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**AGRICULTURE  
FISHERIES &  
FORESTRY -  
AUSTRALIA**



**BIOSECURITY  
AUSTRALIA**

## **A SCIENTIFIC REVIEW OF LEPTOSPIROSIS AND IMPLICATIONS FOR QUARANTINE POLICY**



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## EXECUTIVE SUMMARY

Leptospirosis is a contagious disease of animals and humans caused by the spirochaete *Leptospira* of which there are two forms, the pathogenic species and the non-pathogenic (benign) species. Only the pathogenic *Leptospira* species are considered in this review.\*

In Australia, clinical leptospirosis occurs in cattle (serovars (svs) hardjo, pomona and zanoni) and pigs (pomona, tarassovi and bratislava). Sporadic cases occur in sheep (hardjo), horses (pomona) and dogs (copenhageni and australis). Clinical cases have been reported in humans with svs australis and zanoni predominating in the tropics, and sv hardjo predominating, with some sv pomona and occasionally sv tarassovi, in the temperate regions of Australia.

In other countries, many animal species may be infected with leptospira serovars considered exotic to Australia. Some serovars are highly pathogenic to animals or humans or both. Many exotic serovars are carried by maintenance hosts not found in Australia and consequently may not establish in Australia, despite the perceived possibility of exotic serovars adapting to new hosts and establishing in this country.

Australian quarantine restrictions for leptospirosis are currently confined to the requirement that dogs give a negative result to an antibody test for *L interrogans* sv canicola despite this serovar having been isolated from a human in Australia. In contrast, many of Australia's trading partners impose import conditions for leptospirosis in most livestock species, horses, dogs, cats, and their genetic material.

Because pathogenic leptospire interact with the host and the environment in complex ways, this review discussed a number of leptospira serovars individually. AQIS evaluated the risk associated with several different serovars, host species (including humans) and circumstances.

Leptospirosis is not a notifiable disease of animals in some Australian States and Territories and there is evidence of lack of regulatory action where leptospirosis has been notified. This has impacted on the consequence of entry, establishment and spread of leptospirosis, which is evaluated as 'negligible' to 'low' except where a risk event suggests there could be serious public health risks.

The qualitative method used to derive these risks is described in the review. The results of this evaluation are summarised in Table A.

Australia's *acceptable level of protection* is the level of protection deemed acceptable by Australian public health and veterinary authorities in managing the disease within their territories. Australia has limited requirements for the control of leptospirosis within its territory. Clinical disease in animals and humans generally occurs sporadically, and prevention and control of leptospirosis is not mandatory. Thus where the overall risk is assessed to be negligible or low, the imported animal or animal product satisfies Australia's acceptable level of protection without requiring quarantine measures to further manage the risk of leptospirosis.

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\* Standard nomenclature requires that the species be written in italics, for example, *Leptospira interrogans* or *L. interrogans* but serovars and serogroups be written in standard text.

But where the overall risk is considered to be *moderate* or *high*, that is, for:

- semen and embryos,
- animal derived tissue cultures, and
- pet rodents and other small mammals,

a quarantine measure was considered for each of the risk events.

**Table A.**

<b>Scenario</b>	<b>Risk</b>
Importing a dog from a country with a significant stray dog problem, where canicola infection occurs and dogs are not routinely vaccinated (eg, most African, Asian or South American countries).	<b>Low</b>
Importing a dog from a country where sv canicola has not been isolated from dogs since the 1950's (eg, USA, Canada, and New Zealand).	<b>Low</b>
Importing a pig clinically infected with sv canicola (eg, Republic of South Africa).	<b>Negligible</b>
Importing a ram infected with sv canicola (eg, Portugal).	<b>Low</b>
Importing a dog from a country which reports dogs with antibody titres to svs batavia, bratislava, javanica and cynopteri, none of which has been reported in Australia (eg, Southeast Asia).	<b>Low</b>
Importing a dog from country where infections due to sv bim occurs (eg, the Carribean).	<b>Low</b>
Importing a bull infected with sv hardjoprajitno from a country where this serovar is endemic (eg, Ireland).	<b>Low</b>
Importing a pregnant cow with sv hardjoprajitno infection from a country where this serovar is endemic (eg, Ireland).	<b>Low</b>
Importing a cow shedding either of svs mozdok or kennewicki in urine (eg Europe or North America).	<b>Low</b>
Importing a boar shedding either of svs mozdok or kennewicki in urine (eg Europe or North America).	<b>Low</b>
Importing a non-pregnant racehorse for a temporary stay of 2 months for competition purposes (eg, North America).	<b>Low</b>
Importing a pregnant mare with foetus infected with a serovar exotic to Australia (eg, Europe - sv mozdok and North America – sv kennewicki).	<b>Low</b>
Importing a boar with sg australis infection (eg, Europe).	<b>Low</b>
An infected carrier rat escapes from a ship and enters Australia.	<b>Low</b>
Importing an infected pet rat (eg, Asia).	<b>Moderate</b>
Animal or humans infected with sv lai enter Australia (eg, Asia).	<b>Negligible</b>
Untreated frozen semen from untested donors infected with exotic pathogenic leptospire (eg, Europe, South America)	<b>Negligible</b>
Importing a batch of animal vaccine containing cell lines prepared from infected animal kidneys (eg, Asia).	<b>Moderate</b>

As effective antibiotics are routinely used in the processing of semen, embryos and animal derived tissue cultures, there is no need to impose additional quarantine conditions. For pet rodents and other small mammals, the proposed quarantine requirement is that these mammals must come from colonies tested free from leptospiral strains that are 'exotic' to Australia and relevant to the species.

Several of Australia's trading partners impose quarantine conditions for leptospirosis. Some require animals to be injected with streptomycin/dihydrostreptomycin (S/DHS) prior to export. While S/DHS is usually the most effective antibiotic for treatment of leptospirosis, it does not always sterilise infection and thus it is not an effective quarantine measure. Several countries, including Australia, no longer use S/DHS in food-producing animals as the injection causes local irritation and pain and treatment can cause residue problems in food products. In Australia, it can only be used by permit from the National Registration Authority (NRA) in food-producing animals. Because of the small demand for this antibiotic, pharmaceutical companies are not restocking their supplies and it has becoming increasingly difficult to obtain S/DHS for use in export livestock. Australia will either need to renegotiate those import conditions that require S/DHS treatment for leptospirosis or make arrangements for the importation of S/SDH specifically for use in export livestock. However, the latter option does not address the problem of managing stock treated with S/SDH then withheld from export.

Other quarantine measures for leptospirosis, including those recommended in the OIE *Code*, are also of questionable value. Australia intends to draw this to the attention of other countries and propose revised guidelines for leptospirosis in the OIE *Code*.

# 1. Introduction

Leptospirosis is a contagious disease of animals and humans caused by the spirochaete *Leptospira* of which there are two groups, the pathogenic species and the non-pathogenic (benign) species. Only the pathogenic *Leptospira* species are considered in this review. Over the years, the classification of the genus *Leptospira* has undergone some changes and is now currently classified in two ways:

1. on the basis of agglutinating antigens into over 250 serovars contained within 23 serogroups;
2. on the basis of DNA studies with all 250 plus serovars placed into eight genomospecies.

There are proposals for further changes to the classification system but this will not be considered in this report. There is still considerable confusion on the current taxonomy of the leptospires, largely because serovars were previously grouped into one of 2 species, *interrogans* and *biflexa* and are now grouped into a number of species. Appendix 1 lists the known pathogenic *Leptospira* serovars according to genomospecies and serogroups.

The bacteria can cause polymorphic disease conditions in domestic animals, wildlife and humans. Infections range from asymptomatic or subclinical to acute and fatal. Symptoms of acute leptospirosis in animals include sudden agalactia in the lactating female, icterus and haemoglobinuria in the young, nephritis and hepatitis in dogs, and meningitis. Chronic leptospirosis can cause abortion, stillbirth, runtling, and infertility. Often chronically infected animals remain as asymptomatic carriers for life with the organism localised in the kidneys and in the reproductive organs. Horses can develop periodic ophthalmia as a result of leptospirosis.

In humans, leptospirosis can cause headaches, fever, chills, sweats and myalgia. Other symptoms may include lethargy, aching joints, and long periods of sickness. Some highly pathogenic serovars may cause pulmonary haemorrhaging and death. While mild type leptospirosis is probably the most common form of infection, they can sometimes be chronic in nature and have a 'mental' component to their clinical manifestations.

In Australia, clinical leptospirosis is most common in cattle (svs hardjo and pomona) and pigs (pomona). A number of other serovars have also caused disease in these two species. Sporadic cases occur in sheep (hardjo), horses (tarassovi) and dogs (copenhageni). There are no reports of clinical leptospirosis in camels although leptospira antibodies have been detected in camels. Clinical cases have been reported in humans with svs australis and zaroni predominating in the tropics, and svs hardjo predominating, with some sv pomona and occasionally sv tarassovi, in the temperate regions of Australia.

Leptospirosis is of increasing importance as an occupational disease as intensive farming practices become more widely adopted. During 1999, those working in agricultural industries in Australia accounted for 35.3% of notifications while those working in livestock industries accounted for 22.9% of notifications.<sup>1</sup>

Australian quarantine restrictions for leptospirosis are currently confined to the requirement that dogs give a negative result to an antibody test for *L interrogans* sv canicola. This serovar is considered exotic to Australia despite having been isolated from a human in Australia. In contrast, several of

Australia's trading partners impose import conditions for several leptospira serogroups in livestock species, horses, dogs, cats, and their genetic material.

The aim of this report is

1. to review scientific information on leptospirosis and implications for Australia's quarantine policies,
2. to provide scientific arguments as a basis to negotiate less restrictive conditions for the export of animals and genetic material, and
3. to propose changes to the OIE International Animal Health Code Chapter on leptospirosis.

## 2. National obligations

### a) Commonwealth

The Commonwealth has the legal responsibility to evaluate the quarantine risks of introducing quarantinable diseases, including leptospirosis, into Australia with animals. According to the Australian Government Solicitor, the law recognises that where public bodies carry out statutory functions they must do so in a way which is not negligent. Legal liability is possible where a public body does not perform a statutory function and harm results to others.

### b) State / Territory

Leptospirosis is not a notifiable disease in all States and Territories. However, most States and Territories require donors and other animals in artificial breeding centres to be free from leptospirosis. Table 1 summarises the legislative requirements in different states.

**Table 1 – A summary of legislative requirements for leptospirosis in animals in Australia**

State	Act	Comment
ACT	Animal Diseases Act 1993	Not notifiable
NSW	Stock (Artificial Breeding ) Act 1985	Infected donors used for artificial breeding (AB) must be treated, removed or destroyed.
	Stock Diseases Act 1923	Not notifiable
NT	Stock Diseases Act 1996	Notifiable
	Stock Diseases (Artificial Breeding) Act 1996	Donors for artificial breeding must be free.
QLD	Stock Act 1915	Not notifiable.
SA	Livestock Act 1997	<i>L interrogans</i> sv canicola is notifiable. AB Centres must comply with the <i>Australian Health Standards for Bovine Semen Collection Centres</i> published by SCARM.
TAS	Animal Health Act 1995	<i>Leptospira borgpetersenii</i> sv hardjo, and <i>L interrogans</i> sv pomona, are both List B diseases under the Animal Health Act 1995. All List B diseases are notifiable. (s 28) A person may not knowingly expose an animal to a List B disease (vaccination is exempted from this) (s51) A person may not knowingly give away or sell an animal infected with a list B disease. (s52)
VIC	Livestock Disease Control Act 1994	Notifiable –must notify within 7 days, must not import diseased animal into Victoria, and AI donors must be free from disease
WA	Stock Disease (Regulation) Act 1965	Not notifiable

<b>State</b>	<b>Act</b>	<b>Comment</b>
	Artificial Breeding of Stock Act 1965	AI donors must be free from disease before entering Centres

### 3. International Obligations

Leptospirosis is an Office International des Epizooties (OIE) List B disease. List B diseases are transmissible diseases that are considered to be of socio-economic and/or public health importance within countries and which are significant in the international trade of animals and animal products. Reports of List B diseases are normally submitted once a year.

Australia advises the OIE yearly the status of leptospirosis in Australia. Table 2 summarises Australia's report for leptospirosis to the OIE for the years 1990 to 1999.

**Table 2**

Species Year	Bovine	Canine	Porcine	Caprine	Fauna	Feline
1990	++ Q T V *	++ Qf T V *	++ Q T V *			
1991	++ Q T V *	++ Qf T V *	++ Q T V *			
1992	++ Q T V *	++ Qf T V *	++ Q T V *			
1993	++ Q T V *	++ Qf T V *	++ Q T V *			
1994	++ Q T V *	++ Qf T V *	++ Q T V *			
1995	++ Q T V *	++ Qf T V *	++ Q T V *	+ Q T V	++	+?
1996	++ Q T V *		++ Q T V *			
1997	+ QiV					
1998	+ QiV		+ QiV			
1999	+ V	+ V	+ V	+ V	+ V	+ V

++ enzootic

+ low sporadic occurrence

+? serological evidence only, no clinical disease

Q quarantine, movement control and other precautions at frontier and inside the country

Qi quarantine measures and movement control inside the country

Qf quarantine and other precaution at the frontier

T treatment

V vaccination

\* notifiable disease

Some reports (1990-1996) did not accurately reflect the situation in Australia. There are no reports of quarantine being in place due to leptospirosis in cattle and pigs, despite the occurrence of the disease, nor are there import health requirements for leptospirosis for livestock entering Australia. Treatment and vaccination of livestock is not mandatory. In some States, leptospirosis is not a notifiable disease. The 1999 report accurately reflects Australia's status.

## **4. Australia's surveillance and monitoring systems for *Leptospira*.**

### **a) Australia's disease status**

Leptospirosis is enzootic in Australia, occurring in both animals and humans.

There is no national surveillance system for leptospirosis in animals in Australia. The main sources of data are the veterinary laboratories. However, most veterinary laboratories do have a database of cases reported and some states have a livestock disease management information system, such as Fieldvet in NSW. Leptospirosis occurs in all states and territories. No attempts have been made to map the prevalence of the disease, though it is accepted that prevalence is much higher in wetter tropical areas than in the arid areas of Australia.

In Australia leptospirosis is not a zoonosis of significant economic importance. Australian public health authorities regard Q fever as the most important of all zoonotic diseases in terms of reported numbers of cases. There were 571 notifications of Q fever in 1998 as compared with 197 notifications for leptospirosis despite Q fever being under-reported with only 50% of cases diagnosed by health professionals.<sup>2</sup>

The Animal Industries Public Health Committee (AIPHC), which advises the Standing Committee on Agriculture and Resource Management (SCARM) on public health issues associated with animals and livestock production, has evaluated the nine most critical current and emerging animal industry public health issues in Australia to be anthrax, BSE, Johne's disease, enterohaemorrhagic *E. coli*, salmonella, equine morbillivirus, Australian bat lyssavirus, Japanese encephalitis and Q fever. It appears that leptospirosis is not considered to be a current animal industry public health issue.

### **b) Australia's diagnostic capabilities**

Of the 21 veterinary laboratories in Australia, 11 provide diagnostic tests for leptospirosis. These laboratories are monitored by a quality assurance program, ANQAP – Australian National Quality Assurance Program. During 1997 a total of 269 evaluations covering a range of tests for several diseases were performed and ANQAP expressed serious concerns with the large between-laboratory variation in the 3 micro agglutination tests (MAT) used for leptospirosis (*L hardjo*, *L pomona* and *L tarassovi*). This variation was due to variation in methodology and reagents.

The ASDT (Australian Standard Diagnostic Techniques for Animal Diseases) provides the official standard recognised for diagnosis of animal diseases in Australia. Its purpose is to provide a uniform approach to diagnosis for veterinary laboratories throughout Australia.

According to the Phase 8 ANQAP report, "*elements of the ASDT (CSIRO) require revision as sections of the 1993 ASDT conflict with international practice. Because of these discrepancies, some laboratories have introduced their own modifications or taken on elements of other MAT methods used internationally. The implementation of variations and modifications from the ASDT has been done on an ad hoc basis. Laboratories are now using*

*different methods around the country.... Until the MAT is standardised with the introduction and implementation of a new ASDT, laboratories will not be dis-endorsed for this test.”* Some laboratories were able to increase the sensitivity for the MAT after replacing their cultures (used as a source of antigen) with fresh cultures from the WHO/FAO Reference Laboratory in Brisbane, Queensland.

### **c) Domestic surveillance for human leptospirosis**

Diagnosis of leptospirosis on clinical signs is not definitive and can be confused with several other diseases such as influenza, acute glomerulonephritis, hepatitis, and the various causes of haemorrhagic fevers.

Early diagnosis in humans can be confirmed by culture of blood samples during initial fever, culture of urine samples during recovery from fever or by muscle biopsy with immunostaining for leptospire during muscle pains. One week after onset of symptoms has passed, the MAT or IgM ELISA may be used to detect antibodies in the blood.

The WHO Leptospiral Reference Laboratory in Brisbane maintains an Australian wide surveillance database for leptospirosis notifications. Data are collected from the questionnaires sent by the laboratory to the clinicians of notified cases. Clinicians are required to notify the National Notifiable Disease Surveillance Scheme of confirmed cases of leptospirosis. This data contains specific information regarding different regions within Australia, including the extremely variant epidemiological picture within Queensland. Demographics collected include age, gender, geographical location, animal exposures, occupation, symptoms, previous leptospiral infection status, coexisting infection and recreational activities.

## 5. Current export and import requirements for leptospirosis

### a) Export requirements for leptospirosis

Export requirements for leptospirosis vary considerably, depending on the conditions imposed by the importing countries. Sixty-three countries have imposed health conditions for leptospirosis on Australian animals and their genetic materials. Types of conditions vary considerably and often include one or more of the following:

- disease free certification – variations in requirements include:
  - ◊ for export of livestock - leptospirosis was not diagnosed on the property of origin or the herd of origin for the previous ? months (? is usually 12 months but a few countries require only 6 months);
  - ◊ for semen collection – all animals or the premises were free from leptospirosis at the time of semen collection;
  - ◊ for export of dogs – during the 12 months prior to the date of export there have been no cases of *L interrogans* serovar canicola diagnosed in Australia;
- negative serological test for leptospirosis - variations in requirements include:
  - ◊ MAT (negative at 1/100);
  - ◊ MAT using antigens for *Leptospira* serotypes known to occur in the exporting country;
  - ◊ MAT for certain serovars - a total of 15 serovars involved;
- treatment of animals with antibiotics - variations in requirements include:
  - ◊ two (2) treatments with specific antibiotics not more than 30 days prior to embarking;
  - ◊ dihydrostreptomycin on 2 occasions at a dose rate of 25 mg/kg body weight with an interval of at least 14 days;
  - ◊ dogs treated daily for 5 consecutive days with dihydrostreptomycin at a therapeutic dose rate immediately prior to export;
  - ◊ single intramuscular injection at 15 mg/kg body weight of amoxycillin before export;
- vaccination requirements - variations in requirements include:
  - ◊ livestock to be vaccinated within 30 days of shipment;
  - ◊ cattle vaccinated against leptospirosis (serovars hardjo and pomona) using a vaccine approved by AQIS and in accordance with the vaccine manufacturer's directions;
  - ◊ dogs to be fully vaccinated following manufacturer's recommendation, at least 21 days preceding departure;
  - ◊ dogs to be vaccinated against leptospirosis (bivalent icterohaemorrhagiae and canicola) no less than 14 days or more than 6 months before embarkation.

Some countries require testing for serovars endemic in their country but not for serovars exotic to the importing country but endemic in the exporting country. USA does not have any health requirements for leptospirosis for any animals or their genetic material from Australia except camelids. On the other hand, United Kingdom and New Zealand generally require all animals imported from Australia to be treated prior to export with 2 injections of streptomycin given 14 days apart.

## b) Current import requirements for leptospirosis

The only current import requirement for leptospirosis is that dogs entering Australia must give a negative serologically test for *L interrogans* sv canicola (Figure 1). Prior to 1990, health conditions required that dogs be serologically negative to *L borgpetersenii* sv ballum and *L interrogans* sv canicola or that dogs undergo treatment with dihydrostreptomycin. Around 1990, sv ballum was dropped from the health requirements on the grounds that it posed insignificant risk. In 1997, the option for treatment with streptomycin was dropped on the grounds that dihydrostreptomycin was being banned from use in animals in some countries and alternative antibiotics demonstrated less efficacy than dihydrostreptomycin for the treatment of leptospirosis. Australia did not require vaccination of dogs due to its limitations, especially its lack of effectiveness in removing an existing carrier state. The reason for the dogs to be negative to the serogroup canicola was that it was regarded as an exotic and a significant pathogen. Most likely, it was felt that this serovar did not exist in Australia. However, tests of dogs and humans in Australia in recent years strongly suggest that serovars belonging to the canicola serogroup exist in Australia but there is still no conclusive evidence that serovar canicola is present in dogs in Australia.

**Figure 1**

### Leptospirosis

The dog must be tested for *Leptospira interrogans* var. canicola infection by serum agglutination test, prior to export by EITHER of the following regimes:

- i) a single test within 21 days of export, having less than 50% agglutination at a serum dilution of 1:100 (ie. negative result),  
OR,
- ii) in the case of vaccinated dogs only, two tests, the first within 45 days of export and the second no less than 14 days after the first, each having a positive titre of not more than 1:400, the second sample must show no increase in titre above that of the first test.

The vaccination certificate, when applicable, and the laboratory report, or a copy endorsed by an Official Veterinarian, must be attached to the Veterinary Certificate B or C.

[In the case of a dog from an approved rabies-free island country or territory which does not have an Official Veterinarian this testing or treatment will be performed within the first 7 days of quarantine in Australia at the owner's expense.]

A survey of dogs undergoing pre-import testing for entry into Australia during 1998 and 1999 from south-east Asian countries and the Oceania by the WHO/FAO Reference Laboratory showed that 46 dogs out of 197 dogs were serologically positive (MAT > 100) to leptospirosis (Taylor T, *pers comm*). Nine dogs had MAT titres of 200 or more to sv canicola. In addition, the survey showed a significant number of dogs to show varying degrees of seroconversion to 18 serogroups of *Leptospira* as shown in Table 3. Twenty-eight of 46 dogs had leptospiral titres to more than one serovar.

**Table 3**

<b>WHO/FAO Results of dogs imported into Australia in 1998-99</b>							
<b>Status</b>	<b>Country</b>						
	Hong Kong	Malaysia	PNG	Solomon Is	Fiji	Brunei	Taiwan
Negative	55	37	14	1	10	3	0
Contaminated samples	4	3	0	0	0	0	1
Titres to leptospira	39	21	1	1	4	0	3
<b>Positive (&gt; 1:100)</b>	<b>25</b>	<b>14</b>	<b>1</b>	<b>0</b>	<b>4</b>	<b>0</b>	<b>2</b>
canicola	19	11			3		1
copenhageni	21	15			3		3
ballum	4	1					
hardjo	3				1		
kremastos	4				1		
szwajizak	4						
medanensis	4				1		
australis	1	1		1	2		1
javanica	2	4	1				
panama	1	4	1				
bulgarica	1	4					
cynopteri	1	1					
zanoni		1					2
shermani					1		
djasiman	1						
robinsoni							
pomona							
bataviae	1	1					

## 6. Leptospirosis in Australia

### a) Humans

Leptospirosis in humans is a notifiable disease in all States and Territories but the manner of notification varies considerably between States and Territories. For example, in New South Wales, laboratories must report cases of leptospirosis, while in Western Australia, employers must notify the Commissioner for Occupational Safety and Health if any employee has leptospirosis. In Victoria, notification by both laboratories and practitioners of confirmed cases is required.

The WHO/FAO Leptospirosis Reference Laboratory in Brisbane, Queensland has an Australian Wide Surveillance database for leptospirosis notifications. Data are collected from questionnaires sent by the laboratory to the clinicians of notified cases. Most of the data comes from Queensland where a high incidence is reported, especially in northern Queensland, and where clinicians have an increasing awareness of the disease. In Victoria, especially in wet years there is a history of very high incidence of leptospirosis, almost all sv hardjobovis, in dairy farmers and associated occupations. However, for the last few years, fewer cases were reported, most likely because of the persistent drought and the intensive campaign to routinely vaccinate cattle.

### b) Animals

#### i. Cattle

Two of the most common serovars, *L borgpetersenii* sv hardjo hardjobovis<sup>3</sup> and *L interrogans* sv pomona,<sup>4</sup> occur in cattle in Australia. Other serovars isolated from cattle include *L interrogans* svs australis,<sup>4</sup> grippotyphosa in NSW<sup>5</sup> and zanoni in northern Queensland.<sup>6</sup> *L interrogans* sv hardjo (that is, subtype hardjoprajitno) has not been isolated in Australia and is most likely exotic to Australia.<sup>7</sup> Many early Australian reports refer to sv hardjo. For the purposes of this review, this is presumed in all cases to be subtype hardjobovis. Despite a Victorian survey where 7.8% of cattle had titres to sv tarassovi, this serovar has not yet been isolated from cattle and does not appear to be associated with clinical disease in animals with titres.<sup>8</sup> Two cases of leptospirosis due to infection with a member of the hebdomadis serogroup occurred in farm workers on a Victorian dairy farm, and the source of infection appeared to be the dairy herd which had elevated titres against the hebdomadis serogroup.<sup>9</sup> However analysis of a serological survey of Victorian dairy farmers suggests that the reactions against serogroup hebdomadis were due to exposure to sv hardjo.<sup>10</sup> Often serological surveys show a low prevalence of reactions, usually with low titres to other serogroups, but they are generally regarded as cross-reactions such as the paradoxical reactions known to occur between various serogroups.

A serosurvey in Queensland showed that prevalence of pomona is higher in lower rainfall areas but hardjo is fairly uniformly spread throughout the state. There was a higher prevalence of antibodies to sv hardjo than to sv pomona in cattle while in pigs sv pomona was higher than for svs hardjo or tarassovi.<sup>11</sup> In north Queensland, infections with sv hardjo are usually endemic while infections with svs pomona or zanoni are usually sporadic. Seroconversion to zanoni occurred 15 weeks after initial isolation of the leptospire from the urine.<sup>12</sup>

Leptospirosis in cattle is usually subclinical. Serological titres vary considerably in peak and duration. Leptospire may be excreted in urine, often intermittently, for up to 18 months after infection. Cattle may remain serologically positive to leptospirosis for up to 7 years.<sup>13</sup> Sv hardjo usually causes a sudden decrease in milk production and flaccid or atypical mastitis in cows. Most infected cows return to full milk production within 2 weeks. Heifers may show no clinical signs and this may be due to either a greater tolerance to infection or to a change in pathogenicity of the organism.<sup>14</sup> Other workers have noted that where large outbreaks of leptospirosis due to sv hardjo have occurred, the number of herds becoming infected without clinical signs increased as the outbreak spread.<sup>15</sup>

Serovar pomona can cause haemolytic disease and haemoglobinuria in calves, with interstitial nephritis as a sequel, and late abortion in cows.<sup>16</sup>

Leptospirosis can be a serious disease in pregnant cattle.<sup>17</sup> Infection in pregnant cows may result in abortion that usually occurs after serological response has peaked. Leptospire can be detected in reproductive organs for several months after infection. Maternal serology is a poor indicator of foetal infection with leptospire, particularly with sv hardjo.<sup>18</sup> Leptospiral abortion is usually diagnosed by demonstration of leptospire in foetal tissues and high or rising MAT titre in the dam. However, surveys in the causes of abortion in cattle suggest that leptospire are of minor importance.<sup>19</sup> Experimental induction of abortion by inoculation of sv hardjo is rarely produced.<sup>20</sup> Immunisation against sv hardjo has been associated with a reduction of prenatal losses in beef cattle in Queensland.<sup>21</sup> Svs zannoni<sup>6</sup>, pomona and grippotyphosa<sup>5</sup> are implicated as causes of abortion in dairy cattle.

## **ii. Sheep**

*L borgpetersenii* sv hardjo has been recovered from renal tissues of sheep<sup>22</sup> and can be histologically demonstrated in kidneys with silver staining.<sup>23</sup> Nephritis, hepatitis and leptospiruria have been associated with hardjo infection in Australian sheep.<sup>24</sup> Sheep can act as a maintenance host for this serovar.<sup>25</sup> Serosurveys show that sv hardjo to be most common with low titres to svs tarassovi and pomona<sup>26</sup> and svs australis, autumnalis, copenhageni and grippotyphosa<sup>22</sup> occurring, most likely as crossreactions.

## **iii. Pigs**

Acute leptospirosis in young pigs can cause severe weakness, loss of appetite, jaundice, fever, convulsions, haemorrhaging and can be fatal. Newborn or young piglets are most susceptible to infection while adult non-pregnant animals are usually most resistant.

There is a diversity of leptospire infecting pigs in Australia as listed in the Table below. Leptospirosis in Australian pigs has traditionally been due to either sv pomona or sv tarassovi.<sup>27</sup> However, seroprevalence to the australis serogroup suggested that sv bratislava to be involved as well. Pigs are recognised as the maintenance host for sv pomona and can be carriers, shedding the leptospire in their urine or spreading the disease by sexual contact. The prevalence of nephritis, usually associated with sv pomona infections in pigs, has increased steadily over the past decade. During that time, the prevalence of cases due to sv pomona in humans has decreased.

While attempting to isolate sv bratislava, an extremely fastidious leptospire to culture, sv hurstbridge was isolated.<sup>28</sup> Recent research into the prevalence of sv pomona in kidneys resulted in 2 new serovars isolated. Work is continuing to identify these serovars, one belonging to *L interrogans* and the other *L borgpetersenii*. An isolate belonging to *L kirschneri* was recently recovered from a sow uterus from a herd with poor reproductive performance and awaits identification.<sup>29</sup>

**Table 4 – Leptospire infecting pigs in Australia**

Species	Serovar	Serogroup	Source
<i>L interrogans</i>	pomona bratislava ?	pomona australis	isolate serology isolate
<i>L borgpetersenii</i>	tarassovi ?	tarassovi	isolate isolate
<i>L fainei</i>	hurstbridge	hurstbridge	isolate
<i>L kirschneri</i>	?		isolate

Despite not being able to isolate sv bratislava, the vaccination of sows in a large Australian commercial piggery with sv bratislava significantly reduced the number of stillborn piglets. This suggests that sv bratislava is an important reproductive pathogen in pigs.<sup>30</sup>

#### iv. Feral Pigs

20% of 195 feral pigs (*Sus scrofa*) in New South Wales were found to have seroconverted to leptospirosis. Of this, 63% reacted to sv pomona while only 2 of 195 seroconverted to sv hardjo. The remainder seroconverted to svs canicola, copenhageni, grippotyphosa, szwajizak, tarassovi and/or zanoni.<sup>31</sup>

#### v. Dogs

Clinical leptospirosis is rare in dogs, however, outbreaks have been reported, occurring in greyhounds in Launceston, Tasmania,<sup>32</sup> near Sydney<sup>33</sup> and this year in dogs in northern Queensland.<sup>34</sup> *L interrogans* sv copenhageni was isolated from a greyhound in the Sydney outbreak and isolates recently obtained from the northern Queensland outbreak are expected to be identified as *L interrogans* sv australis.

Serological surveys of dogs have shown seroconversion to a range of serogroups. A survey of dogs in Sydney in 1970-71 showed that 17.5% had positive titres of 1:100 or more to icterohaemorrhagiae, 5% to copenhageni, 1% to pomona, and 1.5% to other serovars (hardjo, tarassovi, australis, grippotyphosa and pyrogenes).<sup>35</sup> A survey of 501 dogs in southeastern Australia showed almost 10% of dogs with positive titres to leptospirosis with 16% to copenhageni, 13% to grippotyphosa, 12% to pomona, 11% to tarassovi, 3% to zanoni and 2% each to australis, canicola and hardjo.<sup>36</sup> A number of titres are probably cross-reactions. All titres to sv canicola in these 2 surveys could be accounted for a cross-reaction with sv copenhageni. A high titre to sv robinsoni was reported in a Sydney dog, which previously resided in Cairns (Qld) and was being tested for

export. This dog was most likely positive to sv robinsoni, even though serogroup pyrogenes, to which this serovar belongs, cross reacts at about 5-10% with serogroup canicola.<sup>37</sup>

Other data provided by unpublished surveys include those listed in Table 5.

**Table 5.:**

<b>No. of dogs tested</b>	<b>Cairns Pound</b>	<b>VPS Laboratory</b>	<b>Serovar (cont)</b>	<b>Cairns Pound</b>	<b>VPS Laboratory</b>
Negative	22	33			
Positive	3	9			
<b>No. of dogs reacting to serovar:</b>					
canicola	0	3	panama	1	0
copenhageni	1	5	bulgarica	0	3
hardjo	0	5	cynopteri	1	0
kremastos	0	3	zanoni	1	3
szwajizak	0	3	djasiman	1	0
medanensis	0	3	robinsoni	1	3
australis	1	5	pomona	0	2

Although literature quotes *L canicola* being isolated from 2 humans in northern Queensland, citing Sinnamon *et al*,<sup>38</sup> this serovar has been neither confirmed nor isolated from dogs in Australia. It was thought that sv canicola also cross-reacted with sv broomi and bindjei, believed to be cycling in bandicoots and rodents in northern Queensland.<sup>39</sup>

The current leptospiral vaccine in use in Australia is based on sv icterohaemorrhagiae. This serovar has common antigenicity with sv copenhageni. It is believed that adequate cross-protection is provided by sv icterohaemorrhagiae.<sup>40</sup>

#### **vi. Cats**

Clinical leptospirosis is much less common in cats despite the apparently greater seroprevalence of leptospiral antibodies in cats than in dogs (16.9% of 59 cats, 9.8% of 501 dogs) in a recent (1988-1990) survey.<sup>36</sup> These cats were positive to sv grippotyphosa, tarassovi, copenhageni, pomona and zanoni. This high prevalence and diversity of serological reaction is not surprising as cats prey upon rodents which are often maintenance hosts and carriers for leptospirosis. An earlier survey done before 1973 showed only 5 of 100 Sydney cats had titres to sv hebdomadis, hardjo, wolffi and grippotyphosa.<sup>41</sup>

#### **vii. Horses**

Seropositivity in horses can reach over 75% of the population in many horse populations around the world. Generally speaking, horses make anti-leptospiral antibodies at titres higher than those found in other species. As a result, this causes considerable cross-reaction with other serovars and confounds interpretation of serosurveys.

In Queensland, the prevalence of reactors to one or more serovars is higher in tropical areas than in subtropical areas but there is no difference in prevalence between coastal and non-coastal areas.<sup>42</sup> Horses in Australia react to svv pomona, icterohaemorrhagicae, tarassovi, hardjo, canicola, grippotyphosa and australis.

Serovar pomona was isolated from a sick foal on a farm experiencing high foal mortalities.<sup>43</sup> However clinical leptospirosis in horses in Australia is rare despite the high percentage of horses surveyed reacting to tests for antibodies to leptospire. 28.6% of 728 horses in southeastern Australia were positive<sup>36</sup> while 24.9% of horses in subtropical Queensland and 53.9% of horses in tropical Queensland reacted to the tests.<sup>42</sup>

### **viii            Other mammals**

A high serological incidence (30%) of leptospirosis, including 2 exotic serovars, has been detected in some flying fox colonies in Western Australia, New South Wales, Northern Territories and Queensland (Smythe L, *pers comm*). Table 11 lists the small mammals from which leptospire have been isolated in Australia.

## 7. International

A number of serovars not reported in Australia occur in other parts of the world. This section includes those serovars isolated and which are known to be pathogenic to animals.

Some serovars have a worldwide distribution, being reported in most countries. For example, cattle throughout the world are infected most frequently with svv hardjo (bovis) and pomona while pigs are most frequently infected with svv pomona, bratislava and tarassovi.

### a) New Zealand

Experience with leptospirosis in New Zealand has shown that some species are maintenance hosts for certain serovars (as listed in Table 6). There is no evidence of svv grippotyphosa and probably icterohaemorrhagiae in New Zealand though significant cross-reaction usually occurs between svv copenhageni and icterohaemorrhagiae. Serovar australis was isolated from a North Island farmer but no evidence of infection could be detected in his stock. Similarly, svv canicola was isolated from a person in Auckland in 1991 but only one of 6,375 dogs surveyed had low titres to this serovar.

**Table 6.**

Serovar	Known maintenance hosts
hardjo (hardjobovis)	cattle sheep
pomona	pigs
balcanica	possums ( <i>Trichosurus vulpecula</i> )
ballum	house mouse ( <i>Mus musculus</i> ) ship rat ( <i>Rattus rattus</i> ) hedgehog ( <i>Erinaceus europaeus</i> )
copenhageni	Norway rats ( <i>Rattus norvegicus</i> )
tarassovi	pigs

#### i. Cattle

The main serovars causing clinical leptospirosis in cattle are svv pomona, copenhageni and hardjo (serotype hardjobovis).<sup>44</sup> Serovar hardjobovis presents a different clinical picture in New Zealand when compared with its Australian counterpart. The reason for this is not known.<sup>45</sup> Serovars copenhageni and ballum have been isolated from the urine of healthy calves. These calves were symptomless carriers of these two serovars.<sup>46</sup> Serovar ballum has been isolated from hedgehogs in dairying districts and they are suspected to be a major reservoir for this serovar in New Zealand.<sup>47</sup> Only svv pomona has been implicated as a cause of leptospiral abortion in cattle and some infected cows were still shedding leptospire a month later.<sup>48</sup> Some rodents trapped near dairies on the North Island were found to carry svv copenhageni and ballum.<sup>49</sup>

In an investigation of leptospirosis at an AI Centre, bulls were found to be infected with sv hardjo and pomona while a survey of wildlife in the area, mainly rabbits, hares, possums and hedgehogs, showed ballum to be the main infecting serovar.<sup>50</sup>

The incidence of human leptospirosis has decreased in New Zealand, especially in dairy farmers for whom it has become a significant occupational risk. Serovars hardjo and pomona are the main serovars infecting humans.<sup>51</sup> Five hundred and eighty-one cases of leptospirosis in humans were reported in 1981.<sup>52</sup> The number of cases (confirmed and probable) dropped to around 200 in 1990 and is now around 100 per year.<sup>53</sup>

The experimental infection of sheep and cattle with sv balcanica, normally found in possums in the North Island, showed that it was possible for balcanica infection to occur sporadically but it was unlikely to be maintained endemically in these two species.<sup>54</sup>

## **ii. Sheep and goats**

Clinical leptospirosis due to sv pomona has occurred in sheep. Outbreaks due to this serovar were reported in lambs, causing haemolytic disease and deaths.<sup>55</sup> It was noted that there were significant differences in the clinical response to the infection in sheep of different haemoglobin types.<sup>56</sup> Serovar hardjo has been isolated from healthy sheep.<sup>57</sup>

Serum samples collected from 428 goats and subjected to the MAT for antibodies to *Leptospira* showed that 70% of all sera had antibody titres, particularly to bratislava, copenhageni and ballum. No isolates were recovered from urine collected from 120 goats.<sup>58</sup>

## **iii. Pigs**

An abortion storm due to sv pomona has been reported in pigs. Neither streptomycin nor oxytetracycline was effective in treating leptospiruria in these pigs.<sup>59</sup> This serovar has also caused clinical leptospirosis, mainly nephritis, in recently introduced grower pigs in several grower houses.<sup>60</sup>

Serovar tarassovi has been isolated from a pig in the North Island.<sup>61</sup> Little is known about the epidemiology of this serovar in pigs in New Zealand. Pigs have shown seropositivity to sv bratislava but attempts to isolate this organism have not been successful.<sup>62</sup>

## **iv. Dogs**

Outbreaks of clinical leptospirosis have been reported in dogs in New Zealand. In one case, four of 35 hounds developed anorexia, jaundice and depression, and two of these died. Twenty-six hounds had MAT titres to sv icterohaemorrhagiae but not to sv tarassovi. Yet sv tarassovi was later isolated from the urine of 4 healthy hounds and a retest of these 4 dogs showed them to have seroconverted to sv tarassovi. In a trial where healthy dogs were injected with sv tarassovi, these dogs developed symptomless leptospiruria persisting for over 7 months with low levels of antibodies. It was suspected that the leptospirosis in the hounds was due to sv copenhageni.<sup>63</sup>

Serovar pomona was isolated from working dogs on a dairy where there was an outbreak of leptospiral abortions due to the same serovar.<sup>51</sup>

Serovar canicola has not been isolated or reported in dogs in New Zealand but this serovar has been isolated from a human.<sup>64</sup> This person had not been overseas, but lived on a small semi-rural block and had been in contact with a variety of animal species.

In a nationwide survey carried out during 1990-91 of more than 5800 dogs to detect antibodies against sv copenhageni, ballum and canicola, only one weak reactor against canicola was found. Even though only a small percentage of dogs seroconverted, sv copenhageni was the most prevalent.<sup>65</sup>

#### **v. Horses**

In 1989, a mare that aborted was found to have high titres to sv copenhageni and lower titres against several other serovars. A serological survey of in-contact horses followed and 2 had high titres against sv hardjo. Another survey involving 762 blood samples collected between 1988 and 1990 showed significant titres against sv pomona, copenhageni, hardjo and bratislava, suggesting the occurrence of sporadic leptospiral infections in horses in New Zealand.<sup>66</sup>

Of 781 equine abortion investigations in New Zealand from 1974 to 1990, 9 were diagnosed as being due to sv pomona.<sup>67</sup>

#### **vi. Deer**

Haemolytic disease and death due to sv pomona has been reported in red deer calves.<sup>68</sup> Leptospiuria in some deer persisted for at least 8 months. Leptospiral abortion was suspected in a mob of red deer and sv copenhageni and hardjo were isolated from the urine of some deer.<sup>69</sup>

Serosurveys of red deer on both North and South Islands showed significant MAT antibody titres to sv ballum, bratislava and copenhageni. Sv bratislava has not been isolated in New Zealand and could be due to a cross-reaction with another strain.<sup>70</sup>

#### **vii. Possums**

There is a high prevalence of sv balcanica in brush tailed possums in the North Island with a survey showing cultures isolated from 38% of 127 possums or 64% of mature adults. It was not possible to recover leptospire from juvenile possums.<sup>71</sup> Possums are believed to be maintenance hosts for this serovar as agglutinating antibodies and leptospiruria in infected animals lasted for over a year. Adult possums appear to be symptomless carriers. Young possums did not react at all to infection with sv hardjo.<sup>72</sup> Serovar balcanica is believed to spread by being transmitted venereally between possums and is being considered as a biological control agent for possums.<sup>73</sup>

Although it was originally identified as sv balcanica, the possum strain has been found to be genetically different from the Bulgarian reference strain and should be renamed to avoid confusion.

## **b) Europe**

Recent research using nucleic acid detection tests has shown that all isolates belonging to the European grippotyphosa serogroup belong to the genomospecies *L kirschneri*. Moreover, studies with monoclonal antibodies have shown that many isolates belonging to sv mozdok can be classified into three different types.<sup>74</sup>

### **i. Cattle**

Sv hardjoprajitno has been isolated from cattle in Northern Ireland and Scotland.<sup>75</sup> The distribution of this serovar in Europe is difficult to determine because of its close antigenic relationship with sv hardjobovis. This serovar was probably associated with abortions in sheep,<sup>76</sup> pigs,<sup>77</sup> and a horse.<sup>78</sup> Sv hardjoprajitno is usually transmitted venereally and has been associated with abortion storms in cattle. There is no indication of this serovar occurring in Switzerland<sup>79</sup> but it appears to occur in Germany where sv hardjo has been isolated from the urine of cattle where 18 of 31 cattle aborted.<sup>80</sup>

*L kirschneri* sv mozdok (sg pomona) is closely related to sv pomona and has been associated with a small outbreak of abortion in southwest England. This serovar appears to be the only serovar belonging to sg pomona in Great Britain while sv pomona has been isolated in Northern Ireland. Sv mozdok has been isolated from rodents in Bulgaria<sup>81</sup> and is regarded as a serovar maintained by free-living species in continental Europe.

A new unidentified leptospire belonging to *L borgpetersenii* sg sejroe was isolated from kidneys of healthy buffaloes from several farms in Central Italy.<sup>82</sup>

### **ii. Pigs**

*L kirschneri* sv mozdok is pathogenic to pigs, causing abortions and stillbirths.<sup>83</sup> It has been associated with significant reproductive losses in pigs in Poland,<sup>84</sup> Portugal<sup>85</sup> and England.<sup>86</sup>

*L interrogans* sv lora (sg australis) was isolated from pigs in the Netherlands following an outbreak of abortions in the last month of pregnancy and the birth of dead or non-viable piglets.<sup>87</sup> Muskrats are probably the maintenance host for this serovar.<sup>88</sup> This serovar has been isolated in Bulgaria.<sup>89</sup>

*L interrogans* sv bratislava and muenchen (sg australis) were associated with a number of abortions and still births in pigs in Northern Ireland during 1981-82, accounting for 91% of isolates from 55 of 78 litters examined.<sup>90</sup> Persistence was observed in renal and genital tissues for up to 147 days after abortion.<sup>91</sup> Leptospire could be isolated from several locations within the urogenital tract of boars from herds with leptospiral abortions. Sv bratislava was isolated from the testes of a boar and sv muenchen was isolated from the urethra of a boar.<sup>92</sup>

*L borgpetersenii* sv guidae (sg tarassovi) has been isolated from a mummified foetus during an investigation into the haematological and nutritional aspects of porcine abortions in the Netherlands.<sup>93</sup>

An unidentified serovar belonging to sg sejroe has been isolated in Germany recently following an investigation into a swine herd where some 40% of sows returned to service, over 10% of piglets were stillborn, and 15% of piglets died.<sup>94</sup>

A recent serosurvey Switzerland has shown svv bratislava and bataviae to be endemic in the swine population while sv pomona seems to have been eliminated.<sup>95</sup>

### iii. Horses

Clinical leptospirosis occurs in horses in Europe. Sv grippotyphosa and an untyped serovar belonging to sg australis have been isolated from the vitreous humour of horses' eyes in Germany.<sup>96</sup> In 1998, leptospire belonging to these two serogroups could be isolated from 33 out of 130 vitreous humour samples.<sup>97</sup> Leptospire belonging to sg grippotyphosa were isolated from the urine of a horse suffering from acute haemolytic anaemia.<sup>98</sup>

In an investigation of 50 abortions that occurred between 1979 and 1987 in the United Kingdom, 24 were positive for leptospiral infection. Leptospire isolated from infected foetuses included *L interrogans* svv bratislava, muenchen, pomona, canicola, hardjo (hardjoprajitno?) and *L borgpetersenii* sv arborea.<sup>99</sup> Horses are possibly maintenance hosts for sv bratislava in Northern Ireland with this serovar having been isolated from a mare and 73% of 650 mares tested positive for this serotype.<sup>100</sup>

In Italy, 31 (73.8%) of 42 horses were positive for leptospirosis. The serotype most commonly involved was bratislava.<sup>101</sup> In 1985-1986, some 108 of 488 horses were seropositive to the MAT. Of the 108 positive horses, 78 (16%) had antibodies to sv bratislava. Leptospire were isolated from the kidneys of 57 (28%) of the 202 slaughter horses.<sup>102</sup>

### iv. Dogs

Clinical *L interrogans* sv canicola infection can occur in dogs in Europe and is often the most common serovar detected serologically, having been isolated from dogs in Scotland<sup>103</sup> and Greece.<sup>104</sup> However a serosurvey of 192 dogs in Switzerland showed only isolated reactions to this serovar.<sup>105</sup>

In Italy, sv sejroe was the cause of asymptomatic renal infection in laboratory dogs with no clinical signs of leptospirosis but with significant titres to this serovar.<sup>106</sup>

### v. Sheep

In Portugal, most sheep are kept outdoors and have regular contacts with dogs owned by their shepherds. This high level of contact is probably the main cause of the high seroprevalence of sv canicola in sheep infected with leptospirosis.<sup>107</sup>

Sv bratislava has been isolated from sheep in both England and Northern Ireland.<sup>108</sup> Sheep develop inapparent infection to leptospire of the australis serogroup.<sup>109</sup>

In Northern Ireland, sv hardjo (probably hardjoprajitno) was detected in aborted fetuses in a dairy herd where abortions began after the introduction of sheep from a flock where some ewes had aborted.<sup>110</sup> Sv hardjo (probably hardjobovis) has caused agalactia in several mobs of ewes in Northern Ireland.<sup>111</sup>

In Switzerland, 17.8% of 1427 sheep on 60.5% of 205 farms were positive to leptospirosis. Of these, 29.5% was due to sv bataviae, 17.3% to sv bratislava and 26.4% to sv hardjo. Twenty-eight percent of 250 goats on 64% of 74 farms were positive to both sv bataviae (66.2% of 71 positive goats) and bratislava (26.8% of 71 positive goats).<sup>95</sup>

#### vi. Other mammals

In a Zurich zoo, 19 Canadian beavers died from acute icterohaemorrhagiae infection over 3.5 years. Rats were considered to be the source of infection.<sup>112</sup>

A leptospire, strongly suspected to belong to sv bratislava, was isolated from the thoracic fluid, aqueous humour and kidney of a cat in Britain (Ulster). Autopsy showed severe pathological changes in several tissues, including lung, liver, kidney and brain.<sup>113</sup>

#### vii. Small mammals

A number of serovars have been isolated from apparently healthy small mammals. Most are symptomless carriers, shedding leptospire in their urine. The common vole (*Microtus arvalis*) is often found on dairy farms in Europe and is possibly a maintenance host for sv grippotyphosa but not for sv hardjo.<sup>114</sup>

Table 7 below is a summary of the serovars isolated from various types of small mammals in Europe.

**Table 7.**

Type of small mammal	Serovar isolated	Countries serovar isolated
Muskrats	saxkoebing grippotyphosa copenhageni lora	Belgium Belgium, Netherlands Belgium, Netherlands Netherlands
Rodents	mozdok	Bulgaria
Rats <i>Rattus norvegicus</i>	icterohaemorrhagiae bratislava copenhageni	Great Britain Northern Ireland Russia
Field mice <i>Apodemus sylvaticus</i>	australis javanica hebdomadis mozdok grippotyphosa	Great Britain Great Britain Great Britain Poland, Portugal Croatia
Voles <i>Microtus agrestis</i> <i>Clethrionomys glareolus</i>	javanica pomona (mozdok) muenchen	Great Britain Great Britain Great Britain

Type of small mammal	Serovar isolated	Countries serovar isolated
	grippotyphosa	Croatia
Hedgehogs <i>Erinaceus europaeus</i>	australis bratislava icterohaemorrhagiae	Great Britain Great Britain, Greece, Italy Greece
Badgers <i>Meles meles</i>	australis javanica hebdomadis	Great Britain Great Britain Great Britain
Coypu <i>Myocastor coypus</i>	icterohaemorrhagiae hebdomadis	Great Britain Great Britain

## c) Africa

### i. Cattle

Serovar hardjoprajitno, slightly different from the reference strain, was isolated from the urine of a butcher serologically positive for leptospirosis in Nigeria. A new strain belonging to sg pyrogenes was recovered from 5 of 6 isolates collected from kidneys of cattle slaughtered at an abattoir in Nigeria.<sup>115</sup> A survey for pathogenic leptospires in kidneys of cattle resulted in the identification of a new serovar, sv nigeria.<sup>116</sup>

In South Africa, sv hardjo (probably hardjoprajitno) was isolated from urine of cattle where an abortion outbreak had occurred as early as at 4 months gestation.<sup>117</sup>

A number of new serovars were isolated from kidneys of healthy ox slaughtered in Zimbabwe and identified. These are listed in Table 8 below and are regarded as African strains of leptospires.<sup>118</sup>

Table 8 Serovars recently identified in Zimbabwe

Genomospecies / Serogroup	<i>L borgpetersenii</i>	<i>L kirschneri</i>
Pyrogenes	kwale mombe strain closely related to sv nigeria	
Hebdomadis	marondera mhou	
Tarassovi	ngavi	
Sejroe	balcanica hardjo	
Icterohaemorrhagiae		zimbabwe
Australis		fugis
Bataviae		paidjan
Pomona		strain closely related to sv mozdok

## ii. Pigs

In South Africa, sv canicola has been isolated from pigs where high mortality, low survival weight at weaning, hypogalactia, infertility and irregular oestrus were observed. One hundred and forty-nine of 240 pigs seroconverted to this serovar. Dogs in the area had emaciation, high fever, anaemia and nephritis and hygiene in all cases were poor.<sup>119</sup>

## iii. Dogs

Serovars canicola and icterohaemorrhagiae were isolated from dogs with clinical leptospirosis in Tanzania.<sup>120</sup>

## iv. Horses

In South Africa, 5 out of the 13 horses (38%) on a farm were seropositive to sv pomona. This serovar was isolated from a porcine foetus and from renal lymph nodes of slaughter pigs with chronic nephritis from the same farm.<sup>121</sup>

## v. Other mammals

The Gambian Giant Pouched Rat (*Cricetomys gambianus*) was found to be symptomless carriers for an unidentified serovar of the icterohaemorrhagiae serogroup.<sup>122</sup> In Zambia, a number of wild animals, including african buffalo, hippopotamus, roan antelope, kudu and waterbuck had antibodies to leptospirosis<sup>123</sup> but there is no report of isolation and identification of leptospire from the numerous species found in Africa.

## d) Asia

*L interrogans* sv lai (sg icterohaemorrhagiae) is the major leptospiral serovar of China<sup>124</sup> and Korea<sup>125</sup> and can cause pulmonary haemorrhaging, a traumatic consequence of leptospirosis, in humans. It has been isolated from field mice (*Apodemus agrarius*) in both countries.<sup>126</sup> A total of 18 serogroups and 70 serovars of pathogenic leptospire, including 35 new serovars, have been identified in China.<sup>124</sup> Rodents appear to be the main reservoir hosts for serogroup icterohaemorrhagiae whilst domestic animals carry the pomona serogroup.<sup>126</sup> A leptospire strongly resembling sv lai has been isolated from a human in the Andaman Islands off India.<sup>127</sup>

In Taiwan, a serosurvey of humans involved in the livestock industry showed that *L santarosai* sv shermani to be the main serovar infecting humans. This serovar can cause chronic renal failure in humans and the main risk factor was humans using water from sources other than tapwater.<sup>128</sup>

Very little work has been done on leptospirosis in the Philippines. Endemic *Leptospira* serovars have not been properly identified however recent serosurveys suggest that *L borgpetersenii* svs poi and tarassovi could be endemic in humans.<sup>129</sup>

In Vietnam *L. santarosai* sv *canalzonae* was isolated from the blood and urine of an American serviceman with clinical hepatitis and who had participated in a study of zoonotic diseases involving the trapping of wild animals.<sup>130</sup>

### i. Cattle

There is very little information on the effects of pathogenic strains in cattle. The following table summarises the pathogenic strains isolated from cattle in some Asian countries.

In India, buffaloes with history of abortions and repeat breeding showed high titres to sv *shermani*.<sup>131</sup>

Table 9.

Country	Species	Pathogenicity	Serovar
Japan	bovine <sup>132</sup>	Not known	kremastos
China	yaks <sup>133</sup>	Clinical disease with 10-20% incidence and mortality of 30-50%	pomona
Israel	dairy cattle <sup>134</sup>	Clinical disease in dairy workers	hardjo
Malaysia	bovine <sup>135</sup>	Healthy animals	unipertama canicola australis javanica ballum pomona hardjo (bovis)
India	buffalo <sup>136</sup>	Clinical jaundice and death	andaman (non-pathogenic spp)

### ii. Pigs

In China, sv *canicola* is the most common leptospire isolated from the urine of breeder pigs. This is in contrast to the common occurrence of sv *pomona* in fattening pigs.<sup>137</sup>

In India, leptospires belonging to serogroups *autumnalis*, *hebdomadis*, *javanica* and *tarassovi* have been isolated from several species of animals, especially pigs.<sup>138</sup> Serovars *icterohaemorrhagiae* and *tarassovi* have been isolated from urine and kidney samples of healthy pigs.<sup>139</sup>

In Sri Lanka, leptospires belonging to serogroups *javanica*, *pomona*, *pyrogenes* and *canicola* have been isolated from pigs, rat and humans.<sup>140</sup>

In Israel, sv canicola has been isolated from rats (*R norvegicus*) trapped in piggeries.<sup>141</sup>

In the Philippines, an unidentified leptospire belonging to serogroup pyrogenes was isolated from a pig.<sup>142</sup> Also sv pomona was isolated from one of the sows that aborted on a farm where an outbreak of clinical leptospirosis had occurred.<sup>143</sup>

### iii. Horses

*L australis* was isolated from a horse in the Philippines.<sup>144</sup>

### iv. Small mammals

The leptospires isolated and identified in small mammals in Asia are listed in the following table.

**Table 10**

Country	Small mammal species	Serovar
India	Rat ( <i>R rattus</i> )	javanica <sup>145</sup> japonica <sup>146</sup>
	Laboratory albino mice	autumnalis
	Laboratory wistar rats	javanica
	Laboratory guinea pigs	autumnalis
	Laboratory rabbits	autumnalis and javanica <sup>147</sup>
Israel	Rats ( <i>R norvegicus</i> )	canicola <sup>141</sup>
Korea	Field mice ( <i>Apodemus agrarius</i> )	lai <sup>125</sup>
Vietnam	Rats ( <i>R norvegicus</i> )	bataviae <sup>148</sup>

### v. Other mammals

Clinical leptospirosis has not been reported in camels (*Camelus dromedarius*). Three of 73 (4.1%) racing camels in the UAE tested for leptospirosis using the slide macroscopic test had leptospiral antibodies.<sup>149</sup> Similar results were obtained in other serosurveys in the UAE where 2.5% of racing mares and 5.6% of breeding camels had leptospiral antibody titres.<sup>150</sup> However, elsewhere, higher incidence of anti-leptospiral antibodies has been reported in camels including Afghanistan (44.4%)<sup>151</sup> and Ethiopia (15.4%).<sup>152</sup>

## e) North America

### i. Cattle

As with Australia, *L borgpetersenii* sv hardjo subtype hardjobovis is widely distributed in cattle in USA and Canada and has not been associated with abortions. While only subtype A has been

isolated in Australia, both subtypes A and B have been isolated in USA. Similarly, sv pomona is widespread in USA but is not as prevalent as hardjobovis. Nevertheless, infection with sv pomona in cattle is due to either subtype kennewicki A or kennewicki B, both of which can cause abortions.<sup>153</sup> Other serovars isolated from cattle in USA include canicola, grippotyphosa, icterohaemorrhagiae, szwajizak and balcanica.<sup>154</sup> Sv szwajizak can cause clinical leptospirosis in 4-week-old heifer calves,<sup>155</sup> mastitis in cows and urinary shedding only during leptospiraemia.<sup>156</sup> However, restriction endonuclease analysis (REA) of svszwajizak and balcanica has resulted in these two serovars being reclassified as svsgorgia and hardjobovis respectively.<sup>157</sup> Subtype hardjobovis is also widespread in Canada while sv pomona infections occur focally in several localities. A serovar antigenically related to sv icterohaemorrhagiae has been detected in bovine specimens.<sup>158</sup>

It has been stated in several references that subtype hardjoprajitno does not occur in North America. However, in USA, it has been isolated from mares that aborted following a flooding incident in San Diego county, near the Mexican border. Some of the floodwaters originated from Mexico where this serovar has been isolated from cattle.<sup>159</sup>

In Canada, calf crop losses of up to 20% have been associated with a high prevalence of antibodies to sv hardjo in cattle herds from which this serovar was isolated.<sup>160</sup> Although this suggests that subtype hardjoprajitno was the likely infective agent, all Canadian isolates belonging to serogroup sejroe were identified as subtype hardjobovis A.<sup>158</sup>

Subtype kennewicki caused clinical leptospirosis in teenage children swimming in an irrigation channel near where a herd of cows were pastured. However, no cases of leptospirosis were reported in any of the cattle in the area.<sup>161</sup>

## **ii. Pigs**

Serovar bratislava was isolated from kidneys and uterus of sows not exhibiting clinical disease<sup>162</sup> and from stillborn and weak piglets and placentas from swine in herds with a history of reproductive losses.<sup>163</sup> Serovars kennewicki and grippotyphosa were isolated from swine herds with sporadic reproductive failure.<sup>164</sup>

In Mexico, antibodies to sv shermani were common and found in 13.5% of 4354 serum samples collected during 1975-1984.<sup>165</sup> Although pomona was the most important serovar affecting pigs in Mexico, antibodies to svspanama, icterohaemorrhagiae, autumnalis, australis, canicola, hardjo, orleans, zanoni, louisiana and arborea were also detected.<sup>166</sup>

## **iii. Horses**

An investigation of equine abortions over three foaling seasons (1991-1993) in Central Kentucky showed that 74 of 2264 abortions were diagnosed as leptospirosis. Leptospire were isolated from 45 of these cases and were identified as svskennewicki (43), grippotyphosa (1) and a serovar antigenically similar to sv pomona.<sup>167</sup> An earlier investigation over four foaling seasons (1987-1990) in the same area showed that 58 of 2266 abortions were due to leptospirosis. Only 3 of 14 isolates

cultured could be identified and were all sv kennewicki. Serology suggested that other serovars, namely grippotyphosa and bratislava were also involved.<sup>168</sup>

Sv hardjoprajitno was isolated from mares that had aborted following severe regional flooding near San Diego. An interesting feature of this outbreak was that many of the samples had high agglutinating antibody titres against sv bratislava and pomona, significant titres to icterohaemorrhagiae and grippotyphosa and low, but positive, titres to hardjo and canicola. Serovar bratislava has never been isolated from horses in USA.

#### **iv. Dogs**

Serovar *canicola* has not been isolated from animals or humans in USA or Canada, though there were reports of dogs infected with svs *canicola* and *icterohaemorrhagiae* in Canada in the late 1940's and early 1950's.<sup>169</sup> There is no evidence of clinical leptospirosis to this serovar although seroconversion has occurred in some animals, including horses in New York State<sup>170</sup> and wolves (*Canis lupus*) in Minnesota.<sup>171</sup> Most dogs that had seroconverted to sv *canicola* also seroconverted to sv *icterohaemorrhagiae*. This suggests that the dogs had been vaccinated against both serovars.

Serovars isolated from dogs include:

- *icterohaemorrhagiae* from a stray dog;<sup>172</sup>
- *grippotyphosa* and *ballum* from a foxhound pup from a litter of pups, all with illthrift;<sup>173</sup>
- *bratislava* from the urine of a dog with clinical leptospirosis. There was a high seroprevalence to this serovar in dogs in Illinois;<sup>174</sup>
- *kennewicki* from a dog with clinical leptospirosis. The source was probably raccoons.<sup>161</sup> This serovar was also isolated from kidneys of red foxes (*Vulpes vulpes*) with nephritis in Canada.<sup>175</sup>

#### **v. Small mammals**

Serovars isolated from small mammals include

- *kennewicki* from skunks.<sup>176</sup>
- *icterohaemorrhagiae* from rats (*R norvegicus*) in Colorado, USA;<sup>177</sup>
- svs *icterohaemorrhagiae*, *ballum* and *sejroe* following the testing of nearly 3000 rodents and mongooses in Hawaii;<sup>178</sup>
- *icterohaemorrhagiae* from a rat on a pig farm in Mexico.<sup>179</sup>

#### **vi. Other mammals and reptiles**

Serovar *tarassovi* was isolated from 12 of 20 kidney suspensions and 6 of 20 cloacal suspensions of turtles (*Pseudemys scripta-elegans*) trapped in sewage settling ponds. Forty-two of 46 turtles were seropositive to this serovar.<sup>180</sup>

*L interrogans* sv *pomona* was isolated repeatedly from Californian sea lions and Northern fur seals during a 5-year study on diseases of pinnipeds.<sup>181</sup>

### **f) South America**

#### **i. Cattle**

In Brazil, an investigation of abortions in 4 Holstein dairy herds showed that 72 of 120 aborted foetuses had evidence of leptospiral infection. Leptospiras isolated from 15 foetuses include *L interrogans* svs *hardjo* (4), *pomona* (3) and *wolffi* (8). Serovar *wolffi* is prevalent in cattle in Brazil.<sup>182</sup> Serovar *castellonis* (belonging to serogroup *ballum*) was isolated from the kidney of an aborted foetus.<sup>183</sup>

A number of serovars have been isolated from healthy slaughter cattle in several countries of South America. These include sv guaicuris, goiano, icterohaemorrhagiae and hardjo in Brazil,<sup>184, 185</sup> hardjobovis A & B and kennewicki in Chile,<sup>186</sup> grippotyphosa, canicola, tarassovi, javanica, pomona, panama and broomi in Cuba,<sup>187, 188</sup> peruviana, hardjo, pomona, hebdomadis, bataviae, and pyrogenes in Peru.<sup>189, 190, 191</sup> Serovar pomona was associated with an abortion outbreak in cattle on a farm in Argentina.<sup>192</sup>

A leptospire cultured from the kidney of an aborted bovine foetus was typed as ballum sv castellanis by endonuclease restriction. It is suggested that ballum may occur sporadically in southern Chile, where pomona and hebdomadis are the prevalent serogroups in cattle.<sup>193</sup>

Leptospiral antibodies against sv shermani occur in cattle on dairy farms in Bolivia.<sup>194</sup> Antibodies to this serovar have also been detected in sheep, goats and dogs in that country and was the most common serovar detected in dogs in the southeast Bolivia.<sup>195</sup>

## **ii. Pigs**

Serovar pomona was isolated from an aborted foetus and was reported to be the cause of an outbreak of abortions and stillbirths in sows on a farm in Brazil.<sup>196</sup> In Peru, 31 of 240 sows aborted over a 5-month period in 1988. Some sows had very high titres to sv canicola and high titres of antibody to other leptospiral serovars (celledoni, pomona, copenhageni and castellanis). Serovar canicola was isolated from the urine of four sows that had aborted and the kidney of one slaughter pig. It was suggested that the infection was brought onto the farm by wild animals.<sup>197</sup>

Serovars isolated from healthy pigs in the various South American countries include:

- tarassovi, pomona and canicola from 70 of 130 kidneys from apparently healthy pigs over 3 years in Argentina;<sup>198</sup>
- pomona, guidae, canicola, icterohaemorrhagiae and tarassovi in Brazil;<sup>199, 200</sup>
- mozdok in Cuba;<sup>201</sup> and
- san-martini, pomona, icterohaemorrhagiae and canicola in Venezuela.<sup>202, 203</sup>

## **iii. Horses**

Three pathogenic leptospire were isolated from apparently normal horse kidneys collected at an abattoir in Argentina. They were antigenically and serologically homologous to serotype hardjo. This is the first known report of an isolation of this serotype from horses. Serological tests were also carried out on randomly collected abattoir serum samples from 245 horses to determine the prevalence of equine leptospirosis. Significant antibody titres (1:100 or greater) were found in 74.6% of the samples. Predominant reactions occurred with the antigens belonging to pomona, hebdomadis, pyrogenes, tarassovi, and canicola serogroups.<sup>204</sup>

In Venezuela, micro-agglutination tests of serum samples from horses with abortion histories showed the predominant serotypes to be pyrogenes, ballum, pomona and canicola.<sup>205</sup>

Serum samples collected in 1979 and 1980 from 1653 racehorses in Brazil had positive leptospiral agglutinin titres in 75 (4.5%) of horses, mainly against serotypes icterohaemorrhagiae and javanica (with small numbers of positive reactions to seven other serotypes). Sv icterohaemorrhagiae was isolated from a foetus with haemorrhagic lesions.<sup>206</sup>

#### **iv. Dogs**

In Barbados, sv bim was isolated from a febrile stray dog and subsequently from 6 human patients with leptospirosis.<sup>207</sup> This serovar can cause acute and sometimes fatal disease with pulmonary haemorrhaging in humans.<sup>208</sup> Several dogs vaccinated against leptospirosis died of, or became severely ill with, leptospirosis and were all seropositive to serogroup autumnalis. Sv bim, which was not included in the vaccine, was strongly suspected to be the cause of infection.<sup>209</sup>

In Brazil, 32 of the 35 isolates collected from 1415 stray dogs were identified as sv canicola, the others were svs copenhageni (2) and pomona (1).<sup>210</sup> It was noted that, generally speaking, sv icterohaemorrhagiae or copenhageni causes icterus and uraemia in dogs with clinical leptospirosis while sv canicola causes uraemia but not icterus.<sup>211</sup>

Serovars isolated from stray dogs in Trinidad were typed as portland-verre, canicola, copenhageni and georgia. Sv canicola was also isolated from a cat.<sup>212</sup>

In Venezuela, micro-agglutination tests of serum samples from dogs showed the predominant serotypes to be canicola, pyrogenes and ballum.<sup>213</sup>

#### **v. Small mammals and reptiles**

Many small mammals native to South America are suspected to be carriers of leptospire. Toads and frogs are possible carriers of some pathogenic leptospire as well. Serovars bim and bajan have been isolated from cane toads (*Bufo marinus*) and whistling frogs (*Eleutherodactylus johnstonei*) in Barbados.<sup>214</sup> Of 198 toads tested with MAT, 21% was seropositive with 50% belonging to sg australis, 23% to sg autumnalis, and 13% to sg panama.<sup>215</sup> However, it is not known what role toads and frogs have in the epidemiology of leptospirosis.

Serum samples collected from poisonous and non-poisonous snakes have shown seropositivity to several pathogenic leptospiral serotypes including javanica, ballum, brasiliensis, grippotyphosa, wolffi, tarassovi, pomona, pyrogenes and shermani serovars.<sup>216, 217</sup> However the role of snakes in the epidemiology of leptospirosis has not been elucidated.

Species of pathogenic leptospire have been isolated from armadillos in Argentina,<sup>218</sup> philander opossums in Brazil<sup>219</sup> and mongooses in Barbados,<sup>220</sup> Trinidad and Grenada.<sup>221</sup> Leptospire have been isolated from rodents in several South American countries, including species exotic to Australia, such as sv fort-bragg in rats in Barbados,<sup>222</sup> mozdok in rats and mice in Cuba<sup>223</sup> and budapest in rats in Peru.<sup>224</sup>

#### **vi. Camelids**

Serological tests of guanacos (*Lama guanicoe*) and sheep sharing a reserve in Argentina for 17 serovars showed the guanacos to be serologically negative to leptospirosis while several sheep were found to have antibody titres to leptospirosis.<sup>225</sup> Alpacas reared on breeding farms in Peru were monitored for leptospirosis from 1993 to 1996. None showed any clinical evidence of leptospirosis yet 6.54% of 810 serum samples were positive to 7 serovars. However it was noted that the proportion of reactants was significantly lower in the dry season than in the wet season.<sup>226</sup>

### **g) Experimental infection in animals**

Experimental infection of cats with svv icterohaemorrhagiae and canicola did not cause clinical disease. Anti-leptospiral agglutinins were detected in 90% of infected cats to both serovars for 8 to 12 weeks, starting 1 week after infection. Only those cats infected with canicola had leptospiruria beginning 2 to 4 weeks after infection and lasting for 2 to 8 weeks.<sup>227</sup>

The experimental infection of lactating goats with svv pomona and hardjo caused mild clinical signs (pyrexia and reduction in milk yield) in some animals. Serovar hardjo was isolated only from the mammary gland of 1 of 4 goats 13 days after infection.<sup>228</sup> In another trial, the experimental infection of goats with svv autumnalis, australis, hardjo and pomona caused a small temperature rise but no other clinical signs or pathological lesions could be detected. However MAT antibody titres were first detected 7 days after infection with autumnalis, australis and hardjo, and on day 15 after infection with pomona. By day 90, the antibody titres were declining in all infected goats.<sup>229</sup>

Sows experimentally infected with sv pomona developed antibody titres 11 days after infection and showed no clinical signs of infection apart from increased body temperature. The titres peaked 12 to 27 days post infection. PM examination showed kidney lesions, atypical mastitis and focal subacute to chronic nephritis in some of the infected sows. Leptospire could be isolated from one of the piglets but none developed antibody titres.<sup>230</sup>

Each group of pregnant gilts was experimentally infected with one of four serovars, muenchen, bratislava, copenhageni and a leptospire from the pomona serogroup. Antibody titres rapidly declined after 33 days with a lower plateau of titres being maintained in the chronically infected pigs. However considerable cross-reactions with other serovars occurred during serological testing. All gilts were susceptible to infection with some showing mild clinical symptoms. Reproductive disease, persistent leptospiruria and severe renal lesions were seen only in those infected with the serovar from the pomona serogroup. The leptospiruria due to other serovars was inconstant and of lower intensity. There was no evidence of transplacental infection with the first three serovars while leptospire of the pomona serogroup could be isolated from the aborted fetuses.<sup>231</sup>

Cows mated to a serologically negative bull and then infected with *L borgpetersenii* sv hardjo by intrauterine infusion all seroconverted to leptospirosis by 35 days after mating but leptospire could not be isolated from daily blood samples, weekly urine samples, kidneys and uterus. Pregnancy rate was not affected.<sup>232</sup>

### **h. Infection in pinnipeds**

Serovar pomona (kennewicki) has been detected as a cause of abortions and deaths among free-living and captive Californian sea lions (*Zalophus californius*) along the California and Oregon coasts in USA since 1970 with MAT levels as high as 1:6400.<sup>233, 234, 235</sup> Serovar hardjo was the most frequently detected serotype in California sea lion pups sampled in several rookeries in the Gulf of California.<sup>236</sup> Other pinnipeds infected by leptospirosis include Pacific harbour seals (*Phoca vitulina richardsii*) at a rehabilitation centre which were suspected to have been infected by sv grippotyphosa. As it is most likely that survival of leptospire in sea water is limited, it is likely pinnipeds become infected as a result of contact with stagnant pools of water contaminated with leptospire or acquired through the food chain.

## 8. Leptospires

While this section concentrates on leptospires found in Australia, it also includes those serovars which our trading partners require Australia to test for prior to exporting livestock.

Although many serovars are recognised globally, only a limited number are usually endemic to a particular region. Pathogenic leptospires are usually found in the proximal convoluted tubules of the kidney and, in some animal hosts, the genital tract (mammary glands, uterus, uterine tube, testes and vesicular glands). Outside the host animals, the optimal conditions for the survival of the leptospires are moist, warm (optimal 28 C), and neutral or mildly alkaline conditions.

The table below lists the serovars isolated in Australia (Smythe L, *pers comm*).

**Table 11: Pathogenic leptospires isolated in Australia**

SEROGROUP	SEROVAR	MAN	ANIMAL HOSTS	
			Domestic	Wild
Australis	australis	+	Cattle	<i>Hydromys chrysogaster</i> <i>Perameles nasuta</i> <i>Isoodon macrourus</i> <i>Rattus sordidus</i> <i>Rattus fuscipes</i> <i>Rattus rattus</i> <i>Mus musculus</i> <i>Uromys caudimaculatus</i>
Autumnalis	bulgarica	+		
Canicola	canicola	+		
	broomi	+		<i>Isoodon macrourus</i> <i>Rattus rattus</i>
	bindjei	+		<i>Melomys burtoni</i> <i>Melomys cervinipes</i>
Celledoni	celledoni	+		<i>Isoodon macrourus</i> <i>Rattus fuscipes</i> <i>Melomys cervinipes</i>
Grippytyphosa	grippytyphosa	+	Cattle	<i>Isoodon macrourus</i> <i>Rattus sordidus</i>
	valbuzzi	+		
Hebdomadis	kremastos			<i>Perameles nasuta</i> <i>Isoodon macrourus</i>
Icterohaemorrhagiae	icterohaemorrhagiae		Dog	
	copenhageni	+	Dog	<i>Rattus norvegicus</i>

SEROGROUP	SEROVAR	MAN	ANIMAL HOSTS	
			Domestic	Wild
	mankarso	+		
Mini	szwajizak perameles	+		<i>Isoodon macrourus</i> <i>Perameles nasuta</i>
Pomona	pomona	+	Cattle Sheep Pigs Horses	<i>Rattus fuscipes</i>
Pyrogenes	zanoni	+	Cattle	<i>Isoodon macrourus</i> <i>Rattus rattus</i> <i>Rattus norvegicus</i> <i>Rattus sordidus</i> <i>Rattus fuscipes</i> <i>Melomys burtoni</i> <i>Melomys cervinipes</i> <i>Uromys caudimaculatus</i> <i>Mus musculus</i>
Sejroe	hardjo medanensis balcanica	+	Cattle Sheep	<i>Isoodon macrourus</i> <i>Trichosurus vulpecula</i>
Tarassovi	tarassovi bakeri	+	Pigs	<i>Rattus fuscipes</i> <i>Uromys caudimaculatus</i> <i>Hydromys chrysogaster</i>
-	hurstbridge		Pigs	

<i>Hydromys chrysogaster</i>	water rat
<i>Isoodon macrourus</i>	northern brown bandicoot
<i>Melomys burtoni</i>	grassland melomys (rat)
<i>Melomys cervinipes</i>	fawn footed melomys (rat)
<i>Mus musculus</i>	domestic mouse
<i>Perameles nasuta</i>	long nosed bandicoot
<i>Rattus fuscipes</i>	bush rat
<i>Rattus norvegicus</i>	brown rat
<i>Rattus rattus</i>	black rat
<i>Rattus sordidus</i>	canefield rat
<i>Trichosurus vulpecula</i>	brushed tailed possum
<i>Uromys caudimaculatus</i>	white tailed rat

A list of serovars sorted by serogroups and genomospecies can be found in Appendix 1.

## **a) Serogroup australis**

Serovars belonging to this serogroup are difficult to isolate and culture, especially from urine.

### **i. *L interrogans sv australis***

This serovar was isolated from cattle<sup>4</sup> and rats<sup>237</sup> in Australia. An outbreak of leptospirosis in dogs in northern Queensland is suspect to be due to this serovar. Isolates are currently undergoing identification.

This serovar can cause severe and sometimes fatal leptospirosis in humans.

### **ii. *L interrogans sv bratislava***

This serovar has been detected serologically in pigs but efforts to isolate this leptospire in Australian and New Zealand pigs have so far been unsuccessful. Pigs, horses and hedgehogs are recognised as being maintenance hosts for this parasite. This serovar can cause significant reproduction losses in pigs but clinical disease is rare in dogs and horses.

## **b) Serogroup ballum**

### **i. *L borgpetersenii sv ballum***

Serovar ballum was isolated from the kidneys of *Rattus norvegicus*, *R rattus* and *Mus musculus* in and around dairy farms in New Zealand.<sup>238</sup>

## **c) Serogroup canicola**

### **i. *L interrogans sv canicola***

Dogs are recognised as the maintenance host for this serovar. It is not known whether dingoes or foxes are also maintenance hosts. In many parts of the world the incidence of clinical infections in dogs appears to be declining. This may be due to the effectiveness of vaccination programs and greater control of stray dog populations.

Clinical leptospirosis due to this serovar can occur in humans, pigs, sheep and horses but is most severe in dogs. Sv canicola has been isolated from healthy cattle.

## **d) Serogroup celledoni**

### **i. *L weilii sv celledoni***

This serovar has been isolated from rats in northern Queensland.<sup>239</sup>

## e) Serogroup grippotyphosa

### i. *L interrogans* sv grippotyphosa

As the European isolates of sv grippotyphosa belong to genomospecies *L. kirschneri* according to nucleic acid tests,<sup>74</sup> it is most likely that the Australian isolate also belong to genomospecies *L. kirschneri* (Smythe, *pers. comm.*).

This serovar was isolated from a healthy pregnant heifer near Tamworth in NSW during experimental trials on leptospirosis (*L borgpetersenii* sv hardjo) transmission.<sup>5</sup> This was an unusual incident in that it was the first and only isolation from domestic livestock in Australia. It occurred in only one of four heifers used in the trial. The authors suspected rodents transmitted the bacterium. Published work suggests that, compared to other serovars, it is rarely isolated from rats and mice being more commonly isolated from other species of small mammals. In Australia, it has been isolated from the canefield rat (*R sordidus*) and the northern brown bandicoot (*I macrourus*), neither of which are found around Tamworth. It has also been isolated from a human in northern Queensland.

The main maintenance hosts for this serovar are raccoons and skunks in North America and the common or field vole (*Microtus arvalis*) and the striped field mouse (*Apodemus agrarius*) in Europe. It is possible that Australian mammals are poor maintenance hosts for this serovar, hence the very low Australian incidence.

Serosurveys of livestock in most countries show a low to high prevalence of agglutinating antibodies to this serovar in several species including cattle, however it is not recognised for causing severe disease in livestock. This serovar has not been found in New Zealand where it is considered to be a quarantine threat (Marshall R, *pers comm*).

This serovar usually causes a mild type of leptospirosis in humans.

## f) Serogroup icterohaemorrhagiae

Rodents are recognised as the maintenance hosts for serovars in this serogroup. Many serovars in this serogroup can cause severe and sometimes fatal leptospirosis in humans.

### i. *L interrogans* sv copenhageni

This serovar has been isolated from man, dog, and the brown rat (*R. norvegicus*) in Australia and from cattle and the brown rat (*R. norvegicus*) in New Zealand. It was strongly suspected to be the cause of clinical leptospirosis in some dogs in New Zealand.<sup>63</sup>

### ii. *L interrogans* sv icterohaemorrhagiae

There are strong similarities between sv's icterohaemorrhagiae and copenhageni. Some workers believe these two serovars to be genetic variants of the same organism. Certainly there is usually very strong cross-reaction between these two serovars during serological testing.

This serovar can cause severe and sometimes fatal disease in dogs.

## **g) Serogroup pomona**

### **i. *L interrogans* sv pomona**

Two genotypes (B and C) are known in Australia, both different from the *pomona* reference strain but closely resembling the *kennewicki* strain using restriction endonuclease analysis (REA). Both are serologically indistinguishable from *pomona*. Only one of the genotype (C) has been recovered from cattle but both occur in pigs and humans in Australia. Genotype A has been isolated from a human in New Zealand. Due to the different genotypes occurring, vaccines may need to be re-evaluated.<sup>240</sup> Recent work using restriction fragment length polymorphism (RFLP) in *pomona* isolates showed even greater genotypic variability among isolates in Australia. This variability may affect the virulence of the genotype and the immunity of the host animal.<sup>241</sup>

Pigs, not rats, are usually the maintenance host for this serovar.<sup>242</sup>

This serovar usually causes mild type of leptospirosis in humans but can cause severe or fatal infections in pigs and cattle.

## **h) Serogroup pyrogenes**

### **i. *L interrogans* sv zanoni**

Corney *et al* 1996 reported that there were slight genotypic variation between *zanoni* isolated from dairy cow in northern Queensland and the *zanoni* reference strain held by the WHO/FAO Leptospirosis Reference Laboratory in Brisbane.

In 1999, in Queensland, 71 Leptospiral isolates were recovered from 436 isolation attempts and 31 (43.8%) isolations were identified as sv *zanoni*.<sup>1</sup>

## **i) Serogroup sejroe**

### **i. *L borgpetersenii* sv balcanica**

Many publications refer to *L interrogans* sv *balcanica* but the correct taxonomy is *L borgpetersenii* sv *balcanica*. This serovar was isolated from kidneys of sexually mature brushtailed possums in southeastern Australia<sup>243</sup> and New Zealand.<sup>244</sup> This serovar is being investigated as a vector for the biological control of brushtailed possums in New Zealand.<sup>245</sup> However experimental infection of calves and sheep in Australia produced neither fever nor clinical disease but a transient leptospiuria lasting up to 9 days in some individuals.<sup>246</sup>

Serovar *balcanica* was also isolated from cattle in New Zealand<sup>247</sup> and USA.<sup>248</sup>

The serovar was originally isolated from cattle, pigs and humans in Eastern Europe but the possum strain found in Australia and New Zealand is genetically different from the Bulgarian strain (Marshall R, *pers comm*).

**ii. *L borgpetersenii* sv hardjo**

There is no apparent genotypic variability in sv hardjobovis in Australia as all isolates from cattle in Queensland,<sup>249</sup> Victoria, New South Wales, from sheep in WA,<sup>250</sup> and from human in Victoria were serotype A.<sup>7</sup> The prevalence of sv hardjobovis infection in cattle is high and pathogenicity is low compared with *L interrogans* sv hardjo (genotype hardjoprajitno).

This serovar usually causes a mild type of leptospirosis in humans.

**iii. *L interrogans* sv hardjo**

Serovar hardjoprajitno, though considered primarily as a bovine pathogen, was originally recovered from a plantation worker in Sumatra. There are no reports of this serovar occurring in Australia, New Zealand or the USA (except on the Mexican border). Serovar hardjoprajitno infections are often associated with abortions and agalactia in Northern Ireland.<sup>251</sup>

**iv. *L interrogans* sv medanensis**

Mexico veterinary authorities require Australia to test livestock for this serovar prior to being exported to Mexico. This serovar appears to be common in Central and South America.<sup>252</sup>

Although this serovar has been isolated in the northern brown bandicoot and man in Australia, it is not considered common in Australia even though the Pan-bio website<sup>253</sup> lists this as a serovar occurring in Australia. As sv medanensis cross-reacts closely with sv hardjobovis, hardjoprajitno and balcanica, the extent and spread of this organism through cattle or sheep populations could be masked (Marshall R, *pers comm*). A small percentage of dogs have seroconverted to this serovar.

**v. *L borgpetersenii* sv sejroe**

There is very little literature on the occurrence of this serovar. It was isolated from kidneys of beagles in Italy that had not shown any clinical signs of leptospirosis but had mild interstitial nephritis.<sup>254</sup>

**vi. *L interrogans* sv wolffi**

This serovar appears to be fairly widespread. It is prevalent among cattle in Brazil and has been isolated from aborted fetuses in dairy cattle,<sup>255</sup> a mouse<sup>256</sup> and man<sup>257</sup> in that country. It was also identified as a pathogenic leptospire in China.<sup>258</sup> Antibodies to this serovar have been detected in snakes in Argentina.<sup>259</sup> There are no reports of this serovar having been isolated in Australia although low titres were recorded in cats in Sydney.<sup>41</sup>

**j) Serogroup tarassovi**

**i. *L borgpetersenii* sv tarassovi**

This serovar has been isolated from Australian and New Zealand pigs.<sup>27, 61</sup>

It usually causes a mild type of leptospirosis in humans.

## 9. Epidemiology

Infection usually occurs directly through mucous membranes or through abraded or water-softened skin. The leptospires may appear in the blood 4 to 10 hours after infection and may remain detectable in blood from only a few hours to 7 days. Clinical signs may not always be evident but fever often occurs with acute leptospirosis. Animals that have recovered from leptospirosis may develop a carrier condition in which leptospires grow in renal tubules for periods of days to years. During this time, leptospires are passed out in the urine. Sometimes leptospires may persist in other organs, particularly the genital tract.

Leptospires cannot survive dry or acidic conditions, preferring wet environments. The main sources of infection are urine, kidneys, contaminated surface waters, mud and soil. Infection is unlikely as a result of ingesting cooked food or inhaling airborne particles but can occur as a result of eating uncooked food or inhaling droplets of infected urine. However, infection can occur when leptospires enter through oral abrasions while ingesting hay contaminated with urine or by inhalation of aerosols of urine.

Infection can occur in many mammalian species and some reptiles. The infected animal often becomes a carrier with a reservoir of leptospires in their kidneys or genital organs. There is no relationship between the severity of the infection and the subsequent carrier status. Animals that do not develop clinical disease but become chronic carriers may be described as maintenance hosts whilst those that develop clinical leptospirosis and then become short-term carriers may be described as accidental hosts. It is not always possible to determine whether an animal is an accidental or maintenance host. Some animals may be accidental hosts for one serovar and maintenance hosts for another, eg, cattle are recognised as a maintenance host for sv hardjobovis and an accidental host for sv pomona. Camelids show very low antibody titres, are not known to excrete leptospires and appear to become neither maintenance nor accidental hosts.

The serovars most associated with acute or fatal leptospirosis in humans are those belonging to serogroup icterohaemorrhagiae (notably svs icterohaemorrhagiae, copenhageni, and lai), and some serovars in serogroups australis (svs australis and bratislava), autumnalis (svs autumnalis and bim), bataviae (sv bataviae) and pyrogenes (pyrogenes and zanoni).

### a) Factors affecting transmission

Transmission can occur as a result of direct or indirect contact with infected animals carrying leptospires. Direct transmission is rare in accidental hosts, especially humans. Congenital transplacental infection, including non-venereal, environmentally acquired infection of pregnant females, can occur as can venereal infection.

Leptospires could be introduced in:

- clinically infected animals,
- healthy animals with leptospiuria,
- animals with venereal infection,
- pregnant females with infected foetus in utero,
- semen, or

- embryos.

Leptospire can be transmitted to a breast fed infant via milk and has been isolated from the milk of a woman during bacteraemic phase.<sup>260</sup>

Indirect transmission occurs when infection arises as a result of contact with

- infected urine,
- water, soil or mud,
- material contaminated by urine, and
- meat and kidneys.

The establishment and maintenance of an endemic focus of leptospirosis depend on the introduction of a particular serovar, the availability of a suitable carrier host, and a suitable host habitat. A maintenance host may best be defined as an animal capable of acting as a natural source of leptospiral infection for its own species while a maintenance population may be defined as a population of an animal species acting as a continuous reservoir of a serovar in a specific ecosystem.<sup>261</sup> Hence, within a maintenance population, direct transmission appears to be more significant than indirect transmission through the environment.<sup>262</sup>

Leptospire do not survive drying under natural conditions and soil and sub-soil type and structure can influence its survival in dry periods. In favourable soil type and acidity, leptospire can persist for long periods. Similarly, leptospire can survive long periods in water, especially in surface water, puddles, sewage, other effluents, swamps, streams and rivers.

Leptospire can enter the host by:

- penetrating through small abrasions in skin or body surfaces,
- inhalation of infected aerosols,
- penetrating the conjunctival sac, or
- being consumed in water or milk.

In highly susceptible animals, less than 10 leptospire are sufficient to cause fatal infection.

Leptospire can multiply quickly before the immune system has time to respond then disrupt the integrity of the endothelial cell membranes lining small blood vessels causing haemorrhage. In acute cases, widespread petechiation can occur in all tissues and organs, with gross bleeding in organs where stretching of blood vessels occur, such as the lungs and pericardium. The damaged blood vessels in the kidneys can cause ischaemia, which in turn can cause renal tubular necrosis. Extensive renal necrosis results in renal failure, sometimes with fatal consequences. Recovery from leptospirosis depends on the extent of the damage to the tissues, the immune response, and the ability of the tissues to recover and function normally.

Leptospire are not pyogenic bacteria as such, as they do not directly cause inflammatory reactions but do so indirectly through secondary tissue reaction. Leptospire can adhere to renal tissues without causing cell damage thus enabling them to survive in the kidneys for long periods.

After recovery leptospire can persist in certain tissues, especially the kidneys. Sometimes leptospire can invade and localise in the brain, the anterior eye chamber and the genital tract of both male and female animals. Leptospire can be isolated from semen of rabbits and bulls following

experimental infection. Specific leptospiral antibodies can also appear in semen of infected animals. Artificial insemination of infected semen can induce leptospirosis in recipients, causing abortion, reduced fertility and stillbirth.<sup>263</sup>

Accidental hosts may excrete the leptospire for several days after recovery but often tend to excrete them intermittently or regularly for months or even years after initial infection.

Second infection with the same serovar is extremely rare due to host immunity arising from the previous infection. However, further infection with an unrelated serovar is possible. Immunity is usually specific for a serovar or a related group of serovars.

## **b) Factors affecting prevalence of leptospirosis**

Risk of infection in an animal is influenced by:

- proximity to carriers,
- concentration of carriers,
- contact with carriers,
- quantity of infective leptospira in environment,
- geographical factors,
- climatic factors, and
- socio-cultural factors affecting animal husbandry and agricultural practices.

Social factors have a profound effect on the occurrence of leptospirosis in humans and animals.

These factors include:

- occupation – high risk occupations include dairy farming, banana farming and abattoir workers;
- lifestyle – those involved with travel, water sports and military activities are at higher risk of exposure;
- economic – wealthy farmers are more likely to have vaccinated animals and less likely to be exposed;
- urban – water supply and sewage drainage systems are both important; and
- agricultural practices – vaccination, rodent control and land drainage.

## **c) Factors affecting severity of leptospirosis in an animal**

The pathogenicity, that is, the host-specific ability of leptospire to cause disease or otherwise to induce pathological change in a susceptible host, is determined by the genotype of the leptospire. On the other hand, virulence is determined by the phenotypic properties of the leptospire. Sometimes, leptospire can lose their virulence but it can be re-established following passing through a suitable host system. The immune system can affect the growth rate and the subsequent spread of leptospire to other tissues.

Leptospirosis can vary in severity according to

- infecting serovar,
- strains within a serovar
- age of the host,

- health of the host, and
- nutrition.

## d) **Distribution of Leptospira species and serovars**

### i. **Geographical distribution**

There is evidence of several serovars, or even species, of leptospire being confined to geographical areas. Serovars belonging to *L santarosai* and *L noguchi* are found almost exclusively in the Americas, while *L weilii* occurs almost only in Europe and Asia. Only leptospire belonging to *L borgpetersenii* and *L kirschneri* have been isolated from cattle in Zimbabwe. However the international movement of animals has led to a change in the distribution of animal hosts, the spread of leptospire carried by these hosts and new host-leptospira relationships.

### ii. **Host distribution**

Certain serovars often have an exclusive association with specific host species. It is likely that every known species of rodent, marsupial or mammal, including aquatic mammals such as platypuses and sea lions, can be a carrier and excretor of at least one leptospiral serovar.

### iii. **Distribution of rodents**

Some 70 species of rodents have been described in Australia, some of which are now considered to be extinct.<sup>264</sup> Over 2,000 species exist in the world. All Australian species fall into the family Muridae. Species belonging to the sub-family Hydromyini, which includes the tree-rats, rock rats, water rats, hopping mice and the arid adapted native mice, are found only in Australia. Many native rodents are a major source of food for carnivorous animals and birds and have suffered reduction in distribution and numbers since European settlement. Many native species now have limited distribution, however some inland species of rodents, such as the long-haired rat, (*Rattus villosissimus*) regularly reach plague proportions following favourable seasonal conditions. Members of the sub-family Hydromyini may be carriers of leptospire but their limited numbers and distribution suggest the risk of infecting animals and livestock to be very low. Of the subfamily Hydromyini, perhaps only two species may be of concern:

- Water rat, *Hydromys chrysogaster*, which probably pose the highest risk, having a widespread distribution throughout the northern, eastern, southeastern parts of Australia, including Tasmania, and in the southwest corner of Western Australia.
- The grassland melomys, *Melomys burtoni*, which, although limited to the coastal areas of northern Australia, is a serious pest in sugarcane

Of the remaining species of rodents in Australia, all belong to subfamily Murinae. Seven species are considered native while the remaining three, the House Mouse (*Mus musculus*), Brown Rat (*Rattus norvegicus*) and the Black Rat (*R rattus*) were introduced by the Europeans during early settlement. The Pacific Rat (*R exulans*), widespread in South-east Asia, the Pacific Islands and parts of New Zealand, is surprisingly not found in mainland Australia, even though there were evidence of these rats on Adele Is and Murray Is. Of the seven native rats:

- Dusky rat (*R colletti*) is abundant only in the far northern portion of Northern Territory;

- Bush rat (*R fuscipes*) is widespread and common along the forested coasts and ranges as it prefers sites where there is dense ground cover;
- Cape York rat (*R leucopus*) is found only in the rainforests of Cape York and New Guinea;
- Swamp rat (*R lutreolus*) is commonly found on the coastal areas of southeastern Australia. Although they can swim through pools of water quite readily, they prefer to live in tunnels made through dense grass, sedge or heath vegetation;
- Canefield rat (*R sordidus*) inhabits tropical grasslands, open forests and grassy patches within forest clearings;
- Pale field rat (*R tunneyi*) is distributed around northern Australia, living in tall grasslands near watercourses, and in hoop pine plantations;
- Long-haired rat (*R villosissimus*), found mainly in arid areas of northern Australia, can plague following good seasons.

Leptospire have been isolated from two species of native rats, *R sordidus* and *R fuscipes*. Thus native rats may be carriers of leptospire but they rarely pose a risk of infecting livestock. However, they may pose a risk to mammalian predators.

The brown rat, *R norvegicus*, has not spread far beyond the major coastal cities in Australia. Serovars icterohaemorrhagiae and zannoni have been isolated from these rats in Australia.

On the other hand, the black rat has spread through much of settled coastal Australia and is now common over most of the agricultural land of southern Australia. Black rats generally inhabit local areas not dominated by other rodents.

The house mouse (*M musculus* or *M domesticus*) occurs throughout Australia and under favourable conditions can form plagues and cause significant crop damage. Leptospire isolated from the house mouse in Australia include svs zannoni and australis.

Very little research has been done on the impact of these three introduced rodents on the health of humans and animals in Australia. There is no report of outbreaks of leptospirosis occurring during mouse or rat plagues in Australia, however, outbreaks of leptospirosis have occurred during prolonged periods of high rainfall and flooding.

While introduced species can be controlled at discretion, native species are protected wildlife and cannot be controlled without permission from wildlife authorities. The use of chemical poisons is constrained by regulations. A number of rodenticides are registered for use around buildings but none is registered for in-crop use except brodifacoum, an anticoagulant, in sugarcane. Special permits are necessary for using rodenticides in or around orchards.

## 10. Risk of introduction, establishment, spread and consequence

### a) Likelihood of entry

Leptospire could enter in:

- animals that:
  - are clinically infected, with or without detectable leptospiral antibodies;
  - were vaccinated, whether against infective serovars or not;
  - have leptospirosis or venereal infection, with or without detectable leptospiral antibodies;
  - are healthy pregnant females with infected foetus; and
  - have recently been given antibiotics;
- semen and embryos;
- imported tissue cultured cell lines derived from animals; and
- materials such as hay and bedding contaminated by urine of imported animals.

#### i. Animals

Imported animals need not have detectable antibodies in their blood to be carriers of leptospire. The MAT, which is the standard test used for detecting anti-leptospiral antibodies, is most useful as a screening test for leptospirosis in a herd but less useful as a diagnostic test in individual animals.

Vaccinated animals are not necessarily free from disease as:

- vaccination offers protection against only those serovars included in the vaccine;
- some vaccines offer only very low levels of protection;
- duration of vaccine protection is limited;
- vaccination is effective only in animals which have not been previously exposed to serovars included in the vaccine;
- vaccination is not an effective treatment for carrier states, and
- some commercial vaccines used in other countries offer poor protection against leptospirosis.

However, sound vaccination programs involving vaccination of young animals and regular booster doses, especially of maintenance hosts, can reduce the incidence of leptospirosis. Regular vaccination of dogs combined with effective stray and feral dog control programs has done much to reduce the incidence of canicola infections in Europe.

Animals treated with antibiotics for leptospirosis may not be free from disease. While leptospire are susceptible to antibiotics except chloramphenicol and rifampicin, antibiotics may not be effective in completely removing leptospire from carriers.

Streptomycin/dihydrostreptomycin (S/DHS) is generally recognised as the most effective antibiotic against leptospirosis in animals, especially in cattle and pigs. However, the high risk of S/DHS residues in milk, meat and offals precludes its use in several countries. A permit is now required for use of S/DHS in food-producing animals in Australia.

## **ii. Semen and embryos**

Leptospire can be recovered from oviductal and uterine fluids in heifers experimentally infected with sv hardjobovis via the uterine, cervical, supraconjunctival or intranasal routes. It was not possible to culture leptospire from the recovered embryos of these heifers although PCR assay were positive for presence of leptospiral DNA on some embryos. None of the recipient heifers developed antibody titres to leptospire.<sup>265</sup>

As any agent that will remove the outer envelope of leptospire is lethal,<sup>266</sup> low concentrations of proteases such as trypsin, commonly used for washing bovine embryos, can quickly remove the outer envelope of leptospire. Continued treatment can lead to the gradual unwinding of the helix to reveal a flat empty tube, which retains some antigens, and contains muramic acid.<sup>267</sup>

The risk of infective leptospire entering Australia via either semen or embryos is negligible as antibiotic use in the processing of semen and embryos is standard practice and most antibiotics are effective against leptospire. Furthermore, trypsin is usually used in the washing of bovine embryos for export and can destroy leptospire before antibiotics are added during processing.

## **iii. Animal derived tissue cultures**

Virulent strains of leptospire can attach effectively to tissue culture cells.<sup>268</sup> Cell lines prepared from kidneys of animals infected with leptospire can be contaminated with the leptospire.<sup>269</sup> A cytopathic effect can occur in kidney cell cultures infected with virulent leptospire.<sup>270</sup>

## **iv. Contaminated material**

Material such as straw and litterbox absorbent used as bedding with imported animals can be contaminated by infected urine of imported animals. Unless animals are individually penned, there is a risk of other susceptible imported animals becoming infected during pre-export quarantine and during shipment. Furthermore, unless such materials are promptly destroyed on arrival in Australia, there is a risk of maintenance hosts, such as rats and mice, accessing these materials and becoming infected.

## **b) Likelihood of exposure**

The exposure of susceptible animals in Australia could result from:

- direct transmission via the urine of a carrier animals to the mucous membrane of another susceptible animal;
- indirect transmission following contamination of the environment by the urine of a carrier animal;
- venereal transmission from a persistently infected uterus;
- both direct and indirect transmission from an aborted infected foetus;
- congenital transplacental infection from an infected pregnant female;
- milk from an infected dam may infect suckling offspring;
- infected semen may infect recipient females;
- infected embryos transferred to recipients, may infect dam and offspring;

- infected animal material may infect carnivores following consumption; and
- contaminated bedding materials can contaminate the environment and cause indirect transmission.

### **c) Likelihood of undesirable consequence**

The exposure of susceptible animals in Australia to introduced leptospires may cause undesirable consequences to the livestock industry, the environment (including natural fauna), and public health.

#### **i. Establishment**

Establishment of leptospirosis may depend on:

- carrier state of imported host, the persistence of infection in the host, and the amount of leptospira being shed;
- availability of maintenance hosts where imported hosts are being kept;
- survival of leptospires outside the host in the environment for indirect transmission to occur, and
- likelihood of direct transmission to other susceptible accidental hosts.

Thus the likelihood of leptospirosis establishing is high if:

- an imported host with persistent leptospiuria is kept in a continually wet or damp area where there is an abundance of suitable maintenance hosts;
- wet areas or concrete pens, used for penning an imported host with persistent leptospiuria, are hosed or drained off to other sites where there are suitable maintenance hosts; and
- an imported host with genital infection is joined to a susceptible host.

#### **ii. Spread**

Spread will be influenced by:

- proximity of susceptible species to carriers;
- concentration of carriers to maintain the infection;
- contact with carriers;
- distribution of carriers;
- movement of infected animals;
- quantity of infective leptospira in environment,
- geographical factors;
- climatic factors, and
- social-cultural factors affecting animal husbandry and agricultural practices.

Under ideal conditions, the likelihood of introduced leptospires spreading can be very high. A careful evaluation of each of the factors listed above is essential to assess the risks of spread. Australia has imported considerable number of animals in recent years. Table 12 is a summary of live animal imports into Australia in 1999. There is no evidence of establishment and spread of exotic leptospirosis as a result of importing live animals in recent years.

Reports of infections due to sv zanoi in both cattle and people in Australia have been confined to northern Queensland. While animals outside northern Queensland have not been routinely tested for this serovar, it appears that infection comes from a wildlife source present only in that part of Queensland. Thus ecological factors may prevent the spread of some exotic serovars if introduced into Australia.

Table 12 Imports in 1999

Species of animal imported	Number imported
Dogs	3270
Cats	2317
Horses	2832
Cattle	54
Other ruminants	14
Cavies (Sep – Jun)	1071
Lab animals (Sep – Jun)	2844
No. cats, dogs and horses quarantined on arrival in Australia	3338
No. dogs, cats and horses imported from New Zealand and not quarantined	5081

### iii. **Biological consequence of agent introduction and disease establishment in Australia**

The entry, establishment and spread of exotic strains of leptospire may cause fatal disease in protected wildlife species and increased public health risks as well as disease in susceptible livestock and other domestic animal species.

There is concern in the scientific community that exotic serovars may eventually become adapted to a new host or carrier in Australia. With some bacteria and viruses, there is evidence of increased virulence with a species change, just as there is evidence of decreased virulence within a species especially during an outbreak. However, there is no evidence of decreased virulence occurring with leptospirosis within a species. Sometimes laboratory cultures lose their virulence and passaging them through a host animal such as hamster can restore the virulence.

There is no clear evidence of a serovar adapting to a new species. Both sv hardjobovis and pomona in Australia are examples. Depending on the nucleic acid detection test being used, several genotypes of pomona have been detected in Australia. Only one hardjobovis (of 18 cattle and 2 human isolates) has been identified. Serovar pomona is believed to have been in Australia considerable longer than hardjobovis and therefore had more opportunity to develop greater genotypic variability. However, the variation among pomona isolates may be due to its introduction on several occasions from different sources. Of the sv pomona, only genotype C has been detected in cattle while both genotypes B and C have been isolated from pigs and humans. Genotype A has been isolated from a human in New Zealand. Genotypes A, B and C are indistinguishable

serologically and cross-protection trials are necessary to confirm the efficacy of leptospiral vaccines containing the pomona strain.

Further, there is no clear evidence of leptospiral serovars suspected to have been introduced into Australia (such as sv *hardjobovis*, pomona, tarassovi and copenhageni) having adverse impact on Australian wildlife.

There is some evidence, however, of increasing pathogenicity of some serovars within some species. An example is the increasing incidence of pulmonary haemorrhaging being reported in human leptospirosis.

While there are unsubstantiated concerns about the risk of exotic serovars entering Australia and adapting to new animal species, introducing conservative quarantine measures to protect against such perceived risk is not consistent with Australia's obligation under the SPS Agreement and is not considered in this review.

#### **iv. Environmental consequence of agent introduction and disease establishment in Australia**

The environmental consequence of leptospirosis may be either favourable or unfavourable. New Zealand is exploring the use of sv *balcanica* as a part of their biological control program to reduce the population of brushtailed possums (*Trichosurus vulpecula*) as they cause significant damage to native forests, threaten populations of native plants and animals, and infect cattle and deer with bovine tuberculosis.

Leptospire can survive long periods in water, soil and mud. The spread of pathogenic leptospire can contaminate the environment and pose increased public health risks.

#### **v. Economic consequence of agent introduction and disease establishment in Australia**

Leptospirosis is a disease of economic importance. The following may cause either reduced income or increased cost of production:

- abortion,
- failure to thrive,
- agalactia,
- veterinary and other livestock management costs,
- hospitalisation expenses,
- rodent control costs,
- management of sugar cane fields and banana plantation and
- food processing.

Renal failure in dogs is common with clinical infection due to canicola but is not often described in other animals, even though interstitial nephritis, especially in pigs, may be extensive. Abortion, still birth, or birth of congenitally infected young may follow weeks or months of maternal leptospirosis and is usually the most economically important form of disease in ruminants and pigs.

Leptospirosis is a public health risk. From January to August 1999, some 61% of 201 reported leptospirosis in Queensland required hospitalisation for 1 to 27 days, depending on the severity of the disease. Some cases, especially those with pulmonary haemorrhaging, required intensive care unit management costing some \$2,000 to \$3,500 per day. During the same period some 20 humans lodged workcover claims resulting in \$23,549 in payments and 271 workdays lost.<sup>271</sup> Fortunately, none died. There have been claims for long-lasting consequences of leptospirosis in workers. One claim resulted in a judgement of over \$400,000 but this was lost on appeal on grounds of responsibility. The claimant had filed claims only against the meat inspectors and not against the field veterinary service. The judge, in handing down his judgment, stated, “*The position may have been different in the case of officers of the Veterinary Field Service but the plaintiff’s case was against the meat inspectors.*”

In New Zealand, a dairy farmer was fined \$15,000 for not taking adequate steps to protect his workers from the risk of becoming infected with leptospirosis.

**d) Overall risk**

**i. Introduction**

Likelihoods have been qualitatively defined to provide an estimate of risk. The nomenclature outlined in Table 13 has been adopted as the standard for the qualitative estimation of risk of introduction (*entry risk*) and establishment and spread (*exposure risk*).

**Table 13. Nomenclature for qualitative likelihoods for entry and exposure risks**

<b>Likelihood</b>	<b>Descriptive definition – Under the conditions described, the defined event is:</b>
High	Likely to occur in most cases.
Moderate	Expected to occur in half the cases.
Low	Unlikely to occur in most cases.
Negligible	Almost certain not to occur at all.

The nomenclature outlined in Table 14 has been adopted as the standard for qualitative estimation of risk of undesirable consequence (*consequence risk*).

**Table 14. Nomenclature for qualitative likelihoods for consequence risk**

<b>Likelihood</b>	<b>Descriptive definition</b>
High	The impact on a given criterion is likely to be significant at a national level, and highly significant within affected zones. This classification implies that the impact would be of national concern. The serious effect on economic stability, societal values or social wellbeing would, however, be limited to a given zone
Moderate	The impact on a given criterion is likely to be recognised at a national level, and significant within affected zones. The impact is likely to be highly significant to directly affected parties
Low	The impact on a given criterion is likely to be recognised within affected zones, and significant to directly affected parties. It is not likely that the impact on the given criterion will be recognised at the national level.

<b>Likelihood</b>	<b>Descriptive definition</b>
Negligible	It is almost certain that there will be no impact at all.

Entry risk and exposure risk are combined using the likelihood matrix as per Table 15. The likelihood of entry and exposure and consequence risk are then integrated using the likelihood matrix (as per Table 16) into the final risk estimate or *overall risk*.

**Table 15. Likelihood matrix for entry and exposure risks.**

<b>Entry risk</b>	<b>High</b>	Low	Moderate	Moderate	High
	<b>Moderate</b>	Low	Low	Moderate	Moderate
	<b>Low</b>	Negligible	Low	Low	Moderate
	<b>Negligible</b>	Negligible	Negligible	Low	Low
		<b>Negligible</b>	<b>Low</b>	<b>Moderate</b>	<b>High</b>
<b>Exposure risk</b>					

**Table 16. Final likelihood matrix for overall risk.**

<b>Likelihood of entry and exposure as derived from Table 15</b>	<b>High</b>	Low	Moderate	Moderate	High
	<b>Moderate</b>	Low	Low	Moderate	Moderate
	<b>Low</b>	Negligible	Low	Low	Moderate
	<b>Negligible</b>	Negligible	Negligible	Low	Low
		<b>Negligible</b>	<b>Low</b>	<b>Moderate</b>	<b>High</b>
<b>Consequence risk</b>					

Interpretation of the overall risk in the light of Australia’s acceptable level of protection is discussed in the next chapter, “Risk reduction”.

Because of the complex interaction of pathogenic leptospires with the host and the environment, a general estimation of risk is not possible. Risk depends on the species of animal or animal products being imported, on leptospiral serovars and on circumstance.

Animals are at high risk of being infected by leptospires, but are at low to moderate risk of developing clinical signs. In cattle, seroprevalence may reach 100%. During outbreaks of clinical leptospirosis, morbidity may range up to 30%, depending on clinical manifestation while case fatality rate is usually low, around 5%. Deaths usually occur only in calves. Abortion storms can occur with certain serovars, resulting in an abortion rate of up to 30%. Seroprevalence usually average 20% in pigs, 30% in horses and up to 100% in sheep and goats.

Humans at greatest risk of clinical leptospirosis in Australia are banana workers (23.4% in 1999), meat workers (8.4%), and dairy farmers (8.4%). The greatest at risk areas were the Northern Peninsula and the Central West Health Regions of Queensland where over 8 humans per 100,000 per year were reported with clinical leptospirosis from 1989 to 1994. Leptospirosis is endemic in the Northern Peninsula Region due to high rainfall and the climatic conditions supporting a large rodent and small marsupial population. Mechanical harvesting has reduced the risk of infection once faced by sugar cane workers.<sup>272</sup>

In some dairy farming regions of Victoria, infection rates of 5% per annum of humans exposed to risk were recorded during 1992 and 1993.<sup>273</sup>

## e) Risk event models

The approach adopted for developing risk reduction strategies was to identify areas where risk may be unacceptably high.

Although there are unsubstantiated concerns about the risk of exotic serovars entering Australia and adapting to new animal species, it is not possible to evaluate such perceived risk. Incorporating this risk in a risk event model for quarantine purposes is not consistent with Australia's obligation under the SPS Agreement and is therefore not considered.

### i. Possible entry and exposure events

#### 1. Risk of introducing "exotic" strains where a maintenance host already exist in Australia

- sv ballum, javanica and bataviae carried by *Rattus* spp;
- sv hardjoprajitno carried by cattle;
- sv canicola carried by dogs;
- *L kirschneri* sv grippotyphosa from Europe (muskrats, field mice – *Apodemus sylvaticus*, and voles) carried by canefield rats and bandicoots, and
- *L kirschneri* sv mozdok from Europe carried by rodents.

However, there are a number of serovars isolated and whose maintenance hosts are unknown

- sv shermani in buffaloes in India, and serologically detected in several animal species in Mexico and Bolivia;
- serovars isolated from healthy cattle in Zimbabwe (Table 8).

The risk where maintenance hosts can be managed, eg, domestic livestock, may differ from the risk where the maintenance host is difficult to manage, eg, rats.

Serotype hardjoprajitno is difficult to detect, as it is antigenically similar to hardjobovis, the most prevalent serovar occurring in cattle worldwide. It is much more difficult to isolate and culture. Its prevalence is very low in areas where it does occur. There is some suggestion of its resistance to streptomycin. Australia has had no quarantine conditions for this serovar, nor does USA, and this serovar has not spread to either country. The risk of its introduction, establishment and spread without any control measures needs to be evaluated.

Serovar canicola has been isolated from a human case in Australia but never reported in dogs, although a case was possible in a dog positive to sv robinsoni in Sydney. Leptospirosis due to this serovar is not a notifiable disease in animals in most of Australia. There is a likelihood of this disease being present in Australia, despite quarantine restrictions. Current quarantine conditions are not adequate to prevent the introduction of this serovar into Australia. A significant stray dog population and favourable dog demography are necessary for canicola to be a concern and Australia has neither of these factors. In the light of these factors, the risk of its introduction, establishment and spread without any control measures needs to be reassessed.

A number of new serovars, belonging to pathogenic strains of leptospire and exotic to Australia, have been isolated from the kidneys of healthy slaughter cattle in Zimbabwe. However, there is no evidence of its pathogenicity in other animals. It may be possible that risk measures are necessary to prevent their introduction into Australia even though there is no evidence that any such introduction, establishment or spread of any of these serovars will result in harm being caused to humans, animals, or other aspects of the environment or economic activities.

2. Risk of introducing new maintenance host species which may in turn carry other exotic strains of leptospire
  - small mammals from Europe as per Table 7.
  - Striped field rat (*Apodemus agrarius*) from Asia carrying sv lai

Some small mammals are carriers of highly pathogenic strains and these pose the highest risk of introduction, establishment and spread of exotic strains that can have an impact on the public health and animal health in Australia. Strict quarantine restrictions may need to be proposed for importation of small mammals known to be carriers of strains highly pathogenic to humans and animals. An appropriate risk reduction strategy may involve requiring only those animals from a colony free from leptospirosis be eligible for export. As investigation for leptospirosis in small mammals may mean the loss of life of test animals, the actual animals for import may not themselves be tested. A high level of surveillance may be necessary to prevent inadvertent introduction of rodents and other small mammals arising from incidents such as:

- rodents jumping off docked ships;
- rodents hiding in containers and other packaging containing imported goods, and
- small mammals smuggled into the country.

3. Risk of importing accidental host having persistent leptospiruria or genital infection
  - sv lora and muenchen with pigs from Europe.

The muskrat is believed to be the maintenance host for sv lora while voles are the likely maintenance host for muenchen. Neither of these animals exists in Australia. Hence risk measures to prevent their introduction into Australia may not be justifiable as there is no evidence that any such introduction, establishment or spread of any of these serovars will result in harm being caused to humans, animals, or other aspects of the environment or economic activities.

There is a risk of direct transmission from animal to animal within the same species where there is genital infection, especially sexual transmission, and with contact with the aborted foetus, stillbirth or

the infected placenta. If a risk reduction strategy is necessary, risk can be reduced to acceptably low levels if only:

- non-pregnant and virgin animals and
- animals with no history of reproductive disorder for the previous 12 months

are eligible for import.

4. Animals that show seroconversion to exotic serovars of leptospires before being imported into Australia

A survey of dogs being tested before being exported from Southeast Asia and Oceania Islands as per Table 3 show a significant number showing seroconversion to exotic serovars such as bataviae, djasiman, shermani, cynopteri, bulgarica, panama, and javanica. It is not clear which dogs were actually imported into Australia. But there are no quarantine measures against such animals and there is no evidence of

- these serovars having been introduced by these dogs;
- dogs being infected with any of these serovars, and
- dogs being infected with other serovars which cross-react with the exotic serovars.

Examples of different risks are given in the following risk events, each according to the animal species or animal product being imported and infecting serovars. The risk events do not necessarily relate to existing health protocols.

5. Risk of importing infected pinnipeds.

Leptospires can infect pinnipeds and occasionally cause clinical disease. There is no evidence to suggest that the risk of importing pinnipeds is higher than the risks associated with importing livestock.

**ii. Possible consequence events**

Leptospirosis is a complex zoonotic disease because different serovars have different impact on different animal species and humans. Each animal species act either as an accidental host or as a maintenance host, depending on the infecting serovar. Thus a range of consequences can arise depending on the serovar introduced and how animals and humans in Australia became exposed.

The major determinant of assessing the consequence of the entry and exposure is what impact will the consequence of the entry and exposure of an 'exotic' serovar have in Australia.

As explained earlier (Chapter 2 – National Obligations), leptospirosis is not a notifiable disease in all States and Territories. It is a notifiable disease of livestock only in Victoria and Northern Territory while two endemic strains, svs hardjo and pomona, are notifiable in Tasmania and one strain, sv canicola, is notifiable in South Australia. It appears that regulatory actions where leptospirosis is diagnosed are not usually enforced. During a court case on a claim for long-lasting consequences of leptospirosis, it transpired that the Victorian veterinary authorities did not take any regulatory action on a pig farm where an outbreak of leptospirosis was notified (*State of Victoria v Richards & Anor [1998] VSCA 103 (11 November 1998)*).

The response of the public health authorities to leptospirosis depends on the type of outbreak. Normally, sporadic leptospirosis cases are not followed up. However, a cluster of leptospirosis (due to svs hardjobovis and pomona), which occurred in eight abattoir workers in the Northern Rivers region of New South Wales in 1998, required an investigation by the regional public health unit. The unit concluded the outbreak “*highlighted a need for continued efforts at prevention. The use of protective clothing for abattoir workers should be encouraged.*”

The discovery of several previously unidentified serovars in Australia in recent years has had no significant national or regional impact, only some local impact. Thus the introduction of an ‘exotic’ serovar will most likely have a negligible to low impact, as defined in Table 13. In some cases, it is possible that the introduction of some ‘exotic’ serovars, highly pathogenic to humans, such as sv bim, found mainly in the Carribean region, may have moderate impact on the public health system in Australia.

There is uniform mandatory requirement for artificial breeding centres for all animals within the centre to test serologically negative for leptospirosis. This is because there is a higher risk of undesirable consequence of semen of an infected donor infecting a large number of recipients over a large geographical area.

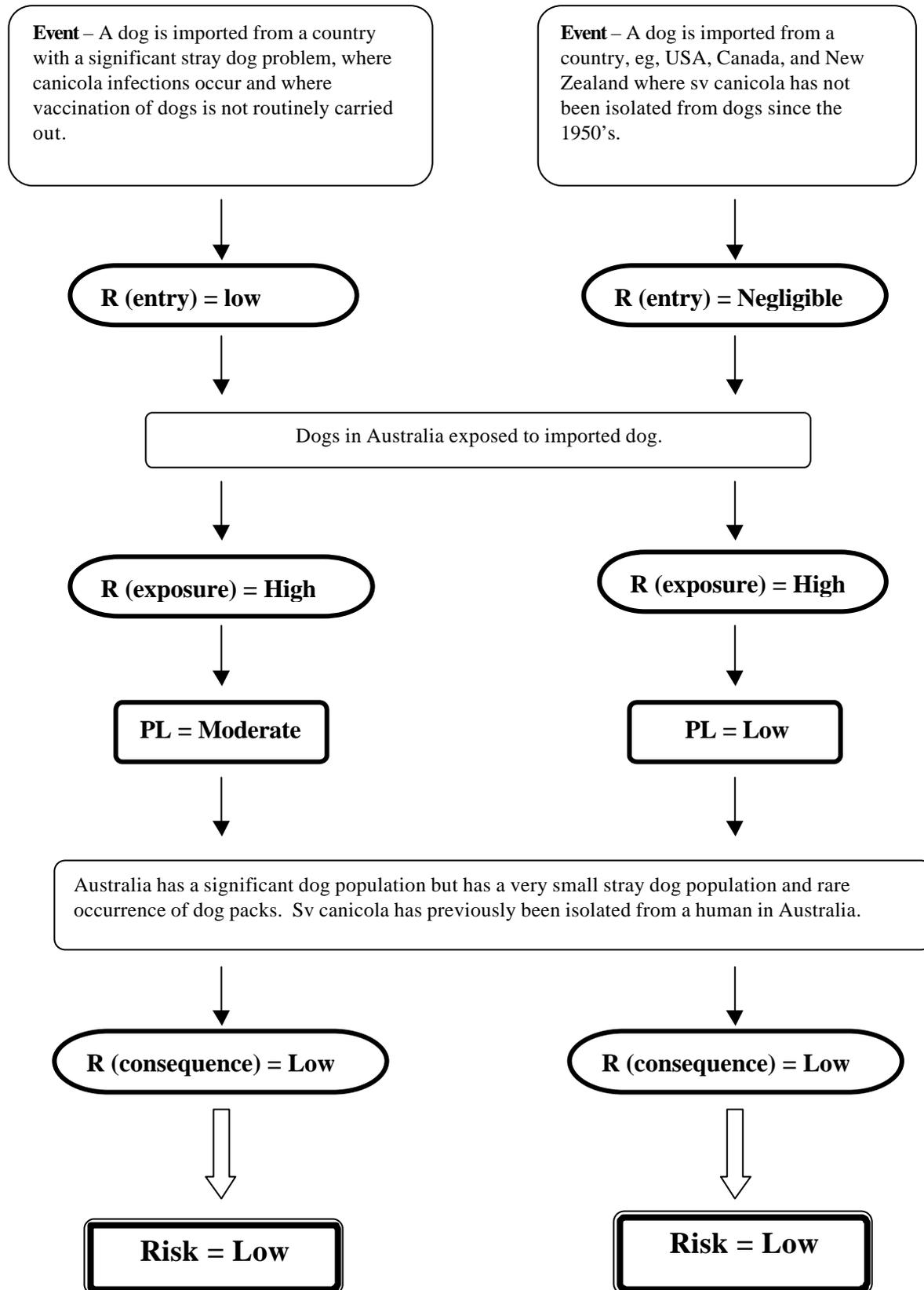
Note: for the following risk events -

R = risk;

PL = progressive likelihood.

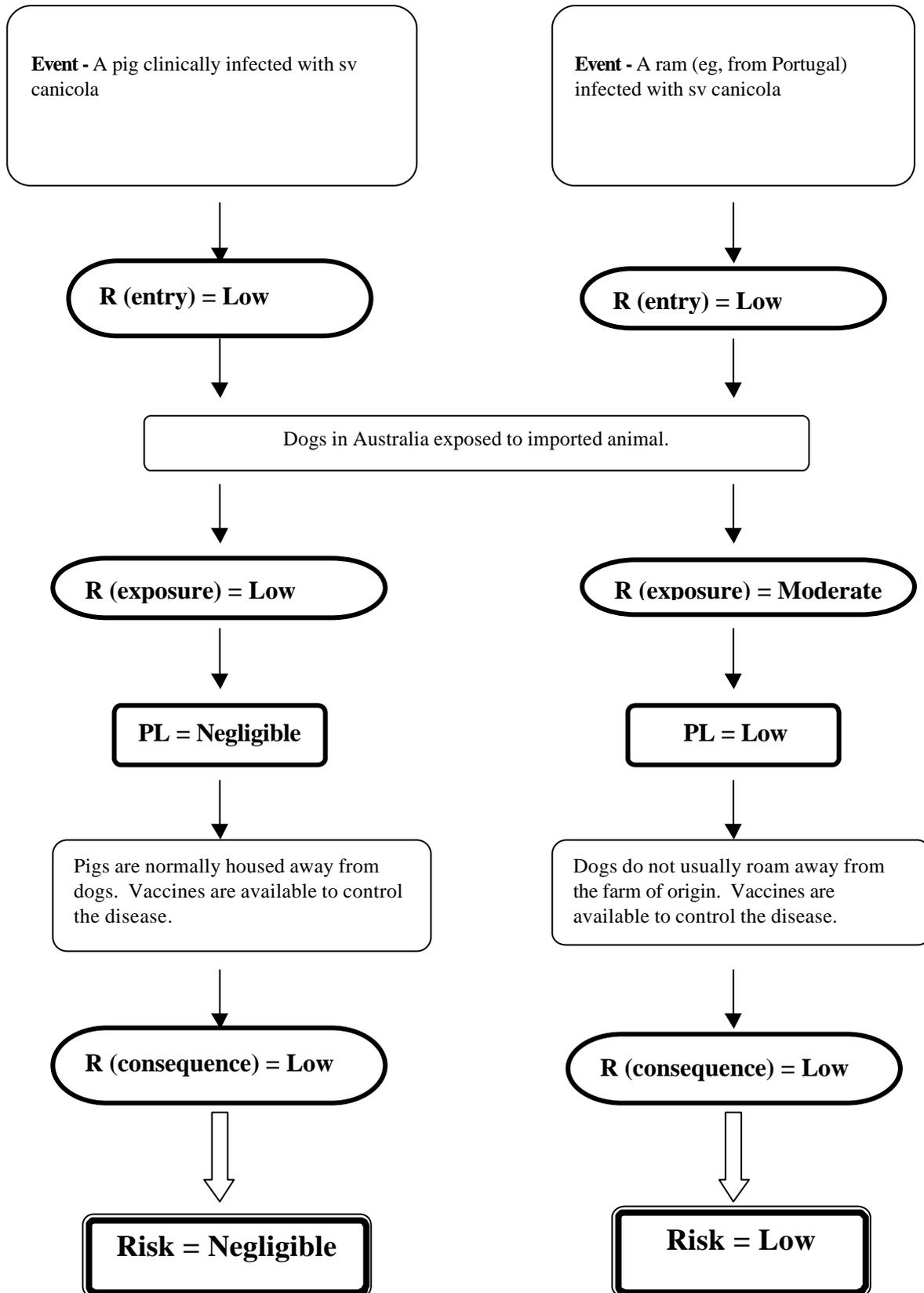
### iii. Importing dogs with sv canicola infection.

Dogs are recognised as the maintenance host for this serovar.



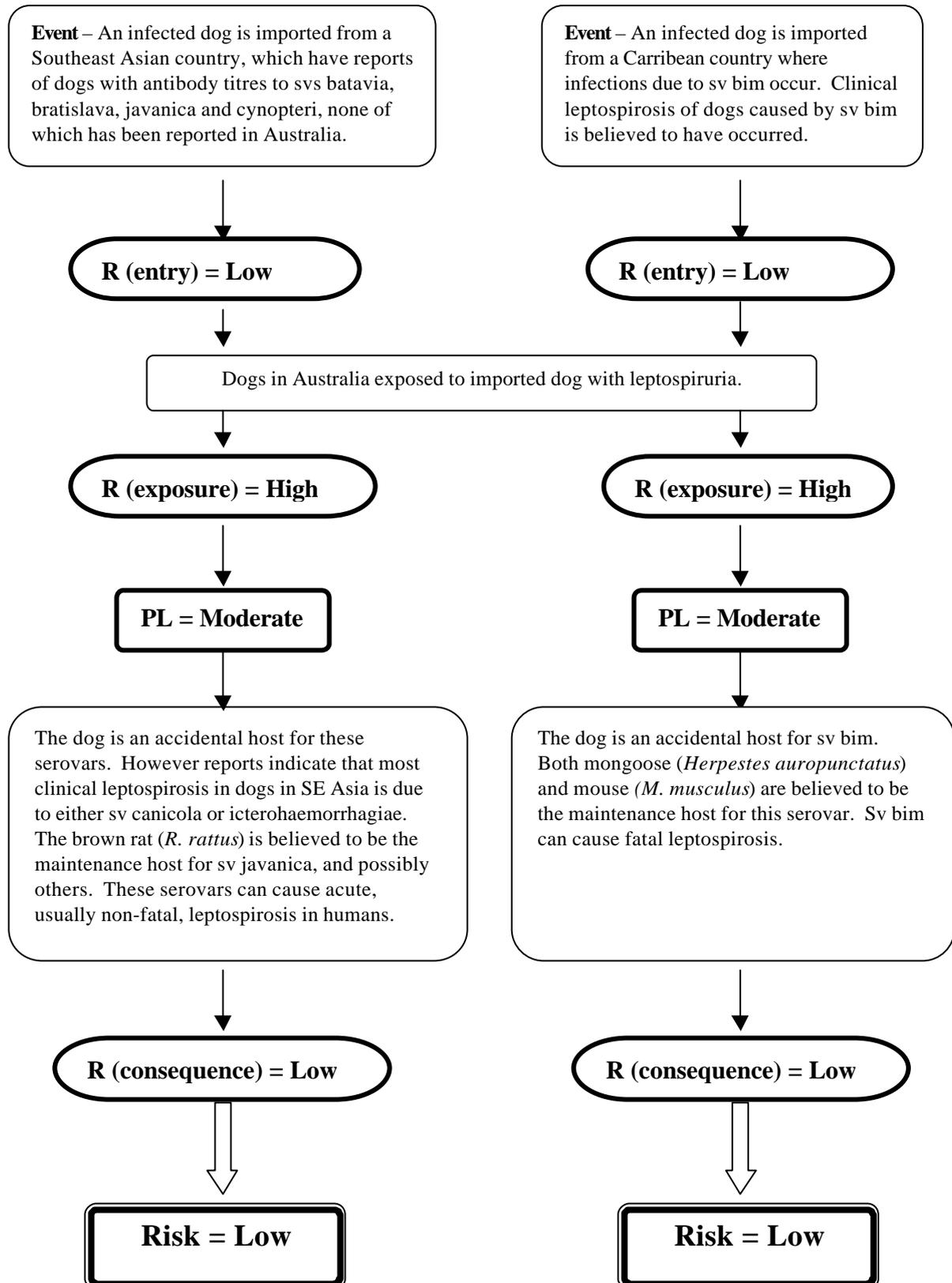
**iv. Importing pigs or sheep with sv canicola infection.**

Dogs are recognised as the maintenance host for this serovar.



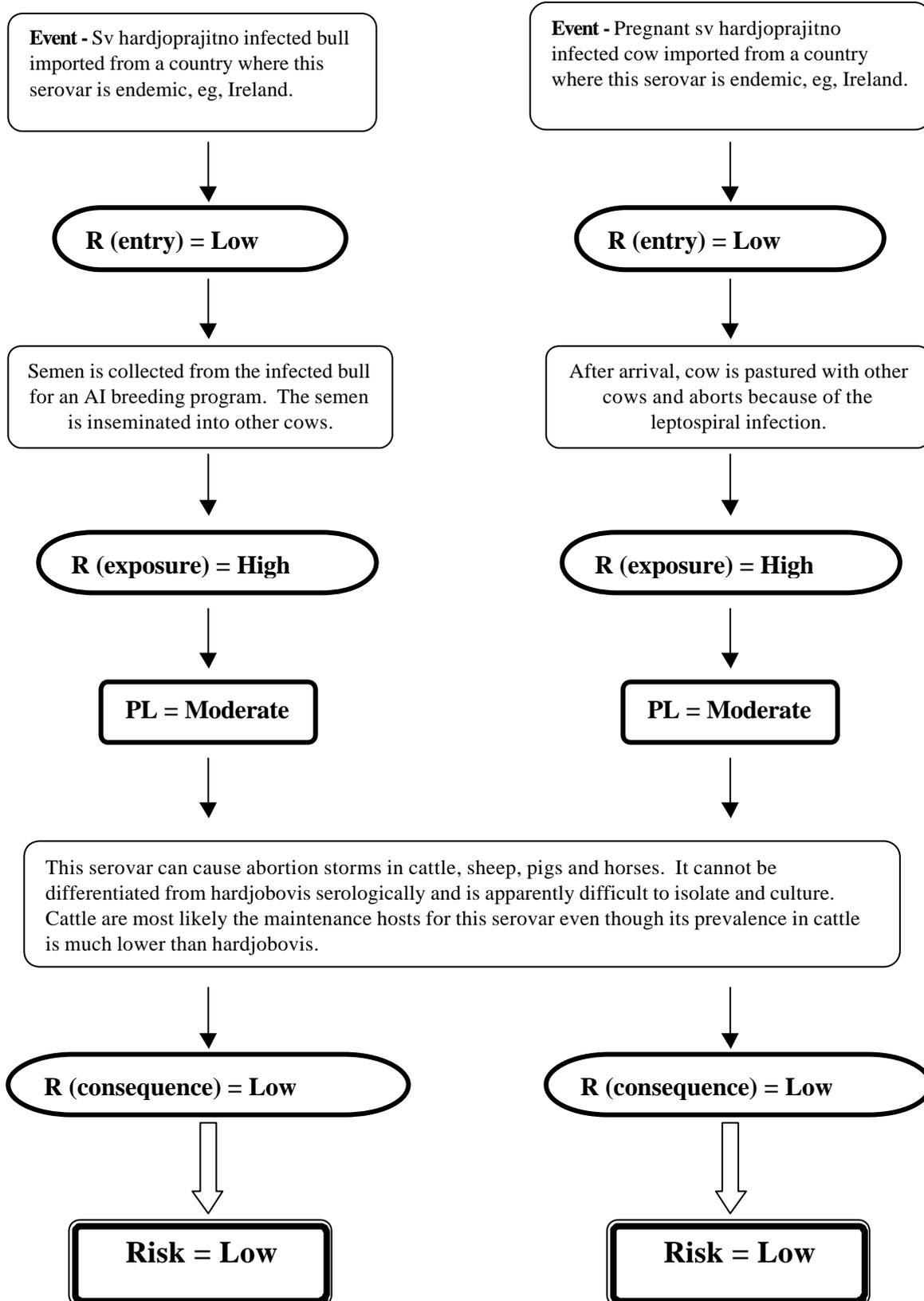
v. **Importing dogs infected with serovars not reported in Australian animals (apart from sv canicola infection.)**

Dogs are not recognised as the maintenance host for any serovars other than sv canicola. Reports of dogs with leptospirosis due to svv other than canicola, icterohaemorrhagiae and australis are rare.



**vi. Importing cattle with sv hardjoprajitno infection**

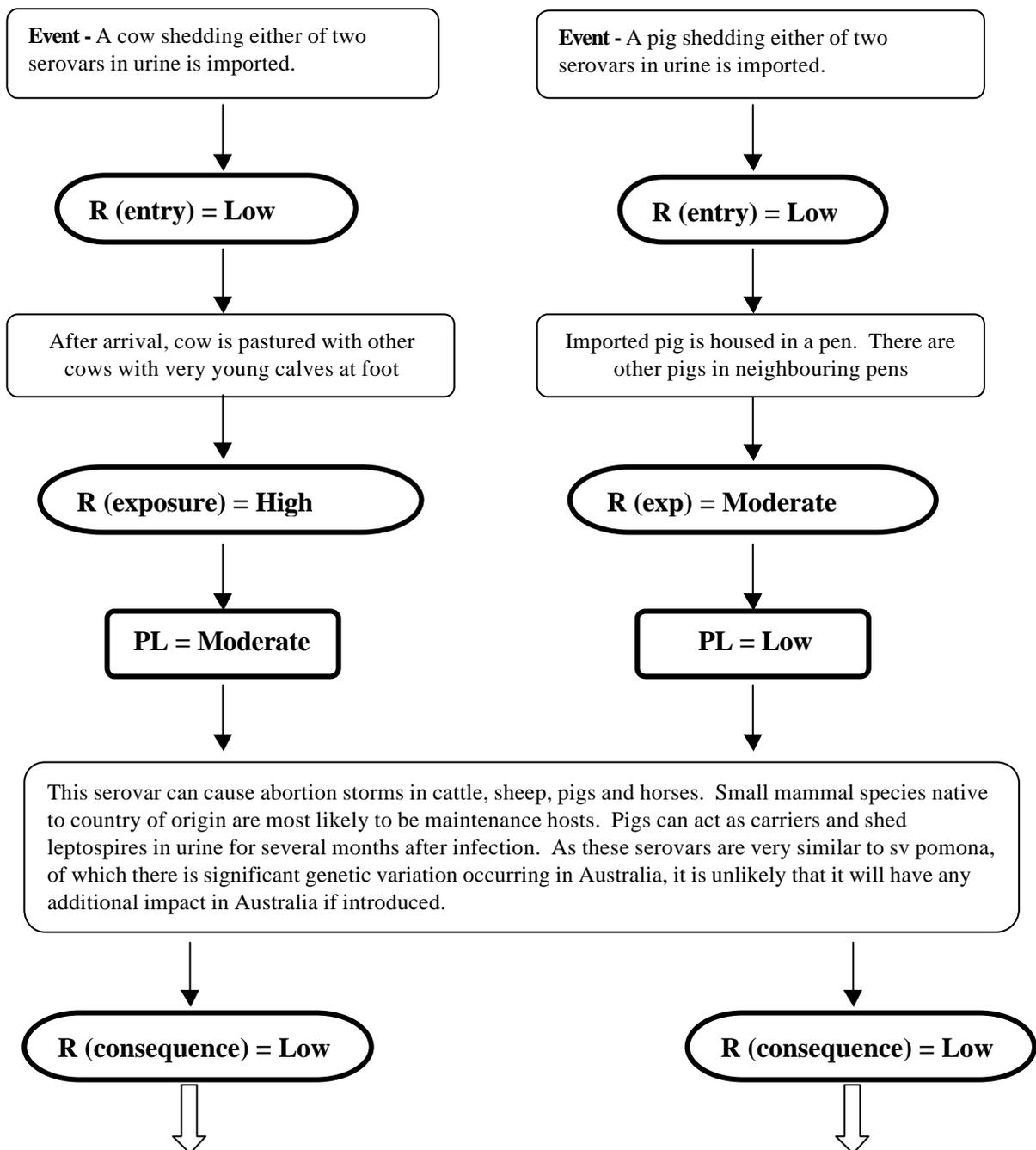
This serovar occurs in cattle in parts of Europe, Africa, and Central America. It was originally isolated from a Sumatran plantation worker and is most likely found in other parts of Asia. It is more pathogenic than its antigenically close relative, subtype hardjobovis and has not been reported in Australia. This disease is spread by venereal transmission.



**vii. Importing livestock with sv kennewicki or mozdok infection**

Serovar pomona and its close relative sv kennewicki, occurring in North America, and sv mozdok, in Europe, usually cause the most severe infections in cattle with sometimes fatal haemolytic anaemia in calves and abortions in cows. Rodents and other small mammals are believed to be the main maintenance hosts, such as house shrews (*Crocidura russula*) in Europe and skunks (*Mephitis mephitis*).

Sheep, goats, deer and camelids are usually less susceptible to clinical leptospirosis than cattle. Sheep are a maintenance host for hardjovovis. Clinical infections are rare in goats, camelids and deer but they do occur.



**Risk = Low**

**Risk = Low**

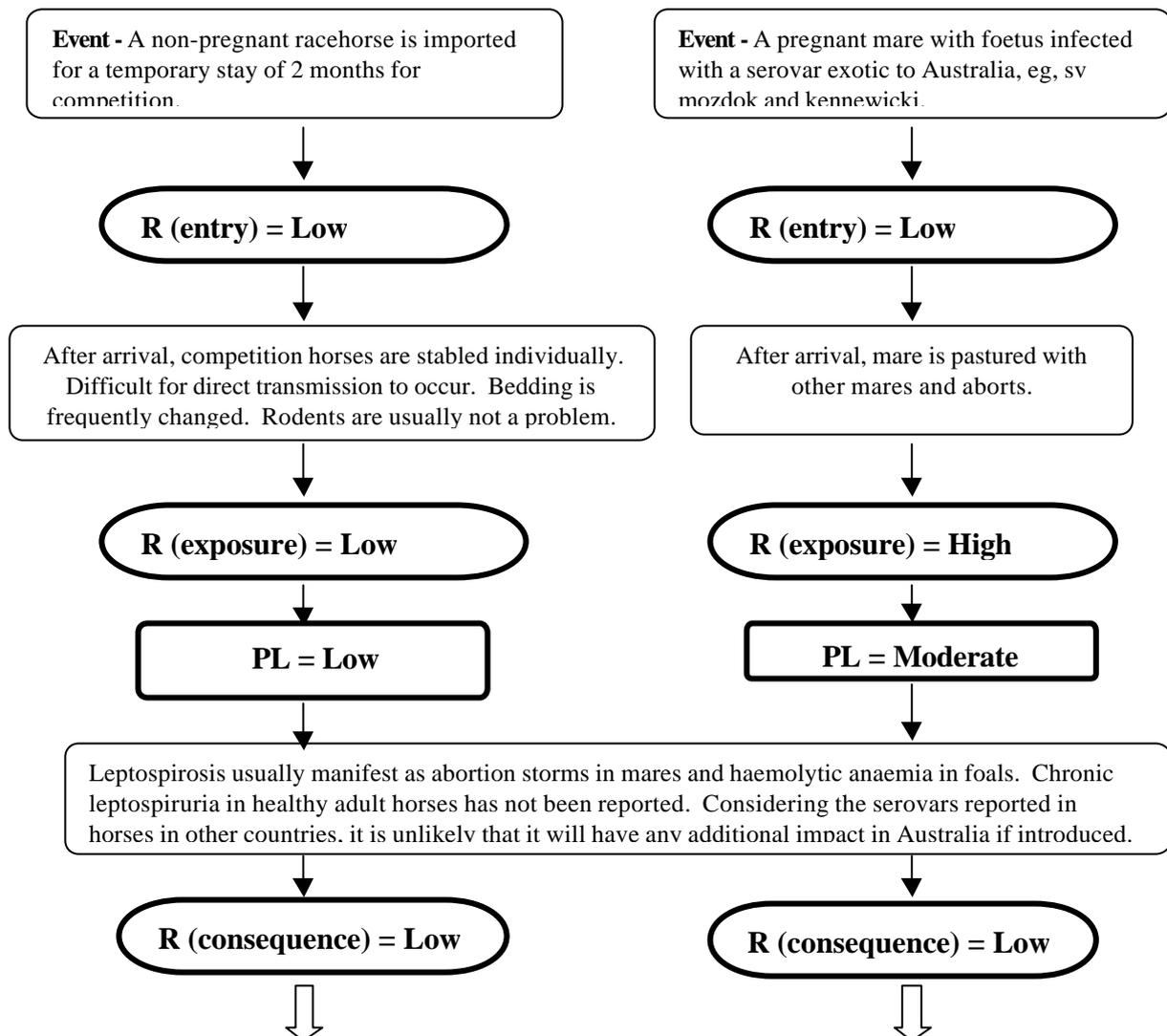
### viii. Importing horses with leptospiral infection

Clinical leptospirosis is uncommon in horses. Leptospirosis was diagnosed in less than 3% of all reported abortions over a 7 year period in Central Kentucky, USA. Yet a significant percentage of horses develop detectable agglutinating antibodies to leptospirosis. Horses are possibly a maintenance host for sv bratislava and a high percentage of European horses seroconvert to this serovar. However, bratislava is a notoriously difficult and fastidious organism to isolate and culture, so it has not been possible to assess the prevalence of bratislava leptospiruria. It has been isolated from an aborted foetus which may be a source of infection. Serovar pomona has been isolated from horses several times in Northern Ireland but no horses, cattle, pigs or sheep had antibodies to pomona despite extensive surveys.<sup>274</sup>

Of all livestock, horses are the most likely to develop recurrent uveitis following infection.

The following serovars have been isolated from horses' urine:

- grippityphosa from a horse with acute haemolytic fever,
- grippityphosa and kennewicki from mares which aborted,
- pomona from a sick foal, and
- hardjoprajitno and pomona 8 weeks after a mare had aborted.



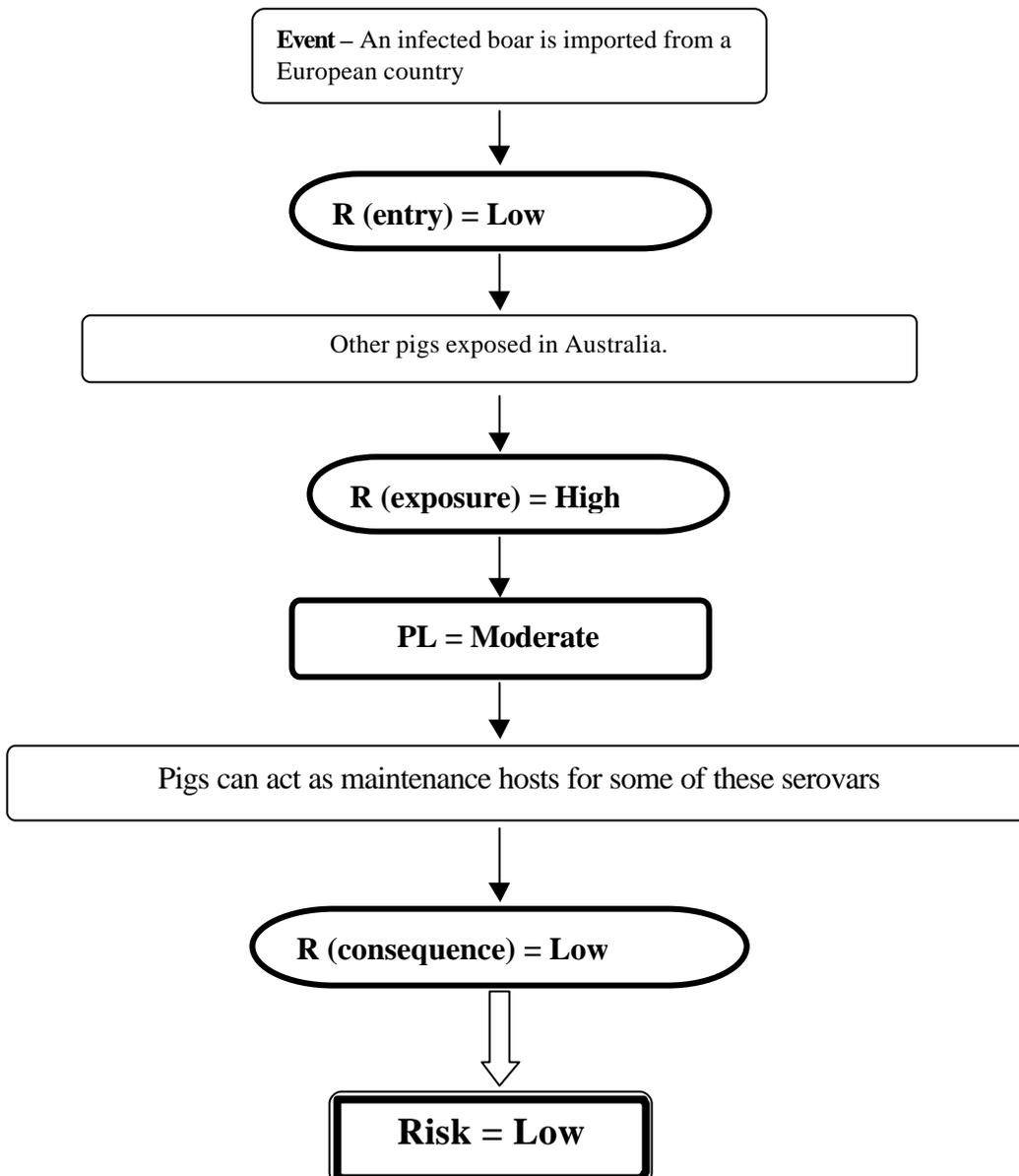
**Risk = Low**

**Risk = Low**

**ix. Live pigs infected with sg australis from Europe**

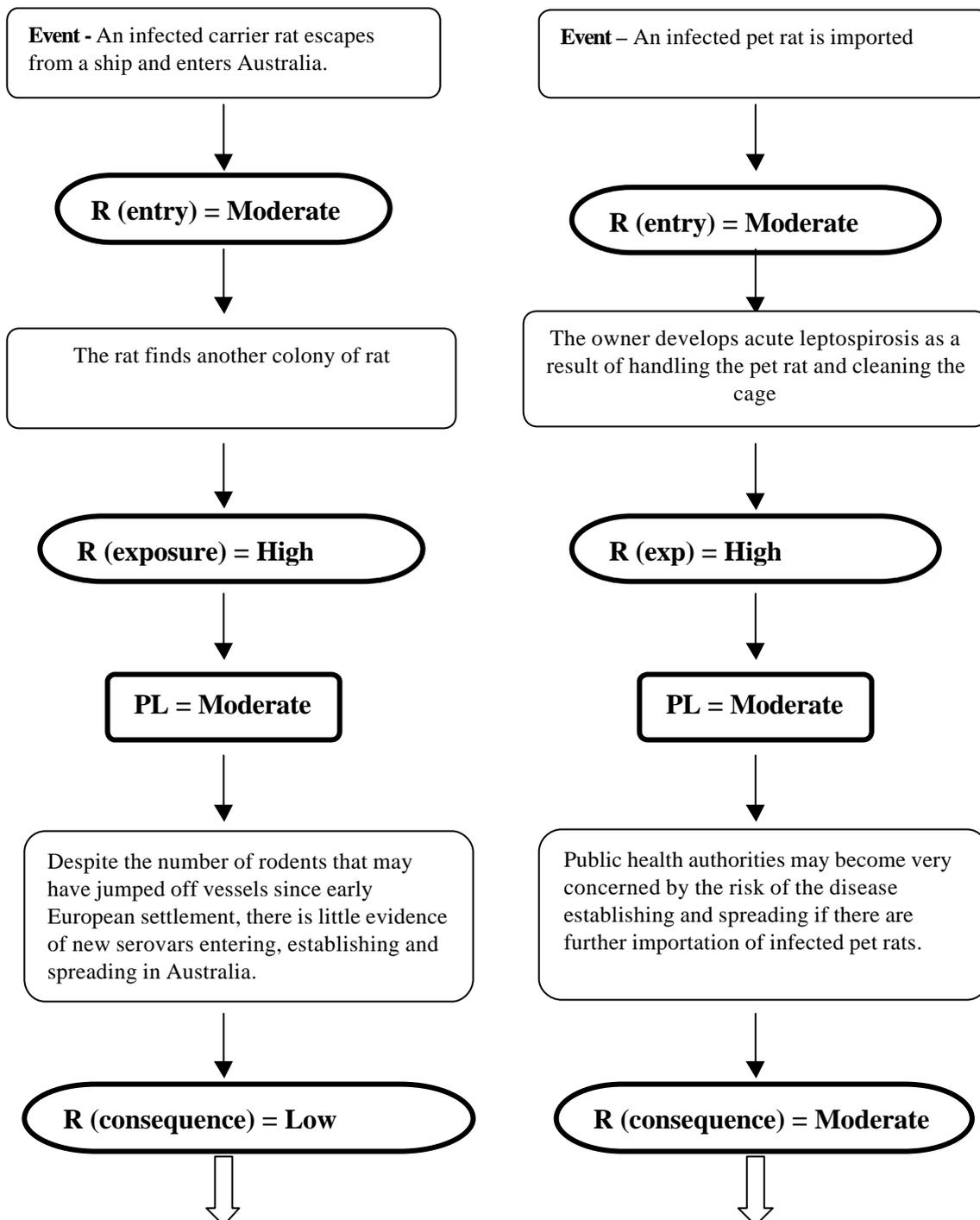
Pigs are recognised as maintenance hosts for several serovars including pomona, bratislava, and tarassovi. Pigs show a high prevalence of antibodies to the australis serogroup, which includes svs bratislava, muenchen and lora and, despite the difficulty of isolating the leptospire of this serogroup, have been confirmed as the cause of abortion and stillbirths in sows. Incidental infections with svs canicola, grippotyphosa, hardjo and others have occurred sporadically. Antibodies to icterohaemorrhagiae are widespread but this serovar rarely causes any significant disease in pigs. The economic consequence of leptospirosis in pigs can be high due to the costs of reproductive losses. Venereal transmission is thought to be very common in pigs.

Pigs have not been introduced into Australia in significant numbers since mid 1980's. Should pigs be imported, eg, from Europe, there is a risk of introducing new serovars of the australis serogroup, namely lora, and muenchen.



**x. Rodents and other small mammals**

Most rodents can act as symptomless carriers to several serovars. Clinical disease is seen mainly in the young animals. There is a risk of entry, establishment and spread is rodents escaping from ships in port.

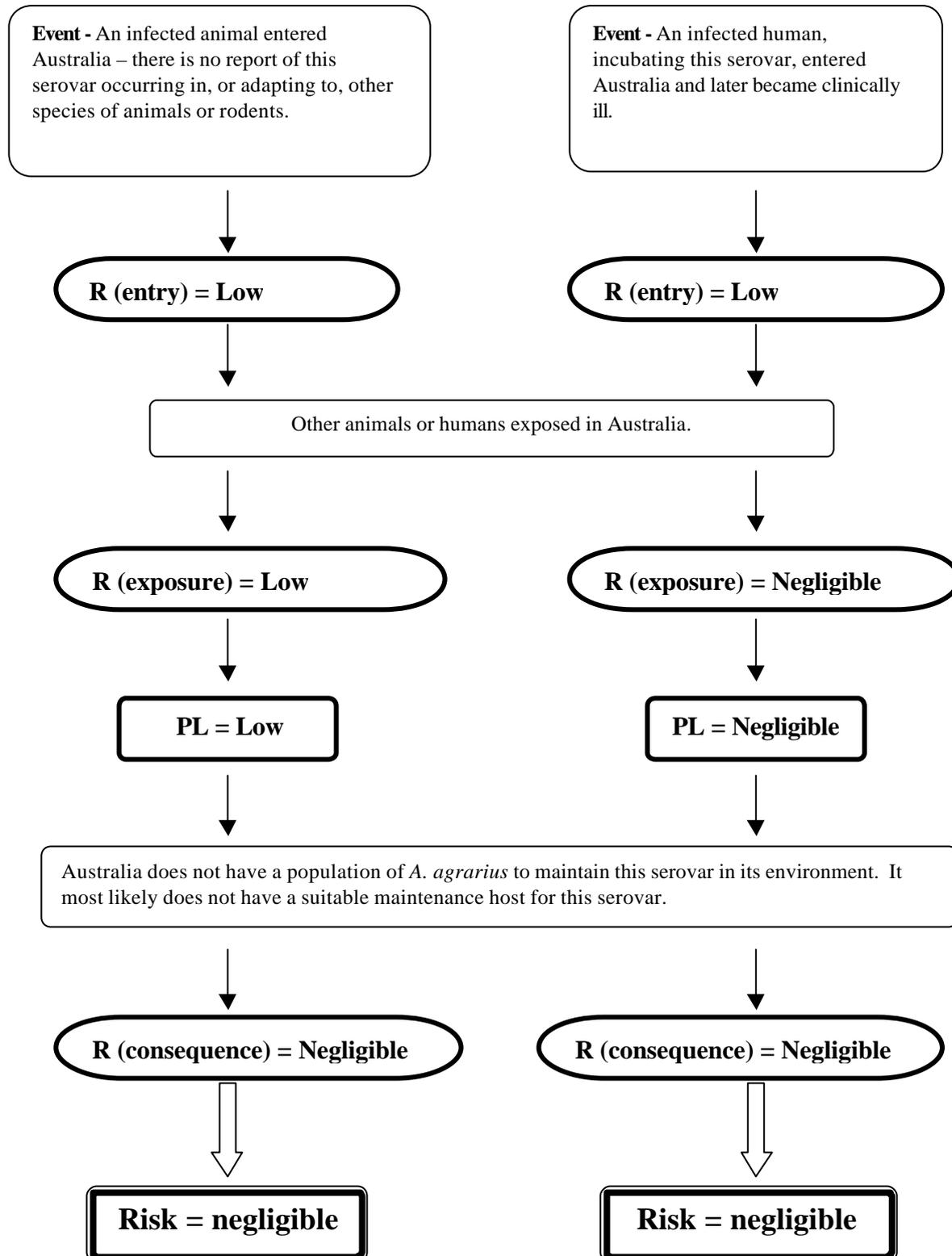


**Risk = Low**

**Risk = Moderate**

**xi. Animals or humans infected with sv lai**

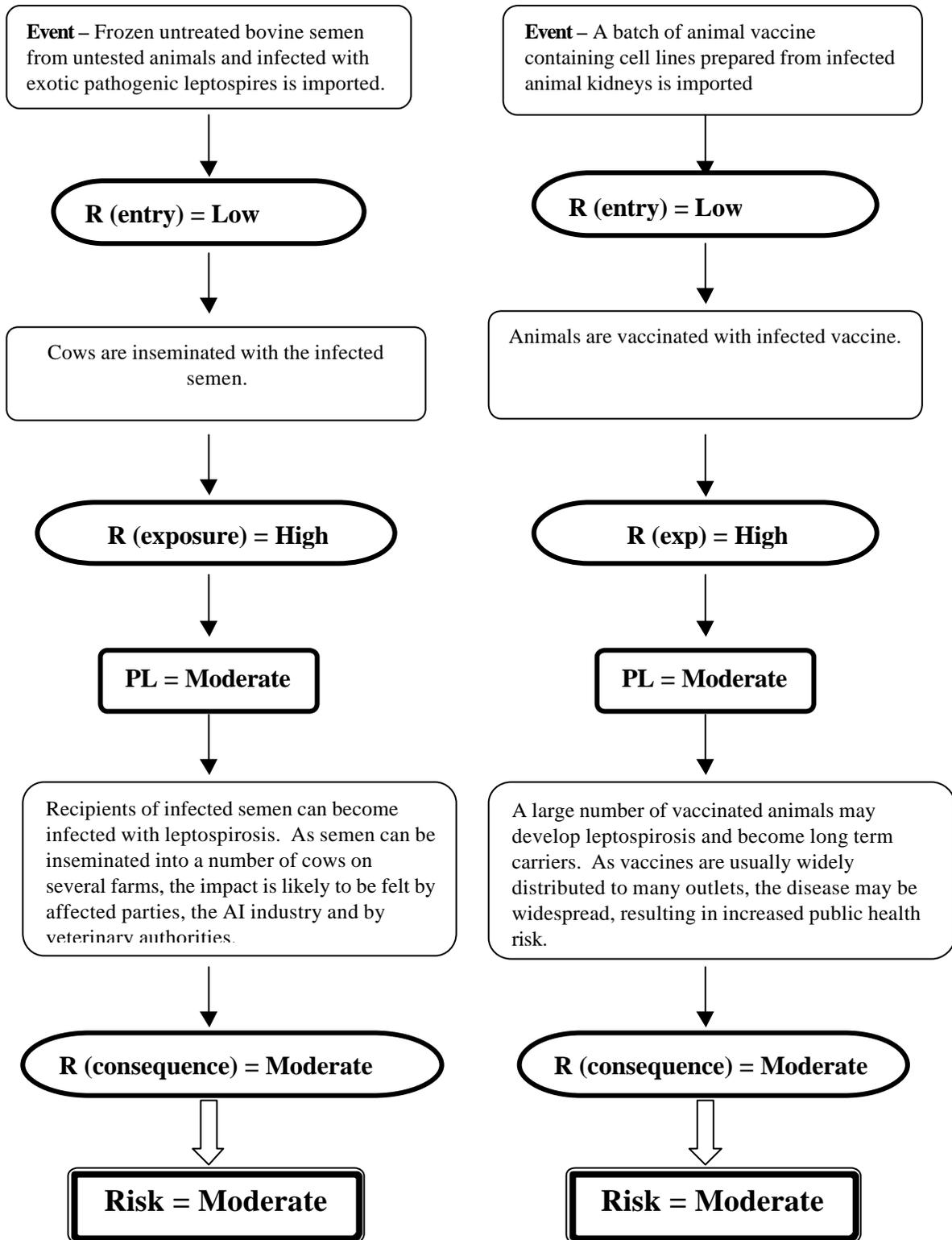
The Asian striped field mouse (*Apodemus agrarius*) is recognised as the maintenance host for sv lai, a member of the icterohaemorrhagiae serogroup. Sv lai is regarded as a serious public health risk in Asia, especially China and Korea.





**xii. Semen, embryos and tissue culture cells.**

Infected animals can shed leptospires in semen and embryos. Tissue cultures are sometimes prepared from kidneys of infected but healthy animals. There is a risk of entry, establishment and spread in untreated semen, embryos and tissue culture cells.





**xiii. Summary of overall risk**

**Table 17. Summary of risk events.**

<b>Scenario</b>	<b>Risk</b>
Importing a dog from a country with a significant stray dog problem, where canicola infection occurs and dogs are not routinely vaccinated (eg, most African, Asian or South American countries).	<b>Low</b>
Importing a dog from a country where sv canicola has not been isolated from dogs since the 1950's (eg, USA, Canada, and New Zealand).	<b>Low</b>
Importing a pig clinically infected with sv canicola (eg, Republic of South Africa).	<b>Negligible</b>
Importing a ram infected with sv canicola (eg, Portugal).	<b>Low</b>
Importing a dog from a country which reports dogs with antibody titres to svb batavia, bratislava, javanica and cynopteri, none of which has been isolated in Australia (eg, Southeast Asia).	<b>Low</b>
Importing a dog from country where infections due to sv bim occurs (eg, the Caribbean).	<b>Low</b>
Importing a bull infected with sv hardjoprajitno from a country where this serovar is endemic (eg, Ireland).	<b>Low</b>
Importing a pregnant cow with sv hardjoprajitno infection from a country where this serovar is endemic (eg, Ireland).	<b>Low</b>
Importing a cow shedding either of svb mozdok or kennewicki in urine (eg Europe or North America).	<b>Low</b>
Importing a boar shedding either of svb mozdok or kennewicki in urine (eg Europe or North America).	<b>Low</b>
Importing a non-pregnant racehorse for a temporary stay of 2 months for competition purposes (eg, North America).	<b>Low</b>
Importing a pregnant mare with foetus infected with a serovar exotic to Australia (eg, Europe - sv mozdok and North America – sv kennewicki).	<b>Low</b>
Importing a boar with sg australis infection (eg, Europe).	<b>Low</b>
An infected carrier rat escapes from a ship and enters Australia.	<b>Low</b>
Importing an infected pet rat (eg, Asia).	<b>Moderate</b>
Animal or humans infected with sv lai enter Australia (eg, Asia).	<b>Negligible</b>
Untreated frozen semen (or embryo) from untested donors infected with exotic pathogenic leptospire (eg, Europe, South America)	<b>Negligible</b>
Importing a batch of animal vaccine containing cell lines prepared from infected animal kidneys (eg, Asia).	<b>Moderate</b>

## 11. Risk evaluation

Australia's *acceptable level of protection* is the level of protection deemed acceptable by Australian public health and veterinary authorities in managing the disease within their territories. The term "acceptable level of protection" is not to be confused with the term "appropriate level of protection (ALOP)". Appropriate level of protection (ALOP) is the term used by Australia when referring to its SPS Agreement (the Agreement on the application of sanitary and phyto-sanitary measures) obligations. The SPS Agreement defines ALOP as the level of protection deemed appropriate by the Member establishing a sanitary or phytosanitary measure to protect human, animal or plant life or health within its territory.

In considering Australia's acceptable level of protection, the following are important:

- Australia has limited requirements for the control of leptospirosis within its territory;
- Australia accepts sporadic occurrence of clinical leptospirosis in animals and humans, and
- prevention and control of this disease is not mandatory.

Evaluating the overall risk in the light of Australia's acceptable level of protection involves determining the significance of the identified hazards in relation to the overall risks to the animals, people or environment concerned with or affected by them.

Where the overall risk is *negligible* or *low*, the animal or animal product meets Australia's acceptable level of protection without requiring quarantine measures. But where the overall risk is considered to be *moderate* or *high*, it may be necessary to impose quarantine measures to effectively reduce the overall risk.

## 12. Risk reduction

### a) Introduction

*Risk reduction strategies* involves identifying and implementing quarantine measures for meeting the importing country's acceptable level of protection, whilst at the same time ensuring that any negative effects on trade are minimised.

Where the subsequently 'reduced risk' derived using a particular risk reduction strategy was *low*, that strategy was considered to be acceptable.

Where the 'reduced risk' derived using a particular risk reduction strategy was *negligible*, the strategy was considered unnecessarily restrictive. Overly restrictive risk reduction strategies were either rejected, or were manipulated, if possible, so as to be less restrictive.

This procedure led to the specification of a set of acceptable risk reduction strategies for each serovar for which the overall risk was considered unacceptable in relation to Australia's acceptable level of protection.

### b) Risk reduction strategies

There are, broadly speaking, four types of risk reduction strategies for leptospirosis:

- detection,
- treatment,
- sound knowledge of history of leptospirosis in the region, and
- prevention.

Methods of detecting animals carrying leptospire include

- serology for detecting anti-leptospira antibodies,
- isolation and culture of leptospire from tissues or urine, and
- detection of leptospiral DNA.

Treatment methods include

- antibiotic therapy,
- adequate supportive symptomatic treatment for clinically ill animals, and
- disinfection of premises.

Sound knowledge of history of leptospirosis in the region include:

- disease history of the animal and
- disease occurrence in the region.

Prevention methods include

- vaccination,
- rodent control, biosecurity,

- isolation from carriers,
- surveillance,
- hygiene measures,
- avoidance and education, and
- protective clothing and occupational hygiene.

### **i. Serology**

Serology for leptospirosis, when used as a diagnostic tool, is not accurate. The immune system of individual infected animals may not respond to infection. Serovar pomona has been isolated from horses several times in Northern Ireland but antibodies to pomona have not been detected in horses, cattle, pigs or sheep despite extensive surveys.

MAT is the standard serological test for leptospirosis. It has a number of disadvantages as outlined earlier (Section 10.a.i):

- there are a number of variations of this test, affecting test sensitivity and specificity;
- it is slow, tedious and potentially biohazardous;
- it is highly subjective and results depend on the skills and experience of the laboratory technician conducting the tests;
- the quality of the live leptospira antigen used in the test can vary, depending on the quality of the culturing procedures used to maintain cultures of leptospire;
- the interpretation of a diagnostic test is difficult as there are different species response to leptospirosis;
- it cannot detect seronegative animals with leptospiuria;
- a large battery of antigens is required to test for all pathogenic serogroups;
- some cross reactions occur with different serovars in different serogroups; and
- it is not a good predictor of infection in individuals.

However, MAT can be a useful tool for diagnosis in animals where specimens collected are spaced over several weeks. Through this protocol of specimen collections, seroconversions, or maintained high level titres, are useful indicators of current infection or transmission risk. A single specimen is generally not useful for determination of a disease status in humans or animals.

The CFT and ELISA are of value in detecting early leptospirosis. The CFT cannot differentiate serovars while the ELISA is still undergoing further development and evaluation.

Due to the disadvantages of the MAT and the limitations of the CFT and ELISA, serology of a single specimen is of little value in detecting infective carriers.

### **ii. Isolation and culture**

The isolation and culture of leptospire is a useful diagnostic tool in the early febrile phase of disease and for confirming diagnosis in aborted foetuses, stillbirths and urine. However, the identification of leptospire in urine is difficult. Disadvantages of using the culture technique are:

- 3 to 20 days for culture in liquid media;
- weeks or even months to identify the leptospire in the culture;

- the necessary high level of asepsis and quality control; and
- difficulties in isolating fastidious leptospire.

Due to the difficulties of isolating leptospire from healthy animals, it is of little value in detecting infective carriers. While it appears that different species may have specific, or differing nutritional requirements, the entire reference collection at the WHO Leptospiral Reference Laboratory in Brisbane, Queensland is maintained on the EMJH (Ellinghausen McCullough Johnson and Harris) commercial media.

### **iii. Detection of leptospiral DNA**

The polymerase chain reaction (PCR) is the recognised detection, or diagnostic method for leptospiral DNA. This technique offers the best hope for rapid and accurate diagnosis. The WHO Leptospiral Reference Laboratory in Brisbane, Queensland is currently developing a “real time” PCR which is expected to be effective and reproducible for detecting leptospiral DNA in most body fluids such as blood, serum, CSF and urine.

Several analysis techniques are currently available for the typing, classification or comparison of isolates, including:

- restriction fragment length polymorphisms (RFLP),
- pulsed-field gel electrophoresis (PGFE),
- arbitrarily primed PCR (APPCR),
- mapped restriction site polymorphisms (MRSP), and
- random amplified polymorphic DNA fingerprinting (RAPD).

Nucleic acid detection technology requires appropriately equipped laboratories stocked with appropriate primers, reference DNA samples and the DNA of the strain to be identified. Few laboratories are appropriately equipped.

Due to the difficulties in accessing well-equipped laboratories and the groundwork required to ensure that appropriate raw materials are available for a successful test, this test is not a viable option. But the likely development of serovar-specific molecular probes amplified by PCR should simplify typing and may become a useful risk reduction tool.

### **iv. Antibiotics**

Leptospirosis can be treated with antibiotics. Laboratory tests show that leptospire are susceptible to all clinically useful antibiotics except chloramphenicol and rifampicin. While treatment failures are common, antibiotic resistance has not been reported. The most recommended antibiotics along with their advantages and disadvantages are:

- penicillin
  - commonly used in humans,
  - high doses are required,
  - problems with hypersensitive patients,
  - procaine penicillin is prohibited in performance animals, and
  - can result in residues in meat and milk of animals;
- erythromycin

- used in human patients hypersensitive to penicillin;
- tetracyclines and oxytetracyclines
  - contraindicated in humans with renal insufficiency, and in pregnant animals and newborns as it can cause yellow staining of teeth,
  - LA Oxytetracycline 200 diffuses into urine and foetus,
  - LA Oxytetracycline 200 withholding period is 42 days for meat and 7 days for milk,
  - LA Oxytetracycline 200 often require divided doses,
  - LA Oxytetracycline 200 may cause anaphylaxis in young cattle,
  - LA Oxytetracycline 200 is contraindicated in horses, dogs and cats,
  - excreted unchanged mainly via the kidneys, and
  - can accumulate in patients with renal dysfunction;
- streptomycin (including dihydrostreptomycin)
  - injection contraindicated in humans as is toxic to the auditory nerve,
  - it is an irritant to muscle tissue when given as an injection,
  - it requires divided doses,
  - long term residues in kidneys and other offals,
  - good efficacy against leptospiruria in cattle and pigs,
  - one step resistance can be produced easily in the laboratory, and
  - banned from use in food producing animals in several countries;
- amoxicillin
  - LA amoxicillin is contraindicated in horses and small herbivores such as rabbits and guinea pigs and in cases of penicillin hypersensitivity,
  - LA amoxicillin withholding period is 30 days for meat and 5 days for milk, and
  - LA amoxicillin require divided doses and often a second dose is necessary in 24 hours for gram-negative bacteria;
- doxycycline
  - no veterinary approved injectable forms currently available,
  - offer longer half-life and higher CNS penetration when compared to tetracyclines,
  - penetrates body tissues and fluids better than tetracyclines
  - used in small animals especially dogs and cats,
  - contraindicated in pregnant animals and newborns as it can cause yellow staining of teeth,
  - not to be used as parenteral injections in horses,
  - need daily doses for 5 to 7 days,
  - excreted mainly via the faeces via non-biliary routes in inactive form, and
  - does not accumulate in patients with renal dysfunction.

The choice of antibiotics for treatment of leptospirosis or leptospiruria is not clearcut. No antibiotic has demonstrated efficacy in all cases. Streptomycin is usually the drug of choice in animals despite its use being forbidden in food-producing animals in several countries. Recent research has shown streptomycin/penicillin to be most effective in treating swine experimentally infected with *sv pomona* and later demonstrating persistent leptospirosis.<sup>275</sup> Unfortunately, there is no work comparing streptomycin/penicillin with streptomycin as commercial preparations of this combination have been banned from use in several countries. Daily injections of oxytetracycline, tylosin and erythromycin

were effective in treating all sows but in doses well above those recommended by the manufacturer. Treatment with ceftiofur, tiamulin or ampicillin was not effective.

Cefotaxime and moxalactam, but not other cephalosporins, were effective in clearing the hamster kidneys of leptospire. Both chlortetracycline and doxycycline were active against the clinical leptospirosis but could not clear all hamster's kidneys of leptospire. All penicillins assessed, ampicillin, bacampicillin, cyclacillin, piperacillin and mezlocillin, were effective against clinical leptospirosis but only ampicillin, bacampicillin and mezlocillin cleared all hamster kidneys of leptospire.<sup>276</sup>

A stallion with clinical leptospirosis and leptospiruria was initially treated with trimethoprim/sulfadiazine and procaine penicillin G for 5 days and then sodium penicillin and gentamycin for 4 days without success. This was followed by intravenous ticarcillin/clavulonic acid combination three times a day for 9 days resulting in the successful treatment of clinical leptospirosis. The horse was discharged with instruction for administration of streptomycin at 10-mg/kg twice daily.<sup>277</sup>

There are significant risks involved with treating animals prior to import into Australia for the following reasons:

- the questionable efficacy of antibiotics;
- the possible undesirable side effects of antibiotic administration;
- streptomycin no longer available in several countries;
- residue problems from the high extra-label dosages required for some antibiotics.

The effectiveness of treating infective carriers with antibiotics depends on a number of factors including:

- suitability of antibiotics for animal species being treated;
- extent of kidney damage affecting excretion of certain antibiotics;
- effectiveness of antibiotics *in vivo*;
- availability of antibiotics; and
- dose rate of antibiotics.

Because of these factors, a blanket recommendation on antibiotic use for treating infective carriers is not possible.

## **v. Disinfection of premises and hygiene**

Leptospire are destroyed by

- heat over 42 C but not by cold or freezing,
- acid under pH 6.5,
- alkali above pH 8.0,
- heavy metals,
- halogens,
- detergents (including soaps, free fatty acids and bile salts), and
- desiccation.

## **vi. Disease history**

While good livestock health and production records may provide details on cases of clinical leptospirosis, serosurveys suggest that most leptospiroses are asymptomatic. Furthermore maintenance hosts are often symptomless carriers of leptospirosis. Thus knowledge of the disease history of animals provides very little information on the state of carriers. As cases of leptospirosis usually occur sporadically they are often unreported. Knowledge of the disease occurrence in a region also provides very little information on the state of carriers.

## **vii. Vaccination**

Vaccines are generally available for:

- dogs
  - canicola
  - icterohaemorrhagiae
- cattle
  - hardjobovis or hardjoprajitno
  - pomona
- pigs
  - pomona
  - tarassovi
  - bratislava

Advantages of vaccination are

- can protect against clinical leptospirosis and leptospiruria caused by serovar included in vaccine in animals not previously exposed to that serovar; and
- useful in controlling disease outbreaks.

Disadvantages of vaccination

- efficacy of vaccine varies considerably from country to country;
- does not treat leptospiruric animals;
- usually protects an animal for up to 6 months;
- causes highly variable immune response, and
- does not confer cross-protection to other serovars.

## **viii. Rodent control and biosecurity measures to ensure isolation from carriers**

Good rodent control and other biosecurity measures help to prevent infection of accidental hosts and spread of disease from infected accidental host to a maintenance host. These factors need to be considered for pre-export quarantine and post-arrival quarantine. In high-risk areas, outbreaks of leptospirosis have been prevented where regular rat control programs are conducted.

## **ix. Surveillance**

Serosurveys consistently show that a proportion of animals in a population to have anti-leptospiral antibodies. They are of no value for demonstrating regional freedom from disease as leptospirosis can occur seasonally and outbreaks can occur during periods of unexpected and extended heavy rains. However, a good epidemiological understanding of leptospirosis can overcome problems with cross-reactions and surveillance using a carefully selected battery of antigens allowing close monitoring of the incidence of particular serovars of leptospirosis.

#### x. Hygiene

Simple hygienic measures such as washing of hands and wearing protective clothing may protect many humans from leptospirosis and can help reduce the risk of transmission. However hygiene is not effective in treating infective carriers.

***Comment:***

There is no single risk reduction strategy apart from excluding all animals from entering a country that will ensure that risk is reduced. Even the combination of several risk management strategies may not necessarily reduce the risk of leptospires in live animals entering, establishing and spreading in a country to the importing country's acceptable level of protection.

Australia has allowed importation of cattle, buffalo, sheep, goats, alpacas, llamas, horses, cats, guinea pigs, hamsters and laboratory rats and mice without any testing requirements for leptospirosis. There are currently no import conditions for live pigs and deer, but there are import protocols requiring dogs to be tested for *canicola*.

### **13. Implications for quarantine policy**

Risk reduction strategies should be based on scientific evidence. There is a school of thought that exotic strains, if introduced, may eventually adapt to a new host within the country and become impossible to eradicate. However, there does not appear to be any scientific evidence or pertinent information to support this hypothesis.

#### **a) Human quarantine policy**

There is concern that humans are a risk of introducing exotic leptospires into Australia. There are no quarantine requirements for leptospirosis for humans entering Australia. Australian public health authorities regard the risk of exotic serovars establishing and spreading in Australia if introduced by infected humans to be negligible. Clinical cases of leptospirosis due to exotic strains have occurred in humans after entering Australia from overseas (Smythe L, *pers comm*). The reasons why risk is considered negligible are:

- humans are regarded as end-hosts,
- humans usually do not become chronic renal carriers or excretors,
- human urine is too acidic for the survival of leptospires, and
- humans adopt more hygienic measures than do animals.

Risk is highest with hospital workers involved with cases with acute leptospirosis. Hypothetically, infected vegetarians whose urine are usually more alkaline are a risk but there is no evidence of spread having occurred in this way.

#### **b) Animal quarantine policy**

The implications for quarantine policy should be considered for each of the risk events described in the previous chapter. According to the overall risk, it is appropriate to require a risk reduction strategy for:

- semen and embryos,
- commodities containing animal derived tissue culture cells, and
- pet rodents and small mammals.

It is also appropriate to consider deleting the current risk reduction strategy for leptospirosis in dogs and maintaining the current quarantine policies for livestock, laboratory rodents, cats and horses.

##### **i. Semen and embryos**

The risk of infective leptospires entering Australia via either semen or embryos is reduced to acceptably low levels if antibiotics are used in the processing of semen and embryos. As this is standard practice and most antibiotics are effective against leptospires, no further risk reduction is necessary except to require certification that the OIE *Code* (Appendices 4.2.1.1. and 4.2.1.2.) guidelines were complied with. Furthermore, trypsin is usually used in the washing of embryos for export and can destroy leptospires before antibiotics are added during processing.

**ii. Animal derived tissue cultures**

As antibiotic use is standard practice in processing of tissue culture cells to remove contaminating bacterial flora, no further risk reduction is necessary.

**iii. Pet rodents and other small mammals**

Risk reduction strategies are very limited for small mammal pets. After drawing the volume of blood required for serology, the small mammal may have insufficient blood for survival. It is difficult, if not impossible, to collect sufficient urine for culturing. Therefore individual testing of rodents and small mammals for leptospirosis is not possible with current technology. However, it is possible to assess breeding colonies by sacrificing a small proportion of animals for testing for leptospirosis. The proposed risk reduction strategy is that pet rodents and other small mammals must come direct from colonies tested free from leptospiral strains 'exotic' to Australia and relevant to the species.

In the near future, it may be possible to test a sample of urine of individual pet rodents or small mammals for leptospiral DNA using "real time" PCR, thus eliminating the need for colony testing.

**c) Public health risks during quarantine and transport**

There is a small risk of people becoming infected with leptospirosis whilst handling animals during pre-export quarantine, transport, and post-arrival quarantine. Health measures should be adopted to prevent infection of humans handling these animals during such periods.

**d) Possible effects of stricter notification requirements for leptospirosis**

If uniform stricter notification and regulatory requirements for leptospirosis were imposed nationally, the consequence assessments would be revised to 'moderate'. Regulatory action need not require quarantine but should allow for less drastic measures such as issuing a treatment notice.

By revising the risk events as summarised in Table 18, overall risk may change:

**Table 18. Summary of modified risk events.**

<b>Scenario</b>	<b>Risk</b>
Importing a dog from a country with a significant stray dog problem, where canicola infection occurs and dogs are not routinely vaccinated (eg, most African, Asian or South American countries).	<b>Moderate</b>
Importing a dog from a country where sv canicola has not been isolated from dogs since the 1950's (eg, USA, Canada, and New Zealand).	<b>Low</b>
Importing a pig clinically infected with sv canicola (eg, Republic of South Africa).	<b>Low</b>
Importing a ram infected with sv canicola (eg, Portugal).	<b>Low</b>

<b>Scenario</b>	<b>Risk</b>
Importing a dog from a country which reports dogs with antibody titres to sv <sub>s</sub> batavia, bratislava, javanica and cynopteri, none of which has been isolated in Australia (eg, Southeast Asia).	<b>Moderate</b>
Importing a dog from country where infections due to sv bim occurs (eg, the Caribbean).	<b>Moderate</b>
Importing a bull infected with sv hardjoprajitno from a country where this serovar is endemic (eg, Ireland).	<b>Moderate</b>
Importing a pregnant cow with sv hardjoprajitno infection from a country where this serovar is endemic (eg, Ireland).	<b>Moderate</b>
Importing a cow shedding either of sv <sub>s</sub> mozdok or kennewicki in urine (eg Europe or North America).	<b>Moderate</b>
Importing a boar shedding either of sv <sub>s</sub> mozdok or kennewicki in urine (eg Europe or North America).	<b>Low</b>
Importing a non-pregnant racehorse for a temporary stay of 2 months for competition purposes (eg, North America).	<b>Low</b>
Importing a pregnant mare with foetus infected with a serovar exotic to Australia (eg, Europe – sv mozdok and North America – sv kennewicki).	<b>Moderate</b>
Importing a boar with sg australis infection (eg, Europe).	<b>Moderate</b>
An infected carrier rat escapes from a ship and enters Australia.	<b>Moderate</b>
Importing an infected pet rat (eg, Asia).	<b>Moderate</b>
Animal or humans infected with sv lai enter Australia (eg, Asia).	<b>Low</b>
Untreated frozen semen (or embryo) from untested donors infected with exotic pathogenic leptospire (eg, Europe, South America)	<b>Negligible</b>
Importing a batch of animal vaccine containing cell lines prepared from infected animal kidneys (eg, Asia).	<b>Moderate</b>

In such case, it may be appropriate to redevelop a quarantine policy that include a risk reduction strategy for:

- dogs for several serovars from some countries,
- livestock, and
- pregnant mares.

## 14. Proposed options for export conditions

Australia is negotiating for deleting test and antibiotic injection requirements for export of animals. The case against these requirements is detailed in “Section 12 – Risk Reduction”. Although negotiations have only recently begun, some countries have already agreed to delete test requirements and/or to change to antibiotics other than dihydrostreptomycin/ streptomycin (S/DHS).

There are some 221 export protocols from 63 countries that have health requirements for leptospirosis for import of animals and their genetic material from Australia.

Table 19 shows the number of livestock exported from Australia in 1999 according to the data provided by the Meat and Livestock Australia. The shaded areas indicate livestock type that has quarantine conditions for leptospirosis imposed by the importing country. Priority should be given to renegotiating these quarantine conditions.

**Table 19: Number of livestock exported from Australia in 1999**

Country	Cattle			Buffalo	Sheep		Goat	
	Breed	Slaughter	Fatten		All	Breed	Slaughter	Breed
Bahrain	0	0	0	0	0	425636	0	0
Brunei	0	2097	0	0	0	0	0	0
China	0	0	0	0	836	0	825	0
Egypt	0	206234	5097	0	0	146037	0	0
Indonesia	34	8124	57707	0	0	250	0	0
Japan	0	135	12023	0	0	0	0	0
Jordan	0	1316	0	0	0	512892	4	0
Korea	5	0	0	0	0	0	0	0
Kuwait	0	959	0	0	121	1138762	0	0
Libya	0	21415	0	0	0	0	0	0
Malaysia	1427	37117	7246	722	0	11704	1163	17111
Mexico	7076	625	0	0	74030	28174	0	0
New Zealand	19	0	0	0	2	0	3	0
Oman	0	0	0	0	0	424181	0	0
Other Middle East	393	1259	0	0	0	223139	0	0
Pacific Islands	0	0	0	0	2	0	0	0
Philippines	847	2513	183974	0	0	0	429	0
Qatar	0	458	0	0	0	264864	0	0
Singapore	0	34	0	0	0	8931	116	1273
Taiwan	0	0	0	0	0	0	67	0
UAE	8	0	0	0	0	742004	0	3771
USA	11	0	0	0	0	0	0	0
Total	9820	282286	266047	722	74991	3926574	2607	22155

United States of America have no leptospirosis requirements for animals imported from Australia except for camelids. As camelids are not known to be a maintenance host, this is probably an oversight by both United States Department of Agriculture (USDA) and Biosecurity Australia (formerly a division of the Australian Quarantine and Inspection Services). An approach should be made to delete this condition.

In 1999, New Zealand had proposed to introduce a requirement that all imported horses either undergo a MAT serology test or be injected with DHS/S or long-acting oxytetracycline. As thousands of horses had already travelled from Australia to New Zealand with minimal restrictions, New Zealand have agreed to delete these requirements for horses from Australia.

## 15. OIE International Animal Health Code Leptospirosis Chapter.

The Office International des Epizooties (OIE) is an intergovernmental organisation created by the International Agreement of 25 January 1924 signed by 28 countries. The headquarters is based in Paris, France. As of December 2000, the OIE comprised 155 Member Countries. The OIE operates under the authority and control of an International Committee formed by permanent Delegates designated by the Governments of the Member Countries.

The Central Bureau, headed by a Director General appointed by the International Committee, conducts the activities of the organisation and implements the resolutions of the Committee. The missions of the OIE are to:

- inform Governments of the occurrence and course of animal diseases throughout the world, and of ways to control these diseases
- coordinate, at the international level, studies devoted to the surveillance and control of animal diseases
- harmonise regulations for trade in animals and animal products among Member Countries.

The International Committee, the highest authority of the OIE, meets for a five day General Session each year. Voting by Delegates on the International Committee respects the democratic principle of 'one country, one vote'. The functions of the International Committee include:

- adopting international standards in the field of animal health, especially for international trade; and
- adopting resolutions on the control of the major animal diseases;

The International Committee has created an International Animal Health Code Commission, a Specialist Commission, to study the problems of epidemiology and control of animal diseases, and issues related to the harmonisation of international regulations. It deals with regulatory rather than scientific matters and assists the Commission with revising and updating the OIE International Animal health Code (*Code*).

Hence, any proposal to change parts of the OIE *Code* must have the support of the International Animal Health Code Commission before being voted on by the General Session of the International Committee.

Leptospirosis is an OIE List B Disease, that is, it is a transmissible diseases which is considered to be of socio-economic and/or public health importance within countries and which are significant in the *international trade of animals* and animal products. The 2000 OIE (*Code*) states:

### CHAPTER 2.2.4. LEPTOSPIROSIS

**Article 2.2.4.1.** Standards for diagnostic tests and vaccines are described in the *Manual*.

**Article 2.2.4.2.** *Veterinary Administrations of importing countries* should require: for domestic ruminants, equines and pigs for breeding or rearing the presentation of an *international veterinary certificate* attesting that the

animals:

- 1) Showed no clinical sign of leptospirosis on the day of shipment;
- 2) were kept in an *establishment* in which no clinical sign of leptospirosis was officially reported during the 90 days prior to shipment;
- 3) were injected twice with 25 mg dihydrostreptomycin per kg of live body weight at an interval of 14 days, the second injection being given on the day of shipment (this point is subject to up-dating);
- 4) When required by the *importing country*, were subjected to a diagnostic test for leptospirosis with negative results.

**Article 2.2.4.3.** *Veterinary Administrations of importing countries* should require: for semen of ruminants and pigs: the presentation of an *international veterinary certificate* attesting that the semen was collected, processed and stored in conformity with the provisions of either Appendix 3.2.1., or Appendix 3.2.2., or Appendix 3.2.3., as relevant.

**Article 2.2.4.4.** *Veterinary Administrations of importing countries* should require: for embryos/ova of ruminants and pigs: the presentation of an *international veterinary certificate* attesting that the embryos/ova were collected, processed and stored in conformity with the provisions of Appendices 3.3.1., 3.3.2. or 3.3.4., as relevant.

This Chapter should be revised for the following reasons:

1. Leptospirosis occurs world-wide. There are no countries known or reported to be free from leptospirosis. Very few countries, if any, consistently adopt sanitary measures within its borders which are in harmony with Article 2.2.4.1. of the OIE *Code*.
  2. The Chapter is restricted to ruminants, equines and pigs and to semen and embryos of ruminants and pigs. Other susceptible animal species such as dogs and animal products such as canine and equine semen can be a source of leptospirosis.
  3. Dihydrostreptomycin is not approved for use in food producing animals in most countries because of antibiotic residues concerns in food and its toxic effects on the human ear and kidney. Also, it is not necessarily the most effective treatment for all serovars in all animal species that can shed leptospores. Further, other antibiotic treatments are available for treating leptospirosis.
- There is a need for country members of the OIE to review the OIE *Code* Chapter on leptospirosis.

The issues being suggested for review include whether:

1. Leptospirosis should continue to be regarded as an OIE List B disease in light of the lack of evidence of leptospirosis being a disease of socio-economic and/or public health importance within countries. In most countries, other zoonotic diseases not listed on the OIE List B disease are regarded to have greater importance. Examples include tick-borne encephalitides, enterohaemorrhagic *E. coli* and bat lyssavirus. Also, the OIE *Code* does not have guidelines for

some zoonotic diseases considered by most countries to be more significant than leptospirosis, for example, Q fever and trypanosomiasis. Further, while some leptospirosis outbreaks are reported by the World Health Organisation (WHO), it is not listed as a disease covered by the WHO Communicable Disease Surveillance and Response Department on the WHO website.

2. Leptospirosis should be a notifiable disease of both humans and animals in the importing country before that country can consider imposing import conditions for leptospirosis on animals entering the country;
3. A disease should be notifiable in the exporting country if the disease is to be officially reported;
4. Appropriate and effective quarantine measures should apply to all animal species that can shed leptospores;
5. The *Code* should specify the type of antibiotics being used for treatment of animals; and
6. The *Code* should include an option for vaccinating animals against leptospirosis.

The review provides information on which Agriculture Fisheries and Forestry Australia (AFFA) will use to formulate proposed changes to the OIE *Code* Chapter on leptospirosis.

**Appendix 1 Table of *Leptospira* serovars sorted by serogroup and genomospecies.**

SERO-GROUP	GENOMOSPECIES							
	Interrogans	borg-petersenii	inadai	Noguchii	santarosai	weilii	kirshneri	meyeri
Australis	Australis Bangkok bratislava fugis hawain jalna lora muenchen wewak	pina		bajan barbadensis nicaragua peruviana rushan			ramisi	
Autumnalis	autumnalis bankinang bulgarica carlos mooris rachmati weerasinghe	srebarna		fortbragg	alice		bim bulgarica butembo erinaceiauriti lambwe	
Ballum		arborea ballum castellonis guangdong kenya soccoestomes			peru?			
Bataviae	bataviae losbanos paidjan	moldoviae		argentiensis claytoni	balboa bataviae brasiliensis kobbe rioja		djatzi bafani	
Canicola	benjamini bindjei broomi canicola dukou jonsis kuwait portlandvere qunjian scheuffneri sumneri		malaya				galtoni kamituga	
celledoni		anhoa				celledoni hainan mengdeng		
cynopteri					naparuca tingomaria		cynopteri	
djasiman	djasiman gurungi sentot			huallaga			agogo	
grippotyphosa	grippotyphosa liangguang muelleri valbuzzi				canalzonae		grippotyphosa ratnapura valbuzzi vanderhoeden	
hebdomadis	hebdomadis kremastos	jules nona worsfoldi			abrahamson borincana figeiro goiano	longnan	kabura kambale	

SERO-GROUP	GENOMOSPECIES							
	Interrogans	borg-petersenii	inadai	Noguchii	santarosai	weilii	kirshneri	meyeri
					kremastos maru sanmartini			
icterohaemorrhagiae	birkini budapest copenhageni gem honghe icterohaemorrhagiae lai mankarso monymusk mwogolo naam nanxi smithi	tonkini	icterohaemorrhagiae				bogvere dakota mwogolo ndahambukuje ndambari	
javanica		ceylonica dehong harbola javanica menoni poi sorexjalna yaan zhenkang			fluminensi may vargonicas	coxi mengma menrun		sofia
louisiana	lanka			louisiana orleans				
lyme			lyme					
manhao			lincang manhao 4			qingshui (manhao 2)		
mini	szwajizak	mini			beye georgia ruparupae szwajizak tabaquite	hekou		perameles
panama			mangus	crisobali panama				
pomona	cornelli kennewicki monjakov pomona			pomona proechimys	dania tropica		kunming mozdok tsaratsova	
pyrogenes	abramis biggis camlo guaratuba manilae pyrogenes robinsoni	hamptoni kwale		mycastoris	alexi bagua cenepa princestown pyrogenes varela	menglian		

SERO-GROUP	GENOMOSPECIES							
	Interrogans	borg-petersenii	inadai	Noguchii	santarosai	weilii	kirshneri	meyeri
ranarum	evansi							ranarum
sarmin	waskurin				machiguengario weaveri	sarmin		
sejroe	geyaweera haemolytica hardjo-(prajitno) jin medanensis recreo ricardi roumanica saxkoebing	balcanica dikkeni hardjo(bovis) istrica nero nyanza polonica sejroe			caribe gorgas guaricurus trinidad	unipertama		
shermani			aguarina	carimagua	babudieri luis shermani			
tarassovi		gengma guidi kanana kisuba moldoviae tarassovi tunis yunxian	kaup		aguatica atchafalaya atlantae bakeri bravo chagres darien gatuni navet rama sulzeriae	langati mogdeni vughia		
unknown					banana 1 peru wawain			(spp <i>fainei</i> – hurstbridge

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