Wildlife disease ecology in changing landscapes: Mesopredator release and toxoplasmosis

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1. Introduction

Wildlife disease is a key threatening process for species conservation. Evidence suggests that diseases are increasing in the face of escalating environmental degradation and worldwide homogenisation of ecosystems (Daszak, 2000). Increased exposure to feral and domestic animals (e.g. Thorne and Williams, 1988; Dobson and Foufopoulos, 2001) and biodiversity loss (Keesing et al., 2010) have both been associated with increased disease risks. Additionally, they have played a primary role in many recent emerging diseases that have resulted in significant population declines (Schrag and Wiener, 1995), including canine distemper in lions (Panthera leo) (Kissui and Packer, 2004) and black footed ferrets (Mustela nigripes) (Thorne and Williams, 1988). There is a critical need to understand the context in which diseases operate in changing ecosystems and the potential threats posed to susceptible populations (Schrag and Wiener, 1995; Woodroffe, 1999; Keesing et al., 2010).

Changes in ecosystems can increase transmission and prevalence of diseases by either changing host or vector ecology, including abundance and behaviour, or by compromising immune function through stress (Schrag and Wiener, 1995; Dobson and Foufopoulos, 2001; Keesing et al., 2010). Hence, biodiversity loss that can reduce competitive pressure on reservoir hosts and thereby increase their population density, has the potential to increase pathogen transmission (Keesing et al., 2010), amplifying the abundance of infectious agents in the environment (Scott, 1988). Feral animals in particular are an important factor...
in disease emergence. These animals can facilitate both the trans-
migration of pathogens to naive hosts through contact with both
domestic species and wildlife as well as the introduction of new
pathogens to naive wild animal populations (Dobson and Foufop-

The obligate coccidian parasite Toxoplasma gondii is particularly
important as an example of a pathogen of conservation significance
spread by feral animals. Members of the cat family (Felidae) are the
only known definitive hosts, but nearly all warm-blooded animals
can act as intermediate hosts. T. gondii infection is widespread and
has been proposed as a conservation threat arising from feral cats,
as indicated by clinical and subclinical infections observed in wild
intermediate host-species ranging from Australian marsupials (e.g.
Attwood et al., 1975; Johnson et al., 1989; Obendorf et al., 1996), to
dolphins (e.g. Inskeep et al., 1990), sea otters (Enhydra lutris nereis)
(Cole et al., 2000; Miller et al., 2002), new world monkeys (e.g.
Dietz et al., 1997) and many avian species (Dubey, 2002). Most
mammals, including humans, which are infected with T. gondii
have asymptomatic infections; however, the virulence of the strain
of the parasite and the susceptibility of the host can affect patho-
genicity within a host (Innes, 1997; Hill et al., 2005; Parameswaran
et al., 2010). T. gondii is of particular concern for immune-compro-
mised individuals, pregnant females (Dubey, 1991; Innes, 1997;
Hill et al., 2005) and wildlife species which have not co-evolved
with felids and their parasites, such as Australian marsupials
(Johnson et al., 1988; Innes, 1997). The transmission route may
be through the ingestion of oocysts, the infective and environmen-
tally resilient stage of the T. gondii parasite, excreted in the millions
by a single cat and remaining viable in vegetation, soil and water
for up to a year in favourable conditions (Dubey, 1991; Hill et al.,
2005). Alternatively, infection can occur through ingestion of tissue
cysts in infected meat (Hill et al., 2005) or by vertical transmission
from mother to offspring (Parameswaran et al., 2009b).

In addition to overt increases in mortality or morbidity, patho-
gen can affect hosts in subtle ways including secondary costs
associated with greater energy investment in immune responses,
changes in anti-predator behaviours or reduced breeding success
and competitive fitness (Scott, 1988; Lafferty et al., 2006), placing
changes in anti-predator behaviours or reduced breeding success
gen can affect hosts in subtle ways including secondary costs
levels (Webster, 1994, 2001; Berdoy et al., 2000). Subclinical infec-
tions have also been linked to an increased incidence of brain can-
dies remained unexposed to
From mother to offspring (Parameswaran et al., 2009b).

Eastern barred bandicoots (Perameles gunnii) have been shown to succumb to experimental T. gondii infection within a
few days, without producing IgG antibodies (Bettiol et al.,
2000), and evidence demonstrates that this species may not have
the ability to maintain a long-term subclinical T. gondii infection
(Obendorf et al., 1996). Seroprevalence of T. gondii in several na-
tive Australian marsupials varies greatly among populations. This
may relate to the local distribution of feral cat populations and
conditions affecting the persistence of oocysts in the environment
(Arundel et al., 1977; Parameswaran, 2008). The individual and
population level impacts of both acute and latent T. gondii infec-
tion on wild Australian native species or on any naive host spe-
cies worldwide are poorly understood.

In Tasmania, the small island state of Australia, a unique oppor-
tunity exists to study disease ecology associated with changes in
definitive host populations. Populations of feral cats are increasing
(Hollings et al., submitted for publication) following a severe dis-
ease-induced population decline of the apex mammalian predator,
the Tasmanian devil (Sarcophilus harrisii), from devil facial tumour
disease (DFTD), an unusual transmissible cancer (Hawkins et al.,
2006; Lachish et al., 2007; McCallum et al., 2007). This increase in
feral cat populations appears to be an instance of mesopredator
release, a phenomenon in which smaller predators increase in
abundance following the decline of an apex predator. Mesopreda-
tor release (sensu Soule, 1988) has been reported from a
range of global ecosystems with wide ranging effects including
species extinctions (Crooks and Soule, 1999; Johnson et al., 2007)
and changes in community composition (Couchamp et al., 1999;
Terborgh et al., 2001; Beschta and Ripple, 2009).

To the best of our knowledge, the direct effects of apex predator
loss and mesopredator release on changing disease dynamics have
not been examined. The aim of this study is to assess whether the
density of feral cats is linked to the seroprevalence of T. gondii
infection in Tasmanian wildlife, to determine whether an increas-
ing feral cat population may correspond to an increased level of
risk to naive native marsupials of contracting acute toxoplasmosis.
We also assess the seroprevalence in different trophic levels and
then assess seroprevalence in a model species in relation to feral
cat densities from two different sample sources: road kill and ani-
mals culled under permit.

2. Materials and methods

2.1. Blood samples

To determine links between cat density and infection with T.
gondii, blood samples were collected from larger marsupial herbiv-
vores across Tasmania: common brush-tail possums (Trichosurus
vulpecula, ~3 kg; n = 14); Tasmanian pademelons (Thylagale bil-
dardierii, males = ~8 kg, females = ~4 kg; n = 228); and Bennetts
wallabies (Macropus rufogriseus males = ~15 kg, females = ~11 kg;
n = 25). Herbivores were considered to be the most suitable species
for this part of the study as exposure is through contact with oo-
cysts in the environment, including vegetation and water, without
the confounding effect of the consumption of infected prey, although
possums are partially omnivorous. Smaller native mam-
mals were not assessed due to difficulties in testing for latent
infection in these species. Blood samples were collected both
opportunistically from fresh road-kill and more systematically
from animals culled by commercial and private shooters holding
permits for crop and forestry grazing protection. Road-kill samples
were taken from the heart within several hours of death, ensuring
that blood was still fluid. Where possible in culling programs, sam-

pars were taken from multiple individuals of the same species from
each localised area, which were at least 5 km apart from each
other. Samples from these animals were collected from free flow-
ing blood from wounds or from the heart immediately after death.
Following initial results, efforts were focused on collecting samples
from Tasmanian pademelons. This species has a previously re-
ported T. gondii antibody prevalence of 18% using an enzyme-
linked immunosorbent assay (ELISA) test (Johnson et al., 1988). Samples for pademelons were taken over the entire northern half of Tasmania within a two year period.

In addition, we collected blood samples from other marsupial species to establish seroprevalence of *T. gondii* at different trophic levels. Samples were tested from three higher-order marsupial carnivores from several locations in Tasmania: the Tasmanian devil (males = ~8 kg, females = ~6 kg; *n* = 18); the spotted-tail quoll (*Dasyurus maculatus*; males = ~4 kg, females = ~2 kg; *n* = 7); and the eastern quoll (*Dasyurus viverrinus*; males = ~1 kg, females = ~0.8 kg; *n* = 24). All samples for these species were obtained by ear vein puncture during live trapping. Tasmanian devil and spotted-tailed quolls samples were collected in relatively low cat density areas mostly in the north-west (Figs. 1 and 2). Eastern quolls were sampled in areas of relatively lower and higher cat density in the north-west and south of the state (Figs. 1 and 2). Additional samples (*n* = 17) were collected from an offshore island, Bruny Island, where Tasmanian devils have never been present. *T. gondii* seroprevalence in these samples was compared to that in eastern quolls from Tasmanian mainland sites but was not included in the overall trophic analysis.

Samples from feral cats, the definitive host of *T. gondii*, were taken opportunistically by commercial shooters. All of the samples were obtained in the north-west region which remains DFTD free and exhibits low cat density (Fig. 1).

2.2. Testing for *T. gondii*

We tested blood samples for IgG antibodies, the class of antibodies representing the secondary immune response, using a modified agglutination test (MAT) (BioMerieux, France). The addition of beta-mercaptoethanol in the MAT test destroys non-specific IgM antibodies, reducing the risk of false-positive results (Desmonts and Remington, 1980). MAT testing has been used extensively for detection of exposure to *T. gondii* in marsupials (Obendorf et al., 1996; Hartley and English, 2005; Eymann et al., 2006; Parameswaran et al., 2009a), as it does not require a species specific reagent and is highly sensitive. The MAT test has high specificity and there is no evidence for cross-reactivity with related organisms (Dubey et al., 1995; Dubey, 1997). Samples were tested at three different sera dilutions: 1:16, 1:64, 1:256. A positive reaction was acknowledged if agglutination occurred at a sera dilution of at least 1:64.
A subsample of blood sera (5%) was retested with the MAT test for verification at the Animal Health Laboratory of the Department of Primary Industries, Parks, Water and Environment (DPIPWE), Tasmania.

2.3. Approximating cat density

We used a long-term state-wide spotlighting dataset to estimate feral cat density across Tasmania. The night-time spotlighting survey is conducted annually by DPIPWE, and covers more than 170 transects, each 10 km long. Individual spotlighting transects are grouped into 29 defined districts based on their proximity to each other, with each district having between 3 and 8 separate transects (Hocking and Driessen, 1992). All non-domestic species are recorded in addition to environmental variables that may affect animal detectability or activity level, including moonlight, wind speed and rainfall (Hocking and Driessen, 1992).

The total number of cats observed over an eight year period on all transects within each spotlight district was divided by the total number of transects surveyed to determine an index of cat density for each district (Fig. 1). The eight year period was chosen to allow for the life expectancy of all the species surveyed for T. gondii infection. This timeframe was considered appropriate as animals may be exposed even before they leave the pouch through vertical transmission (Parameswaran et al., 2009b), and IgG antibodies can remain present for life (Remington et al., 2004). For some districts DFTD was present for varying lengths of time during this 8 year period. The aggregation of data also reduced some of the issues with yearly variation, such as moonlight and rainfall, from surveying transects only once per year.

For a more detailed analysis of seroprevalence variation in response to cat density, all spotlighting transects that fell within a 10 km radius of each blood collection site were identified. An estimate of local cat density was calculated by dividing the number of cats detected on each transect within the 10 km zone during the eight year period by the total number of transects surveyed in the zone. Additionally, environmental variables which were hypothesised to affect the persistence of oocysts in the environment and positively influence cat density were included in the models. These are: the percentage of open vegetation (comprising largely of agricultural land), the average annual rainfall, and the number of address points (a proxy for human settlement and urbanisation), and they were determined for a 2 km buffer around the sites.

2.4. Statistical analysis

2.4.1. Cat density and T. gondii infection

For a broad scale analysis pademelon samples were divided by location into three geographical regions: north-east, central and north-west. These regions were selected to represent different lengths of time DFTD has been present which corresponds to the length of time and magnitude of Tasmanian devil population decline and potentially of mesopredator release of cats. DFTD has been present in the north-east region since (~1996–1999), with declines of up to 94% in some areas (Hawkins et al., 2006;
The disease arrived in the central region between approximately 2000 and 2007 and has also caused substantial population declines (>50%), whereas the north-west region currently remains free of the disease and maintains a large population of Tasmanian devils. Fisher's exact test (in R version 2.11.0) was used to assess evidence of significant differences in \textit{T. gondii} seroprevalence between geographic regions. This analysis was done using all of the pademelon samples and then repeated using only the culled samples to remove the potentially confounding collection method. An index of cat density was obtained for each of the three geographic regions by adding up the total number of cats sighted within the region and dividing by the total number of transects surveyed within the eight year period.

Seroprevalences in culled and road-kill pademelons were compared using Fisher’s exact test. For a finer scale analysis, we used generalised linear models (GLMs) with a binomial error structure and complementary log log link to model the seroprevalence of \textit{T. gondii} in pademelons as a function of cat density, the source of the sample (road-kill or culled) and environmental variables, specifically the geographic region, the percentage of open vegetation, the average annual rainfall and the number of address points.

There were 65 individual samples from a total of 228 which either had no spotlighting transects that fell within the 10 km radius or where we had only approximate co-ordinates; these samples were excluded from the analysis. We used weights \((w_i)\) derived from small sample corrected Akaike Information Criterion (AICc) to evaluate support for alternative models, where the weight indicates the relative support for each model (Burnham and Anderson, 2002). The relative importance of each explanatory variable was quantified by summing the weights of all models containing the variable (Burnham and Anderson, 2002; Rhodes et al., 2006).

### 2.4.2. Trophic level analysis

Fisher’s exact test was also used to compare \textit{T. gondii} seroprevalence among different hosts within a trophic level, i.e. within carnivores and browsers, and then between different trophic levels.

In addition, a generalised mixed model (GLMM) with a binomial error structure and complementary log log link function (using the ‘nlme’ library in R version 2.11.0) was used to assess whether \textit{T. gondii} seroprevalence in the carnivore guild, comprising the Tasmanian devil, spotted-tailed quoll and eastern quoll, was significantly different from the herbivore guild, comprising the brushtail possum, Bennett’s wallaby and Tasmanian pademelon. Seroprevalence was the binary response variable where presence or absence of \textit{T. gondii} infection in the individual was noted as 1 or 0 respectively. In this analysis, the guild was the fixed effect and the species within the guilds was the random effect.

### 3. Results

#### 3.1. Seroprevalence of \textit{T. gondii}

\textit{Toxoplasma} seroprevalences are reported in Fig. 3. Very low \textit{T. gondii} seroprevalence in brushtail possums (0/14 samples analysed) and Bennett’s wallabies (2/25 samples analysed) found in preliminary results would render detection of state-wide regional differences in seroprevalence in these species difficult even with large sample sizes. As a result, we focused subsequent blood sampling on pademelons.

There were significant differences between the three geographic regions in \textit{T. gondii} seroprevalence \((p = 0.035)\), with the north-west having the lowest seroprevalence \((7\%)\), followed by the north-east \((15\%)\) and the highest seroprevalence was recorded in the central region \((19\%)\) (Table 1). This regional difference was even stronger when assessing only culled pademelons \((p = 0.020; \text{north-west } 6\%, \text{central } 20\%, \text{north-east } 22\%)\). Cat density over the last eight years was the lowest in the north-west region, and

<table>
<thead>
<tr>
<th>Region</th>
<th>Number positive (n)</th>
<th>Total number (n)</th>
<th>Percent positive (%)</th>
<th>Est. cat density (cats per transect)</th>
</tr>
</thead>
<tbody>
<tr>
<td>North-east</td>
<td>6</td>
<td>39</td>
<td>15</td>
<td>0.13</td>
</tr>
<tr>
<td>Central</td>
<td>14</td>
<td>74</td>
<td>19</td>
<td>0.13</td>
</tr>
<tr>
<td>North-west</td>
<td>8</td>
<td>115</td>
<td>7</td>
<td>0.07</td>
</tr>
<tr>
<td>Total</td>
<td>28</td>
<td>228</td>
<td>12</td>
<td></td>
</tr>
</tbody>
</table>

Fig. 3. Prevalence of IgG antibodies of Tasmanian mammals to \textit{T. gondii} by trophic level; \(n\) represents the total number of samples tested. Standard error bars are shown.

Table 1

Prevalence of IgG antibodies to \textit{T. gondii} in Tasmanian pademelons. Cat density is the estimated value of the number of cats per transect within the region for data aggregated over 8 years.
The seroprevalence of *T. gondii* in road-kill samples from pademelons was considerably higher (31%, *n* = 16) than culled animals (11%, *n* = 212). The Fisher’s exact test indicated significantly different seroprevalence between the two sample sources (*p* < 0.033). However, the low number of road-kill samples relative to the culling samples means that this result should be interpreted with some caution.

The GLM analysis using the 10 km zones, there were nine models with a ΔAICc of less than two from the best supported model. Cat density, sample source, geographic region and annual rainfall were all contained within these top models, indicating some influence of these predictor variables on *T. gondii* seroprevalence in pademelons (Table 2). The source of the samples and the DFTD region were the two most influential predictor variables with a relative importance of 41% and 44% respectively. Cat density had a relative importance of 47% and rainfall 52%.

### 3.2. Trophic level differences

Different levels of the food web had significantly different seroprevalence levels of *T. gondii* (Fig. 3). Higher order mammalian carnivores, the Tasmanian devil (33%), spotted-tail quoll (71%) and the smaller meso-carnivore, the eastern quoll (58%) had a significantly greater likelihood of being seropositive (Fig. 3) than the omnivorous brushtail possum (0%) and both wallabies, the Tasmanian pademelon (12%) and the larger Bennett’s wallaby (8%). (GLMM model, herbivore est. = −1.50 ± 0.27, *p < 0.0001; Fishers 2 sided, *p < 0.0001). There were no significant differences among species within the higher order carnivore trophic level (*p* = 0.16, Fisher’s 2 sided), or amongst the three species classified as browsers ( *p* = 0.44, Fisher’s 2 sided). The spotted-tail quoll had the highest seroprevalence level of any species except cats, but sample size was relatively small (*n* = 7). High seroprevalence were also recorded in eastern quolls, with 72% at Cradoc (*n* = 11) and 46% at Cradle (*n* = 13). On Bruny Island however, seroprevalence in this species was only 12% (*n* = 17). The largest or top order mammalian predator, the Tasmanian devil, had the lowest seroprevalence of all carnivores overall. The seroprevalence in this host however indicated high levels of site variability between the two sites where Tasmanian devils were surveyed with 60% at West Pencil Pine (*n* = 5) and 23% at the north-west coast (*n* = 13). Similar evidence for large differences in site seroprevalences was observed in eastern quolls. All samples of the two highest order intermediate host species, the Tasmanian devil and spotted-tailed quoll, were taken from the north-west region, where cat density was relatively low. For the definitive host, the feral cat, seven of the eight samples were seropositive for *T. gondii*. The majority of these samples were obtained in the north-west region which has lowest feral cat density (Fig. 1).

### 4. Discussion

Seroprevalence of *T. gondii* in Tasmanian pademelons and cat population density are both higher in the north-east and central regions of Tasmania than in the north-west region. This suggests that the population density of the definitive host might play an important role in the transmission of *T. gondii*. Any increase in feral cat populations through mesopredator release, therefore, has the potential to increase the risk of *T. gondii* exposure in some naive native intermediate host species. Mammals at higher trophic levels exhibited significantly higher seroprevalence. However, neither high prevalence levels nor pathogenicity of parasites are a good gauge of the effect of disease on a population (McCallum and Dobson, 1995), and the population level impact of *T. gondii* infection on naive native host species, or indeed any host species remains unknown.

Feral cats in Tasmania appear to have consistently high seroprevalence of *T. gondii*, and despite the small sample sizes from our study, the results support similar evidence from an earlier study which found *T. gondii* antibodies in 51 of 53 Tasmanian feral cats using the indirect fluorescent-antibody test (Gregory and Munday, 1976). This suggests that feral cats can have a high risk of infection where they occur at low densities. Consequently, lower seroprevalence levels in intermediate hosts in areas with lower cat densities compared with areas of higher cat densities may result from reduced contamination of the environment with oocysts, rather than a minimised risk of infection for cats, for whom carnivornism is likely an important route for acquiring *T. gondii*. Vertical transmission may also play a role in macropods, maintaining high prevalence even in the absence of a definitive host (Parameswaran et al., 2009b), but the implications of this mode of transmission for the persistence of *T. gondii* infections in wildlife populations remains unknown. Further, the high genetic diversity of *T. gondii* in Australian wildlife leaves open the possibility that felids may not be the only definitive host for the genus in Australia (Pan et al., 2012). Because of the limited data available for estimating cat density, we were not able to assess in detail the magnitude of the effect of higher cat densities on *T. gondii* seroprevalence in Tasmanian marsupials and further investigation of this possible link is justified. The spotlighting dataset also has inherent issues with internal variability associated with different observers and weather conditions among other factors (Driessen and Hocking, 1992).

The seroprevalence of *T. gondii* in road-kill animals was significantly higher than in culled animals. This suggests that pademelons with *T. gondii* infection may be more at risk of being killed on the road than uninfected individuals. Unfortunately, the sample size of road-killed animals was small, but this intriguing result warrants further investigation. Latent infections in rodents and humans have revealed evidence of behavioural changes such as slower reaction times. Within the prey size range of feral cats, rats with subclinical *T. gondii* infections are at greater risk of predation (Webster, 1994; Berdoy et al., 2000). Even in humans otherwise asymptomatic infected individuals have decreased reaction times (Havlicek et al., 2001) and have an increased risk of car accidents (Flegler et al., 2002). Although disease has been proposed as a cause of sudden population declines of some Australian marsupials (e.g. Abbott, 2006) subclinical infections are rarely considered. *T. gondii* may be an example of a pathogen that even in the absence of clinical effects can cause behavioural changes that decrease survival, and individuals with subclinical infections could be more susceptible to predation.
Among Tasmanian marsupials the higher order carnivores had a significantly higher seroprevalence of *T. gondii* infection than the herbivores, from which we infer that the risk of infection with *T. gondii* increases at higher levels of the food-web. Exposure to infected wild prey or carrion explains high prevalence in carnivores (Dubey, 1991; Dubey et al., 1999). Carnivores at the top of the food-web may receive continuous and concentrated doses of infectious agents in their diet (Munsen, 2003), likely explaining the increasing vulnerability to parasites with increasing trophic level (Lafferty et al., 2006). Infection in carnivores and scavengers is particularly common with extremely high prevalence reported and infection levels are a good indicator of prevalence of *T. gondii* in the environment (Dubey et al., 1999; Hill et al., 2005). The high seroprevalence of *T. gondii* in carnivores likely reflects its presence in prey and also in the environment. The MAT test, which destroys IgM antibodies, measures exposure to a parasite, rather than a recent infection; this means seroprevalence essentially reflects a lifetime of exposure because IgG antibodies can persist for life (Remington et al., 2004). The marsupial carnivores we surveyed were mostly in the north-west where cat densities are generally lower (Fig. 1) and seroprevalence may be higher in other areas of Tasmania where cat densities are higher. The effects of *T. gondii* infections at both an individual and population level, and whether seroprevalence levels provide an indication of potential risks to these species are unknown.

In coyotes and red foxes in North America, prevalence in the larger more dominant predator (coyote 59%) was lower than in the smaller mesopredator (red fox 86%) (Dubey et al., 1999). In our sample, *T. gondii* prevalence was higher in the smaller native mesopredators than the apex predator, the Tasmanian devil, but this was not statistically significant. With carnivores, the number of prey items may be more important than the density of the definitive host in exposure to *T. gondii* because the primary route of infection may be through eating infected prey, rather than through environmental contamination with oocysts. Smaller, more numerous and varied prey species typical of mesopredators may in part explain the higher seroprevalence for these species. Pathogen transmission is not always linked to host density (Keesing et al., 2010) and prevalence of generalist pathogens may remain high even if the density of any particular host species is low (Packer et al., 2003).

Different host species may exhibit variability in susceptibility to infection by a parasite in terms of resistance (variability in propensity to become infected) and in tolerance (variability in harm caused by a given level of infection) (Raber et al., 2009). For *T. gondii* and naive marsupials, there is evidence of variability in both tolerance and resistance. Jurd (1994) reported that the immune response of marsupials is both slower and less accentuated than in eutharian species. Indeed, Bettiol et al. (2000) found that eastern barred bandicoots had died from primary *T. gondii* infection before detectable antibodies had been produced. *T. gondii* itself may increase susceptibility in some marsupial hosts, following natural selection of strains that are highly transmissible between genetically similar hosts (Parameswaran et al., 2010). When assessing prevalence in Australian marsupials, macropod species may not be equally susceptible to infection, with smaller host species more vulnerable than larger counterparts to acute toxoplasmosis (Dubey and Crutchley, 2008). These differences in susceptibility were also found in the present study. The results are consistent with previous studies, which found prevalence in wild brushtail possums in a major urban area of just over 6% (Eymann et al., 2006) and prevalence in Bennetts wallabies from Tasmania of 3% (Johnson et al., 1988). In pademelons *T. gondii* seroprevalence of 18% has been reported (Johnson et al., 1988) and they have shown evidence of clinical signs in the wild (Obendorf and Munday, 1983; Johnson et al., 1989). Such differences may be a consequence of smaller species grazing closer to the soil increasing their risk of ingesting oocysts or through selection of parasite lineages which have increased transmissibility among hosts with a specific genetic makeup (Parameswaran et al., 2010).

Disease studies using prevalence indices may risk bias from both abiotic and biotic factors which result in differences in detection probabilities (Jennelle et al., 2007). Serological and MAT tests for *T. gondii* in wild animals may underestimate seroprevalence (Owen and Trees, 1998). The under estimation of seroprevalence may result from individuals infected through vertical transmission that can exhibit a tolerance to the infection (Owen and Trees, 1998; Parameswaran et al., 2009b). Age may also be a confounding factor in assessments of seroprevalence as the likelihood of coming into contact with *T. gondii* would increase over the lifetime of a host. Age is unlikely to be a significant factor in this study as methodology was standardised across all areas. Future research on *T. gondii*-induced mortality in marsupial population declines need to consider variations in susceptibility to the parasite, host ecology and environmental stressors.

For diseases that have density dependent transmission dynamics, higher host densities will enhance transmission rates and lead to increased likelihood of elevated parasite loads per individual host and disease outbreaks (Scott, 1988; Dobson and Foufopoulos, 2001). Increased cat abundance can be managed by implementing cat control programs, but caution must be exercised before these are initiated for any species to reduce disease transmission. Culling may have complex effects on host behaviour, for example by increasing animal movements and thus facilitating disease transmission (Donnelly et al., 2003). Currently the implications of management and control practices on the behaviour of feral cats are unknown and should be further investigated.

Species that flourish with environmental degradation and habitat destruction may be those responsible for magnifying transmission of pathogens (Keesing et al., 2010). Feral cats are a common species not only in Australia but worldwide in areas of human habitation and are resilient to environmental changes. They have been shown to respond positively to the loss of larger native predators in a variety of ecological systems (e.g. Crooks and Soule, 1999; Hollings et al., submitted for publication; Kennedy et al., 2012). Ecosystem changes, including loss of the apex predator, could provide an advantage to some parasite species, including *T. gondii*, if they facilitate transmission (Dobson and Foufopoulos, 2001; Horwitz and Wilcox, 2005). Recent epidemics which have significantly reduced populations have highlighted the need for better understanding of wildlife disease ecology. Parasites are an integral part of ecosystems and communities, affecting ecosystem health and regulation. They are susceptible to the same environmental processes as other larger species which dominate conservation programmes. As the environment is continually altered through anthropogenic processes it is likely that infectious diseases will continue to emerge, and the potential of these pathogens to significantly affect the population viability of many endangered species, already suffering from overwhelming environmental pressures will escalate (Cleaveland et al., 2001; Dobson and Foufopoulos, 2001).

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