likelihood of exposure to IHNV is assessed as non-negligible.

16.2.3. Consequence assessment

IHNV is a major disease of salmonids, so introduction into New Zealand would have a significant effect on the New Zealand salmonid aquaculture industry, worth in excess of $60 million in direct exports (Aquaculture New Zealand 2014). Other economic and social consequences include the effects on industries associated with recreational trout and salmonid farming. The Lake Taupo trout fishery and its associated tourist industry alone is valued at $70 to 80 million (Fish & Game 2014).

The consequence of establishment is assessed as non-negligible.

16.2.4. Risk estimation

Since the entry, exposure and consequence assessments for IHNV are non-negligible, the risk estimation is non-negligible. Therefore, IHNV is assessed to be a risk in the commodity and risk management measures may be justified.

16.3. Risk management

IHNV has been assessed to be a risk in the commodity and the OIE *Aquatic Code* (OIE 2016a) provides guidance for importing eviscerated fish from infected countries and the specific processing requirements that would ensure destruction of the virus.

IHNV has generally low host specificity. Over 10 marine and freshwater fish families are considered susceptible (Table 12), but fish of family Gasterosteidae are considered unlikely to be present in the commodity. Species declaration should substantially reduce the occurrence of IHNV in the commodity and be a viable risk management option.

Article 10.6.3 of the OIE *Aquatic Code* (OIE 2016a) states that:

Competent Authorities should not require any conditions related to IHN, regardless of the IHN status of the exporting country, zone or compartment, when authorising the importation or transit of the following aquatic animals and aquatic animal products from the species referred to in Article 10.6.2. which are intended for any purpose and which comply with Article 5.4.1.:

> *a. heat sterilised, hermetically sealed fish products (i.e. a heat treatment at 121°C for at least 3.6 minutes or any time/temperature equivalent);*

b. pasteurised fish products that have been subjected to a heat treatment at 90°C for at least ten minutes (or any time/temperature equivalent which has been demonstrated to inactivate IHNV);
c. mechanically dried, eviscerated fish (i.e. a heat treatment at 100°C for at least 30 minutes or any time/temperature equivalent which has been demonstrated to inactivate IHNV).

Furthermore, Article 10.6.11 states that:

Competent Authorities should not require any conditions related to IHN, regardless of the IHN status of the exporting country, zone or compartment, when authorising the importation or transit of fish fillets or steaks (frozen or chilled) which have been prepared and packaged for retail trade and which comply with Article 5.4.2.

Compliance with Articles 10.6.3 and 10.6.11would eliminate IHNV from the commodity and be a viable risk management option.

IHNV has a worldwide distribution. IHN is an OIE listed disease affecting wild and farmed fish, Approval of a declaration of country/zone freedom through the MPI Country Approval Procedures should substantially reduce the occurrence of IHNV in the commodity and be a viable risk management option. Restriction of the commodity to wild-caught fish (not from aquaculture) would have little effect on pathogen load and is not a viable risk management option.

IHNV is mainly associated with the gill and brain tissue (Stone *et al*. 1997; Bootland & Leong 2011). Removal of the gills should slightly reduce the pathogen load of IHNV in the commodity, while removal of head and gills should moderately reduce pathogen load and be a viable risk management option.

IHNV is resistant to freezing (Stone *et al.* 1997), so frozen storage is not a viable risk management option. It requires high temperatures (to 65°C for at least 15 minutes) for inactivation (Whipple & Rohovec 1994; Stone *et al*. 1997). High-temperature treatment would eliminate IHNV from the commodity and be a viable risk management option.

While DNA vaccines are licenced for the EU, USA, Canada and China (Xu *et al*. 2017), no safe, commercially available vaccine against IHNV is commercially available in other regions (Alonso & Leong 2013). Vaccination is not considered a viable risk management option and is not considered further.

IHNV may be transmitted through the waste products associated with transport, storage and processing of the commodity. The requirement that all wash and wastewater discharges be appropriately chemically treated (e.g., with iodophors) before discharge, and that all solid wastes, tissue scraps and offal be disposed of through a recognised trade waste disposal procedure would be viable management options.

16.3.1. Risk management options

The IHNV virus is reported from fish in families Acipenseridae, Anguillidae, Clupeidae, Embiotocidae, Esocidae, Gadidae, Moronidae, Salmonidae, Scophthalmidae and Sparidae (Table 12), which may be present in the commodity. Other families have not been associated with IHNV. Therefore species declaration indicating the commodity is not originated from any of the above families should substantially reduce the occurrence of IHNV in the commodity.

For the commodities originated from families associated with IHNV, one or a combination of the following additional options could also be considered to effectively manage the risk.

Where country/zone freedom from IHNV is accepted by MPI:

Option 1

Acceptance of country/zone freedom should substantially reduce the occurrence of IHNV, so the commodity may be imported without any further restrictions.

Where country/zone freedom from IHNV is not accepted by MPI or not available:

Option 2

Processing consistent with the conditions of Article 10.6.3 or 10.6.11 of the OIE *Aquatic Code* (OIE 2016a) should eliminate IHNV. Where these provisions are met, the commodity could be imported without any further restrictions.

Where the imported commodity is not consistent with Article 10.6.3 or 10.6.11 of the OIE Aquatic *Code* (OIE 2016a), further processing is necessary:

Option 3

Heat treatment (by cooking to at least 65°C for at least 15 minutes) should eliminate IHNV. Where this provision is met, the commodity could be imported without any further restrictions; or,

Option 4

Further processing (by removal of the head and gills) should moderately reduce the occurrence of IHNV. Where this provision is met, the commodity could be imported without any further restrictions.

16.4. References

Afonso LOB, Richmond Z, Eaves AA, Richard J, Hawley LM, Garver KA (2012). Use of ultraviolet C radiation (UVC) to inactivate infectious hematopoietic necrosis virus (IHNV) and viral haemorrhagic septicaemia virus (VHSV) in fish processing plant effluent. *Journal of Aquaculture Research and Development* 3: 120. doi: 10.4172/2155-9546.1000120 [Online]. Available from: http://omicsonline.org/use-of-ultraviolet-c-uvc-radiation-to-inactivate-infectious-hematopoietic-necrosis-virus-ihnv-and-viral-hemorrhagic-septicemia-virus-vhsv-in%20fish-processing-plant-effluent-2155-9546.1000120.php?aid=3943 [Accessed 24 April 2015].

Alonso M, Leong JA (2013). Licenced DNA vaccines against infectious haematopoietic necrosis virus (IHNV). *Recent Patents in DNA Genetic Sequences* 1-7(1): 62.

Anderson E, Clouthier S, Shewmaker W, Weighall A, LaPatra S (2008). Inactivated infectious haematopoietic necrosis virus (IHNV) vaccines. *Journal of Fish Diseases* 30(10): 729–745.

Anonymous (2010). Biosecurity (Notifiable Organisms) Order 2010. New Zealand Government, Wellington, New Zealand, 13 p.

Aquaculture New Zealand (2014). New Zealand's aquaculture industry key facts [Online]. Available from: http://www.seafoodnewzealand.org.nz/our-industry/key-facts/ [Accessed 26 September 2014].

Bergmann SM, Fichtner D, Skall HF, Schlotfeldt HJ, Olesen NJ (2002). Age- and weight-dependent susceptibility of rainbow trout *Oncorhynchus mykiss* to isolates of infectious haematopoietic necrosis virus (IHNV) of varying virulence. *Diseases of Aquatic Organisms* 55: 205–210.

Bootland LM, Leong JC (2011). Infectious haematopoietic necrosis. pp 66–109. *In:* Woo PTK, Bruno DW (*Eds.*). *Fish Diseases and Disorders: Volume 3 - Viral, Bacterial and Fungal Infections*, 2nd edition. CABI International, Wallingford, United Kingdom, 930 p.

Boustead NC, Meyers TR, Short S (1993). Absence of infectious haematopoietic necrosis virus (IHNV) in New Zealand sockeye salmon, *Oncorhynchus nerka*. *New Zealand Journal of Marine and Freshwater Research* 27: 55–60.

Bovo B, Hill B, Husby A, Hastein T, Michel C, Olesen NJ, Storset A, Midtlyng PJ (2005). Pathogen survival outside the host, and susceptibility to infection. Work Package 3 report, Veso Project 1601. European Commission. Brussels, Belgium, 41 p.

CFIA (2017). List of veterinary biologics licenced in Canada. Canadian Food Inspection Agency [Online] Available from: http://www.inspection.gc.ca/active/scripts/database/vetbio_submit.asp?lang=e&species=9&manufacturer=all [Accessed 14 August 2017].

Dadar M, Dharma K, Vakharia VN, Hoseinfar SH, Karthik K, Tiwari R, Khandia R, Munjal A, Salgado-Miranda C, Joshi SK. (2016). Advances in aquaculture vaccines against fish pathogens: Global status and current trends. *Reviews in Fisheries Science & Aquaculture* 25(3): 184–217.

Dhar Ak, Sikisala RR, Orry A, Allnut FCT (2016). Infectious haematopoietic necrosis viris. p 186 *In*: Muir M (*Ed*.). Mononeaviruses of veterinary importance: Molecular epidemiology and control. CABI International, Wallingford, United Kingdom, 216 p.

Dixon P, Paley R, Alegri-Moran R, Oidtmann B (2016). Epidemiological characteristics of infectious hematopoietic necrosis virus (IHNV): a review. *Veterinary Research* 47(63): 1–26 [Online] Available from: http://www. doi 10.1186/s13567-016-0341-1 [Accessed 8 June 2018].

EMA (2016). First DNA vaccine in the EU recommended for use in salmon. European Medicines Association (EMA). [Online] Available from: http://www.ema.europa.eu/ema/index.jsp?curl=pages/news_and_events/news/2016/04/news_detail_002516.jsp&mid=WC0b01ac058004d5c1 [Accessed 14 August 2017].

Fish and Game (2014). New Zealand Fish and Game Council [Online]. Available from: http://www.fishandgame.org.nz/ [Accessed 24 September 2014].

Fraser DI, Munro PD, Smail DA (2006). Disinfection guide version IV: practical steps to prevent the introduction and minimise the transmission of diseases of fish. Fisheries Research Services Internal Report 13/06. Marine Laboratory, Aberdeen, Scotland, United Kingdom, 14 p.

Hart LM, Traxler GS, Garver KA, Gregg RJ, Grady CA, Kuranth G, Hershberger PK (2011). Larval and juvenile Pacific herring *Clupea pallasii* are not susceptible to infectious haematopoietic necrosis virus under laboratory conditions. *Diseases of Aquatic Organisms* 93(2): 105–110.

Houston R, Moxon S, Nogue F, Papadopoulou N, Ramon M, Waigmann E (2017). Assessment of the potential integration of the DNA plasmid vaccine CLYNAV into the salmon genome. *The EFSA Journal* 15(1): 4689 [Online] Available from: (http://onlinelibrary.wiley.com/doi/10.2903/j.efsa.2017.4689/pdf [Accessed 14 August 2017].

Kahn SA, Beers PT, Findlay VL, Peebles IR, Durham PJ, Wilson DW, Gerrity SE (1999). Import risk analysis on non-viable salmonids and non-salmonid marine finfish. Australian Quarantine and Inspection Service, Department of Agriculture, Fisheries and Forestry, Canberra, Australia, 409 p.

Kent ML, Traxler GS, Kieser D, Richard J, Dawe SC, Shaw RW, Prosperi-Porta G, Ketcheson J, Evelyn TPT (1998). Survey of salmonid pathogens in ocean-caught fishes in British Columbia, Canada. *Journal of Aquatic Animal Health* 10: 211–219.

Munro ES, Midtyling PJ (2011). Infectious pancratic necrosis virus and associated aquatic birnaviruses. pp 1–65 *In:* Woo PTK, Brno DW (*Eds*.) Fish diseases and disorders, Volume 3: Viral Bacterial and fungal infections.CABI International, Wallingford, United Kingdom, 930 p.

OIE (2016a). Aquatic Animal Health Code, 19th Edition. World Organization for Animal Health. Paris, France [Online]. Available from: http://www.oie.int/international-standard-setting/aquatic-code/access-online/ [Accessed 15 March 2017].

OIE (2016b). Manual of diagnostic tests for aquatic animals. World Organization for Animal Health. Paris, France [Online] Available from: http://www.oie.int/international-standard-setting/aquatic-manual/access-online/ [Accessed 17 March 2017].

Rucker RR, Whipple WJ, Parvin JR, Evans CA (1953). A contagious disease of salmon possibly of virus origin. *Fishery Bulletin* 76: 35–46.

Samuelsen OB, Nerland AH, Jorgensen T, Schroder MB, Svasand T, Bergh O (2006). Viral and bacterial diseases of Atlantic cod *Gadus morhua*, their prophylaxis and treatment: a review. *Diseases of Aquatic Organisms* 71: 239–254.

Stone, MAB, MacDiarmid SC, Pharo HJ (1997). Import health risk analysis: salmonids for human consumption. Ministry of Agriculture Regulatory Authority, New Zealand, 269 p.

Traxler GS, Roome JR, Kent ML (1993). Transmission of infectious haematopoietic necrosis virus in seawater. *Diseases of Aquatic Organisms* 16: 111–114.

Tubbs L, Lee P, Diggles B, Jones JB, Sheppard M, Sim-Smith C (2007). A review of aquatic diseases of significance to New Zealand. Final research report for project ZBS 2005-17. National Institute of Water and Atmospheric Research, Wellington, New Zealand, 452 p.

Wang M, Lin X, Ma G, Bai X (2007). Emerging viral diseases of fish and shrimp. pp 35–58. *In*: Lai SK (*Ed.*). Emerging Viral Diseases of Southeast Asia, Volume 4. Issues in Infectious Diseases. Karger Medical, Basel, Switzerland, 149 p.

Whipple MJ, Rohovec JS (1994). The effect of heat and low pH on selected viral and bacterial fish pathogens. *Aquaculture* 123: 179–189.

Wingfield WH, Fryer JL, Pilcher KS (1969). Properties of the sockeye salmon virus (Oregon strain). *Proceedings of the Society for Experimental Biology and Medicine* 130: 1055–1059.

Xu L, Zhao J, Liu M, Kurath G, Ren G, Lapatra SE, Yin J, Liu J, Feng J, Lu T (2017). A effective DNA vaccine against diverse genotype J infectious hematopoietic necrosis virus strains prevalent in China. *Vaccine* 35: 2420–2426 [Online] Available from: http://www.sciencedirect.com/science/article/pii/S0264410X17303638 [Accessed 14 August 2017].

17. Infectious pancreatic necrosis virus

17.1. Hazard identification

17.1.1. Aetiological agent

Infectious pancreatic necrosis virus (IPNV) is a bi-segmented double stranded RNA virus that is the type species for the Family Birnaviridae. It was first identified in brook trout (*Salvelinus fontinalis*) and in 1962 was the first fish virus to be grown in tissue culture (Wolf & Quimby 1962; Munro & Midtlyng 2011). It potentially infects all salmonids, particularly juveniles (Tubbs *et al*. 2007) and has a worldwide distribution (but excluding Australia and New Zealand).

Ten serogroups are known, each with several strains (serotypes) that differ in mortality, depending on the host species as well as water quality (temperature, salinity) and stocking levels (Barja 2004; Crane & Hyatt 2011).

Tasmanian aquabirnavirus (TaBV) is reported from Atlantic salmon (*Salmo salar*) farmed in Australia (McColl *et al.* 2009; B. Jones, *pers. comm*. 2015). The other aquatic birnaviruses that do not infect salmon are collectively called aquabirnaviruses.

Analysis of the viral genome suggests that viral deformity of yellowtail virus (VDV), halibut birnavirus (HBV) and related strains isolated from marine fish are similar to infectious necrosis virus (IPNV) at the genetic level in terms of their biological and serological characteristics (Hosono *et al*. 1996). The junction region on the genome segment-A coding viral capsid protein VP2 and viral protease NS was amplified by PCR in six marine strains. Analysis of nucleotide and the deduced amino acid sequences has revealed that the six marine strains have amino acid variations in the possible amino terminus of NS when compared to IPNV. The six marine strains form a new genogroup, which is distinguished from three previously isolated serotypes of IPNV (Hosono *et al.* 1996; B. Jones, *pers. comm*. 2015).

For this report, the associated birnaviruses HBV (Roberts 2012), TaBV (McColl *et al.* 2009) and VDV (B. Jones, *pers. comm.* 2015) have been regarded as isolates of IPNV.

17.1.2. OIE status

Infection with infectious pancreatic necrosis is not listed by the OIE (OIE 2016a).

17.1.3. New Zealand status

Infectious pancreatic necrosis virus is an exotic, notifiable disease in New Zealand (Johnston 2008; Anon. 2016). Although IPNV-like aquatic birnaviruses have been reported from farmed New Zealand turbot (*Colistium nudipinnis*) and flounder (*Rhombosolea* spp.) and from wild Chinook salmon (*Oncorhynchus tshawytscha*) (Tisdall & Phipps 1987, Anderson 1997; Johnston 2008), these isolates were non-pathogenic to salmon (Horner 2003).

VDV has only been reported from Japan and is considered exotic in New Zealand (Diggles 2002; Tubbs *et al.* 2007).

17.1.4. Epidemiology

IPNV is one of the most serious diseases affecting both wild and farmed fish including salmonids (*Oncorhynchus* spp., *Salmo* spp. and *Salvelinus* spp.) (Ruane *et al.* 2007). Mortality varies widely among serotypes, over 4 orders of magnitude in salmonids (Skjesol *et al.* 2011), but is highest in post-smolt salmon when introduced to sea cages (Barja 2004). Mortality is equally variable (from 5 to 100%) in non-salmonids such as eels (*Anguilla anguilla*), depending on the serotype (Van Beurden *et al.* 2012). Mortality decreases with fish age (Crane & Hyatt 2011).

IPNV also infects other farmed fish including sea bass (*Dicentrarchus labrax*) in France, turbot (*Scophthalmus maximus*) in Norway, France and Spain, dab (*Limanda limanda*) and Atlantic cod (*Gadus morhua*) in Denmark and sole (*Solea* spp.) in Spain, as well as halibut (*Hippoglossus hippoglossus*) (Munro & Midtlyng 2011; Roberts 2012) and yellowtail (*Seriola quinqueradiata*) (Munro & Midtlyng 2011). IPNV is also reported from wild eels (*Anguilla anguilla*) in Spain (Bandin *et al.* 2014) and from *A. japonica* in Japan (McAllister & Owens 1992)

The VDV isolate occurs in yellowtail where mortality may reach 80 to 90% in Japanese aquaculture (Nakajima *et al.* 1998), but clinical infection is rarely associated with wild fish (Kahn *et al.* 1999). It is not considered zoonotic, but may be cross-infective to hosts of YAV (Kahn *et al.* 1999).

The TaBV isolate is only reported from farmed Atlantic salmon (*S. salar*) in Macquarie Harbour, Tasmania, Australia (McColl *et al.* 2009).

Clinical or subclinical infection has been detected in over 100 species of estuarine, marine and freshwater species. A representative, but not exhaustive list is given in Table 13 (USFS 2007; McAlliser 2007; Wallace *et al.* 2008; Anon. 2001; Bandin & Dopazo 2011; Crane & Hyatt 2011; Munro & Midtyling 2011; Skjesol *et al.* 2011; Moreno *et al.* 2014).

IPNV also occurs naturally in mussels (*Mytilus galloprovincialis*) and oysters (*Pinctada fucata*), while scallops (*Pecten* spp.) can be experimentally infected. The life cycle is direct, but infection may involve alternative pathways (Olesen *et al*. 1988; Kitamura & Suzuki 2000; Barja 2004; López-Jimena *et al*. 2010; Munro & Midtlyng 2011). Most isolates from non-salmonid hosts do not generally infect salmonids (Munro & Midtlyng 2011), but experimental infection with isolates derived from infected Atlantic cod (*G. morhua*) resulted in mortalities up to 20% in Atlantic salmon (*S. salar*) (Urquhart *et al.* 2009).

IPNV can be transmitted vertically (via infected eggs), or horizontally, through infected water, fish urine, faeces, and sexual fluids (OIE 2002; Munro & Midtlyng 2011) where healthy sea-cage stock may become infected from adjacent sub-clinically infected wild stocks (Castric 1997). Infection may occur through contaminated water or through fomites (vehicles, nets, utensils and other human activities) in aquaculture or during fish processing (Munro & Midtlyng 2011).

Pathogen entry occurs by ingestion, usually through the intestinal epithelium, or across the gill membranes (Munro & Midtlyng 2011). External clinical signs in salmonids include a distended abdomen and moderate exophthalmia, as well as haemorrhages on the ventral skin surface and base of the ventral fins. The focus of infection is pancreatic, kidney and liver tissues, but IPNV may also be present in the brain and gill tissues (Munro & Midtlyng 2011; Roberts 2012).

Table 13. Families and Species of Fish Susceptible to IPNV and Associated Isolates

Family	Host Species
Acipenseridae	Sturgeons (*Acipens*er spp.)
Amiidae	Bowfins (*Amia calva*)
Ammodytidae	Sand eel (*Ammodytes* spp.)
Anarhichadidae	Atlantic wolf fish (*Anarhinchas lupus*), spotted wolf fish (*A. minor*)
Anguillidae	Eels (*Anguilla* spp.)
Atherinidae	Silversides (*Atherina* spp., *Atherinosoma* spp., *Kestratherina* spp., *Atherinomorus* spp.)
Aulorhynchidae	Tubesnout (*Aulorhynchus flavidus*)
Bothidae	Left-eye flounders (*Bothus* spp.), *Trichopsetta* spp.)
Carangidae	Jacks (*Carangoides* spp.), Japanese amberjack (*Seriola quinqueradiata*, *S. dumerili*), Japanese mackerel (*Trachurus japonicus*) , trevally (*Caranx* spp.), yellowtail (*Seriola lalandi*)
Catostomidae	White sucker (*Catostomus commersonii*)
Centrarchidae	Bluegill (*Lepomis macrochirus*), green sunfish (*Lepomis cyanellus*)
Channidae	Striped snakehead (*Channa striata*)
Cichlidae	Tilapias (*Oreochromis* spp., *Tilapia* spp. *Sarotheron* spp.)
Clupeidae	Atlantic menhadden (*Brevoortia tyrannus*), herrings/sardines (*Clupea* spp., *Sardinops* spp.)
Cobitidae	Loaches (*Acantopsis* spp., *Acanthopsoides* spp., *Botia* spp.)
Cyprinidae	Barbel (*Barbus barbus*), carp[M] (*Cyprinus carpio*), flathead minnow *(Pimephales promelas*), goldfish (*Carassius auratus*), Southwest European nase (*Parachondrostoma toxostoma*), Spanish barbel (*Barbus graellsi*)
Cyprinodontidae	Killifish (*Aphanius* spp.)
Embiotocidae	Shiner perch (*Cymatogaster aggregata*)
Esocidae	Pike (*Esox* spp.)
Gadidae	Atlantic cod (*Gadus morhua*), blue whiting (*Micromesistius poutassou*), coalfish (*Pollachius virens*), haddock (*Melanogrammus aeglefinus*)
Ictaluridae	Bullheads (*Ameiurus* spp.)
Labridae	Goldsinny-wrasse (*Ctenolabrus rupestris*)
Lepisosteidae	Garfish (*Lepisosteus* spp.)
Macrouridae	Roughhead grenadier (*Macrourus berglax*)
Merlucciidae	European hake (*Merlucius merlucius*)
Moronidae	White seabass (*Morone* saxitalis, *Morone* spp.)
Mullidae	Surmullet *(Mullus surmuletus*)
Paralichthyidae	Halibut (*Hippoglossus hippoglossus, H. stenolepis*), summer flounder (*Paralichthys dentatus*), southern flounder (*P. lethostigma*), Bastard halibut (*P. olivaceus*)
Percichthyidae	Murray cod (*Maccullochia peelii*), Macquarie perch (*Macquaria australasica, M. amigua, M. novemaculata)*, Mandarin fish (*Siniperca* spp.), blackfish (*Gadopsis* spp.)
Percidae	Perch (*Perca* spp.), European perch (*Perca fluviatilis*), yellow perch (*P. flavescens*), Balkhash perch (*P. schrenkii*)
Plecoglossidae	Ayu (*Plecoglossus altivelis, P. plecoglossus*)
Pleuronectidae	Common dab (*Limanda limanda*), European plaice (*Pleuronectes platessa*), European flounder (*Platichthys flesus*), dab (*Hippoglossus plattessoides*), flounder (*Pleuronectes* spp.) , greenland halibut (*Reinhardtius hippoglossoides*), lemon sole (*Microstomus kitt*), Pacific halibut (*Hippoglossus stenolepis*), Atlantic halibut (*Hippoglossus hippoglossus*)
Polyodontidae	Paddlefish (*Polyodon spathula*)
Salmonidae	Amago salmon (*Oncorhynchus rhodurus*), Arctic char (*Salvelinus alpinus*), Atlantic salmon (*Salmo salar*) , brook trout (*Salvelinus fontinalis*), brown trout (*Salmo trutta*), rainbow trout (O. mykiss), chinook salmon (O. tshawytscha), chum salmon (*O. keta*), coho salmon (*O. kisutch*), cutthroat trout (*S. clarkii*), Danube salmon (*Hucho hucho*), grayling (*Thymallus thymallus*), lake trout (*Salvelinus. namaycush*), masu salmon (*O. masou*), Pacific salmon (*Oncorhynchus* spp), sockeye salmon (*O. nerka*), whitefish (*Coregonus* spp.,. *Prosopium* spp.)
Sciaenidae	Drums and croakers, including *Aplodinotus* spp., *Atractoscion* spp., meagre (*Arygrosomus* spp.), *Atractoscion* spp., *Atrobucca* spp., *Austronibea* spp., *Bahaba* spp., *Bairdiella* spp., *Boesmania* spp., *Cheilotrema* spp., *Chrysochir* spp., Cilus spp., *Collichthys* spp., *Corvula* spp., *Daysciaenia* spp., *Dendrophysa* spp., *Leiostomus xanthurus*, red drum (*Sciaenops ocellatus Ctenosciaenia spp., Cynoscion spp.,* and over 100 other species
Scophthalmidae	Turbot (*Scophthalmus maxima, Scophthalmus* spp.)
Soleidae	Sole (*Solea* spp.), Senegal sole (*Solea senegalis*)

Sparidae	Axillary seabream (*Pagellus acarne*), black seabream (*Spondyliosoma cantharus*), common pandora (*Pagellus erythrinus*), gilthead seabream (*Sparus aurata*), redbanded seabream (*Pagrus auriga*), sea bream (*Pagellus spp.*), two band bream (*Diplodus* spp.), yellowfin bream (*Acanthopagrus australis*)
Sphyraenidae	Barracouta (*Sphyranea sphyranea*), *Sphyranea* spp.
Triglidae	Grey gurnard (*Eutrigla gurnardus*)

[M] Some authorities recognise subspecies of common carp (*Cyprinus carpio*) including koi carp (*C. carpio koi*) and ghost carp (*C. carpio goi*). For the purposes of this document, these subspecies will be considered as *Cyprinus carpio*, consistent with the taxonomy of Fishbase (2018).

Infection may persist at a subclinical level in surviving fish, which then act as carriers of disease. Infection may persist at low viral titres (Isshiki *et al.* 2001), depending on the virus strain, host species and age, and be influenced by environmental stressors (temperature, salinity, oxygen concentration) (Munro & Midtlyng 1999; Isshiki *et al.* 2001).

Dead and decomposing fish also act as sources of infection with IPNV, while infective viral particles may be transferred by fish eating birds and mammals (Munro & Midtlyng 2011). IPNV may be shed in the faeces of piscivorous birds, such as grey heron (*Ardea cinerea*), for up to 7 days after feeding (McAllister & Owens 1992). The minimum dose for infection is unknown (Munro & Midtlyng 2011).

IPNV and isolates are highly resistant to environmental conditions, remaining viable for several weeks in pond mud at 10°C. IPNV survives for 10 to 12 weeks in filtered water at 4°C, with residual infectivity after 24 weeks (Desautels & MacKelvie 1975), while the VDV isolate survives in Japanese coastal marine waters for at least 15 days (Mortensen *et al.* 1990; Kitamura & Suzuki, 2000; Kitamura *et al.* 2004). IPNV may be spread by transport water as well as by the moist surfaces of contaminated nets, shipping containers and equipment (Munro & Midtlyng 2011).

IPNV is resistant to the environmental conditions likely to be encountered in the human food consumption pathway and commonly associated sewage or effluent treatments (Munro & Midtlyng 2011). It is unaffected by UV light sterilization treatments used in municipal sewerage systems and is even more resistant to the UV light sterilization treatment used in seawater aquaculture (Munro & Midtlyng 2011). It is resistant to saline conditions (from 0 to 40%), to mild acid conditions, being only partially inactivated at pH 2.5 for 1 hour. It is resistant to freezing (Kahn *et al.* 1999) and to heat treatment, requiring 5 hours heating at 60°C, where pH is 7 to 9, but is inactivated after 30 minutes at 60°C under acid conditions (pH 3). IPNV is also unaffected by salinity as infections are reported in both marine and freshwater hosts (Munro & Midtlyng 2011).

It is inactivated by formalin (14 days in 250 ppm), chlorine (40 mg L^{-1} for 30 minutes), alkaline (NaOH: pH 11.9 for 5 minutes) and iodophor (35 mg L^{-1} for 5 minutes) treatments (Tubbs *et al.* 2007; Munro & Midtlyng 2011). No effective commercial vaccines are available for IPNV (Munro & Midtlyng 2011).

The species at risk in New Zealand include farmed salmonids such as rainbow trout (*O. mykiss*) and Chinook salmon (*O. tshawytscha*) (Tubbs *et al.* 2007), as well as eels (*A. dieffenbachii*, *A. australis*, *A. reinhardti*) (Bandin *et al.* 2014). Yellowtail kingfish (*Seriola lalandi*) and Sampson fish (*Seriola hippos*) are also considered susceptible to the VDV isolate (Tubbs *et al.* 2007).

While the associated IPNV-like aquabirnaviruses have a wide host range, eel birnaviruses are not considered likely to cross infect salmonids (B. Jones, *pers. comm.* 2015).

17.2. Risk assessment

17.2.1. Entry assessment

IPNV and its isolates may be present in carrier fish with no external signs of infection (Munro & Midtlyng 2011). These may pass visual inspection and the viral infection present in the brain and gill tissues will be retained after evisceration (Munro & Midtlyng 2011). IPNV is unlikely to be affected by commercial fish processing and storage, remaining viable in fish offal and blood-water discharges in the factory waste stream (Munro & Midtlyng 2011). It may also be spread beyond the immediate processing area on the moist biofilm layers of fomites, including commercial processing, storage and transport equipment (Munro & Midtyling 2011).

The likelihood of entry of IPNV is assessed as non-negligible.

17.2.2. Exposure assessment

To establish infection in the commodity, sufficient infected product would have to become available for consumption by a susceptible marine or freshwater fish host, in sufficient quantity and duration (Kahn *et al.* 1999). IPNV remains viable in discharges from aquaculture (Munro & Midtlying 2011) and remains viable in water for up to 24 weeks (Desautels & MacKelvie 1975), so it may be present in offal and blood-water discharges from factory processing, or be transmitted by fomites (Munro & Midtlying 2011). As IPNV survives passage through the avian digestive system (McAllister & Owens 1992), it may also be spread from landfill disposal sites to the aquatic environment by scavenging birds. IPNV may infect a wide variety of fish and invertebrate hosts including mussels (*Mytilus* spp.) and scallops (*Pecten* spp.).

The likelihood of exposure to IPNV is assessed as non-negligible.

17.2.3. Consequence assessment

The establishment of IPNV in New Zealand would have economic consequences for salmonid exports from the aquaculture industry, which were valued at $63 million in 2011 (Aquaculture New Zealand 2014). There would also be major social and economic effects for the industries associated with recreational and tourist trout and salmon fishing, as well as significant social and environmental costs associated with other domestic fisheries (Fish & Game 2014). While the dollar value of New Zealand's freshwater fisheries is not fully known, the Lake Taupo trout fishery alone is valued at $70 to 80 million (Marsh & Mkwra 2013).

The introduction of IPNV may also affect other fish species in New Zealand. While the severity of disease would be dependent upon the pathogenicity of the IPNV isolate introduced and on the species infected, an outbreak would be likely to incur high mortality in a naïve population under suitable conditions (Munro & Midtlyng 2011). The developing aquaculture industry for yellowtail kingfish (*Seriola lalandi*) (NIWA 2017) and snapper (*Sparus aurata*) (Plant & Food 2016) would be affected by the introduction of IPNV and associated isolates.

The consequences of the introduction of IPNV and its isolates are assessed as non-negligible.

17.2.4. Risk estimation

Since the entry, exposure and consequence assessments are non-negligible, the risk estimate is non-negligible. Therefore, IPNV and its isolates are assessed to be a risk in the commodity and risk management measures may be justified.

17.3. Risk management

IPNV and its isolates have been assessed to be a risk in the commodity. Infection with IPNV is not an OIE-notifiable disease, so the *Aquatic Code* (OIE 2016a) provides no specific guidance for importing eviscerated fish from infected countries, or on specific processing requirements that would ensure the destruction of the virus. It is assumed that compliance with the approved processed states for OIE-listed diseases (Appendix 3) would also eliminate IPNV from the commodity and be a viable risk management option.

IPNV and related isolates occur in over 100 species of marine, estuarine and fresh water fish from 41 families of wild and farmed fish (Table 13). These may be present in the commodity. Species declaration should substantially reduce the pathogen load of IPNV and be a viable risk management option. Restriction of the commodity to wild-caught fish (not from aquaculture) would have no effect on pathogen load and is not a viable risk management option.

IPNV and related isolates have a wide distribution, including Europe, North and South America, Asia and Africa, but no requirements for dedicated monitoring exist. Where country/zone freedom is approved through the MPI Country Approval Procedures, this option is should substantially reduce the occurrence of IPNV and related isolates in the commodity. Approval of country/zone freedom by MPI is a viable risk management option.

Within the commodity, IPNV and related isolates are essentially restricted to the gills and brain tissue of the head, so removal of the gills should slightly reduce this pathogen load. Further removal of the head and gills should moderately reduce pathogen load in the commodity and be a viable risk management option.

IPNV and related isolates are unaffected by freezing, so frozen storage is not a viable risk management option. IPNV is extremely heat stable, requiring high temperatures (to 60°C for 300 minutes (5 hours)) to ensure denaturation. This heat treatment would eliminate IPNV and related isolates from the commodity and be a viable risk management option.

17.3.1. Risk management options

The IPNV virus and associated isolates are reported from fish in families Acipenseridae, Amiidae, Ammodytidae, Anarhichadidae, Anguillidae, Atherinidae, Aulorhynchidae, Bothidae, Carangidae, Catostomidae, Centrarchidae, Channidae, Cichlidae, Clupeidae, Cyprinidae, Cyprinodontidae, Embiotocidae, Esocidae, Gadidae, Ictaluridae, Labridae, Lepisosteidae, Macrouridae, Merlucciidae, Moronidae, Mullidae, Paralichthyidae, Percichthyidae, Percidae, Plecoglossidae, Pleuronectidae, Polyodontidae, Salmonidae, Sciaenidae, Scophthalmidae, Soleidae, Sparidae, Sphyraenidae and Triglidae (Table 13). These families are considered likely to be present in the commodity. Other families have not been associated with IPNV and associated isolates. Therefore species declaration indicating the commodity is not originated from any of the above families should substantially reduce the occurrence of IPNV in the commodity.

For the commodities originated from families associated with IPNV and associated isolates, one or a combination of the following additional options could also be considered to effectively manage the risk.

Where country/zone freedom from IPNV and associated isolates is accepted by MPI:

Option 1

Acceptance of country/zone freedom should substantially reduce the occurrence of IPNV and associated isolates, so the commodity may be imported without any further restrictions.

Where country/zone freedom from IPNV and associated isolates is not accepted by MPI:

Option 2

Processing consistent with the approved states for OIE-listed diseases (Appendix 3) should eliminate IPNV. When these provisions are met, the commodity could be imported without any further restrictions.

Where the imported commodity is not in a state consistent with the approved states for OIE-listed diseases (Appendix 3):

Option 3

Heat treatment (by cooking to at least 60°C for at least 5 hours (300 minutes) should eliminate IPNV. When this provision is met, the commodity could be imported without any further restrictions; or,

Option 4

Further processing (by removal of the head and gills) should moderately reduce the occurrence of IPNV. When this provision is met, the commodity could be imported without any further restrictions.

17.4. References

Anonymous (2001). Loaches, hill stream loaches and algae eaters. Mekong Fisheries Network Newsletter: Mekong Fish Catch and Culture 6(3_10. 2 p. [Online] Available from: http://www.mekonginfo.org/assets/midocs/0002925-biota-loaches-hill-stream-loaches-and-algae-eaters.pdf [Accessed 8 June 2018].

Anonymous (2016). Biosecurity (Notifiable Organisms) Order 2016 SR 2016/73. New Zealand Government, Wellington, New Zealand, 13 p.

Anderson CD (1997). Fish facts from New Zealand. *Microbiology Australia* May 1997: 20–21.

Aquaculture New Zealand (2014). New Zealand's aquaculture industry key facts 2013 [Online] Available from: http://www.seafoodnewzealand.org.nz/our-industry/key-facts/ [Accessed 26 September 2014]..

Bandin I, Souto S, Cutrin JM, López-Vazquez C, Olviera JG, Esteve C, Alcaide E, Dopazo CP (2014). Presence of viruses in wild eels *Anguilla anguilla* L, from the Albufera Lake (Spain). *Journal of Fish Diseases* 37(7): 597–607.

Bandin I, Dopazo CP (2011). Host range, host specificity and hypothesized host shift events among viruses of lower vertebrates. *Veterinary Research* 42:67 [Online] Available from https://doi.org/10.1186/1297-9716-42-67 [Accessed 27 July 2017].

Barja JL (2004). Report about fish viral diseases. pp 91-102. *In*: Alvarez-Pellitero P, Barja JL, Basurco B, Berthe F, Toranzo AE (*Eds*.). Mediterranean aquaculture diagnostic laboratories. Zaragoza: CIHEAM, 2004. *Options Méditerranéennes: Série B. Etudes et Recherches*; n. 49 p.

Castric J (1997). Viral diseases in fish mariculture. *Bulletin of the European Association of Fish Pathologists* 17(6): 220–228.

Crane M, Hyatt A (2011). Viruses of fish: An overview of significant pathogens. *Viruse*s 3: 2025–2046.

Desautels D, MacKelvie RM (1975). Practical aspects of survival and destruction of infectious pancreatic necrosis virus. *Journal of the Fisheries Research Board of Canada* 32(4): 523–531.

Diggles BK, Hine PM, Handley S, Boustead NC (2002). A handbook of diseases of importance to aquaculture in New Zealand. National Institute of Water and Atmospheric Research (NIWA), Auckland, New Zealand. (1st edition).

Fish and Game (2014). New Zealand Fish and Game Council [Online] Available from: http://www.fishandgame.org.nz/ [Accessed 24 September 2014].

Horner GW (2003). Reports from MAF Biosecurity Authority – National Centre for Disease Investigation. *Surveillance* 30(2): 14-15.

Hosono N, Suzuki S, Kusuda R (1996). Genogrouping of birnaviruses isolated from marine fish: a comparison of VP2/NS junction regions on genome segment A. *Journal of Fish Diseases* 19: 295–302.

Isshiki T, Nagano T, Suzuki S (2001). Infectivity of aquabirnavirus strains to various marine fish species. *Diseases of Aquatic Organisms* 46(2): 109–114.

Johnston C (2008). Import risk analysis: Frozen, skinless and boneless fillet meat of *Oreochromis* spp. from China and Brazil for human consumption. MAF Biosecurity New Zealand, 96 p.

Kahn SA, Beers PT, Findlay VL, Peebles IR, Durham PJ, Wilson DW, Gerrity SE (1999). Import risk analysis on non-viable salmonids and non-salmonid marine finfish. Australian Quarantine and Inspection Service, Department of Agriculture, Fisheries and Forestry, Canberra, Australia, 409 p.

Kitamura SI, Suzuki S (2000). Occurrence of marine birnavirus through the year in coastal seawater in the Uwa Sea. *Marine Biotechnology* 2: 188–194.

Kitamura SI, Kamata SI, Nanako S, Suzuki S (2004). Solar UV radiation does not inactivate marine birnavirus in coastal seawater. *Diseases of Aquatic Organisms* 58(2-3): 251–254.

López-Jimena B, Garcia-Rosado E, Infante C, Cano I, Machado M, Castro D, Borrego JJ, Alonso MC (2010). Detection of infectious pancreatic necrosis virus (IPNV) from asymptomatic red banded seabream, *Pagrus auriga* Valenciennes, and common seabream, *Pagrus pagrus* (L.), using a non-destructive procedure. *Journal of Fish Diseases* 33(4): 311–319.

Marsh D, Mkwra L (2013). Review of freshwater non-market value studies. Department of Economics, University of Waikato, Hamilton, New Zealand, 144 p.

McAllister PE, Owens WJ (1992). Assessment of the virulence of fish and molluscan isolates of infectious pancreatic necrosis virus for salmonid fish by challenge of brook trout, *Salvelinus fontinalis* (Mitchell). *Journal of Fish Diseases* 18: 97–103.

McAlliser PE (1997). Infectious Pancreatic Necrosis. Resource sheet, National Health Fishery Laboratory, 9 p [Online] Available from: http://www.afs-fhs.org/perch/resources/14069231202.2.5ipnv2007ref2014.pdf [Accessed 27 July 2017].

McColl KA, Davies KR, Young JG, Crane, MStJ (2009). Aquatic Birnavirus Infections of Finfish. Australia and New Zealand Standard Diagnostic Procedure, Department of Agriculture and Water Resources, Canberra, Australia, 20 p. [Online] Available from: http://www.agriculture.gov.au/SiteCollectionDocuments/animal/ahl/ANZSDP-Aquatic-birnaviruses.pdf [Accessed 15 August 2018].

Moreno P, Olveria JG, Laballa A, Cutrin JM, Baro JC, Borrego JJ, Dopazo CP (2014). Surveillance of Viruses in Wild Fish Populations in Areas around the Gulf of Cadiz (South Atlantic Iberian Peninsula). *Applied and Environmental Microbiology* 80(20): 6560 – 6571.

Mortensen SH, Hjeltnes B, Rodseth O, Krogsrud J, Christie JE (1990). Infectious pancreatic necrosis virus, serotype N1, isolated from Norwegian halibut (*Hippoglossus hippoglossus*), turbot (*Scophthalmus maximus*) and scallops (*Pecten maximus*). *Bulletin of the European Association of Fish Pathologists* 10(2): 42–43.

Munro ES, Midtlyng PJ (2011). Infectious pancreatic necrosis and associated aquatic birnaviruses. pp 1–65. *In*: Woo PTK, Bruno DW (*Eds.*). *Fish Diseases and Disorders: Volume 3 - Viral, Bacterial and Fungal Infections*, 2nd edition. CABI International, Wallingford, United Kingdom, 930 p.

Nakajima K, Inouye K, Sorimachi M (1998). Viral diseases in cultured marine fish in Japan. *Fish Pathology* 33:181–188.

NIWA (2017). Yellowtail Kingfish. National Institute for Water and Atmospheric Research (NIWA), Auckland, New Zealand. [Online] Available from: https://www.niwa.co.nz/aquaculture/aquaculture-species/yellowtail-kingfish [Accessed 14 August 2017].

OIE (2016a). Aquatic Animal Health Code, 19th Edition. World Organization for Animal Health. Paris, France [Online]. Available from: http://www.oie.int/international-standard-setting/aquatic-code/access-online/ [Accessed 15 March 2017].

OIE (2016b). Manual of diagnostic tests for aquatic animals. World Organisation for Animal Health, Paris, France [Online]. Available from: www.oie.int/ international-standard-setting/aquatic-manual/access-online/ [Accessed 15 March 2017].

Olesen NJ, Jorgensen PEV, Bloch B, Mellergaard S (1988). Isolation of an IPN-like virus belonging to the serogroup II of the aquatic birnaviruses from dab (*Limanda limanda*). *Journal of Fish Diseases* 11(5): 449–451.

Plant & Food (2016). Sustainable snapper piped out to sea by Plant and Food [Online] Available from: http://www.stuff.co.nz/business/farming/aquaculture/87522652/sustainable-snapper-piped-out-to-sea [Accessed 14 August 2017].**Roberts R (2012).** Fish Pathology. John Wiley, London, 592 p.

Ruane N, Geoghegan F, Cinneide MO (1997). Infectious pancreatic necrosis virus and its impact on Irish salmon aquaculture and wild fish sectors. Marine Environment and Health Series 30, Marine Institute, Galway, Ireland, 56 p.

Skjesol A, Skjæveland I,Elnæs M, Timmerhaus G, Fredriksen BN, Jørgensen SM, Krasnov A, Jørgensen JB (2011). IPNV with high and low virulence: host immune responses and viral mutations during infection. *Journal of Virology* 8: 396. [Online] Available from: www. doi: 10.1186/1743-422X-8-396 [Accessed 26 July 2017].

Tisdall DJ, Phipps JC (1987). Isolation and characterisation of a marine birnavirus from returning Chinook salmon (*Oncorhynchus tshawytscha*) in the South Island of New Zealand. *New Zealand Veterinary Journal* 35: 217–218.

Tubbs L, Lee P, Diggles B, Jones JB, Sheppard M, Sim-Smith C (2007). A review of aquatic diseases of significance to New Zealand. Final research report for project ZBS 2005-17. National Institute of Water and Atmospheric Research, Wellington, New Zealand, 452 p.

Urquhart K, Bowden TJ, Buckett BE, Fryer RJ, Ellis AE (2009). Experimental study of the susceptibility of Atlantic cod, *Gadus morhua* L, to infection with an IPNV strain pathogenic for Atlantic salmon *Salmo salar* L. *Journal of Fish Diseases* 32: 447–456.

USFWS (2007). Suggested procedures for the detection and identification of certain finfish and shellfish pathogens. Standard procedures for aquatic animal health inspections. AFS-FHS. FHS Blue Book (2007 edition). U.S. Fish and Wildlife Service and American Fisheries Society-Fish Health Section, Bethesda, Maryland, United States of America.

Van Beurden SJ, Engelsma MY, Roozenburg I, Voorbergen-Laarman MA, van Tulden PW, Kerkhoff S, van Nieuwstadt AP, Davidse A. Haenen OLM (2012). Viral diseases of wild and farmed European eel *Anguilla anguilla* with particular reference to the Netherlands. *Diseases of Aquatic Organisms* 101: 69–86.

Wallace IS, Gregory A, Munro ES, Raynard RS (2008). Distribution of infectious pancreatic necrosis virus (IPNV) in wild marine fish from Scottish waters with respect to clinically infected aquaculture sites producing Atlantic salmon, *Salmo salar* L. *Journal of Fish Diseases* 31(3): 177–186.

Wolf K, Quimby MC (1962). Established eurythermic line of fish cells *in vitro*. *Science* 135: 1064.

18. Infectious salmon anaemia virus

18.1. Hazard identification

18.1.1. Aetiological agent

Infectious salmon anaemia virus (ISAV) causes economically significant clinical disease in Atlantic salmon (*Salmo salar*) and Coho salmon (*Oncorhynchus kisutch*), although other fish may be carriers of disease (OIE 2016a, 2016b). Infection with ISAV occurs in two forms: either infection with highly polymorphic region (HPR-deleted ISAV); or infection with HPR0 ISAV (the non-deleted HPR form of ISAV). ISAV is classified in the Genus *Isavirus*, within the Family Orthomyxoviridae (OIE 2016a) and was first isolated from Atlantic salmon (*Salmo salar*) farmed in Norway in 1984 (Rimstad *et al.* 2011). It is now found in most areas where salmonids are farmed (OIE 2016b). Several isolates of each form have been determined, based on geographical area (OIE 2016b).

18.1.2. OIE status

Infection with both the HPR0 and HPR-deleted forms of ISAV are recognised by the OIE and listed as a notifiable disease (OIE 2016a).

18.1.3. New Zealand status

Both forms of ISAV are exotic to New Zealand (Stone *et al.* 1997) and infection with either form of ISAV is a notifiable disease in New Zealand (Anon. 2016). While infection with ISAV was suspected at an aquaculture facility in the Marlborough Sounds during 2012, this was subsequently shown not to be caused by ISAV and New Zealand is still considered free from ISAV (Norman *et al.* 2013).

18.1.4. Epidemiology

HPR0 ISAV

The epidemiology of HPR0 ISAV is currently unclear (OIE 2016a). It has been reported from apparently healthy wild and farmed Atlantic salmon (*Salmo salar*), in most salmon producing areas (OIE 2016a). It causes a transient subclinical infection mainly localised in gill endothelial tissues, although the kidney and heart may also be affected (Christiansen *et al.* 2011; EFSA 2012; Lyngstad *et al.* 2012; OIE 2016b). There is no evidence that HPR0 ISAV causes natural infection and replication in species other than Atlantic salmon (EFSA 2012), but sub-clinical infection occurs in Atlantic salmon, Atlantic cod, brown trout, Coho salmon, rainbow trout, pollock and Pacific herring, usually with no external signs of infection (EFSA 2012).

Mixed infection with both HPR0 and HPR–deleted forms occurs in sea-run Atlantic salmon with no clinical signs of infection, but the infection with the HPR0 form appears to be transient and these fish are not thought to be lifelong carriers of disease (OIE 2016a). Since the HPR0 form has not been detected in diseased Atlantic salmon showing clinical signs consistent with ISAV (OIE 2016a), the possible role of HPR0 ISAV in the re-emergence of the more-pathogenic HPR-deleted form of ISAV has been speculated (Cunningham *et al.* 2002; EFSA 2012). However, the risk factors associated with possible emergence remain unknown and no direct evidence exists, so causality remains unproven (OIE 2016b).

Initial infection occurs mainly through the gill membranes, but also through the intestinal wall following ingestion, or through the skin surface. Ectoparasites including salmon lice (*Lepeophtheirus salmonis*) and sea lice (*Caligus elongatus*) may also spread disease among fish hosts (Nylund *et al.* 1994; Rimstad *et al*. 2011; OIE 2016b).

Transmission is horizontal, through the water column, or by direct contact between fish (OIE 2016a, 2016b). ISAV can spread rapidly in marine waters (EFSA 2012) and may be transferred in transport water or in contaminated fish wastes, mucous, fish blood and wastewater discharges from fish processing factories. It may be also transferred by cross-infection of fishing boats and gear, transport and factory processing equipment (OIE 2016b).

Vertical transmission may also occur (Vike *et al.* 2009; Marshall *et al.* 2014), while transfer may also occur through adhesion to the surface of eggs and embryos, where biosecurity measures are poor (Rimstad *et al.* 2011; OIE 2016b).

The virus remains viable outside the host for extended periods (Rimstad *et al.* 2011) and viable in moist decaying fish (OIE 2016a). It is unaffected by salinity, remains viable in sea and fresh water for up to 10 days at 15°C, or 14 days at 4°C (OIE 2016b). It remains viable in fish tissues held chilled on-ice for at least 6 days (Tubbs *et al.* 2007). It is unaffected by freezing, surviving 5 freeze-thaw cycles of freezing (-20°C) and thawing (20°C) with no loss in viability (Rimstad *et al.* 2011). It survives heat treatment (up to 30 minutes at 56°C) (OIE 2016b). It is tolerant of acidic conditions, surviving at pH 4.5 for more than 24 hours and at pH 12 for up to 24 hours, but is inactivated after 30 minutes at pH 4 (Rimstad *et al.* 2011). It is resistant to the acidic conditions and high temperatures (37°C) found in the avian digestive tract and may be transferred by scavenging piscivorous birds (OIE 2016b). It is inactivated by bleach (100–1000 mg L^{-1} for 10 minutes) and iodophor disinfectants (100 to 200 mg L^{-1} for 5–10 minutes), as well as by ozone (8 mg L^{-1} min^{-1} for 3 minutes) and by UV radiation treatments (Oye & Rimstad 2001; Tubbs *et al.* 2007).

Vaccination (targeting HPR-deleted ISAV) is widely practiced but appears to offer incomplete protection, while the antiviral drug Ribavirin inhibits ISAV replication (OIE 2016b).

Species at risk in New Zealand include members of family Salmonidae, including Atlantic salmon (*S. salar*), Coho salmon (*Oncorhynchus kisutch*), sockeye salmon (*O. nerka*), brown trout (*S. trutta*) and rainbow trout (*O. mykiss*). Brown trout and rainbow trout are widely distributed in New Zealand, but Atlantic salmon are limited to the upper Waiau River catchment (McDowall 1994). Coho and sockeye salmon are virtually extinct in New Zealand natural waters, but some Coho salmon remain as brood stock populations on salmon farms (McDowell 1994; 2000; NIWA 2014a, 2014b).

HPR-Deleted ISAV

Clinical ISAV caused by the HPR-deleted form of ISAV is essentially a disease of Atlantic salmon (Vike *et al*. 2009; OIE 2016a). It has been reported from farmed Atlantic salmon in Norway, the Faroe Islands, the United Kingdom (Scotland), North America (Canada, United States) and South America (Chile) (EFSA 2012; OIE 2016a), while infection is also reported in farmed Chilean Coho salmon (*O. kisutch*) (Kibenge *et al*. 2001; OIE 2016a).

Experimental infection studies are equivocal. Peritoneal injection failed to induce infection in Pacific salmon, including rainbow trout (*O. mykiss*), chum (*O. keta*), Chinook (O. *tshawytscha*), and Coho (O. *kisutch*) (Rolland & Winton 2003). ISAV does not appear to infect Chinook salmon

(*Oncorhynchus tshawytscha*) (Rimstad *et al.* 2011; Norman *et al*. 2013), but rainbow trout (*Oncorhynchus mykiss*) and herring (*Clupea pallasii*) were infected following bath immersion challenge (Biacchesi *et al.* 2007; OIE 2016b) and isolates from these herring subsequently infected Atlantic salmon (Rimstad *et al.* 2011; OIE 2016a). Families and species considered susceptible to ISAV are given in Table 14 (Diggles *et al*. 2002; EFSA 2012; OIE 2016a, 2016b).

Table 14. Families and Species Of Fish Susceptible to HPR0 and HPR-Deleted Infectious Salmon Anaemia Virus (ISAV)

Family	Host Species
Clupeidae	Alewife (*Alosa* (*Clupea*) *pseudoharengus*) , herring (*Clupea pallasii*)
Gadidae	Atlantic cod (*Gadus morhua*), pollock (*Pollachius pollachus, P. virens*)
Salmonidae	Atlantic salmon (*Salmo salar*), brown trout (*Salmo trutta*), rainbow trout (*Oncorhynchus mykiss*), coho salmon (*O. kisutch*)

Natural sub-clinical infection with HPR-deleted ISAV occurs in farmed and wild stocks of Atlantic salmon in both fresh and sea water (OIE 2016b). Infection can be transferred to alewife (*Alosa pseudoharengus*), pollock (*Pollachius* sp., *P. virens*) and Atlantic cod (*Gadus morhua*), when they occur in close proximity to infected Atlantic salmon (*S. salar*) or rainbow trout (*O. mykiss*) (EFSA 2012). These alternative hosts may represent a marine reservoir of infection (OIE 2016b).

HPR-deleted ISAV causes long-term chronic disease with initial daily mortalities of up to 1%. Over several months, these cumulative mortalities may affect up to 90% of the farmed stock (OIE 2016a). Infection usually occurs in juvenile fish following transfer to seawater cages, although infection in fresh water has also been reported from Canada (OIE 2016a).

Clinical signs include exophthalmia and darkening of the skin surface. HPR-deleted ISAV is a systemic disease, affecting blood and the circulatory system of all organs and tissues (Rimstad *et al*. 2011). The focus of infection is the endothelial cells of the gills and major organs including the liver, heart, kidney and spleen (Rimstad *et al*. 2011).

Initial infection occurs through the gill membranes, the intestinal wall following ingestion, or through the skin surface. Ectoparasites including salmon lice (*Lepeophtheirus salmonis*) and sea lice (*Caligus elongatus*) may also spread disease among fish hosts (Nylund *et al.* 1994; OIE 2016a).

Transmission is horizontal, through the water column, or by direct contact between fish (OIE 2016a). Persistent infection is not thought to occur in Atlantic salmon, but may occur in rainbow trout and brown trout (OIE 2016a).

Infection can spread rapidly in marine waters (EFSA 2012) and may be transferred in transport water or in contaminated fish wastes, mucous, fish blood and wastewater discharges from fish processing factories. Transfer may also occur by cross-infection of fishing boats and gear, transport and factory processing equipment (OIE 2016a).

Transmission may be vertical (Vike *et al.* 2009; Marshall *et al.* 2014), but infection can also occur through adhesion to the surface of eggs and embryos, where biosecurity measures are poor (Rimstad *et al.* 2011).

Vaccination is widely practiced but appears to offer incomplete protection, while the antiviral drug ribavirin inhibits ISAV replication (OIE 2016a).

Species at risk in New Zealand include clupeids (Pacific herring *Clupea pallasii*), salmonids (brown trout (*Salmo trutta*) and rainbow trout (*Oncorhynchus mykiss*)), which are widely distributed and may act as carriers (OIE 2016a). Other potential salmonid hosts (including Atlantic salmon *S. salar*), Coho salmon *O. kisutch*), and sockeye salmon (*O. nerka*) are limited in distribution in New Zealand. Atlantic salmon is limited to the upper Waiau River catchment (McDowall 1994). Coho and sockeye salmon are virtually extinct in New Zealand natural waters, but some Coho salmon remain as brood stock populations on salmon farms (McDowall 1994; 2000; NIWA 2014a, 2014b).

18.2. Risk assessment

18.2.1. Entry assessment

HPR0 ISAV

The epidemiology of HPR0 ISAV is currently unclear (OIE 2016a). Sub-clinical infection occurs in Atlantic salmon, Atlantic cod, brown trout, Coho salmon, rainbow trout, pollock and Pacific herring, usually with no external signs of infection (EFSA 2012). Such fish (Table 19) would pass visual inspection and may be present in the commodity (OIE 2016a). The likelihood of entry of HPR0 ISAV in the commodity is assessed as non-negligible.

HPR-Deleted ISAV

HPR-deleted ISAV is relatively host-specific to Atlantic salmon, Coho salmon and rainbow trout but other hosts (Table 19) including Atlantic cod, brown trout, pollock and herring may also act as carriers (OIE 2016a). Clinically infected salmonids with signs of exophthalmia and darkened skin would be unlikely to pass visual inspection and should not be present in the commodity. However, sub-clinically infected salmonids and non-salmonid carrier fish with no external clinical signs may pass visual inspection and be present in the commodity.

The likelihood of entry of HPR-deleted ISAV in the commodity is assessed as non-negligible.

18.2.2. Exposure assessment

HPR0 ISAV

Evisceration would substantially reduce viral titre (Stone *et al.* 1997), but HPR0 ISAV may be present in the gill and eye tissues, fish blood and mucous of infected eviscerated fish (OIE 2016a). To establish infection in New Zealand, infected eviscerated product would have to become available for consumption by a susceptible marine or freshwater fish host, in sufficient quantity and duration (Kahn *et al.* 1999).

HPR0 ISAV may be present in the blood-water discharge from fish processing, as well as in the gill and eye tissues in fish heads discarded as offal after processing (OIE 2016a). It is low-temperature tolerant (Rimstad *et al*. 2011), unaffected by freezing (OIE 2016a) and resistant to environmental extremes likely to be encountered in commercial fish processing (OIE 2016a). It may remain viable in organic material disposed to landfill and possibly be transferred to the marine or aquatic environment by scavenging birds (OIE 2016a). Infection may also occur through cross-contamination of fomites such as fish processing and handling equipment (OIE 2016b).

The likelihood of exposure to HPR0 ISAV is assessed as non-negligible.

HPR-Deleted ISAV

While evisceration would substantially reduce viral titre (Stone *et al.* 1997), HPR-deleted ISAV may be present in the gill and eye tissues, fish blood and mucous of infected eviscerated fish (OIE 2016a). To establish infection in New Zealand, infected eviscerated product would have to become available for consumption by a susceptible marine or freshwater fish host, in sufficient quantity and duration (Kahn *et al.* 1999).

HPR-deleted ISAV may be present in the blood-water discharge from fish processing, as well as in the fish heads and offal discarded after processing (OIE 2016a). HPR-deleted ISAV is low-temperature tolerant (Rimstad *et al*. 2011), unaffected by freezing (OIE 2016a) and is resistant to environmental extremes likely to be encountered in commercial fish processing.

HPR-deleted forms of ISAV may remain viable in organic material disposed to landfill and possibly be transferred to the marine/aquatic environment by scavenging birds (OIE 2016a). Infection may also occur through cross-contamination of fomites, such as fish processing and handling equipment (OIE 2016a).

The likelihood of exposure to HPR-deleted ISAV is assessed as non-negligible.

18.2.3. Consequence assessment

HPR0 ISAV

The HPR0 isolate of ISAV causes sub-clinical infection in Atlantic salmon, Atlantic cod, brown trout, Coho salmon, rainbow trout, pollock and Pacific herring (Table 14), so the consequences of introduction of HPR0 ISAV are likely to be minimal. While the role of HPRO ISA in the re-emergence of the pathogenic HPR-deleted form is supported by the absence of HPR0 isolates in fish clinically infected with ISAV (EFSA 2102) the causality currently remains unproven (OIE 2016a).

Based on current evidence, the consequence of establishment of HPR0 ISAV is assessed as negligible.

HPR-Deleted ISAV

The HPR-deleted isolate of ISAV is the agent of a serious OIE-listed economic disease primarily affecting Atlantic salmon aquaculture (Munday 2002; Rimstad *et al.* 2011; OIE 2016a), while Coho salmon, rainbow trout, brown trout and Pacific herring (Table 19) may also be affected (OIE 2016a).

Salmonid aquaculture in New Zealand is focused on Chinook salmon (*O. tshawytscha*), which is considered resistant to ISAV (Rimstad *et al.* 2011; Norman *et al*. 2013). Atlantic salmon are restricted to the upper Waiau catchment, while Coho salmon are no longer present in New Zealand and sockeye salmon are essentially restricted to isolated brood stocks in hatcheries (McDowall 2000; NIWA 2014a, 2014b).

Freshwater aquaculture in New Zealand is dependent upon clean uncontaminated water supplies (Sim-Smith *et al*. 2014) so the introduction of ISAV may spread rapidly in catchments. It may also spread rapidly in coastal fisheries waters (EFSA 2012), potentially affecting Pacific herring (*C. harengus*).

Introduction of HPR-deleted ISAV may affect rainbow trout (*O. mykiss*) and brown trout (*S. trutta*), particularly in a naïve population (OIE 2016a), with significant social and environmental implications for the recreational and tourist fisheries for these species (Fish & Game 2014). While the dollar value of New Zealand's freshwater fisheries is not fully known, the Lake Taupo trout fishery alone is valued at $70–80 million (Marsh & Mkwra 2013).

The consequence of the introduction of HPR-deleted ISAV in the commodity is assessed as non-negligible.

18.2.4. Risk Estimation

HPR0 ISAV

HPR0 ISAV does not cause clinical disease and the possible risks associated with its reversion to the HPR-deleted form are presently unclear. As the consequence assessment for HPR0 ISAV is negligible, then the risk estimation is negligible and HPR0 ISAV is not assessed to be a risk in the commodity.

HPR-Deleted ISAV

As the entry, exposure and consequence assessments for HPR-deleted ISAV are non-negligible, the risk estimate is non-negligible. HPR-deleted ISAV is assessed to be a risk in the commodity and risk management measures may be justified.

18.3. Risk management

HPR-deleted ISAV has been assessed to be a risk in the commodity. Infection with ISAV is a notifiable disease, so the *Aquatic Code* (OIE 2016a) provides guidance for importing eviscerated fish from infected countries and the specific processing requirements to ensure destruction of the virus.

The HPR-deleted form of ISAV is reported from three families of wild and farmed fish (Table 14), which may be present in the commodity. Species declaration should substantially reduce the occurrence of HPR-deleted ISAV in the commodity and be a viable risk management option. Restriction of the commodity to wild-caught fish (not from aquaculture) would have little, or no effect on pathogen load and is not a viable risk management option.

Article 10.5.3 of the OIE *Aquatic Code* (OIE 2016a) states that:

Competent Authorities should not require any conditions related to ISA, regardless of the ISA status of the exporting county, zone or compartment, when authorising the importation or transit of the following aquatic animals and aquatic animal products from the species referred to in Article 10.6.2. which are intended for any purpose and which comply with Article 5.4.1.:

> *a. heat sterilised, hermetically sealed fish products (i.e. a heat treatment at 121°C for at least 3.6 minutes or any time/temperature equivalent);*
> *b. pasteurised fish products that have been subjected to a heat treatment at 90°C for at least ten minutes (or any time/temperature equivalent which has been demonstrated to inactivate ISAV);*

c. *mechanically dried, eviscerated fish (i.e. a heat treatment at 100°C for at least 30 minutes or any time/temperature equivalent which has been demonstrated to inactivate ISAV).*

Furthermore, Article 10.5.11 states that:

Competent Authorities should not require any conditions related to ISA, regardless of the ISA status of the exporting country, zone or compartment, when authorising the importation or transit of fish fillets or steaks (frozen or chilled) which have been prepared and packaged for retail trade and which comply with Article 5.4.2.

Compliance with Articles 10.5.3 and 10.5.11 would eliminate HPR-deleted ISAV from the commodity and be a viable risk management option.

HPR-deleted ISAV has been reported from Europe, Canada and South America. As an OIE-listed disease, country freedom is well defined. Where a declaration of country/zone freedom is approved through the MPI Country Approval Procedures, this option is likely to substantially reduce the occurrence of HPR-deleted ISAV in the commodity. Approval of country/zone freedom by MPI is a viable risk management option.

In the commodity, HPR-deleted ISAV is mainly associated with fish heads (OIE 2016b), so removal of the gills should slightly reduce pathogen load. Removal of the head and gills should moderately reduce the pathogen load of HPR-deleted ISAV and is a viable risk management option.

HPR-deleted ISAV is resistant to freezing (OIE 2016b), so frozen storage would have no effect on pathogen occurrence. ISAV requires high-temperature treatment for denaturation (OIE 2016a), so heating (to at least 56°C for at least 30 minutes) would eliminate the pathogen from the commodity. Heat treatment is a viable risk management option.

Commercially available vaccines against HPR-deleted ISAV do not provide complete protection (OIE 2016b). Vaccination is not regarded as a viable risk management option and is not discussed further.

HPR-deleted ISAV may be transmitted through the waste products associated with transport, storage and processing of the commodity (OIE 2016a). The requirement that all wash and wastewater discharges be appropriately chemically treated (e.g., with iodophors) before discharge, and that all solid wastes, tissue scraps and offal be disposed of through a recognised trade waste disposal procedure would be viable management options.

18.3.1. Risk management options

The virus HPR-deleted ISAV is reported from fish in families Clupeidae, Gadidae and Salmonidae (Table 14), which are considered likely to be present in the commodity. Other families have not been associated with HPR-deleted ISAV. Therefore, species declaration indicating the commodity is not originated from any of the above families should substantially reduce the occurrence of HPR-deleted ISAV in the commodity.

For the commodities originated from families associated with HPR-deleted ISAV, one or a combination of the following additional options could also be considered to effectively manage the risk.

Where country/zone freedom from HPR-deleted ISAV is accepted by MPI:

Option 1

Acceptance of country/zone freedom by MPI should substantially reduce the occurrence of HPR-deleted ISAV, so the commodity may be imported without any further restrictions.

Where country/zone freedom from HPR-deleted ISAV is not accepted by MPI or not available:

Option 2

Processing consistent with the conditions of Article 10.5.3 or 10.5.11 of the OIE *Aquatic Code* (OIE 2016a) should eliminate HPR-deleted ISAV. When these provisions are met, the commodity could be imported without any further restrictions.

Where the imported commodity is not consistent with Article 10.5.3 or 10.5.11 of the OIE Aquatic *Code* (OIE 2016a), further processing is necessary:

Option 3

Heat treatment (by cooking to at least 56°C for 30 minutes) should eliminate HPR-deleted ISAV. When this provision is met, the commodity could be imported without any further restrictions; or,

Option 4

Further processing (by removal of the head and gills) should moderately reduce the occurrence of HPR-deleted ISAV. When this provision is met, the commodity could be imported without any further restrictions.

18.4. References

Anonymous (2016). Biosecurity (Notifiable Organisms) Order 2016. NZLR/73. New Zealand Government, Wellington, New Zealand, 13 p.

Biacchesi S, Le Berre M, Le Guillou S, Benmansour A, Bremont M, Quillet E, Boudinot P (2007). Fish genotype significantly influences susceptibility of juvenile rainbow trout, *Oncorhynchus mykiss* (Walbaum), to waterborne infection with infectious salmon anaemia virus. *Journal of Fish Diseases* 30: 631–636.

Christiansen DH, Østergaard PS, Snow M, Dale OB, Falk K (2011). A low-pathogenic variant of infectious salmon anaemia virus (ISAV1 - HPR0) is highly prevalent and causes a non-clinical transient infection in farmed Atlantic salmon (*Salmo salar* L.) in the Faroe Islands. *Journal of General Virology* 92: 909–918.

Cunningham CO, Gregory A, Black J, Simpson I, Raynard RS (2002). A novel variant of the infectious salmon anaemia virus (ISAV) haemagglutinin gene suggests mechanisms for virus diversity. *Bulletin of the European Association of Fish Pathologists* 22: 366–374.

Diggles BK, Hine PM, Handley S, Boustead NC (2002). A handbook of diseases of importance to aquaculture in New Zealand. NIWA Science and Technology Series 49. National Institute of Water and Atmospheric Research, Auckland, New Zealand, 200 p.

European Food Safety Authority (EFSA) (2012). Scientific opinion on infectious salmon anaemia (ISA). EFSA Panel on Animal Health and Welfare. *EFSA Journal* 10(11): 2971, 22 p.**Fish and Game (2014).** New Zealand Fish and Game Council [Online] Available from: http://www.fishandgame.org.nz/ [Accessed 24 September 2014].

Kahn SA, Beers PT, Findlay VL, Peebles IR, Durham PJ, Wilson DW, Gerrity SE (1999). Import risk analysis on non-viable salmonids and non-salmonid marine finfish. Australian Quarantine and Inspection Service, Department of Agriculture, Fisheries and Forestry, Canberra, Australia, 409 p.

Kibenge F, Garate O, Johnston G, Arriagada K, Kibenge M, Wadowaka D (2001). Isolation and identification of infectious salmon anaemia virus (ISAV) from Coho salmon in Chile. *Diseases of Aquatic Organisms* 45: 9–18.

Lyngstad TM, Kristoffersen AB, Hjortaas MJ, Devold M, Aspehaug V, Larssen RB, Jansen PA (2012). Low virulent infectious salmon anaemia virus (ISAV) is prevalent and geographically structured in Norwegian salmon farming. *Diseases of Aquatic Organisms* 101: 197–206.

Marsh D, Mkwra L (2013). Review of freshwater non-market value studies. Department of Economics, University of Waikato, Hamilton, New Zealand, 144 p.

Marshall SH, Ramirez R, Labra A, Carmona M. Munoz C (2014). Bona-fide evidence for natural vertical transmission of infectious salmon anaemia virus in freshwater brood stocks of farmed Atlantic salmon (*Salmo salar*) in Southern Chile. *Journal of Virology* 88(11): 6012–6018.

McDowall RM (1994). The origins of New Zealand's Chinook salmon, *Oncorhynchus tshawytscha*. *Marine Fisheries Review* 56(1): 1–7.

McDowall RM (2000). The Reed field guide to New Zealand freshwater fishes. Reed, Auckland, New Zealand, 256 p.

Munday BL (2002). Scientific review of significant disease agents carried by non-viable, non-salmonid freshwater and estuarine finfish and their products. Report prepared for Biosecurity Australia. DAFF (Department of Agriculture, Fisheries and Forestry, Canberra, Australia, 120 p.

NIWA (2014a). Atlantic salmon. NIWA Fish Atlas. National Institute of Water and Atmospheric Research, Auckland, New Zealand [Online] Available from: https://www.niwa.co.nz/freshwater-and-estuaries/nzffd/NIWA-fish-atlas/fish-species/atlantic_salmon [Accessed 15 April 2015].

NIWA (2014b). Sockeye salmon. NIWA Fish Atlas. National Institute of Water and Atmospheric Research, Auckland, New Zealand [Online] Available from: https://www.niwa.co.nz/freshwater-and-estuaries/nzffd/NIWA-fish-atlas/fish-species/sockeye_salmon [Accessed 15 April 2015].

Norman RE, Brosnahan C, Fischer J, Frazer J, Johnston C, Jones B, Keeling S, Pande A, Pulford D, Rawdon T, Williams R (2013). Salmon mortality investigation REW-1017, Pelorus Sound. *MPI Technical Paper* 2013/19. Ministry for Primary Industries, Wellington, New Zealand, 14 p.

Nylund A, Hovland T, Hodneland F, Lovik FN (1994). Mechanism for transmission of infectious salmon anaemia (ISA). *Diseases of Aquatic Organisms* 19: 95–100.

OIE (2016a). Aquatic Animal Health Code, 19th Edition. World Organization for Animal Health. Paris, France [Online]. Available from: http://www.oie.int/international-standard-setting/aquatic-code/access-online/ [Accessed 15 March 2017].

OIE (2016b). Manual of diagnostic tests for aquatic animals. World Organization for Animal Health. Paris, France [Online]. Available from: http://www.oie.int/international-standard-setting/aquatic-manual/access-online/ [Accessed 17 March 2017].

Oye AK, Rimstad E (2001). Inactivation of infectious salmon anaemia virus, viral haemorrhagic septicaemia virus and infectious pancreatic necrosis virus in water using UVC irradiation. *Diseases of Aquatic Organisms* 48: 1–5.

Rimstad E, Dale OB, Dannevig BH, Falk K (2011). Infectious salmon anaemia. pp 143–165. *In*: Woo PTK, Bruno DW (*Eds.).* Fish Diseases and Disorders: Volume 3 - Viral, Bacterial and Fungal Infections, 2nd edition. CABI Publishers, Wallingford, United Kingdom, 930 p.

Rolland JB, Winton JR (2003). Relative resistance of Pacific salmon to infectious salmon anaemia virus. *Journal of Fish Diseases* 26(9): 511–520.

Sim-Smith C, Faire S, Lees A (2014). Managing biosecurity risk for business benefit. Report prepared for the Ministry for Primary Industries by Coast and Catchment Ltd, Auckland, New Zealand, 214 p.

Stone, MAB, MacDiarmid SC, Pharo HJ (1997). Import health risk analysis: salmonids for human consumption. Ministry of Agriculture Regulatory Authority, New Zealand, 269 p.

Tubbs L, Lee P, Diggles B, Jones JB, Sheppard M, Sim-Smith C (2007). A review of aquatic diseases of significance to New Zealand. Final research report for project ZBS 2005-17. National Institute of Water and Atmospheric Research, Wellington, New Zealand, 452 p.

Vike S, Nylund S, Nylund A (2009). ISA virus in Chile: Evidence of vertical transmission. *Archives in Virology* 154: 1–8.

19. Koi herpesvirus

19.1. Hazard identification

19.1.1. Aetiological agent

Koi herpesvirus (KHV), or cyprinid herpesvirus-3 (CyHV-3), is a double-stranded DNA herpesvirus classified within family Alloherpesviridae. It was first reported from the United Kingdom in 1996 and from Israel and Germany in 1998 (Bretzinger *et al.* 1999; Hedrick *et al.* 2000; Perelberg *et al.* 2003). It is closely related to carp-pox virus (CyHV-1) and goldfish haematopoietic necrosis virus (CyHV-2) (OIE 2016b).

Three main isolates are recognised: KHV-J from Japan; KHV-I from Israel; and KHV-U from the United States (Gotesman *et al.* 2013), while further isolates have been identified from Indonesia (Sunarto *et al.* 2011) and South Korea (Han *et al.* 2013). Mortality, host susceptibility and host tissue preference vary among strains (Yuasa *et al.* 2008), although these have become widely distributed through carp broodstock transfer for aquaculture, and the international trade in live ornamental fish (Haenen *et al.* 2004; Sano *et al.* 2004; Bigarré *et al.* 2009; Ilouze *et al.* 2011; Sunarto *et al.* 2011, OIE 2016b).

19.1.2. OIE status

KHV disease is listed by the OIE as a notifiable disease (OIE 2016a).

19.1.3. New Zealand status

KHV disease has not been reported from New Zealand (Johnston 2009) and is considered exotic. Infection with KHV is a notifiable disease in New Zealand (Anon. 2016).

19.1.4. Epidemiology

Infection with KHV has been reported from Continental Europe and the United Kingdom, as well as Romania, Slovenia and Spain. It is also reported from Asia, including China, Hong Kong, Indonesia, Japan, South Korea, Malaysia, Singapore and Taiwan, as well as from Canada, South Africa and the United States. It is likely to be present in other countries but has not yet been identified or reported (OIE 2016b).

KHV is highly infectious, affecting adults and juvenile cyprinids including wild and farmed common and ornamental varieties of carp (*Cyprinus carpio*),. It has caused mass mortalities in farmed carp populations in Israel, Europe and the United States (Gilad *et al.* 2002), Japan (Sano *et al*. 2004) and in ornamental fish populations worldwide (Michel *et al.* 2010; OIE 2016b). It also infects goldfish (*Carassius auratus*) (Gotesman *et al.* 2013) and carp-goldfish hybrids, but at lower rates of mortality (Hedrick *et al*. 2006; Bergmann *et al.* 2010; El-Matbouli & Soliman 2011; Klempter *et al.* 2012; OIE 2016b).

A range of freshwater fish species and families are susceptible to KHV (Table 15) (Kempter & Bergman 2007; Bergmann *et al*. 2009; Kempter *et al.* 2009, 2012; Radosavljević *et al.* 2012; Piačková *et al.* 2013, 2015; Bergmann *et al*. 2016).

Table 15. Families and Species of Fish Susceptible to Koi Herpesvirus (KHV)

Family	Host Species
Acipenseridae	Sturgeon (*Acipenser gueldenstaedtii, A. oxyrinchus, A. ruthenus*)
Cyprinidae	Barbel (*Barbus barbus*), belica (*Leucaspius delineatus*), bighead carp (*Hypophthalmichthys nobilis*), carp (*Cyprinus carpio*), carp bream (*Abramis brama*), chub (*Squalius cephalus*), crucian carp (*Carassius carassius*), dace (*Leuciscus leuciscus*), grass carp (*Ctenopharyngodon idella*), gudgeon (*Gobio gobio*), ide (*Leuciscus idus*), nace (*Chondrostoma nacus*), Prussian carp (*Carassius gibelio*), roach (*Rutilus rutilus*), silver carp (*Hypophthalmichthys molitrix*), tench (*Tinca tinca*), vimba bream (*Vimba vimba*)
Percidae	European perch (*Perca fluviatilis*), ruffe (*Gymnocephalus cernua*)
Siluridae	Sheatfish (*Silurus glanis*)

KHV is reported from invertebrate vectors including rotifers (*Rotifera* spp.), swan mussels (*Anodonta cygnea*) and freshwater shrimp (*Gammarus pulex*) (Kielpinski *et al.* 2010). Filter feeding molluscs and crustaceans passively concentrate KHV in their digestive tract and can function as reservoirs of disease, infecting the carp when these invertebrates are consumed (Kielpinski *et al.* 2010; Gotesman *et al.* 2013).

Infection with KHV affects adult and juvenile fish, typically with high morbidity (up to 100%) and mortality (80–100%) (OIE 2016a). Mortality varies between isolates (Yuasa *et al.* 2008), but is highest where viral replication is optimised at water temperatures between 16°C and 25°C (OIE 2016a). Outside this range (i.e. above 30°C, or below 13°C), KHV becomes dormant and clinical signs generally cease (Gilad *et al.* 2002, 2004), but surviving fish remain latent carriers of disease (Gilad *et al.* 2004; Haenen *et al.* 2004; Gotesman *et al.* 2013; OIE 2016b).

Disease transmission is horizontal, mainly by cohabitation, through the water column (OIE 2016a). The virus is shed in the faeces and urine, or released across the skin and gill mucosa of infected fish (Hartman *et al.* 2014). Vertical transmission may also occur in hatcheries through infected eggs (OIE 2016b).

Pathogen entry after direct contact occurs across the epithelium of the skin, gill, fins (Costes *et al.* 2009), or across the pharyngeal mucosa following the consumption of an infected fish or invertebrate vector (Pikarsky *et al.* 2004; Gotesman *et al.* 2013). Disease onset usually occurs after 8–21 days, although this may occur in 2-3 days when temperatures are optimised (Hedrick *et al.* 2000).

External clinical signs may include excess mucous production, skin ulceration, sunken eyes, necrotic lesions and haemorrhages of fin and gill tissues (Hartman *et al.* 2014; OIE 2016a), although infection may also progress without clinical signs of disease (Li Po 2011).

While the skin and gills are important sites of viral pathogenesis and sources of virus shedding (OIE 2016a), KHV infection progresses to the spleen, liver, gut and interstitial tissues (Hartman *et al.* 2014). KHV has been isolated 62 days post-exposure in the brain, mucus, liver, gill, heart tissues and leucocytes of surviving fish (Gilad *et al.* 2004; OIE 2016a). KHV may remain viable in host brain tissue for at least 360 days post-infection (Yuasa *et al.* 2008) and enter a latent state in leucocytes, re-emerging up to 7 months later, when the host becomes stressed (Gotesman *et al.* 2013; Hartman *et al.* 2014; OIE 2016a).

Herpesviruses are environmentally stable and KHV is unaffected by freezing (to -80°C) (Gilad *et al.* 2002). KHV remains viable for at least 4 hours in water temperatures of 23–25°C (Perelberg *et al*. 2003) and KHV DNA has been reported in river water four months post-infection (Haramoto *et al.* 2007). However, the detection of viral DNA may not indicate the presence of infectious virus

(OIE 2016a). Viral titre is reduced in both water and sediment after 3 days at 15°C, largely through bacterial predation, although viability is enhanced by the presence of fomites, such as organic materials and microbial films on aquaculture and transport equipment (Shimizu *et al.* 2006; Haramoto *et al.* 2007; Rathore *et al.* 2012; OIE 2016a).

KHV may be inactivated by UV radiation (4.0 $\mu Ws/cm^2$), or by high-temperature treatment (cooking to at least 121°C for at least 3.6 minutes, or 90°C for at least 10 minutes). Disinfection also provides effective inactivation, using iodophor treatment (200 mg L^{-1} for 20 minutes); benzalkonium chloride (at 60 mg L^{-1} for 20 minutes); ethyl alcohol (33% for 20 minutes); sodium hypochlorite (at 200 mg L^{-1} for 30 seconds); or quaternary ammonium compounds (500 mg L^{-1} for one hour), at 15°C (Kasai *et al.* 2005; Noga 2010; OIE 2016a).

No effective treatment for KHV exists (OIE 2016b). Early vaccination methods combined with temperature manipulation treatments reduce external signs of disease (Ronen *et al*. 2003), but surviving fish remain carriers (Hartman *et al.* 2014). Attenuated live KHV vaccine confers 80–95% relative survival but this provides only short-term protection (8 months) (Perelberg *et al.* 2003, 2005). The vaccine may revert to the pathogenic form (Rathore *et al*. 2012) and sub-clinically infected vaccinated fish remain carriers of disease (Gotesman *et al.* 2013; Hartman *et al.* 2014).

Susceptible species in New Zealand include introduced common and ornamental carp (*C. carpio*), goldfish (*C. auratus*) and their hybrids, as well as grass carp (*Ctenopharyngodon idella*) (Tubbs *et al.* 2007; OIE 2016a). No data are available on the susceptibility of New Zealand native fish to KHV (Johnston 2009), although recent studies carried out on Australian freshwater fish (McColl *et al*. 2007; B. Jones, MPI, *pers. comm.* 2016) suggest the host range of KHV is limited to the introduced species listed above.

19.2. Risk assessment

19.2.1. Entry assessment

Common carp (*C. carpio*) is the third most important freshwater fish species internationally farmed for human consumption, while ornamental carp varieties are extensively farmed and traded internationally (OIE 2016a). These species are susceptible to infection with KHV, often with high mortality (Gotesman *et al.* 2013; OIE 2016a).

Fish clinically infected with KHV typically show few or no external signs of disease (Rathore *et al.* 2012; Gotesman *et al.* 2013). These may pass visual inspection and enter the human food consumption pathway. KHV is resistant to freezing and chilled storage (OIE 2016b) and it is likely that KHV present in eviscerated fish would remain viable in the commodity.

The likelihood of entry through the commodity is assessed as non-negligible.

19.2.2. Exposure assessment

To establish infection in New Zealand, infected eviscerated product would have to become available for consumption by a susceptible freshwater fish host, in sufficient quantity and duration (Kahn *et al.* 1999). KHV may be present in the blood-water discharge as well as in the offal derived from commercial fish processing, including gill tissues, neural tissues of the brain and spinal cord and the skin, as well as in connective tissue and skeletal muscle (OIE 2016b) discarded

after trimming. KHV remains viable in fresh water and in sediment for up to 4 months. Viability is also enhanced by the presence of biofilms and organic materials and viral material may be transported by fomites (Haramoto *et al.* 2007; OIE 2016a, 2016b).

The likelihood of exposure to KHV is assessed as non-negligible.

19.2.3. Consequence assessment

KHV is an OIE-listed pathogen that is relatively host-specific, affecting common and ornamental varieties of carp (*C. carpio*), goldfish (*Carassius auratus*) and carp-goldfish hybrids. It also affects grass carp (*Ctenopharyngodon idella*), ide (*Leuciscus idus*), ornamental catfish (*Ancistrus* sp.) and sturgeon (*Acipenser gueldenstaedtii, A. oxyrinchus*) (Heydrick *et al*. 2006; Johnston 2008, 2008; Bergmann *et al.* 2010; El-Matbouli & Soliman 2007, 2011; OIE 2016b).

Common carp (*C. carpio*) are not currently farmed in New Zealand while ornamental varieties are prohibited fish (Anon. 2016). Feral carp derived from released ornamental varieties (*C. carpio koi* and *C. carpio goi* and hybrids) are present in the freshwater systems of the North Island (NIWA 2015), but are unwanted organisms under the Biosecurity Act (Anon. 2016).

The consequences of establishment of KHV for New Zealand indigenous freshwater fish are unknown (Johnston 2009). Recent studies on susceptibility of Australian native freshwater fish carried out as part of an assessment of the potential use of KHV as a biological control agent suggest the host range of KHV is limited to the introduced species listed above (McColl *et al.* 2007; Fulton 2013; CSIRO 2015; B. Jones, MPI, *pers. comm.* 2016)

The establishment of KHV in New Zealand would have a significant negative impact upon the freshwater farming industries associated with the acclimatised grass carp (*Ctenopharyngodon idella*), used in New Zealand for aquatic weed control and a significant negative impact on industries associated with production of ornamental goldfish (*Carassius auratus*) (Clayton & Wells 1999; NIWA 2015). The other host species including ide (*Leuciscus idus*), ornamental catfish (*Ancistrus* sp.) and sturgeon (*Acipenser gueldenstaedtii, A. oxyrinchus*) (OIE 2016a) are not present in New Zealand.

The consequences of establishment of KHV is assessed as non-negligible.

19.2.4. Risk estimation

Since the entry, exposure and consequence assessments for KHV are non-negligible, the risk estimate is non-negligible. Therefore, KHV is assessed to be a risk in the commodity and risk management measures may be justified.

19.3. Risk management

KHV is reported from wild and farmed fish from four families (Table 15), which may be present in the commodity (OIE 2016a). Species declaration would substantially reduce the occurrence of KHV in the commodity and be a viable risk management option. Restriction of the commodity to wild-caught fish (not from aquaculture) would have little, or no effect on pathogen load and is not a viable risk management option.

KHV is widely distributed in freshwater systems worldwide (OIE 2016a). As an OIE-listed disease it is assumed that the Competent Authority of the exporting country has a monitoring and

surveillance programme to demonstrate country/zone freedom. Where approved under the MPI Country Approvals Procedures, this option should substantially reduce the occurrence of KHV in the commodity. Approval of a declaration of country/zone freedom by MPI is a viable risk management option.

KHV is unaffected by freezing, so frozen storage is not a viable risk management option.

KHV is likely to be present in the blood-water discharge, as well as in the head (brain and gill tissues), spinal column and skin, as well as in the trimmings of the skeletal musculature and connective tissue that comprise fish offal. Removal of the head and gills would slightly reduce pathogen load of KHV in the commodity. The skin is recognised as an active site of KHV infection, therefore additional processing to the skin-off fillet state would moderately reduce pathogen load and be a viable risk management option.

KHV is a notifiable disease (OIE 2016a), and article 10.7.2 of the *Aquatic Code* provides guidance on importing eviscerated fish of the following species: common carp and subspecies (*Cyprinus carpio*), and common carp hybrids (e.g. *Cyprinus carpio x Carassius auratus*). It is noted that the recommendations also apply to any other susceptible species referred to in the *Aquatic Manual* when traded internationally.

Article 10.7.3 of the Aquatic Code recommends:

For importation or transit of aquatic animals and aquatic animal products for any purpose from a country, zone or compartment not declared free from koi herpesvirus disease:

1. *Competent Authorities should not require any conditions related to KHVD, regardless of the KHVD status of the exporting country, zone or compartment, when authorising the importation or transit of the following aquatic animals and aquatic animal products from the species referred to in Article 10.7.2 which are intended for any purpose and which comply with Article 5.4.1:*
    a. *heat sterilised hermetically sealed fish products (i.e. a heat treatment at 121°C for at least 3.6 minutes or equivalent);*
    b. *pasteurised fish products that have been subjected to heat treatment at 90°C for at least ten minutes (or equivalent);*
    c. *mechanically dried eviscerated fish (i.e. a heat treatment at 100°C for at least 30 minutes or equivalent).*

2. *When authorising the importation or transit of aquatic animals and aquatic animal products of a species referred to in Article 10.7.2, other than those referred to in point 1 of Article 10.7.3, Competent Authorities should require the conditions prescribed in Articles 10.7.7 to 10.7.12 relevant to the KHVD status of the exporting country, zone or compartment.*

3. *When considering the importation or transit of aquatic animals and aquatic animal products of a species not covered in Article 10.7.2 but which could reasonably be expected to pose a risk of spread of KHVD, the Competent Authority should conduct a risk analysis in accordance with the recommendations in Chapter 2.1. The Competent Authority of the exporting country should be informed of the outcome of this assessment.*

Article 10.7.11 of the *Aquatic Code* recommends:

For importation of aquatic animals and aquatic animal products for retail trade for human consumption from a country, zone or compartment not declared free from koi herpesvirus disease:

1. *Competent Authorities should not require any conditions related to KHVD, regardless of the KHVD status of the exporting country, zone or compartment, when authorising the importation or transit of fish fillets or steaks (chilled or frozen) which have been prepared and packaged for retail trade and which comply with Article 5.4.2.*

   Certain assumptions have been made in assessing the safety of the aquatic animal products mentioned above. Member Countries should refer to these assumptions at Article 5.4.2 and consider whether the assumptions apply to their conditions.

   For these commodities Member Countries may wish to consider introducing internal measures to address the risks associated with the commodity being used for any purpose other than for human consumption.

2. *When importing aquatic animals or aquatic animal products, other than those referred to in point 1 above, of species referred to in Article 10.7.2 from a country, zone or compartment not declared free from KHV, the Competent Authority of the importing country should assess the risk and apply appropriate risk mitigation measures.*

Compliance with Articles 10.7.3 and 10.7.11 would eliminate KHV from the commodity and be a viable risk management option.

KHV may be transmitted through the waste products associated with transport, storage and processing of the commodity. The requirement that all wash and wastewater discharges be appropriately chemically treated (e.g. with iodophors) before discharge, and that all solid wastes, tissue scraps and offal be disposed of through a recognised trade waste disposal procedure, would be viable management options.

19.3.1. Risk management options

Koi herpesvirus (KHV) is reported from fish in families Acipenseridae, Cyprinidae, Percidae and Siluridae (Table 15), which are considered likely to be present in the commodity. Other families have not been associated with KHV. Therefore species declaration indicating the commodity is not originated from any of the above families should substantially reduce the occurrence of KHV in the commodity.

For the commodities originated from families associated with KHV, one or a combination of the following additional options could also be considered to effectively manage the risk.

Where country/zone freedom from KHV is accepted by MPI:

Option 1

Acceptance of country/zone freedom should substantially reduce the occurrence of KHV, so the commodity may be imported without any further restrictions.

Where country/zone freedom from KHV is not accepted by MPI or not available:

Option 2

Processing consistent with the conditions of Articles 10.7.3 or 10.7.11 of the OIE Aquatic *Code* (OIE 2016a) should eliminate KHV. When these provisions are met, the commodity could be imported without further restrictions.

Where the imported commodity is not consistent with Article 10.7.3 or 10.7.11 of the OIE Aquatic Code (OIE 2016a), further processing is necessary:

Option 3

Further processing (to the skin-off fillet state) should moderately reduce the occurrence of KHV. When this provision is met, the commodity could be imported without further restrictions.

19.4. References

Anonymous (2016). Biosecurity (Notifiable Organisms) Order 2016. NZLR 2016/73. New Zealand Government, Wellington, New Zealand, 13 p.

Bergmann SM, Kempter J, Riechardt M, Fichtner D (2016). Investigation on the host specificity of koi herpesvirus (KHV) infection [Online] Available from: http://www.moag.gov.il/nr/rdonlyres/cb09d92f-0204-4ae0-b76c-a0ce9a91d157/0/host_specificity.pdf [Accessed 21 January 2016].

Bergmann SM, Sadowski J, Kielpinski M, Bartlomiejczyk M, Fichtner D, Riebe R, Lenk M, Kempter J (2010). Susceptibility of koi x crucian carp and koi x goldfish hybrids to koi herpesvirus and the development of KHV disease. *Journal of Fish Diseases* 33: 267–272.

Bergmann SM, Schutze H, Fischer U, Fichtner D, Riechardt M, Meyer K, Schrudde D, Kempter J (2009). Detection of koi herpes virus (KHV) genome in apparently healthy fish. *Bulletin of the European Association of Fish Pathologists* 29: 145–152.

Bigarré L, Baud M, Cabon J, Antychowicz J, Bergmann SM, Engelsma M, Pozet F, Reichert M, Castric J (2009). Differentiation between cyprinid herpesvirus type-3 lineages using duplex PCR. *Journal of Virological Methods* 158: 51–57.

Bretzinger A, Fisher ST, Oumouma M, Hoffmann R, Truyen U (1999). Mass mortalities in koi, *Cyprinus carpio*, associated with gill and skin disease. *Bulletin of the European Association of Fish Pathologists* 19: 182–185.

Clayton JS, Wells RDS (1999). Some issues in risk assessment reports on grass carp and silver carp. Conservation Advisory Science Notes No. 257. Department of Conservation, Wellington, New Zealand, 14 p.

Costes B, Raj VS, Michel B, Fournier G, Thirion M, Gillet L, Mast J, Lieffrig F, Bremont M, Vanderplasschen A. (2009). The major portal of entry of koi herpesvirus in *Cyprinus carpio* is the skin. *Journal of Virology* 83(7): 2819–2830.

CSIRO (2015). Reducing Australia's carp invasion [Online]. Available from: http://www.csiro.au/en/Research/BF/Areas/Managing-the-impacts-of-invasive-species/Biological-control/Biocontrol-of-carp [Accessed 15 January 2015].

El-Matbouli M, Saleh, M, Soliman H (2007). Detection of cyprinid herpesvirus type 3 in goldfish cohabiting with CyHV-3-infected koi carp (*Cyprinus carpio koi*). *Veterinary Record* 161(23): 792–793.

El-Matbouli M, Soliman H (2011). Transmission of cyprinid herpesvirus-3 (CyHV-3) from goldfish to naïve common carp by cohabitation. *Research in Veterinary Science* 90: 536–539.

European Food Safety Authority (EFSA) (2007). Scientific Opinion of the Panel on Animal Health and Welfare on a request from the European Commission on possible vector species and live stages of susceptible species not transmitting disease as regards certain fish diseases. *EFSA Journal* 584: 1–163.

Fulton W (2013). Pathways to adoption of KHV as a biological control agent for carp in Australia. PestSmart Toolkit publication, Invasive Animals Cooperative Research Centre, Canberra, Australia [Online]. Available from: http://www.pestsmart.org.au/pestsmart-factsheet-carp/ [Accessed 19 January 2016].

Gilad O, Yun S, Andree KB, Adkison MA, Zlotkin A, Bercovier H, Eldar A, Hedrick RP (2002). Initial characteristics of koi herpesvirus and development of a polymerase chain reaction assay to detect the virus in koi, *Cyprinus carpio koi*. *Diseases of Aquatic Organisms* 48(2): 101–108.

Gilad O, Yun S, Zagmutt-Vergara FJ, Leutenegger CM, Bercovier H, Hedrick RP (2004). Concentrations of a Koi herpesvirus (KHV) in tissues of experimentally infected *Cyprinus carpio koi* as assessed by real-time TaqMan PCR. *Diseases of Aquatic Organisms* 60(3): 179–187.

Gotesman M, Kattlun J, Bergmann SM, El-Matbouli M (2013). CyHV-3: the third cyprinid herpesvirus. *Diseases of Aquatic Organisms* 105: 163–174.

Haenen OLM, Way K, Bergmann SM, Ariel E. (2004). The emergence of koi herpesvirus and its significance to European aquaculture. *Bulletin of the European Association of Fish Pathologists* 24: 293–307**.**

Han JE, Kim JH, Renault T, Choresca C Jr, Shin SP, Jun JW, Park SC (2013). Identifying the viral genes encoding envelope glycoproteins for differentiation of cyprinid herpesvirus 3 isolates. *Viruses* 5(2): 568–576.

Haramoto E, Kitajima M, Katayama H, Ohgaki S (2007). Detection of koi herpesvirus DNA in river water in Japan. *Journal of Fish Diseases* 30(1): 59–61.

Hartman KH, Yanong RPE, Pouder DP, Petty BD, Francis-Floyd R, Riggs AC, Waltzek TB (2014). Koi herpesvirus (KHV). University of Florida IFAS Extension. Fish Diseases [Online] Available from: https://edis.ifas.ufl.edu/vm113 [Accessed 12 January 2015].

Hedrick RP, Gilad O, Yun S, Spangenberg JV, Marty GD, Nordhausen RW, Kebus MJ, Bercovier H, Eldar A (2000). A herpesvirus associated with mass mortality of juvenile and adult koi, a strain of a common carp. *Journal of Aquatic Animal Health* 12: 44–57.

Hedrick RP, Waltzek TB, McDowell TS (2006). Susceptibility of koi carp, common carp, goldfish and goldfish x common carp hybrids to cyprinid herpesvirus-2 and herpesvirus-3. *Journal of Aquatic Animal Health* **18:** 26–34.

Ilouze M, Davidovich M, Diamant A, Kotler M, Dishon A (2011). The outbreak of carp disease caused by CyHV-3 as a model for new emerging viral diseases in aquaculture: a review. *Ecological Research* 26: 885–892.

Johnston C (2008). Import risk analysis: Frozen, skinless and boneless fillet meat of *Oreochromis* spp. from China and Brazil for human consumption. MAF Biosecurity New Zealand, 96 p.

Johnston C (2009). Import Risk Analysis: Tropical, Subtropical and Temperate Freshwater and Marine Ornamental Fish and Marine Molluscs and Crustaceans - Review of Submissions and Supplementary Risk Analysis. Ministry of Agriculture and Forestry, Wellington, New Zealand, 157 p.

Kahn SA, Beers PT, Findlay VL, Peebles IR, Durham PJ, Wilson DW, Gerrity SE (1999). Import risk analysis on non-viable salmonids and non-salmonid marine finfish. Australian Quarantine and Inspection Service, Department of Agriculture, Fisheries and Forestry, Canberra, Australia, 409 p.

Kasai H, Muto Y, Yoshimizu M (2005). Virucidal effects of ultraviolet, heat treatment and disinfectants against koi herpesvirus. *Fish Pathology* 40: 137–138.

Kempter J, Bergmann SM (2007). Detection of koi herpesvirus (KHV) genome in wild and farmed fish from Northern Poland. *Aquaculture* 272: 275.

Kempter J, Sadowski J, Schutze H, Fischer U, Dauber M, Fichtner D, Panicz R, Bergmann SM (2009). Koi herpes virus: Do acipenserid restitution programs pose a threat to carp farms in the disease-free zones? *Acta Ichthyologica et Piscatoria* 19: 119–126.

Kempter J, Kielpinski M, Panicz R, Sadowski J, Mysłowski B, Bergmann SM (2012). Horizontal transmission of koi herpes virus (KHV) from potential vector species to common carp. *Bulletin of the European Association of Fish Pathologists* 32: 212–219.

Kielpinski M, Kempter J, Panicz R, Sadowski J, Schutze H, Ohlemeyer S, Bergmann SM (2010). Detection of KHV in freshwater mussels and crustaceans from ponds with KHV history in common carp (*Cyprinus carpio*). *Israel Journal of Aquaculture* 62: 28–37.

Li Po GD (2011). Recent developments in the study and surveillance of koi herpesvirus (KHV) in Asia. pp 13-28. *In:* Bondad-Reantaso MG, Jones JB, Corsin F, Aoki T (*Eds.*). Diseases in Asian Aquaculture VII. Fish Health Section, Asian Fisheries Society, Selangor, Malaysia, 385 p.

McColl KA, Sunarto A, Williams LM, Crane M (2007). Koi herpesvirus: Dreaded pathogen or white knight? *Aquaculture Health International* 9: 4–6.

Michel B, Fournier G, Liffrig F, Costes B, Vanderplasshen A (2010). Cyprinid herpesvirus3. *Emerging Infectious Diseases* 16(12): 1835–1843.

NIWA (2015). Koi carp. New Zealand freshwater fish database [Online] Available from: https://www.niwa.co.nz/our-science/freshwater/tools/fishatlas/species/fish-species/koi_carp [Accessed 12 January 2015].

Noga EJ (2010). Fish disease: diagnosis and treatment, Second Edition. Wiley-Blackwell, New York, United States of America, 536 p.

OIE (2016a). Aquatic Animal Health Code, 19th Edition. World Organization for Animal Health. Paris, France [Online] Available from: http://www.oie.int/international-standard-setting/aquatic-code/access-online/ [Accessed 15 March 2017].

OIE (2016b). Manual of diagnostic tests for aquatic animals. World Organization for Animal Health. Paris, France [Online] Available from: http://www.oie.int/international-standard-setting/aquatic-manual/access-online/ [Accessed 17 March 2017].

Perelberg A, Smirnov M, Hutoran M, Diamant A, Bejerano Y, Kotler M (2003). Epidemiological description of a new viral disease afflicting cultured *Cyprinus carpio* in Israel. *Israel Journal of Aquaculture* 55: 5–12.

Perelberg A, Ronen A, Hutoran M, Smith Y, Kotler M (2005). Protection of cultured *Cyprinus carpio* against a lethal viral disease by an attenuated virus vaccine. *Vaccine* 23: 3396–3403.

Piačková V, Flajšhans M, Pokorová D, Reschová S, Gela D, Čížek A, Veselý T (2013). Sensitivity of common carp, *Cyprinus carpio* L., strains and crossbreeds reared in the Czech Republic to infection by cyprinid herpesvirus 3 (CyHV-3; KHV). *Journal of Fish Diseases* 36: 75–80.

Piačková V, Pospíchal A, Pokorová D, Veselý T (2015). Sensitivity of various fish species to KHV. *Papers from the Carp Conference, Vodnany, 3–4 September 2015* [Online] Available from: http://www.frov.jcu.cz/carpconference/images/pdf/01_Piackova.pdf [Accessed 21 January 2016].

Pikarsky E, Ronen A, Abramowitz J, Levavi-Sivan B, Hutoran M, Shapira Y, Steinitz M, Perelberg A, Soffer D, Kotler M (2004). Pathogenesis of acute viral disease induced in fish by carp interstitial nephritis and gill necrosis virus. *Journal of Virology* 78: 9544–9551.

Radosavljević V, Jeremić S, Ćirković M, Lako B, Milićević V, Potkonjak A, Nikolin V (2012). Common fish species in polyculture with carp as cyprinid herpesvirus 3 carriers. *Acta Veterinaria (Belgrade)* 5–6: 675–681.

Rathore G, Kumar G, Swaminathan TR, Swain P (2012). Koi herpes virus: A review and risk assessment of Indian aquaculture. *Indian Journal of Virology* 23(2): 124–133.

Ronen A, Perelberg A, Abramowitz J, Hutoran M, Tinman S, Bejerano I, Steinitz M, Kotler M (2003). Efficient vaccine against the virus causing a lethal disease in cultured *Cyprinus carpio*. *Vaccine* 21: 4677–4684.

Sano M, Ito T, Kuriata J, Yani T, Wanabe N, Miwa S, Iida T (2004). First detection of koi herpesvirus in cultured common carp *Cyprinus carpio* in Japan. *Fish Pathology* 39: 165–167.

Shimizu T, Yoshida N, Kasai H, Yoshimizu M (2006). Survival of koi herpesvirus (KHV) in environmental water. *Japanese Society of Fish Pathologists* 41: 153–157.

Sunarto A, McColl KA, Crane MS, Sumiati T, Hyatt AD, Barnes AC, Walker PJ (2011). Isolation and characterization of koi herpesvirus (KHV) from Indonesia: Identification of a new genetic lineage. *Journal of Fish Diseases* 34: 87–101.

Tubbs L, Lee P, Diggles B, Jones JB, Sheppard M, Sim-Smith C (2007). A review of aquatic diseases of significance to New Zealand. Final research report for project ZBS 2005-17. National Institute of Water and Atmospheric Research, Wellington, New Zealand, 452 p.

Yuasa K, Ito T, Sano M (2008). Effect of water temperature on mortality and virus shedding in carp experimentally infected with koi herpesvirus. *Fish Pathology* 43: 83–85.

20. New Japan virus

20.1. Hazard identification

20.1.1. Aetiological agent

New Japan virus (NJV) was originally isolated from rainbow trout (*Oncorhynchus mykiss*) in northern Japan during 1991 (Oh *et al.* 1995a; Stone *et al*. 1997). While tentatively considered to be a rhabdovirus (Oh *et al.* 1995a), the virus remains uncharacterised (Kahn *et al.* 1999). No more recent information is available.

20.1.2. OIE Status

Diseases associated with New Japan virus are not listed by the OIE (OIE 2016a).

20.1.3. New Zealand status

New Japan virus is regarded as exotic (Stone *et al*. 1997), since it has not been reported from New Zealand.

20.1.4. Epidemiology

Fish hosts of NJV (Table 16) include species of families Salmonidae and Plecoglossidae in Japanese aquaculture (Oh *et al.* 1995a).

Table 16. Families and Species of Fish Susceptible to New Japan Virus

Family	Host Species
Plecoglossidae	Ayu (*Plecoglossus altivelis*)
Salmonidae	Coho salmon (*Oncorhynchus kisutch*), masu salmon (*O. masou*), rainbow trout (*O. mykiss*), whitespotted char (*Salvelinus leucomaensis*),

New Japan virus appears widespread in northern Japan, occurring in 10% of farmed masou salmon (*O. masou*) (Oh *et al.* 1995a). While Coho salmon occur in both sides of the northern Pacific ocean (from Hokkaido, Japan, around the Bering Sea, to Monterey Bay, California), NJV has only ever been reported from Japanese waters (Kahn *et al*. 1999).

Mortality varies from 6% to 34% for Coho salmon in waterborne experimental infection, but from 30%–63% by intermuscular injection, with mortalities occurring 15–30 days after infection (Oh *et al*. 1995b). Infection may reach 26% in aquaculture (Kahn *et al.* 1999). The minimum infective dose is $10^{3.2}$ $TCID^{50}$ ml^{-1} (tissue culture infective dose is required for 50% infection of host cells) (Oh *et al*. 1995b).

Clinically infected fish show few external signs of disease, other than mild exophthalmia and anorexia (Kahn *et al*. 1999). New Japan virus invades erythrocytes and infection is mainly associated with haematopoietic tissues, with haemorrhage and necrosis of renal tissues. It has also been recovered from the brain, where infection causes encephalopathy (Oh *et al.* 1995b). It also is reported from ovarian fluid and blood of infected fish (Oh *et al.* 1995a), as well as occurring at low titre in somatic muscle tissue (Kahn *et al*. 1999). While evisceration would substantially reduce viral tire, small quantities may remain in the somatic musculature and brain tissue of infected fish (Oh *et al*. 1995a; Kahn *et al.* 1999).

Little is known about the epidemiology of NJV (Oh *et al.* 1995a; Oh *et al.* 1995b; Kahn *et al.* 1999). Fish rhabdoviruses that also invade erythrocytes including viral haemorrhagic septicaemia virus (VHSV) are transmitted horizontally through the water column (OIE 2016b). Infection primarily occurs through the skin or gill epithelium, but infection may also result from the consumption of infected prey fish, or fish tissues (Kim & Faisal 2011). The presence of NJV in ovarian fluid of infected masu salmon suggests that vertical transmission may also occur (Oh *et al.* 1995b).

The optimum temperature for replication is 15°C (Oh *et al*. 1995a). New Japan virus is low temperature stable, remaining viable after storage at -80°C (Oh *et al.* 1995b). Fish rhabdoviruses are typically stable at low temperatures, remaining viable after frozen storage at -20°C for several years and are unaffected by multiple freeze-thaw cycles (Fraenkel-Conrat 2012).

No information is currently available on the viability of NJV after passage through the avian digestive tract in order to determine the likelihood of transfer from land-based offal disposal sites back to the aquatic environment. Fish rhabdoviruses are considered stable in fresh water for up to 5 months, at temperatures less than 20°C (Kim & Faisal 2011; Fraenkel-Conrat 2012). VHSV survived only for 14 days in fresh water and 4 days in sea water, but survival was greatly enhanced where the water contained organic material such as ovarian fluid (Kim & Faisal 2011).

While rhabdoviruses are generally denatured by disinfectants, such as ether, chloroform, formalin, glycerol, sodium hypochlorite and organic iodophor (Fraenkel-Conrat 2012), New Japan virus appears unaffected by ether or chloroform treatment. (Oh *et al*. 1995a). Most rhabdoviruses are denatured by strong acids (pH <4) or alkaline (pH> 10) treatments (Fraenkel-Conrat 2012), but new Japan virus is highly resistant to acids, being only partially inactivated by exposure to pH 1, and mild acids (pH in a range from 2 to 9) had no effect on viral replication.

Most rhabdoviruses are denatured by heat treatment (cooking to 60°C for at least 15 minutes) (Fraenkel-Conrat 2012). Although replication of NJV ceased at 30°C, it remained infective after heat treatment at 60°C for 30 minutes (Kahn *et al*. 1999).

No vaccine is currently available for New Japan Virus.

Potential hosts in New Zealand include rainbow trout (*O. mykiss*), although no information is currently available concerning the viability of viral transfer through the commodity to other potential host species such as Chinook salmon (*O. tshawytscha*) (Kahn *et al*. 1999).

20.2. Risk assessment

20.2.1. Entry assessment

New Japan virus is reported only from Coho salmon (*O. kisutch*), masu salmon (*O. masou*), rainbow trout (*O. mykiss*) and whitespotted char (*S. leucomaensis*) (Salmonidae) and ayu (*P. altivelis*) (Plecoglossidae) (Table 22) in Japanese aquaculture (Oh *et al.* 1995a). Infection appears widespread in Japan (affecting 10% of farmed masu salmon), with low-medium (6–34%) mortality (Oh *et al.* 1995a).

Rhabdoviruses are resistant to freezing and unlikely to be denatured by the standard storage and processing conditions associated with fish processing (Fraenkel-Conrat 2012). Infected fish may not show clinical signs of infection (Kahn *et al.* 1999). Sub-clinically infected fish would be likely to pass visual inspection and may be present in the commodity.

The likelihood of entry is assessed as non-negligible.

20.2.2. Exposure assessment

To establish infection through the commodity, infected eviscerated product would have to become available for consumption by a susceptible fish host, in sufficient quantity and duration (Kahn *et al*. 1999). While evisceration would reduce viral titre, NJV may be present in the brain and somatic muscle tissues of infected eviscerated fish (Kahn *et al*. 1999).

Rhabdoviruses are resistant to environmental extremes (Fraenkel-Conrat 2012) and may remain viable in blood-water discharges, trimmings of somatic musculature, as well as in the brain tissues discarded as offal after processing. No information is currently available on the viability of NJV after passage through the avian digestive tract, to determine the likelihood of transfer from land-based offal disposal sites back to the aquatic environment.

The likelihood of exposure to new Japan virus is assessed as non-negligible.

20.2.3. Consequence assessment

The introduction of NJV is likely to affect the aquaculture of rainbow trout (*O. mykiss*), particularly in a naïve population. This could have significant social and environmental implications for the associated recreational and tourist fisheries for rainbow trout (Fish & Game 2014). While the dollar value of New Zealand's freshwater fisheries is not fully known, the Lake Taupo trout fishery alone is valued at $70–80 million (Marsh & Mkwra 2013).

New Japan virus has only been reported from Japanese salmonid aquaculture (Kahn *et al.* 1999) and Japan was a net importer of salmon in 2012 (Yanagisawa & Ohsumi 2012). While it is unlikely that significant quantities of Japanese salmonid fish would be exported to New Zealand for human consumption, the consequences of establishment are assessed as non-negligible.

20.2.4. Risk estimation

Since the entry, exposure and consequence assessments are non-negligible, the risk estimate is non-negligible. Therefore, NJV is assessed to be a risk in the commodity and risk management measures may be justified.

20.3. Risk management

New Japan virus has been assessed to be a risk in the commodity. Infection with NJV is a non-notifiable disease, so no specific guidance on mitigation measures is provided in the OIE *Aquatic Code* (OIE 2016a). It is assumed that compliance with the approved processed states for OIE-listed diseases (Appendix 3) would also eliminate NJV from the commodity and be a viable risk management option.

New Japan virus is only reported from Japanese aquaculture (Oh *et al.* 1995b), but no requirements for dedicated monitoring exist. Where country/zone freedom is approved through the MPI Country Approval Procedures, this option is likely to substantially reduce the pathogen load of New Japan virus. Approval of a declaration of country freedom by MPI is a viable risk management option.

New Japan virus is reported from two fish families (Table 16), which may be present in the commodity. Species declaration should substantially reduce the pathogen load of NJV in the commodity and be a viable risk management option. This virus is currently uncharacterised and little information is available on its epidemiology (Oh *et al.* 1995a; Oh *et al.* 1995b, Kahn *et al.* 1999). It is however, tentatively considered to be a rhabdovirus (Oh *et al.* 1995a).

New Japan virus is mainly associated with aquaculture in Japan (Oh *et al.* 1995a; Kahn *et al.* 1999). Restriction of the commodity to wild-caught fish (not from aquaculture) would moderately reduce pathogen load and be a viable risk management option.

New Japan virus occurs with brain tissues and viral titre in somatic muscle tissues is likely to be low (Kahn *et al.* 1999). Removal of the gill tissues may slightly reduce the pathogen load of NJV in the commodity. Removal of the head and gills should moderately decrease pathogen load and be a viable risk management option.

New Japan virus is unaffected by freezing (to -80°C (Oh *et al*. 1995b)) so frozen storage is unlikely to be a viable risk management option.

New Japan virus is unaffected by moderate heat treatment (at 60°C for 30 minutes) (Kahn *et al.* 1999), but should be eliminated by the OIE recommended heat treatments (cooking to 121°C for at least 3.6 minutes, or to at least 90°C for 10 minutes) (OIE 2016a). Heat treatment is a viable risk management option.

As no vaccines are available against new Japan virus, a requirement for vaccinated fish is unlikely to be a viable management option.

20.3.1. Risk management options

New Japan virus (NJV) is reported from fish in families Plecoglossidae and Salmonidae (Table 16), which are considered likely to be present in the commodity. Other families have not been associated with NJV. Therefore species declaration should substantially reduce the occurrence of NJV in the commodity.

For the commodities originated from these families, one, or a combination of the following additional options could also be considered to effectively manage the risk.

Where country/zone freedom from NJV is accepted by MPI:

Option 1

Acceptance of country/zone freedom by MPI should substantially reduce the occurrence of NJV, so the commodity may be imported without any further restrictions.

Where country/zone freedom from NJV is not accepted by MPI or not available:

Option 2

Processing to a state consistent with the approved states for OIE-listed diseases (Appendix 3) should eliminate NJV. When these provisions are met, the commodity could be imported without any further restrictions.

Where the imported commodity is not in a state consistent with the approved states for OIE-listed diseases (Appendix 3):

Option 3

Further processing (by removal of the head and gills) should moderately reduce the occurrence of NJV. When this provision is met, the commodity could be imported without any further restrictions; or,

Option 4

Restriction of the commodity to wild-caught fish (not from aquaculture) should moderately reduce the occurrence of NJV. When this provision is met, the commodity could be imported without any further restrictions.

20.4. References

Fish and Game (2014). New Zealand Fish and Game Council [Online] Available from: http://www.fishandgame.org.nz/ [Accessed 24 September 2014].

Fraenkel-Conrat H (2012). Comprehensive virology, Volume 14: Newly characterised vertebrate viruses. Springer Science, New York. United States of America, 544 p.

Kahn SA, Beers PT, Findlay VL, Peebles IR, Durham PJ, Wilson DW, Gerrity SE (1999). Import risk analysis on non-viable salmonids and non-salmonid marine finfish. Australian Quarantine and Inspection Service, Department of Agriculture, Fisheries and Forestry, Canberra, Australia, 409 p.

Kim R, Faisal M (2011). Emergence and resurgence of the viral haemorrhagic septicaemia virus (*Novirhabdovirus*, *Rhabdoviridae*, *Mononegavirales*). *Journal of Advanced Research* 2: 9–23.

Marsh D, Mkwra L (2013). Review of freshwater non-market value studies. Department of Economics, University of Waikato, Hamilton, New Zealand, 144 p.

Oh MJ, Izu Y, Oru M, Ura K, Takahisa K, Yoshio E (1995a). A new virus isolated from salmonid fish. *Fish Pathology* 30(1): 23–32.

Oh MJ, Yoshimura M, Kimura T, Ezura Y (1995b). Pathogenicity of the virus isolated from the brain of abnormally swimming salmonid. *Fish Pathology* 30: 33–38.

OIE (2016). Aquatic Animal Health Code, 19th Edition. World Organization for Animal Health. Paris, France [Online]. Available from: http://www.oie.int/international-standard-setting/aquatic-code/access-online/ [Accessed 15 March 2017].

Stone MAB, MacDiarmid SC, Pharo HJ (1997). Import Risk Analysis: salmonids for human consumption. Ministry of Agriculture Regulatory Authority, New Zealand, 269 p.

Yanagisawa A, Ohsumi K (2012). Overseas market introduction service on the Japanese seafood market. Report OMIS JPN2610, Sea Fish Industry Authority. HMSO, London, 40 p.

21. Nervous necrosis virus and associated nodaviruses

21.1. Hazard identification

21.1.1. Aetiological agent

Nodaviruses are non-enveloped positive stranded RNA viruses that are major pathogens of larval and juvenile fish. Recent reviews of the Picornavirus group based on genomic analysis have resulted in the reclassification of several fish picornaviruses as nodaviruses (Lauber & Gorbalenya 2012).

Nodaviruses generally affect the central nervous system and eye tissues, producing cell vacuolation and necrosis, usually with high mortality (Mundy and Nakai 1997). The type specimen of the Genus *Betanodavirus* is nervous necrosis virus (NNV) (Sano *et al.* 2011). The genus was originally separated into four species: barfin flounder nervous necrosis virus (NNV); red spotted grouper NNV; striped jack NNV; and tiger puffer NNV. Of these, red spotted grouper NNV has the widest distribution (Bandin & Dopazo 2011; Shetty *et al.* 2012). It was first described in 1988 from Japan in parrotfish (*Oplegnathus fasciatus* and *Calotomus japonicus*) (Yoshikoshi & Inoue 1990; Chen *et al.* 2014), but has since been recognised as having a wide host range and distribution (Castric 1997).

Genomic analysis suggests that these viruses represent genotypes of NNV rather than separate species. Five genotypes: striped jack nervous necrosis; barfin flounder nervous necrosis; cold water fish species nervous necrosis; warm water fish nervous necrosis and turbot nervous necrosis group have been proposed (Moody & Horwood 2008; Sano *et al.* 2011). Host specificity appears to be low within each of these genotypes (Munday *et al*. 2002; Sano *et al*. 2011), as isolates of one group may occasionally cause disease in host species that are predominantly infected by isolates of other groups (Tanaka *et al.* 2003).

Other related fish nodaviruses include barramundi nodavirus (Glazebrook *et al.* 1990; Munday *et al*. 1994), turbot encephalomyelitis (Bloch *et al.* 1991), and sea bass nodavirus (Breul *et al*. 1991).

21.1.2. OIE status

Infection with NNV or other nodaviruses is not an OIE listed disease (OIE 2016a).

21.1.3. New Zealand status

NNV, barramundi nodavirus, turbot encephalomyelitis and sea bass nodavirus have not been reported from New Zealand (Hine & Diggles 2005; Tubbs *et al.* 2007; Moody & Horwood 2008). Infection with NNV is not a notifiable disease in New Zealand (Anon. 2010) and it is considered exotic (Tubbs *et al.* 2007).

21.1.4. Epidemiology

Nervous necrosis virus

NNV is the agent of viral nervous necrosis disease, described by the OIE as viral encephalopathy and retinopathy (VER) (OIE 2016b). This causes economically significant disease in a wide range of marine fish species in Chinese, Asian, Australian and European aquaculture (Munday & Nakai 1997; Gomez-Casado *et al*. 2011; Liu *et al*. 2015; Chi *et al.* 2016). Reported mortalities may reach 100% in hatchery-reared larval and juvenile fish (Castric 1997). Susceptibility decreases in older

fish, but significant mortalities may still occur in fish of marketable size (Anderson & Moody 2004). NNV infection is widely reported from Australia, China, India, Southeast Asia, Europe and North America, but has not been reported from Africa, South America or New Zealand (Hine & Diggles 2005; Moody & Horwood 2008; Sano *et al.* 2011). Reported host species occur in 23 families of marine and freshwater fish (Castric 1997; Tubbs *et al.* 2007; Bovo *et al*. 2011; Sano *et al.* 2011; OIE 2016b; Binesh 2013; Olveira *et al.* 2013, Liu *et al.* 2015, Costa & Thompson 2016), as shown in Table 17.

Table 17. Representative Families and Host Species of Fish Susceptible to Nervous Necrosis Virus (NNV) and Associated Nodaviruses

Family	Host Species
Acanthuridae	Surgeonfish (*Acanthurus* spp.)
Acipenseridae	Danube sturgeon (*Acipenser gueldenstaedtii*)
Acropomatidae	Blackthroat seaperch (*Doederleinia berycoides*)
Anarhichadidae	Spotted wolffish (*Anarhichas minor*)
Anguillidae	European eel (*Anguilla anguilla*)
Batrachoididae	Lusitanian toadfish (*Halobatrachus didactylus*)
Bothidae	Left eye flounder (*Crossorhombus kanekonis*)
Carangidae	Bigeye trevally (*Caranx sexfasciatus*), diamond trevally (*Alectis indica*), greater amberjack (*Seriola dumerili*), Japanese scad (*Decapterus maruadsi*), razorbelly scad (*Alepes kleinii*), Samson fish (*Carynx hippos*), shrimp scad (*Alepes djedaba*), striped jack (silver trevally) (*Pseudocaranx dentex*), snub-nosed pompano (*Trachinotus blochii*), yellowstripe scad (*Selaroides leptolepis*), yellow wax pompano (*T. falcatus*), lesser queenfish (*Scomberoides lysan*), Atlantic horse mackerel (*Trachurus trachurus*), Japanese horse makerel (*Trachurus japonicus*)
Centrarchidae	Largemouth black bass (*Micropterus salmoides*)
Chanidae	Milkfish (*Chanos chanos*)
Cichlidae	Tilapia (*Oreochromis niloticus*)
Clupeidae	European pilchard (*Sardina pilchardus*), dotted gizzard shad (*Konosirus punctatus*)
Cynoglossidae	Macao sole (*Cynoglossus sinicus*), spotted tonguesole (*C. punticeps*), sole (*C. robustus*)
Cyprinidae	goldfish (*Carassius auratus*)
Eleotridae	Sleepy cod (*Oxyeleotris lineolata*)
Ephippidae	Orbicular batfish (*Platax orbicularis*)
Gadidae	Atlantic cod (*Gadus morhua*), capelin (*Mallotus villosus*), cod (*Trisopterus capelanus*), haddock (*Melanogrammus aeglefinus*), Pacific cod (*G. macrocephalus*)
Gobiidae	Black goby (*Gobius niger*)
Haemulidae	Crescent sweetlips (*Plectorhinchus cinctus*), gold spotted sweetlips (*P. flavomaculatus*), trout sweetlips (*Plectorhinchus pictus*)
Hapalogenyidae	Barbeled grunter (*Hapalogenys analis*)
Lateolabracidae	Japanese sea bass (*Lateolabrax japonicus*)
Latidae	Asian sea bass/barramundi (*Lates calcarifer*)
Latridae	Striped trumpeter (*Latris lineata*)
Leiognathidae	Berber ponyfish (*Leiognathus berbis*), orangefin ponyfish (*L. bindus*), offshore ponyfish (*Equulites rivulatus*), pugnosed ponyfish (*Secutor insidator*), shortnose ponyfish (*L. brevirostris*), spotnape ponyfish (*Nuchequula nuchalis*)
Lethrinidae	Chinese emperor (*Lethrinus haematopterus*)
Lophiidae	Yellow goosefish (*Lophius litulon*)
Lutjanidae	Blubberlip snapper (*Lutjanus rivulatus*), dory snapper (*Lutjanus fulviflamma*), John's snapper (*L. johnii*), humphead snapper (*L. sanguineus*), mangrove red snapper (*L. argentimaculatus*), crimson snapper (*L. erythropterus),* red snapper *(L. malabaricus*), Russell's snapper (*L. russellii*), black and white snapper (*Macolor niger*)
Macrouridae	Spearnose grenadier (*Coelorinchus multispinulosus*)
Malacanthidae	Japanese tilefish (*Branchiostegus japonicus*)
Monacanthidae	Fanbelly leatherjacket (*Monacanthus chinensis*), threadsail filefish (*Stephanolepis cirrhifer*), black scraper (*Thamnaconus tessellatus*), hair-finned leatherjacket (*Paramonacanthus japonicus*), mudbank filefish (*P. sulcatus*)
Moronidae	European sea bass (*Dicentrarchus labrax*), striped seabass (*Morone saxitalis*), white seabass (*M. chrysops*) and hybrids

Table 17 (Continued)

Family	Host Species
Mugilidae	grey mullet (*Mugil cephalus*, *Chelon auratus*)
Mullidae	Red mullet (*Mullus barbatus*), mullet (*Mullus surmuletus, Mullus* spp.)
Muraenesocidae	Daggertooth pike conger (*Muraenesox cinereus*)
Oplegnathidae	Japanese parrotfish (*Oplegnathus fasciatus*), rock porgy (*O. punctatus*)
Paralichthyidae	Flounder (*Tarphops oligolepis*)**,** cinnamon flounder (*Pseudorhombus cinnamoneus*), *P. levisquamis*), fivespot flounder (*P. pentophthalmus*), *Tephrinectes sinensis*, *Pseudorhombus* sp., Bastard halibut (*Paralichthys olivaceus*)
Percichthyidae	Australian bass (*Macquaria novemaculeata*), golden perch (*Macquaria ambigua*), Macquarie perch (*M. australasica*), Murray cod (*Maccullochella peelii*)
Percidae	Pike-perch (*Sander lucioperca*)
Platycephalidae	Bartailed flathead (*Platycephalus indicus*)
Pleuronectidae	Atlantic halibut (*Hippoglossus hippoglossus*), barfin flounder (*Verasper moseri*), winter flounder ((*Pseudopleuronectes americanus*)
Plotosidae	Australian estuary cobbler (*Cnidoglanis macrocephalus*), Australian freshwater catfish (*Tandanus tandanus*)
Family	**Susceptible Species**
Priacanthidae	Moonsail bullseye (*Priacanthus hamrur*)
Rachycentridae	Cobia (*Rachycentron canadum*)
Salmonidae	Atlantic salmon (*Salmo salar*)
Sciaenidae	Croaker (*Otolithes argenteus*), meagre (*Argyrosomus argentatus*, *A. macrocephalus*, *A. regius*), red drum (*Sciaenops ocellatus*), shi drum (*Umbrina cirrosa*), goatee croaker (*Dendrophysa russelii*), tigertooth croaker (*Otolithes ruber*), white weakfish (*Atractoscion nobilis*), silver croaker (*Pennahia argentata*), bighead croaker (*P. macrocephalus*), donkey croaker (*P. anea*)
Scombridae	Bluefin tuna (*Thunnus thunnus*), Pacific bluefin tuna (*T. orientalis*)
Scophthalmidae	Turbot (*Scophthalmus maximus*)
Sebastidae	white weakfish (*Sebastes oblongus*)
Serranidae	Areolate grouper (*Epinephelus areolatus*), Bleeker's grouper (*E. bleekeri*), brown-spotted grouper (*E. malabaricus*), brown spotted reef cod (*E. chlorostigma*), brown spotted grouper (*E. maculatus*). black/brown spotted grouper (*E. fusogutatus*), chocolate hind (*Cephalopholis boenak*), dotted grouper (*E. epistictus*), kelp grouper (*E. moara*), orange-spotted grouper (*E. coioides*), red spotted grouper (*E. akaara*), rock grouper (*E. fasciomaculatus*), seven band grouper (*E. septemfasciatus*), spinycheek grouper (*E. dicanthus*), three spot grouper (*E. fario*), yellow grouper (*E. awoara*), white grouper (*E. aenus*), grouper (*Epinephelus* spp.)
Siganidae	White-spotted spinefoot (*Siganus canaliculatus*)
Sillaginidae	Trumpeter whiting (*Sillago maculata*), Japanese sillago (*S. japonica*)
Siluridae	Amur catfish (*Silurus asotus*)
Soleidae	Dover sole (*Solea solea*), thickback sole (*S. ovata*), Senegal sole (*S. senegalensis*), oriental sole (*Brachirus orientalis*), zebra sole (*Zebrias zebra*)
Sparidae	Black sea bream (*Acanthopagrus schlegelii),* crimson sea bream (*Parargyrops edita*), gilthead sea bream (*Sparus aurata*), red sea bream (*Pagrus major*), gold-lined seabream (*Rhabdosargus sarba*)
Tetraodontidae	Lunartail puffer (*Lagocephalus lunaris*), golden puffer (*L. spadiceus*), komon-damashi (*Takifugu alboplumbeus*), grass puffer (*T. niphobles*), Japanese pufferfish (*T. rubripes*), yellowfin pufferfish (*T. xanthopterus*)
Terapontidae	Barcoo grunter (*Scortum barcoo*), four lined terapon (*Pelates quadrilineatus*), jarbua terapon (*Terapon jarbua*), silver perch (*Bidyanus bidyanus*)
Triglidae	Tub gurnard (*Chelidonichthys lucerna*), spiny red gurnard (*C. spinosus*)
Zeidae	John dory (*Zeus faber*)
Zoarcidae	Eelpout (*Zoarces gillii*)

Species including mangrove red snapper/milkfish (*Chanos chanos*), Japanese rice fish (*Oryzias latipes*) and Atlantic salmon (*Salmo salar*) may be experimentally infected (Korsnes *et al.* 2005; Sano *et al*. 2011), although infection does not always result in clinical disease (Castric *et al*. 2001; Olveira *et al.* 2013).

The mode of transmission is horizontal, through the water column, where healthy fish in sea cage culture may become infected from adjacent infected wild stocks (Castric 1997), or through infected water used in aquaculture transfer. NNV may also be spread by fomites, including vehicles and fish processing equipment during aquaculture, storage, transport and fish processing (Sano *et al*. 2011). Vertical transmission from brood stock to offspring may also occur (Breuil *et al.* 2002; Chi *et al.* 2016).

Viral entry occurs through the oral route where the virus enters across the intestinal epithelium following ingestion (Chi *et al.* 2016), but infection may follow several routes (Costa & Thompson 2016) The virus may be neuroinvasive in brain, spinal cord and optic tissues causing extensive vacuolation and necrosis in *Oplegnathus fasciatus* (Nguyen *et al*. 2014; Costa & Thompson 2016), while infection in *Caranx* spp. initially affects the spinal cord and swim bladder, before progressing to the brain & retina (Costa & Thompson 2016), but fish typically show no gross external signs of infection (Sano *et al*. 2011). Viral shedding occurs through the gonads and digestive tract (Costa & Thompson 2016). NNV may persist at subclinical levels of infection, where surviving fish can act as carriers of disease (Sano *et al*. 2011; Chi *et al.* 2016). Dead and decomposing fish are also sources of infection (OIE 2016b), although the minimum dose for infection is unknown.

Transmission may be horizontal, as nodavirus outbreaks occur in fish larvae, while NNV is reported from ovarian tissues, sperm and fertilised eggs (Costa & Thompson 2016).

Nodaviruses are highly resistant to environmental conditions, remaining viable for at least 12 months in sea water at up to 15°C (Tubbs *et al.* 2007). NNV is unaffected by mildly acid conditions and remains viable for extended periods in frozen processed fish (OIE 2016b).

NNV is tolerant of high temperatures, with maximum replication occurring at 20 to 26°C. While mild inhibition occurs up to 31°C, inactivation requires temperatures above 60°C for 30 to 60 minutes (Sano et *al*. 2011). NNV is unaffected by salinity, with infections reported in both marine and freshwater hosts (OIE 2016b).

NNV has poor resistance to drying, with 99% inactivation occurring after 7 days (OIE 2016b) and may be inactivated by UV irradiation (1 x 104 µWs mL^{-1}), ozone treatment (3 mg L^{-1} total residual oxidants for 4-7 min) (Tubbs *et al.* 2007). It is inactivated by common disinfectants (chlorine, iodine or benzalkonium chloride at 50 mg L^{-1} (Anderson & Moody 2004), but is chloroform, ether and formalin-resistant (Arimoto *et al.* 1996). Few vaccines are commercially available (OIE 2016b; Shetty *et al.* 2012), although a vaccine for seven-band grouper (*Epinephelus septemfasciatus*) is available in Japan (OIE 2016b) and others are under development (Shetty *et al.* 2012). The recommended treatment is the destruction of infected stock (Biering *et al.* 2005; Sano *et al.* 2011)

Potential species at risk in New Zealand include eels (*Anguilla* spp.), flatfish (Pleuronectidae), jack mackerel (*Trachurus* spp.), snapper (*Pagrus auratus*), striped trumpeter (*Latris lineata*), trevally (*Caranx georgianu*s) and yellowtail kingfish (*Seriola lalandi*).

Barramundi nodavirus

Barramundi nodavirus was previously classed as a picornavirus (Glazebrook *et al.* 1990; Bloch *et al.* 1991; Munday *et al*. 1994), but is now considered as a nodavirus (Anderson & Moody 2004), closely related to NNV. It infects brain and eye tissues and is reported from barramundi (*L. calcarifer*) in Australia and India (Anderson & Moody 2004; Diggles & Hutson 2005; Forrest *et al.*

2011; John *et al.* 2012). Other species may be experimentally infected including golden perch (*Macquaria ambigua*), silver perch (*Bidyanus bidyanus*), sleepy cod (*Oxyeleotris lineolata*) and barcoo grunter (*Scortum barcoo*). The minimum infective doses for nodaviruses in bath infection are $10^{5.1}$ $TCID_{50}$ ml^{-1} (tissue culture infectious dose causing 50% infection) for 2 hours, 3.7 x 10^{7} $TCID_{50}$ ml^{-1} for 1 hour and 6 x $10^{5.2}$ $TCID_{50}$ ml^{-1} for 2 hours, with a cumulative mortality of up to 46%.

Infection progresses with no visible external signs, with vacuolation in brain, olfactory and optic tissues and infection persisted in surviving fish (Anderson & Moody 2004).

Turbot encephalomyelitis

Encephalomyelitis disease was isolated from larval turbot (*Scophthalmus (Psetta) maximus*) (Bloch *et al.* 1991), with high mortality (up to 100%)). Infection causes extensive vacuolation of brain tissue, but the virus is not present in gills, muscle or visceral tissues. The agent was originally classified as a picornavirus, but is now classified as a nodavirus (Anderson & Moody 2004; Olveira *et al.* 2013).

Striped jack nervous necrosis virus (Sea bass picornavirus)

This was originally described as sea bass picornavirus, but is now considered an isolate of NNV (Breuil *et al.* 2001). It occurs in barramundi (*L. calcarifer*), sea bass (*Dicentrarchus labrax*) and striped jack (*Pseudocaranx dentex*) (Breuil *et al.* 1991).

These nodaviruses are considered together with the other NNV isolates in the assessment (Table 18).

21.2. Risk assessment

21.2.1. Entry assessment

NNV is relatively non host-specific (Munday *et al.* 2002), where each genotype infects a wide range of marine and freshwater farmed and wild fish (see Table 19) in a geographical region (Shetty *et al.* 2012). Surviving adult fish act as carriers of disease (Chi *et al.* 2016). NNV is localised in the brain and neural tissues, usually with no external signs of infection (Sano *et al.* 2011), so clinically infected eviscerated fish are likely to pass visual inspection for the human food consumption pathway and be present in the commodity.

NNV is resistant to the environmental conditions likely to be encountered in fish storage, transportation and processing (Sano *et al.* 2011) and is likely to remain viable in eviscerated fish product. The likelihood of entry is assessed as non-negligible.

21.2.2. Exposure assessment

To establish in New Zealand, NNV would have to infect a susceptible fish host in sufficient quantity and duration (Kahn *et al.* 1999). NNV is likely to remain viable in the neural tissues associated with fish heads and fish frames (skeletons with the heads and intestines intact) of moist decaying fish offal discarded in commercial fish processing.

NNV remains viable after passage through the avian digestive system (OIE 2016b) and may be distributed by scavenging piscivorous birds. Infection may also occur through fomites, or

contamination of fish processing equipment (Sano *et al*. 2011). Several genogroups and isolates of NNV have been identified and the severity of disease may be dependent on the virulence, pathogenicity and host specificity of a particular viral genotype (Sano *et al*. 2011). The likelihood of exposure to NNV through the human food consumption pathway is assessed as non-negligible.

21.2.3. Consequence assessment

Nervous necrosis virus/viral encephalopathy and retinopathy is a serious economic disease of marine and freshwater fish aquaculture (Munday *et al.* 2002; Gomez-Casado *et al.* 2011; Sano *et al.* 2011). NNV has low host specificity (Castric 1997; Sano *et al*. 2011).

Introduction of NNV could have economic consequences for developing yellowtail kingfish (*Seriola lalandi*) and groper (*Polyprion oxygeneios*) aquaculture (NIWA 2017a, 2017b). It may have implications for the wild fisheries of several other commercially important fish species, including jack mackerel (export value NZ $39 million), snapper, (export value $33 million), eels (export value $5 million) and trevally (export value $4 million) (Seafood New Zealand 2013), as well as incur significant economic and social costs in the associated recreational and traditional fisheries for these species (MPI 2014). Given the low host specificity of NNV, there are likely to be further consequences for other freshwater endemic and introduced species.

The consequences of the introduction and establishment of NNV are assessed as non-negligible.

21.2.4. Risk estimation

Since the entry, exposure and consequence assessments for NNV and associated nodaviruses are non-negligible, the risk estimate is non-negligible. Therefore, NNV and these associated nodaviruses are assessed to be a risk in the commodity and risk management measures may be justified.

21.3. Risk management

NNV has been assessed as a risk in the commodity. Infection with NNV is a non-notifiable disease, so the *Aquatic Code* (OIE 2016a) provides no guidance in mitigation measures for importing eviscerated fish from infected countries, or specific processing requirements that would ensure the destruction of the pathogen. It is assumed that compliance with the approved processed states for OIE-listed diseases (Appendix 3) should also eliminate NNV from the commodity and be a viable risk management option.

No specific requirements for dedicated monitoring exist. Viral encephalopathy and retinopathy (VER) is however, listed in the OIE Aquatic manual (OIE 2016b) and some mitigation measures are indicated. Families of fish susceptible to NNV and associated nodaviruses (Table 17) may be present in the commodity. Species declaration should substantially reduce the occurrence of NNV in the commodity and be a viable risk management option.

Infection with NNV is uncommon in wild fish stocks, but is a major pathogen of larval and juvenile fish in Chinese, Asian, Australian and European aquaculture (Castric 1997; Chi *et al.* 2016). Restriction of the commodity to wild-caught fish (not from aquaculture) would moderately reduce pathogen load and be a viable risk management option.

NNV is widely distributed in Asia, Australia, Europe and North America, but is absent from Africa and South America. Approval of country/zone freedom from NNV through the MPI Country Approval Procedures should substantially reduce the occurrence of NNV in the commodity. Acceptance of a declaration of country/zone freedom by MPI is a viable risk management option.

NNV is associated with neural tissue of the brain and spinal cord, so removal of the gills would have little or no effect on the pathogen load of NNV. Removal of the head and gills should slightly reduce pathogen load, but virus present in the spinal cord would be retained in the commodity. Further processing to the skin-off fillet state would moderately reduce pathogen load and be a viable risk management option.

NNV is resistant to freezing so frozen storage would have no effect on the pathogen load of NNV. High temperatures are required to ensure inactivation, and heat treatment (by cooking to at least 60°C for 60 minutes) would eliminate NNV from the commodity.

No vaccines are commercially available for NNV.

21.3.1. Risk management options

Nervous necrosis virus (NNV) and related nodaviruses are reported from fish in families Acanthuridae, Acipenseridae, Acropomatidae, Anarhichadidae, Anguillidae, Batrachoididae, Bothidae, Carangidae, Centrarchidae, Chanidae, Cichlidae, Clupeidae, Cynoglossidae, Cyprinidae, Eleotridae, Gadidae, Gobiidae, Haemulidae, Hapalogenyidae, Lateolabracidae, Latridae, Leiognathidae, Lethrinidae, Lophiidae, Lutjanidae, Macrouridae, Malacanthidae, Monacanthidae, Moronidae, Mugilidae, Mullidae, Muraenesocidae, Oplegnathidae, Paralichthyidae, Percichthyidae, Percidae, Platycephalidae, Pleuronectidae, Plotosidae, Priacanthidae, Rachycentridae, Salmonidae, Sciaenidae, Scombridae, Scophthalmidae, Sebastidae, Serranidae, Siganidae, Sillaginidae, Siluridae, Soleidae, Sparidae, Terapontidae, Tetraodontidae, Triglidae, Zeidae, and Zoarcidae Fish (Table 17). These families are considered likely to be present in the commodity. Other families have not been associated with NNV and related nodaviruses. Therefore species declaration indicating the commodity is not originated from any of the above families should substantially reduce the occurrence of NNV and related nodaviruses in the commodity.

For the commodities originated from families associated with NNV and associated nodaviruses, one or a combination of the following additional options could also be considered to effectively manage the risk.

Where country /zone freedom from NNV and associated nodaviruses is accepted by MPI:

Option 1

Acceptance of country/zone freedom by MPI should substantially reduce the occurrence of NNV and associated nodaviruses, so the commodity may be imported without any further restrictions.

Where country/zone freedom from NNV and associated nodaviruses is not accepted by MPI or not available:

Option 2

Processing to a state consistent with the approved states for OIE-listed diseases (Appendix 3) should eliminate NNV and associated nodaviruses. When these provisions are met, the commodity could be imported without any further restrictions.

Where the imported commodity is not in a state consistent with the approved states for OIE-listed diseases (Appendix 3):

Option 3

Heat treatment (by cooking to at least 60°C for at least 60 minutes) should eliminate NNV and associated nodaviruses from the commodity. When this provision is met, the commodity could be imported without any further restrictions; or,

Option 4

Further processing (to the skin-off fillet state) should moderately reduce the occurrence of NNV and associated nodaviruses. When this provision is met, the commodity could be imported without any further restrictions; or,

Option 5

Restriction of the commodity to wild-caught fish (not from aquaculture) should moderately reduce the occurrence of NNV and associated nodaviruses. When this provision is met, the commodity could be imported without any further restrictions.

21.4. References

Anderson IG, Moody N (2004). The effect of barramundi nodavirus on important freshwater fishes. FRDC Report No. 1999/205. Fisheries Research and Development Corporation. Department of Primary Industries, Townsville, Queensland, Australia, 97 p.

Anonymous (2010). Biosecurity (Notifiable Organisms) Order 2010. New Zealand Government, Wellington, New Zealand, 13 p.

Arimoto, M, Sato J, Maruyama K, Mimura G, Furusawa I (1996). Effect of chemical and physical treatments on the inactivation of striped jack nervous necrosis virus (SJNNV). *Aquacultur*e 143: 15–22.

Bandin I, Dopazo CP (2011). Host range, host specificity and hypothesized host shift events among viruses of lower vertebrates. *Veterinary Research* 42: 67 [Online]. Available from: http://www.veterinaryresearch.org/content/42/1/67 [Accessed 12 December 2014].

Biering E, Villoing S, Sommerset I, Christie KE (2005). Update on viral vaccines for fish. *Developmental Biology* 121: 97–113.

Binesh CP (2013). Mortality due to viral nervous necrosis in zebrafish *Danio rerio* and goldfish *Carassius auratus Diseases of Aquatic Organisms* 104: 257–260.

Bloch B, Gravningen K, Larsen JL (1991). Encephalomyelitis among turbot associated with picornavirus-like agent. *Diseases of Aquatic Organisms* 10(1): 65–70.

Bovo G, Gustinelli A, Quaglio F, Gobbo F, Panzarin V, Fusaro A, Mutinelli F, Caffara M, Fioravanti ML (2011). Viral encephalopathy and retinopathy outbreak in freshwater fish farmed in Italy. *Diseases of Aquatic Organisms* 96(1): 45–54.

Breuil G, Bonami JR, Pepin JF, Pichot Y (1991). Viral infection (picorna-like virus) associated with mass mortalities in hatchery-reared sea-bass (*Dicentrarchus labrax*) larvae and juveniles. *Aquaculture* 97, 109–116.

Breuil G, Mouchel O, Fauvel C, Pepin JF (2001). Sea bass *Dicentrarchus labrax* nervous necrosis virus isolates with distinct pathogenicity to sea bass larvae. *Diseases of Aquatic Organisms* 45: 25–31.

Breuil G, Pepin JFP, Boscher S, Thiery R (2002). Experimental vertical transmission of nodavirus from broodfish to eggs and larvae of the sea bass, *Dicentrarchus labrax* (L.). *Journal of Fish Diseases* 25(12): 697–702.

Castric J (1997). Viral diseases in fish mariculture. *Bulletin of the European Association of Fish Pathologists* 17(6): 220–228.

Castric J, Thiéry R, Jeffroy J, de Kinkelin P, Raymond JC (2001). Sea bream *Sparus aurata*, an asymptomatic contagious fish host for nodavirus. *Diseases of Aquatic Organisms* 47: 33–38.

Chen Y, Wang TW, Chen TY (2014). Immunity to betanodavirus infections of marine fish. *Developmental and Comparative Immunity* 43(2): 174–183.

Chi SC, Wu YC, Hog JR (2016). Nodaviruses of fish. pp, 371–393 *In*: Kibenge FSB, Godoy MC (*Eds*.). Aquaculture Virology. Academic Press, New York, United States of America, 549 p.

Colorni A, Diamant A. (2014). Infectious diseases of warm water fish in marine and brackish waters. pp 155–192. *In*: Woo PTK, Bruno DW (*Eds.*). Diseases and disorders of finfish in cage culture, Second edition. CABI, Wallingford, United Kingdom, 354 p.

Costa JZ, Thompson KD (2016). Understanding the interaction between betanodavirus and its host for the development of prophylactic measures for viral encephalopathy and retinopathy. *Fish and Shellfish Immunity* 53: 35–49.

Diggles B, Hutson K (2005). Diseases of kingfish (*Seriola lalandi*) in Australasia. *Aquaculture Health International* 3: 12–14.

Forrest B, Hopkins G, Webb S, Trenblay L (2011). Overview of marine biosecurity risks from finfish aquaculture development in the Waikato region. Cawthron report 1871 prepared for the Waikato Regional Council. Cawthron Institute, Nelson, 78 p.

Glazebrook JS, Heasman MP, de Beer SW (1990). Picorna-like viral particles associated with mass mortalities in larval barramundi, *Lates calcarifer* Bloch. *Journal of Fish Diseases* 13: 245–249.

Gomez-Casado E, Estepa A, Coll JM (2011). A comparative review on European-farmed finfish RNA viruses and their vaccines. *Vaccine* 29(15): 2657–2671.

Hine PM, Diggles BK (2005). Import risk analysis: Ornamental fish. Biosecurity New Zealand, Ministry of Agriculture and Forestry, Wellington, New Zealand, 262 p.

Kahn SA, Beers PT, Findlay VL, Peebles IR, Durham PJ, Wilson DW, Gerrity SE (1999). Import risk analysis on non-viable salmonids and non-salmonid marine finfish. Australian Quarantine and Inspection Service, Department of Agriculture, Fisheries and Forestry, Canberra, Australia, 409 p.

Jaramillo D, Fielder S, Hick P, Go G, Whittington R (2014). Modulating factors in Viral nervous necrosis (VNN). The role of host age and water temperature on disease outcome based on an Australian bass laboratory infection model. Meeting Abstract, World Aquaculture 2014 [Online] Available from: https://www.was.org/meetings/ShowAbstract.aspx?Id=33069 [Accessed 15 August 2017].

John KR, George MR, Jeyatha B, Saravanakumar R, Sundar P, Jithendran KP, Koppang EO (2012). Isolation and characterisation of Indian betanodavirus from infected farm-reared Asian seabass *Lates calcarifer* (Bloch, 1790) juveniles. *Aquaculture Research* 45(9): 1481–1488.

Korsnes K, Devold M, Nerland AH, Nylund A (2005). Viral encephalopathy and retinopathy (VER) in Atlantic salmon *Salmo salar* after intraperitoneal challenge with a nodavirus from Atlantic halibut *Hippoglossus hippoglossus. Diseases of Aquatic Organisms* 68(1): 7–15.

Lauber C, Gorbalenya AE (2012). Towards genetic-based virus taxonomy: Comparative analysis of a genetics-based classification and the taxonomy of picornaviruses. *Journal of Virology* 89(19): 3905–3915.

Liu XD, Huang JN, Weng SP, Hu XQ, Chen WY, Qin ZD, Dong XX, Liu XL, Zhou Y, Asim M, Wang WM, He JG, Lin L (2015). Infections of nervous necrosis virus in wild and cage-reared marine fish from South China Sea with unexpected wide host range. *Journal of Fish Diseases* 38: 533–540.

Ministry for Primary Industries (MPI) (2014). Fisheries Assessment Plenary, May 2014: Stock assessments and stock status. Compiled by the Fisheries Science Group, Ministry for Primary Industries, Wellington, New Zealand, 1381 p.

Moody NJ, Horwood PF (2008). Betanodaviruses in finfish. Australian and New Zealand Standard Diagnostic Procedures: Nodaviruses. Australian Government, Canberra, Australia, 18 p.

Munday BL, Nakai T, Nguyen HD (1994). Antigenic relationship of the picorna-like virus of larval barramundi, *Lates calcarifer* Bloch to the nodavirus of larval striped jack, *Pseudocaranx dentex* (Bloch and Schneider). *Australian Veterinary Journal* 71: 384–385.

Munday BL, Nakai T (1997). Special topic review: nodaviruses as pathogens in larval and juvenile marine finfish. *World Journal of Microbiology and Biotechnology* 13: 375–381.

Munday BL, Kwang J, Moody N (2002). Betanodavirus infections of teleost fish: a review. *Journal of Fish Diseases* 25: 1127–1142.

NIWA (2017a). Yellowtail kingfish: Aquaculture. National Institute for Water and Atmospheric research (NIWA), Auckland, New Zealand [Online] Available from: https://www.niwa.co.nz/aquaculture/aquaculture-species/yellowtail-kingfish [Accessed 15 August 2017].

NIWA (2017b). Hapuku (grouper): Aquaculture. National Institute for Water and Atmospheric research (NIWA), Auckland, New Zealand [Online] Available from: https://www.niwa.co.nz/aquaculture/aquaculture-species/hapuku [Accessed 15 August 2017].

OIE (2016a). Aquatic Animal Health Code, 19th Edition. World Organization for Animal Health. Paris, France [Online] Available from: http://www.oie.int/international-standard-setting/aquatic-code/access-online/ [Accessed 15 March 2017].

OIE (2016b). Manual of diagnostic tests for aquatic animals. World Organization for Animal Health. Paris, France [Online]. Available from: http://www.oie.int/international-standard-setting/aquatic-manual/access-online/ [Accessed 17 March 2017].

Olveira JG, Souto S, Dopazo CP, Bandin I (2013). Isolation of betanodavirus from farmed turbot *Psetta maxima* showing no signs of viral encephalopathy and retinopathy. *Aquaculture* 406–407: 125–130.

Nguyen HD, Nakai T, Muroga K (1996). Progression of striped jack nervous necrosis disease (SJNNV) infection in naturally and experimentally infected striped jack *Pseudocaranx dentex* larvae. *Diseases of Aquatic Organisms* 24: 99–105.

Sano M, Nakai T, Fijan N (2011). Viral diseases and agents of warm water fish. pp 166–244. *In*: Woo PTK, Bruno DW (*Eds.*). *Fish Diseases and Disorders: Volume 3 - Viral, Bacterial and Fungal Infections*, 2nd edition. CABI Publishers, Wallingford, UK, 930 p.

Seafood New Zealand (2013). New Zealand seafood exports: Report 10a. Seafood exports by species by country, calendar year to December 2013 (final). Report prepared by Seafood New Zealand. Wellington, New Zealand, 171 p.

Shetty M, Maiti B, Santhosh KS, Venugopal MN, Karunasagar I (2012). Betanodavirus of marine and freshwater fish: Distribution, genomic organisation, diagnosis and control measures. *Indian Journal of Virology* 23(2): 114–123.

Tanaka S, Kuruyama I, Nakai T, Miyazaki T (2003). Susceptibility of cultured juveniles of several marine fish to the sevenband grouper nervous necrosis virus. *Journal of Fish Diseases* 26(2): 109–115.

Tubbs L, Lee P, Diggles B, Jones JB, Sheppard M, Sim-Smith C (2007). A review of aquatic diseases of significance to New Zealand. Final research report for project ZBS 2005-17. National Institute of Water and Atmospheric Research, Wellington, New Zealand, 452 p.

Yoshikoshi K, Inoue K (1990). Viral nervous necrosis in hatchery-reared larvae and juveniles of Japanese parrotfish, *Oplegnathus fasciatus* (Temminck & Schlegel). *Journal of Fish Diseases* 13(1): 69–77.

22. *Oncorhynchus masou* herpesvirus

22.1. Hazard identification

22.1.1. Aetiological agent

Oncorhynchus masou herpesvirus (OMV), or salmonid herpesvirus 2 (SalHV-2), is a linear double-stranded, enveloped icosahedral DNA virus. It is one of several isolates classified as *Salmonid herpesvirus* 2, within Genus *Salmonivirus* of the Family Herpesviridae (Bandin & Dopazo 2011). It was first described from fry of Japanese salmon *O. masou* (Sano 1976).

22.1.2. OIE status

Infection with *Oncorhynchus masou* herpesvirus (OMV) is not an OIE-listed disease (OIE 2016a). It is listed in the OIE Aquatic Manual (OIE 2016b) as an economically significant aquatic disease.

22.1.3. New Zealand status

Oncorhynchus masou herpesvirus is reported only from Pacific salmon (*Oncorhynchus* spp.). There are no reports of OMV from New Zealand salmonids. Infection with OMV is a notifiable disease in New Zealand (Anon. 2016). It has been assumed to be exotic.

22.1.4. Epidemiology

Oncorhynchus masou herpesvirus is an oncogenic virus also known as coho salmon herpesvirus, coho salmon tumour virus, nerka virus, *Oncorhynchus kisutch* virus, rainbow trout herpesvirus, rainbow trout kidney virus, salmonid herpesvirus 2 and yamame tumour virus (CABI 2016).

Susceptible species are restricted to farmed and wild Salmonidae of genus *Oncorhynchus* (Table 18). All ages of fish may be infected (Kimura *et al*. 1983a) and wild salmonids may act as reservoirs of disease (Furihata *et al.* 2003). *Oncorhynchus masou* herpesvirus was originally reported from the coastal marine waters, rivers and fresh waters of Japan, eastern Asia and Kamchatka, Russia (Yoshimizu & Nomura 2001; OIE 2016b), but has also spread to Kuwait (Anon. 2007). This transfer has occurred largely through the movement of eyed eggs in aquaculture.

Table 18. Families and Species of Fish Susceptible to *Oncorhynchus Masou* Herpesvirus (OMV)

Family	Host Species
Salmonidae	Chum salmon (*Oncorhynchus keta*), coho salmon (*O. kisutch*), cherry or masu salmon (*O. masou masou*), rainbow trout (*O. mykiss*), sockeye salmon (*O. nerka*)

Mortality ranges from 10-80% during OMV epidemics in Japanese aquaculture (Kumagai *et al.* 1994; Yoshimizu 2012). External signs of infection may occur 13 days post-exposure (Anon. 2007). These include darkened coloration of the skin and fins, with the progressive development of haemorrhagic lesions and tumours around the mouth and on the skin surface (Yoshimizu *et al.* 1993). These initially develop as small skin ulcers caused by oedema and haemorrhage of the endothelial cells of the blood capillaries, haematopoietic tissues and hepatocytes, usually with medium to high mortality (Kimura *et al.* 1981).

Infection progresses to a second stage from 4–18 months post-infection, where surviving fish develop antibodies that appear to confer immunity but the serological processes are poorly known (Anon. 2007; CABI 2016). Epitheliomas slowly develop in these fish mainly around the mouth and operculum, the caudal fin and on the skin surface, but with lower mortality. These fish become disfigured and unmarketable (Yoshimizu 1996) causing economic loss in Japanese salmonid aquaculture (Furihata *et al.* 2003). Internal signs of infection include intestinal haemorrhages and white lesions on the liver tissues (Yoshimizu *et al.* 1988). Surviving farmed, feral or wild fish can become covert carriers of infection where OMV is shed via sexual products at the time of spawning (CABI 2016). Under natural conditions, survivors remain persistently infected with virus and they continue to shed the virus until disease maturation (Yoshimizu *et al.* 1993).

Transmission is horizontal, directly through the water column, through a vector or by fomites (Yoshimizu *et al*. 1993). Infectious virus is shed via the faeces, urine, sexual products and skin surface of infected fish. While true vertical transmission does not occur, OMV may be transmitted through contamination of the egg surface in hatcheries (Furihata *et al*. 2003).

Transmission may also occur through other fish species, parasitic invertebrates, as well as by piscivorous birds and mammals (OIE 2016b).

Herpesviruses are relatively environmentally fragile. OMV survives in fresh water for up to 7 days at 10°C and for at least 14 days where temperatures do not exceed 5°C (Yoshimizu *et al.* 2005). OMV remains viable for up to 17 days after freezing to -20°C and is denatured at temperatures above 15°C (Wolf 1988).

Control of OMV is by iodophor treatment of eyed eggs (Stone *et al.* 1997), but an inactivated virus vaccine against OMV is also available (CABI 2016). OMV is denatured by UV light (at 10^4 µW second cm^{-2}) and by antivirals including phosphonoacetate (PA) and acyclovir (ACV) (Kimura *et al*. 1981, 1983a, 1983b). The related channel catfish virus (CCV) is inactivated by heat treatment (to at least 60°C for 1 hour) (Robin & Rodrigue 1980). Smail & Munro (2001) reported that OMV is also heat inactivated and denatured at pH 3.

OMV is a viral disease of Pacific salmon (*Oncorhynchus*) including chum salmon (*O. keta*), coho salmon (*O. kisutch*), cherry or masu salmon (*Oncorhynchus masou masou*), rainbow trout (*Oncorhynchus mykiss*) and sockeye salmon (*Oncorhynchus nerka*). Suseptible Pacific salmon species in New Zealand include the chinook salmon (*O. tshawytscha*), rainbow trout (*O. mykiss*) and sockeye salmon (*O. nerka*) (Roberts *et al*. 2001).

22.2. Risk assessment

22.2.1. Entry assessment

OMV is relatively non-host specific, affecting several *Oncorhynchus* species, but its distribution is restricted to Japan, eastern Russia and eastern Asian waters, as well as Kuwait (Yoshimizu & Nomura 2001; Anon. 2007; OIE 2016b).

Surviving adult fish act as carriers of disease (OIE 2016b). While grossly infected fish would be unlikely to pass visual infection, the virus may be present in the skin and head tissues of sub-clinically infected eviscerated fish and these may be present in the commodity.

OMV is resistant to the environmental conditions likely to be encountered in fish storage, transportation and processing (Sano 1976). It is unaffected by freezing for up to 17 days (Wolf

1988). If viral particles are released in wastewater from commercial processing, they may survive in fresh water for at least 7 days (Yoshimizu *et al.* 2005).

The likelihood of entry is assessed as non-negligible.

22.2.2. Exposure assessment

OMV is predominantly a virus affecting juvenile salmonids (OIE 2016b). New Zealand aquaculture is dependent on supplies of high quality water and little or no treatment of source water is commonly practiced (Sim-Smith *et al.* 2014). Exposure of wild and farmed salmonids may occur if OMV is released in the wastewaters from commercial fish processing, for instance, upstream of a rainbow trout hatchery. Once established in the wild, OMV may be difficult or impossible to control effectively. OMV may be spread by fomites within the aquaculture and transport industries, while vectors including invertebrate pathogens and piscivorous birds (OIE 2016b) may spread OMV between catchments.

The likelihood of exposure is assessed as non-negligible.

22.2.3. Consequence assessment

All salmonids are considered susceptible to OMV (OIE 2016b). New Zealand salmonid stocks have been isolated from external salmonid diseases since the 1900s, so the consequences of the establishment of OMV for this fishery and its related industries including tourism would be extreme, in direct stock losses, in requirements for culling, disinfection and treatment of infected facilities, and in the on-going requirement to treat source water.

The New Zealand aquaculture industry is largely dependent on Chinook salmon (*Oncorhynchus tshawytscha*), with exports valued at $63 million in 2011(Aquaculture New Zealand 2014). In addition, there would also be major social and economic effects for the industries associated with recreational and tourist trout and salmon fisheries for rainbow trout (*O. mykiss*) and brown trout (*Salmo trutta*) (Fish & Game 2014). While the dollar value of New Zealand's freshwater fisheries is not fully known, the Lake Taupo trout fishery alone is valued at $70 to 80 million (Marsh & Mkwra 2013). Rainbow trout and brown trout are not currently farmed for aquaculture in New Zealand (Fish & Game 2014), but are raised in hatcheries in support of the freshwater recreational fisheries. OMV is highly pathogenic, particularly for juvenile fish, and can cause mortalities of up to 100% (OIE 2016b).

The consequences of establishment of OMV are therefore assessed as non-negligible.

22.2.4. Risk estimation

The entry, exposure and consequence assessments for OMV are non-negligible, so the risk estimate is non-negligible. Under the procedures followed in the risk assessment, OMV is assessed to be a risk in the commodity and risk management measures may be developed.

22.3. Risk management

Oncorhynchus masou herpesvirus has been identified as a risk in the commodity. Infection with OMV is no longer a disease notifiable to the OIE (OIE 2016a) and no requirements for dedicated monitoring exist. While the OIE *Aquatic Code* does not provide specific guidance on measures

that would ensure the destruction of the virus, mitigation measures are provided in the OIE Aquatic Manual (OIE 2016b). It is assumed that compliance with the approved processed states for OIE-listed diseases (Appendix 3) would also eliminate OMV from the commodity and be a viable risk management option.

Wild and farmed fish of family Salmonidae are considered susceptible to OMV (Table 18), and all ages of fish may be infected (OIE 2016b). Species declaration would substantially reduce the pathogen load of OMV and be a viable risk management option. Restriction of the commodity to wild-caught fish (not from aquaculture) would have no effect on pathogen load and is not a viable risk management option.

OMV is reported from the coastal marine and fresh waters of Japan, eastern Asia, the Kamchatka Peninsula and Kuwait (Yoshimizu & Nomura 2001; Anon. 2007; OIE 2016b). Where country/zone freedom is approved through the MPI Country Approval Procedures, this option should substantially reduce the occurrence of *Oncorhynchus masou* herpesvirus in the commodity. Acceptance of a declaration of country/zone freedom by MPI is a viable risk management option.

OMV is generally associated with the head and skin, so removal of the gills, or the head and gills, should slightly reduce the pathogen load of OMV in the commodity. However, virus present in the skin of carrier fish would be retained in the commodity, so processing to the skin-off fillet state should moderately reduce the pathogen load of OMV. Processing to the skin-off fillet state is a viable risk management option.

OMV has low resistance to temperature extremes, so either frozen storage (to below -20°C for at least 18 days), or moderate heat treatment (cooking to at least 60°C for 1 hour), would eliminate OMV from the commodity (Robin & Rodrigue 1980; Wolf 1988; Smail & Munro 2001; Anon. 2007). Frozen storage and heat treatment are viable risk management options.

22.3.1. Risk management options

Oncorhynchus masou herpesvirus (OMV) is reported from fish in family Salmonidae, which are considered likely to be present in the commodity. Other families have not been associated with OMV. Therefore species declaration indicating the commodity is not originated from family Salmonidae should substantially reduce the occurrence of OMV in the commodity.

For the commodities originated from family Salmonidae, one or a combination of the following additional options could also be considered to effectively manage the risk.

Where country/zone freedom from OMV is accepted by MPI:

Option 1

Acceptance of country/zone freedom by MPI should substantially reduce the occurrence of OMV, so the commodity may be imported without any further restrictions.

Where country/zone freedom from OMV is not accepted by MPI or not available:

Option 2

Processing to a state consistent with the approved states for OIE-listed diseases (Appendix 3) should eliminate OMV. When these provisions are met, the commodity could be imported without any further restrictions.

Where the imported commodity is not in a state consistent with the approved states for OIE-listed diseases (Appendix 3):

Option 3

Heat treatment (by cooking to at least 60°C for at least 15 minutes) should eliminate OMV. When this provision is met, the commodity could be imported without any further restrictions; or,

Option 4

Frozen storage (to below -20°C for at least 18 days) should eliminate OMV. When this provision is met, the commodity could be imported without any further restrictions; or,

Option 5

Further processing (to the skin-off fillet state) should moderately reduce the occurrence of OMV. When this provision is met, the commodity could be imported without any further restrictions.

22.4. References

Anonymous (2007). *Oncorhynchus masou* virus disease (Salmonid Herpesvirus Type 2 disease). Centre for Food Security and Public Health, Iowa State University, Ames, Iowa, United States of America [Online] Available from: http://www.cfsph.iastate.edu/Factsheets/pdfs/oncorhynchus_masou_virus_disease.pdf [Assessed 16 March 2017].

Anonymous (2016). Biosecurity (Notifiable Organisms) Order 2016 (2016/73). New Zealand Government, Wellington, New Zealand, 4 p.

Aquaculture New Zealand (2014). Seafood New Zealand: Key Facts for Our Industry 2013 [Online] Available from: http://aquaculture.org.nz/industry/overview/ [Accessed 8 June 2014].

Bandin I, Dopazo CP (2011). Host range, host specificity and hypothesized host shift events among viruses of lower vertebrates. *Veterinary Research* 42: 67 [Online] Available from: http://veterinaryresearch.biomedcentral.com/articles/10.1186/1297-9716-42-67 [Accessed 15 March 2017].

CABI (2016). *Oncorhynchus masou* virus disease. Pathogen datasheet. CABI International. Wallingford, United Kingdom [Online] Available from: http://www.cabi.org/isc/datasheet/71788 [Accessed 15 March 2017].

Fish and Game (2014). New Zealand Fish and Game Council [Online] Available from: http://www.fishandgame.org.nz/ [Accessed 24 September 2014].

Furihata M, Hosoe A, Takei K, Kohara M, Nakamura J, Motonishi A, Yoshimizu M (2003). Outbreak of salmonid herpesviral disease in cultured rainbow trout. *Fish Pathology* 38(1): 23–25.

Kimura T, Yoshimizu M, Tanaka M, Sannohe H (1981). Studies on a new virus (OMV) from *Oncorhynchus masou*. I. Characteristics and pathogenicity. *Fish Pathology* 15(3/4): 143–147.

Kimura T, Yoshimizu M, Tanaka M (1983a). Susceptibility of different fry stages of representative salmonid species to *Oncorhynchus masou* virus (OMV). *Fish Pathology* 17: 251–258.

Kimura T, Suzuki S, Yoshimizu M (1983b). *In vitro* antiviral effect of 9-(2-hydroxyethoxymethyl) guanine on the fish herpesvirus *Oncorhynchus masou* virus (OMV). *Antiviral Research* 3: 93–101.

Kimura T, Suzuki S, Yoshimizu M (1983c). *In vivo* antiviral effect of 9-(2-hydroxyethoxymethyl) guanine on experimental infection of chum salmon (*Oncorhynchus keta*) fry with *Oncorhynchus masou virus* (OMV). *Antiviral Research* 3: 103–108.

Kumagai A, Takahashi K, Fukuda H (1994). Epizootics caused by salmonid herpesvirus type 2 infection in maricultured coho salmon. *Fish Pathology* 29: 127–134.

Marsh D, Mkwra L (2013). Review of freshwater non-market value studies. Department of Economics, University of Waikato, Hamilton, New Zealand, 144 p.

OIE (2016a). Aquatic Animal Health Code, 19th Edition. World Organization for Animal Health. Paris, France [Online] Available from: http://www.oie.int/international-standard-setting/aquatic-code/access-online/ [Accessed 15 March 2017].

OIE (2016b). Manual of diagnostic tests for aquatic animals. World Organization for Animal Health. Paris, France [Online].Available from: http://www.oie.int/international-standard-setting/aquatic-manual/access-online/ [Accessed 17 March 2017].

Robin J, Rodrigue A (1980). Resistance of herpes channel catfish virus (HCCV) to temperature, pH, salinity and ultraviolet radiation. *Reviews of Canadian Biology* 39: 153–156.

Sano T (1976). Viral diseases of cultured fish in Japan. *Fish Pathology* 10: 221–226.

Seafood New Zealand (2013). New Zealand seafood exports: Report 10a. Seafood exports by species by country, calendar year to December 2013 (final). Report prepared by Seafood New Zealand. Wellington, New Zealand, 171 p.

Sim-Smith C, Faire S, Lees A (2014). Managing biosecurity risk for business benefit. Report prepared for the Ministry for Primary Industries by Coast and Catchment Ltd, Auckland, New Zealand, 214 p.

Smail DA. Munroe ALS (2001). The Virology of Teleosts. pp 169–253. *In:* Roberts RJ (*Ed.*) Fish Pathology, 3rd Edition, W.B. Saunders, London, 472 p.

Stone MAB, MacDiarmid SC, Pharo HJ (1997). Import health risk analysis: Salmonids for human consumption. Biosecurity New Zealand, MAF Regulatory Authority. Wellington, New Zealand, 269 p.

Wolf K (1988). Fish viruses and fish viral diseases. Cornell University Press, Ithaca, New York, United States of America, 476 p.

Yoshimizu M (1996). Salmonid herpesvirus. *Virus* 46: 53–59.

Yoshimizu M (2012). *Oncorhynchus masou* virus disease. Chapter 2.2.13 American Fisheries Society [Online] Available from: http://www.afs-fhs.org/perch/resources/14069233132.2.13omvdisease2014.pdf [Accessed 22 March 2017].

Yoshimizu M, Takizawa H, Kimura T (1986). U.V. susceptibility of some fish pathogenic viruses. *Fish Pathology* 21: 47–52.

Yoshimizu M, Tanaka M, Kimura T (1988). Histopathological study of tumours induced by *Oncorhynchus masou* virus (OMV) infection. *Fish Pathology* 23(2): 133–138.

Yoshimizu M, Nomura T, Ezura Y, Kimura T (1993). Surveillance and control of infectious hematopoietic necrosis virus (IHNV) and *Oncorhynchus masou* virus (OMV) of wild salmonid fish returning to the northern part of Japan 1976-1991. *Fisheries Research* 17: 163–173.

Yoshimizu M, Nomura T, Furihata M, Montoshi A (2001). Epidemiological study of *Oncorhynchus masou* virus in salmonid fish. Proceedings of the 10th International Symposium on Veterinary Epidemiology and Economics, Vina del Mar, Chile, 5 p.

Yoshimizu M, Yoshinaka T, Hatori S, Kasai H (2005). Survivability of fish pathogenic viruses in environmental water, and inactivation of fish viruses. *Bulletin of the Fisheries Research Agency, Supplement 2*: 47–54.

23. Pilchard orthomyxovirus/Tasmanian salmon orthomyxovirus

23.1. Hazard identification

23.1.1. Aetiological agent

Pilchard orthomyxovirus (POMV) was first identified in 1998 from wild pilchard (*Sardinops sagax/ S. neopilchardus*) in South Australia, but was not associated with clinical disease (SCAHH 2015, Diggles 2016). POMV is genetically identical to Tasmanian salmon orthomyxovirus (SOMV) which was first identified in Atlantic salmon (*Salmo salar*) farmed in Tasmania during 2006. SOMV was not associated with aquatic disease, until 2012 (SCAHH 2015). POMV is considered an unclassified virus within the family Orthomyxoviridae, that is distinct from Infectious salmon anaemia virus (ISAV) (OIE 2016a).

23.1.2. OIE status

Infection with POMV is not an OIE- listed disease (OIE 2016b).

23.1.3. New Zealand status

POMV has not been reported from Atlantic salmon (*S. salar*) in New Zealand. There is no evidence to suggest that POMV occurs in pilchards (*S. sagax*) in New Zealand (Diggles 2016). It is therefore assumed to be exotic.

23.1.4. Epidemiology

POMV is an emerging disease that has previously been reported from farmed Atlantic salmon (*S. salar*) in Macquarie Harbour Tasmania, Australia, as SOMV since 1998 (SCAAH 2015). It has also been reported in wild pilchards in South Australia and Tasmania since 1998, but without clinical signs of infection (SCAAH 2015). It is unlikely that POMV-infected pilchards would be able to migrate from Australia to New Zealand (Diggles 2016). Disease outbreaks in farmed salmon are thought to occur through cohabitation with sub-clinically infected wild pilchards that school around salmon sea-cages (SCAAH 2015).

While POMV is reported from *S. salar* and *S. pilchardus* (Table 19), its host range is effectively unknown (Diggles 2016). However, it is likely that fish known to be susceptible to the related orthomyxovirus ISAV may be susceptible to POMV. Salmonids including brown trout (*Salmo trutta*), rainbow trout (*O. mykiss*), chum (*O. keta*), coho salmon (*O. kitsutch*) and char (*Salvelinus alpinus*) can be experimentally infected with ISAV, together with non-salmonids including herring (*Clupea harengus*), alewife (*Alosa pseudoharengus*) (Rimstad *et al.* 2011) (Table 20). Chinook salmon (*Oncorhynchus tshawytscha*) are considered resistant to the related orthomyxovirus ISAV (Diggles 2016).

Table 19. Families and Species of Fish Potentially Susceptible to Pilchard Orthomyxovirus (POMV)

Family	Species Known to be Susceptible to POMV
Clupeidae	Pacific sardine/pilchard (*Sardinops sagax*), pilchard (*Sardina pilchardus*)
Salmonidae	Atlantic salmon (Salmo salar)
Family	**Species Potentially Susceptible to POMV (may be experimentally infected with related orthomyxovirus ISAV)**
Clupeidae	Alewife (*Alosa pseudoharengus*), Atlantic herring (*Clupea harengus*),
Gadidae	Atlantic cod (*Gadus morhua*)
Salmonidae	Brown trout (*Salmo trutta*), rainbow trout (*O. mykiss*), chum (*O. keta*), coho salmon (*O. kitsutch*), char (*Salvelinus alpinus*)

Infection in farmed *Salmo salar* is generally apparent in juvenile farmed fish, (with reported mortality of up to 1.3 million fish in 2017 (PROMED 2017).

Orthomyxovirus transmission may occur horizontally through the water column by the faecal-oral route, or by direct contact between fish (OIE 2016a, 2016b), while vertical transmission may also occur (Vike *et al.* 2009; Rimstad *et al.* 2011; Marshall *et al.* 2014; OIE 2016b).

Infection can occur across the gill membranes, the intestinal wall following ingestion, or through the skin surface following tissue damage (Rimstad *et al*. 2011; OIE 2016b).

Orthomyxoviruses including ISAV can be transferred through transport melt-water or in contaminated tissue wastes, mucous and fish blood from fish processing discharges. They can also be transferred by cross-infection of fishing boats and gear, transport and factory processing equipment (OIE 2016b).

ISAV remains viable in moist decaying fish (OIE 2016a). It is unaffected by salinity, remaining viable for up to 10 days at 15°C, or 14 days at 4°C in marine or fresh waters (Rimstad *et al.* 2011; OIE 2016b). It is unaffected by freezing (Tubbs *et al.* 2007; Rimstad *et al.* 2011) and survives heat treatment (up to 30 minutes at 56°C) (OIE 2016b). It is tolerant of acidic conditions, surviving at pH 4.5 for more than 24 hours and at pH 12 for up to 24 hours, but is inactivated after 30 minutes at pH 4 (Rimstad *et al.* 2011). It remains viable after passage through the avian digestive tract and may be transferred by scavenging piscivorous birds (OIE 2016b). It is inactivated by bleach (100–1000 mg L^{-1} for 10 minutes), by iodophor disinfectants (100 to 200 mg L^{-1} for 5–10 minutes), by ozone treatment (8 mg L^{-1} min^{-1} for 3 minutes) and by UV radiation (Oye & Rimstad 2001; Tubbs *et al*. 2007).

Other pathways may also allow POMV to enter New Zealand. Pilchard (*Sardinops sagax*) is currently imported into New Zealand for use as fish bait and for processing into pet food (Blackwell 2013). As Orthomyxoviruses are unaffected by freezing (OIE 2016a), POMV is likely to remain viable if present in frozen pilchards imported through these pathways. Entry through fish bait is out of scope of this risk assessment.

Given the lack of knowledge of the host range of POMV (Table 20), marine fish hosts in New Zealand are assumed to include pilchard (*Sardinops sagax*) and sea run salmonids including brown trout (*Salmo trutta*), char (*Salvelinus fontinalis*) and sockeye salmon (*Oncorhynchus nerka*). Sea-run rainbow trout (*O. mykiss*) are uncommon in New Zealand, while Atlantic salmon (*S. salar*) exists in New Zealand in a very small feral landlocked population in the upper Waiau River catchment, and in a broodstock population maintained in a hatchery farm in Wanaka (NIWA 2018).

23.2. Risk assessment

23.2.1. Entry assessment

Orthomyxoviruses including POMV remain viable in fresh chilled or frozen fish (OIE 2016a, 2016b). Sub-clinically infected fish such as pilchards (*S. sagax*) may pass visual inspection and be present in the commodity (Tubbs *et al.* 2007; Diggles 2016).

The likelihood of entry is assessed to be non-negligible.

23.2.2. Exposure assessment

Potential hosts of POMV in New Zealand include pilchards (*Sardinops sagax , Sardina pilchardus*) and sea-run brown trout (*Salmo trutta*). Orthomyxoviruses are environmentally resistant, so wild fish could be exposed through waste water and organic material discarded from commercial fish processing. POMV can be transferred horizontally, by cohabitation between wild sub-clinically infected pilchards (*Sardinops sagax*) and Atlantic salmon (*S. salar*) held in sea cages in Tasmania. While infection in pilchards occurs without clinical signs, it is unlikely that sub-clinically infected pilchards would migrate from Australia to New Zealand (Diggles 2016).

There are no sea-run Atlantic salmon in New Zealand (NIWA 2018). While a potential pathway for infection may exist if the small farmed population were to be fed with infected pilchards, commercial salmonid production in New Zealand uses pelletised feed (Aquaculture New Zealand 2018). In addition, recreational anglers use unbaited fly fishing techniques to target these fish (Fish & Game 2014). The likelihood of exposure of Atlantic salmon (*S. salar*) is therefore so low as to be negligible.

Salmonid aquaculture in New Zealand is focused on Chinook salmon (*O. tshawytscha*), farmed in fresh waters and in sea cages where they are fed with pelletised feed (Aquaculture New Zealand 2018). Chinook salmon are considered resistant to IHNV (OIE 2016a), although their susceptibility to POMV is unknown (Diggles 2016). The likelihood of exposure is assessed to be negligible.

Other potential salmonid hosts occur in fresh waters, but it is considered unlikely that sufficient contact with infectious material would occur that would allow the establishment and maintenance of POMV in New Zealand.

In comparison to the potential exposure through other pathways, such as fish bait, the likelihood of exposure through fish imported for human consumption is assessed to be negligible. Under the procedures followed in this risk assessment, POMV is assessed to not currently be a risk in the commodity. Risk management measures are not necessary.

23.3. References

Aquaculture new Zealand (2018). Feed [Online] Available from: https://www.aquaculture.org.nz/environment/feed-2/ [Accessed 10 July 2018].

Blackwell RG (2013). Baitfish use in New Zealand: recent trends. Memorandum prepared for the Animal Imports team by Animals and Aquatics team, Ministry for Primary Industries, Wellington, New Zealand. 22 p.

Diggles BK (2016). Updated disease risk assessment report – Relocation of salmon farms in Marlborough Sounds, New Zealand. DigsFish Services Report: DF 16-01, 80 p.

DPIPWE (2015). Sampling for POMV, TSRV and Tasmanian RLO PCR. Tasmanian Department of Primary Industries, Parks, Water & Environment Animal Health Laboratory.

Fish and Game (2014). New Zealand Fish and Game Council [Online] Available from: http://www.fishandgame.org.nz/ [Accessed 24 September 2014].

Marshall (2014). Marshall SH, Ramirez R, Labra A, Carmona M. Munoz C (2014). Bona-fide evidence for natural vertical transmission of infectious salmon anaemia virus in freshwater brood stocks of farmed Atlantic salmon (*Salmo salar*) in Southern Chile. *Journal of Virology* 88(11): 6012–6018.

NIWA (2018). Identification guides and fact sheets [Online] Available from: https://www.niwa.co.nz/freshwater-and-estuaries/nzffd/NIWA-fish-atlas/fish-species/species/ [Accessed 10 July 2018].

OIE (2016a). Aquatic Animal Health Code, 19th Edition. World Organization for Animal Health. Paris, France [Online] Available from: http://www.oie.int/international-standard-setting/aquatic-code/access-online/ [Accessed 15 March 2017].

OIE (2016b). Manual of diagnostic tests for aquatic animals. World Organization for Animal Health. Paris, France [Online] Available from: http://www.oie.int/international-standard-setting/aquatic-manual/access-online/ [Accessed 17 March 2017].

Oye AK, Rimstad E (2001). Inactivation of infectious salmon anaemia virus, viral haemorrhagic septicaemia virus and infectious pancreatic necrosis virus in water using UVC irradiation. *Diseases of Aquatic Organisms* 48: 1–5.

PROMED (2017). Pilchard orthomyxovirus – Australia (Tasmania) Farmed salmon [Online] Available from: http://www.promedmail.org/direct.php?id=20171221.5517876 [Accessed 9 July 2018].

Rimstad Ee, Dale OB, Dannevig BH, Faulk K (2011). Infectious salmon anaemia, pp 143–165 *In:* Woo PTK, Leatherland JF, Bruno DW (*Eds*.). Fish Diseases and Disorders (Volume 3), CABI Wallingford, United Kingdom, 930 p.

SCAAH (2015). Sub-committee on Aquatic Animal Health SCAAH Newsletter #5, June 2015 [Online] Available from:. http://apfa.com.au/wp-content/uploads/2015/01/SCAAH-Newsletter-2015-7-July-2015.pdf [Accessed 9 July 2018].

Tubbs L, Lee P, Diggles B, Jones JB, Sheppard M, Sim-Smith C (2007). A review of aquatic diseases of significance to New Zealand. Final research report for project ZBS 2005-17. National Institute of Water and Atmospheric Research, Wellington, New Zealand, 452 p.

Vike S, Nylund S, Nylund A (2009). ISA virus in Chile: Evidence of vertical transmission. *Archives in Virology* 154: 1–8.

24. Piscine aquareovirus, including salmon, Tasmanian salmon, turbot and other related aquareoviruses

24.1. Hazard identification

24.1.1. Aetiological agent

Piscine aquareoviruses (PRV) were first isolated from cyprinid fish (Plumb *et al.* 1979). They are non-enveloped double stranded icosahedral RNA viruses, classified within the family Reoviridae (Lupiani *et al.* 1993; Ke *et al.* 2011). Piscine aquareovirus is currently considered to represent a group of seven closely associated species (Aquareovirus A – Aquareovirus G), each comprised of several isolates (Lupiani *et al.* 1993; Bustos *et al.* 2011; Ke *et al.* 2011; King 2011; Blindheim *et al.* 2015). These have previously been considered as a species complex of genogroups (Genogroup A - Genogroup G) (Fauquet *et al.* 2005) (Table 20).

Associated, but uncharacterised aquareoviruses include chub salmon reovirus (CHRV), golden idle reovirus (GIRV) (Attoui *et al.* 2002), Hubei grass carp reovirus (Fan *et al.* 2013), landlocked salmon reovirus (LSRV), rainbow trout and bluegill reovirus (Meyers 1980, 1983) and tench reovirus (TNRV) (Fauquet *et al.* 2005). The actual relationships between these isolates remain unclear (Palacios *et al*. 2010; Biering & Garseth 2012; Kristoffersen *et al.* 2013).

24.1.2. OIE status

Disease caused by PRV and associated aquareoviruses is not notifiable to the OIE (OIE 2016).

24.1.3. New Zealand status

PRV and associated aquareoviruses are considered exotic (Cobb 2008) and have not subsequently been reported from New Zealand (B. Jones, *pers. comm.* 2015). Infection with PRV is not a notifiable disease in New Zealand (Anon. 2010).

24.1.4. Epidemiology

Aquareovirus diseases are of variable pathogenicity. Mortalities commonly range from 5% and 20% (Hetrick & Hedrick 1993; Aldrin *et al.* 2010; Palacios *et al*. 2010; Roberts 2012), but some aquareoviruses, such as grass carp reovirus (GCRV), may be highly pathogenic (Chen & Jiang 1984; Attoui *et al.* 2002; Crane & Carlile 2008; Zainathan 2012; Zhang & Gui 2015). Mortality may be influenced by factors including husbandry-related stress (Kongtorp *et al*. 2004b), or the onset of higher spring water temperatures (Zainathan 2012). Morbidity is likely to be higher than indicated by field observation, as infection typically occurs with few external signs (Garseth *et al.* 2012).

The piscine aquareovirus species complex represents a disease complex consisting of up to seven isolates (King *et al.* 2011), although the actual relationships between the isolates are unclear (Palacios *et al*. 2010; Biering & Garseth 2012; Kristoffersen *et al.* 2013; Zhang & Jian-Fang 2015). Most research has focussed on salmonid aquareoviruses, but non-salmonid hosts include bass (Moronidae), carp (Cyprinidae), catfish (Ictaluridae), smelt (Osmeridae) and turbot (Scophthalmidae). Some host species may be infected by several isolates (King *et al.* 2011; Chen *et al.* 2015).

Table 20. Relationships Between Piscine Aquareovirus Isolates (A-G) and Associated Fish Aquareoviruses (After King *et al.* 2011)

***Aquareovirus* Species**						
A	**B**	**C**	**D**	**E**	**F**	**G**
Angelfish reovirus (AFRV) Atlantic halibut reovirus (AHRV) Atlantic salmon reovirus (HBRV)/heart and skeletal muscle inflammation (HSMI) Atlantic salmon reovirus 2 (ASRV) Tasmanian Atlantic salmon reovirus TSRV (TasSRV) Chinook salmon reovirus DRC (DRCRV) Chum salmon reovirus CS (CSRV) Masou salmon reovirus MS (MSRV) Smelt reovirus (SRV) Striped bass reovirus (SBRV) Also included: Pacific threadfin reovirus (PTRV) *Scophthalmus maximus* reovirus (SMReV)	Chinook salmon reovirus B (GRCV) Chinook salmon reovirus LBS (LBSV) Chinook salmon reovirus ICR (ICRV) Chinook salmon reovirus YRC (YRCV) Coho salmon reovirus CSR (CSRV) Coho salmon reovirus ELC (ELCV) Coho salmon reovirus SCS (SCSV)	Golden shiner reovirus (GSRV) Grass carp reovirus (GCRV) White bream reovirus (WBRV)	Channel catfish reovirus (CCRV)	Turbot reovirus (TRV)	Chum salmon reovirus (PSRV) Coho salmon reovirus (SSRV)	American grass carp reovirus (AGCRV)

Aquareovirus A isolates

Striped bass reovirus (SBRV) affects striped bass (*Morone saxatalis*) with low pathogenicity (Harrell 1997). Smelt reovirus (SRV) causes high mortality, but is only reported from landlocked freshwater Canadian smelt (*Osmerus mordax*) (Marshall *et al.* 1990). Angelfish reovirus (AFRV) is only reported from angelfish (*Pomacanthus semicirculatus*), a non-commercial or aquarium species (Varner & Lewis 1991). Turbot reovirus (SMReV) is reported from turbot (*Scophthalmus maximus*) aquaculture in China (Ke *et al.* 2011), but with low pathogenicity (Cusack *et al.* 2001).

Atlantic halibut reovirus (AHRV) is only reported from Atlantic halibut (*Hippoglossus hippoglossus*) in Canada, Scotland and Norway (Blindheim *et al.* 2015). As *H. hippoglossus* is a colder-water flatfish species than *S. maximus*, cohabitation is considered unlikely. It is likely that AHRV is a separate pathogen to SMReV (Blindheim *et al.* 2015). These isolates are not considered further.

Atlantic salmon reovirus 1 (HBRV) and Atlantic salmon reovirus 2 (ASRV) are considered identical (Mikalsen *et al.* 2014) and cause heart and skeletal inflammation of Atlantic salmon (HSMI). This has been reported from farmed Atlantic salmon (*S. salar*) in Norway and Scotland (Johansen *et al*. 2008), Chile (Kibenge *et al.* 2013) and Denmark (Mikkelsen *et al*. 2014) with low mortality (20%), but with high morbidity causing myocarditis and red skeletal muscle necrosis (Martinez-Rubio *et al.* 2012). HSMI has recently been identified in farmed *S. salar* in British Columbia, Canada (Di Cicco *et al.* 2017).

Chinook salmon reovirus (DRCRV) affects farmed Chinook salmon (*O. tshawytscha*), sockeye salmon (*O. nerka*) and Atlantic salmon (*S. salar*) in United States aquaculture, usually with low mortality (Garver *et al.* 2016). Chum salmon reovirus (CSRV) affects chum salmon (*O. keta*) and masou salmon reovirus (MSRV) affects masu salmon (*O. masou*) in Japanese aquaculture (Winton *et al.* 1981; Lupiani *et al.* 1995).

Tasmanian salmon aquareovirus (TasSRV) is considered ubiquitous in Tasmania (Winton *et al.* 1989) and an emerging disease of wild and farmed Atlantic salmon (*S. salar*) (Garseth *et al*. 2012; Carlile *et al*. 2014). Infection occurs at a low incidence rate (6 to 15%) with generally low pathogenicity (Crane *et al.* 2000; Zainathan 2012; Carlile *et al.* 2014). High mortalities occur when summer water temperatures approach the upper temperature range for Atlantic salmon, suggesting temperature stress may be a pre-disposing factor to clinical infection (Zainathan 2012; Carlile *et al.* 2014). TasSRV is also reported from wild and farmed rainbow trout (*O. mykiss*) in Tasmania (Zainathan 2012; Carlile *et al.* 2014).

Pacific threadfin reovirus (PTRV) affects the tropical Pacific threadfin (*Polydactylus sexfilis*) farmed in Hawaiian aquaculture (Chambers *et al.* 2001), with high mortality (Crane *et al*. 2010).

Aquareovirus B isolates

Isolates reported from Chinook salmon (*O. tshawytscha*) from the United States include Green River Chinook salmon reovirus B (GRCV), Chinook salmon reovirus LBS (LBSV), Chinook salmon reovirus ICR (ICRV) and Chinook salmon reovirus YRC (YRCV) (King *et al*. 2011; Crane *et al*. 2010; Murphy *et al.* 2012).

Other isolates are reported from coho salmon (*O. kisutch*) aquaculture from the United States, including coho salmon aquareovirus SCS (SCSV) from Oregon (Winton 1989), coho salmon reovirus CSR (CSRV), coho salmon reovirus ELC (ELCV) (King *et al.* 2011), while Simpson

coho salmon reovirus (SCSRV) is reported from Alaskan aquaculture (Dopazo *et al.* 1996; Kibenge & Godoy 2016).

Aquareovirus C isolates

Golden shiner reovirus (GSRV) is highly pathogenic to the cyprinids golden shiner (*Notemigonus crysoleucas*) (Plumb *et al.* 1979), chub minnow (*Semotilus atromaculatus*) and fathead minnow (*Pimephales promelas*) (Crane *et al.* 2010). While farmed extensively as baitfish in the United States, these are minor species for human consumption (Page & Burr 2011). These isolates are not considered further.

Grass carp reovirus (GCRV) includes two genetically distinct isolates (GCRV-JX01 and JX02), causing significant disease, with mortalities of 70–95%. It is reported from farmed grass carp (*Ctenopharyngodon idella*), black carp (*Mylopharyngodon piceus*) and common carp (*Cyprinus carpio*), rare minnow (*Gobiocypris rarus*) and the topmouth gudgeon (*Pseudorasbora parva*) in Chinese and Asian aquaculture (Chen & Jiang 1984; Rangel *et al.* 1989; Ye *et al.* 2012; Wang *et al.* 2013; Liang *et al.* 2014; Zu *et al.* 2014). GCRV is also reported from tench (*Tinca tinca*) and chub (*Squalius cephalus*) in Europe (Ahne & Kolbl 1987). The white bream reovirus (WBRV) isolate is only reported from European white bream (*Blicca bjoerkna*) (Schachner *et al*. 2014).

Aquareovirus D isolates

Two isolates (CCRV and CCRV 730) are reported from channel catfish (*Ameiurus punctatus*), causing significant disease in Canadian aquaculture (Amend *et al.* 1984; Hedrick *et al.* 1984). CCRV has subsequently spread to channel catfish aquaculture in China (Xu *et al.* 2013).

Aquareovirus E isolates

Turbot reovirus (TRV) only occurs in turbot (*Scophthalmus maximus*) where it is associated with a mixed bacterial and viral infection causing low but economically significant mortalities in Spanish marine aquaculture (Lupiani *et al.* 1989). The TRV isolate does not replicate in rainbow trout (*Oncorhynchus mykiss*) (Rivas *et al.* 1995).

Aquareovirus F isolates

Two isolates causing clinical disease are reported from salmonids. Chum salmon reovirus (PSR) occurs in chum salmon (*O. keta*) in Japan (Winton *et al.* 1981). Coho salmon reovirus (SSRV) occurs in coho salmon (*O. kisutch*) (Subramanian *et al.* 1997).

Aquareovirus G isolates

This includes a single isolate, American grass carp reovirus (AGCRV) affecting farmed grass carp (*Ctenopharyngodon idella*) during winter (Goodwin *et al*. 2010; He *et al.* 2013).

Unassigned isolates

Other aquareoviruses include golden idle reovirus (GIRV) (Neukirch *et al.* 1999), which affects the ornamental fish golden ide (*Leuciscus idus melanotus*) (Attoui *et al.* 2002). As this is unlikely to cause significant disease in fish for human consumption, it is not considered further.

Landlocked salmon reovirus (LSRV) affects masu salmon (*O. masou*) only in Taiwanese aquaculture (Crane & Carlile 2010). Hubei grass carp reovirus causes significant disease in grass carp (*P. idella*) in Chinese aquaculture (Fan *et al.* 2013) and chub reovirus (CHRV) affects freshwater chub (*L. cephalus*) in German aquaculture (Murphy *et al*. 2012).

An undescribed reovirus causes significant mortality in rainbow trout (*O. mykiss*) and bluegill (*Lepomis macrochirus*) farmed in United States aquaculture (Meyers 1980, 1983). while tench reovirus (TNRV) affects European tench (*Tinca tinca*) (Fauquet *et al.* 2005). Snakehead reovirus (SKRV) affects snakehead (*Channa striata*) in Thailand (John *et al.* 2001).

PRV infection in salmonids is reported from over 417 marine farms in Norway, the United Kingdom and Chile (Biering & Garseth 2012), Canada and Alaska (Marty *et al.* 2014; Di Cicco *et al*. 2017). Infection in salmonids occurs in fresh water, but clinical signs are typically apparent 5 to 9 months after smolts are transferred to sea cages (Zainathan 2012). PRV has also been reported from freshwater salmonid farms (Garseth *et al*. 2012) and from wild caught marine Atlantic salmon in Ireland, as well as from British Columbia in sea-run brown trout (*S. trutta*), rainbow trout (*O. mykiss*) and cutthroat salmon (*O. clarkii*) (Garseth *et al.* 2012). It is also reported from farmed Atlantic salmon in Chile (Bustos *et al.* 2011), chum salmon (*O. keta*) and masou salmon (*O. masou*) in Japan and Taiwan (Winton *et al.* 1981; Hsu *et al*. 1989).

PRV infection in non-salmonids including wild and farmed stocks of American grass carp (*Ctenopharyngodon idella*), Atlantic horse mackerel (*Trachurus trachurus*), Atlantic herring (*Clupea harengus*), capelin (*Mallotus villosus*), golden idle (*Leuciscus idus melanotus*), silver smelt (*Argentina silas*) and tench (*Tinca tinca*) (Fauquet *et al*. 2005; Wiik-Nielsen *et al.* 2012).

Aquareovirus A has the widest host range, but *Aquareovirus B* appears limited in its host and geographical range. It is not otherwise possible to determine any correlations between aquareovirus species, host and geographical range, and pathogenicity (Crane & Carlile 2010).

The families and host species considered susceptible to PRV and associated aquareoviruses are given in Table 21.

Table 21. Families and Species of Fish Susceptible to Piscine Aquareovirus and Associated Aquareoviruses

Family	Host Species
Argentinidae	Silver smelt (*Argentina silus*)
Carangidae	Atlantic horse mackerel (*Trachurus trachurus*)
Centrarchidae	Bluegill (*Lepomis macrochirus*)
Channidae	Snakehead (*Channa striata*)
Clupeidae	Atlantic herring (*Clupea harengus*)
Cyprinidae	Black carp (*Mylopharyngodon piceus*), chub (*Squalius cephalus*), common carp (*Cyprinus carpio*), European white bream (*Blicca bjoerkna*), ide (*Leuciscus idus*), grass carp (*Ctenopharyngodon idella*), rare minnow (*Gobiocypris rarus*), tench (*Tinca tinca*), topmouth gudgeon (*Pseudorasbora parva*)
Ictaluridae	Channel catfish (*Ameiurus punctatus*)
Osmeridae	Capelin (*Mallotus villosus*)
Polynemidae	Sixfinger threadfin (*Polydactylus sexfilis*)
Salmonidae	Atlantic salmon (*S. salar*), brown trout (*S. trutta*), cutthroat salmon (*O. clarkii*), Chinook salmon (*O. tshawytscha*), chum salmon (*O. keta*), coho salmon (*O. kisutch*), masu salmon (*O. masou*), rainbow trout (*O. mykiss*)
Scopthalmidae	Turbot (*Scophthalmus maximus*)

Infection with aquareoviruses generally occurs with no clinical signs of infection, other than anorexia (Palacios *et al.* 2010; Garseth *et al*. 2012).

Internal signs include swelling of the digestive tract and liver (Castric 1997). Infection is initially focused on the red skeletal and heart muscle, with ventricular lesions, haemorrhage, necrosis and myositis, that may extend into the adjacent white muscle. Haemorrhage and necrosis also occur in

gill tissues, with multifocal necrosis of the liver and swelling of the spleen (Kongtorp *et al*. 2004b) and brain tissue (Wiik-Nielsen *et al.* 2012).

Infection quickly spreads to the liver, kidney, spleen, intestine and musculature, from 6–9 days post-infection (Liang *et al.* 2014), with substantial haemorrhage of the operculum, jaw, fin bases and gills of fish infected with CCRV (Kibenge & Godoy 2016). Infection then spreads to most host tissues, while the TRV isolates commonly invades the host macrophages, which aids the rapid spread of the virus throughout the host (Rivas *et al.* 1996a, 1996b).

Transmission of PRV is horizontal, through the water column (Watanabe *et al.* 2006) but details of transmission and infectivity remain unknown (Aldrin *et al.* 2010). Experimental infection may be induced by injection of tissue homogenates, or through cohabitation with infected fish (Watanabe *et al.* 2006).

The survival of PRV in marine and fresh waters is unknown (Wiik-Nielsen *et al*. 2012), but aquareoviruses are generally resistant to environmental conditions. They can remain viable for over a year (630 days) at 8°C and almost two weeks (13.3 days) at 26°C before T_{90} (90% loss of virus activity) in municipal wastewater, and for more than 66 days in digested sludge at 4°C (Wellings *et al.* 1976). They may remain viable in low-moisture processed food for 24 days at room temperature and for 2 months at 5°C (Pirtle & Beran 1991), as well as survive for almost six months (154 days) at 8°C and almost a week (6.6 days) at 26°C before T_{90} in distilled water was reached (McDaniels *et al.* 1983).

Aquareoviruses are heat labile to 56°C for up to 2 hours or 45°C for 7 days (John *et al.* 2001; Fauquet *et al*. 2005). They may remain viable after municipal sewage treatment or if discharged directly into marine waters (McDaniels *et al.* 1983). Reoviruses dumped in landfill may remain viable for up to 36 days (Ware 1980), but are unlikely to remain viable after passage through the digestive system of scavenging birds (John *et al*. 2001).

Reoviruses are generally ether-resistant, stable over a wide range of pH, although infectivity is reduced at pH < 3.0 (McDaniel *et al.* 1996; John *et al.* 2001). They are resistant to common chemical and physical disinfectants, but are inactivated by treatment with formalin (100 µg L^{-1} for 3 days at 37°C), high concentrations of chlorine (15 mg L^{-1} for 5 minutes), or isopropanol (42.5% for 15 minutes) (Rivas *et al.* 1994).

While vaccine treatment against aquareoviruses are available for carp and salmonids (Beck & Peatman 2015), no commercial vaccines are available for SMReV and TRV aquareoviruses.

Susceptible species in New Zealand include salmonids, silverside (*Argentina elongata*), mackerel (*Trachurus* spp.) and herring (*Clupea harengus*) (Paul 2000; Tubbs *et al.* 2007).

The New Zealand brill (*Colistium guntheri*) and turbot (*C. nudipinnis*) (Pleuronectidae) are related to Northern Hemisphere brill (*Scophthalmus rhombus*) and turbot (*S. maximus*) (Pleuronectidae) (Chanet 2003). Thus, it is reasonable to conclude that exotic diseases of turbot and brill (*Scophthalmus* spp.) may infect endemic brill and turbot species (C. Johnston, *pers. comm.* 2015).

24.2. Risk assessment

24.2.1. Entry Assessment

PRV is primarily associated with farmed Atlantic salmon in sea cages, but infection may also occur in fresh water (Garseth *et al.* 2012) and in wild fish (Carlile *et al.* 2014). Other salmonid species (Winton *et al.* 1981) and other marine fish species may also be affected (Palacios *et al.* 2010). Infection may occur with no external signs (Garseth *et al.* 2012), so these fish may pass visual inspection and be present in the commodity. Infection is mainly associated with the heart and associated red muscle, but commonly spreads to the skeletal musculature, brain and gill tissues (Kongtorp *et al.* 2004b). While evisceration would reduce viral titre, the virus is likely to remain in the eviscerated commodity.

The likelihood of entry of PRV and associated aquareoviruses in the commodity is assessed as non-negligible.

24.2.2. Exposure assessment

To establish infection in New Zealand, infected eviscerated product would have to become available for consumption by a susceptible fish host, in sufficient quantity and duration (Kahn *et al*. 1999). PRV may remain viable in the commodity, in the brain and gill tissues of fish heads, as well as in the skeletal musculature (Kongtorp *et al.* 2004b) and be present in the blood-water discharge and fish offal discarded after fish processing (Kongtorp *et al.* 2004b). While details on the survival of PRV is unknown, Reoviruses are likely to be resistant to the range of environmental conditions encountered in fish processing and storage conditions (Wiik-Nielsen *et al*. 2012). Reoviruses remain viable in low-moisture processed food for 24 days at room temperature and for 2 months at 5°C (Pirtle & Beran 1991), after municipal sewage treatment, or when discharged into marine waters (McDaniels *et al.* 1983). Reoviruses remain viable for up to 36 days in landfill (Ware 1980), but they are unlikely to remain viable after passage through the digestive system of scavenging birds (John *et al*. 2001).

Little information exists to determine the infectivity of piscine aquareoviruses (Garseth *et al.* 2012). Most have been isolated from apparently healthy fish, indicating they are likely to be of low pathogenicity (Crane & Carlile 2010). Aquareovirus A has the widest host range and Aquareovirus B has a limited host and geographical range, but current knowledge precludes determination of correlations between aquareovirus species, host and geographical range, and pathogenicity (Mahy & van Regenmortal 2010). It remains unclear whether farmed fish infected with isolates of HSMIV, PRV or TasSAV will develop clinical disease, or whether wild fish may infect farmed populations (Garseth *et al*. 2012).

The likelihood of exposure to PRV and associated aquareoviruses is assessed as non-negligible.

24.2.3. Consequence assessment

PRV and associated aquareoviruses affect both Atlantic salmon (*Salmo* spp.) and Pacific salmon (*Oncorhynchus* spp.), as well as a variety of other marine and freshwater fish (Garseth *et al.* 2012), although pathogenicity appears dependent on a complex of factors (Winton *et al.* 1981; Garseth *et al.* 2012; Carlile *et al.* 2014). The introduction of these aquareoviruses would have direct economic losses for the Chinook salmon (*O. tshawytscha*) farming industry, valued at $63 million in export earnings in 2011 (Aquaculture New Zealand 2014). An outbreak would also affect recreational and tourist trout and salmon fishing for rainbow trout (*O. mykiss*), as well as incur significant social

and environmental costs associated with other domestic fisheries (Fish & Game 2014). While the dollar value of New Zealand's freshwater fisheries is not fully known, the Lake Taupo trout fishery alone is valued at $70 to 80 million (Marsh & Mkwra 2013).

The introduction of aquareovirus may also affect the industries associated with the farming and use of acclimatised grass carp for aquatic weed control (Clayton & Wells 1999; NIWA 2014). Their introduction may also have a social impact upon the existing recreational coarse fisheries for roach and tench (Fish & Game 2014).

New Zealand brill (*Colistium guntheri*) and turbot (*C. nudipinnis*) (Pleuronectidae) have been identified as species of interest to land-based marine aquaculture, where successful development requires continued access to high quality marine water (Hickman & Tait 2001; Mussely & Goodwin 2012; Sim-Smith *et al*. 2014). The introduction of TRV and SMReV into a naïve population may compromise the continued development of this aquaculture operation.

An outbreak would also affect the recreational fishery for turbot and brill (MPI 2014). An outbreak would incur significant social and environmental costs for these endemic fisheries, although the dollar value of these fisheries is not presently known.

The consequences of the introduction of PRV and associated aquareoviruses is assessed as non-negligible.

24.2.4. Risk Estimation

Since the entry, exposure and consequence assessments are non-negligible, the risk estimate is non-negligible. Therefore, PRV and associated aquareoviruses are assessed to be a risk in the commodity and risk management measures may be justified.

24.3. Risk management

Piscine aquareovirus and associated aquareoviruses have been assessed to be a risk in the commodity. The diseases associated with PRV are non-notifiable to the OIE, so there is no specific guidance in the OIE *Aquatic Code* (OIE 2016a) for importing eviscerated fish from infected countries, or specific processing requirements that would ensure the destruction of the virus. It is assumed that compliance with the approved processed states for OIE-listed diseases (Appendix 3) would also eliminate Piscine aquareovirus and associated aquareoviruses from the commodity and be a viable risk management option.

Susceptible species from eleven families of wild and farmed fish (Table 21) may be present in the commodity. Species declaration should substantially reduce the pathogen load of PRV in the commodity and be a viable risk management option. Restriction of the commodity to wild-caught fish (not from aquaculture) would have no effect on pathogen load and is not a viable risk management option.

The distribution of isolates and genogroups within the PRV species complex is poorly defined and no requirements for dedicated monitoring exist. Approval of country/zone freedom through the MPI Country Approval Procedures should substantially reduce the pathogen load of piscine aquareovirus and associated aquareovirus in the commodity. Acceptance of a declaration of country/zone freedom by MPI is a viable risk management option.

Within the commodity, infection is mainly associated with the brain and gill tissues, so removal of the gills should slightly reduce the pathogen load of PRV an associated aquareoviruses. Further processing to remove the head and gills should moderately reduce the pathogen load and be a viable risk management option.

PRV and associated aquareoviruses are unaffected by freezing, so frozen storage is not a viable management option. These viruses are denatured by cooking (to at least 56°C for at least 120 minutes) to ensure inactivity. Heat treatment is a viable management option.

24.3.1. Risk management options

Piscine aquareovirus (PRV) and associated aquareoviruses are reported from fish in families Argentinidae, Carangidae, Centrarchidae, Channidae, Clupeidae, Cyprinidae, Ictaluridae, Osmeridae, Polynemidae, Salmonidae and Scopthalmidae (Table 21). These families are considered likely to be present in the commodity. Other families have not been associated with PRV and associated aquareoviruses. Therefore species declaration indicating the commodity is not originated from any of the above families should substantially reduce the occurrence of PRV and associated aquareoviruses in the commodity.

For commodities originated from families associated with PRV and associated aquareoviruses, one or a combination of the following additional options could also be considered to effectively manage the risk.

Where country/zone freedom from PRV and associated aquareoviruses is accepted by MPI:

Option 1

Acceptance of country/zone freedom by MPI should substantially reduce the occurrence of PRV and associated aquareoviruses, so the commodity may be imported without any further restrictions.

Where country/zone freedom from PRV and associated aquareoviruses is not accepted by MPI or not available:

Option 2

Processing consistent with the recommended treatments for OIE-listed diseases (Appendix 3) should eliminate PRV and associated aquareoviruses. When these provisions are met, the commodity could be imported without any further restrictions.

Where the imported commodity is not processed to a state equivalent to the recommended treatments for OIE-listed diseases (Appendix 3):

Option 3

Heat treatment (by cooking to at least 56°C for at least 120 minutes) should eliminate PRV and associated aquareoviruses. When this provision is met, the commodity could be imported without any further restrictions; or,

Option 4

Further processing (by removal of the head and gills) should moderately reduce the occurrence of PRV and associated aquareoviruses. When this provision is met, the commodity could be imported without any further restrictions.

24.4. References

Ahne W, Kolbl O (1987). Occurrence of reoviruses in European cyprinid fishes (*Tinca tinca* Lin., *Leuciscus cephalus* Lin.). *Journal of Applied Ichthyology* 3(3): 139–141.

Aldrin M, Storvik B, Frigessi A, Vijiugrein H, Jansen PA (2010). A stochastic model for the assessment of the transmission pathways of heart and skeleton muscle inflammation, pancreas disease and infectious salmon anaemia in marine fish farms in Norway. *Preventative Veterinary Medicine* 93(1): 51–61.

Amend DF, McDowell T, Hedrick RP (1984). Characteristics of a previously unidentified virus from channel catfish (*Ameiurus punctatus*). *Canadian Journal of Fisheries and Aquatic Sciences* 41(5): 807–811.

Anonymous (2010). Biosecurity (Notifiable Organisms) Order 2010. NZSR 210/165. New Zealand Government, Wellington, New Zealand, 13 p.

Aquaculture New Zealand (2014). New Zealand's aquaculture industry key facts [Online].Available from: http://www.seafoodnewzealand.org.nz/our-industry/key-facts/ [Accessed 26 September 2014].

Attoui H, Fang Q, Jaafar FM, Cantaloube JF, Biagini P, de Micco P, de Lamballerie X (2002). Common evolutionary origin of aquareoviruses and orthoreoviruses revealed by genome characterisation of golden shiner reovirus, grass carp reovirus, striped bass reovirus and golden idle reovirus (genus *Aquareovirus*, family *Reoviridae*). *Journal of General Virology* 83: 1941–1951.

Beck BH, Peatman E (2015). Mucosal vaccines, pp 297–295 *In:* Soto E, Griffin MJ, Tobar JA (*Eds.*). Mucosal Health in Aquaculture. Academic Press, New York, 408 p.

Biering E, Garseth AH (2012). Heart and skeletal muscle inflammation (HSMI) of farmed Atlantic salmon (*Salmo salar* L.) and the associated piscine reovirus (PRV). ICES Identification Leaflet for Diseases and Parasites of Fish and Shellfish. No. 58, 6 p.

Blindheim S, Nylund A, Watanabe K, Plarre H, Erstad B, Nylund S (2015). A new aquareovirus causing high mortality in farmed Atlantic halibut fry in Norway. *Archives of Virology* 10: 91–102.

Bustos P, Rozas M, Bohle H, Ildenfonso R, Sandoval A, Gaete A, Araya C, Grothusen H, Tapia E, Gallardo A, Rojas M, Lara M, Labra A, Gálvez C (2011). Primer reporte de Piscine Reovirus (PRV) en salmón del atlántico, *Salmo salar*, cultivado en Chile. *Versión Diferente* 15: 65–69.

Carlile G, East IJ, McColl KA, Ellard K, Browning GF, Crane M St J. (2014). The spatial and temporal variation of the distribution and prevalence of Atlantic salmon reovirus (TSRV) infection in Tasmania, Australia. *Preventive Veterinary Medicine* 116: 214–219.

Castric Y (1997). Viral diseases in fish mariculture. *Bulletin of the European Association of Fish Pathologists* 17(6): 220–228.

Chambers M (2001). Growout Techniques for the Pacific Threadfin Polydactylus sexfilis. Aquafarmer Information Sheet No. 145. The Center for Tropical and Subtropical Aquaculture, The Oceanic Institute, Waimanalo, Hawaii, United States of America, 6 p.

Chanet B (2003). Interrelationships of scophthalmid fishes (Pleuronectiformes: Scophthalmidae). *Cymbium* 27(4): 275–286.

Chen Y, Jiang Y (1984). Morphological and physio-chemical characterisation of the hemorrhagic virus of grass carp. *Chinese Science Bulletin* 29: 832–835.

Chen ZY, Gao XC, Zhang QA (2015). Whole genome analysis of a novel fish reovirus (SMReV) discloses aquareovirus genomic relationship with host in saline environments. *Viruses* 7: 4282–4302.

Clayton JS, Wells RDS (1999). Some issues in risk assessment reports on grass carp and silver carp. Conservation Advisory science Notes No. 257. Department of Conservation, Wellington, New Zealand, 14 p.

Cobb S (2008). Import risk analysis: Fish food. Biosecurity New Zealand, Ministry of Agriculture and Forestry, Wellington, New Zealand, 57 p.

Crane MStJ, Smith PH, Williams LM, Hyatt AD, Eaton LM, Gould A, Handlinger J, Kattenbelt J, Gudkovs N (2000). First isolation of an aquatic birnavirus from farmed and wild fish species in Australia. *Diseases of Aquatic Organisms* 43: 1–14.

Crane MStJ, Carlile F (2010). Aquareoviruses. pp 429–437, *In:* Mahy BWH, van Regenmortel MHV (*Eds*.). Desk Encyclopedia of Animal and Bacterial Virology. Academic Press, New York, United States of America, 646 p.

Cusack R, Groman DG, MacKinnon AM, Kibenge FSB, Wadowska S, Brown M (2001). Pathology associated with an aquareovirus in captive juvenile Atlantic halibut *Hippoglossus hippoglossus* L and an experimental treatment strategy for a concurrent bacterial infection. *Diseases of Aquatic Organisms* 44: 7–16.

Di Cicco E, Ferguson HW, Schulze AD, Kaukinen KH, Li S, Vanderstichel R, Wessel O, Rimstad E, Gardner IA, Hammell KL, Miller KM (2017). Heart and skeletal muscle inflammation (HSMI) disease diagnosed on a British Columbia salmon farm through a longitudinal farm study. *PLoS ONE* 12(2): e0171471. Epub. 2017. 31 p. [Online] Available from: http://www.doi.10.1371/journal.pone.0171471 [Accessed 15 March 2018].

Dopazo CP, Bandin I, Rivas C, Ceped C, Barja JL (1996). Antigenic differences among aquareoviruses correlate with previously established genogroups. *Diseases of Aquatic Organisms* 26: 159–162.

Fan Y, Rao S, Zeng L, Ma J, Zhou Y, Xu J, Zhang H (2013). Identification and genomic characterization of a novel fish reovirus, Hubei grass carp disease reovirus, isolated in 2009 in China. *Journal of General Virology* 94(10): 2266–2277 [Online]. Available from: www.doi: 10.1099/vir.0.054767-0. Epub 2013 Jul 12 [Accessed 22 December 2016].

Fauquet CM, Mayo MA, Maniloff J, Desselberger U, Bal LA (2005). Virus Taxonomy: Eighth Report of the International Committee on the Taxonomy of Viruses. Academic Press, London, United Kingdom, 1162 p.

Fish and Game (2014). New Zealand Fish and Game Council [Online] Available from: http://www.fishandgame.org.nz/ [Accessed 24 September 2014].

Garseth ÅH, Fritsvold C, Opheim M, Skjerve E, Biering E (2012). Piscine reovirus (PRV) in wild Atlantic salmon, *Salmo salar* L and seatrout, *Salmo trutta* L. in Norway. *Journal of Fish Diseases* 36: 483–493.

Garver KA, Marty GD, Cockburn SN, Richard J, Hawley LM, Muller A, Thompson RL, Purcell MK, Saksida S (2016). Piscine reovirus, but not Jaundice Syndrome, was transmissible to Chinook salmon, *Oncorhynchus tshawytscha* (Walbaum), sockeye salmon, *Oncorhynchus nerka* (Walbaum), and Atlantic salmon, *Salmo salar* L. *Journal of Fish Diseases* 39(2): 117–128.

Goodwin AE, Merry GE, Attoui H (2010). Detection and prevalence of the nonsyncytial American grass carp reovirus *Aquareovirus G* by quantitative reverse transcriptase polymerase chain reaction. *Journal of Aquatic Animal Health* 22(1): 8–13.

Harrell RM (1997). Striped bass and other *Morone* culture. Elsevier, New York, United States of America, 325 p.

He Y, Jiang Y, Lu L (2013). Serodiagnosis of grass carp reovirus infection in grass carp *Ctenopharyngodon idella* by a novel Western blot technique. *Journal of Virological Methods* 194(1-2): 14–20.

Hetrick FM, Hedrick RP (1993). New viruses described in finfish from 1988–1992. *Annual Review of Fish Diseases* 3: 187–207.

Hickman RW; Tait MJ (2001). Experimental rearing of juvenile New Zealand turbot *Colistium nudipinnis* (Waite 1910): a potential new aquaculture species. *Aquaculture Research* 32(9): 727–737.

Hsu YL, Chen BS, Wu JL (1989). Characteristics of a new reo-like virus isolated from land-locked salmon (*Oncorhynchus masou* Brevort). *Fish Pathology* 24(1): 37–45.

Johansen R, Kongtorp RT, Bornø G, Skjelstad HR, Olsen AB, Flesjå K, Colquhoun D, Ørpetveit I, Hansen H, Garseth ÅH, Hjeltnes B (2008). The health situation in farmed salmonids 2008. *In*: Farmed fish health report 2008. Norwegian Veterinary Institute, Oslo, Norway, 14 p.

John KR, George RM, Richards RM, Richards RH, Frerichs GN (2001). Characteristics of a new reovirus isolated from epizootic ulcerative syndrome infected snakehead fish. *Diseases of Aquatic Organisms* 46: 83–92.

Kahn SA, Beers PT, Findlay VL, Peebles IR, Durham PJ, Wilson DW, Gerrity SE (1999). Import risk analysis on non-viable salmonids and non-salmonid marine finfish. Australian Quarantine and Inspection Service, Department of Agriculture, Fisheries and Forestry, Canberra, Australia, 409 p.

Ke F, He LB, Pei C, Zhang QY (2011). Turbot reovirus (SMReV) genome encoding a FAST protein with a non-AUG start site. *BMC Genomics* 12: 323, 13 p [Online] Available from: http://www.biomedcentral.com/1471-2164/12/323 [Accessed 12 August 2015].

Kibenge FSB, Godoy M (2016). Aquaculture virology. Academic Press, New York, United States of America, 568 p.

Kibenge MJ, Iwamoto T, Wang Y, Morton A, Godoy MG, Kibenge FS (2013). Whole-genome analysis of piscine reovirus (PRV) shows PRV represents a new genus in family Reoviridae and its genome segment S1 sequences group it into two separate sub-genotypes. *Journal of Virology* 10: 230.

King AMQ, Lefkowitz E, Adams M, Carstens EB (2011). Virus taxonomy: Ninth report of the international Committee on the Taxonomy of Viruses. Elsevier Science, New York, United States, 1338 p.

Kongtorp RT, Kjerstad A, Taksdal T, Guttvik A, Falk K (2004a). Heart and skeletal muscle inflammation in Atlantic salmon, *Salmo salar* L.: a new infectious disease. *Journal of Fish Diseases* 27: 351–358.

Kongtorp RT, Taksdal T, Lyngoy A (2004b). Pathology of heart and skeletal muscle infection (HSMI) in farmed Atlantic salmon *Salmo salar*. *Diseases of Aquatic Organisms* 59(3): 217–224.

Kristoffersen AB, Jensen BB, Jansen PA (2013). Risk mapping of heart and skeletal muscle inflammation in salmon farming. *Preventative Veterinary Medicine* 109(1-2): 136–143.

Liang HR, Li YG, Zeng WW, Wang YY, Wang Q, Wu SQ (2014). Pathogenicity and tissue distribution of grass carp reovirus after intraperitoneal administration. *Virology Journal* 11: 178 [Online] Available from: http://www.doi.10.1186/1743-422X-11-178 [Accessed 22 December 2016].

Lupiani B, Dopazo CP, Ledo A, Fouz B, Barja JL (1989). New syndrome of mixed bacterial and viral aetiology in cultured turbot *Scophthalmus maximus. Journal of Aquatic Animal Health* 1: 197–204.

Lupiani B, Hetrick F, Samal SK (1993). Genetic analysis of aquareoviruses using RNA-RNA blot hybridisation. *Virology* 197(1): 475–479.

Lupiani L, Subramanian K, Samal SK (1995). Aquareoviruses. *Annual Review of Fish Diseases* 5: 175–208.

Marsh D, Mkwra L (2013). Review of freshwater non-market value studies. Department of Economics, University of Waikato, Hamilton, New Zealand, 144 p.

Marshall SH, Samal SK, McPhillips TH, Moore AR, Hetrick FM (1990). Isolation of a rotavirus from smelt, *Osmerus mordax* (Mitchell). *Journal of Fish Diseases* 13: 87–91.

Martinez-Rubio L, Morais S, Evensen O, Wadsworth S, Ruohonen K, Vecino JLG, Bell JG, Tocher DR (2012). Functional feeds reduce heart inflammation and pathology in Atlantic salmon (*Salmo salar* L.) following experimental challenge with Atlantic salmon reovirus (ASRV). *PLoS One* 7(12): e40266 [Online] Available from: www.doi: 10.1371/journal.pone.0040266 [Accessed 20 December 2016].

Marty GD, Morrison DB, Bidulka J, Joseph T, Siah A (2015). Piscine reovirus in wild and farmed salmonids in British Columbia, Canada: 1974–2013. *Journal of Fish Diseases* 38(8): 713–728.

McDaniels AE, Cochran KW, Gannon JJ, Williams GW (1983). Rotavirus and reovirus stability in microorganism free distilled and wastewaters. *Water Research* 17(10): 1349–1353.

McDaniel G, Buck C, Khouri H (1996). Animal Viruses and Antisera: Chlamydiae and Rickettsiae, 7th edition. American Type Culture Collection, Rockville, MD, 1996, 256 p.

Meyers TR (1980) Biochemical characterization of an aquareovirus isolated from coho salmon**.** *Journal of Fish Diseases* 3:187–201.

Meyers TR (1983). Serological and histopathological responses of rainbow trout, *Salmo gairdneri* Richardson, to experimental infection with the 13P2 reovirus. *Journal of Fish Diseases* 6(3): 277–292.

Mikkelsen SS, Arnö J, Bruun MS (2014). PMCV and PRV occurrence in wild and farmed fish in Denmark. pp 38–39. *In:* European Union Reference Laboratory for Fish Diseases Report: 18th Annual Workshop of the National Reference Laboratories for Fish Diseases, Copenhagen, Denmark, June 3–4, 2014.[Online]. Available from: http://www.eurl-fish.eu/Activities/annual-meetings [Accessed 27 Feb 2016].

Murphy FA, Fauquet CM, Bishop DHL, Ghabrial AJ, Jarvis A, Martelli GP, Mayo MA, Summers MD (2012). Virus taxonomy: Classification and nomenclature of viruses. Springer, New York, United States of America, 568 p.

Mussely H, Goodwin E (2012). Feasibility of land-based aquaculture in New Zealand. Cawthron Report No. 2094, 20p.

Neukirch M, Haas L, Lehmann H, von Messeling V (1999). Preliminary characterization of a reovirus isolated from golden ide *Leuciscus idus melanotus. Diseases of Aquatic Organisms* 35: 159–164.

NIWA (2014). Grass carp *Ctenopharyngodon idella* (Valenciennes, 1844) [Online]. Available from: https://www.niwa.co.nz/freshwater-and-estuaries/nzffd/NIWA-fish-atlas/fish-species/grass_carp [Accessed 12 February 2015].

OIE (2016). Aquatic Animal Health Code, 19th Edition. World Organization for Animal Health. Paris, France [Online]. Available from: http://www.oie.int/international-standard-setting/aquatic-code/access-online/ [Accessed 15 March 2017].

Page LM, Burr BM (2011). A field guide to freshwater fishes of North America north of Mexico. Boston : Houghton Mifflin Harcourt, Boston, United States of America, 663p.

Palacios G, Lovoll M, Tengs T, Hornig M, Hutchison S, Hui J, Kongtorp RT, Savji N, Bussetti AV, Solovyov A, Kristoffersen AB, Celone C, Street C, Trifonov V, Hirschberg DL, Rabadan R, Egholm M, Rimstad E, Lipkin WI (2010). Heart and skeletal muscle inflammation of farmed salmon is associated with infection with a novel reovirus. *PLoS ONE* 5(7): e11487. doi: 10.1371/journal.pone.0011487 [Online] Available from: http://journals.plos.org/plosone/article?id=10.1371/journal.pone.0011487 [Accessed 22 April 2015].

Paul LJ (2000). New Zealand Fishes. Reed Books, Second edition. Auckland, New Zealand, 253 p.

Pirtle EC, Beran GW (1991). Virus survival in the environment. *OIE Review of Science and Technology* 10(3): 733–748.

Plumb, JA, Bowser RR, Grizzle JM, Mitchell AJ (1979). Fish viruses: A double- stranded RNA icosahedral virus from a North American cyprinid. *Journal of the Fisheries Research Board of Canada* 36: 1390-1394.

Rangel AAC, Rockemann DD, Hetrick FM, Samal SK (1999). Identification of grass carp haemorrhage virus as a new genogroup of aquareovirus. *Journal of General Virology* 80; 2399–2402.

Rivas C, Bandin I, Cepeda C, Dopazo CP (1994). Efficacy of chemical disinfectants against turbot aquareovirus. *Applied and Environmental Microbiology* 60(6): 2168–2169.

Rivas C, Bandin I, Noya M, Cepeda C, Barja JL, Dopazo CP (1996a). *In vitro* and *in vivo* replication of turbot aquareovirus (TRV) in turbot tissues. *Diseases of Aquatic Organisms* 25: 217–223.

Rivas C, Bandin I, Noya M, Dopazo CP, Cepeda C, Barja JL (1996b). Effect of the turbot aquareovirus on fish macrophages using an *in vitro* model. *Diseases of Aquatic Organisms* 25: 209–216.

Roberts RG (2012). Fish Pathology. John Wiley, London, United Kingdom, 581 p.

Schachner O, Soliman H, Straif M, Schilcher F, El-Matbouli M (2014). Isolation and characterisation of a novel reovirus from white bream *Blicca bjoerkna*. *Diseases of Aquatic Organisms* 112: 131–138.

Sim-Smith C, Faire S, Lees A (2014). Managing biosecurity risk for business benefit. Report prepared for the Ministry for Primary Industries by Coast and Catchment Ltd, Auckland, New Zealand, 214 p.

Subramanian K, Hetrick FM, Samal SK (1997). Identification of a new genogroup of aquareovirus by RNA-RNA hybridization. *Journal of General Virology* 78(6): 1385–1388.

Tubbs L, Lee P, Diggles B, Jones JB, Shepherd M, Sim-Smith C (2007). A review of aquatic diseases of significance to New Zealand. Final research report for project ZBS 2005-17. National Institute of Water and Atmospheric Research, Wellington, New Zealand, 452 p.

Varner PW, Lewis DH (1991). Characterization of a virus associated with head and lateral line erosion syndrome in marine angelfish. *Journal of Aquatic Animal Health* 3(3): 198–205.

Wang T, Li J, Lu L (2013). Quantitative *in vivo* and *in vitro* characterization of co-infection by two genetically distant grass carp reoviruses. *Journal of General Virology* 94(6): 1301–1309 [Online] Available from: doi: 10.1099/vir.0.049965-0. Epub 2013 Feb 20. [Accessed 22 December 2016].

Ware SA (1980). A survey of pathogen survival during municipal solid waste and manual treatment processes. EPA - 600/8-80-034. United States Environmental Protection Agency, Cincinnati, United States of America, 116 p.

Watanabe K, Karlsen M, Devold M, Isdal E, Litlabo A, Nylund A (2006). Virus-like particles associated with heart and skeletal muscle inflammation (HSMI). *Diseases of Aquatic Organisms* 70: 183–192.

Wellings FM, Lewis AL, Mountain CW (1976). Demonstration of solids-associated virus in wastewater and sludge. *Applied and Environmental Biology* 31: 354–358.

Wiik-Nielsen CR, Lovoll M, Sandlund N, Faller R, Wiik-Nielsen J, Jensen BB (2012). First detection of piscine reovirus (PRV) in marine fish species. *Diseases of Aquatic Organisms* 97: 255–258.

Winton JR, Lannan CN, Fryer JL, Kimura T (1981). Isolation of a new reovirus from chum salmon in Japan. *Fish Pathology* 15: 155–162.

Winton JR, Arakawa CK, Lannan CN, Fryer JL (1989). Isolation of a reovirus from coho salmon (*Oncorhynchus kitsuch*) in Oreon, USA. Picornaviruses and retroviruses of fish., p 257 *In:* Ahne W, Kurstak E (*Eds*.). Proceedings of the International Symposium on Viruses of Lower Animals. University of Munich, Federal Republic of Germany, August 22-25, 1988 Springer, Berlin, Germany.

Xu J, Zeng L, Luo X, Wang Y, Fan Y, Gong S (2013). Reovirus infection emerged in cultured channel catfish, *Ameiurus punctatus*, in China. *Aquaculture* 372-375: 39–44.

Ye X, Tian YY, Deng GC, Chi YY, Jiang XY (2012). Complete genomic sequence of a reovirus isolated from grass carp in China. *Virus Research* 163(1): 275–283 [Online] Available from: www.doi: 10.1016/j.virusres.2011.10.014. Epub 2011 Oct 21 [Accessed 22 December 2016].

Zainathan S (2012). Detection of aquareovirus in farmed Tasmanian Atlantic salmon (*Salmo salar*). Ph.D dissertation, University of Tasmania, Australia, 316 p.

Zhang QY, Gui JF (2015). Virus genomes and virus-host interactions in aquaculture animals. Special Topic: Fish biology and biotechnology: A Review. *Science China Life Sciences* 58(2): 156–169.

Zu J, Zeng L, Luo X, Wang Y, Fan Y, Gong S (2014). Reovirus infection emerged in cultured channel catfish, *Ameiurus punctatus*, in China. *Aquaculture* 372: 39–44.

25. Red sea bream iridovirus, infectious spleen and kidney necrosis virus and other megalocytiviruses

25.1. Hazard identification

25.1.1. Aetiological agent

The Genus *Megalocytivirus* is classified within the Family Iridoviridae and consists of four species (OIE 2016a):

- red sea bream iridovirus (RSIV) (two genotypes);
- infectious spleen and kidney necrosis virus (ISKNV) (two genotypes);
- turbot reddish body iridovirus (TRBIV); and
- three spine stickleback iridovirus (TSIV).

Currently, the OIE identify the agents of red sea bream iridoviral disease (RSIVD) as being RSIV and ISKNV, noting that it is unclear whether TRBIV should be included within RSIV (OIE 2016a). Other related iridoviruses, including epizootic haematopoietic necrosis virus (EHNV), European catfish virus (ECV) and grouper iridovirus (GIV)/Singapore grouper iridovirus (SGIV), are considered distinct viruses, as they are not pathogenic to red sea bream (OIE 2016a).

More recent genomic studies suggest *Megalocytivirus* represents a species complex comprising of at least three genotypes, together with isolates and clades that infect a wide range of marine fish species (Sriwanayos *et al.* 2013; Nolan *et al.* 2015). This classification also includes the associated viruses: dwarf gourami iridovirus (DGIV), grass carp iridovirus and white sturgeon iridovirus (B. Jones, *pers. comm*. 2015), as well as Taiwan grouper iridovirus and rock bream iridovirus (Kurita & Nakajima 2012) as clades of RSIV. These isolates are regarded as causing RSIVD or ISKNV in this report. As DGIV is only reported from the ornamental dwarf gourami (*Colisa lalia*) (Rimmer *et al.* 2015) which is unlikely to be present in the commodity, it is not considered further.

RSIVD was first reported in farmed sea bream (*Pagrus major*) in Japan (Inouye *et al*. 1992), and has since been associated with mass mortality epizootics of more than 30 species in Japan, South Korea, China, and Southeast Asia (Inouye *et al.* 1992; Jung & Oh 2000; Kawakami and Nakajima 2002; Wang *et al.* 2003). The disease is reported from Australia, Taiwan, China, Hong Kong, Korea, Malaysia, Philippines, Singapore and Thailand in wild and farmed fish (Song *et al.* 2008; OIE 2016b). An additional clade of megalocytivirus described as scale drop disease virus (SDDV) was identified from barramundi (*Lates calcarifer*) in Indonesian and Malaysian marine aquaculture (De Groof *et al.* 2015). It is not considered further.

25.1.2. OIE status

Red sea bream iridovirus (RSIV) and infectious spleen and kidney necrosis virus (ISKNV) are both recognised as agents of red sea bream iridoviral disease (RSIVD), which is listed by the OIE (OIE 2016a).

25.1.3. New Zealand status

RSIV and ISKNV are exotic organisms that has never occurred in New Zealand (Tubbs *et al*. 2007) and they have not been found in subsequent monitoring (B. Jones, *pers. comm*. 2015). Infection with RSIVD is a notifiable disease in New Zealand (Anon. 2016).

25.1.4. Epidemiology

Megalocytivirus causes economically significant disease in a large number of marine and freshwater fish reported from Australia, Asia and Southeast Asia (Song *et al*. 2008; Sano *et al*. 2011). The fish hosts and families infected with RSIV and ISKNV are listed in Table 22 (Sano et al. 2011; OIE 2016a, 2016b).

RSIV is highly pathogenic, causing 50 to 90% mortality in juvenile red sea bream and groupers in Japanese sea cage culture, although adult fish may also be affected (Sano *et al*. 2011; OIE 2016a). Mortality appears temperature-related, as experimentally infected rock bream (*Oplegnathus fasciatus*) kept for 30 days at 13°C incurred no mortality, but mortality of 100% mortality occurred over 17 days following an increase in water temperature to 25°C (Jun *et al*. 2009).

Table 22. Families and Species of Fish Susceptible to Red Sea Bream Iridovirus (RSIV) and Infectious Spleen and Kidney Necrosis Virus (ISKNV)

Red Sea Bream Iridovirus (RSIV)	
Family	**Susceptible Species**
Carangidae	Greater amberjack (*Seriola dumerili*), Japanese amberjack (*Seriola quinqueradiata*), Japanese jack mackerel (*Trachurus japonicus*), Sampson fish (*Carynx hippos*), snub-nosed pompano (*Trachinotus blochii*), striped jack (*Pseudocaranx dentex*), yellowtail amberjack (*Seriola lalandi*) and hybrids
Centrarchidae	Largemouth bass (*Micropterus salmoides*)
Haemulidae	Chicken grunt (*Parapristipoma trilineatum*), crescent sweetlips (*Plectorhinchus cinctus*)
Kyphosidae	Largescale blackfish (*Girella punctata*)
Lateolabracidae	Japanese sea perch (*Lateolabrax japonicus*, *Lateolabrax* sp.)
Latidae	barramundi or sea bass (*Lates calcarifer*)
Lethrinidae	Chinese emperor (*Lethrinus haemopterus*), Spangled emperor (*Lethrinus nebulosus*)
Moronidae	Striped sea bass (*Morone saxitalis*), white sea bass (*Morone chrysops*) and hybrids
Oplegnathidae	Japanese parrotfish (*Oplegnathus fasciatus*), rock porgy (*Oplegnathus punctatus*)
Paralichthyidae	Bastard halibut (*Paralichthys olivaceus*)
Percichthyidae	Murray cod (*Maccullochella peelii*)
Pleuronectidae	Spotted halibut (*Verasper variegatus*)
Rachycentridae	Cobia (*Rachycentron canadum*)
Sciaenidae	Large yellow croaker (*Larimichthys (Pseudosciaenea) crocea*)
Scombridae	Northern bluefin tuna (*Thunnus thynnus*), chub mackerel (*Scomber japonicus*), Japanese Spanish mackerel (*Scomberomorus niphonius*)
Sebastidae	Rockfish (*Sebastes schlegelii*)
Serranidae	Hong Kong grouper (*Epinephelus akaara*), convict grouper (*Epinephelus septemfasciatus*), Malabar grouper (*Epinephelus malabaricus*), long tooth grouper (*Epinephelus bruneus*), orange-spotted grouper (*Epinephelus coioides*), yellow grouper (*Epinephelus awoara*), greasy grouper (*Epinephelus tauvina*), brown-marbled grouper (*Epinephelus fuscoguttatus*), giant grouper (*Epinephelus lanceolatus*),
Sparidae	Crimson sea bream (*Dentex tumifrons* (=*Evynnis japonica*)), black porgy (*Acanthopagrus schlegelii*), largescale blackfish (*Girella punctata*), red sea bream (*Pagrus major*), silver seabream (*Pagrus auratus*), yellowfin sea bream (*Acanthopagrus latus*)
Terapontidae	Japanese pufferfish (*Takifugu rubripes*),
Infectious spleen and kidney necrosis virus (ISKNV)	
Family	**Susceptible secies**
Cyprinidae	Grass carp (*Ctenopharyngodon idella*)
Mugilidae	Flathead mullet (*Mugil cephalus*)
Percichthyidae	Chinese perch (*Siniperca chuatsi*)

Sciaenidae	Red drum (*Sciaenops ocellatus*)
Serranidae	Groupers (*Epinephelus* spp.)

Megalocytivirus infection is common in Asian aquaculture, where occurrence varies from 13% for sea bream, to 80% for rock bream (Sano *et al.* 2011). RSIV is also reported from the United States and Australia, primarily associated with the release of imported ornamental fish (Kurita & Nakajima 2012; Nolan *et al.* 2015).

Disease transmission is horizontal, by direct contact from viral particles shed in the water, or by feeding on infected fish (Sano *et al.* 2011; OIE 2016a), but vertical transmission does not occur (Subramaniam *et al.* 2012). Megalocytiviral infection is highly contagious (Sano *et al*. 2011).

Infected fish commonly show no external signs (Choi *et al*. 2006; Noga 2010; OIE 2016a). RSIV infection is focused on the heart, stomach, intestine, skeletal musculature, eyes, connective tissues and gills, usually associated with tissue necrosis and petechiae (Whittington *et al.* 2010; Sano *et al.* 2011; OIE 2016a).

Megalocytivirus is stable in water for at least 7 days at 15°C (Ito *et al.* 2013) and resistant to freezing, remaining viable in fish tissue frozen at -80°C (OIE 2016b). It remains viable in frozen whole baitfish tissues commonly used as fish feed in Asian aquaculture (Laijimin *et al.* 2015).

RSIV replicates in a temperature range of 20 to 25°C (Kurita and Nakajima 2012), but is resistant to heat, requiring treatment at 56°C for up to 30 minutes for inactivation (OIE 2016a). It has not been reported from homeothermic vertebrates and is unlikely to be transferred by piscivorous birds (Whittington *et al*. 2010). Megalocytivirus is inactivated by ether and chloroform treatments, as well as by formalin (0.1%) (OIE 2016b).

A formalin-inactivated vaccine for RSIV is available in Japan for red sea bream (*Pagrus major*), striped jack (*Pseudocaranx dentex*), Malabar grouper (*Epinephelus malabaricus*) and orange-spotted grouper (*Epinephelus coioides*) and a vaccine has also been developed in Taiwan for the related Taiwan grouper iridoviral disease (Nakajima *et al.* 1999; Kurita & Nakajima 2012).

Susceptible species in New Zealand include snapper (*Pagrus auratus*), silver trevally (*Pseudocaranx dentex*), trevally (*Caranx georgianus*), jack mackerel (*Trachurus* spp.) and many other marine species (Tubbs *et al*. 2007) as well as freshwater galaxiids (Diggles 2003).

25.2. Risk assessment

25.2.1. Entry assessment

Fish from a wide range of host families may be infected with RSIV and associated isolates (Table 23), typically with no external clinical signs of infection (Noga 2010). These megalocytiviruses are resistant to the normal conditions likely to be encountered in fish storage and processing (OIE 2016a) and may be present in the commodity. While most infected tissue should be removed on evisceration, RSIV and other isolates are likely to remain in the connective tissues, eyes and gill tissues (OIE 2016b), and at low titre in muscle tissue (Johnston 2008). Megalocytiviruses survive in sea water for at least 7 days (Jun *et al*. 2009), are resistant to drying and likely to remain viable even after extended frozen storage (OIE 2016b). The likelihood of entry is assessed as non-negligible.

25.2.2. Exposure assessment

To establish infection in New Zealand, infected organic material would have to become available for consumption by a susceptible fish host, in sufficient quantity and duration (Kahn *et al.* 1999). RSIV remains viable in fish bait used as feed in aquaculture (Laijimin *et al.* 2015) and is likely to remain viable in wash water and the trimmings, head and gill tissues of offal discarded in commercial fish processing for at least 7 days (Ito *et al.* 2013; OIE 2016b). It is resistant to freezing and drying (Sano *et al*. 2011) and may remain viable when dumped in landfill, but is unlikely to be transferred back to the aquatic environment by agents such as piscivorous birds (Whittington *et al*. 2010; Sano *et al*. 2011). The likelihood of exposure to RSIV and associated megalocytiviruses is assessed as non-negligible.

25.2.3. Consequence assessment

The establishment of RSIV and ISKNV and associated isolates may have an economic effect upon several commercial inshore fisheries, including snapper, with exports valued at $62 million, and jack mackerel, with exports of $57 million in 2009 (Statistics New Zealand 2014). The reduction in these key inshore species would incur significant social effects (MPI 2014). Megalocytiviruses may infect freshwater species including galaxiids. These megalocytiviruses may also have an impact upon the aquaculture of yellowtail kingfish (*Seriola lalandi*) and grouper (*Polyprion oxygeneios*), as well as several other potential aquaculture species (Diggles 2003; Kahn *et al*. 2012; Symonds *et al.* 2014). The consequence of the establishment of the associated grouper iridovirus in New Zealand through the live ornamental fish pathway was considered high to catastrophic (Hine & Diggles 2005). The consequence of exposure to RSIV and associated megalocytiviruses is assessed as non-negligible.

25.2.4. Risk estimation

Since the entry, exposure and consequence assessments for RSIV and ISKNV isolates are non-negligible, the risk estimate is non-negligible. Under the procedures followed in this risk assessment, they are assessed to be a risk in the commodity and risk management measures may be developed.

25.3. Risk management

Red seabream iridovirus and infectious spleen and kidney necrosis virus have been assessed to be a risk in the commodity.

Infection with red seabream iridovirus (RSIV) and infectious spleen and kidney necrosis virus (ISKNV) are OIE-listed diseases (OIE 2016a). Article 10.8.2 of the *Aquatic Code* (OIE 2016a) lists the species recognised that need risk management measures to prevent the international spread of these megalocytiviruses:

> *The recommendations in this chapter apply to: red sea bream (Pagrus major), yellowtail (Seriola quinqueradiata), amberjack (Seriola dumerili), sea bass (Lateolabrax sp. and Lates calcarifer), albacore (Thunnus thynnus), Japanese parrotfish (Oplegnathus fasciatus), striped jack (Caranx delicatissimus), mandarin fish (Siniperca chuatsi), red drum (Sciaenops ocellatus), mullet (Mugil cephalus) and groupers (Epinephelus spp.). These recommendations also apply to any other susceptible species referred to in the Aquatic Manual when traded internationally.*

Species declaration should substantially reduce the occurrence of RSIV and ISKNV from the commodity and be a viable risk management measure.

Article 10.8.3 of the *Aquatic Code* recommends:

For the importation or transit of aquatic animals and aquatic animal products for any purpose from a country, zone or compartment not declared free from red sea bream iridovirus:

1. *Competent Authorities should not require any conditions related to RSIVD, regardless of the RSIVD status of the exporting country, zone or compartment, when authorising the importation or transit of the following aquatic animals and aquatic animal products from the species referred to in Article 10.8.2 which are intended for any purpose and which comply with Article 5.4.1:*
    a. *heat sterilised hermetically sealed fish products (i.e. a heat treatment at 121ºC for at least 3.6 minutes or any time/temperature equivalent);*
    b. *pasteurised fish products that have been subjected to heat treatment at 90ºC for at least ten minutes (or any time/temperature equivalent which has been demonstrated to inactivate RSIV);*
    c. *mechanically dried eviscerated fish (i.e. a heat treatment at 100ºC for at least 30 minutes (or any time/temperature equivalent which has been demonstrated to inactivate RSIV).*

2. *When authorising the importation or transit of aquatic animals and aquatic animal products of a species referred to in Article 10.8.2, other than those referred to in point 1 of Article 10.8.3, Competent Authorities should require the conditions prescribed in Articles 10.8.7 to 10.8.12 relevant to the RSIVD status of the exporting country, zone or compartment.*

3. *When considering the importation or transit of aquatic animals and aquatic animal products of a species not covered in Article 10.8.2 but which could reasonably be expected to pose a risk of spread of RSIVD, the Competent Authority should conduct a risk analysis in accordance with the recommendations in Chapter 2.1. The Competent Authority of the exporting country should be informed of the outcome of this assessment.*

Article 10.8.11 of the OIE *Aquatic Code* recommends:

For the importation of aquatic animals and aquatic animal products for retail trade for human consumption from a country, zone or compartment not declared free from RSIV:

1. *Competent Authorities should not require any RSIVD related conditions, regardless of the RSIVD status of the exporting country, zone or compartment, when authorising the importation or transit of fish fillets or steaks (chilled or frozen) which have been prepared and packaged for retail trade and which comply with Article 5.4.2.*

    Certain assumptions have been made in assessing the safety of the aquatic animal products listed above. Member Countries should refer to these assumptions at Article 5.4.2 and consider whether the assumptions apply to their conditions.

For these commodities Member Countries may wish to consider introducing internal measures to address the risks associated with the commodity being used for any purpose other than for human consumption.

2. *When importing aquatic animals or aquatic animal products, other than those referred to in point 1 above, of species referred to in Article 10.8.2 from a country, zone or compartment not declared free from RSIVD, the Competent Authority of the importing country should assess the risk and apply appropriate risk mitigation measures.*

Processing of the commodity in accordance with Articles 10.8.3 and 10.8.11 should eliminate RSIV and ISKNV from the commodity and be a viable risk management option.

RSIVD and ISKNV occur in wild and farmed fish and are considered emerging threats to aquaculture in Australia, Asia and Southeast Asia (OIE 2016a), while alternative infection pathways also exist through imported ornamental fish (Nolan *et al.* 2015). Where country/zone freedom is approved through the MPI Country Approval Procedures, this option is likely to substantially reduce the pathogen load of RSIV and ISKNV in the commodity. Approval by MPI of a declaration of country/zone freedom is a viable risk management option.

Restriction of the commodity to wild-caught fish (not from aquaculture) would have little, or no effect on pathogen load and is not a viable risk management option.

Red seabream iridovirus and ISKNV are mainly associated with eye and gill tissues, so removal of the gills should slightly reduce the pathogen load of RSIV/ISKNV in the commodity. Removal of the head and gills should moderately reduce the pathogen load of RSIV/ISKNV and be an effective risk management option.

Megalocytiviruses are resistant to freezing, so RSIV/ISKNV are not affected by frozen storage. Cooking at high temperatures (such as 100°C for 30 minutes) should eliminate RSIV/ISKNV from the commodity and be a viable risk management option.

25.3.1. Risk management options

The viruses RSIV and ISKNV are reported from fish in families Carangidae, Centrarchidae, Cyprinidae, Haemulidae, Kyphosidae, Lateolabracidae, Latidae, Lethrinidae, Moronidae, Mugilidae, Oplegnathidae, Paralichthyidae, Percichthyidae, Pleuronectidae, Rachycentridae, Sciaenidae, Scombridae, Sebastidae, Serranidae, Sparidae and Terapontidae (Table 22). These families are considered likely to be present in the commodity. Other families have not been associated with RSIV and ISKNV. Therefore species declaration indicating the commodity is not originated from any of the above families should substantially reduce the occurrence of RSIV and ISKNV in the commodity.

For the commodities originated from families associated with RSIV and ISKNV, one or a combination of the following additional options could also be considered to effectively manage the risk.

Where country/zone freedom from RSIV and ISKNV is accepted by MPI:

Option 1

Acceptance of country/zone freedom should substantially reduce the occurrence of RSIV and ISKNV, so the commodity may be imported without any further restrictions.

Where country/zone freedom from RSIV and ISKNV is not accepted by MPI or not available:

Option 2

Processing consistent with the conditions of Article 10.8.3 or 10.8.11 of the OIE Aquatic *Code* (OIE 2016a) should eliminate RSIV and ISKNV. Where these provisions are met, the commodity may be imported without any further restrictions.

Where the imported commodity is not processed to a state equivalent to the conditions of Article 10.8.3 or 10.8.11 of the OIE Aquatic Code (OIE 2016a), further processing is necessary:

Option 3

Heat treatment (by cooking to at least 56°C for at least 30 minutes) should eliminate RSIV and ISKNV from the commodity. Where this provision is met, the commodity may be imported without any further restrictions; or,

Option 4

Further processing (by removal of the head and gills) should moderately reduce the occurrence of RSIV and ISKNV. Where this provision is met, the commodity may be imported without any further restrictions.

25.4. References

Anonymous (2016). Biosecurity (Notifiable Organisms) Order 2016. NZLR 2016/73. New Zealand Government, Wellington, New Zealand, 13 p.

Choi SK, Kwon SR, Nam YK, Kim SK, Kim HK (2006). Organ distribution of red sea bream iridovirus (RSIV) DNA in asymptomatic yearling and fingerling rock bream (*Oplegnathus fasciatus*) and effects of water temperature on transition of RSIV into acute phase. *Aquaculture* 256(1–4): 23–26.

De Groof A, Guelen L, Deijs M, van de Wal Y, Miyata M, Ng KS, Grinsven L, Simmelink B, Biermann Y, Grisez L, van Lent J, de Ronde A, Chang SF, Schrier C, van de Hoek L (2015). A novel virus causes scale drop disease in *Lates calcarifer. PLoS Pathogens* [Online] Available from: http://dx.doi.org/10.1371/journal.ppat.1005074 [Accessed 20 December 2016].

Diggles BK (2003). Import risk assessment: Juvenile yellowtail kingfish (*Seriola lalandi*) from Spencer Gulf Aquaculture, South Australia. NIWA client report WLG 2002/03. National Institute of Water and Atmospheric Research Ltd, Auckland, New Zealand, 54 p.

Gibson-Kueh S, Chee D, Chen J, Wang YH, Tay S, Leong LN, Ng ML, Jones JB, Nicholls PK, Ferguson HW (2012). The pathology of scale drop syndrome in Asian seabass, *Lates calcarifer* Bloch, a first description. *Journal of Fish Diseases* 35: 19–27.

Hine PM, Diggles BK (2005). Import risk analysis: Ornamental fish. Biosecurity New Zealand, Ministry of Agriculture and Forestry, Wellington, New Zealand, 262 p.

Inouye K, Yamano K, Maeno Y, Nakajima K, Matsuoka M, Wada Y, Sorimachi M (1992). Iridovirus infection of cultured red sea bream, *Pagrus major*. *Fish Pathology* 27: 19–27.

Ito T, Yoshiura Y, Kamaishi T, Nakajima K (2013). Dynamics of virus titre and quantity of red sea bream iridovirus DNA using natural and autoclaved artificial sea water under different water temperatures. *Bulletin of the European Association of Fish Pathologists* 33(4): 118–125.

Johnston C (2008). *Import risk analysis:* Frozen, skinless and boneless fillet meat of *Oreochromis* spp. from China and Brazil for human consumption. MAF Biosecurity New Zealand, Ministry of Agriculture and Forestry, Wellington, New Zealand, 96 p.

Jun LJ, Jeong JB, Kim JH, Nam JH, Shin KW, Kim JK, Kang JC, Jeong HD (2009). Influence of temperature shifts on the onset and development of red sea bream iridoviral disease in rock bream *Oplegnathus fasciatus. Diseases of Aquatic Organisms* 84: 201–208.

Jung SJ, Oh MJ (2000). Iridovirus-like infection associated with high mortalities of striped beakperch, *Oplegnathus fasciatus* (Temminck et Schlegel) in southern coastal areas of the Korean peninsula. Journal of Fish Diseases 23: 223–226.

Kahn SA, Beers PT, Findlay VL, Peebles IR, Durham PJ, Wilson DW, Gerrity SE (1999). Import risk analysis on non-viable salmonids and non-salmonid marine finfish. Australian Quarantine and Inspection Service, Department of Agriculture, Fisheries and Forestry, Canberra, Australia, 409 p.

Kawakami H, Nakajima K (2002). Cultured fish species affected by red sea bream iridoviral disease from 1996 to 2000. *Fish Pathology* 37: 45–47.

Kurita J, Nakajima K (2012). Megalocytiviruses. *Viruses* 4: 521–538.

Laijimin S, Razak AA, Denil DJ, Ransagar J, Wahid MEA, Sade A (2015). First detection of megalocytivirus (Iridoviridae) in trash fish used for aquaculture feed in Sabah, Malaysia. *International Journal of Aquatic Science* 6(1): 54–66.

MPI (2014). Fisheries Assessment Plenary, May 2014: Stock assessments and stock status. Compiled by the Fisheries Science Group, Ministry for Primary Industries, Wellington, New Zealand, 1381 p.

Nakajima K, Maeno Y, Honda A, Yokoyama K, Tooriyama T, Manabe S (1999). Effectiveness of a vaccine against red sea bream iridoviral disease in a field trial test. *Diseases of Aquatic Organisms* 36(1): 73–75.

Noga EJ (2010). Fish disease: diagnosis and treatment. John Wiley, New York, United States of America, 378 p.

Nolan D, Stephens F, Crockford M, Jones JB, Snow M (2015). Detection and characterization of viruses of the genus *Megalocytivirus* in ornamental fish imported into an Australian border quarantine premises: an emerging risk to national biosecurity. *Journal of Fish Diseases* 38(2): 187–195.

OIE (2016a). Aquatic Animal Health Code, 19th Edition. World Organization for Animal Health. Paris, France [Online] Available from: http://www.oie.int/international-standard-setting/aquatic-code/access-online/ [Accessed 15 March 2017].

OIE (2016b). Manual of diagnostic tests for aquatic animals. World Organization for Animal Health. Paris, France [Online] Available from: http://www.oie.int/international-standard-setting/aquatic-manual/access-online/ [Accessed 17 March 2017].

Rimmer AE, Becker JA, Tweedie A, Lintermans M, Landos M, Stephens F, Whittington RJ (2015). Detection of dwarf gourami iridovirus (Infectious spleen and kidney necrosis virus) in populations of ornamental fish prior to and after importation into Australia, with the first evidence of infection in domestically farmed Platy (*Xiphophorus maculatus*). *Preventative Veterinary Medicine* 122(1-2): 181–194 Epub 2015 Sep 21 [Online] Available from: http://www.doi: 10.1016/j.prevetmed.2015.09.008 [Accessed 20 Februay 2018].

Sano M, Nakai T, Fijan N (2011) Viral diseases and agents of warm water fish, pp 168–244. *In:* Woo PTK, Bruno DW (*Eds.*). *Fish Diseases and Disorders: Volume 3 - Viral, Bacterial and Fungal Infections*, 2nd edition. CABI Publishers, Wallingford, United Kingdom, 930 p.

Sriwanayos P, Francis-Floyd R, Stidworthy MF, Petty BD, Kelly K, Waltzek TB (2013). Megalocytivirus infection in orbiculate batfish *Platax orbicularis*. *Diseases of Aquatic Organisms* 105:1–8.

Statistics New Zealand (2014). Fish monetary stock account: snapper (*Pagrus auratus*) [Online].Available from: http://www.stats.govt.nz/browse_for_stats/environment/environmental-economic-accounts/fish-monetary-stock-account-1996-2009/results.aspx#snapper [Accessed 10 September 2014].

Subramaniam K, Shariff M, Omar AR, Beho MD (2012). Megalocytivirus in fish. *Reviews in Aquaculture* 4(4): 221–223.

Symonds JE, Walker SP, Pether S, Gublin Y, McQueen D, King A, Irvine GW, Setiawan AN, Forsythe JA, Bruce M (2014). Developing yellowtail kingfish (*Seriola lalandi*) and hapuku (*Polyprion oxygeneios*) for New Zealand aquaculture. *New Zealand Journal of Marine and Freshwater Research* 48(3): 371–384.

Tubbs L, Lee P, Diggles B, Jones JB, Sheppard M, Sim-Smith C (2007). A review of aquatic diseases of significance to New Zealand. Final research report for project ZBS 2005-17. National Institute of Water and Atmospheric Research, Wellington, New Zealand, 452 p.

Wang CS, Shih HH, Ku CC, Chen SN (2003). Studies on epizootic iridovirus infection among red sea bream, *Pagrus major* (Temminck & Schlegel), cultured in Taiwan. *Journal of Fish Diseases* 26(3): 127–133.

Whittington RJ, Becker JA, Dennis MM (2010). Iridovirus infections in finfish – critical review with emphasis on ranaviruses. *Journal of Fish Diseases* 33: 95–122.

26. Salmon alphavirus including salmon pancreatic disease virus and associated togaviruses

26.1. Hazard identification

26.1.1. Aetiological agent

Salmon alphavirus (SAV) is an enveloped, spherical, single-stranded RNA virus classified within the Genus *Alphavirus* in the Family Togaviridae. Three sub-types are recognised, based on genomic analysis (McLoughlin & Graham 2007). Salmonid pancreatic disease virus (PD) was originally identified as a syndrome affecting Scottish salmon (Munro *et al.* 1984) and its viral aetiology was described by Nelson *et al*. (1995). Sleepy virus disease (SVD) was previously described as sleeping disease (SD) of rainbow trout from freshwater farms in the United Kingdom and France (Castric *et al*. 1997). Norwegian salmonid alphavirus (NSAV) was identified from Norwegian salmonids in sea cage culture (Christie *et al*. 1998).

Genomic analysis has identified six genomovars of salmon alphavirus, based on nucleic acid sequences (Graham *et al*. 2011), that share a common epidemiology (McLoughlin & Graham 2007), but vary in pathogenicity (OIE 2016b). Salmon lice (*Lepeophtheirus salmonis*) has been identified as a vector (Petterson *et al*. 2009), although other marine vectors may also transfer this pathogen (Crane & Hyatt 2011).

26.1.2. OIE status

Infection with any sub-type of salmonid alphavirus is listed by the OIE as a notifiable disease (OIE 2016a).

26.1.3. New Zealand status

SAV is considered exotic to New Zealand (Diggles 2011).

26.1.4. Epidemiology

SAV is the only alphavirus infecting fish (Hodneland 2006). Suseptible hosts (Table 23) include Atlantic salmon (*Salmo salar*), brown trout (*Salmo trutta*) and rainbow trout (*Oncorhynchus mykiss*) in marine and fresh waters (Stone *et al*. 1997; McLoughlin & Graham 2007; Graham & McLoughlin 2011). Pancreatic disease is an economically significant salmonid disease (McVicar & Munro 1989; Karlsen *et al.* 2006) reported from Europe (Croatia, France, Germany, Ireland, Italy, Norway, Poland, Spain, Switzerland and the United Kingdom) (OIE 2016a).

Table 23. Families and Species of Fish Susceptible to Salmon Alphavirus (SAV) Including Salmon Pancreatic Disease and Associated Togaviruses

Host Family	Host Species
Gadidae	Pollock (*Pollachius virens*)
Labridae	Ballan wrasse (*Labrus bergylta*)
Pleuronectidae	Common dab (*Limanda limanda*), American plaice (*Hippoglossoides platessoides*), plaice (*Pleuronectes platessa*)
Salmonidae	Atlantic salmon (*Salmo salar*), brown trout (*Salmo trutta*) and rainbow trout (*Oncorhynchus mykiss*)
Scophthalmidae	Megrim (*Lepidorhombus whiffagonis*)

SAV occurs at low levels in wild populations of common dab (*Limanda limanda*), American plaice (*Hippoglossoides platessoides*), megrim (*Lepidorhombus whiffagonis*), plaice (*Pleuronectes platessa*) and pollock/saithe (*Pollachius virens*) in European waters (Graham *et al.* 2006; Snow *et al*. 2010; McCleary *et al.* 2014; OIE 2016b). It has also been detected in cleaner fish used in salmonid aquaculture including the Ballan wrasse (*Labrus bergylta*) (Taksdal *et al.* 2014; Jansen *et al.* 2017). The significance of these hosts as a possible reservoir of infection for salmonids is currently unknown (McCleary *et al*. 2014; Jones *et al.* 2015).

Clinical signs consistent with pancreatic disease have also been reported from Canadian and North American salmon, but no virus was isolated (Kent & Elston 1987; Kibenge *et al*. 2000).

SAV epidemiology is complex and multifactorial, varying significantly between sites, but not necessarily among year classes at the same site (Crockford *et al.* 2007) and all life stages may be affected (OIE 2014). Mortality rates may exceed 50% in farmed fish, but vary widely according to virus sub-type, year and host species (Jensen *et al*. 2012).

SAV is transmitted horizontally in marine and fresh water, and all life stages may be infected, but vertical transmission is considered unlikely (OIE 2016a). Viral particles are released from infected fish in faeces, mucous and fat droplets, while entry occurs through the epidermal cells of the gills and intestine (OIE 2016a). The minimum infective dose is unknown (Kahn *et al.* 1999) and replication occurs in the cell cytoplasm (McLoughlin & Graham 2007). SAV occurs in wild marine fish stocks which may function as reservoirs of infection (McLoughlin & Graham 2007), while the salmon louse is a vector for infection (Hodneland 2006).

Clinical disease manifests when several factors, including temperature and viral titre, are optimised (McLoughlin & Graham 2007; Stene *et al.* 2013) and infection progresses usually with few, or no external signs (OIE 2016a). Clinical signs commonly include necrotic lesions in pancreatic, heart and skeletal muscle tissue, although brain, gill, pseudobranch and kidney tissues may be infected (Stone *et al.* 1997; Graham *et al.* 2012). Infected fish develop pancreatic failure, with some (up to 15%) developing as stunted anorexic "runts" (McLoughlin & Graham 2007; OIE 2014). As SAV infection persists for at least 9 months after infection (Houghton 1994), surviving carrier fish remain infective until slaughter (Anderson *et al*. 2007; OIE 2014).

SAV remains viable for more than 2 months in sea water and hence water currents are important in determining spread of infection, which may be transferred between farms without direct human or animal interference (McLoughlin & Graham 2007). It remains viable at low temperatures, surviving freezing at -80°C for extended periods (OIE 2014). SAV is stable to the environmental conditions likely to occur in fish factory processing and storage, so biosecure slaughter methods, the safe disposal of diseased fish, and the safe disposal of infected offal and effluent are important factors in limiting the spread of SAV (McLoughlin & Graham 2007). SAV replication is inhibited by temperatures above 18°C (Hikke *et al.* 2014), so is unlikely to survive in the digestive system of piscivorous birds.

SAV in eviscerated fish is inactivated by heat (100°C for at least 30 minutes) (OIE 2014, 2016a), chloroform (McLoughlin & Graham 2007), acid conditions (pH < 3.0) or alkaline treatments (pH > 13.0) at 4°C, indicating that composting, ensilage and alkaline hydrolysis are effective treatment methods (OIE 2016b). SAV is also inactivated by disinfectant treatments (Graham *et al*. 2007).

A commercially available inactivated vaccine against SAV-1 is used in Norway, Ireland and Scotland. This confers incomplete protection, and reduces mortality by 50% (OIE 2016a).

Susceptible hosts in New Zealand include salmonids (particularly Atlantic salmon, brown trout and rainbow trout), as well as flatfish species (OIE 2016a).

26.2. Risk assessment

26.2.1. Entry assessment

SAV is a virus of variable pathogenicity reported from wild and farmed salmonids and flatfish (Snow *et al*. 2010). Surviving fish that act as carriers commonly have few external signs of infection (Anderson *et al.* 2007; McLoughlin & Graham 2007). These fish may pass visual inspection and be present in the commodity.

Infection is systemic, mainly occurring in the pancreas, heart, pseudobranch and kidney tissues (McLoughlin & Graham 2007; OIE 2016a), which are removed on evisceration. SAV also occurs in the brain, gills and in the skeletal musculature, which are retained in the commodity (McLoughlin & Graham 2007). The titre of SAV remaining in infected gill and brain tissue is unknown, but it is assumed that it may be sufficient to initiate infection in a suitable host. The likelihood of entry of SAV in eviscerated fish is assessed as non-negligible.

26.2.2. Exposure assessment

To establish infection in New Zealand, infected eviscerated fish product would have to become available for consumption by a susceptible fish host, in sufficient quantity and duration (Kahn *et al.* 1999). SAV may be present in the brain and gill tissues of fish offal and in blood-water discharged from commercial fish processing (Kahn *et al*. 1999; OIE 2016a). SAV is environmentally stable, remaining viable in marine waters for up to 2 months (McLoughlin & Graham 2007), but little information is currently available to determine its infectivity in non-salmonids (OIE 2016a). SAV replication is inhibited above 18°C (Hikke *et al.* 2014) and it is inactivated by composting, ensilage and landfill treatment (Graham *et al.* 2007; OIE 2014; 2016b). As SAV is rendered non-viable by passage through the avian digestive system (Hikke *et al.* 2014), it is unlikely to be redistributed after ingestion by scavenging birds.

The likelihood of exposure to SAV is assessed as non-negligible.

26.2.3. Consequence assessment

The establishment of SAV may have a significant effect on the aquaculture of Chinook salmon (*Oncorhynchus tshawytscha*), valued at $63 million in export earnings in 2011 (Aquaculture New Zealand 2014). An outbreak would also affect recreational and tourist trout and salmon fishing, as well as incur significant social and environmental costs associated with other domestic fisheries (Fish & Game 2014). While the dollar value of New Zealand's freshwater fisheries is not fully known, the Lake Taupo trout fishery alone is valued at $70 to 80 million (Marsh & Mkwra 2013).

The consequences of establishment of SAV are therefore assessed as non-negligible.

26.2.4. Risk estimation

The entry, exposure and consequence assessments for SAV are non-negligible, so the risk estimate is non-negligible. SAV is assessed to be a risk in the commodity and risk management measures may be justified.

26.3. Risk management

SAV has been assessed to be a risk in the commodity. As infection with SAV is an OIE-notifiable disease, the OIE *Aquatic Code* (OIE 2016a) provides guidance on risk management measures to prevent the international spread of SAV and specific processing requirements that would ensure the destruction of the virus.

Article 10.5.2 of the *Aquatic Code* lists the species recognised to need risk management measures to prevent the international spread of SAV:

> *The recommendations in this chapter apply to: Atlantic salmon (Salmo salar), brown trout (S. trutta) and rainbow trout (Oncorhynchus mykiss). These recommendations also apply to any other susceptible species referred to in the Aquatic Manual when traded internationally.*

Article 10.5.3 of the *Aquatic Code* recommends:

For the importation or transit of aquatic animals and aquatic animal products for any purpose from a country, zone or compartment not declared free from infection with salmonid alphavirus

1. *Competent Authorities should not require any conditions related to infection with SAV, regardless of the infection with SAV status of the exporting country, zone or compartment when authorising the importation or transit of the following aquatic animals and aquatic animal products from the species referred to in Article 10.5.2 intended for any purpose and complying with Article 5.4.1:*

    (a) heat sterilised, hermetically sealed fish products (i.e. a heat treatment at 121°C for at least 3.6 minutes or any time/temperature equivalent);
    (b) pasteurised fish products that have been subjected to a heat treatment at 90°C for at least ten minutes (or to any time/temperature equivalent which has been demonstrated to inactivate SAV);
    (c) mechanically dried, eviscerated fish (i.e. a heat treatment at 100°C for 30 minutes or any time/temperature equivalent which has been demonstrated to inactivate SAV).

2. *When authorising the importation or transit of aquatic animals and aquatic animal products of a species referred to in Article 10.5.2., other than those referred to in point 1 of Article 10.5.3., Competent Authorities should require the conditions prescribed in Articles 10.5.7 to 10.5.13, relevant to the SAV status of the exporting country, zone or compartment.*

3. *When considering the importation or transit of aquatic animals and aquatic animal products of a species not covered in Article 10.5.2. but which could reasonably be expected to pose a risk of spread of infection with SAV, the Competent Authority should conduct a risk analysis in accordance with the recommendations in Chapter 2.1. The Competent Authority of the exporting country should be informed of the outcome of this assessment.*

Article 10.5.11 of the *Aquatic Code* recommends:

For the importation of aquatic animals and aquatic animal products for retail trade for human consumption from a country, zone or compartment not declared free from infection with salmonid alphavirus:

1. *Competent Authorities should not require any conditions related to infection with SAV, regardless of the infection with SAV status of the exporting country, zone or compartment, when authorising the importation or transit of fish fillets or steaks (frozen or chilled) which have been prepared and packaged for retail trade and which comply with Article 5.4.2.*

   Certain assumptions have been made in assessing the safety of the aquatic animal products mentioned above. Member Countries should refer to these assumptions at Article 5.4.2 and consider whether the assumptions apply to their conditions.

   For these commodities Member Countries may wish to consider introducing internal measures to address the risks associated with the commodity being used for any purpose other than for human consumption.

2. *When importing aquatic animals or aquatic animal products, other than those referred to in point 1 above, of species referred to in Article 10.5.2 from a country, zone or compartment not declared free from infection with SAV, the Competent Authority of the importing country should assess the risk and apply appropriate risk mitigation measures.*

Compliance with Articles 10.5.3 and 10.5.11 should eliminate SAV from the commodity and be a viable risk management option.

Wild and farmed fish from 5 families (Table 23) are susceptible to SAV and may be present in the commodity. Species declaration should substantially reduce the occurrence of SAV in the commodity and be a viable risk management option. Restriction of the commodity to wild-caught fish (not from aquaculture) would have no effect on pathogen load and is not a viable risk management option.

Infection with SAV is an OIE-listed disease reported from Europe and the United Kingdom, Canada and North America (Kent & Elston 1987; Kibenge *et al*. 2000; OIE 2016b), that also occurs as a sub-clinical infection in non-salmonids (Jones *et al.* 2015, OIE 2016b). Approval of country/zone freedom through the MPI Country Approval Procedures should substantially reduce the occurrence of SAV in the commodity. Acceptance of a declaration of country/zone freedom by MPI is a viable risk management option.

SAV may be present in the brain and gill tissues of infected fish (Stone *et al.* 1997; Graham *et al.* 2012). Gill removal should slightly reduce the pathogen load of SAV, while removal of the head and gills is likely to moderately reduce the occurrence of SAV in the commodity. Processing to remove the head and gills is a viable risk management option.

SAV is inactivated by heat (100°C for at least 30 minutes) (OIE 2014, 2016a), so heat treatment would eliminate SAV from the commodity. Heat treatment is a viable risk management option.

SAV is not affected by freezing (OIE 2014), so frozen storage is not a viable risk management option.

While an inactivated vaccine against SAV-1 is used in Norway, Ireland and Scotland, this confers incomplete protection, and reduces mortality by 50% (OIE 2016a). A requirement for vaccinated fish would be unlikely to be a viable risk management option and is not discussed further.

26.3.1. Risk management options

Salmon alphavirus (SAV) is reported from fish in families Gadidae, Labridae, Pleuronectidae, Salmonidae and Scophthalmidae (Table 23), which are considered likely to be present in the commodity. Other families have not been associated with SAV. Therefore species declaration indicating the commodity is not originated from any of the above families should substantially reduce the occurrence of SAV in the commodity.

For the commodities originated from families associated with SAV, one or a combination of the following additional options could also be considered to effectively manage the risk.

Where country/zone freedom from SAV is accepted by MPI:

Option 1

Acceptance of country/zone freedom by MPI should substantially reduce the occurrence of SAV, so the commodity could be imported without any further restrictions.

Where country/zone freedom from SAV is not accepted by MPI, or not available.

Option 2

Processing consistent with the conditions of Article 10.5.3 or 10.5.11 of the OIE Aquatic *Code* (OIE 2016a) should eliminate SAV. When these provisions are met, the commodity could be imported without any further restrictions.

Where the imported commodity is not consistent with Article 10.5.3 or 10.5.11 of the OIE Aquatic *Code* (OIE 2016a), further processing is necessary:

Option 3

Further processing (by removal of the head and gills) should moderately reduce the occurrence of SAV in the commodity. When this provision is met, the commodity could be imported without any further restrictions.

26.4. References

Anderson L, Bratland A, Hodneland K, Nylund A (2007). Tissue tropism of salmonid alphaviruses (subtypes SAV1 and SAV3) in experimentally challenged Atlantic salmon (*Salmon salar* L.). *Archives of Virology* 152: 1871–1883.

Aquaculture New Zealand (2014). New Zealand's aquaculture industry key facts [Online] Available from: http://www.seafoodnewzealand.org.nz/our-industry/key-facts/ [Accessed 26 September 2014].

Castric J, Baudin LF, Bremont M, Jeffroy J, Le Ven A, Bearzotti M (1997). Isolation of the virus responsible for sleeping disease in experimentally infected rainbow trout (*Oncorhynchus mykiss*). *Bulletin of the European Association of Fish Pathologists* 17: 27–30.

Christie KE, Fyrand K, Holtet L, Rowley HM (1998). Isolation of pancreas disease virus from farmed Atlantic salmon, *Salmo salar* L. in Norway. *Journal of Fish Diseases* 21: 391–394.

Crane M, Hyatt A (2011). Viruses of fish: An overview of significant pathogens. *Viruses* 3(11): 2025–2046.

Crockford T, Menzies FD, McLoughlin MF, Villoing S, Todd D, Knappskog D (2007). Experimental infection of *Salmo salar* pre-smolts by i.p. injection with new Irish and Norwegian pancreatic disease (PDV) isolates – a comparative study. *Diseases of Aquatic Organisms* 75: 13–22.

Diggles BD (2011). Environmental assessment report- Disease risks. Report prepared for New Zealand King Salmon Ltd, DigsFish Services, Queensland, Australia, 68 p.

Fish and Game (2014). New Zealand Fish and Game Council [Online] Available from: http://www.fishandgame.org.nz/ [Accessed 24 September 2014].

Graham DA, Jewhurst H, McLoughlin MF, Sourd P, Rowley HM, Taylor C, Todd D (2006). Sub-clinical infection of farmed Atlantic salmon *Salmo salar* with salmonid alphavirus - A prospective longitudinal study. *Diseases of Aquatic Organisms* 72: 193–199.

Graham DA, Cherry K, Wilson CJ, Rowley HM (2007). Susceptibility of salmonid alphavirus to a range of chemical disinfectants. *Journal of Fish Diseases* 30: 269–277.

Graham DA, Frost P, McLaughlin K, Rowley HM, Gabestad I, Gordon A, McLoughlin MF (2011). A comparative study of marine salmon alphavirus subtypes 1–6 using an experimental cohabitation challenge model. *Journal of Fish Diseases* 34(4): 273–286.

Graham DA, Brown A, Savage P, Frost P (2012). Detection of salmon pancreas disease in the faeces and mucus of Atlantic salmon *Salmo salar* by real-time RT-PCR and cell culture following experimental challenge. *Journal of Fish Diseases* 35: 949–951.

Hikke MC, Verest M, Vlak JS, Pijlman GB (2014). Salmonid alphavirus replication in mosquito cells: towards a novel vaccine production system. *Microbiology and Technology* 7(5): 480–484.

Hodneland K (2006). Salmonid alphavirus: genetic characterisation of a new sub-type, SAV3 and implementation of a novel diagnostic method. Ph.D dissertation. University of Bergen, Norway, 107 p.

Houghton G (1994). Acquired protection in Atlantic salmon *Salmo salar* parr and post-smolts against pancreas disease. *Diseases of Aquatic Organisms* 18: 109–118**.**

Jansen MD, Jensen DD, McLoughlin MF, Rodger HD, Taksdal T. Sindre H, Graham DA, Lillehaug A (2017). The epidemiology of pancreas disease in salmonid aquaculture: a summary of the current state of knowledge. *Journal of Fish Diseases* 40: 141–155.

Jensen BB, Kristoffersen AB, Myr C, Brun E (2012). Cohort study of effect of vaccination on pancreas disease in Norwegian salmon aquaculture. *Diseases of Aquatic Organisms* 102: 23–31.

Jones SRM, Bruno DW, Madsen L, Peeler EJ (2015). Disease management mitigates risk of pathogen transmission from maricultured salmonids. *Aquaculture Environment Interactions* 6: 119–134.

Kahn SA, Beers PT, Findlay VL, Peebles IR, Durham PJ, Wilson DW, Gerrity SE (1999). Import risk analysis on non-viable salmonids and non-salmonid marine finfish. Australian Quarantine and Inspection Service, Department of Agriculture, Fisheries and Forestry, Canberra, Australia, 409 p.

Karlsen M, Hodneland K, Endresen C, Nylund A (2006). Genetic stability within the Norwegian subtype of salmonid alphavirus (family Togaviridae). *Archives of Virology* 151: 861–874.

Kent ML, Elston RA (1987). Pancreas disease in pen-reared Atlantic salmon in North America. *Bulletin of the European Association of Fish Pathologists* 7: 29–31.

Kibenge FSB, Whyte SB, Hanmmell KL, Rainnie D, Kibenge MT, Martin CK (2000). A dual infection of infectious salmon anaemia (ISA) and togavirus-like virus in ISA of Atlantic salmon (*Salmo salar*) in New Brunswick. *Diseases of Aquatic Organisms* 42: 11–15.

Marsh D, Mkwra L (2013). Review of freshwater non-market value studies. Department of Economics, University of Waikato, Hamilton, New Zealand, 144 p.

McCleary SJ, Giltrap M, Henshilwood K, Ruane NM (2014). Detection of salmonid alphavirus RNA in Celtic and Irish Sea flatfish. *Diseases of Aquatic Organisms* 109 (1): 1–7.

McLoughlin MF, Graham DA (2007). Salmon alphaviruses: - a review. *Journal of Fish Diseases* 30: 511–531.

McVicar AH, Munro ALS (1989). Pancreas disease in farmed Atlantic salmon: Evidence for an infectious aetiology pp 70-81. *In*: Aquaculture: A Review of Recent Experience. OCED Paris, France.

Munro ALS, Ellis AE, McVicar AH, McLay AH, Needham EA (1984). An exocrine pancreas disease of farmed Atlantic salmon in Scotland. *Helgolander Meeresunters* 37: 571–586.

Nelson RT, McLoughlin MF, Rowley HM, Platten MA, McCormick JI (1995). Isolation of a toga-like virus from farmed Atlantic salmon *Salmo salar* with pancreas disease. *Diseases of Aquatic Organisms* 22: 25–32.

OIE (2014). Report of the OIE ad-hoc group on the safety of products derived from aquatic animals, February 2014. World Organization for Animal Health, Paris, France, 30 p.

OIE (2016a). Aquatic Animal Health Code, 19th Edition. World Organization for Animal Health. Paris, France [Online] Available from: http://www.oie.int/international-standard-setting/aquatic-code/access-online/ [Accessed 15 March 2017].

OIE (2016b). Manual of diagnostic tests for aquatic animals. World Organization for Animal Health. Paris, France [Online] Available from: http://www.oie.int/international-standard-setting/aquatic-manual/access-online/ [Accessed 17 March 2017].

Petterson E, Sandberg M, Santi N (2009). Salmonid alphavirus associated with *Lepeophtheirus salmonis* (Copepoda: Caligidae) from Atlantic salmon, *Salmo salar* L. *Journal of Fish Diseases* 32: 477–479.

Petterson E, Stormoen M, Evensen O, Mikalsen AB, Haugland O (2013). Natural infection of Atlantic salmon (*Salmo salar*) with salmonid alphavirus 3 generates numerous viral deletion mutants. *Journal of General Virology* 94: 1945–1954.

Snow M, Black I, McIntosh R, Baretto E, Wallace IS, Bruno DW (2010). Detection of salmonid alphavirus RNA in wild marine fish: implications for the origin of salmon pancreas disease in aquaculture. *Diseases of Aquatic Organisms* 91: 177–188.

Stene A, Hellebo A, Viljugrein H, Solvag SE, Aspehaug V (2013). First detection of salmonid alphavirus in liquid fat fractions leaking from dead PD infected salmon. *Journal of Fish Diseases* 37(2): 77–162.

Stone, MAB, MacDiarmid SC, Pharo HJ (1997). Import health risk analysis: salmonids for human consumption. Ministry of Agriculture Regulatory Authority, New Zealand, 269 p.

Taksdal T, Hjortaas MJ, Jansen MD, Jensen BB, Brun E, Røsæg MV, Persson DEL, Olsen AB, Hjeltnes B, Sindre H, Aspehaug V, Devold M., Knapskog D, Breck O. (2014). Hva betyr ny PD-virusvariant for norsk fiskeoppdrett? Karakterisering avvirus, sykdomsutvikling og utbredelse. Preliminary Final Report,Veterinærinstituttet Norway. (In Norwegian, English abstract).

27. Salmon gill poxvirus

27.1. Hazard identification

27.1.1. Aetiological agent

Salmon gill poxvirus (SGPV) is a linear, double stranded DNA virus classified within the Chordopoxvirinae (Upton *et al.* 2003). SGPV is the agent of proliferative gill disease of salmon, viral carp oedema and koi sleepy disease (Lewish *et al.* 2015). It was first identified in Norwegian farmed Atlantic salmon (*Salmo salar*) (Nylund *et al.* 2008), which represented the first record of poxvirus in fish (Gjessing *et al.* 2015). SGPV is regarded as an emerging disease (Gjessing *et al.* 2015; Lewish *et al.* 2015).

27.1.2. OIE status

Infection with SGPV is not listed by the OIE (OIE 2016).

27.1.3. New Zealand status

SGPV has only been reported from Europe and Japan (Gjessing *et al.* 2015; Lewish *et al.* 2015). SGPV has not been reported from New Zealand salmonids (Cobb 2008; B. Jones, MPI, *pers. comm.* 2017) and it is assumed to be exotic. Infection with SGPV is not a notifiable disease in New Zealand (Anon. 2016).

27.1.4. Epidemiology

SGPV is reported from Europe (Austria, France, Germany, Holland, Norway and the United Kingdom) (Nylund *et al*. 2008; Way & Stone 2013; Haenen *et al.* 2016) and from Japan (Ono *et al*. 1986; Wada *et al*. 2008; Gjessing *et al.* 2015).

SGPV is reported from fish of families Cyprinidae, Plecoglossidae and Salmonidae (Table 24). High mortality (up to 100%) has been reported in Norwegian Atlantic salmon (*Salmo salar*) freshwater aquaculture since the 1990s (Nylund *et al.* 2008; Gjessing *et al.* 2015). High mortality (80-100%) is also reported in farmed ayu (*Plecoglossus altivelis*), in European and Japanese freshwater aquaculture since the 1970s (Ono *et al*. 1986; Wada *et al*. 2008; Haenen *et al.* 2014; Gjessing *et al.* 2015; Lewish *et al*. 2015).

Table 24. Families and Species of Fish Susceptible to Salmon Gill Poxvirus (SGPV)

Family	Host Species
Cyprinidae	Common and ornamental carp (*Cyprinus carpio*), grass carp (*Ctenopharyngodon idella*), silver carp (*Hypophthalmichthys molitrix*)
Plecoglossidae	Ayu (*Plecoglossus altivelis*)
Salmonidae	Atlantic salmon (*Salmo salar*)

SGPV has been isolated from wild European cyprinids but not from wild salmonids (Gjessing *et al.* 2015; Lewish *et al.* 2015). Genomic and diagnostic PCR analysis indicates SGPV has recently spread across Europe in wild and farmed fish (Haenen *et al.* 2014; Gjessing *et al.* 2015; Lewish *et al.* 2015). Little is known about the epidemiology of SGPV, but its high virulence suggests a recent spread to new species (Gjessing *et al*. 2015).

SGPV invades gill tissues, with few external signs of disease. SGPV replicates in the gill tissues and infection may progress along several pathways. It causes gill oedema in small juvenile carp and koi sleepy disease in larger juvenile fish, with lamellar cell apoptosis, hypertrophy of the lamellar gill epithelium and haemophagocytosis (Gjessing *et al.* 2015).

SGPV infection may become systemic, with myocarditis in heart muscle. Infection further progresses to multifocal necrotic lesions in the hepatopancreas and spleen, with multifocal leucocyte infiltration between the necrotic fibres (Lewish *et al.* 2015). Infection is chronic in older fish, with lower mortality, and surviving fish become reservoirs of infection (Gjessing *et al.* 2015).

Poxviruses are generally epitheliotrophic, but SGPV infection is not associated with skin lesions in fish, and the viral particles present in skin tissue are most likely shed from the gills (Gjessing *et al.* 2015). Little information is available on the duration of disease. SGPV is generally reported from freshwater fish, but adult carrier salmon may remain infected after smoltification in sea water (Gjessing *et al.* 2015).

Poxviruses are environmentally stable, remaining viable for up to 39 weeks at a moisture of 6.7% at 4°C (Anon. 2011). The infectious dose in water is unknown, but terrestrial poxvirus generally require a titre of 10^8 pfu mL^{-1} to initiate infection (Anon. 2011).

Poxviruses are commonly freeze-dried and appear unaffected by freezing (Gjessing *et al.* 2015). They are heat labile, requiring near-autoclave treatment (dry heat treatment at 95°C for 2 hours, or moist heat at 60°C for 2 hours) for inactivation (Anon. 2011). While denatured by disinfectants (0.02% sodium hypochlorite, 30% isopropanol, 40% ethanol), they are resistant to solvent/detergent combinations, requiring extended contact (from 10 minutes to 24 hours) to ensure inactivation (Anon. 2011).

No vaccines are available for SGPV disease (Gjessing *et al.* 2015).

Susceptible species in New Zealand include salmonids (*Salmo* spp.), carp (*C. carpio*) and grass carp (*C. idella*) in fresh water, although the host range of this emerging pathogen remains poorly defined (Gjessing *et al.* 2015).

27.2. Risk assessment

27.2.1. Entry assessment

SGPV mainly affects fry and juvenile fish with high mortality, but adult carrier fish with no external signs of infection (Gjessing *et al.* 2015; Lewish *et al.* 2015) would be likely to pass visual inspection. SGPV is likely to survive routine handling and storage processes in food preparation (Anon. 2011; Gjessing *et al.* 2015) and may remain viable in the skin and gill tissues retained in the eviscerated commodity (Anon. 2011; Gjessing *et al*. 2015). SGPV is likely to survive in the blood-water and offal waste discharges from factory processing (Anon. 2011; Lewish *et al*. 2015).

The likelihood of entry is assessed as non-negligible.

27.2.2. Exposure assessment

SGPV may be present in the skin and gill tissues retained in the eviscerated commodity (Gjessing *et al.* 2015; Lewish *et al.* 2015). To establish infection, this infected product would have to become available for consumption by a susceptible fish host, in sufficient quantity and duration (Kahn *et*

al. 1999). SGPV may remain viable in blood-water discharge and offal discarded from fish processing (Anon. 2011), although details of the epidemiology of aquatic poxviruses are unknown (Gjessing *et al.* 2015; Lewish *et al.* 2015).

Based on reported host species overseas (Gjessing *et al.* 2015; Lewish *et al.* 2015), potential hosts in New Zealand include Atlantic salmon (*S. salar*), brown trout (*S. trutta*), as well as common and ornamental carp (*C. carpio*), grass carp (*Ctenopharyngodon idella*) and silver carp (*Hypophthalmichthys molitrix*) in fresh water. SGPV causes high mortalities, particularly in naïve populations, with rapid onset of disease (Gjessing *et al.* 2015), but has become established in wild European carp populations and is considered an emerging risk in European freshwater aquaculture (Lewish *et al.* 2015).

The likelihood of exposure to SGPV is assessed as non-negligible.

27.2.3. Consequence assessment

SGPV is considered an emerging pathogen of medium-high mortality mainly affecting salmonids and carp in freshwater aquaculture, but little is known about its host range and epidemiology (Lewish *et al.* 2015). Atlantic salmon (*S. salar*) in New Zealand is limited to the upper Waiau catchment in the South Island of New Zealand and is not commercially farmed (NIWA 2014a, 2014b), so the consequences for this species are likely to be low.

The water supplies in New Zealand aquaculture are commonly untreated, so hatchery facilities are at risk from aquatic pathogens (Sim-Smith *et al.* 2014). The establishment of SGPV would have significant consequences for brown trout (*S. trutta*), which supports major recreational and tourist fisheries (Fish & Game 2014). While the dollar value of New Zealand's freshwater fisheries is not fully known, the Lake Taupo trout fishery alone is valued at $70 to 80 million (Marsh & Mkwra 2013).

SGPV may become established in introduced carp species, including common and ornamental carp (*C. carpio*), which are considered as noxious fish (Anon. 2016). As grass carp (*C. idella*) and silver carp (*H. molitrix*) are farmed for weed management in waterways (Mitchell 1980, 2008; Hofstra 2014; NIWA 2014b), the consequences for these developing fisheries would be severe. The potential for other introduced carp species to act as a reservoir for SGPV (Lewish *et al.* 2015) is unknown.

The consequences of establishment of SGPV are assessed as non-negligible.

27.2.4. Risk estimation

Since the entry, exposure and consequence assessments are non-negligible, SGPV is assessed as a risk in the commodity and risk management measures may be justified.

27.3. Risk management

SGPV has been assessed to be a risk in the commodity. Infection with SGPV is not listed by the OIE (OIE 2016) so the OIE *Aquatic Code* provides no specific guidance on importing eviscerated fish from infected countries, or specific processing requirements that would ensure the destruction of the virus. It is assumed that compliance with the approved processed states for OIE-listed diseases (Appendix 3) should also eliminate SGPV from the commodity and be a viable risk management option.

SGPV has been isolated from three families of wild and farmed fish (Table 24), which may be present in the commodity. Species declaration should substantially reduce the pathogen load of SGPV and be a viable risk management option. However, little is known about the epidemiology and host range of SGPV (Lewish *et al.* 2015). It is considered an emerging risk in European and Japanese freshwater aquaculture (Gjessing *et al.* 2015; Lewish *et al.* 2015). Restriction of the commodity to wild-caught fish (not from aquaculture) would have no effect on pathogen load and is not a viable risk management option.

SGPV is widely reported from Europe (Austria, France, Germany, Holland, Norway and the United Kingdom) (Nylund *et al*. 2008; Way & Stone 2013; Haenen *et al.* 2014) and Japan (Ono *et al*. 1986; Wada *et al*. 2008; Gjessing *et al.* 2015), but no requirements for dedicated monitoring exist. Approval of country/zone freedom through the MPI Country Approval Procedures should substantially reduce the occurrence of SGPV in the commodity. Acceptance of a declaration of country/zone freedom by MPI is a viable risk management option.

SGPV infection is focused on gill tissues, although viral particles may be present on the skin of infected fish (Gjessing *et al.* 2015). Infection may progress to the visceral organs, but does not affect the dermal musculature (Gjessing *et al.* 2015). Removal of the gills should slightly reduce the pathogen load of SGPV, while removal of the head and gills would moderately reduce pathogen load of SGPV in the commodity. Further processing to the skin-off fillet state would substantially reduce the occurrence of SGPV in the commodity and be a viable risk management option.

Poxviruses are resistant to freezing (Anon. 2011), so frozen storage is not a viable risk management option. Inactivation requires high temperature treatment with moist heat (at 60°C for 120 minutes) (Anon. 2011). High temperature treatment would eliminate SGPV from the commodity and be a viable risk management option.

27.3.1. Risk management options

Salmon gill poxvirus (SGPV) is reported from fish in families Cyprinidae, Plecoglossidae and Salmonidae (Table 24), which are considered likely to be present in the commodity. Other families have not been associated with SGPV. Therefore species declaration indicating the commodity is not originated from any of the above families should substantially reduce the occurrence of SGPV in the commodity.

For the commodities originated from families associated with SGPV, one or a combination of the following additional options could also be considered to effectively manage the risk.

Where country/zone freedom from SGPV is accepted by MPI:

Option 1

Acceptance of country/zone freedom by MPI should substantially reduce the occurrence of SGPV, so the commodity may be imported without any further restrictions.

Where country/zone freedom from SGPV is not accepted by MPI or not available:

Option 2

Processing to a state consistent with the approved states for OIE-listed diseases (Appendix 3) should eliminate SGPV. When these provisions are met, the commodity could be imported without any further restrictions.

Where the imported commodity is not in a state consistent with the approved states for OIE-listed diseases (Appendix 3):

Option 3

Heat treatment (by cooking, with moist heat, to at least 60°C for at least 120 minutes) should eliminate SGPV. When this provision is met, the commodity could be imported without any further restrictions; or,

Option 4

Further processing (to the skin-off fillet state) should substantially reduce the occurrence of SGPV. When this provision is met, the commodity could be imported without any further restrictions.

27.4. References

Anonymous (2011). Vaccinia virus: Pathogen safety data sheet. Public Health Agency of Canada [Online]. Available from: http://www.phac-aspc.gc.ca/lab-bio/res/psds-ftss/vaccinia-virus-eng.php [Accessed 28 September 2015].

Anonymous (2016). Biosecurity (Notifiable Organisms) Order 2016. NZLR 2016/73. New Zealand Government, Wellington, New Zealand, 13 p.

Cobb S. (2008). Import risk analysis: Fish food. Biosecurity New Zealand Ministry of Agriculture and Forestry, Wellington, New Zealand. 57 p.

Fish and Game (2014). New Zealand Fish and Game Council [Online]. Available from: http://www.fishandgame.org.nz/ [Accessed 24 September 2014].

Gjessing MC, Yutin N, Tengs T, Senkevitch T, Koonin E, Ronning HP, Alarcon M, Ylving S, Lie KI, Tran L, Moss B, Dale OB (2015). Salmon gill poxvirus, the deepest representative of the Chordopoxvirinae. *Journal of Virology* 89(18): 9348–9367.

Haenen O, Way K, Stone D, Engelsma M (2014). "Koi sleepy disease" found for the first time in koi carps in the Netherlands. *Tijdscher Diergeneeskd* 139: 26–26 (in Dutch).

Hofstra DE (2014). Grass carp Effectiveness and Effects, Stage 2: Knowledge review. NIWA Client Report HAM2014-060, Prepared for the Department of Conservation. National Institute of Water & Atmospheric Research Ltd, Hamilton, New Zealand, 139 p.

Kahn SA, Beers PT, Findlay VL, Peebles IR, Durham PJ, Wilson DW, Gerrity SE (1999). Import risk analysis on non-viable salmonids and non-salmonid marine fish. Australian Quarantine and Inspection Service, Department of Agriculture, Fisheries and Forestry, Canberra, Australia, 409 p.

Lewish E, Gorgoglione B, Way K, El-Matbouli M (2015). Carp edema virus/Koi sleepy sickness: an emerging disease in Central-East Europe. *Transboundary and Emerging Diseases* 62(1): 6–12.

Marsh D, Mkwra L (2013). Review of freshwater non-market value studies. Department of Economics, University of Waikato, Hamilton, New Zealand, 144 p.

Mitchell, C. (1980). Control of water weeds by grass carp in two small lakes. *New Zealand Journal of Marine and Freshwater Research* 14(4): 381–390.

Mitchell, C. (2008) Environmental impact assessment report for the introduction of Grass carp (*Ctenopharyngodon idella*) into Lake Swan, Pouto lakes, North Kaipara Head. Report prepared for the Northland Regional Council. Mitchell and Associates, Auckland, New Zealand.

NIWA (2014a). Salmon. [Online] Available from: https://www.niwa.co.nz/freshwater-and-estuaries/nzffd/NIWA-fish-atlas/fish-species/salmonidae) [Accessed 28 July 2017].

NIWA (2014b). Grass carp *Ctenopharyngodon idella* (Valenciennes, 1844) [Online] Available from: https://www.niwa.co.nz/freshwater-and-estuaries/nzffd/NIWA-fish-atlas/fish-species/grass_carp [Accessed 12 February 2015].

Nylund A, Watanabe K, Nylund S, Karlsen M, Saether PA, Arnesen CE, Karlsbakk E (2008). Morphogenesis of salmonid poxvirus associated with proliferative gill disease in farmed Atlantic salmon (*Salmo salar*) in Norway. *Archives of Virology* 153: 1299–1309.

OIE (2016). Aquatic Animal Health Code, 19th Edition. World Organization for Animal Health. Paris, France [Online] Available from: http://www.oie.int/international-standard-setting/aquatic-code/access-online/ [Accessed 15 March 2017].

Ono S, Nagai A, Sugani N (1986). A histopathological study on juvenile colorcarp *Cyprinus carpio* showing edema. *Fish Pathology* 9: 9.

Sim-Smith C, Faire S, Lees A (2014). Managing biosecurity risk for business benefit. Report prepared for the Ministry for Primary Industries by Coast and Catchment Ltd, Auckland, New Zealand, 214 p.

Upton C, Slack S, Arwen LH, Ehlers A, Roper RL (2003). Poxvirus orthologous clusters: Towards defining the minimum essential poxvirus genome. *Journal of Virology* 77(13): 7590–7600.

Wada S, Kurata O, Hatai K, Ishii H, Kayusa K, Watanabe Y (2008). Proliferative branchitis associated with pathognomonic, atypical gill epithelial cells in cultured ayu *Plecoglossus altivelis. Fish Pathology* 43: 89–91.

Way K, Stone D (2013). Emergence of carp edema virus-like (CEV-like) disease in the U.K. *Finfish News* 15: 32–34.

28. Spring viraemia of carp virus and associated rhabdoviruses

28.1. Hazard identification

28.1.1. Aetiological agent

Spring viraemia of carp (SVC) is an acute and haemorrhagic viral disease of freshwater fish, caused by spring viraemia of carp virus (SVCV). It is a non-segmented, negative sense, single stranded RNA virus, tentatively classified in the Genus *Vesiculovirus*, within the Family Rhabdoviridae (OIE 2016a).

Hydrocephalic disease, or red disease, was first recognised from the fry and fingerlings of pike (*Esox lucius*) (OIE 2016b) and the agent was identified as pike fry rhabdovirus (PFRV) by de Kinkelin *et al.* (1973).

Four genogroups of SVCV are recognised, each containing strains of varying pathogenicity (Haenen & Davidse 1993; Stone *et al.* 2003). The first genogroup includes all previously reported isolates of SVCV from a variety of hosts and geographical areas. The second genogroup includes isolates of pike fry rhabdovirus disease virus (PFRV) specifically from grass carp, while the third and fourth groups include the remaining isolates previously reported as PFRV from a variety of hosts (OIE 2016b). PFRV is therefore considered as an isolate of SVCV in this report.

28.1.2. OIE status

Infection with spring viraemia of carp virus (SVCV) is listed by the OIE as a notifiable disease (OIE 2016a). PFRV is recognised as a strain of SVCV within genogroup II (OIE 2016b).

28.1.3. New Zealand status

SVCV is considered exotic (Tubbs *et al*. 2007) and has not subsequently been reported (B. Jones, *pers. comm*. 2015). SVCV is a notifiable disease in New Zealand (Anon. 2010).

28.1.4. Epidemiology

SVCV has been reported from continental Europe, including Russia and the former Soviet states of Belarus, Georgia, Lithuania, Moldavia and Ukraine (OIE 2016a). It was reported from China in 1994, from Brazil in 1998 and from the United States and Canada in 2006 (OIE 2016a).\

SVCV isolates are reported from a wide range of freshwater fish (Table 25) including rainbow trout (*O. mykiss*) and all cyprinid species in temperate waters (Haenen & Davidse 1993; Svetlana *et al.* 2006; Cipriano *et al.* 2011; OIE 2016a, 2016b). SVCV also occurs in invertebrates, including the Pacific white shrimp (*Litopenaeus vannamei*), and can cause experimentally induced mortality in Pacific blue shrimp (*L. stylirostris*) (OIE 2016a).

While PFRV isolates affect brown trout (*Salmo trutta*) in Europe (Holland and Germany), they are not pathogenic for rainbow trout (*O. mykiss*) (Schaperclaus 1992; Haenen & Davidse 1993; OIE 2016a)

Table 25. Families and Species of Fish Susceptible to Spring Viraemia of Carp Virus and Pike Fry

Rhabdovirus Family	**Host Species**
Centrarchidae	Pumpkinseed (*Lepomis gibbosus*), bluegill (*Lepomis macrochirus*), largemouth black bass (*Micropterus salmoides*)
Cichlidae	Nile tilapia (*Oreochromis niloticus*)
Cyprinidae	Bighead carp (*Hypophthamichthys* (*Aristichthys) nobilis*), bream (*Abramis brama*), catla (*Gibelion catla*), common and ornamental carp (*Cyprinus carpio*), crucian carp (*Carassius carassius*), emerald shiner (*Notropis atherinoides*), goldfish (*Carassius auratus*), grass carp (*Ctenopharyngodon idella*), mrigal (*Cirrhinus mrigala*), orfe (*Leuciscus idus*), roach (*Rutilus rutilus*), rohu (*Labeo rohita*), silver carp (*Hypophthalmichthys molitrix*), tench (*Tinca tinca*), white bream (*Blicca bjoerkna*)
Esocidae	Pike (*Esox lucius*)
Percidae	European perch (*Perca fluviatilis*)
Salmonidae	Brown trout (*Salmo trutta*), rainbow trout (*Oncorhynchus mykiss*)
Siluridae:	Sheatfish/European catfish (*Silurus glanis*).

Disease outbreaks of SVCV are commonly associated with warmer water temperatures (11 to 17°C), but virulence decreases as temperatures increase above 22°C. Disease commonly occurs during the northern hemisphere spring season, as fish recover from over-wintering (Haenen & Davidse 1993; OIE 2016b). Juvenile fish (up to 1 year old) are most susceptible, although SVCV may affect fish of all ages (OIE 2016b). Mortality is significantly lower (1 to 4%) for older fish (OIE 2016a) and surviving fish become carriers with few or no external signs of disease (Haenen & Davidse 1993; Bootsma *et al.* 2006).

The virulence of SVCV varies widely, depending upon the host species, fish age and immune status, the viral strain, together with environmental factors including water temperature and host density (OIE 2016b). Mortality for SVCV isolates usually ranges from 4 to 40%, but may reach 70% during outbreaks (OIE 2016b). Mortalities for PFRV isolates range from 4 to 18% for grass carp (*Ctenopharyngodon idella*), and from 85 to 100% for pike (*Esox lucius*) (Haenen & Davidse 1993).

Initial invasion occurs across the host gill membranes. SVCV initially causes viraemia of the gills, which spreads to the liver, spleen and digestive tissues, then to the skin and external musculature (OIE 2016b).

Infection with the PFRV isolate may occur as one of two forms of disease: hydrocephalic PFRV, or haemorrhagic PFRV (Bootsma 1976). Infection with hydrocephalic PFRV is characterised by hydrocephalic swellings of the third ventricle of the brain, resulting in extensions of the head and distinctive exophthalmia. This form commonly occurs at cooler temperatures (from 10 to 17°C) (Schaperclaus 1992).

Infection with haemorrhagic PFRV is characterised by extensive haemorrhaging of the muscle and connective tissues, resulting in large red swellings at the base of the pelvic fins. This form commonly occurs when temperatures reach 15 to 20°C (Schaperclaus 1992). In both forms, the disease subsequently invades the skeletal musculature, spinal cord, pancreas, kidney, and liver tissues, with lower titres occurring in spleen, gills and brain tissue (OIE 2016b).

Disease transmission is horizontal, through the water, with viral particles shed by infected fish in faeces, urine, gill, and skin mucous and exudates from skin blisters (OIE 2016b).

Vertical transmission also occurs in hatcheries through infected eggs, and infection occurs through ovarian fluid (Haenen & Davidse 1993; OIE 2016b).

SVCV isolates are resistant to freezing, remaining viable after frozen storage (at -30°C or -80°C) for 6 months (de Kinkelin *et al*. 1973; OIE 2016a), and after four freeze-thaw cycles (to -30°C) (OIE 2016b). They are temperature labile, requiring temperatures exceeding 60°C for 10 minutes for inactivation (Tubbs *et al.* 2007). They are resistant to acid pH, being inactivated by pH < 3, for three hours. They are also resistant to alkaline pH, with inactivation requiring treatment of pH >12 for 10 minutes (OIE 2016b).

SVCV isolates are environmentally stable, remaining viable in river water at 10°C for 5 weeks, and in pond mud for 6 weeks at 4°C or 4 weeks at 10°C (OIE 2016b). They may be transferred by vectors including the fish louse (*Argulus foliaceus*) and leech (*Piscicola geometra*). SVCV remained viable for up to 120 minutes after passage through the heron (*Ardea cinerea*). It occurs in invertebrates, where SVCV is over 99% serologically related to rhabdoviral disease of Pacific white shrimp (*Penaeus vannamei*) (OIE 2016b). Rhabdoviruses may also be transferred by fomites, including aquaculture and fish processing equipment (OIE 2016b).

Rhabdoviruses are inactivated by organic iodophor treatment (50 mg L^{-1} for 10 minutes) (Bootsma 1976), chlorine (540 mg L^{-1} for 20 minutes), formalin (3% for 5 minutes), sodium hypochlorite (500 mg L^{-1} for 10 minutes) and sodium hydroxide (2% for 10 minutes). Treatment with ozone gamma or UV radiation is also effective (Tubbs *et al*. 2007; OIE 2016b).

Potential hosts in New Zealand include brown trout (*S. trutta*), common carp (*Cyprinus carpio*), goldfish (*Carassius auratus*), grass carp (*Ctenopharyngodon idella*), rainbow trout (*O. mykiss*), European perch (*P. fluviatilis*) and tench (*Tinca tinca*) (Tubbs *et al*. 2007; OIE 2016a).

28.2. Risk assessment

28.2.1. Entry assessment

SVCV is reported from a wide host range of freshwater fish (Table 26) often with high mortality (Haenen & Davidse 1993; OIE 2016a, 2016b). Fish clinically infected with the hydrocephalic or haemorrhagic forms of the PFRV isolate would be unlikely to pass visual inspection and should not be present in the commodity. Fish sub-clinically infected with the SVCV isolate may be carriers, with few or no external signs of infection. They may enter the human food consumption pathway (Haenen & Davidse 1993; Bootsma *et al*. 2006).

Infection with SVCV typically affects the gills, liver, spleen and viscera, as well as the skin and external musculature. In the hydrocephalic phase SVCV targets the brain and neural tissues (Bootsma 1976; Schaperclaus 1992). While evisceration would substantially reduce viral titre, it is likely that infective material would remain in the gill tissues, the neural tissues of the brain and spinal cord, as well as in the skeletal musculature of chronically infected fish.

The likelihood of entry through the commodity is assessed as non-negligible.

28.2.2. Exposure assessment

To establish infection in New Zealand, infected eviscerated product would have to become available for consumption by a susceptible freshwater fish host, in sufficient quantity and duration (Kahn *et al.* 1999). Viral pathogens such as SVCV and its associated rhabdoviruses may be present in the blood-water discharge as well as in the offal derived from commercial fish processing (OIE 2016a). They are also likely to remain viable in fish frames (gill tissues and neural tissues of the brain and spinal cord); the skin (connective tissue and skin); as well as in the skeletal muscle

discarded after trimming, in commercial food processing (Haenen & Davidse 1993; Bootsma *et al.* 2006).

SVCV and associated rhabdoviruses are resistant to freezing and remain viable in fresh water and sediment for extended periods (OIE 2016b) and can be transferred by invertebrate vectors including the fish louse (*Argulus foliaceus*) and the leech (*Piscicola geometra*). Rhabdoviruses are resistant to the temperature and acid conditions encountered in standard processes for organic material disposal. They remain viable in fish wastes discarded to landfill and can be transferred to the aquatic environment by piscivorous birds such as the grey heron (*Ardea cinerea*) (OIE 2016b). Rhabdoviruses may also be transferred by fomites, including aquaculture and fish processing equipment (OIE 2016b).

The likelihood of exposure to SVCV and associated rhabdoviruses is assessed as non-negligible.

28.2.3. Consequence assessment

The establishment of SVCV and associated rhabdoviruses may have an immediate impact upon the industries associated with the farming and use of acclimatised grass carp (*C. idella*) for aquatic weed control (Clayton & Wells 1999; NIWA 2014). Their introduction may also have a social impact upon the existing recreational coarse fisheries for roach (*R. rutilus*), and tench (*Tinca tinca*) (Rowe 2004; Fish & Game 2014). Other freshwater fish may act as carriers and represent a reservoir for disease.

SVCV isolates are reported from brown trout (*Salmo trutta*) and rainbow trout (*O. mykiss*), but not from Chinook salmon (*O. tshawytscha*) (OIE 2016b). While these viruses infect juveniles of a variety of freshwater fish (Svetlana *et al.* 2006) not all SVCV isolates potentially affect salmonids (OIE 2016b). An outbreak could affect the recreational and tourist trout fisheries for brown and rainbow trout, as well as incur significant social and environmental costs associated with other domestic fisheries (Fish & Game 2014). While the dollar value of New Zealand's freshwater fisheries is not fully known, the Lake Taupo trout fishery alone is valued at $70 to 80 million per annum (Marsh & Mkwra 2013).

The consequence of establishment could be extremely high if SVCV affects the Chinook salmon aquaculture industry, which was worth $63 million in export earnings in 2011 (Aquaculture New Zealand 2014).

The consequences of establishment of SVCV and associated rhabdoviruses are assessed as non-negligible.

28.2.4. Risk estimation

Since the entry, exposure and consequence assessments for SVCV are non-negligible, the risk estimate is non-negligible. Therefore, SVCV and associated rhabdoviruses are assessed to be a risk in the commodity and risk management measures may be justified.

28.3. Risk management

SVCV isolates are reported from several families of wild and farmed freshwater fish (Table 25) which may be present in the commodity. Species declaration should substantially reduce the occurrence of SVCV in the commodity and be a viable risk management option. Restriction of the

commodity to wild-caught fish (not from aquaculture) would have little, or no effect on pathogen load and is not a viable risk management option.

SVCV and isolates are widely distributed in freshwater systems of continental Europe, including Belarus, Georgia, Lithuania, Moldavia, Russia and Ukraine, as well as China, Brazil, the United States and Canada. While infection with SVCV is an OIE-listed disease, isolates also occur in non-cyprinid fish (OIE 2016a). Approval of country/zone freedom through the MPI Country Approval Procedures should substantially reduce the occurrence of SVCV and isolates in the commodity. Acceptance of a declaration of country/zone freedom by MPI is a viable risk management option.

SVCV and isolates are unaffected by freezing, so frozen storage is not a viable risk management option. Inactivation by high temperature treatment (to at least 60°C for 10 minutes) (OIE 2016a), should eliminate SVCV and isolates from the commodity. High-temperature treatment is a viable risk management option.

SVCV and isolates are likely to be present in the blood-water discharge and fish offal routinely discarded from commercial fish processing. They may remain viable in the head and backbone (including neural tissue of the brain and spinal column, and the gill tissues, in the skin, as well as in the trimmings of the skeletal musculature, that comprise fish offal. Removal of the gills, or the head and gills, should slightly reduce the occurrence of SVCV and isolates, while further processing to the skin-off fillet state should moderately reduce the occurrence of SVCV in the commodity and be a viable risk management option.

Article 10.9.2 of the *Aquatic Code* (2016a) provides guidance on importing eviscerated fish of the following species to prevent the international spread of SVCV:

> *Common and ornamental carp (Cyprinus carpio), crucian carp (Carassius carassius), sheatfish or European catfish (Silurus glanis), silver carp (Hypophthalmichthys molitrix), bighead carp (Hypophthamichthys nobilis), grass carp (white amur) (Ctenopharyngodon idella), goldfish (Carassius auratus), orfe (Leuciscus idus), and tench (Tinca tinca). These recommendations also apply to any other susceptible species referred to in the Aquatic Manual when traded internationally.*

Article 10.9.3 of the *Aquatic Code* recommends:

For importation or transit of aquatic animals and aquatic animal products for any purpose from a country, zone or compartment not declared free from spring viraemia of carp:

1. *Competent Authorities should not require any conditions related to SVC, regardless of the SVC status of the exporting country, zone or compartment, when authorising the importation or transit of the following aquatic animals and aquatic animal products from the species referred to in Article 10.9.2 Which are intended for any purpose and which comply with Article 5.4.1:*
    a. *heat sterilised hermetically sealed fish products (i.e. a heat treatment at 121°C for at least 3.6 minutes or equivalent);*
    b. *pasteurised fish products that have been subjected to heat treatment at 90°C for at least ten minutes (or any time/temperature equivalent which has been demonstrated to inactivate SVCV);*

c. *mechanically dried eviscerated fish (i.e. a heat treatment at 100°C for at least 30 minutes or any time/temperature equivalent which has been demonstrated to inactivate SVCV).*

2. *When authorising the importation or transit of aquatic animals and aquatic animal products of a species referred to in Article 10.9.2, other than those referred to in point 1 of Article 10.9.3, Competent Authorities should require the conditions prescribed in Articles 10.9.7 to 10.9.12 relevant to the SVC status of the exporting country, zone or compartment.*

3. *When considering the importation or transit of aquatic animals and aquatic animal products of a species not covered in Article 10.9.2 but which could reasonably be expected to pose a risk of spread of SVC, the Competent Authority should conduct a risk analysis in accordance with the recommendations in Chapter 2.1. The Competent Authority of the exporting country should be informed of the outcome of this assessment.*

Article 10.9.11 of the *Aquatic Code* recommends that:

For importation of aquatic animals and aquatic animal products for retail trade for human consumption from a country, zone or compartment not declared free from spring viraemia of carp

1. *Competent Authorities should not require any conditions related to SVC, regardless of the SVC status of the exporting country, zone or compartment, when authorising the importation or transit of fish fillets or steaks (chilled or frozen) which have been prepared and packaged for retail trade and which comply with Article 5.4.2.*

   Certain assumptions have been made in assessing the safety of the aquatic animal products mentioned above. Member Countries should refer to these assumptions at Article 5.4.2 and consider whether the assumptions apply to their conditions.

   For these commodities Member Countries may wish to consider introducing internal measures to address the risks associated with the commodity being used for any purpose other than for human consumption.

2. *When importing aquatic animals or aquatic animal products, other than those referred to in point 1 above, of species referred to in Article 10.9.2 from a country, zone or compartment not declared free from SVC, the Competent Authority of the importing country should assess the risk and apply appropriate risk mitigation measures.*

Compliance with Articles 10.9.2 and 10.9.11 should eliminate SVCV from the commodity and be a viable risk management option.

SVCV may be transmitted through the waste products associated with transport, storage and processing of the commodity. The requirement that all wash and wastewater discharges be appropriately chemically treated (e.g., with iodophors) before discharge, and that all solid wastes, tissue scraps and offal be disposed of through a recognised trade waste disposal procedure would be viable management options.

28.3.1. Risk management options

Salmon gill poxvirus (SGPV) and related isolates are reported from fish in families Centrarchidae, Cichlidae, Cyprinidae, Esocidae, Percidae, Salmonidae and Siluridae (Table 25), which are considered likely to be present in the commodity. Other families have not been associated with SVCV and related isolates. Therefore species declaration indicating the commodity is not originated from any of the above families should substantially reduce the occurrence of SVCV in the commodity.

For the commodities originated from families associated with SGPV and related isolates, one or a combination of the following additional options could also be considered to effectively manage the risk:

Where country/zone freedom from SVCV is accepted by MPI:

Option 1

Acceptance of country/zone freedom should substantially reduce the occurrence of SVCV, so the commodity may be imported without any further restrictions.

Where country/zone freedom from SVCV is not accepted by MPI or not available:

Option 2

Processing consistent with the conditions of Article 10.9.3 or 10.9.11 of the OIE *Aquatic Code* (OIE 2016a) should eliminate SVCV. When these provisions are met, the commodity could be imported without any further restrictions.

Where the imported commodity is not processed to a state consistent with the conditions of Article 10.9.3 or 10.9.11 of the OIE Aquatic *Code*:

Option 3

Heat treatment (by cooking to at least 60°C for at least 10 minutes) should eliminate SVCV from the commodity. When this provision is met, the commodity could be imported without any further restrictions; or,

Option 4

Further processing (to the skin-off fillet state) should moderately reduce the occurrence of SVCV. When this provision is met, the commodity could be imported without any further restrictions.

28.4. References

Anonymous (2010). Biosecurity (Notifiable Organisms) Order 2010. NZLR 2010/265. New Zealand Government, Wellington, New Zealand, 13 p.

Aquaculture New Zealand (2014). New Zealand's aquaculture industry key facts [Online] Available from: http://www.seafoodnewzealand.org.nz/our-industry/key-facts/ [Accessed 26 September 2014].

Bootsma R. (1976). Studies on two infectious diseases of cultured freshwater fish: rhabdovirus disease of pike fry, *Esox lucius* L. and columnaris disease of carp, *Cyprinus carpio* L. Ph.D dissertation, State University of Utrecht, The Netherlands, 41 p.

Bootsma R, de Kinkelin P, Le Berre M (2006). Transmission experiments with pike fry (*Esox Indus* L.) rhabdovirus. *Journal of Fish Biology* 7(2): 269–276.

Cipriano RC, Bowser PR, Dove A, Goodwin A, Puzach C (2011). Prominent emerging diseases within the United States [Online] Available from: http://www.lsc.usgs.gov/files/Cipriano%20et%20al%202011.pdf [Accessed 22 April 2015].

Clayton JS, Wells RDS (1999). Some issues in risk assessment reports on grass carp and silver carp. Conservation Advisory Science Notes No. 257. Department of Conservation, Wellington, New Zealand, 14 p.

De Kinkelin P, Galimard B, Bootsma R (1973). Isolation and identification of the causative agent of 'red disease' of pike (*Esox lucius* L., 1766). *Nature* 241: 465–467.

Fish and Game (2014). New Zealand Fish and Game Council [Online] Available from: http://www.fishandgame.org.nz/ [Accessed 24 September 2014].

Haenen OLM, Davidse A. (1993). Comparative pathogenicity of two strains of pike fry rhabdovirus and spring viraemia of carp virus for young roach, common carp, grass carp and rainbow trout. *Diseases of Aquatic Organisms* 15: 87–92.

Kahn SA, Beers PT, Findlay VL, Peebles IR, Durham PJ, Wilson DW, Gerrity SE (1999). Import risk analysis on non-viable salmonids and non-salmonid marine finfish. Australian Quarantine and Inspection Service, Department of Agriculture, Fisheries and Forestry, Canberra, Australia, 409 p.

Marsh D, Mkwra L (2013). Review of freshwater non-market value studies. Department of Economics, University of Waikato, Hamilton, New Zealand, 144 p.

NIWA (2014). Grass carp *Ctenopharyngodon idella* (Valenciennes, 1844) [Online] Available from: https://www.niwa.co.nz/freshwater-and-estuaries/nzffd/NIWA-fish-atlas/fish-species/grass_carp [Accessed 12 February 2015].

OIE (2016a). Aquatic Animal Health Code, 19th Edition. World Organization for Animal Health. Paris, France [Online] Available from: http://www.oie.int/international-standard-setting/aquatic-code/access-online/ [Accessed 15 March 2017].

OIE (2016b). Manual of diagnostic tests for aquatic animals. World Organization for Animal Health. Paris, France [Online] Available from: http://www.oie.int/international-standard-setting/aquatic-manual/access-online/ [Accessed 17 March 2017].

Rowe DK (2004). Potential effects of tench (*Tinca tinca*) in New Zealand freshwater ecosystems. NIWA client report HAM 2004-005, prepared for Environment Bay of Plenty, National Institute of Water and Atmospheric Research, Auckland, New Zealand, 28 p.

Schaperclaus W (1992). Fish Diseases, Volume 1. CRC Press, 748 p.

Stone DM, Ahne W, Denham KL, Dixon PF, Lui CT, Sheppard AM, Taylor GR, Way K (2003). Nucleotide sequence analysis of the glycoprotein gene of putative spring viraemia of carp virus and pike fry rhabdovirus isolates reveals four genogroups. *Diseases of Aquatic Organisms* 53: 203–210.

Svetlana J, Ivetic V, Radosavljevic V (2006). *Rhabdovirus carpio* as a causative agent of disease in rainbow trout (*Oncorhynchus mykiss* Walbaum). *Acta Veterinaria (Beograd)* 56: 553–558.

Tubbs L, Lee P, Diggles B, Jones JB, Sheppard M, Sim-Smith C (2007). A review of aquatic diseases of significance to New Zealand. Final research report for project ZBS 2005-17. National Institute of Water and Atmospheric Research, Wellington, New Zealand, 452 p.

29. Viral erythrocytic necrosis

29.1. Hazard identification

29.1.1. Aetiological agent

Viral erythrocytic necrosis (VEN) is caused by erythrocytic necrosis virus (ENV), a member of family Iridoviridae (Emmenegger *et al*. 2014; Purcell *et al.* 2016), possibly representing a new genus (Emmenegger *et al.* 2014; Hick *et al.* 2016). It was originally described from salmonids and herring , as piscine erythrocytic necrosis virus (PEN) (Walker & Sherburne 1977; Winton & Hershberger 2014).

Several morphologically distinct strains of ENV occur among the various host species of fish and geographical locations where it has been recovered (Smail 1982). It has not been isolated in cell culture and the genetic relationships between these strains remain unknown (Winton & Hershberger 2014; Hick *et al.* 2016).

29.1.2. OIE status

Infection with viral erythrocytic necrosis (VEN) is not an OIE-listed disease (OIE 2016).

29.1.3. New Zealand status

VEN is not reported from New Zealand. Other Iridoviridae including OIE-listed diseases red sea bream iridovirus (RSIV) and infectious spleen and kidney necrosis virus (ISKNV) have never been reported from New Zealand (Tubbs *et al.* 2007) or found in subsequent monitoring (B. Jones, *pers. comm.* 2015). Infection with VEN is not a notifiable disease in New Zealand (Anon. 2016).

29.1.4. Epidemiology

VEN is the agent of erythrocytic necrosis, which affects elasmobranch and teleost fish in the Atlantic and Pacific Oceans, as well as Spain, Japan and Australia (Hick *et al*. 2016). It has a wide host range (Table 26), being reported from over 10 families and 18 species of fish (MacMillan *et al*. 1980; Dannevig & Thorud 1999; Davies *et al.* 2009; Winton & Hershberger 2014; Hick *et al*. 2016).

Table 26. Main Families and Species of Fish Considered Susceptible to Viral Erythrocytic Necrosis (VEN)Family	**Susceptible Species**
Balistidae	White-banded triggerfish (*Rhinecanthus aculeatus*)
Carangidae	Yellowtail kingfish (*Seriola lalandi*)
Clupeidae	Alewife (*Alosa pseudoharengus*) Atlantic herring (*Clupea harengus*), Pacific herring (*Clupea pallasii*)

Gadidae	Atlantic cod (*Gadus morhua*)
Gobiidae	Rock goby (*Mauligobius maderensis*)
Liparidae	Atlantic seasnail (*Liparis atlanticus*)
Salmonidae	Atlantic salmon (*Salmo salar*), brown trout (*S. trutta*), chum salmon (*Oncorhynchus keta*), Chinook salmon (*O. tshawytscha*), pink salmon (*O. gorbuscha*), rainbow trout (*O. mykiss*), sockeye salmon (*O. nerka*)
Triakidae	Lesser spotted dogfish (*Scyliorhinus canicula*), dusky smooth-hound (*Mustelus canis*)

VEN affects both wild and farmed fish, causing systemic disease through haemolysis of the red blood cells. Infected cells often contain large intra-cytoplasmic cellular inclusions (McAllister & Stoskopf, 1993; Hick *et al.* 2016). Pathogenicity is highly variable, depending on the strain of the pathogen, route of infection, intensity of exposure, stage of infection and susceptibility of the host (Reno *et al.* 1985; Kahn *et al.* 1999; Winton & Hershberger 2014).

Infection may affect up to 90% of red blood cells in salmonids during epizootics, but sub-clinical infection may be maintained for extended periods and VEN itself rarely causes high morbidity or mortality (Reno *et al*. 1985; Kahn *et al.* 1999). Where the host becomes weakened through anaemia, the onset of clinical disease may be associated with co-infection from other opportunistic pathogens (Kahn *et al.* 1999; Hershberger *et al.* 2006; Glenn *et al.* 2012).

Disease transmission is horizontal, though the water column (Hershberger *et al.* 2006), although vertical transmission may also occur (Rohovec & Amandi 1981). VEN is also reported from an isopod, indicating vector-borne transmission may occur (Davies *et al.* 2009). Cross-species transmission has also been demonstrated experimentally among teleost host species (Winton & Hershberger 2014).

No treatment or vaccine is available for ENV (Winton & Hershberger 2014).

29.2. Risk assessment

29.2.1. Entry assessment

VEN causes systemic disease of red blood cells in a wide range of wild and farmed fish (Winton & Hershberger 2014; Hick *et al*. 2016). Infection may be high in farmed fish, but clinically infected fish with signs of anaemia should not pass visual inspection and be present in the commodity (Kahn *et al*. 1999). VEN present at sub-clinical levels of infection in wild teleost fish stocks may however, be retained in the commodity.

Elasmobranchs processed for human consumption are “trunked” immediately after capture, to prevent ammonia contamination of the blood. This includes evisceration, beheading, de-gilling de-finning and de-tailing, followed by pressure washing of the trunk with seawater (Musick 2005). It is unlikely that VEN would be present in sufficient quantity in the commodity to establish and maintain infection of a suitable host species in New Zealand through the commodity.

The likelihood of entry through elasmobranch fish through the commodity is assessed as negligible. The likelihood of entry of teleost fish is assessed as non-negligible.

29.2.2. Exposure assessment

VEN may be present at sub-clinical levels of infection in teleost fish, but rarely causes high morbidity or mortality (Reno *et al*. 1985; Kahn *et al.* 1999). The likelihood of exposure from sub-

clinical infection through teleost fish in the commodity is assessed as negligible, thus no further assessment needs to be carried out.

29.3. References

Anonymous (2016). Biosecurity (Notifiable Organisms) Order 2016. NZLR 2016/73. New Zealand Government, Wellington, New Zealand, 13 p.

Dannevig BH, Thorud KE (1999). Other viral diseases and agents of cold-water fish: infectious salmon anaemia, pancreas disease and viral erythrocytic necrosis, pp 164–175 *In:* Woo PTK, Bruno DW (*Eds.*). Fish diseases and disorders, volume 3. CABI International, New York. United States of America. 930 p.

Davies AJ, Curtis L, Grutter AS, Smit NJ (2009). Suspected viral erythrocytic necrosis (VEN) in a juvenile blackbar triggerfish *Rhinecanthus aculeatus*, from Lizard Island, Great Barrier Reef, Australia. *Marine Biodiversity Records* 2, e149.

Emmenegger EJ, Glenn JA, Winton JR, Batts WN, Gregg JL, Hershberger PK (2014). Molecular identification of erythrocytic necrosis virus (ENV) from the blood of Pacific herring (*Clupea pallasii*). *Veterinary Microbiology* 174:16–26.

Glenn JA, Emmenegger EJ, Grady CA, Roon SR, Gregg JL, Conway CM, Winton JR, Hershberger PK (2012). Kinetics of viral load and erythrocytic inclusion body formation in Pacific herring artificially infected with erythrocytic necrosis virus. *Journal of Aquatic Animal Health* 24:195–200.

Hershberger P, Hart A, Gregg J, Elder N, Winton J (2006). Dynamics of viral hemorrhagic septicemia, viral erythrocytic necrosis and ichthyophoniasis in confined Pacific herring *Clupea pallasii. Diseases of Aquatic Organisms* 70: 201–208.

Hick P, Becker J, Whittington R (2016). Iridoviruses of fish. pp 127–152 *In:* Kibenge FSB, Godoy MG (*Eds.*). *Aquaculture Virology*, Academic Press, London, United Kingdom, 549 p.

Kahn RA Newman MW (1982). Blood parasites from fish of the Gulf of Maine to Cape Hatteras, Northwest Atlantic Ocean, with notes on the distribution of fish hematozoa. *Canadian Journal of Zoology* 60(3): 396–402 [Online] Available from: https://doi.org/10.1139/z82-054 [Accessed 6 October 2017].

MacMillan JR, Mulcahy D, Landolt M (1980). Viral erythrocytic necrosis: some physiological consequences of infection in chum salmon (*Oncorhynchus keta*). *Canadian Journal of Fisheries and Aquatic Sciences* 37: 799–804.

McAllister PE, Stoskopf MK (1993). Shark viruses, pp 780−782 *In:* Stoskopf MK (*Ed.*). Fish medicine. WB Saunders, Philadelphia, United States of America, 882 p.

Musick JA (2005). Shark utilisation pp 243–251 *In:* Musick JA, Bonfil R (*Eds.*). Management techniques for elasmobranch fisheries. *FAO Fisheries Technical Paper* 474. Food and Agriculture Organisation of the United Nations (FAO). Rome, Italy. 251 p.

Purcell MK, Pearman-Gillman S, Thompson RL, Gregg JL, Hart LM, Winton JR, Emmenegger EJ, Hershberger PK (2016). Identification of the major capsid protein of erythrocytic necrosis virus (ENV) and development of quantitative real-time PCR assays for quantification of ENV DNA. *Journal of Veterinary Diagnostic Investigation* 28(4):382–391 [Online] Available from: http://www.doi: 10.1177/1040638716646411. Epub 2016 May 6. [Accessed 28 April 2018].

Reno PWD, Serreze V, Hellyer SK, Nicholson BL (1985). Hematological and physiological effects of viral erythrocytic necrosis (VEN) in Atlantic cod and herring. *Fish Pathology* 20: 353–360.

Rohovec JS, Amandi A (1981). Incidence of viral erythrocytic necrosis among hatchery reared salmonids in Oregon. *Fish Pathology* 15:135–141.

Smail DA (1982). Viral erythrocytic necrosis in fish: a review. *Proceedings of the Royal Society of Edinburgh* 81B: 169–176.

Tubbs L, Lee P, Diggles B, Jones JB, Sheppard M, Sim-Smith C (2007). A review of aquatic diseases of significance to New Zealand. Final research report for project ZBS 2005-17. National Institute of Water and Atmospheric Research, Wellington, New Zealand, 452 p.

Walker R, Sherburne SW (1977). Piscine erythrocytic necrosis virus in Atlantic cod *Gadus morhua*, and other fish: ultrastructure and distribution. *Journal of the Fisheries Research Board of Canada* 34: 1188–1195.

Winton JR, Hershberger PK (2014). Viral Erythrocytic Necrosis. Chapter 2.2.7. The Blue Book 2014. Fish Health Section, American Fisheries Society [Online] Available from: http://afs-fhs.org/perch/resources/14069231372.2.7ven2014.pdf [Accessed 6 October 2017].

30. Viral haemorrhagic septicaemia virus

30.1. Hazard identification

30.1.1. Aetiological agent

Viral haemorrhagic septicaemia virus (VHSV), a non-segmented, negative sense, single stranded RNA virus, tentatively classified in the Genus *Vesiculovirus*, within the Family Rhabdoviridae (OIE 2016a). Viral haemorrhagic septicaemia (VHS) is an acute haemorrhagic viral disease of marine and freshwater fish, which is considered the most important viral disease in freshwater salmonid farming (OIE 2016b). Other names include Egtved disease, infectious necrotic swelling, abdominal ascites of trout and pernicious anaemia of trout (Tubbs *et al.* 2007). While first isolated in German rainbow trout (*Oncorhynchus mykiss*) aquaculture (Einer-Jensen *et al.* 2004), VHSV is now reported from both marine and freshwater fish hosts (Einer-Jensen *et al*. 2004; Skall *et al.* 2005). VHSV isolates from European freshwater aquaculture appear to have originated from northern Pacific and Atlantic oceanic waters and been transferred through marine fish used as feedstock for freshwater fish aquaculture (Hedrick *et al.* 2003).

Four genotypes of VHSV are recognised from different environments and geographical areas, each with several sub-lineages of varying pathogenicity (OIE 2016b). *Genotype* I isolates are divided into five sub-groups (Ia–Ie) affecting a wide range of marine and freshwater hosts. *Genotype* Ia isolates are mainly reported from European wild and farmed freshwater fisheries including rainbow trout (*Oncorhynchus mykiss*), while *Genotype* Ib mainly occurs in wild and farmed marine fish from the North Sea, Baltic Sea, the English Channel and coastal Norwegian waters (OIE 2016a).

Genotype II isolates are only reported from marine fish from the Baltic Sea (OIE 2016a).

Genotype III isolates are generally reported from marine fish hosts in the North Atlantic, the North Sea and Norwegian coastal waters, but also affect farmed Norwegian rainbow trout (*O. mykiss*) (Dale *et al.* 2009; OIE 2016a).

Genotype IV isolates are divided into two sub-groups, where *Genotype* IVa isolates are widely reported in both wild and farmed fish, in North American, Japanese and Korean waters. *Genotype* IVb isolates are reported from a wide range of predator and forager fish, mainly from the Laurentian Great Lakes and associated watersheds (Amos *et al.* 2010; B. Jones, *pers. comm.* 2015), but have not been detected in farmed fish (OIE 2016a).

30.1.2. OIE status

Infection with VHSV is listed by the OIE as a notifiable disease (OIE 2016a).

30.1.3. New Zealand status

VHSV has not been reported from the Southern Hemisphere (Stone *et al.* 1997b; DAFF 2005, 2014; B. Jones, *pers. comm.* 2015) and it has been assumed exotic. Infection with VHSV is a notifiable disease in New Zealand (Anon. 2016).

30.1.4. Epidemiology

VHS is an OIE-listed and economically significant viral disease affecting marine and freshwater fish (OIE 2016a). It is considered one of the most important diseases affecting rainbow trout (*O. mykiss*) farming in Europe and Asia (Skall *et al.* 2005), as well as channel catfish (*Ictalurus punctatus*) and rainbow trout farming in North America (Skall *et al.* 2005; Bain *et al.* 2010). VHSV is a management concern in recreational freshwater salmonid fisheries in the United States, where movement restrictions have been imposed on several baitfish species as a disease control measure (Skall *et al*. 2005; Bain *et al.* 2010; USDA 2015).

VHS is widely reported in wild fish populations in temperate fresh waters of the Northern Hemisphere including Continental Europe (excluding Portugal, Greece, Norway, Finland, and Sweden) and North America (Canada and the United States). It has been reported in marine hosts from the North Atlantic and Baltic Sea, the Pacific Ocean (from Alaska to California), Japan, Canada, Iran, Kuwait and South Korea (Hedrick *et al*. 2003; Tubbs *et al*. 2007; Amos *et al*. 2010; Spikler 2010; OIE 2016b. It has never been reported from the Southern Hemisphere (DAFF 2005; 2014; B. Jones, *pers. comm*. 2015; OIE 2016b).

VHSV is reported from a wide range of marine and freshwater hosts (Table 27) and all temperate marine species may be susceptible to VHSV under suitable conditions ((Stone *et al.* 1997a; Skall *et al.* 2005; Anon. 2007; EFSA 2008; Bain *et al.* 2010; Kim & Faisal 2011; Kim *et al.* 2013; Anon. 2014; Ogut & Altuntas 2014; OIE 2016b; Anon. 2017).

VHSV is essentially a cold-water disease and viral replication is limited when temperatures exceed 15°C. (Wolf 1988). Mortalities vary widely, from 20 to 100%, where high mortality rates are associated with cooler waters (less than 10°C), while both mortality and morbidity are reduced in warmer waters. Clinical signs of disease are not apparent in waters warmer than 15°C (Munday 2002).

VHSV isolates from European marine fishes are generally considered of low virulence for Atlantic salmon (*Salmo salar*), although Genotype IIIb is pathogenic to rainbow trout (O. mykiss) (Dale *et al.* 2009; OIE 2016b). Other isolates from wild marine fish are pathogenic to farmed turbot (*Scophthalmus maximus*), while isolates from Pacific herring (*Clupea pallasii*) and pilchard (*Sardinops* spp.) are pathogenic to farmed *Salmo salar* in Pacific Ocean aquaculture (Tubbs *et al.* 2007). Caution is advised before assuming a species is resistant to infection (Skall *et al.* 2005a; OIE 2016b).

The outcome of infection with VHSV varies depending upon the host species, and may be acute, nervous, or chronic, while VHSV may also be present in carrier fish (OIE 2016b). Infection may also occur with no external signs where water temperatures exceed 5°C, indicative of a temperature-dependent immune response (Munday 2002). Surviving fish are resistant to re-infection (DAFF 2005).

In acute infection, viral particles may be present in the endothelial cells of the blood capillaries, leucocytes, the haematopoietic tissues of the spleen and liver, as well as in heart and kidney tissues. Infection, which commonly results in extensive necrosis of the musculature of the head and mid-section of the body, is associated with external signs including exophthalmia, together with widespread haemorrhage of the skin, the bases of the fins and in the eyes (DAFF 2005). In the nervous form, viral particles are present in brain and neural tissue (DAFF 2005). Chronic infection may progress with no external clinical signs of disease, but viral particles may be present in all the major internal organs.

Table 27. Families and Species of Fish Considered Susceptible to VHSV

Family	Susceptible Species
Ammodytidae	Pacific sand eel (*Ammodytes personatus*), Pacific sand lance (*Ammodytes hexapterus*), sand eel (*Ammodytes* spp.)
Anguillidae	European eel (*Anguilla anguilla*), American eel (*A. rostrata*)
Anoplopomatidae	Sablefish/blackcod (*Anoplopoma fimbria*)
Argentinidae	Lesser argentine (*Argentina sphyraena*)
Aulorhynchidae	Tubesnout (*Aulorhynchus flavidus*)
Belonidae	Garfish (*Belone belone*)
Carangidae	Mediterranean horse mackerel (*Trachurus mediterraneus*), Japanese amberjack (*Seriola quinqueradiata*)
Catostomidae	River redhorse (*Moxostoma carinatum*), shorthead redhorse (*M. macrolepidotum*), silver redhorse (*M. anisurum*), white sucker (*Catostomus commersonii*)
Centrarchidae	Black crappie (*Pomoxis nigromaculatus*), bluegill (*Lepomis macrochirus*), largemouth bass (*Micropterus salmoides*), pumpkinseed (*L. gibbosus*), rock bass (*Ambloplites rupestris*), smallmouth bass (*M. dolomieu*)
Clupeidae	Atlantic herring (*Clupea harengus*), Pacific herring (*Clupea pallasii*), South American pilchard (*Sardinops sagax*), European pilchard (*Sardina pilchardus*), sprat (*Sprattus sprattus*), gizzard shad (*Dorosoma cepedianum*), pontic shad (*Alosa immaculata*)
Cyprinidae	Bluntnose minnow (*Pimephales notatus*), emerald shiner (*Notropis atherinoides*), fathead minnow (*Pimephales promelas*), Iberian nace (*Pseudochondrostoma polylepis*), Spanish barbel (*Luciobarbus graellsii*), spottail shiner (*N. hudsonius*), zebra danio (*Danio rerio*)
Embiotocidae	Shiner perch (*Cymatogaster aggregata*)
Engraulidae	Anchovy (*Engraulis encrasicolus*)
Esocidae	Pike (*Esox lucius*), muskellunge (*E. masquinongy*)
Fundulidae	Mummichog (*Fundulus heteroclitus*)
Gadidae	Alaska pollock (*Theragra chalcogramma*), Atlantic cod (*Gadus morhua*); blue whiting (*Micromesistius poutassou*), haddock (*Melanogrammus aeglefinus*), Norway pout (*Trisopterus esmarkii*), Pacific cod (*Gadus macrocephalus*), Pacific tomcod (*Microgadus proximus*), poor cod (*Trisopterus minutus*), whiting (*Merlangius merlangus*)
Gobiidae	Round goby (*Neogobius melanostomus*)
Ictaluridae	Brown bullhead (*Ameiurus nebulosus*), channel catfish (*Ameiurus punctatus*)
Lotidae	Burbot (*Lota lota*), four bearded rockling (*Enchelyopus cimbrius*), rockling (*Gaidropsarus* spp.), fivebeard rockling (*Ciliata mustela*)
Merlucciidae	North Pacific hake (*Merluccius productus*)
Moronidae	European sea bass (*Dicentrarchus labrax*), striped bass (*Morone saxitalis*), white bass (*M, chrysops*), white perch (*M. americana*)
Mugilidae	Grey mullet (*Mugil cephalus*)
Mullidae	Red mullet (*Mullus barbatus*)
Ophidiidae	Armoured cusk (*Hoplobrotula armata*)
Osmeridae	Eulachon (*Thaleichthys pacificus*), surf smelt (*Hypomesus pretiosus*)
Paralichthyidae	Bastard halibut (*Paralichthys olivaceus*)
Percidae	European perch (*Perca fluviatilis*), yellow perch (*Perca flavescens*), walleye (*Sander vitreus*)
Percopsidae	Trout-perch (*Percopsis omiscomaycus*)
Pleuronectidae	Atlantic halibut (*Hippoglossus hippoglossus*), blackfin flounder (*Glyptocephalus stelleri*), dab (*Limanda limanda*), European flounder (*Platichthys flesus*), English sole (*Parophrys vetulus*), Greenland halibut (*Reinhardtius hippoglossoides*), marbled flounder (*Pseudopleuronectes yokohame*), plaice (*Pleuronectes platessa*)
Salmonidae	Arctic char (*Salvelinus alpinus*), Arctic grayling (*Thymallus thymallus*), Atlantic salmon (*Salmo salar*), brook trout (*Salvelinus fontinalis*), brown trout (*S. trutta*), coho salmon (*Oncorhynchus kisutch*), Chinook salmon (*O. tshawytscha*), chum salmon (*O. keta*), golden trout (*O. aguabonita*), lake trout (*S. namaycush*), rainbow trout (*O. mykiss*), pink salmon (*Onchorhynchus gorbuscha*), sockeye salmon (*O, nerka*), whitefish (*Coregonus* spp.), lake whitefish (*Coregonus clupeaformis*), *Oncorhynchus* spp. hybrids, *Salvelinus* spp. hybrids
Sciaenidae	Freshwater drum (*Aplodinotus grunniens*), yellow croaker (*Larimichthys polyactis*),
Scombridae	Pacific mackerel (*Scomber japonicus*)
Scophthalmidae	Turbot (*Scophthalmus maximus*)
Scorpaenidae	Black scorpionfish (*Scorpaena porcus*)

Sebastidae	Black rockfish (*Sebastes inerrmis*), Korean rockfish (*Sebastes schlegelii*)
Serranidae	Hongkong grouper (*Epinephelus akaara*)
Sillaginidiae	Whiting (*Sillago ciliata*)
Soleidae	Senegalese sole (*Solea sengalensis*)
Sparidae	Black seabream (*Acanthopagrus schlegelii*), gilthead seabream (*Sparus aurata*), red seabream (*Pagrus major*), yellowback sea bream (*Dentex tumifrons*)
Stromateidae	Silver pomfret (*Pampus argenteus*)
Trichiuridae	Largehead hairtail (*Trichiurus lepturus*)
Uranoscopidae	Atlantic stargazer (*Uranoscopus scaber*)

The minimum infectious dose for VHSV is low ($10^{1.5}$ PFU mL^{-1} for herring *Clupea pallasii*), but infectivity varies both according to host age, host species, and specific virus isolate, and is also strongly influenced by ambient temperature (DAFF 2005).

Viral transmission is horizontal through the water column. Initial infection usually occurs through the epithelial cells of the gills, viscera or skin (Tubbs *et al*. 2007). The course of infection is variable, as viral replication may occur at the site of entry, or be delayed until viral particles reach endothelial cells of the vascular system, spleen and brain (DAFF 2005). Viral particles are shed in the urine, faeces, mucous and sexual fluids of host fish, as well as from the skin ulcers and necrotic gill lesions present in clinically infected fish. While the rate of shedding from carrier fish is unknown, this is likely to be maintained for months or years (DAFF 2005). Although VHSV may survive on the surface of fish eggs in hatcheries, vertical transmission is yet unproven (OIE 2016b).

VHSV appears unaffected by common storage and processing methods and remains viable within processed fish tissues. Homogenates derived from the viscera, brain, gills and musculature of infected rainbow trout carcases cause high mortality of rainbow trout fry by waterborne infection (Oidtmann *et al.* 2011).

Environmental resistance varies between isolates (DAFF 2005), but VHSV is considered resistant to ether, heat and acid (to pH 3) (OIE 2016b). It is highly resistant to freezing, remaining viable for several years at -20°C, and is resistant to the effect of freeze-thaw cycles (Wolf 1988).

VHSV is environmentally stable in both marine and fresh waters. It remains viable for up to 40 hours in filtered sea water, up to 6 hours in blood-water discharged as factory effluent (Afonso *et al.* 2012), and up to 96 hours in foetal bovine solution (DAFF 2005). VHSV remains viable in fresh water for up to 14 days (Grant 2007), but may persist for up to 10 days in sediments at 4°C (Tubbs *et al*. 2007).

VHSV may be spread from one site to another, through ballast water discharges from marine transport (Bain *et al*. 2010), or through regurgitation of food by piscivorous birds, but it is unlikely to remain viable after passage through the avian digestive system (DAFF 2005).

VHSV is inactivated by cooking (to 100°C for 30 minutes), formalin (3% for 5 minutes), drying, iodophor disinfectants (100 ppm active iodine for 5 minutes) and sodium hydroxide (2% for 10 minutes), as well as by strong acid (pH 2.5 for 10 minutes) or alkali (pH 12.2 for 2 hours) treatments (Tubbs *et al*. 2007). While UV treatment is effective, high doses (to 7.87 mJ cm^{-2}) are required to ensure inactivation in blood-water discharge (Afonso *et al*. 2012).

VHSV is readily transferred in baitfish, and the use of whole menhaden (*Brevoortia tyrannus*) as feedstock in Australian aquaculture is restricted to waters where temperatures exceed 14°C (Diggles 2007).

No commercially available vaccine for VHSV is available (Lorenzen & LaPatra 2005; OIE 2016b).

Potential hosts in New Zealand in fresh water include all salmonids (*Oncorhynchus* spp., *Salmo* spp., *Salvelinus* spp.), flatfish (Order Pleuronectiformes) and eel (*Anguilla* spp.), while marine species include pilchard (*Sardinops sagax*), sprat (*Sprattus sprattus*) and kingfish (*Seriola lalandi*) (Tubbs *et al.* 2007), and potentially all marine fish (Stone *et al.* 1997b).

30.2. Risk assessment

30.2.1. Entry assessment

VHSV is an OIE notifiable disease mainly reported from juvenile fish in freshwater aquaculture, but may potentially infect a wide range of juveniles, as well as adult farmed and wild fish (Table 31), from both fresh and marine waters (OIE 2016b). Chronic or acute infection is usually accompanied by visible external signs including pale gills, exophthalmia, darkened skin coloration and bleeding at the base of the fins (DAFF 2005). These fish would be unlikely to pass visual inspection and should not be present in the commodity.

Clinical infection may also progress through all three pathways (acute, nervous or chronic) with no external signs. These carrier fish may pass inspection and be present in the commodity.

While evisceration would significantly reduce viral titre, VHSV is commonly associated with skin, gill and muscle tissues in acute and chronic infection and is concentrated in the brain and neural tissues in nervous tissue infection (OIE 2016b). As these tissues are retained in the commodity, the likelihood of VHSV entry is assessed as non-negligible.

30.2.2. Exposure assessment

To establish infection in New Zealand, infected eviscerated product would have to become available for consumption by a susceptible fish host, in sufficient quantity and duration (Kahn *et al*. 1999). VHSV is likely to remain viable in the offal and blood-water discharge from commercial processing of eviscerated fish, as well as through trimming scraps of skeletal muscle (Oidtmann *et al.* 2011). VHSV is environmentally stable within the range of conditions likely to be encountered in commercial fish processing and storage (OIE 2016b). It is resistant to freezing and remains viable in fresh water and sediment for up to 10 days, or for up to 96 hours in sea water if contaminated with organic material (DAFF 2005).

VHSV infection may be initiated at low viral titre (OIE 2016b). Infected tissue remaining in the gills and brain tissues of eviscerated fish may be sufficient to initiate infection, given the relatively low titre required to initiate infection (Oidtmann *et al.* 2011; OIE 2016b).

It is resistant to high temperatures and mild acid conditions and is likely to remain viable in fish wastes discarded to landfill (OIE 2016a). It is unlikely to be transferred by scavenging birds, as its viability after transfer is unknown (DAFF 2005).

The likelihood of exposure to VHSV is assessed as non-negligible.

30.2.3. Consequence assessment

VHSV is an OIE-listed (OIE 2016a) and New Zealand-notifiable disease (Anon. 2016). Freshwater aquaculture in New Zealand is highly dependent on clean uncontaminated water supplies (Sim-Smith *et al.* 2014), so the introduction of disease into watersheds would have a major economic effect upon fish aquaculture, particularly as Chinook salmon (*O. tshawytscha*) is considered susceptible to VHSV (OIE 2016a). This commodity was valued at $63 million in export earnings in 2011 (Aquaculture New Zealand 2014). VHSV also affects brown trout and rainbow trout, so an outbreak would be likely to affect recreational and tourist trout and salmon fishing, as well as incur significant social and environmental costs associated with other domestic fisheries (Fish & Game 2014). While the dollar value of New Zealand's freshwater fisheries is not fully known, the Lake Taupo trout fishery alone is valued at $70 to 80 million per annum (Marsh & Mkwra 2013).

VHSV has a wide host range in marine and fresh water (Table 28), so its introduction would be likely to have a significant effect upon other native fish species, with extensive political, economic and social consequences (Fish & Game 2014). Given that the virulence of VHSV varies widely between isolates and among host species, the potential effect cannot be determined in advance. The consequences of establishment of VHSV are therefore assessed as non-negligible.

30.2.4. Risk estimation

Since the entry, exposure and consequence assessments for VHSV are non-negligible, the risk estimate is non-negligible. Therefore, VHSV is assessed to be a risk in the commodity and risk management measures may be justified.

30.3. Risk management

VHS is a notifiable disease which affects a wide range of wild and farmed marine and freshwater fish hosts (Table 27). The *Aquatic Code* (OIE 2016a) provides guidance to prevent the international spread of VHSC. Article 10.10.2 of the *Aquatic Code* lists the species recognised to require risk management measures and they apply to:

> *Rainbow trout (Oncorhynchus mykiss), brown trout (Salmo trutta), grayling (Thymallus thymallus), white fish (Coregonus spp.), pike (Esox lucius), turbot (Scophthalmus maximus), herring and sprat (Clupea spp.), Pacific salmon (Oncorhynchus spp.), Atlantic cod (Gadus morhua), Pacific cod (G. macrocephalus), haddock (G. aeglefinus) and rockling (Onos mustelus). These recommendations also apply to any other susceptible species referred to in the Aquatic Manual when traded internationally.*

Article 10.10.3 of the *Aquatic Code* recommends that:

For importation or transit of aquatic animals and aquatic animal products for any purpose from a country, zone or compartment not declared free from viral haemorrhagic septicaemia:

1. *Competent Authorities should not require any conditions related to VHS, regardless of the VHS status of the exporting country, zone or compartment, when authorising the importation or transit of the following aquatic animals and aquatic animal products from the species referred to in Article 10.10.2 which are intended for any purpose and which comply with Article 5.4.1:*
    a. *heat sterilised, hermetically sealed fish products (i.e. a heat treatment at 121°C for at least 3.6 minutes or any time/temperature equivalent);*

b. *pasteurised fish products that have been subjected to a heat treatment at 90°C for at least ten minutes (or to any time/temperature equivalent which has been demonstrated to inactivate VHSV);*
c. *mechanically dried, eviscerated fish (i.e. a heat treatment at 100˚C for at least 30 minutes or any time/temperature equivalent which has been demonstrated to inactivate VHSV);*
d. *naturally dried, eviscerated fish (i.e. sun-dried or wind-dried).*

2. *When authorising the importation or transit of aquatic animals and aquatic animal products of species referred to in Article 10.10.2, other than those referred to in point 1 of Article 10.10.3, Competent Authorities should require the conditions prescribed in Articles 10.10.7 to 10.10.13 relevant to the VHS status of the exporting country, zone or compartment.*
3. *When considering the importation or transit of aquatic animals and aquatic animal products of species not covered in Article 10.10.2 but which could reasonably be expected to pose a risk of spread of VHS, the Competent Authority should conduct a risk analysis in accordance with the recommendations in Chapter 2.1. The Competent Authority of the exporting country should be informed of the outcome of this assessment.*

Article 10.10.11 of the *Aquatic Code* recommends that:

For importation of aquatic animals and aquatic animal products for retail trade for human consumption from a country, zone or compartment not declared free from viral haemorrhagic septicaemia:

- *Competent Authorities should not require any conditions related to VHS, regardless of the VHS status of the exporting country, zone or compartment, when authorising the importation or transit of fish fillets or steaks (chilled or frozen) which have been prepared and packaged for retail trade and which comply with Article 5.4.2.*

  Certain assumptions have been made in assessing the safety of the aquatic animal products mentioned above. Member Countries should refer to these assumptions at Article 5.4.2 and consider whether the assumptions apply to their conditions.

  For these commodities Member Countries may wish to consider introducing internal measures to address the risks associated with the commodity being used for any purpose other than for human consumption.

- *When importing aquatic animals or aquatic animal products, other than those referred to in point 1 above, of species referred to in Article 10.10.2 from a country, zone or compartment not declared free from VHS, the Competent Authority of the importing country should assess the risk and apply appropriate risk mitigation measures.*

Compliance with Articles 10.10.3 and 10.10.11 should eliminate VHSV from the commodity and be a viable risk management option. VHSV is associated with over 40 host fish families (Table 27), which may be present in the commodity. Species declaration should substantially reduce the occurrence of VHSV in the commodity and be a viable risk management option. Restriction of the commodity to wild-caught fish (not from aquaculture) would have little, or no effect on pathogen load and is not a viable risk management option.

VHSV has been widely reported from Continental Europe and from marine waters of the North Atlantic, Baltic Sea and the Pacific Ocean (from Alaska to California), as well as from Japan, Canada, Iran, Kuwait, South Korea, and the continental United States. It has not been reported from the Southern Hemisphere (DAFF 2005). As VHS is an OIE-listed disease, country freedom is well defined. Approval of country/zone freedom through the MPI Country Approval Procedures should substantially reduce the occurrence of VHSV in the commodity. Approval of a declaration of country/zone freedom by MPI is a viable risk management option.

VHSV is commonly associated with skin, gill and muscle tissues in acute and chronic infection and concentrated in the brain and neural tissues in nervous tissue infection. Removal of the gills, or of the head and gills, should slightly reduce the pathogen load of VHSV. Further processing of the commodity to the skin-off fillet state would moderately reduce the occurrence of VHSV in the commodity and be a viable risk management option.

VHSV is inactivated by cooking (to 100°C for 30 minutes) (OIE 2018a), which would eliminate the pathogen from the commodity.

VHSV may be transmitted through the waste products associated with transport, storage and processing of the commodity. The requirement that all wash and wastewater discharges be appropriately chemically treated (e.g., with iodophors) before discharge, and that all solid wastes, tissue scraps and offal be disposed of through a recognised trade waste disposal procedure, would be viable management options.

30.3.1. Risk management options

Viral haemorrhagic septicaemia virus (VHSV) is reported from fish in families Ammodytidae, Anguillidae, Anoplopomatidae, Argentinidae, Aulorhynchidae, Belonidae, Carangidae, Catostomidae, Centrarchidae, Clupeidae, Cyprinidae, Embiotocidae, Engraulidae, Esocidae, Fundulidae, Gadidae, Gobiidae, Ictaluridae, Lotidae, Merlucciidae, Moronidae, Mugilidae, Mullidae, Ophidiidae, Osmeridae, Paralichthyidae, Percidae, Percopsidae, Pleuronectidae, Salmonidae, Sciaenidae, Scombridae, Scorpaenidae, Sebastidae, Serranidae, Sillaginidae, Soleidae, Sparidae, Stromateidae, Trichiuridae and Uranoscopidae (Table 27). These families are considered likely to be present in the commodity. Other families have not been associated with VHSV. Therefore species declaration indicating the commodity is not originated from any of the above families should substantially reduce the occurrence of VHSV in the commodity.

For the commodities originated from families associated with VHSV, one or a combination of the following additional options could also be considered to effectively manage the risk.

Where country/zone freedom from VHS is accepted by MPI:

Option 1

Acceptance of country/zone freedom by MPI should substantially reduce the occurrence of VHSV, so the commodity may be imported without any further restrictions.

Where country/zone freedom from VHSV is not accepted by MPI or not available:

Option 2

Processing consistent with the conditions of Article 10.10.3 and 10.10.11 of the OIE Aquatic *Code* (OIE 2016a) should eliminate VHSV. When these provisions are met, the commodity could be imported without any further restrictions.

Where the imported commodity is not consistent with Article 10.10.3 or 10.10.11 of the OIE Aquatic *Code*:

Option 3

Further processing (to the skin-off fillet state) should moderately reduce the occurrence of VHSV in the commodity. When this provision is met, the commodity could be imported without any further restrictions.

30.4. References

Afonso LOB, Richmond Z, Eaves AA, Richard J, Hawley LM, Garver KA (2012). Use of ultraviolet C radiation (UVC) to inactivate infectious hematopoietic necrosis virus (IHNV) and viral haemorrhagic septicaemia virus (VHSV) in fish processing plant effluent. *Journal of Aquaculture Research and Development* 3: 120 doi: 10.4172/2155-9546.1000120 [Online] Available from: http://omicsonline.org/use-of-ultraviolet-c-uvc-radiation-to-inactivate-infectious-hematopoietic-necrosis-virus-ihnv-and-viral-hemorrhagic-septicemia-virus-vhsv-in%20fish-processing-plant-effluent-2155-9546.1000120.php?aid=3943 [Accessed 24 April 2015].

Amos KH, Bakal RS, Barker AA, Blair MJ, Bouchard DA, Bruneau N, Bowser PR, Egrie PG, Ellis SK, Faisal M, Garver KA, Giray C, Goodwin AE, Gustafson LL, House NL, Karreman GA, Kebus MJ, Klotins KC, LaPatra SE, Marty GD, Merrill PD, Noyes AD, Olesen NJ, Saksida SM, Snow M, St-Hilaire S, Tomlinson SM, Uhland SC, Vennerstrom P, Wagner BA, Warg JV, Whelan GE, Winton JR (2010). Viral haemorrhagic septicaemia virus (VHSV IVb) risk factors and association measures derived by expert panel. *Preventative Veterinary Medicine* 94: 128–139.

Anonymous (2007). Viral Hemorrhagic Septicemia. Information sheet series. Centre for Food Security and Public Health, Iowa State University, Ames, Iowa, United States of America [Online] Available from: http://www.cfsph.iastate.edu/Factsheets/pdfs/viral_hemorrhagic_septicemia.pdf [Accessed 31 July 2017].

Anonymous (2014). Disease strategy: viral haemorrhagic septicaemia (version 2.0), *In:* Australian Aquatic Veterinary Emergency Plan (AQUAVETPLAN), Australian Government Department of Agriculture, Canberra, ACT, Australia [Online] Available from: http://www.agriculture.gov.au/animal-plant-health/aquatic/aquavetplan [Accessed 31 July 2017].

Anonymous (2016). Biosecurity (Notifiable Organisms) Order 2016. NZLR 2016/73. New Zealand Government, Wellington, New Zealand, 13 p.

Anonymous (2017), Viral haemorrhagic septicaemia (VHS). Aquatic Animal Diseases Significant to Australia: Identification Field Guide 4th Edition Department of Agriculture, Fisheries and Forestry. Government of Australia. Canberra, Australia [Online] Available from http://www.agriculture.gov.au/SiteCollectionDocuments/animal-plant/aquatic/field-guide/4th-edition/finfish/viral-haemorrhagic-septicaemia.pdf [Accessed 31 July 2017].

Aquaculture New Zealand (2014). New Zealand's aquaculture industry key facts 2013 [Online] Available from: http://www.seafoodnewzealand.org.nz/our-industry/key-facts/ [Accessed 26 September 2014].

Bain MB, Cornwell ER, Hope KM, Eckerlin GE, Casey RN, Groocock GH, Getchell RG, Bowser PR, Winton JR, Batts WN, Cangelosi A, Casey JW (2010). Distribution of an invasive aquatic pathogen (viral haemorrhagic

septicaemia virus) in the Great Lakes and its relationship to shipping. *PLoS ONE* 5 (4): e10156 [Online] Available from: http://www.ncbi.nlm.nih.gov/pmc/articles/PMC2854150/ [Accessed 23 April 2015].

DAFF (2005). Disease strategy: Viral haemorrhagic septicaemia (Version 1.0), *In*: Australian Aquatic Veterinary Emergency Plan (AQUAVETPLAN), Edition 2. DAFF (Department of Agriculture, Fisheries and Forestry), Canberra, ACT, Australia, 63 p.

DAFF (2014). Viral haemorrhagic septicaemia. Aquatic Animal Diseases Significant to Australia: Identification Field Guide 4th Edition. DAFF (Department of Agriculture, Fisheries and Forestry), Canberra, ACT, Australia. [Online] Available from: http://www.agriculture.gov.au/SiteCollectionDocuments/animal-plant/aquatic/field-guide/4th-edition/aquatic-disease-user-guide-edition-4.pdf.

Dale OB, Orpetveit I, Lyngstad TM, Kahn S, Skall HF, Olesen NJ, Dannevig BH (2009). Outbreak of viral haemorrhagic septicaemia (VHS) in seawater-farmed rainbow trout in Norway caused by VHS virus Genotype III. *Diseases of Aquatic Organisms* 85: 93–103.

Diggles BK (2007). Scientific submission to AQIS supporting an application for an import permit allowing entry of whole, round menhaden (*Brevoortia tyrannus*) for use as lobster bait. DigsFish Services Client Report DF07-01. January 2007, 39 p.

EFSA (2008). Scientific Opinion of the Panel on Animal Health and Welfare (AHAW); Aquatic species susceptible to diseases listed in Directive 2006/88/EC. *The EFSA Journal* 808: 29–144

Einer-Jensen K, Ahrens P, Forsberg R, Lorenzen N (2004). Evolution of the fish rhabdovirus viral haemorrhagic septicaemia virus. *Journal of General Virology* 85(5): 1167–1179.

Fish and Game (2014). New Zealand Fish and Game Council [Online] Available from: http://www.fishandgame.org.nz/ [Accessed 24 September 2014].

Grant B (2007). Fish virus hits the Great Lakes [Online] Available from: http://www.the-scientist.com/?articles.view/articleNo/25137/title/Fish-virus-hits-Great-Lakes/ [Accessed 23 April 2015].

Hedrick RP, Batts WN, Yun S, Traxler GS, Kaufman J, Winton JR (2003). Host and geographic range extensions of the North American strain of viral haemorrhagic septicaemia virus. *Diseases of Aquatic Organisms* 55: 211–220.

Kahn SA, Beers PT, Findlay VL, Peebles IR, Durham PJ, Wilson DW, Gerrity SE (1999). Import risk analysis on non-viable salmonids and non-salmonid marine finfish. Australian Quarantine and Inspection Service, Department of Agriculture, Fisheries and Forestry, Canberra, Australia, 409 p.

Kim WS, Choi SY, Kim DH, Oh MJ)2013). A survey of fish viruses isolated from wild marine fishes from the coastal waters of southern Korea. *Journal of Veterinary Diagnostic Investigation* 25(6): 750–755.

Kim R, Faisal M (2011). Emergence and resurgence of the viral hemorrhagic septicemia virus (Novirhabdovirus, Rhabdoviridae, Mononegavirales). *Journal of Advanced Research* 2(1): 9–23.

Lorenzen N, LaPatra SE (2005). DNA vaccines for aquacultured fish. *OIE Reviews in Science and Technology* 24(1): 201–213.

Marsh D, Mkwra L (2013). Review of freshwater non-market value studies. Department of Economics, University of Waikato, Hamilton, New Zealand, 144 p.

Munday BL (2002). Scientific review of significant disease agents carried by non-viable, non-salmonid freshwater and estuarine finfish and their products. Biosecurity Australia, Department of Agriculture, Fisheries and Forestry, Canberra, Australia, 120 p.

Ogut H, Altunas C (2014). Survey of viral haemorrhagic septicaemia virus in wild fishes in the south-eastern Black Sea.. *Diseases of Aquatic Organisms* 109(2): 99–106

Oidtmann B, Joiner C, Reese RA, Stone D, Dodge M, Dixon P (2011). Risks associated with commodity trade: transmission of viral haemorrhagic septicaemia virus (VHSV) to rainbow trout fry from VHSV-carrying tissue homogenates. *Transboundary and Emerging Diseases* 58: 224–231.

OIE (2016a). Aquatic Animal Health Code, 19th Edition. World Organization for Animal Health. Paris, France [Online]. Available from: http://www.oie.int/international-standard-setting/aquatic-code/access-online/ [Accessed 15 March 2017].

OIE (2016b). Manual of diagnostic tests for aquatic animals. World Organization for Animal Health. Paris, France [Online]. Available from: http://www.oie.int/international-standard-setting/aquatic-manual/access-online/ [Accessed 17 March 2017].

Sim-Smith C, Faire S, Lees A (2014). Managing biosecurity risk for business benefit. Report prepared for the Ministry for Primary Industries by Coast and Catchment Ltd, Auckland, New Zealand, 214 p.

Skall HF, Olesen NJ, Mellergaard S (2005). Viral haemorrhagic septicaemia virus in marine fish and its implications for fish farming – a review. *Journal of Fish Diseases* 28: 509–529.

Spikler AR (2010). Emerging and infectious diseases of animals. Iowa State University Press, 383 p.

Stone DM, Way K, Dixon PF (1997a). Nucleotide sequence of the glycoprotein gene of viral haemorrhagic septicaemia (VHS) viruses from different geographical areas: a link between VHS in farmed fish species and viruses isolated from North Sea cod (*Gadus morhua* L.). Journal of General Virology 78 (6): 1319–1326.

Stone MAB, MacDiarmid SC, Pharo HJ (1997b). Import health risk analysis: salmonids for human consumption. Ministry of Agriculture Regulatory Authority, New Zealand, 269 p.

Tubbs L, Lee P, Diggles B, Jones JB, Sheppard M, Sim-Smith C (2007). A review of aquatic diseases of significance to New Zealand. Final research report for project ZBS 2005-17. National Institute of Water and Atmospheric Research, Wellington, New Zealand, 452 p.

USDA (2015). Viral haemorrhagic septicaemia interim rule [Online] Available from: http://www.regulations.gov/fdmspublic/component/main?main=DocketDetailandd= APHIS-2007-0038 [Accessed 23 April 2015].

Wolf K (1988). Fish viruses and fish viral diseases. Cornell University Press, Ithaca, New York, United States of America, 476 p.

31. *Aeromonas hydrophila* (exotic strains)

31.1. Hazard identification

31.1.1. Aetiological agent

Aeromonas hydrophila are Gram-negative rod-shaped bacteria, classified in Family Vibrionaceae, Subfamily Aeromonadaceae (Quinn *et al*. 1994). *Aeromonas hydrophila* is mesophilic and has been separately classified from *Aeromonas salmonicida*, which is psychrophilic, but more recent genomic analysis indicates these species form part of the *Aeromonas hydrophila* species complex (AHC) (Fuste *et al*. 2012). Within the AHC complex, six main groupings of isolates are recognised: *A. bestiarum, A. hydrophila, A. salmonicida, A. aquariorum, A. piscicola* and *A. popoffii* (Fuste *et al.* 2012; C. Rodgers, *pers.comm.* 2017). Highly pathogenic isolates have a greater number of virulence genes than isolates from healthy fish, or from free-living non-pathogenic isolates (Li *et al.* 2011; Fuste *et al*. 2012; Griffin *et al*. 2013).

Within the *A. hydrophila* group, three clades are recognised: *A. hydrophila hydrophila*; *A. hydrophila ranae*; and *A. hydrophila dhakensis* (Fuste *et al*. 2012). Infection with *Aeromonas hydrophila* causes motile aeromonad septicaemia (MAS) (Austin & Austin 2007). Over 97% of *A. hydrophila* isolates carry at least one virulence gene (*aerA*$^{+}$, *alt*$^{+}$, *ahp*$^{+}$), and the virulence of a particular *A. hydrophila* isolate is highly correlated with the number of virulence genes present in the genome (Li *et al.* 2011; 2011; Sudheesh *et al*. 2012; Griffin *et al*. 2013).

31.1.2. OIE status

Diseases related to *A. hydrophila* are not listed by the OIE (OIE 2016).

31.1.3. New Zealand status

Aeromonas hydrophila is reported from New Zealand (Anderson *et al.* 1994; Diggles *et al.* 2002; Tubbs *et al*. 2007; Anon. 2014; DermNet 2014). Infection with *A. hydrophila* is not a notifiable disease in New Zealand (Anon. 2016).

The recently described highly pathogenic strains, including the Vah strain (Griffin *et al*. 2013) and the ZC1 strain (Hossain *et al.* 2014), have not been reported from New Zealand and are considered exotic (B. Jones, *pers. comm.* 2015). These exotic strains of *A. hydrophila* are included in this assessment.

31.1.4. Epidemiology

Aeromonas hydrophila is a ubiquitous opportunistic contact zoonotic pathogen, affecting wild and farmed teleost and elasmobranch marine and freshwater fish worldwide. It is considered part of the normal microfauna of healthy fish (Austin and Austin 2007; Cipriano & Austin 2011).

The mechanisms controlling infectivity remain unclear, with no simple way to determine whether a particular isolate may be virulent. Several exotic strains are highly pathogenic (Li *et al.* 2011), including the Vah and ZC1 strains which cause epidemics in farmed fish (Table 28).

Table 28. Families and Species of Fish Susceptible *Aeromonas Hydrophila* (Exotic Strains Including Vah and ZC1 Strains)

Family	Host Species
Acipenseridae	Siberian Sturgeon (*Acipenser baerii*)
Cyprinidae	Bighead carp (*Hypophthamichthys nobilis*), Prussian carp (*Carassius gibelio*), grass carp (*Ctenopharyngodon idella*), silver carp (*Hypopthalamichthys molitrix*), Wuchang bream (*Megalobrama amblycephala*)
Ictaluridae	Channel catfish (*Ameiurus punctatus*)
Lotidae	Burbot (*Lota lota*)

The ZC1 strain causes epidemics in farmed cyprinids and sturgeon (Cao *et al.* 2010) in Chinese aquaculture, but has subsequently been reported from farmed channel catfish (*Ameiurus punctatus*) in the United States (Hossain *et al*. 2014). North American channel catfish are also infected with the Vah strain (Griffin *et al.* 2013), while other closely related strains cause epidemics in cyprinids and burbot (*Lota lota*) in Asian aquaculture (Natrah *et al.* 2012; Zhang *et al.* 2014).

Mortality of these highly pathogenic strains may reach 100% in epidemics (Austin & Austin 2007; Cao *et al*. 2010). Infection occurs over a wide temperature range (5°C–35°C), but some isolates remain viable in temperatures up to 70°C (Cipriano & Austin 2011; Haenen *et al.* 2013).

Aeromonas hydrophila isolates are also psychrophilic, remaining viable in chilled (4°C) fish (Daskalov 2006) and surviving in frozen fish for 20 days at -20 °C (Rouf & Rigney 1971; Brady & Vinitnantharat 1990; Yoganth *et al.* 2009).

Infection proceeds in a similar way to lower virulence isolates. *Aeromonas* spp. are transmitted horizontally through the water column, by sloughed off skin cells, faecal material, sexual products, or through the distribution of sub-clinically infected fish that act as reservoirs of infection (Cipriano & Austin 2011).

The disease is initially cutaneous, with visible signs of hyperaemia of the epidermis and underlying muscle, together with haemorrhages on the pectoral fins, liver and intestinal walls and exophthalmia. Infection usually progresses to the visceral organs, causing extensive hepatic and pancreatic necrosis. However, infection may also be latent with no external signs of disease (Grizzle & Kiryu 2011).

Aeromonas spp. may be dispersed in aerosol spray, or through fomites (such as the stainless steel and plastic equipment used in fish farms and fish processing) (Grizzle & Kiryu 2011). They are environmentally resistant, remaining viable for up to 24 days in brackish water, 17 days in fresh water and 8 days in sea water (Grizzle & Kiryu 2011). They can survive in sediments for over 3 months (Tubbs *et al.* 2007). They remain viable after passage through the gut of scavenging birds (Korkoca & Boynukara 2003) and survive routine treatment in municipal sewerage systems (Filipkowska 2003).

Aeromonas hydrophila is heat labile, with inactivation requiring high-temperature treatment (at least 121°C for 30 minutes of moist heat; or at least 170°C for 1 to 2 hours of dry heat) (Anon. 2014).

Vaccination is impractical due to the heterogeneity of the strains likely to be encountered in aquaculture (Goalakannen & Arul 2006).

All New Zealand freshwater fish may be susceptible to *A. hydrophila* infection (Tubbs *et al.* 2007; B. Jones, *pers. comm*. 2015).

31.2. Risk assessment

31.2.1. Entry assessment

Fish with external signs of infection would not be present in the commodity, but sub-clinically infected or carrier fish with few, or no external signs of infection may pass routine inspection and be present in the commodity. *Aeromonas hydrophila* infects the skin and underlying musculature, so it is likely to be retained in the eviscerated commodity (Yoganth *et al*. 2009). It is unaffected by routine processes in fish storage, transport and processing (Brady & Vinitnantharat 1990).

The likelihood of entry of exotic strains of *A. hydrophila* is assessed as non-negligible.

31.2.2. Exposure assessment

To establish infection in New Zealand, infected eviscerated product would have to become available for consumption by a susceptible fish host, in sufficient quantity and duration (Kahn *et al.* 1999). Exotic strains of *A. hydrophila* may be present in the skin and scraps of infected skeletal muscle discarded from commercial fish processing, as well as potentially affecting other fish processing equipment by cross-contamination (Grizzle & Kiryu 2011).

Exotic strains of *A. hydrophila* may remain viable in marine waters and sediments for extended periods, as well as in fresh water and sediments for up to 17 days (Grizzle & Kiryu 2011). They resist high temperature, freezing and mild acid conditions and may remain viable in fish wastes discarded to landfill. They may be transferred from landfill to the aquatic environment by scavenging birds (Korkoca & Boynukara 2003) and remain unaffected by sewerage treatment (Filipkowska 2003). The likelihood of exposure to these exotic strains of *A. hydrophila* is assessed as non-negligible.

31.2.3. Consequence assessment

The exotic and highly pathogenic strains of *A. hydrophila* may potentially affect all freshwater fish (B. Jones, *pers. comm.* 2015), so the consequences of establishment may be extreme. It may affect the aquaculture industry for Chinook salmon (*O. tshawytscha*), valued at $63 million in export earnings in 2011 (Aquaculture New Zealand 2014). An outbreak would also affect recreational and tourist trout and salmon fishing, as well as incur significant social and environmental costs associated with other domestic fisheries (Fish & Game 2014). While the dollar value of New Zealand's freshwater fisheries is not fully known, the Lake Taupo trout fishery alone is valued at $70 to 80 million per annum (Marsh & Mkwra 2013).

These exotic strains would also affect the developing industry for grass carp (*Ctenopharyngodon idella*) and silver carp (*Hypophthalmichthys molitrix*), farmed for waterway management in New Zealand (Mitchell 2009; NIWA 2014).

A. hydrophila is zoonotic, causing skin infections in humans, including folliculitis, impetigo, cellulitis and necrotising fasciitis, as well as gastroenteritis, meningitis and peritonitis (Anon. 2014; DermNet 2014). However, the exotic strains are unlikely to represent a greater risk to human health than the endemic strains already present in New Zealand (Anon. 2014; DermNet 2014).

The consequences of establishment of the exotic strains of *A. hydrophila* are assessed as non-negligible.

31.2.4. Risk estimation

Since the entry, exposure and consequence assessments are non-negligible, the risk estimate is non-negligible. Therefore, exotic strains of *A. hydrophila* are assessed to be a risk in the commodity and risk management measures may be justified.

31.3. Risk management

Exotic strains of *A. hydrophila* have been assessed to be a risk in the commodity. Infection with *A. hydrophila* is a non-notifiable disease, so there is no specific guidance in the OIE *Aquatic Code* (OIE 2016) for importing eviscerated fish from infected countries, or specific processing requirements that would ensure the destruction of the bacterium. It is assumed that compliance with the approved processed states for OIE-listed diseases (Appendix 3) should also eliminate exotic strains of *A. hydrophila* from the commodity and be a viable risk management option.

Exotic strains of *A. hydrophila* are currently reported from four families of fish (Table 28), which may be present in the commodity. While species declaration should substantially reduce the pathogen load and be a viable risk management option, there is no simple way to determine whether a given exotic isolate has the potential to be virulent.

Known exotic strains are reported from China and the United States, but no requirements for dedicated monitoring exist. Approval of country/zone freedom through the MPI Country Approval Procedures should substantially reduce the occurrence of these exotic strains of *A. hydrophila* in the commodity. Approval of a declaration of country/zone freedom by MPI is a viable risk management option.

The exotic virulent strains of *A. hydrophila* are primarily associated with aquaculture in China, Asia and North America (Cao *et al.* 2010; Natrah *et al.* 2012; Zhang *et al.* 2014). Restriction of the commodity to wild-caught fish (not from aquaculture) should moderately reduce pathogen load and be a viable risk management option.

Infection with *A. hydrophila* is associated with the skin and dermal musculature, so removal of the gills, or the head and gills should only slightly reduce the pathogen load of *A. hydrophila* in the commodity. Further processing to the skin-off fillet state would moderately reduce the occurrence of exotic *A. hydrophila* in the commodity and be a viable risk management option.

Aeromonas hydrophila is unaffected by freezing, so frozen storage is not a viable risk management option. Heat inactivation using moist heat (to at least 121°C for 30 minutes) (Anon. 2014) would eliminate exotic *A. hydrophila* from the commodity and be a viable risk management option.

31.3.1. Risk management options

Exotic strains of *Aeromonas hydrophila* are reported from fish in families Acipenseridae, Cyprinidae, Ictaluridae and Lotidae (Table 28), which are considered likely to be present in the commodity. Other families have not been associated with the exotic strains of *A. hydrophila*. Therefore species declaration indicating the commodity is not originated from any of the above families should substantially reduce the occurrence of exotic strains of *A. hydrophila* in the commodity.

For the commodities originated from families associated with exotic strains of *A. hydrophila*, one or a combination of the following additional options could also be considered to effectively manage the risk.

Where country/zone freedom from exotic strains of *A. hydrophila* is accepted by MPI:

Option 1

Acceptance of country/zone freedom by MPI should substantially reduce the occurrence of exotic *Aeromonas hydrophila* strains, so the commodity may be imported without any further restrictions.

Where country/zone freedom from exotic strains of *A. hydrophila* is not accepted by MPI or not available:

Option 2

Processing to a state consistent with the approved states for OIE-listed diseases (Appendix 3) should eliminate exotic strains of *A. hydrophila*. When these provisions are met, the commodity could be imported without any further restrictions.

Where the imported commodity is not in a state consistent with the approved states for OIE-listed diseases (Appendix 3):

Option 3

Heat treatment (by cooking, with moist heat, to at least 121°C for 30 minutes) should eliminate exotic strains of *A. hydrophila*, so the commodity could be imported without any further restrictions; or,

Option 4

Further processing (to the skin-off fillet state) should moderately reduce the occurrence of exotic strains of *A. hydrophila*, so the commodity could be imported without any further restrictions; or,

Option 5

Restriction of the commodity to wild-caught fish (not from aquaculture) should moderately reduce the occurrence of exotic strains of *A. hydrophila*, so the commodity could be imported without any further restrictions.

31.4. References

Anderson CD, Knowles G, de Lisle G W (1994). A survey for *Yersinia ruckeri* and *Aeromonas salmonicida* in farmed and wild fish. *Surveillance* 21: 39–40.

Anonymous (2014). *Aeromonas hydrophila*. Pathogen safety data sheet - infectious substances. Public Health Agency of Canada [Online] Available from: http://www.phac-aspc.gc.ca/lab-bio/res/psds-ftss/aeromonas-hydrophila-eng.php [Accessed 15 December 2014].

Anonymous (2016). Biosecurity (Notifiable Organisms) Order 2016. NZLR 2016/73. New Zealand Government, Wellington, New Zealand, 13 p.

Aquaculture New Zealand (2014). New Zealand's aquaculture industry key facts [Online] Available from: http://www.seafoodnewzealand.org.nz/our-industry/key-facts/ [Accessed 26 September 2014].

Austin B, Austin A (2007). Bacterial fish pathogens: Diseases of farmed and wild fish. Springer, New York, 580 p.

Brady YT, Vinitnantharat S (1990). Viability of bacterial pathogens in frozen fish. *Journal of Aquatic Animal Health* 2(2): 149–150.

Cao H, He S, Lu L, Hou L (2010). Characterisation and phylogenetic analysis of the bitrichous pathogenic *Aeromonas hydrophila* isolated from diseases Siberian sturgeon (*Acipenser baerii*). *The Israeli Journal of Aquaculture* 62(3): 181–188.

Cipriano RC, Austin B (2011). Furunculosis and other aeromonad diseases, pp 424–483 *In*: Woo PTK, Bruno DW (*Eds.*). *Fish Diseases and Disorders: Volume 3 - Bacterial, Viral and Fungal Infections*, 2nd edition. CABI International, Wallingford, United Kingdom, 930 p.

Daskalov H (2006). The importance of *Aeromonas hydrophila* in food safety. *Food Control* 17(6): 474–483.

DermNet (2014). *Aeromonas* skin infection [Online] Available from: http://dermnetnz.org/bacterial/aeromonas.html [Accessed 23 September 2014].

Diggles BK, Hine PM, Handley S, Boustead NC (2002). A handbook of diseases of importance to aquaculture in New Zealand. *NIWA Science and Technology Series* 49, National Institute of Water and Atmospheric Research, Auckland, 200 p.

Filipkowska Z (2003). Sanitary and bacteriological aspects of sewage treatment. *Acta Microbiologica Polonica* 52: 57–66.

Fish and Game (2014). New Zealand Fish and Game Council [Online] Available from: http://www.fishandgame.org.nz/ [Accessed 24 September 2014].

Fuste CM, Farfan M, Minana-Galbis D, Albarral C, Sanglas A, Loren JG (2012). Population genetics of the "*Aeromonas hydrophila* Species Complex", pp 39–54 *In:* Fuste C (*Ed*.) Studies in Population Genetics. Intech Open [Online] Available from: http://www.intechopen.com/books/studies-in-population-genetics/population-genetics-of-the-Aeromonas-hydrophila-species-complex- doi: 10.5772/35584 [Accessed 28 April 2015].

Goalakannen A, Arul V (2006). Immunomodulatory effects of dietary intake of chitin, chitosan and levamisole on the immune system of *Cyprinus carpio* and control of *Aeromonas hydrophila* infection in ponds. *Aquaculture* 255: 179–187.

Griffin MJ, Goodwin AE, Merry GE, Liles MR, Williams MA, Ware C, Waldbieser GC (2013). Rapid quantitative detection of *Aeromonas hydrophila* strains associated with disease outbreaks in catfish culture. *Journal of Veterinary Diagnostic Investigation* 25(4): 473–481.

Grizzle JM, Kiryu Y (2011). Histopathology of gill, liver and pancreas, and serum levels of channel catfish infected with *Aeromonas hydrophila* complex. *Journal of Aquatic Animal Health* 5(1): 36–50.

Haenen OLM, Evans JJ, Berthe F (2013). Bacterial infections from aquatic species; potential for and prevention of contact zoonoses. *OIE Reviews in Science and Technology* 32(2): 497–507.

Hossain MJ, Sun D, McGarey DJ, Wrenn S, Alexander LM, Martino ME, Xing Y, Terhune JS, Liles MR (2014). An Asian origin of virulent *Aeromonas hydrophila* responsible for disease epidemics in United States-farmed catfish [Online] Available from: *MBio* 5(3):e00848-14. doi: 10.1128/mBio.00848-14 [Accessed 28 April 2015].

Kahn SA, Beers PT, Findlay VL, Peebles IR, Durham PJ, Wilson DW, Gerrity SE (1999). Import risk analysis on non-viable salmonids and non-salmonid marine finfish. Australian Quarantine and Inspection Service, Department of Agriculture, Fisheries and Forestry, Canberra, Australia, 409 p.

Korkoca H, Boynukara B (2003). The characterisation of protein profiles of the *Aeromonas hydrophila* and *A. caviae* strains isolated from gull and rainbow trout faeces. *Turkish Journal of Veterinary Animal Science* 27: 1173–1177.

Li J, Ni XD, Liu YJ, Lu CP (2011). Detection of three virulence genes *alt*, *ahp* and *aerA* in *Aeromonas hydrophila* and their relationship with actual virulence to zebrafish. *Journal of Applied Microbiology* 110(3): 823–830.

Marsh D, Mkwra L (2013). Review of freshwater non-market value studies. Department of Economics, University of Waikato, Hamilton, New Zealand, 144 p.

Mitchell C (2009). Profile of white amur (grass carp) and silver amur from the results of releases in New Zealand waterways. Charles Mitchell and Associates, Raglan, New Zealand, 15 p.

Natrah FMI, Alam MI, Pawar S, Harzevili AS, Nevejan N, Boon N, Sorgeloos P, Bossier P, Defoirdt T (2012). The impact of quorum sensing on the virulence of *Aeromonas hydrophila* and *Aeromonas salmonicida* towards burbot (*Lota lota* L.) larvae. *Veterinary Microbiology* 159(1): 77–82.

NIWA (2014). Grass carp *Ctenopharyngodon idella* (Valenciennes, 1844) [Online] Available from: https://www.niwa.co.nz/freshwater-and-estuaries/nzffd/NIWA-fish-atlas/fish-species/grass_carp [Accessed 12 February 2015].

OIE (2016). Aquatic Animal Health Code, 19th Edition. World Organization for Animal Health. Paris, France [Online] Available from: http://www.oie.int/international-standard-setting/aquatic-code/access-online/ [Accessed 15 March 2017].

Quinn PJ, Carter ME, Markey B, Carter GR (1994). Clinical Veterinary Medicine. Mosby, London, United Kingdom, 648p.

Rouf MA, Rigney MM (1971). Growth temperatures and temperature characteristics of *Aeromonas. Applied and Environmental Microbiology* 22(4): 503–506.

Tubbs L, Lee P, Diggles B, Jones JB, Shepherd M, Sim-Smith C (2007). A review of aquatic diseases of significance to New Zealand. Final research report for project ZBS 2005-17. National Institute of Water and Atmospheric Research, Wellington, New Zealand, 452 p.

Yoganth N, Bhakyaraj R, Chanthuru A, Anbalagan T, Nila KM (2009). Detection of virulence genes in *Aeromonas hydrophila* isolated from fish samples using PCR technique. *Global Journal of Biotechnology and Biochemistry* 4(1): 51–53.

Zhang X, Yang W, Wu H, Gong X, Li A (2014). Multilocus sequence typing revealed a clonal lineage of *Aeromonas hydrophila* caused motile *Aeromonas* septicaemia outbreaks in pond-cultured cyprinid fish in an epidemic area in central China. *Aquaculture* 432: 1–6.

32. *Aeromonas salmonicida* subsp. *salmonicida* (atypical strains)

32.1. Hazard identification

32.1.1. Aetiological agent

Aeromonas salmonicida are Gram-negative rod-shaped bacteria, classified in Family Vibrionaceae, Subfamily Aeromonadaceae (Quinn *et al*. 1994). *Aeromonas salmonicida* subsp. *salmonicida* atypical strains are Gram-negative non-motile aeromonad bacteria that represent a group of clades within the *Aeromonas hydrophila* species complex (AHC) (Fuste *et al.* 2012).

Within the *A. salmonicida* group, up to five clades are recognised: *Aeromonas salmonicida achromogenes, A. s. masoucida, A. s. pectinolytica, A. s. salmonicida* and *A. s. smithia,* although the taxonomy of the group is unclear (Austin & Austin 1993; Whittington *et al.* 1995; Wiklund & Dalsgaard 1998; Mundy 2002; Fuste *et al*. 2012). Isolates within this group have a more complex genome with a larger number of pseudogenes than the *A. hydrophila* isolates of the AHC complex (Fuste *et al.* 2012), which is thought to explain their higher pathogenicity to fish (Sudheesh *et al*. 2012).

Isolates of *A. s. salmonicida* vary in response to standard biochemical tests, in their growth characteristics, pigment production and biochemistry in cell culture (Mundy 2002; Kahn *et al.* 1999). The isolates that follow a "typical" response to these standard tests are generally associated with furunculosis of salmonids and cause bacterial septicaemia in other marine and freshwater fish. These "typical" isolates are generally associated with furunculosis of salmonids and are discussed in the following chapter (Chapter 32). The *A.s. salmonicida* isolates that follow an "atypical" response are regarded as the causative agent of bacterial septicaemia, causing cutaneous dermal lesions in many fish species (Wiklund & Dalsgaard 1998). They are considered below.

The terms "typical" and "atypical" specifically apply to the reactions involved with pathology of salmonids and they cause considerable confusion in the literature when applied to non-salmonids (Hidalgo & Figueras 2012). They have been generally used to describe infections of atypical *A. s. salmonicida* in non-salmonids, as well as to describe infection with other subspecies of *A. salmonicida*. In general, separation of these isolates requires use of specific biochemical methods (Hidalgo & Figueras 2012), but recent genomic analysis indicates that the separation of the sub-species into "typical" and "atypical" strains is essentially artificial and does not reflect the complexity inherent within the group (Hidalgo & Figueras 2012; Dallaire-Dufresne *et al*. 2014). However, these separations are maintained in this document largely for clarity in the identification of related fish hosts.

32.1.2. OIE status

Infection with atypical *A. s. salmonicida* is not an OIE-listed disease (OIE 2016a).

32.1.3. New Zealand status

Survey data indicate atypical *Aeromonas salmonicida* strains are not present in New Zealand (Anderson *et al*. 1994; Keeling *et al*. 2013). It has not subsequently been reported and is

considered exotic (Tubbs *et al.* 2007). All isolates of *A. salmonicida* are notifiable diseases in New Zealand (Anon. 2016).

32.1.4. Epidemiology

Atypical strains of *A. salmonicida* have been isolated from a wide variety of marine and freshwater fish from North America, Brazil, Chile, China, Japan, Northern, Central Europe and Southern Europe (Canary Islands), and Australia. Susceptible host species are shown in Table 29 (Whittington & Cullis 1988; Real *et al*. 1994; Humphrey & Ashburner 1993; Yang & Chen 1996; Bergh *et al*. 1997; Wiklund & Dalsgaard 1998; Kahn *et al.* 1999; Pavanelli *et al.* 2000; Castro-Escarpulli *et al.* 2003; Wang & Huang 2006; Tubbs *et al.* 2007; Ronneseth *et al.* 2015; Menenteau-Ledouble *et al.* 2016;C. Rodgers, *pers. comm*. 2017).

Table 29. Families and Species of Fish Susceptible to Atypical Strains of *Aeromonas salmonicida* var. *salmonicida*

Family	Species
Ammodytidae	Sand eels (*Ammodytes lancea*, *Hyperoplus lanceolatus*)
Anarhichadidae	Common wolffish (*Anarhichas lupus*), spotted wolffish (*Anarhichas minor*)
Anguillidae	European eel (*Anguilla anguilla*), Japanese eel (*A. japonica*) , American eel (*A. rostrata*)
Anoplopomatidae	Sablefish (*Anoplopoma fimbria*)
Catostomidae	Golden shiner (*Notemigonus crysoleucas*),white sucker (*Catostomus commersonii*)
Centrarchidae	Smallmouth bass (*Micropterus dolomieu*)
Cichlidae	Nile tilapia (*Oreochromis niloticus*)
Clupeidae	Herring (*Clupea harengus*, *Clupea pallasii*)
Cyclopteridae	Lumpfish (*Cyclopterus lumpus*)
Cyprinidae	Bream (*Abramis brama*), chub (*Squalius cephalus*), common and ornamental carp (*Cyprinus carpio*), creek chub (*Semotilus atromaculatus*), crucian carp (*Carassius carassius*), dace (*Leuciscus leuciscus*), goldfish (*Carassius auratus*), golden shiner (*Notemigonus crysoleucas*), Eurasian minnow (*Phoxinus phoxinus*), roach (*Rutilus rutilus*), silver bream (*Blicca bjoerkna*)
Esocidae	Pike (*Esox lucius*)
Gadidae	Atlantic cod (*Gadus morhua*), haddock (*Melanogrammus aeglefinus*), tomcod (*Microgadus tomcod*), whiting (*Merlangius merlangus*)
Gerreidae	Mojarra (*Gerres ovatus*)
Hexagrammidae	Lingcod (*Ophiodon elongatus*)
Labridae	Corkwing wrasse (*Symphodus melops*), goldsinny wrasse (*Ctenolabrus rupestris*), rock cook (*C. exoletus*), Ballan wrasse (*Labrus bergylta*)
Latridae	Striped trumpeter (*Latris lineata*)
Lotidae	Fourbeard rocking (*Enchelyopus cimbrius*)
Moronidae	Yellow bass (*Morone mississippiensis*), European seabass (*Dicentrarchus labrax*)
Percidae	European perch (*Perca fluviatilis*)
Pleuronectidae	Dab (*Limanda limanda*), flounder (*Platichthys flesus*, *Rhombosolea tapirina*), halibut (*Eopsetta grigorjewi*. *Hippoglossus hippoglossus*, *H. stenolepis*), plaice (*Hippoglossoides platessoides*), *Pleuronectes platessa*)
Salmonidae	Arctic char (*Salvelinus alpinus*), Atlantic salmon (*Salmo salar*), brook trout (*Salvelinus fontinalis*), chum salmon (*Oncorhynchus keta*), Chinook salmon (*O. tshawytscha*), grayling (*Thymallus thymallus*), brown trout (*Salmo trutta*), masu salmon (*O. masou*), pink salmon (*O. gorbuscha*), rainbow trout (*O. mykiss*), whitefish (*Coregonus* sp.)
Scophthalmidae	Turbot (*Scophthalmus maximus*)
Sparidae	Gilthead seabream (*Sparus aurata*)
Terapontidae	Silver perch (*Bidyanus bidyanus*)
Zoarcidae	Viviparous blenny (*Zoarces viviparous*)

Atypical strains of *A. salmonicida* have been regarded as a serious threat to Australian salmonid aquaculture (Carson & Gudkovs 2001), but no outbreaks have been reported since 2008 (Zainathan 2012). Three biovars have previously been reported from Australia. The first biovar, which was isolated from goldfish (*Carassius auratus*) has become established in wild goldfish populations in

Southeast Australia (Trust *et al.* 1980). The second biovar, which was isolated from farmed greenback flounder (*Rhombosolea tapirina*) (Pleuronectidae), is infective to Atlantic salmon (*S. salar*) and striped trumpeter (*Latris lineata*) (Latridae) by cohabitation (Whittington *et al*. 1995; DAFF 2008). Infection occurs with no clinical signs of disease, and these carrier fish may represent a reservoir for disease. The third "Acheron" biovar was isolated from farmed Atlantic salmon (*Salmo salar*) in Tasmania (Carson & Gudkovs 2001), where it is reported as marine aeromonad disease of salmonids (DAFF 2008).

Infection in North American freshwaters has spread from farmed salmon (*Oncorhynchus* spp., *Salmo* spp.) to adjacent wild fish including creek chub (*Semotilus atromaculatus*), golden shiner (*Notemigonus crysoleucas*), goldfish (*Carassius auratus*) and white sucker (*Catostomus commersonii*). These are commonly used as live baitfish in Canada and the United States (Ostland *et al.* 1987), but are not considered as fish for human consumption in this assessment. Atypical *A. salmonicida* infection may be transferred from infected goldfish, common and ornamental carp (*Cyprinus carpio*) to farmed salmonids through cohabitation (Whittington & Cullis 1988; Wiklund & Dalsgaard 1998), or through infected cleaner fish used in salmonid sea cage aquaculture to control salmon louse (*Lepeophtheirus salmonis*) (Gulla *et al.* 2015).

Cleaner fish susceptible to *atypical A. salmonicida* infection include the Ballan wrasse (*Labrus bergylta*), corkwing (*Symphodus melops*), rock cook, (*Centrolabrus exoletus*) and wild goldsinny (*Ctenolabrus rupestris*) (Labridae); as well as the lumpsucker (*Cyclopterus lumpus*) (Cyclopteridae) (Gulla *et al.* 2015). Cleaner fish including *L. bergylta* and *Cyclopterus lumpus* are now being farmed to cope with increasing demands from aquaculture (Gulla *et al.* 2015).

Atypical *A. s. salmonicida* infection also occurs in farmed gilthead seabream (*Sparus aurata*) and European seabass (*Dicentrarchis labrax*) in Spanish aquaculture (C. Rodgers, *pers. comm.* 2017). It is reported from blacktip reef shark *Carcharhinus melanopterus*) held in aquaria (Briones *et al.* 1998). As no subsequent infection has been reported from wild *C. melanopterus*, this infection is assumed to be opportunistic and related to aquarium confinement (C. Rodgers, *pers. comm.* 2017).

Infection with atypical strains of *A. salmonicida* cause "ulcer disease" or "atypical furunculosis." They are less pathogenic than typical strains, with mortalities varying from 15 to 29%, according to the bacterial strain and host species (Kahn *et al.* 1999). However, cumulative mortalities may become significant during epizootics, reaching 25% in carp aquaculture and 65% in Atlantic salmon (*S. salar*) aquaculture (Wiklund & Dalsgaard 1998) and up to 98% for European perch (*Perca fluviatilis*), silver bream (*Blicca bjoerkna*) and Eurasian minnow (*Phoxinus phoxinus*) (Munday 2002). Epizootic outbreaks are generally associated with temperatures in excess of 10°C, often affecting young fish that die with no external signs of disease (Munday 2002).

External signs of infection include exophthalmia and small white patches on the skin and gills (Gudmundsdóttir 1998). These patches progressively form cutaneous ulcerative lesions, which develop into boils with associated septicaemia that may extend deep into the musculature. Chronic infection is commonly associated with degenerative necrosis of the liver, spleen and kidney (Wiklund & Dalsgaard 1998; Cipriano & Austin 2011).

Atypical *A. salmonicida* can also be transferred through movements of dead and frozen fish (Whittington *et al.* 1987; Diggles 2001). Transmission is horizontal, through the water column, with infection occurring by cohabitation, or by ingestion. Pathogen entry occurs through epidermal cells in the skin and gills, or the digestive tract (Farto *et al.* 2011; Vanden Bergh & Frey 2013). Infection may also be transferred between fish by ectoparasites such as *Lepeophtheirus salmonis*, *Argulus corregoni* and *Tetrahymena pyriformis* in fresh water (Cipriano & Bullock 2001).

Atypical *A. salmonicida* is resistant to the environmental conditions likely to be encountered in fish processing and storage (Evelyn 2001). It remains viable in dead fish tissue for up to 32 days, and may survive for a further 8 days in water (Munday 2002). Survival in fresh water is longer (6 to 9 months) than in marine waters (10 days), but may be further extended by entry into a viable, but non-culturable (VBNC) stage that can survive in sediment for up to 3 months (Munday 2002).

Atypical *A. salmonicida* is unaffected by low-temperature storage, surviving for 49 days in visceral tissues and 28 days in kidney tissue stored at 4°C (Munday 2002). It is resistant to freezing, remaining viable for up to 49 days after frozen storage (-10°C) in visceral tissues, but lower temperature storage for 7 days in muscle tissue (frozen to -20°C) resulted in a reduction in the number of viable cells by two orders of magnitude (Munday 2002).

Atypical *A. salmonicida* is can be spread in aerosol form (Wooster & Bowser 1996) or by fomites such as stainless steel or plastic surfaces at the water/air interface in factory processing resulting in cross-contamination of plant and equipment (Cipriano & Bullock 2001; Munday 2002).

Atypical *A. salmonicida* is heat stable and requires cooking at 44°C for 60 minutes, or 48°C for 30 minutes for inactivation (Munday 2002; Ishiguro *et al.* 2013). It is inactivated by UV radiation treatment (1051.02 (±67.54) mJ cm^{-2} for 15 minutes), although its effectiveness is limited where radiation may be shielded by organic material (Munday 2002; Walker *et al.* 2013). It is denatured by iodophors (1g L^{-1} for 30 minutes) (Ishiguro *et al.* 2013). Outbreaks were shown to be initially controlled by antibiotics, such as oxytetracycline, but resistance is reported (Kim *et al.* 2013).

Oil-based vaccines developed for typical strains of *A. s. salmonicida* have been used to control atypical strains in Atlantic salmon aquaculture in Norway since 1992 (Gudmundsdóttir 1998) and in Tasmania, Australia since 2006 (Zainathan 2012). However, vaccines are considered to have limited effectiveness and are not generally available (Gudmundsdóttir & Björnsdóttir 2007).

Atypical *A. salmonicida* is not implicated in human disease (Lowry & Smith 2007).

Potential host species in New Zealand include all salmonids (*Oncorhynchus* spp., *Salmo* spp., *Salvelinus* spp.), as well as a wide variety of fish in marine and freshwater environments. If *A. s. salmonicida* becomes established, eradication and control may be difficult (Munday 2002).

32.2. Risk assessment

32.2.1. Entry assessment

Fish chronically infected with atypical *A. s. salmonicida* would be unlikely to pass visual inspection, but carrier fish with no apparent clinical signs (Munday 2002) may enter the human food consumption pathway. Disease is mainly associated with the skin and dermal musculature, although degenerative necrosis of the liver, spleen and kidney may occur in latently infected carrier fish (Wiklund & Dalsgaard 1998; Tubbs *et al*. 2007; DAFF 2008). While the latter should be removed on evisceration, infection present in the skin and dermal muscle, together with any residual kidney and visceral tissues (C. Rodgers *pers. comm.* 2017) may be present in the commodity.

The likelihood of entry of atypical *A. s. salmonicida* is assessed as non-negligible.

32.2.2. Exposure assessment

To establish infection, sufficient infected eviscerated product would have to become available for consumption by a susceptible fish host, in sufficient quantity and duration (Kahn *et al.* 1999). Atypical *A. s. salmonicida* may be present in the blood-water discharge and in infected skin and muscle tissue discarded after commercial food processing (Munday 2002). It is resistant to freezing (Munday 2002) and may remain viable on dry surfaces, where it can be transferred and distributed in aerosol form (Wooster & Bowser 1996), to contaminate fish processing equipment (Munday 2002).

Atypical *A. s. salmonicida* survives in dead fish tissue for up to 32 days and can be transferred through movements of dead and frozen fish (Evelyn 2001; Diggles 2011). It is resistant to temperature and mild acid conditions (Munday 2002), and it may remain viable in wastewater and fish offal discarded from commercial processing. It remains infective in fresh water for a further 8 days, or in sediment for up to 3 months (Tubbs *et al.* 2007). Infection can be transmitted to adjacent wild fish populations and the infective dose is low (10^3 cfu mL^{-1}) (Bergh *et al.* 1997). Potential freshwater and marine fish hosts are present in New Zealand (Tubbs *et al.* 2007). Therefore, the likelihood of exposure is assessed as non-negligible.

32.2.3. Consequence assessment

Given the variability in virulence between isolates, the consequences of establishment in particular potential host species cannot be determined in advance (Kahn *et al.* 1999). The establishment of atypical *A. s. salmonicida* could directly affect the Chinook salmon (*Oncorhynchus tshawytscha*) farming industry through loss of sales. This industry was valued at $63 million in export earnings in 2011 (Aquaculture New Zealand 2014). An outbreak could also affect recreational and tourist trout and salmon fishing, as well as incur significant social and environmental costs associated with other domestic fisheries (Fish & Game 2014). While the dollar value of New Zealand's freshwater fisheries is not fully known, the Taupo fishery alone is valued at $70 to 80 million per annum (Marsh & Mkwra 2013). Atypical *A. s. salmonicida* is not implicated in human disease (Lowry & Smith 2007).

The consequences of establishment are assessed as non-negligible.

32.2.4. Risk estimation

Since the entry, exposure and consequence assessments for atypical *A. s. salmonicida* are non-negligible, the risk estimate is non-negligible. Therefore, atypical *A. s. salmonicida* is assessed to be a risk in the commodity and risk management measures may be justified.

32.3. Risk management

Atypical *A. s. salmonicida* has been assessed to be a risk in the commodity. As infection with atypical *A. s. salmonicida* is a non-notifiable disease, there is no guidance in the OIE Aquatic *Code* (OIE 2016) for importing eviscerated fish from infected countries, or specific processing requirements that would ensure the destruction of the bacterium. It is assumed that compliance with the approved processed states for OIE-listed diseases (Appendix 3) would also eliminate atypical *A. s. salmonicida* from the commodity and be a viable risk management option.

Atypical *A. s. salmonicida* has been isolated from over 20 families of marine and freshwater fish families (Table 29), which may be present in the commodity. Species declaration should substantially reduce the pathogen load of atypical *A. s. salmonicida* and be a viable risk management option.

Atypical *A. s. salmonicida* has a worldwide distribution including Europe, North America, South America, Asia and Africa. Where country/zone freedom is approved through the MPI Country Approval Procedures, this option is should substantially reduce the pathogen load of atypical *A. s. salmonicida* in the commodity. Approval of a declaration of country/zone freedom by MPI is a viable risk management option.

Atypical *A. s. salmonicida* affects a wide range of fish, often with little or no signs of disease, but is a major pathogen of Australian salmonid aquaculture. Restriction of the commodity to wild-caught fish (not from aquaculture) would moderately reduce pathogen load and be a viable risk management option.

Vaccination is only of limited effectiveness for atypical *A. s. salmonicida* in salmonids and not available for non-salmonid fish (Gudmundsdóttir & Björnsdóttir 2007). It is not considered a viable risk management option and is not discussed further.

Atypical *A. s. salmonicida* may be present in the skin and muscle tissues of carrier fish, so either removal of the gills, or of the head and gills, should slightly reduce pathogen load. Further processing of the commodity to the skin-off fillet state would be likely to moderately reduce the pathogen load of atypical *A. s. salmonicida* as it would reduce the likelihood of waste generation, capable of spreading disease. Processing to the skin-off fillet state is a viable risk management option.

Frozen storage (for at least 7 days at -20°C) only results in a reduction of numbers of viable cells (by two orders of magnitude) (Munday 2002) and is not considered a viable risk management option. Atypical *A. s. salmonicida* is inactivated by heat treatment, so cooking (to 44°C for 60 minutes) is likely to eliminate the pathogen from the commodity. Heat treatment is a viable risk management option.

32.3.1. Risk management options

Atypical *Aeromonas salmonicida* var. *salmonicida* has been reported from fish in families Ammodytidae, Anarhichadidae, Anguillidae, Anoplopomatidae, Centrarchidae, Cichlidae, Clupeidae, Cyclopteridae, Cyprinidae, Esocidae, Gadidae, Gerreidae, Hexagrammidae, Labridae, Latridae, Lotidae, Moronidae, Percidae, Pleuronectidae, Salmonidae, Scophthalmidae, Sparidae, Terapontidae and Zoarcidae (Table 29). These families are considered likely to be present in the commodity. Other families have not been associated with atypical *A. s. salmonicida.* Therefore species declaration indicating the commodity is not originated from any of the above families should substantially reduce the occurrence of atypical *A. s. salmonicida* in the commodity.

For the commodities originated from families associated with atypical *A. s. salmonicida*, one or a combination of the following additional options could also be considered to effectively manage the risk.

Where country/zone freedom from atypical *A. s. salmonicida* is accepted by MPI:

Option 1

Acceptance of country/zone freedom by MPI should substantially reduce the occurrence of atypical *A. s. salmonicida*, so the commodity may be imported without any further restrictions.

Where country/zone freedom from atypical *A. s. salmonicida* is not accepted by MPI or not available:

Option 2

Processing to a state consistent with the approved states for OIE-listed diseases (Appendix 3) should eliminate atypical *A. s. salmonicida*. When these provisions are met, the commodity could be imported without any further restrictions.

Where the imported commodity is not in a state consistent with the approved states for OIE-listed diseases (Appendix 3):

Option 3

Heat treatment (by cooking to 44°C for 60 minutes) should eliminate atypical *A. s. salmonicida.* When this provision is met, the commodity could be imported without any further restrictions; or,

Option 4

Further processing (to the skin-off fillet state) should moderately reduce the occurrence of atypical *A. s. salmonicida.* When this provision is met, the commodity could be imported without any further restrictions; or,

Option 5

Restriction of the commodity to wild-caught fish (not from aquaculture) should moderately reduce the occurrence of atypical *A. s. salmonicida.* When this provision is met, the commodity could be imported without any further restrictions.

32.4. References

Anderson C, Knowles G, De Lisle G (1994). A survey for *Yersinia ruckeri* and *Aeromonas salmonicida* in farmed and wild fish. *Surveillance* 21(3): 38– 40.

Anonymous (2016). Biosecurity (Notifiable Organisms) Register 2016. NZLR 2016/73. New Zealand Government, Wellington, New Zealand, 13 p.

Aquaculture New Zealand (2014). New Zealand's aquaculture industry key facts [Online] Available from: http://www.seafoodnewzealand.org.nz/our-industry/key-facts/ [Accessed 26 September 2014].

Austin BA, Austin DA (1993). Bacterial fish pathogens: Disease of wild and farmed fish. Fifth Edition. Springer, New York, United States of America, 652 p.

Bergh Ø, Hjeltnes B, Skiftesvik AB (1997). Experimental infection of turbot, *Scophthalmus maximus* and halibut *Hippoglossus hippoglossus* yolk sac larvae with *Aeromonas salmonicida* subsp. *salmonicida*. *Diseases of Aquatic Organisms* 29: 13–20.

Briones V, Fernandez A, Blanco M, Ramiro F, de Vicente ML, Garcia J, Mendez JL, Goyache J (1998). Haemorrhagic septicaemia by *Aeromonas salmonicida* subsp. *salmonicida* in a blacktip reef shark (*Carcharhinus melanopterus*). *Zentralblatt fur Veterinarmedizin Reihe* B 45:443–445.

Carson J, Gudkovs N (2001). *Aeromonas salmonicida*: a sleeping giant. *Microbiology Australia* 22(3): 28–30.

Castro-Escarpulli G, Figueras MJ, Aguilera-Arreola G, Soler L, Fernandez-Rendon E, Aparicio GO, Guarro J, Chacon MR (2003). Characterisation of *Aeromonas* spp. isolated from frozen fish intended for human consumption in Mexico. *International Journal of Food Microbiology* 84(1): 41–49.

Cipriano RC, Austin B (2011). Furunculosis and other Aeromonad diseases. pp 424-483. *In*: Woo PTK, Bruno DW *(Eds*.) Fish Diseases and Disorders, Volume 3: Viral, Bacterial and Fungal Infections. CABI International; Wallingford, United Kingdom, 930 p.

Cipriano RC, Bullock GL (2001). Furunculosis and other diseases caused by *Aeromonas salmonicida*. Fish Disease Leaflet 66. National Fish Health Laboratory, US Geological Survey, Kearneyville, West Virginia, United States of America, 33p.

DAFF (2008). Aquatic animal diseases significant to Australia: *Aeromonas salmonicida*. Identification field Guide. Department of Agriculture, Fisheries and Forestry, Canberra, Australia, 29 p.

Dallaire-Dufresne S, Tanak KH, Trudel MV, Lafaille A, Charette SJ (2014). Virulence, genomic features, and plasticity of *Aeromonas salmonicida* subsp. *salmonicida*, the causative agent of fish furunculosis. *Veterinary Microbiology* 169: 1–7.

Diggles BK (2011). Environmental assessment report – Disease risks. DigsFish Report DF 11-02. Final report prepared for New Zealand King Salmon. DigsFish Services Ltd, Queensland, Australia, 68 p.

Evelyn TPT (2001). The effects of chilling, freezing and cold-smoking on the infectious titre of certain microbial fish pathogens that may occasionally be present in marketed salmonid flesh, pp 215-229 *In:* Rodgers CJ (*Ed*.) Risk analysis in aquatic animal health – proceedings of an international conference. World Organisation for Animal Health (OIE); Paris, France.

Farto R., Milton DL, Bermudez MB, Nieto TP (2011). Colonization of turbot tissues by virulent and avirulent *Aeromonas salmonicida* subsp. *salmonicida* strains during infection. *Diseases of Aquatic Organisms* 95: 167–173.

Fish and Game (2014). New Zealand Fish and Game Council [Online] Available from: http://www.fishandgame.org.nz/, [Accessed 24 September 2014].

Fuste MC, Farfan M, Minana-Galbis D, Albarrral V, Sanglas A, Loren JG (2012). Population genetics of the "*Aeromonas hydrophila* Species Complex", pp 39–54 *In*: Fuste MC (Ed.) Studies in Population Genetics. InTech Rijeka, Croatia, 178 p.

Gudmundsdóttir BK (1998). Infections by atypical strains of the bacterium *Aeromonas salmonicida*. *ICEL Journal of Agricultural Science* 12: 61–72.

Gudmundsdóttir BK, Björnsdóttir B (2007). Vaccination against atypical furunculosis and winter ulcer disease of fish. *Vaccine* 25(30): 5512–5523.

Gulla S, Duodu S, Nilsen A, Fossen I, Colquhoun DJ (2015). *Aeromonas salmonicida* infection levels in pre- and post-stocked cleaner fish assessed by culture and an amended qPCR assay. *Journal of Fish Diseases* 39(7): 867–877.

Hidalgo RB, Figueras MJ (2012). Molecular detection and characterisation of furunculosis and other *Aeromonas* fish infections. *In*: *Health and Environment in Aquaculture*, pp 97–132, InTech Open [Online] Available from: http://cdn.intechopen.com/pdfs/35139.pdf [Accessed 29 April 2015].

Humphrey J, Ashburner L (1993). Spread of the fish pathogen *Aeromonas salmonicida* after importation of infected goldfish, *Carassius auratus*, into Australia. *Australian Veterinary Journal* 70: 453–454.

Ishiguro EE, Kay WW, Ainsworth T, Chamberlain JB, Austen RA, Buckley JT, Trust TJ (2013). Loss of virulence during culture of *Aeromonas salmonicida* at high temperature. *Journal of Bacteriology* 148(1): 333–340.

Kahn SA, Beers PT, Findlay VL, Peebles IR, Durham PJ, Wilson DW, Gerrity SE (1999). Import risk analysis on non-viable salmonids and non-salmonid marine finfish. AQIS, Canberra, Australia, 409 p.

Keeling SE, Brosnahan CL, Johnston C, Wallis R, Gudkovs N, McDonald WL (2013). Development and validation of a real-time PCR assay for the detection of *Aeromonas salmonicida. Journal of Fish Diseases* 36: 495–503.

Kim JH, Choresca CH, Shin SP, Han JE, Jun JW, Park SC (2013). Biological control of *Aeromonas salmonicida* subsp. *salmonicida* infection in rainbow trout (*Oncorhynchus mykiss*) using *Aeromonas* phage PAS-1. *Transboundary and Emerging Diseases* 62(1): 81–86.

Lowry T, Smith SA (2007). Vet Med Today: Zoonosis Update. *Journal of the Aquatic Veterinary Medical Association* 6: 876–880.

Marsh D, Mkwra L (2013). Review of freshwater non-market value studies. Department of Economics, University of Waikato, Hamilton, New Zealand, 144 p.

Menenteau-Ledouble S, Kumar G, Saleh M, El-Matbouli M (2016). *Aeromonas salmonicida:* Updates on an old acquaintance. *Diseases of Aquatic organisms* 120(1): 4-–68 [Online] Available from: http://www. doi: 10.3354/dao03006 [Accessed 7 August 2018].

Munday BL (2002). Scientific review of significant disease agents carried by non-viable, non-salmonid freshwater and estuarine finfish and their products. Biosecurity Australia, Department of Agriculture, Fisheries and Forestry, Canberra, Australia, 120 p.

OIE (2016). Aquatic Animal Health Code, 19th Edition. World Organization for Animal Health. Paris, France [Online] Available from: http://www.oie.int/international-standard-setting/aquatic-code/access-online/ [Accessed 15 March 2017].

Ostland VE, Hicks BD, Daley JG (1987). Furunculosis in baitfish and its transmission to salmonids. *Diseases of Aquatic Organisms* 2: 163–166.

Pavanelli GC, Takemoto RM, Eiras JD (2000). Health of fishes. *Informe Agropecuario* (Belo Horizonte) 21(203): 48–56.

Quinn PJ, Carter ME, Markey B, Carter GR (1994). Clinical Veterinary Medicine. Mosby, London, United Kingdom, 648p.

Real F, Acosta B, Deniz S, Oros J, Rodriguez E (1994). *Aeromonas salmonicida* infection *in Sparus aurata* in the Canaries. *Bulletin of the European Association of Fish Pathologists* 14(5): 153–155.

Ronneseth A, Haugland GT, Brudal E, Wergeland HI (2015). Bacterial diseases and vaccination of farmed lumpfish (*Cyclopterus lumpus* l.). p 97 In: Conference Proceedings, 17th International Conference on Diseases of Fish and Shellfish, Las Palmas, Canary Islands, Spain. European Association of Fish Pathologists, 463 p.

Sudheesh PS, Al-Ghabshi A, Al-Mazrooei N, Al-Habsi S (2012). Comparative pathogenomics of bacteria causing infectious diseases in fish. *International Journal of Evolutionary Biology* [Online] Available from: http://www.hindawi.com/journals/ijeb/2012/457264/ [Accessed 29 April 2015].

Trust TJ, Khouri AG, Austen RA, Ashburner LD (1980). First isolation in Australia of atypical *Aeromonas salmonicida*. *FEMS Microbiology Newsletters* 9: 39–42.

Tubbs L, Lee P, Diggles B, Jones JB, Shepherd M, Sim-Smith C (2007). A review of aquatic diseases of significance to New Zealand. Final research report for project ZBS 2005-17. National Institute of Water and Atmospheric Research, Wellington, New Zealand, 452 p.

Vanden Bergh P, Frey J (2013). *Aeromonas salmonicida* subsp. *salmonicida* in the light of its type-three secretion system. *Microbial technology* 7: 381–400.

Walker RW, Markillie LM, Clotelo AH, Geist DR, Gay ME, Woodley CM, Eppard MB, Brown RS (2013). Ultraviolet radiation as disinfection for fish surgical tools. *Animal Biotelemetry* 1(4): 1–11.

Wang GX, Huang ZR (2006). Studies on the pathogen and histopathology of septicaemia in *Esox lucius. Journal of Fisheries of China* 30(3): 383–389.

Whittington R, Cullis B (1988). The susceptibility of salmonid fish to an atypical strain of *Aeromonas salmonicida* that affects goldfish (*Carassius auratus*) in Australia. *Journal of Fish Diseases* 11: 461–470.

Whittington RJ, Djordjevic SP, Carson J, Callinan RB (1995). Restriction endonuclease analysis of atypical *Aeromonas salmonicida* isolates from goldfish *Carassius auratus*, silver perch *Bidyanus bidyanus* and greenback flounder *Rhombosolea tapirina* in Australia. *Diseases of Aquatic Organisms* 22: 185–191.

Whittington RJ, Gudkovs N, Carrigan MJ, Ashburner LD, Thurstan SJ (1987). Clinical, microbiological and epidemiological findings in recent outbreaks of goldfish ulcer disease due to atypical *Aeromonas salmonicida* in south-eastern Australia. *Journal of Fish Diseases* 10: 353–362.

Wiklund T, Dalsgaard I (1998). Occurrence and significance of atypical *Aeromonas salmonicida* in non-salmonid and salmonid fish species: a review. *Diseases of Aquatic Organisms* 32: 49–69.

Wooster G, Bowser P (1996). The aerobiological pathway of a fish pathogen: survival and dissemination of *Aeromonas salmonicida* in aerosols and its implications in fish health management. *Journal of the World Aquaculture Society* 27 (1): 7–14.

Yang XL, Chen YX (1996). The current situation and trends in the development of fish vaccines. *Journal of Fisheries of China* 20(2): 159–167.

Zainathan SC (2012). Detection of aquareovirus in farmed Tasmanian Atlantic Salmon (*Salmo salar*). Ph.D Thesis. University of Tasmania, Australia, 316 p.

33. *Aeromonas salmonicida* subsp. *salmonicida* (typical strains)

33.1. Hazard identification

33.1.1. Aetiological agent

Aeromonas salmonicida subsp. *salmonicida* typical strains are Gram-negative non-motile aeromonad bacteria classified in the Family Aeromonadaceae (Quinn *et al.* 1994), considered as a group of clades within the *Aeromonas hydrophila* species complex (AHC) (Fuste *et al.* 2012; Sudheesh *et al.* 2012).

Aeromonas salmonicida comprises five clades: *A. s. achromogenes, A. s. masoucida, A. s. pecinolytica, A. s. salmonicida* and *A. s. smithia*) although the taxonomy of the group is unclear (Austin & Austin 1993; Whittington *et al.* 1995; Wiklund & Dalsgaard 1998; Munday 2002; Fuste *et al*. 2012).

Isolates of *A. salmonicida* vary in response to standard biochemical tests (Mundy 2002; Kahn *et al.* 1999). Those that follow an "atypical" response are considered in Chapter 31. Isolates which follow a "typical" response are generally associated with furunculosis of salmonids, but also cause bacterial septicaemia in other marine and freshwater fish (Farto *et al.* 2011). These isolates are considered further in this chapter.

The separation into "typical" and "atypical" strains may not properly reflect the complexity within the AHC group (Hidalgo & Figueras 2012; Dallaire-Dufresne *et al*. 2014), particularly when applied to infection of non-salmonids (Hidalgo & Figueras 2012). However, these isolates are considered separately, largely for clarity in the identification of related fish hosts.

33.1.2. OIE status

Infection with *A. s. salmonicida* typical strain is not notifiable to the OIE (OIE 2016).

33.1.3. New Zealand status

Survey data indicate *A. s. salmonicida* typical strain is not present in New Zealand and is considered exotic (Anderson *et al*. 1994; Tubbs *et al.* 2007; Keeling *et al*. 2013). All isolates of *A. salmonicida* are notifiable in New Zealand (Anon. 2016).

33.1.4. Epidemiology

Furunculosis is regarded as one of the most serious diseases of wild and farmed salmonids in Europe (Denmark, Norway, Finland, Scotland and Spain), North America (United States and Canada), South America, Asia and Africa (Kahn *et al.* 1999; Munday 2002).

Infection is not always associated with clinical disease (Munday 1992; Kahn *et al.* 1999; Menenteau-Ledouble *et al.* 2016). Typical strains of *A. s.* s*almonicida* have been isolated from salmonids including rainbow trout (*Oncorhynchus mykiss*) and Chinook salmon (*O. tshawytscha*), although all salmonids (e.g., *Corrregonus* spp., *Hucho* spp., *Oncorhynchus* spp., *Prosopium* spp., *Salmo* spp,. *Salvelinus* spp.), may be affected (Austin & Austin 1993; Kent *et al.* 1998). It has also been reported from marine and freshwater species, as shown in Table 30 (Kent *et al.* 1998; C. Rodgers, *pers.comm*. 2017). It is reported from Chile, Japan, northern and central Europe including

the Mediterranean, the United Kingdom, France, Spain as well as from North America (Canada and the United States) and Australia (Ostland *et al.* 1987; Humphrey & Ashburner 1993; Wiklund & Dalsgaard 1998; Kahn *et al.* 1999; Cipriano & Bullock 2001; Tubbs *et al.* 2007; Vanden Bergh *et al*. 2013; C. Rodgers *pers.com*. 2017).

Table 30. Families and Host Species of Fish Susceptible to Typical *Aeromonas salmonicida* var. *salmonicida*

Family	Species
Catostomidae	White sucker (*Catostomus commersonii*)
Cyprinidae	Creek chub (*Semotilus atromaculatus*), golden shiner (*Notemigonus crysoleucas*), goldfish (*Carassius auratus*), Eurasian minnow (*Phoxinus phoxinus*), silver bream (*Blicca bjornica*)
Esocidae	Pike (*Esox lucius*)
Gadidae	Atlantic cod (*Gadus morhua*), coalfish (*Pollachius virens*)
Labridae	Wrasses (*Centrolabrus* spp., *Coris* spp., *Ctenolabris* spp., *Labroides* spp., *Labrus* spp., *Symphodus* spp.)
Moronidae	Seabass (*Dicentrarchus labrax*)
Pleuronectidae	Halibut (*Hippoglossus hippoglossus, H. stenolepis*)
Salmonidae	All salmonids (*Coregonus* spp., *Hucho* spp., *Oncorhynchus* spp., *Parahucho* spp., *Prosopium* spp., *Salmo* spp,. *Salvelinus* spp., *Salvethymus* spp., *Stenodus* spp., *Thymallus* spp.)
Scophthalmidae	Turbot (*Scophthalmus maximus*)
Sparidae	Sea bream (*Sparus aurata*)

The concentration of typical *A. s. salmonicida* may reach 10^3 to 10^6 cfu g^{-1} in apparently healthy salmonids (Johnston 1999). There are low minimum doses of 10^2 cfu mL^{-1} for Atlantic salmon (*S. salar*) in sea water, but this increases to more than 10^8 cfu mL^{-1} for rainbow trout in fresh water (Johnston 1999), suggesting viable infection pathways exist through cohabitation and ingestion (Stone *et al.* 1997; Johnston 1999).

Infection may follow one of two paths: acute and chronic. Acute infection commonly progresses with few external signs, other than darkening of the skin and reddening of the fin bases, while a progressive septicaemia develops through the internal organs leading to death through haemorrhage and circulatory failure. Acute infection may be marked with white skin patches, representing growth of *A. s. salmonicida* in the musculature that develop into necrotic lesions or furuncles (Bruno & Ellis 1996).

In chronic infection, the furuncles progressively develop into deep necrotic lesions, which may extend through the musculature, allowing protrusion of the viscera. In older fish, this is combined with necrotic septicaemia of the musculature, internal organs and heart, enlargement of the spleen and liquefaction of the kidney tissues. Death occurs though septicaemia and general circulatory failure (Bruno & Ellis 1996; Tubbs *et al.* 2007). Disease progression is complex, depending on factors including the health, age and species of the host (Dallaire-Dufresne *et al.* 2014).

Virulence varies among strains and is dependent upon the structure of the A-layer, the production of extracellular proteases, lipases and enzymes, as well as the type III secretion system that transfers toxins to the host cytoplasm (Dallaire-Dufresne *et al.* 2014). Virulence varies from < 10 cells per fish for Atlantic salmon (*Salmo salar*) injected with a goldfish (*Carassius auratus*) isolate (LD_{50}= 7.4 x 10^{-3} cells $fish^{-1}$), to 1.7 x 10^9 cfu $fish^{-1}$ for rainbow trout (*O. mykiss*) injected with isolates from infected silver bream (*Blicca bjornica*) (Wiklund & Dalsgaard 1998).

Clinical disease occurs in wild fish, or in fish farmed in the absence of antibiotics or vaccines (Bergh *et al.* 1997), although antibiotic resistance has been reported (Kim *et al.* 2013) and vaccines have practical limitations (Dallaire-Dufresne *et al.* 2014). Infection may be epizootic, with mortality reaching 54% for Atlantic salmon (*S. salar*) (Jarp *et al.* 2006), and up to 73% for

rainbow trout (*O. mykiss*), but this varies among bacterial strains and hosts (Bullock & Stuckey 1975; Dallaire-Dufresne *et al.* 2014). Optimal growth is reported within a temperature range of 22 to 25°C, but virulence decreases when temperatures exceed 22°C (Dallaire-Dufresne *et al.* 2014).

While effective vaccines have been developed (Midtlyng *et al.* 1996a, 1996b; Vanden Bergh *et al.* 2013; Veso 2015), these are expensive and may cause side effects leading to lower productivity (Midtlyng 1997). The immune protection decreases over time, while the genomic plasticity inherent in this group has proven to be a limitation in the use of a generalised vaccine and in the very real possibility that disease may be transmitted through the prophylactic treatment itself (Cipriano & Austin 2011). Bacteriophage treatments are under development but their effectiveness against furunculosis is uncertain (Kim *et al*. 2013).

Transmission is horizontal, by shedding infected material through the water, or by physical contact. Surviving carrier fish may pass infected material via the gill and skin mucosa or through faeces from infections in visceral tissues (Tubbs *et al*. 2007). Disease may be transmitted by ectoparasites including *Lepeophtheirus salmonis, Argulus coregoni* and *Tetrahymena pyriformis* in fresh water (Cipriano & Bullock 2001). Wild fish stocks including creek chub (*Semotilus atromaculatus*), golden shiner (*Notemigonus crysoleucas*) and white sucker (*Catostomus commersonii*) may act as reservoirs of infection for farmed salmonids in Canada and the United States, where these species are commonly used as freshwater live baitfish (Ostland *et al.* 1987).

Aeromonas salmonicida salmonicida is resistant to the environmental conditions likely to be encountered in fish processing and storage. It can be transferred in dead and frozen fish tissues (Whittington *et al.* 1987; Diggles 2011). It survives in dead fish tissue for up to 32 days, remaining viable for 28 days in kidney tissues stored at 4°C (Munday 2002). It survives freezing (to -10°C for 49 days) in visceral tissues, although lower temperature storage (to -20°C for 7) has resulted in a two log reduction in viable cell numbers in frozen muscle tissue (Munday 2002).

Aeromonas salmonicida salmonicida survives for 8 days in water (Munday 2002) and can be transmitted through the air in aerosol form, or by contact with contaminated surfaces such as stainless steel or plastics in factory processing (Cipriano & Bullock 2001; Munday 2002).

Once established, eradication and control may be difficult, due to multiple hosts (Munday 2002) and its long survival in fresh water (6 to 9 months). While survival is shorter in marine waters (10 days), this may be extended where it can enter a viable, but non-culturable (VBNC) resistant stage that may remain in sediment for an additional 3 months (Munday 2002).

Aeromonas salmonicida salmonicida is heat stable, with some virulent strains surviving at 30°C (Ishiguro *et al.* 2013) and requires heat treatment at 44°C for 60 minutes or 48°C for 30 minutes for inactivation (Munday 2002). It is resistant to UV radiation treatment at 1051.02 (±67.54) mJ cm^{-2} (Munday 2002; Walker *et al.* 2013) and to iodophors, requiring 1000 mg L^{-1} for 30 minutes for inactivation (Ishiguro *et al.* 2013).

Current vaccines provide limited protection (reducing cumulative mortality from 90% to 25% in epizootics) (Romstad *et al.* 2014; Veso 2015) and have side-effects (Villumsen *et al.* 2012). While formalin-killed bath immersion vaccines are under development, these are not yet commercially available (Villumsen & Raida 2013).

Aeromonas salmonicida salmonicida is not considered to cause human disease (Lowry & Smith 2007).

Potential host species in New Zealand include all salmonids (Tubbs *et al.* 2007), as well as a wide variety of fish in marine and freshwater environments.

33.2. Risk assessment

33.2.1. Entry assessment

With the advent of antibiotics and vaccination treatment, the incidence of clinical furunculosis has declined, but developing antibiotic resistance (Kim *et al.* 2013), the variable effectiveness of vaccination and the incidence of disease in non-salmonid fish (Dallaire-Dufresne *et al.* 2014) has meant that chronically infected fish may be present in the commodity. While fish with furuncles and other obvious skin infections would be unlikely to pass visual inspection, carrier fish with no apparent clinical signs may enter the human food consumption pathway (Munday 2002). Furunculosis is associated with the mucous and epidermis of the skin and gills, as well as with the musculature of latently infected carrier fish (Tubbs *et al*. 2007; DAFF 2008). These tissues would be retained after evisceration and may be present in the commodity (Dallaire-Dufresne *et al.* 2014).

The likelihood of entry of typical *A. s. salmonicida* is assessed as non-negligible.

33.2.2. Exposure assessment

Potential hosts of typical *A. s. salmonicida* include all salmonids and a wide variety of non-salmonid host species in marine and fresh waters of New Zealand (Tubbs *et al*. 2007). To establish infection through the human food consumption pathway, infected eviscerated product would have to become available for consumption by a susceptible fish host, in sufficient quantity and duration (Kahn *et al*. 1999). *A. s. salmonicida* may be present in blood-water discharge as well as in the offal discarded after commercial food processing. Infective tissues may be present in the skin, gill tissues and muscle trimmings discarded in processing. It is resistant to freezing (Munday 2002) and remains viable on dry surfaces, where it may be transferred and distributed in aerosol form (Wooster & Bowser 1996), or through contact with fish processing equipment (Munday 2002). It may remain viable in dead fish tissue for up to 32 days and from then remain infective in fresh water for a further 8 days, or in sediment for up to 3 months (Tubbs *et al.* 2007), where infection may be transmitted to adjacent wild fish populations (Bergh *et al.* 1997). It is resistant to temperature and mild acid conditions and remains viable in fish wastes discarded from commercial processing as wastewater (Munday 2002). The infective dose required to initiate infection is low (10^3 cfu mL^{-1}) and potential freshwater and marine hosts are present in New Zealand (Tubbs *et al.* 2007).

The likelihood of exposure for typical *A. s. salmonicida* is assessed as non-negligible.

33.2.3. Consequence assessment

The establishment of typical *A. s. salmonicida* could affect all salmonids in New Zealand. This may result in direct economic losses for the Chinook salmon (*O. tshawytscha*) farming industry, through loss of sales due to infected product. This was valued at $63 million in export earnings in 2011 (Aquaculture New Zealand 2014). An outbreak could also affect recreational and tourist trout and salmon fishing, as well as incur significant social and environmental costs associated with other domestic fisheries (Fish & Game 2014). While the dollar value of New Zealand's freshwater fisheries is not fully known, the Lake Taupo trout fishery alone is valued at $70 to 80 million per annum (Marsh & Mkwra 2013).

Other species in marine and fresh waters may be impacted (Tubbs *et al.* 2007), but given the variability in virulence among isolates of *A. salmonicida*, the effect on particular potential host species cannot be determined in advance. Björnsdóttir 2007).

Typical *A. s. salmonicida* is not implicated in human disease (Lowry & Smith 2007).

The consequences of establishment are therefore assessed as non-negligible.

33.2.4. Risk estimation

Since the entry, exposure and consequence assessments for typical *A. s. salmonicida* are non-negligible, the risk estimate is non-negligible. Therefore, typical *A. s. salmonicida* is assessed to be a risk in the commodity and risk management measures may be justified.

33.3. Risk management

Typical *A. s. salmonicida* has been assessed to be a risk in the commodity. As infection with *A. s. salmonicida* is a non-notifiable disease, there is no specific guidance in the OIE *Aquatic Code* (OIE 2016) for importing eviscerated fish from infected countries, or specific processing requirements that would ensure the destruction of the pathogen. It is assumed that compliance with the approved processed states for OIE-listed diseases (Appendix 3) would also eliminate typical *A. s. salmonicida* from the commodity and be a viable risk management option.

Ten families of fish are susceptible to the typical strain of *A. s. salmonicida* (Table 30) and these may be present in the commodity. Species declaration should substantially reduce the pathogen load of typical *A. s. salmonicida* in the commodity and be a viable risk management option.

Typical *A. s. salmonicida* occurs at low rates of incidence in wild stocks, but is a major disease of farmed fish worldwide. Restriction of the commodity to wild-caught fish (not from aquaculture) would moderately reduce pathogen load and be a viable risk management option.

While vaccines for typical *A. s. salmonicida* are available, they have limited effectiveness and are only available for a narrow range of species. Certification of vaccination is not considered a viable management option and is not considered further.

Typical *A. s. salmonicida* has a worldwide distribution that includes Europe, North America, South America, Asia and Africa. Where country/zone freedom is approved through the MPI Country Approval Procedures, this option is likely to substantially reduce the pathogen load of typical *A. s. salmonicida* in the commodity. Approval of a declaration of country/zone freedom by MPI is a viable risk management option.

While chronically infected fish with obvious infection of the skin and musculature would be unlikely to be present in the commodity, carrier fish may be present in the commodity. Removal of the gills, or the head and gills, should slightly reduce pathogen load of typical *A. s. salmonicida* in the commodity. Further processing to the skin-off fillet state would moderately reduce pathogen load of typical *A. s. salmonicida* in the commodity as it would reduce the likelihood of waste generation, capable of spreading disease. Processing to the skin-off fillet state is a viable risk management option.

Typical *A. s. salmonicida* is not affected by freezing, so frozen storage is not a viable management option. It is inactivated by heat treatment, so cooking (to 44°C for 60 minutes or 48°C for 30

minutes) is likely to eliminate the pathogen from the commodity. Heat treatment is a viable risk management option.

33.3.1. Risk management options

Typical *A. s. salmonicida* is reported from fish in families Catostomidae, Cyprinidae, Esocidae, Labridae, Moronidae, Pleuronectidae, Salmonidae, Scopthalmidae and Sparidae (Table 30), which are considered likely to be present in the commodity. Other families have not been associated with typical *A. s. salmonicida.* Therefore species declaration indicating the commodity is not originated from any of the above families should substantially reduce the occurrence of typical *A. s. salmonicida* in the commodity.

For the commodities originated from families associated with typical *A. s. salmonicida,* one or a combination of the following additional options could be considered to effectively manage the risk.

Where country/zone freedom from typical *A. s. salmonicida* is accepted by MPI:

Option 1

Acceptance of country/zone freedom by MPI should substantially reduce the occurrence of typical *A. s. salmonicida,* so the commodity may be imported without any further restrictions.

Where country/zone freedom from typical *A. s. salmonicida* is not accepted by MPI or not available:

Option 2

Processing to a state consistent with the approved states for OIE-listed diseases (Appendix 3) should eliminate typical *A. s. salmonicida*. When these provisions are met, the commodity could be imported without any further restrictions.

Where the imported commodity is not in a state consistent with the approved states for OIE-listed diseases (Appendix 3):

Option 3

Heat treatment (by cooking to at least 44°C for 60 minutes) should eliminate typical *A. s. salmonicida*. When this provision is met, the commodity could be imported without any further restrictions; or,

Option 4

Further processing (to the skin-off fillet state) should moderately reduce the occurrence of typical *A. s. salmonicida.* When this provision is met, the commodity could be imported without any further restrictions; or,

Option 5

Restriction of the commodity to wild-caught fish (not from aquaculture) should moderately reduce the occurrence of typical *A. s. salmonicida.* When this provision is met, the commodity could be imported without any further restrictions.

33.4. References

Anderson C, Knowles G, De Lisle G (1994). A survey for *Yersinia ruckeri* and *Aeromonas salmonicida* in farmed and wild fish. *Surveillance* 21(3): 38–40.

Anonymous (2016). Biosecurity (Notifiable Organisms) Register 2016. NZLR 2016/73. New Zealand Government, Wellington, New Zealand, 13 p.

Aquaculture New Zealand (2014). New Zealand's aquaculture industry key facts [Online] Available from: http://www.seafoodnewzealand.org.nz/our-industry/key-facts/ [Accessed 26 September 2014].

Austin BA, Austin DA (1993). Bacterial fish pathogens: Disease of wild and farmed fish. Fifth Edition. Springer, New York, United States of America, 652 p.

Bergh Ø, Hjeltnes B, Skiftesvik AB (1997). Experimental infection of turbot, *Scophthalmus maximus* and halibut *Hippoglossus hippoglossus* yolk sac larvae with *Aeromonas salmonicida* subsp. *salmonicida*. *Diseases of Aquatic Organisms* 29: 13–20.

Bruno DE, Ellis AE (1996). Salmonid Disease Management, pp 739–832 *In:* Pennell W, Barton BA (*Eds*.) Developments in Aquaculture and Fisheries Science: Volume 29, Principles of Salmonid Culture, Elsevier, New York, United States of America, 1039 p.

Bullock GL, Stuckey HM (1975). *Aeromonas salmonicida*: Detection of asymptomatically infected trout. *The Progressive Fish-Culturist* 37: 237–239.

Cipriano RC, Austin B (2011). Furunculosis and other aeromonad diseases, pp 424–483 *In:* Woo PTK, Bruno DW (*Eds.*). *Fish Diseases and Disorders: Volume 3 - Bacterial, Viral and Fungal Infections*, 2nd edition. CABI International, Wallingford, United Kingdom, 930 p.

Cipriano RC, Bullock GL (2001). Furunculosis and other diseases caused by *Aeromonas salmonicida*. Fish Disease Leaflet 66. National Fish Health Laboratory, US Geological Survey, Kearneyville, West Virginia, United States of America, 33p.

DAFF (2008). Aquatic animal diseases significant to Australia: *Aeromonas salmonicida*. Identification field Guide. Department of Agriculture, Fisheries and Forestry, Canberra, Australia, 29 p.

Dallaire-Dufresne S, Tanaka KH, Trudel MV, Lafaille A, Charette S (2014). Virulence, genomic features, and plasticity of *Aeromonas salmonicida* subsp. *salmonicida*, the causative agent of fish furunculosis. *Veterinary Microbiology* 169: 1–7.

Diggles BK (2011). Environmental assessment report – Disease risks. DigsFish Report DF 11-02. Final report prepared for New Zealand King Salmon. DigsFish Services Ltd, Queensland, Australia, 68 p.

Farto R., Milton DL, Bermudez MB, Nieto TP (2011). Colonization of turbot tissues by virulent and avirulent *Aeromonas salmonicida* subsp. *salmonicida* strains during infection. *Diseases of Aquatic Organisms* 95: 167–173

Fish and Game (2014). New Zealand Fish and Game Council [Online] Available from: http://www.fishandgame.org.nz/ [Accessed 24 September 2014].

Fuste CM, Farfan M, Minana-Galbis D, Albarral C, Sanglas A, Loren JG (2012). Population genetics of the "*Aeromonas hydrophila* Species Complex", pp 39–54 *In:* Fuste C (*Ed.*). Studies in Population Genetics. Intech, [Online]. Available from: http://www.intechopen.com/books/studies-in-population-genetics/population-genetics-of-the-Aeromonas-hydrophila-species-complex- doi.10.5772/35584 [Accessed 28 April 2015].

Gulla S, Lund V, Kristoffersen AB, Sorum H, Colquhoun DJ (2015). *vapA* (A-layer) typing differentiates *Aeromonas salmonicida* subspecies and identifies a number of previously undescribed subtypes. *Journal of Fish Diseases* (in press 2015) [Online] Available from: http://onlinelibrary.wiley.com/doi/10.1111/jfd.12367/full [Accessed 21 September 2015].

Hidalgo RB, Figueras MJ (2012). Molecular detection and characterisation of furunculosis and other *Aeromonas* fish infections. *In*: *Health and Environment in Aquaculture*, pp 97–132, InTech Open [Online]. Available from: http://cdn.intechopen.com/pdfs/35139.pdf [Accessed 29 April 2015].

Humphrey J, Ashburner L (1993). Spread of the fish pathogen *Aeromonas salmonicida* after importation of infected goldfish, *Carassius auratus*, into Australia. *Australian Veterinary Journal* 70: 453–454.

Ishiguro EE, Kay WW, Ainsworth T, Chamberlain JB, Austen RA, Buckley JT, Trust TJ (2013). Loss of virulence during culture of *Aeromonas salmonicida* at high temperature. *Journal of Bacteriology* 148(1): 333–340.

Jarp J, Gjevre AG, Olsen AB, Bruheim T (2006). Fisk factors for furunculosis, infectious pancreatic necrosis and mortality in post-smolt of Atlantic salmon *Salmo salar* L. *Journal of Fish Diseases* 18(1): 67–76.

Johnston C (1999). Supplementary import risk analysis: Head-on, gill-in Australian salmonids for human consumption. Biosecurity Authority, Wellington, New Zealand, 24 p.

Kahn SA, Beers PT, Findlay VL, Peebles IR, Durham PJ, Wilson DW, Gerrity SE (1999). Import risk analysis on non-viable salmonids and non-salmonid marine finfish. Australian Quarantine and Inspection Service, Department of Agriculture, Fisheries and Forestry, Canberra, Australia, 409 p.

Keeling SE, Brosnahan CL, Johnston C, Wallis R, Gudkovs N, McDonald WL (2013). Development and validation of a real-time PCR assay for the detection of *Aeromonas salmonicida. Journal of Fish Diseases* 36: 495–503.

Kent ML, Traxler GS, Kieser D, Richard J, Dawe SC, Prosperi-Porter G, Ketheson J, Evelyn TPT (1998). Survey of salmonid pathogens in ocean-caught fishes in British Columbia, Canada. *Journal of Aquatic Animal Health* 10(2): 211–219.

Kim JH, Choresca CH, Shin SP, Han JE, Jun JW, Park SC (2013). Biological control of *Aeromonas salmonicida* subsp. *salmonicida* infection in rainbow trout (*Oncorhynchus mykiss*) using *Aeromonas* phage PAS-1. *Transboundary and Emerging Diseases* 62(1): 81–86.

Lowry T, Smith SA (2007). Vet Med Today: Zoonosis Update. *Journal of the Aquatic Veterinary Medical Association* 6: 876–880.

Marsh D, Mkwra L (2013). Review of freshwater non-market value studies. Department of Economics, University of Waikato, Hamilton, New Zealand, 144 p.

Menenteau-Ledouble S, Kumar G, Saleh M, El-Matbouli M (2016). *Aeromonas salmonicida:* Updates on an old acquaintance. *Diseases of Aquatic organisms* 120(1): 4–-68 [Online] Available from: http://www. doi: 10.3354/dao03006 [Accessed 7 August 2018].

Midtlyng PJ, Reitan LJ, Lillehaug A, Ramstad A. (1996a). Protection, immune responses and side effects in Atlantic salmon (*Salmo salar* L.) vaccinated against furunculosis by different procedures. *Fish and Shellfish Immunology* 6(8): 599–613.

Midtlyng PJ, Reitan LJ, Speilberg L (1996b). Experimental studies on the efficacy and side-effects of intraperitoneal vaccination of Atlantic salmon (*Salmo salar* L.) against furunculosis. *Fish and Shellfish Immunology* 6(5): 335–350.

Midtlyng PJ (1997). Vaccination against furunculosis. pp 382–404. *In*: Bernoth EM, Ellis AE, Midtlyng PJ, Olivier G, Smith P (*Eds.*) Furunculosis. Multidisciplinary Fish Disease Research, Academic Press, San Diego, United States of America, 529 p.

Munday BL (2002). Scientific review of significant disease agents carried by non-viable, non-salmonid freshwater and estuarine finfish and their products. Biosecurity Australia, Department of Agriculture, Fisheries and Forestry, Canberra, Australia, 120 p.

OIE (2016). Aquatic Animal Health Code, 19th Edition. World Organization for Animal Health. Paris, France [Online] Available from: http://www.oie.int/international-standard-setting/aquatic-code/access-online/ [Accessed 15 March 2017].

Ostland VE, Hicks BD, Daley JG (1987). Furunculosis in baitfish and its transmission to salmonids. *Diseases of Aquatic Organisms* 2: 163–166.

Quinn PJ, Carter ME, Markey B, Carter GR (1994). Clinical Veterinary Medicine. Mosby, London, United Kingdom, 648p.

Romstad AB, Reintan LJ, Midtlyng P, Gravningen K, Emilsen V, Evensen O (2014). Comparison of a serological potency assay for furunculosis vaccines (*Aeromonas salmonicida* subsp. *salmonicida*) to intraperitoneal challenge in Atlantic salmon (*Salmo salar* L.) *Biologicals* 42(2): 86–90.

Stone, MAB, MacDiarmid SC, Pharo HJ (1997). Import health risk analysis: salmonids for human consumption. Ministry of Agriculture Regulatory Authority, New Zealand, 269 p.

Tubbs L, Lee P, Diggles B, Jones JB, Shepherd M, Sim-Smith C (2007). A review of aquatic diseases of significance to New Zealand. Final research report for project ZBS 2005-17. National Institute of Water and Atmospheric Research, Wellington, New Zealand, 452 p.

Vanden Bergh P, Heller M, Braga-Lagache S, Frey J (2013). The *Aeromonas salmonicida* subsp. *salmonicida* exoproteome: global analysis, moonlighting proteins and putative antigens for vaccination against furunculosis. *Proteome Science* 11: 44 [Online] Available from: https://proteomesci.biomedcentral.com/articles/10.1186/1477-5956-11-44 [Accessed 18 August 2017].

Veso (2015). Atlantic salmon/rainbow trout – *Aeromonas salmonicida.* Veso Vikan [Online] Available from: http://www.veso.no/174768 [Accessed 23 September 2015].

Villumsen KR, Dalsgaard I,Holten-Andersen L, Raida MK (2012). Potential Role of Specific Antibodies as Important Vaccine Induced Protective Mechanism against Aeromonas salmonicida in Rainbow Trout. *PLoS ONE* 7(10): e46733. doi:10.1371/journal.pone.0046733.

Villumsen KR, Raida MK (2013). Long-lasting protection induced by bath vaccination against *Aeromonas salmonicida* subsp. *salmonicida* in rainbow trout. *Fish and Shellfish Immunology* 35(5): 1649–1653.

Walker RW, Markillie LM, Clotelo AH, Geist DR, Gay ME, Woodley CM, Eppard MB, Brown RS (2013). Ultraviolet radiation as disinfection for fish surgical tools. *Animal Biotelemetry* 1(4): 1–11.

Whittington RJ, Gudkovs N, Carrigan MJ, Ashburner LD, Thurstan SJ (1987). Clinical, microbiological and epidemiological findings in recent outbreaks of goldfish ulcer disease due to atypical *Aeromonas salmonicida* in south-eastern Australia. *Journal of Fish Diseases* 10: 353–362.

Whittington RJ, Djordjevic SP, Carson J, Callinan RB (1995). Restriction endonuclease analysis of atypical *Aeromonas salmonicida* isolates from goldfish *Carassius auratus*, silver perch *Bidyanus bidyanus* and greenback flounder *Rhombosolea tapirina* in Australia. *Diseases of Aquatic Organisms* 22: 185–191.

Wiklund T, Dalsgaard I (1998). Occurrence and significance of atypical *Aeromonas salmonicida* in non-salmonid and salmonid fish species: a review. *Diseases of Aquatic Organisms* 32: 49–69.

Wooster G, Bowser P (1996). The aerobiological pathway of a fish pathogen: survival and dissemination of *Aeromonas salmonicida* in aerosols and its implications in fish health management. *Journal of the World Aquaculture Society* 27(1): 7–14.

34. *Edwardsiella* spp.

34.1. Hazard identification

34.1.1. Aetiological agent

Edwardsiella spp. are a species complex of zoonotic Gram-negative rod-shaped filamentous bacteria currently classified in the Enterobacteriaceae (Farmer & McWhorter 1984) and originally described from humans and snakes (Evans *et al.* 2011). *Edwardsiella tarda* was first reported from Japanese eels (*Anguilla japonica*) as *Paracolobacterium anguillimortiferum* (Hoshinae 1962) and then reclassified as *E. tarda* (Farmer & McWhorter 1984). It is the agent of *Edwardsiella* septicaemia of carp, catfish and eels in fresh waters, but is also reported from marine fish (Evans *et al*. 2011).

Four serotypes were recognised: A, B, C and D, which differ in their responses to environmental conditions (including salinity, temperature and pH tolerance), as well as in their host specificity, target organs, drug resistance and virulence (Park *et al.* 1983).

Recent genomic analysis indicates isolates previously considered as *Edwardsiella tarda* represent a species complex of genetically distinct, but phenotypically ambiguous taxa (*E. ictaluri*, *E. tarda* , *E. piscida*, *E. hoshinae* and *E. anguillarum*) (Reichley *et al.* 2018). The high phenotypic variation and interogenomic heterogeneity within the species complex have resulted in considerable confusion between isolates within the species complex in both host range and distribution. For the purposes of this report these species and isolates are considered together as *Edwardsiella* spp.

34.1.2. OIE status

Infection with *Edwardsiella* spp. is not listed by the OIE (OIE 2016).

34.1.3. New Zealand status

Edwardsiella tarda has twice been isolated in New Zealand, from migratory cetaceans and seals (Stan Fenwick, Massey University, reported in Stone *et al.* 1997). *Edwardsiella* spp. is considered exotic to New Zealand (Duignan *et al.* 2003; Melville & Johnston 2010) but is not listed as a notifiable disease (Anon. 2016).

34.1.4. Epidemiology

Edwardsiella septicaemia is an economically significant and zoonotic disease (Park *et al*. 2012) affecting wild and farmed freshwater fish from over 30 countries, including Africa (Democratic Republic of the Congo, South Africa), Antarctica, Asia (Japan, South Korea, Singapore, Taiwan, Thailand), Australia, India, North America (United States), South America (Venezuela) and Europe.

Edwardsiella spp. has a wide host range, with *E. tarda* reported from 20 families and at least 40 species of marine and freshwater fish, including eels, channel catfish and salmonids (see Table 31) (Miyazaki & Egusa 1976; Evans *et al*. 2011; Park *et al*. 2012). It also occurs in other vertebrates (including amphibians, reptiles, birds, cattle, swine and marine mammals) and in marine and freshwater invertebrates (Muratori *et al.* 2000; Evans *et al*. 2011; CABI 2015). Experimental

infection of climbing perch *Anabas testudinensis* (Anabatidae), by injection is also reported (Sahoo *et al.* 2000).

Edwardsiella spp. are zoonotic, but rarely cause significant disease (Hirai *et al.* 2015). However, *Edwardsiella tarda* bacteraemia (ETB) causes severe gastroenteritis and extra intestinal infection including endocarditis, empyema, hepatobiliary infections, peritonitis, intra-abdominal abscesses, osteomyelitis, wound infections and meningitis (Bockemuhl *et al.* 1971; Lowry & Smith 2007; Evans *et al.* 2011), with clinical signs and risk factors similar to infection with *Aeromonas* spp., typhoid fever (*Salmonella enterica* serotype Typhi) and *Vibrio vulnificus* infection. ETB infection may be fatal, with an overall mortality rate of 44.6% (from 72 reviewed cases). This mortality rate increased to 61% for patients with liver cirrhosis (Hirai *et al.* 2015). ETB occurs in Europe and Japan, but has not been reported from the southern hemisphere (Hirai *et al.* 2015).

Other mammals including pigs may also be infected. Infection generally occurs following the consumption of under-cooked contaminated fish products (Anon. 2011; Sun *et al.* 2012), but skin infections also occur in factory workers who process infected fish product (Evans *et al.* 2011).

Edwardsiella spp. is a widespread opportunistic non-obligate zoonotic pathogen. It was recovered from 88% of processed catfish fillets in United States aquaculture, and was present in 30% of fillets imported into the United States (Evans *et al.* 2011).

Table 31. Families and Species of Fish Susceptible to *Edwardsiella* spp.

Family	Species
Anabantidae	Climbing perch (*Anabas testudineus*)
Anguillidae	Australian eel (*Anguilla reinhardtii*), European eel (*A. anguilla*), Japanese eel (*A. japonica*)
Batrachoididae	Toadfish (*Barchatus cirrhosus*), oyster toadfish (*Opsanus tau*)
Carangidae	Yellowtail (*Seriola quinqueradiata*)
Centrarchidae	Largemouth bass (*Micropterus salmoides*), smallmouth bass (*M. dolomieu*)
Channidae	Spotted snakehead (*Channa punctata*)
Cichlidae	Blue tilapia (*Oreochromis aureus*), Malawi blue cichlid (*Maylandia zebra*), Mozambique tilapia (*O. mossambicus*), Nile tilapia (*O. niloticus*), red tilapia (*O. niloticus* hybrid)
Clariidae	North African catfish (*Clarias gariepinus*),walking/Asian catfish (*C. batrachus*)
Cyprinidae	Common and ornamental carp (*Cyprinus carpio*), emerald shiner (*Notropis atherinoides*), goldfish (*Carassius auratus*), grass carp (*Ctenopharyngodon idella*), Indian carp (*Gibelion catla*), rohu (*Labeo rohita*)
Dasyatidae	Round ribbontail ray (*Taeniurops meyeni*)
Eleotridae	Marble goby (*Oxyeleotris marmorata*)
Gobiidae	Sand goby (*Pomatoschistus minutus*)
Haemulidae	Blue striped grunt (*Haemulon sciurus*)
Ictaluridae	Brown bullhead (*Ameiurus nebulosus*), channel catfish (*Ameiurus punctatus*)
Latidae	Barramundi (*Lates calcarifer*)
Moronidae	European sea bass (*Dicentrarchus labrax*), striped bass (*Morone saxatalis*)
Mugilidae	Mullet (*Mugil cephalus*)
Osphronemidae	Three-spot gourami (*Trichogaster trichopterus*)
Paralichthyidae	Bastard halibut (*Paralichthys olivaceus*)
Pleuronectidae	American flounder (*Pleuronectes putnami*), winter flounder (*Pseudopleuronectes americanus*)
Salmonidae	Atlantic salmon (*Salmo salar*), brook trout (*Salvelinus fontinalis*), brown trout (*Salmo trutta*), Chinook salmon (*Oncorhynchus tshawytscha*), rainbow trout (*O. mykiss*)
Scophthalmidae	Turbot (*Scophthalmus maximus*)
Serranidae	White grouper (*Epinephelus aeneus*)
Siluridae	Far Eastern catfish (*Silurus asotus*), Wels catfish/sheatfish (*S. glanis*)
Sparidae	Gilthead sea bream (*Sparus aurata*), crimson sea bream (*Dentex tumifrons*), red sea bream (*Pagrus major*)

Initial entry commonly occurs in fish across the digestive epithelium following ingestion, or through the gills, fin bases and skin, generally because of tissue damage. Mortality may be high (up to 90%) in Bastard halibut (*Paralichthys olivaceus*) in acute infection, with pathological lesions in kidney and liver tissues. Chronic infection is more common, where surviving fish may act as carriers of infection (Evans *et al.* 2011). It may remain inert in the gut lumen, becoming pathogenic only when fish are stressed (Evans *et al.* 2011; CABI 2015). The LD_{50} for the North African catfish (*Clarias gariepinus*) varied from 8.52×10^6 for the PB_B strain, to 1.68×10^7 for the PB_p strain (Abraham *et al.* 2015).

External signs of chronic infection vary widely among host species. Infection may occur with no external signs, while Saleh (2005) found low concentrations of *E. tarda* in the musculature of Nile tilapia (*Oreochromis niloticus*) following IP injection.

Infection may follow either a renal or hepatic pathway. In the renal form, infection of the trunk kidney progresses into abscesses in the spleen, liver, heart and mucosa of the digestive tract, as well as the lateral musculature (Egusa 1976a, Miyazaki & Kaige 1985, Plumb 1999). In the hepatic form, the liver is initially infected, then infection spreads through the hepatocytes and blood vessels into the surrounding tissues (Miyazaki & Kaige 1985). In other fish, small cutaneous lesions, whitish skin patches and petechiae may be present. These may develop into large necrotic skin abscesses, while further disease progression includes the development of abscesses in the spleen, liver, epicardium, stomach, gills and musculature. These abscesses commonly become liquefied and give off a putrid odour when excised, which renders the product unmarketable (Evans *et al.* 2011).

Disease transmission is horizontal, through the water column. Infection occurs at low to medium pathogen concentrations, with the median lethal dose (LD_{50}) ranging from 1.7×10^2 to 1.3×10^6 CFU mL^{-1} (colony forming units) in experimental infection by oral exposure (Baya *et al.* 1997). Infection may also be enhanced by co-infection with protozoans such as *Trichodina* spp., or *Tetrahymena pyriformis* (Evans *et al.* 2011).

Edwardsiella spp. are environmentally stable. *Edwardsiella tarda* survives for at least 76 days in marine and fresh waters and tolerates a wide temperature range, from 45°C (Evans *et al.* 2011) to Antarctic waters (Park *et al.* 2012). It may persist for extended periods in a viable but non-culturable state in sediments and at low temperatures (Evans *et al.* 2011). It is resistant to drying and may be transmitted by aerosol contamination (Anon. 2011). *Edwardsiella tarda* may be found in the intestinal tract of a wide range of aquatic organisms, including piscivorous birds which also act as mechanical vectors of disease (Berg & Anderson 1972). It may become ubiquitous in endemic areas (Buller 2004, Wiedenmayer *et al.* 2006).

Edwardsiella tarda is resistant to cold temperatures, remaining viable after 30–50 days frozen storage at -20° (Melville & Johnston 2010; Brady & Vinitnantharat 2011). Physical inactivation requires freezing (to -80°C), or heat treatment with either moist heat (121°C for 15 minutes), or dry heat (160–170°C for at least 60 minutes) (Anon. 2011).

There are no effective commercially available vaccines, so control of *Edwardsiella* spp. in aquaculture depends mainly on the use of antibiotics (Evans *et al.* 2011).

Susceptible species in New Zealand include salmonids (*Salmo* spp., *Oncorhynchus* spp., *Salvelinus* spp.) and the introduced brown bullhead catfish (*Ameiurus nebulosus*), while marine species include snapper (*Chrysophrys auratus*) (Sparidae), hapuku (*Polyprion oxygeneios*)

(Polyprionidae), flounders (Order Pleuronectiformes) and yellowtail kingfish (*Seriola lalandi*) (Carangidae) (Johnston 2008; Evans *et al.* 2011).

34.2. Risk assessment

34.2.1. Entry assessment

Fish showing clinical signs of infection would not pass visual inspection for the human food consumption pathway, but sub-clinically infected fish, and healthy fish environmentally exposed to *Edwardsiella* spp. may be present in the commodity (Johnston 2008). *E. tarda* remains viable after long-term frozen storage (Brady & Vinitnantharat 2011). If transportation to New Zealand and storage took less than 50 days, then viable *Edwardsiella* spp. could remain in the commodity (Johnston 2008).

The likelihood of entry of *E. tarda* is assessed as non-negligible.

34.2.2. Exposure assessment

Edwardsiella spp. may occur in several marine and freshwater fish species likely to be imported into New Zealand (Table 31). To establish, infected eviscerated product would have to become available for consumption by a susceptible fish host, in sufficient quantity and duration (Kahn *et al*. 1999). *Edwardsiella* spp. may occur in blood-water discharge, in skin and muscle tissues discarded after trimming, as well as in the skin, brain and gill tissues of fish heads discarded as offal.

Edwardsiella spp. remains viable on dry surfaces and may cross-contaminate other product through contact with infected fish processing equipment (Evans *et al.* 2011), or through aerosol contamination (Anon. 2011). The presence of undetected muscle lesions of *Edwardsiella* spp. in catfish can contaminate processing equipment, requiring production to be halted, until the equipment is cleaned (Johnston 2008).

Edwardsiella spp. are non-obligate pathogens, resistant to environmental extremes including freezing. *Edwardsiella tarda* remains viable in discarded fish offal, or discarded wastewater and survives in sediments for at least 76 days (Brady & Vinitnantharat 2011; Evans *et al.* 2011). It is resistant to high temperatures (Anon. 2011) while the infective dose is low to moderate (1.7×10^2 – 1.3×10^6 (Baya *et al.* 1997).

Previous reports of *Edwardsiella* spp. from migratory cetaceans and seals (Fenwick, *unpublished,* reported in Stone *et al.* 997) suggest that alternative pathways for infection may exist. Potential hosts are present in New Zealand (Tubbs *et al*. 2007).

The likelihood of exposure to *Edwardsiella* spp. in the commodity (eviscerated fish) is assessed as non-negligible.

34.2.3. Consequence assessment

Edwardsiella spp. are significant exotic pathogens of several introduced freshwater species, including brown bullhead carp (*Ameiurus nebulosus*), eels (*Anguilla* spp.), rainbow trout (*Oncorhynchus mykiss*) and Chinook salmon (*O. tshawytscha*) (Table 31), as well as several families of commercially important marine fish (CABI 2015).

The consequences of introduction would be extremely serious for the salmonid fishery, which was valued at $63 million in export earnings in 2011 (Aquaculture New Zealand 2014). An outbreak could also affect recreational and tourist trout and salmon fishing, as well as incur significant social and environmental costs associated with other domestic fisheries, including eels (Fish & Game 2014). While the dollar value of New Zealand's freshwater fisheries is not fully known, the Lake Taupo trout fishery alone is valued at $70 to 80 million per annum (Marsh & Mkwra 2013).

Establishment of *Edwardsiella* spp. may negatively impact the developing aquaculture industry for grass carp (*Ctenopharyngodon idella*) and silver carp (*Hypophthalmichthys molitrix*), currently used for weed control in freshwater lakes and rivers, with many of the hatcheries located in northern New Zealand waters (Clayton & Wells 1999; NIWA 2014a).

Edwardsiella spp. may also infect wild stocks of several commercially important inshore marine fish (Tubbs *et al.* 2007; Evans *et al.* 2011; MPI 2014). These include snapper (*C. auratus*), with exports valued at NZ$ 6.5 million and jack mackerel (*Trachurus* spp.) valued at NZ$ 64 million in exports in 2012 (Anon. 2014).

The introduction of *Edwardsiella* spp. may have social and economic consequences for other lower value fish, such as grey mullet (*M. cephalus*). These species support several inshore target commercial fishery, as well as significant recreational and traditional Maori fisheries (MPI 2014; NIWA 2014b). Any reduction in abundance due to disease would be likely to have major social and environmental costs.

Edwardsiella spp. are considered zoonotic, but rarely cause significant disease (Hirai *et al.* 2015). Most infection results in local skin lesions and minor gastroenteritis. Infection may also be chronic, particularly in immunocompromised or otherwise at-risk patients (such as with liver cirrhosis) (Hirai *et al.* 2015). Here, systemic infection with *Edwardsiella tarda* bacteraemia (ETB) results in severe gastroenteritis, with extra intestinal infection including endocarditis, empyema, hepatobiliary infection, peritonitis, intra-abdominal abscesses, osteomyelitis, consequential wound infections and meningitis (Bockemuhl *et al*. 1971; Lowry & Smith 2007; Evans *et al.* 2011). ETB infection has a mortality rate from 44 %, rising to 61% for patients with liver cirrhosis (Hirai *et al.* 2015). ETB is not reported from the southern hemisphere (Hirai *et al.* 2015).

The control or eradication of *Edwardsiella* spp. in marine or freshwater fisheries would be ineffective (Kahn *et al*. 1999).

The consequences of establishment of *Edwardsiella* spp. are assessed as non-negligible.

34.2.4. Risk estimation

Johnston (2008) considered for frozen, skin-off fillets of tilapia (*Oreochromis niloticus*), that the likelihood of exposure of *E. tarda* was negligible, due in part to the low titre in skeletal musculature (Saleh 2005). In the present assessment, the entry, exposure and resulting consequence assessments for *Edwardsiella* spp. in eviscerated fish are non-negligible, so the risk estimate is non-negligible. Therefore, under the procedures followed in this risk assessment, *Edwardsiella* spp. is assessed to be a risk in the commodity and risk management measures may be developed.

34.3. Risk management

Edwardsiella spp. have been assessed to be a risk in the commodity. Infection with *Edwardsiella* spp. is a non-notifiable disease, so there is no specific guidance in the OIE *Aquatic Code* (OIE 2016) for importing eviscerated fish from infected countries, or specific processing requirements that would ensure the destruction of the pathogen. It is assumed that compliance with the approved processed states for OIE-listed disease (Appendix 3) would also eliminate *Edwardsiella* spp. from the commodity and be a viable risk management option.

Edwardsiella spp. may be present in over 20 families of wild and farmed marine and fresh water fish (Table 31) that may be present in the commodity. Species declaration should substantially reduce the pathogen load of *Edwardsiella* spp. in the commodity and be a viable risk management option. Restriction of the commodity to wild-caught fish (not from aquaculture) would have little, or no effect on pathogen load and is not a viable risk management option.

Infection with *Edwardsiella* spp. is reported from marine and fresh waters of Africa (Democratic Republic of the Congo, South Africa), Antarctica, Asia (Japan, Korea, Singapore, Taiwan, Thailand), Australia, India, North America (United States), South America (Venezuela) and Europe (Evans *et al*. 2011; Park *et al*. 2012). Where country/zone freedom is approved through the MPI Country Approval Procedures, this option is likely to substantially reduce the pathogen load of *Edwardsiella* spp. in the commodity. Approval of a declaration of country/zone freedom by MPI is a viable risk management option.

Edwardsiella spp. are likely to be associated with the gills and epicardium of carrier fish. Removal of the gills, or the head and gills, should slightly reduce the pathogen load of *Edwardsiella* spp. (Johnston 2008). Further processing to the skin-off fillet state should moderately reduce the pathogen load of *Edwardsiella* spp. in the commodity and be a viable risk management option.

Edwardsiella spp. are unaffected by frozen storage, with *E. tarda* requiring freezing (to -80°C) for deactivation (Anon. 2011). Low-temperature storage is not a viable risk management option.

High-temperature inactivation requires cooking (to at least 121°C, with moist heat, for 15 minutes), which is likely to eliminate *Edwardsiella* spp. from the commodity. High-temperature treatment is a viable risk management option.

No vaccine for *Edwardsiella* spp. is commercially available (Evans *et al*. 2011), so vaccination is not a risk management option.

34.3.1. Risk management options

Edwardsiella spp. are reported from fish in families Anabantidae, Anguillidae, Batrachoididae, Carangidae, Centrarchidae, Channidae, Cichlidae, Clariidae, Cyprinidae, Dasyatidae, Eleotridae, Gobiidae, Haemulidae, Ictaluridae, Latidae, Moronidae, Mugilidae, Osphronemidae, Paralichthyidae, Pleuronectidae, Salmonidae, Scophthalmidae, Serranidae, Siluridae and Sparidae (Table 31). These families are considered likely to be present in the commodity. Other families have not been associated with *Edwardsiella* spp. Therefore species declaration indicating the commodity is not originated from any of the above families should substantially reduce the occurrence of *Edwardsiella* spp. in the commodity.

For the commodities originated from families associated with *Edwardsiella* spp., one or a combination of the following additional options could also be considered to effectively manage the risk.

Where country/zone freedom from *Edwardsiella* spp. is accepted by MPI:

Option 1

Acceptance of country/zone freedom by MPI should substantially reduce the occurrence of *Edwardsiella* spp., so the commodity may be imported without any further restrictions.

Where country/zone freedom from *Edwardsiella* spp. is not accepted by MPI or not available:

Option 2

Processing to a state consistent with the approved states for OIE-listed diseases (Appendix 3) should eliminate *Edwardsiella* spp. When these provisions are met, the commodity could be imported without any further restrictions.

Where the imported commodity is not in a state consistent with the approved states for OIE-listed diseases (Appendix 3):

Option 3

Heat treatment (by cooking, in moist heat, to at least 121°C for at least 15 minutes) should eliminate *Edwardsiella* spp. When this provision is met, the commodity could be imported without any further restrictions; or,

Option 4

Further processing (to the skin-off fillet state) should moderately reduce the occurrence of *Edwardsiella* spp. When this provision is met, the commodity could be imported without any further restrictions.

34.4. References

Abraham TJ, Mallick PK, Adikesavalu H, Banerjee S (2015). Pathology of *Edwardsiella tarda* infection in African catfish, *Clarias gariepinus* (Burchell 1822), fingerlings. *Archives of Polish Fisheries* 23: 141–148.

Anonymous (2011). *Edwardsiella tarda*. Pathogen safety data sheets – infectious substances. Public Health Agency of Canada, Ottawa, Canada, 5 p [Online] Available from: http://www.phac-aspc.gc.ca/lab-bio/res/psds-ftss/edwardsiella-tarda-eng.php [Accessed 8 August 2015].

Anonymous (2014). New Zealand's seafood industry [Online] Available from: http://www.stats.govt.nz/tools_and_services/newsletters/price-index-news/apr-13-article-seafood.aspx [Accessed 6 April 2015].

Anonymous (2016). Biosecurity (Notifiable Organisms) Register 2016. NZLR 2016/73. New Zealand Government, Wellington, New Zealand, 13 p.

Aquaculture New Zealand (2014). Seafood New Zealand: Key Facts for Our Industry 2013 [Online] Available from: http://aquaculture.org.nz/industry/overview/ [Accessed 8 June 2014].

Baya AM, Romalde JL, Green DE, Navarro RB, Evans JJ, May EB, Toranzo AE (1997). Edwardsiellosis in wild striped bass from the Chesapeake Bay. *Journal of Wildlife Diseases* 33: 517–525.

Berg RW, Anderson AW (1972). *Salmonellae* and *Edwardsiella tarda* in gull feces: a source of contamination in fish processing plants. *Applied Microbiology* 24: 501–503.

Bockemuhl, J, Pan-Urai R, Burkhardt F (1971). *Edwardsiella tarda* associated with human disease. *Pathologia et Microbiologia* 37: 393–401.

Brady YJ, Vinitnantharat S (2011). Communications: Viability of bacterial pathogens in frozen fish. *Journal of Aquatic Animal Health* 2(2): 149–150.

Buller NB (2004). Bacteria from Fish and Other Aquatic Animals: A Practical Identification Manual. CABI International; Wallingford, Oxfordshire, (1st edition), 361 p.

CABI (2015). *Edwardsiella* septicaemia (*Edwardsiella tarda* infection). Pathogen data sheet. CABI International, Wallingford, United Kingdom [Online] Available from: http://www.cabi.org/isc/datasheet/84398 [Accessed 12 August 2015].

Clayton JS, Wells RDS (1999). Some issues in risk assessment reports on grass carp and silver carp. *Conservation Advisory Science Notes* No. 257. Department of Conservation, Wellington, 16 p.

Duignan PJ, Hine PM, Joy M, Gibbs N, Jones GW, Okeoma C (2003). Disease surveillance in freshwater fish from the lower North Island. *Surveillance* 30(3): 6–8.

Egusa S (1976). Some bacterial diseases of freshwater fishes in Japan. *Fish Pathology* 10:103–114.

Evans JJ, Klesius PH, Plumb JA, Shoemaker CA (2011). *Edwardsiella* septicaemias. pp 512–569. *In:* Woo PTK, Bruno DW (*Eds.*). *Fish Diseases and Disorders: Volume 3 - Viral, Bacterial and Fungal Infections*, 2nd edition. CABI International, Wallingford, United Kingdom, 930 p.

Farmer JJ, McWhorter AC (1984). Genus X. *Edwardsiella*, pp 486–491 *In:* Kersters K, Vanacanneyt M (*Eds.*) Bergey's Manual of Systematic Bacteriology. Baltimore, Maryland, United States of America.

Fish and Game (2014). New Zealand Fish and Game Council [Online] Available from: http://www.fishandgame.org.nz/ [Accessed 24 September 2014].

Harai Y, Ashata-Tago S, Ainoda Y, Fujita T, Kikuchi K (2015). *Edwardsiella tarda* bacteremia. A rare but fatal water- and foodborne infection: Review of the literature and clinical cases from a single centre. *Canadian Journal of Infectious Diseases and Medical Microbiology* 26(6): 313–318 [Online] Available from https://www.ncbi.nlm.nih.gov/pmc/articles/PMC4692300 [Accessed 26 June 2018].

Hoshinae T (1962). On a new bacterium, *Paracolobacterium anguillimortiferum* sp. nov. *Bulletin of the Japanese Society of Scientific Fisheries* 28: 162–164.

Johnston C (2008). Import risk analysis: Frozen, skinless and boneless fillet meat of *Oreochromis* spp. from China and Brazil for human consumption. MAF Biosecurity New Zealand, Ministry of Agriculture and Forestry, Wellington, New Zealand, 96 p.

Kahn SA, Beers PT, Findlay VL, Peebles IR, Durham PJ, Wilson DW, Gerrity SE (1999). Import risk analysis on non-viable salmonids and non-salmonid marine finfish. Australian Quarantine and Inspection Service, Department of Agriculture, Fisheries and Forestry, Canberra, Australia, 409 p.

Lowry T, Smith SA (2007). Vet Med Today: Zoonosis Update. *Journal of the Aquuatic Veterinary Medical Association* 6: 876–880.

Marsh D, Mkwra L (2013). Review of freshwater non-market value studies. Department of Economics, University of Waikato, Hamilton, New Zealand, 144 p.

Melville V, Johnston C (2010). Supplementary Risk Analysis: Extension of countries eligible to export frozen, skinless, boneless fillet meat of *Pangasius* spp. under the import health standard fisfilic.spe. Ministry of Agriculture and Forestry, Wellington, New Zealand, 15 p.

Ministry for Primary Industries (MPI) (2014). Grey mullet. pp 364–369. *In:* Fisheries Assessment Plenary, May 2014: stock assessments and stock status. Compiled by the Fisheries Science Group, Ministry for Primary Industries (MPI), Wellington, New Zealand, 1381 p.

Muratori MCS, De Oliveira AL, Ribeiro LP, Leite RC, Costa APR, Da Silva MCC (2000). *Edwardsiella tarda* isolated in integrated fish farming. *Aquaculture Research* 31(6): 481–483.

Miyazaki T, Egusa S (1976). Histopathological studies of Edwardsiellosis of the Japanese eel (*Anguilla japonica*)-I. Suppurative interstitial nephritis form. *Fish Pathology* 11: 33–43.

Miyazaki T, Kaige N (1985). Comparative histopathology of edwardsiellosis in fishes. *Fish Pathology* 20: 219–227.

NIWA (2014a). Grass carp *Ctenopharyngodon idella* (Valenciennes, 1844) [Online] Available from: https://www.niwa.co.nz/freshwater-and-estuaries/nzffd/NIWA-fish-atlas/fish-species/grass_carp [Accessed 12 February 2015].

NIWA (2014b). Grey mullet [Online] Available from: https://www.niwa.co.nz/freshwater-and-estuaries/nzffd/NIWA-fish-atlas/fish-species/atlantic_salmon [Accessed 28 April 2015].

OIE (2016). Aquatic Animal Health Code, 19th Edition. World Organization for Animal Health. Paris, France [Online] Available from: http://www.oie.int/international-standard-setting/aquatic-code/access-online/ [Accessed 15 March 2017].

Park SB, Takashi A, Jung TS (2012). Pathogenesis of and strategies for preventing *Edwardsiella tarda* infection in fish. *Veterinary Research* 43(67), 11 p [Online] Available from: http://www.veterinaryresearch.org/content/43/1/67 [Accessed 8 August 2015].

Park SI, Wakabayashi H, Watanabe Y (1983). Serotypes and virulence of *Edwardsiella tarda* isolated from eels and their environment. *Journal of Fish Pathology* 18(2): 85–89.

Plumb JA (1999). Health maintenance and principal microbial diseases of cultured fishes. Iowa State University Press, Ames, Iowa, United Sates of America, 328 p.

Reichley SR, Ware C, Steadman J, Gaunt PS, Garcia JC, LaFrentz BR, Thachil A, Waldbieser JC, Stine CB, Arias CR, Loch T, Welch TJ, Cipriano RC, Greenway TE, Khoo1 LH, Wise DJ, Lawrence ML, Griffin MJ (2018). Comparative phenotypic and genotypic analysis of *Edwardsiella* spp. isolates from different hosts and geographic origins, with an emphasis on isolates formerly classified as *E. tarda* and an evaluation of diagnostic methods. *Journal of Clinical Microbiology* [Online] Available from: http://www. doi:10.1128/JCM.00970-17 [Accessed 25 June 2018].

Sahoo PK, Swain P, Sahoo SK, Mukherjee SC, Sahu AK (2000). Pathology caused by the bacterium *Edwardsiella tarda* in *Anabas testudineus* (Bloch). *Asian Fisheries Science* 13: 357–362.

Saleh WD (2005). Isolation and identification of *Edwardsiella tarda* from infected Nile tilapia fish "*Oreochromis niloticus*". *Bulletin of Faculty of Agriculture, Cairo University* 56(4): 839–846.

Sun Y, Zheng WJ, Hu YH, Sun BG, Sun L (2012). *Edwardsiella tarda* Eta1, an *in vivo*-induced antigen that is involved in host infection. *Infection and Immunology* 80(8): 2948–2955.

Tubbs L, Lee P, Diggles B, Jones JB, Shepherd M, Sim-Smith C (2007). A review of aquatic diseases of significance to New Zealand. Final research report for project ZBS 2005-17. National Institute of Water and Atmospheric Research, Wellington, New Zealand, 452 p.

Wiedenmayer AA, Evans JJ, Klesius PH (2006). Experimental *Edwardsiella tarda* infection in non-abraded channel catfish *Ameiurus punctatus* by immersion. *Fisheries Science* 72(5): 1124–1126.

35. *Flavobacterium columnare* (exotic strains)

35.1. Hazard identification

35.1.1. Aetiological agent

Flavobacterium columnare is a Gram-negative rod-shaped filamentous bacterium currently classified in the Flavobacteriaceae (Bernardet *et al.* 1996), which was first recovered from sockeye salmon (*Oncorhynchus nerka*) (Ordal & Rucker 1944). It has previously been classified as *Flexibacter columnaris*, *Bacillus columnare*, *Chondrococcus columnaris* and *Cytophaga columnaris* (Starliper & Schill 2011). It is the agent of freshwater columnaris disease (Lafrentz *et al.* 2014).

Three genomovars of *F. columnare* are recognised: Genomovars I, II, and III, which differ in their responses to environmental conditions (such as salinity, temperature and pH tolerance), as well as in their host specificity, drug resistance and virulence (Tekedar *et al.* 2012; Cai *et al.* 2013; Lafrentz *et al.* 2014).

Genomovar I represents the main agent of columnaris disease and contains the most widespread isolates (Kunttu 2010). While genomovars I, II, and III are reported from Asia and in the USA (Michel *et al.* 2002; Kunttu 2010; Lafrentz *et al.* 2014), only genomovars I and II have been found in Europe (Kunttu 2010) and in India (Verma & Rathore 2013). The highly pathogenic isolates are included in genomovar II (Lafrentz *et al.* 2014).

35.1.2. OIE status

Infection with *Flavobacterium columnare* is not notifiable to the OIE (OIE 2016).

35.1.3. New Zealand status

Flavobacterium columnare is present in New Zealand (Diggles 2011) and columnaris disease has been reported from ornamental fish and salmon aquaculture (Anderson 1996; Duignan *et al.* 2003). However, the virulent infection characteristic of genomovar II (Michel *et al.* 2002; Kunttu, 2010) has not been reported, and this genomovar is considered exotic (Anderson 1996; Tubbs *et al.* 2007; B. Jones, *pers. comm.* 2015).

Genomovar II is included in this assessment because of the wide species range of possible hosts, together with the higher virulence and the severity of the disease caused by the isolates of this group (Michel *et al.* 2002; Tubbs *et al*. 2007; Johnston 2008a, 2008b).

Columnaris disease reported from marine fish is caused by *Tenacibaculum maritimum* (=*Flexibacter maritimus*, or *Cytophaga marina*) (FAO 2014), which is considered ubiquitous in marine waters (Kahn *et al.* 1999; Diggles 2011; C. Johnston, *pers. comm.* 2015). It is not considered further.

35.1.4. Epidemiology

Flavobacterium columnare is a ubiquitous, opportunistic pathogen affecting a wide range of freshwater fish, including salmonids, eels and cichlids (Plumb & Hanson 2011; Starliper & Schill 2011), in a wide temperature range (from 12 to 30°C), with some isolates able to survive in temperatures up to 37°C (Starliper & Schill 2011).

Freshwater columnaris disease, also known as fin rot or tail rot (Starliper & Schill 2011), is attributed to genomovars I and III of *F. columnare* (Table 33), which cause cumulative mortality of up to 60% and continuing economic losses in channel catfish (*Ictalurus punctatus*) and trout (*Oncorhynchus* spp., *Salmo* spp.) aquaculture in the United States (Lafrentz *et al.* 2014; Loch & Faisal 2014); carp (*Gibelion catla*) aquaculture in India (Verma *et al.* 2011; Verma and Rathore 2013); and cichlid (*Oreochromis* spp.) aquaculture in Asia, despite the widespread use of antibiotics (Dong *et al.* 2014).

Columnaris disease initially infects epidermal cells causing necrotic skin lesions, which progressively develop into red-orange coloured ulcers (Decostere 2002). Lesions may also form on the fins (fin rot), gills (gill rot), or along the dorsal fin (saddleback lesion), but these progressively develop into systemic ulcerative dermatitis that eventually invades the dermal musculature (Decostere 2002), commonly resulting in death through loss of osmotic integrity (Declercq *et al.* 2013). The disease progression for the low pathogenic strains (genomovar I and genomovar III) is slow, with clinical signs developing over several days or weeks post-infection.

Fish of all ages can be infected, but pathogenicity is influenced by salinity, pH and host species (Starliper & Schill 2011; Verma *et al.* 2011; Meepagala *et al.* 2013), as well as by external factors including water quality, stocking levels, or water temperature (Buller 2004; Hine & Diggles 2005; Starliper & Schill 2011; Tekedar *et al.* 2012). Genomovar I isolates infect fish in cold and temperate waters, but virulence is generally lower in colder waters (Lafrentz *et al.* 2012).

Infection with genomovar II isolates have been reported from the United States, India and Asia (Michel *et al.* 2002; Verma & Rathore 2013; Mohammed & Arias 2014). Disease progression is similar to the lower virulence isolates, but is characterised by high mortality (typically 75–100%). The onset of disease is rapid, particularly if fish are stressed under poor husbandry conditions and death often occurs within 48 hours of exposure (Olivares-Fuster 2010).

Genomovar II isolates have been reported mainly from warm water freshwater fish including the families Centrarchidae, Cichlidae, Cyprinidae and Percidae (Table 32) (Fujihara & Nakatani 1971; Anderson & Thomas 1995; Bernardet *et al.* 1996; Michel *et al.* 2002; Arias *et al.* 2004; Figueiredo *et al.* 2005; Olivares-Fuster *et al.* 2007; Herbert & Graham 2008; Johnston 2008a, 2008b; Starliper & Schill 2011; Verma *et al.* 2011; Declercq *et al.* 2013; Verma & Rathore 2013; Dong *et al.* 2014 Mohammed & Arias 2014).

Salmonids (*Salmo* spp., *Oncorhynchu*s spp., *Salvelinus* spp.) are naturally infected only with genomovar I, but rainbow trout (*O. mykiss*) may be experimentally infected with genomovar II (Lafrentz *et al.* 2012). *Flavobacterium columnare* has also been reported from cichlids (*Oreochromis* spp., *Sarotherodon* spp., *Tilapia* spp.), yellow perch (*Perca flavescens*), golden perch (*Macquaria ambigua*) and barramundi (*Lates calcarifer*) (Herbert & Graham 2008). These infections have been reasonably assumed to include genomovar II isolates (Johnston 2008a, 2008b, Melville and Johnston 2010) but available disease descriptions do not provide definitive genomic data for these species.

Epidemiological analysis of genomovar II infection in channel catfish (*Ictalurus punctatus*), blue catfish (*I. furcatus*), bluegill (*Lepomis macrochirus*), Nile tilapia (*O. niloticus*) and largemouth bass (*Micropterus salmoides*), in United States aquaculture suggests that host species is the least influential parameter for infection (Mohammed & Arias 2014). The higher virulence attributed to genomovar II isolates appears to be related to a more efficient initial infection of the host through better adhesion and higher persistence at the site of infection, as well as to higher levels of chemotaxis, when compared to the other less virulent genomovars (Decostere *et al.* 2001, Decostere 2002; Declercq *et al.* 2013).

Table 32. Families, Species and Location of Fish Susceptible to *Flavobacterium Columnare,* Ordered by Genomovar

Family	Species	Genomovar	Location
Anguillidae	Japanese eel (*Anguilla japonica*)	I	Japan
Clupeidae	Threadfin shad (*Dorosoma petenense*)	I	United States
Cyprinidae	Grass carp (*Ctenopharyngodon idella*)	I	China
Ictaluridae	Black bullhead (*Ameiurus melas*)	I	France
Percidae	Yellow perch (*Perca flavescens*)	I	United States
Salmonidae	Brown trout (*Salmo trutta*)	I	France
Salmonidae	Rainbow trout (*Oncorhynchus mykiss*), Chinook salmon (*O. tshawytscha*)	I	United States
Serrasalmidae	Tambaqui, pacu (*Colossoma macropomum*)	I	Brazil
Centrarchidae	Largemouth bass (*Micropterus salmoides*), bluegill (*Lepomis macrochirus*)	II	United States
Cichlidae	Nile tilapia (*Oreochromis niloticus*)	II	Brazil
Cichlidae	Red tilapia (O. *mossambicus* x *O. niloticus*)	II	Thailand
Cichlidae	Cichlid (*Oreochromis* spp., *Sarotherodon* spp., *Tilapia* spp.)	II?	Asia
Clariidae	Walking catfish *Clarias batrachus*)	II	Thailand, India
Cyprinidae	Common and ornamental carp (*Cyprinus carpio*), Indian carp (*Gibelion catla*)	II	Thailand, India
Cyprinidae	Golden shiner (*Notemigonus crysoleucas*)	II	United States
Cyprinidae	Grass carp (*Ctenopharyngodon idella*)	II	China
Ictaluridae	Channel catfish (*Ictalurus punctatus*), blue catfish (*I. furcatus*)	II	United States
Latidae	Barramundi (*Lates calcarifer*)	II?	Australia, Asia
Pangasiidae	Striped catfish (*Pangasianodon hypothalamus*)	II	Vietnam
Percichthyidae	Golden perch (*Macquaria ambigua*)	II?	Australia
Percidae	Yellow perch (*Perca flavescens*)	II?	United States
Salmonidae	Rainbow trout (*Oncorhynchus mykiss*), Chinook salmon (*O. tshawytscha*)	II*	United States
Serrasalmidae	Tambaqui , pacu (*Colossoma macropomum*)	II	Brazil
Acipenseridae	Chinese sturgeon (*Acipenser sinensis*)	III	China
Bryconidae	Matrinxa (*Brycon amazonicus*)	III	Brazil
Cyprinidae	Grass carp (*Ctenopharyngodon idella*)	III	China
Ictaluridae	Channel catfish (*I. punctatus*), blue catfish (*I. furcatus*)	III	United States
Percidae	Chinese perch (*Siniperca chuatsi*)	III	China
Plecoglossidae	Ayu (*Plecoglossus altivelis*)	III	Japan
Salmonidae	Rainbow trout (*Oncorhynchus mykiss*), Chinook salmon (*O. tshawytscha*)	III	United States

* by experimental infection
? lacks definitive genomic information

Genetic variation may improve resistance as hybrid catfish ((*I. punctatus* x *I. furcatus*) appear more resistant to genomovar II infection than either parent (Arias *et al.* 2012), while genomovar

classification appears unrelated to mortality of hybrid tilapia (*O. niloticus* x *O. aureus*) in experimental infection (Shoemaker & Lafrentz 2015).

Disease transmission is direct and horizontal through the water column (Starliper & Schill 2011), with carrier rainbow trout releasing up to 5 x 10^3 cfu mL^{-1} mainly through the gill tissues (Fujihara & Nakatani 1971). Entry into the host occurs mainly through the epidermal cells of the skin and gills (Declercq *et al.* 2013), although infection rates are facilitated by injuries and abrasions (Starliper & Schill 2011).

F. columnare is infective at low concentrations, as exposure of Chinook salmon (*O. tshawytscha*) for 25 minutes at 2.5 x 10^5 cfu mL^{-1} (colony forming units) resulted in 100% mortality (Fujihara & Nakatani 1971). Reported LD_{50} range from 1.3 x 10^3 cfu mL^{-1} for barramundi (*Lates calcarifer*), to 1.7 x 10^5 cfu mL^{-1} for goldfish (*Carassius auratus*) (Bernardet *et al.* 1996) or $10^{4.5}$ cfu mL^{-1} for walking catfish *Clarias batrachus* (Verma *et al.* 2011). Wild stocks may function as disease reservoirs (Austin & Austin 2007), but their effect on the infection rates of farmed fish is unproven (Reynard *et al.* 2007).

Chronically infected and surviving fish function as carriers, usually with few external signs of infection (Starliper & Schill 2011; Kunttu *et al.* 2012). Infected rainbow trout (*Oncorhynchus mykiss*) may release bacteria at a rate of 5 x 10^3 cfu mL^{-1} (Fujihara & Nakatani 1971; Hawke & Thun 1992). *Flavobacterium columnare* also proliferates on moribund and dead fish, which spread infection on average 10x higher than living fish (Kunttu 2010, Starliper & Schill 2011; Declercq *et al.* 2013).

F. columnare is environmentally stable in conditions likely to be encountered in fish processing and storage. It is unaffected by freezing (to -20°C) and remains viable in chilled fish product (Suomalainen 2005). It can persist in biofilms found in processing facilities, which may represent a source of cross-infection (Kunttu 2010). The biofilm provides protection from desiccation, as well as protection from the toxic effects of antibiotic treatments (Cai *et al.* 2013). *Flavobacterium columnare* can form resistant microcysts that can survive in fresh water without nutrients for over two years, while retaining infectivity for at least 5 months (Kunttu *et al.* 2012). It can remain viable in sediments for up to five months (Schachte 1983; Declercq *et al.* 2013; Lanto *et al.* 2014).

Flavobacterium columnare cannot survive in salinities greater than 5%, or in marine waters (Kunttu 2010; Starliper & Schill 2011). It is inactivated by heat treatment of at least 65°C for 25 minutes (Decostere *et al.* 2001), as well as by antibiotics including sulphonamides, florfenicol and oxytetracycline (Declercq *et al*. 2013).

Vaccines developed for genomovar I disease in salmonids are available, but have limited effectiveness on genomovar II isolates (Olivares-Fuster &Arias 2012). *Flavobacterium* spp. are not considered zoonotic (Sudheesh *et al.* 2012).

Flavobacterium columnare genomovar II is environmentally stable and infective at low pathogen density. The potential host range includes almost all New Zealand freshwater fish.

35.2. Risk assessment

35.2.1. Entry assessment

Genomovar II of *F. columnare* causes systemic infection of the gills, skin and musculature of infected fish (Hawke & Thun 1992; Olivares-Fuster *et al.* 2011; Starliper & Shill 2011). Most

infected fish would not pass visual inspection, but carrier fish with little or no external signs of infection may be present in the commodity (Olivares-Fuster *et al.* 2011).

Flavobacterium columnare survives in dead fish tissues (Kunttu 2010, Starliper & Schill 2011; Declercq *et al.* 2013) and is unaffected by freezing or cold storage (Suomalainen 2005; Johnston 2008a, 2008b). It persists in biofilms and may be spread through contamination of processing equipment (Kunttu 2010).

The likelihood of entry is assessed as non-negligible.

35.2.2. Exposure assessment

Flavobacterium columnare genomovar II may be present in fresh, chilled and frozen eviscerated fish. To establish infection, infected eviscerated product would have to become available for consumption by a susceptible fish host, in sufficient quantity and duration (Kahn *et al.* 1999). It may be present in the blood-water discharge and in the gills, skin and muscle tissue trimmings discarded as offal following commercial fish processing, as well as in the storage and packaging materials, including ice used to transport the commodity (Munday 2002). It may be transferred during processing by cross-contamination of plant and equipment (Munday 2002). It can remain viable in discarded organic material and wastewater for at least 5 months and may survive in a resistant microcyst form in aquatic sediments for over 90 days (Schachte 1983; Kunttu *et al.* 2012; Declercq *et al.* 2013).

Flavobacterium columnare is an opportunist pathogen and genomovar I isolates are already present in New Zealand (Anderson 1996; Duignan *et al.* 2003). The infective dose required to initiate infection is low (Fujihara & Nakatani 1971) and *F. columnare* can survive in fresh water outside of its host for extensive periods (Schachte 1983; Declercq *et al.* 2013; Lanto *et al.* 2014).

Genomovar II strains are essentially pathogens of warm water fish and their pathogenicity decreases in cooler waters (Lafrentz *et al.* 2012; Mohammed & Arias 2014), so establishment is unlikely in the waters of southern temperate New Zealand. Freshwater salmonid aquaculture for Chinook salmon (*O. tshawytscha*) is essentially confined to the South Island where water temperatures may reach 8-11°C in winter (Hine & Diggles 2005; Tubbs *et al*. 2007).

However, the grass carp (*Ctenopharyngodon idella*) aquaculture industry which has established in the warmer waters (>12°C) of northern New Zealand may be vulnerable to infection. Genomovar II isolates are highly pathogenic to rainbow trout (Lafrentz *et al*. 2012), so salmonid hatcheries for sports fishing in these areas may also be affected.

Flavobacterium columnare infection is generally associated with poor husbandry, overcrowding and high water temperatures (Hine & Diggles 2005). Exposure to genomovar II isolates may occur, where biosecurity is sub-optimal, such as through contamination of water supplies (Sim-Smith *et al.* 2014).

The likelihood of exposure to the exotic isolates of *F. columnare* genomovar II for susceptible species in northern New Zealand waters is assessed as non-negligible.

35.2.3. Consequence assessment

The consequences of the introduction of the exotic isolates of *F. columnare* are likely to be limited, as establishment is considered unlikely in the waters of central and southern New Zealand (Hine & Diggles 2005).

Establishment of genomovar II isolates may negatively impact the developing aquaculture industry for grass carp (*C. idella*) currently used for weed control in freshwater lakes and rivers (Clayton & Wells 1999), as many of the hatcheries are located in northern New Zealand waters.

The consequences of establishment in northern waters are therefore assessed as non-negligible.

35.2.4. Risk estimation

Since the entry, exposure and consequence assessments for the exotic genomovars of *F. columnare* are non-negligible, the risk estimate is non-negligible. Therefore, exotic genomovars of *F. columnare* are assessed to be a risk in the commodity and risk management measures may be justified.

35.3. Risk management

The exotic genomovar II of *F. columnare* has been assessed to be a risk in the commodity. As infection with *F. columnare* is a non-notifiable disease, the OIE *Aquatic Code* (OIE 2016) provides no specific guidance on importing eviscerated fish from infected countries, or specific processing requirements that would ensure the destruction of the pathogen. It is assumed that compliance with the approved processed states for OIE-listed diseases (Appendix 3) would also eliminate the genomovar II isolates of *F. columnare* from the commodity and be a viable risk management option.

Genomovar II isolates of *F. columnare* have been positively identified from several families of fish (Table 32), which may be present in the commodity. Species declaration is likely to substantially reduce the pathogen load of *F. columnare* genomovar II in the commodity and be a viable risk management option.

The genomovar II isolates have been widely reported from Asia, Europe and North and South America, mainly associated with aquaculture. Restriction of the commodity to wild-caught fish (not from aquaculture) would moderately reduce pathogen load and be a viable risk management option.

Where country/zone freedom is approved through the MPI Country Approval Procedures, this option is likely to substantially reduce the pathogen load of the genomovar II isolates of *F. columnare* in the commodity. Declaration of country/zone freedom is a viable risk management option.

Genomovar II isolates of *F. columnare* are likely to be present in the gills, skin and dermal musculature. Removal of the gills, or the head and gills, should result in a slight reduction in pathogen load. Further processing to the skin-off fillet state should moderately reduce pathogen load and be a viable risk management option.

Flavobacterium columnare is unaffected by freezing (to -20°C), so low-temperature storage is not considered a viable management option. *Flavobacterium columnare* is inactivated by heat

treatment (cooking to at least 65°C for 25 minutes), so this heat treatment would eliminate the pathogen from the commodity. Heat treatment is a viable risk management option.

35.3.1. Risk management options

The virulent genomovar II isolates of *Flavobacterium columnare* are reported from fish in families Centrarchidae, Cichlidae, Clariidae, Cyprinidae, Ictaluridae, Latidae, Pangasiidae, Percichthyidae, Percidae, Salmonidae and Serrasalmidae (Table 32), which are considered likely to be present in the commodity. Other families have not been associated with the genomovar II isolates of *F. columnare*. Therefore species declaration indicating the commodity is not originated from any of the above families should substantially reduce the occurrence of the genomovar II isolates of *F. columnare* in the commodity.

For the commodities originated from families associated with the genomovar II isolates of *F. columnare*, one or a combination of the following additional options could also be considered to effectively manage the risk.

Where country/zone freedom from genomovar II isolates of *F. columnare* is accepted by MPI:

Option 1

Acceptance of country/zone freedom by MPI should substantially reduce the occurrence of the genomovar II isolates of *F. columnare*, so the commodity may be imported without any further restrictions.

Where country/zone freedom from genomovar II isolates of *Flavobacterium columnare* is not accepted by MPI or not available:

Option 2

Processing to a state consistent with the approved states for OIE-listed diseases (Appendix 3) should eliminate the genomovar II isolates of *F. columnare*. When these provisions are met, the commodity could be imported without any further restrictions.

Where the imported commodity is not in a state consistent with the approved states for OIE-listed diseases (Appendix 3):

Option 3

Heat treatment (by cooking to 65°C for at least 25 minutes) should eliminate the genomovar II isolates of *F. columnare*. When this provision is met, the commodity could be imported without any further restrictions; or,

Option 4

Further processing (to the skin-off fillet state) should moderately reduce the occurrence of the genomovar II isolates of *F. columnare.* When this provision is met, the commodity could be imported without any further restrictions; or,

Option 5

Restriction of the commodity to wild-caught fish (not from aquaculture) should moderately reduce the occurrence of genomovar II isolates of *F. columnare*. When this provision is met, the commodity could be imported without any further restrictions.

35.4. References

Anderson CD (1996). Distribution of salmonid diseases in New Zealand. *Surveillance* 23 (4): 23–24.

Anderson IG, Thomas A (1995). Recent advances in the bacterial diseases of barramundi. *Proceedings of the Australian Barramundi Farming Workshop, 15–16 June, 1995*. Walkamin, Queensland, 44 p.

Arias CR, Cai W, Peatman E, Bullard SA (2012). Catfish hybrid *Ameiurus puncatus* x *I. furcatus* exhibits higher resistance to columnaris disease than the parental species. *Diseases of Aquatic Organisms* 100: 77–81.

Arias CR, Welker TL, Shoemaker CA, Abernathy JW, Klesius PH (2004). Genetic fingerprinting of *Flavobacterium columnare* isolates from cultured fish. *Journal of Applied Microbiology* 97: 421–428.

Austin B, Austin A (2007). Bacterial fish pathogens: Diseases of farmed and wild fish. Springer, New York, United States of America, 580 p.

Bernardet JFP, Segers M, Vancanneyet M, Berth M, Kersters, K, Vandamme, P. (1996). Cutting a Gordian knot: emended classification and description of genus *Flavobacterium.* Emended description of the family Flavobacteriaceae and proposal of *Flavobacterium hydatis* nom. nov. (Basonym *Cytophaga aquatilis* Strohl and Tait 1978). *Journal of Systematic Bacteriology* 46: 128–148.

Buller NB (2004). Bacteria from Fish and Other Aquatic Animals: A Practical Identification Manual. CABI International, Wallingford. Oxfordshire, United Kingdom, 390 p.

Cai W, De La Fuente L, Arias CR (2013). Biofilm formation by the fish pathogen *Flavobacterium columnare*: Development and parameters affecting surface attachment. *Applied and Environmental Microbiology* 79(18): 5633–5642.

Clayton JS, Wells RDS (1999). Some issues in risk assessment reports on grass carp and silver carp. Conservation Advisory Science Notes No. 257. Department of Conservation, Wellington, 16 p.

Declercq AM, Haesebrouck F, Van den Broek W, Bossier P, Decostere A (2013). Columnaris disease in fish: a review with emphasis on bacterium:host interactions. *Veterinary Research* 44 (27): 1–17.

Decostere A (2002). *Flavobacterium columnare* infections in fish: the agent and its adhesion to gill tissue. *Verhandelingen Koninkliijke Acamie veer Geneeskunde van Belgie* 64(6): 421–430.

Decostere A, Haesebrouk F, Van Driessche E, Charlier G, Ducatelle R (2001). Characterisation of the adhesion of *Flavobacterium columnare* (*Flexibacter columnaris*) to gill tissue. *Journal of Fish Diseases* 22(6): 465–474.

Diggles B (2011). Risk analysis – Aquatic animal diseases associated with domestic bait translocation. Fisheries Research and Development Corporation Project Report 2009/072. Government of Australia, Canberra, Australia.

Dong HT, Lafrentz B, Pirat N, Rodkhum C (2014). Phenotypic characterization and genetic diversity of *Flavobacterium columnare* isolated from red tilapia, *Oreochromis* sp., in Thailand. *Journal of Fish Diseases* [Online]. Available from: http://onlinelibrary.wiley.com/doi/10.1111/jfd.12304/abstract [Accessed 27 April 2015].

Duignan PJ, Hine PM, Joy M, Gibbs N, Jones GW, Okeoma C (2003). Disease surveillance in freshwater fish from the lower North Island. *Surveillance* 30(3): 6–8.

Figueiredo HCP, Klesius PH, Arias CR, Shoemaker CA, Peira DJ, Peixota MTD (2005). Isolation and characterisation of strains of *Flavobacterium columnare* from Brazil. *Journal of Fish Diseases* 28(4): 199–204.

Food and Agriculture Organisation (FAO) (2014). *Lates calcarifer* Block, 1790. Cultured aquatic species information programme. Fisheries and Aquaculture Department, Food and Agriculture Organisation of the United Nations [Online] Available from: http://www.fao.org/fishery/culturedspecies/Lates_calcarifer/en [Accessed 24 March 2014].

Fujihara MP, Nakatani RE (1971). Antibody protection and immune responses of rainbow trout and coho salmon to *Chondrococcus columnaris*. *Journal of the Fisheries Research Board of Canada* 28: 1253–1258.

Hawke JP, Thun RL (1992). Systemic isolation and antimicrobial susceptibility of *Cytophaga columnaris* from commercially reared channel catfish. *Journal of Aquatic Animal Health* 4: 109–113.

Herbert B, Graham P (2008). Tetrahymenosis, columnaris disease and motile aeromonad septicaemia in golden perch, *Macquaria ambigua* (Richardson), from Australia. pp 179–192. *In*: Bondad-Reantaso MG, Mohan CV, Crumlish M, Subasinghe RP (*Eds*.). Diseases in Asian Aquaculture VI. Fish Health Section, Asian Fisheries Society, Manila, Philippines, 505 p.

Hine PM, Diggles BK (2005). Import risk analysis: Ornamental fish. Biosecurity New Zealand, Ministry of Agriculture and Forestry, Wellington, New Zealand, 264 p.

Johnston C (2008a). Import risk analysis: frozen skinless and boneless fillet meat of *Pangasius* spp. fish from Vietnam for human consumption. MAF Biosecurity New Zealand, Wellington, 68 p.

Johnston C (2008b). Import risk analysis: frozen skinless and boneless fillet meat of *Oreochromis* spp. fish from China and Brazil for human consumption. MAF Biosecurity New Zealand, Wellington, 106 p.

Kahn SA, Beers PT, Findlay VL, Peebles IR, Durham PJ, Wilson DW, Gerrity SE (1999). Import risk analysis on non-viable salmonids and non-salmonid marine finfish. Australian Quarantine and Inspection Service, Department of Agriculture, Fisheries and Forestry, Canberra, Australia, 409 p.

Kunttu HM (2010). Characterizing the bacterial fish pathogen *Flavobacterium columnare*, and some factors affecting its pathogenicity. *Jyväskylä Studies in Biological and Environmental Science* 206, University of Jyväskylä, Finland, 81 p.

Kunttu HM, Sunberg LR, Pulkkinen K, Valtonen ET (2012). Environment may be the source of *Flavobacterium columnare* outbreaks at fish farms. *Environmental Microbiology Reports* 4(4): 398–402.

Lafrentz BR, LaPatra SE, Shoemaker CA, Klesius PH (2012). Reproducible challenge model to investigate the virulence of *Flavobacterium columnare* genomovars in rainbow trout *Oncorhynchus mykiss*. *Diseases of Aquatic Organisms* 101(2): 115–121.

Lafrentz BR, Waldbieser GC, Welsh TJ, Shoemaker CA (2014). Intergenomic heterogeneity in the 165r RNA genes of *Flavobacterium columnare* and standard protocols for genomovar assignment. *Journal of Fish Diseases* 37: 657–669.

Lanto E, Pettinen RK, Bamford JHK, Sundberg LR (2014). Comparing the different morphotypes of a fish pathogen – implications for key virulence factors in *Flavobacterium columnare*. *BMC Microbiology* 14: 170–175.

Loch TP, Faisal M (2014). Emerging flavobacterial infections in fish: a review. *Journal of Advanced Research* 6(3): 283–300.

Meepagala KM, Schrader KL, Burandt CL (2013). Antibacterial compounds from Rutaceae with activities against *Flavobacterium columnare* and *Streptococcus iniae*. *Journal of Agricultural Chemistry and Environment* 2(4): 90–100.

Melville V, Johnston C (2010). Supplementary risk analysis: extension of countries eligible to export frozen, skinless, boneless fillet meat of *Pangasius* spp. under the import health standard fisfilis.spe. MAF Biosecurity New Zealand, Wellington, 15 p.

Michel C, Messiaen S, Bernardet JF (2002). Muscle infections in imported neon tetra *Paracheirodon innesi* Myers: Limited occurrence of microsporidia and predominance of severe forms of columnaris disease caused by an Asian genomovar of *Flavobacterium columnare*. *Journal of Fish Diseases* 25: 253–263.

Mohammed HH, Arias CR (2014). Epidemiology study of columnaris disease affecting fish species within the same watershed. *Diseases of Aquatic Organisms* 109(3): 201–211.

Munday BL (2002). Scientific review of significant disease agents carried by non-viable, non-salmonid freshwater and estuarine finfish and their products. Biosecurity Australia, Department of Agriculture, Fisheries and Forestry, Australia, 71 p.

OIE (2016). Aquatic Animal Health Code, 19th Edition. World Organization for Animal Health. Paris, France [Online] Available from: http://www.oie.int/international-standard-setting/aquatic-code/access-online/ [Accessed 15 March 2017].

Olivares-Fuster O, Baker JL, Terhune JS, Shoemaker CA, Klesius PH, Arias CR (2007). Host-specific association between *Flavobacterium columnare* genomovars and fish species. *Systematic and Applied Microbiology* 30(8): 624–633.

Olivares-Fuster O, Terhune JS, Shoemaker CA, Arias CR (2010). Cloning, expression, and immunogenicity of *Flavobacterium columnare* heat shock protein dna. *Journal of Aquatic Animal Health* 22(2): 78–86.

Olivares-Fuster O, Bullard SA, McElwain A, Llosa MJ, Arias CR (2011). Adhesion dynamics of *Flavobacterium columnare* to channel catfish *Ameiurus punctatus* and zebrafish *Danio rerio* after immersion challenge. *Diseases of Aquatic Organisms* 96(3): 221–227.

Olivares-Fuster O, Arias CR (2012). Development, characterisation and early evaluation of new modified live vaccine against columnaris disease. Proceedings of the 6th Annual Symposium for Aquatic Animal Health, Tampa, Florida, September 5–9, 2010.

Ordal EJ, Rucker RR (1944). Pathogenic myxobacteria. *Proceedings of the Society for Experimental Biology and Medicine* 56: 15–18.

Plumb J, Hanson L (2011). Health maintenance and principal microbial diseases of cultured fishes, 3rd Edition. Wiley-Blackwell, Oxford, United Kingdom, 400 p.

Reynard R, Wahli T, Vatsos I, Mortensen S (2007). Review of disease interactions and pathogen exchange between farmed and wild finfish and shellfish in Europe (DIPnet). Final VESO report 1655.2007 for the European Commission. IFREMER, Paris, France, 460 p.

Schachte JH (1983). *Flavobacterium columnare*. Chapter 23. *In*: A Guide to Integrated Fish Health. Special Publication 83(2). Great Lakes Fishery Commission, Canada, 262 p.

Shoemaker CA, Lafrentz BR (2015). Lack of association between *Flavobacterium columnare* genomovars and virulence in hybrid tilapia (*Oreochromis niloticus* x *O. aureus* (Steindachner). *Journal of Fish Diseases* 38(5): 491–498.

Sim-Smith C, Faire S, Lees A (2014). Managing biosecurity risk for business benefit. Report prepared for the Ministry for Primary Industries by Coast and Catchment Ltd, Auckland, New Zealand, 214 p.

Starliper CE, Schill WB (2011). Flavobacterial diseases: columnaris disease, coldwater disease and bacterial gill disease. pp 606-631. *In:* Woo PTK, Bruno DW (*Eds.*). *Fish Diseases and Disorders: Volume 3 - Viral, Bacterial and Fungal Infections*, 2nd edition. CABI Publishers, Wallingford, United Kingdom, 930 p.

Suomalainen LR (2005). *Flavobacterium columnare* in Finnish fish farming: Characterisation and putative disease management strategies. *Jyvaskyla Studies in Biological and Environmental Science* 155. University of Jyvaskyla, Finland, 54 p.

Tekedar HC, Karsi A, Gillaspy AF, Dyer DW, Benton NR, Zaitshik J, Vamenta S, Banes MM, Gülsoy N, Aboko-Cole M, Waldbieser GC, Lawrence ML (2012). Genome sequence of the fish pathogen *Flavobacterium columnare* ATCC 49512. *Journal of Bacteriology* 194(10): 2763–2764.

Tubbs L, Lee P, Diggles B, Jones JB, Sheppard M, Sim-Smith C (2007). A review of aquatic diseases of significance to New Zealand. Final research report for project ZBS 2005-17. National Institute of Water and Atmospheric Research, Wellington, New Zealand, 452 p.

Verma, DK, Rathore G (2013). Molecular characterisation of *Flavobacterium columnare* isolated from a natural outbreak of columnaris disease in farmed fish, *Catla catla* from India. *Journal of General and Applied Microbiology* 59: 417–424.

Verma V, Prasad Y, Singh BR (2011). Effect of pH and salinity on pathogenicity of *Flavobacterium columnare* and *Myxobacterium* sp. in Indian catfish, *Clarias batrachus* (Linn.) and *Heteropneustes fossilis* (Bloch). *Journal of Environmental Biology* 32: 573–577.

36. *Francisella* spp.

36.1. Hazard identification

36.1.1. Aetiological agent

Francisella spp. are strictly aerobic, irregular shaped motile Gram-negative intracellular bacteria, classified within the Proteobacteriaceae, in Family Francisellaceae (Arkush & Bartholomew 2011). They were first described as a *Rickettsia*-like bacterium from farmed three lined grunt (*Parapristipoma trilineatum*) in Japan and from tilapia (*Oreochromis* spp.) in Taiwan (Kamaishi *et al*. 2005; Hsieh *et al*. (2006).

Four species were provisionally recognised (Mikalsen *et al.* 2007) but genomic analysis suggests *Francisella* represents a species complex with two major lineages: *F. tularensis* and *F. philomiragia*, but the host relationships and distribution in both marine and freshwater within this species complex remain unclear (Ottem *et al.* 2009; Arkush & Bartholomew 2011; Colquhoun & Duodu 2011). Some isolates are zoonotic (Colquhoun & Duodu 2011).

36.1.2. OIE status

Infection with *Francisella* spp. is not listed by the OIE (OIE 2016).

36.1.3. New Zealand status

Francisella spp. are considered exotic (Johnston 2008) and have not been reported from New Zealand. Infection with *Francisella* spp. is not a notifiable disease in New Zealand (Anon. 2016).

36.1.4. Epidemiology

Francisella spp. represent a species complex of *Rickettsia*-like proteobacteria that cause systemic, chronic, granulomatous infections in marine and fresh water fish (Johnston 2008). *Francisella philomiragia noatunensis* infects fish in Norway, while *F. p. orientalis* is reported from Asian and Chinese aquaculture (Ottem *et al.* 2008; Arkush & Bartholomew 2011; Colquhoun & Duodu 2011). A distinct, but yet undescribed species of *Francisella* occurs in Atlantic cod (*Gadus morhua*) farmed in Norway, as well as from farmed tilapia (*Oreochromis* spp.) in North, Central and South America, Hawaii, and the United Kingdom, (Mikalsen *et al.* 2007; Colquhoun & Duodu 2011)

Francisella spp. isolates affect a wide host range of farmed fish, as well as from wild stocks caught adjacent to aquaculture facilities (Ottem *et al.* 2008). Susceptible species (Table 33) include members of the families Cichlidae, Gadidae, Moronidae, Pleuronectidae, Salmonidae and Scombridae in aquaculture (Nylund & Ottem 2006; Ostland *et al*. 2006; Mikalsen *et al.* 2007; Johnston 2008; Arkush & Bartholomew 2011; Zerihun *et al.* 2011).

Infection also occurs in invertebrates including abalone (*Haliotis discus discus* and *H. discus hannai*) and blue mussels (*Mytilus edulis*) (Mollusca); and crabs (*Cancer pagurus*) (Crustacea) when collected adjacent to infected marine farms (Ottem *et al*. 2009; Colquhoun & Duodu 2011). *Francisella* sp. isolates are also reported from protozoans collected in free environmental sampling. These alternative pathways may represent reservoirs of infection (Nylund & Ottern 2006; Arkush & Bartholomew 2011; Zerihun *et al.* 2011).

Table 33. Families and Species of Fish Susceptible to *Francisella* Spp., by Geographical Location

Family	Host Species	Locations
Cichlidae	Nile Tilapia (*Oreochromis niloticus*), Mozambique tilapia (*O. mossambicus*), blue tilapia (*O. aureus*), redbelly tilapia (*Coptodon zillii*), wami tilapia (*O. urolepis*)	China, Southeast Asia, Japan, Taiwan, Europe, North, Central and South America
Gadidae	Atlantic cod (*Gadus morhua*)	China, Southeast Asia, Japan, Taiwan, North and South America, Europe
Gadidae	Saithe (*Pollachius virens*), pollock (*P. pollachius*)	Europe
Moronidae	European sea bass (*Dicentrarchus labrax*), striped bass (*Morone* spp.)	North America
Pleuronectidae	European plaice (*Pleuronectes platessa*)	Europe
Salmonidae	Atlantic salmon (*Salmo salar*)	South America
Sciaenidae	White weakfish (*Atractoscion nobilis*)	North America
Scombridae	Atlantic mackerel (*Scomber scombrus*)	Europe
Scophthalmidae	Megrim (*Lepidorhombus whiffiagonis*)	Europe

Francisella spp. is an emerging pathogen of marine farming worldwide and fish of all ages may be affected (Colquhoun & Duodu 2011).

Mortality is highly variable, from 30% to 95% for tilapia (Johnston 2008) and up to 40% in Atlantic cod (*Gadus morhua*) (Olsen *et al*. 2006). Clinically infected fish may have dark skin coloration due to haemorrhaging of the underlying musculature, necrotic granulomas of the skin, mouth, gill and nasal tissue, and opacity of the eyes (Arkush & Bartholomew 2011).

Internal signs of infection in fish include enlargement of the spleen, kidney and heart, together with the formation of whitish granulomas throughout the internal organs (Olsen *et al.* 2006; Arkush & Bartholomew 2011).

Clinical infection may also occur with no apparent signs of clinical disease (Nylund & Ottem 2006). Despite a mortality rate of up to 50% and the presence of extensive muscular granulomas, market-sized tilapia in Central American aquaculture showed few or no external signs of infection (Johnston 2008). These Central American isolates of *Francisella* were shown to preferentially target splenic and renal tissues in 82% and 93% of infected *Tilapia* spp., respectively, while affecting the musculature in only 10% of sampled fish (Mauel *et al*. 2007). Other isolates have differing preferences for muscle tissue, so *Francisell*a spp. could be present in muscle tissue without obvious granulomata (Johnston 2008).

Disease transmission is horizontal, through the water column. Entry may occur through the visceral epidermis, or directly through skin epidermal cells (Johnston 2008; Colquhoun & Duodu 2011), although the details of transmission and infection remain unclear (Arkush & Bartholomew 2011). Infection, transmission and morbidity are temperature-limited below 4°C, although bacterial growth increases up to the maximum that the host fish can survive (Colquhoun & Duodu 2011). Vertical transmission is suspected, but still unproven (Colquhoun & Duodu 2011).

Francisella spp. are environmentally stable and unaffected by salinity, surviving for long periods in marine and fresh waters. They may be transported on fish processing equipment, with potential for cross-infection (Arkush & Bartholomew 2011). They survive in fresh water (at 4°C) for 30 days at 8°C and 16 days at 12°C (Colquhoun & Duodu 2011), and may enter a resistant viable but non-culturable (VBNC) state in sediments (Ottem *et al*. 2009).

Some isolates are zoonotic. *Francisella philomiragia* rarely causes opportunistic infections in humans (Colquhoun & Duodu 2011). As isolates derived from fish appear to have lower

temperature tolerances than those infecting mammals, it appears unlikely that reported human infections have resulted from the consumption of infected fish (Arkush & Bartholomew 2011).

Francisella spp. are unaffected by freezing, since they survive at -80°C with no loss of infectivity (Colquhoun & Duodu 2011), while related species survive for up to 75 days in frozen sheep muscle stored at -20°C (Johnston 2008). The infective dose is as low as 10 cfu (colony forming units) per fish (Nylund & Ottem 2006; Colquhoun & Duodu 2011).

Francisella spp. are highly heat tolerant, requiring moist heat at 121°C for 15 minutes for inactivation (Anon. 2014).

Given the intracellular nature of infection, antibiotics are of limited effectiveness, although oxytetracycline is commonly used in aquaculture (Colquhoun & Duodu 2011). No vaccines are commercially available for *Francisella* spp. infections in fish (Colquhoun & Duodu 2011).

Potential hosts in New Zealand include Atlantic salmon (*S. salar*) and brown trout (*S. trutta*), as well as mackerel (*Scomber* spp.) and flatfishes (Order Pleuronectiformes) (Colquhoun & Duodu 2011). *Francisella* spp. have previously been identified as a risk in imported oreochromid fish (Johnston 2008).

36.2. Risk assessment

36.2.1. Entry assessment

Granulomatous infections caused by *Francisella* spp. have been reported from farmed fish from Asia, Australia, Europe, North, Central and South America (Johnston 2008; Ottem *et al.* 2009; Arkush & Bartholomew 2011; Colquhoun & Duodu 2011).

While fish with obvious external signs of infection should not enter the human food consumption pathway, up to 10% of those infected typically show few, or no, external signs (Johnston 2008; Arkush & Bartholomew 2011). Infection with some isolates is concentrated in the viscera (Mauel *et al.* 2007), and evisceration would substantially reduce the bacterial concentration. Other isolates preferentially infect the musculature (Johnston 2008a; Ottem *et al*. 2009) and these may be retained in the skin, gills and musculature of eviscerated fish. They are likely to be present in the commodity.

The likelihood of entry is assessed as non-negligible.

36.2.2. Exposure assessment

Francisella spp. may be present in the commodity (Johnston 2008). To establish infection, infected product would have to become available for consumption by a susceptible fish host, in sufficient quantity and duration (Kahn *et al.* 1999). *Francisella* spp. may remain viable in the blood-water discharge, as well as in the gills, brain tissue, skin and muscle trimmings discarded after commercial food processing (Johnston 2008; Arkush & Bartholomew 2011; Colquhoun & Duodu 2011), and may be present in the storage and packaging materials, including ice, used to transport the commodity (Johnston 2008).

While evisceration is likely to reduce pathogen concentration, the level of infection of the musculature varies among isolates (Johnston 2008) and infected tissue is likely to be retained in the gills, pharyngeal cavity, brain and eyes of eviscerated fish (Arkush & Bartholomew 2011).

The organism remains viable in dead fish tissues (Colquhoun & Duodu 2011) and is likely to survive in the environmental conditions present in commercial fish processing. It is resistant to freezing (to -80°C) with no loss of infectivity and may survive in fresh water for up to 30 days (Johnston 2008; Colquhoun & Duodu 2011), while it can persist in a VBNC state for extended periods in sediments (Ottem *et al.* 2009). The infective dose required to initiate infection is extremely low at 10 cfu fish^{-1} (Nylund & Ottem 2006). Potential marine and freshwater hosts are present in New Zealand (Johnston 2008).

The likelihood of exposure to *Francisella* spp. is assessed as non-negligible.

36.2.3. Consequence assessment

Francisella spp. is a recognised pathogen of salmonids (Arkush & Bartholomew 2011). The consequences of introduction would be extremely serious for the salmonid fishery, which was valued at $63 million in export earnings in 2011 (Aquaculture New Zealand 2014). An outbreak could also affect recreational and tourist trout and salmon fishing, as well as incur significant social and environmental costs associated with other domestic fisheries (Fish & Game 2014). While the dollar value of New Zealand's freshwater fisheries is not fully known, the Lake Taupo trout fishery alone is valued at $70 to 80 million per annum (Marsh & Mkwra 2013).

Francisella spp. may also infect marine species (Table 33), with potential economic consequences for several marine fisheries, including flatfishes (Order Pleuronectiformes) and mackerel (Scombridae). The mackerel fishery alone was valued at $NZ 64 million in 2012 (Statistics New Zealand 2013). Given the variability in virulence of *Francisella* spp., the effect on particular potential host species cannot be determined in advance.

Francisella spp. isolates associated with fish are considered of low to minimal zoonotic significance (Colquhoun &Duodo 2011).

The consequences of establishment are assessed as non-negligible.

36.2.4. Risk estimation

Since the entry, exposure and consequence assessments for *Francisella* spp. are non-negligible, the risk estimate is non-negligible. Therefore, *Francisella* spp. is assessed to be a risk in the commodity and risk management measures may be justified.

36.3. Risk management

Francisella spp. has been assessed to be a risk in the commodity. As infection with *Francisella* spp. is a non-notifiable disease, the *Aquatic Code* (OIE 2016) provides no specific guidance for importing eviscerated fish from infected countries, or specific processing requirements that would ensure the destruction of the pathogen. There are no specific requirement for reporting of non-OIE listed diseases. It is assumed that compliance with the approved processed states for OIE-listed diseases (Appendix 3) should also eliminate *Francisella* spp. from the commodity and be a viable risk management option.

Francisella spp. infection is reported from eight families of marine and freshwater fish (Table 33), which may be present in the commodity. Species declaration should substantially reduce the pathogen load of *Francisella* spp. in the commodity and be a viable risk management option.

Francisella spp. are reported from marine and fresh waters of Asia, Europe, North, Central and South America, and Australia. Where country/zone freedom is approved by through the MPI Country Approval Procedures, this option is likely to substantially reduce the pathogen load of Francisella spp. in the commodity. Approval of a declaration of country/zone freedom by MPI is a viable risk management option.

Francisella spp. is mainly associated with farmed fish (Ottem *et al.* 2008). Restriction of the commodity to wild-caught fish (not from aquaculture) would moderately reduce pathogen load in the commodity and be a viable risk management option.

Francisella spp. may be present in the skin, skeletal muscle and heads of eviscerated fish. Removal of the gills, or of the head and gills should slightly reduce pathogen load of *Francisella* spp. in the commodity. As there are no literature reports that describe the spread of any proteobacteria, including *Francisella* spp., through skin-off filleted fish imported for human consumption (Johnston 2008), further processing to the skin-off fillet state should moderately reduce the pathogen load of *Francisella* spp. Processing to the skin-off fillet state is a viable risk management option.

Francisella spp. is resistant to freezing, so frozen storage is unlikely to reduce pathogen load in the commodity and is not considered a viable risk management option.

Thermal inactivation requires high temperatures (by cooking to 121°C, with moist heat, for 15 minutes). This heat treatment is likely to eliminate *Francisella* spp. from the commodity and be a viable risk management option.

36.3.1. Risk management options

Francisella spp. is reported from fish in families Cichlidae, Gadidae, Moronidae, Pleuronectidae, Salmonidae, Sciaenidae, Scombridae and Scophthalmidae (Table 33), which are considered likely to be present in the commodity. Other families have not been associated with *Francisella* spp. Therefore species declaration indicating the commodity is not originated from any of the above families should substantially reduce the occurrence of *Francisella* spp. in the commodity.

For commodities originated from families associated with *Francisella* spp., one or a combination of the following additional options could also be considered to manage the risk.

Where country/zone freedom from *Francisella* spp. is accepted by MPI:

Option 1

Acceptance of country/zone freedom by MPI should substantially reduce the occurrence of *Francisella* spp., so the commodity may be imported without any further restrictions.

Where country/zone freedom from *Francisella* spp. is not accepted by MPI or not available:

Option 2

Processing to a state consistent with the approved states for OIE-listed diseases (Appendix 3) should eliminate *Francisella* spp. When these provisions are met, the commodity could be imported without any further restrictions.

Where the imported commodity is not in a state consistent with the approved states for OIE-listed diseases (Appendix 3):

Option 3

Heat treatment (by cooking, with moist heat, to at least 121°C for 15 minutes) should eliminate *Francisella* spp. When this provision is met, the commodity could be imported without any further restrictions; or,

Option 4

Further processing (to the skin-off fillet state) should moderately reduce the occurrence of *Francisella* spp. When this provision is met, the commodity could be imported without any further restrictions; or,

Option 5

Restriction of the commodity to wild-caught fish (not from aquaculture) should moderately reduce the occurrence of *Francisella* spp. When this provision is met, the commodity could be imported without any further restrictions.

36.4. References

Anonymous (2014). *Francisella tularensis* – Material safety data sheets (MSDS). Public Health Agency of Canada [Online] Available from: http://www.phac-aspc.gc.ca/lab-bio/res/psds-ftss/msds68e-eng.php [Accessed 8 December 2014].

Anonymous (2016). Biosecurity (Notifiable Organisms) Order 2016. NZLR 2016/73. New Zealand Government, Wellington, 13 p.

Aquaculture New Zealand (2014). New Zealand's aquaculture industry key facts 2013 [Online] Available from: http://www.seafoodnewzealand.org.nz/our-industry/key-facts/ [Accessed 26 September 2014].

Arkush KD; Bartholomew JL (2011). *Piscirickettsia*, *Francisella* and epitheliocystis, pp 302–337 *In*: Woo PTK, Bruno DW (*Eds.*). Fish Diseases and Disorders: Volume 3 - Viral, Bacterial and Fungal Infections, 2nd edition. CABI International, Wallingford, United Kingdom, 930 p.

Colquhoun DJ, Duodu S (2011). *Francisella* infections in farmed and wild aquatic organisms. *Veterinary Research* 42(1): 47 [Online] Available from: http://www.veterinaryresearch.org/content/42/1/47 [Accessed 24 September 2014].

Fish and Game (2014). New Zealand Fish and Game Council [Online] Available from: http://www.fishandgame.org.nz/ [Accessed 24 September 2014].

Hsieh CY, Tung MC, Tu C, Chang CD, Tsai SS (2006). Enzootics of visceral granulomas associated with *Francisell*a-like organism infection in tilapia (*Oreochromis* spp.). *Aquaculture* 254: 129–138.

Johnston C 2008. Import risk analysis: frozen, skinless and boneless fillet meat of *Oreochromis* spp. from China and Brazil for human consumption. MAF Biosecurity, Ministry of Agriculture and Forestry, Wellington, New Zealand, 106 p.

Kahn SA, Beers PT, Findlay VL, Peebles IR, Durham PJ, Wilson DW, Gerrity SE (1999). Import risk analysis on non-viable salmonids and non-salmonid marine finfish. Australian Quarantine and Inspection Service, Department of Agriculture, Fisheries and Forestry, Canberra, Australia, 409 p.

Kamaishi T, Fukuda Y, Nishiyama M, Kawakami H, Matsuyama T, Yoshinaga T, Oseko N (2005). Identification and pathogenicity of intracellular *Francisella* bacterium in three lined grunt *Parapristipoma trilineatum. Fish Pathology* 40: 67–71.

Marsh D, Mkwra L (2013). Review of freshwater non-market value studies. Department of Economics, University of Waikato, Hamilton, New Zealand, 144 p.

Mauel MJ, Soto E, Moralis JA, Hawke J (2007). A piscirickettsiosis-like syndrome in cultured Nile tilapia in Latin America with *Francisella* spp. as the pathogenic agent. *Journal of Aquatic Animal Health* 19: 27–34.

Mikalsen J, Olsen AB, Tengs T, Colquhoun DJ (2007). *Francisella philomiragia* subsp. nov. isolated from farmed Atlantic cod (*Gadus morhua* L.). *International Journal of Systemic and Evolutionary Microbiology* 57: 1960–1965.

Nylund A, Ottem KF (2006). Francisellosis a new cod (*Gadus morhua*) disease caused by *Francisella* n. sp. DIPnet (Disease interactions and pathogen exchange between farmed and wild aquatic animal populations – a European network), Newsletter 45. VESO (Norwegian Veterinary Centre for Contract Research), 2 p.

OIE (2016). Aquatic Animal Health Code, 19th Edition. World Organization for Animal Health. Paris, France [Online] Available from: http://www.oie.int/international-standard-setting/aquatic-code/access-online/ [Accessed 15 March 2017].

Olsen AB, Mikalsen J, Rode M, Alfjorden A, Hoel E, Straum-Lie K, Haldorsen R, Colquhoun DJ (2006). A novel systemic granulomatous infection in farmed Atlantic cod *Gadus morhua* L., associated with a bacterium belonging to the genus *Francisella. Journal of Fish Diseases* 29: 307–311.

Ostland VE, Stannard JA, Creek JJ, Hedrick RP, Ferguson HW, Carlberg JM, Westerman ME (2006). Aquatic *Francisella*-like bacterium associated with mortality of intensively cultured hybrid striped bass *Morone chrysops* × *M. saxatilis. Diseases of Aquatic Organisms* 72(2): 135–145.

Ottem KF, Nylund A, Isaksen TE, Karlsbakk E, Bergh O (2008). Occurrence of *Francisella piscicida* in farmed and wild Atlantic cod, *Gadus morhua* L., in Norway. *Journal of Fish Diseases* 31: 525–534.

Ottem KF, Nylund A, Karlsbakk E, Fris-Moller A, Kamaishi T (2009). Elevation of *Francisella philomiragia* subsp. *noatunensis* Mikalsen *et al.* (2007) to *Francisella noatunensis* comb. nov. [syn. *Francisella piscicida* Ottem *et al*. (2008) syn. nov.] and the characterisation of *Francisella noatunensis* subsp. *orientalis* subsp. nov., two important fish pathogens. *Journal of Applied Microbiology* 106: 1231–1243.

Statistics New Zealand (2013). New Zealand's seafood industry. Statistics New Zealand, Wellington, New Zealand [Online] Available from: http://www.stats.govt.nz/tools_and_services/newsletters/price-index-news/apr-13-article-seafood.aspx [Accessed 12 April 2015].

Zerihun MA, Feist SW, Bucke D, Olsen AB, Tandstad NM, Colquhoun DJ (2011). *Francisella noatunensis* subsp. *noatunensis* is the aetiological agent of visceral granulomatosis in wild Atlantic cod *Gadus morhua*. *Diseases of Aquatic Organisms* 95: 65–71.

37. *Moritella viscosa*

37.1. Hazard identification

37.1.1. Aetiological agent

Moritella viscosa, a non-spore forming bacterium, is the causative agent of "winter ulcer disease" in salmonids (Gudmundsdóttir & Björnsdóttir 2007). Several distinct strains have been identified from Norway and Iceland, affecting salmonid fry, smolts and adult fish (Björnsdóttir 2011). Four genotypes are recognised. The first genotype is found exclusively in Atlantic cod (*Gadus morhua*) and Atlantic salmon (*Salmo salar*), farmed in Norway. The second genotype occurs in Icelandic, Norwegian and Scottish farmed rainbow trout (*Oncorhynchus mykiss*). The third genotype is isolated from Icelandic and Canadian farmed Atlantic salmon. The fourth genotype represents a low virulence isolate from Icelandic lumpsucker (*Cyclopterus lumpus*), which also occurs occasionally in Atlantic salmon (Grove *et al*. 2010; Björnsdóttir 2011).

37.1.2. OIE status

Infection with *Moritella viscosa* is not notifiable to the OIE (OIE 2016).

37.1.3. New Zealand status

Moritella viscosa has not been reported from New Zealand and is considered exotic (Tubbs *et al.* 2007). Infection with *Moritella viscosa* is not a notifiable disease in New Zealand (Anon. 2016).

37.1.4. Epidemiology

Winter ulcer disease has been reported from northern European marine farming where water temperatures fall below 8°C, or during the winter smoltification of cultured freshwater salmonids (Björnsdóttir 2011).

Moritella spp. is reported from wild and farmed fish from several families (Table 34) including the Cyclopteridae, Gadidae, Mugilidae, Pleuronectidae, Salmonidae and Scophthalmidae (Benediktsdóttir *et al*. 2000; Toranzo *et al.* 2005; Gudmundsdóttir *et al.* 2006; Tubbs *et al.* 2007; Björnsdóttir 2011). It has also been reported in invertebrates including the parasitic salmon louse (*Lepeophtheirus salmonis*) (Björnsdóttir 2011), as well as jellyfish (*Cyanea lamarckii*) when associated with salmonid sea cages in Scotland (Schuett & Hilke 2010).

Table 34. Families and Species of Fish Susceptible to *Moritella* Spp.

Family	Host Species
Cyclopteridae	Lumpsucker (*Cyclopterus lumpus*)
Gadidae	Atlantic cod (*Gadus morhua*)
Mugilidae	Grey mullet (*Mugil cephalus*)
Pleuronectidae	Atlantic halibut (*Hippoglossus hippoglossus*), European plaice (*Pleuronectes platessa*)
Salmonidae	All salmonids (*Coregonus* spp., *Hucho* spp., *Oncorhynchus* spp., *Parahucho* spp., *Prosopium* spp., *Salmo* spp., *Salvelinus* spp., *Salvethymus* spp., *Stenodus* spp., *Thymallus* spp.)
Scophthlmidae	turbot (*Scophthalmus maximus*)

An unidentified deep-sea reservoir of infection may also exist in North Atlantic waters (Björnsdóttir 2011).

Moritella viscosa was originally restricted to European waters, but is now reported from grey mullet (*Mugil cephalus*) (Muglidae) in Indian waters (Ghosh *et al.* 2014). Grey mullet are widely distributed in tropical and sub-tropical brackish, estuarine and coastal marine waters, including New Zealand (Paul 2000; NIWA 2014).

Mortality varies from 10% to 90% for Atlantic salmon (*S. salar*) smolts (Veso 2014), while morbidity may reach 50%, particularly under high stocking densities (Björnsdóttir 2011). The virulence factors of *M. viscosa* are complex (Björnsdóttir *et al.* 2011; Karlsen *et al*. 2014), and vary among isolates (Veso 2014).

Disease transmission is horizontal, through the water column (Tubbs *et al*. 2007), with pathogen entry occurring across the epidermal cells of the skin and gill lamellae (Karlsen *et al*. 2012), although the details of infection remain unclear (Björnsdóttir 2011). *Moritella viscosa* strains are not host-specific, and infection may occur at concentrations as low as 14 cfu (colony forming units) per fish (Björnsdóttir *et al*. 2011).

Disease onset is usually rapid (Björnsdóttir 2011) although longer-term chronic disease is also reported (Karlsen *et al*. 2014). Infected fish commonly have pale gills with haemorrhaging and exophthalmia of the eyes and fin rot. As the disease progresses, pinhead sized skin spots form on the skin and gill surface that develop into ulcerative lesions (Björnsdóttir 2011).

The external skin lesions may heal if water temperatures increase above 8°C (Olsen *et al.* 2011) but this masks the continued progression of the disease into a chronic phase affecting the internal organs and dermal musculature (Tunsjo *et al.* 2009; Björnsdóttir 2011). Further disease development includes extensive necrosis of kidney, spleen and liver tissues and the development of visible ascites, together with degeneration and necrosis of the dermal musculature (Lovoll *et al.* 2009). Death occurs usually through extensive internal haemorrhaging of the host (Benediktsdóttir *et al.* 2000; Olsen *et al.* 2011).

Moritella viscosa is unaffected by frozen storage (to -80°C) and may survive for over a year in marine and brackish waters (more than 30% salinity) (Irwin 2010). It remains virulent over a wide temperature range (4 to 15 °C) and is reported from wild fish populations (Björnsdóttir 2011). It is inactivated in fish tissues by heat treatment (to at least 85°C for 25 minutes) (Nesse *et al*. 2012).

Moritella viscosa is resistant to main antibiotic treatments used in aquaculture (Björnsdóttir *et al.* 2012). A vaccine is only available for Atlantic salmon (*S. salar*), but this is of limited effectiveness (Korsnes 2007).

Susceptible species in New Zealand include salmonids (*Salmo* spp., *Oncorhynchus* spp., *Salvelinus* spp.), grey mullet (*M. cephalus*), flatfish (Order Pleuronectiformes) as well as other marine species when reared in sea cages in cold waters (Tubbs *et al*. 2007).

37.2. Risk assessment

37.2.1. Entry assessment

Moritella viscosa is mainly associated with farmed fish, but also occurs in wild fish stocks, including grey mullet (Björnsdóttir 2011; Ghosh *et al.* 2014). Clinically infected fish show few external signs of infection (Björnsdóttir 2011) and may pass visual inspection for the human food consumption pathway. While evisceration would significantly reduce pathogen concentration, *M. viscosa* also occurs in the musculature, gills and skin tissues, which are retained in the commodity

(Tunsjo *et al*. 2009; Björnsdóttir 2011; Karlsen *et al.* 2014). It is resistant to the environmental conditions likely to be encountered in commercial fish processing, storage and transport (Johnston 2008; Lovoll *et al.* 2009; Irwin 2010).

The likelihood of entry of *M. viscosa* through the commodity is assessed as non-negligible.

37.2.2. Exposure assessment

For disease to establish, sufficient infected uncooked fish scraps or offal would have to be discharged into the environment, at a level to become self-sustaining in the population (Kahn *et al.* 1999). This may occur through discharges from fish processing facilities where the material can infect susceptible fish or other carriers (Schuett & Hilke 2010; Björnsdóttir 2011). *Moritella viscosa* is known to survive in frozen fish and may be present in the blood-water discharge, as well as in the gills, skin and muscle trimmings discarded as offal from commercial processing. It can survive in marine waters for over 12 months (Tunsjo *et al.* 2009; Björnsdóttir 2011) and is infective at low concentration (Björnsdóttir 2011).

Susceptible species in New Zealand include all salmonids (*Oncorhynchus* spp., *Salmo* spp., *Salvelinus* spp.), flatfish (Order Pleuronectiformes) and grey mullet (*Mugil cephalus*) (Tubbs *et al.* 2007).

The likelihood of exposure to *M. viscosa* is assessed as non-negligible.

37.2.3. Consequence assessment

Moritella viscosa is a pathogen of medium mortality in farmed fish, but is of high economic significance because product quality is affected even at low rates of infection (Björnsdóttir 2011), and control measures including antibiotic treatment and vaccination have limited effectiveness (Korsnes 2007; Björnsdóttir *et al.* 2012).

Moritella viscosa is pathogenic to salmonids including rainbow trout (*O. mykiss*) (Björnsdóttir 2011). Its introduction would have severe economic and significant social and environmental consequences associated with the recreational and tourist trout and salmon fisheries in New Zealand (Fish & Game 2014). While the dollar value of New Zealand's freshwater fisheries is not fully known, the Taupo fishery alone is valued at $70 to 80 million per annum (Marsh & Mkwra 2013). The direct consequences for Atlantic salmon (*S. salar*) are likely to be low, as this species is limited to the upper Waiau catchment in the South Island of New Zealand and is not commercially farmed (NIWA 2014).

Moritella viscosa also affects marine fish (Table 34). Grey mullet (*M. cephalus*) supports an important inshore commercial target fishery, and is important for recreational and traditional Maori fisheries (Ministry for Primary Industries 2014; NIWA 2014). Introduction of *M. viscosa* would have social and economic consequences for these fishing activities in New Zealand. Once established, control or eradication of *M. viscosa* in these fisheries would likely be ineffective (Kahn *et al*. 1999).

Little is known about the epidemiology of *M. viscosa* (Karlsen *et al.* 2014). Pathogenicity, host range and distribution vary between isolates (Björnsdóttir 2011; Ghosh *et al*. 2014). Isolates cause economic disease in rainbow trout (*O. mykiss*) and infect cell lines of Chinook salmon (*O. tshawytscha*) (Tunsjo *et al*. 2009). Whilst no clear evidence exists for natural infection in Chinook salmon, the consequences of introduction would be extreme for the salmon farming industry,

through loss of sales due to infected product. This industry was valued at $63 million in export earnings in 2011 (Aquaculture New Zealand 2014).

The consequences of establishment are assessed as non-negligible.

37.2.4. Risk estimation

Since the entry, exposure and consequence assessments are non-negligible, the risk estimate for *M. viscosa* is non-negligible. Therefore, *M. viscosa* is assessed to be a risk in the commodity and risk management measures may be justified.

37.3. Risk management

Moritella viscosa has been assessed to be a risk in the commodity. Infection with *M. viscosa* is a non-notifiable disease, so the *Aquatic Code* (OIE 2016) provides no specific guidance for importing eviscerated fish from infected countries, or specific processing requirements that would ensure the destruction of the pathogen. It is assumed that compliance with the approved processed states for OIE-listed diseases (Appendix 3) would also eliminate *M. viscosa* from the commodity and be a viable risk management option.

Moritella viscosa is reported from six families of wild and farmed fish (Table 34) that may be present in the commodity. Species declaration should substantially reduce the occurrence of *M. viscosa* in the commodity and be a viable risk management option. Restriction of the commodity to wild-caught fish (not from aquaculture) would have little, or no effect on pathogen load and is not a viable risk management option.

Moritella viscosa infection is reported from northern Europe (Iceland, Norway and Faeroe Islands), Scotland, Atlantic Canada and India. Where country/zone freedom is approved through the MPI Country Approval Procedures, this option is likely to substantially reduce the pathogen load of *M. viscosa* in the commodity. Approval of a declaration of country/zone freedom by MPI is a viable risk management option.

Moritella viscosa infection occurs in the musculature, gills and skin tissues. Removal of the gills, or the head and gills, should slightly reduce the occurrence of *M. viscosa* in the commodity. Further processing of the eviscerated commodity to the skin-off fillet state should moderately reduce the occurrence of *M. viscosa* in the commodity and be a viable risk management option.

Moritella viscosa is resistant to freezing, and unaffected by frozen storage so this is not a viable risk management option. High temperatures are required for inactivation (by cooking to at least 85°C for 25 minutes) which should eliminate *M. viscosa* from the commodity. High-temperature treatment is a viable risk management option.

37.3.1. Risk management options

Moritella viscosa is reported from fish in families Cyclopteridae, Gadidae, Mugilidae, Pleuronectidae, Salmonidae and Scophthlmidae (Table 34), which are considered likely to be present in the commodity. Other families have not been associated with *M. viscosa.* Therefore species declaration indicating the commodity is not originated from any of the above families should substantially reduce the occurrence of *M. viscosa* in the commodity.

For the commodities originated from families associated with *M. viscosa*, one or a combination of the following additional options could also be considered to effectively manage the risk.

Where country/zone freedom from *M. viscosa* is accepted by MPI:

Option 1

Acceptance of country/zone freedom by MPI should substantially reduce the occurrence of *M. viscosa*, so the commodity may be imported without any further restrictions.

Where country/zone freedom from *M. viscosa* is not accepted by MPI or not available:

Option 2

Processing to a state consistent with the approved states for OIE-listed diseases (Appendix 3) should eliminate *M. viscosa*. When these provisions are met, the commodity could be imported without any further restrictions.

Where the imported commodity is not in a state consistent with the approved states for OIE-listed diseases (Appendix 3):

Option 3

Heat treatment (by cooking to at least 85°C for 25 minutes) should eliminate *M. viscosa*. When this provision is met, the commodity could be imported without any further restrictions; or,

Option 4

Further processing (to the skin-off fillet state) should moderately reduce the occurrence of *M. viscosa*. When this provision is met, the commodity could be imported without any further restrictions.

37.4. References

Anonymous (2016). Biosecurity (Notifiable Organisms) Order 2016. NZLR 2016/73. New Zealand Government, Wellington, 13 p.

Aquaculture New Zealand (2014). Seafood New Zealand: Key Facts for Our Industry 2013 [Online] Available from: http://aquaculture.org.nz/industry/overview/ [Accessed 8 June 2014].

Benediktsdóttir E, Verdonck L, Spröer C, Helgason S, Swings J (2000). Characterization of *Vibrio viscosus* and *Vibrio wodanis* isolated at different geographical locations: a proposal for reclassification of *Vibrio viscosus* as *Moritella viscosa* (comb. nov.). *International Journal of Systematic and Evolutionary Microbiology* 50: 479–488.

Björnsdóttir B (2011). *Moritella viscosa* Virulence: Extracellular products and host-pathogen interaction. Ph.D thesis, Faculty of Medicine, University of Iceland, 184 p.

Björnsdóttir B, Gudmundsdóttir T, Gudmundsdóttir BK (2011). Virulence properties of *Moritella viscosa* extracellular products. *Journal of Fish Diseases* 35(5): 333–343.

Björnsdóttir B, Hjerde E, Bragason BT, Gudmundsdottir T, Willasen N, Gudmundsdottir BK (2012). Identification of type IV secretion systems in *Moritella viscosa. Veterinary Microbiology* 158: 436–422.

Fish and Game (2014). New Zealand Fish and Game Council [Online]. Available from: http://www.fishandgame.org.nz/, [Accessed 24 September 2014].

Ghosh S, Ringo E, Selvam DG, Rahiman KM, Sathyan N, Nifty J, Hatha AM (2014). Gut associated lactic acid bacteria isolated from the estuarine fish *Mugil cephalus*: Molecular diversity and antibacterial activities against pathogens. *International Journal of Aquaculture* 4(1): 1–11.

Grove S, Wiik-Nielsen CR, Lunder T, Tunsjo HS, Tandstad NM, Reitan LJ, Marthinussen A, Sorgaard M, Olsen AB, Colquhoun DJ (2010). Previously unrecognised division within *Moritella viscosa* isolated from fish farmed in the North Atlantic. *Diseases of Aquatic Organisms* 93(1): 51–61.

Gudmundsdóttir BK, Björnsdóttir B (2007). Vaccination against atypical furunculosis and winter ulcer disease of fish. *Vaccination* 25 (30): 5512–5523.

Gudmundsdóttir BK, Björnsdóttir B, Gudmundsdóttir S, Bambir SH (2006). A comparative study of susceptibility and induced pathology of cod, *Gadus morhua* (L.), and halibut, *Hippoglossus hippoglossus* (L.), following experimental infection with *Moritella viscosa*. *Journal of Fish Diseases* 29: 481-487.

Irwin JA (2010). Extremophiles and their application to veterinary medicine. *Environmental Technology* 31 (8-9): 857–869.

Johnston C (2008). Import risk analysis: Frozen, skinless and boneless fillet meat of *Pangasius* spp. fish from Vietnam, for human consumption. Ministry of Agriculture and Forestry, Wellington, New Zealand, 60 p.

Kahn SA, Beers PT, Findlay VL, Peebles IR, Durham PJ, Wilson DW, Gerrity SE (1999). Import risk analysis on non-viable salmonids and non-salmonid marine finfish. Australian Quarantine and Inspection Service, Department of Agriculture, Fisheries and Forestry, Canberra, Australia, 409 p.

Karlsen C, Sorum H, Willassen NP, Asbakk K (2012). *Moritella viscosa* bypasses Atlantic salmon epidermal keratocyte clearing activity and might use skin surfaces as a port of infection. *Veterinary Microbiology* 154(3-4): 353–362.

Karlsen C, Vanberg C, Mikkelsen H, Sørum H (2014). Co-infection of Atlantic salmon (*Salmo salar*) by *Moritella viscosa* and *Aliivibrio wodanis*, development of disease and host colonization. *Veterinary Microbiology* 171: 112–121.

Korsnes K (2007). *Moritella viscosa.* pp 70–72. *In:* Raynard R, Wahli T, Vatsos I, Mortensen S (*Eds.*). Review of disease interactions and pathogen exchange between farmed and wild finfish and shellfish in Europe (DIPnet). VESO Project 165, IFREMER, Paris, France, 460 p.

Lovoll M, Wiik-Nielsen CR, Tunsjo HS, Colquhoun D, Lunder T, Sorum H, Grove S (2009). Atlantic salmon bath challenged with *Moritella viscosa*: Pathogen invasion and host response. *Fish and Shellfish Immunology* 26(6): 877884.

Marsh D, Mkwra L (2013). Review of freshwater non-market value studies. Department of Economics, University of Waikato, Hamilton, New Zealand, 144 p.

Ministry for Primary Industries (2014). Grey mullet. pp 364–369, *In:* Fisheries Assessment Plenary, May 2014: stock assessments and stock status. Compiled by the Fisheries Science Group, Ministry for Primary Industries, Wellington, New Zealand, 1381 p.

Nesse LV, Rimstad E, Ringo E, Rosnes JT (2012). Risk assessment regarding processing requirements of by-products from aquaculture for use as fish feed. Report 11/603 to the Panel on Animal Feed. Norwegian Scientific Committee for Food Safety (VKM), 50 p.

NIWA (2014). Grey mullet [Online]. Available from: https://www.niwa.co.nz/freshwater-and-estuaries/nzffd/NIWA-fish-atlas/fish-species/atlantic_salmon [Accessed 28 April 2015].

OIE (2016). Aquatic Animal Health Code, 19th Edition. World Organization for Animal Health. Paris, France [Online]. Available from: http://www.oie.int/international-standard-setting/aquatic-code/access-online/ [Accessed 15 March 2017].

Olsen AB, Nilsen H, Sandlund N, Mikkelsen H, Sorum H, Colquhoun DJ (2011). *Tenacibaculum* sp. associated with winter ulcers in sea-reared Atlantic salmon *Salmo salar*. *Diseases of Aquatic Organisms* 94: 189–199.

Paul LJ (2000). New Zealand fishes: Identification, natural history and fisheries. Reed Publishing, New Zealand, 253 p.

Schuett C, Hilke D (2010). Endobiotic bacteria and their pathogenic potential in cnidarian tentacles. *Helgoland Marine Research* 64(3): 205–212.

Toranzo AE, Magarinos B, Romalde J (2005). A review of the main bacterial fish diseases in mariculture systems. *Aquaculture* 246(1): 37–61.

Tubbs L, Lee P, Diggles B, Jones JB, Sheppard M, Sim-Smith C (2007). A review of aquatic diseases of significance to New Zealand. Final research report for project ZBS 2005-17. National Institute of Water and Atmospheric Research, Wellington, New Zealand, 452 p.

Tunsjo HS, Paulsen SM, Berg K, Sorum H, L'Abee-Lund TM (2009). The winter ulcer bacterium *Moritella viscosa* demonstrates adhesion and cytotoxicity in a fish cell model. *Microbiology and Pathology* 47: 134–142.

Veso (2014). Atlantic salmon – *Moritella viscosa*. Veso Vican [Online]. Available from: http://www.veso.no/174764 [Accessed 3 October 2014].

38. *Piscirickettsia salmonis* species complex

38.1. Hazard identification

38.1.1. Aetiological agent

Piscirickettsia salmonis is a Gram-negative, non-motile, coccoid, intracellular bacterial pathogen classified in Class Gammaproteobacteria, Order Thiotrichales, Family Piscirickettsiaceae (Arkush & Bartholomew 2011). It is the agent of piscirickettsiosis, or salmon rickettsial septicaemia (Corbeil & Crane 2009). This was first isolated from diseased Coho salmon (*Oncorhynchus kisutch*) in Chilean marine aquaculture (Fryer *et al*. 1990, 1992), but has subsequently isolated from a wide range of salmonids and other species (Stone *et al.* 1997).

Piscirickettsiosis affects a wide range of fish hosts over a wide geographical range (Corbeil & Crane 2009), but *P. salmonis* refers to specific strains isolated from Chilean Coho salmon (Kahn *et al.* 1999). Other strains isolated from other areas, other fish, or other vertebrates, as well as invertebrates, including molluscs, are provisionally classified as *Piscirickettsia salmonis*-like organisms (PLOs) or rickettsia-like organisms (RLOs) (Hine 2002; Corbeil & Crane 2009; Arkush & Bartholomew 2011). Their current status as sub-species is in dispute (Rozas & Enríquez 2014) and *P. salmonis* is likely to represent a species complex (Kahn *et al*. 1999; Corbeil & Crane 2009).

Tasmanian rickettsia-like organism (Tasmanian RLO) was first reported from Atlantic salmon farmed in Tasmania (Corbeil *et al.* 2005). While this differs at the genetic level from strains of *P. salmonis* described overseas (Corbeil & Crane 2009), it is considered as part of this species complex for the purposes of this risk assessment.

38.1.2. OIE status

Infection with *P. salmonis* is not notifiable to the OIE (OIE 2016).

38.1.3. New Zealand status

Rickettsia-like organisms are reported from New Zealand molluscs (Hine 2002), but neither systemic rickettsial infection including *P. salmonis*, nor Tasmanian RLO have been reported from New Zealand (Corbeil & Crane 2009). They are considered exotic (Tubbs *et al*. 2007; Johnston 2008a; Corbeil & Crane 2009). Infection with *P. salmonis* is not a notifiable disease in New Zealand (Anon. 2016).

38.1.4. Epidemiology

Piscirickettsia salmonis is the agent of salmon rickettsial syndrome (SRS), also known as Coho salmon septicaemia, piscirickettsiosis or salmon rickettsial septicaemia (Mauel & Miller 2002; Corbeil & Crane 2009). It affects salmonids (Table 35), including Atlantic salmon (*Salmo salar*), coho salmon (*Oncorhynchus kisutch*), pink salmon (*O. gorbuscha*), Chinook salmon (*O. tshawytscha*) and rainbow trout (*O. mykiss*) (Kahn *et al.* 1999).

Table 35. Families and Species of Fish Susceptible to the *Piscirickettsia Salmonis* Species Complex Including *Piscirickettsia*-Like (PLO) and R*ickettsia*-Like (RLO) Organisms

Host Family	Susceptible Fish Species
Cichlidae	Tilapia (*Oreochromis* spp., *Tilapia* spp., *Sarotherodon* spp.)
Moronidae	European sea bass (*Dicentrarchus labrax*)
Salmonidae	Atlantic salmon (*Salmo salar*), coho salmon (*Oncorhynchus. kisutch*), pink salmon (*O. gorbuscha*), Chinook salmon (*O. tshawytscha*), rainbow trout (*O. mykiss*)
Sciaenidae	White weakfish (*Atractoscion nobilis*)
Serranidae	Groupers (*Epinephelus* spp.)
Tetraodontidae	Globe fish (*Tetraodon lineatus*)

Non-salmonids may be infected by other strains of the *P. salmonis* species complex (Table 36). These are widely distributed, infecting farmed tilapia (Cichlidae) in freshwater and marine fish (Moronidae, Sciaenidae and Serranidae) in Hawaii, the continental United States and in Taiwan (Mauel & Miller 2002; Anthanassopoulou *et al*. 2004; Mauel *et al*. 2005; Rozas & Enríquez 2014). They also occur in farmed puffer fish (*Tetraodon fahaka*) in Egypt, as well as in farmed salmonids in Australia, Canada, Chile, Ireland, Norway and Scotland (Corbeil & Crane 2009; Rozas & Enríquez 2014).

While *P. salmonis* does not replicate outside of a susceptible host (Kahn *et al*. 1999), wild fish may act as reservoirs for infection (Fryer & Hedrick 2003). Marine and freshwater invertebrates may act as vectors, including the isopod *Ceratothoa gaudichaudii* in Chile, or the copepod *Caligus rogercresseyi* in Patagonia (Arkush & Bartholomew 2011) but lack of a suitable vector may limit pathogen spread in the freshwater environment (Rozas & Enríquez 2014).

Caligus rogercresseyi is not present in New Zealand (Costello 2009). Endemic caligids including *C. elongates* (Lester and Hayward 2006); and isopods, including *C. gaudichaudii*, reported from the Chilean jack mackerel (*Trachurus symmetricus murphyi*) (Taylor 1999), represent potential vectors in New Zealand marine waters.

Mortalities associated with *P. salmonis* vary widely among isolates, between host species, and across geographical locations. The virulence and pathogenicity factors are poorly understood (Bohle *et al.* 2014; Rozas & Enríquez 2014). While mortalities of 10 to 20% occur in Atlantic salmon and rainbow trout in United States aquaculture, mortalities of 80 to 90% are reported for Atlantic salmon farmed in Chile, and in tilapia (*O. niloticus*) farmed in Taiwan (Kahn *et al*. 1999; Johnston 2008a). Experimental intraperitoneal infection of Atlantic cod (*Gadus morhua*) has resulted in a mortality rate of 77% over 122 days (Johnston 2008b).

Transmission is horizontal, through the water column, and *P. salmonis* is shed in fish bile, faeces and urine (Johnston 2008b), while entry occurs by ingestion, across the gill epithelium, or directly through the skin surface where it may be facilitated by scale damage (Tubbs *et al*. 2007). The details of the life cycle remain essentially unknown (Rozas & Enríquez 2014). The presence of *P. salmonis* in ovaries, coelomic fluid and testicles of infected fish indicates vertical transmission is possible (Rozas & Enríquez 2014).

The onset of disease may follow several pathways. In most fish, infection is initially focused on the haematopoietic tissues. Fish with systemic infection typically show signs of anorexia, with distended abdomens and exophthalmia. Coloration is usually dark, with haemorrhages at the base of the fins, periocular and perianal regions. This systemic disease progresses to splenomegaly, renomegaly and necrosis of the liver, myocardium, and intestinal epithelium, often with severe haemorrhagic meningoencephalitis (Reid *et al.* 2004; Arkush & Bartholomew 2011; Rozas & Enríquez 2014).

In some fish, infection may proceed with granulomatous inflammation and thrombosis of the skeletal musculature, leading to the formation of large exudate-filled cavities within the skeletal musculature. Infection is often accompanied by multifocal hyperplasia, necrosis and haemorrhaging of the gill lamellae (Arkush & Bartholomew 2011; Rozas & Enríquez 2014).

In other fish, neural infection is described in the brain and nervous tissues leading to severe haemorrhagic meningoencephalitis. Mortality may occur in these fish with no external signs of infection (Rozas & Enríquez 2014).

Piscirickettsia salmonis is environmentally resistant, remaining viable for up to 50 days in sea water at 5°C, although survival is limited to 10 days at 10°C and further reduced to 7 days at 20°C (Rozas & Enríquez 2014). It is rapidly inactivated by fresh water, unless protected in the host tissues or other biological materials (Lannan & Fryer 1994).

The minimum infectious dose is unknown, but it likely to be low, based on experimental data where 100% mortality occurred at $10^{1.9}$ $TCID_{50}$ (tissue culture infective dose) for coho salmon and $10^{2.1}$ $TCID_{50}$ for rainbow trout in intraperitoneal injection challenge (Kahn *et al.* 1999).

Piscirickettsia salmonis is unaffected by long-term frozen storage at -20°C (Johnston 2008b), but is inactivated by the freeze-thaw cycle (99% reduction in infectivity after one freeze-thaw cycle from -70°C) (Kahn *et al.* 1999). It is inactivated by chlorine (500 mg L^{-1} for 20 minutes) or iodine (100 ppm for 15 minutes) treatments (Tubbs *et al.* 2007). Inactivation by heat treatment occurs by heating to at least 100°C for 30 minutes (Rozas & Enríquez 2014).

Disease control by antibiotics is poor, with treatment used for prophylaxis in Chile leading to the development of antibiotic resistance (Henríquez *et al*. 2015). Although commercial vaccines have been available for a decade, vaccination is only partially successful. The vaccines are strain-specific, confer short-term resistance and side effects include a lowered resistance to more virulent isolates (Rozas & Enríquez 2014).

Potential host species in New Zealand include Chinook salmon (*O. tshawytscha*) and coho salmon (*O. kisutch*) and many native and introduced non-salmonid species in marine and fresh waters (Tubbs *et al*. 2007).

38.2. Risk assessment

38.2.1. Entry assessment

Piscirickettsia salmonis is primarily associated with juvenile farmed salmonids and adult fish are less likely to have clinical disease, However, other strains within the species complex infect a wide range of juvenile and adult fish, including tilapia, Atlantic cod, groupers and sea bass in marine, brackish and fresh waters (Arkush & Bartholomew 2011).

Clinically infected fish with external skin lesions, body abnormalities or exophthalmia (Arkush & Bartholomew 2011; Rozas & Enríquez 2014) would fail visual inspection, while evisceration would remove infected tissues associated with the heart, liver, kidney, spleen and digestive system (Rozas & Enríquez 2014).

Clinical infection may also occur with no external signs of infection, typically when associated with myeloencephalitis of the brain and nervous tissues (Reid *et al.* 2004; Rozas & Enríquez 2014), hyperplasia and lamellar fusion of the gill tissues, or inflammation and necrosis of the

skeletal musculature (Arkush & Bartholomew 2011; Rozas & Enríquez 2014). These tissues are retained upon evisceration and would be present in the commodity (Johnston 2008b). In addition, *P. salmonis* is likely to remain viable in chilled or frozen fish product during storage and transport (Johnston 2008b),

The likelihood of entry of the *P. salmonis* species complex is assessed as non-negligible.

38.2.2. Exposure assessment

Little is known about the mode of transfer of *P. salmonis* and associated PLOs (Rozas & Enríquez 2014). Infective tissues may be present in the trimmings of the skeletal musculature, as well as in the brain and gills of the fish heads discarded as offal after commercial fish processing. For disease to establish, sufficient infected uncooked fish scraps or offal would have to be discharged into the environment, at a level to become self-sustaining in the population (Kahn *et al.* 1999). This may occur through blood-water, or through offal discarded in commercial fish processing (Johnston 2008b; Rozas & Enríquez 2014).

Piscirickettsia salmonis is inactivated by exposure to fresh water (Arkush & Bartholomew 2011), but survival is extended where organic material is present (Rozas & Enríquez 2014). *Piscirickettsia salmonis* does not replicate outside a susceptible host (Kahn *et al*. 1999) and the lack of vectors in fresh water is considered a limiting factor to disease establishment in the freshwater environment (Rozas & Enríquez 2014). Therefore, the likelihood of exposure to the *P. salmonis* species complex and establishment in fresh water is assessed as negligible.

Piscirickettsia salmonis remains viable in frozen fish tissues (Kahn *et al.* 1999) and survives for up to 50 days in sea water (Rozas & Enríquez 2014). The minimum infective dose for fish is low (Kahn *et al.* 1999), while the distribution, host range and infectivity may be influenced by the presence of invertebrate vectors, such as the isopod *Ceratothoa gaudichaudii* or the copepod *Caligus rogercresseyi*. *C. gaudichaudii* is present in New Zealand, and endemic caligids may also function as vectors to transfer *P. salmonis* to salmon in sea cages. (Costello 2009).

The likelihood of introduction of the *P. salmonis* species complex in marine waters is assessed as non-negligible.

38.2.3. Consequence assessment

The establishment of the *P. salmonis* species complex in New Zealand would have significant economic consequences for salmonid aquaculture, through loss of sales due to infected product. This industry was valued at $63 million in export earnings in 2011 (Seafood New Zealand 2014). An outbreak would also affect recreational and tourist trout and salmon fishing, as well as incur significant social and environmental costs associated with other domestic fisheries (Fish & Game 2014). While the dollar value of New Zealand's freshwater fisheries is not fully known, the Taupo fishery alone is valued at $70 to 80 million per annum (Marsh & Mkwra 2013).

The consequences of establishment of the *P. salmonis* species complex in marine waters are assessed as non-negligible.

38.2.4. Risk estimation

As the entry, exposure and consequence assessments are non-negligible, the risk estimation is non-negligible. Therefore, *P. salmonis* species complex is assessed as a risk in the commodity and risk management measures may be justified.

38.3. Risk management

Piscirickettsia salmonis and the associated (RLO-like or PLO-like) organisms within the species complex have been assessed to be a risk in the commodity. Infection with *P. salmonis* is a non-notifiable disease. There is no specific guidance in the OIE *Aquatic Code* (OIE 2016) for importing eviscerated fish from infected countries, or specific processing requirements that would ensure the destruction of these pathogens. It is assumed that compliance with the approved processed states for OIE-listed diseases (Appendix 3) would also eliminate *P. salmonis* and associated species from the commodity and be a viable risk management option.

Piscirickettsia salmonis and associated species within the species complex have been reported from six families of marine and fresh water fish (Table 35), which may be present in the commodity. Species declaration should substantially reduce the pathogen load of the *P. salmonis* species complex in the commodity and be a viable risk management option.

Piscirickettsia salmonis and other organisms within this species complex have been reported from aquaculture in Europe (Ireland, Norway and Scotland), Egypt, North and South America (Chile), Asia (Taiwan), Hawaii and Tasmania. Restriction of the commodity to wild-caught fish (not from aquaculture) would have little, or no effect on pathogen load and is not a viable risk management option.

Where country/zone freedom is approved through the MPI Country Approval Procedures, this option is likely to substantially reduce the pathogen load of the *P. salmonis* species complex in the commodity. Approval of a declaration of country/zone freedom by MPI is a viable risk management option.

Infection is focused in the skin, brain, gills and skeletal muscle tissues of marine and fresh water hosts (Johnston 2008a), so removal of the gills, or the head and gills, should slightly reduce the pathogen load of the *P. salmonis* species complex in the commodity. Further processing to the skin-off fillet state should moderately reduce the pathogen load of the *P. salmonis* species complex in the commodity. Processing to the skin-off fillet stage is a viable risk management option.

Piscirickettsia salmonis is unaffected by freezing (to -24°), so frozen storage is not a viable risk management option. Members of the *P. salmonis* species complex are inactivated by cooking (to at least 100°C for 30 minutes) and this heat treatment is likely to eliminate these pathogens from the commodity. Heat treatment is a viable risk management option.

38.3.1. Risk management options

The *P. salmonis* species complex (including RLO-like or PLO-like organisms are reported from fish in families Cichlidae, Moronidae, Salmonidae, Sciaenidae, Serranidae and Tetraodontidae (Table 35), which are considered likely to be present in the commodity. Other families have not been associated with the *P. salmonis* species complex. Therefore species declaration indicating the

commodity is not originated from any of the above families should substantially reduce the occurrence of the *P. salmonis* species complex in the commodity.

For the commodities originated from families associated with the *P. salmonis* species complex, one or a combination of the following additional options could also be considered to effectively manage the risk.

Where country/zone freedom from the *P. salmonis* species complex is accepted by MPI:

Option 1

Acceptance of country/zone freedom by MPI should substantially reduce the occurrence of the *P. salmonis* species complex, so the commodity may be imported without any further restrictions.

Where country/zone freedom from the *P. salmonis* species complex is not accepted by MPI or not available:

Option 2

Processing to a state consistent with the approved states for OIE-listed diseases (Appendix 3) should eliminate the P. *salmonis* species complex. When these provisions are met, the commodity could be imported without any further restrictions.

Where the imported commodity is not in a state consistent with the approved states for OIE-listed diseases (Appendix 3):

Option 3

Heat treatment (by cooking to at least 100°C for at least 30 minutes) should eliminate the *P. salmonis* species complex. When this provision is met, the commodity could be imported without any further restrictions; or,

Option 4

Further processing (to the skin-off fillet state) should moderately reduce the occurrence of the *P. salmonis* species complex. When this provision is met, the commodity could be imported without any further restrictions.

38.4. References

Anonymous (2016). Biosecurity (Notifiable organisms) Order 2016 NZLR 2016/73. New Zealand Government, Wellington, 13 p.

Anthanassopoulou F, Groman D, Prapas T, Sabatakou O (2004). Pathological and epidemiological observations on rickettsiosis in cultured sea bass (*Dicentrarchus labrax* L.) from Greece. *Journal of Applied Ichthyology* 20: 525–545.

Arkush KD, Bartholomew JL (2011). *Piscirickettsia, Francisella* and epitheliocystis. pp 302–337. *In*: Woo PTK, Bruno DW (*Eds.*). *Fish Diseases and Disorders: Volume 3 - Viral, Bacterial and Fungal Infections*, 2nd edition. CABI International, Wallingford, United Kingdom, 930 p.

Bohle H, Henríquez P, Grothusen H, Navas E, Sandoval A, Bustamante F, Bustos P, Mancilla M (2014). Comparative genome analysis of two isolates of the fish pathogen *Piscirickettsia salmonis* from different hosts reveals major differences in virulence-associated secretion systems. *Genome Announcements* [Online] Available from: http://www.ncbi.nlm.nih.gov/pmc/articles/PMC4271152/ [Accessed 12 April 2015].

Corbeil S, Crane MS (2009). *Piscirickettsia salmonis. Australia New Zealand Standard Operating Procedure*. SCAHLS (Sub-committee on Aquatic Animal Health laboratory standards). Government of Australia, Canberra, Australia, 20 p.

Corbeil S, Hyatt AD, Crane M StJ (2005). Characterisation of an emerging rickettsia-like organism in Tasmanian farmed Atlantic salmon *Salmo salar. Diseases of Aquatic Organisms* 64: 37–44.

Costello MJ (2009). How sea lice from salmon farms may cause wild salmon declines in Europe and North America and be a threat to fishes elsewhere. *Proceedings of the Royal Society B*. 276: 3385–3394.

Fish and Game (2014). New Zealand Fish and Game Council [Online] Available from: http://www.fishandgame.org.nz/ [Accessed 24 September 2014].

Fryer JL, Hedrick RP (2003). *Piscirickettsia salmonis*: a Gram-negative intracellular pathogen of fish. *Journal of Fish Diseases* 26: 251–262.

Fryer JL, Lannan CN, Garces LH, Larenas JJ, Smith PA (1990). Isolation of a rickettsiales-like organism from diseased coho salmon (*Oncorhynchus kisutch*) in Chile. *Fish Pathology* 25(2): 107–114.

Fryer JL, Lannan CN, Giovannoni SJ, Wood ND (1992). *Piscirickettsia salmonis* gen. nov., sp. nov., the causative agent of an epizootic disease in salmonid fishes. *International Journal of Systematic Bacteriology* 42: 120–126.

Henríquez P. Bohle H, Bustamante F, Bustos P, Mancilla M (2015). Polymorphism in *gyrA* is associated with quinolone resistance in Chilean *Piscirickettsia salmonis* field isolates. *Journal of Fish Diseases* 38: 415–418.

Hine MP (2002). Results of a survey on shellfish health in New Zealand in 2000. *Surveillance* 29(1): 3–7.

Johnston C (2008a). Import risk analysis: frozen skinless and boneless fillet meat of *Pangasius* spp. fish from Vietnam for human consumption. MAF Biosecurity New Zealand, Wellington, New Zealand, 68 p.

Johnston C (2008b). Import risk analysis: frozen skinless and boneless fillet meat of *Oreochromis* spp. fish from China and Brazil for human consumption. MAF Biosecurity New Zealand, Wellington, New Zealand, 106 p.

Kahn SA, Beers PT, Findlay VL, Peebles IR, Durham PJ, Wilson DW, Gerrity SE (1999). Import risk analysis on non-viable salmonids and non-salmonid marine finfish. Australian Quarantine and Inspection Service, Department of Agriculture, Fisheries and Forestry, Canberra, Australia, 409 p.

Lannan C, Fryer J (1994). Extracellular survival of *Piscirickettsia salmonis. Journal of Fish Diseases* 17: 545–548.

Lester RJG, Hayward CL (2006). Phylum Arthropoda, pp 466–565 *In:* Woo PTK (*Ed.*). Fish Diseases and Disorders, Volume 1: Protozoan and Metazoan infections, 2nd edition. CABI International, Wallingford, United Kingdom, 791 p.

Mauel MJ, Miller DL (2002). Piscirickettsiosis and piscirickettsiosis-like infections in fish: a review. *Veterinary Microbiology* 87: 279–289.

Mauel MJ, Miller DL, Styer E, Pouder DB, Yanong RPE, Goodwin AE, Schwedler TE (2005). Occurrence of piscirickettsiosis-like syndrome in tilapia in the continental United States. *Journal of Veterinary Diagnostic Investigation* 17: 601–605.

Marsh D, Mkwra L (2013). Review of freshwater non-market value studies. Department of Economics, University of Waikato, Hamilton, New Zealand, 144 p.

OIE (2016). Aquatic Animal Health Code, 19th Edition. World Organization for Animal Health. Paris, France [Online] Available from: http://www.oie.int/international-standard-setting/aquatic-code/access-online/ [Accessed 15 March 2017].

Reid HL, Griffen AA, Birbeck TH (2004). Isolates of *Piscirickettsia salmonis* from Scotland and Ireland show evidence of clonal diversity. *Applied and Environmental Microbiology* 70: 4393–4397.

Rozas M, Enríquez R (2014). Piscirickettsiosis and *Piscirickettsia salmonis* in fish: a review. *Journal of Fish Diseases* 37: 163–188.

Seafood New Zealand (2014). Seafood New Zealand: Key facts for our Industry. Seafood New Zealand, Wellington, New Zealand, 6 p.

Stone MAB, MacDiarmid SC, Pharo HJ (1997). Import Risk Analysis; salmonids for human consumption. Ministry of Agriculture Regulatory Authority, New Zealand, 269 p.

Taylor PR (1999). Stock structure and population biology of the jack mackerel *Trachurus symmetricus murphyi*. Final Research Report for project JMA9802. National Institute of Water and Atmospheric Research, Auckland, New Zealand, 83 p.

Tubbs L, Lee P, Diggles B, Jones JB, Shepherd M, Sim-Smith C (2007). A review of aquatic diseases of significance to New Zealand. Final research report for project ZBS 2005-17. National Institute of Water and Atmospheric Research, Wellington, New Zealand, 452 p.

39. *Pseudomonas anguilliseptica*

39.1. Hazard identification

39.1.1. Aetiological agent

Pseudomonas anguilliseptica is a Gram-negative, rod-shaped motile bacterium with polar flagella. It was first described as "red spot" disease in Japanese eels (Wakabayashi & Egusa 1972), but has subsequently been reported from many other marine and freshwater hosts (Daly & Aoki 2011). Genomic analysis indicates three different serotypes exist, but these show high phenotypical similarity regardless of their host or geographical distribution (López-Romalde *et al*. 2003). Serotype 1 (K-) has been isolated from Japanese eels (*Anguilla japonica*). Serotype 2 (K+) has been isolated from ayu (*Plecoglossus altivelis*), Atlantic salmon (*Salmo salar*), brown sea trout (*Salmo trutta*) and European whitefish (*Coregonus lavaretus*). Serotype 3 is reported from a wide range of other hosts and geographical areas (Wiklund & Bylund 1990; Daly & Aoki 2011).

39.1.2. OIE status

Infection with *P. anguilliseptica* is not listed by the OIE (OIE 2016).

39.1.3. New Zealand status

Pseudomonas anguilliseptica has not been reported from New Zealand and is considered exotic (Duignan *et al.* 2003). Infection with *P. anguilliseptica* is not a notifiable disease in New Zealand (Anon. 2016).

39.1.4. Epidemiology

Pseudomonas anguilliseptica is relatively difficult to culture and easily confused with other marine pathogens, such as *Tenacibaculum maritimum* (Austin & Austin 2007; Austin 2011). It has been regarded as an opportunist pathogen of barramundi (*Lates calcarifer*) and estuarine grouper (*Epinephelus tauvina*) in Malaysian sea cages (Nash *et al.* 1987). This was based on the relatively high lethal dose, the importance of stress factors such as water quality and stocking levels in moderating infection and pathogenicity, as well as feeding with trash fish associated with infection.

Pseudomonas anguilliseptica is an emerging pathogen (Daly 1999; López-Romalde *et al*. 2003; Magi *et al*. 2009; Daly & Aoki 2011) causing "winter disease" (Berthe *et al*. 1995) in a wide range of fish in marine, brackish and fresh water (Table 36) (Domenech *et al*. 1997; Ferguson *et al*. 2004; Magi *et al*. 2009; Daly & Aoki 2011; Wheeler 2012).

Serotype 1 (K-) occurs only in eels (*Anguilla* spp.), but serotype 2 (K+) has been reported from Japanese ayu (*Plecoglossus altivelis*) and from Atlantic salmon (*Salmo salar*), sea-run brown trout (*S. trutta*) and whitefish (*Corregonus* sp.) in Finnish sea-cage culture (Wiklund & Bylund 1990). Serotype 3 has been reported from fish (Table 37) from Australia, Canada, Denmark, Egypt, Finland, France, India, Japan, Malaysia, Portugal, Spain, Taiwan and the United Kingdom (Berthe *et al.* 1995; Daly 1999; Balboa *et al.* 2003; Wheeler 2012; Andree *et al*. 2013; Mastan 2013). Genetically similar (99%) isolates also occur in deep-sea microbial communities, and in hydroids and ascidians off the coast of Fiji (Romanenko *et al.* 2008). Isolates show phenotypic homogeneity, regardless of the host from which they were isolated (López-Romalde *et al.* 2003).

Other fish may be experimentally infected, including bluegill sunfish (*Lepomis macrochirus*), goldfish (*Carassius auratus*), carp (*Cyprinus* spp.) and loach (*Misgurnus anguillicaudatus*) (Daly & Aoki 2011).

Table 36. Families and Species of Fish Susceptible to *Pseudomonas Anguilliseptica,* by Serotype

Family	Species	Serotype
Anguillidae	European eel (*Anguilla anguilla*), Japanese eel (*A. japonica*)	I
Centrarchidae	Bluegill (*Lepomis macrochirus*)*	I
Cobitidae	Pond loach (*Misgurnus anguillicaudatus*)*	I
Cyprinidae	Carp (*Cyprinus* sp.), crucian carp (*Carassius carassius*), goldfish (*C. auratus*)*	I
Salmonidae	Rainbow trout *Oncorhynchus mykiss*), amago (*O. rhodurus*), kokanee salmon (*O. nerka*) , whitespotted char (*Salvelinus leucomaensis*)	I
Plecoglossidae	Ayu (*Plecoglossus altivelis*)	II
Salmonidae	Atlantic salmon (*Salmo salar*), rainbow trout (*Oncorhynchus mykiss*), brown trout (*Salmo trutta*), whitefish (*Coregonus* sp.)	II
Carangidae	Striped jack (*Pseudocaranx dentex*)	III
Cichlidae	Nile tilapia (*Oreochromis niloticus*)	III
Clupeidae	Atlantic herring (*Clupea harengus*), Baltic herring (*C. harengus membras*)	III
Cyprinidae	Rohu (*Labeo rohita*), carp (*Cyprinus carpio*)	III
Gadidae	Atlantic cod (*Gadus morhua*)	III
Latidae	Barramundi (*Lates calcarifer*)	III
Moronidae	Sea bass (*Dicentrarchus labrax*)	III
Plecoglossidae	Ayu (*Plecoglossus altivelis*)	III
Plotosidae	Australian eel tailed catfish (*Tandanus tandanus*)	III
Salmonidae	Atlantic salmon (*Salmo salar*), rainbow trout (*O. mykiss*), brown trout (*Salmo trutta*), whitefish (*Coregonus* sp.)	III
Scophthalmidae	Turbot (*Scophthalmus maximus*)	III
Serranidae	Orange-spotted grouper (*Epinephelus coioides*), greasy grouper (*E. tauvina*)	III
Sparidae	Black spot sea bream (*Pagellus bogaraveo*), blackhead sea bream (*Acanthopagrus schlegelii*), gilthead sea bream (*Sparus aurata*)	III

Where * indicates experimental infection

Mortality varies among species and isolates (Sakr & El-Rhman 2008), while all sizes of fish may be infected (Jansson & Vennerstrom 2014). Mortalities range from 2% in farmed Atlantic cod (*S. salar),* up to 50% in salmonids and may reach 80% in tilapia (*Oreochromis* spp.) (Wiklund & Bylund 1990; Sakr & El-Rhman 2008; Jansson & Vennerstrom 2014; Fadel *et al.* 2018). Pathogenicity is temperature-related, but varies among serotypes. Infection with serotype 1 can be eliminated in eels where temperatures are maintained above 26°C, although this temperature is too high for salmonid aquaculture (Daly & Aoki 2011). Infection with serotype 3 does not cause economic disease when temperatures are maintained above 16°C (Jansson & Vennerstrom 2014).

The infective dose varies widely among host species, from 1 x 10^3 to 1 x 10^8 cfu $fish^{-1}$ (colony forming units) ((Nash *et al*. 1987); Romalde *et al.* 2004; Magi *et al*. 2009), as determined by intraperitoneal challenge and surviving fish can act as carriers (Romalde *et al*. 2004). Serotype 1 isolates from European eels (*Anguilla anguilla*) are highly pathogenic to Japanese eel (*A. japonica*), pond loach (*Misgurnus anguillicaudatus*) and sunfish (*Lepomis macrochirus*), but of lower pathogenicity to cyprinids (Muroga *et al.* 1975) and salmonids (Wiklund & Bylund 1990). Serotype 2 isolates from Baltic herring (*C. harengus membras*) are of low pathogenicity to rainbow trout (*O. mykiss*), but able to establish a carrier state (Lonnstrom *et al.* 1994). Serotype 3 isolates infect a wide variety of hosts (Daly & Aoki 2011).

External signs of clinical infection include loose scales, skin haemorrhages and ulcers, anorexia, exophthalmia and abdominal distention, while petechial haemorrhaging causes the skin and fin

bases to become reddened (Kahn *et al*. 1999; Magi *et al*. 2009; Mastan 2013). The fins may be eroded and the gills can develop extreme hyperplasia, haemorrhaging and oedema of the filaments (Sakr & El-Rhman 2008).

Disease progression may follow several pathways. Systemic infection, which first develops in the gills, quickly spreads to the remainder of the body (Ferguson *et al*. 2004; Magi *et al.* 2009), with necrotic vacuolation and degeneration of the liver, kidney and spleen tissue (Sakr & El-Rhman 2008). This progresses to the connective tissue, skeletal musculature, brain and cartilage, with pyogranulomatous inflammation and myolysis of the musculature occurring after 7 days post-infection (Nakai 1985; Sakr & El-Rhman 2008).

In cephalic infection, *P. anguilliseptica* preferentially infects brain tissue. Infected fish develop corneal opacity, cephalic osteochondritis and meningitis after 9-10 days post-infection (Magi *et al*. 2009). Cephalic tissue was infected in 40% of surviving carrier gilt-head sea bream (*Sparus aurata*) (Romalde *et al*. 2004; Fadel *et al.* 2018), while ocular tissue infection was also reported in 33% of infected Baltic herring (*C. h. membras*) (Lonnstrom *et al*. 1994) and both conditions may occur in addition to visceral infection.

Disease transmission is horizontal, with initial pathogen entry occurring across the digestive or gill epidermal cells, although virulence details are poorly understood (Austin 2006). Wild fish stocks, such as Baltic herring (*C. h. membras*) and Atlantic cod (*S. salar*), may act as a reservoir for infection of adjacent farmed species (Lonnstrom *et al*. 1994).

Pseudomonas anguilliseptica remains viable on dead fish tissue (Sakr & El-Rhman 2008). It is resistant to environmental conditions, surviving for long periods in fresh and marine waters (Ciric *et al*. 2009).

Pseudomonas anguilliseptica is cold-tolerant, remaining viable after freezing at -70°C (Magi *et al*. 2009). Infectivity declines with rising temperature and no growth occurs above 37°C (Ferguson *et al*. 2004). It is inactivated by heat treatment to 80°C for 20 minutes, or 50°C for 30 minutes (Nakai 1985) and by ozone treatment (Tian *et al*. 2014).

Pseudomonas anguilliseptica can be denatured by florfenicol, ciprofloxacin, nitrofurantoin and oxytetracycline (Wiklund & Bylund 1990; Fadel *et al*. 2018), but resistance to tetracycline and ciprofloxacin is reported (Kholil *et al*. 2015). No commercial vaccine is currently available (Daly & Aoki 2011).

Potential hosts in New Zealand include all salmonids (*Oncorhynchus* spp., *Salmo* spp., *Salvelinus* spp.), flatfish (Order Pleuronectiformes) and herrings (Family Clupeidae) (Tubbs *et al.* 2007).

39.2. Risk assessment

39.2.1. Entry assessment

Clinically infected fish with external signs of infection, such as haemorrhagic skin lesions, or fin wasting, should be rejected in visual examination (Kahn *et al*. 1999), but carrier fish may show few or no external signs of infection (Sakr & El-Rhman 2008; Ferguson *et al*. 2009). These may pass visual inspection and be present in the commodity.

Infection with *P. anguilliseptica* may affect brain, gill, eye, neurological, skin and muscle tissues (Lonnstrom *et al*. 1994; Romalde *et al*. 2004), that are likely to be retained in the eviscerated commodity.

Pseudomonas anguilliseptica is resistant to freezing (Magi *et al*. 2009), survives in dead fish tissue (Sakr & El-Rhman 2008) and may remain viable in marine and fresh waters for extended periods (Ciric *et al*. 2009). It is reported from wild fish hosts including eels, herring and cod, which may function as a reservoir for subsequent infection of farmed fish (Lonnstrom *et al.* 1994; Daly and Aoki 2011). Potential marine and brackish water fish hosts are present in New Zealand (Tubbs *et al.* 2007).

The likelihood of entry is assessed as non-negotiable.

39.2.2. Exposure assessment

For disease to establish, sufficient infected blood-water, fish scraps or offal would have to be discharged from fish processing facilities into the environment, at a level to become self-sustaining in the population (Kahn *et al.* 1999; Johnston 2008). *Pseudomonas anguilliseptica* is known to survive in dead fish tissue and is environmentally stable (Daly & Aoki 2011). Repeated high doses of *P. anguilliseptica* discharged in blood-water or organic offal from a fish processing facility may be infective to susceptible hosts, including eels (*Anguilla* spp.) in fresh water (Hine & MacDiarmid 1994), or herrings (*Clupea harengus*) in marine waters (Lonnstrom *et al*. 1994).

Pseudomonas anguilliseptica may remain viable in offal dumped to landfill and has been recovered from groundwater (Tian *et al.* 2014). It is denatured by temperatures above 37°C (Ferguson *et al.* 2004), and is unlikely to survive passage through the avian digestive system.

The likelihood of exposure to *P. anguilliseptica* is assessed as non-negligible.

39.2.3. Consequence assessment

The consequence of an outbreak of *P. anguilliseptica* would be dependent on the virulence and pathogenicity of the introduced strain (López-Romalde *et al.* 2003; Sakr & El-Rhman 2008, Jansson & Vennerstrom 2014). Treatment measures are largely ineffective and it is poorly controlled by antibiotics (Daly & Aoki 2011).

Aquaculture in New Zealand is heavily dependent on high quality water supplies (Sim-Smith *et al.* 2014). An outbreak would be likely to affect the native eel population, with significant social and environmental costs (Fish & Game 2014). The control or eradication of *P. anguilliseptica* in these fisheries would be difficult (Kahn *et al*. 2009).

Pseudomonas anguilliseptica is pathogenic to salmonids (Romalde *et al*. 2004; Magi *et al.* 2009). An outbreak could affect recreational and tourist fisheries for brown trout (*S. trutta*) and rainbow trout (*O. mykiss*) and incur significant social and environmental costs to other domestic fisheries (Fish & Game 2014). While the dollar value of New Zealand's freshwater fisheries is not fully known, the Lake Taupo trout fishery alone is valued at $70 to 80 million per annum (Marsh & Mkwra 2013).

Pseudomonas anguilliseptica infects several *Oncorhynchus* hybrids (Wiklund & Bylund 1990) but there is no clear evidence of natural infection in Chinook salmon (*O. tshawytscha*). The potential effect of an outbreak in Chinook salmon would be significant for the salmon farming industry, through loss of sales due to infected product. This was valued at $63 million in export earnings in 2011 (Seafood New Zealand 2014).

Pseudomonas anguilliseptica may infect wild stocks of commercially important inshore marine species (Lonnstrom *et al*. 1994), including snapper (*Pagrus auratus*), valued at NZ$ 6.5 million,

and jack mackerel (*Trachurus* spp.) valued at NZ$ 64 million in exports in 2012 (Statistics New Zealand 2014). Other species such as herring (*Clupea pallasii*), support smaller commercial, recreational and traditional Maori fisheries (MPI 2014). A reduction in their abundance due to disease would be likely to incur social and environmental costs.

The consequences of establishment are assessed as non-negligible.

39.2.4. Risk estimation

Since the entry, exposure, and consequence assessments are non-negligible, the risk estimation is non-negligible. Therefore, *P. anguilliseptica* is assessed to be a risk in the commodity and risk management measures may be justified.

39.3. Risk management

Pseudomonas anguilliseptica has been assessed to be a risk in the commodity. Infection with *P. anguilliseptica* is a non-notifiable disease, so the *Aquatic Code* (OIE 2016) provides no guidance for importing eviscerated fish from infected countries, or specific processing requirements that would ensure the destruction of the pathogen. It is assumed that compliance with the approved processed states for OIE-listed diseases (Appendix 3) would also eliminate *P. anguilliseptica* from the commodity and be a viable risk management option.

Several families of marine and fresh water fish (Table 36) that are susceptible to *P. anguilliseptica* may be present in the commodity. Species declaration should substantially reduce the pathogen load of *P. anguilliseptica* in the commodity and is a viable risk management option. As clinical disease is mainly associated with aquaculture (Nash *et al.* 1987), restriction of the commodity to wild-caught fish (not from aquaculture) should moderately reduce pathogen load and be a viable risk management option.

Pseudomonas anguilliseptica is widely reported from Asia (Japan, Malaysia and Taiwan), Australia, Canada and Europe (Denmark, Egypt, Finland, France, Portugal, Spain, and the United Kingdom). Where country/zone freedom is approved through the MPI Country Approval Procedures, this option is likely to substantially reduce the pathogen load of *P. anguilliseptica* in the commodity. Approval of a declaration of country/zone freedom by MPI is a viable risk management option.

Pseudomonas anguilliseptica is associated with connective tissue, brain and gill tissue of marine and freshwater fish, so removal of the gills would only slightly reduce the pathogen load of *P. anguilliseptica* in the commodity. Removal of the head and gills should moderately reduce the pathogen load of *P. anguilliseptica*, as residual levels of the pathogen may remain in the skeletal musculature. Removal of the head and gills is a viable risk management option.

Pseudomonas anguilliseptica is unaffected by freezing, so low-temperature storage is not a viable risk management option. High-temperature treatment (by cooking to 80°C for 20 minutes) will denature *P. anguilliseptica* and eliminate it from the commodity. High-temperature treatment is a viable risk management option.

39.3.1. Risk management options

Pseudomonas anguilliseptica is reported from fish in families Anguillidae, Carangidae, Centrarchidae, Cichlidae, Clupeidae, Cobitidae, Cyprinidae, Gadidae, Latidae, Moronidae,

Plecoglossidae, Plotosidae, Salmonidae, Scophthlmidae, Serranidae and Sparidae (Table 36), which are considered likely to be present in the commodity. Other families have not been associated with *P. anguilliseptica*. Therefore species declaration indicating the commodity is not originated from any of the above families should substantially reduce the occurrence of *P. anguilliseptica* in the commodity.

For the commodities originated from families associated with *P. anguilliseptica*, one or a combination of the following additional options could be considered to effectively manage the risk.

Where country/zone freedom from *P. anguilliseptica* is accepted by MPI:

Option 1

Acceptance of country/zone freedom by MPI should substantially reduce the occurrence of *P. anguilliseptica*, so the commodity may be imported without any further restrictions.

Where country/zone freedom from *P. anguilliseptica* is not accepted by MPI or not available:

Option 2

Processing to a state consistent with the approved states for OIE-listed diseases (Appendix 3) should eliminate *P. anguilliseptica.* When these provisions are met, the commodity could be imported without any further restrictions.

Where the imported commodity is not in a state consistent with the approved states for OIE-listed diseases (Appendix 3):

Option 3

Heat treatment (by cooking to at least 80°C for 20 minutes) should eliminate *P. anguilliseptica*. When this provision is met, the commodity could be imported without any further restrictions; or,

Option 4

Further processing (by removal of the head and gills) should moderately reduce the occurrence of *P. anguilliseptica*. When this provision is met, the commodity could be imported without any further restrictions; or,

Option 5

Restriction of the commodity to wild-caught fish (not from aquaculture) should moderately reduce the occurrence of *P. anguilliseptica.* When this provision is met, the commodity could be imported without any further restrictions.

39.4. References

Anonymous (2016). Biosecurity (Notifiable Organisms) Order 2016 SR 2016/73. New Zealand Government, Wellington, 13 p.

Andree KB, Rodgers CJ, Furones D, Gisbert E (2013). Co-infection with *Pseudomonas anguilliseptica* and *Delftia acidovorans* in the European eel, *Anguilla anguilla* (L.): a case history of an illegally trafficked protected species. *Journal of Fish Diseases* 36: 647–656.

Austin B (2006). Bacterial pathogens of marine fish, pp 307–330 *In*: Belken S, Colwell RR (*Eds*.). Oceans and Health: Pathogens in the marine environment. Springer Science, New York, United States of America, 478 p.

Austin B (2011). Taxonomy of bacterial fish pathogens. *Veterinary Research* 42: 20 [Online] Available from: http://www.biomedcentral.com/content/pdf/1297-9716-42-20.pdf [Accessed 2 October 2015].

Austin B, Austin DA (2007). Bacterial Fish Pathogens: Disease in Farmed and Wild Fish. Springer, New York, United States of America, 457 p.

Balboa S, Ferguson HW, Romalde JL (2003). Phenotypic, serological and genetic characterisation of *Pseudomonas anguilliseptica* strains isolated from cod, *Gadus morhua* L., in northern Europe. *Journal of Fish Diseases* 30: 657–664.

Berthe FCJ, Michael C, Bernardet JF (1995). Identification of *Pseudomonas anguilliseptica* isolated from several species in France. *Diseases of Aquatic Organisms* 21: 151–155.

Ciric L, Philp JC, Whiteley S (2009). Hydrocarbon utilisation within a diesel-degrading bacterial consortium. *FEMS Microbiology Letters* 303(2): 116–122.

Daly JG (1999). Other bacterial pathogens. pp 577 –597. *In*: Woo PTK, Bruno DW (*Eds*.) Fish Diseases and Disorders. Vol. 3, Viral, Bacterial and Fungal infections. CABI International, London, New York, United States of America, 930 p.

Daly JG, Aoki T (2011). Pasteurellosis and other bacterial diseases. pp 648–668. *In:* Woo PTK Bruno DW (*Eds.*). *Fish Diseases and Disorders: Volume 3 - Viral, Bacterial and Fungal Infections*, 2nd edition. CABI International, Wallingford, United Kingdom, 930 p.

Domenech A, Fernandez-Garayzabal JF, Lawson P, Garcia JA, Cutuli JA, Blanco-Gutierrez M, Gibello A, Moreno MA, del Collins MD, Dominguez L (1997). Winter disease outbreak in sea bream (*Sparus aurata*) associated with *Pseudomonas anguilliseptica* infection. *Aquaculture* 156 (3–4): 317–326.

Duignan PJ, Hine PM, Joy M, Gibbs N, Jones GW, Okeoma C (2003). Disease surveillance in freshwater fish from the lower North Island. *Surveillance* 30(3): 6–8.

Fadel A, Mabrok M, Aly S (2018). Epizootics of *Pseudomonas anguilliseptica* among cultured seabream (*Sparus aurata*) populations: Control and treatment strategies. *Microbial Pathogenesis* 121: 1–8 [Online] Available from: https://doi.org/10.1016/j.micpath.2018.04.021 [Accessed 8 August 2018].

Ferguson HW, Collins RO, Moore M, Coles M, MacPhee DD (2004). *Pseudomonas anguilliseptica* infection in farmed cod, *Gadus morhua* L. *Journal of Fish Diseases* 27: 249–253.

Fish and Game (2014). New Zealand Fish and Game Council [Online] Available from: http://www.fishandgame.org.nz/ [Accessed 24 September 2014].

Hine PM, MacDiarmid SC (1994). Contamination of fish products: risks and prevention. *OIE Review of Science and Technology* 16(1): 135–145.

Jansson E, Vennerstrom P (2014). Infectious diseases of cold-water fish in marine and brackish waters. pp 15–59. *In:* Woo PTK, Bruno DW (*Eds*.) Diseases and disorders of finfish in cage culture, 2nd edition. CABI International, Wallingford, United Kingdom, 354 p.

Johnston C (2008). Import risk analysis: frozen skinless and boneless fillet meat of *Oreochromis* spp. fish from China and Brazil for human consumption. MAF Biosecurity New Zealand, Wellington, 106 p.

Kahn SA, Beers PT, Findlay VL, Peebles IR, Durham PJ, Wilson DW, Gerrity SE (1999). Import risk analysis on non-viable salmonids and non-salmonid marine finfish. Australian Quarantine and Inspection Service, Department of Agriculture, Fisheries and Forestry, Canberra, Australia, 409 p.

Kholil MI, Hossain MM, Neowajh S, Islam MS, Kabir M (2015). Comparative efficiency of some commercial antibiotics against *Pseudomonas* infection in fish. *International Journal of Fisheries and Aquatic Studies.* 2(3): 114–117.

Lonnstrom L, Wiklund T, Bylund G (1994). *Pseudomonas anguilliseptica* isolated from Baltic herring *Clupea harengus membras* with eye lesions. *Diseases of Aquatic Organisms* 18: 143–147.

López-Romalde SL, Magarinos B, Nunez S, Toranzo AE, Romalde JL (2003). Phenotypic and genetic characterisation of *Pseudomonas anguilliseptica* strains isolated from fish. *Journal of Aquatic Animal Health* 15: 39–47.

Magi GE, López-Romalde S, Magarinos GE, Lamas L, Toranzo AE, Romalde L (2009). Experimental *Pseudomonas anguilliseptica* infection in turbot *Psetta maxima* (L.): a histopathological and immunohistochemical study. *European Journal of Histochemistry* 53(2) [Online] Available from: http://www.ncbi.nlm.nih.gov/pmc/articles/PMC3167283/ [Accessed 18 December 2014].

Marsh D, Mkwra L (2013). Review of freshwater non-market value studies. Department of Economics, University of Waikato, Hamilton, New Zealand, 144 p.

Mastan SA (2013). *Pseudomonas* septicaemia in *Labeo rohita* (Ham.) and *Cyprinus carpio* (Linn.) in Andhra Pradesh – natural occurrence and artificial challenge. *International Journal of Pharmacy and Pharmaceutical Sciences* 5(2): 564–568.

Ministry for Primary Industries (MPI) (2014). Fisheries Assessment Plenary, May 2014: stock assessments and stock status. Compiled by the Fisheries Science Group, Ministry for Primary Industries, Wellington, New Zealand, 1381 p.

Muroga K, Jo Y, Sadawa T (1975). Studies on red spot disease of pond cultured eels. II: Pathogenicity of the causative bacterium *Pseudomonas anguilliseptica. Fish Pathology (Japan)* 9(2): 107–114.

Nakai T (1985). Resistance of *Pseudomonas anguilliseptica* to bactericidal action of fish serum. *Bulletin of the Japanese Society of Scientific Fisheries* 51(9): 1431–1436.

Nash G, Anderson IA, Shariff M, Shamsudin M (1987). Bacteriosis associated with epizootic in the giant sea perch, *Lates calcarifer*, and the estuarine grouper *Epinephelus tauvina*, cage culture in Malaysia. *Aquaculture* 67 (1–2): 105–111.

OIE (2016). Aquatic Animal Health Code, 19th Edition. World Organization for Animal Health. Paris, France [Online] Available from: http://www.oie.int/international-standard-setting/aquatic-code/access-online/ [Accessed 15 March 2017].

Romalde JL, López-Romalde SL, Ravelo C, Magarinos B, Toranzo AE (2004). Development and validation of a PCR- based protocol for the detection of *Pseudomonas anguilliseptica. Fisheries Pathology* 19: 33–41.

Romanenko L, Uchino M, Tebo RM, Tanaka N, Frolova GM, Mikhailov VV (2008). *Pseudomonas marincola* sp. nov., isolated from marine sediments. *International Journal of Systematic and Evolutionary Microbiology* 58: 706–710.

Sakr SFM, El-Rhman AMM (2008). Contribution on *Pseudomonas* septicaemia caused by *Pseudomonas anguilliseptica* in cultured *Oreochromis niloticus*. pp 1177–1197. *In*: From the Pharoes to the Future: Proceedings of the Eighth International Symposium on Tilapia in Aquaculture, October 12–14, Cairo, Egypt [Online]. Available from: http://ag.arizona.edu/azaqua/ista/ ISTA8/ProceedingsISTA8.htm [Accessed 3 October 2014].

Seafood New Zealand (2014). Seafood New Zealand: Key facts for our Industry. Seafood New Zealand, Wellington, New Zealand, 6 p.

Sim-Smith C, Faire S, Lees A (2014). Managing biosecurity risk for business benefit. Aquaculture biosecurity practices research report prepared for Ministry for Primary Industries, by Coast and Catchment Ltd, New Zealand Wellington, New Zealand, 214 p.

Statistics New Zealand (2014). New Zealand's seafood industry [Online]. Available from: http://www.stats.govt.nz/tools_and_services/newsletters/price-index-news/apr-13-article-seafood.aspx [Accessed 6 April 2015].

Tian J, Lu J, Zhang Y, Cheng-Li J, Chen-Sun L, Li-Hu Z (2014). Microbial community structures and dynamics in the 0_3/BAC drinking water treatment process. *International Journal of Environmental Research and Public Health* 11(6): 6281–6290.

Tubbs L, Lee P, Diggles B, Jones JB, Shepherd M, Sim-Smith C (2007). A review of aquatic diseases of significance to New Zealand. Final research report for project ZBS 2005-17. National Institute of Water and Atmospheric Research, Wellington, New Zealand, 452 p.

Wakabayashi H, Egusa S (1972). Characteristics of a *Pseudomonas* sp. from an epizootic of pond-cultured eels (*Anguilla japonica*). *Bulletin of the Japanese Society of Scientific Fisheries* 38: 577–587.

Wheeler M (2012). Perth Metro lakes surveyed for introduced fish. Fisheries and Water, Science Network WA, Australia, Government of Western Australia [Online] Available from: http://sciencewa.net.au/topics/fisheries-a-water/item/2764-perth-metro-lakes-surveyed-for-introduced-fish [Accessed 3 October 2014].

Wiklund T, Bylund G (1990). *Pseudomonas anguilliseptica* as a pathogen of salmonid fish in Finland. *Diseases of Aquatic Organisms* 8: 13–19.

40. *Renibacterium salmoninarum*

40.1. Hazard identification

40.1.1. Aetiological agent

Renibacterium salmoninarum is a coryneform rod-shaped, non-motile Gram-positive bacterium that is the only species in the Genus *Renibacterium* (Sanders & Fryer (1980) and classified within the *Micrococcus-Arthrobacter* subdivision of the Actinomycetes (Wiens 2011). It is the causative agent of bacterial kidney disease (BKD), also known as cornyobacterial kidney disease, Dee disease, salmonid kidney disease or white boil disease (Wiens 2011). It was first described from Scottish salmonids (Mackie *et al*. 1933). BKD is a disease of farmed and wild salmonids in fresh water (Kahn *et al.* 1999). Genomic analysis has identified two separate lineages of *R. salmoninarum*. Lineage 1 has been widely distributed through live fish movements, while Lineage 2 is endemic throughout Europe and the Eastern Atlantic salmon (*Salmo salar*) stocks. Isolates within each lineage are recovered from many different salmonids (*Salvelinus* spp., *Salmo* spp., *Oncorhynchus* spp.) and are almost genetically indistinguishable, indicating free inter-species transmission (Brynildsrud *et al.* 2013).

40.1.2. OIE status

Infection with *R. salmoninarum* is not listed by the OIE (OIE 2016).

40.1.3. New Zealand Status

Renibacterium salmoninarum has not been reported from surveys in New Zealand (Anderson and Knowles 1999), and is considered exotic (Johnston & Knowles 1999; Tubbs *et al*. 2007; Diggles 2011). BKD is a notifiable disease in New Zealand (Anon. 2016).

40.1.4. Epidemiology

BKD has been reported from most areas where salmonids are cultured, including Western Europe, Scandinavia, Iceland, North America, South America, and Japan (Johnston & Knowles 1999). It is not reported from Australia or New Zealand (Kahn *et al.* 1999; Diggles 2011; Anon. 2017).

BKD is primarily a disease affecting salmonids (Table 37) , particularly Chinook salmon (*Oncorhynchus tshawytscha*) and Atlantic salmon (*Salmo salar*) (Mitchum *et al*. 1979; Wiens 2011). The disease occurs in fresh water, but fish may remain infected after smoltification in sea cage aquaculture (Johnston & Knowles 1999).

Natural infection with BKD is also reported from non-salmonids which may be associated with aquaculture (Kahn *et al*. 1999). These include the Plecoglossidae (ayu, *Plecoglossus altivelis*) from Europe; the Hexagrammidae (greenling, *Hexagrammos otakii*) and Platycephalidae (flathead, *Platycephalus indicus*) from Japan, and the Merlucciidae (Pacific hake, *Merluccius productus*) from Canada. These opportunistic infections are likely to represent reservoirs for infection (Wiens 2011; Hershberger *et al.* 2013).

Other fish can be experimentally infected including the Anoplopomatidae (sablefish, *Anoplopoma fimbria*), Clupeidae (Pacific herring, *Clupea harengus*), Cyprinidae (common shiner, *Notropis cornutus*, flathead minnow, *Pimephales promelas*), and Embiotocidae (shiner perch,

Cymatogaster aggregata), although no natural infections are reported in these species (Bell *et al.* 1990; Kahn *et al.* 1999; Wiens 2011).

Table 37. Families and Species of Fish Susceptible to *Renibacterium salmoninarum*

Family	Host Species
Anoplopomatidae	Sablefish (*Anoplopoma fimbria*)*
Clupeidae	Atlantic herring (*Clupea harengus*)*
Cyprinidae	Common shiner (*Notropis cornutus*), flathead minnow (*Pimephales promelas*)*
Embiotocidae	Shiner perch (*Cymatogaster aggregata*)
Hexagrammidae	Fat greenling (*Hexagrammos otakii*)
Merlucciidae	Pacific hake (*Merluccius productus*)
Platycephalidae	Flathead (*Platycephalus indicus*)
Plecoglossidae	Ayu (*Plecoglossus altivelis*)
Salmonidae	Salmon (*Hucho* spp., *Oncorhynchus* spp., *Salmo* spp., *Salvelinus* spp., grayling (*Thymallus* spp.), whitefish (*Coregonus* spp.)

Where * indicates experimental infection

Renibacterium salmoninarum affects fish of all ages (Kahn *et al*. 1999; Wiens 2011), with reported mortalities of 40% for Scottish Atlantic salmon (*S. salar*) and 80% in Canadian Chinook salmon (*Oncorhynchus tshawytscha*) (Wiens 2011).

Epizootic infections also occur in wild salmonids (Mitchum *et al*. 1979), but sub-clinical infection is widely reported from Continental Europe, North America, Scotland, Scandinavia, South America, Turkey and Japan (Wiens 2011; Hershberger *et al.* 2013; Ozturk & Altinok 2014), with no clinical signs of infection. Reported prevalence varies widely, from 3%–100% in wild Icelandic Arctic charr (*Salvalinus alpinus*) and from 6% to 81% in wild brown trout (*Salmo trutta*) (Jónsdottir *et al.* 1998).

Horizontal infection occurs through direct contact with diseased or carrier fish, through faecal-oral transmission (Kent *et al.* 1998; Wiens 2011), although details of the epidemiology are unclear (Hershberger *et al.* 2013). Disease transmission may also be vertical, through infected eggs from broodstock in hatcheries, while ovarian fluid may also be infected (Wiens 2011).

Pathogen entry occurs through the skin epidermal cells, across the mucous membranes of the gills, eye, or digestive tract, and bacterial cells are present in host phagocytes within 45 minutes of intraperitoneal injection (Densmore 1997; Wiens 2011).

Clinical disease progresses slowly, typically requiring at least 9 months before external signs become apparent (Kahn *et al.* 1999; Hershberger *et al.* 2013) and infected fish may show darkening of the skin, with exophthalmia (Kahn *et al*. 1999; Wiens 2011). As the disease becomes systemic, further external signs include petechial haemorrhages, skin lesions and ulcers (Wiens 2011). Internal infection is characterised by the development of inflammatory granulomas and necrosis of kidney, liver, skeletal musculature and subcutaneous tissues (Johnston & Knowles 1999; Wiens 2011).

Infection in non-salmonids usually progresses with no external signs of infection (Kahn *et al.* 1999), while sea lice may act as vectors (Tubbs *et al*. 2007).

Renibacterium salmoninarum remains viable for up to 20 weeks in water and sediments (Hirvela-Koski 2005) and can survive in the tissues of dead infected fish when used as fish food (Wiens 2011). It is resistant to freezing and extremely temperature stable. Growth is optimised at 15°C, but inhibited at temperatures above 22°C, and totally inhibited at 37°C (Densmore 1997). It survives for more than 15 minutes at 65°C and requires either two stage heat treatment (10 minutes

at 71°C, followed by 10 minutes at 82°C), or pasteurisation (heated to a core temperature of 100°C for 30 minutes), to ensure inactivation (Whipple & Rohovec 1994; Hirvela-Koski 2005).

Renibacterium salmoninarum is difficult to control due to the intracellular nature of infection, which limits the effect of potential chemotherapeutic control measures (Elliott *et al*. 1989) and antibiotic resistance is a developing issue (Densmore 1997; Wiens 2011). The single commercially available vaccine is of variable efficacy (Johnston & Knowles 2009; Wiens 2011; Elliott *et al.* 2014).

Potential host species in New Zealand include rainbow trout (*Oncorhynchus mykiss*) and Chinook salmon (*O. tshawytscha*), although all New Zealand salmonids (*Oncorhynchus* spp., *Salmo* spp., *Salvelinus* spp.) may be affected. Other potential hosts include Pacific herring (*C. harengus*) (Tubbs *et al*. 2007).

40.2. Risk assessment

40.2.1. Entry assessment

BKD is a long-term systemic disease and clinically infected fish commonly show no visible signs of infection (Wiens 2011). Infected fish may pass visual inspection and be present in the commodity. *Renibacterium salmoninarum* may be present in the muscle, gill and brain tissues of eviscerated fish (Densmore 1997; Wiens 2011). It is resistant to the environmental conditions likely to be encountered in fish storage and processing (Wiens 2011).

The likelihood of entry through the commodity is assessed as non-negligible.

40.2.2. Exposure assessment

For disease to establish, sufficient infected blood-water, fish scraps or offal would have to be discharged into the environment, at a level to become self-sustaining in the population (Kahn *et al.* 1999; Johnston 2008). *Renibacterium salmoninarum* is environmentally stable, surviving in dead fish tissues (Wiens 2011). Suitable host species are present in New Zealand (Tubbs *et al.* 2007).

Bacterial kidney disease primarily affects salmonids, while natural infection of non-salmonids appears rare and opportunistic, occurring only in proximity to infected salmonids under aquaculture conditions (Johnston & Knowles 1999; Wiens 2011). The role of non-salmonids as reservoirs of infection remains equivocal (Wiens 2011; Hershberger *et al.* 2013).

Renibacterium salmoninarum may be present in blood-water discharge, while the muscle trimmings, brain and gill tissues are likely to remain infective in the fish offal discarded from commercial fish processing. It remains viable in marine or fresh waters and in sediment for up to 20 weeks (Hirvela-Koski 2005) and infection commonly occurs through discarded infected fish wastes (Wiens 2011). The establishment of disease in wild salmonid stocks would provide a reservoir for infection and be difficult to control (Mitchum *et al.* 1979; Wiens 2011).

The likelihood of exposure to *R. salmoninarum* through the commodity is assessed as non-negligible.

40.2.3. Consequence assessment

Renibacterium salmoninarum is one of the most important salmonid diseases worldwide, so the loss of New Zealand's BKD-free status would have significant impacts on aquaculture exports (Anderson & Knowles 1999).

Renibacterium salmoninarum particularly affects rainbow trout (*O. mykiss*) and Chinook salmon (*O. tshawytscha*) (Wiens 2011). Its establishment would result in significant economic losses for the Chinook salmon farming industry through loss of sales due to infected product. This was valued at $63 million in export earnings in 2011 (Seafood New Zealand 2014). An outbreak could also affect recreational and tourist trout and salmon fishing, as well as incur significant social and environmental costs associated with other domestic fisheries (Fish & Game 2014). While the dollar value of New Zealand's freshwater fisheries is not fully known, the Lake Taupo trout fishery alone is valued at $70 to 80 million per annum (Marsh & Mkwra 2013).

As the water supplies in New Zealand aquaculture are commonly untreated, these facilities are at risk from aquatic pathogens (Sim-Smith *et al.* 2014). *Renibacterium salmoninarum* is one of the most difficult fish diseases to control (Wiens 2011). As a systemic, intracellular pathogen, it responds poorly to chemotherapeutics and antibiotic resistance is increasing (Elliott *et al*. 1989; Densmore 1997; Wiens 2011). The single licenced vaccine is of variable efficacy (Johnston & Knowles 1999; Wiens 2011; Elliott *et al.* 2014).

The consequence of establishment in salmonid fish is assessed as non-negligible.

40.2.4. Risk estimation

Since the entry, exposure, and consequence assessments are non-negligible, the risk estimation is non-negligible. Therefore, *R. salmoninarum* is assessed to be a risk in the commodity and risk management measures may be justified.

40.3. Risk management

Renibacterium salmoninarum has been assessed to be a risk in the commodity. Infection is a non-notifiable disease, so the OIE *Aquatic Code* (OIE 2016) does not provide specific guidance in importing eviscerated fish from infected countries, or specific processing requirements that would ensure the destruction of the pathogen. It is assumed that compliance with the approved processed states for OIE-listed diseases (Appendix 3) would also eliminate *R. salmoninarum* from the commodity and be a viable risk management option.

While primarily affecting salmonids, *R. salmoninarum* is reported from nine host families of fish in marine and fresh waters (Table 37), which may be present in the commodity. Species declaration should substantially reduce the pathogen load of *R. salmoninarum* in the commodity and be a viable risk management option. Clinical infection is mainly associated with aquaculture. A restriction of the commodity to wild-caught fish (not from aquaculture) should moderately reduce the pathogen load of *R. salmoninarum* and be a viable risk management option.

Renibacterium salmoninarum is widely reported from Europe, Iceland, North and South America and Japan. Where country/zone freedom is approved through the MPI Country Approval Procedures, this option is likely to substantially reduce the pathogen load of *R. salmoninarum* in

the commodity. Approval of a declaration of country/zone freedom by MPI is a viable risk management option.

As infection is associated with the brain, gills and blood, removal of the gills should slightly reduce the pathogen load of *R. salmoninarum.* Removal of the head and gills together should moderately reduce pathogen load in the commodity, although *R. salmoninarum* may be present in the musculature. Removal of the head and gills is a viable risk management option.

Renibacterium salmoninarum is unaffected by freezing, so frozen storage is not a viable management option. Denaturation requires high temperatures (by cooking to at least 100°C for 30 minutes), which should eliminate *R. salmoninarum* from the commodity. Temperature treatment is a viable risk management option.

40.3.1. Risk management options

Renibacterium salmoninarum is reported from fish in families Anoplopomatidae, Clupeidae, Cyprinidae, Embiotocidae, Hexagrammidae, Merlucciidae, Platycephalidae, Plecoglossidae and Salmonidae (Table 37), which are considered likely to be present in the commodity. Other families have not been associated with *R. salmoninarum.* Therefore species declaration indicating the commodity is not originated from any of the above families should substantially reduce the occurrence of *R. salmoninarum* in the commodity.

For the commodities originated from families associated with *R. salmoninarum*, one or a combination of the following additional options could also be considered to effectively manage the risk.

Where country/zone freedom from *R. salmoninarum* is accepted by MPI:

Option 1

Acceptance of country/zone freedom by MPI should substantially reduce the occurrence of *R. salmoninarum*, so the commodity may be imported without any further restrictions.

Where country/zone freedom from *R. salmoninarum* is not accepted by MPI or not available:

Option 2

Processing to a state consistent with the approved states for OIE-listed diseases (Appendix 3) should eliminate *R. salmoninarum.* When these provisions are met, the commodity could be imported without any further restrictions.

Where the imported commodity is not in a state consistent with the approved states for OIE-listed diseases (Appendix 3):

Option 3

Heat treatment (by cooking (to at least 100°C for 30 minutes), should eliminate *R. salmoninarum*. When this provision is met, the commodity could be imported without any further restrictions; or,

Option 4

Further processing (by removal of the head and gills) should moderately reduce the occurrence of *P. salmoninarum*. When this provision is met, the commodity could be imported without any further restrictions.

Option 5

Restriction of the commodity to wild-caught fish (not from aquaculture) should moderately reduce the occurrence of *P. salmoninarum*. When this provision is met, the commodity could be imported without any further restrictions.

40.4. References

Anderson CD, Knowles G (1999). Surveillance of New Zealand salmonids for *Renibacterium salmoninarum. Surveillance* 26(4): 8-9.

Anonymous (2016). Biosecurity (Notifiable Organisms) Order 2016 NZLR 2016/73. New Zealand Government, Wellington, 13 p.

Anonymous (2017). Bacterial kidney disease. Aquatic Animal Diseases Significant to Australia: Identification Field Guide 4th Edition. [Online] Available from www.agriculture.gov.au/SiteCollectionDocuments//bacterial-kidney-disease.doc [Accesses 23 August 2017].

Bell GR, Hoffmann RW, Brown LL (1990). Pathology of experimental infections of the sablefish, *Anoplopoma fimbria* (Pallis), with *Renibacterium salmoninarum*, the agent of bacterial gill disease in salmonids. *Journal of Fish Diseases* 13: 355–367.

Brynildsrud O, Feil EJ, Bohlin J, Castillo-Ramirez S, Colquhoun D, McCarthy U, Matejusova IM, Rhodes LM, Wiens GD, Verner-Jeffreys DW (2013). Microevolution of *Renibacterium salmoninarum*: evidence for intercontinental dissemination associated with fish movements. *The ISME Journal* 8: 746–756.

Densmore CL (1997). Bacterial kidney disease and its effect on the salmonid immune response. Ph.D dissertation, Virginia Polytechnic Institute, Blacksburg, Virginia, United States of America, 149 p.

Diggles BK (2011). Environmental Assessment Report –Disease Risks. Report prepared for New Zealand King Salmon. DigsFish Services, Queensland, Australia. [Online] Available from: http://www.epa.govt.nz/Publications/Appendix%2011%20Disease%20Risk%20Report.pdf [Accessed 23 August 2017].

Elliott DG, Pascho RJ, Bullock GL (1989). Developments in the control of bacterial kidney disease of salmonid fishes. *Diseases of Aquatic Organisms* 6: 201–215.

Elliott DG, Wiens GD, Hamell KL, Rhodes LD (2014). Vaccination against bacterial kidney disease. pp 255–274. *In:* Gudding R, Lillehaug A, Evensen O (*Eds.*), Fish Vaccination. John Wiley, Chichester, United Kingdom, 404 p.

Fish and Game (2014). New Zealand Fish and Game Council [Online] Available from: http://www.fishandgame.org.nz/ [Accessed 24 September 2014].

Hershberger P, Rhodes L, Kurath G, Winton J (2013). Infectious diseases of fishes in the Salish Sea. *Fisheries* 38(9): 402.

Hirvela-Koski V (2005). Fish pathogens *Aeromonas salmonicida* and *Renibacterium salmoninarum*: diagnostic and epidemiological aspects. B. VSci. Dissertation, University of Helsinki, Finland, 92 p.

Johnston C (2008). Import Risk Analysis: Frozen, skinless and boneless fillet meat of *Pangasius* spp. fish from Vietnam for human consumption. Ministry of Agriculture and Forestry, Wellington, New Zealand, 60 p.

Johnston C, Knowles G (1999). Surveillance of New Zealand salmonids for *Renibacterium salmoninarum. Surveillance* 26(4): 8–9.

Jónsdottir H, Malmquist HJ, Snorrason SS, Gudbergsson G, Gudmundsdóttir S (1998). Epidemiology of *Renibacterium salmoninarum* in wild Arctic charr and brown trout in Iceland. *Journal of Fish Biology* 53(2): 322–339.

Kahn SA, Beers PT, Findlay VL, Peebles IR, Durham PJ, Wilson DW, Gerrity SE (1999). Import risk analysis on non-viable salmonids and non-salmonid marine finfish. Australian Quarantine and Inspection Service, Department of Agriculture, Fisheries and Forestry, Canberra, Australia, 409 p.

Kent ML, Traxler GS, Kieser D, Dawe RJ, Shaw RW, Prosperi-Porta G, Ketchenson J, Evelyn TPT (1998). Survey of salmonid pathogens in ocean-caught fishes in British Columbia, Canada. *Journal of Aquatic Animal Health* 10: 211–219.

Mackie T, Arkwright J, Pryce-Tannatt T, Mottram J, Johnston W, Menzies W (1933). Furunculosis committee, Second Interim Report. HMSO, Edinburgh, United Kingdom, 81 p.

Marsh D, Mkwra L (2013). Review of freshwater non-market value studies. Department of Economics, University of Waikato, Hamilton, New Zealand, 144 p.

Mitchum DL, Sherman LE, Baxter GT (1979). Bacterial kidney disease in feral populations of brook trout (*Salvelinus fontinalis*) and rainbow trout (*Salmo gairdneri*). *Journal of the Fisheries Research Board of Canada* 36: 1370–1376.

OIE (2016). Aquatic Animal Health Code, 19th Edition. World Organization for Animal Health. Paris, France [Online] Available from: http://www.oie.int/international-standard-setting/aquatic-code/access-online/ [Accessed 15 March 2017].

Ozturk RC, Altinok I (2014). Bacterial and viral fish diseases on Turkey. *Turkish Journal of Fisheries and Aquatic Sciences* 14: 275–279.

Sanders JE, Fryer JL (1980). *Renibacterium salmoninarum* gen. nov., sp. nov., the causative agent of bacterial gill disease in salmonid fishes. *International Journal of Systemic Bacteriology* 30(2): 496–502.

Seafood New Zealand (2014). Seafood New Zealand: Key facts for our Industry. Seafood New Zealand, Wellington, New Zealand, 6 p.

Sim-Smith C, Faire S, Lees A (2014). Managing biosecurity risk for business benefit. Aquaculture biosecurity practices research report prepared for Ministry for Primary Industries, by Coast and Catchment Ltd, New Zealand Wellington, New Zealand, 214 p.

Tubbs L, Lee P, Diggles B, Jones JB, Shepherd M, Sim-Smith C (2007). A review of aquatic diseases of significance to New Zealand. Final research report for project ZBS 2005-17. National Institute of Water and Atmospheric Research, Wellington, New Zealand, 452 p.

Whipple MJ, Rohovec JS (1994). The effect of heat and low pH on selected viral and bacterial fish pathogens. *Aquaculture* 123(3-4): 179–189.

Wiens GD (2011). Bacterial kidney disease (*Renibacterium salmoninarum*). pp 338–374. *In:* Woo PTK, Bruno DW (*Eds*.). *Fish Diseases and Disorders: Volume 3 - Viral, Bacterial and Fungal Infections*, 2nd edition. CABI Publishing, Wallingford, United Kingdom, 930 p.

41. *Streptococcus* (exotic group B species including *S. agalactiae* (serotype 3: 283) and *S. iniae*)

41.1. Hazard identification

41.1.1. Aetiological agent

Streptococcus agalactiae and *S. iniae* are non-motile spherical or ovoid shaped *β*-haemolytic Gram-positive facultative anaerobic bacteria. They are members of a pan-genomic species complex (where at least 80% of genomic material is shared), that is classified within group B of the Family Streptococcaceae, Order Lactobacillales (Johnston 2008; Kim *et al.* 2014). Pathogenicity and host specificity vary widely between the hundreds of identified genomic strains, which include free-living and commensal organisms (Rajendram *et al.* 2016).

Streptococcus agalactiae was originally described (as *S. difficule*) from cultured seabream (*Sparus auratus*) and wild mullet (*Liza klunzingeri*) in Kuwait (Evans *et al*. 2002). Within the *S. agalactiae* group, five pathogenic serotypes (1a, Ib, II, III and V) are recognised as causative agents of streptococcidiosis in fish (Tettelin *et al.* 2005; Salati 2011). Several strains are zoonotic, including the serotype III: 283 strain reported from fish (Channidae, Cyprinidae) in Singapore (Mehershahi *et al*.2015).

Streptococcus iniae was previously considered as part of the *S. agalactiae* grouping, as routine tests have only recently been able to separate these groups (Evans *et al.* 2006; Salati 2011). *Streptococcus iniae* is a causative agent of streptococcosis, or red boil disease, affecting marine and freshwater fish (Salati 2011). Up to five serotypes of *S. iniae* are now recognised, that vary in pathogenicity and host specificity (Zhou *et al.* 2008; Kim *et al.* 2014).

41.1.2. OIE status

Streptococcidiosis (infection with *Streptococcus agalactiae* or *S. iniae*) is not an OIE listed disease (OIE 2016).

41.1.3. New Zealand status

Streptococcus agalactiae is endemic, causing streptococcidiosis in agriculture (mastitis) and low levels of infection (streptococcidiosis) in fish (Hine & Diggles 2005; Johnston 2008). The zoonotic strain of *S. agalactiae* (serotype III: 283) (Mehershahi *et al.* 2015), has not been reported from New Zealand. It is assumed to be exotic.

Streptococcus iniae has never been reported from surveys in New Zealand (Hine & Diggles 2005; Johnston 2008) and is considered to be exotic. Infection with *Streptococcus* spp. is not a notifiable disease in New Zealand (Anon. 2016).

41.1.4. Epidemiology

Streptococcus agalactiae is a common and widely distributed commensal organism that may become pathogenic in fish (Rajendran *et al.* 2016). It is an agent of streptococcosis, together with other members of the *Streptococcus* species complex (*S. dysgalactiae, S. faecium, S. iniae* and *S. faecialis* (Zhou *et al.* 2011). The exotic, zoonotic strain of *Streptococcus agalactiae* (serotype III:

283) is reported from farmed snakehead, *Channa* spp. (Channidae), and Asian bighead carp, *Hypophthalmichthys molitrix* (Cyprinidae), in Singapore (Rajendram *et al*. 2016; Kalimudden *et al.* 2017; Barkham 2018).

Streptococcus iniae is a major pathogen in marine and freshwater aquaculture, in Australia, Bahrain, Korea, Iran, Israel, Italy, Japan, South Africa, Spain and North America (Buchanan *et al*. 2008; Zhou *et al.* 2011). It is reported from a wide range of farmed marine and freshwater fish (Table 38) (Pier & Madin 1976; Kusuda *et al.* 1978; Zlotkin *et al*. 1998; Yuasa *et al*. 1999; Colorn *et al.* 2002; Darwish 2007; Park *et al.* 2009; El-Aamri *et al.* 2010; Suanyuk *et al.* 2010; Salati 2011). While epizootics occurred in Japanese eel (*Anguilla japonica*) aquaculture during the 1970s *S. iniae* is not currently a significant pathogen of farmed Japanese eels (Kusuda *et al.* 1978; C. Rodgers *pers.comm*. 2017).

Streptococcus iniae causes epizootics in wild fish, including gilthead sea bream (*Sparus aurata*), princess parrotfish (*Scarus taeniopterus*), redband parrotfish (*Sparisoma aurofrenatum*), red porgy (*Pagrus pagrus*), red snapper (*Lutjanus campechanus*), red hind (*Epinephelus guttatus*), stingray (*Dasyatis* spp.) and yellowtail snapper (*Ocyurus chrysurus*) (Pier & Madin 1976; Eldar & Ghittino 1999; Evans *et al.* 2004; El-Aamri *et al.* 2014; Keirstead *et al.* 2014). *Streptococcus iniae* is also reported from the Amazonian freshwater dolphin (*Inia geoffrensis*) (Salati 2011).

Table 38. Families and Species of Fish Susceptible to *Streptococcus Agalactiae* (Exotic Serotype III: 283) and *Streptococcus Iniae*

Host Family	Susceptible Fish Species
Anguillidae	Japanese eel (*Anguilla japonica*)
Ariidae	Hardhead sea catfish (*Ariopsis felis*)
Carangidae	Yellowtail kingfish (*Seriola quinqueradiatus*)
Channidae	Snakehead (*Channa* spp.)
Cichlidae	Tilapia (*Tilapia* spp., *Coptodon* spp.), Nile tilapia (*Oreochromis niloticus*)
Clupeidae	Sardine (*Sardinops sagax*), Atlantic menhaden (*Brevoortia patronus*)
Cyprinidae	Asian bighead carp (*Hypophthalmichthys molitrix*), Golden shiner (*Notemigonus crysoleucas*)
Haemulidae	Striped piggy (*Pomadasys stridens*)
Ictaluridae	Channel catfish (*Ameiurus punctatus*)
Latidae	Barramundi (*Lates calcarifer*)
Lutjanidae	Caribbean red snapper (*Lutjanus campechanus*), yellowtail snapper (*Ocyurus chrysurus*)
Moronidae	European sea bass (*Dicentrarchus labrax*), striped bass (*Morone saxatilis*), sunshine bass (*M. saxitalis* x *M. chrysops*)
Mugilidae	Mullet (*Mugil cephalus*), Klunzinger's mullet (*Liza klunzingeri*)
Paralichthyidae	Bastard halibut (*Paralichthys olivaceus*)
Salmonidae	Rainbow trout (*Oncorhynchus mykiss*)
Scaridae	Redband parrotfish (*Sparisoma aurofrenatum*)
Sciaenidae	Atlantic croaker (*Micropogon undulatus*), red drum (*Sciaenops ocellatus*), silver seatrout (*Cynoscion nothus*), spot croaker (*Leiostomus xanthurus*)
Serranidae	Red hind (*Epinephelus guttatus*)
Siganidae	White-spotted spinefoot (*Siganus canaliculatus*)
Sparidae	Red porgy (*Pagrus pagrus*), gilthead sea bream (*Sparus aurata*), pinfish (*Lagodon rhomboides*)
Synodontidae	Lizardfish (*Synodus variegatus*)

Disease transmission is horizontal and direct. Infection commonly occurs through the oral pathway, following the consumption of infected fish or faecal material (Diggles 2011), but *S. iniae* may also be transmitted by vectors such as *Gyrodactylus niloticus* (Shoemaker 2008).

Initial entry occurs through the epithelial cells by transcytosis and the disease is internalised through invasion of macrophage cells (Evans *et al*. 2000; Baiano & Barnes 2009).

Clinical infection without external signs of disease is reported from channel catfish (*Ictalurus punctatus*) in Chinese aquaculture, where infected fish may act as carriers for infection (Baiano & Barnes 2009). Wild stocks that occur in close proximity to aquaculture may act as reservoirs of disease (Zlotkin *et al.* 1998). Multiple genotypes may be present among the different host species, both between and within fish farms (Baiano & Barnes 2009).

External clinical signs of infection include bilateral exophthalmia, anorexia and dark skin coloration caused by haemorrhage of the skin and inflammation of the eyes. Petechiae are commonly present on the inner opercula, caudal fin and mouth areas (Perera *et al.* 1998; Johnston 2008; Chen *et al*. 2011, Salati 2011).

Internal signs include cellular infiltration and inflammation of the eye and kidney tissues, granulomatous meningitis, epicarditis and myocarditis of the liver and spleen (Johnston 2008; Salati 2011). *Streptococcus iniae* infection commonly occurs as concurrent infection with *Lactococcus garvieae* (Salati 2011) and correct identification requires specific PCR based assay techniques (Fadaeifard *et al.* 2012; Suebsing *et al.* 2013; Park *et al.*, 2013).

The disease progression of *S. iniae* is not fully understood (Wang *et al.* 2015) and pathogenicity appears related to the ability to survive within host phagocytes (Buchanan *et al.* 2008). Chen *et al.* 2011). The minimum infectious dose varies from 1 x 10^3 cfu mL^{-1} (colony forming units) for barramundi (*L. calcarifer*) (Bromage & Owens 2002), to 2 x 10^7 cfu mL^{-1} for tilapia (*O. niloticus*) (Evans *et al.* 2000; Shoemaker *et al.* 2000).

Infection is initially focused on the viscera, causing hepatitis and splenitis in the liver and spleen, together with haemorrhagic lesions and necrosis of the kidneys and intestine (Chen *et al.* 2011; Salati 2011). The disease then progresses to the gills, brain and nervous system, causing mengioencephalitis of the brain and nervous tissues, together with the detachment of the respiratory epithelial tissues of the gills, resulting in anaemia (Buchanan *et al*. 2008). Bacterial concentration in the somatic musculature is comparatively low (Chen *et al.* 2004; Johnston 2008).

Mortality may exceed 75% in epizootics of tilapia (*Oreochromis* spp.) aquaculture, but pathogenicity varies among strains of *S. iniae*. Pathogenicity in farmed fish is also influenced by external factors including water quality, stocking density and the widespread use of infected trash fish as feed (Shoemaker *et al.* 2000; Johnston 2008; Salati 2011).

Streptococcus agalactiae (serotype III: 283) and S. *iniae* are also emerging zoonotic pathogens (Weinstein *et al.* 1997; Sun *et al.* 2007; Baiano & Barnes 2009; Haenen *et al.* 2013; Mehershahi *et al.* 2015). Infection in humans generally results from consumption of improperly cooked carrier fish (Salati 2011), or following soft tissue injuries (Baiano & Barnes 2009). While infection of the skin on the hands is generally mild, the infection may become internalised by macrophage invasion (Salati 2011). Systemic infection results in endocarditis, meningitis, arthritis, sepsis, pneumonia, osteomyelitis and toxic shock. Systemic infection is usually fatal if left untreated, while the mortality rate of 25% is higher, particularly in immunocompromised individuals (Baiano & Barnes 2009; Ambrosioni *et al.* 2017).

Streptococcus spp. are resistant to freezing. *Streptococcus iniae* remains viable in frozen fish for up to 6 months (Hine & Diggles 2005), while *S. agalactiae* remains viable in frozen mullet (*M. cephalus*) and tilapia (*Oreochromis* spp.) after freezing for 9 months (Evans *et al.* 2004). *Streptococcus iniae* can survive in fish processing wastes (Skall & Olesen 2011).

Streptococcus iniae is inactivated by alkaline disinfection (pH 12 for 20 minutes contact time), or by acidic disinfection (pH 4 for 24 hours contact time) (Dixon *et al.* 2003), but is resistant to chlorine-based treatments (Skall & Olesen 2011). *Streptococcus agalactiae* requires treatment with moist heat (at 121°C for at least 15 minutes), or dry heat (at 160 to 170°C for at least 1 hour) for inactivation (Anon. 2011).

Streptococcus iniae can be inactivated by antibiotics, including amoxicillin, enrofloxacin, erythromycin, furazolidone and oxytetracycline (Agnew & Barnes 2007). Control by vaccination has been unsuccessful, due to complex serotypic variation (Agnew & Barnes 2007), while effectiveness is reduced where fish are co-infected by parasites, including *Trichodina* spp., *Gyrodactylus* spp. and *Ichthyophthirius multifiliis* (Martins *et al.* 2011).

Potential marine and freshwater hosts occur in New Zealand, including eels (*Anguilla* spp.), yellowtail kingfish (*Seriola lalandi*) and snapper (*Pagrus auratus*) (Tubbs *et al*. 2007). Infection of wild fish populations in marine or fresh water may establish a reservoir for infection of farmed species (Diggles 2011).

41.2. Risk assessment

41.2.1. Entry assessment

Most fish clinically infected with *S. iniae* or *S. agalactiae* would fail visual inspection and not be present in the commodity, but carrier fish may show no external signs of infection (Chen *et al*. 2011). These fish may pass visual inspection and be present in the commodity.

Streptococcus iniae and *S. agalactiae* survive in dead fish muscle tissue (Skall & Olesen 2011; Barkham *et al.* 2018) as well as in the gills, brain and nervous tissue, the inner opercular walls and mouth of eviscerated fish (Johnston 2008; Chen *et al*. 2011). *Streptococcus agalactiae* and *S. iniae* are resistant to freezing and other processes likely to be encountered in commercial fish processing (Hine & Diggles 2005; Tubbs *et al*. 2007).

The likelihood of entry is assessed as non-negligible.

41.2.2. Exposure assessment

For disease to become established, sufficient infected effluent would have to be discharged from a processing facility to infect susceptible wild marine or freshwater fish, and for this infection to become self-sustaining in the wild population (Kahn *et al.* 1999). *Streptococcus agalactiae* and *S. iniae* may be present in the blood-water discharge during factory processing and may survive in the fish offal (skin, brain, gills, oral and buccal epidermis) discarded after commercial processing (Skall & Olesen 2011).

Streptococcus iniae is an exotic opportunistic pathogen of marine and freshwater-farmed fish (Buchanan *et al.* 2008) and infection in wild fish occurs in close proximity to aquaculture (Zlotkin *et al.* 1998; Eldar & Ghittino 1999). However, the importance of infection of wild stocks may be under-estimated. Epizootic outbreaks may go unnoticed, because infected wild fish have lower survival due to predation (Fadaeifard *et al.* 2012; Suebsing *et al.* 2013; Park *et al.* 2013; Keirstead *et al.* 2014).

Both *S. agalactiae* and *S. iniae* may cause zoonotic infection following the consumption of raw or under-cooked fish products, as well as through handling of infected fish (Chen *et al*. 2011; Skall & Olesen 2011; Barkham *et al.* 2018).

A wide range of potential marine and freshwater hosts, including eels (*Anguilla* spp.), yellowtail kingfish (*Seriola lalandi*) and snapper (*Pagrus auratus*) occur in New Zealand (Tubbs *et al*. 2007).

The likelihood of exposure to *S. iniae* is assessed as non-negligible.

41.2.3. Consequence assessment

Streptococcus iniae is associated with significant losses in the aquaculture of a wide range of marine and freshwater host fish (El-Aamri *et al.* 2014) and causes epizootic infection in wild fish stocks (Keirstead *et al.* 2014).

Potential commercially significant host species include New Zealand snapper (*Pagrus auratus*), which was valued at NZ$ 6.5 million in 2012 (Statistics New Zealand 2014), while there is a developing aquaculture industry for yellowtail kingfish (NIWA 2014). *Streptococcus iniae* is pathogenic to rainbow trout (Salati 2011). An outbreak could also affect recreational and tourist trout and salmon fishing, as well as incur significant social and environmental costs associated with other domestic fisheries (Fish & Game 2014). While the dollar value of New Zealand's freshwater fisheries is not fully known, the Lake Taupo trout fishery alone is valued at $70 to 80 million per annum (Marsh & Mkwra 2013).

Eel farming (*Anguilla australis*, *A. dieffenbachii*) is not commercially practiced in New Zealand (NIWA 2015). *Streptococcus iniae* has previously caused epizootics in Japanese eel farming (Kusuda *et al*. 1978), but has not subsequently been reported. The establishment of *S. iniae* could have possible consequences for wild eel fish stocks (Tubbs *et al.* 2007). Eels are a national Taonga (MPI 2017) and support significant non-commercial fisheries. Any reductions in stock levels would have social consequences for recreational and traditional Maori eel fisheries.

The introduction of *S. iniae* may lead to direct losses in aquaculture production and indirect losses through potential impacts on trade. Historically, the New Zealand aquaculture industry has been largely free of any serious pests or diseases. However, it is realised that biosecurity practices in New Zealand can be strengthened by improvements such as in treatment of water supplies (Sim-Smith *et al.* 2014). The establishment of *S. iniae* in wild populations could create a reservoir for infection of important commercial wild marine and freshwater fisheries and aquaculture species, which once established, would be difficult to control (Kahn *et al.* 1999).

Exotic *Streptococcus agalactiae* serotype III: 283 and *S. iniae* are emerging zoonotic pathogens. There is no evidence to indicate the exotic strains of *S. agalactiae* have a higher zoonotic potential than endemic strains that are already present in New Zealand. *Streptococcus iniae* is an exotic zoonotic pathogen with associated mortalities of 25% and higher, particularly for immunocompromised individuals (Baiano & Barnes 2009; Salati 2011; Kalimudden et al. 2017; Keirstead et al. 2014; Barkham et al. 2018).

The consequence of establishment of *S. iniae* and *S. agalactiae* serotype III: 283 are assessed as non-negligible.

41.2.4. Risk estimation

Since the entry, exposure, and consequence assessments are non-negligible, the risk estimation is non-negligible. Therefore, under the procedures followed in this risk assessment, *S. iniae* and *S. agalactiae* serotype III: 283 are assessed to be risks in the commodity and risk management measures may be developed.

41.3. Risk management

Streptococcus agalactiae serovar III: 283 and *S. iniae* have been assessed to be a risk in the commodity. Infection with *Streptococcus* spp. is a non-notifiable disease, so there is no specific guidance in the OIE *Aquatic Code* (OIE 2016) for importing eviscerated fish from infected countries, or specific processing requirements that would ensure the destruction of these pathogens. It is assumed that compliance with the approved processed states for OIE-listed diseases (Appendix 3) would also eliminate *S. agalactiae* serovar III: 283 and *S. iniae* from the commodity and be a viable risk management option.

Streptococcus agalactiae serovar III: 283 and *S. iniae* are reported from over 20 families of wild and farmed marine and fresh water fish (Table 38) which may be present in the commodity. Species declaration is likely to substantially reduce the pathogen load of these exotic *Streptococcus* spp. in the commodity and be a viable risk management option. Restriction of the commodity to wild-caught fish (not from aquaculture) would have little, or no effect on pathogen load and is not a viable risk management option.

Streptococcus agalactiae serovar III: 283 is reported from freshwater fish farming in Singapore, while *S. iniae* is widely distributed in the marine and fresh waters of Australia, Bahrain, Korea, Iran, Israel, Italy, Japan, South Africa, Spain and North America.

Where country/zone freedom is approved through the MPI Country Approval Procedures, this option is likely to substantially reduce the pathogen load of *Streptococcus agalactiae* serovar III: 283 and *S. iniae* in the commodity. Approval of a declaration of country/zone freedom by MPI is a viable risk management option.

Streptococcus agalactiae serovar III: 283 and *S. iniae* may be concentrated in the skin, gills, brain and the epidermal tissues of the mouth and buccal cavity of infected eviscerated fish. Removal of the gills, or the head and gills should slightly reduce their pathogen load in the commodity. These bacteria are present in comparatively lower densities in the somatic musculature (Chen *et al.* 2004; Johnston 2008), so further processing to the skin-off fillet state should moderately reduce pathogen load of these exotic *Streptococcus* spp. in the commodity and be a viable risk management option.

Streptococcus agalactiae and *S. iniae* are unaffected by frozen storage, so this is not a viable risk management option.

Streptococcus agalactiae serovar III:283 and *S. iniae* are zoonotic pathogens that are denatured by heat (by cooking to 121°C, using moist heat, for at least 15 minutes), so heat treatment should eliminate them from the commodity. Heat treatment is a viable risk management option.

41.3.1. Risk management options

Streptococcus agalactiae serovar III:283 and *S. iniae* are reported from fish in families Anguillidae, Ariidae, Carangidae, Channidae, Cichlidae, Clupeidae, Cyprinidae, Haemulidae,

Ictaluridae, Latidae, Lutjanidae, Moronidae, Mugilidae, Paralichthyidae, Salmonidae, Scaridae, Sciaenidae, Serranidae, Siganidae, Sparidae and Synodontidae (Table 38). These families are considered likely to be present in the commodity. Other families have not been associated with the exotic serovar III:283 strain of *S. agalactiae*, or with *S. iniae*. Therefore species declaration indicating the commodity is not originated from any of the above families should substantially reduce the occurrence of *S. agalactiae* serovar III:283 and of *S. iniae* in the commodity.

For commodities originating from families associated with *S. agalactiae* serovar III:283 and with *S. iniae*, one or a combination of the following options could also be considered to effectively manage the risk:

Where country/zone freedom from *S. agalactiae* serovar III:283 and from *S. iniae* is accepted by MPI:

Option 1

Acceptance of country/zone freedom by MPI should substantially reduce the occurrence of *S. agalactiae* serovar III:283 and *S. iniae*, so the commodity may be imported without any further restrictions.

Where country/zone freedom from *S. agalactiae* serovar III:283 and from *S. iniae* is not accepted by MPI or not available:

Option 2

Processing to a state consistent with the approved states for OIE-listed diseases (Appendix 3) should eliminate *S. galactiae* III:283 and *S. iniae*. When these provisions are met, the commodity could be imported without any further restrictions.

Where the imported commodity is not in a state consistent with the approved states for OIE-listed diseases (Appendix 3):

Option 3

Heat treatment (by cooking, with moist heat, to 121°C for at least 15 minutes) should eliminate *S. galactiae* III:283 and *S. iniae.* When this provision is met, the commodity could be imported without any further restrictions.

Option 4

Further processing (to the skin-off fillet state) should moderately reduce the occurrence of *S. galactiae* III:283 and *S. iniae*. When this provision is met, the commodity could be imported without any further restrictions.

41.4. References

Agnew W, Barnes AC (2007). *Streptococcus iniae*: an aquatic pathogen of global veterinary significance and a challenging candidate for reliable vaccination. *Veterinary Microbiology* 122(1–2): 1–15.

Ambrosioni J, Hernandez-Meneses M, Téllez A, Pericàs J, Falces C, Tolosana JM, Vidal B, Almela M, Quintana E, Llopis J, Moreno A, Miro JM (2017). The Changing Epidemiology of Infective Endocarditis in the Twenty-First Century. *Current infectious disease reports* 19 (5): 21 [Online] Available from http://www.doi:10.1007/s11908-017-0574-9. PMID 28401448 [Accessed 6 August 2018].

Anonymous (2011). *Streptococcus iniae*. Pathogen safety data sheets. Public Health Agency of Canada [Online] Available from: http://www.phac-aspc.gc.ca/lab-bio/res/psds-ftss/streptococcus-agalactiae-eng.php [Accessed 23 December 2014].

Anonymous (2016). Biosecurity (Notifiable Organisms) Order 2016 SR 2016/73. New Zealand Government, Wellington, 13 p.

Baiano JC, Barnes AC (2009). Towards control *of Streptococcus iniae. Emerging and Infectious Diseases* 15(12): 1891–1896.

Barkham T, Sheppard A, Jones N, Chen SL (2018). *Streptococcus agalactiae* that caused meningitis in healthy adults in 1998 are ST283, the same type that caused a food-borne outbreak of invasive sepsis in 2015: an observational molecular epidemiology study. *Clinical Microbiology and Infection* 24(8): 923–925.

Bromage ES, Owens L (2002). Infection of barramundi *Lates calcarifer* with *Streptococcus iniae*: effects of different routes of exposure. *Diseases of Aquatic Organisms:* 52(3):199–205.

Buchanan JT, Colvin KM, Viknar MR, Patel SK, Timmer AM, Nizet VT (2008). Strain-associated virulence factors of *Streptococcus iniae* in hybrid striped bass. *Veterinary Microbiology* 131: 145–153.

Chen CY, Wooster GA, Bowser PR (2004). Comparative blood chemistry and histopathology of tilapia infected with *Vibrio vulnificus* or *Streptococcus iniae* or exposed to carbon tetrachloride, gentamicin, or copper sulphate. *Aquaculture* 239 421–443.

Chen D, Wang K, Geng Y, Wang J, Huang X, He M (2011). Pathological changes in cultured channel catfish *Ameiurus punctatus* spontaneously infected with *Streptococcus iniae*. *Diseases of Aquatic Organisms* 95: 203–208.

Colorn A, Diamant A, Eldar A, Kvitt H, Zlotkin A (2002). *Streptococcus iniae* infections in Red Sea cage-cultured and wild fishes. *Diseases of Aquatic Organisms* 49(3): 165–170.

Darwish AM (2007). Laboratory Efficacy of Florfenicol against *Streptococcus iniae* Infection in Sunshine Bass. *Journal of Aquatic Animal Health* 19(1): 1–7.

Diggles B (2011). Risk analysis – Aquatic animal diseases associated with domestic bait translocation. Final Report for FRDC Project 2009/072. Fisheries Research and Development Corporation, Canberra, Australia.

Dixon PF, Smail DA, Algoet M, Hastings T, Bayley A, Byrne H, Dodge M, Garden A, Joiner C, Roberts E, Thompson F (2003). Inactivation of fish pathogenic viruses and bacteria for waste disposal purposes: laboratory and field studies. 12th International Conference of the European Association of Fish Pathologists [Abstract O-030], p 35.

El-Aamri F, Caballero MJ, Real F, Acosta F, Deniz S, Roman J, Padilla D (2014). *Streptococcus iniae* in gilthead sea bream (*Sparus aurata* L.) and red porgy (*Pagrus pagrus*, L.): ultrastructural analysis. *Veterinary Pathology* 52(1): 209–212.

El-Aamri F, Padilla D, Acosta F, Caballero MJ, Roo J, Bravo J, Vivas J, Real F (2010). First report of *Streptococcus iniae* in red porgy (*Pagrus pagrus* L.). *Journal of Fish Diseases* 33(11): 901–905.

Eldar A, Ghittino C (1999). *Lactococcus garvieae* and *Streptococcus iniae* infections in rainbow trout (*Oncorhynchus mykiss*): similar, but different diseases. *Diseases of Aquatic Organisms* 36(1): 227–231.

Evans JJ, Klesius PH, Shoemaker CA (2006). *Streptococcus* in warm water fish. *Aquaculture Health International* 7: 10–14.

Evans JJ, Wiedenmeyer AA, Klesius P, Shoemaker C (2004). Survival of *Streptococcus agalactiae* from frozen fish following natural and experimental infections. *Aquaculture* 233(1–4): 15–21.

Evans JJ, Klesius PH, Gilbert PM, Shoemaker CA, Al Sarawi MA, Landsberg J, Duremdez R, Al Marzouk A, Al Zenki S (2002). *Characterization of β-haemolytic Group B Streptococcus agalactiae* in cultured seabream, Sparus auratus L., and wild mullet, *Liza klunzingeri* (day), in Kuwait. *Journal of Fish Diseases* 25: 505–513 [Online] Available from: http://www. doi:10.1046/j.1365-2761.2002.00392 [Accessed 24 July 2018].

Evans JJ, Shoemaker CA, Klesius PH (2000). Experimental *Streptococcus iniae* infection of striped bass (*Morone chrysops x Morone saxatilis*) and tilapia (*Oreochromis niloticus*) by nares inoculation. *Aquaculture* 189 (3-4): 197–210.

Fadaeifard F, Momtaz H, Rahimi E, Mirzakhani A (2012). Detection of *Streptococcus iniae* and *Lactococcus garvieae* by multiplex polymerase chain reaction (PCR) in some rainbow trout farms of Iran. *African Journal of Biotechnology* 11(2): 260–263.

Fish and Game (2014). New Zealand Fish and Game Council [Online] Available from: http://www.fishandgame.org.nz/ [Accessed 24 September 2014].

Haenen OLM, Evans JJ, Berthe F (2013). Bacterial infections from aquatic species: potential for and prevention of contact zoonoses. *Revue Scientifique et Technique (International Office of Epizootics*) 32(2): 497–507.

Hine MP, Diggles BK (2005). Import Risk Analysis: Ornamental fish. Biosecurity New Zealand. Ministry of Agriculture and Forestry, Wellington, New Zealand, 264 p.

Johnston C (2008). Import risk analysis: Frozen, skinless and boneless fillet meat of *Oreochromis* spp. from China and Brazil for human consumption. MAF Biosecurity New Zealand, Ministry of Agriculture and Forestry, Wellington, New Zealand. 96 p.

Kahn SA, Beers PT, Findlay VL, Peebles IR, Durham PJ, Wilson DW, Gerrity SE (1999). Import risk analysis on non-viable salmonids and non-salmonid marine finfish. Australian Quarantine and Inspection Service, Department of Agriculture, Fisheries and Forestry, Canberra, Australia, 409 p.

Kalimuddin S, Chen SL, Lim CT, Koh TH, Tan TY, Kam M, Wong CW, Mehershahi KS, Chau ML, Ng LC, Tang WY, Badaruddin H, Teo J, Apisarnthanarak A, Suwantarat N, Ip M, Holden MTG, Hsu LY, Barkham T (2017). 2015 epidemic of severe *streptococcus agalactiae* sequence type 283 infections in Singapore associated with the consumption of raw freshwater fish: A detailed analysis of clinical, epidemiological, and bacterial sequencing data. *Clinical and Infectious Diseases* 15 (64, suppl2): 145–152 [Online] Available from http://www.doi: 10.1093/cid/cix021. [Accessed 24 July 2017].

Keirstead ND, Brake JW, Griffin MJ, Halliday-Simmonds I, Thrall MA, Soto E (2014). Fatal septicaemia caused by the zoonotic bacterium *Streptococcus iniae* during an outbreak in Caribbean reef fish. *Veterinary Pathology* 51(5): 1035–1041.

Kim MS, Jin JW, Han HJ, Choi HS, Hong S, Cho JY (2014). Genotype and virulence of *Streptococcus iniae* isolated from diseased olive flounder *Paralichthys olivaceus* in Korea. *Fisheries Science* 80(6): 1277–1284.

Kusuda R, Komatsu I, Kawai K (1978). *Streptococcus* sp isolated from an epizootic of cultured eels. *Bulletin of the Japanese Society of Scientific Fisheries* 44: 295.

Marsh D, Mkwra L (2013). Review of freshwater non-market value studies. Department of Economics, University of Waikato, Hamilton, New Zealand, 144 p.

Martins ML, Shoemaker CA, Xu D, Klesius PH (2011). Effect of parasitism on vaccine efficiency against *Streptococcus iniae* in Nile tilapia. *Aquaculture* 314(1–4): 18–23.

Mehershahi KS, Hsu LY, Koh TH, Chen SL (2015). Complete genome sequence of *Streptococcus agalactiae* serotype III, multilocus sequence type 283 strain SG-M1. Singapore *Streptococcus agalactiae* Working Group. *Genome Announcements* 3(5):e01188-15 [Online] Available from: http://www.doi:10.1128/genomeA.01188-15 [Accessed 24 July 2018].

Ministry for Primary industries (MPI) (2017). Tangata Whenua: Maori Customary Fisheries [Online] Available from http://fs.fish.govt.nz/Page.aspx?pk=128&tk=592 [Accessed 2 August 2017].

NIWA (2014). Yellowtail kingfish [Online]. Available from: https://www.niwa.co.nz/aquaculture/aquaculture-species/yellowtail-kingfish [Accessed 12 April 2015].

NIWA (2015). Tuna aquaculture -New Zealand [Online]. Available from: https://www.niwa.co.nz/te-kuwaha/tuna-information-resource/pressures-on-new-zealand-populations/tuna-in-aquaculture/new-zealand [Accessed 7 October 2015].

OIE (2016). Aquatic Animal Health Code, 19th Edition. World Organization for Animal Health. Paris, France [Online]. Available from: http://www.oie.int/international-standard-setting/aquatic-code/access-online/ [Accessed 15 March 2017].

Park SB, Kwon K, Cha IS, Jang HB, Nho SW, Fern, Fagutao F, Kim YK, Yu JE, Jung TS (2013). Development of a multiplex polymerase chain reaction for the detection of *Edwardsiella tarda*, *Streptococcus parauberis* and *Streptococcus iniae* in olive flounder (*Paralichthys olivaceus*). *Journal of Veterinary Science* 15(1): 163–166.

Park YK, Nho SW, Shin GW, Park SB, Jang H, Cha IH, Ha M, Kim Y, Dalvi RS, Kang BJ, Jung TS (2009). Antibiotic susceptibility and resistance of *Streptococcus iniae* and *Streptococcus parauberis* isolated from olive flounder (*Paralichthys olivaceus*). *Veterinary Microbiology* 136(1/2): 76–81.

Perera RP, Fiske RA, Johnson SK (1998). Histopathology of hybrid tilapias *Tilapia niloticus* x *T. aurea* infected with a biotype of *Streptococcus iniae. Journal of Aquatic Animal Health* 10: 294–299.

Pier GB, Madin SH (1976). *Streptococcus iniae* sp. nov., a beta-hemolytic streptococcus isolated from an Amazon freshwater dolphin, *Inia geoffrensis*. *International Journal of Systematic Bacteriology* 26 (4): 545–553.

Rajendram P, Kyaw WM, Leo YS, Ho H, Chen WK, Lin R, Pratim P, Badaruddin H, Ang B, Barkham T, Chow A (2016). Group B *Streptococcus* sequence type 283 disease linked to consumption of raw fish, Singapore. *Emerging Infectious Diseases* 22(11): 1974–1977 [Online] Available from: http://dx.doi.org/10.3201/eid2211.160252 [Accessed 24 July 2018].

Ringo E, Gatesoupe FJ (1998). Lactic acid bacteria in fish: a review. *Aquaculture* 160: 177–203.

Salati F (2011). *Enterococcus seriolicida* and *Streptococcus* spp. (*S. iniae*, *S. agactiae* and *S. dysgalactiae*). pp 375–396. *In*: Woo PTK, Bruno DW (*Eds*.). *Fish Diseases and Disorders: Volume 3 - Viral, Bacterial and Fungal Infections*, 2nd edition. CABI Publishers, Wallingford, United Kingdom, 930 p.

Shoemaker CA, Evans JJ, Klesius PH (2000). Density and dose: Factors affecting mortality of *Streptococcus iniae* infected tilapia (*Oreochromis niloticus*). *Aquaculture* 188: 229–235.

Shoemaker C (2008). Concurrent infections (parasitism), pp 1365-1375 *In:* From the pharaohs to the future. Eighth International Symposium on Tilapia in Aquaculture. Proceedings. Cairo, Egypt, 12-14 October 2008. AQUAFISH Collaborative Research Support Program, Corvallis, USA [Online] Available from: http://ag.arizona.edu/azaqua/ista/ISTA8/ProceedingsISTA8.htm [Accessed 7 October 2015].

Sim-Smith C, Faire S, Lees A (2014). Managing biosecurity risk for business benefit. Aquaculture biosecurity practices research report prepared for Ministry for Primary Industries, by Coast and Catchment Ltd, New Zealand Wellington, New Zealand, 214 p.

Skall HF, Olesen NJ (2011). Treatment of wastewater from fish slaughterhouses. Report to the National Veterinary Institute, Denmark, 111 p.

Statistics New Zealand (2014). New Zealand's seafood industry [Online] Available from: http://www.stats.govt.nz/tools_and_services/newsletters/price-index-news/apr-13-article-seafood.aspx [Accessed 6 April 2015].

Suanyuk N, Sukkasame N, Tanmark N, Yoshida T, Itami T, Thune RL, Tantikitti C, Supamattaya K (2010). *Streptococcus iniae* infection in cultured Asian sea bass (*Lates calcarifer*) and red tilapia (*Oreochromis* sp.) in southern Thailand. *Songklanakarin Journal of Science and Technology* 32(4): 341–348.

Suebsing R, Kampeera J, Tookdee B, Withyachumnarnkul B, Turner W, Kiatpathomchai W (2013). Evaluation of colorimetric loop-mediated isothermal amplification (LAMP) assay for visual detection of *Streptococcus agalactiae* and *Streptococcus iniae* in tilapia. *Letters in Applied Microbiology* 57(4): 317–324.

Sun JR, Yan JC, Yeh CY, Lee SY, Lu JJ (2007). Invasive infection with *Streptococcus iniae* in Taiwan. *Journal of Medical Microbiology* 56: 1246–1249.

Tubbs L, Lee P, Diggles B, Jones JB, Shepherd M, Sim-Smith C (2007). A review of aquatic diseases of significance to New Zealand. Final research report for project ZBS 2005-17. National Institute of Water and Atmospheric Research, Wellington, New Zealand, 452 p.

Wang J, Wang K, Chen D, Geng Y, Huang X, He Y, Ji L, Lui T, Wang E, Yang Q, Lai W (2015). Cloning and characterisation of surface-localised α-enolase of *Streptococcus iniae*, an effective protective antigen in mice. *International Journal of Molecular Science* 16(7): 14490–14510.

Weinstein MR, Litt M, Kertesz DA, Wyper P, Rose D, Coulter M, McGeer A, Facklam R, Ostach C, Wiley BM, Borcryk A, Low DE (1997).. Invasive infections due to a fish pathogen, *Streptococcus iniae*. *New England Journal of Medicine* 337: 589–594.

Yuasa K, Kitancharoen N, Kataoka Y, Al-Murbaty FA (1999). *Streptococcus iniae*, the causative agent of mass mortality in rabbitfish *Siganus canaliculatus* in Bahrein. *Journal of Aquatic Animal Health* 11(1): 87–93.

Zhou SM, Fan Y, Zhu, XQ, Xie, MQ, Li AX (2011). Rapid identification of *Streptococcus iniae* by specific PCR assay utilizing genetic markers in ITS rDNA. *Journal of Fish Diseases* 34(4): 265–271.

Zhou SM, Xie M, Zhu XQ, Ma Y, Tan ZL, Li AX (2008). Identification and genetic characterisation of *Streptococcus iniae* strains isolated from diseased fish in China. *Journal of Fish Diseases* 31(11): 869–875.

Zlotkin A, Hershko H, Eldar A (1998). Possible transmission of *Streptococcus iniae* from wild fish to cultured marine fish. *Applied Environmental Microbiology* 64: 4065–4067.

42. *Yersinia ruckeri* (Hagerman and other exotic strains)

42.1. Hazard identification

42.1.1. Aetiological agent

Yersinia ruckeri is a Gram-negative bacterium classified within the Enterobacteriaceae (Ewing *et al*. 1978), first isolated from rainbow trout (*Oncorhynchus mykiss*) in the 1950s (Rucker 1966). The genus *Yersinia* includes 18 species, several of which are zoonotic, including three major human pathogens: *Yersinia pestis,* the agent of plague, while *Y. pseudotuberculosis* and *Y. enterocolitica* cause enteric yersiniosis, which may be fatal in elderly or immunocompromised patients. In comparison, *Y. ruckeri* is considered a minor zoonotic pathogen, causing skin infections, generally associated with tissue damage (Le Guern *et al*. 2016).

Several strains of *Y. ruckeri* have been isolated based on biotype, serotype and outer-membrane proteins (Tobback *et al.* 2009). Four serotypes (01, 02, 03 and 04) are recognised as the agents of enteric redmouth disease (ERM) and yersiniosis in fish (Davies 1991a; Austin & Austin 1993; Carson & Wilson 2009).

Serotype 01 represents a heterogeneous assembly of isolates with respect to pathogenicity and host (Romalde *et al.* 1993). It is further divided into serotypes 01a and 01b (Davies 1991a, 1991b; Verner-Jeffreys *et al.* 2011). Highly virulent strains of serotype 01a cause enteric redmouth (ERM) epizootics in salmonid aquaculture (Busch 1978; Romalde *et al.* 1993; Kahn *et al.* 1999; Barnes 2011). Serotype 01b has been recovered from diseased vaccinated farmed *S. salar* in Chile (Bastardo *et al.* 2011; Navas *et al.* 2014) and in farmed trout (*Oncorhynchus mykiss*) in Spain (Fouz *et al.* 2006). While the 01b isolates are reported from Atlantic salmon (*Salmo salar*) in Australian aquaculture (Kahn *et al.* 1999; Carson & Wilson 2009; Barnes 2011) and from Chinook salmon (*O. tshawytscha*) in New Zealand, these are serologically distinct from the northern hemisphere 01b isolates (Barnes *et al.* 2016).

While some type 02 serovars may cause virulent infection in certain conditions (De Grandis *et al*. 1988), the 02, 03 and 04 serotypes are of generally lower pathogenicity and have a wide host range with a worldwide distribution, including Australia and New Zealand (Kahn *et al*. 1999; Carson & Wilson 2009; Wheeler *et al.* 2009; Barnes 2011; Verner-Jeffreys *et al*. 2011). The 02, 03 and 04 serotypes are not considered further.

The 01a serotype includes several highly virulent clonal types (Barnes 2011), including the Hagerman strain (McDaniel 1971), later identified as clonal type 5 (Carson & Wilson 2009; Pekala *et al.* 2010; Barnes 2011). Other new and emerging virulent clonal types have also been identified, but variability between different serotyping schemes, laboratory methods and standards limit valid comparisons of their host specificity and pathogenicity (Barnes 2011).

42.1.2. OIE status

Infection with *Yersinia ruckeri* is not notifiable to the OIE (OIE 2016).

42.1.3. New Zealand status

Yersiniosis occurs in New Zealand salmonids, caused by low-pathogenic serotypes of *Yersinia ruckeri* including serotype 01b (Anderson *et al.* 1997; Carson & Wilson 2009) and is often isolated from healthy fish (Anderson *et al.* 1994). Serotype 01b is not considered further.

Serotype 01a, including the highly pathogenic Hagerman strain, and other virulent serotypes causing ERM, are considered exotic (Diggles *et al.* 2002; Carson & Wilson 2009). Infection with ERM is a notifiable disease in New Zealand (Anon. 2016).

42.1.4. Epidemiology

The virulent exotic clonal types of the 01a serotype of *Y. ruckeri*, including the Hagerman strain, is reported from salmonid aquaculture in Europe (Bulgaria, Denmark, Finland, France, Italy, Poland, Portugal, Spain, Switzerland, Turkey, United Kingdom and Germany); North America (Canada and the United States); and South America (Chile and Peru) (Davies 1991a, 1991b; Carson & Wilson 2009; Bastardo *et al*. 2011; C. Rodgers, *pers.comm*. 2017). It is exotic to Australia (Barnes *et al.* 2016).

Serotype 01a primarily affects rainbow trout (*Oncorhynchus mykiss*), but all salmonids may be affected (Busch 1978; Barnes 2011; Barnes *et al.* 2016). Epizootic outbreaks are reported in farmed Atlantic salmon (*Salmo salar*) in Norwegian and Scottish marine and freshwater aquaculture (Bruno 1990), as well as from a wide range of marine and freshwater fish , as shown in Table 39 (Kahn *et al.* 1999; Tubbs *et al.* 2007; Carson & Wilson 2009; Barnes 2011). Serotype 01a occurs in non-fish hosts, including shellfish, crustaceans (such as freshwater crayfish), mammals (such as otters *Lutra lutra*) and birds, including the kestrel (*Falco* spp.) and gulls (*Laridae*). These hosts act as carriers with no external signs of disease (Tubbs *et al*. 2007; Barnes 2011; Kumar *et al*. 2015).

Table 39. Families and Species of Fish Susceptible to the Hagerman and Other Exotic, Virulent Strains of *Yersinia ruckeri*

Host Family	Susceptible Fish Species
Acipenseridae	Sturgeon (*Acipenser* spp.)
Anguillidae	European eel (*Anguilla anguilla*), Japanese eel (*A. japonica*)
Cichlidae	Nile tlapia (*Oreochromis niloticus*)
Cyprinidae	Bighead carp (*Hypophthamichthys nobilis*), carp (*Cyprinus carpio*), goldfish (*Carassius auratus*), emerald shiner (*Notemigonus atherinoides*), Catla (*Gibelion catla*), mrigal carp *Cirrhinus mrigala*), Indian mrigal carp (*C. cirrhosus*), rohu carp (*Labeo rohita*), fathead minnow (*Pimephales promelas*), rudd (*Scardinius erythrophthalmus*), roach (*Rutilus rutilus*), silver carp (*Hypophthalmichthys molitrix*)
Gadidae	Atlantic cod (*Gadus morhua*)
Ictaluridae	Channel catfish (*Ameiurus punctatus*)
Lotidae	Burbot (*Lota lota*)
Percidae	European perch (*Perca fluviatilis*)
Salmonidae	Atlantic salmon (*Salmo salar*), brown trout (*S. trutta*), Chinook salmon (*Oncorhynchus tshawytscha*), Coho salmon (*O. kisutch*), cisco (*Coregonus artedi*), rainbow trout (*O. mykiss*), peled (*Coregonus peled*), muksun (*Corregonus muksun*)
Scophthalmidae	Turbot (*Scophthalmus maximus*)
Soleidae	Sole (*Solea solea*)

Fish of all ages may be infected, but epizootics of serotype 01a are more common in younger salmonids (Busch 1978). Mortality in juvenile rainbow trout (*O. mykiss*) may reach 2% per week, incurring cumulative losses of up to 35% in a production cycle (Carson & Wilson 2009).

The life cycle of *Y. ruckeri* is direct and disease transmission is horizontal through the water column. The rate of shedding from infected fish is highly variable and strongly influenced by external stressors including water quality, nutrition and overcrowding (Barnes 2011).

Initial infection of *Y. ruckeri* occurs through the faecal-oral route (Busch 1978), although infection may also occur in aquaculture through contact with contaminated water, handling or cross-contamination of processing equipment (Jonkers *et al*. 2010). Pathogen entry of *Y. ruckeri* occurs across the gill membranes of the host fish (Tobback *et al*. 2009; Kumar *et al*. 2015).

Disease progression of serotype 01a is rapid, with initial mortalities occurring after a 4-8 day incubation period (Busch 1978). External signs include reddening and darkening along the base of the fins, around the mouth and anus, caused by subcutaneous haemorrhaging. Infected fish commonly develop exophthalmia associated with the development of lesions in the choroid gland of the eye (Busch 1978).

After 7 days, infection follows an intracellular phase, with invasion of macrophages (Ryckaert *et al.* 2010). There is progressive haemorrhage and oedema of the visceral organs, particularly of the liver and spleen, leading to generalised septicaemia and tissue necrosis (Busch 1978). Infection may spread to the lateral musculature causing petechial haemorrhages (Lehmann *et al.* 1987), but bacterial concentration of disease in fish flesh is low (Kahn *et al.* 1999).

Chronic infection with serotype 01a develops in 50 to 75% of surviving fish and localises in the lower intestines, brain, gills and eye tissues (Busch & Lingg 1975; Bruno 1990; Barnes 2011). This may persist in aquaculture for up to 102 days after infection (Busch & Lingg 1975; Busch 1978). As the volume of infective cells released by these carrier fish when unstressed is low, their ability to infect healthy fish has been assumed to be limited (Kahn *et al.* 1999). However, carrier fish have been implicated in serotype 01a-related epizootics in Canadian and Scottish aquaculture (Hunter *et al.* 1980), as well as in disease transmission in hatcheries, resulting from secretions associated with the eggs and ova of infected broodstock (Barnes 2011).

Serotype 01a remains viable after freezing for 6 months in salmon carcasses (Anderson *et al.* 1994) and in blood-water discharged from factory processing (Flogstad *et al.* 1991; Hartung & Gerigk 1991). The organism's ability to survive outside the host is uncertain (Barnes 2011). It can remain dormant, or even slowly reproduce in a VBNC (viable, but non-culturable) state for up to three months in cold (less than 6°C) fresh and estuarine waters, as well as sediments and biofilm surfaces (Bruno 1990; Hartung & Gerigk 1991; Tobback *et al.* 2009). Growth occurs from 0°C, but is inhibited by temperatures above 18°C (Hartung & Gerigk 1991; Kahn *et al*. 1999).

The Serotype 01a strain remains viable for up to four months in sea water (salinity greater than 35 ppt.), at lower temperatures (less than 10°C) and marine sediments (Thorsen *et al*. 1992; Romalde *et al.* 1994; Barnes 2011), but survival is reduced at temperatures greater than 20°C (Fernandez *et al*. 1992).

Serotype 1a is acid and temperature tolerant, surviving passage through the avian digestive system (Romalde *et al.* 1994), where it can be transferred between waterways by piscivorous birds or aquatic invertebrates (Barnes 2011). Wild finfish can act as reservoirs of disease for adjacent aquaculture (Romalde *et al.* 1994).

Denaturation of *Y. ruckeri* in fish offal requires ensilage (heat treatment at 60°C for 5 hours) (Smail *et al.* 1990), or heat treatment (cooking to 40°C for 7–8 hours, or 1 hour at 49°C) (Hine & MacDiarmid 1999). Other successful methods include ferric salt and chlorine treatment (100 mg L^{-}

[1]), chlorine treatment (250 mg L^{-1}), formic acid treatment (pH 2.0 for 3 minutes), or alkaline treatment (pH >12 using sodium hydroxide) (Flogstad *et al*. 1991). Inactivation is achieved by antiseptics, including chloramine-T (1000 mg L^{-1} for 24 hours), iodophor treatment with Betadine or Wescodyne (25 ppm iodine for 15 seconds), or sulphonamide Ro5-0037 (50 mg kg^{-1} for 5 days), as treatment for plant and equipment (Kahn *et al.* 1999; Tubbs *et al*. 2007).

UV radiation treatment even at high doses (250m $Wscm^{-2}$) is ineffective (Kahn *et al.* 1999).

Antibiotic and single-strain vaccination programmes have controlled serotype 01a since the 1970s (Bruno 1990), but recent vaccine failures are associated with the emergence of novel strains of serotype 01a and 01b that are not controlled by current vaccines (Barnes 2011; Bastardo *et al.* 2011; Calvez *et al*. 2014; Navas *et al*. 2014).

Fish hosts in New Zealand include all salmonids, goldfish and introduced freshwater carp, perch, rudd and roach (Cyprinidae). Hosts in marine waters include flatfish such as sole (*Solea* spp.) (Soleidae) (Diggles *et al.* 2002; Tubbs *et al*. 2007).

Yersinia ruckeri is a minor zoonotic pathogen (CABI 2018), causing opportunistic human wound infection, where skin is exposed to river water. The spores of *Y. ruckeri* remain viable in freshwater for up to 4 months, and *Yersinia* sp. survives in biofilm and sediments (Tobback *et al.* 2009). Little is known about its pathogenicity in human hosts (de Keukeleire *et al.* 2014), as the source of yersiniosis infection in humans is infrequently reported (Le Guern *et al.* 2016). *Yersinia ruckeri* represented less than 0.01% of the 19,670 human cases of yersiniosis reported in France between 1951 and 2013 (Le Guern *et al*. 2016). There is no evidence to indicate the exotic strains of *Y. ruckeri* have a higher zoonotic potential than endemic strains that are already present in New Zealand.

Other potential infection pathways exist. *Yersinia ruckeri* is reported from goldfish (*C. auratus*) and may be imported through the ornamental finfish pathway (Hine & Diggles 2005).

42.2. Risk assessment

42.2.1. Entry assessment

Infected fish with obvious external signs of infection should not pass visual inspection, but carrier fish with no external signs may be present in the commodity (Kahn *et al*. 1999). *Yersinia ruckeri* disseminates throughout body tissues during septicaemia and will be present in the brain, eye, gill and lateral muscle tissues of eviscerated fish (Barnes 2011). The organism is unaffected by the standard conditions likely to be encountered in commercial fish processing (Kahn *et al.* 1999).

The likelihood of entry of serotype 01a of *Y. ruckeri* is assessed as non-negligible.

42.2.2. Exposure assessment

Rainbow trout is the most susceptible salmonid host of serotype 01a, although other freshwater and coastal marine species could be infected (Tubbs *et al*. 2007). To establish infection, sufficient infected eviscerated product would have to become available for consumption by a susceptible fish host, in sufficient quantity and duration (Kahn *et al.* 1999). Serotype 01a may be present in the blood-water discharged during commercial processing, as well as being associated with the brain, eye, gill and muscle tissue trimmings discarded as offal after processing (Lehmann *et al.* 1987; Anderson *et al.* 1994; Bruno 1990; Kahn *et al.* 1999; Jonkers *et al*. 2010; Barnes 2011). Infection

may also occur through fomites such as the cross-contamination of processing equipment (Romalde *et al.* 1994).

Serotype 01a remains viable for two months in fresh water and up to four months in marine waters. It can survive for several months in a VBNC state (Fernandez *et al*. 1992; Romalde *et al.* 1994), including surviving disposal to landfill. It can be redistributed back into the marine or aquatic environment by scavenging birds (Romalde *et al.* 1994) and suitable host species exist in both marine and fresh water (Tubbs *et al*. 2007).

The likelihood of exposure to serotype 01a of *Y. ruckeri* is assessed as non-negligible.

42.2.3. Consequence assessment

The degradation of water quality through the introduction of serotype 01a would have serious environmental consequences. The organism can remain viable in the aquatic environment for 2-4 months. It survives for extended periods in aquatic sediments and in landfill, where it can be redistributed between waterways by scavenging birds (Horn & Barnes 1999; Thorsen *et al*. 1992; Romalde *et al.* 1993, 1994; Barnes 2011).

Freshwater salmonid aquaculture largely depends on unpolluted waterways, as little or no controls exist on water quality (Sim-Smith *et al*. 2014). Infection can spread from wild to farmed fish (Hunter *et al.* 1980) and be transferred to hatcheries through infected broodstock (Barnes 2011).

The New Zealand Chinook salmon (*O. tshawytscha*) aquaculture is susceptible to the exotic strains of *Y. ruckeri* (Barnes 2011). This industry was valued at $63 million dollars in export earnings in 2011 (Seafood New Zealand 2014). The introduction of serotype 01a may reduce access to markets where serotype 01a is a notifiable disease and incur additional development costs for new vaccines against the 01a serotype (Calvez *et al*. 2014). The use of antibiotics is not acceptable for many export markets (Barnes 2011).

The introduction of serotype 01a would affect the hatchery rearing programmes of the recreational and tourist fisheries for rainbow trout and brown trout. While the dollar value of New Zealand's freshwater fisheries is not fully known, the Lake Taupo trout fishery alone is valued at $70 to 80 million per annum (Marsh & Mkwra 2013). This introduction would also incur social and environmental costs associated with the recreational sports fisheries for introduced cyprinids including roach (*R. rutilus*) and rudd (*S. erythrophthalmus*) (Fish & Game 2014). Its introduction would have significant social and economic consequences for the traditional Maori fisheries for eel (*Anguilla* spp.), as well as affecting the developing aquaculture of grass carp (*Ctenopharyngodon idella*) and silver carp (*H. molitrix*) farmed for weed management of waterways (Mitchell 2009; NIWA 2014)

An outbreak in marine waters may affect several recreational and commercial inshore fisheries, including flatfish (Soleidae) (Tubbs *et al.* 2007).

While *Y. ruckeri* is a minor, opportunistic zoonotic pathogen (Le Guern *et al*. 2016; Cabi 2018), there is no evidence to suggest the exotic serotype 1a strains have a higher risk of human infection than the existing endemic strains present in New Zealand (Anderson *et al.* 1994),

The consequences of the introduction of the 01a serotype of *Y. ruckeri* are assessed as non-negligible.

42.3. Risk management

The 01a serotype of *Y. ruckeri* is not an OIE-listed disease (OIE 2016), so no specific guidance is available on mitigation measures. It is assumed that compliance with the approved processed states for OIE-listed diseases (Appendix 3) would also eliminate the 01a serotype of *Y. ruckeri* from the commodity and be a viable risk management option.

The 01a serotype of *Y. ruckeri* is reported from several families of wild and farmed fish in marine and fresh waters (Table 39), that may be present in the commodity. Species declaration is likely to substantially reduce the occurrence of the *Y. ruckeri* 01a serotype in the commodity and be a viable management option. Restriction of the commodity to wild-caught fish (not from aquaculture) would have little, or no effect on pathogen load and is not a viable risk management option.

The 01a serotype of *Y. ruckeri* is reported from Europe (Bulgaria, Denmark, Finland, France, Italy, Poland, Portugal, Spain, Switzerland, Turkey, United Kingdom and Germany), North America (Canada and the United States) and South America (Chile and Peru). Where country/zone freedom is approved through the MPI Country Approval Procedures, this option is likely to substantially reduce the pathogen load of the 01a serotype of *Y. ruckeri* in the commodity. Approval of a declaration of country/zone freedom by MPI is a viable risk management option.

Yersinia ruckeri is associated with tissues of the brain, eye, gill and lateral musculature (Barnes 2011), so removal of the gills, should slightly reduce *Y. ruckeri* occurrence. As the concentration of *Y. ruckeri* in muscle tissue is likely to be comparatively low, further processing to remove the head and gills should moderately reduce the occurrence of *Y. ruckeri* in the commodity and be a viable risk management option.

Yersinia ruckeri is not affected by freezing so frozen storage is not a viable risk management option. Inactivation requires heating (to at least 49°C for 60 minutes) (Hine & MacDiarmid 1999). Heat treatment would eliminate *Y. ruckeri* from the commodity and be viable risk management options.

42.3.1. Risk management options

The 01a serotype of *Y. ruckeri* is reported from fish in families Acipenseridae, Anguillidae, Cichlidae, Cyprinidae, Gadidae, Ictaluridae, Lotidae, Percidae, Salmonidae, Scophthalmidae and Soleidae (Table 39). These families are considered likely to be present in the commodity. Other families have not been associated with *Y. ruckeri* serotype 01a. Therefore species declaration indicating the commodity is not originated from any of the above families should substantially reduce the occurrence of *Y ruckeri* serotype 01a in the commodity.

For commodities originated from families associated with *Y. ruckeri* serotype 01a, one or a combination of the following additional options could also be considered to effectively manage the risk.

Where country/zone freedom from *Y. ruckeri* serotype 01a is accepted by MPI:

Option 1

Acceptance of country/zone freedom by MPI should substantially reduce the occurrence of *Y. ruckeri* serotype 01a, so the commodity may be imported without any further restrictions.

Where country/zone freedom from *Y. ruckeri* serotype 01a is not accepted by MPI or not available:

Option 2

Processing to a state consistent with the approved states for OIE-listed diseases (Appendix 3) should eliminate *Y. ruckeri* serotype 01a. When these provisions are met, the commodity could be imported without any further restrictions.

Where the imported commodity is not in a state consistent with the approved states for OIE-listed diseases (Appendix 3):

Option 3

Heat treatment (by cooking to at least 49°C for 60 minutes) should eliminate *Y. ruckeri* serotype 01a. When this provision is met, the commodity could be imported without any further restrictions; or,

Option 4

Further processing (by removal of the head and gills) should moderately reduce the occurrence of *Y. ruckeri* serotype 01a. When this provision is met, the commodity could be imported without any further restrictions.

42.4. References

Anderson C, Knowles G, De Lisle G (1994). A survey for *Yersinia ruckeri* and *Aeromonas salmonicida* in farmed and wild fish. *Surveillance* 21(3): 39–40.

Anderson SA, Turner SJ, Lewis GD (1997). Enterococci in the New Zealand environment: implications for water quality monitoring. *Water Science and Technology* 35: 325–331.

Anonymous (2016). Biosecurity (Notifiable Organisms) Order 2016. NZLR 2016/73. New Zealand Government, Wellington, 13 p.

Austin B, Austin DA (1993). Bacterial fish pathogens: Diseases in farmed and wild fish. 4th edition, Springer, New York., United States of America, 552 p.

Barnes AC (2011). Enteric redmouth disease (ERM) (*Yersinia ruckeri*)., pp 484–511 *In:* Woo PTK, Bruno DW (*Eds*.). *Fish Diseases and Disorders: Volume 3 - Viral, Bacterial and Fungal Infections*, 2nd edition. CABI International, Wallingford, United Kingdom, 930 p.

Barnes AC, Deboutteville JD, Gudkovs N, Brosnahan C, Morrison R, Carson J (2016). Whole genome analysis of *Yersinia ruckeri* isolated over 27 years in Australia and New Zealand reveals geographical endemism over multiple lineages and recent evolution under host selection. *Microbial Genomics* 2(11): e000095 [Online] Available from www.doi: 10.1099/mgen.0.000095 [Accessed 23 August 2017].

Bastardo A, Sierralta V, León J, Ravelo C, Romalde JL (2011). Phenotypical and genetic characterization of *Yersinia ruckeri* strains isolated from recent outbreaks in farmed rainbow trout *Oncorhynchus mykiss* (Walbaum) in Peru. *Aquaculture* 317(1–4): 229–232.

Bruno DW (1990). Enteric redmouth disease. Aquaculture Information Service Publication 10, 5 p.

Busch RA (1978). Enteric red mouth disease (Hagerman strain). *Marine Fisheries Review* 40(3): 42–51.

Busch RA, Lingg AJ (1975). Establishment of an asymptomatic carrier state infection of enteric redmouth disease in rainbow trout (*Salmo gairdneri*). *Journal of the Fisheries Research Board of Canada* 32: 2429–2432.

CABI (2018). Enteric redmouth disease. Invasive species compendium, datasheet [Online] Available from http://www.cabi.org/isc/datasheet/83811 [Accessed 31 July 2018].

Calvez S, Gantelet H, Blanc G, Douet DG, Daniel P (2014). *Yersinia ruckeri* biotypes 1 and 2 in France: presence and antibiotic susceptibility. *Diseases of Aquatic Organisms* 109: 117–126.

Carson J, Wilson T (2009). Yersiniosis in fish. Australian New Zealand Standard Diagnostic Procedures. Sub-Committee on Animal Health Laboratory Standards. Commonwealth of Australia, Canberra [Online]. Available from: http://www.scahls.org.au/Procedures/Documents/AQANZSDP/Yersiniosis.pdf [Accessed 12 December 2014].

Davies RL (1991a). Clonal analysis of *Yersinia ruckeri* based on biotypes, serotypes and outer membrane protein-types. *Journal of Fish Diseases* 14: 221–228.

Davies RL (1991b). Virulence and serum-resistance in different clonal groups and serotypes of *Yersinia ruckeri*. *Veterinary Microbiology* 2(3-4): 289–297.

De Grandis SA, Krell PJ, Flett DE, Stevenson RMW (1988). Deoxyribnucleic acid relatedness of serovars of *Yersinia ruckeri*, the enteric redmouth bacterium. *International Journal of Systemic Bacteriology* 38(1): 49–55.

De Keukeleire S, de Bel A, Jansen Y, Jansesens M, Wauters G, Piérard D (2014). *Yersinia ruckeri*, an unusual microorganism isolated from a human wound infection. *New Microbes New Infections* 2(4): 134–135 [Online] Available from: www.doi: 10.1002/nmi2.56 PMCID: PMC4184584 [Accessed 23 August 2017].

Diggles BK, Hine PM, Handley S, Boustead NC (2002). A handbook of diseases of importance to aquaculture in New Zealand. NIWA Science and Technology Series No. 49. National Institute of Water and Atmospheric Research Ltd, Auckland, New Zealand, 200 p.

Ewing WH, Ross AJ, Brenner DJ, Fanning GR (1978). *Yersinia ruckeri* sp. nov., the redmouth (RM) bacterium. *International Journal of Systemic Bacteriology* 28(1): 37–44.

Fernandez AIG, Rodriguez LA, Nieto TP (1992). Survival of bacterial fish pathogens in seawater. *Aquaculture* 107: 271–276.

Fish and Game (2014). New Zealand Fish and Game Council [Online] Available from: http://www.fishandgame.org.nz/ [Accessed 24 September 2014].

Fløgstad H, Schei I, Torgersen Y, Røttereng PJ (1991). Inactivation of pathogens in salmon slaughtery effluents. *In*: DePauw N, Joyce J (*Eds*.), Aquaculture and the Environment. Proceedings of Aquaculture Europe, 10-12 June 1991, Dublin, Ireland. *European Aquaculture Society Special Publication* 14: 104–105.

Fouz B, Zarza C, Amaro C (2006). First description of non-motile *Yersinia ruckeri* serovar I strains causing disease in rainbow trout, *Oncorhynchus mykiss* (Walbaum), cultured in Spain. *Journal of Fish Diseases* 29: 339–346 [Online] Available from: www.doi:10.1111/j.1365-2761.2006.00723.x.Fouz [Accessed 23 August 2016].

Hartung M, Gerigk K (1991). *Yersinia* in effluents from the food processing industry. *OIE Reviews in Science and Technology* 19(3): 799–811.

Hine PM, MacDiarmid SC (1999). Contamination of fish products: risks and prevention. *OIE Review of Science and Technology* 16(1): 135–145.

Hine MP, Diggles BK (2005). Import risk analysis: Ornamental fish. Biosecurity New Zealand. Ministry of Agriculture and Forestry, Wellington, New Zealand, 270 p.

Hunter VA, Kittel MD, Fryer JL (1980). Stress-induced transmission of *Yersinia ruckeri* infection from carriers to recipient steelhead trout *Salmo gairdneri* Richardson. *Journal of Fish Diseases* 3(6): 467–472.

Jonkers ART, Sharkey KJ, Thrush MA, Turnbull JF, Morgan KL (2010). Epidemics and control strategies for diseases of farmed salmonids: A parameter study. *Epidemics* 2: 195–206.

Kahn SA, Beers PT, Findlay VL, Peebles IR, Durham PJ, Wilson DW, Gerrity SE (1999). Import risk analysis on non-viable salmonids and non-salmonid marine finfish. Australian Quarantine and Inspection Service, Department of Agriculture, Fisheries and Forestry, Canberra, Australia, 409 p.

Kumar G, Menanteau-Ledouble S, Saleh M, El-Matbouli M (2015). *Yersinia ruckeri*, the causative agent of enteric redmouth disease in fish. *Veterinary Research* 46 (103): 2–10 [Online] Available from: doi: 10.1186/s13567-015-0238-4 [Accessed 12 October 2015].

LeGuern AS, Martin L, Savin C, Carniel E (2016). Yersiniosis in France: overview and potential sources of infection. *International Journal of Infectious Diseases* 46: 1–7 [Online] Available from: http://www.dx.doi.org/10/1016/j.ijid.2016.03.008 [Accessed 31 July 2017].

Lehmann J, Stüerenberg FJ, Mock D (1987). The changes in haemogram of rainbow trout (*Salmo gairdneri* Richardson) to an artificial and natural challenge with *Yersinia ruckeri*. *Journal of Applied Ichthyology* 3: 174–183.

Marsh D, Mkwra L (2013). Review of freshwater non-market value studies. Department of Economics, University of Waikato, Hamilton, New Zealand, 144 p.

McDaniel DW (1971). Hagerman redmouth: a new look at an old fish problem. *American Fish Farmer and United States Trout News* 1: 14.

Mitchell C (2009). Profile of white amur (grass carp) and silver amur (silver carp) from the results of releases in New Zealand waterways. Charles Mitchell and Associates, Raglan, New Zealand, 15 p.

Navas E, Bohle H, Henríquez P, Grothusen H, Bustamante F, Bustos P, Mancilla M (2014). Draft genome sequence of the fish pathogen *Yersinia ruckeri* strain 37551, serotype O1b, isolated from diseased, vaccinated Atlantic salmon (*Salmo salar*) in Chile. *Genome Announcements* 2(4):e00858-14 [Online] Available from: www.doi:10.1128/genomeA.00858-14 [Accessed 23 August 2017].

NIWA (2014). Grass carp [Online] Available from: https://www.niwa.co.nz/freshwater-and-estuaries/nzffd/NIWA-fish-atlas/fish-species/grass_carp [Accessed 28 April 2015].

OIE (2016). Aquatic Animal Health Code, 19th Edition. World Organization for Animal Health. Paris, France [Online] Available from: http://www.oie.int/international-standard-setting/aquatic-code/access-online/ [Accessed 15 March 2017].

Pekala A, Kozinska A, Antychowicz J (2010). Serological variation among Polish isolates of *Yersinia ruckeri*. *Bulletin of the Veterinary Institute Pulawy* 54: 305–308.

Romalde JL, Barja JL, Magarinos B, Toranzo A (1994). Starvation-survival process of the bacterial fish pathogen *Yersinia ruckeri*. *Systematic and Applied Microbiology* 17: 161–168.

Romalde JL, Magarinos B, Barja JL, Toranzo AE (1993). Antigenic and molecular characterisation of *Yersinia ruckeri* proposal for a new intraspecies classification. *Systematic and Applied Microbiology* 16(3): 411–419.

Rucker RR (1966). Redmouth disease in rainbow trout (*Salmo gairdneri*). *Bulletin Office International Des Epizooties* 66: 825–830.

Ryckaert J, Bossier P, D'Herde K, Diez-Fraile A, Sorgeloos P, Haesbrouck F, Pasmans F (2010). Persistence of *Yersinia ruckeri* in trout macrophages. *Fish and Shellfish Immunology* 29(4): 648–655.

Seafood New Zealand (2014). Seafood New Zealand: Key facts for our Industry. Seafood New Zealand, Wellington, New Zealand, 6 p.

Sim-Smith C, Faire S, Lees A (2014). Managing biosecurity risk for business benefit. Aquaculture biosecurity practices research report prepared for Ministry for Primary Industries, by Coast and Catchment Ltd, New Zealand Wellington, New Zealand, 214 p.

Smail DA, Huntley PJ, Munro ALS (1990). Fate of four fish pathogens after exposure to an ensiling mixture containing fish farm mortalities. ICES CM 1990/F: 50. Mariculture Committee. International Council for the Exploration of the Sea, Copenhagen, Denmark, 7 p.

Thorsen BK, Enger O, Norland S, Hoff KA (1992). Long-term starvation survival of *Yersinia ruckeri* at different salinities studied by microscopical and flow cytometric methods. *Applied and Environmental Microbiology* 58(5): 1624–1628.

Tobback E, Decostere A, Hermans K, Ryckaert J, Duchateau L, Haesebrouck F, Chiers K (2009). Route of entry and tissue distribution of *Yersinia ruckeri* in experimentally infected rainbow trout *Oncorhynchus mykiss. Diseases of Aquatic Organisms* 84(3): 219–218.

Tubbs L, Lee P, Diggles B, Jones JB, Shepherd M, Sim-Smith C (2007). A review of aquatic diseases of significance to New Zealand. Final research report for project ZBS 2005-17. National Institute of Water and Atmospheric Research, Wellington, New Zealand, 452 p.

Verner-Jeffreys DW, Haig SJ, Welch TJ, Pond MJ, Stone D, Davies RL, Gardner R (2011). Characterisation of a serotype O1 *Yersinia ruckeri* isolate from the Isle of Man: further evidence that O antigen serotype is not a reliable indicator of virulence. *Bulletin of the European Association of Fish Pathologists* 31(3): 86–91.

Wheeler R, Davies RL, Dalsgaard I, Garcia J, Welch TW, Wagley S, Bateman KS, Verner-Jeffreys DW (2009). *Yersinia ruckeri* biotype 2 isolates from mainland Europe and the UK likely represent different clonal groups. *Diseases of Aquatic Organisms* 84: 25–33.

43. *Aphanomyces invadans*

43.1. Hazard identification

43.1.1. Aetiological agent

Aphanomyces invadans (= *A. piscicida*) is a water mould, first reported from ayu (*Plecoglossus altivelis*) in Japan (Egusa & Masuda 1971). In some references (e.g. Kahn *et al.* 1999), water moulds are classified as fungi within the Genus *Oomycetida*, of the Family Saprolegniaceae (Kahn *et al.* 1999). However, they have be re-classified as pseudofungi and grouped with diatoms and brown algae, in Phylum Bigyra of the Superphylum Chromista (Stramenopiles) (Gordon 2007; Bruno *et al.* 2011; OIE 2016a).

A single clone of *A. invadans* exists (Afzali *et al*. 2015), which is widely distributed (Lilley *et al.* 1997). It is the primary agent of the epizootic ulcerative syndrome (EUS), also known as red spot disease (RSD), mycotic granulomatosis (MG) and ulcerative mycosis (UM) (OIE 2016b). Since EUS is a syndrome, other pathogens, including *Aeromonas* spp. or viruses, may also be involved (Diggles *et al.* 2002; Vandersea *et al.* 2006).

43.1.2. OIE status

Infection with *A. invadans* is listed by the OIE (OIE 2016a).

43.1.3. New Zealand Status

Aphanomyces invadans is considered exotic (Diggles *et al*. 2002; Tubbs *et al*. 2007; Johnston 2008a), and infection with *A. invadans* is a notifiable disease in New Zealand (Anon. 2016).

43.1.4. Epidemiology

Aphanomyces invadans is considered a warm-water pathogen found in fresh and estuarine waters of over 20 countries across four continents, including Africa (Botswana, Namibia, South Africa and Zambia), Asia (Japan, Papua-New Guinea), Southeast Asia, West Asia (including Pakistan), Australia, North America (United States and Canada), and South America (Kahn *et al.* 1999; Hine & Diggles 2005; OIE 2016b). It is also reported from temperate waters in Japan (Hine & Diggles 2005).

Aphanomyces invadans has low host specificity and is reported from over 94 species of marine, estuarine and freshwater fish (Table 40) (Lilley *et al.* 1998; Noga *et al*. 1991; Chinabut & Roberts 1999; OIE 2016b; Kar 2015). Hosts including common carp (*Cyprinus carpio*), milkfish (*Chanos chanos*) and Nile tilapia (*Oreochromis niloticus*) previously been considered resistant (Lilley *et al.* 1998) now considered to have lower susceptibility, rather than being refractory to disease (Johnston 2008; OIE 2016b).

Aphanomyces invadans is an exotic opportunistic pathogen of fish, where both hyphae and spore stages are infective (Johnston 2008a). Disease transmission is direct and horizontal (Hine & Diggles 2005). Entry occurs through open wounds and abrasions, parasitic attack, or by direct invasion of skin or gill epidermal cells (Hine & Diggles 2005; Bruno *et al.* 2011).

The life cycle is complex, involving alternating sexual and asexual stages, together with additional cycles of primary, secondary and tertiary zoospores and resistant cysts, and zoospores. The details of release, routes of transmission, or mechanisms of survival outside the host, are essentially unknown (Oidtmann *et al*. 2008, Oidtmann 2011).

Table 40. Families and Species of Fish Susceptible to *Aphanomyces invadans*

Family	Host Species and Common Name
Achiridae	Hogchoker (*Trinectes maculatus*)
Alestidae	Striped robber (*Brycinus lateralis*), tigerfish (*Hydrocynus vittatus*)
Ambassidae	Chanda perch (*Ambassis agassizii*),
Anabantidae	Climbing perch (*Anabas testudineus*)
Anguillidae	Eels (*Anguilla* spp.)
Ariidae	Fork-tailed catfish (*Arius* spp.)
Bagridae	Bagrid catfish (*Sperata aor*), dwarf catfish (*Mystus vittatus*) (all Bagridae)
Belonidae	Long tom (*Strongylura krefftii*)
Carangidae	Jacks (*Alepes* spp., *Caranx* spp.)
Centrarchidae	Bluegill (*Lepomis macrochirus*), largemouth black bass (*Micropterus salmoides*)
Channidae	Great snakehead (*Channa marulius*), spotted snakehead (*C. punctata*), striped snakehead (*C. striata*)
Cichlidae	Banded tilapia (*Tilapia sparrmanii*), longfin tilapia (*Oreochromis machrochir*), green happy (*Sargochromis codringtonii*), Nile tilapia (*O. niloticus*), yellow-belly bream (*Serranochromis robustus*), pink happy (*Sargochromis giardi*), rainbow happy (*Sargochromis. carlottae*), redbreast tilapia (*Coptodon rendalli*), thinface cichlid (*Serranochromis angusticeps*), three-spotted tilapia (*Oreochromis andersonii*)
Clariidae	Blunt toothed African catfish (*Clarias ngamensis*), sharptooth African catfish (*C. gariepinus*), walking catfish (*C. batrachus*), catfish (*Clarias* spp.)
Clupeidae	American shad (*Alosa sapidissima*), Atlantic menhaden (*Brevoortia tyrannus*), Australian river gizzard shad (*Nematalosa erebi*), hickory shad (*Alosa mediocris*)
Cyprinidae	Barbs (*Barbus* spp., *Puntius* spp., *Systomus* spp.), catla (*Gibelion catla*), dashtail barb (*Enteromius poechii*), flying barb (*Esomus* sp.), goldfish (*Carassius auratus*), Keti-Bangladeshi (*Osteobrama* spp.), mrigal (*Cirrhinus mrigala*), pool barb (*Puntius sophore*), redeye labeo (*Labeo cylindricus*), rohu (*Labeo rohita*), rosy bitterling (*Rhodeus ocellatus*), rudd (*Scardinius erythrophthalmus*), silver barb (*Barbonymus gonionotus*), slender barb (*Enteromius unitaeniatus*), straightfin barb (*E. paludinosus*), Thamalakane barb (*E. thamalakanensis*), upper Zambezi labeo (*L. lunatus*), Kuria labeo (*L. gonius*)
Eleotridae	Marble goby (*Oxyeleotris marmorata*), sleepy cod (*O. lineolata*)
Exocoetidae	Fflying fish (*Cheilopogon* spp., *Cypselurus* spp., *Exocoetus* spp., *Fodiator* spp., *Hirundichthys* spp., *Parexocoetus* spp., *Prognichthys* spp.)
Gobiidae	Gobies (*Glossogobius* spp., *Gobius* spp.), bar-eyed goby (*G. giuris*), dusky tripletooth goby (*Tridentiger obscurus*)
Ictaluridae	Black bullhead (*Ameiurus melas*), brown bullhead (*A. nebulosus*), channel catfish (*Ameiurus punctatus*)
Kurtidae	Nursery fish (*Kurtus gulliveri*)
Latidae	Barramundi (*Lates calcarifer*)
Lutjanidae	Mangrove jack (*Lutjanus argentimaculatus*)
Mastacembelidae	Spiny eel (*Macrognathus aral, M. pentophthalmos*)
Moronidae	Striped bass (*Morone saxatalis*)
Mugilidae	Mullet (*Mugil* spp. *Liza* spp.), grey mullet (*M. cephalus*), white mullet (*M. curema*)
Notopteridae	Clown knifefish (*Chitala chitala*)
Osphronemidae	Dwarf gourami (*Trichogaster lalius*), giant gourami (*Osphronemus goramy*), snakeskin gourami (*Trichopodus pectoralis*), three-spot gourami (*T. trichopterus*)
Percichthyidae	Australian bass (*Macquaria novemaculeata*), golden perch (*M. ambigua*), freshwater cod (*Maccullochella peelii*)
Platycephalidae	Dusky flathead (*Platycephalus fuscus*)
Plecoglossidae	Ayu (*Plecoglossus altivelis*)
Pomatomidae	Bluefish (*Pomatomus saltatrix*)

Psettodidae	Indian spiny turbot (*Psettodes erumei*), spottail spiny turbot (*Psettodes belcheri*), spiny turbot (*P. bennettii*)
Salmonidae	Rainbow trout (*Oncorhynchus mykiss*)
Scatophagidae	Spotted scat (*Scatophagus argus*), striped scat (*Selenotoca multifasciata*)
Schilbeidae	African butter catfish (*Schilbe mystus*), silver catfish (*S. intermedius*)
Sciaenidae	Atlantic croaker (*Micropogonias undulatus*), spot (*Leiostomus xanthurus*), black drum (*Pogonias* spp.), drum (*Bairdiella chrysoura*), weakfish (*Cynoscion regalis*)
Sillaginidae	Sand whiting (*Sillago ciliata*)
Siluridae	Wels catfish/sheatfish (*Silurus glanis*), wallago catfish (*Wallago attu*)
Soleidae	Narrow banded sole (*Synclidopus macleayanus*)
Sparidae	Goldsilk bream (*Acanthopagrus berda*), pinfish (*Lagodon rhomboides*), sheepshead (*Archosargus probatocephalus*), yellowfin sea bream (*Acanthopagrus australis*)
Synbranchidae	Asian swamp eel (*Monopterus albus*)
Terapontidae	Barcoo grunter (*Scortum barcoo*), silver perch (*Bidyanus bidyanus*), spangled perch (*Leiopotherapon unicolor*), banded grunter (*Amniataba percoides*), terapon (*Terapon theraps*), tiger bass (*T. jarbua*)

Infective primary zoospores are released from spore clusters at the tips of sporangia formed by the vegetative (asexual) fungus-like mycelial structures parasitic on the host tissues (Bruno *et al*. 2011; OIE 2016b). The primary zoospore germinates in water to release a mobile flagellated secondary zoospore, which actively seeks out a new host and remains infective for at least 19 days (Lilley *et al*. 1998). This encysts and germinates to form a new mycelial structure on the host (Lilley *et al.* 1998).

If no suitable host is found, the secondary zoospore can encyst on a suitable substrate, developing further tertiary generations of zoospores (OIE 2016b). While *A. invadans* appears to undergo only one additional cycle in the absence of a suitable substrate, its ability to function as a saprophytic organism is likely, but this is currently unproven (Oidtmann 2011). Fish may be a terminate host, but other, yet unknown hosts and sources of infection may also occur (Vandersea *et al.* 2006).

All zoospore stages can be horizontally transmitted through the water column, but only the secondary zoospores are invasive (OIE 2016b). The periodicity associated with EUS epizootics may relate to the development time required for other encysted stages to transform into the mobile infective stage, before further infection can occur (OIE 2016b). *Aphanomyces invadans* can be spread in fresh waters downstream by river currents, or by flooding (OIE 2016b).

Aphanomyces invadans affects juvenile and young adult fish, causing epizootic mortalities of 50–100% in aquaculture (Kiryu *et al.* 2002; Hine & Diggles 2005; OIE 2016b). Mortalities in wild fish vary from 0.2% for flounder to 1.2% for Atlantic menhaden, and infection may be spread from wild fish to aquaculture stock (Levine *et al.* 1990; Johnston *et al.* 2004).

Infection occurs over a wide temperature range (OIE 2016a), but sporulation is facilitated by high temperatures (18–30°C) and heavy rainfall conditions (Oidtmann 2011; OIE 2016b). Host response is delayed at temperatures of 17–19°C (Catap & Munday 1998; OIE 2016b). Growth *in vitro* does not occur over 37°C, but the effective temperature range is greater than indicted by experimental data, because of the resistant spore stage in the life cycle (Chinabut & Roberts 1999; Oidtmann 2011).

The minimum infectious dose is low, but susceptibility varies among host species (Oidtmann 2011). From experimental data, infection with 1–3 spores per fish results in 80% mortality in grey mullet (*M. cephalus*) and menhaden (*B. tyrannus*) (Sosa *et al*. 2007), but an infective dose of 1,000 spores per fish were required to infect rainbow trout (Oidtmann *et al*. 2008).

Infection progresses with few external signs (Oidtmann 2011; OIE 2016b). Subclinically infected fish act as carriers, but mortality is high, so few fish survive to be lifelong carriers of disease (Hine & Diggles 2005; OIE 2016b).

The rate of disease progression varies from 7 days for susceptible species (such as menhaden *B. tyrannus*), to 30 days or longer for resistant species (such as snakehead *Channa* spp.) (Hine & Diggles 2005). Infection targets the skeletal musculature, beginning with a small circular skin lesion around the point of entry, typically located in the abdomen and anal region. This progresses into necrotic skin ulcers, which develop into mycotic granulomas that eventually reach the spinal cord and skeletal musculature (Johnston *et al*. 2004; Bruno *et al*. 2011). Invasive hyphae ramify through the skeletal musculature, brain and spinal cord, causing extensive necrosis (Chinabut *et al.* 1995; Oidtmann 2011). These injuries are unsightly, usually making the commodity unmarketable (Hutson 2013). Host death results from haemodilution and osmotic imbalance due to loss of epithelial integrity (Bruno *et al*. 2011). *Aphanomyces invadans* continues to develop and release infective spores from moribund and dead host tissues (Oidtmann 2011).

Infective spores are transmitted through blood-water discharges, and spores and pseudohyphae are retained in the tissue trimmings and offal discarded from fish processing. Infection also occurs by cross-infection of plant and handling equipment, or through factory personnel under poor hygiene and handling conditions (Bruno *et al*. 2011).

Aphanomyces invadans is inactivated by salinities greater than 2 ppt (OIE 2016b), suggesting transitory marine species may be infected when in estuarine waters (Johnston *et al*. 2004). It is inactivated by general disinfection chemicals commonly used in aquaculture, such as iodophors (OIE 2016b), hydrogen peroxide or Proxitane 0510 (Diggles *et al*. 2002).

Aphanomyces invadans is inactivated by freezing at -18°C for 72 hours, and inactivation of the related *A. astaci* occurs after freezing at -5°C for 72 hours, although the latter species has thinner pseudohyphae (Oidtmann *et al.* 2002; Johnston 2008; OIE 2016b).

Aphanomyces invadans requires high-temperature treatment to ensure inactivation in mechanically dried eviscerated product (121°C for at least 3.6 minutes, or equivalent); pasteurised product (90°C for at least 10 minutes, or equivalent); or dry heat treated product (100°C for at least 30 minutes, or equivalent) (OIE 2016b).

No vaccine is currently available and control in natural waters is probably impossible (OIE 2016b).

Susceptible freshwater fish in New Zealand include salmonids (*Salmo* spp., *Oncorhynchus* spp.), eels (*Anguilla* spp.), carp (*Cyprinu*s spp.) and many other freshwater fish species (Diggles *et al.* 2002; Tubbs *et al*. 2007). Flounders (Pleuronectidae, Soleidae), grey mullet (*M. cephalus*) and many other species in estuarine waters may also be affected (Diggles *et al.* 2002; Fraser *et al*. 1992).

43.2. Risk assessment

43.2.1. Entry assessment

Although clinically infected fish with obvious external skin lesions would fail visual inspection and not be present in the commodity, infective carrier fish are likely to remain viable in the commodity. *Aphanomyces invadans* present in the skin, musculature, head and gill tissues of

infected eviscerated fish (Bruno *et al.* 2011; OIE 2016b) is likely to remain infective in the commodity.

Aphanomyces invadans is rendered inactive by freezing (to below -5°C for 72 hours) (Oidtmann *et al.* 2002; OIE 2016b) so the likelihood of entry through frozen eviscerated fish is negligible.

The likelihood of entry for fresh or fresh-chilled eviscerated fish is assessed as non-negligible.

43.2.2. Exposure assessment

Aphanomyces invadans has a wide host range in fresh and estuarine waters (OIE 2016a). It remains viable within the temperature range likely to be encountered in commercial fish processing and can survive for extended periods in waters of less than 2 ppt salinity (Bruno *et al*. 2011; Oidtmann 2011).

For an infection to be established in a host population, sufficient infected product would have to become available for consumption by a susceptible estuarine or freshwater fish host, in sufficient quantity and duration (Kahn *et al.* 1999). *Aphanomyces invadans* can be transferred in blood-water discharges, as well as in the offal and fish heads discharged from fish processing facilities (OIE 2016b) into the estuarine or freshwater environment (Oidtmann 2011). Infective spores are environmentally stable and may be spread downstream by river currents, or by flooding (OIE 2016b). The minimum infective dose of 1–3 cell per fish is very low (Sosa *et al.* 2007). Susceptible species include grey mullet (*M. cephalus*) and flounders (Pleuronectidae and Soleidae) in estuarine waters; and salmonids (*Salmo* spp., *Oncorhynchus* spp.) and carp (*Cyprinus* spp.) in fresh water (Tubbs *et al.* 2007).

The likelihood of exposure to *A. invadans* is assessed as non-negligible.

43.2.3. Consequence assessment

The consequences of the establishment of *A. invadans* would be extreme on a wide range of fish species in the aquatic environment, due to its extensive host range, ease of transmission, resilience to environmental conditions and lack of effective treatment (OIE 2016b). The effects on individual host species would be variable, as pathogenicity varies widely (Oidtmann 2011).

Establishment of *A. invadans* would have extreme consequences for freshwater aquaculture (Tubbs *et al*. 2007; Fish & Game 2014), which depends on high quality untreated water supplies (Sim-Smith *et al.* 2014). Affected industries would include Chinook salmon (*O. tshawytscha*) aquaculture, through loss of sales due to infected product. Chinook salmon was valued at $63 million in export earnings in 2011 (Seafood New Zealand 2014).

Establishment would have significant economic and social costs for the recreational rainbow trout (*O. mykiss*) and brown trout (*S. trutta*) fisheries, as well as major economic costs in tourism and fishing-related industries. While the dollar value of New Zealand's freshwater fisheries is not fully known, the Lake Taupo trout fishery alone is valued at $70–80 million per annum (Marsh & Mkwra 2013). Establishment could affect the farming of grass carp (*Ctenopharyngodon idella*) aquaculture (NIWA 2014), used for weed control in New Zealand freshwater lakes and rivers (Hofstra 2014).

Establishment would have social and economic consequences for estuarine and brackish water fish, such as flounders (Pleuronectidae, Soleidae) and grey mullet (*M. cephalus*) (Tubbs *et al*. 2007; OIE 2016a). Flatfish species include yellow-belly flounder (*Rhombosolea leporina*), sand

flounder (*R. plebeian*), black flounder (*R. retiaria*), greenback flounder (*R. tapirina*), lemon sole, (*Pelotretis flavilatus*), New Zealand sole (*Peltorhamphus novaezeelandiae*), brill (*Colistium guntheri*) and turbot (*C. nudipinnis*) that had combined commercial landings of 2,792 t, while 871 t of grey mullet were landed commercially during 2012–13 (MPI 2014).

The consequences of establishment are assessed as non-negligible.

43.2.4. Risk estimation

Since the entry, exposure and consequence assessments are non-negligible, the risk estimate is non-negligible. Therefore, *A. invadans* is assessed to be a risk in the commodity and risk management measures may be justified.

43.3. Risk management

Aphanomyces invadans is the agent of EUS, an OIE-listed disease, so the *Aquatic Code* (OIE 2016a) provides specific guidance for importing eviscerated fish from infected countries, or specific processing requirements that would ensure the destruction of the pathogen.

Aphanomyces invadans affects a wide range of fish in estuarine, brackish and fresh waters, as listed in Table 40. Article 10.2.2 of the *Aquatic Code* recognises the following species to be susceptible:

> Yellowfin sea bream (*Acanthopagrus australis*), climbing perch (*Anabas testudineus*), eels (*Anguillidae*), bagrid catfishes (*Bagridae*), silver perch (*Bidyanus bidyanus*), Atlantic menhaden (*Brevoortia tyrannus*), jacks (*Caranx* spp.), catla (*Gibelion* (*Catla*) *catla*), striped snakehead (*Channa striata*), mrigal (*Cirrhinus mrigala*), torpedo-shaped catfishes (*Clarias* spp.), flying fishes (*Exocoetidae*), tank goby (*Glossogobius giuris*), marble goby (*Oxyeleotris marmorata*), gobies (*Gobiidae*), rohu (*Labeo rohita*), rhinofishes (*Labeo* spp.), barramundi (*Lates calcarifer*), striped mullet (*Mugil cephalus*), mullets (*Mugil* spp. and *Liza* spp.), ayu (*Plecoglossus altivelis*), pool barb (*Puntius sophore*), barcoo grunter (*Scortum barcoo*), sand whiting (*Sillago ciliata*), wells catfishes (*Siluridae*), snakeskin gourami (*Trichopodus pectoralis*), common archer fish (*Toxotes chatareus*), silver barb (*Barbonymus gonionotus*), spotted scat (*Scatophagus argus*), giant gourami (*Osphronemus goramy*), dusky flathead (*Platycephalus fuscus*), spiny turbot (*Psettodes* sp.), rosy bitterling (*Rhodeus ocellatus*), Keti-Bangladeshi (*Osterobrama* sp.), rudd (*Scardinius erythrophthalmus*), terapon (*Terapon* sp.) and three-spot gourami (*Trichopodus trichopterus*).

Species declaration is likely to substantially reduce *A. invadans* from the commodity and be a viable risk management option. *Aphanomyces invadans* occurs at low levels of occurrence in wild fish stocks but is a major fish pathogen in aquaculture (OIE 2018a). Restriction of the commodity to wild-caught fish (not from aquaculture) would moderately reduce pathogen load and be a viable risk management option.

Infection with *A. invadans* (EUS) is an OIE-listed disease reported from over 20 countries worldwide (OIE 2016a). Where country/zone freedom is approved through the MPI Country Approval Procedures, this option is likely to substantially reduce the pathogen load of *A. invadans*

in the commodity. Approval of a declaration of country/zone freedom by MPI is a viable risk management option.

Infection progresses with few or no visible signs (Oidtmann 2011). As *A. invadans* is present in the skin, musculature, gills and head tissues of infected fish, removal of the gills, or the head and gills, would slightly reduce the pathogen load of *A. invadans* in the commodity. Further processing to the skin-off fillet state should moderately reduce the pathogen load of *A. invadans* in the commodity and would be a viable risk management option.

Aphanomyces invadans is denatured by heat treatment, so application of heat treatment as defined in Article 10.2.3 of the OIE *Aquatic Code* (2016a) would eliminate the pathogen from the commodity.

Article 10.2.3 of the *Aquatic Code* states:

1. *Competent Authorities should not require any conditions related to infection with A. invadans, regardless of the infection with A. invadans status of the exporting country, zone or compartment, when authorising the importation or transit of the following aquatic animals and aquatic animal products from the species referred to in Article 10.2.2. which are intended for any purpose and which comply with Article 5.4.1.:*
    - *heat sterilised hermetically sealed fish products (i.e. a heat treatment at 121°C for at least 3.6 minutes or any time/temperature equivalent);*
    - *pasteurised fish products that have been subjected to heat treatment at 90°C for at least ten minutes (or any time/temperature equivalent which has been demonstrated to inactivate A. invadans);*
    - *mechanically dried eviscerated fish (i.e. a heat treatment at 100°C for at least 30 minutes or any time/temperature equivalent which has been demonstrated to inactivate A. invadans);*
    - *frozen eviscerated fish;*
    - *frozen fish fillets or steaks.*

2. *When authorising the importation or transit of aquatic animals and aquatic animal products of a species referred to in Article 10.2.2., other than those referred to in point 1 of Article 10.2.3., Competent Authorities should require the conditions prescribed in Articles 10.2.7. to 10.2.11 relevant to infection with A. invadans status of the exporting country, zone or compartment.*

3. *When considering the importation or transit of aquatic animals and aquatic animal products from an exporting country, zone or compartment not declared free from infection with A. invadans of a species not covered in Article 10.2.2. but which could reasonably be expected to pose a risk of spread of infection with A. invadans, the Competent Authority should conduct a risk analysis in accordance with the recommendations in Chapter 2.1. The exporting country should be informed of the outcome of this assessment.*

Article 10.2.11 of the *Aquatic Code* states:

1. *Competent Authorities should not require any conditions related to infection with A. invadans, regardless of the infection with A. invadans status of the exporting country, zone or compartment, when authorising the importation or transit of fish fillets or steaks*

(chilled) which have been prepared and packaged for retail trade and which comply with Article 5.4.2.

Certain assumptions have been made in assessing the safety of the aquatic animal products mentioned above. Member Countries should refer to these assumptions at Article 5.4.2. and consider whether the assumptions apply to their conditions.

For these commodities Member Countries may wish to consider introducing internal measures to address the risks associated with the commodity being used for any purpose other than for human consumption.

2. *When importing aquatic animals or aquatic animal products, other than those referred to in point 1 above, of species referred to in Article 10.2.2. from a country, zone or compartment not declared free from infection with A. invadans, the Competent Authority of the importing country should assess the risk and apply appropriate risk mitigation measures.*

Compliance with Articles 10.2.3 and 10.2.11 should eliminate *A. invadans* from the commodity and be a viable risk management option.

Aphanomyces invadans is inactivated by frozen storage (to -18°C for at least 72 hours) (Oidtmann *et al.* 2002; Johnston 2008), although the OIE Aquatic *Code* provides no specific time/temperature recommendations (OIE 2016a). Frozen storage would also eliminate *A. invadans* from the commodity and be a viable risk management option.

Aphanomyces invadans may be transmitted through the waste products associated with transport, storage and processing of the commodity. The requirement that all wash and wastewater discharges be appropriately chemically treated (e.g., with iodophors) before discharge, and that all solid wastes, tissue scraps and offal be disposed of through a recognised trade waste disposal procedure, would be viable management options.

43.3.1. Risk management options

Aphanomyces invadans is reported from fish in families Achiridae, Alestidae, Ambassidae, Anabantidae, Anguillidae, Ariidae, Bagridae, Belonidae, Carangidae, Centrarchidae, Channidae, Cichlidae, Clariidae, Clupeidae, Cyprinidae, Eleotridae, Exocoetidae, Gobiidae, Ictaluridae, Kurtidae, Latidae, Lutjanidae, Mastacembelidae, Moronidae, Mugilidae, Notopteridae, Osphronemidae, Percichthyidae, Platycephalidae, Plecoglossidae, Pomatomidae, Psettodidae, Salmonidae, Scatophagidae, Schilbeidae, Sciaenidae, Sillaginidae, Siluridae, Soleidae, Sparidae, Synbranchidae and Terapontidae (Table 40). These families are considered likely to be present in the commodity. Other families have not been associated with *A. invadans*. Therefore species declaration indicating the commodity is not originated from any of the above families should substantially reduce the occurrence of *A. invadans* in the commodity.

For the commodities originated from families associated with *A. invadans*, one or a combination of the following options could also be considered to effectively manage the risk.

Where country/zone freedom from *A. invadans* is accepted by MPI:

Option 1

Acceptance of country/zone freedom by MPI should substantially reduce the occurrence of *A. invadans*, so the commodity may be imported without any further restrictions.

Where country/zone freedom from *A. invadans* is not accepted by MPI or not available:

Option 2

Processing consistent with the conditions of Article 10.2.3 or 10.2.11 of the OIE *Aquatic Code* (OIE 2016a) should eliminate *A. invadans.* When these provisions are met, the commodity could be imported without any further restrictions.

Where the imported commodity is not consistent with Articles 10.2.3 or 10.2.11 of the OIE Aquatic Code (2016a):

Option 3

Frozen storage (to below -20°C for 72 hours) should eliminate *A. invadans*. When this provision is met, the commodity could be imported without any further restrictions.

Option 4

Further processing (to the skin-off fillet state) should moderately reduce the occurrence of *A. invadans.* When this provision is met, the commodity could be imported without any further restrictions.

Option 5

Restriction of the commodity to wild-caught fish (not from aquaculture) should moderately reduce the occurrence of *A. invadans*. When this provision is met, the commodity could be imported without any further restrictions.

43.4. References

Afzali F, Mohd-Daud HH, Shankar S, Shuaib-Kahn M, Afzali S (2015). Detecting *Aphanomyces invadans* in pure cultures and EUS-infected fish lesions by applying PCR. *Malaysian Journal of Medical and Biological Research* 2/2 (5): 136–146 [Online] Available from: http://jmbr-my.weebly.com/uploads/1/3/4/5/13455174/5.10.pdf [Accessed 14 September 2015].

Anonymous (2016). Biosecurity (Notifiable Organisms) Order 2016. NZLR 2016/73. New Zealand Government, Wellington, New Zealand, 13 p.

Bruno DW, VanWest P, Beakes GW (2011). *Saprolegnia* and other oomycetes. pp 669–720. *In*: Woo PTK, Bruno DW (*Eds.*). *Fish Diseases and Disorders: Volume 3 - Viral, Bacterial and Fungal Infections*, 2nd edition. CABI International, Wallingford, United Kingdom, 930 p.

Catap ES, Munday BL (1998). Effects of variations in water temperature and dietary lipids on the expression of epizootic ulcerative syndrome (EUS) in sand whiting, *Sillago ciliata*. *Gyobyo-Kenku* 33: 327–335.

Chinabut S, Roberts RJ (1999). Pathology and histopathology of Epizootic Ulcerative Syndrome (EUS). Aquatic Animal Health Research Institute, Dept. of Fisheries Bangkok, Thailand, 33 p.

Chinabut S, Roberts RJ, Willoughby GR, Pearson MD (1995). Histopathology of snakehead, *Channa striatus* (Bloch), experimentally infected with the specific *Aphanomyces* fungus associated with epizootic ulcerative syndrome (EUS), at different temperatures. *Journal of Fish Diseases* 18: 41–47.

Diggles BK, Hine PM, Handley S, Boustead NC (2002). A handbook of diseases of importance to aquaculture in New Zealand. NIWA Science and Technology Series 49. National Institute of Water and Atmospheric Research, Wellington, New Zealand, 200 p.

Egusa MN, Masuda N (1971). A new fungal disease of *Plecoglossus altivelis. Fish Pathology* 6: 41–46.

Fish and Game (2014). New Zealand Fish and Game Council [Online] Available from: http://www.fishandgame.org.nz/, [Accessed 24 September 2014].

Fraser GC, Callinan RB, Calder LM (1992). *Aphanomyces* species associated with red spot disease: an ulcerative disease of estuarine fish from eastern Australia. *Journal of Fish Diseases* 15: 173–181.

Gordon D (2007). Kingdom Chromista in New Zealand. *Water and Atmosphere* 15(4): 14–15.

Hine PM, Diggles BK (2005). Import Risk Assessment: Ornamental Fish. Biosecurity New Zealand. Ministry of Agriculture and Forestry, Wellington, New Zealand, 264 p.

Hofstra DE (2014). Grass carp effectiveness and effects. Stage 2: Knowledge review. NIWA Client Report HAM2014-060, prepared for the Department of Conservation, Wellington, New Zealand, 140 p.

Hutson KS (2013). Infectious Diseases of Asian Seabass and Health Management, Chapter 6. pp 102–134. *In:* Jerry DR (*Ed*.) Biology and Culture of Asian Seabass *Lates calcarifer.* CRC Press, New York, United States of America, 326 p.

Johnston C (2008). Import risk analysis: frozen skinless and boneless fillet meat of *Oreochromis* spp. fish from China and Brazil for human consumption. MAF Biosecurity New Zealand, Wellington, New Zealand, 106 p.

Johnston RA, Zabrecky J, Kiryu Y, Shields JD (2004). Infection experiments with *Aphanomyces invadans* in four species of estuarine fish. *Journal of Fish Diseases* 27: 287–295.

Kahn SA, Beers PT, Findlay VL, Peebles IR, Durham PJ, Wilson DW, Gerrity SE (1999). Import risk analysis on non-viable salmonids and non-salmonid marine finfish. Australian Quarantine and Inspection Service, Department of Agriculture, Fisheries and Forestry, Canberra, Australia, 409 p.

Kar D (2015). Epizootic Ulcerative Fish Disease Syndrome. Academic Press, New York, United States of America. 314 p.

Kiryu Y, Shields JD, Vogelbein WK, Zwerner DE, Kator H, Blaser VS (2002). Induction of skin ulcers in Atlantic menhaden by injection and aqueous exposure to the zoospores of *Aphanomyces invadans. Journal of Aquatic Animal Health* 14: 11–24.

Levine JF, Hawkins JH, Dykstra MJ, Noga EJ, Moye DW, Cone RS (1990). Species distribution of ulcerative lesions on finfish in the Tar-Pamlico estuary, North Carolina. *Diseases of Aquatic Organisms* 8: 1–5.

Lilley JH, Callinan RB, Chinabut S, Kanchanakhan S, MacRae IH, Phillips MJ (1998). Epizootic ulcerative syndrome (EUS) technical handbook. Aquatic Animal Health Research Institute, Bangkok, Thailand, 88 p.

Lilley JH, Hart D, Richards RH, Roberts RJ, Cerenius L, Soderhall K (1997). Pan-Asian spread of single fungal clone results in large-scale fish kills. *Veterinary Record* 140: 653–654.

Marsh D, Mkwra L (2013). Review of freshwater non-market value studies. Department of Economics, University of Waikato, Hamilton, New Zealand, 144 p.

Ministry for Primary Industries (MPI) (2014). Grey mullet, pp 364–369 *In:* Fisheries Assessment Plenary, May 2014: stock assessments and stock status. Compiled by the Fisheries Science Group, Ministry for Primary Industries, Wellington, New Zealand, 1381 p.

NIWA (2014) Grass carp - *Ctenopharyngodon idella* (Valenciennes, 1844) [Online] Available from: https://www.niwa.co.nz/freshwater-and-estuaries/nzffd/NIWA-fish-atlas/fish-species/grass_carp [Accessed 12 October 2015].

Noga EJ, Wright JF, Levine JF, Dykstra MJ, Hawkins JH (1991). Dermatological diseases affecting fish of the Tar-Pamlico estuary, North Carolina. *Diseases of Aquatic Organisms* 10: 87–92.

Oidtmann B (2011). Review of biological factors relevant to import risk assessments for epizootic ulcerative syndrome (*Aphanomyces invadans*). *Transboundary and Emerging Diseases* 59: 26–39.

Oidtmann B, Steinbauer P, Geiger S, Hoffmann RW (2008). Experimental infection and detection of *Aphanomyces invadans* in European catfish, rainbow trout and European eel. *Diseases of Aquatic Organisms* 82: 195–207.

Oidtmann B, Heitz BE, Rogers D, Hoffmann RW (2002). Transmission of crayfish plague. *Diseases of Aquatic Organisms* 52: 159–167.

OIE (2016a). Aquatic Animal Health Code, 19th Edition. World Organization for Animal Health. Paris, France [Online] Available from: http://www.oie.int/international-standard-setting/aquatic-code/access-online/ [Accessed 15 March 2017].

OIE (2016b). Manual of diagnostic tests for aquatic animals. World Organization for Animal Health. Paris, France [Online] Available from: http://www.oie.int/international-standard-setting/aquatic-manual/access-online/ [Accessed 17 March 2017].

Seafood New Zealand (2014). Seafood New Zealand: Key facts for our Industry. Seafood New Zealand, Wellington, New Zealand, 6 p.

Sim-Smith C, Faire S, Lees A (2014). Managing biosecurity risk for business benefit. Report prepared for the Ministry for Primary Industries by Coast and Catchment Ltd, Auckland, New Zealand, 214 p.

Sosa ER, Landsberg JH, Kiryu Y, Stephenson CM, Cody TT, Dukeman AK, Wolfe HP, Vandersea MW, Litaker RW (2007). Pathogenicity studies with the fungi *Aphanomyces invadans*, *Achlya bisexualis*, and *Phialemonium dimorphosum*: induction of skin ulcers in striped mullet. *Journal of Aquatic Animal Health* 19: 41–48.

Tubbs L, Lee P, Diggles B, Jones JB, Shepherd M, Sim-Smith C (2007). A review of aquatic diseases of significance to New Zealand. Final research report for project ZBS 2005-17. National Institute of Water and Atmospheric Research, Wellington, New Zealand, 452 p.

Vandersea MW, Litaker RW, Yonnish B, Sosa E, Landsberg JH, Pullinger C, Moon-Butzin P, Green J, Morris JA, Kator H, Noga EJ, Testor PA (2006). Molecular assays for detecting *Aphanomyces invadans* in ulcerative mycotic skin lesions. *Applied Environmental Microbiology* 72: 1551–1557.

44. *Ichthyophonus hoferi*

44.1. Hazard identification

44.1.1. Aetiological agent

Ichthyophonus hoferi is the agent of ichthyophoniasis, a disease affecting marine and freshwater fish, which causes tissue spoilage even at low pathogen concentration (McVicar 2011; Conway *et al.* 2014). *Ichthyophonus hoferi* is considered a pseudofungus and is currently classified within the Family Ichthyosporea of the Class Mesomycetozoea (Bruno *et al.* 2011; McVicar 2011; Jones 2013).

Ichthyophonus hoferi is a species complex reported from reptiles, amphibians, birds and invertebrates. While distinct genomovars may infect freshwater, anadromous or marine fish (Conway *et al.* 2014), the taxonomy of this group is uncertain (McVicar 2011).

44.1.2. OIE status

Infection with *I. hoferi* is not listed by the OIE (OIE 2016).

44.1.3. New Zealand status

Ichthyophonus hoferi has not been reported from New Zealand (Stone *et al.* 1997) and is considered exotic (Johnston 2008; Diggles 2011). Infection with *I. hoferi* is not a notifiable disease in New Zealand (Anon. 2016).

44.1.4. Epidemiology

Ichthyophonus hoferi is reported from northern Atlantic waters of Europe and North America, the Northern and North-western Pacific Ocean, including Alaska and Russia, China, Japan and Australia. It is considered to have a global distribution in marine waters (McVicar 2011).

Ichthyophonus hoferi is considered primarily as a pathogen of marine fish in coastal and open ocean waters, but the occurrence of *I. hoferi* in freshwater fish appears related to the spawning movements of infected anadromous fish (Jones 2013), or as a historical consequence of the use of marine fish as feed in freshwater aquaculture (McVicar 2011). *Ichthyophonus hoferi* is also an opportunistic pathogen on moribund fish (McVicar 2011).

Ichthyophonus hoferi has low host specificity (McVicar 2011) The major host species are listed in Table 41, but *I. hoferi* has been reported from over 100 coastal and deep sea marine and freshwater fish species (Agius 1978; Egusa 1983; Eaton *et al.* 1991; Athanassopoulou 1992; Rahimian & Thulin 1998; Mikaelian *et al*. 2000; Hershberger *et al*. 2002; Jones & Dawe 2002; Kent *et al*. 2005; Rohde 2005; Rasmussen *et al*. 2010; Hendricks 2011; McVicar 2011), as well as from elasmobranchs, crustaceans (calanoid copepods), amphibians, reptiles and piscivorous birds (Torgersen *et al*. 2002; McVicar 2011; Hamazaki *et al.* 2013).

Infection may follow several pathways (Hershberger 2012). It may result in acute disease and mortality, chronic disease that reduces host condition and performance, or sub-clinical infection. Disease prevalence increases with host size and age, and is inevitably fatal to the host (Hershberger 2012).

In acute infection, occurrence of *I. hoferi* may reach 78% in wild herring, with mortalities ranging from 20–50% (Jones 2013). These mortality rates are likely to remove infected fish from wild populations during epizootics. The high rates of infection and mortality have been blamed for the stock collapse of the Canadian Pacific herring fishery (Patterson 1996; Marty *et al.* 2003; McVicar 2011).

Table 41. Major Families and Species of Fish Susceptible to *Ichthyophonus hoferi* (Infection has been Reported From More Than 100 Fish Species)

Family	Species
Carangidae	Yellowtail kingfish (*Seriola quinqueradiata*)
Clupeidae	Atlantic herring (*Clupea harengus*), Pacific herring (*Clupea pallasii*), alewife (*Alosa pseudoharengus*), sprat (*Sprattus sprattus*)
Cyprinidae	Carp (*Cyprinus carpio*), goldfish (*Carassius auratus*)
Gadidae	Atlantic cod (*Gadus morhua*), haddock (*Melanogrammus aeglefinus*), Pacific tomcod (*Microgadus proximus*), walleye pollock (*Theragra chalcogramma*)
Lutjanidae	Crimson snapper (*Pristipomoides filamentosus*)
Moronidae	European sea bass (*Dicentrarchus (Morone) labrax*)
Mugilidae	Mullet (*Mugil cephalus*)
Osmeridae	Surf smelt (*Hypomesus pretiosus*)
Paralichthyidae	Speckled sand dab (*Citharichthys stigmaeus*)
Pleuronectidae	European flounder (*Pleuronectes flesus*), yellowtail flounder (*Limanda (Pleuronectes) ferruginea*), European plaice (*Pleuronectes platessa*), Pacific halibut (*Hippoglossus stenolepis*)
Salmonidae	Chinook salmon (*Oncorhynchus tshawytscha*), rainbow trout (*O. mykiss*), brown trout (*Salmo trutta*)
Scombridae	Atlantic mackerel (*Scomber scombrus*)
Sebastidae	Rockfish (*Sebastes* spp.)
Sparidae	Sea bream (*Sparus aurata*)
Trichiuridae	Black scabbard fish (*Aphanopus carbo*)

Wild stocks of marine fish act as a reservoir of infection for farmed marine fish (Diggles 2011), but infection of estuarine species and anadromous fish is initiated in marine waters (McVicar 2011). Infection also occurs in freshwater aquaculture (McVicar 2011) and in wild freshwater fish stocks, but the source of infection remains unknown. No natural disease transfer between farmed and wild stocks in freshwater aquaculture has been substantiated (McVicar 2011).

Transmission follows multiple pathways. This may be horizontal and direct, where schizonts present in the water column are directly ingested, or where infected fish tissue is directly consumed (McVicar 2011).

Horizontal transmission may be indirect, where invertebrate vectors containing endospores are consumed (McVicar 2011), or it may follow complex routes. While planktonic feeding fish become infected by direct ingestion of schizonts, carnivorous species become infected through the consumption of infected fish, partially decomposed fish, or by the ingestion of infected invertebrate vectors (Kocan *et al*. 2010).

Infection may be acute, chronic, or passive. In acute infection, fish commonly show exophthalmia and emaciation, often with curvature of the spine (Stone *et al.* 1997). The skin surface is initially marked with small white papules that indicate the exit points of invasive pseudohyphae that extend from the skeletal and cardiac musculature, which release viable schizonts directly into the water column (Kocan *et al.* 2010; Jones 2013). As disease progresses, these invasive hyphae ramify through the viscera (liver, kidney, gut, spleen), heart and muscle tissues (McVicar 2011) causing granulomatous lesions in the musculature that result in poor flesh quality (softness and malodour) and product rejection (Johnston 2008; McVicar 2011). Infective schizonts are also released directly

from the pseudohyphae through the gills, skin, or faecal discharges, and follow the death of the host (Kent 1992; Rahimian & Thulin 1998).

In chronic infection, the effects on surviving fish at a population level are difficult to quantify (Hamazaki *et al.* 2013). Chronic infection in Chinook salmon (*O. tshawytscha*) results in reduced swimming performance and reduction in spawning success (Rahimian & Thulin 1998; Kocan *et al.* 2009). Chronically infected adult herring (*C. harengus*) have significantly lower body weight and reduced reproductive capacity (Kocan 2001). These fish are less capable of migrating to the feeding and spawning grounds and their overall spawning success is reduced (Kramer-Schadt *et al*. 2010).

Passive clinical infection occurs with no external clinical signs (Jones 2013), but endospores present throughout the host tissues remain inactive until after host death. These germinate within 15–30 minutes of host death, each producing a germination tube that releases infected mobile spores (schizonts) into the surrounding water (Kent 1992). *Ichthyophonus hoferi* is pleomorphic and other pathogen life stages remain to be fully described (Conway *et al.* 2014).

Ichthyophonus hoferi is environmentally tolerant. Endospores and pseudohyphae are unaffected by the standard conditions encountered in fish processing. They remain viable for up to 2 days in chilled (stored at -8 °C) fish flesh (McVicar 2011). They survive in decomposing fish tissue for at least 29 days post-mortem, after which they appear to follow a saprozoic life phase (Kocan *et al*. 2014) and they continue to proliferate in great numbers (McVicar 2011).

Schizonts remain viable for extended periods in fresh water (Hershberger *et al*. 2008) and up to 2 years in sea water (McVicar 2011). They are unaffected by strong to mild acid conditions (pH 3–7) or salinity (ranging from 0–6% NaCl) (Spanggaard & Huss 1996).

Ichthyophonus hoferi is inactivated by freezing (-20 °C for 3 minutes) (Roberts 2013). It remains viable at temperatures up to 25°C, but is inactivated by mild heat treatment (40°C for 3 minutes) (Spanggaard & Huss 1996; Jones 2013).

Growth is inhibited by higher concentrations of salt, such as used in pickling and salting fish products (Spanggaard & Huss 1996), and hyphal growth continues in fish products gas-packaged with CO_2. These processes are ineffective for denaturation of *I. hoferi* (Spanggaard & Huss 1996).

No vaccination or antibiotic treatment is effective against *I. hoferi* and the disease is eventually fatal to infected fish (McVicar 2011).

Susceptible species in New Zealand potentially include most marine fish, all salmonids, and several possible freshwater species hosts (Diggles 2011).

44.2. Risk assessment

44.2.1. Entry assessment

Fish infected with *I. hoferi* may show external signs, including exophthalmia, extensive skin papules and lesions (McVicar 2011). While these fish would be unlikely to pass visual inspection, infection may also be unapparent in the early stages of active infection, or in passively infected fish (McVicar 2011). Such fish would be likely to pass visual inspection and may enter the human food consumption pathway.

Infection is systemic, with pseudohyphae and endospores that ramify through the visceral organs and musculature (Jones 2013). *Ichthyophonus hoferi* is likely to remain viable in the skin, musculature, head and gill tissues of the eviscerated commodity.

Ichthyophonus hoferi is quickly rendered inactive by freezing (to below -20 °C) (Spanggaard & Huss 1996), so the likelihood of entry for frozen eviscerated fish in the commodity is negligible.

However, the likelihood of entry of *I. hoferi* through fresh and fresh-chilled (0°C to 4°C) fish in the eviscerated commodity is assessed as non-negligible.

44.2.2. Exposure assessment

For an infection to be established in a host population, sufficient infected product would have to become available for consumption by a susceptible marine or freshwater host, in sufficient quantity and duration (Kahn *et al.* 1999). *Ichthyophonus hoferi* has a very wide host range in fresh, estuarine and marine waters, including invertebrates as well as fish hosts (McVicar 2011). The severity of an outbreak would depend on the particular host species, but *I. hoferi* remains viable within the temperature range likely to be encountered in commercial fish processing (McVicar 2011).

Ichthyophonus hoferi has a variable life cycle that optimises the success of infecting a wild vertebrate or invertebrate host (McVicar 2011). It may be transferred by the direct release of schizonts from infected muscle or skin scraps, by the germination of dormant endospores present in these tissues, or where schizonts are transferred in the wash water or blood-water discharges into the aquatic environment (Kocan *et al*. 2014). The schizonts are environmentally resistant (Torgersen & Håstein 1995) and *I. hoferi* can survive as a saprophyte for extended periods in estuarine and fresh waters (Kocan *et al*. 2010, 2014).

The likelihood of exposure to *I. hoferi* is assessed as non-negligible.

44.2.3. Consequence assessment

Ichthyophonus hoferi has a wide host range including many inshore coastal marine and estuarine species. Epizootics have been implicated in population-level effects on wild marine fish populations (Patterson 1996; Marty *et al*. 2003), while these outbreaks represent a reservoir of infection for marine fish aquaculture (Diggles 2011).

In the coastal marine environment, for example, infection may affect jack mackerel (*Scomber australasicus*), with an export value of NZ$ 39 million in 2012 (Seafood New Zealand 2014). It could also affect the commercial fishery for flatfish (Pleuronectidae, Soleidae), which comprises yellow-belly flounder (*Rhombosolea leporina*), sand flounder (*R. plebeia*), black flounder (*R. retiaria*), greenback flounder (*R. tapirina*), lemon sole (*Pelotretis flavilatus*), New Zealand sole (*Peltorhamphus novaezeelandiae*), brill (*Colistium guntheri*) and turbot (*C. nudipinnis*). Combined flatfish landings were 2,792 t during 2012–13 (MPI 2014). An outbreak could affect the grey mullet (*Mugil cephalus*) fishery which had commercial landings of 871 t during 2012–13 (MPI 2014). An outbreak in these species would also have major social and environmental costs associated with their significant recreational and Maori target fisheries (Tubbs *et al*. 2007; MPI 2014).

Chinook salmon (*O. tshawytscha*) is susceptible to *I. hoferi* in freshwater aquaculture and in sea cage farming (Kocan *et al*. 2009; Seafood New Zealand 2014). An outbreak could result in direct

economic losses for the salmon farming industry, through loss of sales due to infected product. This industry was valued at NZ$ 63 million in export earnings in 2011 (Seafood New Zealand 2014).

An outbreak in fresh waters may incur environmental costs associated with wild and farmed domestic fisheries (Tubbs *et al*. 2007; Fish & Game 2014). Freshwater aquaculture in New Zealand is based on untreated water supplies (Sim-Smith *et al.* 2014), so an outbreak could have significant economic costs associated with the provision of appropriate water treatment (Seafood New Zealand 2014).

Rainbow trout (*O. mykiss*) and brown trout (*S. trutta*) are also susceptible to *I. hoferi* in fresh water (McVicar 2011). An outbreak in these species would cause social and economic losses in the recreational trout and salmon fisheries, well as in the tourism and other fishing-related industries. While the dollar value of New Zealand's freshwater fisheries is not fully known, the Taupo fishery alone is valued at NZ$70–80 million per annum (Marsh & Mkwra 2013). An outbreak could also affect the farming of grass carp (*Ctenopharyngodon idella*) used for weed control in New Zealand freshwater lakes and rivers (Hofstra 2014; NIWA 2014).

The consequences of establishment are assessed as non-negligible.

44.2.4. Risk estimation

Since the entry, exposure and consequence assessments are non-negligible, the risk estimate is non-negligible. Therefore, *I. hoferi* is assessed to be a risk in the commodity and risk management measures may be justified.

44.3. Risk management

Ichthyophonus hoferi is not an OIE-listed disease (OIE 2016), so no specific guidance is available on mitigation measures. It is assumed that compliance with the approved processed states for OIE-listed diseases (Appendix 3) would also eliminate *I. hoferi* from the commodity and be a viable risk management option.

Ichthyophonus hoferi is mainly associated with 15 families of wild and farmed marine and fresh water fish (Table 41), but it is actually reported from over 100 species of marine and freshwater fish (Hendricks 2011; McVicar 2011). Species declaration is likely to substantially reduce the pathogen load of *I. hoferi* in the commodity and be a viable risk management option. Restriction of the commodity to wild-caught fish (not from aquaculture) would have little, or no effect on pathogen load and is not a viable risk management option.

While no requirements for dedicated monitoring exist, *I. hoferi* is reported from Europe and North America (United States and Canada), the Northern and North-western Pacific Ocean, including Alaska and Russia, China, Japan and Australia (McVicar 2011). Where country/zone freedom is approved through the MPI Country Approval Procedures, this option is likely to substantially reduce the pathogen load of *I. hoferi* in the commodity. Approval of a declaration of country/zone freedom by MPI is a viable risk management option.

Ichthyophonus hoferi infection is systemic, with infection occurring in the gills, skin, head and musculature of infected fish (Kocan *et al.* 2014). Removal of the gills, or the head and gills, should result in a slight reduction in pathogen load of *I. hoferi* in the commodity. Further processing to the

skin-off fillet state should moderately reduce the pathogen load of *I. hoferi* in the commodity. Processing to the skin-off fillet state would be a viable risk management option.

Ichthyophonus hoferi is inactivated by freezing (-20 °C for 3 minutes) (Roberts 2013), so frozen storage is likely to eliminate *I. hoferi* from the commodity. As *I. hoferi* is inactivated by heat treatment, cooking (to at least 40°C for 3 minutes) (Spanggaard & Huss 1996) is also likely to eliminate it from the commodity. Frozen storage and heat treatment are viable risk management options.

44.3.1. Risk management options

Ichthyophonus hoferi is reported from fish in families Carangidae, Clupeidae, Cyprinidae, Gadidae, Lutjanidae, Moronidae, Mugilidae, Osmeridae, Paralichthyidae, Pleuronectidae, Salmonidae, Scombridae, Sebastidae, Sparidae and Trichiuridae (Table 41), which are considered likely to be present in the commodity. Other families have not been associated with *I. hoferi.* Therefore species declaration indicating the commodity is not originated from any of the above families should substantially reduce the occurrence of *I. hoferi* in the commodity.

For the commodities originated from families associated with *I. hoferi*, one or a combination of the following options could also be considered to effectively manage the risk.

Where country/zone freedom from *I. hoferi* is accepted by MPI:

Option 1

Acceptance of country/zone freedom by MPI should substantially reduce the occurrence of *I. hoferi*, so the commodity may be imported without any further restrictions.

Where country/zone freedom from *I. hoferi* is not accepted by MPI or not available:

Option 2

Processing to a state consistent with the approved states for OIE-listed diseases (Appendix 3) should eliminate *I. hoferi.* When these provisions are met, the commodity could be imported without any further restrictions.

Where the imported commodity is not in a state consistent with the approved states for OIE-listed diseases (Appendix 3):

Option 3

Heat treatment (by cooking to at least 40°C for 3 minutes) should eliminate *I. hoferi.* When this provision is met, the commodity could be imported without any further restrictions; or,

Option 4

Frozen storage (to below -20°C for at least 3 minutes) should eliminate *I. hoferi.* When this provision is met, the commodity could be imported without any further restrictions; or,

Option 5

Further processing (to the skin-off fillet state) should moderately reduce the occurrence of *I. hoferi*. When this provision is met, the commodity could be imported without any further restrictions.

44.4. References

Agius C (1978). Infection by an *Ichthyophonus*-like fungus in the deep-sea scabbard fish *Aphanopus carbo* (Trichiuridae) in the North East Atlantic, pp 191–193 *In:* Kinne O. (*Ed.*), Diseases of Marine Animals. Inter-Research, Oldendorf, Germany, 541 p.

Anonymous (2016). Biosecurity (Notifiable Organisms) Order 2016. NZLR 2016/73. New Zealand Government, Wellington, New Zealand, 13 p.

Athanassopoulou, F. (1992). Ichthyophoniasis in sea bream, *Sparus aurata* (L.), and rainbow trout**,** *Oncorhynchus mykiss* (Walbaum), from Greece. *Journal of Fish Diseases* 15(5): 437–441**.**

Bruno DW, VanWest P, Beakes GW (2011). *Saprolegnia* and other oomycetes. pp 669–720. *In:* Woo PTK, Bruno DW (*Eds.*). *Fish Diseases and Disorders: Volume 3 - Viral, Bacterial and Fungal Infections*, 2nd edition. CABI International, Wallingford, United Kingdom, 930 p.

Conway CM, Purcell MK, Elliott DG, Hershberger PK (2014). Detection of *Ichthyophonus* by chromogenic *in situ* hybridisation. *Journal of Fish Diseases* [Online] Available from: http://onlinelibrary.wiley.com/doi/10.1111/jfd.12300/pdf [Accessed 2 May 2015].

Diggles BK (2011). Environmental assessment report – disease risks. DigsFish Services Report DF 11–02. Report prepared for New Zealand King Salmon. DigsFish Services, Bribie Island, Queensland, Australia, 68 p.

Eaton WDM, Kent ML, Meyers TR (1991). Coccidia, X-cell pseudotumors and *Ichthyophonus* sp. infections in walleye pollock (*Theragra chalcogramma*) from Auke Bay, Alaska. *Journal of Wildlife Diseases* 27**(**1): 140–143.

Egusa S (1983). Disease problems in Japanese yellowtail, *Seriola quinqueradiata*, culture: a review. *Journal du Conseil* 182: 10–18.

Fish and Game (2014). New Zealand Fish and Game Council [Online] Available from: http://www.fishandgame.org.nz/ [Accessed 24 September 2014].

Hamazaki T, Kahler E, Borba BM, Burton T (2013). Impact of *Ichthyophonus* infection on spawning success of Yukon River Chinook salmon *Oncorhynchus tshawytscha*. *Diseases of Aquatic Organisms* 106: 207–215.

Hendricks JR (2011). Two new host species for the parasitic fungus *Ichthyophonus hoferi* in the northwest Atlantic. *Journal of the Fisheries Research Board of Canada* 29(12): 1776–1777.

Hershberger PK (2012). Ichthyophonus disease (Ichthyophoniasis), 13 p [Online] Available from: http://afs-fhs.org/perch/resources/14069249443.2.18ichthyophonus2014.pdf [Accessed 12 April 2015].

Hershberger PK, Pacheco CA, Gregg JL, Purcell MK, LaPatra SE (2008). Differential survival of *Ichthyophonus* isolates indicates parasite adaptation to its host environment. *Journal of Parasitology* 94(5): 1055–1959.

Hershberger PK, Stick K, Bui B, Carrol C, Fall B, Mork JA, Perry E, Sweeny E, Wittouck J, Winton J, Kocan R (2002). Incidence of *Ichthyophonus hoferi* in Puget Sound fishes and its increase with age of Pacific herring. *Journal of Aquatic Animal Health* 14: 50–56.

Hofstra DE (2014). Grass carp effectiveness and effects. Stage 2: Knowledge review. NIWA Client Report HAM2014-060, prepared for the Department of Conservation, Wellington, New Zealand, 140 p.

Johnston C (2008). Import risk analysis: frozen skinless and boneless fillet meat of *Oreochromis* spp. fish from China and Brazil for human consumption. MAF Biosecurity New Zealand, Wellington, 106 p.

Jones SRM (2013). *Ichthyophonus*, a systemic mesomycetozoan pathogen of fish. *ICES Identification Leaflet* 3, 7 p.

Jones SRM, Dawe SC (2002). *Ichthyophonus* in Pacific herring and Chinook salmon. *Journal of Fish Diseases* 25: 415–421.

Kahn SA, Beers PT, Findlay VL, Peebles IR, Durham PJ, Wilson DW, Gerrity SE (1999). Import risk analysis on non-viable salmonids and non-salmonid marine finfish. Australian Quarantine and Inspection Service, Department of Agriculture, Fisheries and Forestry, Canberra, Australia, 409 p.

Kent ML (1992). Diseases of seawater net pen-reared salmonid fishes in the Pacific Northwest. *Canadian Special Publications in Fisheries and Aquatic Science* 116, 76 p.

Kent ML, Heidel JR, Marie A. Moriwake A, Moriwaken V, Alexander B, Watral V, Kelley CD (2005). Diseases of Opakapaka *Pristipomoides filamentosus*, pp 183-195 *In*: Walker P, Lester R, Bondad-Reantaso MG (*Eds.*). Diseases in Asian Aquaculture V, Fish Health Section, Asian Fisheries Society, Manila.

Kocan R (2001). Incidence of *Ichthyophonus hoferi* in Puget Sound fishes and its increase with age of Pacific herring. *Journal of Aquatic Animal Health* 14: 50–56.

Kocan RM, Hart L, Lewandowski N, Hershberger P (2014). Viability and infectivity of *Ichthyophonus* sp. in post-mortem pacific herring, *Clupea pallasii*. *Journal of Parasitology* 100(6): 790–796 [Online] Available from: http://www.dx.doi.org/10.1645/14-518.1) [Accessed 12 April 2015].

Kocan RM, Gregg JL, Hershberger PK (2010). Release of infected cells from epidermal ulcers in *Ichthyophonus* sp. – infected Pacific herring (*Clupea pallasii*): Evidence for multiple mechanisms of transmission. *Journal of Parasitology* 96(2): 348–352.

Kocan RM, Hershberger P, Sanders G, Winton J (2009). Effects of temperature on disease progression and swimming stamina in *Ichthyophonus*-infected rainbow trout, *Oncorhynchus mykiss* (Walbaum). *Journal of Fish Diseases* 32(10): 835–843.

Kramer-Schadt S, Holst JC, Skagen D (2010). Analysis of variables associated with the *Ichthyophonus hoferi* epizootics in Norwegian spring spawning herring, 1992-2008. *Canadian Journal of Fisheries and Aquatic Science* 67(8): 1862–1873.

Marsh D, Mkwra L (2013). Review of freshwater non-market value studies. Department of Economics, University of Waikato, Hamilton, New Zealand, 144 p.

Marty GD, Quinn TJ, Carpenter G, Meyers TR, Willits NH (2003). Role of disease in abundance of a Pacific herring (*Clupea pallasi*) population. *Canadian Journal of Fisheries and Aquatic Sciences* 60(10): 1258–1265.

McVicar AH (2011). *Ichthyophonus.* pp 721–747. *In*: Woo PTK, Bruno DW (*Eds.*). *Fish Diseases and Disorders: Volume 3 - Viral, Bacterial and Fungal Infections*, 2nd edition. CABI International, Wallingford, United Kingdom, 930 p.

Mikaelian I, Ouellet M, Pauli B, Rodrigue J, Harshbarger JC, Green DM (2000). *Ichthyophonus*-like infection in wild amphibians from Quebec, Canada. *Diseases of Aquatic Organisms* 40: 195–201.

Ministry for Primary Industries (MPI) (2014). Fisheries Assessment Plenary, May 2014: Stock assessments and stock status. Compiled by the Fisheries Science Group, Ministry for Primary Industries, Wellington, New Zealand, 1381 p.

NIWA (2014). Grass carp *Ctenopharyngodon idella* (Valenciennes, 1844) [Online] Available from: https://www.niwa.co.nz/freshwater-and-estuaries/nzffd/NIWA-fish-atlas/fish-species/grass_carp [Accessed 15 October 2015].

OIE (2016). Aquatic Animal Health Code, 19th Edition. World Organization for Animal Health. Paris, France [Online] Available from: http://www.oie.int/international-standard-setting/aquatic-code/access-online/ [Accessed 15 March 2017].

Patterson KR (1996). Modelling the impact of disease-induced mortality in an exploited population: the outbreak of the fungal parasite *Ichthyophonus hoferi* in the North Sea herring (*Clupea harengus*). *Canadian Journal of Fisheries and Aquatic Sciences* 53*:* 2870–2887.

Rahimian H, Thulin J (1998). Epizootiology of *Ichthyophonus hoferi* in herring populations off the Swedish west coast. *Diseases of Aquatic Organisms* 27: 187–195.

Rasmussen C, Purcell MK, Gregg JL, LaPatra SE, Winton JR, Hershberger PK (2010). Sequence analysis of the internal transcribed spacer (ITS) region reveals a novel clade of *Ichthyophonus* sp. from rainbow trout. *Diseases of Aquatic Organisms* 89: 179–183.

Roberts HE (2013). Fungal diseases, pp 24–26 *In*: Mayer JM, Donnelly TM (*Eds*.). Clinical Veterinary Adviser, Birds and Exotic Pests, Part 1. Elsevier Science, New York, United States of America, 752 p.

Rohde H (2005). Marine Parasitology. CSIRO Publishing, Canberra, Australia, 592 p.

Seafood New Zealand (2014). Seafood New Zealand: Key facts for our Industry. Seafood New Zealand, Wellington, New Zealand, 6 p.

Sim-Smith C, Faire S, Lees A (2014). Managing biosecurity risk for business benefit. Aquaculture biosecurity practices research report prepared for Ministry for Primary Industries, by Coast & Catchment Ltd, New Zealand Wellington, New Zealand, 214 p.

Spanggaard B, Huss HH (1996). Growth of the fish parasite *Ichthyophonus hoferi* under food relevant conditions. *Food Science Technology* 31(5): 427–432.

Stone, MAB, MacDiarmid SC, Pharo HJ (1997). Import health risk analysis: salmonids for human consumption. Ministry of Agriculture Regulatory Authority, New Zealand, 269 p.

Torgersen Y, Håstein T (1995). Disinfection in aquaculture. *OIE Reviews in Science and Technology* 14(2): 419–434.

Torgersen T, Karlsbakk E, Kaartvedt S (2002). Deviating vertical distribution and increased conspicuousness of parasitized *Calanus. Limnology and Oceanography* 47(4): 1187–1191.

Tubbs L, Lee P, Diggles B, Jones JB, Shepherd M, Sim-Smith C (2007). A review of aquatic diseases of significance to New Zealand. Final research report for project ZBS 2005-17. National Institute of Water and Atmospheric Research, Wellington, New Zealand, 452 p.

45. *Sphaerothecum destruens* (Rosette agent)

45.1. Hazard identification

45.1.1. Aetiological agent

Sphaerothecum destruens is an obligate intracellular pathogen of freshwater fish, originally described as the "Rosette agent" in Chinook salmon (*Oncorhynchus tshawytscha*), rainbow trout (*O. mykiss*) and the invasive topmouth gudgeon (*Pseudorasbora parva*), in North America (Harrell *et al*. 1986; Stone *et al.* 1997; Arkush *et al*. 2003; Gozlan *et al.* 2009).).

Sphaerothecum destruens is a pseudofungus, currently classified within the Order Dermocystida, class Mesomycetozoea (Mendoza *et al.* 2002; Gozlan *et al.* 2009; Diggles 2011; Andreou *et al*. 2012).

45.1.2. OIE status

Infection with *S. destruens* is not listed by the OIE (OIE 2016).

45.1.3. New Zealand status

Sphaerothecum destruens has not been reported from the Southern Hemisphere (Diggles 2011) and is considered exotic to New Zealand. Infection with *S. destruens* is not a notifiable disease in New Zealand (Anon. 2016).

45.1.4. Epidemiology

Sphaerothecum destruens is reported from North America, Europe and the United Kingdom (Andreou *et al.* 2012). While most reports are from the aquaculture of freshwater cyprinid and salmonid fish, *S. destruens* also affects wild fish stocks. It occurs in 32% of returning adult Chinook salmon (*O. tshawytscha*) in the Sacramento River in California (Arkush *et al.* 1998). Major fish hosts include the Cyprinidae and Salmonidae (Table 42) (Harrell *et al.* 1986; Andreou *et al.* 2011; Gozlan *et al.* 2014), but *S. destruens* is now regarded as a generalist pathogen, potentially affecting almost all wild and farmed freshwater fish (Andreou *et al.* 2012; Spikmans *et al.* 2013).

Table 42. Main Families and Species of Fish Susceptible to *Sphaerothecum destruens* (All Freshwater Fish may be Susceptible to Infection)

Family	Species
Cyprinidae	Bream (*Abramis brama*), common carp (*Cyprinus carpio*), fathead minnow (*Pimephales promelas*), roach (*Rutilus rutilus*), rudd (*Scardinius erythrophthalmus*), topmouth gudgeon (*Pseudorasbora parva*)
Salmonidae	Atlantic salmon (*Salmo salar*), brown trout (*S. trutta*), brook trout (*Salvelinus fontinalis*) Chinook salmon (*Oncorhynchus tshawytscha*), coho salmon (*O. kisutch*), rainbow trout (*O. mykiss*)

Sphaerothecum destruens has a complex intracellular life cycle, although details are presently unclear (Andreou 2010; Paley *et al*. 2012). Clinical infection may be either acute or chronic, with mortalities of up to 90%, usually with no external signs of infection (Gozlan *et al.* 2009; Andreou 2010). Identification is complex, requiring specific PCR analysis (Spikmans *et al.* 2013).

Mortality in acute infection ranges between 32% and 95% during salmonid epizootics, but infection in surviving fish may remain in a latent state, which may later become chronic (Kahn *et al.* 1999; Gozlan *et al.* 2009; Andreou *et al*. 2012). Surviving fish function as unapparent carriers of disease, while wild populations of brook trout, brown trout, rainbow trout, topmouth gudgeon and sunbleak represent reservoirs of disease for freshwater aquaculture (Andreou *et al*. 2012).

Chronic infection acts over several months, where the on-going mortality may be low (typically 8% in carp, *C. carpio*), but the cumulative mortality is high (Stone *et al.* 1997; Andreou 2010; Andreou *et al*. 2012). Infection may be underestimated or remain undetected in wild freshwater populations due to the chronic mortality pattern of infection (Harrell *et al.* 1986; Andreou 2010; Gozlan *et al.* 2010; Fisher *et al.* 2012; Gozlan *et al.* 2014; Al-Shorbaji *et al.* 2015). It is likely to be under-reported, but is capable of population-level effects on its host species (Spikmans *et al.* 2013).

Disease progression may follow either a nodular, or a systemic (disseminated) pathway (Andreou *et al*. 2011; Paley *et al.* 2012).

In the nodular form, necrotic granulomatous nodules are present in the visceral tissues (liver, kidney, spleen and gut) (Arkush *et al*. 2003; Spikmans *et al.* 2013). Within these nodules, the infective endospores of *S. destruens* occur in intracellular and extra-cellular forms (Arkush *et al.* 2003).

In the systemic form, infection is characterised by the presence of small rosettes of infected cells. These occur throughout the host tissues (including the gill, heart, brain, eye, kidney, liver, spleen, swim bladder, ovary, testis, hind gut and skeletal muscle), usually with little apparent granulomatous host response (Spikmans *et al*. 2013).

Disease transmission is horizontal, through the water column (Paley *et al.* 2012). Endospores of *S. destruens* are released by the lysis of host tissue cells and are excreted by the host in bile, urine, faecal, seminal, or ovarian secretions. Endospores are environmentally tolerant, remaining viable in fresh water for several weeks at 4°C and entering a dormant phase when conditions are unfavourable. Growth occurs over a wide temperature range (4–30°C). While endospores release several mobile uniflagellate zoospores, the rate of release increases with warmer temperatures within this range (Andreou *et al*. 2009; McVicar 2011; Gozlan *et al.* 2014).

Zoospores remain infective in cell culture for up to 44 days at 5°C (Kahn *et al.* 1999). Initial infection occurs across the gill, gut or skin epithelium (Arkush *et al*. 2003), but the details of penetration are unknown and further life cycle stages may be necessary for infection (Paley *et al*. 2012; Gozlan *et al.* 2014). *Sphaerothecum destruens* is considered a primary fish pathogen, but the life cycle is likely to include saprozoic stages that function as opportunistic pathogens, following the death of the host (Spikmans *et al.* 2013; Gozlan *et al*. 2014).

Little information exists on processes for denaturation of *S. destruens* (Spikmans *et al.* 2013). The related *Ichthyophonus hoferi* can be denatured by freezing (-20°C for 72 hours), or by cooking (40°C for 3 minutes) (Jones 2013), so it is assumed that these processes would also be effective against *S. destruens*.

There are no specific sterilisation treatments for *S. destruens.* As an intracellular pathogen, most industry-standard antiseptic treatments are ineffective. The available options, including bronopol, malachite green, formalin and sodium peroxide, present significant health and welfare issues when used at dosage rates required to eliminate the pathogen (Spikmans *et al.* 2013; Gozlan *et al*. 2014).

Potential hosts in New Zealand include all salmonids, particularly Chinook salmon and rainbow trout, and cyprinid species including common carp, roach and rudd, which may act as reservoirs for infection (Gozlan *et al*. 2014). However, *S. destruens* is a generalist pathogen (Andreou *et al*. 2012) and almost all freshwater fish may potentially be affected, or act as reservoirs for infection.

45.2. Risk assessment

45.2.1. Entry assessment

Fish clinically infected with *S. destruens* typically show no external signs (Gozlan *et al*. 2009). These would pass visual inspection and may be present in the commodity. As infection with the nodular form of *S. destruens* is largely confined to the viscera, this should be removed upon evisceration (Spikmans *et al*. 2013). However, infection with the systemic form is likely to affect all tissues and viable endospores present in the skin, musculature, head and gill tissues of infected fish will be retained in the commodity (Spikmans *et al*. 2013).

Sphaerothecum destruens is tolerant to the standard temperatures employed in fish processing and storage of chilled fish (Kahn *et al.* 1999).

The likelihood of entry of *S. destruens* is assessed as non-negligible.

45.2.2. Exposure assessment

To establish infection through commercial fish processing, sufficient infected product would have to become available for consumption by a susceptible fish host in sufficient quantity and duration (Kahn *et al.* 1999). *Sphaerothecum destruens* is resistant to industry-standard antiseptic treatments (Gozlan *et al.* 2014). It may remain viable in the blood-water discharges from factory processing, in the absence of direct contact with infected hosts (Gozlan *et al.* 2014).

Sphaerothecum destruens may remain viable in the trimmings of skin and skeletal muscle, together with the gills, brain and eye tissues associated with fish offal discarded after processing (Paley *et al*. 2012; Spikmans *et al.* 2013).

Endospores are environmentally tolerant, remaining viable in fresh water for several weeks at 4°C, but are able to enter a dormant phase when conditions are unfavourable (Andreou *et al*. 2009; McVicar 2011; Gozlan *et al.* 2014). Growth continues over a wide temperature range (4–30°C) and the infective zoospores remain viable for at least 44 days (Kahn *et al.* 1999).

Reported fish hosts (Table 43) include the Salmonidae, as well as common carp, roach and rudd (Cyprinidae) (Gozlan *et al.* 2014). However, *S. destruens* may be a generalist fish pathogen (Peeler *et al.* 2011; Andreou *et al.* 2012), so practically all freshwater fish could potentially be infected.

The likelihood of exposure to *S. destruens* is assessed as non-negligible.

45.2.3. Consequence assessment

Sphaerothecum destruens is an important emerging generalist pathogen, potentially affecting all freshwater fish and capable of creating population-level effects on wild freshwater fish stocks (Gozlan *et al.* 2010; Peeler *et al.* 2011; Andreou *et al.* 2012), so the environmental consequences of establishment could be extreme (Spikmans *et al.* 2013). The severity of an outbreak would depend on the host species, as virulence and pathogenicity varies widely (Andreou *et al.* 2012).

Sphaerothecum destruens may easily be underestimated or remain undetected in wild freshwater populations due to the chronic mortality pattern of infection (Harrell *et al.* 1986; Andreou 2010; Fisher *et al.* 2012; Gozlan *et al.* 2014; Al-Shorbaji *et al.* 2015), but which ultimately results in host cell death (Andreou *et al.* 2012).

The economic consequences would also be extreme for freshwater aquaculture. As Chinook salmon (*O. tshawytscha*) is particularly susceptible to *S. destruens* (Kahn *et al.* 1999; Andreou *et al.* 2012), a direct effect would be caused by loss of sales of infected product. The Chinook salmon industry was valued at $63 million in export earnings in 2011 (Seafood New Zealand 2014).

Freshwater salmonid aquaculture largely depends on pure untreated water supplies (Sim-Smith *et al.* 2014) and wild fish stocks act as reservoirs of infection (Gozlan *et al.* 2014). An indirect effect of the introduction of *S. destruens* would be the additional costs of water treatments, such as filtration or UV sterilization.

An outbreak would also affect the recreational and tourist fisheries for rainbow trout (*O. mykiss*) and brown trout (*S. trutta*), and incur significant social and environmental costs for associated industries (Fish & Game 2014). While the dollar value of New Zealand's freshwater fisheries is not fully known, the Lake Taupo trout fishery alone is valued at $70–80 million per annum (Marsh & Mkwra 2013). An outbreak would also affect the developing fishery for grass carp and silver carp reared for weed control in rivers and lakes (NIWA 2014).

The consequences of establishment are assessed as non-negligible.

45.2.4. Risk estimation

Since the entry, exposure and consequence assessments are non-negligible, the risk estimate is non-negligible. Therefore, *S. destruens* is assessed to be a risk in the commodity and risk management measures may be justified.

45.3. Risk management

Infection with *S. destruens* is not an OIE-listed disease (OIE 2016a), so no specific guidance is available on mitigation measures and no requirements for dedicated monitoring exist. It is assumed that compliance with the approved processed states for OIE-listed diseases (Appendix 3) would also eliminate *S. destruens* from the commodity and be a viable risk management option.

While the major known hosts of *S. destruens* include wild and farmed salmonid and cyprinid species (Table 42), *S. destruens* is not host-specific and is regarded as an exotic generalist with a potentially wide host range in fresh waters. Species declaration would have little or no effect on the occurrence of *S. destruens* in the commodity and is not considered a viable risk management option. Restriction of the commodity to wild-caught fish (not from aquaculture) would moderately reduce pathogen load in the commodity and is a viable risk management option,

Sphaerothecum destruens is reported from Europe and the United Kingdom, as well as from North America (Gozlan *et al.* 2010; Andreou *et al*. 2012). Where country/zone freedom is approved through the MPI Country Approval Procedures, this option is likely to substantially reduce the pathogen load of *S. destruens* in the commodity. Approval of a declaration of country/zone freedom by MPI is a viable risk management option.

Sphaerothecum destruens occurs in the skin, musculature, head and gill tissues of infected fish, so removal of the gills, or the head and gills should slightly reduce the occurrence of *S. destruens* in the commodity. Further processing to the skin-off fillet state should moderately reduce the occurrence of *S. destruens* and is a viable risk management option.

Sphaerothecum destruens is denatured by freezing (-20°C for 72 hours) (Jones 2013). Frozen storage should eliminate the pathogen from the commodity and be a viable risk management option. *Sphaerothecum destruens* is also denatured by heat (by cooking to at least 40°C for 3 minutes) (Jones 2013) which would eliminate it from the commodity. High-temperature treatment is a viable risk management option.

45.3.1. Risk management options

Sphaerothecum destruens is a generalist pathogen of freshwater fish, mainly reported from fish in families Cyprinidae and Salmonidae (Table 42). Susceptible freshwater fish families, including families Cyprinidae and Salmonidae are considered likely to be present in the commodity. Families of marine fishes have not been associated with *S. destruens*, but no distinction is made in this risk assessment between marine and freshwater fish. Therefore species declaration is likely to have no effect on the occurrence of *S. destruens* in the commodity.

For all families of fish, one or a combination of the following options could be considered to effectively manage the risk:

Where country/zone freedom from *S. destruens* is accepted by MPI:

Option 1

Acceptance of country/zone freedom by MPI should substantially reduce the occurrence of *S. destruens*, so the commodity may be imported without any further restrictions.

Where country/zone freedom from *S. destruens* is not accepted by MPI or not available:

Option 2

Processing to a state consistent with the approved states for OIE-listed diseases (Appendix 3) should eliminate *S. destruens*. When these provisions are met, the commodity could be imported without any further restrictions.

Where the imported commodity is not in a state consistent with the approved states for OIE-listed diseases (Appendix 3):

Option 3

Frozen storage (to below -20°C for at least 72 hours) should eliminate *S. destruens*. When this provision is met, the commodity could be imported without any further restrictions; or,

Option 4

Heat treatment (by cooking to at least 40°C for at least 3 minutes) should eliminate *S. destruens*. When this provision is met, the commodity could be imported without any further restrictions; or,

Option 5

Further processing (to the skin-off fillet state) should moderately reduce the occurrence of *S. destruens*. When this provision is met, the commodity could be imported without any further restrictions; or,

Option 6

Restriction of the commodity to wild-caught fish (not from aquaculture) should moderately reduce the occurrence of *S. destruens*. When this provision is met, the commodity could be imported without any further restrictions.

45.4. References

Al-Shorbaji FN, Gozlan RE, Roche B, Britton JR, Andreou D (2015). The alternative role of direct and environmental transmission in fungal infectious disease in wildlife: threats for biodiversity conservation. *Scientific Reports* [Online] Available from: http://www.ncbi.nlm.nih.gov/pmc/articles/PMC4438611/, doi: 10.1038/srep10368 [Accessed 12 April 2015].

Anonymous (2016). Biosecurity (Notifiable Organisms) Order 2016. NZLR 2016/73. New Zealand Government, Wellington, New Zealand, 13 p.

Andreou D (2010). *Sphaerothecum destruens*: life history traits and host range. Ph.D thesis, University of Cardiff, Cardiff, United Kingdom, 202 p.

Andreou D, Arkush KD, Guegan JF, Gozlan RE (2012). Introduced pathogens and native freshwater biodiversity: A case study of *Sphaerothecum destruens. PLoS ONE* 7(5): e36998. doi: 10.1371/journal.pone.0036998.

Andreou D, Gozlan RE, Stone D, Martin P, Bateman K, Feist SW (2011). *Sphaerothecum destruens* in cyprinids. *Diseases of Aquatic Organisms* 95(2): 145–151.

Andreou D, Gozlan RE, Paley R (2009). Temperature influence on production and longevity of *Sphaerothecum destruens* zoospores. *Journal of Parasitology* 95(6): 1539–1531.

Arkush KD, Mendoza L, Adkison MA, Hedrick RP (2003). Observations on the life stages of *Sphaerothecum destruens* n. g., n. sp., a mesomycetozoan fish pathogen formerly referred to as the rosette agent (correction). *Journal of Eukaryotic Microbiology* 50(6): 430–438.

Arkush KD, Frasca S, Hedrick RP (1998). Pathology associated with the rosette agent, a systemic protist infecting salmonid fishes. *Journal of Aquatic Animal Health* 10: 1–11.

Diggles BK (2011). Environmental assessment report – disease risks. DigFishServices Report DF 11–02. Report prepared for New Zealand King Salmon. DigsFish Services, Bribie Island, Queensland, Australia, 68 p.

Fish and Game (2014). New Zealand Fish and Game Council [Online] Available from: http://www.fishandgame.org.nz/, [Accessed 24 September 2014].

Fisher MC, Henk DA, Briggs CJ, Brownstein JS, Madoff LC, McCraw SL, Gurr SJ (2012). Emerging fungal threats to animal, plant and ecosystem health. *Nature* 484(7393): 186–194.

Gozlan RE, Whipps CM, Andreou D, Arkush KD (2009). Identification of a rosette-like agent as *Sphaerothecum destruens*, a multi-host fish pathogen. *International Journal for Parasitology* 39(10): 1055–1058.

Gozlan RE, Andreou D, Asaeda T, Beyer K, Bouhadad R, Burnard D, Caiola N, Cakic P, Djikanovic V, Esnmaeili HR, Falka I, Golicher D, Harka A, Jeney G, Kovac V, Musil J, Nocita A, Povz M, Poulet N, Viribickas T, Wolter C, Tarkan AS, Tricarcio E, Trichkova T, Verreycken H, Witkowski A, Zhang C, Zweimueller I, Britton JR (2010). Pan-continental invasion of *Pseudorasbora parva*: towards a better understanding of freshwater fish invasions. *Fish and Fisheries* 11: 315–340.

Gozlan RE, Marshall WL, Lije O, Jessop CN, Gleason FH, Andreou D (2014). Current ecological understanding of fungal-like pathogens of fish: what lies beneath? *Frontiers in Microbiology* 5(62): 1–16.

Harrell LW, Elston RA, Scott TM, Wilkinson MT (1986). A significant new systemic disease of net-pen reared Chinook salmon (*Oncorhynchus tshawytscha*) brood stock. *Aquaculture* 55: 249–262.

Jones SRM (2013). *Ichthyophonus*, a systemic mesomycetozoan pathogen of fish. Leaflet No. 3. ICES Identification Leaflet series for Diseases and Parasites of Fish and Shellfish. International Council for the Exploration of the Sea, Copenhagen, Denmark, 5 p.

Kahn SA, Beers PT, Findlay VL, Peebles IR, Durham PJ, Wilson DW, Gerrity SE (1999). Import risk analysis on non-viable salmonids and non-salmonid marine finfish. Australian Quarantine and Inspection Service, Department of Agriculture, Fisheries and Forestry, Canberra, Australia, 409 p.

Marsh D, Mkwra L (2013). Review of freshwater non-market value studies. Department of Economics, University of Waikato, Hamilton, New Zealand, 144 p.

McVicar (2011). *Ichthyophonus.* pp 721–747. *In*: Woo PTK, Bruno DW (*Eds*.). *Fish Diseases and Disorders: Volume 3 - Viral, Bacterial and Fungal Infections*, 2nd edition. CABI International, Wallingford, United Kingdom, 930 p.

Mendoza L, Taylor JW, Ajello L (2002). The Class Mesomycetozoea: a heterogeneous group of microorganisms at the animal-fungal boundary. *Annual Review of Microbiology* 56: 315–344.

NIWA (2014). Grass carp *Ctenopharyngodon idella* (Valenciennes, 1844) [Online] Available from: https://www.niwa.co.nz/freshwater-and-estuaries/nzffd/NIWA-fish-atlas/fish-species/grass_carp [Accessed 15 October 2015].

OIE (2016). Aquatic Animal Health Code, 19th Edition. World Organization for Animal Health. Paris, France [Online] Available from: http://www.oie.int/international-standard-setting/aquatic-code/access-online/ [Accessed 15 March 2017].

Paley RK, Andreou D, Bateman KS, Feist SW (2012). Isolation and culture of *Sphaerothecum destruens* from sunbleak (*Leucaspius delineatus*) in the UK and pathogenicity experiments in Atlantic salmon (*Salmo salar*). *Parasitology* 139: 904–914.

Peeler EJ, Oidtmann BC, Midtlyng PJ, Miossec L, Gozlan RE (2011). Non-native aquatic animal introductions have driven disease emergence in Europe. *Biological Invasions* 13(6): 1291–1303.

Seafood New Zealand (2014). Seafood New Zealand: Key facts for our Industry. Seafood New Zealand, Wellington, New Zealand, 6 p.

Sim-Smith C, Faire S, Lees A (2014). Managing biosecurity risk for business benefit. Report prepared for the Ministry for Primary Industries by Coast and Catchment Ltd, Auckland, New Zealand, 214 p.

Spikmans F, van Tongeren T, Van Alen TA, Van de Velde G, den Camp HJ (2013). High prevalence of the parasite *Sphaerothecum destruens* in the invasive topmouth gudgeon *Pseudorasbora parva* in the Netherlands, a potential threat to native freshwater fish. *Aquatic Invasions* 8: 1–6.

Stone, MAB, MacDiarmid SC, Pharo HJ (1997). Import health risk analysis: salmonids for human consumption. Ministry of Agriculture Regulatory Authority, New Zealand, 269 p.

46. Microsporidian pathogens

46.1. Hazard identification

46.1.1. Aetiological agents

The intracellular parasites *Glugea plecoglossi, Pleistophora* (*Heterosporis*) *anguillarum, Kabatana arthuri, Microsporidium seriolae*, *Nucleospora* (*Enterocytozoon*) *salmonis* and *Tetramicra brevifilum* are obligate pathogens of marine and freshwater fish (Dykova 2006). They are currently classified as highly specialised fungi (Keeling & Fast 2002; Anane & Attouchi 2010; Ghosh & Weiss 2011; Didier *et al.* 2014).

46.1.2. OIE status

Infection with microsporidian disease is not listed by the OIE (OIE 2016).

46.1.3. New Zealand status

The microsporidians *Glugea plecoglossi*, *Kabatana arthuri*, *Nucleospora* s*almonis* and *Pleistophora anguillarum* are considered exotic to New Zealand (Stone *et al*. 1997; Hine *et al*. 2000; Duignan *et al.* 2003; Hine & Diggles 2005; Johnston 2008). While little is known about the microsporidian fauna of New Zealand (Jones 1979), it appears reasonable to assume that *Microsporidium seriolae and Tetramicra brevifilum*, which have yet to be reported, are also exotic to New Zealand. Infection with microsporidian pathogens is not a notifiable disease in New Zealand (Anon. 2016).

46.1.4. Epidemiology

Microsporidians are opportunistic pathogens or agents of tissue spoilage with a wide geographical distribution, including Africa, Asia, Europe and North America (Dykova 2006; Abowei & Ezekiel 2011; Jithendran *et al*. 2011). While most are host-specific, at least to the genus level (Shaw 1999), others show broad host specificity and infect a larger number of fish hosts (Dykova 2006; Leiro *et al*. 2012). *Pleistophora salmonis* and *Kabatana arthuri* are agents of economic disease in salmonid and seriolid fish aquaculture, respectively (Dykova 2006; Johnston 2008).

Infected fish may become emaciated, and juvenile mortality is high (Dykova 2006). Mortality is generally lower in adult fish, even when heavily infected (Hine & Diggles 2005; Abowei & Ezekiel 2011; Leiro *et al.* 2012). The major effect of microsporidian infection is post-mortem myoliquifaction resulting from the breakdown of the macroscopic cysts during spore release. This commonly results in rejection of the commercial fish product (Leiro *et al*. 2012; Mackenzie & Kalavati 2014).

The genera *Pleistophora, Kabatana* and *Microsporidium* predominantly occur in muscle, nervous, or gill tissues (Dykova 2006). Others including genera *Glugea* and *Tetramicra* show low organ specificity and may be present throughout the host tissues (Dykova 2006). These would be retained upon evisceration and may be present in the commodity (Dykova 2006; Johnston 2008). Zenoma-forming microsporidia do not seem to be strictly host specific (Lom & Dykova 2005).

Fish families and species associated with these microsporidian pathogens are given in Table 43 (Takahashi 1978; Lom *et al*. 2000; Lom 2002; Lee *et al.* 2004; Dykova 2006; Johnston 2008).

Table 43. Families and Species of Fish Susceptible to Muscle Encysting Microsporidia *Glugea plecoglossi, Pleistophora anguillarum, Kabatana arthuri, Microsporidium seriolae, Nucleospora salmonis* and *Tetramicra brevifilum*

Host Family	Host Species	Pathogen
Anguillidae	Japanese eel (*Anguilla japonica*), European eel (A. anguilla)	*Pleistophora anguillarum*
Carangidae	Yellowtail kingfish (*Seriola quinqueradiata*)	*Microsporidium seriolae*
Lophiidae	Black bellied angler (*Lophius budegassa*)	*Tetramicra brevifilum*
Pangasiidae	Swai catfish (*Pangasius sutchi*)	*Kabatana arthuri*
Plecoglossidae	Ayu (*Plecoglossus altivelis*)	*Glugea plecoglossi*
Pleuronectidae	American plaice (*Hippoglossoides* (*Pleuronectes*) *platessoides*), spotted halibut (Verasper varigatus)	*Microsporidium seriolae*
Salmonidae	Rainbow trout (*Oncorhynchus mykiss*), pink salmon (*O. gorbuscha*), Masu salmon (*O. masou*), chum salmon (*O. keta*), white spotted char (*Salvelinus leucomaenis*), brook trout (*Salvelinus fontinalis*), brown trout *(Salmo trutta*),	*Glugea plecoglossi, Nucleospora salmonis*
Scopthalmidae	Turbot (*Scophthalmus maximus*)	*Tetramicra brevifilum*

Microsporidians infecting fish have been assumed to have a simple direct horizontal fish-to-fish life cycle, although details are usually incompletely known (Dykova 2006; Hedrick *et al*. 2012; Mackenzie & Kalavati 2014). Some life cycles involve a secondary molluscan host (Dykova 2006; Mackenzie & Kalavati 2014), while vertical transmission occurs in *Nucleospora salmonis* (Hedrick *et al*. 2012).

Infection occurs either by ingestion of a microspore in the water column, or after predation of an infected host (Dykova 2006; Lee *et al.* 2004). Infected fish typically show no external signs of infection (Hedrick *et al.* 2012).

The microspore releases an invasive amoeboid spiroplasma, which invades the gut epithelium (Sibley 2011). This migrates via the bloodstream where it invades a neutrophil, or invades a host cell in a body organ, in the connective, epithelial or muscle tissues (Dykova 2006). Further development is intracellular, where replication occurs by multiple cycles of sporogony and merogony (Dykova 2006).

Clinical disease typically follows one of two pathways (Dykova & Lom 1980; Hedrick *et al*. 2012). In the most common pathway, infected cells progressively lose their internal structure by hypertrophy and the cell contents are replaced by spores through sporogony and merogony (Dykova 2006). Infection progresses to adjacent healthy cells by the extrusion of sporangial plasmodia and the secretion of organoleptic discharges, which break down adjacent cell walls (Dykova 2006). This initiates a host response and creates a necrotic granulomatous boil-type lesion (the xenoma) (Dykova & Lom 2000; Abowei & Ezekiel 2011). These xenomas, which are created as a result of the host-pathogen reaction, significantly reduce the functionality of the infected organ (Dykova 2006) and predispose the host to secondary infection (Lom & Dykova, 2002). The presence of xenomas renders the flesh unfit for human consumption (Dykova 2006).

Infection may follow a second pathway, which elicits less host reaction (Dykova 2006). In *Kabatana* spp., *Pleistophora* spp., *Microsporidium* spp. and *Nucleospora* spp. infections, invasive spiroplasmas invade individual muscle cells (Brown & Kent 2002; Dykova 2006), creating a network of diffuse granulomatous lesions throughout the muscle and connective tissue. Each lesion also grows by individually secreting organoleptic enzymes, resulting in a more generalised host response, but still renders the tissues unfit for human consumption (Dykova 2006).

Spores generally remain in the host tissues and are released on the death of the host (Dykova 2006), unless the necrosis reaches the epidermis and forms a surface lesion (Dykova & Lom 2000).

The spores are also released during the evisceration and processing of infected fish (Leiro *et al*. 2012).

The spores survive temperature extremes, variations in pH and salinity, for periods of days to several years at 4°C (Dykova 2006). They remain viable after passage through the avian gut and are transferred between waterways by piscivorous birds (Dykova 2006; Weiser 2014), as well as being present in groundwater (Leiro *et al*. 2012).

Most spores are inactivated by freezing (-20°C for 72 hours (Takahashi 1978; Dykova 2006; Leiro *et al*. 2012), but some are more resistant. The spores of *Pleistophora* spp., which are commonly found in Alaska pollock (*Gadus chalcogramma*), require freezing at -20°C for several weeks to ensure inactivation (Priebe 1986; Leiro *et al*. 2012).

Spores can be inactivated by heating (60°C for 15 minutes), or by microwave treatment (750 W for 60 seconds) (Leiro *et al*. 2012). They are inactivated by chemical treatment, including formaldehyde (1%), ethanol (70%), phenol or hydrogen peroxide based treatments (Leiro *et al*. 2012).

Pleistophora spp. and *Microsporidium* spp. are emerging opportunistic zoonotic pathogens, primarily causing acute keratitis and diahorrea in healthy individuals (Han & Weiss 2017). Chronic microsporidiosis is more serious in the elderly, organ transplant patients, or otherwise immunocompromised individuals. Infection occurs through consumption of raw or under-cooked fish products (Kotler & Orenstein 1998; Lores *et al*. 2002; Anane & Attouchi 2010; Cali *et al.* 2011; Stentiford *et al*. 2013; Didier *et al.* 2014; Han & Weiss 2017). The epidemiology of microsporidian zoonotic infection remains uncertain, but microsporidiosis may affect almost every tissue and organ (Mathis *et al.* 2005; Freeman 2015; Stentiford 2015). Symptoms include enteropathy, keratoconjunctivitis, sinusitis, tracheobronchitis, encephalitis, interstitial nephritis, hepatitis, cholecystitis, osteomyelitis, and myositis. Individuals commonly suffer chronic wasting diahorrea and malabsorption, which are potentially fatal if untreated (Kotler & Orenstein 1998).

Suitable hosts in New Zealand include members of the families Anguillidae (*Anguilla* spp.), Salmonidae (*Oncorhynchus* spp., *Salmo* spp., *Salvelinus* spp.), Carangidae (*Seriola* spp.) and Pleuronectidae (*Rhombosolea* spp.) (Dykova 2006; Johnston 2008).

46.2. Risk assessment

46.2.1. Entry assessment

Fish heavily infected with microsporidians in the skin and musculature would be likely to have epidermal lesions or externally visible xenomas (Dykova 2006). These would be unlikely to pass visual inspection, but sub-clinically infected fish may be present in the commodity (Johnston 2008).

Pleistophora anguillarum, *K. arthuri*, *M. seriolae* and *N. salmonis* may be present in muscle tissues; while *G. plecoglossi* and *T. brevifilum* may occur in connective tissue, typically with no external signs of infection (Dykova 2006). These fish may pass visual inspection and be present in the commodity (Dykova 2006; Johnston 2008; Leiro *et al*. 2012).

The likelihood of entry of *Glugea plecoglossi*, *Kabatana arthuri*, *Microsporidium seriolae*, *Nucleospora salmonis*, *Pleistophora anguillarum* and *Tetramicra brevifilum* through the commodity is assessed as non-negligible.

46.2.2. Exposure assessment

To establish in New Zealand, infected product would have to become available for consumption by a susceptible fish host in sufficient quantity and duration (Kahn *et al.* 1999). Most microsporidians are host-specific, at least to genus level (Dykova 2006), so the establishment of *K. arthuri* or *T. brevifilum* is unlikely as suitable hosts are not present in New Zealand (Johnston 2008). These taxa are therefore eliminated from further consideration in the assessment.

Establishment is possible for *P. anguillarum*, *G. plecoglossi*, *M. seriolae* and *N. salmonis*, as alternative hosts occur in New Zealand (e.g., eels, salmonids and kingfish) (Dykova 2006; Johnston 2008).

Microsporidian spores are considered environmentally resistant (Dykova 2006) and unaffected by the low temperatures (4°C) of chilled fish storage and processing (Leiro *et al.* 2012). The spores may be introduced through wash water and blood-water discharged during processing, or through the infected skin, muscle trimmings, or fish heads and gill tissues discarded as offal after processing. They may be transferred between waterways by piscivorous birds and may be present in groundwater (Johnston 2008; Leiro *et al*. 2012; Weiser 2014).

The likelihood of establishment of *Glugea plecoglossi*, *Pleistophora anguillarum, Microsporidium seriolae and Nucleospora salmonis* through the commodity is assessed as non-negligible.

46.2.3. Consequence assessment

The consequences of establishment of microsporidian pathogens may be economically significant (Dykova 2006). Microsporidian epizootics were implicated in the population-level collapse of the North American ocean pout (*Zoarces americanus*) fishery in the 1940s (Kent *et al*. 2014). Epizootic infections of *Nucleospora salmonis* are reported in coho salmon (*Oncorhynchus kisutch*) and Chinook salmon (*O. tshawytscha*) aquaculture in North America (Kent *et al.* 1989, 2014).

Freshwater aquaculture in New Zealand is dependent on pure uncontaminated water and is poorly protected against downstream contamination by microorganisms (Sim-Smith *et al.* 2014). The introduction of *Nucleospora salmonis* would result in direct economic losses in the freshwater Chinook salmon farming industry, through loss of sales due to infected product. The Chinook salmon industry was valued at $63 million in export earnings in 2011 (Seafood New Zealand 2014). An outbreak would also affect recreational and tourist fisheries for other trout and salmon species, While the dollar value of New Zealand's freshwater fisheries is not fully known, the Lake Taupo trout fishery alone is valued at $70–80 million per annum (Marsh & Mkwra 2013).

The introduction of *Pleistophora anguillarum* would impact on the commercial fishery valued at $4.9 million per annum (Jellyman 2012). Eels support small domestic and recreational fisheries and represent a significant taonga for Maori (Parliamentary Commissioner for the Environment 2013). The eel fishery resource is considered to be stressed at current commercial catch levels (MPI 2014).

In marine waters, the introduction of *Glugea plecoglossi* would impact on the inshore fisheries for flounders (Feist & Longshaw 2008), while *Microsporidium seriolae* could impact the developing aquaculture for yellowtail kingfish (*Seriola lalandi*) (NIWA 2017).

Several microsporidians are opportunistic zoonotic pathogens, with potentially serious consequences for elderly and immunocompromised individuals (Kotler & Orenstein 1998; Lores *et*

al. 2002; Anane & Attouchi 2010; Cali *et al.* 2011; Stentiford *et al*. 2013; Didier *et al.* 2014; Han & Weiss 2017).

The consequences of establishment of *Glugea plecoglossi*, *Pleistophora anguillarum*, *Microsporidium seriolae*, and *Nucleospora salmonis* are assessed as non-negligible.

46.2.4. Risk estimation

Since the entry, exposure and consequence assessments are non-negligible, the risk is estimated to be non-negligible. Therefore *Glugea plecoglossi*, *Pleistophora anguillarum*, *Microsporidium seriolae*, and *Nucleospora salmonis* are assessed to be a risk in the commodity and risk management measures may be justified.

46.3. Risk management

Infection with microsporidian pathogens is not an OIE-listed disease (OIE 2016), so no specific guidance is available on mitigation measures and no requirements for dedicated monitoring exist.

It is assumed that compliance with the approved processed states for OIE-listed diseases (Appendix 3) would also eliminate microsporidian pathogens from the commodity and be a viable risk management option.

Microsporidian pathogens are generally host-specific to the genus level (Shaw 1999; Dykova 2006). The muscle-encysting microsporidians *Glugea plecoglossi*, *Pleistophora anguillarum*, *Microsporidium seriolae*, and *Nucleospora salmonis* are associated with 8 families of fish (Table 43), which are likely to be present in the commodity. Species declaration should substantially reduce the pathogen load of these microsporidians in the commodity and be a viable risk management option. Clinical infection is associated with aquaculture and the incidence of microsporidian infection is generally lower in wild fish (Dykova 2006; Johnston 2008). Restriction to wild-caught fish (not from aquaculture) should moderately reduce pathogen load and be a viable risk management option.

The microsporidians *Glugea plecoglossi*, *Pleistophora anguillarum*, *Microsporidium seriolae*, and *Nucleospora salmonis* are widely reported, including Africa, Asia, India, Europe and North America (Dykova 2006). Where country/zone freedom is approved through the MPI Country Approval Procedures, this option is likely to substantially reduce the pathogen load of these exotic microsporidian pathogens in the commodity. Approval of a declaration of country/zone freedom by MPI is a viable risk management option.

Glugea plecoglossi, *Pleistophora anguillarum*, *Microsporidium seriolae*, and *Nucleospora salmonis* are associated with gill tissues, connective or nervous tissues and the somatic musculature, so removal of the gills, or the head and gills should slightly reduce the pathogen load in the commodity. Further processing to the skin-off fillet state should moderately reduce the pathogen load of these microsporidians in the commodity and be a viable risk management option.

Microsporidian pathogens may require frozen storage for several weeks to ensure inactivation (Priebe 1986). Extended freezing (-20°C for at least 28 days) should eliminate *Glugea plecoglossi*, *Pleistophora anguillarum*, *Microsporidium seriolae*, and *Nucleospora salmonis* from the commodity and be a viable risk management option.

Denaturation requires high temperatures (to at least 60°C for 15 minutes) (Leiro *et al.* 2012) which should eliminate microsporidian pathogens from the commodity. Heat treatment is a viable risk management option.

46.3.1. Risk management options

The microsporidian pathogens *Glugea plecoglossi*, *Pleistophora anguillarum*, *Microsporidium seriolae*, and *Nucleospora salmonis* are reported from families Anguillidae, Carangidae, Plecoglossidae, Pleuronectidae and Salmonidae (Table 43), which are considered likely to be present in the commodity. Other families have not been associated with these microsporidian pathogens. Therefore species declaration indicating the commodity is not originated from any of the above families should substantially reduce the occurrence of *Glugea plecoglossi*, *Pleistophora anguillarum*, *Microsporidium seriolae*, and *Nucleospora salmonis* in the commodity.

For the commodities originated from families associated with *Glugea plecoglossi*, *Pleistophora anguillarum*, *Microsporidium seriolae*, and *Nucleospora salmonis*, one or a combination of the following options could also be considered to effectively manage the risk.

Where country/zone freedom from *Glugea plecoglossi*, *Pleistophora anguillarum*, *Microsporidium seriolae* and *Nucleospora salmonis* is accepted by MPI:

Option 1

Acceptance of country/zone freedom by MPI should substantially reduce the occurrence of these pathogens, so the commodity may be imported without any further restrictions.

Where country/zone freedom from *Glugea plecoglossi*, *Pleistophora anguillarum*, *Microsporidium seriolae*, and *Nucleospora salmonis* is not accepted by MPI or not available:

Option 2

Processing to a state consistent with the approved states for OIE-listed diseases (Appendix 3) should eliminate these microsporidian pathogens. When these provisions are met, the commodity could be imported without any further restrictions.

Where the imported commodity is not in a state consistent with the approved states for OIE-listed diseases (Appendix 3):

Option 3

Heat treatment (by cooking to at least 60°C for 15 minutes) should eliminate these microsporidian pathogens. When this provision is met, the commodity could be imported without any further restrictions; or,

Option 4

Frozen storage (to below -20°C for 28 days). should eliminate these microsporidian pathogens. When this provision is met, the commodity could be imported without any further restrictions; or,

Option 5

Further processing (to the skin-off fillet state) should moderately reduce the occurrence of these microsporian pathogens. When this provision is met, the commodity could be imported without any further restrictions; or,

Option 6

Restriction of the commodity to wild-caught fish (not from aquaculture) should moderately reduce the occurrence of these microsporidian pathogens.

46.4. References

Abowei JFN, Ezekiel EN (2011). A review of Myxosporea, Microspora and Monogenea infections in African fish. *British Journal of Pharmacology and Toxicology* 2(5): 236–250.

Anane S, Attouchi H (2010). Microsporidiosis: Epidemiology, clinical data and therapy. *Gastroenterologie Clinique et Biologique* 34: 450–464.

Anonymous (2016). Biosecurity (Notifiable Organisms) Order 2016. NZLR2016/73. New Zealand Government, Wellington, 13 p.

Brown AMV, Kent ML (2002). Molecular diagnostics for *Loma salmonae* and *Nucleospora salmonis* (Microsporidia), pp 267–283 *In:* Cunningham CO (*Ed*.). Molecular diagnosis of salmonid diseases. Academic Press, New York, United States of America, 345 p.

Cali A, Neafie RC, Takvorian PM (2011). Microsporidiosis. Topics on the Pathology of Protozoan and Invasive Arthropod Diseases. Rutgers University Department of Biological Sciences, New Jersey, United States of America, 25 p.

Corradi N, Keeling PJ (2009). Microsporidia: a journey through radical taxonomic revisions. *Fungal Biology Reviews* 23(1–2): 1–8.

Didier ES, Becnel JJ, Kent ML, Sanders JL, Weiss LM (2014). Microsporidia, pp 115–129 *In:* McLauchlin DJ, Spatafora JW (*Eds.*). The Mycota: Systematics and Evolution, VII Part A, 2nd Edition. Springer-Verlag, Berlin, Germany, 461 p.

Diggles BK (2003). Some pathological abnormalities of New Zealand fishes. *New Zealand Journal of Marine and Freshwater Research* 37(4): 705–713.

Duignan PJ, Hine PM, Joy M, Gibbs N, Jones GW, Okeoma C (2003). Disease surveillance in freshwater fish from the lower North Island. *Surveillance* 30(3): 6–8.

Dykova I (2006). Phylum Microspora. *In*: Woo PTK (2006). (*Ed.*) Fish Diseases and Disorders. Volume 1: Protozoan and Metazoan Infections (Second Edition). CABI International, Wallingford, United Kingdom, 791 p.

Dykova I, Lom J (2000). Histopathology of *Kabatana arthuri* (Microspora) infection in sutchi catfish *Pangasius sutchi*. *Folia Parasitologica* 47: 161–166.

Dykova I, Lom J (1980). Tissue reactions to microsporidian infections in fish. *Journal of Fish Diseases* 3(4): 265–283.

Feist SW, Longshaw M (2008). Histopathology of fish parasite infections: importance for populations. *Journal of Fish Biology* 73(9): 2143–2160.

Freeman M (2015). Wild and cultured fish as potential sources of zoonotic infections in humans, pp 79-85 *In:* Microsporidia in the animal to human food chain: An international symposium to address chronic epizootic disease. OECD Co-operative Research Programme. [Online] Available from: http://www.sipweb.org/docs/Proceedings.pdf [Accessed 4 August 2017].

Ghosh K, Weiss LM (2011). T-cell response and persistence of the microsporidia. *FEMS Microbiology Reviews* 36(3): 748–760.

Han B, Weiss LM (2017). Microsporidia: Obligate Intracellular Pathogens within the Fungal Kingdom. *Microbiology Spectrum* 5(2) [Online] Available from: http://www. doi:10.1128/microbiolspec.FUNK-0018-2016 [Accessed 9 August 2018].

Hedrick RP, Purcell MK, Kurobe T (2012). Salmonid intracellular microsporidiosis. Section 3.2.17, Blue Book, American Fisheries Society (AFS) [Online] Available from: http://afs-fhs.org/perch/resources/3.2.17-nucleospora-2014.pdf [Accessed 12 November 2014].

Hine M, Diggles BK (2005). Import risk analysis: Ornamental fish. Ministry of Agriculture and Forestry, Wellington, New Zealand, 264 p.

Hine PM, Jones JB, Diggles BK (2000). A checklist of the parasites of New Zealand fishes, including previously unpublished records. NIWA Technical Report 75. National Institute of Water and Atmospheric Research Ltd., Auckland, New Zealand, 93 p.

Jellyman D (2012). The status of longfin eels in New Zealand – an overview of stocks and harvest. NIWA client report CHC2012-006. Report prepared for the Parliamentary Commissioner for the Environment by the National Institute of Water and Atmospheric Research. Christchurch, New Zealand, 78 p.

Jithendran KP, Vijayan KK, Kailasam M (2011). Microsporidian (*Glugea* sp.) infection in the grey grouper *Epinephelus tauvina* (Forskal 1997). *Indian Journal of Fisheries* 58(3): 125–127.

Johnston C (2008). Import Risk Analysis: Frozen skinless and boneless fillet meat of *Pangasius* spp. fish from Vietnam for human consumption. Biosecurity New Zealand, Ministry of Agriculture and Forestry, Wellington, New Zealand, 60 p.

Jones JB (1979). Trouble with Microsporidia. *New Zealand Journal of Zoology* 6: 648.

Kahn SA, Beers PT, Findlay VL, Peebles IR, Durham PJ, Wilson DW, Gerrity SE (1999). Import risk analysis on non-viable salmonids and non-salmonid marine finfish. Australian Quarantine and Inspection Service, Department of Agriculture, Fisheries and Forestry, Canberra, Australia, 409 p.

Keeling PJ, Fast NM (2002). Microsporidia: Biology and evolution of highly reduced intracellular parasites. *Annual Reviews in Microbiology* 56: 93–116.

Kent ML, Shaw RW, Sanders JL (2014). Microsporidia in fish, pp 493-520 *In:* Weiss LM, Becnel JJ (*Eds*.). Microsporidia: Pathogens of Opportunity, John Wiley, New York, United States of America. [Online] Avalable from: https://doi.org/10.1002/9781118395264.ch20 [Accessed 9 August 2018].

Kent ML, Elliott DG, Groff JM, Hedrick RP (1989). *Loma salmonae* (Protozoa: Microspora) infections in seawater reared coho salmon *Oncorhynchus kisutch*. *Aquaculture* 80(3-4): 211–222 [Online] Available from https://doi.org/10.1016/0044-8486(89)90169-5 [Accessed 8 August 2018].

Kotler DP, Orenstein JM (1998). Clinical syndromes associated with microsporidiosis. *Advances in Parasitology* 40: 321–349.

Lee SJ, Yokoyama H, Ogawa K (2004). Modes of transmission of *Glugea plecoglossi* (Microspora) via the skin and digestive tract in an experimental infection model using rainbow trout, *Oncorhynchus mykiss* (Walbaum). *Journal of Fish Diseases* 27(8): 435–444.

Leiro JM, Piazzon C, Dominguez B, Mallo N, Lamas J (2012). Evaluation of some physical and chemical treatments for inactivating microsporidian spores isolated from fish. *International Journal of Food Microbiology* 156: 152–160.

Lom J (2002). A catalogue of described genera and species of microsporidians parasitic in fish. *Systematic Parasitology* 53: 81–99.

Lom J, Dykova I, Wang CH, Lo CF, Kou GH (2000). Ultrastructural justification for the transfer of *Pleistophora anguillarum* Hoshina 1959 to the genus *Heterosporis* Schubert 1969. *Diseases of Aquatic Organisms* 43: 225–231.

Lom J, Dykova I (1992). Protozoan Parasites of Fishes. *Developments in Aquaculture and Fisheries Science* (Book 26), Elsevier Science, New York, United States of America, 315 p.

Lores B, del Aguila C, Arias C (2002) *Enterocytozoon bieneusi* (microsporidia) in faecal samples from domestic animals from Galicia, Spain. *Memoirs of the Oswaldo Cruz Institute* 97(7): 941–945.

Marsh D, Mkwra L (2013). Review of freshwater non-market value studies. Department of Economics, University of Waikato, Hamilton, New Zealand, 144 p.

Mathis A, Weber R, Deplazes P (2005). Zoonotic Potential of the Microsporidia. Clinical Microbiology Reviews 18(3): 423–445.

Ministry for Primary Industries (MPI) (2014). Fisheries Assessment Plenary, May 2014: Stock assessments and stock status. Compiled by the Fisheries Science Group, Ministry for Primary Industries, Wellington, New Zealand, 1381 p.

NIWA (2017). Yellowtail kingfish: Aquaculture. National Institute for Water and Atmospheric research (NIWA), Auckland, New Zealand [Online] Available from: https://www.niwa.co.nz/aquaculture/aquaculture-species/yellowtail-kingfish [Accessed 15 August 2017].

OIE (2016). Aquatic Animal Health Code, 19th Edition. World Organization for Animal Health. Paris, France [Online] Available from: http://www.oie.int/international-standard-setting/aquatic-code/access-online/ [Accessed 15 March 2017].

Parliamentary Commissioner for the Environment (2013). On a pathway to extinction? An investigation into the status and management of the longfin eel. Parliamentary Commission for the Environment, Wellington, New Zealand, 95 p.

Priebe K (1986). Muscle parasites of the Alaska pollock *Theragra chalcogramma*, pp 235–242 *In:* Anonymous. Arbeitstsgung des Arbeitgebietes "Lebensnittelhygiene" Grmisch-Partenkirchen, 9–12 September (cited in Johnston 2008).

Seafood New Zealand (2014). Seafood New Zealand: Key facts for our Industry. Seafood New Zealand, Wellington, New Zealand, 6 p.

Shaw RW, Kent ML (1999). Fish Microsporidia, pp 418-446 *In:* Wittner M, Weiss L (*Eds.*). The Microsporidia and Microsporidiosis. American Society for Microbiology (ASM) Press, Washington, DC, United States of America [Online] Available from: www.doi: 10.1128/9781555818227.ch13 [Accessed 15 March 2017].

Shinn AP, Pratoomyot J, Bron JE, Paladini G, Brooker EE, Brooker AJ (2014). Economic costs of protistan and metazoan parasites to global mariculture. *Parasitology* Special Issue Article Cambridge University Press, United Kingdom, 75 p [Online] Available from: http://www.doi.10.10107/S0031182014111437 [Accessed 15 March 2017].

Sibley LD (2011). Invasion and intracellular survival by protozoan parasites. *Immunological Review* 240(1): 72–91.

Sim-Smith C, Faire S, Lees A (2014). Managing biosecurity risk for business benefit. Report prepared for the Ministry for Primary Industries by Coast and Catchment Ltd, Auckland, New Zealand, 214 p.

Stentiford GD (2015). The *Enterocytozoon* clade of Microsporidians: Invertebrate, fish and human pathogens pp 86-92. *In:* Microsporidia in the animal to human food chain: An international symposium to address chronic epizootic disease. OECD Co-operative Research Programme [Online] Available from: http://www.sipweb.org/docs/Proceedings.pdf [Accessed 4 August 2017].

Stentiford GD, Feist SW, Stone DM, Bateman KS, Dunn AM (2013). Microsporidia: diverse, dynamic and emergent pathogens in aquatic systems. *Trends in Parasitology* 29(11): 567–578.

Stone, MAB, MacDiarmid SC, Pharo HJ (1997). Import health risk analysis: salmonids for human consumption. Ministry of Agriculture Regulatory Authority, New Zealand, 269 p.

Takahashi S (1978). Microsporidiosis in fish with emphasis on glugeosis of ayu. *Fish Pathology, Japan* 13(1): 9–16.

Weiser J (2014). Microsporidia in invertebrates: Host-parasite relations at the organismal level. pp 164–175. *In:* Buklla LA, Cheong TC (*Eds.*). Biology of the Microsporidia. Springer Science, New York, United States of America, 371 p.

Vossbrinck CR, Debrunner-Vossbrinck BA, Weiss LM (2014). Phylogeny of the Microsporidia, pp 203–220 *In:* Weiss LM, Becnel JJ (*Eds.*). Microsporidia: Pathogens of Opportunity, 1st Edition, Wiley, Chichester, United Kingdom, 709 p. [Online] Available from: www. doi: 10.1002/9781118395264.ch6 [Accessed 4 August 2017].

Wongtavatchaia J, Conrad PA, Hedrick RP (1995). Effect of the microsporidian *Enterocytozoon salmonis* on the immune response of chinook salmon. *Veterinary Immunology and Immunopathology* 48 (3–4): 367–374.

Yokoyama H, Yokoyama F, Zhang JY, Tsurukoa K, Ogawa K (2008). Microsporidian infection in the trunk muscle of hatchery-bred juvenile spotted halibut *Verasper varigatus. Fish Pathology* 43(4): 137–143.

47. Myxosporean pathogens

47.1. Hazard identification

47.1.1. Aetiological agents

The genera *Ceratomyxa*, *Enteromyxum*, *Henneguya*, *Kudoa*, *Myxobolus*, *Parvicapsula*, *Sphaerospora*, *Thelohanellus* and *Unicapsula* are classified in subclass Myxosporea, of class Myxozoa (feist & Longshaw 2006).

47.1.2. OIE status

Diseases associated with myxosporean pathogens are not listed by the OIE (OIE 2016a).

47.1.3. New Zealand status

Several myxosporeans are reported from New Zealand marine and freshwater fish, including species of *Auerbachia*, *Ceratomyxa*, *Chloromyxum*, *Myxodavisia*, *Henneguya*, *Myxidium*, *Myxobolus* and *Thelohanellus* (Hine *et al.* 2000).

47.1.4. Epidemiology

Myxosporean pathogens are generally of low pathogenicity where chronic infection causes little apparent harm. The genera *Ceratomyxa*, *Enteromyxum*, *Henneguya*, *Kudoa*, *Myxobolus*, *Parvicapsula*, *Sphaerospora*, *Thelohanellus* and *Unicapsula* are recognised fish pathogens, while myxosporean pseudocysts present in the musculature can taint the flesh, rendering the commodity unsuitable for marketing and sale (Feist & Longshaw 2006).

The myxosporean life cycle is complex, and poorly known for the majority of species (Feist & Longshaw 2006). Some species are capable of direct (fish-to-fish) transmission (e.g., *Enteromyxum* spp. (Feist & Longshaw 2006)), however most have a two-host life cycle that alternates between a vertebrate intermediate host (typically a fish) and an invertebrate definitive host (typically an annelid) (Rangel *et al.* 2017).

Myxosporeans are generally host specific at the fish family level (Feist & Longshaw 2006; Forro & Eszerbauer 2016), but the level of specificity for invertebrate hosts is unclear. There are two infective life stages–myxospores and actinospores–which are released from the vertebrate and invertebrate hosts, respectively.

In fish, infection is initiated when the actinospore penetrates the epidermis. Further development is complex and poorly understood, but involves proliferative extrasporogonic stages that invade both the surrounding connective tissues and red blood cells of the host. Infection then spreads through the circulatory system to potentially infect all organ systems. While the site of encystment may be variable, myxosporean species are generally associated with a particular organ system (Feist & Longshaw 2006). Each secondary cell finally develops into a pseudocyst. Species that preferentially encyst in the viscera, internal organs and reproductive systems of their host (Benz & Bullard 2004; Feist & Longshaw 2006, Caira *et al.* 2012; Mckenzie & Kalavati 2014) should not be present in the commodity and are not considered further.

Myxosporean pseudocysts present in the cartilaginous backbone of elasmobranchs, or the central nervous system, gills and skeletal musculature of fish, rarely result in pathological disease (Borucinska & Frasca 2002; Feist & Longshaw 2006; Garner 2013). However, they may become pathogenic at high levels of infection, or when the host is under environmental stress (Sinderman 1990). Pseudocysts of *Ceratomyxa* spp., *Enteromyxum* spp., *Henneguya* spp., *Kudoa* spp., *Myxobolus* sp., *Sphaerospora* sp. and *Unicapsula* spp. are commonly associated with the gills, epidermis or skeletal musculature of fish. While infection of the skin and dermal musculature is rarely pathogenic (Benz & Bullard 2004), the macroscopic pseudocysts within the musculature are commonly associated with tissue myoliquifaction (Gleeson *et al.* 2010).

A representative, but not exhaustive, list of fish families and species associated with infection with these myxosporeans is given in Table 44 (Davis 1924; Lester 1982; Sarkar 1984, 1999; Naidjenova & Zaika 1970; Lom & Dykova 1992, Kostoingue & Toguebaye 1994; Yokoyama *et al.* 1998; Diebakate *et al.* 1999; Eszterbauer *et al.* 2000; Aseeva & Krasin 2001; Feist & Longshaw 2006; Lom & Dykova 2006; Alama-Bermejo *et al.* 2009; Diggles 2011; Abe & Maehara 2013; Miller & Adlard 2013; Yokoyama *et al.* 2014; Kasai *et al.* 2017).

Myxosporeans may be spread by the translocation of their fish host. The introduction of *Myxobolus cerebralis* to North America, New Zealand, South Africa and Australia is thought to have occurred through transfer of non-viable chilled fish for human consumption, but transfer can also occur by the live transport of their intermediate (fish) or definitive (annelid) host through the pet and ornamental fish trade (Feist & Longshaw 2006).

Myxosporean actinospores are resistant to cold and are environmentally stable (Hoffman 1990). *Myxobolus cerebralis* spores in sediments and suspension remained infective to *Tubifex* sp. intermediate hosts after being held at 5°C for 7 days, but their viability decreased (log reduction = 4.3) after longer storage (2 months at 20°C) (Hedrick *et al.* 2008). *Myxobolus cerebralis* was denatured by freezing (to below or -80°C for 7 days), but the effectiveness of frozen storage in pathogen mitigation may be reduced at high pathogen concentrations (Hedrick *et al.* 2008) and zoonotic infection is reported following consumption of previously frozen fillets (Boreham *et al.* 1998).

Myxosporeans survive passage through avian digestive systems (El-Matbouli & Hoffmann 1991). *Myxobolus cerebralis* spores survive in sand and water, remaining infective to *Tubifex* sp. hosts for up to 12 months (Nehring *et al.* 2015). Myxosporeans encysted in the musculature of their fish host commonly remain viable until the decomposition of the host tissues, and other fish may be infected directly by consumption of infected tissues (Feist & Longshaw 2006). Myxosporeans can withstand heat treatment, requiring cooking to at least 75°C for 5 minutes (Iwashita *et al.* 2013) to ensure denaturation. They may also be denatured by desiccation (105 minutes to at least 42°C) and by the standard UV treatment (at 40 mJ cm^{-2}) used in aquaculture, but the effectiveness of UV may be limited by organic content in the water (Hedrick *et al.* 2008).

Myxosporeans are resistant to chemical treatments such as chlorination and high concentrations (500 mg L^{-1} for 15 minutes) are required for effective treatment (Feist & Longshaw 2006; Hedrick *et al.* 2008). *Myxobolus cerebralis* requires a 6-day treatment with fumagillin (3 mg kg^{-1} bodyweight per day) or with quaternary ammonium disinfectants (at 1,500 mg L^{-1}) (Feist & Longshaw 2006).

Table 44. Families and Species of Fish Susceptible to Muscle Encysting Myxosporean Pathogens

Host Family	Fish Species	Pathogen
Anguillidae	European eel (*Anguilla anguilla*)	*Myxobolus portucalensis*
Berycidae	Alfonsino (*Beryx splendens*)	*Kudoa thyrsites*
Carangidae	Yellowtail kingfish (*Seriola lalandi*)	*Kudoa neothunni, Unicapsula seriolae*
Carcharhinidae	Blacknose shark (*Carcharhinus acronotus*), blacktip shark (*C. limbatus*), blacktip reef shark (*C. melanopterus*), nervous shark (*C. cautus*), pigeye shark (*C. amboinensis*)	*Kudoa* spp., *Unicapsula* spp.
Chirocentridae	Wolf herring (*Chirocentrus dorab*)	*Unicapsula chirocentrusi*
Cichlidae	*Oreochromis* spp.	*Henneguya piaractus*
Clariidae	*Clarias* spp.	*Myxobolus* spp., *Thelohanellus* spp.
Clupeidae	Atlantic menhaden (*Brevoortia tyrannus*)	*Kudoa clupeidae*
Clupeidae	South American pilchard (*Sardinops sagax*), Bali sardinella (*Sardinella lemuru*), fringe-scale round herring (*Spratelloides robustus*)	*Kudoa thyrsites*
Coryphaenidae	Mahi mahi (*Coryphaena hippurus*)	*Kudoa thyrsites*
Cyprinidae	Bighead carp (*Hypophthamichthys nobilis*), silver carp (*Hypophthalmichthys molitrix*)	*Myxobolus drjagini, M. pavlovski*
Cyprinidae	Common and ornamental carp (*Cyprinus carpio*)	*Myxobolus* spp., *M. aeglefini, M. articus, M. artus, M. buckei, M. koi, M. portucalensis, M. pseudodispar, Sphaerospora molnari*, *S. chinensis*, *S. renicola, Thelohanellus hovorkai*, *T. nikolskii*
Cyprinidae	Carp bream (*Abramis brama*)	*Myxobolus bramae*, *M. hungaricus*, *M. macrocapsularis*, *M. parviformis*
Cyprinidae	Chub (*Squalius cephalus),* dace *Leuciscus leuciscus*), roach (*Rutilus rutilus*)	*Myxobolus intimus, M. pfeifferi*, *M. pseudodispar*
Cyprinidae	Ide (*Leuciscus idus*)	*Myxobolus carassii*
Cyprinidae	Goldfish (*Carassius auratus*)	*Myxobolus cultus, Sphaerospora molnari*, *S. chinensis*, *S. renicola*
Cyprinidae	Barbel (*Barbus barbus*)	*Myxobolus ergensi*
Dasyatidae	Bluespotted ribbontail ray (*Taeniura lymma*), bluespotted stingray (*Neotrygon kuhlii*), estuary stingray (*Dasyatis fluviorum*)	*Kudoa* spp., *Unicapsula* spp.
Distichodontidae	*Grass eater (Distichodus rostratus*), perch (*D. engycephalus*)	*Myxobolus* spp.
Engraulidae	Australian anchovy (*Engraulis australis*),	*Kudoa thyrsites*
Esocidae	Pike (*Esox lucius*)	*Henneguya creplini*
Gadidae	Haddock (*Melanogrammus aeglefinus*)	*Myxobolus* spp., *M. aeglefini*
Gempylidae	Barracouta (*Thyrsites atun*)	*Kudoa thyrsites*
Glaucostegidae	Giant shovelnose ray (*Glaucostegus typus*)	*Kudoa* spp., *Unicapsula* spp.
Ictaluridae	Channel catfish (*Ameiurus punctatus*)	*Henneguya ictaluri*, *H. exilis*
Latidae	Nile perch (*Lates niloticus*)	*Henneguya ghaffari*
Lutjanidae	Russell's grouper (*Lutjanus russellii*)	*Unicapsula andersenae*
Macrouridae	Giant grenadier (*Albatrossia pectoralis*)	*Unicapsula pacifica, U. schulmani*
Merlucciidae	Pacific hake (*Merluccius productus*)	*Kudoa paniformis*
Mullidae	Rosy goatfish (*Parupeneus rubescens*)	*Unicapsula galatea*
Nemipteridae	Monocle bream (*Scolopsis monogramma*), Japanese threadfin (*Nemipterus japonicus*)	*Unicapsula pyramidata*
Orectolobidae	Ornate wobbegong (*Orectolobus ornatus*), Western wobbegong (*Orectolobus hutchinsi*)	*Kudoa* spp., *Unicapsula* spp.
Paralichthyidae	Bastard halibut (*Paralichthys olivaceus*)	*Kudoa septempunctata*
Percidae	European perch (*Perca fluviatilis*)	*Henneguya creplini*
Pimelodidae	Long whiskered catfish (*Pimelodus maculatus*)	*Myxobolus* spp., *Thelohanellus* spp.
Pleuronectidae	Flounder (*Pleuronectes* spp.)	*Myxobolus aeglefini*
Pleuronectidae	Pacific halibut (*Hippoglossus stenolepis*)	*Unicapsula muscularis, U. stenolepis*
Pleuronectidae	English sole (*Parophrys vetulus*)	*Kudoa* sp.

Table 44 (Continued)

Host Family	Fish Species	Pathogen
Polynemidae	Fourfinger threadfin (*Eleutheronema tetradactylum*)	*Unicapsula andersenae*
Polynemidae	Giant African threadfin (*Polydactylus quadrifilis*)	*Unicapsula marquesi*
Rhinobatidae	Eastern shovelnose ray (*Aptychotrema rostrata*)	*Kudoa* spp., *Unicapsula* spp.
Salmonidae	Salmon (*Oncorhynchus* spp., *Salmo* spp., (*Salvelinus* spp.) whitefish (*Coregonus* spp.)	*Ceratomyxa shasta, Henneguya nuesslini, H. salmonicola, H. zschokkei, Kudoa thyrsites, Myxobolus arcticus, Parvicapsula pseudobranchicola*
Sciaenidae	Japanese meagre (*Argyrosomus japonicus*)	*Unicapsula andersenae*
Sciaenidae	Ganges jewfish (*Nibea coibor*)	*Unicapsula maxima*
Scophthalmidae	Turbot (*Scophthalmus maximus*)	*Enteromyxum scophthalmi*
Serranidae	Malabar grouper (*Epinephelus malabaricus*), convict grouper *(E. septemfasciatus*)	*Unicapsula seriolae*
Sillaginidae	Sand sillago (*Sillago ciliata*)	*Unicapsula andersenae*
Siluridae	*Clarias* spp., *Pimelodus maculatus*	*Henneguya* spp., *Myxobolus* spp., *Thelohanellus* spp.
Sparidae	Picarel (*Spicara smaris*), sand steenbras (*Lithognathus mormyrus*)	*Unicapsula pflugfelderi*
Sparidae	Gilthead snapper (*Sparus aurata*)	*Enteromyxum leei*
Sparidae	Surf bream (*Acanthopagrus australis*)	*Unicapsula andersenae*
Sphyraenidae	Red barracuda (*Sphyraena pinguis*)	*Kudoa thyrsites*
Sphyrnidae	Bonnethead shark (*Sphyrna tiburo*)	*Kudoa* spp., *Unicapsula* spp.
Tetraodontidae	Japanese puffer fish (*Takifugu rubripes*)	*Enteromyxum fugu*

Myxosporeans may be zoonotic, generally causing mild gastrointestinal infection, which may become serious in immunocompromised patients (Hessen & Zamzame 2004; De Velasco *et al.* 2008; Kaur 2014). Human infection with *Myxobolus plectroplites* is reported after consumption of cooked, previously frozen fillets of golden perch (*Macquaria ambigua*) (Percichthyidae) in Australia (Boreham *et al.* 1998). Infection with *Henneguya salminicola* and *Myxobolus* sp. is associated with consumption of salmon (Salmonidae), *Kudoa septempunctata* is associated with Bastard halibut (*Paralichthys olivaceous* (Paralichthyidae), while *K. hexapunctata* and *K. neothunni* infection is associated with consumption of tuna (*Thunnu*s spp.; Scombridae) in Columbia (McClelland *et al.* 1997; Moncada *et al.* 2001).

A wide range of susceptible elasmobranch and teleost fish, as well as invertebrate cogeneric or alternative intermediate hosts are present in New Zealand (NZOR 2017).

Alternative infection pathways exist. Myxosporeans may also be present in live ornamental fish imports into New Zealand (Hine & Diggles 2005).

47.2. Risk assessment

47.2.1. Entry assessment

Myxosporean infection associated with the muscle, skin, gill, connective and nervous tissues typically occurs with few external signs (Feist & Longshaw 2006), so infected fish may pass visual inspection. Myxosporean spores are unaffected by the death of the fish host and are resistant to the temperature extremes of commercial fish transport, storage and processing, including frozen storage (Hedrick *et al.* 2008). These cysts and spores are also resistant to chilling and freezing temperatures (Nehring *et al.* 2015).

The likelihood of entry is assessed as non-negligible.

47.2.2. Exposure assessment

Myxosporean pathogens that infect the musculature may only be discovered during fish processing, which would lead to rejection of the product as offal. Myxosporeans that infect the head, gills and skin of fish may not be noticed during processing and are also likely to be present in the skin, trimmings, wash water and offal discarded during commercial operations.

Little is known about the myxosporean life cycle and host specificity, but myxosporeans infecting fish are considered to be host-specific at the family level (Mackenzie & Kalavati 2014), but less specific for their invertebrate hosts (Feist &Longshaw 2006).

Suitable fish hosts may be infected by direct consumption of infected fish offal (Feist & Longshaw 2006). Myxosporeans are likely to remain viable in wastewater discharges from commercial fish processing and could infect alternative intermediate hosts, such as bryozoans or annelids, including *Tubifex* sp., while myxosporeans present in solid wastes may be redistributed to the aquatic environment by scavenging birds, such as seagulls (*Larus* spp.) (El-Matbouli & Hoffmann 1991).

A wide range of susceptible marine and freshwater fish may be affected, while alternative or cogeneric *Tubifex* species that could potentially function as intermediate hosts are present in New Zealand (NZOR 2017).

The likelihood of exposure to myxosporean pathogens is assessed as non-negligible.

47.2.3. Consequence assessment

Myxosporean infection of the muscle tissues appears to be of low pathogenicity, but the pseudocysts of *Kudoa* spp. and *Unicapsula* spp. taint the flesh post-mortem (myoliquifaction) upon cooking. This makes the flesh unsuitable for marketing and sale, resulting in the economic rejection of the commodity (Sitja-Bobadilla 2009; Sirin & Toksen 2014). Infection of the gonads may result in castration of the host, while infection of the gill filaments results in lowered respiratory efficiency (Feist & Longshaw 2006).

Myxosporeans are recognised as significant pathogens in marine aquaculture, including the developing industries for snapper (*Sparus aurata*) (Hine & Jones 1994; Sitja-Bobadilla 2009; Sirin & Toksen 2014) and yellowtail kingfish (*Seriola lalandi*) (NIWA 2014a). Snapper is one of New Zealand's most important coastal fish fisheries (MPI 2014), which was valued at $62 million in 2009 (Statistics New Zealand 2014). It also supports significant recreational and Maori fisheries (MPI 2014).

Freshwater aquaculture in New Zealand is highly dependent upon pure water supplies as little or no water treatment is generally practiced (Sim-Smith *et al.* 2014). The establishment of myxosporean pathogens in New Zealand could result in direct economic losses for the Chinook salmon (*Oncorhynchus tshawytsha*) farming industry in marine sea cages and freshwater farms, through loss of sales due to infected product and additional costs related to the water treatments required. This industry was valued at $63 million in export earnings in 2011 (Seafood New Zealand 2014). An outbreak could also affect the recreational freshwater trout and salmon fisheries, together with their tourism and other associated support industries. While the dollar value of New Zealand's freshwater fisheries is not fully known, the Lake Taupo trout fishery alone is

valued at $70–80 million per annum (Marsh & Mkwra 2013). Disease outbreaks may also incur significant social and environmental costs associated with other domestic freshwater fisheries, including eels and grass carp.

Eel farming (*Anguilla australis*, *A. dieffenbachii*) is not currently practiced in New Zealand, but the establishment of myxosporean pathogens would have major social consequences for recreational and traditional Maori eel fisheries (Fish & Game 2014; MPI 2014). Grass carp (*Ctenopharyngodon idella*) are farmed and used for weed control in New Zealand freshwater lakes and rivers (Hofstra 2014; NIWA 2014b), while infection of wild introduced cyprinids may represent a potential reservoir for infection of aquaculture species.

Zoonotic infection with myxosporeans appears essentially opportunistic, resulting in minor gastrointestinal infection (Moncada *et al.* 2001), but pathogenicity may be higher for immunocompromised patients (Hessen & Zamezame 2004). Human infection is associated with fresh chilled tuna (Scombridae), cooked olive flounder (Paralichthyidae), and cooked (previously frozen) fillets of golden perch (Percicthyidae) (McClelland *et al.* 1997; Boreham *et al.* 1998; Moncada *et al.* 2001; Kaur 2014). Myxosporeans are present in New Zealand fish (Hine *et al.* 2000) and no evidence exists to suggest that exotic myxozoan species are likely to be more pathogenic to humans than those already present in New Zealand.

The consequences of the establishment of myxosporean pathogens are assessed as non-negligible.

47.2.4. Risk estimation

Myxosporeans are generally of low pathogenicity, but can cause tissue spoilage (myoliquifaction) upon cooking resulting in potentially significant economic losses for New Zealand fisheries and aquaculture. The life cycle, mode of transmission and definitive host range remain uncertain for many species. As the entry, exposure and consequence assessments are non-negligible, under the procedures followed in this risk assessment, exotic myxosporean pathogens are assessed to be a risk in the commodity and risk management measures may be developed.

47.3. Risk management

Infection with exotic myxosporean pathogens is not an OIE-listed disease, so the *Aquatic Code* (OIE 2016), provides no specific guidance on importing eviscerated fish from infected countries, or specific processing requirements that would ensure the destruction of the pathogens. It is assumed that compliance with the approved processed states for OIE-listed diseases (Appendix 3) would also eliminate exotic myxosporean pathogens from the commodity and be a viable risk management option.

The classification of myxosporeans to genus or species level is contentious, and the host specificity of most species is uncertain, but no requirements for dedicated monitoring exist.. Exotic myxosporeans from genera *Ceratomyxa*, *Enteromyxum*, *Henneguya*, *Kudoa*, *Myxobolus*, *Parvicapsula*, *Sphaerospora*, *Thelohanellus* and *Unicapsula* are reported from over 40 families of wild and farmed marine and freshwater fish (Table 44), which may be present in the commodity. Species declaration should moderately reduce the pathogen load of these myxosporeans in the commodity and be a viable risk management option. Myxosporean incidence and pathogenicity is relatively low in wild-caught fish (Feist & Longshaw 2006). Restriction to wild-caught fish should moderately reduce pathogen load in the commodity and be a viable risk management option.

Myxosporean genera *Ceratomyxa, Enteromyxum, Henneguya, Kudoa, Myxobolus, Sphaerospora* and *Unicapsula* have a widespread distribution. Where country/zone freedom is approved through the MPI Country Approval Procedures, this option should substantially reduce the occurrence of these exotic myxosporean pathogens in the commodity. Declaration of country/zone freedom is a viable risk management option.

Myxosporean genera *Ceratomyxa, Enteromyxum, Henneguya, Kudoa, Myxobolus, Sphaerospora* and *Unicapsula* may be present in the musculature, head, gills and skin tissues, so removal of the gills, or the head and gills, should slightly reduce the pathogen load of these myxosporeans in the commodity. Further processing to the skin-off fillet state should moderately reduce the pathogen load of these myxosporeans in the commodity. Processing to the skin-off fillet state is a viable risk management option.

Myxosporeans are resistant to freezing and are not affected by frozen storage. Frozen storage is not considered a viable risk management option.

Myxosporean genera including *Ceratomyxa, Enteromyxum, Henneguya, Kudoa, Myxobolus, Sphaerospora* and *Unicapsula* are denatured by heat treatment, so cooking (to at least 75°C for 5 minutes) should eliminate them from the commodity. Heat treatment is a viable risk management option.

47.3.1. Risk management options

Myxosporean genera *Ceratomyxa, Enteromyxum, Henneguya, Kudoa, Myxobolus, Sphaerospora* and *Unicapsula* are reported from fish in families Anguillidae, Berycidae, Carangidae, Carcharhinidae, Chirocentridae, Cichlidae, Clariidae, Clupeidae, Coryphaenidae, Cyprinidae, Dasyatidae, Distichodontidae, Engraulidae, Esocidae, Gempylidae, Glaucostegidae, Ictaluridae, Latidae, Lutjanidae, Macrouridae, Merlucciidae, Mullidae, Nemipteridae, Orectolobidae, Paralichthyidae, Percidae, Pimelodidae, Pleuronectidae, Polynemidae, Rhinobatidae, Salmonidae, Sciaenidae, Scophthalmidae, Serranidae, Sillaginidae, Siluridae, Sparidae, Sphyraenidae, Sphyrnidae and Tetraodontidae (Table 44). These families are considered likely to be present in the commodity. Other families have not been associated with myxosporean genera *Ceratomyxa, Enteromyxum, Henneguya, Kudoa, Myxobolus, Sphaerospora* and *Unicapsula*. Therefore species declaration indicating the commodity is not originated from any of the above families should substantially reduce the occurrence of these myxosporean pathogens in the commodity.

For the commodities originated from families associated with myxosporean genera *Ceratomyxa, Enteromyxum, Henneguya, Kudoa, Myxobolus, Sphaerospora* and *Unicapsula*, one or a combination of the following additional options could also be considered to effectively manage the risk.

Where country/zone freedom from genera *Ceratomyxa, Enteromyxum, Henneguya, Kudoa, Myxobolus, Sphaerospora* and *Unicapsula* is accepted by MPI:

Option 1

Acceptance of country/zone freedom should substantially reduce the occurrence of these myxosporean pathogens, so the commodity may be imported without any further restrictions.

Where country/zone freedom from genera *Ceratomyxa, Enteromyxum, Henneguya, Kudoa, Myxobolus, Sphaerospora* and *Unicapsula* is not accepted by MPI or not available:

Option 2

Processing to a state consistent with the approved states for OIE-listed diseases (Appendix 3) should eliminate these pathogens. Where these provisions are met, the commodity could be imported without any further restrictions.

Where the imported commodity is not in a state consistent with the approved states for OIE-listed diseases (Appendix 3):

Option 3

Heat treatment (by cooking to at least 75°C for 5 minutes) should eliminate these myxosporean pathogens. When this provision is met, the commodity could be imported without any further restrictions; or,

Option 4

Further processing (to the skin-off fillet state) should moderately reduce the occurrence of these myxosporean pathogens. When this provision is met, the commodity could be imported without any further restrictions; or,

Option 5

Restriction of the commodity to wild-caught fish (not from aquaculture) should moderately reduce the occurrence of these myxosporean pathogens. When this provision is met, the commodity could be imported without any further restrictions.

47.4. References

Abe N, Maehara T (2013). Molecular characterization of kudoid parasites (Myxozoa: Multivalvulida) from somatic muscles of Pacific bluefin (*Thunnus orientalis*) and yellowfin (*T. albacores*) tuna. *Acta Parasitologica* 58(2): 226–230 [Online] Available from: http://www.doi: 10.2478/s11686-013-0130-1. Epub 2013 May 11. [Accessed 20 April 2018].

Alama-Bermejo G, Cuadrado M, Raga JA, Holzer AS (2009). Morphological and molecular redescription of the myxozoan *Unicapsula pflugfelderi* Schubert, Sprague & Reinboth 1975 from two teleost hosts in the Mediterranean. A review of the genus *Unicapsula* Davis 1924. *Journal of Fish Diseases* 32: 335–350.

Aseeva NL, Krasin VK (2001). On new records of the family Trilosporidae (Myxosporida: Multivalvulida) from fishes of Pacific Ocean. *Parazitologiya* 35:353–356.

Benz GW, Bullard SA (2004). Metazoan parasites and associates of chondrichthyans with emphasis on taxa harmful to captive hosts, pp 325–416 *In:* The Elasmobranch Husbandry Manual: Captive Care of Sharks, Rays and Their Relatives. Ohio Biological Survey, Inc. Columbus, Ohio, United States of America, 588 p.

Boreham RE, Hendrick S, O'Donoghue PJ, Stenzel DJ (1998). Incidental finding of *Myxobolus* spores (Protozoa: Myxozoa) in stool samples from patients with gastrointestinal symptoms. *Journal of Clinical Microbiology* 36: 3728–3730.

Borucinska JD, Frasca S (2002). Naturally occurring lesions and micro-organisms in two species of free-living sharks: the spiny dogfish, *Squalus acanthias* L., and the smooth dogfish, *Mustelus canis* (Mitchill), from the north-western Atlantic. *Journal of Fish Diseases*, 25: 287–298 [Online] Available from: http://www.doi:10.1046/j.1365-2761.2002.00373.x [Accessed 16October 2017].

Caira JN, Healy CJ, Jensen K (2012). An updated look at elasmobranchs as hosts of metazoan parasites, pp 548–650 *In:* Carrier JC, Musick JA, Heithaus MR (*Eds*.) Biology of sharks and their relative, 2nd Edition. CRC Press, 666 p.

Davis HS (1924). A new myxosporidian parasite, the cause of "wormy" halibut. Appendix VIII to the Report of United States Commission on Fisheries 1923 Doc. 957: 1–5.

Diebakate C, Fall M, Faye N, Toguebaye BS (1999). *Unicapsula marquesi* n. sp. (Myxosporea, Multivalvulida) a gill parasite of *Polydactylus quadrifilis* (Cuvier, 1829) (Pisces, Polynemidae) from Senegalese coast (West Africa). *Parasite* 6: 231–235.

Diggles BK (2011). Environmental Assessment Report: Disease Risk. Digsfish Services Report DF 11-02. Digsfish Services, Queensland, Australia, 70 p.

El-Matbouli M, Hoffmann RW (1991). Effects of freezing, aging and passage through the alimentary canal of predatory animals on the viability of *Myxobolus cerebralis* spores. *Journal of Aquatic Animal Health* 3: 260–262.

Eszterbauer E, Szekely C, Molnar K, Ferenc B (2000). Development of *Myxobolus bramae* (Myxosporea: Myxobolidae) in an oligochaete alternate host, *Tubifex tubifex. Journal of Fish Diseases* 23(1):19–25

Feist SW, Longshaw M (2006). Phylum Myxozoa, pp 230–296 *In:* Woo PTK (*Ed.*).Fish Diseases and Disorders, Volume 1. CABI International, Wallingford, United Kingdom, 791 p.

Fish and Game (2014). New Zealand Fish and Game Council [Online].Available from: http://www.fishandgame.org.nz/ [Accessed 24 September 2014].

Forro B, Eszterbauer E (2016). Correlation between host specificity and genetic diversity for the muscle-dwelling fish parasite *Myxobolus pseudodispar*: examples of myxozoan host-shift? *Folia Parasitica* 2: 63: 2016–2019.

Garner MM (2013). A retrospective study of disease in elasmobranchs. *Veterinary Pathology* 50(3): 377–389.

Gleeson RJ, Bennett MB, Adlard RD (2010). First taxonomic description of multivalvulidan myxosporean parasites from elasmobranchs: *Kudoa hemiscylli* n. sp. and *Kudoa carcharhini* n. sp. (Myxosporea: Multivalvulidae). *Parasitology* 137: 1885–1898.

Hedrick RP, McDowell TS, Mukkatira K, MacConnell E, Petri B (2008). Effects of freezing, drying, ultraviolet irradiation, chlorine and quaternary ammonium treatments on the infectivity of myxospores of *Myxobolus cerebralis* for *Tubifex tubifex. Journal of Aquatic Animal Health* 20: 116–125.

Hessen EM, Zamzame ML (2004). *Myxobolus* sp: a possible new opportunistic parasite in immunocompromised patients in Ismailia. *Journal of the Egyptian Society of Parasitology* 34(3): 925–930.

Hine PM, Jones JB (1994). *Bonamia* and other aquatic parasites of importance to New Zealand. *New Zealand Journal of Zoology* 21(1): 49–56.

Hine PM, Jones JB, Diggles BK (2000). A checklist of the parasites of New Zealand fishes, including previously unpublished records. *NIWA Technical Report 75*. National Institute of Marine and Freshwater Research, Wellington, New Zealand, 93 p.

Hine MP, Diggles BK (2005). Import risk analysis: Ornamental fish. Biosecurity New Zealand. Ministry of Agriculture and Forestry, Wellington, New Zealand. 270 p.

Hoffman GL (1990). *Myxobolus cerebralis*, a worldwide cause of salmonid whirling disease. *Journal of Aquatic Animal Health* 2: 30–37.

Hofstra DE (2014). Grass carp effectiveness and effects. Stage 2: Knowledge review. NIWA Client Report HAM2014-060, prepared for the Department of Conservation, Wellington, New Zealand, 140 p.

Iwashita Y, Kamijo Y, Nakahashi S, Shindo A, Yokoyama K, Yamamoto A, Yukinari O, Ishikura K, Fujioka M, Hatada T, Takeda T, Maruyama K, Hiroshi I (2013). Food poisoning associated with *Kudoa septempunctata. Journal of Emerging Medicine* 44: 943–945.

Kalavati C, Narasimhamurti CC (1985). Histopathological changes in the gills of *Channa punctatus* BL. infected with *Henneguya waltairensis. Archiv für Protistenkunde* 129:199–202 [Online] Available from: http://www. doi: 10.1016/S0003-9365(85)80023-3 [Accessed 15 August 2018].

Kasai A, Tsuduki H, Jimenez LA, Li YC, Tanaka S, Sato H (2017). Incidence of three *Kudoa* spp., K. *neothunni, K. hexapunctata*, and *K. thunni* (Myxosporea: Multivalvulida), in *Thunnus* tunas distributed in the western Pacific Ocean. *Parasitological Research* 116: 1137–1150 [Online] Available from: http://www.DOI 10.1007/s00436-016-5369-7 [Accessed 21 February 2018].

Kaur H (2014). Myxozoan Infestation in Freshwater Fishes in Wetlands and Aquaculture in Punjab (India). *Advances in Animal and Veterinary Sciences* 2 (9): 488-502 [Online] Available from: http://dx.doi.org/10.14737/journal.aavs/2014/2.9.488.502 [Accessed 31 July 2018].

Kostoingue B, Toguebaye BS (1994). Le genre *Myxobolus* (Myxozoa: Myxosporea) chez les poissons d'eau douce du Tchad avec la description de trois nouvelles especes. *Bulletin of the Scienific Institute of French West-Africa* 47:63–71.

Lester RJG (1982). *Unicapsula seriolae* n. sp. (Myxosporea, Multivalvulida) from Australian Yellowtail Kingfish *Seriola lalandi*. Journal of Protozoology 29: 584–587.

Lom J, Dykova I (2006). Myxozoan genera: definition and notes on taxonomy, lifecycle terminology and pathogenic species. *Folia Parasitolica* 53: 1–36.

Lom J, Dykova I (1992). Protozoan Parasites of Fishes. Developments in Aquaculture and Fisheries Science (Book 26). Elsevier Science, New York, United States of America, 315 p.

Marsh D, Mkwra L (2013). Review of freshwater non-market value studies. Department of Economics, University of Waikato, Hamilton, New Zealand, 144 p.

Martinez de Velasco G, Rodero M, Cuellar C, Chivato T, Mateos JM, Laguna R (2008). Skin prick test of *Kudoa* sp. antigens in patients with gastrointestinal and/or allergic symptoms related to fish ingestion. *Parasitological Research* 103: 713–715.

McClelland RS, Murphy DM, Cone DK (1997). Report of spores of *Henneguya salminicola* (Myxozoa) in human stool specimens: possible source of confusion with human spermatozoa. Journal of Clinical Microbiology 35: 2815–2818.

Mackenzie K, Kalavati C (2014). Myxosporean parasites of marine fishes: their distribution in the world's oceans. *Parasitology* 141: 1709–1717.

Miller TL, Adlard RD (2013). *Unicapsula* species (Myxosporea: Trilosporidae) of Australian marine fishes, including the description of *Unicapsula andersenae* n. sp. in five teleost families off Queensland, Australia. *Parasitology Research* 112(8): 2945–2957.

Moncada LI, Lopez MC, Murcia MI, Nicholis SLF, Guio OL, Corredor A (2001). *Myxobolus* sp., another opportunistic parasite in immunosuppressed patients. *Journal of Clinical Microbiology* 39(5): 1938–1940.

MPI (2014). Fisheries Assessment Plenary, May 2014: Stock assessments and stock status. Compiled by the Fisheries Science Group, Ministry for Primary Industries, Wellington, New Zealand, 1381 p.

Naidjenova NN, Zaika VE (1970). Three new genera of Myxosporidia - fish parasites from the Indian Ocean. *Zoologicheskyi Zhurnal* 49: 451–454.

Nehring BB, Schisler G, Chiaramonte L, Horton A, Poole B (2015). Assessment of the long-term viability of the myxospores of *Myxobolus cerebralis* as determined by production of the actinospores by *Tubifex tubifex. Journal of Aquatic Animal Health* 27(1): 50–56.

NIWA (2014a). Yellowtail kingfish [Online] Available from: https://www.niwa.co.nz/aquaculture/aquaculture-species/yellowtail-kingfish [Accessed 20 October 2015].

NIWA (2014b). Grass carp *Ctenopharyngodon idella* (Valenciennes, 1844) [Online] Available from: https://www.niwa.co.nz/freshwater-and-estuaries/nzffd/NIWA-fish-atlas/fish-species/grass_carp [Accessed 12 February 2015].

NZOR (2017). Tubifex. New Zealand Organisms Register [Online] Available from: http://www.nzor.org.nz/names/b7dde507-5f19-408c-bebf-74348f9ea93c [Accessed 22 March 2017].

OIE (2016). Aquatic Animal Health Code 19th Edition. World Organisation for Animal Health, Paris, France [Online] Available from: http://www.oie.int/en/international-standard-setting/aquatic-code/access-online/ [Accessed 21 March 2017].

Rangel LF, Rocha S, Casal G, Castro R, Severino R, Azevedo C, Cavaleiro F, Santos MJ (2017). Life cycle inference and phylogeny of *Ortholinea labracis* n. sp. (Myxosporea: Ortholineidae), a parasite of the European seabass *Dicentrarchus labrax* (Teleostei: Moronidae), in a Portuguese fish farm. *Journal of Fish Diseases* 40: 243–62.

Sarkar NK (1999). Some new Myxosporidia (Myxozoa: Myxosporea) of the genera *Myxobolus* Butschli, 1882 *Unicapsula* Davis, 1942 *Kudoa* Meglitsch, 1947 *Ortholinea* Shulman, 1962 and *Neoparvicapsula* Gaevskaya, Kovaleva and Shulman, 1982. *Proceedings of the Zoological Society of Calcutta* 52: 38–48.

Sarkar NK (1984). A species of *Unicapsula* Davis, 1924 (Myxozoa) infecting a fish of the southern coast of India. *International Journal of the Academy of Ichthyology*, Modinagar 5:187–189.

Seafood New Zealand (2014). Seafood New Zealand: Key facts for our Industry. Seafood New Zealand, Wellington, New Zealand, 6 p.

Sim-Smith C, Faire S, Lees A (2014). Managing biosecurity risk for business benefit. Report prepared for the Ministry for Primary Industries by Coast and Catchment Ltd, Auckland, New Zealand, 214 p.

Sirin C, Toksen E (2014). Several internal myxozoan parasites on cultured sea bass, *Dicentrarchus labrax* and gilthead sea bream, *Sparus aurata* in the Mediterranean region. *Journal of Science and Technology* 2(1): 65–72.

Sitja-Bobadilla A (2009). Can myxosporean parasites compromise fish and amphibian reproduction? *Proceedings of the Royal Society B* (276) 2861–2870.

Statistics New Zealand (2014). New Zealand's commercial fish resource, 2009: snapper [Online]. Available from: http://www.stats.govt.nz/browse_for_stats/environment/environmental-economic-accounts/fish-monetary-stock-account-1996-2009/results.aspx [Accessed 12 September 2014].

Yokoyama H, Abe N, Maehara T, Suzuki J (2014). First record of *Unicapsula seriolae* (Myxozoa: Multivalvulida) from the muscle of Malabar grouper *Epinephelus malabaricus* in Japan.. *Parasitology International* 63(3): 561–563. [Online] Available from www. doi: 10.1016/j.parint.2014.03.002 [Accessed 27 June 2017].

Yokoyama H, Liyanage YS, Sugai A, Wakabayashi H (1998). Hemorrhagic thelohanellosis of color carp caused by *Thelohanellus hovorkai* (Myxozoa:Myxosporea). *Fish Pathology* 33: 85-89.

48. *Anguillicola crassus*

48.1. Hazard identification

48.1.1. Aetiological agent

Anguillicola crassus, the Asian eel swimbladder worm is classified within family Dracunculidae of the Phylum Nematoda (Molnar *et al.* 2006). It was first described in the Japanese eel (*Anguilla japonica*) but has subsequently spread to Europe and the United Kingdom. The increasing consumption of raw fish has highlighted the importance of nematodes as zoonotic pathogens (Molnar *et al.* 2006).

48.1.2. OIE status

Infection with pathogenic nematodes is not listed by the OIE (OIE 2016).

48.1.3. New Zealand status

Pathogenic nematodes are reported from New Zealand fish including the related nematode *Anguillicola australiensis* (Hine *et al.* 2000; Johnston 2008), but *A. crassus* is not reported from New Zealand fish. Infection with nematode pathogens is not a notifiable disease in New Zealand (Anon. 2016).

48.1.4. Epidemiology

The Asian eel nematode *Anguillicola crassus* matures in the swim bladder of eels (*Anguilla* spp.). It was originally reported from the Japanese eel (*Anguilla japonica*), but has been spread across four continents (Africa, America, Asia and Europe), largely through the live eel trade (Szekely 1994).

Nematode fish pathogens have a complex life cycle which involves at least one invertebrate intermediate host. A wide range of other marine and freshwater fish may function as intermediate, paratenic (transport, or alternative) and final hosts (Molnar *et al.* 2006).

Unlike most nematodes infecting fish in the adult stage, *A. crassus* is pathogenic to its final host, with infection causing reduced swim bladder performance and a reduction in stamina and swimming ability. Eels undergo extensive migrations to offshore spawning grounds and *A. crassus* has been implicated in population-level abundance decreases in *A. anguilla* in Europe and *A. rostrata* in the United States (Lefebre *et al.* 2012).

The host reaction may be stronger when a new (naïve) host species is infected (Molnar *et al.* 2006) and *Anguillicola crassus* is more pathogenic to endemic American eels (*A. rostrata*) and European eels (*A. anguilla*), than to the Asian eel (*A. japonica*) (Szekely 1994; Heitlinger *et al.* 2013). *Anguillicola crassus* has even displaced the endemic Australasian eel nematode (*A. australiensis*), following the accidental introduction of both species in Italian eel farming (Dangel *et al*. 2015). *Anguillicola crassus* is reported from over 30 other fish species (Table 45), which function as paratenic hosts (Szekely 1994).

The first three intermediate larval stages of *A. crassus* occur in a range of invertebrates in marine and fresh waters, including crustaceans (copepods (*Cyclops* spp.) and ostracods) and molluscs

Table 45. Families and Species of Fish Susceptible to Larval *Anguillicola crassus*

Host Family	Host Species
Anguillidae	American eel (*Anguilla rostrata*), European eel *(A. anguilla*), Japanese eel (*Anguilla japonica*)
Centrarchidae	Pumpkinseed (L*epomis gibbosus*)
Cichlidae	Nile tilapia (*Oreochromis niloticus*)
Cyprinidae	Asp (*Leuciscus aspius*), bleak (*Alburnus alburnus*), European bitterling (*Rhodeus amarus*), freshwater bream (*Abramis brama*), common and ornamental carp (*Cyprinus carpio*), Prussian carp (*Carassius gibelio*), gudgeon (*Gobio gobio*),), rain bleak (*Leucaspius delineatus*), roach (*Rutilus rutilus*), rudd (*Scardinius erythrophthalmus*), tench (*Tinca tinca*), white bream (*Blicca bjornica*)
Esocidae	Pike (*Esox lucius*)
Gobiidae	Monkey goby (*Neogobius fluviatilis*)
Osmeridae	European smelt (*Osmerus eperlanus*)
Percidae	European perch (*Perca fluvitilus*), ruffe (*Gymnocephalus cernua*), pike perch (*Sander lucioperca*)
Siluridae	European catfish (*Silurus glanis*)

(*Lymnaea* spp.). It is also reported from insects including the Megaloptera (*Sialis lutaria*), Odonata (*Coenagrion puella, Sympetrum sanguinum*) and Trichoptera (*Oligotricha striata*). *Anguillicola crassus* also infects amphibians (*Triturus vulgaris, Bombina bombina*) (Thomas & Ollevier 1992; Moravec & Skorikova 1998).

Initial infection in fish results from the consumption of a crustacean, annelid, coelenterate, fish or mollusc infected with a third-stage nematode larvae. This migrates from the gut lumen, to encyst in the muscle, or connective tissues, of its fish intermediate host (Yanong 2011).

When an infected fish is consumed by another fish, the larvae excyst in the gut lumen of the new host and migrate to the muscle or connective tissues of their new host. The migration process involves dissolution of host tissue, which releases immunogenic molecules into the fish hosts. These chemicals, which are unaffected by storage and processing, cause severe allergic reactions in some people when the infected fish product is consumed (Molnar *et al.* 2006).

The life cycle is completed when the fish is consumed by the definitive eel host (*Anguilla* spp.) (Yanong 2011). The larvae migrate from the gut lumen, to the swim bladder and develop into the adult stage, which then releases eggs into the lumen of the swim bladder. *Anguillicola crassus* is pathogenic to its final host, causing tissue damage (anisakidosis) to the epidermis of the swim bladder (Molnar *et al*. 2006). This reduces its use as a hydrostatic organ and consequently decreases the host ability to withstand high water temperatures and reduces swimming ability required to successfully complete the extensive offshore migrations of the eel life cycle (Kennedy 2007).

Nematode larvae remain inactive in the fish tissues, throughout the body of their fish host (Johnston 2008). They survive throughout the lifetime of their host and their numbers can accumulate as the fish gets older (FAO 2014).

Nematodes survive in dead fish tissue stored at room temperature, or as chilled product, for several days (Molnar *et al.* 2006). Fish, particularly when sourced from wild species, are commonly stored as chilled product, where evisceration and processing occur later. Larval nematodes present in the gut or connective tissues can migrate to the muscle tissues after the death of their host. These larvae are colourless, cryptic and are unlikely to be noticed even by extensive visual examination or "candling" of the product (Johnston 2008). The "worm tracks" caused by larval migration (larval migrans) through the muscles of the intermediate host may predispose the host to secondary

infection (Yanong 2011), as well as reducing the product value of the commodity (Molnar *et al.* 2006).

Processing including cold smoking, salting or pickling is unlikely to deactivate nematode larvae. *Anguillicola crassus* larvae remain viable in fish tissues after salting, marinating (in lemon juice, soy sauce or coconut milk), or after cold smoke treatments. They may survive 28 days in 80° salinometer brine (21% salt by weight) (FAO 2014). They survive in the storage tanks and handling equipment used in eel farming and transport (Kennedy 2007).

Adult and larvae of *A. crassus* are destroyed by hot smoke processing (to 80°C for 10 minutes), or by heat treatment (to 60°C for 10 minutes) (FAO 2014). Frozen storage (to at least -20°C for 60 hours) is also an effective treatment for adult and larval nematodes present in eviscerated fish product (Johnston 2008; EFSA 2010; FDA 2014). while all nematodes in whole sockeye salmon were shown to be killed by freezing (-35°C for 24 hours) (Howgate 1998).

No effective chemical treatments exist for fish pathogenic nematodes (Molnar *et al.* 2006).

Anguillicola crassus may be zoonotic, where humans can act as paratenic hosts. Infection is usually associated with consumption of raw or partly processed fish, causing larval migrans, anisakiasis or allergic reactions in humans (Fratamico *et al.* 2005; FAO 2014; Noga 2014).

Potential intermediate host species in New Zealand exist for *A. crassus*, including copepods (*Cyclops* spp.), ostracods and molluscs (*Lymnaea* sp.). A wide range of fish can function as paratenic hosts (Moravec 1996; Wurtz *et al.* 1998). Introduced cyprinids (Paulin *et al.* 2001) may also function as paratenic hosts in New Zealand (Szekely 1994).

48.2. Risk assessment

48.2.1. Entry assessment

Infective larvae may be present in the skin, musculature, connective tissue, gills and eyes of wild and farmed marine and freshwater fish, usually with no external signs of infection (Molnar *et al.* 2006). These can migrate into the musculature following the death of the host (Molnar *et al.* 2006; Johnston 2008). As the swim bladder may be incompletely removed upon evisceration, adult *A. crassus* present in remaining infected swimbladder tissue can remain viable in the commodity (Johnston 2008).

The likelihood of entry of *A. crassus* in the commodity is assessed as non-negligible.

48.2.2. Exposure assessment

The life cycle of *A. crassus* is complex, involving fish as intermediate and final hosts (Molnar *et al.* 2006). Both adults and larvae of *Anguillicola crassus* may remain viable in fresh-chilled eviscerated fish (Molnar *et al.* 2006). To establish an infection in New Zealand, sufficient infected product would have to become available for consumption by susceptible primary and intermediate hosts in sufficient quantity and duration (Kahn *et al.* 1999).

Anguillicola crassus is unaffected by the low temperatures (4°C) commonly associated with chilled fish storage and processing (Molnar *et al.* 2006), but nematode larvae and eggs are effectively inactivated by freezing (-20°C for 60 h) (FAO 2014), so exposure through frozen eviscerated fish is assessed as negligible.

Anguillicola crassus can remain viable in the wastewater discarded during fish processing, as well as in the skeletal muscle trimmings and offal discarded after processing (FAO 2014), or in fresh-chilled fish following cold-smoking, salting or pickling treatments (FDA 2014). *Anguilla crassus* has a wide host range and many potential intermediate and paratenic hosts including cyprinid and percid fish are present in New Zealand (Paulin *et al.* 2001)

The likelihood of exposure of *A. crassus* through the commodity is assessed as non-negligible.

48.2.3. Consequence assessment

Anguillicola crassus is pathogenic to a wide variety of fish, including eels, and causes economic disease (Szekely 1994; Molnar *et al.* 2006).

Anguillicola crassus is significantly more pathogenic to eels than the endemic *A. novaezelandiae*, and has replaced *A. novaezelandiae* in Italian eel population when both species were introduced following transfers of live eels in European eel aquaculture (Dangel *et al*. 2015). Introduction of *A. crassus* into New Zealand may adversely affect an already stressed endemic eel population (Jellyman 2012). The New Zealand eels (*A. australis, A. dieffenbachii* and *A. reinhardtii*) are iconic and highly valued species, supporting a commercial eel fishery valued at $4.9 million (Jellyman 2012). Eels are also important for non-commercial subsistence eel fishers and are considered a taonga by Maori (Parliamentary Commissioner for the Environment 2013).

Anguillicola crassus potentially affects a wide number of freshwater fish hosts, including several species caught for human consumption in New Zealand such as rudd (*Scardinius erythrophthalmus*) and tench (*Tinca tinca*) (NIWA 2017a, 2017b).

Nematode infections can be zoonotic (Fratamico *et al.* 2005; FAO 2014; Noga 2014). *Anguillicola crassus* causes larval migrans and allergic reactions in humans that act as paratenic hosts (Noga 2014). The increased presence of nematode zoonotic pathogens may have human health consequences, particularly given the increasing market for fresh-chilled fish products, such as sushi and sashimi (Molnar *et al.* 2006).

The introduction of *A. crassus* would have significant social, economic and human health consequences. The consequence assessment is assessed to be non-negligible.

48.2.4. Risk estimation

Since the entry, exposure assessments and consequence assessments are non-negligible, the risk is estimated to be non-negligible. Therefore, *A. crassus* is assessed to be a risk in the commodity and risk management measures may be justified.

48.3. Risk management

Infection with *Anguillicola crassus* is not an OIE-listed disease, so the *Aquatic Code* (OIE 2016) provides no specific guidance on mitigation measures for importing eviscerated fish from infected countries, or specific processing requirements that would ensure the destruction of the pathogen. It is assumed that compliance with the approved processed states for OIE-listed diseases (Appendix 3) would also eliminate *A. crassus* from the commodity and be a viable risk management option.

Anguillicola crassus is reported from nine families of fish (Table 45), which may be present in the commodity. Species declaration should substantially reduce the pathogen load of *A. crassus* in the commodity and be a viable risk management option.

Anguillicola crassus occurs in wild and farmed fish, including paratenic hosts Szekely 1994; Molnar *et al*. 2006). Restriction of the commodity to wild-caught fish (not from aquaculture) would have little, or no effect on pathogen load and is not a viable risk management option.

Anguillicola crassus is widely reported from African, American, Asian and European waters. Where country/zone freedom is approved through the MPI Country Approval Procedures, this option should substantially reduce the pathogen load of *A. crassus* in the commodity. Approval of a declaration of country/zone freedom by MPI may be a viable risk management option.

Anguillicola crassus may be present in the swimbladder and skeletal musculature of eels and in the skin, dermal musculature, gills and eyes of other paratenic hosts (Table 45). Larval *A. crassus* are likely to migrate to the musculature after the death of the host (Molnar *et al.* 2006). Removal of the gills, or the head and gills, would be likely to slightly reduce pathogen load in the commodity. As the larvae may remain viable in the musculature, additional processing to the skin-off fillet state would only slightly reduce the pathogen load, but is not likely to eliminate *A. crassus* from the commodity. Processing to the skin-off fillet state is not a viable risk management option.

Anguillicola crassus is zoonotic, causing larval migrans and allergic reactions in humans that act as paratenic hosts. As it is likely to remain viable in fresh chilled skin-off fillets (such as sushi and sashimi), further treatment to ensure denaturation may be necessary (Molnar *et al* 2006; Noga 2014). Nematodes are denatured by freezing (to below -20°C for at least 60 hours) (FAO 2014), so frozen storage would eliminate *A. crassus* from the commodity. Frozen storage is a viable risk management option.

Nematode larvae are also destroyed by high-temperature heat treatments, including hot smoke processing (where temperatures exceed 80°C for 10 minutes), or by cooking (to at least 60°C for 10 minutes) (FAO 2014). These heat treatments would eliminate *A. crassus* from the commodity and are viable risk management options.

48.3.1. Risk management options

Anguillicola crassus is reported from fish in families Anguillidae, Centrarchidae, Cichlidae, Cyprinidae, Esocidae, Gobiidae, Osmeridae, Percidae and Siluridae (Table 45), which are considered likely to be present in the commodity. Other families have not been associated with *A. crassus*. Therefore species declaration indicating the commodity is not originated from any of the above families should substantially reduce the occurrence of *A. crassus* in the commodity.

For the commodities originated from families associated with *A. crassus,* one or a combination of the following options could also be considered to effectively manage the risk.

Where country/zone freedom from *A. crassus* is accepted by MPI:

Option 1

- Acceptance of country/zone freedom by MPI should substantially reduce the occurrence of *A. crassus*, so the commodity may be imported without any further restrictions.

Where declaration of country/zone freedom from *A. crassus* is not accepted by MPI or not available:

Option 2

Processing to a state consistent with the approved states for OIE-listed diseases (Appendix 3) should eliminate *A. crassus*. When these provisions are met, the commodity could be imported without any further restrictions.

Where the imported commodity is not in a state consistent with the approved states for OIE-listed diseases (Appendix 3):

Option 3

Frozen storage (to below -20°C for 60 hours) should eliminate *A. crassus*. When this provision is met, the commodity could be imported without any further restrictions; or,

Option 4

Heat treatment (by cooking to at least 60°C for 10 minutes); or by hot smoke processing (to at least 80°C for 10 minutes) should eliminate *A. crassus*. When this provision is met, the commodity could be imported without any further restrictions.

48.4. References

Anonymous (2016). Biosecurity (Notifiable Organisms) Order 2016. NZLR 2016/73. New Zealand Government, Wellington, 13 p.

Dangel KC, Keppel M, De Grabner TTY, Sures B (2015). Competing invaders: Performance of two Anguillicola species in Lake Bracciano. *International Journal for Parasitology: Parasites and Wildlife* 4(1): 119–124 [Online] Available from: http://www. doi: 10.1016/j.ijppaw.2014.12.010

EFSA (2010). Treatment to kill viable parasites in fishery products for human consumption (EU 2976/2011). *Official Journal of the European Union* 327/39, 3p.

FAO (2014). Round worms in fish. Torrey Research Station. FAO Corporate Document Repository [Online] Available from: http://www.fao.org/wairdocs/tan/x5951e/x5951e01.htm [Accessed 5 December 2014].

FDA (2014). Parasites. pp 91–98. *In:* Fish and Fishery Products Hazards and Controls Guidance, 4th edition. U.S. Department of Health and Human Services, Food and Drug Administration, Washington D.C. United States of America, 468 p.

Fratamico PM, Bhunia AK, Smith JL (2005). Foodborne pathogens: Microbiology and molecular biology. Horizon Scientific, New York, United States of America, 453 p.

Heitlinger E, Bridgett S, Montazam A, Taraschewski H, Blaxter M (2013). The transcriptome of the invasive eel swimbladder nematode parasite Anguillicola crassus. *BMC Genomics* 14(87) [Online] Available from www. **DOI:** 10.1186/1471-2164-14-87 [Accessed 11 July 2017].

Hine PM, Jones JB, Diggles BK (2000). A checklist of the parasites of New Zealand fishes, including previously unpublished records. *NIWA Technical Report* 75. National Institute of Water and Atmospheric Research Ltd., Auckland, New Zealand, 93 p.

Howgate P (1998). Freezing to kill nematode parasites in fish products: Implications for HACCP. May 1998 [Online] Available from: http:www.seafood.ucdavis.edu/Pubs/nematodes.htm [Accessed 12 January 2015].

Jellyman D (2012). The status of longfin eels in New Zealand – an overview of stocks and harvest. NIWA Client Report prepared for the Parliamentary Commissioner for the Environment. National Institute of Water and Atmospheric Research, Christchurch, New Zealand. 78 p.

Johnston C (2008). Import Risk Analysis: Frozen skinless and boneless fillet meat of *Oreochromis* spp. fish from Brazil and China for human consumption. Biosecurity New Zealand, Ministry of Agriculture and Forestry, Wellington, New Zealand, 96 p.

Kahn SA, Beers PT, Findlay VL, Peebles IR, Durham PJ, Wilson DW, Gerrity SE (1999). Import risk analysis on non-viable salmonids and non-salmonid marine fish. Australian Quarantine and Inspection Service, Department of Agriculture, Fisheries and Forestry, Canberra, Australia, 409 p.

Lefebre F, Wielgoss S, Nagasawa K, Moravec F (2012). On the origin of *Anguillicoides crassus*, the invasive nematode of anguillid eels. *Aquatic Invasions* 7(4): 443–453.

Molnar K, Buchmann K, Szekely C (2006). Phylum Nematoda. pp 417–443. *In:* Woo PTK (*Ed*.) Fish Diseases and Disorders. Volume 1: Protozoan and Metazoan Infections (Second Edition). CABI International, Wallingford, United Kingdom, 791 p.

Moravec F (1996). Aquatic invertebrates (snails) as new paratenic hosts of *Anguillicola crassus* (Nematoda: Dracunculoidea) and the role of paratenic hosts in the life cycle of this parasite. *Diseases of Aquatic Organisms* 27: 237–239.

Moravec F, Skorikova B (1998). Amphibians and larvae of aquatic insects as new paratenic hosts of *Anguillicola crassus* (Nematoda: Dracunculoidea), a swimbladder parasite of eels. *Diseases of Aquatic Organisms* 34: 217–222.

NIWA (2017a). Rudd. Information guides and fact sheets. National Institute for Water and Atmospheric Research, Auckland, New Zealand [Online] Available from https://www.niwa.co.nz/freshwater-and-estuaries/nzffd/NIWA-fish-atlas/fish-species/rudd [Accessed 12 July 2017].

NIWA (2017b). Tench. Information guides and fact sheets. National Institute for Water and Atmospheric Research, Auckland, New Zealand [Online] Available from https://www.niwa.co.nz/freshwater-and-estuaries/nzffd/NIWA-fish-atlas/fish-species/tench [Accessed 12 July 2017].

Noga EJ (2014). Fish disease: Diagnosis and treatment. John Wiley, New York, United States of America, 536 p.

OIE (2016). Aquatic Animal Health Code, 19th Edition. World Organization for Animal Health. Paris, France [Online] Available from: http://www.oie.int/international-standard-setting/aquatic-code/access-online/ [Accessed 15 March 2017].

Parliamentary Commissioner for the Environment (2013). On a path to extinction? An investigation into the status and management of the longfin eel. Parliamentary Commissioner for the Environment, Wellington, New Zealand, 95 p.

Paulin C, Stewart A, Roberts C, McMillan P (2001). New Zealand Fish: A complete guide. Te Papa Press, Wellington, New Zealand, 279 p.

Szekely C (1994). Paratenic hosts for the parasitic nematode *Anguillicola crassus* in Lake Balaton, Hungary. *Diseases of Aquatic Organisms* 18: 11–20.

Thomas K, Ollevier F (1992). Paratenic hosts of the swimbladder nematode *Anguillicola crassus. Diseases of Aquatic Organisms* 13: 165–174.

Wurtz J, Knopf K, Taraschewski H (1998). Distribution and prevalence of *Anguillicola crassus* (Nematoda) in eels *Anguilla anguilla* of the rivers Rhine and Naab, Germany. *Diseases of Aquatic Organisms* 32: 137–143.

Yanong RPE (2011). Nematode (roundworm) infections in fish. Circular 91, Fisheries and Aquatic Services Department, UF/IFAS Extension. Institute of Food and Aquatic Sciences, Gainesville, Florida, United States of America, 8 pp [Online] Available from: https://edis.ifas.ufl.edu/fa091 [Accessed 12 February 2015].

49. *Gyrodactylus salaris* and related monogeneans

49.1. Hazard identification

49.1.1. Aetiological agent

Monogenean flatworms are classified in subclass Monogenea, within Class Trematoda of the Phylum Platyhelminthes. Monogenean pathogens of fish show a high degree of host specificity and specialisation. There may be over 25,000 pathogenic monogenean species, of which fewer than 3,000 species have been described (Whittington 1998).

The family Dactylogyridae includes several species complexes affecting marine fish and freshwater catfish (Ictaluridae, Siluridae, and Pangasiidae) in neotropical waters (Mendoza-Palmero *et al.* 2015). The *Benedenia seriolae* species complex affects marine fish of the family Carangidae in diverse geographical locations, including New Zealand (Hine *et al.* 2000; Sepulveda & Gonzalez 2014). The *Neobenedenia melleni* species complex also has a wide host range and distribution (Whittington 2004, 2011). The salmonid ectoparasites *Gyrodactylus salaris* and *G. thymalli* represent a species complex consisting of at least six clades (Gilmore *et al.* 2012; Fromm *et al.* 2014).

These include the OIE-listed *Gyrodactylus salaris*, which causes economic disease in fish (Buchmann & Bresciani 2006). These monogenean species complexes have important implications for species definitions, host specificity and geographical distribution (Meinila *et al.* 2004), but no consensus has yet been reached on their implications for disease management and trade (Fromm *et al.* 2014).

49.1.2. OIE status

Infection with *Gyrodactylus salaris* is listed by the OIE (OIE 2016a).

49.1.3. New Zealand status

Several species of monogenean ectoparasites are reported from New Zealand, including *Gyrodactylus* sp. and *B. seriolae* (Boustead 1982; Hine *et al*. 2000). The identification of a species complex for *B. seriolae* (Sepulveda & Gonzalez 2014) indicates more virulent strains exist overseas. *Gyrodactylus salaris* has not been reported and is considered exotic (Johnston 2008a, 2008b). It is a notifiable organism in New Zealand (Anon. 2016).

49.1.4. Epidemiology

Ectoparasitic monogeneans are generally host specific for a particular microhabitat (such as the fins, skin and gill tissues). They may occur within this microhabitat across one, or a range of different, but usually related, fish species (Buchmann & Bresciani 2006).

While widely reported, endoparasitic monogeneans are rarely pathogenic in wild fish stocks, as the incidence per fish is generally low (Buchmann & Bresciani 2006). Monogeneans are generally small, often cryptic or transparent when alive and may not be noticed on visual examination (Hutson 2010).

Monogeneans may cause economic disease in farmed fish (Buchmann & Bresciani 2006), so those reasonably expected to potentially infect farmed marine and freshwater fish in New Zealand are considered further.

The monogeneans likely to be associated with fish of families Anguillidae, Carangidae, Cyprinidae, Salmonidae, Serranidae and Sparidae, together with other fish families where these monogeneans are reported, are given in Table 46 (Whittington 1998; Bullard *et al.* 2000; Hansen *et al.* 2003; Montero *et al*. 2003; Whittington 2004; Buchmann & Bresciani 2006; Johnston 2008a; Sitjà-Bobadilla & Alvarez-Pellitero 2009; Antonelli *et al.* 2010; Paladini *et al.* 2011; Woo & Buchmann 2012; Chaudhary *et al.* 2013; Faruque *et al.* 2013; Zahradníčková *et al*. 2014; Mahmoud *et al.* 2015).

Monogenean infection causes sequential loss of epidermal integrity, associated with the hooks and suckers used for attachment and by feeding activity (Whittington 2011). At each attachment of these mobile parasites, their marginal hooklets cause puncture wounds to the host epithelial cells (Buchmann & Bresciani 2006). Infestation rates can reach thousands of individuals per host, facilitating secondary infection and causing significant mortality results through loss of osmoregulatory integrity (Buchmann & Bresciani 2006).

Monogeneans present on the gills can reduce gill efficiency and cause hyperplasia (clubbing and fusion of gill filaments), resulting in anaemia (Buchmann & Bresciani 2006).

Mortality may be high following introduction into naïve populations. *Gyrodactylus salaris* present on Atlantic salmon (*Salmo salar*) caused high mortality in the resident population following their introduction into Norwegian and Scottish rivers (Buchmann & Bresciani 2006).

The life cycle is simple and direct although transmission and distribution patterns may be influenced by predator-prey relationships between host species (Strona 2014). Monogeneans are hermaphroditic and most are ovoviviparous (Buchmann & Bresciani 2006), but some, including *Gyrodactylus* spp., are viviparous (producing live offspring), each of which contains a maturing embryo (Buchmann & Bresciani 2006). These life cycles provide for rapid growth in infection, where fecundity may reach 100 eggs per hour per adult (Whittington 2011), although the actual reproduction rate is temperature related (Ernst *et al*. 2005). Monogenean eggs are distributed by water currents and may be concentrated by physical structures, including nets and cages (Whittington 2011). The eggs have a sticky pad to facilitate attachment to the substrate and their mucous coating is effectively impervious to chemical treatment and to environmental extremes (Reed *et al*. 2009). Hatching generally occurs within hours and releases an infective miracidium larval stage, which generally locates a host in 2–4 hours. The miracidium then migrates to its chosen location on the skin or gill filaments (Reed *et al*. 2009). Sexual maturity occurs in 3–8 days (Reed *et al.* 2009), but development at each life cycle stage can be delayed for up to 6 months, under adverse conditions. The drivers for host location, as well as the triggers for hatching and subsequent development, are complex and poorly understood (Buchmann & Bresciani 2006; Umeda *et al*. 2006; Reed *et al*. 2009).

Monogeneans present on the skin generally detach from their hosts soon after host death and are unlikely to remain viable for more than 48 hours after their removal from water (Kahn *et al*. 1999). Monogeneans on the gills are protected from desiccation by the gill operculum and host mucous secretions. They continue to feed and reproduce for up to 15 days at 3°C, although viability is reduced to 3 days at 18°C (Olstad *et al.* 2006).

Table 46. Families and Species of Fish Susceptible to *Gyrodactylus salaris* and Related Monogeneans

Family	Fish Host	Pathogen
Acanthuridae	Doctorfish (*Acanthurus chirurgus*)	*Neobenedenia melleni*
Anguillidae	American eel (*Anguilla rostrata*), Japanese eel (*A. japonicus*, *Anguilla* spp.)	*Pseudodactylogyrus anguillae*, *P. bini*, *Gyrodactylus anguillae*
Carangidae	Creville jack (*Caranx hippos*), horse-eye jack (*C. latus*), yellowtail kingfish (*Seriola quinqueradiata*), greater amberjack (*S. dumerili*), yellowtail amberjack (*S. lalandi*), longfin yellowtail (*S. rivoliana*), white trevally (*Pseudocaranx dentex*)	*Allencotyla mcintoshi, Allopyragraphorus hippos*, *Cemocotyle noveboracensis*, *Heteraxine hetercerca, Neobenedenia melleni, Protomicrocotyle mirabilis*
Catostomidae	White sucker (*Catostomus commersonii*)	*Neobenedenia melleni*
Channidae	Spotted snakehead (*Channa punctata*)	*Dactylogyrus* sp., *Gyrodactylus* sp.
Cichlidae	Blue tilapia (*Oreochromis aureus*), Mozambique tilapia (*O. mossambicus*), Nile tilapia (*O. niloticus*), redburst tilapia (*Coptodon rendalli*), *Oreochromis* spp.	*Bifurcohaptor* spp., *Cichlidogyrus* spp., *C. tilapae*, *Gyrodactylus cichlidarum*, *Neobenedenia melleni*, *Scutogyrus longicornis*, *Silurodiscoides* spp., *Thaparocleidus* spp.
Congridae	Spotted garden-eel (*Heteroconger hassi*)	*Neobenedenia melleni*
Cyprinidae	Barbell (*Barbus barbus*), Balkan barbell (*B. peloponnesius*), bata carp (*Labeo bata*), bleak (*Alburnus alburnus*) , boga carp (*L. boga*), calbasu carp (*L. calbasu*), labeo carp (*L. panguisa*), labeo carp (*L. rohita*), bighead carp (*Hypophthalmichthys nobilis*), chaguni (*Chagunius chagunio*), common carp (*Cyprinus carpio*), fathead minnow (*Pimephales promelas*), flying barb (*Esomus danrica*), gudgeon (*Gobio gobio*), goldfish (*Carassius auratus*, crucian carp (*Carassius carassius*), Indian carp (*Gibelion catla*), mad barb *(Leptobarbus hoevenii),* Mediterranean barbell (*B. meridionalis*), Indian carp (*Cirrhinus cirrhosus),* Mrigal carp (*C. mrigala*), silver barb *(Barbonymus gonionotus*), silver carp (*Hypophthalmichthys molitrix*), transcaucasian carp (*Capoeta capoeta*)	*Ancyrocephalus chakrabartii, Dactylogyrus anchoratus, D. aristichthys, D. batae, D. bifurcatus, D. brevitubus, D. calbasi, D. chagunionis , D. chauhani, D. ctenopharyngodonis, D. extensus, D. fotedari, D. glossogobii, D. hypophthalmichthys, D. inexpectatus, D. kalyanensis, D. labei, D. lamellatus, D. lampam, D. leptobarbus, D. lohanii, D. minutus, D. mrigali, D. nobilis, D. scriabini, D. speciosus, D. suchengtaii, D. vastator, D. vicinus, D. yogendrai, Diplozoon indicum, Dogielius catalius, Gyrodactylus elegans, G. katharineri Haplocleidus vachi, Mazocraes mamaevi, M. singhi, Paradactylogyrus catalius, Paramazocraes gorakhanati, Singhiogyrus exotica, Thaparogyrus lucknowius*
Embiotocidae	Black surfperch (*Embiotoca jacksoni*), striped seaperch (*E. lateralis*)	*Neobenedenia melleni*
Ephippidae	Atlantic spadefish (*Chaetodipterus faber*)	*Neobenedenia melleni*
Hexagrammidae	Kelp greenling (*Hexagrammos decagrammus*)	*Neobenedenia melleni*
Ictaluridae	Brown bullhead (*Ameiurus nebulosus*), channel catfish (*Ameiurus punctatus*), white catfish (*A. catus*)	*Cleidodiscus pricei*
Kyphosidae	Opaleye (*Girella nigricans*), halfmoon (*Medialuna californiensis*)	*Neobenedenia melleni*
Labridae	California sheephead (*Semicossyphus pulcher*)	*Neobenedenia melleni*
Latidae	Asian sea bass/barramundi (*Lates calcarifer*)	*Diplectanum* spp., *Neobenedenia melleni*
Lateolabracidae	Japanese seabass (*Lateolabrax japonica*)	*Neobenedenia melleni*
Pangasiidae	*Pangasius* spp., *Pangasianodon* spp.	*Pangasitrema* spp., *Thaparocleidus* spp., *T. pangasi, T. siamensis, T. caecus*
Polyprionidae	Wreckfish (*Polyprion americanus*), hapuku (*P. oxygeneios*)	*Calicobenedenia polyprioni*
Salmonidae	*Oncorhynchus* spp., *Salmo* spp., *Salvelinus* spp.	*Discocotyle sagittata, Gyrodactylus bychowskii, G. colemanensis*, *G. salaris*, *G. derjavini, G. salmonis, Tetraonchus awakurai*, *T. oncorhynchi*
Scaridae	Bumphead parrotfish (*Scarus perrico*)	*Neobenedenia melleni*
Sebastidae	Cape redfish (*Sebastes capensis*), black rockfish (*S. melanops*), Korean rockfish (*Sebastes schlegelii)*	*Microcotyle sebastis, Neobenedenia melleni*
Serranidae	Grouper (*Epinephelus* spp.), gag (*Mycteroperca microlepis*), leopard coral grouper (*Plectropomus leopardus*), snowy grouper (*Epinephelus niveatus*)	*Neobenedenia melleni, Pseudorhabdosynochus* spp.

Sparidae	Gilt-head sea bream (*Sparus aurata*), sheepshead (*Archosargus probatocephalus*), red sea bream (*Pagrus major*)	*Anoplodiscus tai, Encotyllable spari, Lamellodiscus echeneis, Sparicotyle chrysophrii, Choriocotyle chrysophrii, Gyrodactylus longipipes, G. orecchiae, Neobenedenia melleni*

Monogeneans remain viable where released into fresh water. *Gyrodactylus derjavini* will survive and continue to reproduce for up to 30 hours at 25°C, 48 hours at 17°C or 112 hours at 3°C (Buchmann & Bresciani 1999). *Gyrodactylus salaris* which occurs in fresh and brackish waters, survives for up to 240 hours at 10 ppt salinity, or 42 hours at 20 ppt salinity (OIE 2016b). This tolerance provides for wide distribution in river systems, where wild stocks represent a reservoir of infection for aquaculture species (Buchmann & Bresciani 2006). *Gyrodactylus salaris* primarily affects Atlantic salmon (*S. salar*), but other fish species may act as vectors in waters where the parasite is present (OIE 2016b).

Control measures in freshwater include removal of all fish from entire catchments using rotenone, followed by fallowing and restocking. Recirculating aquaculture systems are treated by drainage and chemical sterilisation (Buchmann & Bresciani 2006). Several applications are usually required, as monogenean eggs are resistant to potassium permanganate, trichlorfon, methylene blue, sodium chloride, chlorine, organophosphate and benzimidazole (Schmahl 1991; Umeda *et al.* 2006; Reed *et al.* 2009). As these chemicals may accumulate in fish, their use is not approved for food production in many countries (Buchmann & Bresciani 2006).

Control in marine waters is not possible and wild fish stocks represent a reservoir for infection (Okawa 2014). For example, the Japanese strain of *B. seriolae* has been distributed between kingfish (*Seriola quinqueradiata*) sea cages for up to 1 km by marine currents (Chambers & Ernst 2003). Given its high fecundity, if 15,000 kingfish were each infected with just five individuals, then 180 million eggs could be released every day from an infected sea cage alone. Densities of 64,000 *B. seriolae* eggs per metre have been reported from floating netting material (Ernst *et al.* 2002).

The infective egg and larval stages of monogeneans, including *G. salaris*, are inactivated by heat treatment (to 60°C for one hour) or by freezing (to below -20°C for at least 24 hours) (Umeda *et al.* 2006; Fraser *et al*. 2008; OIE 2016b).

Potential host species in New Zealand include the families Acanthuridae (*Acanthurus dussumieri, Prionurus maculatus*), Anguillidae (*A. dieffenbachii, A. australis* and *A. rostrata*), Carangidae (*Caranx* spp.), Chaetodontidae (*Amphichaetodon howensis*), Congridae (*Conger verreauxii*), Cyprinaceae (*Carassius aurata*), Echenidae (*Remora* spp.), Ictaluridae (*Ameiurus nebulosus*), Kyphosidae (*Scorpis lineolatus*), Labridae (*Notolabrus fucicola*), Mugilidae (*Mugil cephalus*), Polyprionidae (*Polyprion oxygenios*), Salmonidae (Salmo spp.), Scaridae (*Leptoscarus vaigiensis*), Serranidae (*Epinephelus* sp., *Lepidoperca magna*), Soleidae (*Aseraggodes bahamondei*), Sparidae (*Sparus aurata*) and Tetraodontidae (*Contusus richei*) (Paulin *et al.* 2001; Diggles 2008).

49.2. Risk assessment

49.2.1. Entry assessment

Monogenean hyper-infection is uncommon in wild fish stocks, but is common in aquaculture species (Diggles 2008; Reed *et al*. 2009). Infected fish usually show no external signs, even when heavily infected. As monogeneans are cryptic and transparent and firmly attached to their hosts

with hooks and suckers (Buchmann & Bresciani 2006; Hutson 2010), they are likely to be retained in the commodity.

Monogeneans survive in fresh-chilled eviscerated fish, continuing to feed and reproduce for up to 15 days in chilled product at 4°C (Olstad *et al.* 2006), but have little resistance to freezing. All life stages are killed by freezing and cold storage (to below -20°C for 24 hours) (Fraser *et al.* 2008; OIE 2016b).

Monogeneans which detach from their hosts after death and removal from water can survive for up to 48 hours (Kahn *et al*. 1999; OIE 2016b), but those infecting the gills may continue to feed on their host and remain viable for extended periods (Olstad *et al.* 2006). The eggs are resistant to temperature and salinity variations and may enter the aquatic environment through the factory wastewater stream (Reed *et al*. 2009).

The likelihood of entry of *G. salaris* and related monogeneans through the commodity is assessed as non-negligible.

49.2.2. Exposure assessment

To establish an infection in New Zealand through fresh-chilled eviscerated fish, monogenean infective eggs or larvae would have to contact a susceptible alternative fish host in sufficient quantity and duration (Kahn *et al*. 1999; Chambers & Ernst 2003).

Monogeneans may remain viable in fish tissues for up to 15 days at 3°C (Olstad *et al*. 2006) and survive for up to 48 hours at 17°C, so they can be distributed through discarded fish offal, or in the factory wastewater discharges. The egg stages are resistant to environmental extremes (OIE 2016b) and can survive for up to 6 months in the aquatic environment (Reed *et al*. 2009).

The numbers required to initiate infection are low and monogeneans may cause epizootics through their high fecundity and the viviparous release of offspring (Buchmann & Bresciani 1999).

Once established, clinical disease may occur rapidly in downstream populations and other fish species can also act as vectors (OIE 2016b). Monogeneans are now widely distributed in European freshwater river systems (Buchmann & Bresciani 1999) and are common for at least 1 km from the infection site in marine waters (Chambers & Ernst 2003; OIE 2016b).

The likelihood of exposure of *G. salaris* and related monogeneans through the commodity is assessed as non-negligible.

49.2.3. Consequence assessment

The consequences of establishment of monogenean pathogens vary widely depending upon the availability of primary or alternative host species (Buchmann & Bresciani 2006). Monogenean pathogenicity is low at lower levels of incidence (Buchmannn & Bresciani 2006), so the consequences of establishment are likely to be negligible for wild New Zealand fish.

Monogeneans, including *G. salaris,* can become pathogenic to farmed fish and wild stocks may represent a reservoir for infection (OIE 2016b). The consequence of introduction of a pathogen likely to be associated with farmed marine and freshwater fish in New Zealand would incur direct costs for aquaculture. The potential effect of the introduction of *G. salaris* in Scottish salmonid

aquaculture, included a likely mortality of 50% with a projected economic loss of £633 million in 2006 (Riddington *et al*. 2006).

Farmed fish in New Zealand include eels (Anguillidae), salmon, trout (Salmonidae) and carp (Cyprinidae). Freshwater aquaculture in New Zealand largely depends upon the availability of uncontaminated water (Sim-Smith *et al.* 2014). The consequences of establishment to salmonid aquaculture would include direct losses due to mortality, as well as indirect losses resulting from the introduction of an OIE-listed disease. Freshwater and marine salmonid aquaculture was valued at $63 million in export earnings in 2011 (MPI 2014; Seafood New Zealand 2014). The hatcheries producing salmonids for the recreational and tourist trout and salmon fisheries would also be affected, with significant social and environmental costs. While the dollar value of New Zealand's freshwater fisheries is not fully known, the Taupo fishery alone is valued at $70–80 million per annum (Marsh & Mkwra 2013).

Introduced monogenean pathogens may affect farmed cyprinid species including grass carp (*Ctenopharyngodon idella*). This would have significant consequences for the industries associated with the use of cyprinids in weed control in New Zealand's freshwater lakes and rivers (Hofstra 2014; NIWA 2014a).

Farmed marine species in New Zealand include grouper (Polyprionidae), snapper (Sparidae), salmon (Salmonidae) and yellowtail kingfish (Carangidae). *Gyrodactylus salaris* causes major economic effects in the sea cage aquaculture of salmonids including Atlantic salmon (*S. salar*) (OIE 2016b).

Monogenean infection is a limiting factor in the sea-cage aquaculture of amberjack (*Seriola quinqueradiata*) in Japanese marine farms, where *Benedenia seriolae* causes production losses up to 22% (Okawa 2014). Exotic monogeneans may have economic consequences for farming of yellowtail kingfish (*S. lalandi*) (NIWA 2014b; Symonds *et al.* 2014). In addition to the direct costs of freshwater bath or chemical treatment (Okawa 2014), poorly coordinated treatments between Japanese farms resulted in rapid re-infection and provided only short-term economic relief (Ernst et *al.* 2002). *Calicobenedenia polyprion* may affect the developing sea cage aquaculture for hapuku (*Polyprion oxygeneios*) and kingfish (*S. lalandi*), through lost production and additional costs of treatment (Hutson 2010; NIWA 2014b; Symonds *et al.* 2014).

The consequences of establishment of *G. salaris* and related monogeneans are assessed as non-negligible.

49.2.4. Risk estimation

As the entry, establishment and consequence assessments are non-negligible, *G. salaris* and related monogenean ectoparasites are assessed to be a risk in the commodity and risk management measures may justified.

49.3. Risk management

Monogeneans can remain viable on the skin and gills of fish. The risk of establishment is primarily associated with fresh-chilled marine and freshwater fish from a wide range of species and geographical locations (Buchmann & Bresciani 2006).

Infection with *G. salaris* is an OIE-listed disease (OIE 2016a), so the Aquatic *Code* provides specific guidelines for the importation of aquatic animals or animal products from any country or

zone not declared free from infection. These recommendations apply to susceptible species listed in section 10.3.2 of the *Code:*

Atlantic salmon (S. salar), rainbow trout (O. mykiss), Arctic char (S. alpinus), North American brook trout (S. fontinalis), grayling (T. thymallus), North American lake trout (S. namaycush) and brown trout (S. trutta). They also apply to other fish species from waters where the parasite is present because these species may carry the parasite and act as vectors. The *Code* provides guideline for the importation of aquatic animals and animal products from countries/zones/compartments not declared free from *G. salaris.*

Article 10.3.3 of the *Code* states:

1. *Competent Authorities should not require any related conditions related to infection with G. salaris, regardless of the infection with G. salaris status of the exporting country, zone or compartment, when authorising the importation or transit of the following aquatic animals and aquatic animal products from the species referred to in Article 10.3.2. which are intended for any purpose and which comply with Article 5.4.1.:*
    a. *heat sterilised, hermetically sealed fish products (i.e. a heat treatment at 121˚C for at least 3.6 minutes or any time/temperature equivalent);*
    b. *pasteurised fish products that have been subjected to a heat treatment at 63˚C for at least 30 minutes (or any time/temperature equivalent which has been demonstrated to inactivate G. salaris);*
    c. *mechanically dried, eviscerated fish (i.e. a heat treatment at 100˚C for at least 30 minutes or any time/temperature equivalent which has been demonstrated to inactivate G. salaris);*
    d. *naturally dried, eviscerated fish (i.e. sun-dried or wind-dried);*
    e. *frozen eviscerated fish that have been subjected to minus 18°C or lower temperatures;*
    f. *frozen fish fillets or steaks that have been subjected to minus 18°C or lower temperatures;*
    g. *chilled eviscerated fish that have been harvested from seawater with a salinity of at least 25 parts per thousand (ppt);*
    h. *chilled fish fillets or steaks derived from fish that have been harvested from seawater with a salinity of at least 25ppt;*
    i. *chilled fish products from which the skin, fins and gills have been removed;*
    j. *fish roe.*

2. *When authorising the importation or transit of aquatic animals and aquatic animal products of a species referred to in Article 10.3.2., other than those referred to in point 1 of Article 10.3.3., Competent Authorities should require the conditions prescribed in Articles 10.3.7 to 10.3.11 relevant to the infection with G. salaris status of the exporting country, zone or compartment.*

3. *When considering the importation or transit of aquatic animals and aquatic animal products of a species not covered in Article 10.3.2. but which could reasonably be expected to pose a risk of spread of infection with G. salaris, the Competent Authority should conduct a risk analysis in accordance with the recommendations in Chapter 2.1. The Competent Authority of the exporting country should be informed of the outcome of this assessment.*

Compliance with Articles 10.3.3 and 10.3.11 should eliminate *G. salaris* and all other monogenean pathogens from the commodity and be a viable risk management option.

Monogeneans are reported from a wide range of fish hosts and some species may be reasonably expected to be pathogenic to species farmed in New Zealand. Many monogeneans are host-specific, but naïve infection may occur across several, generally related, host species (Ernst *et al.* 2002; Buchmann & Bresciani 2006; Sepulveda & Gonzalez 2014; Mendoza-Palmero *et al.* 2015; OIE 2016b). Over 20 families of marine and freshwater fish are associated with monogenean pathogens (Table 46), that may be present in the commodity. Species declaration should substantially reduce the pathogen load of these monogeneans, including *Gyrodactylus salaris*, in the commodity and is a viable risk management option. Infection in wild fish occurs at low levels of incidence and pathogenicity (Buchmann & Bresciani 2006). Restriction of the commodity to wild-caught fish (not from aquaculture) should moderately reduce pathogen load and be a viable risk management option.

As infection with *Gyrodactylus salaris* is an OIE-listed disease, the Competent Authority of the exporting country is expected to have a monitoring and surveillance programme (OIE 2016a), but no specific monitoring programmes are required for other monogenean pathogens. Where country/zone freedom is approved through the MPI Country Approval Procedures, this option is likely to substantially reduce the pathogen load of *G. salaris* and other monogenean pathogens. Declaration of country/zone freedom is a viable risk management option.

Monogeneans are ectoparasites mainly located on the skin or the gill tissues (Buchmann & Bresciani 2006). Removal of the gills should slightly reduce monogenean pathogen load, while removal of the head and gills should moderately reduce pathogen load in the commodity. Further processing to the skin-off fillet state would be expected to substantially reduce monogenean pathogen load in the commodity. This processing requirement was proposed for imported catfish (*Pangasius* spp.) and tilapia (*Oreochromis* spp.) (Johnston 2008a, 2008b).

Monogeneans are denatured by heat treatment (to at least 60°C for 60 minutes) (OIE 2016b), so heat treatment would eliminate these pathogens from the commodity. Heat treatment is a viable risk management option.

Monogeneans are also destroyed by freezing (to below -20°C for at least 24 hours) (Fraser *et al.* 2008), so frozen treatment would eliminate these pathogens from the commodity. Frozen storage is a viable risk management option.

49.3.1. Risk management options

Monogenean ectoparasites including *Gyrodactylus salaris* are reported from fish in families Acanthuridae, Anguillidae, Carangidae, Catostomidae, Chaetodontidae, Channidae, Cichlidae, Congridae, Cyprinidae, Embiotocidae, Hexagrammidae, Ictaluridae, Kyphosidae, Labridae, Lateolabracidae, Latidae, Pangasiidae, Polyprionidae, Salmonidae, Scaridae, Sebastidae, Serranidae and Sparidae (Table 46) which are considered likely to be present in the commodity. Other families have not been associated with these monogenean parasites. Therefore species declaration indicating the commodity is not originated from any of the above families should substantially reduce the occurrence of monogenean ectoparasites, including *Gyrodactylus salaris* in the commodity.

For the commodities originated from families associated with monogenean ectoparasites, including *Gyrodactylus salaris*, one or a combination of the following additional options could also be considered to effectively manage the risk.

Where country/zone freedom from *G. salaris* and related monogeneans is accepted by MPI:

Option 1

Acceptance of country/zone freedom by MPI should substantially reduce the occurrence of *G. salaris* and related monogeneans, so the commodity may be imported without any further restrictions.

Where country/zone freedom from *G. salaris* and related monogeneans is not accepted by MPI or not available:

Option 2

Processing consistent with the conditions of Article 10.3.3 or 10.3.11 of the OIE Aquatic *Code* (OIE 2016a) should eliminate *G. salaris* and related monogeneans. Where these provisions are met, the commodity could be imported without further restrictions.

Where the imported commodity is not consistent with Article 10.3.3 or 10.3.11 of the OIE Aquatic *Code* (OIE 2016a) and further processing is necessary:

Option 3

Frozen storage (to below -20°C for 24 hours) should eliminate *G. salaris* and related monogeneans. When this provision is met, the commodity could be imported without any further restrictions; or,

Option 4

Further processing (to the skin-off fillet state) should substantially reduce the occurrence of *G. salaris* and related monogeneans. When this provision is met, the commodity could be imported without any further restrictions; or,

Option 5

Restriction of the commodity to wild-caught fish (not from aquaculture) should moderately reduce the occurrence of *G. salaris* and related monogeneans. When this provision is met, the commodity could be imported without any further restrictions.

49.4. References

Anonymous (2016). Biosecurity (Notifiable Organisms) Order 2016. NZLR 2016/73. New Zealand Government, Wellington, 13 p.

Antonelli L, Quilichini Y, Marchland B (2010). *Sparicotyle chrysophrii* (Van Beneden and Hesse 1863) (Monogenea: Polyopisthocotylea) parasite of cultured Gilthead sea bream *Sparus aurata* (Linnaeus 1758) (Pisces: Teleostei) from Corsica: ecological and morphological study. *Parasitological Research* 107: 389–398.

Boustead NC (1982). Fish diseases recorded in New Zealand, with a discussion on potential sources and certification procedures. Occasional Publications No. 34. Fisheries Research Division, Ministry of Agriculture and Fisheries, Wellington, New Zealand, 19 p.

Buchmann K, Bresciani J (1999). Rainbow trout leucocyte activity: influence on the ectoparasitic monogenean *Gyrodactylus derjavini. Diseases of Aquatic Organisms* 35: 13–22.

Buchmann K, Bresciani J (2006). Monogenea (Phylum Platyhelminthes). pp 297–342. *In*: Woo PTK (*Ed.*). Fish Diseases and Disorders. Volume 1: Protozoan and Metazoan Infections (Second Edition). CABI International, Wallingford, United Kingdom, 791 p.

Bullard SA, Benz GW, Overstreet RM, Williams EH, Hemdal Y (2000). Six new host records and an updated list of wild hosts for *Neobenedenia melleni* (MacCallum) (Monogenea: Capsalidae). *Comparative Parasitology* 67(2): 190–196.

Chambers C, Ernst I (2003). Effect of tidal current on monogenean egg dispersal and infection rates at a kingfish farm in Australia. *In*: Van As (*Ed.*) Proceedings of the Sixth International Symposium on Fish Parasitology, Bloemfontein, South Africa, September 22–26, University of Free State, Bloemfontein, South Africa.

Chaudhary A, Verma C, Manu-Varma S, Singh HS (2013). A review of Monogenean diversity in India: Pathogens of fish diseases. *Journal of Coastal Life Medicine* 1(2): 151–168.

Diggles BK (2008). Transfer of parasites between wild and cultured fishes with respect to aquaculture development in the Firth of Thames, New Zealand. DigsFish Services Pty Ltd. (Appendix 2 in Kelly S (*Ed*.). Potential environmental effects associated with the proposed shift from mussel to finfish farming in the Firth of Thames. *Environment Waikato Technical Report* 2008/38, 22p.

Ernst I, Whittington ID, Corneillie S, Talbot C (2002). Monogenean parasites in sea-cage aquaculture. *Austasia Aquaculture* February/March 2002: 46–48.

Ernst I, Whittington ID, Corneillie S, Talbot C (2005). Effects of temperature, salinity, desiccation and chemical treatments on egg embryonation and hatching success of *Benedenia seriolae* (Monogenea: Capsalidae), a parasite of farmed *Seriola* spp. *Journal of Fish Diseases* 28: 157–164.

Faruque Miah M, Deb M, Ali H, Quddus MMA, Ahmed K (2013). Comparative surveillance of parasitic infestation in *Channa punctatus* (Osteichthyes: Channidae) collected from open and closed waters in Sylhet, Bangladesh. *Advances in Zoology and Botany* 1(1): 17–23.

Fraser DI, Munro PD, Small DA (2008). Disinfection Guide Version IV: Practical steps to prevent the introduction and minimise transmission of diseases of fish. Internal Report 13/06. Fisheries Research Services, Aberdeen, Scotland, 24 p.

Fromm B, Burow S, Hahn C, Bachmann L (2014). MicroRNA loci support conspecificity of *Gyrodactylus salaris* and *Gyrodactylus thymalli* (Platyhelminthes: Monogenea). *International Journal of Parasitology* 44: 787–793.

Gilmore SR, Cone DK, Lowe G, King SK, Jones SRM, Abbott CL (2012). Molecular phylogeny of *Gyrodactylus* (Monogenea) parasitizing fishes in fresh water, estuarine, and marine habitats in Canada. *Canadian Journal of Zoology* 90: 776–786.

Hansen H, Bachmann L, Bakke TA (2003). Mitochondrial DNA variation of *Gyrodactylus* spp. (Monogenea, Gyrodactylidae) populations infecting Atlantic salmon, grayling, and rainbow trout in Norway and Sweden. *International Journal of Parasitology* 33(13): 1471–1478.

Hine PM. Jones JB, Diggles BK (2000). A checklist of the parasites of New Zealand fishes, including previously unpublished records. NIWA Technical Report 75. National Institute of Water and Atmospheric Research Ltd., Auckland, New Zealand, 93 p.

Hofstra DE (2014). Grass carp effectiveness and effects. Stage 2: Knowledge review. NIWA Client Report HAM2014-060, prepared for the Department of Conservation, Wellington, New Zealand, 140 p.

Hutson K (2010). Parasites of hapuku and seabass (*Polyprion oxygeneios* and *P. americanus*) [Online] Available from: http://www.marineparasites.com/pdf/Hapuku.pdf [Accessed 18 May 2015].

Johnston C (2008a). Import Risk Analysis: Frozen skinless and boneless fillet meat of *Oreochromis* spp. fish from Brazil and China for human consumption. Biosecurity New Zealand, Ministry of Agriculture and Forestry, Wellington, New Zealand, 96 p.

Johnston C (2008b). Import Risk Analysis: Frozen skinless and boneless fillet meat of *Pangasius* spp. fish from Vietnam for human consumption. Biosecurity New Zealand, Ministry of Agriculture and Forestry, Wellington, New Zealand, 60 p.

Kahn SA, Beers PT, Findlay VL, Peebles IR, Durham PJ, Wilson DW, Gerrity SE (1999). Import risk analysis on non-viable salmonids and non-salmonid marine finfish. Australian Quarantine and Inspection Service, Department of Agriculture, Fisheries and Forestry, Canberra, Australia, 409 p.

Mahmoud NE, Mahmoud AM, Fahmy MM (2015). Parasitological and comparative pathological studies on monogenean infestation of cultured sea bream (*Sparus aurata*, Sparidae) in Egypt. *Oceanography* 2: 129 [Online] Available from: doi: 10.4172/2332-2632.1000129 [Accessed 19 October 2015].

Marsh D, Mkwra L (2013). Review of freshwater non-market value studies. Department of Economics, University of Waikato, Hamilton, New Zealand, 144 p.

Meinila M, Kussela J, Zietara MS, Lumme J (2004). Initial steps of speciation by geographical isolation and host switch in salmonid pathogen *Gyrodactylus salaris* (Monogenea: Gyrodactylidae). *International Journal for Parasitology* 34: 515–526.

Mendoza-Palmero CA, Blaso-Costa I, Scholz T (2015). Molecular phylogeny of Neotropical monogeneans (Platyhelminthes: Monogenea) from catfishes (Siluriformes). *Parasites and Vectors* 8: 164 [Online] Available from: http://www.parasitesandvectors.com/content/pdf/s13071-015-0767-8.pdf [Accessed 18 June 2015].

Montero FE, Aznar FJ, Fernández M, Raga JA (2003). *Pseudoallencotyla* gen. n., a new genus for *Allencotyla pricei* (Kritsky, Noble et Moser 1978) (Monogenea: Heteraxinidae), with a key to genera of Heteraxininae. *Folia Parasitologica* 50: 43–47.

Ministry for Primary Industries (MPI) (2014). Fisheries Assessment Plenary, May 2014: Stock assessments and stock status. Compiled by the Fisheries Science Group, Ministry for Primary Industries, Wellington, New Zealand, 1381 p.

NIWA (2014a). Grass carp *Ctenopharyngodon idella* (Valenciennes, 1844) [Online] Available from: https://www.niwa.co.nz/freshwater-and-estuaries/nzffd/NIWA-fish-atlas/fish-species/grass_carp [Accessed 12 February 2015].

NIWA (2014b). Yellowtail kingfish [Online]. Available from: https://www.niwa.co.nz/aquaculture/aquaculture-species/yellowtail-kingfish [Accessed 20 October 2015].

OIE (2016a). Aquatic Animal Health Code, 19th Edition. World Organization for Animal Health. Paris, France [Online] Available from: http://www.oie.int/international-standard-setting/aquatic-code/access-online/ [Accessed 15 March 2017].

OIE (2016b). Manual of diagnostic tests for aquatic animals. World Organization for Animal Health. Paris, France [Online] Available from: http://www.oie.int/international-standard-setting/aquatic-manual/access-online/ [Accessed 17 March 2017].

Okawa K (2014). Diseases of cultured marine fishes caused by Platyhelminthes (Monogenea, Digenea, Cestoda). *Parasitology* 142: 178–195.

Olstad K, Cable J, Robertson G, Bakke TA (2006). Unpredicted transmission strategy of *Gyrodactylus salaris* (Monogenea: Gyrodactylidae): survival and infectivity of parasites on dead hosts. *Parasitology* 133(1): 33–41.

Paladini G, Hanson H, Fioravanti ML, Shinn AP (2011). *Gyrodactylus longipes* n. sp. (Monogenea: Gyrodactylidae) from farmed gilthead seabream (*Sparus aurata* L.) from the Mediterranean. *Parasitology International* 60(4): 410–418.

Paulin C, Stewart A, Roberts C, McMillan P (2001). New Zealand Fish: A complete guide. Te Papa Press, Wellington, New Zealand. 279 p.

Reed P, Francis-Floyd R, Klinger RE, Petty D (2009). Monogenean parasites of fish. Fisheries and Aquatic Sciences Report 28, Institute of Food and Agricultural Science, University of Florida, Gainesville, Florida, United States of America, 10 p.

Riddington G, Radford A, Paffrath S, Bostock J, Shinn A (2006). An economic evaluation of the impact of the salmon parasite *Gyrodactylus salaris* (Gs) should it be introduced into Scotland. Report prepared for the Scottish Executive Environmental and Rural Affairs Department. Institute of Aquaculture, University of Stirling, Glasgow, Scotland, 132 p.

Schmahl G (1991). The chemotherapy of monogeneans, which parasitize fish: A review. *Folia Parasitologica* 38: 97–108.

Seafood New Zealand (2014). Seafood New Zealand: Key facts for our Industry. Seafood New Zealand, Wellington, New Zealand, 6 p.

Sepulveda FA, Gonzalez MT (2014). Molecular and morphological analyses reveal that the pathogen *Benedenia seriolae* (Monogenea: Capsalidae) is a complex species: Implications for yellowtail *Seriola* spp. aquaculture. *Aquaculture* 418–419: 94–100.

Sim-Smith C, Faire S, Lees A (2014). Managing biosecurity risk for business benefit. Report prepared for the Ministry for Primary Industries by Coast and Catchment Ltd, Auckland, New Zealand, 214 p.

Sitjà-Bobadilla A, Alvarez-Pellitero P (2009). Experimental transmission of *Sparicotyle chrysophrii* (Monogenea: Polyopisthocotylea) to gilthead seabream (*Sparus aurata*) and histopathology of the infection. *Folia Parasitologica* 56(2): 143–151

Strona G (2014). The underrated importance of predation in transmission ecology of direct lifecycle parasites. *Oikos* 124(6): 685–690.

Symonds JE, Walker SP, Pether S, Gublin Y, McQueen D, King A, Irvine GW, Setiawan AN, Forsythe JA, Bruce M (2014). Developing yellowtail kingfish (*Seriola lalandi*) and hapuku (*Polyprion oxygeneios*) for New Zealand aquaculture. *New Zealand Journal of Marine and Freshwater Research* 48(3): 371–384.

Umeda N, Nibe H, Hara T, Hirazawa N (2006). Effects of various treatments on hatching of eggs and viability of oncomiracidia of the monogeneans *Pseudodactylogyrus anguillae* and *Pseudodactylogyrus bini. Aquaculture* 253(1–4): 148–153.

Whittington ID (2011). *Benedenia seriolae* and *Neobenedenia* species. pp 225 –244. *In:* Woo PTK, Buchmann K (*Eds*.) Parasites: Pathobiology and protection. CABI, Wallingford, United Kingdom, 395 p.

Whittington ID (2004). The Capsalidae (Monogenea: Monopisthocotylea): a review of diversity, classification and phylogeny with a note about species complexes. *Folia Parasitologica* 51: 109–122.

Whittington ID (1998). Diversity "down under": Monogeneans in the antipodes Australia with a prediction of monogenean diversity worldwide. *International Journal for Parasitology* 28: 1481–1493.

Woo PTK, Buchmann L (Eds.) (2012). Fish Parasites: Pathobiology and protection. CABI, Wallingford, United Kingdom, 383 p.

Zahradníčková PL, Barson M, Luus-Powell WJ, Gelnar M, Přikrylová I (2014). Study of species diversity of viviparous monogeneans parasitizing African cichlids: looking for a needle in a haystack. Session Four: Monogeneans and nematodes – ecological and veterinary parasitology, pp 53–54 *In:* Parasites in the Heart of Europe. V4 Parasitological Meeting, Congress Centre, Stara Lesna, Slovakia, May 25–30, 2014.

50. Digenean pathogens (including muscle-encysting larvae)

50.1. Hazard identification

50.1.1. Aetiological agent

Digenean trematodes are internal parasites classified within the Class Digenea of Phylum Platyhelminthes (Paperna & Dzikowski 2006). Their complex life cycle involves several intermediate larval stages that infect invertebrates and vertebrates, which may function as paratenic (alternative transport) hosts. Many of these larval stages are incompletely known, or incorrectly described (Paperna & Dzikowski 2006).

50.1.2. OIE status

Infection with digenean pathogens is not listed by the OIE (OIE 2016).

50.1.3. New Zealand status

Digeneans are reported from marine fish in New Zealand, but species causing clinical disease of economic importance in fish, or of zoonotic concern, are regarded as exotic (Boustead 1982; Hine *et al*. 2000; Johnston 2008a, 2008b). Infection with digenean parasites is not a notifiable disease in New Zealand (Anon. 2016).

50.1.4. Epidemiology

The digenean life cycle is complex, involving at least three hosts. At least one intermediate host is an invertebrate (mollusc, crustacean, annelid), while other invertebrates or vertebrates, including fish, may function as intermediate or paratenic hosts (Paperna & Dzikowski 2006). When digenean trematodes (other than blood flukes such as *Sanguinicola* spp.) occur in their adult state in fish, they rarely cause clinical disease (Johnston 2008a).

Where the digenean intermediate stages occur in fish, the adult stage generally occurs in piscivorous birds or mammals (Paperna & Dzikowski 2006). Digenean eggs are usually released with the host faeces or secretions and hatch to release a free-swimming miracidium larvae, which may be consumed by a range of invertebrate hosts.

The complex life cycle involves several asexual sporocyst and redia larval stages in the intermediate hosts (Paperna & Dzikowski 2006) that serve to increase larval numbers and allow the progressive release of the infective larvae from the intermediate host (Paperna & Dzikowski 2006). The incidence of infection may exceed 80% (Kajiyama *et al.* 1979), while an invertebrate host may remain infective for at least 5 years. Up to 3,300 infective larvae may be released per day from an intermediate host (Paperna & Dzikowski 2006). Additional progenetic sexually mature life cycle stages may also occur in the intermediate annelid or crustacean host, which co-exist, or alternate with, the asexually developing larval stages. These progenetic stages produce eggs that are released on the death or predation of the intermediate host and release infective larvae to continue the cycle (Paperna & Dzikowski 2006).

Carnivorous fish generally become infected through consumption of an infected invertebrate, while herbivorous fish become infected through consumption of cercaria attached to plant material or other suitable substrates but infection may also occur by direct invasion across the epidermis of the

skin, gills and fin bases (Paperna & Dzikowski 2006). Within the fish host, the cercaria larvae actively migrate to their chosen location (such as the musculature, skin, eyes, gills or viscera) where they encyst as a metacercaria larva.

In most cases, digenean larvae have little effect on their host, even when present in brain tissues (Betterton 1974; Chappell 1995; Paperna & Dzikowski 2006; Simoes *et al.* 2009), but the presence of digenean larvae in the flesh results in tissue spoilage, rendering the commodity unmarketable (FDA 2017).

While infection in wild fish stocks rarely occurs in sufficient numbers to cause significant disease (Paperna & Dzikowski 2006; Diggles 2008), the reported occurrence may be high. *Metagonimus yokogawai* is present in over 50% of susceptible wild freshwater fish in Taiwan (Li *et al.* 2013).

The life cycle is completed upon consumption of the fish by the final or paratenic host, where the metacercaria excysts and develops into an adult (Paperna & Dzikowski 2006).

Digenean metacercaria affecting fish usually have a wide host range, although some species appear to be preferred hosts. Their distributions are effectively limited to either marine or fresh waters by the salinity tolerance limits of their hosts. Where the definitive host is a land vertebrate, the fish intermediate host generally occurs in coastal inshore or estuarine waters (Paperna & Dzikowski 2006). Digeneans infecting widely distributed fish hosts, such as mullet (*Mugil cephalus*), generally have the widest range of alternative intermediate hosts (Paperna & Dzikowski 2006).

A representative, but not exhaustive, list of digenean fish ectoparasites that may potentially establish in New Zealand, is given in Table 47 (Wells & Randall 1955, Noda 1959; Berrie 1960; Aleem 1987; Phillips & Lambert 1989; Cribb *et al.* 1992; Rim *et al.* 1996; Arthur & Lumanlan-Mayo. 1997; Bargues & Mas-Coma 1997; Fan 1998; Musiba & Nkwengulila 1998; Scholz 1999, Slifco *et al.* 2000; Venable *et al.* 2000; Roberts 2001; Kennedy 2004; Paperna & Dzikowski 2006; Shelaby *et al.* 2006; Reynard *et al.* 2007; Abou-Eisha *et al.* 2008, Armignacco *et al*. 2008; Bullard *et al.* 2008; Chi *et al.* 2008; Johnston 2008a, 2008b; EFSA 2010; Lobna *et al.* 2010, Arocena & Rodriguez 2011; Cho *et al*. 2012; Hung *et al.* 2013; Li *et al.* 2013; Melo *et al*. 2013; Anon. 2014; Krailas *et al.* 2014; Mayo-Hernandez *et al.* 2015; Ozer & Kirca 2015; Rosser *et al*. 2016; Nareaho *et al*. 2017; Santos *et al*. 2018).

The spread of digenean metacercaria infection in the United States is related to the distribution and consumption of raw, salted, fermented or chilled fish (FDA 2017). Encysted digenean cercariae are resistant to the temperatures likely to be encountered during the routine storage and processing of chilled fish. *Clonorchis* sp. cercariae remain viable in chilled (3–6°C) eviscerated fish product after 16 days, with viability declining to 50% after 40 days storage (Bargues & Mas-Coma 1997).

Digenean metacercaria are unaffected by cold-smoke or brine processing. *Clonorchis* spp. required high salt concentrations (3 g salt g^{-1} fish at 26°C for 15 days) to ensure non-viability (Fan 1998; EFSA 2010; FDA 2017).

Digenean metacercaria in raw fish intended for human consumption were inactivated by frozen storage at -35°C for 15 hours (FDA 2017), or at -20°C for 168 hours (Johnston 2008a).

Table 47. Representative List of Families and Species of Fish Susceptible to Muscle Encysting Digeneans

Host Family	Fish Species	Pathogen
Channidae	Snakehead (*Channa punctata, C. striata*)	*Euclinostomum heterostomum, Lophosicyadiplostomum* sp., *Posthodiplostomum spp., Polylelithum* sp.
Cichlidae	Redbelly tilapia (*Coptodon zilli*), Nile tilapia (*Oreochromis niloticus*)	*Ascocotyle ascolonga, Clinostomum* spp., *Clinostomum tilapae, Euclinostomum heterostomum, Heterophyes* spp., *Mesostephanus* spp., *Moedlingeria amphoraeformis, Pharyngostoma* spp*., Polylekithum sp. Posthodiplostomum sp., Procerovum* spp., *Prohemistomum vivax, Pygidiopsis* spp.,
Clupeidae	Shad (*Konosirus punctatus*)	*Heterophyes* spp.
Cyprinidae	Barbs (*Barbus* spp.), bleak (*Alburnus* spp.), bighead carp (*Hypophthalmichthys nobilis*), black carp (*Mylopharyngodon piceus*), common and ornamental carp (*Cyprinus carpio*), bleaker (*Opsariichthys evolans*), freshwater minnow (*Zacco platypus*), (fathead minnow (*Pimephales promelas*), freshwater bream *(A. brama*), golden shiner (*Notemigonus crysoleucas*), flying barb (*Esomus metallicus*), goldfish (*Carassius auratus*), grass carp (*Ctenopharyngodon idella*), Hampala barb (*Hampala dispar*), Java barb (*Systomus rubripinnis),* minnow (*Phoxinus* spp.), mrigal (*Cirrhinus mrigal*), roach (*Rutilus rutilus*), rohu (*Labeo rohita*), rudd (*Scardinius erythrophthalmus*), silver carp (*Hypophthalmichthys molitrix*), snow trout (*Schizopyge niger, Schizothoraxesocinus*), sunbleak (*Leucaspius* spp.), tench (*Tinca tinca*), topmouth culter (*Culter alburnus*), topmouth gudgeon (*Pseudorasbora parva*), white bream (*Blicca bjoerkna*), Zope bream (*Ballerus ballerus*), and over 150 other species of fish	*Centrocestus formosanus, Clonorchis* sp., *Diplostomum* spp., *Echinochasmus* spp., *Metorchis bilis, Metagonimus* spp., *Opisthorchis* spp.,*Posthodiplostomum* spp.
Elotridae	Pacific fat sleeper (Dormitator latifrons)	*Clinostomum* spp.
Esocidae	Northern pike (*Esox lucius*)	*Metorchis conjunctus*
Ictaluridae	Catfish (*Ameiurus punctatus*)	*Bolbophorus* spp*., Centrocestus formosanus*
Lateolabracidae	Japanese seabass (*Lateolabrax japonicus*)	*Clinostomum* spp.
Mugilidae	mullet (*Mugil cephalus*), redlip mullet (*Planiliza haematocheila*)	*Ascocotyle longa, Clonorchis* sp., *Diplostomum* sp., *Heterophyes* spp., *Heterophyopsis continua, Metorchis bilis*, *Metagonimus* spp., *Posthodiplostomum sp., Pygidiopsis* spp*., Transversotrema* spp.
Percidae	European perch (*Perca fluviatilis*), Yellow perch (*Perca flavescens*)	*Bolbophorus* spp., *Clinostomum* spp., *Diplostomum* spp., *Metorchis conjunctus*
Salmonidae	brown trout (*Salmo trutta*), rainbow trout (*Oncorhynchus mykiss*), lake trout (*Salvelinus fontinalis*.)	*Diplostomum* spp., *Haplorchis* spp., *Metorchis conjunctus*
Serralsalmidae	Cachama (*Collosoma macropomum*)	Undescribed digenean metacercaria
Serranidae	Grouper (*Epinephelus* spp.)	*Heterophyopsis continua, Procerovum varium*
Siluridae	Wallago (*Wallago attu*)	*Euclinostomum heterostomum, Lophosicyadiplostomum sp., Posthodiplostomum spp., Polylekithum* sp.
Soleidae	*Solea vulgaris*	*Heterophyes* sp.
Sparidae	black sea bream (*Acanthopagrus schlegelii*)	*Heterophyes* spp.

In other studies, larvae of the families Cyathocotylidae (*Mesostephanus* spp. and *Prohemistomum* sp.) and Heterophyidae (*Pygidiopsis* spp., *Centrocestus* spp., *Heterophyes* spp., *Haplorchis* spp., *Metagonimus* spp., and *Phagicola* spp.) present in *Oreochromis* spp. fillet product were inactivated by freezing at -10°C for 3 days, or for 2 days at -20°C (Elnawawi *et al.* 2000). The cercaria of *Heterophyes* spp. in fish fillets required freezing for 8 hours at -20°C to ensure inactivation (Wiwanitkit *et al.* 2001), while *Clonorchis* spp. required 72 hours at -12°C (Fang *et al.* 2003) and *Pygidiopsis* spp. required 20 days of frozen storage at -12°C (Yousef *et al*. 1981; Fan 1998). *Heterophyes heterophyes* required 30 hours of frozen storage at either -10°C or -20°C to ensure inactivation (Hamed & Elias 1970).

For whole fish, rather than fillets, freezing for 3 days at -26°C inactivated the cercaria of cyathacotylid and heterophyid digeneans (Rácz & Zemankovics 2002), while *Opisthorchis* sp. required lower temperatures (-28°C for 20 hours) (Fattakhov 1989), or longer storage (-22°C for 92 hours) to ensure inactivation (Fattakhov 1985).

Digenean metacercaria are killed by heat treatment (cooking for 20 minutes at 70°C, or 2 hours at 50°C (Borges *et al.* 2015), while the zoonotic Heterophyid *Ascocotyle* spp. are denatured by cooking (to at least 56ºC for 2-3 minutes (Rodriguez *et al.* 2015).

Digenean trematodes may be agents of zoonotic disease (Slifco *et al.* 2000; Belizario *et al.* 2004; Cywinska 2005; WHO 2005; Paperna & Dzikowski 2006; Athokpam & Tandon 2012; Petney *et al.* 2013; Doanh & Nawa 2016) but they appear to be under-reported and under-diagnosed in many countries (Slifco *et al.* 2000). Where raw or under-cooked fish are consumed, metacercaria present in the flesh are released in the intestine and migrate to the liver or bile duct (Hung *et al.* 2013). Fish-transmitted zoonotic trematodes include 59 species in four families (Opisthorchiidae, Echinostomatidae, Heterophyidae and Nanophyetidae) (Hung *et al.* 2013; Hegazi *et al.* 2014; Rodriguez *et al.* 2015), as well as digenean families Allocreadidae, Clinostomidae, Diplostomidae (Athokpam & Tandon 2012). Major zoonotic pathogens include *Ascocotyle* spp., *Clonorchis sinensis*, *Opisthorchis viverrini* and *O. felineus*, as well as *Metorchis conjunctus*, *M. albidus* and *Amphimerus pseudofelineus* (Kennedy 2004). Infection with *Ascocotyle longa* resulting from the consumption of under-cooked or raw mullet (Mugil *spp.*) is common in Brazil (Rodriguez *et al.* 2015).

Zoonotic infection with *Clonorchis* spp. is endemic in China, Hong Kong, Japan, the Philippines and Taiwan, affecting between 10% and 48% of the population in some areas (Belizario *et al.* 2004; Ko 2006; Hung *et al.* 2013). Most infections are asymptomatic, but moderate infections may cause diarrhoea and splenomegaly. Where disease is endemic and parasites may be progressively acquired, heavy infection may cause symptoms including choleocystitis, hepatitis and tachycardia which may be fatal. In these areas, even a low proportion of heavily infected individuals may result in increased morbidity (Muller 2002). Infection with *Euclinostomum heterostomum* (Clinostomidae); *Lophosicyadiplostomum* sp. and *Posthodiplostomum* sp. (Diplostomidae); and *Polylekithum* sp. (Allocreadiidae) causes significant zoonotic disease in India (WHO 1995, 2009; Athokpam & Tandon 2012).

The establishment and distribution of digenean pathogens is largely dependent upon the availability of conspecific, or alternative intermediate host species. Potential fish hosts in New Zealand freshwater aquaculture include eels (*Anguilla* spp.), salmon (*Salmo* spp., *Oncorhynchus* spp., and *Salvelinus* spp.) as well as cyprinids including grass carp (*Ctenopharyngodon idella*) in fresh water (Paperna & Dzikowski 2006; Johnston 2008a, 2008b). Other freshwater fish hosts include common carp (*Cyprinus carpio*), catfish (*Ameiurus punctatus*, European perch (*Perca fluviatilis*), goby (*Acanthogobius flavimanus*), goldfish (*Carassius auratus*), grass carp

(*Ctenopharyngodon idella*), yellow perch (*Perca flavescens*), roach (*Rutilus rutilus*), rudd (*Scardinius erythrophthalmus*), silver carp (*Hypophthalmichthys molitrix*), tilapia (*Oreochromis* spp.) and topmouth gudgeon (*Pseudorasbora parva*) (Paulin *et al.* 2001).

Freshwater mollusc species, including *Assiminea* spp., *Bulinus* spp., *Lymnaea* spp., *Melanoides tuberculata, Planorbis* spp. and *Potamopyrgus* spp., may potentially harbour digeneans, while piscivorous birds, including cattle egrets (*Bubulcus ibis*), gulls (*Larus* spp.) and herons (*Ardea* spp.) could function as final hosts (Winterborn 1973; Phillips & Lambert 1989; Paperna & Dzikowski 2006; Johnston 2008a, 2008b). Overseas control treatments have focused on the removal of the intermediate host, as anthelminthic treatments such as praziquantel or bayluscide are restricted-use pesticides (Hung *et al.* 2013).

Little information is currently available to assess the likelihood of introduction of digeneans in marine waters (Paperna & Dzikowski 2006), as the host-pathogen relationships are complex. Although the baitfish species used in Indonesian grouper (*Epinephelus* spp.) mariculture contained digenean larvae, infection was not transferred to *Epinephelus* spp. However, other larval digeneans associated with invertebrate intermediate hosts also present on these sea-cage structures successfully initiated infection in *Epinephelus* spp. (Ruckert *et al*. 2009). The life cycle details, intermediate hosts and host preferences of digenean cestodes are essentially unknown (Li-Po & Lim 2014), so the potential of these marine digeneans to affect fish in sea-cage aquaculture in New Zealand is also unknown.

50.2. Risk assessment

50.2.1. Entry assessment

Digenean life cycles are complex and generally host-specific, at least to the genus level (Paperna & Dzikowski 2006). Fish infected with digenean metacercaria in the musculature show no external signs of infection (Paperna & Dzikowski 2006). These would pass visual inspection and may be present in the commodity (Johnston 2008a).

Encysted digeneans can survive for at least 48 hours following the death and processing of the host (Kahn *et al.* 1999) and are tolerant of the temperatures involved in fish processing and storage (Paperna & Dzikowski 2006; Johnston 2008a).

The likelihood of entry through fresh-chilled eviscerated fish is assessed as non-negligible.

50.2.2. Exposure assessment

Many digeneans have a wide geographical distribution, often because of the range expansion of their host through aquaculture, and the trade in chilled or lightly processed fish for human consumption (Bargues & Mas-Coma 1997; Paperna & Dzikowski 2006). Their establishment and distribution is limited to the availability of usual, or alternative invertebrate hosts (Paperna & Dzikowski 2006; Johnston 2008a).

To establish in New Zealand, infected eviscerated product would have to become available for consumption by a susceptible intermediate invertebrate or fish host, in sufficient quantity and duration (Kahn *et al.* 1999). Encysted digenean larvae may be retained in the trimmings of muscle tissue, as well as being associated with the gills, brain, skin, spinal column and eye tissues discarded as offal after processing (Johnston 2008b).

Digenean larval stages are resistant to environmental extremes. Invertebrates could potentially act as intermediate hosts, while piscivorous birds could act as alternative final hosts, as well as acting as a distribution mechanism between waterways (Paperna & Dzikowski 2006; Johnston 2008a, 2008b).

Infection can be initiated by a low number of parasites and the replication inherent in digenean life cycle can result in the rapid development of disease, particularly under aquaculture conditions (Paperna & Dzikowski 2006).

The likelihood of exposure to digenean parasites is assessed as non-negligible.

50.2.3. Consequence assessment

Over 60 species of digeneans may be zoonotic, including *Ascototyle* spp., *Clonorchis sinensis*, *Euclinostomum* spp., *Haplorchis* spp., *Posthodiplostomum* spp., *Opisthorchis viverrini* and *O. felineus*. While low level infection causing mild-severe diarrhoea and splenomegaly is likely to be under-reported (Athokpam & Tandon 2012), heavy infection causes ccholeocystitis, hepatitis and tachycardia which can be fatal (WHO 1995; Muller 2002; De *et al.* 2003; Ko 2006; WHO 2009; Athokpam & Tandon 2012; Hung *et al*. 2013; Nareaho *et al.* 2017).

Infection in wild fish rarely causes significant disease (Paperna & Dzikowski 2006), but wild stocks may represent a reservoir of infection for farmed conspecific or alternative host fish species (Rosser *et al.* 2016). The economic effect of digenean infection mainly relates to production losses due to contaminated product. The establishment of digeneans, such as *Centrocestus* spp. or *Haplorchis* spp., would cause direct economic losses for freshwater salmonid aquaculture in New Zealand, including Chinook salmon (*O. tshawytscha*) (Johnston 2008a, 2008b). Salmonid aquaculture was valued at $63 million in export earnings in 2011 (Seafood New Zealand 2014).

Freshwater aquaculture in New Zealand largely depends upon the availability of uncontaminated water (Sim-Smith *et al.* 2014), so the requirement for water treatment would represent additional financial consequences of the introduction of digenean pathogens.

The introduction of digenean parasites would also affect recreational and tourist trout and salmon fisheries, as well as causing significant social and environmental costs associated with other domestic fisheries (Fish & Game 2014). While the dollar value of New Zealand's freshwater fisheries is not fully known, the Lake Taupo trout fishery alone is valued at $70–80 million per annum (Marsh & Mkwra 2013).

The major fish aquaculture species in the marine environment is Chinook salmon (*O. tshawytscha*), although aquaculture development continues for snapper (*Pagrus auratus*) and yellowtail kingfish (*Seriola lalandi*) (Diggles 2008; NIWA 2014). While pathogenicity is low, the consequences of establishment for marine species are essentially unknown.

The consequences of establishment are assessed as non-negligible.

50.2.4. Risk estimation

As the entry, establishment and consequence assessments are non-negligible, digenean larvae are considered as a risk in the commodity. Further risk management measures may be justified.

50.3. Risk management

Infection with digenean larvae is not an OIE-listed disease, so the *Aquatic Code* (OIE 2016a) provides no specific guidance on mitigation measures for importing eviscerated fish from infected countries, or specific processing requirements that would ensure the destruction of the pathogen. It is assumed that compliance with the general OIE processing and heat treatment provisions (Appendix 4) would eliminate these digenean larvae from the commodity and be a viable risk management option.

Fish from several families of marine and fresh water fish (Table 47) are hosts of digenean parasites with the potential to cause significant social and economic effects to New Zealand. Species declaration should substantially reduce the pathogen load of these digeneans in the commodity and be a viable risk management option. The incidence in wild-caught fish is generally low (Paperna & Dzikowski 2006; Diggles 2008), so restriction of the commodity to wild-caught fish (not from aquaculture) should moderately reduce pathogen load and be a viable risk management option.

As these parasites are widely distributed and infection with digeneans is not an OIE-listed disease, no requirements for dedicated monitoring exist. Where country/zone freedom is approved through the MPI Country Approval Procedures, this option is likely to substantially reduce the pathogen load of digenean larvae in the commodity. Approval of a declaration of country/zone freedom by MPI is a viable risk management option.

Digenean larvae may be present in the head, gill tissues and the musculature (Paperna & Dzikowski 2006; Johnston 2008a, 2008b), so removal of the gills, or the head and gills, would slightly reduce the pathogen load in the commodity. Further processing to the skin-off fillet state would be expected to moderately reduce the pathogen load of larval digeneans in the commodity and be a viable risk management option.

Digenean larvae are inactivated by freezing (to below -20°C for 168 hours) (Johnston 2008a, 2008b; FDA 2017), so frozen storage would eliminate digenean larvae from the commodity and be a viable risk management option.

Digenean larvae are inactivated by moderate heat treatment (by cooking to at least 70°C for 20 minutes) (Borges *et al.* 2015). Heat treatment should eliminate digenean larvae from the commodity and is likely to be a viable risk management option.

50.3.1. Risk management options

Digenean larvae are reported from fish in families Channidae, Cichlidae, Clupeidae, Cyprinidae, Eleotridae, Esocidae, Ictaluridae, Lateolabracidae, Mugilidae, Percidae, Salmonidae, Serrasalmidae, Serranidae, Siluridae, Soleidae and Sparidae (Table 47), which are considered likely to be present in the commodity. Other families have not been associated with digenean larvae. Therefore species declaration indicting the commodity is not originated from any of the above families should substantially reduce the occurrence of digenean larvae in the commodity.

For the commodities originated from families associated with digenean larvae, one or a combination of the following additional options could also be considered to effectively manage the risk.

Where country/zone freedom from digenean larvae is accepted by MPI:

Option 1

Acceptance of country/zone freedom by MPI should substantially reduce the occurrence of digenean larvae, so the commodity may be imported without any further restrictions.

Where country/zone freedom from digenean larvae is not accepted by MPI or not available:

Option 2

Processing consistent with the approved states for OIE-listed diseases (Appendix 3) should eliminate digenean larvae. When this provision is met, the commodity could be imported without any further restrictions.

Where the imported commodity is not in a state consistent with the approved states for OIE-listed diseases (Appendix 3):

Option 3

Frozen storage (to below -20°C for 168 hours) should eliminate digenean larvae. When this provision is met, the commodity could be imported without further restrictions; or,

Option 4

Heat treatment (by cooking (to at least 70°C for 20 minutes) should eliminate digenean larvae. When this provision is met, the commodity could be imported without further restrictions; or,

Option 5

Further processing (to the skin-off fillet state) should moderately reduce the occurrence of digenean larvae. When this provision is met, the commodity could be imported without further restrictions; or,

Option 6

Restriction of the commodity to wild-caught fish (not from aquaculture) should moderately reduce the occurrence of digenean larvae. When this provision is met, the commodity could be imported without further restrictions.

50.4. References

Abou-Eisha AM, Saleh RE, Fadel HM, Youseff EM, Helmy YA (2008). Role of freshwater fishes in the epidemiology of some zoonotic trematodes in Ismailia Province (Egypt). *Suez Canal Veterinary Medicine Journal* 13(2): 653–676.

Aleem SO (1987). An assessment of tobacco waste for control of the gastropod *Tympanotonus fuscatus* (Linnaeus) in brackish water fishponds. FAO Working Paper African Regional Aquaculture Centre Working Paper ARAC/87/WP/7. Fisheries and Aquaculture Department, Food and Agriculture Organisation, United Nations, Rome, Italy, 8 p.

Anonymous (2014). *Echinochasmus perfoliatus.* Information sheet, American Association of Veterinary Pathologists [Online]. Available from: http://www.aavp.org/wiki/trematodes-2/trematodes-small-intestine/echinostomatidae/echinochasmus-perfoliatus/ [Accessed 2 December 2014].

Anonymous (2016). Biosecurity (Notifiable Organisms) Order 2016. NZLR 2016/73. New Zealand Government, Wellington, 13 p.

Armignacco O, Caterini L, Marucci G, Ferri F, Bernadini G, Raponi GN, Ludovisi A, Bossu T, Morales MA, Pozio E (2008). Human illnesses caused by *Opisthorchis felineus* flukes, Italy. *Emerging Infectious Diseases* 14(12): 1902–1905.

Arocena R, Rodriguez L (2011). Small-scale structure of macrophytes and macrobenthos in coastal 'Laguna de Rocha'. *Pan-American Journal of Aquatic Sciences* 6(2): 148–160.

Arthur JR, Lumanlan-Mayo S (1997). Checklist of the parasites of fishes of the Philippines. FAO Fisheries Technical Paper 369. Fish Health Section, Asian Fisheries Society, Food and Agriculture Organisation, United Nations, Rome, Italy, 102 p.

Athokpam VD, Tandon V (2015). A survey of metacercarial infections in commonly edible fish and crab hosts prevailing in Manipur, Northeast India. *Journal of Parasitic Diseases* 39(3): 429–440 [Online] Available from: http://www. 10.1007/s12639-013-0360-z [Accessed 9 July 2018].

Bargues MD, Mas-Coma S (1997). Phylogenetic analysis of Lymnaeid snails based on 18S rDNA sequences. Molecular Biology and Evolution 14(5): 569–577.

Belizario VY, De Leon WU, Bersabe MJJ, Baird JK, Bangs MJ (2004). A focus of human infection by *Haplorchis taichui* (Trematoda, Heterophyidae) in the southern Philippines. *Journal of Parasitology* 90(5): 1165–1169.

Berrie AD (1960). The influence of various definitive hosts on the development of *Diplostomum phoxini* (Strigeida: Trematoda). *Journal of Helminthology* 34(3-4): 205–210.

Betterton C (1974). Studies on the host specificity of eye fluke, *Diplostomum spathaceum*, in brown and rainbow trout. *Parasitology* 69: 11-29.

Borges JN, Skov J, Bahool QZM, Moller OS, Kania PW, Santos CP, Buchmann K (2015). Viability of *Cryptocotyle ligula* metacercariae from Atlantic cod (*Gadus morhua*) after exposure to freezing and heating in the temperature range from -80°C to 100°C. *Food Control* 50: 371–377.

Boustead NC (1982). Fish diseases recorded in New Zealand with a discussion on potential sources and certification processes. Fisheries Research Division, new Zealand Ministry of Agriculture and Fisheries, 19 p.

Chappell LH (1995). The biology of diplostomatid eye flukes of fishes. *Journal of Helminthology* 69: 97–101.

Chi TT, Dalsgaard A, Turnbill JF, Yuan PA, Murrell KD (2008). Prevalence of zoonotic trematodes in fish from a Vietnamese fish-farming community. *Journal of Parasitology* 94(2): 423–428.

Cho SH, Kim IS, Hwang EJ, Kim TS, Na BK, Sohn WM (2012). Infection status of estuarine fish and oysters with intestinal fluke metacercaria in Muan-gun, Jeollanam-do, Korea. *Korean Journal of Parasitology* 50(3): 215–220.

Cribb TH, Bray RA, Barker SC (1992). A review of the Family Transversotrematidae (Trematoda: Digenea) with the description of a new Genus, *Crusziella*. *Invertebrate Taxonomy* 6: 909–935.

De NV, Murrell KD, Cong LD, Cam PD, Chau LV, Toan ND, Dalsgaard A (2003). The food-borne trematode zoonoses of Vietnam. *Southeast Asian Journal of Tropical Medicine and Public Health* 34(1-2): 12–34.

Diggles BK 2008. Transfer of parasites between wild and cultured fishes with respect to aquaculture development in the Firth of Thames, New Zealand. DigsFish Services Pty Ltd. (Appendix 2 in Kelly S (2008). Potential environmental effects associated with the proposed shift from mussel to finfish farming in the Firth of Thames. Environment Waikato Technical Report 2008/38, 22 p.).

Doanh PN, Nawa Y (2016). *Clonorchis sinensis* and *Opisthorchis* spp. in Vietnam: current status and prospects. *Transactions of the Royal Society for Tropical Medicine and Hygiene* 110(1):13–20 [Online] Available from: http://www.doi: 10.1093/trstmh/trv103 [Accessed 29 January 2018].

Elnawawi FA, Tawfik MAA, Shaapan RM (2000). Some methods of inactivation or killing of encysted metacercariae in tilapia muscles. *Egyptian Journal of Veterinary Science* 34: 31–38.

European Food Safety Authority (EFSA) (2010). Regulation (EC) N 853/2004 of the European Parliament and Council. Hygiene rules for food of animal origin. *EFSA Journal* 8(4): 1543, 91 p.

Fan PC (1998). Viability of metacercariae of *Clonorchis sinensis* in frozen or salted freshwater fish. *International Journal for Parasitology* 28(4): 603–605.

Fang Y, Dai C, Jun N, Hui M (2003). Study on the effects of freezing and irradiation on the survival of *Clonorchis sinensis* metacercaria in freshwater fish. *China Food Health Journal* 5: 410–411.

Fattakhov RG (1985). Low temperature regimes for treating fish containing *Opisthorchis* larvae. *Meditinskaya Parazitologiya i Parazitarnye Bolezni* 5: 63–64.

Fattakhov RG (1989). The effect of low temperatures on the viability of *Opisthorchis felineus* metacercariae. *Meditinskaya Parazitologiya i Parazitarnye Bolezni* 6: 37–38.

FDA (2017). Safe practices for food processes. Processing parameters needed to control pathogens in cold smoked fish, Chapter 5. Potential hazards in cold smoked fish: Parasites. Food and Drug Administration, Washington D.C., United States of America [Online] Available from: http://www.fda.govt/Food/FoodScience/SafePracticesFor FoodProcesses/ucm094578.htm [Accessed 20 January 2017].

Fish and Game (2014). New Zealand Fish and Game Council [Online] Available from: http://www.fishandgame.org.nz/ [Accessed 24 September 2014].

Hamed MGE, Elias AN (1970). Effect of food processing methods upon survival of the trematode *Heterophyes* sp. in flesh of mullet caught in brackish Egyptian waters. *Journal of Food Science* 35: 386.

Hegazi MA, Hassan AT, Al-Nashar TM, Abo-Elkheir OI, El-Lessi FM (2014). Encysted metacercariae of family Heterophyidae in infected fish in Dakahlia Governorate, an endemic focus in Egypt. *Journal of the Egyptian Society for Parasitology* 44(3):547–558.

Hine PM. Jones JB, Diggles BK (2000). A checklist of the parasites of New Zealand fishes, including previously unpublished records. *NIWA Technical Report* 75. National Institute of Water and Atmospheric Research Ltd., Auckland, New Zealand, 93 p.

Hung NM, Madsen H, Fried B (2013). Global status of fish-borne zoonotic trematodiasis in humans. *Acta Parasitologica* 58(3): 231–258.

Johnston C (2008a). Import Risk Analysis: Frozen skinless and boneless fillet meat of *Oreochromis* spp. from China and Brazil for human consumption. Biosecurity New Zealand, Ministry of Agriculture and Forestry, Wellington, New Zealand, 96 p.

Johnston C (2008b). Import Risk Analysis: Frozen skinless and boneless fillet meat of *Pangasius* spp. fish from Vietnam for human consumption. Biosecurity New Zealand, Ministry of Agriculture and Forestry, Wellington, New Zealand, 60 p.

Kahn SA, Beers PT, Findlay VL, Peebles IR, Durham PJ, Wilson DW, Gerrity SE (1999). Import risk analysis on non-viable salmonids and non-salmonid marine finfish. Australian Quarantine and Inspection Service, Department of Agriculture, Fisheries and Forestry, Canberra, Australia, 409 p.

Kajiyama M, Kajiyama M, Suzuki N (1979). Studies on *Pharyngostomum cordatum* Diesling 1850. 1: The first intermediate host in Japan. *Japanese Journal of Parasitology* 284: 235.

Kennedy M (2004). Flatworm Cysts in Fish: the North American Liver Fluke Alberta Agriculture and Forestry, Government of Alberta, Canada [Online] Available from: https://www.agriculture.alberta.ca/app21/ministrypage [Accessed 10 August 2018].

Ko RC (2006). Fish-borne parasitic zoonoses. pp 592–628. *In*: Woo PTK (*Ed.*) Fish Diseases and Disorders, Volume 1: Protozoan and Metazoan Infections. CAB International, Wallingford, United Kingdom, 791 p.

Krailas D, Namchote S, Koonchornboon T, Dechruksa W, Boonmekam D (2014). Trematodes obtained from the thiarid freshwater snail *Melanoides tuberculata* (Müller, 1774) as vector of human infections in Thailand. *Zoosystematics and Evolution* 90(1): 57–86.

Li MH, Huang HI, Chen PL, Huang CH, Chen YH, Ooi HK (2013). *Metagonimus yokogawai*: metacercariae survey in fishes and its development to adult worms in various rodents. *Parasitological Research* [Online] Available from: http://www.doi.10.1007/s00436-013-3320 [Accessed 10 August 2018].

Li-Po GD, Lim LHS (2014). Infectious diseases of warmwater finfish in fresh water. pp 193–252. *In:* Woo PTK, Bruno DW (*Eds.*). Diseases and disorders of finfish in cage culture, 2nd edition. CABI International, Wallingford, United Kingdom, 354 p.

Lobna SM, Metawea YF, Elsheikha HM (2010). Prevalence of heterophyiosis in Tilapia fish and humans in Northern Egypt. *Parasitology Research* 107(4): 1029–1034.

Marsh D, Mkwra L (2013). Review of freshwater non-market value studies. Department of Economics, University of Waikato, Hamilton, New Zealand, 144 p.

Mayo-Hernandez E, Peñalver J, García-Ayala A, Serrano E, Muñoz P, Ruiz de Ybáñez R (2015). Richness and diversity of helminth species in eels from a hypersaline coastal lagoon, Mar Menor, south-east Spain. *Journal of Helminthology* 89(3): 345–351.

Melo FT, e Silva JP, Costa-Goncalves E, Furtado AP, Giese EG, Santos CP, Santos JN (2015). Taxonomic status and redescription of the genus *Brasicystis* Thatcher, 1979 (Digenea: Didymozoidae). *Parasitology International* 62: 201–214.

Muller R (2002). Worms and human disease (2nd edition). CABI International, Wallingford, United Kingdom, 310 p.

Musiba MJ, Nkwengulila G (1998). Occurrence of metacercariae of *Diplostomum* and *Tylodelph*ys species (Diplostomidae) in *Clarias* species (Clariidae) from Lake Victoria. *Tanzanian Journal of Science* 32(1): 88–98.

Nareaho A, Eriksson-Kallio AM, Heikken P, Snellman A, Sukura A, Koski P (2017). High prevalence of zoonotic trematodes in roach (*Rutilus rutilus*) in the Gulf of Finland. *Acta Veterinaria Scandanavica* 59: 75 [Online] Available from: http://www.doi.10.1186/s13028-017-0343-7 [Accessed 20 July 2018].

NIWA (2014) Yellowtail kingfish [Online] Available from: https://www.niwa.co.nz/aquaculture/aquaculture-species/yellowtail-kingfish [Accessed 24 April 2015].

Noda K (1959). The larval development of *Stellantchasmus falcatus* (Trematoda: Heterophyidae) in the first intermediate host. *Journal of Parasitology* 45(6): 635–642.

OIE (2016). Aquatic Animal Health Code, 19th Edition. World Organization for Animal Health. Paris, France [Online] Available from: http://www.oie.int/international-standard-setting/aquatic-code/access-online/ [Accessed 15 March 2017].

Ozer SA, Kirca Y (2015). Parasite fauna of the grey mullet *Mugil cephalus* L. 1758, and its relationship with some ecological factors in Lower Kızılırmak Delta located by the Black Sea, Turkey. *Journal of Natural History* 49 (15-15): 933–956.

Paperna I, Dzikowski R (2006). Digenea (Phylum Platyhelminthes), pp 345-390 *In*: Woo PTK (*Ed.*) Fish Diseases and Disorders, Volume 1: Protozoan and Metazoan Infections. CAB International, Wallingford, United Kingdom, 791 p.

Paulin C, Stewart A, Roberts C, McMillan P (2001). New Zealand Fish: A complete guide. Te Papa Press, Wellington, New Zealand, 279 p.

Petney TN, Andrews RH, Saijuntha W, Wenz-Mucke A, Sithithaworn P (2013). The zoonotic, fish-borne liver flukes *Clonorchis sinensis*, *Opisthorchis felineus* and *Opisthorchis viverrini. International Journal of Parasitology* 43 (12–13): 1031–1046.

Phillips NR, Lambert DM (1989). Genetics of *Potamopyrgus antipodarum* (Gastropoda: Prosobranchia): evidence for reproductive modes. *New Zealand Journal of Zoology* 16: 435–445.

Rácz OZ, Zemankovics E (2002). Survival of metacercariae of *Metagonimus yokogawai* (Digenea: Heterophyidae) on fish from River Danube. *Magyar Allatorvosok Lapja* 124(7): 437–444.

Reynard R, Wahli T, Vatsos I, Mortensen S (2007). Review of disease interactions and pathogen exchange between farmed and wild finfish and shellfish in Europe. VESO Report 1655 produced for the European Commission. IFREMER, Paris, France, 460 p.

Rim HJ, Kim KH, Joo KH, Kim SJ, Eom KS, Chung MS (1996). The infestation states and changing patterns of human infecting metacercariae in freshwater fish in Kyongsang-do and Kyonggi-do, Korea. *Korean Journal of Parasitology* 34(2): 95–105.

Roberts RI (2001). The parasitology of teleosts, pp. 254-296 *In:* Roberts RI (*Ed*.), Fish Pathology, Saunders, London, United Kingdom, 592 p.

Rodrigues MV, de Pérez ACA, Machado TM, Orisaka FM, Kurissio JK, Lafisca A (2015). Research of *Ascocotyle* (*Phagicola*) *longa* in Heat Treated Fillets of *Mullet* (*Mugil platanus*). *Fisheries and Aquaculture Journal* 6: 115 [Online] Available from: http://www.doi:10.4172/2150-3508.10000115 [Accessed 27 July 2018].

Rosser TG, Baumgartner WA, Alberson NR, Woodyard ET, Reichley SR, Wise DJ, Pote LM, Griffin MJ (2016). *Austrodiplostomum* sp., *Bolbophorus* sp. (Digenea: Diplostomidae), and *Clinostomum marginatum* (Digenea: Clinostomidae) metacercariae in inland silverside *Menidia beryllina* from catfish aquaculture ponds, with notes on the infectivity of *Austrodiplostomum* sp. cercariae in channel catfish *Ameiurus punctatus*. *Parasitological Research* 115(11): 4365–4378.

Ruckert S, Klimpel S, Al-Quraishy S, Melhorn H, Palm HW (2009). Transmission of fish parasites into grouper mariculture (Serranidae: *Epinephelus coioides* (Hamilton 1822)) in Lampung Bay, Indonesia. *Parasitological Research* 104: 523–532.

Santos MA, peixoto JS, Madi RR, da silva Esposito T (2018). Protozoan and metazoan parasites of juvenile tambaqui *Colossoma macropomum* farmed in the Lower São Francisco, Brazil. *Acta of Fisheries and Aquatic Resources* 6(1): 29–34.

Seafood New Zealand (2014). Seafood New Zealand: Key facts for our Industry. Seafood New Zealand, Wellington, New Zealand, 6 p.

Shalaby SL, Anter O, Hassan N, El- Mahdy M, Mohamed I, Gupta N (2006). Clinical manifestations of experimentally induced fish-borne trematode infection in experimental animals. *International Journal of Current Science* 9: 443– 448.

Scholz T (1999). Taxonomic study of *Ascocotyle* (*Phagicola*) *longa* Ransom, 1920 (Digenea:Heterophyidae) and related taxa. *Systematic Parasitology* 43: 147–158.

Simoes SBE, Barbosa HS, Santos CP (2009). The life history of *Pygiopsis macrostomum* (Travassos 1928) (Digenea: Heterophyidae). *Memoirs of the Oswald Cruz Institute* 104(1): 106–111.

Sim-Smith C, Faire S, Lees A (2014). Managing biosecurity risk for business benefit. Report prepared for the Ministry for Primary Industries by Coast and Catchment Ltd, Auckland, New Zealand, 214 p.

Slifco TR, Smith HV, Rose JB (2000). Emerging parasite zoonoses associated with water and food. *International Journal for Parasitology* 30: 1379–1393.

Venable DL, Gaudé AP, Klerks PL (2000). Control of the trematode *Bolbophorus confusus* in channel catfish *Ameiurus punctatus* in ponds using salinity manipulation and polyculture with black carp *Mylopharyngodon piceus*. *Journal of the World Aquaculture Society* 31: 158-166.

Vo, DV, Murrell D, Dalsgaard A, Bristow G, Nguyen D, Bui T, (2008). Prevalence of Zoonotic Metacercariae in Two Species of Grouper, *Epinephelus coioides* and *Epinephelus bleekeri*, and Flathead Mullet, *Mugil cephalus*, in Vietnam. *The Korean Journal of Parasitology* 46: 77–82 [Online] Available from: http://www.doi.10.3347/kjp.2008.46.2.77 [Accessed 10 July 2018].

Wells WH, Randall BH (1955). New hosts of the genus *Heterophyes* in Egypt. *The Journal of Parasitology* 42(3): 287–292.

WHO (1995). Control of food borne trematode infections. WHO Technical Report 849. World Health Organisation of the United Nations, Rome, Italy, 157 p.

WHO (2009). Foodborne trematode infections and taeniasis/cysticercosis. Report of the WHO expert consultation, Vientiane, Lao PDR, World Health Organisation of the United Nations, Rome, Italy, 50 p.

Winterborn MJ (1973). A guide to the freshwater Mollusca of New Zealand. *Tuatara* 20(3): 141–158.

Wiwanitkit V, Nithiuthai S, Suwansaksri J, Chongboonprasert C, Tangwattakanont K (2001). Survival of heterophyid metacercariae in uncooked Thai fish dishes. *Annals of Tropical Medicine and Parasitology* 95(7): 725–727.

Yousef M, Mansour NS, Hammouda NA, Awadalla HN, Boulos LM (1981). Effect of freezing and grilling on *Pygidiopsis genata* metacercariae in Tilapia. *Journal of the Egyptian Society of Parasitology* 11(2): 425–428.

51. Cestode pathogens (muscle-encysting larvae)

51.1. Hazard identification

51.1.1. Aetiological agent

Cestodes are classified within the Class Cestoda of Phylum Platyhelminthes (Dick *et al*. 2006). Fish commonly function as intermediate, or paratenic hosts, while the adult stages infect piscivorous birds and mammals, including humans. Cestodes have a complex life cycle, but many larval stages are incompletely known, or incorrectly described (Dick *et al.* 2006). Cestodes of economic importance affecting fish occur in Orders Amplilinidea, Caryophyllidea, Pseudophyllidea, Proteocephalidea, Tetraphyllidea and Tetrarhyncha (Dick *et al.* 2006).

51.1.2. OIE status

Infection with cestode pathogens is not listed by the OIE (OIE 2016).

51.1.3. New Zealand status

Cestode fish pathogens occur in New Zealand (Hine *et al.* 2000), but many species causing clinical disease of economic importance in fish, or of zoonotic concern, are regarded as exotic (Boustead 1982; Hine *et al.* 2000; Johnston 2008a, 2008b).

Infection with cestode parasites is not a notifiable disease in New Zealand (Anon. 2016).

51.1.4. Epidemiology

The cestode life cycle involves at least three hosts. Host specificity can be variable, involving a wide range of species that can function as alternative and paratenic intermediate hosts (Dick *et al.* 2006). Maturity may occur in several alternative, but usually related species (such as within Elasmobranchs, piscivorous birds, or marine mammals (Scholz & Kuchta 2016)). The introduction of exotic cestode pathogens can significantly alter the topology of food webs, often with complex and unexpected outcomes at the population ecology level (Amundsen *et al.* 2013).

The first intermediate host is usually a copepod crustacean (such as *Acanthodiaptomus*, *Arctodiaptomus*, *Boeckella*, *Cyclops*, *Diaptomus*, *Eudiaptomus*, *Eurytemora*, and *Mesocyclops*), while others may be invertebrates or vertebrates, including fish (Felix 2013).

For cestode species where the intermediate stages occur in fish, the adult stage usually occurs in the gut, kidneys or gonads of piscivorous birds or mammals (Dick *et al.* 2006). Cestode eggs are released with the host faeces or secretions, or less commonly, after the death of the definitive host. The eggs hatch, releasing a free-swimming (coracidium) larvae which infects an invertebrate (such as *Cyclops* spp., *Diaptomus* sp., or *Eudiaptomus* spp.) and develops into a procercoid within the haemocoel. Upon consumption by a fish, this migrates from the gut, through the circulatory system to encyst as an unencapsulated plerocercoid larvae either in an organ system, or the musculature. Following consumption by the final host (usually piscivorous birds, such as gulls (*Larus* spp.), or marine mammals, such as fur seals (*Arctocephalus* spp.)), it develops into the adult stage within 2-3 months (Heynemann 1996; Dick *et al.* 2006; Scholz *et al.* 2009). Alternative hosts include dogs (*Canis* sp.), or humans, following consumption of raw or under-cooked fish (Torres *et al.* 2004; Kuchta *et al.* 2013, Shamsi *et al.* 2018).

If the fish host is consumed by another fish, the plerocercoid migrates from the viscera, to encyst in the musculature of its new paratenic host (Dick *et al.* 2006; Scholz *et al.* 2009). Cestodes commonly develop in a wide range of fish, although some species appear to be preferred hosts (Dupont & Gabrion 1987). The incidence in wild-caught fish is generally low, but occurrence may be high in farmed fish (Dick *et al.* 2006). Cestodes are effectively limited in distribution to either marine or fresh waters by the salinity tolerance limits of their hosts (Dick *et al.* 2006). Where the definitive host is a land vertebrate, the fish intermediate host occurs in freshwaters, coastal inshore or estuarine waters (Dick *et al.* 2006). Cestodes infecting widely distributed fish hosts, such as mullet (*Mugil cephalus*), generally have the widest range of alternative intermediate hosts (Dick *et al.* 2006).

Infection with proteocephalid cestodes do not generally cause economic effects, while cestode species infecting oceanic deep-water and pelagic fish usually have a cosmopolitan distribution, including New Zealand (Hine *et al.* 2000) that matches the range of their hosts (Dick *et al*. 2006). They are not considered further.

Pseudophyllidean cestodes including the genera *Diphyllobothrium*, *Triaenophorus* and *Ligula* cause stunting of their host, particularly in aquaculture (Dick *et al.* 2006; Ehab & Faisal 2008). While mortality is rare, infection of farmed fish resulted in lower growth rates, when compared to uninfected fish, and this growth differential increased over time (Saksvik *et al*. 2001; Dick *et al.* 2006).

A representative, but not exhaustive, list of major muscle-infecting cestode parasites (Order and Species) and their fish intermediate hosts (Family and Species) with the potential to infect farmed fish in New Zealand is given in Table 48 (Overstreet 1977; Peterson *et al.* 1983; Korting 1984; Moravec 1985; Durborow *et al.* 1988; Nawa *et al.* 1995; Ibrahim 2000; Rausch & Adams 2000; Kino *et al.* 2002; Khanna 2004; Chai *et al.* 2005; Dick *et al.* 2006; Wicht *et al.* 2008; Scholz *et al.* 2009, 2012; Felizaro *et al.* 2010; Haseli *et al.* 2010; Justine *et al.* 2010, 2012; D'Silva et al. 2012; Zarger *et al*. 2012; Morsey *et al.* 2013; Ahmadiara *et al.* 2014; Beveridge *et al.* 2014; Bhuiyan *et al*. 2014; Soroka *et al.* 2014; Mayo-Hernandez *et al.* 2015; Kuchta *et al.* 2015; Scholz & Kuchta 2016; Waeschenbach *et al*. 2017).

Plerocercoids lack definitive adult features and historically have been poorly classified, or mis-classified (Kuchta *et al.* 2014). Many records are reported as *Scolex pleuronectes*, which indicates the plerocercoid has been provisionally identified to Order or Family level and the species remains undescribed (Jensen & Bullard 2010; Kuchta *et al.* 2014).

Cestodes within orders Amphilinidea, Caryophyllidea, Pseudophyllidea and Proteocephalidea affect fish and may cause zoonotic disease (Chung *et al.* 1995; Dick *et al.* 2006; Scholz 1997; Kuchta *et al.* 2013; Scholtz & Kuchta 2016). While muscle-encysting larvae may cause significant product spoilage in fish, they appear under-reported and under-diagnosed in many countries (Dick *et al.* 2006).

Cestode eggs are highly resistant to environmental extremes, while plerocercoid larvae are unaffected by the temperatures likely to be encountered during the routine storage and processing of chilled fish.

The optimum survival temperature range (28°C to 38°C) for intramuscular *Diphyllobothrium latum* plerocercoids corresponds with the mean summer temperature of the host fish and variations in temperature from this range reduces survival time (Wardle 1933). Plerocercoids remained viable across a wide temperature range (from −8° to 55°C) when bathed in 0·2 molar sodium chloride

Table 48. Representative Families and Species of Fish Susceptible to Muscle-Encysting Cestodes

Host Family	Host Species	Cestode Species
Anguillidae	American eel (*Anguilla rostrata*), European eel (*A. anguilla*)	*Bothriocephalus claviceps, Diphyllobothrium* sp.
Clupeidae	Ilish (*Tenualosa ilisha*), Japanese anchovy (*Engraulis japonica*), Japanese sardine (*Sardinops melanostictus*)	*Diphyllobothrium balaenopterae, Otobothrium* spp.
Cyprinidae	Bream (*Abramis brama*), carp (*Cyprinus carpio*)	*Digramma interrupta, Otobothrium* spp., *Triaenophorus nodulosus*
Esocodae	Pike (*Esox lucius*)	*Triaenophorus nodulosus*
Lotidae	Burbot (*Lota lota*)	*Triaenophorus nodulosus*
Mullidae	Red mullet (*Mullus barbatus*)	*Nybelinia bisulcata*
Paralichthyidae	Flounder (*Paralichthys isoceles*), bay whiff (*Citharichthys spilopterus*), lenguado (*Xystreurys rasile*), Patagonian flounder (*Paralichthys patagonicus*)	*Diphyllobothrium* sp., *Heteronybelinia nipponica, .Nybelinia lingualis, Otobothrium* spp., *Pterobothrium kingstoni, P. crassicolle*
Percidae	Creole perch (*Percichthys trucha*), European perch (*Perca fluviatilis*), pikeperch (*Sander lucioperca*), ruffe (*Gymnocephalus cerna*), *sauger* (*S. canadensis*) walleye (*S. vitreus*), yellow perch *(P. flavescens)*	*Bothriocephalus claviceps, .Dibothriocephalus dendriticus, D. latus, Triaenophorus nodulosus*
Salmonidae	Atlantic salmon, Pacific salmon (*Salmo* spp., *Oncorhynchus* spp.), brook trout (*Salvelinus fontinalis*), char (*Salvelinus* spp.), cisco (*Coregonus sardinella*), whitefish (*Coregonus* sp.)	*Dibothriocephalus ursi, D. dalliae, D. dendriticus, Poecilancistrium caryophyllum, P. dipsacum, Pyramicocephalus phocarum, Triaenophorus nodulosus, T. crassus*
Sciaenidae	Atlantic croaker (*Micropogonias undulatus*), black drum (*Pogonias cromis*), cassava croaker (*Pseudotolithus senengalensis*), red drum (*Sciaenops ocellata*), sand weakfish (*Cynoscion arenarius*), silver perch (*Bairdiella chrysura*), Southern kingcroaker (*Menticirrhus americanus*), Spotted weakfish (*Cynoscion nebulosa*)	*Poecilancistrium caryophyllum*
Scombridae	Skipjack tuna (*Katsuwonas pelamis*), Atlantic Spanish mackerel (*Scomberomorus maculatus*)	*Diphyllobothrium balaenopterae*, *Otobothrium* spp.
Sparidae	Red porgy *(Pagrus pagrus)*	*Kotorella pronosoma, Nybelinia narinari, Pseudogrillotia* sp.

solution), surviving for 60 hours at 20°C. They are unaffected by chilling (surviving for over 24 hours at 0°C) (Wardle 1933), but are denatured by heat treatment (to at least 60°C for at least 1 minute (EFSA 2010), or to at least 54°C for 15 minutes (Wardle 1933).

Plerocercoids are inactivated by frozen storage (to below -18°C for 24 hours) (Salminen 1970), but are unaffected by stomach acid and digestive juices for the length of time they would normally be subject to canine or human gastric and duodenal digestion (Wardle 1933). The plerocercoids are likely to remain viable in the trimmings of fish musculature discarded during commercial processing. Most *Diphyllobothrium* species show low host specificity at the adult stage, so humans may become infected with species that would normally mature in marine mammals or piscivorous birds (Scholz *et al.* 2009). Where these waste products are consumed by piscivorous birds, the life cycle may be completed in these alternative hosts (Dick *et al.* 2006).

Plerocercoids are unaffected by cold-smoke or brine processing (FDA 2011), but infection risk is eliminated if the fish is fried, boiled, or adequately smoked (Salminen 1970). Low dose irradiation (0.15 kGy or less) may prevent further development of the plerocercoid larvae in fish products, but significantly higher levels (up to 10kGy) may be necessary to ensure inactivation (FDA 2011, 2017). In general terms, the FDA recommendation for all fishery product likely to be consumed raw is that the product be frozen (to below -20°C for 7 days, or to below -35°C for 15 hours) prior

to salting or brine processing. The cold-storage time should be further extended for larger fish (i.e. where flesh thickness exceeds 15 cm), to ensure tissue penetration and destruction of the pathogen (FDA 2011, 2017).

Bothriocephalus spp. cestodes are generally non-specific for intermediate hosts, including cyclopoid and calanoid copepods. Potential marine and freshwater hosts occur in New Zealand, including cyclopoid copepods (*Cyclops* spp., *Diacyclops* spp.) and calanoid copepods, as well as *Boeckella* spp., *Skistodiaptomus pallidus*, *Sinodiaptomus valkanovi*, and *Daphnia dentifera* introduced through the ornamental aquarium trade (Duggan *et al.* 2010). *Sinodiaptomus valkanovi* is a known host for the endemic cestode *Ligula intestinalis* (Webber *et al.* 2010).

Human infection (diphyllobothriasis) is an emerging risk internationally, largely due to the increased consumption of raw salted, fermented marinated, under-cooked or fresh-chilled fish (Nawa *et al.* 1995; FDA 2011, 2017; Kuchta *et al.* 2017). Cestode infection has not been reportable in the United States since 1983 (Ruttenber *et al.*1984), but the increasing occurrence in the Asia-Pacific region suggests 20 million human cases may currently occur worldwide (Scholz *et al.* 2009).

Potential marine and freshwater fish hosts, including introduced cichlids, cyprinids, percids and salmonids are widely distributed in New Zealand (Paulin *et al.* 2001).

51.2. Risk assessment

51.2.1. Entry assessment

Plerocercoid larvae commonly occur in the musculature and organ systems of a wide range of marine and freshwater fish. The incidence is generally low in wild fish, but may be higher for farmed stocks (Dick *et al.* 2006). Infected fish generally show no external signs of infection (Dick *et al.* 2006). These would pass visual inspection and may be present in the commodity.

The likelihood of entry is assessed as non-negligible.

51.2.2. Exposure assessment

Cestode parasites have a wide geographical distribution, often because of the range expansion of their host through aquaculture, and in the increased popularity of chilled or lightly processed fish for human consumption (Dick *et al.* 2006).

Plerocercoid larvae remain viable in the temperature range (0°C to 4 °C) associated with storage and transport of the commodity They survive for at least 48 hours in fish tissues following the death of the host, and are unaffected by cold-smoking or brine processing techniques (FDA 2011, 2015). They actively migrate during storage and processing (Dick *et al.* 2006; FDA 2011, 2017) and may be present in muscle, gill, brain, skin, spinal column and eye tissue (Dick *et al.* 2006).

Cestode larval stages are resistant to environmental extremes and exposure may occur through several pathways. To establish an infection in New Zealand, fish product infected with viable plerocercoid larvae would have to be consumed by a susceptible final host (such as a piscivorous bird, or marine mammal) in sufficient quantity and duration (Kahn *et al.* 1999). The cestode would then have to release eggs that could infect a suitable invertebrate host (Dick *et al.* 2006). Potential crustacean hosts such as *Cyclops* spp., *Boeckella* spp. and *Sinodiaptomus valkanovi* occur in New Zealand waters (Duggan *et al.* 2010; Webber *et al.* 2010).

Piscivorous birds may act as alternative final hosts (Scholz *et al.* 2009), as well as acting as a distribution mechanism between waterways (Dick *et al.* 2006). Cestodes can use paratenic hosts to increase their host range and their geographical distribution (Dick *et al.* 2006). While alternative fish intermediate hosts are present in fresh, brackish and coastal marine waters of New Zealand, it is unlikely that exotic cestodes associated with deep-water marine species would be exposed in sufficient quantities and occurrence to establish and maintain infection through the human consumption pathway. These are not considered further.

The likelihood of exposure through the commodity is assessed as non-negligible.

51.2.3. Consequence assessment

Cestodes such as *Diphyllobothrium latum* may cause zoonotic infection where humans represent an accidental paratenic host (Dick *et al.* 2006). While some cestodes can mature in humans, the ingestion of plerocercoid larvae can also trigger allergenic processes (Pelayo *et al.* 2009; Felizardo *et al.* 2010; Abdelsalam *et al*. 2016). The incidence of such infection is increasing globally, due in part to the increase in consumption of raw or partly-processed fish products (Kuchta *et al*. 2017).

The presence of plerocercoid larvae in the commodity may have direct and indirect economic consequences for fisheries and aquaculture in New Zealand, including product rejection due to larval contamination (Dick *et al.* 2006). As the incidence in wild fishstocks is likely to be low, the consequence of introduction in capture (non-aquaculture) fisheries is likely to be negligible.

Pseudophyllid cestodes can have a significant indirect economic effect by reduction in the growth rate of infected fish (Saksvik *et al*. 2001; Dick *et al.* 2006). They may also become pathogenic at high levels of infection (Ehab & Faisal 2008).

Freshwater aquaculture in New Zealand largely depends upon the availability of uncontaminated water (Sim-Smith *et al.* 2014). The establishment of cestodes such as *Triaenophorus crassus* or *Diphyllobothrium latum* would cause direct economic losses for freshwater salmonid aquaculture in New Zealand, including Chinook salmon (*O. tshawytscha*) (Johnston 2008a, 2008b). Salmonid aquaculture was valued at $63 million in export earnings in 2011 (Seafood New Zealand 2014). The introduction of exotic cestode parasites would also affect recreational and tourist trout and salmon fisheries (*Salmo* spp., *Oncorhynchus* spp. and *Salvelinus* spp.) fisheries, as well as incur significant social and environmental costs associated with other domestic fisheries (Fish & Game 2014). While the dollar value of New Zealand's freshwater fisheries is not fully known, the Lake Taupo fishery alone is valued at $70–80 million per annum (Marsh & Mkwra 2013).

The introduction of exotic cestodes may also affect other significant freshwater species. Freshwater eels (*Anguilla dieffenbachii, A. australis, A. reinhardti*) are iconic and highly valued species, with a commercial fishery valued at $4.9 million in 2010 (Jellyman 2012). Eels are considered stressed at current commercial catch levels (MPI 2014), but also support significant non-commercial and subsistence fisheries (MPI 2014). They are considered a taonga by Maori (Parliamentary Commissioner for the Environment 2013) and support a developing aquaculture industry (Watene 2003; NIWA 2017).

Exotic cestodes may establish in freshwater percids including European perch (*Perca fluviatilis*) and cyprinids including common and ornamental carp (*Cyprinus carpio*), goldfish (*Carassius auratus*), tench (*Tinca tinca*), rudd (*Scardinius erythrophthalmus*) and orfe (*Leuciscus idus*) which are widely distributed in New Zealand waterways. These could act as reservoirs of disease for significant freshwater fisheries.

Cestodes may also affect marine aquaculture species, notably sea-run Chinook salmon (*O. tshawytscha*) as well as the developing aquaculture for yellowtail kingfish (*Seriola lalandi*) and snapper (*Sparus aurata*) (Diggles 2008; Plant & Food 2016), but the economic consequences are essentially unknown.

The establishment of exotic cestodes would have significant social, economic and human health consequences. The consequences of establishment are assessed as non-negligible.

51.2.4. Risk estimation

Since the entry, exposure and consequence assessments are non-negligible, muscle-encysted cestode larvae are assessed as representing a risk in the commodity and risk management measures may be justified. The risks associated with the introduction of cestode pathogens in wild (capture) fisheries is assessed to be negligible. However, the entry, exposure and consequence assessments for cestode pathogens affecting farmed fish species of families Anguillidae, Cyprinidae, Salmonidae and Sparidae (Table 48) are non-negligible, and so the risk is estimated to be non-negligible. Therefore, cestode pathogens affecting these fish families are assessed as being risks in the commodity.

The incidence of plerocercoid larvae is likely to be low in wild-caught marine fish, such as from families Clupeidae, Esocidae, Lotidae, Mullidae, Paralichthyidae, Percidae and Scombridae (Table 48), so risks associated with these fish are assessed as negligible.

Fish of families Anguillidae, Cyprinidae, Percidae, Salmonidae and Sparidae (Table 48) are assessed as being risks in the commodity, so further management measures may be justified.

51.3. Risk management

Infection with cestode larvae is not an OIE-listed disease, so the Aquatic *Code* (OIE 2016), provides no specific guidance on mitigation measures for importing eviscerated fish from infected countries, or specific processing requirements that would ensure the destruction of the pathogen. It is assumed that compliance with the general OIE processing and heat treatment provisions (Appendix 4) would eliminate exotic cestode larvae from the commodity and be a viable risk management option.

While several families of fish susceptible to exotic cestodes may be present in the commodity (Table 48), farmed fish of families Anguillidae, Cyprinidae, Percidae, Salmonidae and Sparidae are considered likely to host larval cestodes likely to cause significant social and economic effects in New Zealand. Species declaration would substantially reduce the pathogen load of larval cestodes in the commodity and be a viable risk management option.

Infection with cestode larvae is uncommon in wild-caught fish (Dick *et al*. 2006), so restriction of the commodity to wild-caught fish (not from aquaculture) should moderately reduce pathogen load and be a viable risk management option.

Exotic cestode larvae affecting fish of families Anguillidae, Cyprinidae, Percidae, Salmonidae and Sparidae are widely distributed (Dick *et al.* 2006). Where country/zone freedom is approved through the MPI Country Approval Procedures, this option should substantially reduce the pathogen load of cestode larvae in the commodity. Approval of a declaration of country/zone freedom by MPI is a viable risk management option.

Plerocercoid larval cestodes may encyst in any organ system as well as in the musculature of fish (Dick *et al.* 2006). They may also move from the viscera into the musculature post-mortem, but before fish are processed (Shamsi *et al.* 2018). Removal of the gills, or removal of the head and gills would be likely to have no effect on the occurrence of these muscle-encysting cestodes in the commodity. As these larvae may remain viable in the musculature, further processing (to the skin-off fillet state) would only slightly reduce pathogen load, but is unlikely to eliminate them from the commodity. Further processing to the skin-off fillet state is not a viable risk management option.

Muscle-encysting cestode larvae are inactivated by heat treatment (by cooking to at least 60ºC for at least 1 minute) (EFSA 2010), or by frozen storage (to below -20°C for 168 hours) (FDA 2011, 2017). Either heat treatment or frozen storage would eliminate these muscle-encysting cestodes from the commodity and be viable risk management options.

Other processing methods, including salting, cold-smoking and pickling are likely to have little or no effect on the occurrence of muscle-encysted larval cestodes (Salminen 1970; FDA 2011) and not considered viable risk management options.

Muscle-encysting cestode larvae are also inactivated by high dose (up to 10kGy) irradiation (FDA 2011, 2017). While this would also eliminate these pathogens from the commodity, this treatment is not generally available and is unlikely to be a viable risk management option.

51.3.1. Risk management options

Muscle-encysting cestode larvae are reported from fish in families Anguillidae, Cyprinidae, Percidae, Salmonidae and Sparidae (Table 48), which are considered likely to be present in the commodity. Other families have not been associated with these cestode larvae. Therefore species declaration indicating the commodity is not originated from any of the above families should substantially reduce the occurrence of muscle-encysting cestode larvae in the commodity.

For the commodities originated from families associated with muscle-encysting cestode larvae, one or a combination of the following additional options could also be considered to effectively manage the risk.

Acceptance of country/zone freedom from muscle-encysting cestode larvae by MPI:

Option 1

Acceptance of country/zone freedom by MPI should substantially reduce the occurrence of muscle-encysting cestode larvae, so the commodity may be imported without any further restrictions.

Where country/zone freedom from muscle-encysting cestode larvae is not accepted by MPI or not available:

Option 2

Processing to a state consistent with the approved states for OIE-listed diseases (Appendix 3) should also eliminate muscle-encysting cestode larvae. When these provisions are met, the commodity could be imported without any further restrictions.

Where the imported commodity is not in a state consistent with the approved states for OIE-listed diseases (Appendix 3):

Option 3

Heat treatment (by cooking to at least 60°C for 1 minute) should eliminate muscle-encysting cestode larvae. When this provision is met, the commodity could be imported without any further restrictions; or,

Option 4

Frozen storage (to below -20°C for 168 hours) should eliminate muscle-encysting cestode larvae. When this provision is met, the commodity could be imported without any further restrictions; or,

Option 5

Restriction of the commodity to wild-caught fish (not from aquaculture) should moderately reduce the occurrence of muscle-encysting larvae. When this provision is met, the commodity could be imported without any further restrictions.

51.4. References

Abdelsalam M, Abdel-Gaber R, Mamoud MA, Mahdy OA, Khafaga NIM, Wada M (2016). Morphological, molecular and pathological appraisal of *Callitetrarhynchus gracilis* plerocerci (Lacistorhynchidae) infecting Atlantic little tunny (*Euthynnus alletteratus*) in Southeastern Mediterranean. *Journal of Advanced Research* 7(2): 317–326 [Online] Available from: http://www.doi. 10.1016/j.jare.2015.07.004 [Accessed 3 August 2018].

Ahmadiara E, Hosseini SH, Jaloisian F, Mousavi EHA, Sakhaiifar S, Sadeghia GA (2014). The study of the plerocercoid of diphyllobothriidae (cestoda,pseudophyllidea) in two cyprinid hosts, *Abramis brama* and *Alburnoides bipunctatus* from north and northwest of Iran. *Iranian Journal of Veterinary Medicine* 7(2): 103–109.

Amundsen PA, Lafferty KD, Knudsen R, Primicerco R, Krisofferson R, Klemesen A, Kuris AM (2013). New parasites and predators follow the introduction of two fish species to a subarctic lake: implications for food-web structure and functioning. *Oecologia* 171(4): 993–1002 [Online] Available from http://www.doi.10.1007/s00442-012-2461-2 [Accessed 3 August 2018].

Anonymous (2016). Biosecurity (Notifiable Organisms) Order 2016. NZLR 2016/73. New Zealand Government, Wellington, New Zealand, 13 p.

Athokpam VD, Tandon V (2015). A survey of metacercarial infections in commonly edible fish and crab hosts prevailing in Manipur, Northeast India. *Journal of Parasitic Diseases* 39(3): 429–440. [Online] Available from: http://www. 10.1007/s12639-013-0360-z [Accessed 9 July 2018].

Beveridge I, Bray RA, Cribb TH, Justine JL (2014). Diversity of trypanorhynch metacestodes in teleost fishes from coral reefs off eastern Australia and New Caledonia. *Parasite* 21: 60 [Online] Available from: http://www.doi,10.1051/parasite/2014060 [Accessed 24 July 2017].

Bhuiyan AI (2013). Endoparasitic helminths of *Tenualosa ilisha* in Bangladesh in relation to sex, seasons and habitat type. *Bangladesh Journal of Zoology* 41(2): 153–163.

Boustead NC (1982). Fish diseases recorded in New Zealand with a discussion on potential sources and certification processes. Fisheries Research Division, New Zealand Ministry of Agriculture and Fisheries, 19 p.

Chai J-Y, Murrell KD, Lymbery AJ (2005). Fish-borne parasitic zoonoses: status and issues. *International Journal of Parasitology* 35: 1233–1254.

Chung DI1, Kong HH, Moon CH, Choi DW, Kim TH, Lee DW, Park JJ (1995). The first human case of *Diplogonoporus balaenopterae* (Cestoda: Diphyllobothriidae) infection in Korea. Korean Journal of Parasitology 33(3): 225–230.

da Fonseca MCG, de São Clemente SC, Felizardo NN, Gomes DC, Knoff M (2012). Trypanorhynch cestodes of hygienic-sanitary importance infecting flounders *Paralichthys patagonicus* Jordan, 1889 and *Xystreurys rasile* (Jordan, 1891) of the Neotropical region, Brazil. *Parasitological Research* 111(2): 865–874 [Online] Available from: https://doi.org/10.1007/s00436-012-2912-z [Accessed 2 August 2018].

Dick TA, Chambers C, Isinguzo I (2006). Cestoidea (Phylum Platyhelminthes)., pp 391–416 *In:* Woo PTK (*Ed*.) Fish Diseases and Disorders, Volume 1: Protozoan and metazoan infections. CABI International, Wallingford, United Kingdom, 791 p.

Diggles BK (2008). Transfer of parasites between wild and cultured fishes with respect to aquaculture development in the Firth of Thames, New Zealand. DigsFish Services Pty Ltd. (Appendix 2 in Kelly S (2008). Potential environmental effects associated with the proposed shift from mussel to finfish farming in the Firth of Thames. Environment Waikato Technical Report 2008/38. 22 p).

D'Silva J, Bhuiyan AI, Bristow GA (2012). Distribution of helminth parasites in size groups and organs of hilsa shad, *Tenualosa ilisha*. *Dhaka University Journal of Biological Science* 21(1): 55–65.

Durborow RM, Rogers WA, Klesius PH (1988). Interaction of bass tapeworm, *Proteocephalus ambloplitis*, and *Neoechinorhynchus* sp. (Acanthocephala) in largemouth bass, *Micropterus salmoides. Journal of Parasitology* 74(6): 1056–1059.

Duggan IC, Green JD, Burger DF (2010). First New Zealand records of three non-indigenous Zooplankton species: *Skistodiaptomus pallidus*, *Sinodiaptomus valkanovi*, and *Daphnia dentifera. New Zealand Journal of Marine and Freshwater Research* 40: 561–569.

European Food Safety Authority (EFSA) (2010). Risk assessment on parasites in fishery products. *EFSA Journal* 8(4): 1543.

Ehab E, Faisal M (2008). Interactions between *Proteocephalus ambloplitis* and *Neoechinorhynchus* sp. in Largemouth Bass, *Micropterus salmoides*, collected from Inland Lakes in Michigan, USA. *Journal of American Science* 4(4):50–57.

Felizardo NN, Torres EJL, Fonseca GMC, Pinto RM, Gomes CD, Knoff M (2010). Cestodes of the flounder *Paralichthys isosceles* Jordan, 1890 (Osteichthyes - Paralichthyidae) from the state of Rio de Janeiro, Brazil. *Neotropical Helminthology* 4(2): 113–125.

Food and Drug Administration (**FDA) (2017).** Safe practices for food processes. Processing parameters needed to control pathogens in cold smoked fish, Chapter 5. Potential hazards in cold smoked fish: Parasites. Food and Drug Administration, Washington D.C., United States of America [Online] Available from: http://www.fda.govt/Food/FoodScience/SafePracticesFor FoodProcesses/ucm094578.htm [Accessed 20 January 2017].

Food and Drug Administration (**FDA) (2011).** Fish and Fishery Products Hazards and Controls Guidance, 4th Edition. Department of Health and Human Services, Food and Drug Administration (FDA), Silver Spring, Maryland, United States of America [Online] Available from: www.fda.gov/Food/Guidances [Accessed 1 March 2017].

Felix J (2013). Reported incidences of parasitic infections in marine mammals from 1892 to 1978. Zea Books, University of Nebraska, Lincoln, Nebraska, United States of America [Online] Available from: Http:///www.digitalcommons.unl.edu/zeabook/] Accessed 20 July 2017].

Felizardo NN, Torres EJL, Fonseca GMC, Pinto RM, Gomes CD, Knoff M (2010). Cestodes of the flounder *Paralichthys isosceles* Jordan, 1890 (Osteichthyes - Paralichthyidae) from the state of Rio de Janeiro, Brazil. *Neotropical Helminthology* 4(2): 113–125.

Fish and Game (2014). New Zealand Fish and Game Council [Online]. Available from: http://www.fishandgame.org.nz/ [Accessed 24 September 2014].

Haseli M, Malek M, Valinasasb T, Palm HW (2010). Trypanorhynch cestodes of teleost fish from the Persian Gulf, Iran. *Journal of Helminthology* (2010): 1–10 [Online] Available from http://www. doi:10.1017/S0022149X10000519 [Accessed 25 July 2017].

Heynemann D (1996). Cestodes. Chapter 89. *In*: Baron S (*Ed.*). *Medical Microbiology*, 4th Edition. University of Texas, Galveston. United States of America [Online] Available from: https://www.ncbi.nlm.nih.gov/books/NBK8399 [Accessed 1 March 2017].

Hine PM. Jones JB, Diggles BK (2000). A checklist of the parasites of New Zealand fishes, including previously unpublished records. NIWA Technical Report 75. National Institute of Water and Atmospheric Research Ltd., Auckland, New Zealand, 93 p.

Ibrahim MM (2000). Histopathology of Trypanorhyncha plerocercoids (Cestodes) in some Marine Fish from Waters of the Arabian Gulf. *Kau University Journal of Marine Science* 11: 59–73.

Jellyman D (2012). The status of longfin eels in New Zealand – an overview of stocks and harvest. NIWA report prepared for the Parliamentary Commissioner for the Environment, January 2012. National Institute for Water and Atmospheric Research, Auckland, New Zealand.

Jensen K, Bullard SA (2010). Characterization of a diversity of tetraphyllidean and rhinebothriidean cestode larval types, with comments on host associations and life-cycles. *International Journal for Parasitology* 40(8): 889–910.

Johnston C (2008a). Import Risk Analysis: Frozen skinless and boneless fillet meat of *Oreochromis* spp. from China and Brazil for human consumption. Biosecurity New Zealand, Ministry of Agriculture and Forestry, Wellington, New Zealand, 96 p.

Johnston C (2008b). Import Risk Analysis: Frozen skinless and boneless fillet meat of *Pangasius* spp. fish from Vietnam for human consumption. Biosecurity New Zealand, Ministry of Agriculture and Forestry, Wellington, New Zealand, 60 p.

Justine JL, Beveridge I, Boxshall GA, Bray RA, Miller TL, Moravec F, Trilles J-P, Whittington ID (2012). An annotated list of fish parasites (Isopoda, Copepoda, Monogenea, Digenea, Cestoda, Nematoda) collected from Snappers and Bream (Lutjanidae, Nemipteridae, Caesionidae) in New Caledonia confirms high parasite biodiversity on coral reef fish. *Aquatic Biosystems* 8 (22) [Online] Available from http://www.aquaticbiosystems.org/content/8/1/22 [Accessed 21 July 2017].

Justine JL, Beveridge I, Boxshall GA, Bray RA, Moravec F, Trilles JP, Whittington ID (2010). An annotated list of parasites (Isopoda, Copepoda, Monogenea, Digenea, Cestoda and Nematoda) collected in groupers (Serranidae, Epinephelinae) in New Caledonia emphasizes parasite biodiversity in coral reef fish. *Folia Parasitologica* 57(4): 237–262.

Kahn SA, Beers PT, Findlay VL, Peebles IR, Durham PJ, Wilson DW, Gerrity SE (1999). Import risk analysis on non-viable salmonids and non-salmonid marine finfish. Australian Quarantine and Inspection Service, Department of Agriculture, Fisheries and Forestry, Canberra, Australia, 409 p.

Khanna DR (2004). Biology of helminth. Discovery Press, London, United Kingdom, 456 p.

Kino H, Hori W, Kobayashi H, Nakamura N, Nagasawa K (2002). A mass occurrence of human infection with *Diplogonoporus grandis* (Cestoda: Diphyllobothriidae) in Shizuoka Prefecture, central Japan. *Parasitology International* 51(1): 73–79 [Online] Available from: https://doi.org/10.1016/S1383-5769(01)00106-4 [Accessed 27 July 2018].

Korting W (1984). Larval cyclophyllidean cestodes in carp and tench. *Bulletin of the European Association of Fish Pathologists* 4(3): 40–41.

Kuchta R, Oros M, Ferguson J, Scholz T (2017). *Diphyllobothrium nihonkaiense* tapeworm larvae in salmon from North America. *Emerging Infectious Diseases* 23(2): 351–353.

Kuchta R, Esteban JG, Brabec J, Scholz T (2014). Misidentification of *Diphyllobothrium* Species Related to Global Fish Trade, Europe. *Emerging Infectious Diseases* 20(11): 1955–1957.

Kuchta R, Brabec J, Kubakova P, Scholtz T (2013). Tapeworm *Diphyllobothrium dendriticum* (Cestoda)--neglected or emerging human parasite? *PLoS Neglected Tropical Diseases* Dec 26; 7(12):e2535 [Online] Available from: http://www.doi.10.1371/journal.pntd.0002535. eCollection 2013 [Accessed 1 March 2017].

Marsh D, Mkwra L (2013). Review of freshwater non-market value studies. Department of Economics, University of Waikato, Hamilton, New Zealand, 144 p.

Mayo-Hernandez E, Peñalver J, García-Ayala A, Serrano E, Muñoz P, Ruiz de Ybáñez R (2015). Richness and diversity of helminth species in eels from a hypersaline coastal lagoon, Mar Menor, south-east Spain. *Journal of Helminthology* 89(3): 345–351.

Moravec F (1985). Occurrence of endoparasitic helminths in eels (*Anguilla anguilla* L) from the Macha Lake fishpond system, Czechoslovakia. *Folia Parasitologica* 32: 113–125.

Morsey K, Bashtar AR, Ghaffar FA, Al Quraishy S, Al Ghamdi A, Mostafa N (2013). First identification of four trypanorhynchid cestodes: *Callitetrarhynchus speciosus, Pseudogrillotia* sp. (Lacistorhynchidae), *Kotorella pronosoma* and *Nybelinia bisulcata* (Tentaculariidae) from Sparidae and Mullidae fish. *Parasitology Research* 112(7): 2523–2532.

Ministry for Primary Industries (MPI) (2014). Freshwater eels (SFE, LFE), pp 276–301 *In:* Fisheries Assessment Plenary, May 2014: stock assessments and stock status. Compiled by the Fisheries Science Group, Ministry for Primary Industries, Wellington, New Zealand, 1381 p.

Nawa Y, Hatz C, Blum J (1995). Sushi delights and parasites: The risk of fish borne and foodborne parasitic zoonoses in Asia. *Clinical and Infectious Diseases* 41(9): 1297–1303.

NIWA (2017). Aquaculture species: Eels. National Institute of Water and Atmospheric Research, Auckland, New Zealand [Online] Available from: https://www.niwa.co.nz/aquaculture/aquaculture-species/eel [Accessed 3 August 2018].

OIE (2016). Aquatic Animal Health Code, 19th Edition. World Organization for Animal Health, Paris, France [Online] Available from: http://www.oie.int/international-standard-setting/aquatic-code/access-online/ [Accessed 15 March 2017].

Overstreet RM (1977). *Poecilancistrium caryophyllum* and other trypanorhynch cestode plerocercoids from the musculature of *Cynoscion nebulosus* and other sciaenid fishes in the Gulf of Mexico. *Journal of Parasitology* 63(5): 780–789.

Palm HW, Overstreet RM (2000). New records of trypanorhynch cestodes from the Gulf of Mexico, including *Kotorella pronosoma* (Stossich, 1901) and *Heteronybelinia palliata* (Linton, 1924) comb. n. *Folia Parasitica* 47: 293–302.

Parliamentary Commissioner for the Environment (2013). On a pathway to extinction? An investigation into the status and management of the longfin eel. Parliamentary Commission for the Environment, Wellington, New Zealand, 95 p.

Paulin C, Roberts C, Stewart A, McMillan P (2001). New Zealand Fish: A complete Guide. Te Papa Press, Wellington, New Zealand. 279 p.

Pelayo V, Garcia-Hernandez P, Puente P, Rodero M, Cuebilar C (2009). Seroprevalence of anti-*Gymnorhynchus gigas* (Trypanorhyncha, Gymnorhynchidae) antibodies in a Spanish population. *Journal of Parasitology* 95(): 778–780.

Petersen F, Palm H, Moller H, Cuzi MA (1993). Flesh parasites of fish from central Philippine waters. *Diseases of Aquatic Organisms* 15: 81–86.

Plant & Food (2016). Sustainable snapper piped out to sea by Plant and Food [Online] Available from: http://www.stuff.co.nz/business/farming/aquaculture/87522652/sustainable-snapper-piped-out-to-sea [Accessed 14 August 2017].

Rausch RL, Adams AM (2000). Natural transfer of helminths of marine origin to freshwater fishes, with observations on the development of *Diphyllobothrium alascense*. *The Journal of Parasitology* 86(2): 319–327.

Ruttenber AJ, Weniger BG, Sorvillo F (1984). Diphyllobothriasis associated with salmon consumption in Pacific coast states. *American Journal of Tropical Medicine and Hygiene* 33: 455–459.

Saksvik M, Nilsen F, Nylund A, Berland B (2001). Effect of marine *Eubothrium* sp. (Cestoda: Pseudophyllidea) on the growth of Atlantic salmon, *Salmo salar* L. Journal of Fish Diseases 24(2): 111–119.

Salminen K (1970). The infestiveness of heat and cold exposed *Diphyllobothrium latum* plerocercoids on golden hamster. *Acta Veterinaria Scandinavia* 11: 247–253.

Scholz T (1997). Life-cycle of *Bothriocephalus claviceps*, a specific parasite of eels. *Journal of Helminthology* 71(3): 241–248.

Scholz T, Kuchta R (2016). Fish-borne, zoonotic cestodes (*Diphyllobothrium* and relatives) in cold climates: A never-ending story of neglected and (re)-emergent parasites. *Food and Waterborne Parasitology* 4: 23–38.

Scholz T, Kuchta R, Williams C (2012). "*Bothriocephalus acheilognathi*", pp 282–297 *In:* Woo PTK, Buchmann K (*Eds*.). Fish Parasites: Pathobiology and Protection. CABI International, Wallingford, United Kingdom.

Scholz T, Garcia HH, Kuchta R, Wicht B (2009). Update on the human broad tapeworm (genus *Diphyllobothrium*), including clinical relevance. *Clinical Microbiology Reviews* 22(1): 146–160.

Schultz ET, Topper M, Heins DC (2006). Decreased reproductive investment of female threespine stickleback *Gasterosteus aculeatus* infected with the cestode *Schistocephalus solidus*: parasite adaptation, host adaptation, or side effect? *Oikos* 114: 303–310.

Seafood New Zealand (2014). Seafood New Zealand: Key facts for our Industry. Seafood New Zealand, Wellington, New Zealand, 6 p.

Shamsi S, Steller E, Chen Y (2018). New and known zoonotic nematode larvae within selected fish species from Queensland waters in Australia. *International Journal of Food Microbiology* 272: 73–82 [Online] Available from https://www.doi.org/10.1016/j/ijfoodmicro.2018.03.007 [Accessed 26 June 2018].

Sim-Smith C, Faire S, Lees A (2014). Managing biosecurity risk for business benefit. Report prepared for the Ministry for Primary Industries by Coast and Catchment Ltd, Auckland, New Zealand, 214 p.

Soroka NM, Safarova FE, Akraomova FD, Shakarboev EB, Azimov DA, Mengliyev GA, Halikov SR, Mamatiminov SH (2014). Cestodes of Cypriniformes fishes in the waterbodies of the midstream of the River Syrdarya. *Biological Resources and Nature Management* 6(3-4): 88–91.

Torres P, Cuevas C, Tang M, Barra M, Franjola R, Navarrete N, Montefusco A, Otth L, Wilson G, Puga S, Figueroa L, Cerda O (2004). Introduced and native fishes as infection foci of *Diphyllobothrium* spp. in humans and dogs from two localities at Lake Panguipulli in Southern Chile. *Comparative Parasitology* 71: 111–117.

Valtonen FT, Rintamaki P, Lappalainen M (1989). *Triaenophorus nodulosus* and *T. crassus* in fish from northern Finland. *Folia Parasitica* 36(4): 351–370.

Waeschenbach A, Brabec J, Scholz T, Littlewood DTJ, Kuchta R. (2017). The catholic taste of broad tapeworms – multiple routes to human infection. *International Journal for Parasitology.* 47(13): 831–843 [Online] Available from: https://doi.org/10.1016/j.ijpara.2017.06.004 [Accessed 10 April 2018].

Wardle RA (1933). Significant factors in the plerocercoid environment of *Diphyllobothrium latum* (Linn.). *Journal of Helminthology* 11(1): 25–44.

Watene E (2003). Potential for Commercial Eel Aquaculture in Northland. NIWA Client Report AKL2003-032. NIWA, National Institute of Water and Atmospheric Research, Auckland, New Zealand. 36 p.

Webber W, Fenwick G, Bradford-Grieve J, Eagar S, Buckeridge J, Poore G, Dawson E, Watling L, Jones J, Wells J, Bruce N, Ahyong S, Larsen K, Chapman M, Olesen J, Ho J, Green J, Shiel R, Rocha C, Lorz A, Bird G, Charleston W (2010). Phylum Arthropoda Subphylum Crustacea: shrimps, crabs, lobsters, barnacles, slaters, and kin', pp 98-232 *In:* Gordon DP (*Ed*.) *New Zealand Inventory of Biodiversity:* Volume Two: Kingdom Animalia - Chaetognatha, Ecdysozoa, Ichnofossils, Canterbury University Press, New Zealand [Online] Available from: https://researchbank.rmit.edu.au/view/rmit:16047 [Accessed 1 March 2017].

Wicht B, Scholz T, Peduzzi R, Kuchta R (2008). First record of human infection with the tapeworm *Diphyllobothrium nihonkaiense* in North America. *American Journal of Tropical Medicine and Hygiene* 78(2): 235–238.

Zarger UR, Chishti MZ, Yousuf AR, Ahmed F (2012). Infection level of the Asian tapeworm (*Bothriocephalus acheilognathi*) in the cyprinid fish, *Schizothorax niger*, from Anchar Lake, relative to season, sex, length and condition factor. *Parasitology Research* 110(1): 427–435 [Online] Available from: http://www.doi.10.1007/s00436-011-2508-z [Accessed 10 August 2018].

52. Appendix 1 - Determination of Pathways for Diseases Associated with Fish Imports

52.1. Current requirements for imported eviscerated fish

Currently, marine fish may be imported as whole, round fish (MPI 2008), while fish sourced from freshwater (including anadromous fish) require further treatment. Salmon may be imported as headed, gilled and gutted product, while Nile perch (*Lates niloticus*), tilapia (*Oreochromis* spp.) and catfish (*Pangasius* spp.) can be imported as skin-off fillets, from approved countries, while other freshwater fish must be cooked (to 85°C for 15 minutes). Fish may also be imported for human consumption in a consumer-ready form, or imported as a bulk commodity for further processing in New Zealand in a licenced Transitional Facility (TF) for subsequent retail sale (MPI 2000, 2004a, 2004b, 2011, 2016). Fish organic wastes in the human food consumption pathway may thus be derived from commercial processing, or from household processing by domestic consumers.

52.2. Commercial processing of imported fish product

The Ministry for Primary Industries (MPI) has developed specific requirements for TFs, in addition to those of the import health standard and/or import permit required. There is currently a specific TF requirement for management and disposal of animal products, including fish (MPI 2016). This may also include requirements for biosecure disposal of any imported packaging material, as well as animal product wastes, such as off-cuts from trimming, liquids from fish thawing, and the disposal of any associated unwanted animal products (offal).

52.3. Domestic processing of imported fish product

During 2010, an estimated 2.531 million tonnes of waste was disposed of in the 71 municipal landfills in New Zealand, representing approximately half of the total organic wastes produced by New Zealand households (Hogg *et al.* 2010). Of this, over 50% is derived from kitchen wastes, the remainder consisting of garden and landscaping waste material (Waste Not Consulting 2009).

The remainder of the domestic waste is disposed of by composting (13%) and as domestic sewage following in-sink or waste master maceration (13%), whereas the rest (71%) is collected by local Authorities. This suggests that almost 85% of New Zealand organic waste is disposed of in landfills or by disposal into domestic sewerage (Hogg *et al.* 2010).

Few data are available on the composition of organic waste materials from New Zealand domestic households. However, data from United Kingdom households (Ventour 2008) is generally assumed to reasonably approximate disposal patterns for New Zealand households (Jo Berry, MPI, *pers. comm*. 2014).

Meat and fish wastes in the United Kingdom have been estimated to account for only 8.4% (by weight) of the total domestic food waste. Of this, less than 20% (18.5%) was discarded in a fresh or uncooked state. Most meat and fish waste was shown to be derived from poultry (55%), and only 4.2% was derived from fish products (Ventour 2008).

52.4. Summary

Regulatory mechanisms are available to control the likely pathways for exotic pathogens in imported fish products derived from the commercial processing of bulk-imported fish products.

As the quantity of fish product wastes derived from domestic food processing is so low as to be negligible, this pathway is considered insignificant.

52.5. References

Hogg D, Wilson D, Gibbs A, Holmes A, Eve L (2010). Earthcare Environmental Ltd & Envirofert Ltd. Household Organic Waste Cost Benefit Analysis. Report to Greenfingers Garden Bags/Eunomia Research and Consulting Ltd, Auckland, New Zealand, 76 p.

MPI (2000). Import health standard for the importation into New Zealand of frozen Nile perch (*Lates niloticus*) skinless, boneless fillets for human consumption from Kenya, Tanzania or Uganda (Lake Victoria), FISNPEIC.ALL. Ministry for Primary Industries, Wellington, New Zealand, 2 p.

MPI (2004a). Import health standard for the importation into New Zealand of cooked fish from all countries FISCOOIC.ALL. Ministry for Primary Industries, Wellington, New Zealand, 6 p.

MPI (2004b). Import health standard for the importation into New Zealand of commercial consignments of fresh/frozen/processed salmonids for human consumption from the European Community, FISSALIC.EEC. Ministry for Primary Industries, Wellington, New Zealand, 7 p.

MPI (2008). Import health standard for the importation of marine fisheries products for human consumption from all countries, FISMARIC.ALL. Ministry for Primary Industries, Wellington, New Zealand, 5 p.

MPI (2011). Import health standard for processed tilapia and catfish for human consumption, FISFILIC.SPE. Ministry for Primary Industries, Wellington, New Zealand, 7 p.

MPI (2016). Facility Standard: Transitional Facilities for animal products, MPI-STD-ANIPRODS, Ministry for Primary Industries, Wellington, New Zealand, 18 p. [Online]. Available from: https://www.mpi.govt.nz/dmsdocument/12774/send [Accessed 15 April 2019].

Ventour L (2008). The food we waste. Report for project code RBC405-0010. Waste and Resources Action Programme (WRAP). Banbury, Oxon., United Kingdom, 237 p.

Waste Not Consulting (2009). Household Sector Waste to Landfill in New Zealand; Report to the Ministry for the Environment, 39 p.

53. Appendix 2: Pathogen incidence by fish host family

Key to Abbreviations for Viral Pathogens

EHNV	Epizootic haemopoietic necrosis virus
EVE	European eel virus
EEHV	European eel herpesvirus
GCHV	Grass carp haemorrhagic virus
GIV	Grouper iridovirus
IHNV	Infectious haemopoietic necrosis virus
IPNV	Infectious pancreatic necrosis virus
ISAV	Infectious salmon anaemia virus
KHV	Koi herpesvirus
HIRRV	Marine hirame rhabdovirus
NJV	New Japan virus
NNV	Nervous necrosis virus (NNV) and associated nodaviruses
OMV	*Oncorhynchus Masou* herpesvirus
PRV	Piscine and related aquareoviruses
RSIV	Red seabream iridovirus
SAV	Salmon alphavirus and associated alphaviruses
SGPV	Salmon gill poxvirus
SVCV	Spring viraemia of carp virus and associated rhabdoviruses
VHSV	Viral haemorrhagic septicaemia virus

Appendix 2a: Viral Pathogens and Fish Host Families

Fish Host Family	EHNV/ECV/ESV	EVE	EEHV	GCHV	GIV	HIRRV	IHNV	IPNV	ISAV	KHV	NJV	NNV	OMV	PRV	RSIV/ISKNV	SAV	SGPV	SVCV	VHSV
Acanthuridae												✓							
Achiridae																			
Acipenseridae							✓	✓		✓		✓							
Acropomatidae												✓							
Alestidae																			
Amiidae								✓											
Ambassidae																			
Ammodytidae								✓											✓
Anabantidae																			
Anarhichadidae								✓				✓							
Anguillidae		✓	✓				✓	✓				✓							✓
Anoplopomatidae																			✓
Apogonidae																			
Ariidae																			
Argentinidae														✓					✓
Atherinidae								✓											
Aulorhynchidae								✓											✓
Bagridae																			
Batrachoididae												✓							
Belonidae																			✓
Berycidae																			
Bothidae								✓				✓							
Callionymidae																			
Carangidae								✓				✓		✓	✓				✓
Carcharhinidae																			
Catostomidae								✓											✓
Centrarchidae								✓				✓		✓	✓			✓	✓
Centropomatidae																			
Chaetodontidae																			
Characidae																			
Chanidae												✓							
Channidae								✓						✓					
Chirocentridae																			
Cichlidae		✓						✓				✓						✓	
Clariidae																			
Clupeidae						✓	✓	✓	✓			✓		✓					✓
Cobiditidae																			
Congridae																			
Coryphaenidae																			
Cottidae																			
Cyclopteridae																			
Cynoglossidae												✓							
Cyprinidae				✓				✓		✓		✓		✓	✓		✓	✓	✓
Cyprinodontidae								✓											
Dasyatidae																			
Distichodontidae																			
Eleotridae												✓							
Embiotocidae							✓	✓											✓

Fish Host Family	EHNV/ECV/ESV	EVE	EEHV	GCHV	GIV	HIRRV	IHNV	IPNV	ISAV	KHV	NJV	NNV	OMV	PRV	RSIV/ISKNV	SAV	SGPV	SVCV	VHSV
Engraulidae																			✓
Esocidae	✓						✓	✓										✓	✓
Exocoetidae																			
Fundulidae																			✓
Gadidae							✓	✓	✓			✓				✓			✓
Galaxiidae	✓																		
Gasterosteidae																			
Gempylidae																			
Gerreidae																			
Glaucostegidae																			
Gobiidae												✓							✓
Haemulidae												✓			✓				
Hapalogenyidae												✓							
Helostomatidae																			
Hexagrammidae																			
Ictaluridae	✓							✓						✓					✓
Kyphosidae															✓				
Kurtidae																			
Labridae								✓								✓			
Lateolabracidae						✓						✓			✓				
Latidae															✓				
Latridae												✓							
Leiognathidae												✓							
Lepisosteidae						✓		✓											
Lethrinidae												✓			✓				
Liparidae																			
Lophiidae												✓							
Lotidae																			✓
Lutjanidae												✓							
Macrouridae								✓				✓							
Malacanthidae												✓							
Mastacembelidae																			
Merlucciidae								✓											✓
Monacanthidae												✓							
Mormyridae																			
Moronidae							✓	✓				✓			✓				✓
Mugilidae												✓			✓				✓
Mullidae								✓				✓							✓
Muraenesocidae												✓							
Nemipteridae																			
Notopteridae																			
Oplegnathidae												✓			✓				
Ophidiidae																			✓
Orectolobidae																			
Osmeridae														✓					✓
Osphronemidae																			
Pangasiidae																			
Paralichthyidae						✓		✓				✓			✓				✓
Percichthyidae	✓				✓			✓				✓			✓				

Fish Host Family	EHNV/ECV/ESV	EVE	EEHV	GCHV	GIV	HIRRV	IHNV	IPNV	ISAV	KHV	NJV	NNV	OMV	PRV	RSIV/ISKNV	SAV	SGPV	SVCV	VHSV
Percidae	✓							✓		✓		✓						✓	✓
Percopsidae																			✓
Pimelodidae																			
Platycephalidae												✓							
Plecoglossidae						✓		✓			✓						✓		
Pleuronectidae						✓		✓				✓			✓	✓			✓
Plotosidae	✓											✓							
Poeciliidae																			
Polynemidae														✓					
Polyodontidae								✓											
Polyprionidae																			
Pomatomidae																			
Priacanthidae												✓							
Psettodidae																			
Rachycentridae												✓			✓				
Rhinobatidae																			
Salmonidae	✓	✓				✓	✓	✓	✓		✓	✓	✓	✓		✓	✓	✓	✓
Scaridae																			
Scatophagidae																			
Schilbeidae																			
Sciaenidae								✓				✓			✓				✓
Scombridae												✓			✓				✓
Scophthalmidae							✓	✓				✓		✓		✓			
Scorpaenidae																			✓
Sebastidae						✓						✓			✓				✓
Serrasalmidae																			
Serranidae					✓							✓			✓				✓
Siganidae												✓							
Sillaginidae												✓							✓
Siluridae										✓		✓						✓	
Soleidae								✓				✓							✓
Sparidae						✓	✓	✓				✓			✓				✓
Sphyraenidae								✓											
Sphyrnidae																			
Stichaeidae						✓													
Stromateidae																			✓
Synbranchidae																			
Synodontidae																			
Tetraodontidae												✓							
Terapontidae	✓											✓			✓				
Trichiuridae																			✓
Triglidae								✓				✓							
Umbridae																			
Uranoscopidae																			✓
Zeidae												✓							
Zoarcidae												✓							

Appendix 2b: Bacterial Pathogens and Fish Host Families

Fish Host Family	*A. hydrophila*	*Atypical A. salmonicida*	*Typical A. salmonicida*	*Edwardsiella tarda*	*F. columnare*	*Francisella spp.*	*Moritella viscosa*	*Piscirickettsia salmonis*	*Pseudomonas anguilliseptica*	*Renibacterium salmoninarum*	*S. agalactiae, S. iniae*	*Yersinia ruckeri*
Acanthuridae												
Achiridae												
Acipenseridae	✓											✓
Acropomatidae												
Alestidae												
Amiidae												
Ambassidae												
Ammodytidae		✓										
Anabantidae				✓								
Anarhichadidae		✓										
Anguillidae		✓		✓					✓		✓	✓
Anoplopomatidae		✓								✓		
Apogonidae												
Ariidae											✓	
Argentinidae												
Atherinidae												
Aulorhynchidae												
Bagridae												
Batrachoididae				✓								
Belonidae												
Berycidae												
Bothidae												
Callionymidae												
Carangidae				✓					✓		✓	
Carcharhinidae												
Catostomidae			✓									
Centrarchidae		✓		✓	✓				✓			
Centropomatidae												
Chaetodontidae												
Characidae												
Chanidae												
Channidae				✓							✓	
Chirocentridae												
Cichlidae		✓		✓	✓	✓		✓	✓		✓	✓
Clariidae				✓	✓							
Clupeidae		✓							✓	✓	✓	
Cobiditidae									✓			
Congridae												
Coryphaenidae												
Cottidae												
Cyclopteridae		✓					✓					
Cynoglossidae												
Cyprinidae	✓	✓	✓	✓	✓				✓	✓	✓	✓
Cyprinodontidae												
Dasyatidae				✓								
Distichodontidae												
Eleotridae				✓								

Fish Host Family	*A. hydrophila*	*Atypical A. salmonicida*	*Typical A. salmonicida*	*Edwardsiella tarda*	*F. columnare*	*Francisella spp.*	*Moritella viscosa*	*Piscirickettsia salmonis*	*Pseudomonas anguilliseptica*	*Renibacterium salmoninarum*	*S. agalactiae, S. iniae*	*Yersinia ruckeri*
Embiotocidae										✓		
Engraulidae												
Esocidae		✓	✓									
Exocoetidae												
Fundulidae												
Gadidae		✓				✓	✓		✓			✓
Galaxiidae												
Gasterosteidae												
Gempylidae												
Gerreidae		✓										
Glaucostegidae												
Gobiidae				✓								
Haemulidae				✓							✓	
Hapalogenyidae												
Helostomatidae												
Hexagrammidae		✓								✓		
Ictaluridae	✓			✓	✓						✓	✓
Kyphosidae												
Kurtidae												
Labridae		✓	✓									
Lateolabracidae												
Latidae				✓	✓				✓		✓	
Latridae		✓										
Leiognathidae												
Lepisosteidae												
Lethrinidae												
Liparidae												
Lophiidae												
Lotidae	✓	✓										✓
Lutjanidae											✓	
Macrouridae												
Malacanthidae												
Mastacembelidae												
Merlucciidae										✓		
Monacanthidae												
Mormyridae												
Moronidae		✓	✓	✓		✓		✓	✓		✓	
Mugilidae				✓			✓				✓	
Mullidae												
Muraenesocidae												
Nemipteridae												
Notopteridae												
Oplegnathidae												
Ophidiidae												
Orectolobidae												
Osmeridae												
Osphronemidae				✓								
Pangasiidae					✓							
Paralichthyidae				✓							✓	

Fish Host Family	*A. hydrophila*	*Atypical A. salmonicida*	*Typical A. salmonicida*	*Edwardsiella tarda*	*F. columnare*	*Francisella spp.*	*Moritella viscosa*	*Piscirickettsia salmonis*	*Pseudomonas anguilliseptica*	*Renibacterium salmoninarum*	*S. agalactiae, S. iniae*	*Yersinia ruckeri*
Percichthyidae					✓							
Percidae		✓			✓							✓
Percopsidae												
Pimelodidae												
Platycephalidae										✓		
Plecoglossidae									✓	✓		
Pleuronectidae		✓	✓	✓		✓	✓					
Plotosidae									✓			
Poeciliidae												
Polynemidae												
Polyodontidae												
Polyprionidae												
Pomatomidae												
Priacanthidae												
Psettodidae												
Rachycentridae												
Rhinobatidae												
Salmonidae		✓	✓	✓	✓	✓	✓	✓	✓	✓	✓	✓
Scaridae											✓	
Scatophagidae												
Schilbeidae												
Scatophagidae												
Sciaenidae						✓		✓			✓	
Scombridae						✓						
Scophthalmidae		✓	✓	✓		✓	✓		✓			✓
Scorpaenidae												
Sebastidae												
Serrasalmidae												
Serranidae				✓				✓	✓		✓	
Serrasalmidae					✓							
Siganidae											✓	
Sillaginidae												
Siluridae				✓								
Soleidae												✓
Sparidae		✓	✓	✓					✓		✓	
Sphyraenidae												
Sphyrnidae												
Stichaeidae												
Stromateidae												
Synbranchidae												
Synodontidae											✓	
Tetraodontidae								✓				
Terapontidae		✓										
Trichiuridae												
Triglidae												
Umbridae												
Uranoscopidae												
Zeidae												
Zoarcidae		✓										

Appendix 2c: Other Pathogens and Fish Host Families

Fish Host Family	*Aphanomyces invadans*	*Ichthyophonus hoferi*	*Sphaerothecum destruens*	*Microsporidians*	*Myxozoans*	*Anguillicola crassus*	*Monogeneans*	*Digeneans*	*Cestodes*
Acanthuridae							✓		
Achiridae	✓								
Acipenseridae									
Acropomatidae									
Alestidae	✓								
Amiidae									
Ambassidae	✓								
Ammodytidae									
Anabantidae	✓								
Anarhichadidae									
Anguillidae	✓			✓	✓	✓	✓		✓
Anoplopomatidae									
Apogonidae									
Ariidae	✓								
Argentinidae									
Atherinidae									
Aulorhynchidae									
Bagridae	✓								
Batrachoididae									
Belonidae	✓								
Berycidae					✓				
Bothidae									
Callionymidae									
Carangidae	✓	✓		✓	✓		✓		
Carcharhinidae					✓				
Catostomidae							✓		
Centrarchidae	✓					✓			
Centropomatidae									
Chaetodontidae							✓		
Characidae									
Chanidae									
Channidae	✓						✓	✓	
Chirocentridae					✓				
Cichlidae	✓				✓	✓	✓	✓	
Clariidae	✓				✓				
Clupeidae	✓	✓			✓			✓	
Cobiditidae									
Congridae							✓		
Coryphaenidae					✓				
Cottidae									
Cyclopteridae									
Cynoglossidae									
Cyprinidae	✓	✓	✓		✓	✓	✓	✓	✓
Cyprinodontidae									
Dasyatidae					✓				
Distichodontidae					✓				
Eleotridae	✓							✓	
Embiotocidae							✓		

Fish Host Family	*Aphanomyces invadans*	*Ichthyophonus hoferi*	*Sphaerothecum destruens*	*Microsporidians*	*Myxozoans*	*Angullicola crassus*	*Monogeneans*	*Digeneans*	*Cestodes*
Engraulidae					✓				
Esocidae					✓	✓		✓	
Exocoetidae	✓								
Fundulidae									
Gadidae		✓							
Galaxiidae									
Gasterosteidae									
Gempylidae					✓				
Gerreidae									
Glaucostegidae					✓				
Gobiidae	✓					✓			
Haemulidae									
Hapalogenyidae									
Helostomatidae									
Hexagrammidae							✓		
Ictaluridae	✓				✓		✓	✓	
Kyphosidae							✓		
Kurtidae	✓								
Labridae							✓		
Lateolabracidae							✓	✓	
Latidae	✓				✓		✓		
Latridae									
Leiognathidae									
Lepisosteidae									
Lethrinidae									
Liparidae									
Lophiidae									
Lotidae									
Lutjanidae	✓	✓			✓				
Macrouridae					✓				
Malacanthidae									
Mastacembelidae	✓								
Merlucciidae					✓				
Monacanthidae									
Mormyridae									
Moronidae	✓	✓							
Mugilidae	✓	✓						✓	
Mullidae					✓				
Muraenesocidae									
Nemipteridae					✓				
Notopteridae	✓								
Oplegnathidae									
Ophidiidae									
Orectolobidae					✓				
Osmeridae		✓				✓			
Osphronemidae	✓								
Pangasiidae							✓		
Paralichthyidae		✓			✓				
Percichthyidae	✓								

Fish Host Family	*Aphanomyces invadans*	*Ichthyophonus hoferi*	*Sphaerothecum destruens*	*Microsporidians*	*Myxozoans*	*Angullicola crassus*	*Monogeneans*	*Digeneans*	*Cestodes*
Percidae					✓	✓		✓	✓
Percopsidae									
Pimelodidae					✓				
Platycephalidae	✓								
Plecoglossidae	✓			✓					
Pleuronectidae		✓		✓	✓				
Plotosidae									
Poeciliidae									
Polynemidae					✓				
Polyodontidae									
Polyprionidae							✓		
Pomatomidae	✓								
Priacanthidae									
Psettodidae	✓								
Rachycentridae									
Rhinobatidae					✓				
Salmonidae	✓	✓	✓	✓	✓		✓	✓	✓
Scaridae							✓		
Scatophagidae	✓`								
Schilbeidae	✓								
Sciaenidae	✓				✓				
Scombridae		✓							
Scophthalmidae					✓				
Scorpaenidae									
Sebastidae		✓					✓		
Serrasalmidae								✓	
Serranidae					✓		✓	✓	
Siganidae									
Sillaginidae	✓				✓				
Siluridae	✓				✓	✓			
Soleidae	✓							✓	
Sparidae	✓	✓			✓		✓	✓	✓
Sphyraenidae					✓				
Sphyrnidae					✓				
Stichaeidae									
Stromateidae									
Synbranchidae	✓								
Synodontidae									
Tetraodontidae					✓				
Terapontidae	✓								
Trichiuridae		✓							
Triglidae									
Umbridae									
Uranoscopidae									
Zeidae									
Zoarcidae									

54. Appendix 3: Risk management options by risk organism

Identified Risk Organism	OIE Listed	Restrict Commodity to Wild-Caught Fish	Processing Options	OIE-Listed, Approved States and Treatments See Footnote	Heat Treatment: Other Commodities (From Literature)	Frozen Storage (From Literature)
Viral Pathogens						
EHNV/ECV/ESV	Y	N	HGU	Y	60°C/ 15 m	NA
EVE	N	N	GGU	Y	56°C/ 120 m	NA
EEHV	N	N	HGU	Y	56°C/ 120 m	NA
GCHV	N	N	HGU	Y	56°C/ 120 m	NA
GIV	N	Y	HGU	Y	56°C/ 30 m	NA
HIRRV	N	N	SKF	Y	65°C/ 15 m	NA
IHNV	Y	N	HGU	Y	65°C/ 15 m; 82°C for 5 m	NA
IPNV	N	N	HGU	Y	60°C/ 300 m (5 hrs)	NA
ISAV	Y	N	HGU	Y	56°C/ 30 m	NA
KHV	Y	N	SKF	Y	NA	NA
NJV	N	Y	HGU	Y	NA	NA
NNV	N	Y	SKF	Y	60° C/ 60m	NA
OMV	N	N	SKF	Y	60° C/ 15m	-20°C/ 18 d
PRV	N	N	HGU	Y	56°C/ 120 m	NA
RSIV	Y	N	HGU	Y	56°C/ 30 m	NA
SAV	Y	N	HGU	Y	NA	NA
SGPV	N	N	SKF	Y	95°C/120 m	NA
SVCV	Y	N	SKF	Y	60° C/ 10m	NA
VHSV	Y	N	SKF	Y	NA	NA
Bacterial Pathogens						
Aeromonas hydrophila (exotic strains)	N	Y	SKF	Y	170°C/ 120 m (moist heat)	NA
Aeromonas salmonicida (atypical strains)	N	Y	SKF	Y	44° C/ 60m	NA
Aeromonas salmonicida (typical strains)	N	Y	SKF	Y	44° C/ 60m	NA
Edwardsiella spp. (species complex)	N	N	SKF	Y	121° C/ 15m (moist heat)	NA
Flavobacterium columnare (exotic strains)	N	Y	SKF	Y	65°C/ 25m	NA
Francisella spp.	N	Y	SKF	Y	121° C/ 15m	NA
Moritella viscosa	N	N	SKF	Y	85° C/ 25m	NA
Piscirickettsia salmonis species complex	N	N	SKF	Y	100°C/ 30 m	NA
Pseudomonas anguilliseptica	N	Y	HGU	Y	80° C/ 20m	NA
Renibacterium salmoninarum	N	Y	HGU	Y	100° C/ 30m	NA
Streptococcus species complex (exotic strains)	N	N	SKF	Y	121° C/ 15m (moist heat)	NA
Yersinia ruckeri (exotic strains)	N	N	HGU	Y	49° C/ 60m	NA

Identified Risk Organism	OIE Listed	Restrict Commodity to Wild-caught Fish	Processing	Heat Treatment OIE-Listed, Approved Commodities (See Footnote)	Heat Treatment: Other commodities (From Literature)	Frozen Storage (From Literature)
Other Pathogens						
Aphanomyces invadans	Y	Y	SKF	Y		-20 C° /72 hrs
Ichthyophonus hoferi	N	N	SKF	Y	40° C/ 3m	-20 C° / 3m
Sphaerothecum destruens	N	Y	SKF	Y	40° C/ 3m	-20 C° /72 hrs
Microsporidian pathogens	N	Y	SKF	Y	60° C/ 15m	-20 C° /28 days
Myxosporean pathogens	N	Y	SKF	Y	75° C/ 5m	NA
Anguillicola crassus	N	N	NA	Y	60° C/ 10m; or 80°C/ 10 m (hot smoking)	-20 C° /60 hrs
Gyrodactylus salaris and related monogenean	Y	Y	SKF	Y	NA	-20 C° /24 hrs
Digenean pathogens	N	Y	SKF	Y	70° C/ 20m	-20 C° /168 hrs
Cestode pathogens	N	Y	NA	Y	60° C/ 1m	-20 C° /168 hrs

Footnote to Appendix 3

Time	m = minutes, hrs=hours, d = day
OIE-listed	Disease is listed in the OIE Aquatic *Code* (2016a)
Country freedom	Declaration of country freedom from the Competent Authority of the exporting country may be accepted through the MPI Country Approval Procedures.
Restrict to wild-caught fish	Restrict commodity to wild-caught fish (not from aquaculture) (Y) yes, (N) no
Processing	GGU Head-on, gills in, gutted product (where gutted means "evisceration" for teleost fish, "trunking" for chimaera, sharks, guitarfish and sawfish, and the retention of the "wings" for skates and rays) HGU Headed, gilled and gutted product SKF Processed to skin-off fillet product NA Further processing is unlikely to substantially reduce pathogen occurrence (refer to appropriate pathogen chapter)
General OIE recommended treatments	For OIE-listed diseases heat treatments and approved commodities (OIE 2016a), include: • Heat-sterilised hermetically sealed product (cooked to 121°C for at least 3.6 minutes) or, • Pasteurised products (cooked to 90°C for at least 10 minutes) or, • Mechanically dried eviscerated fish (cooked to 100°C for at least 30 minutes, or, any other time/temperature combination that has been shown to inactivate the pathogen (refer to appropriate OIE Aquatic *Code* chapter). • Frozen fish fillets • Frozen fish fillets Negligible risk is likely where processing and commodity types for non-OIE listed pathogens are consistent with these OIE guidelines.
Alternative heat treatments	Alternative heat treatments from the literature may be available. Where no treatment is available (NA), the OIE–listed heat treatments should be used
Frozen storage	Frozen storage temperatures are provided from the literature in the relevant pathogen chapter. Where identified risk organisms are unaffected by low temperatures, frozen storage is not an effective risk management option (Not applicable, NA).
Microsporidian pathogens	Muscle encysting microsporidians (*Glugea plecoglossi, Pleistophora* (*Heterosporis*) *anguillarum, Kabatana arthuri, Microsporidium seriolae, Nucleospora* (*Enterocytozoon*) *salmonis* and *Tetramicra brevifilum*)
Myxosporean pathogens	Muscle encysting myxosporeans of genera *Ceratomyxa, Enteromyxum, Henneguya, Kudoa, Myxobolus, Parvicapsula, Sphaerospora, Thelohanellus* and *Unicapsula*

55. Appendix 4. Evaluation of risk management options

This summarises the qualitative evaluation of the expected reduction in pathogen load associated with each risk management option in the relevant risk organism chapter, where the OIE- recommended heat treatments and commodity definitions are provided in Appendix 3. The estimated levels of reduction in occurrence are:

Eliminate	> 95% reduction in pathogen load
Substantial	71-95% reduction in pathogen load
Moderate	51-70% reduction in pathogen load
Slight	< 50% reduction in pathogen load

Appendix 4a. Viral Pathogens: OIE-Listed Diseases

General information on pathogens and level of reduction in pathogen load by various risk management measures, where "No effect" means a risk management option is unlikely to reduce pathogen occurrence and NA means a risk management option is not applicable. Definitions of pathogens are provided in Appendix 1 and the OIE- recommended heat treatments and commodity definitions are provided in Appendix 3.

	EHNV/ ECV/ ESV	IHNV	ISAV	KHV	RSIV	SAV	SVCV	VHSV
	General Pathogen Information							
No. susceptible fish families	8	11	3	4	21	5	7	42
	Risk Management Measures Based on OIE Recommendations							
Country freedom accepted	Substantial	Substantial	Substantial	Substantial	Substantial	Substantial	Substantial	Substantial
OIE approved processed state	Eliminate	Eliminate	Eliminate	Eliminate	Eliminate	Eliminate	Eliminate	Eliminate
OIE approved heat treatment	Eliminate	Eliminate	Eliminate	Eliminate	Eliminate	Eliminate	Eliminate	Eliminate
Species declaration	Substantial	Substantial	Substantial	Substantial	Substantial	Substantial	Substantial	Substantial
Restrict to wild-caught fish	No effect	No effect	No effect	No effect	No effect	No effect	No effect	No effect
	Additional Processed States (values determined from reference material)							
Removal of gills (GGU)	Slight	Slight	Slight	Slight	Slight	Slight	Slight	Slight
Removal of head and gills (HGU)	Moderate	Moderate	Moderate	Slight	Moderate	Moderate	Slight	Slight
Skin-off fillet (SKF)	NA	NA	NA	Moderate	NA	NA	Moderate	Moderate
Alternative heat treatment	Eliminate	Eliminate	Eliminate	NA	Eliminate	NA	Eliminate	NA
Frozen storage	No effect	No effect	No effect	No effect	No effect	No effect	No effect	No effect

Appendix 4b. Viral Pathogens: Non-OIE-Listed Diseases.

General information on pathogens and level of reduction in pathogen load by various risk management measures, where "No effect" means a risk management option is unlikely to reduce pathogen occurrence and NA means a risk management option is not applicable. Definitions of pathogens are provided in Appendix 1 and the OIE-recommended heat treatments and commodity definitions are provided in Appendix 3.

	EVE	EEHV	GCHV	GIV	HIRRV	IPNV
	General Pathogen Information					
Affects salmonids (Y/N)	Y	N	N	N	Y	Y
No. susceptible fish families	3	1	1	2	9	41
	Risk Management Measures Based on OIE Recommendations					
Country freedom accepted	Substantial	Substantial	Substantial	Substantial	Substantial	Substantial
OIE approved processed states	Eliminate	Eliminate	Eliminate	Eliminate	Eliminate	Eliminate
OIE approved heat treatment	Eliminate	Eliminate	Eliminate	Eliminate	Eliminate	Eliminate
Species declaration	Substantial	Substantial	Substantial	Substantial	Substantial	Substantial
Restrict to wild-caught fish	No effect	No effect	No effect	Moderate	No effect	No effect
	Additional Processed States (values determined from reference material)					
Removal of gills (GGU)	Substantial	Slight	Slight	Slight	Slight	Slight
Removal of head and gills (HGU)	NA	Moderate	Moderate	Moderate	Slight	Moderate
Skin-off fillet (SKF)	NA	NA	NA	NA	Moderate	NA
Alternative heat treatment	Eliminate	Eliminate	Eliminate	Eliminate	Eliminate	Eliminate
Frozen storage	No effect	No effect	No effect	No effect	No effect	No effect

Appendix 4b (Continued)

	NJV	NNV	OMV	PRV	SGVP
	General Pathogen Information				
Affects salmonids (Y/N)	Y	Y	Y	Y	Y
No. susceptible affected fish families	2	59	1	11	3
	Risk Management Measures Based on OIE Recommendations				
Country freedom accepted	Substantial	Substantial	Substantial	Substantial	Substantial
OIE approved processed states	Eliminate	Eliminate	Eliminate	Eliminate	Eliminate
OIE approved heat treatment	Eliminate	Eliminate	Eliminate	Eliminate	Eliminate
Species declaration	Substantial	Substantial	Substantial	Substantial	Substantial
Restrict to wild-caught fish	Moderate	Moderate	No effect	No effect	No effect
	Additional Processed States (values determined from reference material)				
Removal of gills (GGU)	Slight	No effect	Slight	Slight	Slight
Removal of head and gills (HGU)	Moderate	Slight	Slight	Moderate	Moderate
Skin-off fillet (SKF)	NA	Moderate	Moderate	NA	Substantial
Alternative heat treatment	NA	Eliminate	Eliminate	Eliminate	Eliminate
Frozen storage	No effect	No effect	Eliminate	No effect	No effect

Appendix 4c. Bacterial Pathogens: Non-OIE-Listed Diseases

General information on pathogens and level of risk reduction by various risk management measures. General information on pathogens and level of reduction in pathogen load by various risk management measures, where "No effect" means a risk management option is unlikely to reduce pathogen occurrence and NA means a risk management option is not applicable. Definitions of pathogens are provided in Appendix 1 and the OIE- recommended heat treatments and commodity definitions are provided in Appendix 3. Aeromonas hydrophila occurs generally in salmonids, but exotic strains are only reported from non-salmonids. These strains may potentially affect all freshwater fish.

	***Aeromonas hydrophila* (exotic Strains)**	***Aeromonas salmonicida* (atypical Strains)**	***Aeromonas salmonicida* (typical Strains)**	***Edwardsiella* spp. species complex**	***Flavobacterium columnare* (exotic Strains)**	***Francisella* spp.**	***Moritella viscosa***
General Pathogen Information							
Affects salmonids	N	Y	Y	Y	Y	Y	Y
No. susceptible fish families	4	25	10	25	11	8	6
Risk Management Measures Based on OIE Recommendations							
Country freedom accepted	Substantial	Substantial	Substantial	Substantial	Substantial	Substantial	Substantial
OIE-approved processed states	Eliminate	Eliminate	Eliminate	Eliminate	Eliminate	Eliminate	Eliminate
OIE-approved heat treatment	Eliminate	Eliminate	Eliminate	Eliminate	Eliminate	Eliminate	Eliminate
Species declaration	Substantial	Substantial	Substantial	Substantial	Substantial	Substantial	Substantial
Restrict to wild-caught fish	Moderate	Moderate	Moderate	No effect	Moderate	Moderate	No effect
Additional Processed States (values determined from reference material)							
Removal of gills (GGU))	Slight	Slight	Slight	Slight	Slight	Slight	Slight
Removal of head and gills (HGU)	Slight	Slight	Slight	Slight	Slight	Slight	Slight
Skin-off fillet (SKF)	Moderate	Moderate	Moderate	Moderate	Moderate	Moderate	Moderate
Alternative heat treatment)	Eliminate	Eliminate	Eliminate	Eliminate	Eliminate	Eliminate	Eliminate
Frozen storage	No effect	No effect	No effect	No effect	No effect	No effect	No effect

Appendix 4c (Continued)

	Piscirickettsia salmonis, Tasmanian and other Rickettsia-like bacteria	*Pseudomonas anguilliseptica*	*Renibacterium salmoninarum*	*Streptococcus* species complex (exotic strains)	*Yersinia ruckeri* (exotic strains)
	General Pathogen Information				
Affects salmonids (Y/N)	Y	Y	Y	Y	Y
No. susceptible fish families	6	16	9	21	11
	Risk Management Measures Based on OIE Recommendations				
Country freedom accepted	Substantial	Substantial	Substantial	Substantial	Substantial
OIE-approved processed states	Eliminate	Eliminate	Eliminate	Eliminate	Eliminate
OIE-approved heat treatment	Eliminate	Eliminate	Eliminate	Eliminate	Eliminate
Species declaration	Substantial	Substantial	Substantial	Substantial	Substantial
Restrict to wild-caught fish	No effect	Moderate	Moderate	No effect	No effect
	Additional Processed States (values determined from reference material)				
Removal of gills (GGU)	Slight	Slight	Slight	Slight	Slight
Removal of head and gills (HGU)	Slight	Moderate	Moderate	Slight	Moderate
Skin-off fillet (SKF)	Moderate	NA	NA	Moderate	NA
Alternative heat treatment)	Eliminate	Eliminate	Eliminate	Eliminate	Eliminate
Frozen storage	No effect	No effect	No effect	No effect	No effect

Appendix 4d. Other Pathogens: OIE and Non-OIE-Listed Diseases

General information on pathogens and level of reduction in pathogen load by various risk management measures, where "No effect" means a risk management option is unlikely to reduce pathogen occurrence and NA means a risk management option is not applicable. Definitions of pathogens are provided in Appendix 1 and the OIE- recommended heat treatments and commodity definitions are provided in Appendix 3. *Sphaerothecum destruens* is an exotic pathogen reported from cyprinids and salmonids, that may potentially affect all freshwater fish

	Aphanomycesinvadans **(OIE-listed)**	*Ichthyophonus hoferi* (Not OIE-listed)	*Sphaerothecum destruens* (Not OIE-listed)	Microsporidian pathogens (muscle encysting) (not OIE-listed)	Myxosporean pathogens (muscle encysting) (not OIE-listed)
	General Pathogen Information				
Affects salmonids (Y/N)	Y	Y	Y	Y	Y
No. susceptible fish families	42	15	All freshwater fish	8	42
	Risk Management Measures Based on OIE Recommendations				
Country freedom accepted	Substantial	Substantial	Substantial	Substantial	Substantial
OIE approved processed states	Eliminate	Eliminate	Eliminate	Eliminate	Eliminate
OIE approved heat treatment	Eliminate	Eliminate	Eliminate	Eliminate	Eliminate
Species declaration	Eliminate	Moderate	No effect	Substantial	Moderate
Restrict to wild-caught fish	Moderate	No effect	Moderate	Moderate	Moderate
	Additional Processed States (values determined from reference material)				
Removal of gills (GGU)	Slight	Slight	Slight	Slight	Slight
Removal of head and gills (HGU)	Slight	Slight	Slight	Slight	Slight
Skin-off fillet (SKF)	Moderate	Moderate	Moderate	Moderate	Moderate
Alternative heat treatment	NA	Eliminate	Eliminate	Eliminate	Eliminate
Frozen storage	Eliminate	Eliminate	Eliminate	Eliminate	No effect

Appendix 4d (Continued)

	Anguillicola crassus (not OIE-listed)	*Gyrodactylus salaris* (OIE-listed) and related monogeneans	digenean pathogens (including muscle encysting larvae) (not OIE-listed)	Cestode larvae (muscle encysting) (not OIE-listed)
	General Pathogen Information			
Affects salmonids	N	Y	Y	Y
No. susceptible fish families	9	23	16	12
	Risk Management Measures Based on OIE Recommendations			
Country freedom accepted	Substantial	Substantial	Substantial	Substantial
OIE approved processed states	Eliminate	Eliminate	Eliminate	Eliminate
OIE approved heat treatment	Eliminate	Eliminate	Eliminate	Eliminate
Species declaration	Substantial	Substantial	Substantial	Substantial
Restrict to wild-caught fish	No effect	Moderate	Moderate	Moderate
	Additional Processed States (values determined from reference material)			
Removal of gills (GGU)	Slight	Slight	Slight	No effect
Removal of head and gills (HGU)	Slight	Moderate	Slight	No effect
Skin-off fillet (SKF)	Slight	Substantial	Moderate	Slight
Alternative heat treatment	Eliminate	NA	Eliminate	Eliminate
Frozen storage	Eliminate	Eliminate	Eliminate	Eliminate