Scientific Evaluation of Bovine Post Mortem Examination Procedures in New Zealand

Part B

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November 2011

Ministry of Agriculture and Forestry
Te Manatū Ahuwhenua, Ngāherehere
BIOSECURITY NEW ZEALAND
NEW ZEALAND FOOD SAFETY AUTHORITY

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1. Executive summary

This document describes a scientific evaluation of the likely contribution of routine post mortem examination of selected lymph nodes of cattle slaughtered in New Zealand to food safety and to food suitability. Currently all carcass lymph nodes are required to be examined by intensive examination while the mesenteric lymph nodes are examined by viewing and palpation. In addition to questions over food safety aspects, there is scientific consensus that intensive routine examination of lymph nodes is likely to increase cross-contamination of some tissues.

This evaluation is based on: review of the scientific literature, unpublished MAF reports on meat hygiene, comparison with regulatory requirements in other countries, and expert opinion. The evaluation cannot be considered a risk assessment because of the lack of quantitative data on actual changes in levels of hazards consequential to different post mortem procedures. However, there is clear scientific evidence of the likely value of different procedures relative to overall food safety and suitability outcomes achieved by post mortem meat inspection.

The study also evaluated the contribution that routine post mortem examination of the selected lymph nodes makes to animal health surveillance for bovine tuberculosis.

The study indicates that the current intensity of routine examination for some of the lymph nodes of cattle is not scientifically justified. For specified lymph nodes, there is negligible evidence of food safety and/or food suitability benefit arising from routine examination for grossly-detectable abnormalities. Further, the specified lymph nodes do not provide an indicator function for other tissues in terms of judgements of food safety and/or food suitability.

Routine examination of the mesenteric lymph nodes was the only procedure of these lymph nodes of benefit to animal health surveillance for bovine tuberculosis.

The recommendations for post mortem examination of cattle slaughtered in New Zealand that arise from this study are:

- The atlantal, lumbar chain, renal, subiliac and superficial cervical lymph nodes should not be routinely examined.
- The mesenteric lymph nodes should be routinely examined by viewing.

Given the limited information available on the possible value of the internal iliac, superficial inguinal and supramammary lymph nodes, as indicators for judgement of tissues from which they receive efferent lymphatic drainage, current procedures should be maintained pending further studies.

Table 1: A summary of procedures carried out in New Zealand and other countries and recommendations for change to the New Zealand procedures (V - view, P- palpate and I - incise)

Lymph node	NZ current	NZ proposed	Aus	Canada	US	EU
Atlantal (in head &/or c/c)	I (in head &/or c/c	none in head or c/c	I (in head only) or V or none	I (in head only)	I (in head only)	I (in head only)
Internal iliac	I	I (interim)	V or P	none	P	none
Lumbar chain	I	none	none	none	none	none
Mesenteric	VP	V	V	V	V	VP
Renal	I	none	none	none	none	none
Subiliac	P & I	none	none	none	none	none
Superficial cervical	P & I	none	none	none	none	none
Superficial inguinal	I	I (interim)	V or P	none	P	none
Supramammary	I	I (interim)	V or P	none	P	V

2. Background

There is an overwhelming consensus that in contemporary meat hygiene systems, the main threats to public health are from a) bacterial zoonotic agents that can be carried and excreted via faeces by animals without symptoms and b) the presence of residues of pharmacologically-active substances (authorised and unauthorised) and/or contaminants. Traditional post mortem meat examination is increasingly shown as being of very little value in mitigating these risks (Hathaway and McKenzie, 1991; Hathaway, 1993; Edwards *et al*., 1997; Mousing *et al*., 1997; Hamilton *et al*., 2002; Sofos, 2008).

The most relevant biological hazards in the context of safety of meat and meat products of bovine origin include *Escherichia coli* O157, *Salmonella, Campylobacter,* and *Yersinia*; zoonotic pathogens usually transmitted through faecal contamination or cross-contamination of the carcass during slaughter and dressing. These hazards are rarely associated with grossly-detectable abnormalities and therefore public health risks are not mitigated by post mortem examination of carcasses. In fact, some examination tasks may even increase cross-contamination with these pathogens.

Post mortem examination of cattle in New Zealand was the subject of an early scientific study of procedures for the head and tongue (Anon, 1999). In 2011 MAF initiated a much more comprehensive body of work to scientifically evaluate all post mortem procedures. Part A involved a small range of tissues that were examined in New Zealand but not in other countries and this work is now completed (Anon.a, 2011)

This report documents Part B of the study; a scientific evaluation of the likely contribution of routine post mortem examination of selected lymph nodes of cattle in assuring food safety and suitability. Currently all carcass lymph nodes are required to be intensively examined (palpation or incision) except the mesenteric lymph nodes which are examined by viewing and palpation. The selected lymph nodes were those that have virtually no value in animal health surveillance, as informed by a detailed MAF epidemiological study on the distribution of *Mycobacterium bovis* lesions in cattle (Anon.b, 2011).

Part C of the study will incorporate all other tissues and organs subject to post mortem examination in New Zealand.

3. Methodology

This scientific evaluation is based on:

- Review of the scientific literature
- Unpublished MAF reports
- AsureQuality inspection databases
- Comparison with regulatory requirements in other countries
- Expert opinion

3.1. Procedures

The current procedures for selected lymph nodes of cattle slaughtered in New Zealand are shown in Table 2 and these were compared with procedures in other countries, as detailed in Appendix 1).

Table 2: Post mortem examination procedures for selected lymph nodes of cattle slaughtered in New Zealand

Lymph nodes	Procedures	Comment
Atlantal	Incise and view	Either on the head or carcass
Internal iliac	Incise and view	
Lumbar chain	Incise and view	
Mesenteric	View and palpate	Incise in cattle with lesions suspected of being tuberculosis and any cattle designated as specified for veterinary disposition (SPVD).
Renal	Incise and view	
Subiliac	Palpate or incise	Palpate in prime cattle and lightly muscled young bulls. Incise in prime cattle and young bulls which are designated SPVD at ante mortem or with tuberculous or actinoform lesions, and in young bulls if overlying tissue prevents effective palpation and all other cattle.
Superficial cervical	Palpate or incise	Palpate in prime cattle and lightly muscled young bulls. Incise in prime cattle and young bulls which are designated SPVD at ante mortem or with tuberculous or actinoform lesions, and in young bulls if overlying tissue prevents effective palpation and all other cattle.
Superficial inguinal	Incise and view	
Supramammary	Incise and view	

3.2. Anatomy of selected nodes

To support evaluation of different procedures, the anatomy of each selected lymph node was described in detail (Section 4). A description of the afferent and efferent lymphatic flow to and from each set of lymph nodes especially supports evaluation of a possible indicator function.

3.3. Biological hazards

Hazards that may be present in cattle (Table 3) were compiled from a variety of sources as well as the published scientific literature (Untermann, 1998; Schlundt, 2004; Goldsmid, 2005; Alonso *et al.*, 2010) Hazards not present in New Zealand were then excluded from further consideration. These included agents never reported in New Zealand (bovine spongiform encephalopathy, *Sarcocystis hominis)* and those eradicated from the country *(Brucella abortus, Echinococcus granulosus)*.

All hazards (biological, chemical and physical) that might be present in the lymph nodes of cattle slaughtered in New Zealand were then considered. The list was differentiated relative to hazards that may cause grossly-detectable abnormalities and cross-checked with other studies in the scientific literature (Tauxe, 2002; Dorny, 2009; Hugas and Liebana, 2009; Alonso *et al*., 2010).

To evaluate the value of routine post mortem meat examination, each hazard / abnormality combination was assessed in terms of:

- Systemic / generalised food safety conditions
- Localised food safety conditions
- Indicator value

Food safety considerations were evaluated relative to viewing, palpation and incision as possible procedural options for examination of lymph nodes. .The indicator function of any grossly detectable abnormalities in the lymph nodes was evaluated in terms of possible assistance to the meat inspector when making a disposition for tissues other than the lymph node itself. A commentary was also provided on the possibility of the hazard being present in the lymph node in the absence of grossly-detectable abnormalities.

The study cannot be considered a full risk assessment because the public health outcomes subsequent to different examination procedures were not modelled in a quantitative sense. However, the methodology provides for a clear scientific evaluation of the likely food safety (and suitability) value in terms of the changes in levels of hazards in meat and meat products that might be achieved from different post mortem examination regimes.

3.4. Suitability

A separate evaluation was undertaken for abnormalities that might be of value in the judgement of suitability, using the above approach.

3.5. Animal health considerations

The value of routine post mortem examination of the selected lymph nodes for animal health surveillance was taken into account in the study.

4. Anatomy of selected lymph nodes

Lymph nodes that occupy key positions in the lymphatic system of an animal have traditionally been considered as useful indicators of food safety on the basis of afferent vessels draining specified regions of the carcass or viscera.

4.1. Atlantal lymph nodes

The name of these lymph nodes (i.e. atlantal) used in relation to post mortem examination of cattle in New Zealand has continued from that used in text books from early last century (Sisson, 1930). The nomenclature is further confused by conflicting descriptions within the two standard meat inspection textbooks referring to these lymph nodes as the lateral and median retropharyngeal lymph nodes respectively (Thornton, 1962; Collins, 1970).

Thornton does not use the name atlantal but describes the lymph node that lies beneath the wing of the atlas as the lateral retropharyngeal lymph node and furthermore states that these lymph nodes are usually located at the neck end of the dressed carcass. However, this description is in contrast to Collins who describes the lateral retropharyngeal lymph node as being situated on the roof of the pharynx and being commonly left attached to the base of the tongue when that organ is removed from the head by the butcher. Collins describes the median retropharyngeal lymph node as lying in loose facia above the pharynx and being of up to 40 to 50 mm in length and that these lymph nodes were commonly left under the scrag of the neck when the head was removed by butchers. This corresponds to the lateral retropharyngeal lymph node described by Thornton.

More recent anatomical text books (Sisson and Grossman, 1975; Dyce *et al*., 2010) use the name lateral retropharyngeal to describe these lymph nodes. In Nomina Anatomica Veterinaria (2005), an internationally recognised authority on anatomical nomenclature, these lymph nodes are described as *Lnn. retropharyngei laterales*.

According to Sisson, the atlantal lymph node is situated ventral to the wing of the atlas, on the cleido-mastoideus tendon, and partly under cover of the mandibular salivary gland. It is related ventrally to the carotid artery, is usually discoid and from 40 to 50 mm in length. One or more small lymph nodes may occur near the large constant one, and with small haemolymph glands commonly present. The afferent vessels come from the tongue, the sublingual and mandibular salivary glands, the gums in part, the cervical part of the thymus and most of the hyoid and cervical muscles. They also come from the parotid, mandibular (syn. submaxillary) and medial retropharyngeal lymph nodes. The efferent vessels, which number from 3 to 6, converge to form the tracheal lymph duct.

These differences in anatomical nomenclature and position create difficulties when interpreting the intent and application of post mortem examination rules in different countries. Because of a lack of common understanding on anatomical location, as well as biological variability in anatomy, it is likely that actual implementation of procedures is highly variable. Further to this, the atlantal lymph node may be:

- Left intact on the head
- Transected by the dressing incision used to remove the head from the carcass

- Left on the distal end of the neck (the most common situation in New Zealand).

It is notable that the retropharyngeal lymph node referred to in the New Zealand bovine post mortem procedures is described as the internal retropharyngeal lymph node by Thornton, lateral retropharyngeal lymph node by Collins and medial retropharyngeal lymph node by Sisson.

4.2. Internal iliac lymph nodes

Nomina Anatomica Veterinaria describes these lymph nodes as *Lnn. iliaci interni* and part of the lymphocentrum iliosacrale (*lymphocentrum* refers to a lymph node or group of lymph nodes that occur in the same region of the body and receive afferent vessels from approximately the same region in most domestic animals).

The internal iliac lymph node is situated on the brim of the pelvis 5 to 8 cm in front of and medial to the deep inguinal lymph node. The lymph node may be composed of up to several nodes from 12 to 50 mm in length and may be exposed by an incision level with the junction of the sacrum and last lumbar vertebrae, lying some 175 mm from the vertebrae. Their afferent vessels come chiefly from muscles of the sublumbar region, pelvis, tail, thigh, the genital organs, the kidneys, bladder and urethra. They also receive vessels which are efferents of the popliteal, ischiatic, deep inguinal, external iliac and subiliac lymph nodes, as well as from the sacral and coxal lymph nodes while their efferents go to the lumbar lymphatic trunk which leads to the cisterna chyli.

In prime animals the internal iliac lymph nodes may be covered wholly or to some degree by fat while in young bulls and aged cows, the fat cover is much reduced or absent.

The internal iliac lymph nodes may act as post mortem examination indicators of infection and pathology in the hind legs as only the deep inguinal and the external iliac lymph nodes have any alternate lymphatic connection to the lumbar lymphatic trunk.

4.3. Lumbar chain lymph nodes

In Nomina Anatomica Veterinaria these lymph nodes are described as *Lnn. lumbales aortici* and *Lnn. lumbales proprii* as part of the lymphocentrum lumbale. This group is also considered to include *Lnn. renales*.

These lymph nodes are situated in the fat covering the lumbar muscles and are related anatomically to the aorta and posterior vena cava as well as in some carcasses occupying the spaces between the transverse processes. Some nodes may be described as superficial, while others are embedded in the loin suet, and haemo-lymph nodes are common in this region. The afferent vessels come from the sublumbar, abdominal and serratus dorsalis muscles, the lumbo-dorsal fascia, the paired viscera (such as the kidneys and adrenals), the peritoneum and the lumbar vertebrae while the efferent vessels go to the lumbar lymphatic trunk and the cisterna chyli.

Thornton considers that these lymph nodes also receive efferent vessels from the external and internal iliac, sacral and popliteal lymph nodes, while Collins and Sisson do not share this opinion.

4.4. Mesenteric lymph nodes

In Nomina Anatomica Veterinaria these lymph nodes are considered to belong to two groups, the lymphocentrum mesentericum craniale (including *Lnn. mesenterici craniales*, *Lnn. jejunales* and *Lnn. ileocaecales*) and lymphocentrum mesentericum caudale (*Lnn. mesenterici caudales*).

The mesenteric lymph nodes comprise a large number of elongated nodes which lie between the peritoneal folds of the mesentery and receive lymph from the intestines. Various authors have divided these lymph nodes into between two (Collins), three (Thornton) and seven (Sisson) subgroups according to their distribution along the small and large intestine. However the post mortem procedures used in New Zealand and our major trading partners do not distinguish between the various anatomical subgroups of the mesenteric lymph nodes and consequently there is no practical gain from a precise anatomical description of all nodes in the bovine mesentery.

In the anterior mesentery, there are lymph nodes that drain the duodenum, from which the efferent lymphatics pass to the portal nodes of the liver. The jejuno-ilieal lymph nodes lie in part of the mesentery to which the coils of the jejunum and ileum are attached. They vary in number from 10 to 50 and in length from 5 mm to 1.2 m. and form the main chain parallel to, and some 50 mm from, the intestine. There are also several caecal, colic and rectal lymph nodes in the posterior mesentery.

Generally the long narrow nodes are in the peripheral section of the mesentery, while numerous small nodes are scattered throughout the mesentery and extend centrally to the coils of the colon. Their afferent vessels come from the mesenteric part of the small intestine while the efferent vessels converge to form a large common efferent vessel. This receives efferent vessels of the caecal and colic lymph node groups, and runs upward and forward on the right side of the spiral mass of the colon, and reaches the ventral face of the posterior vena cava just behind the anterior mesenteric artery. Here it unites with the common efferent vessel of the gastric lymph nodes to form the intestinal lymphatic trunk.

4.5. Renal lymph nodes

In Nomina Anatomica Veterinaria these lymph nodes are described as *Lnn. renales* as part of the lymphocentrum lumbale.

The renal lymph nodes are situated between the posterior medial border of the kidney and the posterior vena cava and usually covered in fat. Sisson regarded the renal lymph nodes as being part of the lumbar chain. The lymph node can be exposed by making an incision lengthwise through the overlying blood vessel and continuing the incision 25 mm deep into the lumbar suet. The renal lymph node drains the kidney and adrenal body and conveys lymph to the cisterna chyli. They vary in size and number and Collins asserts that in addition to receiving drainage from the kidney and adrenal, the renal lymph nodes accept drainage from the peritoneum and certain visceral organs. This view is not shared by Thornton or Sisson.

4.6. Subiliac lymph nodes

In Nomina Anatomica Veterinaria these lymph nodes are described as *Lnn. subiliaci* as part of the lymphocentrum inguinofemorale.

The subiliac lymph node (also known as the precrural or prefemoral lymph node) is embedded in fat in the conspicuous fold of the flank below the stifle joint in the hanging carcass. It is situated on the aponeurosis of the obliquus abdominis externus at the anterior border of the tensor fascia lata and about 125 mm above the patella. It has an elongated elliptical outline and is flattened. Its average length is from 8 to 10 cm and its width around 25 mm in the adult bovine. It may be incised from either side of the fold but is usually approached through the edge of the tensor fascia lata by an incision made about 175 mm down from the apex of this muscle.

It receives afferent vessels from the skin of the posterior part of the thorax, the abdomen, pelvis, thigh and leg and also from the tensor fascia lata and prepuce. The efferent vessels ascend on the deep face of the tensor fascia lata and end chiefly in the deep inguinal lymph node, but in some animals go directly to the internal iliac lymph nodes, although some vessels may convey lymph to the external iliac lymph node

4.7. Superficial cervical lymph nodes

In Nomina Anatomica Veterinaria these lymph nodes are described as *Lnn. cervicales superficiales* as part of the lymphocentrum cervicale superficiale which also includes *Lnn. cervicales superficiales accessorii*.

The superficial cervical lymph node (also known as the prescapular lymph node) is situated at the anterior border of the supraspinatus muscle, a little above the level of the shoulder joint. It is covered by the omo-transversarius and brachiocephalicus muscles. It is elongated in shape and may obtain a length of 10 to 12 cm and a width of up to 3 cm. Its deep face has a long and distinct hilus. According to Collins, it receives from 150 to 200 afferents altogether from the skin of the neck, shoulder, part of the ventral and lateral surfaces of the thorax, and the thoracic limb (from the poll to a vertical line passing through the back of the elbow in the standing animal), from the muscles of the shoulder girdle, and from the external scapular muscles, the tendons of the muscles of the forelimb, the fascia of the forelimb and from the joints of the carpus and digits. The efferent vessel descends over the scalenus muscle and opens on the right side into the end of the right tracheal duct, and on the left side into the terminal part of the thoracic duct or left tracheal duct.

4.8. Superficial inguinal and supramammary lymph nodes

In Nomina Anatomica Veterinaria these lymph nodes are individually described as *Lnn. scrotales* and *Lnn. mammarii*, but collectively known as *Lnn. inguinales superficiales* as part of the lymphocentrum inguinofemorale.

The *superficial inguinal lymph node* is positioned in the male bovine below the prepubic tendon and in the narrow interfemoral space. It lies in the mass of fat about the neck of the scrotum and behind the spermatic cord, and is covered in part by the retractor of the prepuce. It is usual to find one or two on each side of the penis. The afferent vessels come from the external genital organs, except for the testicles, the skin of the adjacent region, the medial and posterior surface of the thigh, and the medial surface of the leg. The efferent vessels ascend through the inguinal canal to the deep inguinal lymph node at the side of the pelvic inlet and from there to the internal iliac lymph node.

The *supramammary lymph nodes* lie above and behind the udder and there are usually two present on each side. The larger nodes are in apposition medially and are sometimes united while the smaller nodes are above or in front of the large ones. The larger lymph nodes are often 6 to 10 cm in length with the smaller ones from a quarter to a half as large. The alignment of the two nodes changes from the heifer to the aged cow.

The afferent vessels come from the udder, the external genital organs, and part of the skin of the thigh and leg while the efferent vessels converge to two or three large trunks which go to the deep inguinal glands at the side of the pelvic inlet.

5. Identification of hazards, suitability characteristics and associated abnormalities

5.1. Biological hazards

5.1.1. Association with grossly-identifiable abnormalities

Biological hazards and their association with the likelihood of grossly-identifiable abnormalities are shown in Table 3

Some microorganisms, previously described as potential foodborne hazards in the scientific literature, were included in the listing. However because of strong contemporary evidence that there is a negligible risk of transmission via the selected lymph nodes of cattle, or lack of scientific consensus as to their zoonotic potential, some were deemed a negligible health risk. *Mycobacterium bovis, Mycobacterium avium* subsp. p*aratuberculosis* and *Fasciola hepatica* are in this category and a scientific justification for their exclusion is presented in 5.1.2.

Table 3: Biological hazards and their likely association with grossly-detectable abnormalities in lymph nodes of cattle slaughtered in New Zealand

Hazard	Possible presence in lymph nodes	Possible grossly detectable abnormality
Salmonella spp	+	+
E.coli STEC	+	-
Campylobacter spp	+	-
Yersinia spp	+	-
Listeria monocytogenes	+	-
Mycobacterium bovis	+	NR
Mycobacterium avium subsp. *paratuberculosis*	+	NR
Clostridium spp	+	-
Cryptosporidium parvum	+	-
Fasciola hepatica	-	NR
Taenia saginata	-	-
Toxoplasma gondii	+	+

NR- deemed to constitute a negligible public health risk

5.1.2. Biological hazards potentially present in selected lymph nodes of cattle categorised as negligible risk

Mycobacterium bovis

M. bovis is a potential food borne zoonosis and unpasteurised milk from infected cows presents a risk to public health (Cressey *et al*., 2006) However, the likelihood of this zoonosis being transmitted by meat from cattle slaughtered in New Zealand is negligible. Clinical cases do not occur due to the very advanced state of the tuberculosis surveillance programme in New Zealand and therefore the possibility of bacteraemic cattle or contaminated milk in slaughter cattle is extremely low. Further, there is an extremely low number of cattle confirmed as infected with *M. bovis* at slaughter in New Zealand (less than 80 out of a 2.4 million cattle slaughter for 2010, P. Livingstone, pers. comm.) and a recent MAF report (Anon. b, 2011; Appendix 3) details the extremely low likelihood of *M. bovis* being present in the selected lymph nodes.

***Mycobacterium avium* subsp. *paratuberculosis* (MAP)**

As yet there is no scientific consensus as to whether MAP should be regarded as a zoonosis. An association between Johne’s disease in animals and Crohn’s disease in humans has been established but causality has yet to be shown (Ryan and Campbell, 2006; Appendix 4). Notwithstanding this, Johne’s disease is regarded as a significant disease of cattle and that post mortem examination may have a role to play in the detection and control of the disease.

Fasciola hepatica

The possibility of the meatborne zoonotic potential of *F. hepatica* has been suggested based on the successful oral inoculation of mice and piglets with metacercariae and immature flukes (Taira, 1991) Although the results of the experiments suggest that humans consuming raw dishes, such as pâté, prepared from raw fresh bovine livers infected with immature *Fasciola* spp. could become infected with liver fluke, the most common food borne transmission route of this parasite to humans is via the ingestion of watercress contaminated with encysted cercariae (Slifko, 2000). Irrespective of epidemiological considerations, the presence of metacercariae in lymph nodes would not be detected by routine gross examination.

5.2. Chemical hazards

Chemical hazards may be present in the carcasses and offal of slaughtered animals but are highly unlikely to be detected by organoleptic examination of lymph nodes. There have been reports of reactive lymph nodes in response to an irritant injection (Montgomery, 1997).

5.3. Physical hazards

Physical hazards that can be detected in the carcass include (i) broken needles, or fragments of needles, that are used for veterinary purposes and (ii) fragments of bullets, pellets or similar projectiles that are used for hunting (Horchner et al., 2006) However, apart from possible associated infection that may extend to a regional lymph node, these physical hazards will not be present in lymph nodes

5.4. Suitability characteristics

Suitability characteristics that may be detected by routine post mortem examination of lymph nodes are listed in Table 4. Grossly detectable abnormalities may be restricted to a single node or may involve multiple nodes.

Table 4: Suitability characteristics detectable by post mortem examination of lymph nodes in cattle slaughtered in New Zealand

Non-specific abscessation
Caseation
Eosinophilic infiltration
Haemorrhage
Lymphadenopathy
Lymphadenitis
Necrosis
Neoplasms - benign
Parasitic infestation
Pigmentation
Reaction to irritant injections

5.4.1. Abscessation

Abscessation from distal limb infection may progress to regional lymph node abnormalities with reactive lymph nodes being the normal response to the presence of antigen. Infectious agents that may give rise to subcutaneous, muscular and mammary abscessation in cattle include *Arcanobacterium* (syn. *Actinomyces* or *Corynebacterium*) *pyogenes, Actinobacillus lignieresi*, *Fusobacterium necrophorum*, *Pseudomonas* spp., and *Staphylococcus* spp. and of these, only *A. pyogenes* is considered potentially zoonotic (Goldsmid, 2005). However, human infections with this organism are believed to be through close association with animals rather than food borne (Ramos *et al*., 2009).

Mastitis in New Zealand cattle is generally caused by *Staphylococcus* spp., *Streptococcus* spp. and some gram-negative species and can lead to localised involvement of the supramammary lymph nodes. *Staphylococcus aureus* is not regarded as a zoonosis (Goldsmid, 2005) and although one zoonotic species from this genus, *S. intermedius,* has been isolated from cow's milk (Chaffer *et al.*, 1998), the significance of this organism as a foodborne zoonosis has not been established and the risk to consumers is more closely correlated to improper food handling and storage enabling growth-related toxin production.

Bacillus cereus can occasionally cause severe necrotising mastitis (Parkinson *et al*., 1999). *Clostridium perfringens* has occasionally been isolated from cows with severe necrotising mastitis while *B. licheniformis* is another rare cause of mastitis (Graham, 1998). Any cow with severe necrotising mastitis will not be presented for slaughter.

5.4.2. Other suitability characteristics

Other suitability characteristics that may be detected by routine post mortem inspection of lymph nodes are described in Section 7.

6. General evaluation of hazards and grossly detectable abnormalities in lymph nodes of slaughtered cattle

Hazards associated with grossly detectable abnormalities in the lymph nodes of slaughtered cattle are limited to systemic or generalised conditions[1].

6.1. *Salmonella* species

There have been a number of highly publicised salmonellosis outbreaks in humans following ingestion of contaminated food (Lynch, 2006) and among the outbreaks, for which the aetiology was determined, bacterial pathogens caused the largest percentage (55%) and of the bacterial pathogens, *Salmonella* serotype Enteritidis accounted for the largest number of outbreaks and outbreak-related cases. Within New Zealand, recent studies have indicated the predominance of food pathways as a source of infection and disease is for serovar (*S.* Typhimurium DT156) (French *et al*., 2011).

Salmonella has been reported in New Zealand cattle since 1948 (Josland, 1950). In 1970 the predominant *Salmonella* serovars in New Zealand cattle were reviewed (Robinson, 1970) and found to be *S.* Typhimurium and *S.* Bovismorbificans*,* which were considered important and common causes of enteritis in New Zealand cattle. Since then, many serovars have been isolated from cattle including; Agona, Albany, Anatum, Bovismorbificans, Brandenburg, Bredeney, Choleraesuis, Enteritis, Hadar, Havana, Hafia, Heidelberg, Hindmarsh, Infantis, Johannesburg, Kentucky, Kiambu, Lexington, London, Orion, Saintpaul, Senftenberg, Typhimurium and Uganda (Clark *et al*., 2002). Many serovars have also been isolated from humans although the epidemiological links between human foodborne outbreaks and serovars present in domestic animals have been difficult to quantify. The passage of *Salmonella* from enteric organisms to being detected within mesenteric lymph nodes in cattle without clinical symptoms or gross pathology has long been recognised (Moo *et al*., 1980; Samuel *et al*., 1981) along with the potential for these lymph nodes to be a potential reservoir of Salmonella that could be transferred to meat and meat products.

S. Brandenburg has become an occupational hazard for farmers and veterinarians but has not yet been established as a foodborne pathogen, although the potential exists. Zoonotic pathogenic bacteria, such as *S. enterica* are known to be present in the lower gastro-intestinal tract of cattle and cause human illnesses through carcass contamination (Dowd *et al.,* 2008).

Animals in the septicaemic phase of salmonellosis, in the unlikely situation of being presented for slaughter, will have pathological evidence of infection throughout the carcass and viscera. Most carcass lymph nodes will be reactive along with petechial haemorrhages throughout the intestinal tract, especially the ileum. The gall bladder is likely to be inflamed and thickened with an enlarged liver. The mesenteric lymph nodes are usually enlarged and oedematous, the heart frequently has small haemorrhages under the epicardium and there may be blood stained fluid within the pericardium and peritoneal cavity.

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[1] Localised abscessation not caused by zoonotic micro-organisms, see Table 4

Gross pathology associated with salmonellosis is typical of acute muco-necrotic enteritis and occurs especially in the ileum and large intestine. The disease is more commonly seen in calves than adult cattle. The intestinal wall may be thickened, oedematous and covered with yellow-grey necrotic material overlying a hyperaemic, granular surface. The spleen may be enlarged and congested, soon replaced by an acute splenitis with miliary foci of necrosis or reactive nodules (Jubb *et al*., 1993) while the mesenteric lymph nodes are enlarged and oedematous (Wray and Wray, 2000;Thompson, 2004). The liver is often pale and presents with many pin head sized necrotic foci while in chronic cases of salmonellosis, there is almost always an anterior bronchopneumonia, usually with adhesions and small abscesses (Jubb *et al*., 1993). The organism has been isolated from the spleen, liver, lungs and thoracic lymph nodes after inoculation of cattle with *S.* Dublin (Hall and Jones, 1977) and it would seem likely that enteric infections would precede infiltration of the mesenteric lymph nodes by this organism.

The National Microbiological Database (NMD) has been compiled in New Zealand since 1997 and shows the prevalence of *Salmonella* detected in beef to be extremely low, at 0.01% of sampled carcasses. The prevalence of septicaemic-like conditions **of all causes** detected by meat inspectors in New Zealand cattle is around 0.04% over the last decade (AsureQuality database)

It is clear that routine post mortem examination of the carcass and offal of slaughter cattle is important to detect systemic / generalised salmonellosis. Such cases are extremely rare and are much more likely to be identified at ante mortem cf. post mortem inspection. However, if such an animal is presented for post mortem inspection, the gross signs are readily apparent in many organs and tissues as well as the lymph nodes and are fully evident by viewing. Viewing of the mesenteric lymph nodes may provide a contribution to overall judgment of generalised / systemic salmonellosis.

6.2. Pathogenic *Escherichia coli* (STEC)

Escherichia coli species are part of the normal gut microflora in healthy animals although some cattle are known to carry more pathogenic strains such as *E. coli* O157:H7 without exhibiting any clinical signs. The bacterial and host factors influencing colonisation and pathogenicity in cattle and humans remain poorly defined and, although human infections may arise through a variety of routes; since the organism has been frequently isolated from cattle, this species is considered a primary reservoir of infection.

Ruminants are also believed to be the major reservoir of *E. coli* O157:H7 in New Zealand (Bunčić and Avery, 1997; Cookson *et al*., 2006), but very little information is available about the epidemiology of carriage of STEC in animal reservoirs in this country (French *et al*., 2011). Recent genotyping studies have indicated that only a clinical subset of STEC 0157:H7 is important for human infection and that while clinical types 1 and 3 were predominant in both human cases and ground beef, types 5 and 6 were common in cattle faeces but relatively uncommon in human cases and ground beef (Whitworth *et al*., 2008).

E. coli O157:H7 has been isolated from the mesenteric lymph nodes from a position close to the anterior root of the mesentery, the middle of the small intestine, and the ileal caecal colic node with no gross pathological lesions (Cray and Moon, 1995).

As there is a lack of gross pathology associated with this disease in cattle, routine examination of lymph nodes will not mitigate foodborne risks.

6.3. *Campylobacter* species

Campylobacter species are commonly reported as zoonoses with *C. jejuni* the most common cause of human foodborne illness (Anon.a, 2010). These organisms generally cause subclinical intestinal infections in a variety of ruminants, and human infection may occur by oral transmission with an infective dose that is relatively low. The organism has been reported as being isolated from the mesenteric lymph nodes from apparently health cattle (Garcia *et al.,* 1985).

Human infection in New Zealand has shown an association with dairy cattle, however spatial and temporal patterns suggest that many rural cases are associated with environmental rather than food pathways (French *et al*., 2011). Although consumption of poultry is considered the major contributor to human infection by *Campylobacter* species, current attribution studies (Anon.a, 2010) are evaluating other potential exposure pathways for humans.

As there is a lack of gross pathology associated with this disease in cattle, routine examination of lymph nodes will not mitigate any foodborne risks.

6.4. *Yersinia* species

Yersinia species are ubiquitous organisms known to be isolated from soil, water and the gastrointestinal tracts of many animals. The species most associated with disease in humans is *Y. enterocolitica,* a heterogeneous species that is divided phenotypically into five biotypes and numerous serotypes; many of which are considered environmental and non pathogenic. Clinical disease of animals by *Yersinia* spp. is generally in young animals and almost all adult animals from which the organism can be isolated will be asymptomatic.

An early European study isolated *Y. enterocolitica* from the mesenteric lymph nodes of cattle but many more from faecal samples (Wauters *et al*., 1971) A later New Zealand study demonstrated that *Y*. *pseudotuberculosis* was commonly found in the faeces of clinically healthy cattle (Hodges, 1985) although a subsequent study carried out to investigate the presence of *Yersinia* species in the rectal contents of adult cattle in New Zealand (Bullians, 1987) was not able to isolate *Y. enterocolitica*. Only 2 out of 330 cull cows were positive for other Yersinia species (*Y. pseudotuberculosis* and *Y. intermedia*). Fenwick (1997) isolated potentially pathogenic strains of *Y. enterocolitica* from asymptomatic animals, which reinforces the importance of preventing faecal contamination of product during dressing.

There is a lack of gross pathology associated with lymph nodes of cattle and routine examination of lymph nodes will not mitigate any possibility of foodborne risks.

6.5. Listeria monocytogenes

L. *monocytogenes* is responsible for severe foodborne infections in humans and can also cause invasive disease in many different animal species, including cattle. In ruminants, *L. monocytogenes* primarily causes meningoencephalitis and uterine infections (Nightingale *et al.,* 2004) and the uterine infections are characterized by late-term abortions or septicaemia in neonates, while the meningoencephalitic form of animal listeriosis is characterized by neurological signs, including circling, excessive salivation, and unilateral facial paralysis (Vermunt, 2000). An acute septicaemic form of listeriosis can cause anorexia, emaciation and diarrhoea in animals; however these cases are highly unlikely to be presented for slaughter.

In contrast with *Salmonella* and *E. coli* O157:H7 infections, raw animal-derived food products are considered rarely a direct source of human listeriosis (Nightingale *et al.,* 2004).

L. monocytogenes has been isolated from the retropharyngeal and supramammary lymph nodes in cattle (Bunčić, 1991, Winter *et al*., 2004).

There is a lack of gross pathology associated with lymph nodes of cattle and routine examination of lymph nodes will not mitigate any possibility of foodborne risks.

6.6. *Clostridium species*

Clostridium difficile can be found in the intestinal tracts of a variety of animal species, including food animals, such as cattle and pigs (Weese *et al*., 2009) but without any accompanying clinical signs. The same study demonstrated the presence of *C. difficile* in retail meat, and that it is an important cause of enteric disease in humans. Other *Clostridia* that have been isolated from the bovine intestinal tract contents include *C. perfringens, C. tetani,* and *C. botulinum* while *C.perfringens* has been isolated from the caecal lymph nodes of cattle in Australia (Moo *et al*., 1980). In one study, commensal species of *Clostridia* made up 20% of the gut microflora in healthy cattle (Dowd *et al*., 2008) and were the most common and diverse species identified.

C. perfringens is the predominant cause of gas gangrene-like traumatic and wound infection, especially when localised in muscle tissue. It is also believed to play an auxiliary role in enteric conditions (Niilo, 1980).

As carriage of Clostridia spp. in cattle is asymptomatic, routine examination of lymph nodes has no value in mitigating any potential foodborne risks

6.7. *Toxoplasma gondii*

Toxoplasmosis is a zoonosis that occurs in livestock, although infection appears to be rare in cattle (Hall *et al*., 2001). Older serological tests to detect *T. gondii* antibodies in bovines gave false positives and greatly overestimated prevalence. Cattle are also not believed to remain persistently infected as long as do sheep, and epidemiological studies point to consumption of raw or undercooked mutton and pork as the most important risk factors for human transmission during pregnancy (Kapperud *et al*., 1996; Tenter *et al*., 2000). The risk of toxoplasmosis as a foodborne zoonosis of meat or meat products from cattle is unknown.

Dissemination of *Toxoplasma* within the host occurs in lymphocytes, macrophages, granulocytes, and as free forms in plasma. From the intestine the organism may either spread via the lymph to regional nodes and from there to the bloodstream, or by portal circulation to the liver and from there to the systemic circulation (Jubb *et al*., 1993)

Lesions in lymph nodes are often associated with pathology of the corresponding organ. They are characterised by irregular areas of coagulative necrosis, mainly in the cortex and a moderate inflammatory reaction may be evident at the periphery of the necrotic areas (Jubb *et al*., 1993).However, such pathological changes are unremarkable when lymph nodes are grossly examined; are not prominent visually and are unlikely to be seen by routine post mortem examination.

Due to the limited and unremarkable pathology associated with occasional infection of cattle with Toxoplasma spp., routine post mortem inspection of lymph nodes is highly unlikely to identify infected animals. Further, the public health benefit of identification of such infections is unknown.

6.8. *Cryptosporidium parvum*

C. parvum is the only zoonotic species of *Cryptosporidia* and is a common enteropathogen of cattle globally. It is generally associated with diarrhoea in calves of less than one month of age, and is most often found in the distal small intestine accompanied by atrophy and fusion of villi, and metaplasia of the surface epithelium (Heine *et al*., 1984). It is rarely found in adult cattle and if so, clinical signs are mild.

Consumption of meat as a vehicle for foodborne infection by *Cryptosporidia* of humans has not been identified (Smith *et al*., 2007) with contaminated water and poor food handling practices considered to contribute most to human infection.

While in extremely rare cases of C.parvum may be present in the mesenteric lymph nodes of adult cattle, routine examination of lymph nodes will not mitigate any foodborne risks.

7. General evaluation of suitability characteristics in lymph nodes of slaughtered cattle

7.1. Pathology and other conditions

7.1.1 Lymphadenitis

Lymphadenitis results when an infectious agent is present in the lymph node which may be the result of drainage to the node of products from the distant inflammatory process, which then progresses to involve the lymph node directly. The condition may be acute or chronic, suppurative, caseous or granulomatous.

Post mortem examination of regional lymph nodes may provide an indicator function for an infectious condition within the area of drainage that is not grossly evident on initial post mortem examination but this is considered to be a rare occurrence.

7.1.2. Abscessation

Abscessation from distal limb inoculation and infection may progress to regional lymph node abnormalities with reactive lymph nodes being the normal initial response to the presence of micro-organisms. Any haematogenous borne pathogen may accumulate in lymph nodes and organisms that have been isolated from carcass and visceral lymph nodes in cattle include *Actinomyces bovis*, *Actinobacillus lignieresi*, *Arcanobacter pyogenes*, *Pseudomonas aeruginosa*, *Staphylococcus aureus*, *M. bovis* and *M. avium subsp. paratuberculosis (*Momotani *et al.*, 1993)

Post mortem examination of regional lymph nodes may provide an indicator function for a suppurative condition within the area of drainage that is not grossly evident on initial post mortem examination but this is considered to be a rare occurrence

Arcanobacterium (syn.Corynebacterium, Actinomyces) pseudotuberculosis has occasionally been recorded as causing cutaneous abscessation in dairy cows which led to regional lymph node involvement (Yeruham *et al*., 1997).The condition did not progress to a generalised lymphadenitis and lymphangitis was not observed. Other pathogenic bacteria isolated from cutaneous abscesses in this report included *A. pyogenes*, *Staphylococcus aureus* and *Micrococcus* species.

Mannheimia granulomatis has been isolated from milk of a cow with subclinical mastitis and with no gross pathology (Blum *et al*., 2010), and it has been suggested by others that this organism could be responsible for the previously described “actino” lesions of the tongue and oral cavity in Australian cattle (Blackall *et al*., 2002). It is not regarded as a zoonosis.

Actinobacillis lignieresii is regarded as a commensal of the oropharynx of cattle (Kokotovic *et al*., 2011) but occasionally inadvertent inoculation of the oral cavity and/or tongue gives rise to discrete granulomatous lesions, known as woody tongue. Very rarely, cutaneous abscesses in other areas have been reported, such as on the lower hind limb (Aslani *et al*., 1995) These lesions are generally restricted to the tongue although rarely a primary infection of the oral cavity and tongue may give rise to involvement of the efferent lymph nodes, in particular the parotid, retropharyngeal and bronchial lymph nodes (Hungerford, 1975; Jubb *et al*., 1993).

The condition of "woody tongue" is identified from the primary lesion and gross pathology in the lymph nodes of the head will be secondary to this detection

A review of lesions encountered by meat inspectors that resembled bovine tuberculosis in New Zealand (Montgomery, 1997) considered that lesions caused by *Rhodococcus equi* could be confused with those of bovine tuberculosis. *R. equi* is a common inhabitant of the gastrointestinal tract but occasionally produces lesions in lymph nodes, in particular the mediastinal lymph nodes. Lesions vary from abscesses to calcified caseous lesions with epithelioid cells and Langhans cells as found in tubercular lesions. These lesions are distinguished from those caused by *M .bovis* by culture.

Johne's disease occurs in New Zealand cattle and infection by *M. avium subsp. paratuberculosis* is characterised by chronic, granulomatous degenerative enteritis that causes intermittent but persistent diarrhoea, progressive weight loss, and eventual death. Advanced cases are unlikely to be presented for slaughter due to their very poor condition. Animals presented for slaughter with long standing Johne's disease are often detected by examination of the gastrointestinal tract, mesentery and mesenteric lymph nodes with thickening and corrugation of the jejunal wall a feature of this disease (Thompson, 2004). In less advanced cases, lesions are generally confined to small areas of the intestine and individual lymph nodes and may only be diagnosed by histopathology (Worthington, 2004). Although in some scientific quarters there is support for the treatment of this organism as a zoonosis due to its association with Crohn's disease, no causation has been established (Ryan and Campbell, 2006). It has not been established as a foodborne zoonosis.

Detection of Johne's disease at post mortem examination is primarily due to visible changes in the intestine, spreading to the mesentery and lymph nodes. Viewing of the mesenteric lymph nodes may provide some value in detecting this infection.

Routine organoleptic examination of readily available carcass and mesenteric lymph nodes of slaughtered cattle has been routinely employed to detect *M. bovis* as caseation and calcification were common features of those lesions with liquefaction observed less frequently (Collins, 1970). Aggregations of tubercles may result in the formation of large masses of tuberculous tissue that are frequently a distinctive golden yellow colour.

Pathological changes in lymph nodes that may result from infection with M. bovis in cattle in New Zealand are not considered to constitute a food-borne public health risk.

7.1.3. Lymphadenopathy

Lymphadenopathy is defined as a regional or generalised enlargement of lymph nodes of unknown or unspecified cause (Jubb *et al*., 1993) and local enlargement usually reflects pathology limited to the drainage area, particularly inflammatory or neoplastic disease. The essential architecture of the node is mostly preserved. Generalised enlargement of lymph nodes may be the result of a primary tumour such as lymphoma, or an infection such as malignant catarrhal fever which has animal health importance only.

7.1.4. Neoplasms

Squamous cell carcinoma is the most common neoplasm detected in New Zealand cattle and the primary sites are poorly pigmented mucocutaneous junctions of the body; usually adjacent to the eye and very rarely, the vulva. The gross appearance depends not only on the anatomical tumour site, but also the stage of malignancy. In general, pre-malignant lesions are small, greyish-white, elevated, hyperplastic plaques or papilloma-like structures. Malignant tumours are more irregular, nodular, pink, erosive, and necrotic in nature and such tumours may metastasize along draining lymphatics to regional lymph nodes such as the parotid, retropharyngeal and submaxillary lymph nodes in cases of ocular squamous cell carcinoma (Vermunt, 2002) and the internal iliac lymph node in cases of squamous cell carcinoma of the vulva. The prevalence of neoplasms detected at post mortem examination of cattle in New Zealand averaged just under 0.08% in 2010 (AsureQuality data base). The largest proportion of these neoplasms are squamous cell carcinoma and if there is any invasion of the surrounding bony orbit or metastatic spread to lymph nodes, the carcass and all parts are condemned (around 51% of such cases in 2010, AsureQuality database) on the grounds of suitability.

Lymphosarcoma is detected mainly in young cattle of less than two year of age (Shortridge and Cordes, 1971) and predominantly occurs as a single case within a herd. The thymic and multicentric forms are the most common. The initial lesion in a lymph node may be large with the capsule usually permeated and obliterated with infiltration to adjacent tissue (Jubb *et al*., 1993). The condition is generally extensive and obvious upon observation. Unlike adenocarcinoma in sheep, this neoplasm in cattle has not been associated with the small intestine. It is known to be associated with bovine leukaemia virus and transmission requires prolonged and close contact with infected animals or the inoculation of infected lymphocytes. Prevailing husbandry practices and lack of widespread winter housing are likely to account for the current low prevalence of this disease in New Zealand.

Mesotheliomas are rare tumours that arise from cells of the serous linings of the pericardial, pleural and peritoneal cavities, frequently involving all these locations. The tumour occurs most commonly as a congenital neoplasm in foetal or young cattle, although there are a few reports of acquired mesothelioma in adult cattle (Girard and Cécyre, 1995). These cases have included metastatic involvement with the tracheobronchial lymph node.

Small intestinal carcinomas has very rarely been reported in New Zealand cattle (Johnstone *et al*., 1983) and gross pathology included thickening and proliferation of the mucosa to form nodular or polypoid masses, which were often associated with areas ulceration and necrosis. Where partial obstruction to the flow of ingesta was present, the thickness of the intestine was increased by dilatation or muscular hypertrophy. The mesenteries and omentum were thickened and nodular while the mesenteric lymph nodes were swollen and nodular. Organs that contained focal nodular metastatic lesions included the liver, kidneys, lungs, ovaries and uterus.

Urinary bladder tumours are rarely reported in New Zealand but in some countries are commonly associated with the grazing of bracken fern by cattle (Pinto *et al*., 2000) and may constitute a major cause for carcass condemnation. Some neoplasms of the alimentary tract of cattle are believed to be caused by an interaction of the bracken fern carcinogen with the bovine papilloma virus (Jarret *et al*., 1978).

Cutaneous lymphomas have been reported in cattle and all cases have progressed to multiple lymphadenopathy (Schweizer *et al*., 2003) with detectable lesions in regional lymph nodes. This condition should be readily apparent by visual examination.

Post mortem examination of lymph nodes may assist in identification of neoplasms in slaughter cattle. e.g. lymphadenopathy (defined as a regional or generalised enlargement of lymph nodes of unknown or unspecified cause) and lymphosarcoma in young animals.

Squamous cell carcinoma are not primarily identified via lymph node examination, but further examination of lymph nodes may assist in determining the extent of metastasis

7.1.5. Pigmentation

A number of conditions that give rise to abnormal pigmentation of the carcass and viscera of cattle, such as icterus, lipofuscinosis, melanosis, ochronosis and xanthosis but very few affect the lymph nodes and none are restricted to lymph nodes. Dark brown pigments from obstructive jaundice in cases of sever fascioliasis have been reported as producing discoloured hepatic lymph nodes but always with concurrent pathology in the liver.

7.1.6. Haemorrhage

Lymph nodes that exhibit haemorrhages if accompanied by other systemic gross pathology such as generalised lymphadenitis may indicate infection by organisms such as *Pasteurella multocida.*

However most incidental haemorrhage seen post mortem in lymph nodes is post agonal in origin and of no consequence to the consumer.

7.1.7. Reaction to irritant injections

In New Zealand cattle, instances of these were restricted to early vaccines developed against Johne's disease in which attenuated organisms were suspended in an oily adjuvant. Initially in some animals, injections of the vaccine gave rise to a lesion that had most of the features of a classical tubercule, with caseation, central calcification, peripheral epithelioid cells, Langhans giant cells and often a zone of lymphocytes. These lesions were not within lymph nodes and generally confined to the neck musculature.

These initial outcomes were reduced by changing both the oil used as an adjuvant and the site of vaccination from the anterior neck to the brisket to reduce any confusion between these lesions and tubercular lymph nodes (Montgomery, 1997).

8. Evaluation of hazards and associated abnormalities in selected lymph nodes of cattle slaughtered in New Zealand

8.1. Atlantal lymph nodes

8.1.1. Food safety

These nodes may be involved in generalised or systemic conditions but are highly unlikely to provide any additional information to that seen in more prominent groups of lymph nodes throughout the carcass and viscera.

There are no specific food safety abnormalities in the atlantal lymph nodes and no specific indicator functions.

8.1.2. Food suitability

The atlantal nodes may be affected by a range of suitability abnormalities. However, their presence is likely to be rare and the sensitivity of any post mortem inspection procedure is likely to be highly variable. Further, the anatomy and position of the atlantal lymph nodes are such that the submaxillary (syn. mandibular), parotid and medial retropharyngeal lymph nodes are likely to have a much higher level of abnormalities from oral inoculation and buccal cavity infection.

There is no specific indicator function of the atlantal nodes for suitability characteristics involving other tissues.

In the case of squamous cell carcinoma of the head or tongue, further examination of the head nodes may assist in determining extent of metastasis. Metastatic spread of ocular squamous cell carcinoma from the eye and surrounding tissue is usually found in the parotid lymph node (Jubb *et al.*, 1993), with fewer metastases in the submaxillary, medial retropharyngeal lymph nodes or atlantal lymph nodes.

8.1.3. Animal health surveillance

Examination of the atlantal lymph nodes has negligible usefulness in the bovine tuberculosis eradication programme. The recent study of distribution of grossly-detectable lesions in slaughter cattle in New Zealand (Anon.b, 2011) showed that around one third had lesions in the head lymph nodes with 28% of these occurring in the medial retropharyngeal lymph nodes. Of the animals with only one lesion, only 0.69% of cases involved the atlantal node. The study concluded that the sensitivity of examination for bovine tuberculosis would still be 99.78% that of the current system if examination of these lymph nodes was removed.

8.1.4. Other considerations

Differences in anatomical nomenclature and position create difficulties when interpreting the intent and application of post mortem examination rules in different countries. Because of a lack of common understanding on anatomical location, as well as biological variability in anatomy, it is likely that actual implementation of procedures is highly variable.

Consistent routine examination of the atlantal lymph nodes has anecdotally been difficult to achieve. After the head has been removed from the carcass during dressing, these lymph nodes are occasionally left on the head, or more commonly either transected with parts attached to both head and neck, or left intact at the distal end of the neck. The range of outcomes is affected by variation in both anatomical location of the lymph nodes and disarticulation techniques by slaughtermen. To ensure consistent and intensive examination of these lymph nodes requires diligence and communication between the head and carcass examiners for each carcass. This required level of carcass by carcass communication is not facilitated by many modern beef slaughter establishments where there is substantial separation between the head and carcass chain, which has given rise to variable implementation of this specific examination procedure.

It is noted that examination of these lymph nodes by incision is not required on high speed lines by one overseas competent authority (Canada).

8.1.5. Scientific evaluation

Routine examination of the atlantal lymph nodes in cattle slaughtered in New Zealand has no value for food safety, food suitability or animal health surveillance.

8.2. Internal iliac lymph nodes

8.2.1. Food safety

These nodes may be involved in generalised or systemic conditions but are unlikely to provide any additional information to that seen in more prominent groups of lymph nodes throughout the carcass and viscera.

There are no specific food safety abnormalities affecting the internal iliac lymph nodes and there are no specific indicator functions. However, due to their location, the internal iliac lymph nodes may have some indicator function for conditions that have occurred distally in the hind limbs, with a unilateral reactive change indicating an infection in one limb. For systemic conditions both lymph nodes are likely to show similar gross pathological change. Compared to more peripheral lymph nodes (such as the popliteal lymph nodes) the internal iliac lymph nodes may have a greater indicator function for localised conditions being strategically placed between the hind limb, the popliteal, ischiatic, superficial inguinal, supramammary, deep inguinal, external iliac and subiliac lymph nodes; and the distal end of the lumbar lymphatic trunk. Any enlargement of these lymph nodes may be a result of infection in the afferent limb, such as arthritis, abscessation or conditions of the feet, and indicate the need for further examination. Any noticeable difference between the gross appearances of the two lymph nodes may be investigated further, as should any bilateral abnormality.

8.2.2. Food suitability

The internal iliac nodes may be affected by a range of suitability abnormalities. However, their presence is likely to be rare and the sensitivity of any post mortem inspection procedure is likely to be highly variable.

There is no specific indicator function for suitability characteristics involving other tissues. The general comments on a potential indicator function for hazards as above apply.

Metastatic spread of squamous cell carcinoma from the poorly pigmented muco-cutaneous junctions adjacent to the vulva may occur in the internal iliac lymph node. Compared to ocular squamous cell carcinoma, this site is very rarely affected and consequently metastases are extremely infrequent in efferent lymph nodes. Scientific research (Burdin, 1964) into this condition has indicated that cattle characterised by unpigmented areas of skin over the vulva, such as the Ayrshire breed, were more prone to this particular neoplasm than cattle with pigmented skin. Ayrshires are uncommon dairy cattle in New Zealand (around 2%).

8.2.3. Animal health surveillance

Examination of the internal iliac lymph nodes has negligible usefulness in the bovine tuberculosis eradication programme. The recent study on distribution of grossly-detectable lesions in slaughter cattle in New Zealand (Anon. b, 2011) showed that only 0.09% of submissions were from the internal iliac lymph nodes. Of those animals with only one lesion, only 0.06% came from these lymph nodes.

8.2.4. Other considerations

There is significant divergence between countries on examination requirements for this lymph node, with some recommending viewing and some recommending incision..

Initial field studies indicate that if routine examination is carried out, incision may be more practical than physical exteriorisation of the lymph nodes from fatty tissue for viewing.

8.2.5. Scientific evaluation

Field studies are needed to further evaluate a potential indicator function of the internal iliac lymph nodes for food safety and/or food suitability conditions of the hind limb and other areas of the carcass.

8.3. Lumbar chain lymph nodes

8.3.1. Food safety

These nodes may be involved in generalised or systemic conditions but are highly unlikely to provide any additional information to that seen in more prominent groups of lymph nodes throughout the carcass and viscera. Note that *Salmonella* infection is primarily enteric in location and since this chain of lymph nodes does not receive efferent vessels from the gastrointestinal tract, it is highly unlikely that they will have any indicator of enteric infection.

There are no specific food safety abnormalities in the lumbar lymph nodes and no specific indicator function. Infections of the peritoneum may lead to reactive lumbar chain lymph nodes but evidence of this will be apparent through observation of the peritoneum on the carcass and viscera.

8.3.2. Food suitability

The lumbar nodes may be affected by a range of suitability abnormalities. However, their presence is likely to be rare and the sensitivity of any post mortem inspection procedure is likely to be highly variable.

There is no specific indicator function of the lumbar lymph nodes for suitability characteristics involving other tissues.

8.3.3. Animal health surveillance

Examination of the lumbar lymph nodes has no usefulness in bovine tuberculosis surveillance. Not one submission has been made from these nodes in the New Zealand programme

8.3.4. Scientific evaluation

Routine examination of the lumbar lymph nodes in cattle slaughtered in New Zealand has no value for food safety, food suitability or animal health surveillance

8.4. Mesenteric lymph nodes

8.4.1. Food safety

These nodes may be involved in generalised or systemic conditions and their anatomical prominence contributes to their value as indicators of such conditions.

There are no specific food safety abnormalities in the mesenteric lymph nodes. However, they will act as indicators of specific conditions affecting the gastro-intestinal tract since the afferent lymphatic drainage is from the entire length of the intestinal tract. Abnormalities associated with salmonellosis include enlargement and oedema (Thompson, 2004) which will be visually apparent along with concurrent changes in the gastro intestinal tract.

There is considerable evidence of enteric pathogens being isolated from mesenteric lymph nodes and enteric material (Moo *et al*., 1980; Cray and Moon, 1995; Hoar *et al*., 2001; Dowd *et al*., 2008) and this indicates that routine incision of these lymph nodes could potentially cross-contaminate other tissues.

8.4.2. Food suitability

The mesenteric nodes may be affected by some suitability abnormalities. There is no specific indicator function of the mesenteric lymph nodes for suitability characteristics involving other tissues.

Advanced cases of Johne's disease are accompanied by emaciation, atrophy of fat depots, intermandibular oedema and ascites. These animals are unlikely to be presented for slaughter. Less advanced cases will present with lesions in the gastro-intestinal tract, with thickening and corrugation of the jejunal wall (Thompson, 2004) especially adjacent to the ileocaecal junction which may be enlarged, and extending to the caecum and colon while the mesenteric lymph nodes will be markedly enlarged, pale and oedematous, especially in the medulla (Jubb *et al*., 1993). In less advanced cases, lesions are generally confined to small areas of the intestine and individual lymph nodes and may only be diagnosed by histopathology (Worthington, 2004)

8.4.3. Animal health surveillance

Examination of the mesenteric lymph nodes is useful for bovine tuberculosis surveillance Of the laboratory confirmed cases of bovine tuberculosis found in New Zealand in recent years (Anon.b, 2011) over 7% of cases were from lesions in the ileojejunal or ileocaecal lymph nodes with most (6.3%) detected in the former. However, most of these cases involved multiple lesion sites from the carcass and viscera, and only 2.4% of submitted samples were from animals with only one lesion. The study also indicated that ileojejunal lesions were more common in reactor animals, which will continue to receive an enhanced intensity of examination by incision of lymph nodes

8.4.4. Scientific evaluation

The mesenteric lymph nodes of cattle slaughtered in New Zealand should be routinely examined by viewing.

8.5. Renal lymph nodes

8.5.1. Food safety

These nodes may rarely be involved in generalised or systemic conditions

There are no specific food safety abnormalities in the renal lymph nodes.

Leptospira species infections of cattle do occur and generally affect the kidneys with interstitial nephritis being the usual gross pathology. However some studies indicate that in many cattle in which the organism can be isolated, gross lesions are present but may be inconspicuous with only microscopic foci of interstitial nephritis present (Kingscote, 1985; Prescott *et al*., 1987). The renal lymph nodes have been reported as being enlarged and hyperaemic in acute leptospirosis in cattle (Hungerford, 1975) with concurrent and obvious congestion of the kidneys. The renal lymph nodes provide no indicator value in these cases.

Tumours of bovine renal cells are rarely encountered in slaughtered cattle and of these, in a retrospective study of cattle so affected over 11 years, only one case showed any metastasis involvement of the renal lymph node with concurrent abnormalities in the peritoneum and mesentery (Kelley *et al*., 1996).

8.5.2. Food suitability

The renal nodes may be affected by some suitability abnormalities. There is no specific indicator function for suitability characteristics involving other tissues.

8.5.3. Animal health surveillance

Examination of the renal lymph nodes has no usefulness in the bovine tuberculosis surveillance Not one submission has been made from these nodes in the New Zealand programme

8.5.4. Scientific evaluation

Routine examination of the renal lymph nodes in cattle slaughtered in New Zealand has no value for food safety, food suitability or animal health surveillance

8.6. Subiliac lymph nodes

8.6.1. Food safety

These nodes may rarely be involved in generalised or systemic conditions

There are no specific food safety abnormalities in the subiliac lymph nodes.

Abscessation occasionally occurs in the hind limb and due to their location and afferent vessels from the skin of the posterior part of the thorax, the abdomen, pelvis, thigh and leg and also from the tensor fascia lata and prepuce, the subiliac lymph nodes may have some indicator function towards conditions that may have occurred distally in the hind limbs, However, efferent vessels from the subiliac lymph nodes lead to the internal iliac lymph nodes, and reactive changes would also be expected in the latter lymph nodes from any severe peripheral challenge.

8.6.2. Food suitability

The subiliac lymph nodes may be rarely affected by some suitability abnormalities. There is no specific indicator function for suitability characteristics involving other tissues.

8.6.3. Animal health surveillance

Examination of the subiliac lymph nodes has no usefulness in the bovine tuberculosis surveillance. The recent MAF study (Anon.b, 2011) has shown that of the laboratory confirmed cases of bovine tuberculosis that were included in the databases, the prevalence of affected subiliac lymph nodes was 0.27%. Of those animals with only one lesion, only 0.06% of submissions were from these lymph nodes.

8.6.4. Scientific evaluation

Routine examination of the subiliac lymph nodes in cattle slaughtered in New Zealand has no value for food safety, food suitability or animal health surveillance

8.7. Superficial cervical lymph nodes

8.7.1. Food safety

These nodes may rarely be involved in generalised or systemic conditions

There are no specific food safety abnormalities in the superficial cervical lymph nodes.

8.7.2. Food suitability

The superficial cervical lymph nodes may be rarely affected by some suitability abnormalities. There is no specific indicator function for suitability characteristics involving other tissues.

Abscessation may occasionally occur in the fore limb and the superficial cervical lymph nodes receives afferent vessels from the skin of the neck, shoulder, part of the ventral and lateral surfaces of the thorax, and the thoracic limb. On this basis, the superficial cervical lymph node might provide an indicator function for a localised and non-observable peripheral condition such as arthritis, or infection of the foot or lower forelimb, but there is no anecdotal evidence of the value of routine post mortem inspection for detecting such cases.

8.7.3. Animal health surveillance

Examination of the superficial cervical lymph nodes has negligible usefulness in the bovine tuberculosis surveillance. The recent MAF study has shown that of the laboratory confirmed cases that were included in the databases, the prevalence of affected superficial cervical lymph nodes was 2.52%. Most of thesesubmissions also involved three or more other carcass sites..

8.7.4. Scientific evaluation

Routine examination of the superficial cervical lymph nodes in cattle slaughtered in New Zealand has no value for food safety, food suitability or animal health surveillance

8.8. Superficial inguinal and supramammary lymph nodes

8.8.1. Food safety

These nodes may be involved in generalised or systemic conditions but are unlikely to provide any additional information to that seen in more prominent groups of lymph nodes throughout the carcass and viscera.

There are no specific food safety abnormalities affecting the superficial inguinal and supramammary lymph nodes.

There are no specific indicator functions for these lymph nodes although a range of micro-organisms may be present, usually without accompanying grossly detectable abnormalities.

Organisms that may be isolated from milk and mammary tissue of cows in New Zealand include *Staphylococci* spp., *Streptococci* spp., coliforms, *Pseudomonas* spp., *Arcanobacterium* spp., *Bacillus* spp., *Clostridium* spp. and *Mannheimia* spp. Within cattle, these organisms are generally restricted to a localised infection of the mammary tissue as opposed to systemic infection. Other microflora that have been isolated from affected mammary tissue in New Zealand include *Aeromonas hydrophilia*, *Nocardia asteroides*, *Candida* spp., *Aspergillus* spp., *Pseudoallescheria* spp. and *Trichosporon* spp. although it was not known whether these organisms were pathogens or contaminants (Vermunt, 2000). The likelihood of *M. bovis* being isolated from cow's milk in New Zealand is extremely low. *M. avium* subsp. *paratuberculosis* has been isolated from the milk of asymptomatic cattle in the US (Sweeney, 1992) and in these animals it was believed to have disseminated from enteric infections.

8.8.2. Food suitability

The superficial inguinal and supramammary lymph nodes may be affected by a range of suitability abnormalities. However, the prevalence is likely to be low and the sensitivity of any post mortem inspection procedure is likely to be highly variable.

There is no specific indicator function for suitability characteristics involving other tissues. However, the general comments on a potential indicator function for hazards as above apply.

8.8.3. Animal health surveillance

Examination of the superficial inguinal and supramammary lymph nodes has no value in the bovine tuberculosis eradication programme. The recent study on distribution of grossly-detectable lesions in slaughter cattle in New Zealand (Anon. b, 2011) showed that only one submission of a suspect lesion was made from these nodes.

8.8.4. Other considerations

For udders that are saved for human consumption, the supramammary lymph nodes may provide information to the meat inspector as to the likelihood of the presence of localised conditions such as mastitis. In New Zealand, the mammary tissue of cows is not routinely saved for human consumption and, along with associated lymph nodes and surrounding fat, is usually removed prior to the carcass leaving the slaughterfloor. Any spillage of milk from the udder is regarded as contamination and is controlled by the application of GHP.

In male cattle, the cod fat and tissue surrounding and including the superficial inguinal lymph node are routinely trimmed prior to the carcass leaving the slaughterfloor. The afferent drainage to the superficial lymph nodes does not include that from the testicles or epididymi and so these lymph nodes provide no information about any localised condition restricted to these particular tissues.

The examination of the superficial inguinal and supramammary lymph nodes will only provide information about material that is usually discarded, such as external genital organs, udder or skin. In the event of the penis and/or udder being saved for human consumption, these tissues will be required to undergo further examination. Conditions that have spread from the external genitalia and mammary tissue should be detected by examination of the internal iliac lymph nodes and adjacent tissue on the carcass.

While there is little information available regarding the prevalence of abnormalities in the superficial inguinal and supramamary lymph nodes, aged dairy cows constitute a significant proportion of the manufacturing beef production of New Zealand.

There is divergence between countries on examination requirements for this lymph node, with some recommending viewing and some recommending incision.

8.8.5. Scientific evaluation

Field studies are needed to further evaluate a potential indicator function of the superficial inguinal and supramammary internal iliac lymph nodes for food safety and/or food suitability conditions.

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Appendix 1: Summary of post mortem examination requirements

Lymph node	NZ current	NZ proposed	Aus	Canada	US	EU
Atlantal (in head &/or c/c)	I (in head &/or c/c	none in head or c/c	I (in head only) or #	I (in head only) Φ	I (in head only)	I (in head only)
Internal iliac	I	I (interim)	V or P∂	none	P	none
Lumbar chain	I	none	none	none	none	none
Mesenteric	VP	V	V	V	V	VP
Renal	I	none	none	none	none	none
Subiliac	P 1 & I 2	none	none	none	none	none
Superficial cervical	P 1 & I 2	none	none	none	none	none
Superficial inguinal	I	I (interim)	V or P†	none	P	none
Supramammary	I	I (interim)	V or P†	none	P	V *

I – Examine by incision and viewing

V – Examine by viewing only,

VP – Examine by viewing and palpation

P – Examine by palpation only

none - Examination not specified

P 1 – Palpate in prime cattle and young bulls

I 2 – Incise in all other cattle, including prime cattle and young bulls which are SPVD on ante mortem or with tuberculous or actinoform lesions, and young bulls if overlying tissue prevents effective palpation.

V or P∂ - Examination of the internal iliac lymph nodes by viewing in all cattle, apart from bulls and mature cows, where they are required to be palpated.

V or P† - Palpate the superficial inguinal and internal iliac lymph nodes or, for animals which require minimal risk inspection for tuberculosis (other than animals subject to conditional slaughter or emergency slaughter), an equivalent procedure is to observe the nodes (other than in bulls or mature females).

V* - Visual examination, and if necessary, palpation and incision of the udder and the supramammary lymph nodes. In cows, each half of the udder must be opened by a long, deep incision as far as the lactiferous sinuses and the lymph nodes of the udder must be incised, except when the udder is excluded from human consumption.

- For animals for which minimal inspection procedures are required for tuberculosis, equivalent procedures are either 1) observe only or 2) excise and discard without inspection. Australia does not distinguish between procedures for medial and lateral retropharyngeal lymph nodes.

Φ - Under the High Line Speed Inspection System (HLIS), examination of the lateral retropharyngeal is by observation only.

Appendix 2: Possible foodborne pathogens of cattle (after Alonso et al., 2010)

Bacteria	Parasites and protozoa	Viruses, rickettsia and prions
Campylobacter spp * *Salmonella* spp * *Yersinia* spp *	*Trichinella* spp * *Taenia saginata* (C. bovis) * *Taenia solium* (C. cellulosae) *	TSEs / vCJD * *Adenoviridae* Astrovirus
VTEC *	*Echinococcus* (hydatidosis) * †	Enterovirus
*Mycobacterium bovis** *Mycobacterium avium* subsp. *paratuberculosis* *	*Sarcocystis suihominis* * *Sarcocystis hominis* *	Norovirus Rotavirus
Streptococcus suis * *Staphylococcus aureus* *Bacillus* spp *Arcanobacter* spp *Aeromonas* spp Shigella *Brucella* spp *Vibrio cholerae* *Listeria monocytogenes* *Clostridium botulinum* *Erysipelothrix rhusiopathiae* *Clostridium perfringens* *Streptococcus* spp *Francisella tularensis*	*Toxoplasma gondii* * *Fasciola hepatica* * † *Giardia* spp *Cryptosporidium* spp *Balantidium coli* *Isospora belli* *Capillaria hepatica*	Sapporo-like virus Flavivirus Hepatitis A *Coxiella burnetii*

† Hazard not meatborne but for which meat is an essential components of transmission.

Appendix 3: The contribution of routine post mortem inspection of slaughter cattle to animal health surveillance

In 2010 a study was undertaken by NZFSA on the distribution of lesions in cattle caused by *Mycobacterium bovis*. (Anon.b, 2011) The objective of this project was to evaluate, relative to current practices, the impact of alternative post mortem examination regimes for tuberculosis in cattle presented for slaughter in New Zealand. This included not only the scope of the examination of carcases, but also the possible use of animal, herd and farm factors to target high prevalence tissues. In the investigation there were three sequential phases. Firstly, a retrospective analysis of slaughterhouse post mortem data from between 1989 and 2010 was conducted. Secondly, using the findings from part one, a model of the sensitivity of gross post mortem examination with respect to detecting both infected animals and slaughter lines containing infected animals was developed, and thirdly, the outcomes of alternative post mortem examination procedures were evaluated using the model.

This study showed that of the laboratory confirmed cases of bovine tuberculosis that were included in the databases, irrespective of the number of detected suspect lesions; around one third (33%) had lesions in the head lymph nodes, almost two thirds (62%) had lesions in the thorax and 7% had lesions in the mesenteric lymph nodes. The most commonly affected lymph nodes were the mediastinal lymph nodes (44%), followed by the retropharyngeal nodes (28%) and the tracheobronchial nodes (24%). Just over 92% of positively identified cases had lesions in head, thorax or mesenteric lymph nodes and these areas represent the primary sites for bovine tuberculosis.

In animals with single or multiple lesions the prevalence of affected tissues did not increase in a proportional manner. Lesions of the mediastinal, retropharyngeal and tracheobronchial lymph nodes still predominated in both classes, but in those with multiple lesions; lung, liver and the ileojejunal nodes were, proportionally, more commonly affected. The distribution of lesions in animals with single or multiple lesions differed. In the former, only 2.4% had lesions in abdominal viscera while in the latter it was 19% of animals.

The study considered the likely reduction in sensitivity of post mortem examination of cattle with regard to the detection of bovine tuberculosis if particular tissues and lymph nodes were removed from the list of required procedures. The order in which these tissues and lymph nodes were added to the list of dropped procedures began with those that had the lowest recorded prevalence of confirmed bovine tuberculosis and successively included those tissues and lymph nodes in which the prevalence increased.

A model which accurately simulated the current observed lesion prevalence was developed. It was found that approximately 50% of lesion sites could be dropped from the examination procedures with only a 2% loss of sensitivity while a group of ‘operationally relevant’ tissues could be dropped with only a 1% loss of sensitivity.

When the following organs and lymph nodes were dropped from routine bovine post mortem procedures: kidney, lumbar chain, superficial inguinal and supramammary, renal, ischiatic, internal iliac, popliteal, prepectoral, precrural, hepatic, ileocaecal and atlantal lymph nodes; the reduction of probability of detection of an infected animal was less than 2 %.

When the probability of detection of an infected line was calculated, with the above tissues and lymph nodes removed from the routine procedures, the reduction in probability of detection of an infected line was even less than that for an individual animal, at 1.5%.

It is estimated that the overall sensitivity of gross post mortem examination is around 85% (i.e. the probability that visible lesions will occur in an *M. bovis* infected animal) and the decrease in sensitivity that would occur as a result of dropping around 50% of current sites is small in comparison with this.

Since bovine tuberculosis is not considered a significant foodborne zoonosis in red meat but a disease that New Zealand wishes to eradicate, the primary importance of routine organoleptic meat examination at slaughter is to detect the disease in those animals in lines not already subject to movement control, such as positive reactors and animals from herds with an infected status.

The study concluded that a more intensive examination of lymph nodes draining the oropharynx and thoracic tissues would yield a higher sensitivity than the current procedures.

Appendix 4: Mycobacterium avium subsp. paratuberculosis in slaughter cattle

As yet there is no scientific consensus as to whether this organism should be regarded as a zoonosis. An association between Johne's disease in animals and Crohn's disease in humans has been reported but causality has yet to be shown (Ryan and Campbell, 2006). Notwithstanding this uncertainty, Johne's disease is regarded as a significant production limiting disease of cattle and post mortem examination has a role to play in the detection and control of the disease.

The bacterium *Mycobacterium avium* subspecies *paratuberculosis* (MAP) has been reported as a possible food contaminant, particularly in dairy products. Despite there being no general evidence that MAP causes human foodborne disease, it has been suggested it has a role in Crohn's disease, a chronic inflammatory condition of the lower bowel. Parallels have been drawn with Johne's disease in livestock, a condition accepted as being caused by MAP.

Mycobacteria are a large, varied group of organisms, which include some significant pathogens (such as *M. tuberculosis, M. bovis, and M. leprae*) and many free-living non-pathogens. An important sub-group that can survive outside an animal host is the *M. avium* complex; MAP falls within this group. Although MAP is considered an obligatory parasite, it can survive outside the animal for prolonged periods (6 to 12 months in moist, pH neutral conditions). Two major strains of MAP are recognised, the so-called sheep and cattle strains with a high degree of host preference. Characteristic differences in genome of these strains have been demonstrated.

A feature of MAP infection or 'paratuberculosis' or Johne's disease, as it is commonly called, in ruminants is a prolonged latency; i.e. infection can be demonstrated but the animal exhibits no clinical signs. The typical picture of an advanced clinical case in cattle and sheep is one of a granulomatous enteritis, principally in the lower small intestine and its draining lymph nodes. In the multibacillary form of Johne's disease, large numbers of acid-fast staining bacilli are present in macrophages in the granulomas. In cattle it has also been shown that a proportion of advanced cases may also shed small numbers of organisms in milk.

There is general agreement that there is insufficient evidence to conclude that MAP is the cause of Crohn's disease, and data on Johne's and Crohn's diseases are not consistent with a simple cause and effect relationship, as is seen, for example, with brucellosis in cattle and undulant fever in humans.

A comprehensive report of the 'Scientific Committee on Animal Health and Welfare' (EC 2000) covers the microbiology, pathology, epidemiology and diagnosis of Johne's and Crohn's diseases. The potential for exposure via milk and drinking water is also reviewed. There are references to 372 scientific papers and articles. The authors reached two main conclusions.

First, the currently evidence is insufficient to confirm or disprove that MAP is a causative agent of at least some cases of Crohn's disease. Second, there are sufficient grounds for concern to warrant increased and urgent research activity to resolve the issue. A number of supporting conclusions are also included, as follows:

- Crohn's disease is most likely a multifactorial condition
- The incidence is more common in the developed world, in families where there have been other cases and in homes where hygiene in early life has been good
- Although there are similarities between Johne's and Crohn's diseases, there are also some significant differences
- MAP is a relatively common environment contaminant and its association with Crohn's disease may be as a causative agent, pathogenic secondary invader, or non-pathogenic coloniser of changed bowel conditions
- The public could be exposed directly via contact with infected animals or unpasteurised milk. Other possible lower probability sources are pasteurised milk and infected wildlife
- A simple relationship between exposure and development of Crohn's disease does not exist
- If MAP is involved in the causation of Crohn's disease, other factors are required

There have been a range of proposed bacterial and viral aetiologies of Crohn's disease. In addition to mycobacteria, they include *Escherichia coli, Listeria monocytogenes, Klebsiella pneumonia, Yersinia* species, *Streptococcus* species, measles, cytomeglovirus, and Epstein-Barr virus.