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Renal Coccidiosis with Cystic Tubular Dilatation in Four Bats

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Abstract. Renal coccidiosis was diagnosed in four bats of different species (*Pipistrellus pipistrellus, Myotis mystacinus, M. nattereri,* and *Nyctalus noctula*). Multiple white and partly indented foci up to 2 mm in diameter were visible on the renal surface. Histologically, the foci appeared as cystic dilated tubules with proliferated epithelium. Asexual and sexual coccidian stages were seen in the epithelial cells, and the extremely distended tubular lumina were filled with schizonts, free zoites, microgamonts, macrogamonts, and unsporulated oocysts. Because the majority of the renal tissue appeared uninvolved in the disease process at the gross and histologic levels and there was no evidence for uremia in other organs, renal function was probably not impaired. Precise classification of the coccidia was impossible because no sporulated oocysts were available. The parasite morphology and the hitherto unreported cystic dilatation of infected tubules containing all developmental stages differ from renal coccidioses reported previously and therefore suggest an undescribed coccidian species.

Key words: Bats; renal coccidiosis.

Renal coccidiosis has been reported in a wide variety of animals, including mammals, birds, and amphibians. Numerous species from different coccidian genera are known to infect the kidney as their main target organ. Coccidia of the genus *Eimeria* have been found, e.g., in the kidneys of waterfowl.[3,7] *Klossiella* spp. have been described in mice,[10] equids,[5] guinea pigs,[8] the Australian water rat (*Hydromys chrysogaster*)[9] and two bats (*Myotis sodalis*).[2] *Isospora* spp. have been reported in the kidneys of frogs.[6] In the renal coccidioses described previously, parasitic stages developed in glomeruli or tubular epithelium with no or only mild dilatation of the tubules. We describe renal coccidiosis with cystic tubular dilatation in four insectivorous bats belonging to four different species. The infection pattern in the kidneys differs completely from that of renal coccidioses reported previously, and therefore a hitherto unknown form of renal coccidiosis may occur in bats.

Fourteen bats belonging to six different insectivorous species were found dead and were submitted for necropsy examination to the Institute of Pathology, Hannover School of Veterinary Medicine, Hannover, Germany, between July 1993 and August 1994. In four of these bats (No. 1: *Pipistrellus pipistrellus,* male, juvenile, 5 g; No. 2: *Myotis mystacinus,* female, juvenile, 4 g; No. 3: *Myotis nattereri,* male, adult, 8 g; No. 4: *Nyctalus noctula,* female, adult, 23 g), severe renal coccidiosis was diagnosed. Macroscopically, the external surfaces of the kidneys had one to numerous irregularly distributed and partly indented white foci up to 2 mm in diameter. In kidney sections, these areas were identified as cystic spaces. All organs of the bats were fixed by immersion in 10% formaldehyde, embedded in paraffin, cut at 4 μm, and stained with hematoxylin and eosin (HE). After light microscopic examination, several selected areas on the formalin-fixed renal tissue sections were prepared for electron microscopy by the detachment technique.[1]

Light microscopic examination revealed numerous cystic dilatations of the tubules in the renal medulla and cortex (Fig. 1) ranging from 0.1 to 2 mm in diameter. Most of these

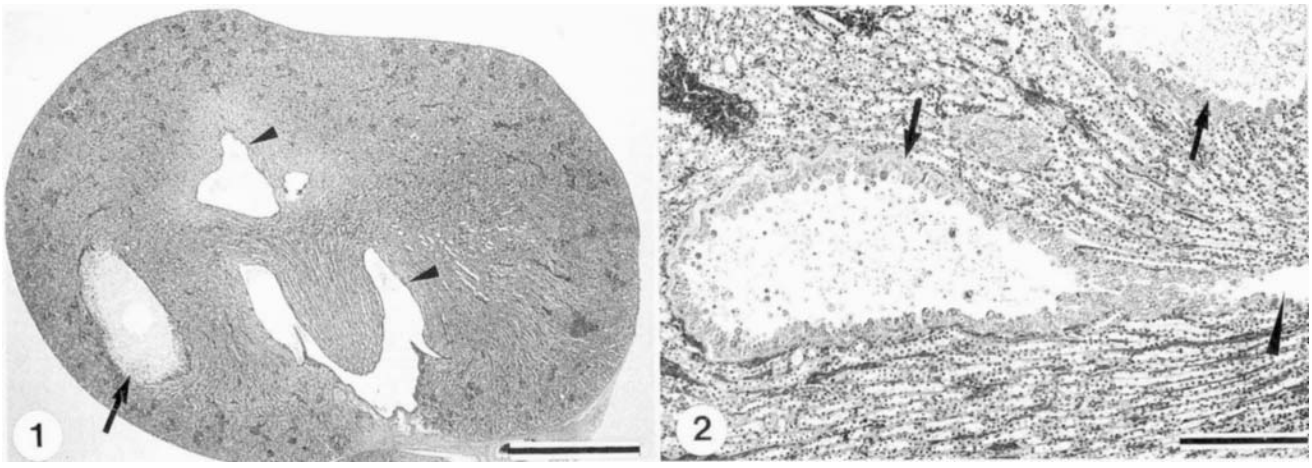

Fig. 1. Kidney; bat No. 3, with coccidiosis. Note cystic dilatation of a tubule (arrow). (arrowheads = renal pelvis). HE. Bar = 1 mm.

Fig. 2. Kidney; bat No. 3, with coccidiosis. Note cone-shaped dilated tubule without inflammatory reaction. The proliferated epithelium and the cystic tubular lumen (arrows) contain numerous parasitic stages. No parasites or proliferation are present in the epithelium of the collecting ducts beyond the infected tubule (arrowhead). HE. Bar = 0.25 mm.

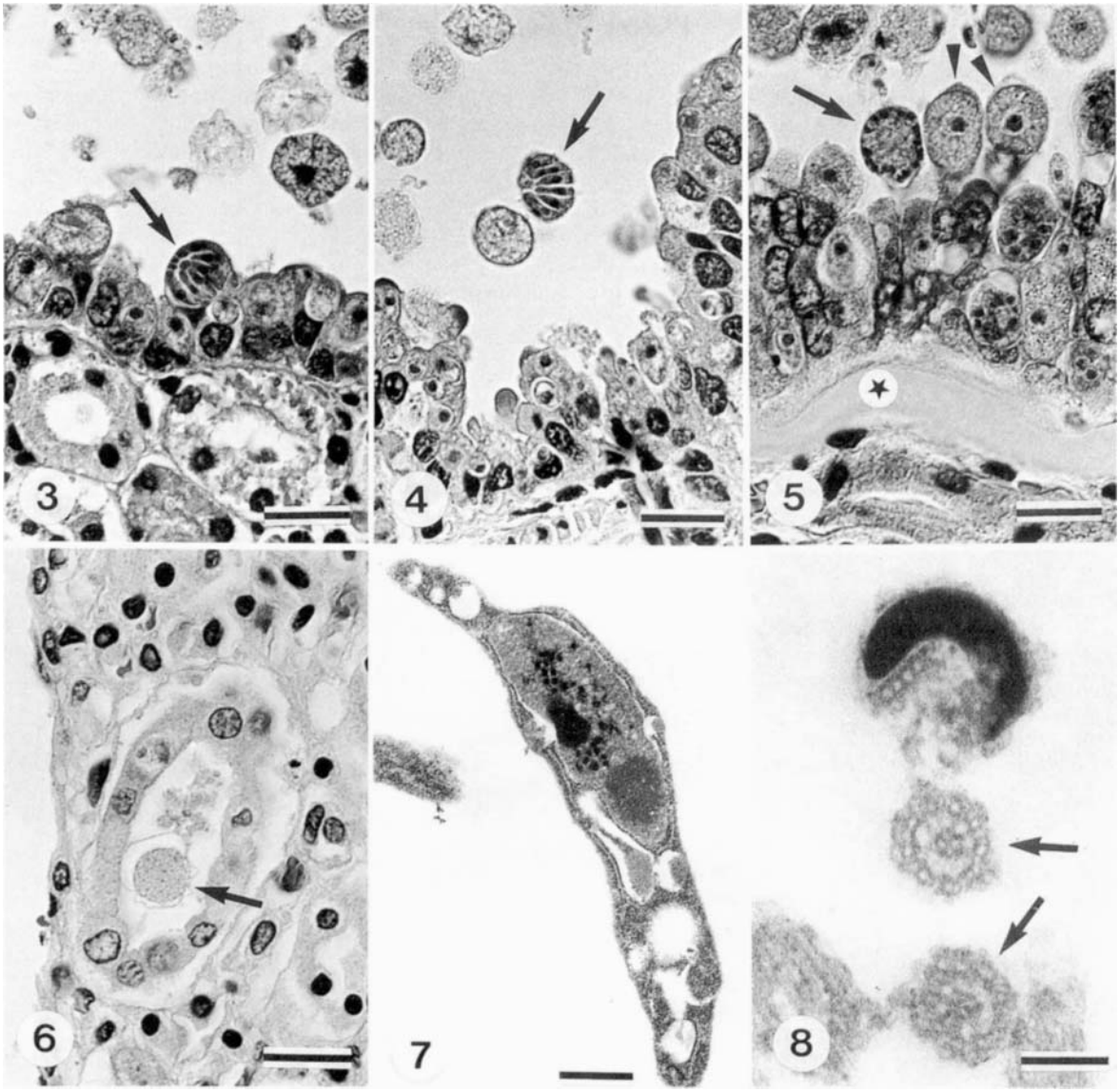

Fig. 3. Kidney; bat No. 1, with coccidiosis. Note schizont (arrow) budding from a tubular epithelial cell. HE. Bar = 20 μm.

Fig. 4. Kidney; bat No. 4, with coccidiosis. Note schizont (arrow) free in the lumen of a dilated tubule. HE. Bar = 20 μm.

Fig. 5. Kidney; bat No. 2, with coccidiosis. Note microgamont (arrow) with no nucleus and macrogamonts (arrowheads) released from tubular epithelial cells. The proliferated tubular epithelium is demarcated from the adjacent parenchyma by a thickened basement membrane (*). HE. Bar = 20 μm.

Fig. 6. Kidney; bat No. 1, with coccidiosis. Note suggested unsporulated oocyst (arrow) in a collecting duct near the renal pelvis. HE. Bar = 20 μm.

Fig. 7. Electron micrograph. Kidney; bat No. 2, with coccidiosis. Note free zoite without refractile bodies (merozoite). Formalin-fixed, paraffin-embedded tissue processed with the detachment technique.[1] Bar = 1 μm.

Fig. 8. Electron micrograph. Kidney; bat No. 2, with coccidiosis. Note Microgamete with flagellae (arrows). Formalin-fixed, paraffin-embedded tissue processed with the detachment technique.[1] Bar = 0.1 μm.

dilatations were located deep in the renal cortex or medulla, and only a few were in contact with the capsule and were accompanied by slight indentation of the renal surface, which was attributed to a partial drainage of these cysts. The cystic dilated tubules were almost completely filled with both asexual and sexual developmental stages of coccidia. Only a few contained solely necrotic, amorphous, and partly calcified material. The wall of the cystic dilatation was composed of one to three layers of an epithelium resembling tubular epithelium with proliferated tall and irregularly shaped cells (Figs. 2–5). Their apices were severely distended by tiers of developing parasites. In two bats (Nos. 2, 3), the dilated

tubules were surrounded by an eosinophilic, hyaline, periodic acid–Schiff-positive membrane up to 20 μm thick (Fig. 5), which was identified by electron microscopy to be the thickened tubular basement membrane characterized as a marked increase of an amorphous matrix in the lamina densa. Coronal sectioning of the tissues revealed that the lumina of some cone-shaped cysts ended in the collecting ducts of the renal medulla (Fig. 2). Dilatation of the tubules, proliferation of the tubular epithelium, and the occurrence of parasitic stages decreased in deeper areas of the renal medulla. Minimal to no proliferation of tubular epithelium and single unsporulated oocysts were identified in the lumina of collecting ducts near the renal papilla (Fig. 6). Glomeruli and epithelium of the renal pelvis were not affected. Except for moderate compression of the adjacent tubules and glomeruli, no changes occurred around the cystic cavities that contained intact parasites. A few necrotic and partly calcified cysts were surrounded by thin fibrous walls, macrophages, lymphocytes, and a few plasma cells.

Both asexual and sexual developmental stages were seen in the tubular epithelium and lumina. Schizogony, gamogony, and unsporulated oocysts were seen as developmental stages. Schizonts (13–19 μm in diameter; Figs. 3, 4), macrogamonts (12–18 μm in diameter; Fig. 5), and microgamonts (12–15 μm in diameter; Fig. 5) were released into the cystic tubular lumina from superficial epithelial cells. With the electron microscope, the macrogametes were identified by their characteristic peripherally located osmiophilic, electron-dense wall-forming bodies that surrounded lipid bodies and polysaccharide granules.[4] Fragments of schizonts and microgamonts were seen with release of numerous merozoites (1.5–2.3 × 8–10 μm; Fig. 7) or microgametes (0.2–0.4 × 2.0–3.5 μm; Fig. 8), respectively, into the tubular lumina. Microgametes were associated with two flagellae each (Fig. 8). Schizonts (Figs. 3, 4) contained 16–22 banana-shaped merozoites, as seen with the electron microscope. No refractile bodies were seen in the free zoites (1.5–2.3 × 7–10 μm; Fig. 7), suggesting that only merozoites and no sporozoites were present in the dilated tubular lumina.[4] Only a few structures suggestive of unsporulated oocysts (11–17 μm in diameter), characteristically missing wall-forming bodies when viewed with the electron microscope[4] were found in distal tubules and collecting ducts (Fig. 6).

It is not known whether this renal coccidian was pathogenic to the bats or merely a commensal organism. Fat reserves and body conditions were good in all four bats, and only a few intestinal parasites (trematodes, cestodes, nematodes) were found in bat Nos. 3 and 4. However, except for bat No. 2 that died of septicemia after accidental wound infection, the cause of death remains unclear in these bats. The cystic tubules occupied no more than 25% of the kidney volume, and no indication for uremia was seen in other organs. Thus, renal failure seemed to be unlikely. In a study of 270 guinea pigs with renal coccidiosis caused by *Klossiella cobayae*, the authors determined that fatal uremia in guinea pigs only occurs in animals with massive renal involvement.[8]

The infection pattern and histomorphologic features in the four bats were distinctly different from those of renal coccidioses in bats and other animal species described previously. Marked cystic dilatation of the infected tubules has not been reported in other renal coccidioses, in which tubules were not (*Klossiella* sp., *Isospora* sp.)[2,5,6,8–10] or were only mildly (*Eimeria* sp.)[3,7] distended. In the one previous report on renal coccidiosis in bats (*Myotis sodalis*),[2] only parasitic stages of sporogony without schizogony or gametogony were described. In addition, the infected tubules were normal or only mildly extended, without the cystic tubular dilatation seen in the bats in the present study and no statements were made on gross renal lesions or parasite life cycle.[2] Simultaneous infection of a tubular segment with both asexual and sexual developmental stages as seen in these bats is a rare finding in renal coccidiosis and has only been reported in *Eimeria* infections.[3] In renal coccidiosis associated with *Klossiella* sp. or *Isospora* sp., different parasitic stages are restricted to specific segments, e.g., the glomeruli, tubular epithelium, or collecting ducts.[2,5,6,8,9] No urine was collected from these bats, so precise classification of the parasites was impossible because unsporulated oocysts were not available.[4] Thus, no statements can be made concerning the life cycle of this parasite. Nevertheless, excretion of oocysts via the urine with contamination of the bats environment seems plausible.

The mechanism for tubular dilatation remains speculative, but proliferation of the epithelium as a sequel to parasitic infestation and subsequent distension of the lumina is conceivable. Obstruction of the distal tubules seems unlikely because it would impair the release of mature parasitic stages, which were noted in distal tubules and collecting ducts.

The consistent morphology of the parasite and the severely cystic dilated renal tubules containing asexual and sexual developmental stages are features distinctly different from those of previously reported renal coccidioses in bats and other animal species. It appears that these four bats were infected with a hitherto undescribed renal coccidian species.

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