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A New *Eimeria* Species (Apicomplexa: Eimeriidae) Infecting *Onychomys* Species (Rodentia: Muridae) in New Mexico and Arizona

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A NEW *EIMERIA* SPECIES (APICOMPLEXA: EIMERIIDAE) INFECTING *ONYCHOMYS* SPECIES (RODENTIA: MURIDAE) IN NEW MEXICO AND ARIZONA

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ABSTRACT: Fecal samples from 3 species of *Onychomys* (Rodentia: Muridae) captured in New Mexico and Arizona were examined for coccidia. Six of the 59 (10%) were infected with a new species of *Eimeria.* Sporulated oocysts (n = 105) of this new species are subspheroidal, 17.4 × 16.1 (14–21 × 13–19) μm, with ellipsoidal sporocysts 10.4 × 5.7 (9–12 × 5–8) μm. This species occurred in 3 of 24 (13%) *Onychomys arenicola,* 2 of 31 (6%) *Onychomys leucogaster* from New Mexico, and 1 of 4 (25%) *Onychomys torridus* from Arizona. Isolates recovered from *O. leucogaster* and *O. torridus* were inoculated into *O. leucogaster* (n = 5) and produced infections with a prepatent period of 7 days and a patent period of 7–23 days.

There are 3 species of *Onychomys*, or grasshopper mice, but to date only 1 coccidian has been described from these predatory rodents, i.e., *Eimeria onychomysis* Levine, Ivens, and Kruidenier, 1957. Recent parasitological work on grasshopper mice has included studies on helminths (Pfaffenberger et al., 1985) and ectoparasites (Pfaffenberger and DeBruin, 1986; Thomas, 1988), but the only recent survey for coccidia of *Onychomys* included just 2 individuals, and neither was shedding oocysts (McAllister et al., 1991). Herein, we describe a new species of *Eimeria* that was recovered from the feces of all 3 species of *Onychomys* that were trapped in the southwestern United States.

MATERIALS AND METHODS

Feces were taken from the intestines of *Onychomys leucogaster* and *Onychomys arenicola* that were collected as part of The University of New Mexico's Long Term Ecological Research (LTER) project on the Sevilleta National Wildlife Refuge, Socorro County, New Mexico during 1993–1996 (see Wilson et al., 1997). All voucher specimens of hosts from the LTER were deposited in the Museum of Southwestern Biology, the University of New Mexico (MSB). In addition, in December 1996, feces were collected from live *Onychomys torridus* that were trapped on permanent plots located at the Cave Creek Bajada, San Simon Valley, near Portal, Arizona (see Brown and Munger, 1985). Procedures for experimental inoculations of captive-reared *O. leucogaster* followed Upton et al. (1992); inoculation doses ranged from ~10 to 1,000 oocysts. All procedures for preserving fecal material as well as measuring and photographing oocysts were described earlier (Duszynski et al., 1982; Stout and Duszynski, 1983). The species description is based on guidelines by Duszynski and Wilber (1997); measurements are in μm, with size ranges in parentheses following the means.

DESCRIPTION

Eimeria sevilletensis n. sp.
(Figs. 1–4)

Description: Oocyst subspheroidal, occasionally spheroidal, with wall 1.3 thick (0.9–2.2) (n = 103), composed of 2 layers; outer layer smooth, colorless to light yellow-brown, ~½ of total thickness, inner layer smooth, colorless to light yellow-green; micropyle and oocyst residuum absent, but 1–2 (occasionally 0) relatively large polar granules present, 1.8 × 1.3 (1.0–2.5 × 0.5–2.5) (n = 75); sporulated oocysts (n = 105), 17.4 × 16.1 (14–21 × 13–19), with L:W ratio 1.1 (1.0–1.2); sporocysts ellipsoidal, 10.4 × 5.7 (9–12 × 5–8), with L:W ratio 1.8 (1.5–2.2); Stieda body present, small and button-like, but sub- and parastieda bodies absent; sporocyst residuum typically a large, compact mass of many fine granules located between and sometimes obscuring the sporozoites; sporozoites elongate, with 1 end rounded and the other tapering, lacking refractile globules.

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Taxonomic summary

Symbiotype: Onychomys arenicola Mearns, 1896, Mearn's grasshopper mouse.

Other hosts: Onychomys leucogaster (Wied-Neuwied, 1841), northern grasshopper mouse; *O. torridus* (Coues, 1874), southern grasshopper mouse (scorpion mouse).

Type locality: U.S.A., New Mexico, Socorro Co., Sevilleta National Wildlife Refuge, Grassland—east trapping site, Web 1 (106° 43′31″W, 34° 20′7″N).

Other localities: U.S.A., New Mexico, Socorro Co., Sevilleta National Wildlife Refuge, Creosote-east trapping site, Web 2 (106° 43′57″W, 34° 19′52″N), Web 4 (106° 44′22″W, 34° 20′3″N); Creosote-west trapping site, Web 1 (106° 55′26″W, 34° 17′42″N); Grassland-east trapping site, Web 4 (106° 43′13″W, 34° 20′10″N); Arizona, Cochise Co., San Simon Valley, Cave Creek Bajada, 6.5 km E, 2 km N of Portal.

Prevalence: Onychomys arenicola: 3 of 24 (13 %) in New Mexico, 1995–1996; *O. leucogaster*: 2 of 31 (6%) in New Mexico, 1995–1996; *O. torridus*: 1 of 4 (25%) in Arizona, 1996.

Site of infection: Unknown, oocysts recovered from intestinal contents.

Prepatent period: Seven days after inoculation (DAI) in *O. leucogaster* (experimental; n = 5).

Patent period: Seven to 23 DAI in *O. leucogaster* (experimental; n = 5).

Pathogenicity: The feces of 3 animals, each receiving ~1,000 sporulated oocysts, became loose but formed and darker in color within 6–9 DAI and remained so throughout the patent period.

Cross-immunity: Two animals, each inoculated with the proposed species within 5–7 days after the last day of patency resulting from experimental infections with *Eimeria onychomysis,* developed infections whose prepatencies, patencies, and intensities were indistinguishable from those of experimentally infected animals with no prior exposure to *E. onychomysis.*

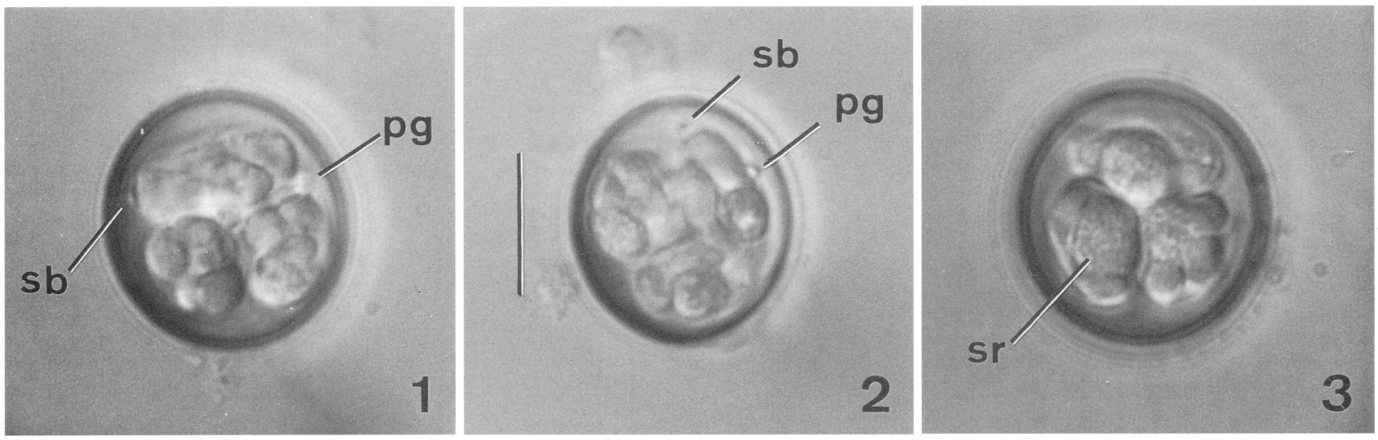
Sporulation time: Two to 3 days at ~25 C.

Material deposited: Phototype (see Bandoni and Duszynski, 1988) of sporulated oocysts in the United States National Parasite Collection (USNPC), no. 88173. Symbiotype (see Frey et al., 1992): *O. arenicola* in the MSB no. 85073 (NK 40134, male, 21 May 1996).

Etymology: The nomen triviale is derived from the collection locality where we first encountered this coccidian.

Remarks

Eimeria sevilletensis is the second *Eimeria* species to be described from the genus *Onychomys*; the first was *E. onychomysis* Levine, Ivens, and Kruidenier, 1957, collected from *O. leucogaster* from Arizona (Levine et al., 1957). The new species differs from *E. onychomysis* in the mean size of its oocyst (17.4 × 16.1 vs. 20 × 19) and sporocyst (10.4 × 5.7 vs. 11 × 8), in lacking an oocyst residuum, and in having a button-like and less prominent Stieda body. Among *Eimeria* from other hosts in the subfamily Sigmodontinae, *E. sevilletensis* is most similar to *Eimeria knoxjonesi* Duszynski and McAllister, 1995, from *Peromyscus pectoralis* from Texas (Duszynski and McAllister, 1995). Like *E. knoxjonesi,* the new species has a smooth, bilayered wall, a small button-like Stieda body, lacks an oocyst residuum, and is similar in the



FIGURES 1–3. Photomicrographs of sporulated oocysts of *Eimeria sevilletensis* n. sp. recovered from the feces of *Onychomys arenicola* captured at the Sevilleta National Wildlife Refuge, Socorro County, New Mexico, U.S.A. Scale bar = 10 μm. Abbreviations: pg, polar granule; sb, Stieda body; sr, sporocyst residuum.

mean length of its oocyst (17.4 vs. 16.5) and mean dimensions of its sporocyst (10.4 × 5.7 vs. 9.1 × 4.7). However, it differs from *E. knoxjonesi* in oocyst shape (subspheroid vs. ellipsoid), mean width of oocyst (16.1 vs. 11.9), and L:W ratio of oocyst (mean 1.1, range 1.0–1.2 vs. mean 1.4, range 1.3–1.6). In addition, the oocyst wall of *E. knoxjonesi* appears thinner at one end, its sporocyst residuum is usually a small rosette of 3–4 granules, and its sporozoites contain 1–2 refractile bodies; in contrast, the oocyst wall of *E. sevilletensis* has no areas of thinning, its sporocyst residuum is a large, compact mass of many fine granules, and its sporozoites do not appear to contain refractile globules. Among *Eimeria* from other hosts in the Muridae, the new species most closely resembles *Eimeria falciformis* (Eimer, 1870) Schneider, 1875, from *Mus musculus*. The oocysts and sporocysts of both species overlap in shape and size (see Pellérdy, 1974; Levine and Ivens, 1990), their oocyst walls are smooth, and their Stieda bodies and sporocyst residua appear similar. However, the oocyst wall of the new species is obviously bilayered, whereas that of *E. falciformis* has been reported to have either 1 or 2 layers. In addition, *E. sevilletensis* consistently lacks an oocyst residuum, whereas the oocyst residuum of *E. falciformis* is variably present, the sporozoites of the proposed species lack refractile bodies, and those of *E. falciformis* contain a refractile globule at the broad end. Finally, an experimental inoculation of ~1,000 *E. falciformis* oocysts into an *O. leucogaster* (previously unexposed to any coccidia) did not result in a patent infection (data not shown).

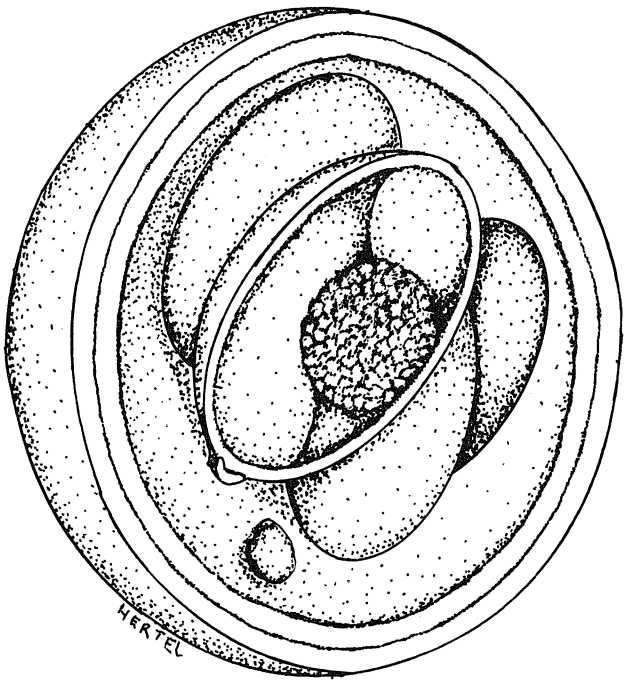


FIGURE 4. Composite line drawing of sporulated oocyst of *Eimeria sevilletensis* found in the feces of *Onychomys* species. Scale bar = 10 μm.

Although the mean dimensions of the oocysts and sporocysts of *E. sevilletensis* collected from *O. arenicola* were statistically smaller than those isolated from *O. leucogaster* and *O. torridus*, and the mean dimensions of the oocysts collected from *O. leucogaster* were statistically smaller than those from *O. torridus* (Table I), oocysts and sporocysts from the 3 hosts overlapped in measurements and were identical in qualitative traits. In addition, oocysts isolated from *O. torridus* produced patent infections in experimentally infected *O. leucogaster*. For these reasons, we consider the 3 forms to represent a single species.

DISCUSSION

Members of the genus *Onychomys* can be found from southern Manitoba and eastern Washington state, through the southwestern United States, and into northern Mexico, and typically are found in desert scrub and shortgrass prairie habitats within these regions (Nowak, 1991). Nevertheless, at present, only 2 coccidia have been documented from these rodents, the first of which was described 40 yr ago. The apparently low diversity of coccidia in these hosts may reflect the fact that few surveys for coccidia have included *Onychomys* spp.; this may be because they, like most predators, occur at relatively low densities (Nowak, 1991) and are thus less likely to be included in sizable numbers in surveys of the coccidia of rodents (McAllister et al., 1991).

In addition, the species we are describing may have escaped prior detection because, on cursory examination at lower magnifications, it may be confused with *E. onychomysis*. This was demonstrated when an experimental infection with a sample that had been misidentified as *E. onychomysis* yielded oocysts of the new species, a serendipitous result that prompted the infection experiments reported on above. This example is also noteworthy because the experimental infection was done in Jan-

TABLE I. Comparison of *Eimeria sevilletensis* oocysts from 3 *Onychomys* host species for 4 quantitative characters (in μm) with mean values followed by ranges in parentheses; sample sizes refer to the number of oocysts and sporocysts measured.*

Character	*O. arenicola* (n = 43)	*O. leucogaster* (n = 37)	*O. torridus* (n = 25)
Oocyst length	16.5 (13.6–21.0)	17.5 (15.0–19.0)	19.0 (17.0–21.0)
Oocyst width	15.3 (12.8–19.0)	16.2 (13.5–18.0)	17.0 (15.0–19.0)
Sporocyst length	9.9 (9.0–12.0)	10.5 (9.0–12.0)	11.0 (10.0–12.0)
Sporocyst width	5.5 (4.5–7.5)	5.7 (5.0–7.0)	6.0 (5.0–7.0)

* Underscored character mean indicates no significant difference between those means (Fisher's LSD multiple comparisons test, $\alpha = 0.05$).

uary 1997, using ~10–15 oocysts that were isolated and stored in June 1993, i.e., that were ~3.5 yr old, yet still viable. Pellérdy (1974) stated that, in general, the viability of oocysts stored at 4–6 C declines so that after 1–1.5 yr, the oocysts are useless for experiments. Nevertheless, our experience is that at least some of the *Eimeria* species of rodents may remain viable for 3–4 yr when stored in 2% (w/v) aqueous potassium dichromate at ~4 C.

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LITERATURE CITED

BANDONI, S. M., AND D. W. DUSZYNSKI. 1988. A plea for improved presentation of type material for coccidia. Journal of Parasitology **74:** 519–523.

BROWN, J. H., AND J. C. MUNGER. 1985. Experimental manipulation of a desert rodent community: Food addition and species removal. Ecology **66:** 1545–1563.

DUSZYNSKI, D.W., G. EASTHAM, AND T. L. YATES. 1982. *Eimeria* from jumping mice (*Zapus* spp.): A new species and genetic and geographic features of *Z. hudsonicus luteus.* Journal of Parasitology **68:** 1146–1148.

———, AND C. T. MCALLISTER. 1995. Coccidian parasites of *Peromyscus attwateri* and *P. pectoralis* in Texas with a description of a new species from *P. pectoralis laceianus.* Occasional Papers of the Museum of Texas Tech University **156:** 1–8.

———, AND P. G. WILBER. 1997. A guideline for the preparation of species descriptions in the Eimeriidae. Journal of Parasitology **83:** 333–336.

FREY, J. K., T. L. YATES, D. W. DUSZYNSKI, W. L. GANNON, AND S. L. GARDNER. 1992. Designation and curatorial management of type host specimens (symbiotypes) for new parasite species. Journal of Parasitology **78:** 930–932.

LEVINE, N. D., AND V. IVENS. 1990. The coccidian parasites of rodents. CRC Press, Inc., Boca Raton, Florida, 228 p.

———, ———, AND F. J. KRUIDENIER. 1957. New species of *Eimeria* from Arizona rodents. Journal of Protozoology **4:** 80–88.

MCALLISTER, C. T., S. J. UPTON, J. V. PLANZ, AND T. S. DEWALT. 1991. New host and locality records of coccidia (Apicomplexa, Eimeriidae) from rodents in the southwestern and western United States. Journal of Parasitology **77:** 1016–1019.

NOWAK, R. M. 1991. Walker's mammals of the world, 5th ed., Vol. 2. The Johns Hopkins University Press, Baltimore, Maryland, 1629 p.

PELLÉRDY, L. P. 1974. Coccidia and coccidiosis, 2nd ed. Akademiai Kiado, Budapest, Hungary, 959 p.

PFAFFENBERGER, G.S., AND D. DEBRUIN. 1986. Ectoparasitic overlap between sympatric *Dipodomys ordii* and *Onychomys leucogaster* (Rodentia) in eastern New Mexico, U.S.A. Journal of Medical Entomology **23:** 201–207.

———, K. KEMETHER, AND D. DEBRUIN. 1985. Helminths of sympatric populations of kangaroo rats (*Dipodomys ordii*) and grasshopper mice (*Onychomys leucogaster*) from the high plains of eastern New Mexico. Journal of Parasitology **71:** 592–595.

STOUT, C. A., AND D. W. DUSZYNSKI. 1983. Coccidia from kangaroo rats (*Dipodomys* spp.) in the western United States, Baja California and northern New Mexico with descriptions of *Eimeria merriami* sp. n. and *Isospora* sp. Journal of Parasitology **69:** 209–214.

THOMAS, R. E. 1988. A review of flea collection records from *Onychomys leucogaster* with observations on the role of grasshopper mice in the epizootiology of wild rodent plague. Great Basin Naturalist **48:** 83–95.

UPTON, S. J., C. T. MCALLISTER, D. B. BRILLHART, D. W. DUSZYNSKI, AND C. D. WASH. 1992. Cross-transmission studies with *Eimeria arizonensis*-like oocysts (Apicomplexa) in New World rodents of the genera *Baiomys, Neotoma, Onychomys, Peromyscus,* and *Reithrodontomys* (Muridae). Journal of Parasitology **78:** 406–413.

WILSON, W. D., J. A. HNIDA, AND D. W. DUSZYNSKI. 1997. Parasites of mammals on the Sevilleta National Wildlife Refuge, Socorro, New Mexico: *Cuterebra austeni* and *C. neomexicana* (Diptera: Oestridae) from *Neotoma* and *Peromyscus* (Rodentia: Muridae), 1991–1994. Journal of Medical Entomology **34:** 359–367.