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Cat fur mite in New Zealand Detection tools for brown marmorated stink bug Recreational vessels as a vector of exotic ant incursions

Ministry for Primary Industries Manatū Ahu Matua







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**Surveillance** is published as the Ministry for Primary Industries' authoritative source of information on the ongoing biosecurity surveillance activity and the health status of New Zealand's animal and plant populations in both terrestrial and aquatic environments. It reports information of interest both locally and internationally and complements New Zealand's international reporting.

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#### **Editorial**

# 2016: A year of international co-operation for animal health and agriculture

This has been an exciting year for international animal health and agricultural co-operation. The Ministry for Primary Industries (MPI) actively engages with its trading partners in bilateral, plurilateral and multilateral forums to help improve the trading environment. One way that it does this is through agricultural co-operation. Agricultural co-operation may contribute to:

- fulfilling obligations to which New Zealand is committed under trade agreements;
- building greater understanding and recognition of NZ biosecurity, food safety and farming systems;
- promoting our goods and services export sectors; and
- building our capacity and capability through collaboration.

As part of these co-operation agreements, members of MPI's Investigation and Diagnostic Centres and Response Directorate (IDC&R), including staff from the Animal Health Laboratory, the Surveillance and Incursion Investigation Group and the Response Group, have successfully established collaborative relationships with a number of international partners.

For example, April 2016 saw the inaugural Korea New Zealand Animal Health and Veterinary Epidemiology Workshop, held in Seoul, South Korea. This workshop was established under the NZ-Korea Free Trade Agreement (NZK-FTA) with the objective of sharing of animal health and veterinary epidemiological expertise between the two countries for mutual benefit. This workshop was highly successful, with a follow-up workshop in the advanced stages of planning for early 2017 and will be hosted in New Zealand.

A similar initiative has begun with China. New Zealand is developing a closer relationship with China in the area of biosecurity through collaboration. MPI and China's Ministry of Agriculture signed a work plan of co-operation in animal disease prevention and control, in November 2016. The scope of the work involves four collaborating organisations: MPI, Massey University's Epi-Centre, China's Animal Health and Epidemiology Centre and China's Animal Disease Control Centre. Collaboration among these agencies will centre on training, exchanges and research.

MPI has also assisted the OIE/FAO global effort against foot and mouth disease (FMD) through support of the South East Asia and China Foot-and-Mouth Disease Control Programme (SEACFMD). MPI has a Memorandum of Understanding with the OIE to provide technical expertise to this programme. Recent activities have been focused in Lao PDR and Myanmar. New Zealand is officially free of FMD but globally the disease is hugely significant and can be highly challenging to control.

Additionally, MPI works with other countries to help build understanding and confidence in our primary sector and systems. In response to multiple requests for similar information, MPI has developed a Supply Chain Integrity Programme, which identifies the key components of a supply chain in areas such as biosecurity, food safety and animal welfare. It explains in specific detail why we are a trusted, respected and beneficial trading partner. Under this programme of work, MPI has hosted a number of overseas delegations from counterpart agencies (e.g., in Brazil and Colombia) and explored the key elements and principles necessary to ensure the integrity of the supply chain (robust and verifiable systems) for different sectors. The programme looks at both regulatory and non-regulatory systems.

The initiatives described above are only a few of the multiple streams of work being undertaken in animal health and agricultural co-operation by MPI. Working with our international partners on information sharing and disease control programmes provides significant benefit for all involved. The year 2017 looks like being an exciting one for international co-operation in animal health and agriculture and we look forward to building further on the relationships developed in 2016.

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# First report of the cat fur mite *Lynxacarus radovskyi* in New Zealand

In Northland during June 2016 unusual mites were discovered on a stray farm kitten that had specks of dandruff-like material on its back. The veterinarian suspected these were cat fur mites, Lynxacarus radovskyi (Tenorio 1974), and notified MPI about the unusual find. The identification was subsequently confirmed as *L. radovskyi* by a parasitologist. This mite had not previously been reported from the NZ cat population, although in 1999 it was detected on an imported cat in quarantine. This article provides the first report of this species from the NZ postborder space.

#### **Global distribution**

*L. radovskyi* is a widely distributed parasite of cats and has been reported in many parts of the world, including the southern US, Fiji, Brazil, Malaysia and Australia (**Figure 1**). It was first discovered in Hawai'i four decades ago (Tenorio 1974), and was found in Australia soon after (Bowman & Domrow 1978). Reports in the literature are relatively few, presumably because the mite typically keeps a low profile on its hosts, and because its small size prevents easy detection except when it reaches high numbers. Clinical signs of mite infestation (pruritis, alopecia, hairballs) overlap substantially with those of the cat flea (*Ctenocephalides felis*). Cat fleas are ubiquitous and are easier to see than mites, which helps explain how mite infection can be missed during co-infection with cat fleas. In addition, another fur mite, *Cheyletiella blakei* (Smiley, 1970), can cause a similar dandruff-like appearance.

#### Epidemiology of infection

Detection of *L. radovskyi* is infrequent even within populations of cats that are not treated regularly with parasiticides. For example, in Brazil only one percent of untreated stray cats were found to be infested, compared with 60 percent infested with the cat flea (Mendesde-Almeida et al. 2011). In Malaysia, various cat populations were surveyed and the prevalence of infested cats was 1.5-5 percent (Jeffery et al. 2012), though in most cases the number of mites found was very low (one or two). The mite's life cycle is not well known, but transmission is assumed to occur by direct contact between animals, and has been suggested to occur through fomites Clare et al. 2004).

#### **Clinical signs and treatment**

Infection with *L. radovskyi* is often subclinical, but has also been associated



Figure 1: *Lynxacarus radovskyi* cat fur mites from an infested kitten in Northland. Some of the mites are clinging to hair shafts. (Courtesy of Leeann Ruddell and Jane Nichols, Northland Veterinary Group.)

with pruritis, alopecia, increased hairballs, irritability and other signs. There are a few single clinical case reports in the literature, but only one report of a population-level outbreak of disease (Foley 1991), which describes an epidemic of itching, hair loss and secondary signs among about 300 cats in a Florida Keys neighbourhood. In the same report one cat owner reported itchiness, which resolved after their cat was treated. L. radovskyi is reportedly susceptible to commonly used parasiticides, for example fipronil eliminated mites from 100 percent of naturally infected cats in one study (Clare et al. 2004).

#### Situation in New Zealand

Based on its reported distribution, the mite seems to favour a warm, humid climate (Craig et al. 1993). The present find from Northland is consistent with this pattern. It is not known how many NZ cats or what populations and regions are affected. However, to date no other reports have been generated by passive surveillance via commercial diagnostic laboratories or the exotic pest and disease hotline. Given the global distribution of the parasite, and its cryptic, dandrufflike appearance, it could be widespread within the cat population without having previously been detected. MPI's Import Health Standard for cats requires that imported animals be examined for external parasites and treated with parasiticides. In 1999, mites were detected during quarantine of a cat recently imported from Western Samoa (Heath & Mariadass 1999), indicating that a pathway exists but suggesting that quarantine measures are robust enough to catch at least some cases. The origin of the mite population reported here is unclear. The host kitten was a stray that had no history of contact with imported cats and could be one of the first of its kind to be infected. However, it is more likely that the present find represents the first detection of a parasite that has been present for some time.

#### Conclusion

This case of cat fur mite is considered to represent a baseline surveillance find, and is considered unlikely to significantly impact on either primary industry or on companion animal health. However, veterinary professionals may see occasional cases and are encouraged to report these to MPI through the exotic pest and disease hotline in order to increase our awareness of the distribution of this parasite.

#### Acknowledgements

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# Avian influenza: epidemiology and surveillance in New Zealand

#### Avian influenza epidemiology

Avian influenza (AI) refers to infection of birds with avian influenza A viruses of the family Orthomyxoviridae. These RNA viruses are widespread, highly contagious and extremely variable. AI viruses are most frequently recorded in waterfowl (defined for this paper as members of the order Anseriformes - ducks, geese, and swans), which are considered to be the biological and genetic reservoirs of all AI viruses and the primordial reservoir of all influenza viruses for birds and mammals (Webster et al. 1992; Stallknecht 1998; Perdue et al. 2000). Carriage of AI is also seen to a lesser extent among two families within the order Charadriiformes: the Laridae (gulls and terns), and Scolopacidae (waders). The Charadriiformes are considered of secondary importance, given that AI viruses are only detected seasonally and at low prevalence when compared to the Anseriformes (Olsen et al. 2006; Munster et al. 2007; Whitworth et al. 2007). Wild birds, particularly migratory waterfowl, may play a major role in the initial introduction of AI viruses into commercial poultry (Halvorson et al. 1985; Hinshaw et al. 1986).

#### New Zealand official status

New Zealand is free from high-pathogenicity avian influenza (HPAI) and has never had a case of low-pathogenicity avian influenza (LPAI) in poultry. New Zealand's claim of freedom from HPAI is based on the historical freedom provisions of Article 1.4.6a of the OIE *Terrestrial Animal Health Code* (Anon 2015). Addressing the specific requirements detailed in Article 1.4.6a:

- New Zealand has never had an occurrence of HPAI;
- HPAI has been notifiable for at least the last 10 years;
- an early detection system is in place for all susceptible species;
- measures to prevent the introduction of HPAI are in place;
- no vaccination against HPAI has been carried out; and

• infection with HPAI is not established among wild birds in New Zealand. There has never been a case of HPAI in wildlife, although LPAI has been detected sporadically in wild waterfowl (Stanislawek *et al.* 2002; Stanislawek *et al.* 2016)

However, once AI becomes established in commercial poultry, wild birds have very little or no role in secondary dissemination (Nettles *et al.* 1985).

The influenza A virus is subtyped based on serologic reactions to the haemagglutinin (HA) and neuraminidase (NA) surface glycoproteins (WHO Expert Committee 1980). Sixteen subtypes of HA and nine subtypes of NA are recognised. The distribution of virus subtypes varies by year, geographic location and host species (Swayne & Halvorson 2008). The surface glycoproteins are the major targets of the host immune response. There is little or no cross-protection between different HA or NA types (Anon 2014; Swayne *et al.* 2008).

The diversity of influenza A viruses and their tendency for ongoing genetic change cause a considerable challenge



Figure 1: The nine major migratory bird flyways of the world

as two viruses that share a subtype may be only distantly related. Some of this variability results from a process known as antigenic drift - the gradual accumulation of mutations. As a consequence of this, the viral HA or NA surface glycoproteins may change sufficiently for the immune responses generated against these glycoproteins to no longer be effective. A more rapid change can occur through a process known as genetic reassortment, which occurs when two different influenza viruses infect the same cell. In this situation, gene segments from both viruses may be packaged into a single novel viron. If genetic reassortment results in the acquisition of new HA or NA glycoproteins, this can cause an antigenic shift among the viruses circulating in a species. This may be sufficient for the reassortant virus to evade existing immunity or to significantly change its virulence for birds or mammals. (Anon 2014; Swayne et al. 2008).

There are two well-recognised lineages of avian influenza viruses: Eurasian and North American. The amount of reassortment between these lineages differs between regions, with very few reassortant viruses detected in some areas or among some wild bird populations, but significant reassortment where there is overlap between migratory flyways, such as in Alaska (**Figure 1**). Viruses in wild birds are more likely to be transferred between hemispheres in the latter regions (Anon. 2014)

#### **OIE definitions**

The OIE defines AI as an infection of poultry caused by any influenza A virus of the H5 or H7 subtypes, or by any influenza A virus with an intravenous pathogenicity index (IVPI) greater than 1.2 (or alternatively, at least 75 percent mortality), as described below. These viruses are further divided into two categories: HPAI and LPAI viruses.

#### **HPAI viruses**

These are influenza A viruses that have an IVPI in six-week-old chickens greater than 1.2; or alternatively, that cause at least 75 percent mortality in fourto eight-week-old chickens infected intravenously. H5 and H7 viruses that do not have an IVPI of greater than 1.2 or cause less than 75 percent mortality in an intravenous lethality test should be sequenced to determine whether multiple basic amino acids are present at the cleavage site of the haemagglutinin molecule (HA0). If the amino-acid motif is similar to that observed for other highpathogenicity avian influenza isolates, the isolate being tested should be considered as HPAI virus.

#### LPAI viruses

These are all influenza A viruses of H5 and H7 subtypes that are not highpathogenicity avian influenza viruses (Anon. 2015).

For international reporting purposes the OIE requires reporting of cases of LPAI and HPAI (as defined above) in poultry, and infection with HPAI in birds other than poultry (including wild birds). The OIE defines poultry as "all domesticated birds, including backyard poultry, used for the production of meat or eggs for consumption, for the production of other commercial products, for restocking supplies of game, or for breeding these categories of birds, as well as fighting cocks used for any purpose" (Anon. 2015).

LPAI infection of domestic poultry can result in mild to severe respiratory signs including coughing, sneezing, rales, rattles and excessive lacrimation. Generalised clinical signs such as huddling, ruffled feathers, depression, lethargy and occasionally diarrhoea have also been described. Layers may show decreased egg production. High morbidity and low mortality are normal for LPAI infections (Swayne & Halvorson 2008). Intratracheal inoculation of poultry with LPAI can result in localised infection of the upper and lower respiratory tract (tracheitis, bronchitis, airsaccultitis and pneumonia), with histological lesions and viral antigen distribution restricted to the lungs and trachea, although pancreatic necrosis is also reported (Swayne et al. 1992; Shalaby et al. 1994; Mo et al. 1997; Capua et al. 2000). Intravenous inoculation of poultry with LPAI results in swollen and mottled kidneys, with necrosis of the renal tubules and interstitial nephritis noted on histopathology and high viral titres in kidney tissues (Slemons & Swayne 1990; Swayne & Slemons 1990; Slemons & Swayne 1992; Swayne & Slemons 1992; Shalaby et al. 1994; Swayne & Alexander 1994; Swayne et al. 1994; Swayne & Slemons 1995). However, this renal tropism is strain-specific and is most consistently associated with

experimental intravenous inoculation studies (Swayne & Halvorson 2008), though Alexander & Gough (1986) did report the recovery of H10N4 LPAI from the kidneys of hens presenting with nephropathy and visceral gout. Salpingitis associated with a nonpathogenic H7N2 virus was described by Zielger *et al.* (1999).

In contrast, most cases of HPAI infection of domestic poultry are associated with severe disease, with some birds being found dead before clinical signs are noticed. Clinical signs such as tremors, torticollis and opisthotonus may be seen for three to seven days before death. Precipitous drops in egg production in breeders and layers are reported. Morbidity and mortality are usually very high (Swayne & Halvorson 2008). HPAI of poultry results in necrosis and inflammation of multiple organs including the cloacal bursa, thymus, spleen, heart, pancreas, kidney, brain, trachea, lung, adrenal glands and skeletal muscle (Mo et al. 1997; Swayne 1997; Perkins & Swayne 2001). Histopathological lesions described include diffuse nonsuppurative encephalitis, necrotising pancreatitis and necrotising myositis of skeletal muscles (Acland et al. 1984). Viral infection of the vascular endothelium is suggested as the mechanism for the pathogenesis of HPAI infections in poultry, especially the CNS lesions (Kobayashi et al. 1996a; Kobayashi et al. 1996b). Viral antigen can be detected in multiple organs, most commonly the heart, lung, kidney, brain and pancreas (Mo et al. 1997).

#### Epidemiology of AI in New Zealand

New Zealand lies at the southeastern extremity of the East Asian-Australasian Flyway (Figures 1 and 2), which is an important consideration regarding the likelihood of introduction of any new AI viruses into the country. Because of this geographical isolation, relatively few migratory birds reach the country. In total, about 200 000 birds of 47 species arrive as annual migrants, but more than 90 percent of these are just two species of wader (Scolopacidae): bar-tailed godwits (Limosa lapponica) and red (lesser) knots (Calidris canutus) (Williams et al. 2006). Most godwits are believed to fly here directly from Alaska, while red knots have stopovers along the Asian coast as they migrate from the Siberian



Figure 2: The East Asian-Australasian flyway. Source: Adrian Riegen, Miranda Shorebird Centre brochure. From http://www.teara.govt.nz/en/map/7218/the-east-asian-australasian-flyway

Arctic. Additionally, small numbers of other waders such as the Pacific golden plover (*Pluvialis fulva*) and red-necked stint (*Calidris ruficollis*) migrate to New Zealand via a number of stops in the Pacific. These birds share summer breeding grounds in the Arctic regions of Siberia and Alaska with species from Eurasia and the Americas (**Figure 2**) (Bulach *et al.* 2010).

In contrast to the Americas and Europe, where regular migrations of waterfowl are established, New Zealand is not on any waterfowl migration pathway, so our populations are isolated from all others except occasional Australian vagrants (Williams *et al.* 2006). When considering the movement of avian influenza viruses around the world this is important, as previous studies have suggested that members of the order Anseriformes, and particularly the family Anatidae, play an important role as the natural reservoirs of AI virus (Gilbert *et al.* 2006, Hars *et al.* 2008, Bulach *et al.* 2010)

Further minor migration links include

those of pelagic (ocean-ranging) seabirds such as petrels and albatrosses, which breed on and around the New Zealand coast during the southern hemisphere summer. These birds migrate to maritime regions of the northern Pacific associated with Japan, Russia, and Alaska, with some travelling as far as the west coasts of North and South America (Bulach *et al.* 2010).

Some trans-Tasman migration of seabirds between New Zealand and Australia also takes place. For example, the Australasian gannet (*Morus serrator*) and whitefronted tern (*Sterna striata*) breed in New Zealand, then migrate to Australian coastal areas for their adolescence or during the non-breeding season (Marchant & Higgins 1990). On their return to New Zealand, they associate at roosts or nesting sites on beaches, headlands and small offshore islands, mingling with adult gannets and terns that have overwintered in New Zealand waters (Williams *et al.* 2006).

With respect to the importance of the migratory species reaching New Zealand as a source of new AI viruses, Bulach et al. (2010) speculated that the pelagic seabirds and waders that often migrate into North America do not play a role in the movement of influenza viruses to Australia and New Zealand. This conclusion was based on the closer relationship of the Australian and New Zealand H7 isolates to Eurasian AI virus isolates than to North American isolates, despite the flyways that link the Australia, New Zealand and North America regions. Further support for this conclusion has come from surveys carried out by the Ministry for Primary Industries (MPI) National Animal Health Laboratory during the first six years of the avian influenza surveillance

programme (2004–2010). These surveys targeted migratory birds, in particular the bar-tailed godwit and red (lesser) knot. No AI virus was isolated, indicating that migratory birds pose a very low risk for the introduction of AI to New Zealand (Stanislawek *et al.* 2015). This conclusion is supported by findings from similar surveillance of migratory birds in Australia (Curran *et al.* 2014).

Phylogenetic analysis of the HA gene of the New Zealand H5 viruses shows that these cluster together and appear to be genetically closer to the lowpathogenic North American H5 wild bird viruses than to H5 wild bird viruses from Eurasian lineage (Stanislawek, unpublished data). While this does not fully support the theory developed for New Zealand H7 viruses by Bulach et al. (2010), the genetic differences in the relationship of the H5 and H7 viruses in New Zealand could be explained by the early introduction of mallard ducks (Anas platyrhynchos) that were most likely already infected with AI viruses, from Europe in the 1860s and from North America in the 1930s and 1940s (Heather & Robertson 1996).

In conclusion, New Zealand is geographically isolated from the migratory species that are influential in the spread of avian influenza viruses around the world. In contrast to the situation in North America, Asia and Europe, the circulating subtype H5 and H7 avian influenza viruses in New Zealand appear to have undergone an extended period of genetic isolation. This is supported by the work undertaken by Bulach *et al.* (2010) and Stanislawek (unpublished data).

#### **MPI AI surveillance**

New Zealand's AI surveillance programme is multi-faceted, incorporating active surveillance of resident wild birds, and enhanced passive surveillance. New Zealand has never had a case of HPAI in wild birds or poultry, or a case of LPAI in poultry.

#### Wild bird surveillance

From 2004 to 2015, MPI, in conjunction with the New Zealand Fish and Game Councils, the Department of Conservation and other stakeholders, carried out surveillance for AI on targeted migratory and resident birds (**Table 1**). The first six years of surveillance focused on migratory birds, in particular the bar-tailed godwit, and red (lesser) knots, on their arrival from late September to November, at Miranda,

Table 1: Active surveillance for H5 and H7	7 subtypes of AI in wild birds	in New Zealand, 2004–2015	

				RT/PCR pos	sitive	
Year	Species sampled	Cloacal samples tested	Oropharyngeal samples tested	H5	H7	Confirmed H5 or H7 isolates
2004	Mallard duck, red knot, bar-tailed godwit	469	0	0	0	0
2005	Mallard duck, paradise duck, red knot, bar- tailed godwit	1 089	0	0	0	0
2006	Mallard duck, red knot, bar-tailed godwit, ruddy turnstone	826	0	0	0	0
2007	Mallard duck, paradise duck, grey teal, red knot, bar-tailed godwit	950	174	2	0	2xH5*
2008	Mallard duck, paradise duck, red knot, bar- tailed godwit	1 484	343	37	0	5xH5*
2009	Mallard duck, red knot, bar-tailed godwit, black-billed gull, black-backed gull	1 480	1 480	4	0	0
2010	Mallard duck, black-billed gull, black-backed gull, wrybill, little blue penguin, yellow-eyed penguin, sooty shearwater	1 991	1 991	5	17	2xH7*
2011	Mallard duck	790	790	25	0	3xH5*
2012	Mallard duck	790	790	9	0	0
2013	Mallard duck	960	960	49	0	1xH5*
2014	Mallard duck	880	880	7	0	0
2015	Mallard duck	1 065	1 065	0	9	5xH7*
Total		12 774	8 473	138	26	11xH5*; 7xH7*

\* All H5 and H7 detections were low-pathogenicity avian influenza. All detections were in resident waterfowl.

their main North Island arrival site. Findings from surveillance undertaken from 2004 to 2010 indicate that migratory birds pose a very low risk for the introduction of AI to New Zealand, as no AI virus was isolated. These birds were targeted because of their migration pathway, along which AI viruses may be present: directly from the Arctic regions of Asia and North America in the case of the godwit, and from Arctic regions via the Pacific coast of Asia in the case of the knot. Subsequently, from 2010 to 2015 surveillance focused on resident birds, mainly waterfowl (Stanislawek et al. 2012) (Figure 3).

New Zealand is not on a migration pathway for waterfowl as observed in the northern hemisphere, although vagrant waterfowl from Australia are occasionally encountered. Nevertheless, since 2004, non-migratory waterfowl (predominantly mallard ducks) have also been sampled in the summer months throughout New Zealand, with a particular focus on coastal areas close to migratory shorebird habitats (Stanislawek *et al.* 2015) (**Figure 4**). This surveillance programme is ongoing.

# Enhanced passive surveillance

MPI operates a 24/7 toll-free exotic pest and disease emergency hotline and receives calls regarding sick and dead wild and domestic birds from members of the public, veterinarians, regional laboratory pathologists and others (Tana 2014; Stanislawek *et al.* 2015). Where reports relate to native birds, they are handled collaboratively with the Department of Conservation (DOC).



Figure 3: Locations of active surveillance for H5 and H7 subtypes of AI in wild birds, 2004–2015



Figure 4: Samples being collected from wild ducks in the Hauraki Plains area (left) and the mouth of the Kaituna River (right).

A risk assessment determines whether there is a need to investigate the report further. Key information used in the assessment includes:

- history of the event (numbers affected and timeline of events);
- presenting syndrome in dying birds;
- species of bird(s) affected;
- availability of fresh samples (where unavailable, follow-up is instigated);
- location; and
- epidemiological trends over space and time.

Based on the risk assessment, the report is either stood down or investigated further for a potential exotic or emerging disease aetiology (Stanislawek *et al.* 2015).

A rapid field service is in place for sample collection and submission of unexplained bird deaths (Rawdon et al. 2007; van Andel et al. 2010), using MPI-approved suppliers. A standardised investigation protocol, co-ordinated by the MPI Investigation and Diagnostic Centre at Wallaceville, is applied to submissions. The investigation protocol includes necropsy and sample collection for histology, bacteriology and virology. The presence of AI is assessed using influenza A real-time RT-PCR TaqMan (Spackman et al. 2003), with follow-up using realtime H5 and H7 RT/PCR TaqMan assays to exclude H5 and H7 subtypes (Slomka et al. 2007; Sidoti et al. 2010). Virus isolation is performed on samples that are positive in PCR assays (Stanislawek et al. 2002).

Reports on avian disease and mortality investigations are published quarterly in *Surveillance* as part of the IDC report of suspect exotic disease investigations.

Mortalities of threatened or critically endangered native birds are also monitored. Necropsies are performed by veterinary wildlife pathologists contracted to DOC and where required further diagnostic testing is undertaken to help make a definitive diagnosis. MPI also collects data from approved veterinary diagnostic laboratories on avian submissions from veterinary practitioners. Table 2 summarises submission data from the MPI passive surveillance system. Annual reports for the MPI AI surveillance programme can be found at the NZVA SciQuest site http://www.sciquest.org.nz/surveillance

#### Table 2: Avian submissions to the MPI Passive Surveillance System, 2004–2015

Year	Submissions from approved veterinary diagnostic labs	Wildlife submissions	MPI pest and disease hotline notifications	MPI investigations
2004	116	-	30	8
2005	340	-	85	8
2006	360	-	154	24
2007	33	-	60	14
2008	120	-	37	10
2009	163	-	*151	7
2010	174	-	25	7
2011	142	-	19	7
2012	290	16	19	8
2013	664	178	19	6
2014	385	141	30	13
2015	503	95	45	14

\*The greatly elevated number of bird mortality reports in 2009 was due to a toxicity event in August of that year caused by grey side-gilled sea slugs (Pleurobranchaea maculata) in the Auckland region. This event created considerable media interest and increased the number of calls to MPI's hotline from the public.

#### Industry surveillance

Surveillance for AI within the New Zealand poultry industry is multifaceted and supports the MPI passive surveillance system.

Previous work undertaken has included serological surveillance (2006-2007) and virological surveillance (2008-2009) for AI virus H5 and H7 subtypes in commercial poultry. This surveillance work was conducted by the then MAF Biosecurity New Zealand (now MPI), with the support of the Poultry Industry Association of New Zealand (PIANZ) and the Egg Producers Federation (EPF). The survey was designed using the 2005 OIE guidelines for AI surveillance. The serological survey used a crosssectional two-stage stratified design to ensure representative sampling of all poultry sectors and proportional regional representation of poultry farms. Production sectors included initially were broiler, caged/barn layer, free-range layer, pullet rearer and turkey broiler farms (Rawdon et al. 2010). Active surveillance was extended in 2008-2009 to ducks, quail, pheasants, partridges and guinea fowl produced for meat, eggs, or release in game reserves (Frazer et al. 2009; Frazer et al. 2010). This survey found no evidence of active infection with either H5 or H7 AI subtypes, and provided evidence of absence at a between-farm prevalence of 5 percent (95 percent confidence).

At present surveillance is conducted to meet the requirements of an MPI import

health standard (IHS). Breeder birds in donor flocks are tested as per the IHS before hatching eggs are imported. Further testing is conducted where any mortalities occur (as required by the IHS) and a sample of live chicks born in quarantine is also tested. AI surveillance is also undertaken to meet the trading partner's requirements for the export of table eggs, hatching eggs and day-old chicks. This aspect of industry surveillance is reported each year in the Surveillance Annual Report, http://www. sciquest.org.nz/node/123576). Neither LPAI nor HPAI has ever been detected in the New Zealand commercial poultry flock as a result of these surveillance activities.

Commercial poultry companies employ veterinarians to develop health plans for the flocks they are responsible for. These plans include the establishment, supervision and interpretation of routine testing for flock health and also the investigation of poor performance in a flock. Where exotic diseases such as AI are suspected, MPI is also notified via the exotic pest and disease hotline (see Enhanced passive surveillance, above) and, under the direction of MPI Incursion Investigators, further diagnostics may be carried out.

Poultry veterinarians are supported by two specialist poultry pathology laboratories that conduct avian lab tests for the commercial poultry industry and are audited to the NZS ISO/ IEC standard 17025:2005 "General requirements for the competence of testing and calibration laboratories" by International Accreditation New Zealand (IANZ) (http://www.ianz.govt.nz/). Both laboratories are also Recognised Laboratories under the MPI Export Laboratory Programme (https://mpi. govt.nz/exporting/overview/generalrequirements/export-laboratoryprogramme/). Poultry veterinarians can also use the private veterinary laboratory network for disease investigation

To further enhance the passive surveillance system, PIANZ and the EPF provide all their commercial poultry farmer members (who produce about 95 percent of all commercial poultry in New Zealand) with AI information posters for their poultry sheds (**Figure 5**). The objective is to raise awareness and provide information regarding early warning signs and the requirement to call the MPI exotic pest and disease hotline whenever there is a suspicion of AI (or other exotic diseases).

#### Import health standards

Import risk analysis (IRA) is a scientific discipline that transparently accommodates known facts, knowledge

gaps and uncertainty (Vose, 2008; World Organisation for Animal Health 2010). MPI uses IRA to identify pre-border hazards such as pathogens that may be associated with imported animals, and to assess the likelihood and consequences of introducing those hazards. IRA also informs control measures to manage the identified risks, and helps communicate the risks to others (Cobb & MacDiarmid, 2014).

An early study revealed that AI virus persisted in refrigerated muscle tissue for 287 days, although feeding meat or blood from a viraemic bird to a susceptible bird did not transmit infection (Purchase 1931). More recently, Swayne & Beck (2005) confirmed this when they demonstrated that LPAI virus could not be found in the blood, bone marrow, breast or thigh meat of experimentally infected poultry, and that feeding breast or thigh meat to a susceptible bird did not transmit infection. However, experimental infection of poultry with HPAI resulted in detectable virus in blood, bone marrow and breast and thigh meat. A H5N2 isolate was found to achieve only low viral titres in muscle tissue (10<sup>2.2-3.2</sup> EID50 virus/g), and

feeding this meat to susceptible birds did not transmit infection, whereas an H5N1 isolate achieved a much higher titre in muscle tissue (10<sup>7.3</sup> EID50 virus/g), which was sufficient to achieve transmission in a feeding trial. This study also demonstrated that AI virus vaccination prevented HPAI virus replication in muscle tissue. The authors concluded that the potential for LPAI virus to appear in the meat of infected chickens was negligible, while the potential for having HPAI virus in meat from infected chickens was high; but proper use of vaccines could prevent HPAI from being present in meat.

LPAI cannot be transmitted to susceptible birds by feeding meat from an infected bird. Following natural infection, LPAI virus replication is limited mainly to the respiratory tract tissues, although some infectivity may be associated with the pancreas, kidneys and reproductive tract. HPAI viruses replicate in a wide range of tissues, and studies have shown that feeding meat from an HPAIinfected bird can transmit the virus to a susceptible bird.

AI virus has been isolated from the internal contents of eggs from naturally

Although the symptoms of AI can be similar to those of other common poultry diseases, if you suspect your poultry are infected with AI then

An experienced exotic disease investigator with access to poultry

• the MPI Exotic Pest and Disease Hotline on 0800 80 99 66

### Symptoms of Avian Influenza (AI) in poultry

you must call:

your vet, or your technical advisor, or

specialists will answer your call.

#### Clinical signs of AI are highly variable.

They may be influenced by the strain of the virus, the age and species of bird affected, and the presence or absence of other diseases and prevailing environmental conditions.

The main symptoms include:

- sudden and unexplained deaths
- a rapid spread of the disease throughout the flock
- depression and loss of appetite
- a drop in egg production
- nervous signs
- facial swelling
- coughing, sneezing and diarrhoea









Swollen wattles & comb



Blue discoloration of comb in chicken on left, normal chicken on right

Figure 5: The AI information poster produced by PIANZ and EPF for poultry farmers

infected layer and breeder flocks with clinical disease, and from an infected layer flock with no clinical signs (Cappucci *et al.* 1985). Unpublished work by Brugh (cited by Swayne & Beck 2004) identified HPAI virus in 85–100 percent of eggs laid on days 3 and 4 following experimental inoculation. Although no reports of transmission of infection to chicks via infected eggs have been located, movement of egg trays and associated fomites was a significant risk factor in the spread of AI infection during an epidemic in the Netherlands in 2003 (Thomas *et al.* 2005).

New Zealand's import health standards for poultry products include sanitary measures for HPAI in poultry meat and all strains of avian influenza in poultry hatching eggs. For example, poultry meat or meat products must be derived from birds kept in a country, zone or compartment free from HPAI since hatching, or for 21 days before slaughter for export, with current OIE Code surveillance requirements being met to claim freedom from HPAI. Otherwise they must be cooked in accordance with the Code recommendations for inactivation of avian influenza virus in meat.

#### Conclusion

New Zealand is free from HPAI and has never had a case of LPAI in poultry. Internationally, waterfowl play a pivotal role in the epidemiology of AI and are responsible for its movement globally via overlapping migratory flyways. New Zealand is not on a migratory pathway for waterfowl, with only occasional vagrants arriving from Australia. Import health standards are in place to effectively manage the risks associated with live birds and poultry products. Active surveillance is undertaken in wild birds, enabling characterisation of the endemic viruses (Stanislawek et al. 2015) and the passive surveillance system allows for rapid detection of HPAI should it ever occur (Tana 2014).

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# Quarterly report of diagnostic cases: July to September 2016

#### Gribbles Veterinary Pathology

#### Bovine

In a mob of 100 ten-to-14-day-old dairy calves from the Auckland region, five had died after about a day of malaise and 10 were scouring with blood. Faecal testing of four showed that three were positive for *Cryptosporidium* on ELISA, and small-intestine histopathology of a sacrificed moribund calf showed marked villus blunting with crypt abscesses and basophilic objects attached to the epithelium, consistent with **cryptosporidiosis**.

A one-week-old dairy calf in a mob of 100 from the Waikato collapsed and died after scouring overnight. Faecal testing produced a culture of *Salmonella* Bovismorbificans, and a faecal *Cryptosporidium* ELISA test was positive, suggesting the animal had combined salmonellosis and cryptosporidiosis.

Two cases of Mortierella wolfii abortion occurred on the West Coast, both associated with the feeding of poorquality silage. In one case 14 cows had aborted and three of them had died. At postmortem one had a diffuse pneumonia and histologically there was acute pneumonia with massive numbers of fungal hyphae. Four of these cows had a fungal placentitis. In the other case, four cows had aborted and lesions of fungal placentitis were present in one placenta examined. Fungal encephalitis was present in one of the calves, and fungal hepatitis in another. The histological appearance of the fungal hyphae in all cases was consistent with Mortierella.

A few cases of **bovine neonatal pancytopenia** were noted this spring on the same South Canterbury farms that had been affected in previous years. The cases were confirmed on sternal bone marrow histology that showed a total or near-total lack of haematopoietic tissue.

Four two-year-old heifers suffered lateterm abortion in a mob of 50 heifers from a South Canterbury beef herd. **Ureaplasma diversum** was detected by PCR in the stomach contents of the two fetal calves that were sampled. Histologically, both had peribronchiolar lymphoid infiltrates and one had perivascular infiltrates of plasma cells in the brain and in portal areas of the liver. This infection has been suspected in the past but until now the lack of any means to readily detect the agent has made diagnosis difficult.

One outbreak of **sporadic bovine encephalomyelitis** has been seen this spring in a group of 90 calves on a North Canterbury dairy farm. The calves were a mixture of Friesian and Friesian crossbreds around two months old. Ten were affected and were unsteady and knuckling on their hind legs. Three developed severe neurological signs and died. Histological examination of the brains of two calves revealed lesions consistent with a diagnosis of sporadic bovine encephalomyelitis.

A mob of beef cows on an Otago farm had an outbreak of deaths in newborn calves. Twelve calves died within 24 hours of birth. No significant gross lesions were found on necropsy of two dead calves but one had fed and walked while the other had not. Bacterial cultures of the stomach contents of both were negative but **bovine viral diarrhoea** (**BVD**) virus was found in both by a BVD PCR test of the heart blood. The herd not been vaccinated against this virus.

In July, three pregnant heifers on an Otago dairy farm developed unilateral humeral fractures within a few days of each other. These animals were euthanased and their livers were tested for copper. Liver copper concentrations were very low, at 29, 32 and 47 umol/kg (adequate concentration > 95), confirming severe **copper deficiency**.

**Prototheca zopfii** was identified as the cause of a mastitis outbreak in a Waikato dairy herd. This alga had also caused a severe mastitis outbreak on the same property the previous year. More than 30 cows were affected on this occasion.

Salmonella Bovismorbificans was

isolated in outbreaks of calf diarrhoea from four different Waikato dairy properties during a seven-day period in August. It is not known whether there were any specific links between these two properties.

Over a period of three weeks, three mixed-age lactating Friesian cows died unexpectedly on a Bay of Plenty dairy farm. The cows had received selenium injections prior to death. Histopathological examination of the liver from one cow revealed mild chronic sporidesmin damage and subtle hepatocellular swelling. This animal had a normal liver copper concentration (96 µmol/kg; reference range 95–3000) but the liver selenium concentration was high, at 402 460 nmol/kg (reference range 850–15 000), confirming a diagnosis of **acute selenium toxicity.** 

One of 90 nine-month-old Friesian heifer calves on a Wairarapa farm was found recumbent with corneal oedema, mucopurulent nasal discharge and a fever of 40.5° C. The heifer was euthanased and the brain collected and processed for histopathology. Microscopic changes were characterised by widespread lymphohistiocytic vasculitis, confirming the clinical suspicion of **malignant catarrhal fever.** The causative agent of this disease in New Zealand is **ovine herpesvirus-2**, shed in highest quantities by lambs aged six to nine months.

Three of a mob of 70 eighteen-monthold beef bulls on a Central Hawke's Bay farm developed nervous signs and blindness, and then died within days after introduction to a brassica crop. The brain was collected from one of the dead animals. There were small foci of green fluorescence of the cortical gyri under ultraviolet light. Microscopic evaluation revealed tracts of necrosis in the deep cortical laminae, characterised by rarefaction of the neuropil, shrinkage and pyknosis of neurons, and hyperplasia of capillary endothelial cells. The pathological findings confirmed a diagnosis of polioencephalomalacia, which is typically induced by thiamine deficiency caused by dietary alteration, sulphur excess, or dietary thiaminases.

Outbreaks of bovine adenoviral enteritis occurred on two North Island farms during July. On one farm, three out of 80 one-year-old Friesian heifers were dead when checked at a run-off. An outbreak of adenovirus-related deaths had occurred on the same farm the previous year. No signs of illness were observed prior to death. Most of the alimentary tract was too autolysed to interpret. However, there were interstitial inflammatory changes accompanied by basophilic intranuclear inclusion bodies in endothelial cells of the kidney, compatible with adenoviral infection. In the other outbreak, two of 20 yearling Friesian calves presented moribund with sunken eyes, hypothermia and haemorrhagic diarrhoea. One calf died and the other was euthanased. Samples were collected from one animal and examined microscopically. There were areas of congestion, haemorrhage and erosion of the abomasal mucosa, small intestine and large intestine. Many associated vascular endothelial cells were swollen and contained large basophilic intranuclear inclusions. The kidney contained interstitial inflammation and endothelial inclusions. Culture of intestinal contents was negative for Salmonella spp.

#### **Ovine**

Several mixed-age Romney ewes from Northland were ill-thrifty and dying. A moribund ewe was euthanased. Post-mortem findings were a marked granulomatous enteritis and lymphangitis, with many large histiocytes containing acid-fast bacilli, consistent with **Johne's disease**.

A one-year-old ewe hogget on a Hawke's Bay sheep farm was growing poorly and had red urine. Post-mortem examination was remarkable for multiple 2–5-mm pale foci through the renal cortices. Histological findings in the kidney included multifocal lymphoplasmacytic interstitial nephritis with scattered regenerating tubules. The liver contained scattered single-cell hepatocellular necrosis and lymphoplasmacytic portal inflammation. Leptospires were detected in the urine by PCR, supporting a clinical suspicion of **leptospirosis**.

Ten of 1 500 hoggets grazing swedes on a Southland sheep farm were found dead after a short period of being unwell. Rapid post-mortem autolysis was noted. They had been vaccinated against clostridial diseases before going onto the crop. An aqueous humour nitrate test was negative. A heavy growth of *Salmonella* Typhimurium was cultured from the intestinal contents of one of the dead sheep. This bacterium is a rare cause of death in this age group.

Thirty of a mob of 200 hoggets were found recumbent but otherwise bright and alert three days after being shorn on a Central Otago farm. Previously they had spent two months on a paddock of swedes, then been driven (on foot) 5 km to the shearing shed. Two affected hoggets had very high serum creatinine phosphokinase (> 20 000 IU/L; normal range < 150) and very low glutathione peroxidase ( < 2 kU/L; adequate > 5), confirming a diagnosis of white muscle disease caused by selenium deficiency. Contrary to normal practice, these hoggets had not been given selenium before going on to the crop in July.

#### Caprine

Two adult Saanen does from the Auckland region showed central neurological signs and were euthanased. Histopathology of their brains showed variably severe suppurative to necrotising meningoencephalitis, with microabscesses containing Gram-positive bacilli. These changes are consistent with **listeriosis**. As in other ruminants, **listerial meningoencephalitis** can be seen in goats fed silage or baleage, especially if the feed was contaminated with soil or did not reach a low pH during fermentation.

#### Cervine

There were two outbreaks of acute copper toxicity in mobs of nine-monthold deer on Otago deer farms in mid-August. On both farms the deer had been taken off a brassica crop and given a copper injection at the recommended dose. On one farm 20 of 200 deer subsequently died within three days. On the other farm three of 30 deer died within 48 hours. The diagnosis was confirmed after very high kidney copper concentrations were found in dead deer from both farms. Copper toxicity can manifest as a consequence of other stresses (risk factors) occurring at the time of copper supplementation. These risk factors are not well defined but may include yarding, environmental stressors, and administering several treatments concurrently.

Two adult hinds in Central Otago presented with a severe hindlimb ataxia. The signs were present for about a month in one deer and a week in the other. Serum copper was very low (< 3 µmol/L; adequate range 8–22) in both animals, confirming a diagnosis of **enzootic ataxia**, the result of a severe **copper deficiency**.

Six 10-month-old female red deer were found dead over a 10-day period. The deer were part of a mob of 300 on a farm in the Rangitikei District. Post-mortem examination was carried out on one animal. Gross findings included numerous adult lungworms in bronchi throughout the caudal lung lobes. Histology confirmed the presence of numerous adult nematodes in bronchi and bronchioles, and eggs with developing larvae. There was severe eosinophilic interstitial pneumonia. Liver selenium concentration was adequate (3 350 nmol/kg; reference range 850–15 000) but copper was inadequate (60 µmol/kg; reference range 100-2 000). Verminous pneumonia and copper deficiency were diagnosed.

#### Equine

A 10-month-old Thoroughbred horse from Auckland had a history of chronic pneumonia. A trans-tracheal wash showed marked neutrophilic inflammation with intracellular bacteria, and *Streptococcus equi* ssp. *zooepidemicus* was cultured, consistent with opportunistic **streptococcal pneumonia**.

A one-year-old female Arab horse from Taranaki developed crusting skin lesions and raised welts on the distal legs, progressing to involve the trunk. Blood test results showed increased serum amyloid A (251 µg/mL; reference range < 10), increased globulins (51 g/L; reference range 20-39) and increased fibrinogen (6 g/L; reference range 2-4), consistent with inflammatory disease. Skin scrapes were stained with methylene blue and cleared with KOH to assess for Dermatophilus congolensis and dermatophytes, respectively. Both tests were negative, as was dermatophyte culture. Histological examination of skin biopsies revealed neutrophilic crusts containing numerous roundedup and dissociated keratinocytes (acantholytic cells). No pathogens were identified. These findings were considered consistent with a diagnosis of

**pemphigus foliaceus**, the most common autoimmune skin disease of horses.

#### Feline

A four-month-old female desexed Balinese kitten presented with a history of vomiting and weight loss over a period of two months. The kitten was dehydrated and anorexic, and vomiting malodorous intestinal contents. It died in spite of supportive treatment. Gross postmortem changes included reddening of the stomach and small intestinal serosa and mucosa, and congestion of the kidneys. Histological evaluation showed severe regionally extensive necrotising enteritis with marked villus blunting and fusion and attempted regeneration of the few remaining crypts. These findings were considered compatible with a diagnosis of feline panleukopenia.

#### Marine mammal

A swab from the blowhole of a young orca (*Orcinus orca*) yielded a heavy growth of the bacterium *Moraxella* sp. The significance is unclear but Shotts *et al.* (1990), who cultured large numbers of lesional and non-lesional skin tissue and swab samples from bowhead whales (*Balaena mysticetus*), found that 94 percent of their *Moraxella* sp. isolates were associated with lesional skin. It is therefore possible that this isolate may have been clinically significant.

#### Avian

A 30-month-old hen from Northland lost weight and died. At necropsy, abundant peritoneal fluid was found in association with thickened intestines, and nodules within the liver and body wall. Histopathology identified infiltrating neoplastic epithelial cells within the liver, pancreas, intestinal wall and body wall, consistent with a metastatic **adenocarcinoma**, probably of ovarian or perhaps pancreatic origin.

A six-year-old gentoo penguin (*Pygoscelis papua*) in an Auckland zoological collection died suddenly. The previous death of a king penguin (*Aptenodytes patagonicus*) kept in the same exhibit had been linked to a change of diet, and the submitter was concerned about the possibility of salt poisoning. A sample of brain tissue contained 2 150 ppm of sodium. In pigs, sodium toxicosis can be tentatively diagnosed with brain sodium concentrations greater than 1800 ppm, and therefore it seemed that **salt poisoning** was likely in this case.

#### New Zealand Veterinary Pathology

#### Bovine

A two-month-old calf in the Kaipara district had acute scour. Several other calves had similar clinical signs and there had been one death previously. This animal died and was necropsied within a few hours of death. A faecal egg count revealed large numbers of coccidial oocysts. *Yersinia pseudotuberculosis* was isolated from faeces and the mesenteric lymph node. **Coccidiosis** with concurrent **yersiniosis** was diagnosed.

About 15–20 animals out of a mob of 150 eleven-month-old bull calves in the Hauraki region had very loose faeces despite regular drenching. Testing showed the faecal egg counts were not sufficient to suggest parasitism as a cause of clinical disease, but two of six animals tested positive for *Yersinia pseudotuberculosis*. Yersiniosis was diagnosed.

A herd in the South Waikato had several recently calved cows that were skinny and had a reduced appetite, with decreased milk production. Culture of samples from three affected cows revealed that they all had *Salmonella* Bovismorbificans infection. An outbreak of salmonellosis was diagnosed.

Two out of a group of ten 5–6-month-old calves in the Waipa district had chronic ill-thrift and scouring, despite regular anthelminthic treatment. Faecal egg counts confirmed that parasitism was not an issue, but culture of faeces was positive for *Yersinia pseudotuberculosis* in both animals tested. **Enteric yersiniosis** was diagnosed.

At least ten young cows in a 300-cow dairy herd in the Matamata-Piako district developed severe, bloody scours. One animal died. Testing for Johne's disease was negative, but faecal samples from three animals were positive on culture for *Salmonella* Bovismorbificans.

Salmonella Bovismorbificans was also a common isolate from outbreaks of neonatal calf diarrhoea this season, and was identified on numerous properties, especially in the Matamata-Piako district. This agent sometimes occurs in concert with others such as rotavirus and cryptosporidium.

Thirty dairy cows out of a herd of 600 in the Waikato district suffered from

lameness clinically resembling digital dermatitis. One of the cows had a biopsy taken from a lesion on the heel at the coronet. Histologically, the biopsies featured regions of hyperkeratosis and suppurative inflammation with numerous bacteria in a "railroad" formation, consistent with *Dermatophilus congoliensis*. **Dermatophilosis** was diagnosed.

Seven cows out of a 700-cow dairy herd in the Waipa district had late-term abortions and four died, with clinical signs of acute pneumonia following the abortion. Postmortem examination was carried out on one of these cows and samples were taken. Histological examination revealed the presence of necrosuppurative placentitis in addition to a marked necrotising interstitial pneumonia. Special stains demonstrated the presence of branching fungal hyphae associated with the lesions in the placenta and the lung, confirming a systemic fungal infection most likely caused by Mortierella wolfii.

A herd of 30 beef cows in Whakatane experienced three late-term abortions within a week. A calf necropsied by the submitting veterinarian had no significant gross abnormalities. Histology from the aborted fetus revealed scattered aggregates of leukocytes within the blood vessels of the brain. Small amounts of meconium were present within pulmonary alveoli. A pure growth of *Mannhemia haemolytica* was cultured from stomach contents, suggesting that this agent was the cause of a **placentitis leading to abortion.** 

Two heavily pregnant beef cows in the Rotorua district were recumbent and crawling. They had been dosed with an oral magnesium product before sampling, but blood samples still revealed a marked **hypophosphataemia** (0.36 and 0.68 mmol/L; reference range 1.1–2.80), and a marked **hypocalcaemia** (1.18 and 1.57 mmol/L; reference range 2.00–2.60).

A two-year-old beef steer on Coromandel Peninsula had a large visible mass on the left side of its neck. No other clinical abnormalities were present. Biopsy revealed a pyogranulomatous cellulitis with intralesional club colonies, consistent with actinobacillosis. Gram staining of the histologic sections showed that the bacteria involved were Gram negative, favouring an aetiologic diagnosis of *Actinobacillus lignieresii*, the causative agent of **woody tongue**. While frequently causing infections in the mouth, this agent may also be involved in other soft tissue infections like this one.

A mob of two-year-old heifers on a beef property in Taupo experienced increased mortalities amongst calves, with five out of 110 newborn calves dying within the first four days of life. Samples from one of these calves were submitted for histology. Abnormal histology findings were few, but there was evidence of meconium aspiration and mild pneumonia. PCR testing of the spleen revealed the presence of bovine viral diarrhoea virus, suggesting that the increased mortalities in this group could have been due to **persistent bovine viral diarrhoea infection**.

A property in south Waikato diagnosed *Salmonella* Bovismorbificans as a cause of enteritis and death in their calves earlier in the season. More recently, two cows were seen sick, with one dying suddenly. Four calves were born dead at full term on the same day when the sick cows were observed. A faecal sample was submitted and *S*. Bovismorbificans was isolated. It was postulated that *Salmonella* had spread from the calves to the pregnant and calving cows, resulting in an **outbreak of abortions**.

A herd of cows in Ashburton exhibited abortions, accompanied by diarrhoea in the dams and some deaths. A complete set of tissues including placenta from two aborted dams and one aborted fetus was submitted. Histology revealed a marked necrosuppurative placentitis on both placentas. Culture from both, and enrichment culture from the fetal tissues, revealed the presence of *Salmonella Brandenburg*, which was also isolated from the faeces of three of the four dams. Enteric salmonellosis with septicaemia and subsequent abortions was diagnosed.

Routine milk cultures were submitted from three cows with mastitis on a dairy property at Waitomo, in the King Country district. A heavy pure growth of *Bergeyella zoohelcum* was isolated from one of the cows, suggesting that this organism was the cause of the **mastitis** in that particular animal. *B. zoohelcum* is considered a normal commensal found in the oral cavity of cats and dogs. It has been identified in association with a number of infections in humans, including cellulitis, tenosynovitis, septicaemia, pneumonia and meningitis. Infection in humans is typically the result of an animal bite. A dog bite is one possible underlying cause of the mastitis noted in this cow.

Two Hereford beef calves in the Waikato District exhibited nervous signs, with anorexia, ataxia, salivation and bruxism. One animal appeared blind and developed convulsions before dying. The blood lead level in the surviving animal was > 0.70 mg/L (toxic level > 0.35), consistent with a diagnosis of **lead poisoning** (Meldrum & Ko, 2003).

#### Ovine

Ten sheep up to two years of age died over the winter on a property in the Whangarei District. The animals initially had ataxia with circling and recumbency and then died. Brains from two affected animals were submitted for histopathology, which revealed a subacute suppurative and lymphoplasmacytic meningoencephalitis, typical of **listeriosis**.

Three animals out of a 200-ewe stud flock in south Waikato aborted in late gestation. The ewes were run in two different mobs. Aborted and weak-born lambs and placentas were sampled from two ewes. The flock had a long vaccination history for both campylobacter and toxoplasmosis. Histologic examination revealed multifocal cotyledonary necrosis with low numbers of Toxoplasma cysts containing multiple tachyzoites that were visible histologically. Toxoplasmosis was diagnosed. This infection does occur occasionally in vaccinated flocks, as the live vaccine is sensitive to temperature fluctuations during storage and transport, which can decrease its effectiveness.

Three young lambs from the Manawatu district developed severe swellings over the top and bottom lips over a few days, with crusting. The worst-affected lamb was euthanased and histology was performed on the lesions. There was a marked ballooning hyperplasia of the epidermis or mucosal epithelium. Single-cell necrosis and apoptosis was present at all levels of the epithelium, which also had marked cytoplasmic swelling and clearing of keratinocytes with margination of nuclear chromatin. Histology was considered most

### consistent with **contagious pustular dermatitis (orf)**.

A flock of hoggets at a property in Marlborough experienced late-term abortions, with one or two sheep per day losing their lambs. The flock was vaccinated for toxoplasma and campylobacter. Histology revealed random multifocal hepatic necrosis with suppurative inflammation surrounding colonies of Gram positive short bacilli. In the lung, there was evidence of a suppurative bronchiolitis, again with similar colonies of Gram positive short bacilli. *Listeria ivanovii* was isolated from stomach contents.

Samples from aborted twin lambs were submitted from the Taupo district. Histology revealed multifocal necrosis with suppurative inflammation and numerous colonies of coccobacilli. In both feti there was also an acute suppurate bronchopneumonia, consistent with placentitis. Culture of fetal stomach contents yielded *Listeria ivanovii*, a likely cause of **abortion** in this case.

A property in south Waikato had an abortion storm in 2015, with 30 percent of hoggets aborting in the third trimester (out of a mob of 600). The animals were well vaccinated for campylobacter and toxoplasmosis and no diagnosis was established at that time. A leptospirosis outbreak had occurred at the same time in cows on the property. This year, samples from two recently aborted hoggets were submitted for leptospirosis serology. One had a *Leptospira* Pomona MAT titre of 1:800, suggesting recent exposure to this serovar.

Samples of aborted lamb feti from the Waitaki district were submitted to investigate the cause of abortions. There was moderate autolysis, but histology revealed a fetal pneumonia and placentitis. Culture of stomach contents from one of the lambs yielded a heavy growth of **Yersinia pseudotuberculosis**, the likely cause of abortion in this case.

#### Caprine

Three animals in a mob of 40 mixed-age goats in the Tasman district had severe diarrhoea for several months and two died. Tissues from one were submitted for histopathology, which revealed an **acute necrotising enteritis** consistent with bacterial enteritis caused by *Salmonella* or *Yersinia*.

Eight out of a mob of 280 dairy goats in the Matamata-Piako district had clinical evidence of pneumonia and diarrhoea. Three animals were necropsied. Faeces were negative for *Yersinia* spp. on culture, but *Mannheimia haemolytica* was isolated from the lungs of all three. **Pneumonic mannheimiosis** was diagnosed.

A 170-goat dairy herd in the Matamata-Piako district had three young animals with skin lesions. One animal had more generalised exudation and crusting present over much of the body, and two others had similar lesions affecting the nose and ears. Cytology was performed and revealed no evidence of *Dermatophilus* infection. Histology from one animal revealed a marked proliferative and hyperplastic dermatitis with ballooning of keratinocytes and neutrophilic crusting, typical of **contagious pustular dermatitis (orf)** infection.

Increased mortality was noted among a group of 15-day-old goat kids at a research facility in the Waikato district. Tissues from four animals were submitted for testing. There was a marked fibrinosuppurative bronchopneumonia in all animals examined histologically. Some also had areas of type II pneumocyte hyperplasia, suggesting the possibility of concomitant viral infection. One goat had evidence of an acute suppurative synovitis. A mixed culture was isolated from the lungs of three of the goats, with Mannheimia haemolytica prominent on all of the plates. Acute suppurative bronchopneumonia caused by Mannheimia haemolytica was diagnosed, accompanied by septicaemia/ bacteraemia in one kid.

#### Equine

A three-year-old Standardbred filly had severe chronic dermatitis with numerous scaly crusts, matted hairs and pruritis. Lesions started under the halter and harness, then spread to involve much of the hair coat. Biochemistry revealed a moderate hypergammaglobulinemia, likely reflecting the chronic inflammation. Fungal culture of affected hair coat yielded a heavy growth of *Trichophyton equinum*. **Dermatophytosis** was diagnosed.

A Thoroughbred horse in the Matamata-Piako district aborted at late term. Histologic examination of the fetus revealed a multifocal necrotising hepatitis with intranuclear inclusion bodies visible in hepatocytes at the margins of the necrotic foci. Within the lung there was a necrotising bronchiolitis with numerous intranuclear inclusion bodies visible within damaged bronchiolar epithelium. **Abortion caused by equine herpesvirus-1** was diagnosed.

A 24-year-old horse had a two-week history of anterior uveitis affecting the right eye. The cornea was grossly ulcerated and the anterior chamber was filled with pus. Culture of a swab from the lesion revealed the presence of *Aspergillus fumigatus*. Mycotic keratitis was diagnosed.

An 18-year-old gelding from Hamilton City had a history of a markedly increased respiratory rate accompanied by weight loss. The horse was euthanased. At gross postmortem there was generalised consolidation of the lungs, which failed to collapse and did not float in formalin, and there was enlargement of the pulmonary lymph nodes. Histology revealed a severe generalised chronic pulmonary fibrosis with oedema and in some areas there was suppurative inflammation. Rare macrophages had nuclear changes suggestive of intranuclear inclusion bodies. Equine multinodular pulmonary fibrosis was the major differential diagnosis based on histopathology, and PCR for equine herpesvirus-5 was positive, confirming infection.

#### Porcine

Tissues from a piglet about four weeks from the Franklin district were submitted for histopathology. Little history was available, but histology revealed moderate to marked lymphoid depletion in the spleen and lymph node, with numerous large intracytoplasmic botryoid inclusions visible within sinusoidal macrophages of the lymph node. **Postweaning multisystemic wasting syndrome** associated with **porcine circovirus-2** was diagnosed. This piglet was younger than the usual age (five to 12 weeks) at which pigs are usually affected by this disease.

#### Cervine

A three-year-old red deer stag in Marlborough died suddenly and tissues were submitted for histology. Examination of the brain revealed a marked nonsuppurative meningoencephalitis with a vasculitis, consistent with **malignant catarrhal fever (MCF).** MCF in New Zealand is commonly associated with infection of non-host ruminants by ovine herpesvirus-2 or caprine herpesvirus-2.

#### Camelid

A one-year-old female alpaca in the Rodney district had ill-thrift with a low rate of weight gain despite being dewormed frequently. Biochemistry and haematology were within normal limits but serum copper was less than  $2 \mu mol/L$  (the lower limit of detection for the assay used). Normal serum copper levels in this species are 5.0–16.0 µmol/L (Ellison 2006). **Severe copper deficiency** was diagnosed.

An alpaca in the Hauraki district was recumbent and in shock with severe oral ulceration when attended by a veterinarian. The animal died and histopathology was performed on samples taken at necropsy. There was diffuse severe hepatic fibrosis with multifocal suppurative inflammation and an ulcerative stomatitis. The hepatic fibrosis suggested a previous hepatic insult. Sporodesmin toxicity was unlikely owing to the diffuse (not periportal) distribution of the fibrosis. Alternative causes of hepatopathy, including toxic plant ingestion, were considered. The accompanying suppurative inflammation suggested the presence of a bacteremia, possibly a result of the ulcerative stomatitis. However, the initiating cause of the ulcerative stomatitis was not determined.

#### Canine

An 18-month-old dog from the Auckland region had persistent, intractable diarrhoea. Faeces was submitted for a full faecal panel, including culture for Salmonella, Campylobacter and Yersinia and testing for Giardia, Cryptosporidia (by antigen ELISA) and a faecal egg count. The Giardia antigen ELISA test was positive, and culture was also positive for Yersinia enterocolitica. As *Y. enterocolitica* is not an uncommon isolate from clinically normal dogs, it is difficult to assess its role in this dog's enteritis. However, Y. enterocolitica is an important human pathogen, and the presence of severe diarrhoea (which would increase faecal environmental contamination) in this patient raises the concern for potential zoonotic risk.

A litter of five-week-puppies in the Thames–Coromandel district had severe mucoid diarrhoea. The bitch was also affected. Faecal egg count revealed 1 200 ascarid eggs per gram in the faeces of an affected pup. Severe **roundworm infection** was diagnosed.

An 11-week-old puppy in Whakatane presented with lethargy, weight loss, reduced appetite and polydipsia/polyuria. The urine specific gravity was 1.009. The pup was azotaemic, had moderate icterus, and had an alkaline phosphatase level of 524 IU/L (reference range 0–185). Based on the evidence of renal failure and hepatic compromise, testing for leptospirosis was recommended. The *Leptospira* Copenhageni MAT titre was 1:1600, consistent with recent infection by this organism. Leptospirosis was diagnosed.

#### Lagomorph

A group of pet rabbits in an outside hutch in Tauranga had increased mortalities, with three rabbits dying suddenly over a short period. Liver histology revealed widespread degeneration and necrosis of hepatocytes, consistent with **calicivirus** infection. **Rabbit haemorrhagic disease** was diagnosed.

#### Reptile

A bearded dragon (Pogona sp.) from a captive facility in the Rotorua district died suddenly and was submitted for necropsy. Histology revealed a diffuse severe hepatic lipidosis accompanied by a multifocal granulomatous hepatitis. Large aggregates of heterophilic inflammation were also present in the colonic mucosa, often replacing normal mucosal glands. PAS stains of the intestine demonstrated the presence of numerous large broad-based branching fungal hyphae within the areas of intestinal and hepatic inflammation. Systemic fungal infection, likely due to a zygomycete, was diagnosed.

#### Avian

A zebra finch (*Taeniopygia guttata*) in a captive facility in Auckland was found dead. Necropsy and histology revealed a marked granulomatous enteritis, with sheets of macrophages present within the mucosa. Scattered infiltrates of macrophages and granulomas were also present in the liver, crop and lung. Acid-fast staining revealed that these granulomas and macrophages contained large aggregates of acid-fast

bacilli. Disseminated mycobacteriosis was diagnosed.

Marked ill-thrift was noted in young hens (less than 18 months old) in a flock of 40 Silky domestic chickens in the Thames-Coromandel district. Seven animals were affected and five had died. Histologic examination of two hens revealed marked infiltrates of neoplastic round cells in a number of organs, including the kidney, liver, spleen, pancreas, gastrointestinal tract and oviduct. One bird also had evidence of a necrotising typhlitis and hepatitis with intralesional bacterial colonies visible histologically. The other also had evidence of a multifocal suppurative hepatitis. Lymphoid leucosis was diagnosed. This disease also frequently causes immunosuppression, predisposing these birds to bacterial typhlitis and septicaemia.

A flock of 1 500 brown laying hens aged 44 weeks in the Palmerston North district suffered an increase in losses over a one-week period, with an associated loss of production. Necropsy of three birds by the submitting veterinarian revealed evidence of coalescing regions of splenic necrosis, multifocal to coalescing hepatic necrosis, and bacterial endocarditis. Culture of the spleen from two birds revealed the presence of **Pasteurella multocida**.

#### Zoo animal

A group of kiore or Pacific rats (Rattus exulans) collected from an offshore island nature reserve had white patches visible on the surface of their livers. Histology revealed a chronic granulomatous cholangiohepatitis, centred around numerous ovoid bioperculate thickwalled eggs consistent with Capillaria spp. Capillaria hepatica infection was diagnosed. C. hepatica is a parasite with worldwide distribution and is the only nematode to inhabit the liver at the adult stage of its life cycle. Eggs are deposited in the hepatic parenchyma and only released after the host dies. Embryonation of the eggs requires exposure to oxygen and moisture. Eggs become infective if the liver is consumed by a predator or scavenger and are then deposited in the faeces, allowing embryonation to occur (Mowat et al., 2009). Animals infected by adult nematodes do not have evidence of Capillaria eggs in the faeces, making premortem diagnosis challenging.

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# Quarterly report of investigations of suspected exotic diseases

### Exotic vesicular diseases ruled out

A veterinarian notified MPI via the exotic pest and disease hotline about mouth ulcers in a calf, which had been reported by a farmer. The five-week-old calf, one of 140 on a dairy run-off in the Waikato, had a large ulcer in the rostroventral mucosa, surrounding the incisors. A second calf was noted one day later with milder mouth lesions. The AsureQuality Initial Investigating Veterinarian (IIV) system was activated because of the clinical presentation and the fact that no veterinarian had evaluated the case. Exotic vesicular disease such as foot -and-mouth disease, and many endemic infectious and non-infectious diseases. can cause mouth lesions in ruminants. The IIV examined the mob and was able to rule out exotic vesicular disease based on risk factors, epidemiology and lack of classic clinical lesions. Minor reddening or small ulcers were found in 17 calves (about 23 percent) of 73 examined. There was no disease in the adjacent dairy herd. Lesions in calves were mostly on the floor of the mouth near the incisors. The most likely cause was considered to be some factor associated with the calf feeder, such as a sharp edge, followed by secondary bacterial infection, e.g., by Fusobacterium necrophorum (the cause of calf diphtheria). No infectious disease such as bovine viral diarrhoea or bovine papular stomatitis was present, and the investigation was stood down. Presentation was different in several respects from the idiopathic disease known as calf ulcerative stomatitis (O'Connell 2016; Anon 2016), which has been recognised sporadically since 2015 in weaned calves in New Zealand but was ruled out based on lesion localisation and the lack of other clinical signs such as diarrhoea and drooling. Exotic disease was excluded and the investigation stood down.

In a similar case, a veterinarian notified MPI via the exotic pest and disease hotline of stomatitis seen in a calf during routine disbudding. The three-monthold calf, one of about 130 on a dairy run-off in South Waikato, had a large Exotic disease investigations are managed and reported by MPI Investigation and Diagnostic Centre (IDC) and Response, Wallaceville. The following is a summary of investigations of suspected exotic disease during the period from July to September 2016.

ulcer with necrotic edges affecting the majority of the rostral third of hard palate, and a smaller proliferative area of granulation tissue on the ventral tongue. The calf was otherwise well and drinking. Exotic vesicular disease such as footand-mouth disease, and many endemic infectious and non-infectious diseases, can cause mouth lesions in ruminants. Of particular interest in this age group is calf ulcerative stomatitis. Under the direction of the MPI Incursion Investigator, the veterinarian examined the remainder of the mob of 39 bull calves, and a mob of about 90 heifer calves. No lesions were identified in any other calves. Photographs were taken and sent to the Incursion Investigator. Given the extent and depth of the hard-palate lesion, the most likely cause was considered to be trauma followed by secondary infection. Presentation in this case differed in several respects from CUS (O'Connell 2016; Anon 2016), which was ruled out based on the nature of the lesion, the fact that only a single animal was affected, and the lack of other clinical signs such as diarrhoea and drooling. The calf responded to routine antibiotic therapy and no other cases were reported. Exotic disease was excluded and the investigation stood down.

A regional laboratory pathologist called the MPI exotic pest and disease hotline to report mouth ulcers in an 18-monthold dairy heifer that had returned a negative BVD antigen ELISA test. Under the direction of an MPI Incursion Investigator, the farm's veterinarian revisited the property to re-examine the affected heifer and inspect the group of 110 in-contact heifers. The affected animal had lost weight and had a watery scour but no fever. Ulcerative lesions were limited to the mouth, presenting as linear areas of superficial ulceration

along the lateral edges of the tongue, and two small (about 5 mm) round ulcerated areas below the incisors or in front of the ventral molars. No lesions were identified on the feet, udder or perineum, and no lesions were identified in the remainder of the mob. Exotic vesicular disease was excluded on clinical and epidemiological grounds. Serum biochemistry indicated mild hepatic changes but was otherwise unremarkable. Intestinal parasitism or Johne's disease appeared unlikely as the cause of the scour, given a negative faecal egg count, normal serum pepsinogen and a negative *Mycobacterium avium* ssp. paratuberculosis ELISA test. Molecular testing for bovine viral diarrhoea virus gave negative results, and a molecular assay for ovine herpesvirus-2 (the cause of malignant catarrhal fever) was also negative. Although the cause of the condition was not determined, the heifer recovered over the following two weeks after receiving broad-spectrum anthelminthic and antibiotic therapy. Exotic disease was excluded and the investigation was stood down.

A veterinarian called the MPI exotic pest and disease hotline to report teat lesions followed by severe mastitis seen over a period of 7-10 days in four of about 50 recently calved heifers. Teat lesions can be associated with exotic vesicular or poxviral diseases. The farm veterinarian sent pictures of the lesions to an MPI Incursion Investigator. The appearance of the lesions and the absence of any other clinical signs in the herd enabled exotic disease to be excluded on clinical and epidemiological grounds. A diagnosis was pursued because the lesions were so severe, including severe swelling, haemorrhage and necrosis of the udder, and teat degeneration in the chronic cases with the loss of one or more entire teats. The herd was inspected and a further

early case was identified with mild areas of teat wall and teat canal epithelial necrosis forming a dry scab. The early lesions were considered potentially consistent with traumatic injury (e.g., by the milking machine) or photosensitivity. Serum samples from two heifers with lesions did not have significantly elevated gamma-glutamyl transferase, excluding liver damage associated with secondary photosensitisation. Staphylococcus aureus and Streptococcus dysgalactiae were cultured from both milk and a dryscab tissue sample from the early case, and from affected teat tissue in a heifer with chronic lesions. The presentation was considered to be a gangrenous ("black") mastitis associated with a toxinproducing strain of Staphylococcus aureus following an initial insult to the teat and/or teat canal, possibly resulting from milking-machine damage. No further cases occurred and the investigation was stood down.

#### Anthrax excluded

A veterinarian contacted MPI via the exotic pest and disease hotline to request exclusion of anthrax after six calves in a mob of 50 died acutely and were found with bloody discharge from the nostrils and anus. Whole-blood EDTA samples were collected from the peripheral blood vessels of two affected animals and blood smears were made. No organisms consistent with Bacillus anthracis were seen after staining with polychrome methylene blue at the MPI Animal Health Laboratory (AHL). The blood samples were negative for B. anthracis on culture. No historic cases of anthrax affecting calves had occurred in the Gisborne region for more than 50 years. No further acute deaths subsequently occurred on the property. Anthrax was excluded and the investigation stood down.

A veterinarian called the MPI exotic pest and disease hotline to request exclusion of anthrax after two adult cows from a dairy herd of about 150 died acutely and were found with bloody discharge from various orifices. The herd was in Canterbury, a region that historically has not experienced anthrax. Whole-blood EDTA and serum samples were collected from the peripheral blood vessels and smears were made of the bloody discharge, in both cows. No organisms consistent with *Bacillus anthracis* were seen after staining with polychrome methylene blue at the MPI AHL. The blood samples were negative for *B. anthracis* on culture, and molecular assays excluded type 2 bovine virus diarrhoea virus. Bacterial culture of the blood for four days under aerobic, anaerobic and in a 5 percent  $CO_2$  atmosphere was negative. No further deaths occurred on the property. Anthrax was excluded as a cause of the clinical signs observed and the investigation was stood down.

# Ulcerative posthitis in rams investigated

A veterinarian reported via the MPI exotic pest and disease hotline multiple rams with severe inflammation and ulceration of the penis. The affected rams were two-tooth Perendales that had been introduced into a commercial breeding flock. Six of the 10 rams present in the two-tooth mob were affected clinically. None of the other 20 mixed age rams present in the flock were affected. Blood samples were taken from the affected rams. There were no significant changes in the serum biochemistry or haematology. Several of the rams were euthanased because of the severity of the lesions. Samples collected during necropsy showed severe suppurative and necrotising balanitis with evidence of mixed bacteria and possible viral inclusions.

Swabs were collected from the penises of the 10 rams in the affected cohort and four were positive by PCR for mycoplasma. DNA sequencing confirmed the species was *Mycoplasma bovigenitalium*. While the inclusion bodies seen in cells on histology could have indicated parapox virus infection, all swabs tested negative by PCR for this agent. A number of bacterial species were grown from swabs but the predominant species was *Escherichia coli*.

No single causative agent was identified from laboratory testing. It is likely that the cause of the lesions was multifactorial, including a number of environmental factors. While *M. bovigenitalium* was identified in a previous investigation of balanitis in Perendale rams (Crosbie *et al.*, 2008), as in this case the significance remains unknown.

#### Scrapie ruled out

A veterinary pathologist called the MPI exotic pest and disease

hotline to report a case of suspected Nor98/atypical scrapie in an older ewe. The ewe had shown central nervous signs including vacancy, slight ataxia and nystagmus, so her brain was submitted for testing under the MPI Transmissible Spongiform Encephalopathy Surveillance Programme. By histology, the cerebellum contained intraneuron vacuolation consistent with Nor98/atypical scrapie, a sporadic disease that is distinct from the reportable form. Because Nor98/atypical scrapie is not reported frequently, this case was followed up to rule out the classical form of scrapie. A scrapie ELISA test on brainstem was negative, and follow-up testing confirmed Nor98/atypical scrapie. The investigation was stood down.

#### **Brucellosis ruled out**

A veterinarian called the MPI exotic pest and disease hotline to report a suspect case of fistulous withers in a one-year-old pony filly. A swollen, draining wound on the withers had been treated with antibiotics but had not resolved. Fistulous withers can be caused by the exotic agent Brucella abortus and, less frequently, by B. suis; both these agents also cause a suppurative bursitis of the supraspinus bursa. Brucella infections in horses are also associated with poll evil, which is inflammation of the supra-atlantal bursa. Serum and an abscess swab were collected and submitted to the Animal Health Laboratory (Wallaceville). Exotic disease was ruled out after the serum tested negative by competitive ELISA for antibodies to the smooth brucellae (including *B. abortus* and *B. suis*). Bacterial culture identified a range of Gram-positive and Gram-negative isolates including E. coli, Bacteroides uniformis, Enterococcus spp. and others. Possible causes of this type of lesion include bacterial infection secondary to a penetrating wound or foreign body in the subcutaneous tissues of the withers. Exotic disease was excluded and the investigation was stood down.

#### Equine influenza ruled out

A pathologist called the MPI exotic pest and disease hotline to discuss an outbreak of coughing in a group of performance horses. Over a five-week period, 11 of 22 horses ranging from 1–14 years old had presented with coughing and nasal discharge. While none of the horses were imported,

they could have come in contact with imported horses at equestrian events. Equine influenza (EI) and equine viral arteritis (EVA) are exotic differentials for coughing and nasal discharge in horses. Blood samples and nasal swabs were submitted by the primary veterinarian to theAnimal Health Laboratory (Wallaceville), where EI was ruled out by hemagglutination inhibition and PCR tests, and EVA by VNT. The endemic agents Streptococcus equi equi (strangles), equine adenovirus and equine rhinitis virus (types A and B) were ruled out by culture or serology. However, serological tests for the endemic equine herpesviruses (EHV-1 and EHV-4) suggested that one of these agents had played a role in this respiratory disease outbreak. The horses had not been vaccinated for EHV.

#### **EVA** excluded

An exported breeding stallion was reported to MPI via the exotic pest and disease hotline after the MPI Animal Exports team was notified of a positive equine viral arteritis (EVA) antibody test on arrival in the UK. The horse had been exported two months previously and EVA is present in the UK but not in New Zealand. The horse was not showing any clinical signs consistent with the disease. Historical antibody titre test results held at the AHL (Wallaceville) were consistent with the horse having been seronegative before it was vaccinated in 2010, and there was evidence of a positive post-vaccination titre. Virus isolation for EAV had been conducted on paired semen samples before export and these were negative. In light of these results, the positive antibody titre found in the UK was considered to be due to vaccination.

#### **EVA and EIA excluded**

An 18-month-old Standardbred horse with anaemia, oedema and significant inflammatory signs on bloodwork was reported to the MPI exotic pest and disease hotline by a veterinary pathologist. The horse had first become ill a month previously with gastrointestinal disease. Over the following week the clinical signs relating to colitis resolved but on complete blood count testing the anaemia had progressed, ongoing systemic inflammation was evident, and ventral and limb oedema had developed. Repeated blood testing two weeks later

showed the same results and MPI was notified. The horse's history did not include travel or other risk exposures for exotic disease. Serum and EDTA blood samples from the three sampling events were submitted to the AHL (Wallaceville) for exclusion of exotic disease consistent with this presentation. Equine viral arteritis, equine infectious anaemia, and infection with Anaplasma phagocytophilum or piroplasms (Theileria and Babesia spp.) were excluded after negative results by VNT, AGID and PCR respectively. The horse made a slow recovery, with indicators of systemic inflammation gradually diminishing.

# Equine chlamydial abortion excluded

A veterinary pathologist called the MPI exotic pest and disease hotline to report endometritis in a seven-yearold Thoroughbred mare imported a few months previously from the Hunter Valley, Australia. The mare had recently given birth to a small foal with contracted tendons and thickened umbilicus, and the placenta had appeared inflamed. The foal died a few days later and was submitted for necropsy and histopathology. Given that a number of Chlamydia-associated abortions had been reported in the Hunter Valley, the AHL carried out tests to exclude Chlamydia abortus and C. psittaci, which although present in New Zealand have not been associated with equine abortion here. Molecular testing at the AHL of lung tissue from the foal and two uterine swabs taken three days apart from the mare excluded Chlamydia spp. Routine bacterial culture of the uterine swab identified a heavy growth of Streptococcus equi ssp. zooepidemicus (commonly associated with endometritis in mares) and a mild growth of Escherichia coli. The endometritis resolved uneventfully after routine antibacterial therapy. Emerging disease was excluded and the investigation stood down.

### *Echinococcus granulosus* ruled out

A hunter from Northland called the MPI exotic pest and disease hotline to report possible lesions consistent with hydatids seen in the liver of a feral pig. Hydatids, the larval stage of the tapeworm *Echinococcus granulosus*, are unwanted and notifiable. The liver, which had been frozen, was sent to the AHL, where samples were taken for histopathology and bacterial culture. During sampling it was noted that the liver contained about twenty solid to cystic pale nodules 1-3 cm long scattered throughout the parenchyma. Another exotic cause of cysts can be the tapeworm *Cysticercus cellulosae/Taenia solium.* Endemic causes of the lesions include parasitism from migrating roundworms (e.g., Ascaris suis) and tuberculosis. Bacteriology revealed moderate mixed bacterial growth, which was considered insignificant. Histopathology showed that lesions consisted of a predominantly eosinophilic infiltrate, which contained occasional parasite sections consistent with larval trematodes (flukes). As part of the fluke lifecyle (e.g. Fasciola hepatica, a common endemic species), migration of young flukes takes place through the liver parenchyma before reaching the biliary tract, where they mature. It was considered most likely that this was a normal parasite migration through the liver of this pig. The investigation was stood down.

A veterinary clinic called the MPI exotic pest and disease hotline to report the presence of cystic masses resembling parasites in the liver of a dog recently imported from Australia. The seven-yearold dog was originally from New Zealand but had lived in urban Melbourne for the past year before being re-imported to New Zealand. Prior to re-importation a neck mass had been identified, which fine-needle aspiration found to be highly inflamed, with strange mesenchymal cells thought to be most consistent with reactive fibroblasts. The dog tested free of exotic disease and was imported to New Zealand, where it was seen for the neck mass by a specialist. A CT scan revealed about 10-20 hepatic cystic masses containing apparent "bodies". Cystic parasites in dogs are rare, since canids (and dogs in particular) are the definitive host of Echinococcus granulosus, the causative agent of hydatidosis in sheep and humans. However, there are rare case reports of E. multilocularis lesions and Taenia spp. cysts in dogs, and there are anecdotal reports of immunosuppressed dogs developing hydatids disease. Therefore, owing to the strange presentation and recent importation this case was investigated as a possible exotic parasitic disease.

The neck mass, liver lesions and

hepatic lymph nodes were aspirated and cytology indicated that all three contained similar poorly differentiated erythrophagocytic mesenchymal cells with high nuclear and cellular variability. This was consistent with a mesenchymal cell tumour, and most consistent with histiocytic neoplastic disease. Although other samples were collected, testing was deemed unnecessary following diagnosis of the neoplastic disease. It was considered that metastases of the tumour rather than exotic parasites were the cause of the cystic liver lesions, and the investigation was closed.

### Canine *Leishmania* confirmed

Incursion Investigators were contacted by an AHL scientist following a positive Leishmania antibody test in a five-yearold government working dog bound for export to Australia. L. infantum is a trypanosomal disease transmitted primarily by phlebotamine sandflies, and is common in many parts of the world, particularly southern Europe. It is considered exotic to New Zealand, although imported dogs are occasionally found carrying the parasite, which can remain dormant for years. Infection with L. infantum is not considered curable, though active infection can be treated. The New Zealand import health standard for dogs does not require testing for Leishmania, owing to poor test sensitivity and the absence of phlebotamine sandflies here. An investigation was undertaken to determine whether infection had been acquired within New Zealand. The dog's history revealed that it had been bred in the Netherlands before being imported in 2014. Given its foreign origin and the lack of testing prior to importation, it was considered most likely that the infection originated outside of New Zealand, and the investigation was closed. Owing to Australian import requirements, the dog will not be able to take its anticipated trip.

#### Canine Leishmania excluded

A veterinary pathologist called the MPI exotic pest and disease hotline to report a suspected exotic disease in a dog imported three weeks prior from Perth, Australia. The presentation was similar to leishmaniasis, although this disease is not thought to be endemic in Australia. The dog initially presented with bilateral uveitis and generalised muscle wastage,

reduced appetite and vomiting. It had a history of weight loss (8 kg in 2-3 weeks). A large splenic mass was identified on ultrasound examination. Despite treatment, the dog continued to deteriorate and presented one week later with ataxia, tachycardia, bilateral blindness and marked depression, and was subsequently euthanased. Histopathology showed multi-organ inflammation with a predominant histiocytic component. There was a prominent meningoencephalitis, splenitis, enteritis, pancreatitis, myocarditis and nephritis. Some organs showed evidence of a vasculitis. No organisms were seen after special staining, but the highly inflammatory pattern of disease was possibly consistent with infectious disease or, more likely, an unusual histiocytic neoplasm. Given these findings, affected tissues were sent for subcontracted immunohistochemistry. A battery of histiocytic and lymphocytic immunolabelling tests indicated that the invading cells were in fact a neoplastic proliferation of histiocytes most closely resembling the disease systemic histiocytosis. Based on the confirmation of neoplastic disease as the underlying cause of the multisystemic organ infiltration, leishmaniasis was excluded and the investigation was stood down.

#### Canine heartworm excluded

A veterinarian contacted MPI to report a coughing five-year-old Blue Heeler dog recently released from quarantine after importation from South Africa. The veterinarian believed the dog had infectious tracheobronchitis (kennel cough) but was concerned that canine heartworm (Dirofilaira immitis) was a potential aetiology. Whole blood and serum samples were submitted to MPI's Animal Health Laboratory. Testing was negative for canine heartworm antigen and microfilariae by ELISA and Knotts concentration test, respectively. The dog responded well to treatment. Exotic disease was excluded and the investigation stood down.

#### Brucella canis excluded

A veterinarian reported via the MPI exotic pest and disease hotline a fiveyear-old working dog with unilateral orchitis. The New Zealand-bred dog had not travelled overseas or been used for mating. A blood sample was collected from the dog and submitted to the AHL (Wallaceville). *Brucella canis* was excluded after serum tested negative using the *B. canis* card agglutination test.

A veterinarian rang the MPI exotic pest and disease hotline after diagnosing unilateral epididymitis in a rescued adult male Siberian Husky. A bacterial swab of the lesion was taken, and both testes and attached epididymis were put into formalin and sent to the AHL. Brucella canis is a common cause of epididymitis in dogs worldwide, but is exotic to New Zealand. Serology was negative for anti-Brucella antibodies on the B. canis card test. Bacterial culture of the lesion yielded no bacteria. Histopathology showed an apparently sterile, spermatic granuloma at the head of the epididymis. Possible causes of the spermatic granuloma in this dog include an idiopathic condition, a resolved bacterial infection and an uncommon condition, known as spermatic granuloma of the epididymal head. Brucellosis was ruled out in this case and the investigation was closed.

A pathologist called the MPI exotic pest and disease hotline to report a dog showing signs of epididymitis where canine brucellosis was a differential diagnosis. The dog was an eight-year-old Huntaway born and bred in New Zealand. After the epididymis was surgically removed, the dog was determined to be negative to *Brucella canis*. Histology revealed a suppurative exudate in the epididymal duct, meaning the lesions were likely the result of an ascending bacterial infection.

A veterinarian called the MPI exotic pest and disease hotline after neutering a dog and identifying a unilateral testicular abscess. The dog was a three-year-old New Zealand-bred Huntaway and had not been used for breeding. Serum and a bacterial swab were submitted to the AHL (Wallaceville), where *Brucella canis* was excluded after a negative *B. canis* rapid slide agglutination test. Exotic disease was excluded and no further action was required.

#### Ehrlichia canis confirmed

A scientist from the AHL(Wallaceville) notified an Incursion Investigator of a dog with a positive pre-export immunofluorescent antibody test result for *Ehrlichia canis* (>=1:640). The dog was a 3½-year-old Staffordshire Bull Terrier that had been imported into New Zealand, having lived in a number of Asian countries including Vietnam and Taiwan (where E. canis is known to be present). Thus it was considered that the dog had been imported with subclinical infection. E. canis testing is not a requirement of our import health standard for dogs because its vector, the brown dog tick Rhipicephalus sanguineus, is not present in New Zealand. The endemic tick Haemaphysalis longicornis may feed on dogs, but is not a vector of Ehrlichia spp. A positive PCR (carried out on blood) confirmed that the dog was infected with *E. canis* and it was subsequently treated with doxycycline 10 mg/kg for six weeks.

# *Pythium insidiosum* excluded

A veterinarian called the MPI exotic pest and disease hotline to report a possible case of pythiosis in a New Zealandborn 13-year-old female Cairn Terrier. This disease is caused by the funguslike organism Pythium insidiosum and has been reported mostly in dogs, horses and humans - mainly in tropical and subtropical areas. Infection is acquired through ingestion or contact with wetlands, swamps or pond water containing the organism, and in dogs can manifest as a gastroenteritis or dermatitis (Gaastra et al. 2010). Pythiosis does not appear to be contagious or zoonotic. While a human case of pythiosis has been described in New Zealand (Murdoch & Parr 1997), it has not been described here in animals.

The dog had presented with retching, inappetance and cranial abdominal pain of three weeks' duration. A CT scan had identified a pattern of infiltration in the distal oesophagus, lower oesophageal sphincter and gastric wall. This was deemed most compatible with neoplasia such as leiomyosarcoma, adenocarcinoma or lymphoma but could also be consistent with pythiosis. The dog's condition deteriorated and it was euthanased before further diagnostics could be carried out. It was submitted to a regional commercial pathology laboratory for necropsy. Leiomyosarcoma was confirmed on histopathological exam and the investigation closed.

#### **Brown dog tick confirmed**

A brown dog tick (*Rhipicephalus* 

sanguineus) was found by a veterinarian during examination of a client's dog, and the identification was confirmed by an MPI entomologist. The dog had never travelled overseas. A field visit by an Incursion Investigator and an expert parasitologist found no further ticks at properties that had a risk association with the dog. Blood tested negative to agents vectored but not present in New Zealand. Tests were negative by serology (IFAT) for Babesia canis, B. gibsoni and Ehrlichia canis, and PCR was negative for Anaplasma phagocytophilum, Babesia spp., Borrelia spp. and *Ehrlichia* spp. The dog had been sleeping in a building where machinery had been imported, but this was considered to be a low risk for introduction of ticks. A biosecurity response was initiated including surveillance but no further ticks were found.

#### **Exotic ticks identified**

A traveller returned from Perth, Australia alerted MPI via the exotic pest and disease hotline after having a tick removed by their doctor. The tick was sent to the Plant Health and Environment Laboratory, Tamaki, for identification by an entomologist. It was a nymphal *Ixodes amblyomma*, but because it was missing mouth parts and scutum no further morphological identification was possible.

A veterinarian called the MPI exotic pest and disease hotline to report finding a single small tick on a New Zealand-bred dog from a Hawke's Bay farm. The tick was submitted to the Plant Health and Environment Laboratory (Tamaki) and identified as a nymph of the New Zealand cattle tick (*Haemaphysalis longicornis*). Exotic ticks were excluded and the investigation was stood down.

#### **Exotic parasites excluded**

A member of the public called the MPI exotic pest and disease hotline to report fleas that she thought might be exotic. She had two houses that she said were infested with the fleas, which persisted despite numerous treatment measures by herself and professional exterminators. Specimens were sent to the Plant Health and Environment Laboratory (Tamaki), where they were determined not to be fleas, but fragments of rolled lint and other debris. The caller was advised accordingly, and the local Medical Officer of Health was also alerted because of the possibility that the caller was suffering from delusional parasitosis. Exotic insects were excluded and the investigation was stood down.

# Infectious bursal disease ruled out

A poultry industry veterinarian undertaking routine monitoring during processing reported via the MPI exotic pest and disease hotline cases of gross bursal changes including bursal atrophy or enlargement with mild haemorrhage. Changes affected about 5 percent of the bursae from a single barn on a commercial housed broiler operation. MPI's Incursion Investigation team followed up with sampling and testing of birds from the flock. In addition, mortality monitoring and bursal evaluation of daily mortalities was carried out by the farm's veterinarian, with no abnormalities detected. At processing, samples of serum (20) and fresh (20) and fixed (10) bursae were collected from any birds showing evidence of atrophied or enlarged bursae. Serological screening using the IDEXX IBD ELISA identified no reactors and molecular assays for IBD virus were negative. Histological findings were normal for the age of the birds, including mild epithelial hyperplasia in some bursae and moderately cellular lymphoid follicles. Exotic disease was excluded and the investigation was stood down.

# Honey bee mortalities investigated

The manager of a commercial apiary in the Bay of Plenty region called the MPI exotic pest and disease hotline to report finding 36 dead hives in an apiary near to maize crops. No other apiaries were affected. The notifier believed systemic poisons used on the maize crops were killing his bees. Mostly young bees seemed to be dying, and could be found at the front of hives. Besides poisoning by agricultural toxins, bee deaths may be caused by other factors such as viral stress, the microsporidian parasite Nosema ceranae, wasps and other pests. The exotic tracheal mite (Acarapis woodi) can also cause bee deaths. Fresh dead bees were collected by the notifier and submitted to the Animal Health Laboratory. Testing for pathogens confirmed the presence of deformed wing virus (DWV) and N. ceranae. A subcontracted panel for more than a

hundred persistent organic pollutants only detected measurable residues of the chemical fluvalinate (at 0.41 mg/kg). Fluvalinate is the active ingredient in Apistan strips, used commonly as a hive treatment to control the parasitic mite Varroa destructor. The level of fluvalinate detected was considered significant, and likely to affect the health of queens and young bees. In this case, the combination of infectious agents and high levels of fluvalinate likely played a role in the bee deaths. Apistan strips left in hives for longer than the recommended interval were thought to be a possible cause. No other toxins were detected, and exotic disease was ruled out by confirmation of several likely endemic causes. The investigation was stood down.

A beekeeper called the MPI exotic pest and disease hotline to report a mass dieoff of bees in about 150 hives within a number of apiaries in close proximity. Based on the acuity of effects, poisoning was considered to be the most likely cause. However, testing of bees and honey for a wide array of chemicals (including carbaryl, fiprinol and imidacloprid) using a multi-residue analysis did not detect chemical toxins. Tracheal mite (Acarapis woodi) was not detected by PCR and the levels of Nosema ceranae were relatively low. Exotic disease was excluded and although unconfirmed, the presumptive diagnosis for the mortality event was from an unidentified poison.

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#### MARINE AND FRESHWATER

# Quarterly report of investigations of suspected exotic marine and freshwater pests and diseases

#### Sabella spallanzanii excluded

An MPI staff member found a polychaete in Wellington Harbour that resembled the exotic pest Mediterranean fanworm (*Sabella spallanzanii*). After phoning the MPI exotic pest and disease hotline, the samples were submitted to the Marine Invasive Taxonomic Service (MITS) and identified as the native species *Pseudobranchiomma grandis*. No biosecurity risk was associated with the find, so the investigation was closed.

# *Styela clava* range extension confirmed

A member of the public notified MPI after finding an exotic sea squirt on mussel farm spat lines in the Tasman district. Specimens were collected and submitted to MITS for identification. A taxonomist confirmed that the specimen was the invasive sea squirt, Styela clava. A reproductive assessment revealed that one of specimens had spawned. S. clava was first reported in New Zealand from Auckland in 2005 and was subsequently found to be present throughout the Hauraki Gulf and at the Port of Lyttelton. Since then it has become established in several other places and also found on fouled vessels throughout the country. As this report represents a range extension, the investigation was referred to the Response Team to liaise with affected parties.

#### **Exotic ascidian excluded**

A member of the public observed what was described as "an orangey-yellow, slimy mass" on rocks at low tide at a beach in Lyttelton Harbour. Concerned that it might be the result of a pollution event, they notified the Canterbury Regional Council, which had already received several reports of the slime and notified MPI. A sample was collected and submitted to MITS, where it was identified as an indigenous colonial ascidian, *Aplidium adamsi*. As there was no biosecurity risk, the investigation was closed. Exotic marine pest and aquatic disease investigations are managed and reported by MPI Investigation and Diagnostic Centre and Response, Wallaceville. The following is a summary of investigations of suspected exotic marine diseases and pests during the period from July to September 2016.

#### Mollusc mortality events investigated

The CEO of Aquaculture New Zealand rang the MPI pest and disease hotline to report a 5 percent mortality of large green-lipped mussels (Perna canaliculus) in farms on the Coromandel Peninsula. This is a high mortality rate for large mussels as the baseline mortality is very low. Samples were sent to the MPI Animal Health Laboratory to rule out the presence of pathogenic agents. Samples consisted of both preserved specimens for histology and chilled whole mussels for molecular testing and bacteriology. Unfortunately the histology specimens were inappropriately fixed, showing autolysis and tissue degradation. There was also evidence of bacterial overgrowth, suggesting the mussels had already been dead when fixed. One of the six preserved mussels was heavily infested with an unidentified microcell haplosporidian. Haplosporidians are obligate parasites and some species have caused significant bivalve mortality overseas. Histologically, the haplosporidian had the morphology of a Bonamia-like organism, although Bonamia-specific PCR probes and generic haplosporidian probes gave negative results. Specific PRC probes for Marteilia sydneyi, M. refringens, Haplosporidium nelsoni and Microcytos roughleyi were also negative. Attempts to identify the haplosporidian by TEM were unsuccessful owing to the poor fixation of the tissues. Because it was present in only one mussel, the haplosporidian was considered an incidental finding. Bacterial isolates included endemic Vibrio spp., but no primary pathogenic agents were identified. The cause of

the mortality event was likely to be the unusually warm adverse environmental conditions, causing the mussels to be immunocompromised and succumbing to bacterial infection. No biosecurity risk was identified, so the investigation was closed.

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### PLANTS AND ENVIRONMENT Developing tools for the detection of brown marmorated stink bug in imports

# The risk of incoming diapausing BMSB

In recent years, the number of brown marmorated stink bug or BMSB (Halyomorpha halys) interception events at the New Zealand border has increased, especially during the southern hemisphere summer months (Figure 1(b)). This is due to BMSB's overwinter behaviour: the bugs settle into overwintering sites during the northern hemisphere autumn and may subsequentially "hitchhike" to NZ on items they have settled upon. The three types of incoming items most used by overwintering BMSB in the past two years have been motor vehicles (both new and used), travellers' personal effects and freight containers. Increasing efforts to better understand the behaviour of preoverwintering BMSB and overwintering aggregations could lead to improved procedures for managing BMSB

incursions and preventing establishment. Numerous research projects are being carried out in this field.

#### Defensive responses of diapausing BMSB

When threatened or agitated, stink bugs release aldehydes that create an unpleasant odour. Until recently this behaviour in overwintering BMSB had not been confirmed. However, it has now been shown not only that overwintering BMSB are capable of this response, but they are more likely than active BMSB to use it when agitated. A PhD research project at the Bio-Protection Research Centre, Lincoln University (BPRC), funded by Better Border Biosecurity (B3), is investigating whether detecting these defensive compounds could be a tool for finding BMSB associated with imported freight at the border. Sensitive analytical methods are being developed and tested





Figure 1: Number of BMSB interception events at the NZ border (a) during risk periods since 2005; (b) by month. Source: Catherine Duthie, pers. comm. 28 Surveillance 43 [4] 2016

to identify these compounds in large and complex spaces. Research in the US has shown that trained sniffer dogs can detect as few as two overwintering BMSB, even when the bugs are not agitated (Lee *et al.* 2014). This suggests that overwintering bugs are continuously releasing volatile compounds that do not necessarily have a defensive purpose. If identified, these compounds have the potential to broaden this investigation into using chemical techniques for monitoring freight in areas such as sea ports that may not be suitable for using sniffer dogs.

#### Effect of transportation on diapausing BMSB

The BMSB research at the BPRC has led to a collaboration with a group of research entomologists at the USDA-ARS Appalachian Fruit Research Station in West Virginia. The challenge this group is currently working on is to determine how real-life shipping conditions affect the responses of overwintering BMSB. For this, the team is using a robot (Figure 2) to simulate agitation associated with shipping aggregations of overwintering BMSB over week-long periods. This should establish whether such agitation is sufficient to trigger a defensive response from the bugs. In another simulation, overwintering BMSB are exposed to temperature fluctuations over 26 days, the time a cargo ship takes to travel from the east coast of the US to New Zealand. Determining BMSB responses to such simulations should give better information on their biological state when they arrive in NZ (e.g., in overwinter, active, mobile), the proportion surviving within an imported population, and the feasibility of using chemical detection to monitor high-risk pathways.

#### Vibrational communication of BMSB in pre-diapausing clusters

Research has shown that BMSB rely on vibrational signals for intraspecific communication such as sexual competition, mate location and predator/ prey interactions. Recent findings from



Figure 2: Robot arm programmed to simulate ship movements. The four metal boxes attached are designed to contain populations of overwintering BMSB during their simulated journey.



Figure 3: Spectrogram of a male BMSB vibration during the aggregation process. The acquired vibration is composed of two different types of syllables with a dominant frequency around 70 Hz.

vibrations may play a significant role in pre-overwinter aggregation behavior, though the mechanisms that regulate this are unknown. However, recently several types of vibrations, an example of which is shown in **Figure 3**, have been recorded among established and growing clusters of pre-overwintering BMSB. If these vibrations are consistently associated with the process of cluster formation, it may be possible to develop an acoustic detection or pest management tool using an attract-and-kill strategy.

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# Super-yachts and other recreational vessels as a vector of exotic ant incursions

Over the past 10 years New Zealand has experienced a steady growth in the numbers of private yachts and motor vessels, both small and large, arriving from overseas (Tourism New Zealand 2015).

Correspondingly, the marine services industry has also seen an increase in refit and repair work being done on these vessels. Many of them also travel the coastline of NZ, visiting a number of places along the way. This means they are often in the country for long periods of time and in a number of places.

Biosecurity risks are associated with all vessels arriving in NZ and these are managed by the Ministry for Primary Industries (MPI). All such vessels are subject to MPI border clearance inspections (MPI 2016), but private yachts and motor vessels pose a particular risk of exotic ant incursions because they offer a favourable environment to harbour structureinfesting ants. Vessels typically have many internal cavities and crevices suitable for ants to make their nests. Engines and systems such as water pipes and wiring provide warmth and allow access throughout the vessel. Food stores are often large, in order to supply crews for extended periods, and larger vessels (so-called super-yachts) often have large commercial-style galleys (kitchens) that provide an abundance of food for foraging ants. These environments, combined with the cryptic nature of some species, mean that nests of ants are often present on vessels but remain undetected for some time.

It is therefore not surprising that a number of exotic ant infestations have recently been found on vessels arriving in NZ. Sometimes crews report the infestations themselves; quarantine officers discover infestations during arrival inspections, and infestations may be uncovered during refit and repair work, when the internal structure of the vessel is disassembled.

Table 1 lists recent ant incursions foundin private vessels arriving in NZ. Themost common species found was theghost ant (*Tapinoma melanocephalum*)(Figure 1), with carpenter ant(*Camponotus* sp.) the second mostcommon. Other species foundinclude brown crazy ant (*Paratrechina longicornis*), parrot ant (*Nylanderia* sp.),white-footed ant (*Technomyrmex* sp.)and the red imported fire ant (RIFA,Solenopsis invicta).

Most infestations have been found in the Auckland region (**Table 1**) as this appears to be the most popular region for arrivals. The Auckland and Northland regions are also a hub for refit and maintenance work catering for super-yachts and other large vessels.

#### Table 1: Records of exotic ants found in private vessels arriving in New Zealand

Date	Location	Species	Vessel type	Ports previously visited	Discovered by
Sep 2016	Westhaven Marina, Auckland	Tapinoma melanocephalum, Camponotus sp.	Super-yacht	Pacific Islands	Refit workers
Oct 2015	Viaduct Harbour, Auckland	Tapinoma melanocephalum	Motor launch	Fiji	Crew
Oct 2015	Westhaven Marina, Auckland	Tapinoma melanocephalum, Camponotus sp.	Super-yacht	Pacific Islands	MPI inspection, refit workers
Nov 2014	Gulf Harbour Marina, Auckland	Tapinoma melanocephalum	Yacht	Fiji	Owner
Apr 2014	Silo Park Marina, Auckland	<i>Technomyrmex</i> sp.	Super-yacht	Caribbean, Panama, Pacific Islands	Crew
Feb 2012	Nelson Marina, Nelson	Tapinoma melanocephalum	Yacht	Pacific Islands	MPI inspection
Nov 2010	Viaduct Harbour, Auckland	<i>Nylanderia</i> sp.	Super-yacht	New Caledonia	Refit workers
Nov 2010	Chaffer's Marina, Wellington	Camponotus sp.	Yacht	Vanuatu	Owner
Oct 2009	Westhaven Marina, Auckland	Tapinoma melanocephalum, Camponotus sp. (Figure 1)	Large luxury motor launch	Pacific Islands	MPI inspection
Mar 2007	Viaduct Harbour, Auckland	Paratrechina Iongicornis	Motor launch	Fiji	Crew
Feb 2007	Westhaven Marina, Auckland	Solenopsis invicta	Yacht	Caribbean, Pacific Islands	Owner

\* Source: unpublished reports by FBA Consulting.



Figure 1: Ghost ant (Tapinoma melanocephalum) feeding on bait aboard a motor launch at Westhaven Marina, October 2009. Photo: Brett Rawnsley

In several ways the biosecurity risk associated with ants on private vessels differs from that with commercial shipping. Private vessels typically spend longer in each port - often weeks and months rather than few hours or days. Also the locations of vessel stopovers are greater in number and more geographically widespread. In contrast, commercial ships mostly visit just one or two of 10 major deepwater ports on each voyage to NZ. After first arrival and biosecurity clearance, smaller vessels can travel to or stay at any location around the country, and frequently visit many places during their time here. If inspection at the port of arrival fails to discover an infestation of ants on board, this may increase the potential for ants to get ashore discreetly in an unmonitored place that is not traditionally considered high risk. However, the National Invasive Ant Surveillance programme does undertake surveillance at what are considered high-risk marinas around the country.

This underlines the importance of ongoing border surveillance for invasive ants by close inspection of vessels and marinas all around the country. It also highlights the need for MPI to continue engaging with marine service industries to raise awareness and report biosecurity issues that may arise during the course of their work. Many of the ant interceptions have been reported via passive surveillance, which highlights the importance of ongoing engagement with the industry by MPI.

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### Plants and environment investigation report

#### Fanwort in stream

The Auckland City Council reported in July that fanwort (Cabomba caroliniana), an unwanted organism under the Biosecurity Act 1993, had invaded ponds in West Auckland. Fanwort is a submerged invasive aquatic plant that can form dense mats at the water surface. The site consisted of two stormwater retention ponds on the Paremuka Stream, Henderson, with a total area of around 2.5 ha. It is uncertain how fanwort came to be growing in the ponds, but dumping of aquarium matter is a likely cause. Fanwort was dominating these ponds and appeared to be the only submerged species, along with emergent parrot's feather (Myriophyllum aquaticum). This appeared to be a recent incursion, with no record of fanwort from an ecological survey conducted in 2013. A survey of the stream found no further fanwort plants. C. caroliniana is a weed of national significance in Australia and problematic in many states there. In New Zealand it has been sold in the aquarium trade for more than 40 years and previously was only known in the wild at one site - a tiny spring-head feeding into Meola Creek at Western Springs, Auckland, where it grew beneath a number of other aquarium/pond species. In 2007 the National Institute of Water and Atmospheric Research (NIWA) assessed the competitive performance of fanwort in pairwise competition trials and based on the results no management action was recommended, nor was the plant added to the National Plant Pest Accord (NPPA) list. The NPPA is designed to prevent the sale, distribution and propagation of listed pest plants which, if allowed to spread further, could seriously damage New Zealand's economy and environment. Since there is an infestation it may be appropriate to revisit the plant's status and add it to the NPPA list. A site assessment was undertaken by an Incursion Investigator, MPI botanist and the Auckland City Council. A rapid assessment report outlining management options was submitted to the response team and a response was initiated.

The Ministry for Primary Industries' (MPI) Incursion Investigation (Plants & Environment) and Plant Health Environment Laboratory (PHEL) teams investigate and diagnose suspect exotic pests and diseases in the plant and environment sectors. Investigators and scientists are based in Auckland and Christchurch. These teams provide field investigation, diagnostic testing and technical expertise with regard to new pests and diseases affecting plants and the environment. They also have surveillance and response functions and carry out research and development to support surveillance and incursion response activities.

#### **Termites on yacht**

The Western drywood termite Incisitermes minor was identified from a yacht during a refit at Waipapa, 20 km north of Kerikeri. I. minor is an unwanted organism under the Biosecurity Act 1993. While most drywood termites only infest dead trees, others are economically important pests because they nest in wooden framing and studs in homes or in commodities such as wooden furniture. In New Zealand, I. minor would pose a risk to wooden structures including buildings and wooden taonga. The present detection would devalue the current owner's investment in the vessel and affect his ability to sail it to countries where I. minor is a regulated pest. The yacht was built in 1990, had been purchased by its present owner five months previously and as far as he knew it had not left New Zealand waters in 22 years. The yacht had in the past sailed in US waters including Hawaii and Los Angeles, California, via Tonga and Palmyra Atoll. The boatbuilder carrying out the refit had found dead termites inside the yacht, and these were collected by a Quarantine Officer from Whangarei and identified at MPI's Plant Health and Environment Laboratory (PHEL) as alates (reproductive caste) of I. minor. An Incursion Investigator inspected the yacht and found more alates and shed wings throughout the vessel. The boatbuilder was asked to keep the yacht sealed to prevent the escape of alate termites into the environment. The vacht was constructed using the native timber kahikitea (Dacrycarpus dacrydioides), with a fibreglass skin over the hull and

superstructure. A Rapid Assessment Report was submitted to initiate a response and the vessel was fumigated under tarpaulin using methyl bromide at an approved rate. The investigation was closed.

#### Termites in picture frames

Wooden picture frames shipped to New Zealand, along with other household items from a property in Bali, Indonesia, showed evidence of termite damage when one of the picture frames was being rehung in an Auckland apartment. There had been a five-week period during which the frame was not hung, and termite leads were seen at the time of rehanging, along with the shed wings of alate termites. The consignment, which also included hardwood carvings, had been fumigated before shipment and cleared by MPI on arrival, though no actual inspection had been undertaken. An Incursion Investigator visited the apartment to view the insect activity and collected a picture frame for removal to the Plant Health and Environment laboratory (PHEL) Tamaki for destructive sampling to collect specimens for identification. An interim identification of the adult termites was *Cryptotermes* sp., possibly *C. sumatrensis*. PHEL does not have specimens in its collection or sequences of C. sumatrensis to compare with, so it would be difficult to confirm the identification. To mitigate the risk of establishment the woodware associated with this consignment was collected and sent for fumigation with methyl bromide at an approved rate. The investigation was closed.

#### Borer in bed slats

A live borer-beetle larva was found in a wooden slat while unpacking a Zephyr brand bed purchased from Harvey Norman in Porirua. The timber of the bed slats had much tunnelling evident on the surface. Harvey Norman had purchased the bed unit (ends, base and slats) from Synargy Corporation Ltd in Christchurch. Synargy, which imports at least one container of furniture a month from the Chinese company Trendworld Industrial Corporation Ltd, received the Zephyr beds from Trendworld's factory in Vietnam.

Two larvae were found, one sent by the submitter and the other (also alive) after destructive sampling of the wooden slats at the Plant Health and Environment Laboratory (PHEL) Christchurch. Initial identification based on morphological characteristics was only to subfamily level (Coleoptera: Bostrichidae: Lyctinae). The larvae were sent to PHEL Tamaki for further identification by molecular methods but the results were inconclusive with determination only to family level (Bostrichidae - auger beetles or false powderpost beetles). Harvey Norman replaced all the bed slats with ones from a showroom model and Synargy Corporation Ltd shipped the infested bed slats back to its Christchurch warehouse to destroy them by burning. The investigation was closed.

#### Catnip seeds sold online

A man in the United Kingdom was operating a business called Buysend, selling "Cosmic Catnip" seeds, a type of growable grass for cats on the Trademe website. The product did not meet New Zealand's standards for importation of seed for sowing because it contained other seeds including barley (Hordeum vulgare), oats (Avena sativa) and wheat (Triticum sp.), all of which require a phytosanitary certificate with additional declarations. The seller was contacted and informed about MPI's requirements and regulations. He was sent information on the Import Health Standard - Seed for Sowing 155.02.05. and regulations on importing seed into New Zealand (via MPI's website).

There was a similar case in 2015 where the same business (Buysend) was selling doff grass and wild flower seeds on Trade Me On this occasion they were also informed about New Zealand's seed importation requirements. These types of cases illustrate that online trading opens up pathways for biosecurity risk goods to come into New Zealand. The case was closed as the seller agreed to remove his listing from the Trademe website.

#### Bloodlike substance in a banana

A member of the public reported a red, bloodlike substance in a Dole banana imported from the Philippines and purchased from Pak'n'Save in Papakura. In the past, bananas showing similar symptoms have been associated with Colletotrichum musae (a common banana pathogen) or Burkholderia cenocepacia (a ubiquitous bacterium established in New Zealand). Fungal isolates from the banana were identified as Fusarium *musae* and *Gliomastix polychroma*, both species not known to be present here. Pathogenicity tests conducted in Europe confirmed that F. musae causes leaf spots on banana fruits. This species is widely known from the Canary Islands, Central America, Ecuador and the Philippines. It, has only been isolated from banana plants (Van Hove et al. 2011) and is not known to affect any other plant species of economic or environmental significance in New Zealand.

Limited information is available about this species, but biosecurity impacts are considered to be low or negligible based on information gathered so far. While F. musae has been isolated from human tissues, it appears to be difficult to distinguish morphologically from typical Fusarium verticillioides strains in routine mycological tests (Hirata et al. 2001). G. polychroma also appears unable to produce the mycotoxin fumonisin because it lacks most fumonisin biosynthesis genes (Glenn et al. 2008). However, recent research (Triest et al. 2015) shows that some F. musae strains isolated from blood samples, biopsies of immune-suppressed patients and from the clinical environment were misidentified as F. verticillioides. These were suggested to have a role in causing the human disease fusariosis. The study also suggested that imported bananas might carry F. musae spores and cause infection in humans; or, alternatively, that the natural distribution of F. musae is far broader than originally suspected and it is also present on other plant hosts. Bananas are fast-turnover fresh produce and it was expected that the shipment from which this case was reported

had been sold and consumed before pathology tests could be concluded. Relevant MPI teams were advised to enhance awareness and follow-up. No further action was recommended in terms of post-border biosecurity risk management. The investigation was closed but the biosecurity risk remains.

#### Termites found at Prime Port, Timaru

MPI Quarantine Officers were alerted to termites in wharf timbers at Prime Port in Timaru. A sample of the termites was sent to MPI's Plant Health & Environment Laboratory (PHEL), Christchurch and identified as the Australian dampwood termite (Porotermes adamsoni). P. adamsoni has been found at several wharves around New Zealand, is an established species and is not an unwanted organism. It was first found here in imported host material about 70 years ago and has since been intercepted many times on infested timber of Australian origin. In Australia, P. adamsoni is not considered a significant pest in houses and other wooden structures. However, it has historically been reported as a pest of forestry because it can attack live eucalyptus trees. The case was closed.

#### Suspect myrtle rust

A person walking the Paekakariki Escarpment track on the Kapiti Coast noticed a whiteywood/mahoe (Melictyus ramiflorus) tree with yellow rust pustules on its leaves. This species is not in the family Myrtaceae, which are susceptible to myrtle rust (Puccinia psidii). Myrtle rust spores are microscopic and can easily spread across large distances by wind. It is thought that they could cross the Tasman Sea from Australia, a pathway MPI cannot manage. Spores could also be transported on clothing, equipment and by insects. On investigation, none of the characteristic myrtle rust pustules or uredinospores were seen on the sample, though the host was infected with a number of fungal diseases which were identified as Peyronellaea pinodella (syn. Phoma pinodella), Colletotrichum torulosum and Pestalotiopsis sp. *P. pinodella* was considered the most likely primary pathogen as it has been reported to cause foliar blight similar to the symptoms observed on whiteywood. This is a new host record for this species in New Zealand. C. torulosum has

previously been recorded from *Solanum melongena* (aubergine/eggplant) and this too is a new host record in New Zealand. *Pestalotiopsis* sp. is a secondary pathogen. All three fungal species are established in New Zealand. The investigation was closed.

#### Borer holes in kwila

The notifier had bought about 600 metres of kwila decking timber from Mitre 10 in Rangiora. While building the deck they noticed borer damage in one piece of timber and took it back for replacement. The retailer advised that all its imported timber was either fumigated or kilndried so the borer damage was old and inactive. However, subsequently the notifier noticed more borer damage on the timber. The live insect was identified by the Plant Health & Environment Laboratory as Cartodere bifasciatus, the brown scavenger beetle, which is present in New Zealand but not actually a timber pest. The sawdust or frass seen was indicative of the manufacturing process and not from insect activity. The investigation was closed.

#### New New Zealand Alydidae (Heteroptera)

A Northland resident found an unusual insect on the beach at Henderson Bay and posted a photograph on the website NatureWatch New Zealand. The image uploaded provided enough diagnostic detail to confidently identify the specimen as Melanacanthus margineguttatus (Hemiptera: Alydidae). This species was not previously known to be present in New Zealand, though it had been previously found at Tahunanui, Nelson, in 1928 on marram grass (Psamma arenaria) near the sea. This population was believed to have been destroyed in a fire when the area was burnt off and the species had not been recorded here since.

Information on the impact of this species is limited but it has been recorded on rattlepods *Crotalaria quinquefolia* (Leguminose), pigeon pea *Cajanus cajan* (Fabaceae), belly ache bush *Jatropha gossypifolia* (Euphorbiaceae), wild orange *Capparis mitchellii* (Capparidaceae) and paperbarks *Melaleuca* spp. (Myrtaceae). Most members of this family feed on legumes. In Australia, another species, *M. scutellaris*, is an infrequent but major pest of many legume crops, such as mung beans, soybeans, navy beans and adzuki beans.

A Northland Entomologist was contracted to visit Henderson Bay to look for more *M. margineguttatus* but found none and MPI's Plant Health and Environment laboratory (PHEL) decided to wait until November to carry out another survey. This second survey also yielded no specimens of *M. margineguttatus*. This line of enquiry will remain open to determine the establishment status and potential impacts of *Melanacanthus margineguttatus* in New Zealand.

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### PEST WATCH: 1 July – 23 November 2016

Biosecurity is about managing risks: protecting New Zealand from exotic pests and diseases that could harm our natural resources and primary industries. MPI's Investigation & Diagnostic Centres and Response (IDC & R) directorate devotes much of its time to ensuring that new organism records come to its attention, and to following up as appropriate. This information was collected from 1 July 2016 to 23 November 2016. The plant information is held in the MPI Plant Pest Information Network (PPIN) database. Wherever possible, common names have been included. Records in this format were previously published in the now discontinued magazine *Biosecurity*. To report suspect new pests and diseases to MPI phone 0800 80 99 66.

#### Validated new to New Zealand reports

Туре	Organism	Host	Location	Submitted by	Comments
fungus	Capronia epimyces no common name	<i>Pinus radiata</i> radiata pine	Auckland	IDC & R (General Surveillance)	
fungus	Cytospora diatrypelloidea no common name	<i>Eucalyptus leucoxylon</i> white ironbark	Auckland	IDC & R (High Risk Site Surveillance)	This species is previously known only from Eucalyptus in Australia.
fungus	<i>Disculoides</i> sp. no common name	<i>Eucalyptus leucoxylon</i> white ironbark	Auckland	IDC & R (High Risk Site Surveillance)	This isolate represents an undescribed species of <i>Disculoides</i> .
fungus	Holocryphia eucalypti no common name	<i>Eucalyptus leucoxylon</i> white ironbark	Auckland	IDC & R (High Risk Site Surveillance)	This species has been found on various <i>Eucalyptus</i> species overseas.
fungus	Pezicula neocinnamomea no common name	<i>Eucalyptus leucoxylon</i> white ironbark	Auckland	IDC & R (High Risk Site Surveillance)	This fungus is likely to be an endophyte.
fungus	Phialocephala bamuru no common name	<i>Cynodon dactylon</i> Bermuda grass	Auckland	IDC & R (General Surveillance)	Isolated from grass roots but it is uncertain whether the fungus is causing any disease.
fungus	Pseudophloeospora eucalyptorum no common name	Eucalyptus leucoxylon white ironbark	Auckland	IDC & R (High Risk Site Surveillance)	This species has been associated with leaf spots on <i>Eucalyptus</i> in Australia.
fungus	<i>Pseudoplagiostoma</i> sp. no common name	<i>Eucalyptus leucoxylon</i> white ironbark	Auckland	IDC & R (High Risk Site Surveillance)	This isolate represents an undescribed <i>Pseudoplagiostoma</i> sp.
fungus	Thielaviopsis ethacetica no common name	<i>Triticum aestivum</i> wheat	Dunedin	IDC & R (General Surveillance)	This fungus was previously known by the synonym Ceratocystis paradoxa.
fungus	Tuber dryophilum no common name	<i>Pinus</i> sp. pine	Mid Canterbury	IDC & R (General Surveillance)	An ectomycorrhizal fungus forming edible truffle-like fruiting bodies.
insect	<i>Chrysomphalus</i> sp. cf. <i>pinnulifer</i> Armoured scale insect	Ficus benjamina weeping fig	Auckland	S E Thorpe (General Surveillance)	Scales feeding on leaves.
insect	Eucalymnatus tessellatus Tessellated scale	<i>Rhopalostylis sapida</i> nikau	Auckland	IDC & R (High Risk Site Surveillance)	Has a broad range of hosts (more than 50 plant families).
insect	<i>Migneauxia lederi</i> Latridiid beetle	Found on grass clippings	Auckland	S E Thorpe (General Surveillance)	Cosmopolitan species present in Europe, Africa, Asia, South America and the US.
insect	Paropsisterna beata Eucalyptus leaf beetle	<i>Eucalyptus macarthurii</i> Paddy's River box	Wellington	IDC & R (General Surveillance)	Native to Australia. Only known hosts are <i>Eucalyptus</i> spp.
insect	Paropsisterna variicollis Eucalyptus variable beetle	<i>Eucalyptus globulus</i> Southern blue gum	Hawke's Bay	SCION (General Surveillance)	Native to Australia. Adults and larvae feed exclusively on <i>Eucalyptus</i> leaves.
insect	Stenchaetothrips bambusae no common name	<i>Bambusa</i> sp. bamboo	Auckland	IDC & R (General Surveillance)	Feeds on leaves, causing leaf rolling. This species is recorded in Burma, India and Malaysia.
mite	<i>Carpoglyphus</i> sp. no common name	Cordyline obtecta (syn. C. kaspar) Three Kings cabbage tree	Wellington	High Risk Site Survey	New to science species. The genus <i>Carpoglyphus</i> currently includes five species.
virus	Snowdrop virus Y SnVY	<i>Clivia miniata</i> clivia	Auckland	IDC & R (General Surveillance)	Chlorotic stripes on leaves observed.
virus	<i>Spiranthes mosaic virus 3</i> SpMV3	Phlox stolonifera phlox	Mid Canterbury	IDC & R (General Surveillance)	No obvious symptoms were observed.

If you have any enquiries regarding this information please contact surveillance@mpi.govt.nz

To report suspected exotic land, freshwater and marine pests, or exotic diseases in plants or animals, call:

#### 0800 80 99 66

Investigation and Diagnostic Centre – Wallaceville 66 Ward Street Upper Hutt Tel: 04 526 5600

Investigation and Diagnostic Centre – Tamaki 231 Morrin Road St Johns Auckland Tel: 09 909 3568

Investigation and Diagnostic Centre – Christchurch 14 Sir William Pickering Drive Christchurch Tel: 03 943 3209 Veterinary Diagnostic Laboratories

#### GRIBBLES VETERINARY PATHOLOGY

- AUCKLAND Courier: 37–41 Carbine Road, Mount Wellington, Auckland 1060 Postal: PO Box 12049, Penrose, Auckland 1642 Tel: 09 574 4701 Fax: 09 574 5304
- HAMILTON Courier: 57 Sunshine Ave, Hamilton 3240 Postal: PO Box 195, Hamilton 3240 Tel: 07 850 0777 Fax: 07 850 0770
- PALMERSTON NORTH Courier: 840 Tremaine Avenue, Palmerston North 4440 Postal: PO Box 536, Palmerston North 4440 Tel: 06 356 7100 Fax: 06 357 1904
- CHRISTCHURCH Courier: 7 Halkett Street, Christchurch 8140 Postal: PO Box 3866, Christchurch 8140 Tel: 03 379 9484 Fax: 03 379 9485
- DUNEDIN Courier: Invermay Research Centre, Block A, Puddle Alley, Mosgiel, Dunedin 9053 Postal: PO Box 371, Dunedin 9053 Tel: 03 489 4600 Fax: 03 489 8576

#### NEW ZEALAND VETERINARY PATHOLOGY

- AUCKLAND Courier: NZCCM, Gate 2, Auckland Zoo, Motions Road, Western Springs, Auckland 1022 Postal: PO Box 44 422, Point Chevalier, Auckland 1246
- HAMILTON Courier: Cnr Anglesea and Knox Streets, Hamilton Postal: PO Box 944, Hamilton Tel: 07 839 1470 Fax: 07 839 1471
- PALMERSTON NORTH Courier: IVABS Building, 1st Floor, Massey University, Tennant Drive, Palmerston North Postal: PO Box 325, Palmerston North Tel: 06 353 3983 Fax: 06 353 3986



#### New Zealand Government