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Invited review

Parasites, emerging disease and wildlife conservation

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1 Invited Review

2 **Parasites, emerging disease and wildlife conservation**

3

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5

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18

19 **Abstract**

20

21 In this review some emerging issues of parasite infections in wildlife, particularly
22 in Australia, are considered. We discuss the importance of understanding parasite
23 biodiversity in wildlife in terms of conservation, the role of wildlife as reservoirs of
24 parasite infection, and the role of parasites within the broader context of the ecosystem.
25 Using a number of parasite species, the value of undertaking longitudinal surveillance in
26 natural systems using non-invasive sampling and molecular tools to characterize
27 infectious agents is illustrated in terms of wildlife health, parasite biodiversity and
28 ecology.

29

30 *Keywords:* Wildlife; *Giardia*; *Cryptosporidium*; *Blastocystis*; *Echinococcus*;
31 *Leishmania*; *Trypanosoma*; *Toxoplasma*

32

33

34 **1. Introduction**

35 In terms of conservation, it is unfortunate that the emergence of infectious
36 diseases with zoonotic potential has dominated investigations and commentary on
37 wildlife pathogens in recent years (e.g. McCallum and Dobson, 1995; Holmes, 1996;
38 Daszak et al., 2000; Rhyan and Spraker, 2010). By doing so, not only have studies on the
39 biodiversity and ecology of wildlife parasites been neglected, but control efforts have also
40 been hampered. This is because with the focus on threats to humans or domestic animals,
41 we have yet to gain a broader understanding of how these emergent pathogens interact
42 with the assemblage of cohabiting organisms in the ecosystem. Most importantly,
43 however, the potential impacts of infectious diseases on wildlife populations have often
44 been overshadowed by the apparently more pressing anthropocentric issues. For example,
45 the ongoing debate as to whether or not the culling of badgers (*Meles meles*) is an
46 effective means of controlling Tuberculosis (TB) in cattle in the United Kingdom (UK)
47 shows how a failure to fully understand the issues at hand can result in a polarization
48 within political and scientific communities (Jenkins et al., 2010).

49 In this review we will focus on some emerging issues of parasite infections in
50 wildlife, particularly in Australia, with emphasis on their impact on conservation.
51 Information obtained in the past has been limited and often acquired opportunistically
52 from sporadic culling, autopsy of individual cases or examinations of road-kills. More
53 recently, longitudinal surveillance of live trapped animals using molecular tools to aid
54 parasite characterization is providing valuable, novel data. In particular, the ability to
55 amplify DNA directly from minute parasite stages recovered from faeces, tissue or
56 environmental samples by PCR has been of enormous value in characterizing parasites.

57 As well as discussing the threats posed to wildlife by emerging parasitic diseases, we will
58 briefly consider the role of parasites within the broader context of a healthy, functioning
59 ecosystem.

60

61 **2. Parasites and parasitic disease in threatened animal populations – the need for** 62 **surveillance**

63 The fact that parasites can have a significant impact on the population dynamics
64 of wildlife has emerged as a critical issue in the conservation of threatened species
65 (Hudson et al., 1992, 1998; Tompkins and Begon, 1999; Daszak et al., 2000; Albon et al.,
66 2002; Newey and Thirgood, 2004; Hawlena et al., 2007; Møller and Nielsen, 2007;
67 Pedersen et al., 2007; Aguirre and Tabor, 2008; Burthe et al., 2008). Most authorities
68 would now agree that insufficient attention has been given to the role of infectious
69 diseases in conservation, even though there is mounting evidence that infectious agents
70 can impact significantly on local populations by causing temporary or permanent declines
71 (Daszak et al., 2000; Harvell et al., 2002; Smith et al., 2006). For example, the fungal
72 disease chytridiomycosis is devastating amphibian populations worldwide with the
73 decline and possible extinction of at least 14 species of frogs in Australian rainforests
74 (Retallick et al., 2004). In addition to such new, emerging diseases, a major concern to
75 wildlife diversity is the inadvertent introduction, by translocation or release of captive
76 bred animals, of known ‘animal’ pathogens into populations that have no previous
77 exposure and therefore no effective immunity (Wyatt et al., 2008). Similarly, the impact
78 of spill-over of ‘human’ parasites to naïve species of wildlife is another emerging threat
79 that is not well understood yet such spill-overs are likely to increase in the future,

80 establishing novel spill-back reservoirs of potential public health and economic
81 significance, as well as threatening wildlife.

82 Clearly, understanding the role of infectious agents in wildlife endangerment,
83 declines and extinctions requires accurate data on the diversity and abundance of
84 potential pathogens in natural systems, particularly at a local level (Smith et al., 2009a).
85 Although the theoretical and historical evidence is often compelling (de Castro and
86 Bolker, 2005; Gerber et al., 2005), surveillance of native fauna is urgently required in
87 order to document pathogen diversity, establish their potential importance in the
88 aetiology of disease and identify the circumstances of when this is most likely to occur
89 (Smith et al., 2009b). For example, local extinctions in Australian rainforest frogs were
90 first attributed to disease on the basis of the epidemiological pattern of the declines
91 (Laurance et al., 1996), but the inference of disease impact was strongly challenged until
92 the isolation and identification of a fungal pathogen (*Batrachochytrium dendrobaties*)
93 from morbid frogs (Berger et al., 1998).

94 Australia's long isolation has resulted in the evolution of a unique, extremely rich
95 and varied native fauna and one that is potentially susceptible to introduced parasites
96 (Freeland, 1994; Abbott, 2006). For example, concern has been expressed that should
97 *Trypanosoma evansi*, the causative agent of Surra, be introduced into Australia it could
98 devastate native mammalian fauna (Thompson et al., 2003). Many small and medium
99 sized mammal species, for example, that were once widespread across the continent are
100 now restricted to isolated areas in the south west or on off-shore islands. In the last 10
101 years there has been a further decline of many of these native mammals, with evidence
102 that parasitic diseases may be implicated (Start and Mawson, 2005; Smith et al., 2008).

103 Similarly, the highly endemic freshwater fish fauna of Australia is threatened by a range
104 of anthropogenic processes, such as river regulation, secondary salinisation and
105 eutrophication (Barmuta, 2003). In recent years, it has become clear that parasitic
106 diseases, particularly those introduced with exotic fish species, may also pose a threat to
107 native fishes (Chapman et al., 2006; Marina et al., 2008; Lymbery et al., 2010).

108

109 **3. Wildlife as reservoirs of parasite infection**

110 Wildlife have often been incriminated as being the source of parasite diseases in
111 humans or livestock, and acting as reservoirs for such infections. It is interesting
112 therefore, to examine a few important parasites of public health significance where
113 wildlife have been identified as reservoirs and the circumstances of how this happened.

114

115 *3.1. Giardia*

116 When the World Health Organization initially listed the common enteric
117 protozoan parasite *Giardia* as a zoonosis over 25 years ago it was as a result of
118 epidemiological observations suggesting that giardiasis in campers in Canada was caused
119 by drinking stream water contaminated with *Giardia* from beavers (Thompson, 2004).
120 The assumption was that beavers shared a parasite with humans but this was not the case.
121 Epidemiological investigations revealed that only beavers downstream from sewage
122 works were infected and more recent studies have confirmed that beavers are susceptible
123 to infection with zoonotic strains of *Giardia* (Thompson, 2004). Thus beavers that

124 contract infection from accidentally ingesting *Giardia* of human origin can serve to
125 amplify the parasite, thereby increasing the numbers of *Giardia* cysts in stream and river
126 water. It is not known whether beavers harbour their own 'strain' of *Giardia*.

127 *Giardia* has been found in numerous species of mammalian wildlife but very little
128 information is available on the species and strains/genotypes that occur naturally in
129 mammals in the wild (Appelbee et al., 2005; Kutz et al., 2009; Thompson et al., 2009a).
130 Three species have been described from rodents, *Giardia muris* (mice), *Giardia simondi*
131 (rats) and *Giardia microti* (microtine rodents), which are all genetically distinct
132 (reviewed in Thompson and Monis, 2004), but little is known about their host range,
133 prevalence of infections and geographical distribution. More recently, a novel genotype
134 of *Giardia* was described in an Australian marsupial, a bandicoot known as the quenda
135 (*Isoodon obesulus*), and on the basis of genetic characteristics would appear to represent
136 a distinct species that may be endemic within Australian native fauna (Adams et al.,
137 2004). However, mice, rats, microtine rodents and bandicoots are also susceptible, like
138 beavers, to infection with zoonotic strains/species of *Giardia*. In the majority of cases,
139 where appropriate tools for parasite characterisation have been applied, the type of
140 *Giardia* found in free-ranging terrestrial and aquatic mammals has usually proved to be
141 of human origin, i.e. zoonotic genotypes/assemblages of *Giardia duodenalis* (Thompson,
142 et al., 2009b). Apart from beavers in North America, this has also been demonstrated in
143 presumed pristine and/or isolated environments involving wildlife species such as
144 primates in Africa, muskoxen in the Arctic, house mice on remote islands, Australian
145 marsupials and marine cetaceans in various parts of the world (Graczyk et al., 2002;
146 Moro et al., 2003; Sulaiman et al., 2003; Appelbee et al., 2005; Dixon et al., 2008; Kutz

147 et al., 2008; Teichroeb et al., 2009; Thompson et al., 2010). In all of these cases,
148 epidemiological evidence supports humans as the source of infection through
149 environmental contamination, either directly or indirectly via domestic animal hosts. The
150 impact of these zoonotic species of *Giardia* on wildlife is not known, but there is some
151 evidence of an association with clinical disease in primates (Graczyk et al., 2002).

152

153 3.2. *Cryptosporidium*

154 The emergence of *Cryptosporidium* as a significant parasite of humans,
155 particularly in patients whose immune system is compromised, stimulated considerable
156 research on the surveillance and characterization of the parasite in wildlife species, in
157 order to identify possible reservoirs of waterborne infection to humans (Thompson et al.,
158 2005). The outcome of these activities has been the identification of numerous, novel,
159 host-adapted species and strains of *Cryptosporidium* that pose little if any significance to
160 public health (Caccio et al., 2005; Hunter and Thompson, 2005; Slapeta, 2009). This
161 incredible diversity of *Cryptosporidium* spp. in wildlife clearly warrants further study in
162 terms of ecology, evolutionary biology and potential impact on wildlife health. However,
163 this is unlikely to happen if the priorities for research on *Cryptosporidium* continues to be
164 driven by public health and water quality issues.

165

166 3.3. *Blastocystis*

167 As with research on the epidemiology and biodiversity of *Giardia* and
168 *Cryptosporidium*, the application of molecular tools has provided much new information
169 on the ubiquitous enteric protozoan *Blastocystis*. This parasite has proved to be far more
170 common in wildlife than previously considered, due to the limitations of previous
171 diagnostic techniques and the lack of surveillance in wildlife (Parkar et al., 2006;
172 Stensvold et al., 2007). An enormous genetic diversity has been found between isolates of
173 the parasite from wildlife but no clear picture has as yet emerged on the degree of host
174 specificity (Parkar et al., 2006, 2010). Interestingly, genotypes of the parasite isolated
175 from humans have also been found in wildlife from pristine environments and it is
176 unclear if there is any relationship between their occurrence and zoonotic transmission.
177 *Blastocystis* was long considered to be of no clinical significance to humans but opinion
178 has changed and there is a growing consensus that infection with *Blastocystis* is associated
179 with a variety of gastrointestinal disorders including irritable bowel syndrome
180 (Dogruman-Al, et al., 2009; Yamamoto-Furusho and Torijano-Carrera, 2010). The
181 potential impact of *Blastocystis* on wildlife health has yet to be determined.

182

183 3.4. *Echinococcus*

184 Wildlife are essential for maintaining the life cycle of some species of
185 *Echinococcus* in nature. For *Echinococcus multilocularis*, foxes and microtine rodents
186 serve this role whereas for *Echinococcus canadensis* it is wolves and cervids (Thompson,
187 2008). Unfortunately, with *Echinococcus granulosus*, anthropogenic activities have led to
188 the establishment on the Australian mainland of a cycle involving native wildlife that not

189 only impacts on public health and livestock industries but also on wildlife health (Jenkins
190 et al., 2005; Thompson et al., 2009b).

191 It is believed that *E. granulosus* was first introduced into Australia with sheep
192 during early European settlement in the late 1700's. The parasite is now widespread with
193 the larval cystic stage infecting many species of macropod marsupials (wallabies and
194 kangaroos) throughout the Australian mainland, a cycle perpetuated in the wild with
195 dingoes acting as the definitive host. From an ecological perspective, the recent
196 introduction of *Echinococcus* into Australia with domestic livestock has thus resulted in
197 the establishment of a sylvatic life cycle that can affect predator-prey relationships, as
198 well as host survival directly. In marsupials, as with *E. canadensis* in moose, *E.*
199 *granulosus* has a predilection for the lungs and can result in massive infections. Dingoes
200 hunt a range of macropods from small wallabies to large kangaroos (Corbett, 2001) and it
201 has long been considered that hydatid cysts in the lungs of macropods could weaken the
202 animal, making it easier for capture by dingoes (Durie and Riek, 1952) a similar situation
203 to that seen in the predation of infected moose by wolves (Mech, 2002; Joly and Messier,
204 2004; Thompson et al., 2009b). Recent studies suggest that hydatid disease reduces
205 effective lung volume in wallabies by ~ 55% in males and ~ 70-80% in females (Barnes
206 et al., 2007). These authors consider that such reductions impair the fitness of the animals
207 to a degree seldom seen in sheep, where infection is usually asymptomatic (Schwabe,
208 1986; Eckert et al., 2001).

209 Apart from enhancing susceptibility to predation, it has been suggested that the
210 presence of hydatid disease might be fatal and a threat to the survival of endangered small
211 macropod species that exist in small isolated colonies with small home ranges (Barnes et

212 al., 2007). This could be significant in Australia, where many species of marsupial are
213 under threat. In addition, the establishment of a dingo-macropod cycle, which effectively
214 maintains parasite transmission, also acts as a ‘spill-back’ reservoir of infection for sheep
215 and cattle and is a major problem for control strategies that focus on education and
216 husbandry activities to break the domestic ‘dog-sheep’ cycle on the Australian mainland
217 (Jenkins et al., 2005).

218

219 **4. Impact of parasite infection on wildlife**

220 Parasites of wildlife have been neglected in terms of conservation and
221 biodiversity. As emphasised earlier, interest has been dominated in recent years by
222 anthropomorphic issues, particularly the role of wildlife as reservoirs of zoonotic and
223 non-zoonotic diseases, and the consequences of such diseases to public health, livestock
224 industries and as a loss of human dietary protein (Rhyan and Spracker, 2010). Such issues
225 have dominated discussion and directed research effort. Fortunately, the importance of
226 generating data on the diversity of infectious agents harboured by wildlife populations is
227 now understood, in terms of conservation and ecosystem health. However, the effects of
228 endemic infections that circulate within wildlife populations can be difficult to detect and
229 even counterintuitive in their impact. For example, cowpox virus infection in wood mice
230 and bank voles in the UK appears to have a significantly negative effect during winter
231 months, resulting in reduced survival rates (Telfer et al., 2002). However, at the
232 population level, infected individuals appear to have increased survival rates over the
233 summer months. This unusual pattern is explained by the fact that during the summer

234 infected individuals tend not to breed or breed less often than uninfected individuals and
235 therefore do not suffer the added physiological costs associated with reproduction.
236 During the winter months, when all other factors are equal, the cost of infection becomes
237 apparent (Telfer et al., 2002).

238 We have selected trypanosomes and *Toxoplasma* in Australia as case studies to
239 illustrate the range of potential impacts on wildlife conservation but also, and
240 importantly, how localized, targeted investigations can yield valuable and often
241 unexpected data. Trypanosomes and *Toxoplasma* also provide valuable examples of how
242 little we understand about the ecological relationships between parasite and host and their
243 impact on wildlife health.

244

245 4.1. *Leishmania*

246 *Leishmania* is a vector-borne protozoan parasite transmitted by sandflies. The
247 genus consists of numerous species and subspecific variants that affect a variety of
248 wildlife mammalian hosts. In addition, humans and domestic dogs are susceptible to
249 infection with several species, which often results in serious disease. *Leishmania* has a
250 broad geographical distribution but South East Asia and Australasia have never, until
251 recently, been considered as endemic areas. Therefore, the discovery of *Leishmania* in
252 kangaroos and other macropods in the Northern Territory of Australia (Rose et al., 2004;
253 Dougall et al., 2009) raises a number of issues. Initially, and perhaps predictably, media
254 and government focussed in different ways on speculation that the kangaroos could be a
255 source of infection to humans (Stark et al., 2006). However, a systematic investigation of

256 the parasites isolated from the lesions of all affected macropods, including molecular
257 characterisation, has demonstrated that they all belong to the genus *Leishmania* but not to
258 any species so far described (Rose et al., 2004). This indicates that macropods and
259 possibly other native mammalian fauna in Australia harbour a novel species of
260 *Leishmania* that has perhaps evolved over thousands of years and adapted to its marsupial
261 host. All cases so far investigated have been in captive animals. This may be an important
262 factor in the expression of the disease since the stress associated with captivity may be
263 more likely to lead to overt clinical disease with the development of lesions than in
264 animals in the wild. Although the pathogenic significance of this species to wildlife is not
265 known and may be minimal to animals in the wild, it raises the question of how
266 *Leishmania* is transmitted between kangaroos. Presumably, there are species of sandflies,
267 or possibly other biting flies, capable of acting as vectors of *Leishmania* in Australia. If
268 so, these vectors could also transmit other species of *Leishmania*. It is interesting that in
269 Europe, there is increasing debate over the vectorial significance of other arthropods,
270 apart from sandflies, such as fleas and ticks, in the life cycle of *Leishmania* (Ferreira et
271 al., 2009; Dantas-Torres et al., 2010 ; Otranto and Dantas-Torres, 2010).

272 Pathogenic species of *Leishmania* have been reported to enter Australia regularly
273 in infected humans or dogs from endemic areas of the world (Thompson et al., 2003;
274 Stark et al., 2006; Konecny and Stark, 2007). Until recently, it has been assumed that
275 such infections represent a minimal biosecurity risk since Australia was considered not to
276 have vectors capable of transmitting the parasite. The discovery of the parasite in
277 kangaroos demonstrates that this is not the case, and thus imported cases of *Leishmania*
278 pose a risk of being transmitted to humans, their pets and, importantly, to wildlife.

279 Wildlife could become a significant reservoir, as well as suffer the potentially more
280 serious clinical consequences associated with exposure to a novel introduced pathogen
281 likely to be of human origin, in view of the increasing number of introduced cases in
282 immigrants to Australia and returning soldiers from endemic areas (Thompson et al.,
283 2003).

284

285 4.2. *Trypanosoma*

286 The situation with *Leishmania* in Australian wildlife demonstrates how little we
287 know about systemic and blood parasites of native wildlife in Australia. For example, we
288 are only just beginning to understand the diversity of trypanosomes in Australian
289 marsupials, their evolutionary biology, transmission, their potential impact on the health
290 of wildlife and the relationship indigenous trypanosomes may have to exotic, human
291 pathogenic trypanosomes that could establish a reservoir in native wildlife (Smith et al.,
292 2008). Until recently, little was known about the extent and diversity of infection with
293 *Trypanosoma* spp. in Australian wildlife. However, longitudinal molecular ecological
294 studies have demonstrated an overall pattern of widespread distribution, with
295 *Trypanosoma* genotypes/species occurring in many different host species, often at high
296 prevalences, on the mainland of Australia as well as on offshore islands (Averis et al.,
297 2009).

298 In South America and Africa, the trypanosomes that cause disease in humans and
299 domestic animals are primarily the result of spill-back from wildlife reservoirs (e.g.
300 *Trypanosoma cruzi* and *Trypanosoma brucei*). In Australia, however, human activities

301 have almost certainly been responsible for introducing trypanosomes from one wildlife
302 population to another through translocation, yet we do not understand whether parasites
303 associated with closely related host species pose a disease threat to translocated animals.
304 For example, within Western Australia, different *Trypanosoma* spp. infect the woylie
305 (*Bettongia penicillata*) and the closely related boodie (*Bettongia lesueur*), but nothing is
306 known regarding the potential for cross-infection and the consequences of increased
307 pathogenicity should this occur (Averis et al., 2009). Similarly, it has also become
308 apparent that geographically distinct populations of some Australian native species
309 harbour different *Trypanosoma* genotypes, which will almost certainly be infective to
310 other individuals should the populations, and presumably their vectors which have yet to
311 be identified, become mixed as the result of translocation (Averis et al., 2009).

312 Furthermore, the establishment of an exotic trypanosome cycle within Australian
313 wildlife would be greatly facilitated if specific arthropod vectors were inadvertently
314 introduced at the same time. For example, the introduction of flea-infested ship rats onto
315 Christmas Island (approximately AD 1900) resulted in the spread of the pathogenic
316 trypanosome *Trypanosoma lewisi* into the native rat population which was described as
317 'morbid' on subsequent visits and extinct within 25 years (Pickering and Norris, 1996;
318 Wyatt et al., 2008). Recent DNA-based findings indicate that native rats were devoid of
319 any trypanosome-like infection prior to arrival, suggesting *T. lewisi* was probably
320 maintained initially in a reservoir of ship rats and flea vectors, and subsequently spread
321 by contact between infected fleas and naïve native rats (Wyatt et al., 2008). The spread of
322 *T. lewisi* would have been enhanced if ectoparasites associated with native rats were
323 biologically capable of acting as more than just mechanical vectors.

324 Although multiple trypanosome infections within any single individual host have
325 not been found in Australian marsupials, there is evidence to suggest that some species
326 can be infected by multiple genotypes within the same location. For example, in the
327 carnivorous chuditch (*Dasyurus geoffroii*) two distinct genotypes occur in one isolated
328 conservation park (Averis et al., 2009). There is also evidence to suggest that some
329 genotypes are geographically widespread and occur in multiple host species, while others
330 appear to be host specific and/or confined to certain geographic areas (Fig. 1). For
331 example, as mentioned above, isolates detected in woylies from geographically different
332 populations, including a population in eastern Australia, consistently group together
333 within a single clade (Averis et al., 2009). Those from the closely related boodie appear
334 not only to be genetically different to genotypes from woylies, but to vary considerably
335 both within and between geographically distinct populations, suggesting that some host
336 species may be susceptible to infection from multiple *Trypanosoma* spp. and therefore
337 possibly susceptible to exotic species should they be introduced into Australia.

338 *Trypanosoma lewisi* was first reported in Australia by Bancroft (1888) and has
339 previously been found in bush rats (*Rattus fuscipes*) and water rats (*Hydromys*
340 *chrysogaster*) (Mackerras, 1959). Its presence within native wildlife raises questions
341 regarding its origin, specifically whether it was introduced along with black and brown
342 rats on ships arriving from Europe or was present before their arrival. It also raises
343 questions regarding its impact on other native wildlife species. Although *T. lewisi* is
344 usually considered to be a non-pathogenic trypanosome in its natural host (*Rattus rattus*),
345 it has been shown to vary in its clinical impact in different aged rats, with higher
346 incidences of anaemia and weight loss, and significantly greater mortality rates in

347 juveniles than adults (Brown, 1914, 1915). Similar observations have been made on
348 *Trypanosoma microti* in microtine rodent populations in England (Smith et al., 2005).
349 This apparent plasticity in virulence may also be influenced by concomitant infections or
350 the general condition of the host (Cox, 2001; Brown et al., 2003). For example, Smith et
351 al. (2008) reported a higher prevalence and intensity of trypanosome infection in a
352 declining population of woylies compared with that observed in a stable non-declining
353 population in the southwest of Western Australia. They suggested that the impact of
354 infection was only of significance for animals in the declining population because they
355 were also exposed to high levels of the parasite *Toxoplasma gondii* (Arrea et al., 1998). It
356 is also possible that the initial introduction of *T. lewisi* into Australia was associated with
357 adverse clinical impacts in native fauna but that over time some adaptation has occurred
358 in the host-parasite relationship in native fauna. It is also possible that less virulent forms
359 of *T. lewisi* have evolved in Australia. For example, in an isolated region in the south of
360 Western Australia, several species of native mammal were all found to be infected with a
361 closely related *T. lewisi*- like trypanosome (Averis et al., 2009).

362 The vectors of Australian trypanosomes are not known. Given the role of fleas as
363 vectors for *T. lewisi* they are considered likely to play a role with some of the
364 trypanosomes of native Australian wildlife since phylogenetic analysis of genotypes from
365 several ecologically similar wildlife hosts including bush rat (*R. fuscipes*), dibbler
366 (*Parantechinus apicalis*), and ash-grey mouse (*Pseudomys albocinereus*) suggests a
367 potentially similar mode of transmission (Hamilton et al., 2007; Averis et al., 2009; see
368 Fig. 1). Jakes et al. (2001) have proposed tabanid flies as possible vectors for kangaroo
369 trypanosomes, although ixodid ticks are also commonly found on kangaroos and are

370 thought to be involved in the transmission of trypanosomes in Japan (Thekiso et al.,
371 2007). Consideration should perhaps also be given to the potential role of triatomines as
372 vectors of trypanosomes in native wildlife. Little research has been undertaken on their
373 host range in Australia nor whether any bite and feed on the blood of mammals although
374 *Triatoma leopoldi* is thought to occur in northern Australia and is a vector of *T. cruzi* in
375 South America (Monteith, 1974). This is also of significance to biosecurity issues of
376 public health since if Australian triatomines act as vectors of native species of
377 *Trypanosoma*, they could presumably transmit *T. cruzi* should it enter Australia. Recent
378 reports have raised concerns on this issue, with evidence that thousands of migrants from
379 Chagas-disease endemic countries are now living in Australia (Schmunis, 2007; Gascon
380 et al., 2009; Schmunis and Yadon, 2009).

381 A more comprehensive understanding of the diversity of *Trypanosoma* spp.
382 associated with native wildlife may well contribute to the conservation efforts and
383 translocation programmes of endangered species in Australia. It is also important to
384 understand the current range of infections that exist within wildlife populations to be able
385 to predict their suitability to act as competent reservoirs for introduced exotic parasites in
386 the event that climate change (Polley and Thompson, 2009) leads to Australia becoming a
387 suitable environment for establishment (Thompson et al., 2009b). In this respect, an
388 exotic and unrelated trypanosome to those endemic in Australia, *Trypanosoma evansi*,
389 has long been considered a biosecurity risk because of its known clinical impact on
390 livestock. *Trypanosoma evansi* has a low mammalian host specificity and non-specific
391 mechanical vectorial requirements (Cleland, 1908; Thompson et al., 2003). The parasite's
392 geographical proximity to northern Australia in eastern Indonesia and potential spread via

393 Irian Jaya and Papua New Guinea is seen as a serious biosecurity issue. This is because if
394 *T. evansi* were to enter Australia, feral animals, particularly wild pigs, could act as
395 excellent reservoirs due to their abundance and the lack of clinical symptoms associated
396 with infection in pigs (Thompson et al., 2003). Although native mammalian fauna were
397 also considered for many years as providing an important reservoir of infection, the focus
398 was on their role in maintaining a parasite that could spread to livestock. However, recent
399 studies in experimentally infected macropods have demonstrated that in nature they are
400 likely to suffer severely and rapidly die from the clinical consequences of infection (Reid,
401 2002).

402

403 4.3. *Toxoplasma*

404 In Australia, the impact of *Toxoplasma* on wildlife has been an issue of speculation for
405 many years and it is only recently that a better understanding of the parasite's ecology
406 and relationship to the hosts that maintain it has been obtained. Over the years, infection
407 with *T. gondii* has been linked to wildlife declines of several species and the rapid
408 disappearance of native species in certain areas of Australia, although the evidence is
409 largely circumstantial (Shepherd, N.C., Mahood, I.T., 1978. The potential effect of feral
410 cats and dogs on Australian native fauna. Australian Adv Vet Sci, Proc. 55th Annual
411 Conf. Aust. Vet. Assoc, Sydney pp. 108; Braithwaite and Griffiths, 1994; Obendorf et al.,
412 1996; Abbott, 2006; Smith et al., 2008; Parameswaran et al., 2009a,b). The parasite is
413 known to cause severe, often fatal disease in Australian native fauna but most reported
414 cases have been in captive animals where the effects of stress are likely to have

415 exacerbated the consequences of infection (Brathwaite and Griffiths, 1994; Thompson et
416 al., 2009b). However, *Toxoplasma* infection has been frequently reported in many species
417 of free-ranging Australian wildlife not associated with any overt clinical symptoms
418 (Attwood et al., 1975; Speare et al., 1983; Turni and Smales, 2001; Henderson, 2009;
419 Parameswaran et al., 2009b, 2010). As such, the physiological effects of stress and
420 *Toxoplasma* infections may play an important role in modifying the host parasite
421 relationship whether caused by captivity, the presence of introduced predators such as
422 cats and foxes, poor nutrition, competition for food and resources or the effects of habitat
423 degradation. These are all thought to contribute to both host susceptibility and severity of
424 infection (McCallum and Dobson, 2002; Davey et al., 2006; Pedersen and Grieves,
425 2008), and must be considered in the context of *Toxoplasma* and wildlife declines, as
426 should enhanced predation through behavioural changes induced by chronic *Toxoplasma*
427 infection (Berdoy et al., 2000).

428 Molecular epidemiological studies of *Toxoplasma* infections are providing new
429 information on transmission and have revealed a much greater degree of genetic diversity
430 than expected, particularly in wildlife populations (Thompson et al., 2007). Our current
431 understanding of the genetic diversity and virulence of *T. gondii* has been driven by the
432 remarkably clonal population structure of the parasite in domestic transmission cycles in
433 Europe and North America, with just three predominant clonal lineages, referred to as
434 Types I, II and III, accounting for >95% of strains isolated from humans and domestic
435 animals (Howe and Sibley 1995; Grigg et al., 2001; Boyle et al., 2006). These three
436 lineages appear to have arisen within the last 10,000 years, from crosses among a few
437 genetically very similar parental strains (Grigg et al., 2001; Su et al., 2003; Boyle et al.,

438 2006). In contrast to this clonal population structure in European and North American
439 domestic cycles, much more genetic diversity has been found when *T. gondii* is sampled
440 from wildlife in North America and South America (Ajzenberg et al., 2004; Lehmann et
441 al., 2006). This diversity appears to be driven by regular cycles of sexual reproduction,
442 with occasional expansion of clonal lineages, suggesting an epidemic population structure
443 (i.e. one where reticulate relationships arising from frequent recombination are obscured
444 by the clonal expansion of a few genotypes) in South America and parts of North
445 America where wildlife plays a major role in transmission of the parasite (Ajzenberg et
446 al., 2004; Khan et al., 2007).

447 Until recently, almost nothing was known of the genetic diversity of *T. gondii* in
448 Australia. Our studies have found that *T. gondii* is common and widely distributed among
449 the native fauna of Western Australia, both geographically and in terms of the number of
450 host species infected. They have also revealed a level of genetic diversity that is much
451 greater than previously found anywhere else in the world, with the majority (67%) of
452 marsupial *T. gondii* infections caused by atypical strains, with several novel alleles
453 (Parameswaran et al., 2010). The abundance of atypical strains that preliminary studies in
454 Australian wildlife have revealed (Parameswaran et al., 2010), have the potential to
455 radically change the way we view *T. gondii* and toxoplasmosis. Of particular significance
456 is the relationship between genetic diversity, frequent recombination and virulence and
457 the potential for the rapid appearance of virulent strains that could be associated with
458 wildlife declines.

459

460 **5. The conservation value of parasite biodiversity**

461 We typically think of parasite infections of wildlife in a negative context, as a
462 cause of wildlife endangerment and possibly extinction. This is often the case,
463 particularly when exotic parasites are introduced into a naive host population (Daszak et
464 al., 2000; Lafferty and Kuris, 2005; Pedersen et al., 2007). This totally negative view of
465 parasites is, however, an oversimplification and there are two main reasons why we need
466 a more comprehensive understanding of the complex relationships between parasites and
467 their wildlife hosts.

468 First, parasites are an important component of biodiversity in their own right.
469 There are at least as many species of parasites as there are of free living organisms (Price,
470 1980; Toft, 1986) and there is increasing recognition that the extinction of free living
471 species is often accompanied by the coextinction of many parasite species, especially
472 those that are host-specific (Koh et al., 2004; Dobson et al., 2008; Dunn et al., 2009).
473 This is of particular concern for the parasites of wildlife hosts because we generally have
474 very incomplete knowledge of what parasite species exist in these hosts, or of their life
475 history and biology (Colwell et al., 2009; Lymbery et al., 2010). When a wildlife species
476 becomes extinct, we will, in many cases, not even know how many species of parasites
477 we have lost through coextinction. Lymbery et al. (2010), for example, found 44 putative
478 species of parasites in freshwater fishes of south-western Australia; all but two of these
479 species appeared to be native and most were undescribed.

480 Second, parasites are integral components of biological communities and have
481 pervasive effects throughout community food webs. The inclusion of parasites changes
482 the topological pattern of food webs, dramatically increasing connectance and nestedness,
483 which is expected to alter food web stability (Hudson et al., 2006; Lafferty, 2008).

484 Parasites may influence the behaviour of individual hosts (Lefèvre et al., 2009), regulate
485 host population sizes through direct effects on host birth and death rates (Møller 2005),
486 mediate competitive interactions among hosts (Thomas et al., 2005) and act as ecosystem
487 engineers (Thomas et al., 1999). Reductions in parasite population sizes and the loss of
488 parasite species from an ecosystem may therefore have important impacts on ecosystem
489 functioning and a cascading influence on other members of the biological community
490 (Thomas et al., 2005; Dobson et al., 2008).

491 In the face of all this, what advice should we give to wildlife managers about how
492 to deal with parasite infections? Unfortunately, there is no simple solution which will be
493 generally applicable. Certainly, there will be situations where parasitic disease will pose a
494 threat to endangered wildlife populations and treatment is essential. There are also likely
495 to be situations, however, where the continued presence of parasites is important to the
496 health of individual hosts, for example by stimulating acquired immunity, or to the health
497 of the population of hosts or the broader ecosystem of which that population is a part. The
498 great challenge is to better understand the diversity of parasites found in wildlife and the
499 ecological role they play in natural ecosystems. Only then will we be in a position to offer
500 more useful advice to wildlife managers.

501

502 **6. Concluding comments**

503 The collection and analysis of data on the diversity and ecology of parasites in
504 natural systems, and the nature of the host/parasite relationship, are clearly very
505 important in terms of conservation. However, this is a daunting task when one considers
506 that “we lack even species lists of parasites and pathogens for most, if not all wild

507 animals” (Mathews, 2009). Human encroachment of wildlife habitats, land degradation
508 and climate change are all exacerbating the problem with an ever increasing number of
509 threatened species being identified throughout the world. There is an increasing need for
510 initiating translocation programmes but without baseline data on the pathogen fauna of
511 source and recipient populations such programmes may be compromised. Such data will
512 also allow predictions to be made on the susceptibility of wildlife populations to exotic
513 pathogens.

514 A major impediment to the large-scale surveillance of wildlife populations has
515 been the difficulties of assessing infection status in living animals. This has been
516 alleviated with the development of molecular tools that can provide sufficient information
517 through non-invasive sampling of living animals. Gathering the information needed will
518 require a global effort of collaboration between conservation agencies, ecologists and
519 infectious disease specialists. This has to start at a regional level, as with the studies
520 described above on trypanosomes and *Toxoplasma*. These studies illustrate the value of
521 such surveillance, both in assisting with wildlife conservation and in providing a wealth
522 of new information about parasite biodiversity and ecology. We are only now starting to
523 appreciate just how important this information on parasite biodiversity may prove to be,
524 given that a healthy ecosystem may be one that is rich in parasites (Hudson et al., 2006).

525

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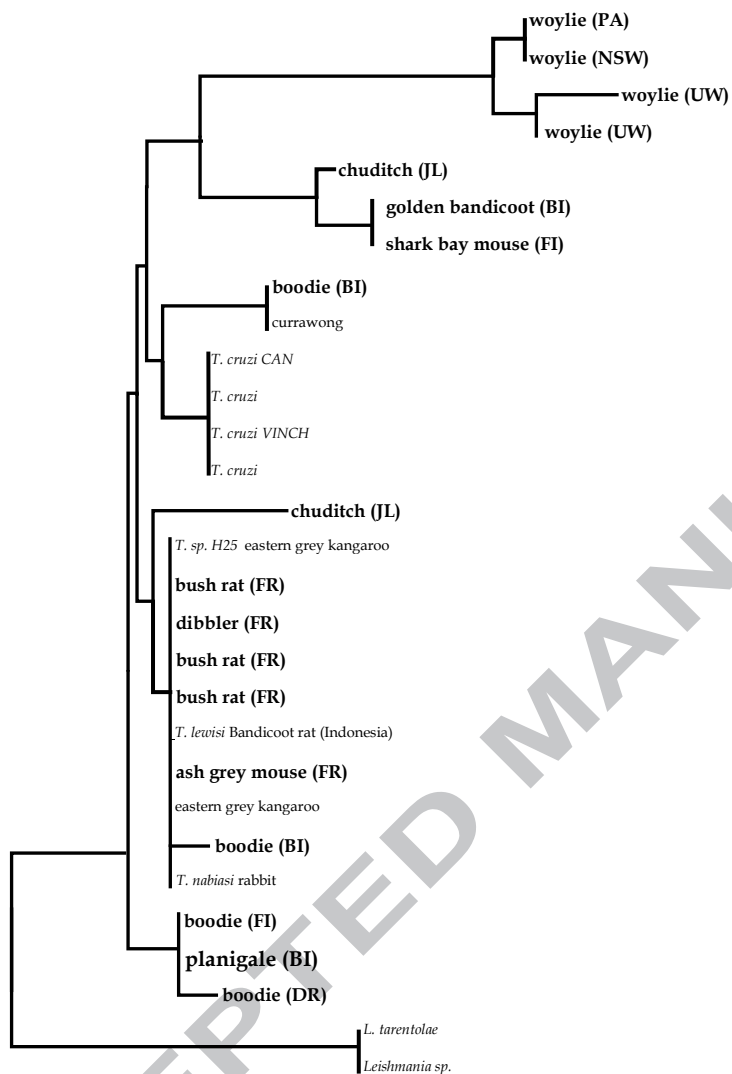
1004 **Figure legend**

1005 Fig. 1. Phylogram showing the relationship at the 18S rRNA gene between some
1006 Australian isolates of *Trypanosoma* (bold) and reference isolates. The relationship was
1007 inferred using the neighbour-joining method. Host species are indicated after
1008 *Trypanosoma* sp. or isolate code. Genotypes sequenced during this study are shown in
1009 bold. The letters in parentheses refer to sampling locality: BI, Barrow Island; FR,
1010 Fitzgerald River National Park; JL, Julimar Conservation Park; DR, Dryandra; FI, Faure
1011 Island; PA, Paruna; NSW, New South Wales; UW, Upper Warren (details in Averis et al.,
1012 2009).

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