Salmonella in Australian marsupials

Fact Sheet

March 2025

Key points

- Bacteria of the genus *Salmonella* have been detected in a range of wild and captive Australian marsupials.
- Disease associated with *Salmonella* infection (salmonellosis) can result in diarrhoea, lethargy, anorexia and dehydration. In Australian marsupials, clinical disease is typically only observed in captive animals (particularly hand-reared macropod joeys), or in individuals experiencing stress or other diseases.
- Both wild and domestic animals that carry *Salmonella* spp. pose a zoonotic risk to humans via faecal-oral transmission.
- Appropriate hygiene in animal care, handling and food preparations helps to reduce zoonotic spillover risk.

Aetiology

Salmonella are Gram-negative, rod-shaped bacilli belonging to the family *Enterobacteriaceae*, genus *Salmonella* [1]. *Salmonella* nomenclature is complex and evolving. Currently, the genus *Salmonella* consists of two species, *Salmonella enterica* and *S. bongori*. *Salmonella enterica* is further divided into six subspecies, which are referred to by a Roman numeral and a name [2, 3]. Between the two species, there are over 2,500 recognised serovars of *Salmonella*, the majority belonging to *Salmonella enterica* [4]. The individual names of each serovar may be used for identification; antigenic formulae are used for unnamed serovar[1]. Salmonellosis refers to clinical disease associated with *Salmonella* infection (in either humans or animals).

One Health implications

Wildlife and the environment: Although *Salmonella* are present in a range of wild species, both in Australia and around the world, infection is not typically associated with disease in wildlife [5]. Salmonellosis most often occurs in captive wildlife or in individuals experiencing stress or co-morbidities. Although it is unlikely to directly pose a significant threat to the conservation of

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[1] At the first citation of a named serovar or "serotype", the genus name is followed by the word "serovar" / "serotype" or the abbreviation "ser." or "var.", then the serovar name. Subsequently, the name may be written with the genus followed directly by the serovar name. The serovar name is capitalised and not italicised.

Australian marsupials, *Salmonella* may exacerbate other stressors and has the potential to increase the vulnerability of wild populations to other diseases [6, 7].

Domestic animals: *Salmonella* can be carried by all livestock species, and have most commonly been identified in cattle, swine and poultry [8]. Many of these infections are asymptomatic, although disease can occur in any livestock species, particularly in young chickens and turkeys [1]. *Salmonella* can also infect and cause disease in domestic pet species [9].

Humans: Salmonellosis in humans, although often mild, can result in severe gastro-intestinal symptoms [10] and has been identified as a major cause of diarrhoea globally, with hundreds of thousands of fatalities recorded annually, most often in children under four years old [11]. There have been several outbreaks in Australia, some associated with marsupials [12-14]. Globally, wildlife (primarily birds and reptiles) have been identified as an important reservoir for *Salmonella* spillover into human populations [15, 16]. Those who have direct contact with marsupials or their faeces are considered to be at the highest risk for infection.

Natural hosts

Salmonella have been identified in species from all classes of vertebrates (fish, amphibians, reptiles, birds and mammals) and in invertebrates. Serovars may have a predilection for certain species. For example, *Salmonella* ser. Choleraesuis usually infects pigs, while *S. enterica* subsp. *arizonae* is usually found in cold-blooded vertebrates. However, most *Salmonella* serovars can infect a broad range of hosts [1].

World distribution

Salmonella have a world-wide distribution, and appear to be most prevalent in areas of intensive animal husbandry, particularly where pigs, calves and poultry are housed in confinement [3]. *Salmonella* have been detected in opossums (marsupials; family *Didelphidae*) throughout the Americas [17-19]. The isolation of multiple *Salmonella* serovars from pet sugar gliders (*Petaurus breviceps*) overseas also indicates a potential susceptibility of gliders in Australia to infection [20].

Occurrences in Australia

Within Australian marsupials, *Salmonella* has been isolated most commonly from hand-reared macropod joeys, pet kangaroos, kangaroos held in captivity and quokkas (*Setonix brachyurus*) [6, 13, 21, 22]. *Salmonella* has been detected at low prevalence in wild western grey kangaroo (*Macropus fuliginosus*) populations [23].

Salmonella has been detected in wild Australian marsupials including long-nosed bandicoots (*Perameles nasuta*), northern brown bandicoots (*Isoodon macrourus*), numbats (*Myrmecobius fasciatus*), long-nosed potoroos (*Potorous tridactylus*), northern quolls (*Dasyurus hallucatus*), eastern quolls (*D. viverrinus*), burrowing bettongs (*Bettongia lesueur*), brushtail possums (*Trichosurus vulpecula*), ringtail possums (*Pseudocheirus peregrinus*), scaly-tailed possums (*Wyulda squamicaudata*), yellow-footed antechinuses (*Antechinus flavipes*), common planigales (*Planigale maculata*), Tasmanian devils (*Sarcophilus harrisii*), dunnarts (genus: *Sminthopsis*), koalas (*Phascolarctos cinereus*), and common wombats (*Vombatus ursinus*) [7, 14, 24-30]. Marsupials in the

Kimberley region of WA were found to carry significantly more *Salmonella* serovars than their eutherian counterparts [28].

The National Enteric Pathogens Surveillance Scheme reports frequent isolations from quokkas, kangaroos and wallabies [31]. The majority of reports in these taxa have been of subspecies I serovars, with *S.* Muenchen and *S.* Typhimurium reported most frequently in kangaroos, *S.* Typhimurium reported most frequently in wallabies; and., *S.* Muenchen, *S.* Adelaide and *Salmonella* subspecies II serovar Wandsbek most frequently reported in quokkas [31].

Epidemiology

Salmonella spp. are gut-associated bacteria that can be shed in the faeces either continuously or intermittently with transmission occurring faecal-orally [1]. The bacteria are persistent in the environment and can survive in abiotic (non-living) reservoirs such as groundwater as well as in animal populations [32].

Although theoretically possible under conditions of environmental stress, there have been no confirmed reports of primary clinical disease in macropods in their natural habitat and the morbidity and mortality rates of salmonellosis in wild macropods are believed to be negligible [33-35]. Instances of salmonellosis are equally rare in other wild Australian marsupials. The isolation of *Salmonella* spp. from three sick free-ranging brushtail possums (*Trichosurus vulpecula*) in north Qld between 1978–98 is the only reported instance of disease in this group [36]. Animals in captivity may be exposed to *Salmonella* as a result of consumption of contaminated feed or water [37]. Orphaned and hand-raised macropod joeys are considered the most likely marsupial to develop salmonellosis, presumably due to lowered immune function. Disease has also been reported in captive possums and bandicoots [7]. In any species, faecal shedding of *Salmonella* is often intermittent and may not be related to the level of stress to which the animal is exposed. Shedding can occur within 24 hours of a host becoming infected [37].

The prevalence of infection may be subject to seasonal variation. Wild quokkas on Rottnest Island, WA have low infection rates (0-30%) over winter and high infection rates (70-100%) over summer [22]. This change occurs over only a few weeks and is attributed to an increase in diet-related stress over summer due to the reduced availability of high-quality feed, leading to a disruption in digestive physiology. A relationship between *Salmonella* prevalence and feeding may also be present in western grey kangaroos across WA, as infections are detected most frequently following periods of greatest rainfall and therefore increased abundance of grazing options [23]. The sudden change in available food may have altered faecal shedding or increased exposure to any bacteria present in groundwater during grazing [23].

Host characteristics such as sex and age do not appear to impact prevalence in wild marsupials, with lower body condition being the only trait that is potentially predictive of an individual within an exposed population being infected [33]. This is likely due to the connection with diet described above.

Zoonotic spillover may occur via direct contact with marsupials or their faeces in the urban environment, through unsafe consumption of kangaroo meat, or from animal handling and husbandry of wildlife (e.g. by rehabilitators, owners of native pets or zookeepers) [13].

Clinical signs

Most wild and many captive marsupials that carry *Salmonella* exhibit no clinical signs [5, 37]. Clinical signs, when present, include loose faeces (anywhere from soft and semi-formed through to haemorrhagic diarrhoea) in association with lethargy, anorexia and dehydration, and death. These changes are not pathognomonic and other disease processes should also be considered. Animals may also be found dead with no observed ante mortem clinical signs. Orphaned joeys often present with abdominal pain characterised by teeth grinding and abdominal guarding.

Diagnosis

The two most common methods used to diagnose *Salmonella* spp. infection in marsupials are bacterial culture and PCR testing [6]. Culture allows for identification to the serovar-level more frequently (at this stage PCR can only detect 10% of known *Salmonella* serovars) but is more time-consuming than PCR and may be less sensitive; *Salmonella* can enter a viable but non-culturable state, potentially leading to false-negative results [6, 38]. However, in cases where serovar identification is required, bacterial culture is still recommended and may be supported by serological testing [6, 18, 39].

Laboratory diagnostic specimens and procedures

Diagnosis of *Salmonella* spp. infection is based on isolation of the organism from aseptically collected tissue samples (primarily the mesenteric and ileocecal lymph nodes, or less commonly the liver, lung, kidney, heart, abdomen and spleen) postmortem or from faeces/rectal swabs antemortem. Samples should be collected during the acute phase of disease or as soon after death as possible. It is also preferable to collect samples prior to the commencement of any antibiotic treatment [3]. Samples should be stored at 4°C until processing [6].

Culture: Bacterial culture of *Salmonella* is specialised and typically requires selective enrichment of samples (tissue or faecal matter) [6, 40]. Isolates can be characterised using specific biochemical and serological techniques or by matrix-assisted laser desorption-ionization-time-of-flight mass spectrometry (MALDI-TOF MS) systems [41].

PCR: Tissue samples (fresh or embedded in paraffin) and faecal samples (including swabs) may be used for qPCR [24, 26].

Clinical and other pathology

In general, haematological and serum biochemical changes in marsupials are not well correlated to the severity of underlying disease and results may be unremarkable despite significant bacterial infection. Fibrinogen may be increased with salmonellosis. Neutrophilia and lymphocytosis may be present, but absence of these changes does not rule out bacterial infection [5]. Infected animals may

display lower red blood cell concentration (RBC), haemoglobin concentration (HGB), and packed cell volume (PCV) than their uninfected conspecifics [6].

There are few descriptions of pathology associated with salmonellosis in marsupials. Changes include focal hepatic necrosis with and without cholangitis, inflammation in the muscularis of the intestine, intussusception of the intestine, suppurative bronchopneumonia, haemorrhagic enteritis, and mucosal and submucosal necrosis [29, 42].

Differential diagnoses

In captive marsupials, differential diagnoses of salmonellosis include other causes of gastroenteritis and diarrhoea, including coccidiosis and campylobacteriosis [5]. Toxoplasmosis should be considered in the event of acute death. *Strongyloides* may cause diarrhoea and death [43, 44]. In hand-reared joeys, physiological stress, coccidiosis, yeast/candidiasis, roundworms, antibiotic administration and cryptosporidia are all differential diagnoses of diarrhoea [45]. Other causes of bacterial diarrhoea, including *E. coli, Klebsiella* spp.*, Clostridium* spp. and *Yersinia pseudotuberculosis* should also be considered [46, 47].

Treatment

Antibiotics are generally used to treat clinical salmonellosis in humans and animals although their efficacy in marsupials has not yet been formally investigated. Where possible, culture and sensitivity may inform the most effective antibiotic to use [5]. For clinically affected animals, analgesia is recommended if abdominal pain is apparent, as well as supportive care in the form of subcutaneous or intravenous fluids and removal of stressors [5]. There is no requirement to treat asymptomatic infection in marsupials, although the potential disease risk to the host, and to humans and other animals, should be considered and managed, through appropriate hygiene practices (see below).

Prevention and control

Minimising physiological stress is key to preventing and controlling salmonellosis in marsupials in captivity, especially joeys in care [5, 46]. Strict hygiene, including handwashing, hygiene of pouches, feeding equipment and the environment should be maintained whenever any marsupials, and particularly macropod joeys, are in care. Orphaned marsupials taken in for hand-raising should be considered at high risk of carrying and shedding *Salmonella*, particularly those in poor health as clinically affected animals will shed higher bacterial loads than asymptomatic carriers. A severely ill joey is likely to excrete more *Salmonella* than a healthy joey, with a likely greater zoonotic risk to the carers [21]. Appropriate PPE, including facemasks and disposable gloves, should be worn by those working with high-risk animals to minimise zoonotic risk. There should be no direct or indirect contact between marsupials in care or captivity and domestic or feral animals. *Salmonella* are susceptible to many disinfectants and can also be killed by moist heat (121°C for a minimum of 15 min) or dry heat (160-170°C for at least 1 hour) [1].

Hygiene should be maintained during processing of kangaroo carcasses. General food safety practices should be followed when handling and storing kangaroo meat to avoid contamination of

other foodstuffs. Thorough cooking of kangaroo meat prior to consumption, reduces the likelihood of transmission through the oral route and improved carcass handling procedures have significantly reduced the prevalence of *Salmonella* in harvested kangaroo meat [23, 34, 48].

Research

There is a need to better understand the source of *Salmonella* infection in free living Australian mammals and the potential links between environmental conditions and infection [33]. Further research on the prevalence and species/serovars of *Salmonella* present in wild marsupials (such as the work already done on western grey kangaroos and quokkas in WA) would provide a better picture of the natural levels of asymptomatic infection in these animals [6, 23]. More investigation into the link between human cases and domestic animals, wildlife and animal products would be valuable [36]. Research should also continue into the development of diagnostic methods, especially the ability of PCR to identify more *Salmonella* serovars.

Surveillance and management

Since 1978, the National Enteric Pathogens Surveillance Scheme has collected data on human and non-human isolations of enteric bacterial pathogens in Australia, including information on *Salmonella* serotypes identified in Australian animals and wildlife [31].

The Australian Government Department of Health OzFoodNet network (https://www.health.gov.au/our-work/ozfoodnet-network) undertakes national investigations into gastroenteritis outbreaks and clusters of disease in people, if they are potentially related to food products. Results are announced quarterly, and annual reports have been published in CDI since 2002 and their "Human and Non-human Annual Reports" are available on their website or on request.

Wildlife Health Australia administers Australia's general wildlife health surveillance system, in partnership with government and non-government agencies. Wildlife health data is collected into a national database, the electronic Wildlife Health Information System (eWHIS). Information is reported by a variety of sources including government agencies, zoo-based wildlife hospitals, sentinel veterinary clinics, universities, wildlife rehabilitators, and a range of other organisations and individuals. Targeted surveillance data is also collected by WHA. See the WHA website for more information https://wildlifehealthaustralia.com.au/Our-Work/Surveillance and https://wildlifehealthaustralia.com.au/Our-Work/Surveillance/eWHIS-Wildlife-Health-Information-System.

We are interested in hearing from anyone with information on this condition in Australia, including laboratory reports, historical datasets or survey results that could be added to the National Wildlife Health Information System. Negative data are also valuable. If you can help, please contact us at admin@wildlifehealthaustralia.com.au.

Cases of *Salmonella* infection and of disease associated with the presence of *Salmonella* in macropods are reported in eWHIS, primarily in hand-raised juveniles.

WHA provides reports on wildlife isolates to the NEPSS, which maintains a summary database of *Salmonella* isolates from wildlife in Australia. People seeking detailed information on *Salmonella* isolates from wild animals in Australia are referred to this source.

Acknowledgements

We are grateful to the people who contributed to this Fact Sheet and would like to thank Abbey Bestall, Peter Holz, Geoff Hogg, Mary Valcanis, Katie Fullerton, Cheryl Sangster and Amy Naicker.

Wildlife Health Australia recognises the Traditional Custodians of Country throughout Australia. We respectfully acknowledge Aboriginal and Torres Strait Islander peoples' continuing connection to land, sea, wildlife and community. We pay our respects to them and their cultures, and to their Elders past and present.

Please cite this Fact Sheet as: Wildlife Health Australia (2025) "*Salmonella* in Australian marsupials – Fact Sheet", published by Wildlife Health Australia, Canberra, available at https://wildlifehealthaustralia.com.au/Resource-Centre/Fact-Sheets.

Updated: March 2025

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