
Coccidian Parasites of Eastern Cottontail, *Sylvilagus floridanus* (Lagomorpha: Leporidae) from Oklahoma, with a Summary of Coccidians (Apicomplexa: Eimeriidae) from Mammals of the State

Chris T. McAllister*

Division of Natural Sciences, Northeast Texas Community College, Mt. Pleasant, TX 75455

John A. Hnida

Department of Microbiology and Immunology, Midwestern University, Glendale, AZ 85308

Henry W. Robison

602 Big Creek Drive, Sherwood, AR 72120

Abstract: A single eastern cottontail, *Sylvilagus floridanus*, collected opportunistically in January 2023 from McCurtain County, Oklahoma, was examined for coccidian parasites. Two coccidians were passed in feces of this host, including *Eimeria neoirresidua* and *Eimeria poudrei*. Ellipsoidal to ovoidal oocysts of *E. neoirresidua* averaged (L × W) 28.6 × 17.5 μm; a polar granule and an oocyst residuum were absent but a micropyle was present. Ellipsoidal sporocysts of *E. neoirresidua* averaged 14.4 by 7.1 μm and a sporocyst residuum was present. Ovoidal to ellipsoidal oocysts of *E. poudrei* averaged 24.9 × 17.1 μm; a polar granule was absent but a micropyle and an oocyst residuum were present. Ellipsoidal to ovoidal sporocysts of *E. poudrei* averaged 13.2 × 6.5 μm and a sporocyst residuum was present. Here, we provide the first report of these two coccidians from *S. floridanus* in Oklahoma, including a summary of the coccidians documented from mammals of the state.

Introduction

Coccidians are apicomplexan parasites that are accountable for an intestinal disease that affects several different vertebrates, including rabbits (Lagomorpha). As such, they are hosts of one of the most common North American protozoan infections.

The eastern cottontail, *Sylvilagus floridanus* (J.A. Allen) has been the subject of several coccidian studies, including harboring 16 species

*Corresponding author: emcallister@ntcc.edu

(Duszynski and Couch 2013). However, we are not aware of previous reports of coccidians from *S. floridanus* in Oklahoma. Here, we document two species of *Eimeria* from a single *S. floridanus* collected from southeastern Oklahoma. In addition, we provide a summary of the coccidians documented from mammals of the state.

Methods

On 30 January 2023, a single adult *S. floridanus* was found freshly dead on the road (DOR) in Broken Bow, McCurtain County.

It was taken to the lab and examined for coccidians. A mid-ventral incision was made to expose the lower gastrointestinal tract and a fresh fecal sample as well as rectal contents was placed in an individual vial containing 2.5% (w/v) aqueous potassium dichromate ($K_2Cr_2O_7$). The sample was further examined for coccidia via flotation in a 15-ml conical centrifuge tube (with centrifugation) containing Sheather's sugar solution (Ricca Chemical Company, Arlington, Texas; specific gravity: 1.25) using an Olympus BX43 light microscope (Olympus Corporation, Center Valley, Pennsylvania). Partially sporulated oocysts were placed in a Petri dish containing a small layer of $K_2Cr_2O_7$ for 48–72 hr to allow complete sporulation. All morphological measurements are reported in micrometers (μm) with the means followed by the ranges in parentheses. Oocysts were ca. 90 days old from the time they were found in host feces to the time when they were measured and photographed using Nomarski interference-contrast optics at $\times 1,000$ magnification. Oocyst and sporocyst descriptions follow the standard guidelines of Wilber et al. (1998) including 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), sub-Stieda body (SSB), para-Stieda body (PSB), sporocyst residuum (SR), sporozoites (SZ) anterior (ARB) and posterior (PRB) refractile bodies, and nucleus (N).

A photovoucher of *S. floridanus* was accessioned into the Eastern Oklahoma State Collection, Idabel, Oklahoma. Photovouchers of sporulated oocysts were accessioned into the Harold W. Manter Laboratory of Parasitology (HWML), Lincoln, Nebraska.

Results and Discussion

Two coccidians were recovered from the fecal sample as follows: *Eimeria neoirresidua* and *Eimeria poudei*. Data are provided on each in an annotated format below.

Eimeria neoirresidua Duszynski and Marquardt, 1969 (Figs. 1A–B)

Description of sporulated oocyst: Oocyst shape (n = 15): ellipsoidal to ovoidal; bilayered wall, ~ 1.3 (1.0–1.5) outer layer smooth (n = 10) or lightly pitted (n = 5), colorless to $\sim 2/3$ total thickness; darker inner layer. L \times W (n = 15): 28.6×17.5 (26–31 \times 16–19); L/W ratio: 1.6 (1.5–1.8); M present, 4.0 (3.0–5.0); OR: and PG both absent.

Description of sporocyst and sporozoites: Sporocyst shape: ellipsoidal; L \times W (n = 15): 14.4×7.1 (16–18 \times 6–8); L/W ratio: 2.0 (1.9–2.3); nipple-like SB present, SSB, PSB: both absent; SR: present; SR characteristics: spheroidal to irregular mass of large granules. Sporozoite shape: elongate with large PRB and small central N.

Taxonomic Summary

Host: Eastern cottontail, *Sylvilagus floridanus* (J. A. Allen, 1890); photovoucher host deposited in the EOSC collection.

New geographic distribution: USA: Oklahoma: McCurtain County, off Hatchery Road, Broken Bow (34.008114°N, -94.756989°W).

Type host and locality: Desert cottontail, *Sylvilagus audubonii* (Baird); USA: Colorado, Larimer County, near Ft. Collins (Duszynski and Marquardt 1969).

Other localities: USA: Pennsylvania (Wiggins et al. 1980); Italy: Province of Alessandria (Bertolino et al. 2010).

Prevalence: 1/1.

Sporulation: Oocysts were passed partially sporulated and completed sporulation in 24–48 hr in $K_2Cr_2O_7$.

Site of infection: Unknown; oocysts passed in feces.

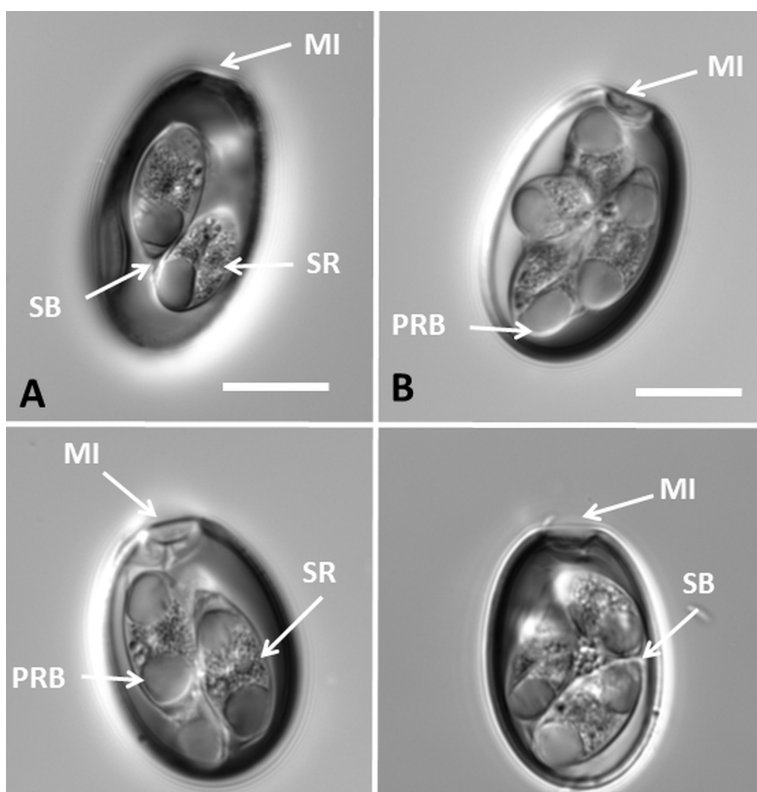


Figure 1. Sporulated oocysts of two coccidians from *Sylvilagus floridanus*. (A–B) *Eimeria neoirresidua*. (C–D) *Eimeria poudrei*. Scale bars = 10 μm . Abbreviations: MI (micropyle); PRB (posterior refractile body); SB (Stieda body); SR (sporocyst residuum).

Materials deposited: Photovoucher of sporulated oocysts are deposited as HWML 217042.

Remarks

Oocysts from the present sample were not significantly different from those described originally by Duszynski and Marquardt (1969) from *S. audubonii* from Colorado and those described by Wiggins et al. (1980) from *S. floridanus* from Pennsylvania. Our oocysts were, on average, slightly greater in length (28.6 vs. 25.7 μm) than those reported by Duszynski and Marquardt (1969) and slightly greater in length and width (28.6 \times 17.5 vs. 25.2 \times 16.8 μm) than those reported by Wiggins et al. (1980). Average L \times W sizes of our sporocysts were nearly identical (14.4 \times 7.1 vs. 14.5 \times 6.4 μm) to those reported by Duszynski and Marquardt (1969) and slightly larger (14.4 \times 7.1 vs. 12.6 \times 6.4 μm) than those reported by Wiggins et al. (1980). All other morphological and mensural

characteristics of the oocysts from the present sample were similar to those previously reported for *E. neoirresidua*.

Eimeria poudrei Duszynski and Marquardt, 1969

(Figs. 1C–D)

Description of sporulated oocyst: Oocyst shape (n = 15): ovoidal to ellipsoidal; bilayered wall, \sim 1.3 (1.3–1.5) outer layer smooth, colorless to \sim 2/3 total thickness; darker inner layer. L \times W (n = 15): 24.9 \times 17.1 (20–30 \times 16–19); L/W ratio: 1.5 (1.3–1.7); M present, 3.9 (3.0–4.5); OR present, irregular to spheroidal mass of medium to large granules; PG absent.

Description of sporocyst and sporozoites: Sporocyst shape: ellipsoidal to ovoidal; L \times W (n = 15): 13.2 \times 6.5 (10–16 \times 6–8); L/W ratio: 2.1 (1.7–2.2); nipple-like SB present, SSB, PSB: both absent; SR: present; SR characteristics:

Irregular mass of medium to large granules. Sporozoite shape: elongate with large PRB and small central N.

Taxonomic Summary

Host: Eastern cottontail, *Sylvilagus floridanus* (L., 1758); photovoucher host deposited in the EOSC collection.

New geographic distribution: USA: Oklahoma: McCurtain County, off Hatchery Road, Broken Bow (34.008114°N, -94.756989°W).

Type host and locality: Desert cottontail, *Sylvilagus audubonii* (Baird); USA: Colorado, Larimer County, near Ft. Collins.

Other localities: Italy: Province of Alessandria (Bertolino et al. 2010).

Prevalence: 1/1.

Sporulation: Oocysts were passed partially sporulated and completed sporulation in 24–48 hr in $K_2Cr_2O_7$.

Site of infection: Unknown; oocysts passed in feces.

Materials deposited: Photovoucher of sporulated oocysts are deposited as HWML 217043.

Remarks

Oocysts from the present sample were not significantly different from those described originally by Duszynski and Marquardt (1969) from *S. audubonii* from Colorado. Average L×W sizes of our oocysts (24.9 × 17.1 vs. 26.0 × 18.1 μm) and sporocysts (13.2 × 6.5 vs. 14.4 × 6.4 μm) were nearly identical to those reported by Duszynski and Marquardt (1969) as well as all other mensural and morphological characteristics fitting *E. poudeiri*.

Interestingly, Bertolino et al. 2010 reported that *S. floridanus* from Italy harbored both *E. poudeiri* (40% prevalence), and *E. neoirresidua* (49% prevalence). They found a total of eight

eimerians in 113 *S. floridanus*, of which all but one had North American origins, and were presumably introduced into Italy together with imported cottontails.

Including the mammalian host species included herein, 11 species of coccidians have been reported in six species of Oklahoma mammals, including those in two chiropterans, and a single each of eulipotyphlan and lagomorph, as well as two rodents (Table 1). As Oklahoma supports 108 native species of mammals (Caire et al. 2019) and only 6% have been reported with coccidia to date, much more survey work remains in determining the coccidian fauna of this class of vertebrates, including salvaging DOR specimens as in the present case.

Acknowledgements

The Oklahoma Department of Wildlife Conservation issued Scientific Collecting Permit No. 1551646 to CTM. Drs. Scott L. Gardner and Gabor Racz (HWML) are acknowledged for expert curatorial assistance.

References

- Bertolino S, Hofmannová L, Girardello M, Modry D. 2010. Richness, origin and structure of an *Eimeria* community in a population of eastern cottontail (*Sylvilagus floridanus*) introduced into Italy. *Parasitology* 137:1179–1186.
- Caire W, Loucks LS, Haynie ML, Coyner BS, Braun JK. 2019. Updated and revised checklist of the mammals of Oklahoma, 2019. *Proc Oklahoma Acad Sci* 99:1–6.
- Duszynski DW, Couch L. 2013. The biology and identification of the coccidia (Apicomplexa) of rabbits of the world. London (UK): Elsevier Science. 352 p.
- Duszynski DW, Marquardt WC. 1969. *Eimeria* (Protozoa: Eimeridae) of the cottontail rabbit, *Sylvilagus audubonii*, in northeastern Colorado with descriptions of three new species. *J Protozool* 16:128–137.

Table 1. Coccidians reported from Oklahoma mammals.*

Coccidian	Host	Prevalence†	County Locality	Reference
<i>Eimeria catronensis</i>	<i>Myotis septentrionalis</i>	1/4 (25)	Le Flore	McAllister et al. (2012)
<i>Eimeria lukfataensis</i>	<i>Sciurus carolinensis</i>	1/3 (33)	McCurtain	McAllister et al. (2019)
<i>Eimeria macyi</i>	<i>Perimyotis subflavus</i>	1/2 (50)	Delaware	McAllister et al. (2016)
<i>Eimeria neoirresidua</i>	<i>Sylvilagus floridanus</i>	1/1 (100)	McCurtain	This study
<i>Eimeria poudeiri</i>	<i>S. floridanus</i>	1/1 (100)	McCurtain	This study
<i>Eimeria roperi</i>	<i>Sigmodon hispidus</i>	4/30 (13)	Payne	Faulkner and Lochmiller (1997)
<i>Eimeria sigmodontis</i>	<i>S. hispidus</i>	15/30 (50)	Payne	Faulkner and Lochmiller (1997)
<i>Eimeria tkachi</i>	<i>Blarina carolinensis</i>	2/3 (67)	McCurtain	McAllister and Seville (2017)
<i>Eimeria tumlisoni</i>	<i>M. septentrionalis</i>	1/4 (25)	Le Flore	McAllister et al. (2012)
<i>Eimeria tuskegenesis</i>	<i>S. hispidus</i>	3/30 (10)	Payne	Faulkner and Lochmiller (1997)
<i>Eimeria webbae</i>	<i>S. hispidus</i>	2/30 (7)	Payne	Faulkner and Lochmiller (1997)

*Not including large game (deer, elk, bears, pronghorn antelopes) and farm animals (goats, sheep, cattle, pigs).

†Prevalence = number infected/number examined (%).

- Faulkner BC, Lochmiller RL. 1977. New locality records of coccidian parasites (Apicomplexa: Eimeriidae) of the hispid cotton rat (*Sigmodon hispidus*) from Oklahoma. Proc Oklahoma Acad Sci 77:131–132.
- McAllister CT, Connior MB, Bursley CR, Durden LA, Seville RS, Robison HW, Trauth SE. 2016. Parasites (Coccidia, Trematoda, Acari) of tri-colored bats, *Perimyotis subflavus* (Chiroptera: Vespertilionidae): New geographical records for Oklahoma. Proc Oklahoma Acad Sci 96:63–69.
- McAllister CT, Seville RS. 2017. A new eimerian (Apicomplexa: Eimeriidae) from southern short-tailed shrews, *Blarina carolinensis* (Bachman) (Soricimorpha: Soricidae: Soricinae) from southeastern Oklahoma, USA. Syst Parasitol 94:711–716.
- McAllister CT, Seville RS, Roehrs ZP. 2012. A new species of *Eimeria* (Apicomplexa: Eimeriidae) from the northern myotis, *Myotis septentrionalis* (Chiroptera: Vespertilionidae), in Oklahoma. J Parasitol 98:1003–1005.
- McAllister CT, Motriuk-Smith D, Seville RS. 2019. A new species of *Eimeria* (Apicomplexa: Eimeriidae) from eastern gray squirrel, *Sciurus carolinensis* (Rodentia: Sciuridae: Sciurinae: Sciurini) from Oklahoma, USA. Syst Parasitol 96:417–421.
- Wiggins JP, Cosgrove M, Rothenbacher H. 1980. Gastrointestinal parasites of the eastern cottontail (*Sylvilagus floridanus*) in central Pennsylvania. J Wildl Dis 16:541–544.
- Wilber PG, Duszynski DW, Upton SJ, Seville RS, Corliss JO. 1998. A revision of the taxonomy and nomenclature of the *Eimeria* spp. (Apicomplexa: Eimeriidae) from rodents in the Tribe Marmotini (Sciuridae). Syst Parasitol 39:113–135.

Submitted August 22, 2023 Accepted October 30, 2022