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A new species of *Eimeria* (Apicomplexa: Eimeriidae) from tri-colored bats, *Perimyotis subflavus* (Chiroptera: Vespertilionidae), from the Ouachitas of Arkansas

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Abstract

Between February 2013 and October 2013, eleven tri-colored bats, *Perimyotis subflavus* were collected from Marion, Polk, and Searcy counties, Arkansas, and their faeces examined for coccidian parasites. Two of eleven (18%) harboured an eimerian that we describe here as new. Oocysts of *Eimeria mcdanieli* sp. n. were ellipsoidal to elongate with a bi-layered wall and measured (length $\times$ width, L $\times$ W) 28.3 $\times$ 17.9 μ m, with an L/W ratio of 1.6. A micropyle and oocyst residuum were absent but a single polar granule was present. Sporocysts were ovoidal, 12.6 $\times$ 8.3 μ m, with an L/W ratio of 1.5. A pronounced, nipple-like Stieda body was present as well as a substieda body. A sporocyst residuum was present as dispersed bubble-like granules. This is the third coccidian described from tri-colored bats and the sixth species reported from Arkansas chiropterans. In addition, both infected bats harbored a concurrent infection of *Eimeria heidti* McAllister, Burt, Seville, and Robison, 2011.

Keywords

Apicomplexa; Coccidia; taxonomy; oocysts; Chiroptera; Vespertilionidae; *Eimeria heidti*; *Eimeria mcdanieli*; Arkansas; *Perimyotis subflavus*

Introduction

The tri-colored bat, *Perimyotis subflavus* (F. Cuvier) is a small bat with dark brown to blackish wing membranes that ranges from southeastern Minnesota and Nova Scotia south to eastern Texas and northern Florida, then further southward to eastern Mexico and northern Honduras (Reid 2006). In Arkansas, *P. subflavus* occurs statewide in woodland or mixed farmland habitat in treetops, crevices of cliffs, in caves and mines, and occasionally buildings; it is the most abundant chiropteran in the state (Sealander and Heidt 1990).

Two eimeriid coccidians have been described previously from *P. subflavus*, *Eimeria macyi* Wheat from bats in Alabama and *Eimeria heidti* McAllister, Burt, Seville, and Robison from Arkansas specimens (Wheat 1975; McAllister et al. 2011). Herein, we provide a description of a third species of *Eimeria* from *P. subflavus* and report *E. heidti* for the second time from *P. subflavus*.

Materials and Methods

Between February 2013 and October 2013, 11 adult *P. subflavus* were collected by hand from Marion ($n = 1$), Polk ($n = 5$) and Searcy ($n = 5$) counties, Arkansas, and examined for coccidia. Fresh faecal samples were placed in individual vials containing 2.5% (w/v) aqueous potassium dichromate ($K_2Cr_2O_7$). Samples were examined for coccidia by light microscopy after flotation in Sheather's sugar solution (specific gravity = 1.30). Measurements were taken on 10 sporulated oocysts from a single bat using a calibrated ocular micrometer and reported in micrometres (μm) with means followed by the ranges in parentheses; photographs were taken using Nomarski interference-contrast optics. Oocysts were ~125 days old when measured and photographed. Descriptions of oocysts and sporocysts follow guidelines of Wilber et al. (1998) as follows: oocyst length (L) and width (W), their ranges and ratios (L/W), micropyle (M), oocyst residuum (OR), polar granule(s) (PG), sporocyst length (L) and width (W), their ranges and ratio (L/W), sporocyst (SP), Stieda body (SB), substieda body (SSB), parastieda body (PSB), sporocyst residuum (SR), sporozoites (SZ) anterior (ARB) and posterior (PRB) refractile bodies, and nucleus (N). A host voucher was accessioned into the Henderson State University Collection (HSU), Arkadelphia, Arkansas, USA. Photosyntypes of sporulated oocysts were accessioned into the United States National Parasite Collection (USNPC), Beltsville, Maryland, USA.

Results

Two of 11 (18%) *P. subflavus* were found to be passing coccidian oocysts that we describe as new. In addition, both bats were concurrently infected with *Eimeria heidti*.

Eimeria mcdanieli sp. nov. (Figs. 1–4)

Description of sporulated oocyst: Oocyst with 4 sporocysts; shape ellipsoidal—elongate; bi-layered wall, colourless, ~ 1.6 thick, heavily textured outer layer ~1.0 thick, smooth inner layer ~0.6 thick; L $\times$ W: 28.3 $\times$ 17.9 (25–30 $\times$ 16–20); L/W: 1.6 (1.5–1.9); M, OR, both absent; PG: one present.

Description of sporocyst and sporozoites: SP ovoidal, with a smooth uni-layered wall, ~ 0.5 thick; L $\times$ W: 12.6 $\times$ 8.3 (11–14 $\times$ 8–9); L/W: 1.5 (1.3–1.6); SB pronounced, nipple-like, SSB body present, PSB body absent; SR: bubble-like, composed of large-sized granules in a compact mass or dispersed along perimeter of SP; SZ: (not measured) sausage-shaped with subspheroidal ARB and elongate PRB; single N slightly posterior to midpoint.

Taxonomic summary

Type host: Tri-colored bat, *Perimyotis subflavus* (F. Cuvier, 1832) (Chiroptera: Vespertilionidae). Collected 19 October 2013.

Type specimens: Symbiotype host in the Henderson State University Collection (HSU No. 702). Photosyntype of sporulated oocyst deposited in the USNPC as No. 108163.

Type locality: Pipistrelle Mine, 17.8 km SSE of Mena off county road 80, Ouachita National Forest, Polk County, Arkansas, USA (34.42163°N, 94.20639°W, elev. 303 m).

Prevalence: In two of 11 (18%) of the type host; two of five (40%) Polk County.

Sporulation: Exogenous. All oocysts were passed unsporulated or partially sporulated and became fully sporulated within five days at ~23 C.

Prepatent and patent periods: Unknown.

Site of infection: Unknown, oocysts recovered from faeces.

Etymology: The specific epithet is given in honour of Dr. Van Rick McDaniel (1945–2009), for his work on Arkansas bats and other mammals. Rick also served as CTM's M.S. advisor at Arkansas State University.

Remarks: When the new species is compared to other eimerians described from *P. subflavus* (*E. macyi* and *E. heidti*), there are very few morphological similarities. Both are considerably smaller (19×18 and 26.1×20.5 μm , respectively) and neither have the pronounced ellipsoidal to elongate oocyst shape of *E. mcdanieli*. Other eimerians from bats from Arkansas include: *Eimeria catronensis* Scott and Duszynski from northern myotis, *Myotis septentrionalis*, and *Eimeria dowleri* McAllister and Upton and *Eimeria sealanderi* McAllister and Upton from eastern red bats, *Lasiurus borealis* (McAllister et al. 2004; McAllister and Upton 2009). The former possesses a micropyle (see McAllister et al. 2012) not found in the new species while the latter two possesses subspheroidal to ellipsoidal oocysts that are considerably smaller (24.7×22.0 and 16.7×14.4 μm , respectively) than the new species. Further comparison of the new species with other bat eimerians (see Duszynski 2002, Figs. 1–44) show none with the elongate oocyst shape that results in an increased range of L/W ratios (up to 1.9) characteristic of the new species.

We also found that both bats were concurrently infected with *Eimeria heidti* McAllister, Burt, Seville, and Robison, 2011 (Figs. 5–6). Oocysts ($n = 11$) were ovoidal and measured 24.5×18.9 ($22\text{--}27 \times 17\text{--}21$) μm with an L/W of 1.3 (1.1–1.4). These measurements are well within the ranges reported for oocysts by McAllister et al. (2011) and all other morphological characters mirrored those of *E. heidti*.

Discussion

Duszynski (2000) provided a summation on the coccidia of bats of the world. Surprisingly, only 27 of 925 (3%) bat species have been reported to serve as hosts for 31 named coccidian species. Since then, using molecular methodologies, the number of species of bats has risen to over 1,200 (Simmons 2010), yet only a few additional coccidians have been reported from these mammals. In addition, it is not yet known how bats become infected with coccidian oocysts. Duszynski (2000) speculated that *Eimeria* spp. of aerial insectivorous vespertilionid bats (like those from *P. subflavus*), might utilize intermediate or transport

hosts to bridge the food gap for these aerial feeders. However, future studies will be necessary to help answer this and other questions given the paucity of current knowledge of the coccidia of chiropterans.

In Arkansas, six species of *Eimeria* have now been reported from three species of Arkansas bats (Table 1). Prevalence of infection with these coccidians from bats of Arkansas tends to be rather low (~ 20%) except for *E. doweri* from a limited sample size. McAllister et al. (2011, 2012) recently noted that it was difficult to obtain fecal samples from bats in Arkansas because of restrictions on collections due to fear of spreading white-nose syndrome, caused by the fungus *Pseudogymnoascus destructans* (Frick et al. 2010). However, now that bats in the state have recently been found to harbor this fungus (<http://www.whitenosesyndrome.org>), the USDA Forest Service has placed a complete moratorium on bat collections from mines and caves with private landowners being instructed to prohibit access to their caves. In addition, one bat (*M. septentrionalis*) reported to harbor coccidia in both Arkansas and Oklahoma (McAllister et al. 2004, 2012) is proposed to be placed on the endangered species list (Federal Register 2013). Therefore, the study of bat coccidians in Arkansas, and elsewhere in the United States where white-nose syndrome is a concern, will be seriously affected and we expect only select species to be examined in the near future. Yet, the potential for new species of coccidia to be discovered in bats highlights the need to continue research on bats.

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References

- Duszynski DW. Coccidia (Apicomplexa: Eimeriidae) of the mammalian order Chiroptera. Special Publications of the Museum of Southwestern Biology. 2002; 5:1–45.
- Federal Register. Endangered and threatened wildlife and plants; 12-month finding on petition to list the eastern small-footed bat and the northern long-eared bat as endangered or threatened species; listing the northern long-eared bat as an endangered species; proposed rule. Federal Register. 2013; 78(191):61046–610802.
- Frick WF, Pollock JF, Hicks AC, Langwig KE, Reynolds DS, Turner GR, Butchoski CM, Kunz TH. An emerging disease causes regional population collapse of a common North American bat species. Science. 2010; 329:679–682. [PubMed: 20689016]
- McAllister CT, Burt S, Seville RS, Robison HW. A new species of *Eimeria* (Apicomplexa: Eimeriidae) from the eastern pipistrelle, *Perimyotis subflavus* (Chiroptera: Vespertilionidae), in Arkansas. Journal of Parasitology. 2011; 97:896–898. [PubMed: 21506799]
- McAllister CT, Seville RS, Roehrs ZP. A new species of *Eimeria* (Apicomplexa: Eimeriidae) from the northern myotis, *Myotis septentrionalis* (Chiroptera: Vespertilionidae), in Oklahoma. Journal of Parasitology. 2012; 98:1003–1005. [PubMed: 22509940]
- McAllister CT, Upton SJ. Two new species of *Eimeria* (Apicomplexa: Eimeriidae) from eastern red bats, *Lasiurus borealis* (Chiroptera: Vespertilionidae), in Arkansas and North Carolina. Journal of Parasitology. 2009; 95:991–993. [PubMed: 20050004]
- McAllister CT, Upton SJ, Bursey CR. Parasites (Coccidia, Trematoda, Nematoda) from selected bats of Arkansas. Journal of the Arkansas Academy of Science. 2004; 58:133–136.

- McAllister CT, Upton SJ, Trauth SE, Allard DW. A redescription of *Eimeria macyi* (Apicomplexa: Eimeriidae) from the eastern pipistrelle, *Pipistrellus subflavus* (Mammalia: Chiroptera), from Arkansas. *Journal of the Arkansas Academy of Science*. 2001; 55:181–183.
- Reid, FA. A field guide to mammals of North America. New York: Houghton Mifflin; 2006. p. 579
- Sealander, JA.; Heidt, GA. Arkansas mammals: Their natural history, classification, and distribution. Fayetteville, Arkansas: University of Arkansas Press; 1990. p. 308
- Simmons, N. Estimating diversity: How many species of bats are there?. 15th Annual International Bat Research Conference; 23–27 August 2010; Prague, Czech Republic. 2010.
- Wheat BE. *Eimeria macyi* sp. N. (Protozoa: Eimeriidae) from the eastern pipistrelle, *Pipistrellus subflavus* from Alabama. *Journal of Parasitology*. 1975; 61:920–922. [PubMed: 1185436]
- Wilber PG, Duszynski DW, Upton SJ, Seville RS, Corliss JO. A revision of the taxonomy and nomenclature of the *Eimeria* spp. (Apicomplexa: Eimeriidae) from rodents in the Tribe Marmotini (Sciuridae). *Systematic Parasitology*. 1998; 39:113–135.

Figs. 1–3.

Nomarski interference-contrast photomicrographs of oocysts of *Eimeria mcdanieli* n. sp.

Abbreviations: oocyst wall (OW), Stieda body (SB), substieda body (SSB). Scale bars = 10 μ m for all figures.

Fig. 4.
Composite line drawing of oocyst of *Eimeria mcdanieli* n. sp. Scale bar = 10 μ m.

Figs. 5–6.

Nomarski interference-contrast photomicrographs of oocysts of *Eimeria heidi*. Scale bars = 10 µm for both figures.

Table 1

Coccidia reported from Arkansas bats.

Coccidian	Host	Prevalence [*]	Reference
<i>Eimeriacatronensis</i>	<i>Myotis septentrionalis</i>	4/20 (20%)	McAllister et al. (2004)
<i>Eimeriadowleri</i>	<i>Lasiurus borealis</i>	4/5 (80%)	McAllister and Upton (2009)
<i>Eimeriaheidti</i>	<i>Perimyotis subflavus</i>	2/20 (10%)	McAllister et al. (2011)
		2/11 (18%)	This study
<i>Eimeriamacyi</i>	<i>P. subflavus</i>	2/5 (20%)	McAllister et al. (2001)
<i>Eimeriamcdanieli</i>	<i>P. subflavus</i>	2/11 (18%)	This study
<i>Eimeriasealanderi</i>	<i>L. borealis</i>	2/11 (18%)	McAllister and Upton (2009)

* Number infected/number examined (%).