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HELMINTHS OF NORTHERN LEOPARD FROGS, *RANA PIPIENS* (RANIDAE), FROM NORTH DAKOTA AND SOUTH DAKOTA

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Key words: Rana pipiens, *Ranidae, helminths, North Dakota, South Dakota.*

The northern leopard frog, *Rana pipiens* Schreber 1782, is widely distributed in North America from the northeast Atlantic coast through Hudson Bay, Canada, to eastern Washington, Oregon, and California, south to northern Virginia through Nebraska to New Mexico and central Arizona (Stebbins 1985). Trematodes and cestodes of *R. pipiens* have been summarized by Prudhoe and Bray (1982), nematodes by Baker (1987). Additional *R. pipiens* helminth records are included in Dyer (1991) and McAlpine and Burt (1998). Because to date there are no published records of helminths for *R. pipiens* from North Dakota and South Dakota, we undertook an investigation to report helminths harbored by *R. pipiens* from these states.

One hundred forty *R. pipiens* from 6 collection sites (2 in North Dakota and 4 in South Dakota) were examined (Appendix). The frogs were collected from 1995 to 1998 (collection dates are in the Appendix), fixed in 10% formalin, and preserved in 70% ethanol. After opening the body cavity, we removed the gastrointestinal tract. The lungs, esophagus, stomach, small intestine, large intestine, bladder, and body cavity of each frog were then examined separately with a dissecting microscope for helminths. Nematodes were cleared on a glass slide in glycerol; selected cestodes and trematodes were stained with hematoxylin and mounted in balsam. We made identifications from these preparations. Frogs were deposited in the herpetology collection of the Natural History Museum of Los Angeles County (LACM), Los Angeles, California (Appendix). Representative specimens were placed in vials of ethanol and deposited in the United States National Parasite Collection, USDA, Beltsville, Maryland (Appendix).

We found gravid individuals representing 5 species of Trematoda: *Cephalogonimus americanus* Stafford 1902; *Glypthelmins quieta* (Stafford 1902), Stafford 1905; *Gorgoderina attenuata* (Stafford 1902), Stafford 1905; *Haematoloechus varioplexus* Stafford 1902; *Megalodiscus temperatus* (Stafford 1905) Harwood 1932. We also found 5 species of Nematoda: *Cosmocercoides variabilis* (Harwood 1930) Travassos 1931; *Falcaustra ranae* (Walton 1941) Chabaud and Golvan 1957; *Oswaldocruzia pipiens* Walton 1929; *Rhabdias ranae* Walton 1929; *Spinitectus gracilis* Ward and Magath 1917. In addition, metacercariae representing 4 species of Trematoda *(Apharyngostrigea pipientis, Alaria* sp., *Fibricola* sp., Ochetosomatidae gen. sp.), plerocercoids of 1 species of cestoda *(Proteocephalus* sp.), and 3rd stage larvae of 2 species of Nematoda (*Physaloptera* sp., *Physocephalus* sp.) were found. Prevalence and mean intensity for each helminth species are given in Table 1.

One hundred three of 140 (74%) frogs harbored helminths: 49 of 71 (69%) female frogs and 54 of 69 (78%) males. There was no significant difference in prevalence between female and male frogs ($\chi^2 = 1.53$, 1 df, $P > 0.05$). A total of 2057 helminths were found (20.0 ± 32.8 *s* helminths per infected frog). Of 103 infected frogs, 48 harbored 1 species of helminth, 24 harbored 2 species, 24 harbored 3 species, 5 harbored 4 species, and 2 harbored 5 species (1.9 ± 1.0 *s* helminth species per infected frog).

Helminths we found in this study fall into 3 categories: phoresis (resting stages), anuran

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TABLE 1. Prevalence (P) and mean intensity ($\bar{x} \pm s$) of helminths from 140 *Rana pipiens* from North and South Dakota.

	North Dakota				South Dakota							
	Barnes Co. ($N = 20$)		Sargent Co. ($N = 20$)		Beadle Co. ($N = 20$)		Brown Co. ($N = 31$)		Hamlin Co. ($N = 26$)		Hand Co. ($N = 23$)	
Helminth	P	$\bar{x} \pm s$	P	$\bar{x} \pm s$	P	$\bar{x} \pm s$	P	$\bar{x} \pm s$	P	$\bar{x} \pm s$	P	$\bar{x} \pm s$
TREMATODA												
Cephalogonimus americanus	—	—	—	—	—	—	—	—	—	—	4	2
Glypthelmins quieta	25	1.4 ± 0.5	40	2.3 ± 0.7	20	3.3 ± 1.7	—	—	4	1	—	—
Gorgoderina attenuata	5	1	—	—	—	—	—	—	—	—	—	—
Haematoloechus varioplexus	30	5.7 ± 4.1	25	1.2 ± 0.4	95	8.6 ± 9.9	16	1.4 ± 0.9	27	2.6 ± 1.9	30	2.6 ± 2.3
Megalodiscus temperatus	20	2.5 ± 1.3	—	—	—	—	3	1	—	—	—	—
mesocercariae, *Alaria* sp.	55	55 ± 52	20	65 ± 81	15	2.0 ± 1.0	19	12.2 ± 5.2	—	—	7	4.0 ± 0.0
metacercariae, *Apharyngostrigea pipientis*	—	—	—	—	20	6.8 ± 4.2	—	—	—	—	—	—
metacercariae, *Fibricola* sp.	10	1.5 ± 0.7	15	21 ± 26	50	21 ± 27	—	—	15	14.3 ± 23.3	—	—
metacercariae, Ochetosomatidae gen. sp.	5	1	5	20	5	24	16	16.0 ± 15.4	16	22.3 ± 26.5	13	1.0 ± 0.0
CESTODA												
plerocercoids, *Proteocephalus* sp.	30	2.0 ± 1.3	—	—	—	—	—	—	—	—	—	—
NEMATODA												
Cosmocercoides variabilis	20	1.5 ± 1.0	5	6	—	—	32	4.6 ± 3.6	19	3.4 ± 2.3	—	—
Falcaustra ranae	—	—	—	—	—	—	—	—	4	2	—	—
Oswaldocruzia pipiens	—	—	—	—	—	—	3	1	23	1.8 ± 1.0	—	—
Rhabdias ranae	40	3 ± 2.1	—	—	60	3.6 ± 2.8	3	4	—	—	—	—
Spinitectus gracilis	—	—	—	—	—	—	29	5.7 ± 9.8	—	—	—	—
larvae, *Physaloptera* sp.	—	—	5	1	—	—	3	2	—	—	—	—
larvae, *Physocephalus* sp.	—	—	25	5.8 ± 2.6	—	—	—	—	—	—	—	—

parasites, and dietary artifacts. Of 2057 helminths, 1575 (76%) were larvae of species that do not reach maturity in frogs (1531 metacercariae, 12 plerocercoids, 29 larval nematodes in cysts) and 3 were larval nematodes representing dietary artifacts. For these metacercariae, mammals are definitive hosts for *Alaria* sp. and *Fibricola* spp., birds for *Apharyngostrigea* spp., and snakes for Ochetosomatidae gen. sp.; frogs serve as paratenic hosts (Schell 1985). Species of *Proteocephalus* are primarily parasites of fish with intermediate stages in microcrustaceans (Olsen 1974). Species of *Physaloptera* and *Physocephalus* require an insect intermediate host (Anderson 1992); and in anurans, larvae of *Physocephalus* are most often found in cysts while larvae of *Physaloptera* pass from the body without further development.

The remaining 482 helminths represent species reaching maturity in anurans. Thirty-nine (55%) female and 44 (64%) male frogs were infected. There was no significant difference in prevalence between female and male frogs ($\chi^2 = 1.13$, 1 df, $P > 0.05$; 1.4 ± 0.6 *s* species per infected frog, 5.8 ± 7.6 *s* helminths per infected frog). Thus, anuran parasites represent 23% of the helminth load of these frogs.

With the exception of *S. gracilis,* these helminths have been reported previously in *R. pipiens* and are common parasites of amphibians of the American Midwest (see Dyer 1991). *Spinitectus gracilis,* a parasite of fish, has been reported from *Bufo woodhousii* from Illinois (Jilek and Wolff 1978) and *Rana catesbeiana* from Michigan and Oklahoma (Trowbridge and Hefley 1934, Muzzall 1991).

Rana pipiens represents a new host record for *Spinitectus gracilis.* North and South Dakota are new locality records for each helminth listed in Table 1.

Rana pipiens from North Dakota were collected under permit 102038938 issued by the North Dakota Game and Fish Department to RGM; frogs from South Dakota were collected under permit 25 issued by the Department of Game, Fish and Parks of South Dakota to RGM. Frogs were collected with support, in part, of grant 2675BR1 from the Council for Tobacco Research–USA, Inc. to RGM.

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APPENDIX

MUSEUM ACCESSION NUMBERS

Collection data for *Rana pipiens* from North Dakota and South Dakota. NORTH DAKOTA: $N = 20$, snout-vent length (SVL) = 54.9 mm ± 9.8 *s*, 11 female, 9 male, Sheyenne River, Little Yellowstone State Park, Barnes County (46°38′N, 97°55′W, 381 m elevation), collected September 1998, LACM 145306–145325; $N = 20$, SVL = 64.5 mm ± 6.2 *s*, 11 female, 9 male, Silver Lake, Sargent County (46°01′N, 97°34′W, 372 m elevation), collected September 1998, LACM 145326–145345. SOUTH DAKOTA: $N = 20$, SVL = 59.8 mm ± 9.9 *s*, 12 female, 8 male, Lake Cavour, Beadle County (44°24′N, 98°03′W, 399 m elevation), collected July 1996, LACM 143822–143841; $N = 31$, SVL = 61.4 mm ± 5.0 *s*, 11 female, 20 male, Richmond Lake, Brown County (45°32′N, 98°35′W, 414 m elevation), collected October 1995, LACM 143791–143821; $N = 26$, SVL = 55.7 mm ± 5.9 *s*, 15 female, 11 male, Lake Poinsett, Hamlin County (44°35′N, 97°04′W, 502 m elevation), collected October 1995, LACM 143845–143870; $N = 23$, SVL = 59.5 mm ± 6.2 *s*, 10 female, 13 male, Jones Lake, Hand County (44°28′N, 98°56′ 494 m elevation), collected July 1996, LACM 143871– 143893.

Helminths from *Rana pipiens* deposited in the United States National Parasite Collection. *Cephalogonimus americanus* USNPC 88784, *Glypthelmins quieta* USNPC 88785, *Gorgoderina attenuata* USNPC 88786, *Haematoloechus varioplexus* USNPC 88787, *Megalodiscus temperatus* USNPC 88788, *Alaria* sp. (mesocercariae) USNPC 88789, *Apharyngostrigea pipientis* (metacercariae) USNPC 88790, *Fibricola* sp. (metacercariae) USNPC 88791, Oechetosomatidae gen. sp. (metacercariae) USNPC 88792, *Proteocephalus* sp. (plerocercoids) USNPC 88793, *Cosmocercoides variabilis* USNPC 88794, *Oswaldocruzia pipiens* USNPC 88795, *Rhabdias ranae* USNPC 88797, *Spinitectus gracilis* USNPC 88796, *Physaloptera* sp. (larvae) USNPC 88798, *Physocephalus* sp. (larvae) USNPC 88799.