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Occurrence of tongue worm, *Linguatula cf. serrata* (Pentastomida: Linguatulidae) in wild canids and livestock in south-eastern Australia

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ABSTRACT

Pentastomids are obligate zoonotic arthropod parasites utilising canids and vulpids as their definitive hosts and several herbivorous species as their intermediate hosts. Reported only 10 times in Australia over the last 150 years as incidental findings, adult Pentastomids referred to as *Linguatula serrata* have been encountered in nasal cavities of domestic and wild dogs, and foxes. Nymphs have been reported in cattle and rabbits. In the present study, a number of potential definitive hosts, including red foxes (*Vulpes vulpes*), wild dogs (*Canis lupus dingo* and *C.l. dingo* x *C. familiaris*) and feral cats (*Felis catus*), and intermediate hosts cattle (*Bos taurus*), sheep (*Ovis aries*), feral pigs (*Sus scrofa*), rabbits (*Oryctolagus cuniculus*), goats (*Capra hircus*) and a European hare (*Lepus europaeus*), from the highlands of south-eastern Australia were examined. Of the animals examined 67.6% of wild dogs (n = 37), 14.5% of red foxes (n = 55) and 4.3% of cattle (n = 164) were found to be infected with Pentastomids, herein identified as *Linguatula cf. serrata*. The common occurrence of the parasite in wild dogs and less frequently in foxes suggests these wild canids have potential to act as a reservoir for infection of livestock, wildlife, domestic dogs and possibly humans. The unexpected high frequency of the parasite in wild dogs and foxes in south-eastern Australia suggests the parasite is more common than previously realised. Of the potential intermediate hosts in the region, only 4.3% of cattle were found to be infected with pentastomid nymphs which suggest the search for the host(s) acting as the main intermediate host in the region should continue. Future studies should investigate transmission patterns, health impacts on hosts and whether the parasite has zoonotic significance in Australia.

1. Introduction

Members of the genus *Linguatula,* also known as tongue worms due to their resemblance to the mammalian tongue, are obligate arthropod parasites which inhabit the upper respiratory tract of canids such as domestic dogs, foxes and wolves. After fertilisation, gravid females release millions of eggs during their mature lifetime. These eggs are expelled into the environment in nasal secretions and/or swallowed and passed in faeces (Riley, 1986). Most herbivores, including ruminants such as sheep, cattle and camels may serve as intermediate hosts for *Linguatula* species, becoming infected through accidental consumption of pasture contaminated with eggs resulting in visceral linguatulosis in the herbivore host (Tavassoli et al., 2007). Following ingestion of eggs by an intermediate host, the primary larvae emerge into the lumen of the intestine and penetrate the intestinal wall. Following a phase of migration, the larvae encyst in visceral tissues of the intermediate host such as the liver, lungs and mesenteric lymph nodes where they complete several moults before becoming infective nymphs (Riley, 1986; Paré, 2008). To complete the life cycle, infective nymphs must be consumed by a canid. This usually occurs as a result of predator/prey interaction or scavenging. Following ingestion the nymphs move from the digestive tract, up the oesophagus to the nasal cavity where they develop into mature adults (Riley, 1986; Paré, 2008). Zoonotic cases of infection with *L. serrata* have been reported from several countries (Self and Kuntz, 1967; Riley, 1986; Bowman, 1995; Lazo et al., 1999; Paré, 2008; Koehsler et al., 2011; Bhende et al., 2014; Oluwasina et al., 2014).

In Australia, knowledge of *L. serrata* is poor. The parasite has been reported only 10 times over the past 150 years, almost always as incidental findings. Adult pentastomids, identified as *L. serrata* having been reported in the nasal cavities of dingoes, domestic

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dogs and foxes (Johnson, 1910; Pullar, 1946; Durie and Riek, 1952; Riley et al., 1985) and nymphs have been encountered in cattle and rabbits (Ralph, 1865; Barnard and Park, 1893; Johnston and Cleland, 1910; Johnston, 1911; Pullar, 1936; Durie and Riek, 1952). None of these studies provide a detailed morphological description which makes specific identification of the parasite according to current criteria difficult. Wild dogs predate on livestock (mainly sheep) which in some areas is a major agricultural issue (Allen and Fleming, 2004), but their diet mainly consists of small macropodid marsupials, particularly swamp wallabies (*Wallabia bicolor*) (Newsome et al., 1983; Robertshaw and Harden, 1986). Wild dogs, and to a lesser extent foxes, play a key role in transmission of parasites of veterinary and human health importance in Australia, particularly *Echinococcus granulosus* (Stevenson and Hughes, 1988; Saunders et al., 1995; Jenkins and Morris, 2003; Jenkins et al., 2005). In parts of the world where *Linguatula* spp. occur commonly in canids, prevalence of the parasite in livestock is also high. This transmission cycle between domestic hosts (dogs and livestock) makes these species significant reservoirs for potential zoonotic infection. The lack of reports of *Linguatula* infection in Australian wild or domestic canids suggests the parasite occurs rarely. However, the nasal cavity of canids are rarely examined during post mortem examinations, especially those of wild dogs and foxes and the parasites although present, may simply have been overlooked. The aim of this study was to undertake a preliminary investigation into the occurrence and distribution of adult *Linguatula* spp in wild canids and nymphal stages in domestic livestock in south-eastern Australia.

2. Materials and methods

2.1. Wildlife definitive hosts

Wild dogs (*Canis lupus dingo* and *C.l. dingo* x *Canis familiaris*), foxes (*Vulpes vulpes*) and feral cats (*Felis catus*) were obtained from professional vertebrate pest control officers of the Australian Capital Territory (ACT) Parks and Conservation Service, New South Wales (NSW) Forests, NSW Local Lands Services and the Victorian Department of Environment, Land, Water and Planning. These animals were trapped and shot by these officers during the normal course of their duties. The heads of the animals were removed, packed in labelled plastic bags and kept frozen until examined.

2.2. Collection of mesenteric lymph nodes from cattle

Mesenteric lymph nodes from cattle (*Bos taurus*) were collected by meat inspectors in a local abattoir. Since there were no recent data on the occurrence and prevalence of *Linguatula* spp in cattle in south eastern Australia, collection of lymph nodes was restricted to cattle that were most likely to have been grazing in rough bush pasture, areas most likely also to be inhabited by wild dogs. Between 1 and 7 mesenteric lymph nodes were collected from each animal. All lymph nodes collected from a single animal were placed into labelled plastic bag and stored at 4 °C until examined.

2.3. Other potential intermediate hosts examined

Mesenteric lymph nodes from small numbers of a range of other potential intermediate hosts were also examined. These animals were collected opportunistically or donated from several sources. Mesenteric lymph nodes from sheep (*Ovis aries*) were collected in a local abattoir. The sheep were from a property near Dubbo and one near Holbrook, both locations in NSW. The rabbits (*Oryctolagus cuniculus*) were provided by the NSW forests vertebrate pest control officer, Tumbarumba. The hare (*Lepus europaeus*) was found as road kill on the Charles Sturt University Campus, Wagga Wagga. The two feral pigs (*Sus scrofa*) were found dumped on the side of the road between Wagga Wagga and Coolamon, NSW, but their origin was unknown. The two feral goats (*Capra hircus*) were shot on a property at Mangoplah, NSW and donated by the property owner.

2.4. Parasite collection

The skulls of dogs, cats and foxes were split into two halves using a hatchet and a hammer. This unsophisticated procedure enabled us to cleave the skull whilst not damaging tongue worms that may be been present. It also enabled us to obtain a clear view into the right and left sides of the nasal cavity and to see any tongue worms that were present (Fig. 1). Each side of the nasal cavity was extensively searched macroscopically for adult tongue worms by carefully removing any tissue or structures such as the conchae with forceps (Fig. 1). After removal of the tissue and any clearly visible tongue worms the nasal cavities were irrigated with running water into a 300μ sieve and all additional tongue worms dislodged (usually the small males) were backwashed from the sieve into a dish and collected (Fig. 2). All tongue worms collected were rinsed in distilled water before being preserved in ethanol (70%) or 10% formalin solution. Mesenteric lymph nodes from cattle, sheep, pigs, hare, rabbit and goat were cut longitudinally using a scalpel. Nymphs contained in their capsules could be easily observed macroscopically as distinct white round masses about 2–3 mm in diameter (Fig. 1). Nymphs were released from the capsule tissue surrounding them and viewed microscopically and then preserved in 70% ethanol or 10% formalin. Parasite data, including number of parasites, developmental stages, geographic location, host, host age and location in the host were recorded in an Excel spreadsheet. Fisher's exact test was employed to test the correlation between prevalence of infection between dogs and foxes.

2.5. Faecal egg count

Faeces from a Tumbarumba wild dog infected with one male and two female tongue worms were examined. Four flotations were prepared from the faecal sample but only 1 to 3 eggs were recovered from each flotation. No faeces from foxes were examined. One gram of faeces was placed in the base container of a Faecalizer® (EVSCO Pharmaceuticals, NJ, USA) with approximately 2 ml of saturated sodium nitrate flotation solution (SG 1.25) and mixed well. The green sieve insert was placed firmly into the Faecalizer® before it was filled with saturated sodium nitrate flotation solution until a meniscus was achieved. A glass cover slip was floated on the meniscus and allowed to stand for 10 min. After 10 min the cover slip was carefully lifted off and placed on a slide. The slide was scanned microscopically for parasite eggs under ×4 and ×10 magnifications.

3. Results

3.1. Parasite identification

Adult specimens in the present study were placed in the family Linguatulidae based on their general morphology, including a fluke-like flattened body, and the location in which they were found (nasal cavity). In the present study parasite specimens are referred to as *Linguatula cf. serrata* until detailed morphological and molecular studies are done.

3.2. Prevalence in potential definitive hosts

A total of 37 wild dogs, 55 foxes and 5 feral cats were examined. A Fisher's exact test showed that there was a significant difference

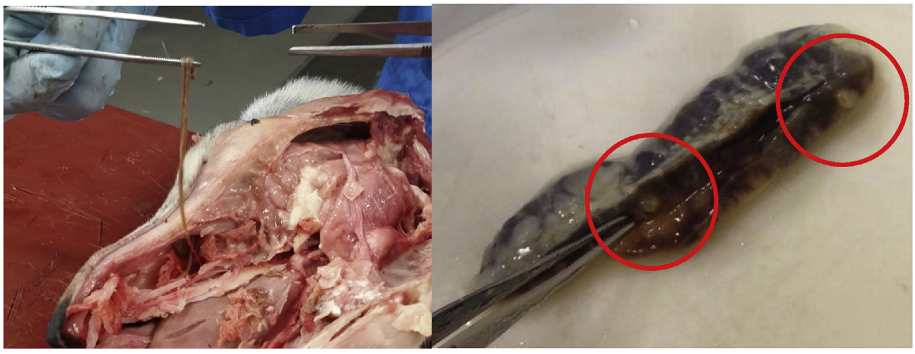

Fig. 1. Left: A female tongue worm being removed from the nasal cavity of a wild dog; Right: Two encapsulated tongue worm nymphs in a bovine mesenteric lymph node.

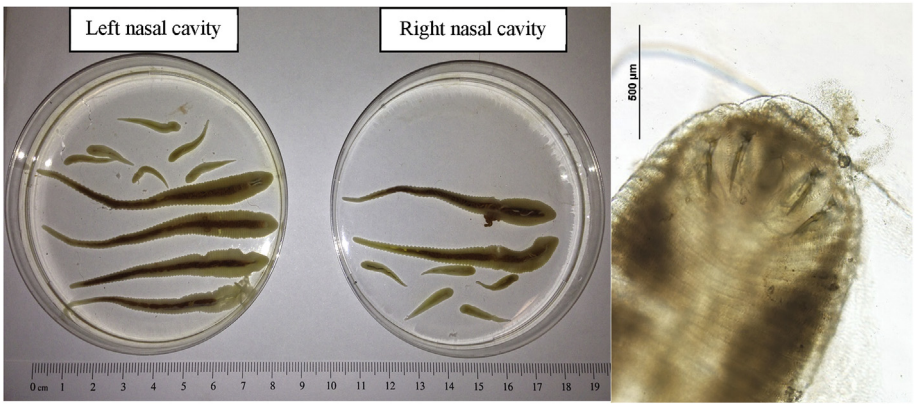

Fig. 2. In the petri dishes, adult males and females removed from the left and right nasal cavities of a wild dog; Right image shows the anterior end of a nymph removed from lymph node of a cow.

between prevalence of the parasite in dogs and foxes. Sixty-seven percent of wild dogs were found to be infected with tongue worms (Table 1). They were from five, two and one locations in NSW, ACT and Victoria, respectively. Of the 25 wild dogs infected with tongue worms, a total of 95 tongue worms were recovered (42 females and 53 males).

In foxes the infection rate was significantly lower, 14.5% (Table 1). Of the 34 foxes collected in NSW, six (25.0%) of the infected animals were collected from three locations in NSW (Fig. 3). Two of the 19 foxes collected in the ACT were also infected. To date no infected foxes have been collected in Victoria. Eighteen *Linguatula cf. serrata* consisting of nine females and nine males were removed from eight infected foxes (Table 1). One fox contained a single parasite, four harboured two and all other individuals had three tongue worms present. None of the five cats was found to be infected with tongue worms.

Approximate age was determined by the size of the skull and wear on teeth, broken into four broad categories; young, young adult, adult and old. Older animals were commonly found to be infected with greater numbers of tongue worms than the younger animals (Table 2). Of the wild dogs classified as old, the number of tongue worms in each individual varied from four to 17 compared to the younger animals that had one to six tongue worms. It should be noted that most of the heads examined in the present study (both foxes and wild dogs) were received already detached from the body with no information as to the sex and body condition of the animals. All infected foxes were young adult or adult with the one exception of a single fox that was determined to be old. This individual had a large skull and extremely worn and decaying teeth. Interestingly, the female tongue worms infecting this animal were considerably larger (7.5 cm - 8.5 cm) than the female tongue worms infecting all the younger foxes (3–5 cm). In fact, the female tongue worms recovered from this old fox were comparable in length to the largest (9 cm) female tongue worms removed from infected wild dogs.

3.3. Prevalence in potential intermediate hosts

Four hundred and ninety four mesenteric lymph nodes from 164 cattle were examined. Parasites were found in lymph nodes from seven animals (4.3%). A total of 19 nymphs were recovered (Table 1). Of the 164 cattle examined, 112 originated from seven locations in Victoria and the remainder were from farms in NSW (Table 1). The majority of nymphs recovered were encapsulated (Fig. 1). The mesenteric lymph nodes from 34 sheep from NSW were examined: 11 from Holbrook and 23 from Dubbo. These sheep comprised both lambs and older mutton sheep, however no infections were detected. Nymphs were not recovered in any of the other potential intermediate hosts examined, including feral pigs ($n = 2$), rabbits ($n = 8$), hare ($n = 1$) and goats ($n = 2$) examined.

3.4. Faecal egg count

The number of eggs recovered ranged from 0 to 3 per gram of faeces. The eggs were 130-133 x 80-83-3 ($n = 10$) micron in size and a developing nymph was clearly visible inside the egg. Other noticeable features included a thick egg shell wall and presence of the nymph's two pairs of hooks (Fig. 4). Each egg was also enclosed in a translucent membrane-like envelope.

Table 1
Locations & occurrence of tongue worms in various hosts collected in south-eastern Australia.

Host	Locality (State)	Number of hosts examined	Number of hosts infected (%)	No of parasites found (females + males)
Wild dog	Booroomba (ACT)	1	1 (100) [a]	1 + 4
	Bullen Range (ACT)	1	0 (0) [a]	0
	Limestone (NSW)	7	5 (71)	6 + 8
	Brindabella (NSW)	7	5 (71)	14 + 19
	Mullion (NSW)	3	2 (67)	7 + 8
	Bago/Maragle forest (Tumbarumba,NSW)	14	11 (79)	14 + 12
	Wee Jasper (NSW)	1	1 (100) [a]	0 + 2
	Orbost (Vic)	3	0 (0)	0
	Totals	**37**	**25 (67.6)**	**42 + 53**
Fox	Booroomba (ACT)	12	0 (0)	0
	Bullen Range (ACT)	7	2 (29)	0 + 4
	Limestone (NSW)	4	0 (0)	0
	Brindabella (NSW)	6	1 (17)	0 + 1
	Perisher Valley (NSW)	2	0 (0)	0
	Mullion (NSW)	8	3 (38)	6 + 3
	Bago/Maragle forest (Tumbarumba, NSW)	7	2 (29)	3 + 1
	Wee Jasper (NSW)	6	0 (0)	0
	Creighton's Creek (NSW)	1	0 (0) [a]	0
	Black Mountain (Vic)	1	0 (0) [a]	0
	Orbost (Vic)	1	0 (0) [a]	0
	Totals	**55**	**8 (14.5)**	**9 + 9**
Feral cat	Tumbarumba (NSW)	1	0 (0) [a]	0
	Perisher Valley (NSW)	3	0 (0)	0
	Booroomba (ACT)	1	0 (0) [a]	0
	Totals	**5**	**0 (0)**	**0**
Cattle	Corryong (Vic)	75	4 (5.3%)	12
	Mitta Mitta (Vic)	8	1 (12.5%)	1
	Tallangatta (Vic)	6	1 (16.7%)	5
	Towong (Vic)	20	0 (0)	0
	Wabonga (Vic)	1	0 (0) [a]	0
	Wangaratta (Vic)	1	0 (0) [a]	0
	Wodonga (Vic)	1	0 (0) [a]	0
	Braidwood (NSW)	6	0 (0)	0
	Carcoar (NSW)	6	0 (0)	0
	Holbrook (NSW)	5	0 (0)	0
	Tumbarumba (NSW)	5	1 (20.0%)	1
	Wagga Wagga (NSW)	30	0 (0)	0
	Totals	**164**	**7 (4.3)**	**19**
Sheep	Dubbo (NSW)	23	0 (0)	0
	Holbrook (NSW)	11	0 (0)	0
	Totals	**34**	**0 (0)**	**0**
Feral pig	Tumbarumba (NSW)	2	0 (0)	0
Rabbit	Wagga Wagga (NSW)	8	0 (0)	0
Goat	Holbrook (NSW)	2	0 (0)	0
Hare	Mangoplah (NSW)	1	0 (0) [a]	0

[a] From sample size of one, therefore, unlikely to represent the true percentage of infection in the population.

4. Discussion

This is the first major study to determine the occurrence and distribution of *Linguatula cf. serrata* over a wide geographical area in Australia. As shown in our results, tongue worms were present in 25 of 37 (67%) wild dogs and eight of 55 (14.5%) foxes collected in NSW and the ACT indicating the prevalence of the parasite is much higher in this region than previously thought. This is also only the second time tongue worm has been reported in European red foxes in Australia (the first time being Pullar (1946)) and the first time the parasite has been reported from the ACT.

Occurrence of *Linguatula cf. serrata* appears widespread in wild dogs in the highlands of south eastern Australia with at least one individual being infected from each of the five collection sites. Infection with *Linguatula cf. serrata* in foxes appears to be less widespread than in wild dogs which is worthy of further investigation but may be a reflection of different dietary preferences of foxes compared to wild dogs. The first report of *Linguatula serrata* in a canid in Australia was from an experimental infection of a dog using nymphs collected from cattle in NSW (Johnston and Cleland, 1910). There has also been one report (Pullar, 1936) of adult parasites in a domestic dog in Victoria following the spontaneous expulsion of a mature female *L. serrata* from the nose of this dog in 1935 on a property in Derrinallum. The parasite has been reported in dingoes from South Australia (Johnson, 1910) and Queensland (Durie and Riek, 1952) and these early data may be a useful basis for future studies in both jurisdictions. Pullar (1946) noted that there was no correlation between age, body condition or sex with tongue worm infection in foxes. In the present study, in wild dogs, animals that appeared to be oldest were the most heavily infected, suggesting infection with tongue worm may accumulate over time. Similarly, a study of 143 stray dogs in Iran found the relationship between infection with *L. cf. serrata* and age of the dogs was statistically significant. The infection rates in dogs two to three years old or younger, four years old or five years old were 44%, 76.7% and 70.8%, respectively (Meshgi and Asgarian, 2003). Soulsby (1982) noted that tongue worms live for approximately 15 months in canids after which time clinical signs such as sneezing, coughing, mucus discharge and difficulty in breathing are said to resolve.

Apart from the present study, in which tongue worms were quantified, only Pullar (1946) reported counts (of 1-3 per fox), and to date there have been only five reports of adult *L. serrata* in

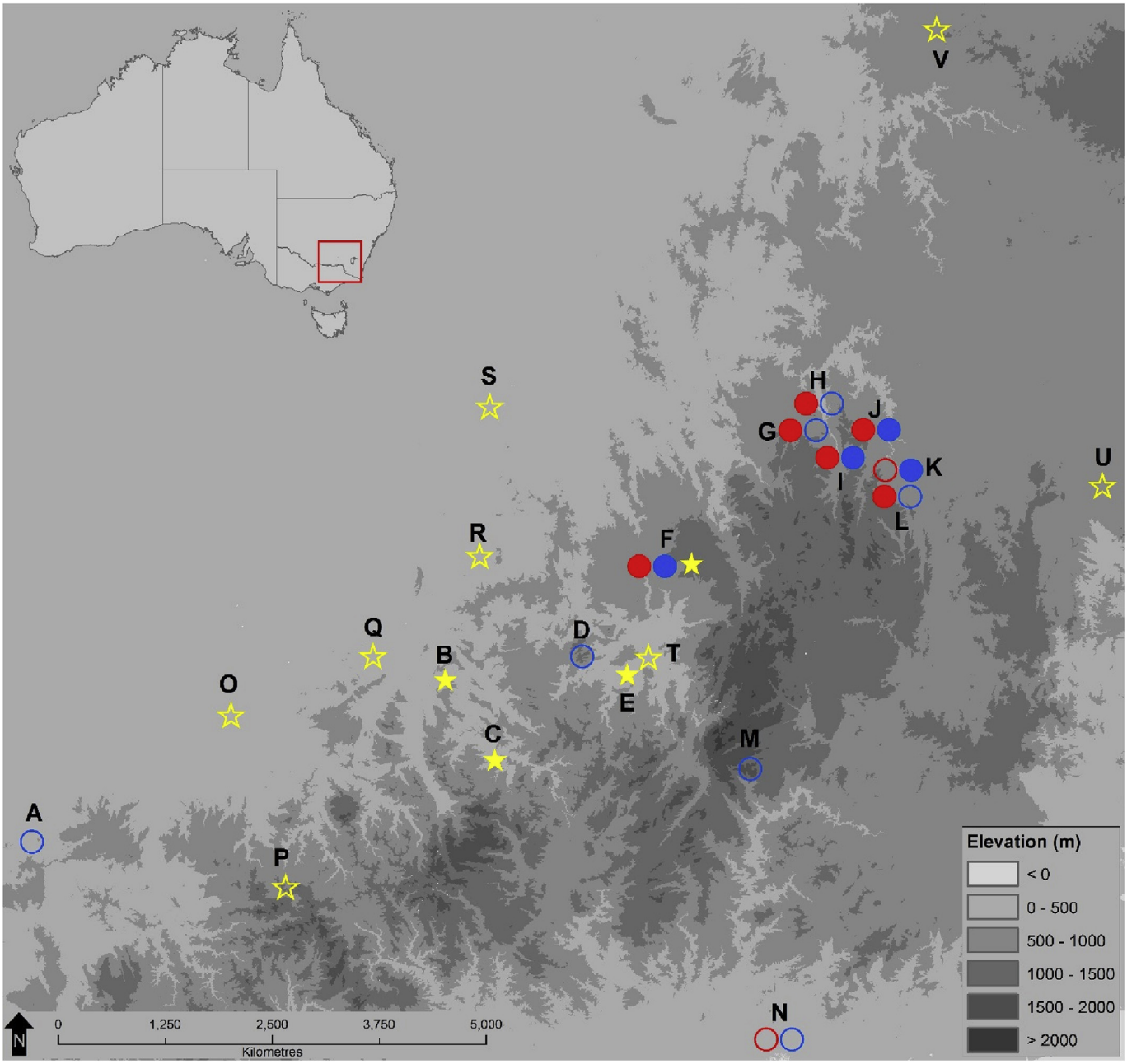

Fig. 3. Localities where animals infected with *L. cf. serrata* were found. Open and solid symbols represent uninfected and infected animals, respectively. The stars, the red circles and the blue circles are cattle, wild dogs and foxes, respectively. Those animals that have been collected opportunistically have been excluded from this map. A) Creighton's Creek, B) Tallangatta, C) Mitta Mitta, D) Black Mountain, E) Corryong, F) Tumbarumba, G) Limestone, H) Wee Jasper, I) Brindabella, J) Mullion, K) Bullen Range, L) Booroomba, M) Perisher Valley, N) Orbost, O) Wabonga, P) Wangaratta, Q) Wodonga, R) Holbrook, S) Wagga Wagga, T) Towong, U) Braidwood, V) Carcoar. (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.)

Australia (Johnson, 1910; Johnston, 1911; Pullar, 1936; Durie and Riek, 1952; Riley et al., 1985) merely referring to the presence of the parasite in wild dogs and foxes. Pullar (1936) suggested the reason for the rarity of reports of tongue worms in wild canids in Australia may be simply due to the infrequent examination of the nasal cavity during post mortem examinations. Therefore, there is no benchmark to determine if tongue worms are more common in wild dog populations of south-eastern Australia today as compared to 50–100 years ago.

One potential factor influencing the infection variation between the two canine species observed in the present study, i.e., 67.6% in wild dogs and 14.5% in foxes, is likely to be diet. Numerous herbivorous mammals have been reported to act as the intermediate host for tongue worm in other parts of the world. These species include but are not limited to ruminants, such as cattle, sheep, goats and deer, lagomorphs, pigs and horses (Acha and Szyfres, 2003). In Australia, cattle and rabbits have been reported as intermediate hosts (Pullar, 1936; Durie and Riek, 1952) and other potential candidates include sheep, goats, deer, hares, pigs and macropods. Pullar (1936) suggested a sylvatic cycle of transmission of tongue worm was occurring in Victoria between foxes and rabbits and Durie and Riek (1952) acknowledged a similar cycle may be taking place in Queensland between dingoes and wallabies. Turni and Smales (2001) recorded the occurrence of a single

Table 2
Number of parasites found in different age groups of dogs and foxes in the present study. Animals were aged approximately by skull size and teeth wear.

Age	No. of animals (no of infected animals)	No of parasites (females, males)	Mean[a] (range) per infected individual
Dogs			
Young	2 (0)	0 (0, 0)	0 (0–0)
Young Adult	2 (0)	0 (0, 0)	0 (0–0)
Adult	33 (24)	78 (36, 42)	3.2 (1–10)
Old	2 (1)	17 (6, 11)	17 (17–17)
Foxes			
Young	4 (0)	0 (0, 0)	0 (0–0)
Young Adult	10 (5)	11 (7, 4)	2.2 (1–3)
Adult	40 (2)	4 (0, 4)	2 (2–2)
Old	1(1)	3 (2, 1)	3 (2–2)

[a] Mean = total number of parasites/total number of infected animals.

Fig. 4. Tongue worm eggs found in the faeces of a wild dog in the present study. Arrows indicate a pair of hooks.

pentastome nymph suggested to be of the Linguatulidae family in a bridled nailtail wallaby (*Onychogalea fraenata*) in Queensland, however, they did not further identify or describe any morphological details of this parasite.

While the diet of wild dogs and foxes overlap to a degree, differences in size and hunting behaviour leads to considerable variation in the prey each species consumes (Corbett, 2001; Fleming et al., 2001; Davis et al., 2015). For example, wild dogs are larger and can adjust their hunting tactics such as working together in groups, enabling them to bring down large kangaroos and calves (Corbett, 2001). Conversely, foxes are solitary hunters that more commonly feed on rabbits, rodents, ground-nesting birds, insects and fruit. However, if the opportunity presents itself foxes will also feed on livestock species including lambs and goat kids. Foxes may also gain access to adult livestock and macropod species through scavenging carcasses of animals that have died of natural causes or scavenging on carcasses of native wildlife and domestic livestock killed by wild dogs (Corbett, 2001; Mitchell and Banks, 2005).

In a study by Coman (1973), the stomach contents of 1229 foxes collected in Victoria found rabbits (*Oryctolagus cuniculus*) to be the most common dietary item, occurring in 38.8% of individuals. The second most frequent dietary item were sheep (31.3%), mainly carrion, of which 45% were thought to be lambs up to first shearing. Among larger native wildlife species potentially consumed as carrion were wombats (*Vombatus ursinus*), grey kangaroos (*Macropus giganteus*), and swamp wallabies (*Wallabia bicolor*). A comparative study by Saunders et al. (2004) examined the stomach contents of 240 foxes before the spread of rabbit haemorrhagic disease and 269 foxes following its arrival in south-eastern Australia. Prior to the spread of the disease the study recorded sheep, rabbit and macropod material in 40%, 20.8% and 2.5% of fox intestines, respectively. The post rabbit haemorrhagic disease sample showed little variation in fox intestinal contents with sheep, rabbit and macropod material occurring at 34.9%, 19.3% and 2.6%, respectively.

Corbett (2001) stated that while wild dogs will predate on approximately 200 species in Australia, around 80% of their diet is made up of only 10 species. Of these frequently consumed species, examination of 2063 faecal and stomach samples of wild dogs from the coastal mountains in south-east Australia revealed swamp wallabies and wallabies of unidentified species comprised 33.7% of the diet. In Corbett's study, red-necked wallabies (*Macropus rufogriseus*) were a smaller portion of the wild dog diet, occurring in 5.3% of the animals sampled and rabbit was present in 10.5% of the animals examined. The examination of 1993 faecal samples of wild dogs from the humid coastal mountains of east Australia revealed a similar dietary composition. According to Corbett (2001) the most common diet components included swamp wallaby (30.5%), red-necked wallaby (11.1%) and rabbit (6.4%). For both regions the occurrence of sheep and cattle in the diet was small, consisting of $\leq$1.4%.

A recent study in south-eastern Australia by Davis et al. (2015) examined 5875 wild dog scats and found that rabbits and swamp wallabies each composed > 10% of the diet. Rabbits were also found to contribute >10% of the diet from faecal samples of 11,569 foxes. More generally, Davis et al. (2015) reported that medium and large mammals occur more frequently in the diet of wild dogs than do small mammals. In contrast, large mammal remains occur less frequently in the diet of foxes than in wild dogs. In view of the

infrequency of wild dogs killing and eating livestock, and their preference for eating swamp wallabies and rabbits, the high frequency of infection with tongue worm in wild dogs suggests macropods and/or rabbits maybe the main intermediate host(s) in transmission of tongue worm in Australian wildlife. Further studies exploring all potential wildlife intermediate hosts for tongue worm in Australia need to be undertaken to confirm this hypothesis.

In Australia, the diet of foxes and feral cats (*Felis catus*) has been shown to largely overlap (Catling, 1988). There have been conflicting reports of cats acting as a definitive host for tongue worm. Paré (2008) stated that cats are not "adequate" hosts but provides no data to support this claim. However, Esmaeilzadeh et al. (2008) reported the presence of a tongue worm nymph encysted in the lung following the necropsy of a stray cat in Iran. None of the five feral cats in the present study was infected with pentastomids. However, in order to confirm or exclude Australian feral cats as a definitive host of the parasite, further studies using larger sample sizes, collected from several different geographical areas where infection in wild dogs and foxes has been confirmed, need to be undertaken.

Tongue worm eggs are described as oval with a length of 70–90 μm and a thick chitinous shell (Soulsby, 1982). Each egg is surrounded by a bladder-like envelope and contains a larva that bears two pairs of hooks (Bowman, 1995; Baker, 2007). Eggs recovered from a faecal sample in this study (Fig. 4) were larger in size (130–133 μm). In faecal examination of canids a differential diagnosis for tongue worm eggs is highly recommended. Morphological features of nymphs can be seen through the wall of tongue worm eggs such as the presence of small hooks, a useful diagnostic feature for the identification of tongue worm eggs in faecal floats.

In conclusion the high frequency of infection in wild dogs and foxes in the highlands of south-eastern Australia suggests tongue worms occur commonly in this region of Australia. Future studies should continue to investigate the geographical distribution of tongue worm in definitive hosts (both wildlife and domestic) in other regions and also investigate the range of wildlife and domestic animal species acting as intermediate hosts.

Conflict of interest

The authors declare that there is no conflict of interest.

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