J. Parasitol., 93(1), 2007, pp. 89–92

KLOSSIELLA QUIMRENSIS (APICOMPLEXA: KLOSSIELLIDAE) CAUSES RENAL COCCIDIOSIS IN WESTERN BARRED BANDICOOTS *PERAMELES BOUGAINVILLE* (MARSUPIALIA: PERAMELIDAE) IN WESTERN AUSTRALIA

M. D. Bennett, L. Woolford, A. J. O'Hara, P. K. Nicholls, K. S. Warren, J. A. Friend*, and R. A. Swan
School of Veterinary and Biomedical Science, Murdoch University, South Street, Murdoch, Western Australia, Australia, 6150.
e-mail: m.bennett@murdoch.edu.au

ABSTRACT: Previous studies have described a range of *Klossiella* species parasitic in marsupial hosts. *Klossiella quimrensis* is the etiologic agent of renal coccidiosis in the peramelid marsupial hosts *Isoodon obesulus* and *Perameles gunnii* in Eastern Australia, but there is no previous report of klossiellosis in Western Australian peramelids. This study describes klossiellosis diagnosed by histology of renal tissue sections collected during necropsy of 20 *Perameles bougainville* between 2000 and 2005. Sporonts, sporoblasts, and macrogametes were identified within parasitophorous vacuoles of epithelial cells located near the renal corticomedullary junction. The prevalence of renal coccidiosis in *P. bougainville* diagnosed by renal histology is estimated at 30%. Only a single unsporulated sporocyst was detected by examination of cystocentesis-collected urine, indicating that microscopic evaluation of urine samples is an insensitive diagnostic test for detection of *K. quimrensis* in *P. bougainville*. This infection in *P. bougainville* is indirectly associated with mild multifocal interstitial lymphohistiocytic nephritis and is likely to be only minimally pathogenic in otherwise healthy individuals. Our study also extends the host and geographic range of *K. quimrensis* to include *P. bougainville* and Western Australia.

Klossiellidae and *Klossiella* were erected for *Klossiella muris*, a renal coccidian parasite of mice. *Klossiella* spp. parasitize vertebrates and usually infect the kidneys (Taylor et al., 1979). There are currently 18 named *Klossiella* spp., mammalian hosts of which occur across the world in 3 eutherian orders, Perissodactyla, Rodentia, and Chiroptera, and 3 marsupial orders, Didelphimorphia, Diprotodontia, and Peramelemorphia (Levine, 1988). *Klossiella quimrensis* has been described in the kidneys of the peramelid hosts *Isoodon obesulus* (Shaw, 1797) and *Perameles gunnii* Gray, 1838, from Maria Island, Tasmania, Australia (Barker et al., 1975). The present study reports on the morphology, pathology, and prevalence of *K. quimrensis* infection in another peramelid host, the endangered western barred bandicoot, *Perameles bougainville* Quoy and Gaimard, 1824, from Western Australia.

MATERIALS AND METHODS

Western barred bandicoots were housed at the Kanyana Wildlife Rehabilitation Centre in small enclosures approximately 1.5 m × 1.5 m and at Dryandra Woodland in large enclosures approximately 400 m × 500 m. During necropsies conducted on 20 western barred bandicoots from these 2 captive breeding colonies between 2000 and 2005, kidneys were examined grossly, and representative samples of kidney tissue were collected into 10% neutral buffered formalin. Once fixed, these tissues were embedded in paraffin, sectioned at 4 μm, and stained routinely with hematoxylin and eosin (H&E). Sections of renal tissue were examined for evidence of klossiellosis and any associated renal pathology using an Olympus BX51 light microscope. Whenever possible, urine was collected by cystocentesis during necropsy and screened microscopically for evidence of urinary sporocysts. All measurements were made using an ocular micrometer.

RESULTS

Six of 20 (30%) kidneys examined histologically were found to be positive for renal coccidiosis. Whenever multiple slides were produced from the same infected kidney, it is noteworthy that each slide did not always include *K. quimrensis* life cycle stages. Only 1 sporocyst was detected by microscopic examination of urine collected by cystocentesis. This unsporulated urinary sporocyst was ellipsoidal (17 μm × 11 μm), with a rough, dark outer sporocyst wall and a micropyle at the more acutely tapered pole.

Our description follows the example of Gardiner et al. (1998) for the nomenclature of *Klossiella* sp. tissue stages. Parasitophorous vacuoles containing *K. quimrensis* life cycle stages are detectable within renal epithelial cells located near the corticomedullary junction. Round-to-oval basophilic macrogametes, 5.5 μm in diameter, are contained within a round-to-irregular eosinophilic parasitophorous vacuole up to 23 μm in diameter (Figs. 1–3). Microgametes were not identified in any renal tissue sections examined. Sporonts are round to ovoid, approximately 25 μm in diameter, amphophilic to basophilic, and dotted circumferentially with 12–16 deeply basophilic nuclei (Figs. 1, 2). These nuclei bud from the sporont, producing ovoid sporoblasts (5 μm × 7.5 μm) arranged around a residual body of amorphous cytoplasm (Fig. 2). Mature sporoblasts are amphophilic ovoid structures (7.5 μm × 10 μm) with multiple dotted basophilic nuclei. Numbers of mature sporoblasts found within individual parasitophorous vacuoles range from 7 to 24 (Fig. 3).

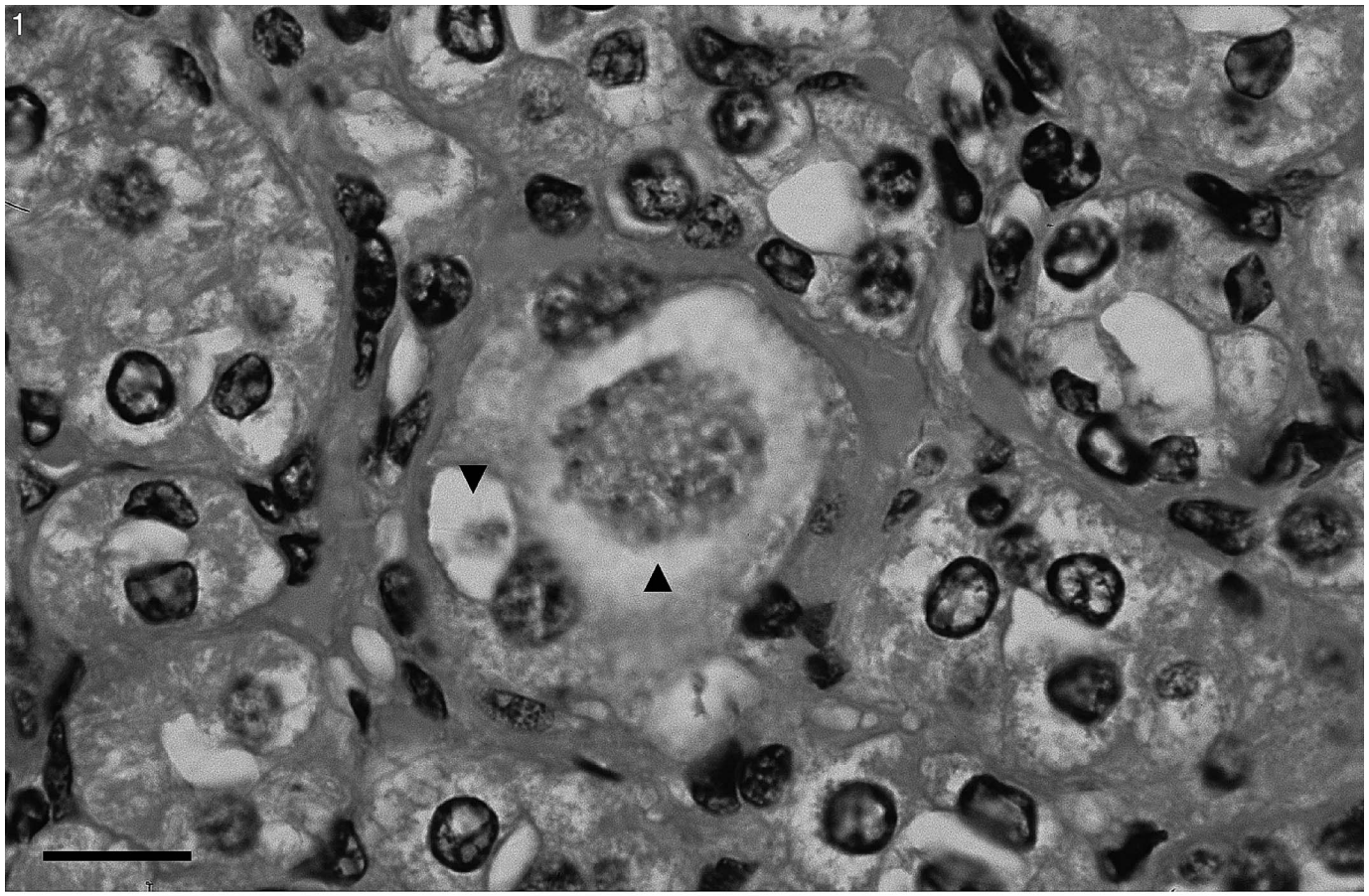
Klossiella quimrensis life cycle stages contained within parasitophorous vacuoles were not associated with any adjacent host inflammatory response. However, kidneys containing *K. quimrensis* were affected by mild multifocal chronic interstitial lymphohistiocytic nephritis.

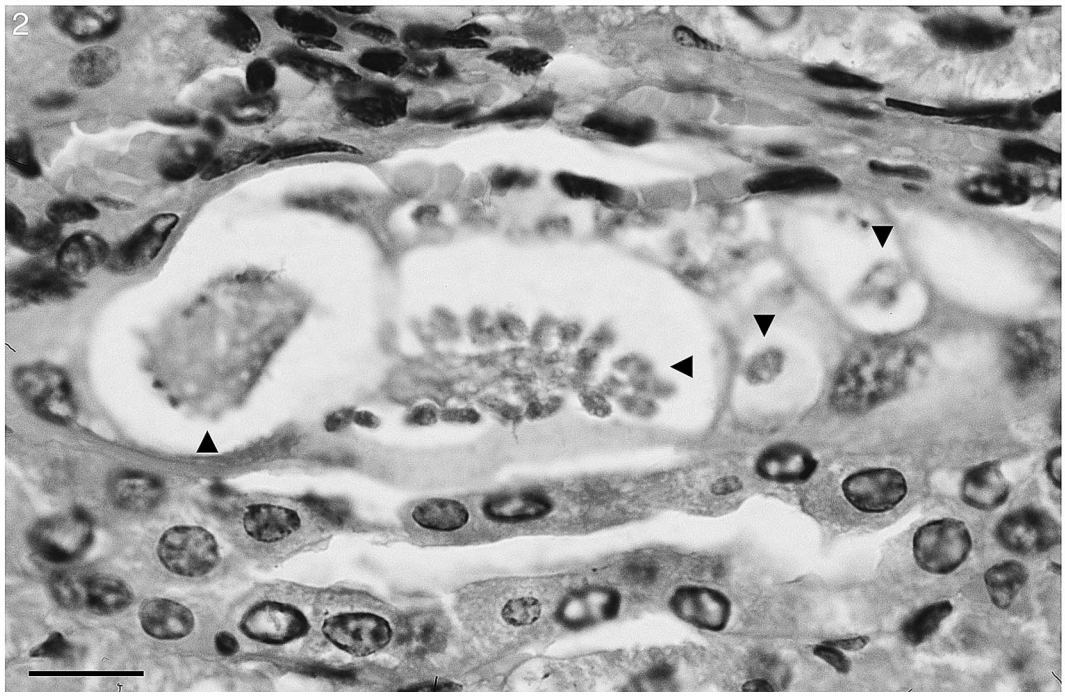
Renal tissue slides, demonstrating klossiellosis in *P. bougainville*, are deposited with the Australian Registry of Wildlife Health, Taronga Zoo, Mosman, NSW, Australia, ARWH 5055.2.

DISCUSSION

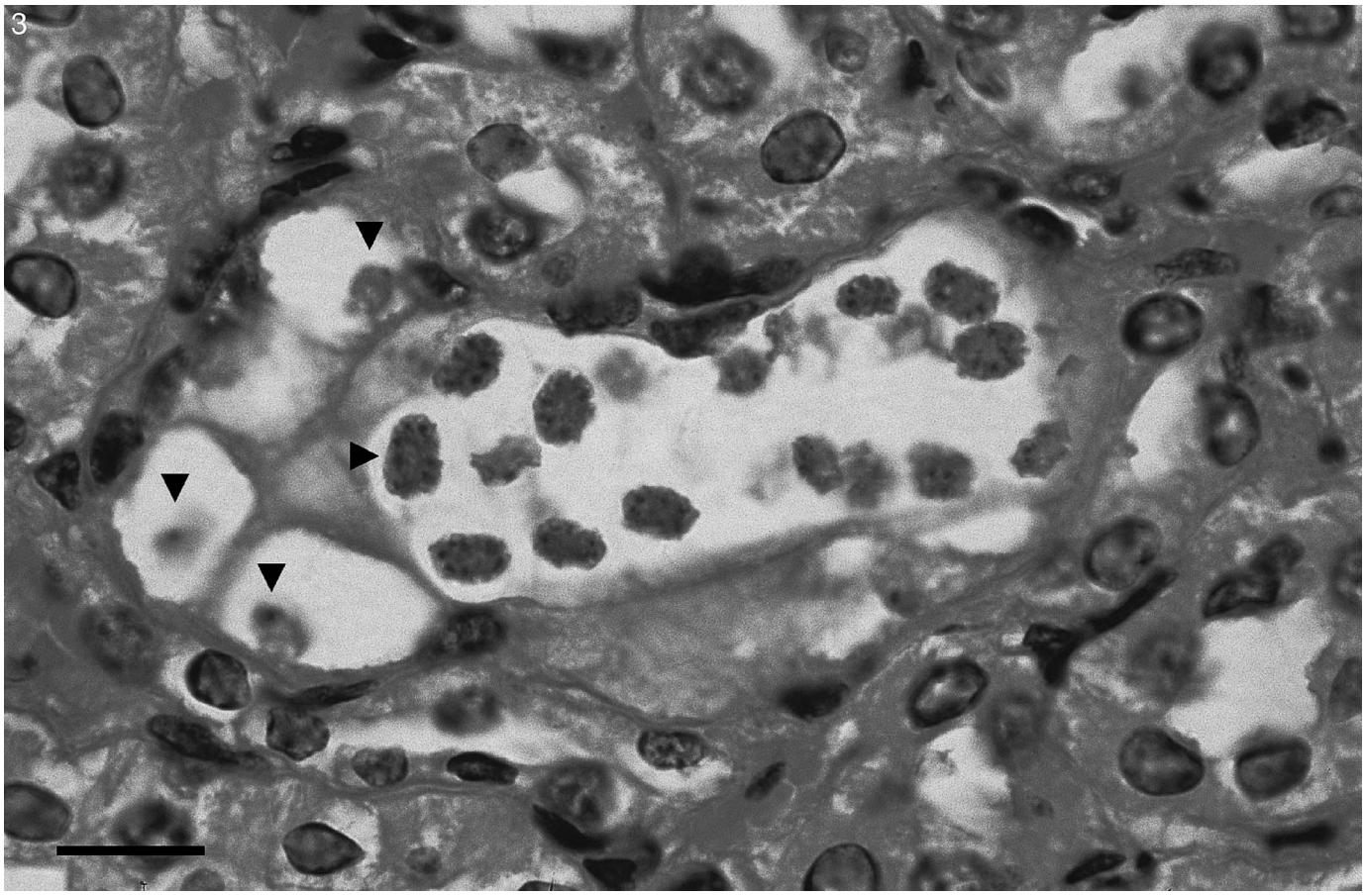
Eleven of the 18 named *Klossiella* spp. are found in marsupials, 10 of which occur in Australian marsupials (Barker et al., 1975, 1984). The 7 remaining named species occur in rodents, equids, bats, and the *Boa constrictor* (Levine, 1988). An unnamed *Klossiella* sp. has been reported in primates (Schoeb, 1984) and another in an owl (Helmboldt, 1967).

Received 2 March 2006; revised 4 August 2006; accepted 15 August 2006.
* Department of Environment and Conservation, PO Box 525 Albany, Western Australia, Australia, 6331.





FIGURES 1–3. Photomicrographs of *K. quimrensis* life cycle stages in renal tissue from *P. bougainville* in Western Australia. Bar = 15 μm. (**1**) Early sporont (▲) demonstrating the circumferential disposition of the nuclei and a macrogamete (▼) contained within parasitophorous vacuoles. (**2**) Macrogametes (▼), an early sporont (▲), and a late budding sporont (◀) within parasitophorous vacuoles. (**3**) Free mature sporoblasts (▶) and macrogametes (▼) within parasitophorous vacuoles.



FIGURES 1–3. Continued.

Our observations of *Klossiella* sp. in western barred bandicoots are closely comparable to the description given by Barker et al. (1975) for *K. quimrensis* in *P. gunnii* and *I. obesulus* (Table I). *Perameles bougainville* once occurred across much of arid southern mainland Australia prior to the recent severe decline in its range, attributed to habitat destruction and introduced species, particularly introduced predators (Friend, 1990). Therefore, *P. bougainville* may formerly have been sympatric with *I. obesulus* in New South Wales, Victoria, South Australia, and Western Australia (Ashby et al., 1990; Friend, 1990; Kemper, 1990; Menkhorst and Seebeck, 1990). The similarities in life cycle stage morphology and possible formerly overlapping natural range led us to propose that *K. quimrensis* be considered the etiologic agent of renal coccidiosis in *P. bougainville*.

TABLE I. Comparison of *K. quimrensis* life cycle stages in *P. bougainville* from Western Australia and observations by Barker et al. (1975) of *K. quimrensis* from *I. obesulus*.

	P. bougainville	*I. obesulus*
Macrogametes (μm diam.)	5.5	7
Macrogamete parasitophorous vacuoles (μm diam.)	<23	<28
Sporonts (μm diam.)	25	30
Number of sporont nuclei	12–16	ND*
Number of sporoblasts	7–24	>20
Sporoblast dimensions (μm)	5–7.5 × 7.5–10	3–5.5 × 7–11
Microgametes (μm diam.)	ND*	1.5
Urinary sporocyst dimensions (μm)	11 × 17	ND*

* ND = No data.

The complete life cycle of *K. quimrensis* remains unknown. Most studies investigating the life cycle, epidemiology, and pathology of *Klossiella* spp. have involved *K. muris* and have been reviewed (Taylor et al., 1979).

There is no reliable ante-mortem test available to diagnose renal coccidiosis in *P. bougainville*. Sporocysts of *Klossiella* spp. are detected in urine infrequently and may be refractory to traditional laboratory concentration methods (Reppas and Collins, 1995). Therefore, microscopic examination of urine cannot be relied upon to diagnose klossiellosis, nor can mild-to-moderate renal coccidiosis be detected by macroscopic inspection of kidneys. Histology of multiple sections of kidney tissue remains the gold standard for definitive diagnosis of klossiellosis (Taylor et al., 1979).

We have not yet encountered a clinical case of klossiellosis in *P. bougainville*. All *K. quimrensis*–infected kidney sections examined demonstrated mild to focally moderate parasite burdens that would not, of themselves, severely compromise renal function. There was no inflammatory response directly associated with intracellular life cycle stages of *K. quimrensis* in *P. bougainville*; however, mild multifocal chronic interstitial lym-

phohistiocytic nephritis was evident in the infected kidneys. We consider klossiellosis to be an incidental finding in this host.

ACKNOWLEDGMENTS

This work is funded by the Australian Research Council in partnership with Murdoch University and the Western Australian Department of Environment and Conservation (DEC) under Linkage Project LP0455050. We gratefully acknowledge Science Division and Narrogin DEC officers and the Kanyana Wildlife Rehabilitation Centre for establishing, monitoring, and maintaining captive colonies of western barred bandicoots at Kanyana and Dryandra Woodland. We are indebted to Michael Slaven and Gerard Spoelstra for production of the histopathology slides.

LITERATURE CITED

ASHBY, E., D. LUNNEY, J. ROBERTSHAW, AND R. HARDEN. 1990. Distribution and status of bandicoots in New South Wales. *In* Bandicoots and bilbies, J. H. Seebeck, P. R. Brown, R. L. Wallis, and C. M. Kemper (eds.). Surrey Beatty and Sons, Sydney, Australia, p. 43–50.

BARKER, I. K., B. L. MUNDAY, AND K. E. HARRIGAN. 1975. *Klossiella* spp. in the kidneys of peramelid, petaurid and macropodid marsupials. Zeitschrift für Parasitenkunde **46:** 35–41.

———, ———, AND W. J. HARTLEY. 1984. *Klossiella* (Apicomplexa, Klossiellidae) in petaurid and macropodid marsupials in Australia. Journal of Protozoology **32:** 520–522.

FRIEND, J. A. 1990. The distribution and status of bandicoots in Western Australia. *In* Bandicoots and bilbies, J. H. Seebeck, P. R. Brown, R. L. Wallis, and C. M. Kemper (eds.). Surrey Beatty and Sons, Sydney, Australia, p. 73–84.

GARDINER, C. H., R. FAYER, AND J. P. DUBEY. 1998. Klossiella. *In* An atlas of protozoan parasites in animal tissues, 2nd edition. Armed Forces Institute of Pathology, Washington, D.C., p. 61–62.

HELMBOLDT, C. F. 1967. An unidentified protozoan parasite in the kidney of the Great-Horned Owl (*Bubo virginianus*). Bulletin of the Wildlife Disease Association **3:** 23–25.

KEMPER, C. 1990. Status of bandicoots in South Australia. *In* Bandicoots and bilbies, J. H. Seebeck, P. R. Brown, R. L. Wallis, and C. M. Kemper (eds.). Surrey Beatty and Sons, Sydney, Australia, p. 67–72.

LEVINE, N. D. 1988. The coccidia: Adeleinorina. *In* The protozoan phylum Apicomplexa. CRC Press Inc., Boca Raton, Florida, p. 133–134.

MENKHORST, P. W., AND J. H. SEEBECK. 1990. Distribution and conservation status of bandicoots in Victoria. *In* Bandicoots and bilbies, J. H. Seebeck, P. R. Brown, R. L. Wallis and C. M. Kemper (eds.). Surrey Beatty and Sons, Sydney, Australia, p. 51–60.

REPPAS, G. P., AND G. H. COLLINS. 1995. *Klossiella equi* infection in horses: Sporocyst stage identified in urine. Australian Veterinary Journal **72:** 316–318.

SCHOEB, T. R. 1984. *Klossiella* sp. in a galago. Journal of the American Veterinary Medical Association **185:** 1381–1382.

TAYLOR, J. L., J. E. WAGNER, D. F. KUSEWITT, AND P. C. MANN. 1979. *Klossiella* parasites of animals: A literature review. Veterinary Parasitology **5:** 137–144.