PARASITES OF THE NORTHERN HAIRY-NOSED WOMBAT *LASIORHINUS KREFFTII:* IMPLICATIONS FOR CONSERVATION

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Gerhardt K, Smales LR and McKillup SC, 2000. Parasites of the northern hairy-nosed wombat *Lasiorhinus krefftii:* Implications for conservation. *Australian Mammalogy* **22**: 17-22.

The northern hairy-nosed wombat *Lasiorhinus krefftii* is now restricted to a single population of less than 70 individuals at Epping Forest National Park, North Queensland, Australia, and is listed as critically endangered. We examined six trapped animals for ectoparasites, and 197 faecal samples for endoparasites. All ectoparasites (the tick *Ambylomma triguttatum*, the flea *Echidnophaga cornuta* and the louse *Boopia dubia*) were new host records. Nematode eggs and larvae were found in every faecal sample and the number of eggs varied significantly among months sampled. Cestode proglottids were also found. There was no indication that parasites were causing disease and few species were detected. This last remaining population of *L. krefftii* may be relatively immunologically naïve, and we suggest that removing them from their natural environment to other areas as part of a captive breeding program should be attempted with caution.

Key words: *Lasiorhinus krefftii*, parasites, pathological effects, conservation, wombats.

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THE northern hairy-nosed wombat, *Lasiorhinus krefftii* (Owen 1872) (Marsupialia: Vombatidae) is the largest of the three extant wombat species found in Australia. The forest-dwelling common wombat, *Vombatus ursinus* (Shaw 1800) is relatively abundant across south eastern Australia (McIlroy 1995) and the southern hairy-nosed wombat *L. latifrons* (Owen 1845) is common, but has a limited distribution, in the semi-arid plains of South Australia and across the Nullabor into Western Australia (Wells 1989). Although *L. krefftii* was recorded from two regions after European arrival (the Moonie River in Southern Queensland and Deniliquin in New South Wales) it is now restricted to a single population at Epping Forest National Park in Central Queensland (Crossman *et al*. 1994). This reduction in geographic range and numbers is thought to have occurred over the past 120 years (Taylor *et al*. 1994). Estimates indicate that the population may have declined to as few as 35 individuals in 1982 (Crossman *et al.* 1994) but is now thought to have increased to some 65 individuals (Horsup and Davidson 1994). *L. krefftii* is currently listed as critically endangered (Maxwell *et al*. 1996). Distinguishing features of the population of *L. krefftii* are its isolation, small size and contracted range. It is not an exaggeration to say that *L. krefftii* is close to extinction. Measures employed in an attempt to conserve this species include the exclusion of cattle from the park, restriction of park access to managers and researchers only, and maintenance of all fire breaks in the park (Horsup and Davidson 1994). Future management plans include the establishment of a captive breeding program (Horsup and Davidson 1994).

The population receives no immigrant wombats and is therefore unlikely to be exposed to new wombat parasites, but animals selected for captive breeding or relocation in the future could be at risk of infection. Previous records of parasites occurring in *L. krefftii* are from post-mortem examination of two individuals and reports from workers involved in studies of behaviour and ecology since 1985. One nematode *Oesophagostomoides eppingensis* Smales, 1994 and one cestode recorded as *Paramoniezia suis* Maplestone and Southwell 1923 are the only known helminths (Spratt *et al*. 1991; Smales 1994). Of the ectoparasites "mange" as a minor skin condition, possibly caused by *Sarcoptes scabiei* Linn., 1758 (see Crossman 1988) is the only record.

The vulnerability of *L. krefftii* to extinction makes unsuitable direct methods to investigate its parasite burden and the potential for parasites to cause disease. Therefore, two other strategies were used to obtain data for this study. Animals trapped as part of the ongoing monitoring and research program

of the Queensland Department of Environment (DOE) and the recovery program funded by the Australian Nature Conservation Agency were examined for ectoparasites. Faecal samples from the wombat population were collected in Epping Forest National Park for examination since parasite propagules are useful indicators of parasite groups present (Watve and Sukumar 1995). This sampling can also reveal trends in infectivity and prevalence of parasite infection (McCallum 1994). Our findings are discussed in terms of their significance for the development of effective management strategies for the host as well as providing insight into the biogeography of both host and parasites.

MATERIALS AND METHODS

The extant *L. krefftii* population is in Epping Forest National Park (EFNP hereafter) 120 km north-west of Clermont, Central Queensland. The wombats occupy a series of large burrows dug along the banks of a sandy gully close to a supply of perennial native grasses. Burrows, with single or multiple entrances, are arranged in loose clusters each of which may be used by up to 10 wombats (Johnson and Crossman 1991) (Fig. 1). While it is possible to associate an individual wombat with a burrow cluster, frequent exchange of burrows by the animals makes it impossible to assign a known individual to a particular burrow (Johnson and Crossman 1991). Wombats emerge from their burrows after dark and spend between 2 and 6 hours per night above ground (Johnson 1991) during which they feed on pastures of low nutritive value.

Burrow entrances are marked with piles of fresh faecal pellets, and smaller piles are deposited along paths between burrows, but not elsewhere (Johnson and Crossman 1991). Fresh, still moist, pellets collected at dawn can therefore provide a sample from the population over a 24 hour period. Each pile of dung collected may represent the total output of a single individual, but where several piles of fresh dung are found in the same area they may be from one or several individuals.

During a monitoring program being implemented as part of the Northern Hairy-Nosed Wombat Recovery Plan (Horsup and Davidson, 1994), six different individuals were trapped on emerging from their burrows after dark. Two were trapped in January, 1 in March, 1 in June, and 2 in September 1996. Each was anaesthetised in the trap with Zoletil 20 following the recovery plan protocols and was examined for ectoparasites, (lice, ticks, fleas and mites) at the trap site prior to release. This phase of the study was carried out as a permitted activity within the recovery plan. Any ectoparasites present were removed from the host as quickly as possible

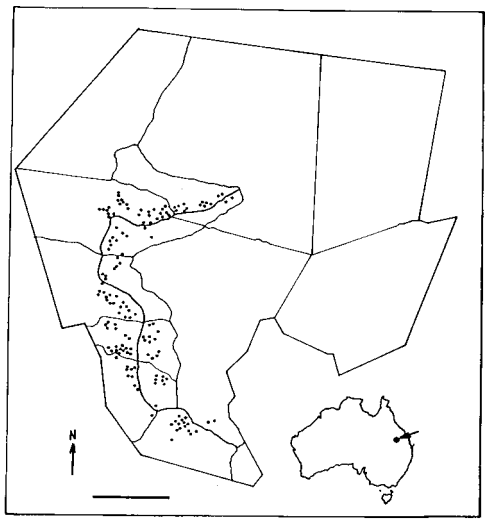

Fig. 1. Epping Forest National Park showing relict watercourse (meandering line), along which all current burrows (●) occur, together with roads and tracks. Scale bar indicates 1 km.

with tweezers or fingernails and stored in 70% ethanol; any lesions on the animals were also noted.

A total of 197 faecal samples, 24 - 32 per sampling event representing about 40% of the wombat population, were collected at monthly intervals from February to August 1996 inclusive, under permit from the DOE. The entrances to all wombat burrows were checked at dawn and each pile of fresh, still moist, faecal pellets was collected as a separate sample, placed in a specimen jar on ice, transported back to the laboratory and stored at 4°C prior to examination within two days.

To determine the number of eggs per gram of faeces (epg), counts were carried out using the standard McMaster technique for nematode and cestode eggs. Replicate samples were counted until the standard error did not exceed 25% of the mean (Arundel and Rickard 1980). A zinc sulphate solution (specific gravity of 1.5) was used to float trematode eggs (Kaufmann 1996) and a concentration technique, including centrifugation and flotation in salt solution (specific gravity 1.2) (O'Callaghan, pers. comm.), was used to separate and count protozoan oocysts. The coarse residue from sieved faeces was examined for cestode fragments and any found were stored in 70% ethanol. A Baermann technique (Kaufmann 1996) was used to extract strongyloid and other nematode larvae from aliquots of the faecal samples. All measurements were made using an ocular micrometer.

Rainfall data (the only accurate weather data available) were taken from Walthum Station just south of EFNP.

One-way analysis of variance was used to determine any significant differences among mean numbers of epg per month after preliminary testing for normality (Anderson Darling test) and equality of variances (Bartlett's test) and *a posteriori* Tukey tests were used to compare particular means when significant differences were found.

RESULTS

Rainfall data

In 1996, rainfall was similar to the average for the previous five years (Table 1). Although detailed data for temperatures in EFNP during the study period were not available, anecdotal evidence suggests they conformed to the usual pattern of winter temperatures (maximum 33°C and minimum of 9°C).

	1992	1993	1994	1995	1996
January	40	15	30	175	75
February	35	38	15	102	20
March	0	0	155	30	0
April	30	0	0	30	45
May	28	20	0	32	0
June	0	0	0	0	18
July	0	0	8	15	0
August	20	35	20	65	35
September	10	8	0	0	0

Table 1. Rainfall (mm) at Walthum Station, south of Epping Forest National Park, Central Queensland from January to September in 1992-1996 inclusive.

Ectoparasites

Five adult and nine juvenile ticks *Ambylomma triguttatum* Koch, 1844 (Acari: Ixodidae) were found. Of these, all adults and seven juveniles were found on one wombat in January and the two remaining juveniles on another wombat in September. Ticks were found only on the undersides of the wombats (ventral abdominal area and legs).

Fleas *Echidnophaga cornuta* Wagner, 1936 (Siphonaptera: Pulicidae) in infestation levels ranging from sparse (< 50) to medium (50 – 100), were found on the stomach area of the 3 wombats trapped in January and June. Rough skin and small scars resembling flea feeding sites were common on all 6 wombats trapped. Six specimens of the louse *Boopia dubia* Werneck and Thompson, 1940 (Mallophaga: Boopiidae) were found on a wombat in June. No mites were detected and no obvious lesions of the type that could have been caused by mites were noted, so no skin scrapings were taken. *L. krefftii* is a new host record for each of the above ectoparasites.

Faecal examination

No cestode or trematode eggs and no protozoan oocysts were found. Nematode eggs were found in every sample. The eggs all had a similar appearance and were typical strongylid eggs, elipsoidal in shape with transparent shells 88.5-92 µm long by 42.5-51 µm wide. Mean monthly epg are given in Fig 2. Data for all months except February showed a normal distribution and since ANOVA is robust in terms of minor departures from normality (Zar 1984) a one-way analysis was done. There was a highly significant difference among months ($F_{6,188} = 7.87$, $p < 0.001$) and a Tukey test showed that there were three groups: (a) March to June inclusive and August; (b) February and (c) July. No significant association was found between rainfall and egg count (Pearson correlation $r^2 = 0.25$, $p > 0.05$).

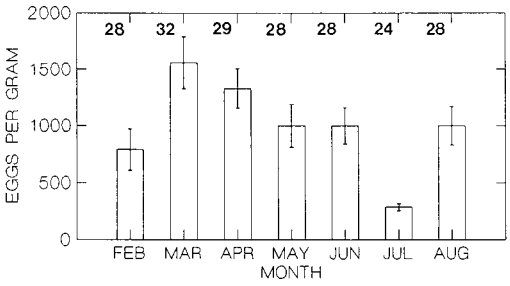

Fig. 2. The number of eggs per gram of faecal samples and numbers of samples taken each month from Feb – Aug 1996. Vertical bars show the standard error of each mean.

On three separate occasions (May, July and August) a cestode fragment was found associated with a pile of faecal pellets. There were 9 - 25 proglottids per fragment, each measuring 10 - 5 mm long by 55 - 56 mm wide. Eggs taken from the proglottids were all 49.5 µm in diameter.

Nematode larvae, *Strongyloides* sp. were found in all samples and ranged from 24% prevalence in April to 68% prevalence in May, with an average prevalence of 51% during the study period. Larvae were 470 µm long by 20 µm wide. This is a new host record.

DISCUSSION

Of the ectoparasites found, the tick *A. triguttatum* has not previously been recorded on any wombat species, but has been reported throughout Queensland and New South Wales. Adults are most prevalent in the warmer months and although its normal hosts are the larger macropodid marsupials (kangaroos) it commonly occurs on cattle, horses, dogs (including dingos) pigs and humans (Roberts 1970). Most ticks

were found in January. This may have been because rain which had fallen a few weeks before the January sampling had encouraged the growth of long buffel grass around the burrows. The long grass could have sheltered ticks which then easily transferred to the ventral surfaces of wombats as they brushed through.

The flea *E. cornuta* has not previously been recorded from *L. krefftii* but has been found on *L. latifrons* (Doube 1981). In South Australia, five species of *Echidnophaga (E. calabyi, E. cornuta, E. eyeri, E. octotricha* and *E. perilis)* have been collected from wombats. *E. perilis* and *E. eyeri* have also been recorded from other hosts (e.g. *E. eyeri* has been found on *V. ursinus* and the rat *Rattus norvegicus).* Further description of the host range of *E. perilis* was not found in the literature but *E. octotricha, E. calabyi* and *E. cornuta* are known only from *L. latifrons* (Dunnet and Mardon 1974). It has been suggested that *E. cornuta* is specific to wombats, especially *L. latifrons* (Dunnet and Mardon 1974). There has been no known overlap of the distribution of *L. latifrons* and *L. krefftii* of recent times. *Lasiorhinus* spp. fossils, however, have been found from Balladonia in Western Australia, to Lake Menindee in New South Wales and the Darling Downs in Queensland (Wells 1989). If *E. cornuta* found on *L. krefftii* was acquired from *L. latifrons* (or vice versa) it may have been associated with *L. krefftii* for a very long time. The possibility of chance contamination by researchers transporting field equipment used with *L. latifrons* to EFNP is remote but cannot be totally discounted.

The louse, *B. dubia,* found on *L. krefftii* has also previously been known only from *L. latifrons.* This adds further weight to the possibility that *L. latifrons* and *L. krefftii* may have acquired the same parasite species when the host genus was more widely dispersed across Australia.

Despite mange being one of the major threats to the southern wombat species (Doube 1981), no clinical disease was found in the population of *L. krefftii* at EFNP. Crossman (1988) has previously reported mange in *L. krefftii*, but only as a minor skin condition in winter which was not present in the spring/summer. Consequently, mange-causing mites may exist as a latent infection in this host. The disease is linked to overcrowding, habitat degradation and the presence of foxes and feral dogs (Booth 1994). Therefore, if an animal becomes immunocompromised the possibility of a severe infection cannot be discounted. Confirmation of mites being present in the park remains one of the important questions that should be answered for management purposes.

Although there are obvious deficiencies in trying to infer the possible worm burden of a host from faecal egg counts, this technique is still one of the most widely used methods for monitoring parasite loads. It does show when parasites are present and can give some indication of pasture contamination (Grenfell *et al*. 1995). When it is not practical to use more invasive techniques this method remains the only useful determinant of infection levels in a host individual or population. At EFNP the arrangement of burrows, clustered together along an old water course, allowed regular sampling of the total wombat population. This distribution, together with the unique behaviour of the wombats, ensured that freshly voided faecal samples, each from a single individual, could be collected although a particular host could not be linked to a particular sample or tracked from one sampling period to the next.

Gastro-intestinal nematodes, as indicated by consistently high faecal egg counts, were present every time the population was sampled. All samples contained eggs that appeared to be morphologically identical to *O. eppingensis;* the only species of nematode known from *L. krefftii* (Smales 1994). Although the eggs of related species are often morphologically indistinguishable (Crofton 1966), it seems probable that *O. eppingeisis* was the only gastro-intestinal nematode present. This is congruent with the number of gastro-intestinal species (only two) which occur in its congener *L. latifrons*, (Smales 1998) despite the less severe constraints on the distribution of the latter host.

Gastro-intestinal nematodes of wombats, with the exception of *Strongyloides* sp. discussed below, are considered non-pathogenic (Booth 1994), and therefore probably not a health problem. Larvae, identified as *Strongyloides* sp., similar in dimensions to the third stage filariform larvae of *S. spearei* (480-560 µm long) described by Skerrat (1995), were found at relatively high prevalences. Monitoring the population for intensity of *Strongyloides* sp. infections could provide a useful indicator of potential risk to the population since *Strongyloides* spp. have the potential to become serious pathogens of marsupials (Skerrat 1995).

The lack of trematode eggs in faecal samples was expected. With no freely available water in the Park it would be difficult for a trematode to establish itself or persist in the area, since trematodes require moist to wet conditions suitable for molluscs (usually snails) that are their intermediate hosts.

Finding cestode proglottids rather than eggs in the faecal pellets is indicative of a survival strategy that provides added protection to eggs in a hot dry climate. The only cestode identified from *L. krefftii* is *Paramoniezia suis* which is normally found in pigs,

but doubt has been cast on this identification by Beveridge (1976). Wombats were commonly mistaken for wild pigs in Queensland earlier this century (Horsup and Davidson 1994) and were even referred to as "bush pigs" by early settlers (Beveridge 1976). Therefore Beveridge (1976) suggested that the cestode identified as *P. suis* from *L. krefftii* is more likely to be the non-pathogenic *P. johnstoni*, Beveridge, 1976, normally found in *L. latifrons* and *V. ursinus*. Presidente (1979) also noted lesions associated with migrating *Taenia hydatigena* larvae in *V. ursinus.* It would be prudent therefore to ensure that any relocation program excludes the possibility of exposing *L. krefftii* to pastures potentially contaminated by this latter cestode, whose life cycle includes dogs, sheep or deer.

Since protozoan oocysts were not detected, they may have been present in extremely low numbers, or absent. If the latter is the case, this population of *L. krefftii* may have no immunological defenses against coccidia such as *Eimeria. Toxoplasma* species, present in other wombats (Doube 1981; Booth 1994), can only be detected by tissue biopsy. Any plans to translocate wombats and/or attempt a captive breeding program will, however, need to be formulated with the possibility of such infections occurring if animals are moved to environments where there is an increased likelihood of infection. Although coccidiosis is rare it may appear in naïve or immunologically compromised wombats introduced to environments where the parasites may exist in higher levels than the animal has previously experienced (Hum *et al*. 1991). If such infections do not occur in the population, it raises the question of whether *L. krefftii* was never infected or if an infection has been lost because of contraction of the geographic range of the host.

In this study there was no indication of parasites causing disease and the number of parasite species found was few. Therefore, since *L. krefftii* at EFNP are the only extant population, which is isolated in a discrete geographic area, the threat of parasitic disease is slight. It would be prudent, however, to consider the consequences of allowing exotic diseases into the park. This could occur, for example, if animals became infected during a captive breeding program. They could then act as carriers, transmitting disease when they were released back into the wild (Viggers *et al*. 1993).

Records are already available of parasites that have caused fatalities in other species of wombat (Doube 1981; Booth 1994). Measures should be taken to ensure that in the future these diseases do not affect either the Epping Forest or any newly established colony. Exotic diseases caused by *Toxoplasma gondii*, *Sarcoptes scabiei* and *F. hepatica* are endemic in certain areas of Australia (Johnson *et al*. 1988; Booth 1994) and constitute a real threat to the wombat if translocation of these animals were to take place without proper protective measures.

ACKNOWLEDGEMENTS

We thank Dr N. Chilton, Ms E. Evans and Dr S. Barker for assistance with identification of the ecoparasites found in this study. This project would not have been possible without the co-operation and help of the Wombat Recovery Team and staff of the Dept. of Environment, Queensland, especially Dr A. Horsup, who facilitated logistical support and provided helpful advice.

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