

New Host and Distributional Records for Hematozoan and Helminth Parasites of Midland

Watersnake, *Nerodia sipedon pleuralis* (Ophidia: Natricidae), from Western Arkansas

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Abstract: Four Midland watersnakes, *Nerodia sipedon pleuralis*, collected in June 2024 and May and August 2025 from Polk County, Arkansas, were examined for hematozoan and helminth parasites. A single snake was infected with an intraerythrocytic hematozoan, *Hepatozoon* sp., as well as the digenetic trematodes, *Styphlodora magna* Byrd and Denton, 1938, and *Dasymetra conferta* Nicoll, 1911. Another harbored several helminths, including the digenetic trematodes, *Renifer aniarum* (Leidy, 1890) and *Paralechthiorchis natricis* (Holl and Allison, 1935), a cestode, *Mesocostoides* sp., and a nematode, *Ophidascaris ashi* Walton, 1927; one other snake was infected with only *R. aniarum* and the other specimen was negative. We document some new host and geographic distributional records for these parasites.

Introduction

The Midland watersnake, *Nerodia sipedon pleuralis* (Cope) ranges from southern Illinois and Indiana south through western Ten-

nessee, western Kentucky, Arkansas, eastern Oklahoma, most of Mississippi and Alabama, and eastward to Georgia and South Carolina (Powell et al. 2016). In Arkansas, this snake occurs throughout the Interior Highlands and West Gulf Coastal Plain with sporadic records from the

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Mississippi Alluvial Plain (Trauth et al. 2004). It feeds mostly on fish but the diet also includes amphibians (Ernst and Barbour 1989; Mount 1975).

Most of the information on hematozoans (Fantham and Porter 1954; Hull and Camin 1960; Smith et al. 1994; Smith and Dessler 1997) and helminths (Ernst and Ernst 2006; Walley et al. 2012) of *N. sipedon* is provided on the nominal subspecies, the northern watersnake, *N. sipedon sipedon* (L.) from Ohio, Ontario, and Québec, Canada. However, McAllister et al. (2017) reported a hematozoan, *Hepatozoon* sp. from *N. s. pleuralis* from Missouri, and Detterline et al. (1984) documented helminths of *N. s. pleuralis* from Alabama. Here, we report a hematozoan and several helminths from a small sample of *N. s. pleuralis* from western Arkansas.

Methods

On 21–22 June 2024 ($n=2$), 27 May 2025 ($n=1$), and 30 August 2025 ($n=1$), four adult *N. s. pleuralis* (mean $\pm$ sd snout-vent length [SVL] = 550 ± 76.8 , range 460–670 mm) were collected from Butcherknife Creek at Big Fork off St. Hwy 8, Polk County, Arkansas ($34^{\circ}28'5.109''\text{N}$, $93^{\circ}56'52.1772''\text{W}$) (Fig. 1) with a snake tong or in a baited minnow trap and examined for helminth parasites. Specimens were euthanized with an intraperitoneal injection of concentrated tricaine methanesulfonate (SIH 2004). We follow McAllister et al. (2017) in processing snakes for hematozoan parasites in the blood. The oral cavity was examined for trematodes, and a mid-ventral incision was made to expose the viscera. The entire gastrointestinal tract was removed and split lengthwise, rinsed in 0.9% (v/v) saline, and examined as well as other organs, including the heart, lungs, liver, and gonads. Trematodes and cestodes were fixed in nearly boiling tap water without coverslip pressure, stained with aceto-carmine, dehydrated in a graded ethanol series, cleared in methyl salicylate, and mounted in Canada balsam. Nematodes were fixed in hot tap water and studied as temporary mounts on a microscopic slide in a drop of glycerol. Parasites were either retained for future molecular studies or deposited as vouchers in the Harold W. Manter Laboratory of Parasitology (HWML), University

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of Nebraska-Lincoln. Host voucher specimens were deposited in the Northeast Texas Community College (NTCCVC) Vertebrate Collection, Mt. Pleasant, Texas. Host common and scientific names follow Nicholson (2025).



Figure 1. Habitat of *Nerodia sipedon pleuralis* at Butcherknife Creek, Polk County, Arkansas. Photo by CTM.

Results and Discussion

Three of four *N. s. pleuralis* harbored parasites. A single *N. s. pleuralis* was infected with an intraerythrocytic hematozoan parasite as well as two trematodes. Two others harbored helminth parasites as follows: one harbored multiple infection of two trematodes, a cestode, and a nematode; another was infected with a trematode; one specimen was negative. Detailed information on each parasite is provided below in an annotated format.

APICOMPLEXA: ADELEINA: HEPATIZOIDAE

Hepatozoon sp. – one specimen, a 670 mm SVL individual collected from a minnow trap on 30 August 2025 from the Butcherknife

Creek site was found to harbor an intraerythrocytic hematozoan of the genus *Hepatozoon*. Intensity (parasitemia) was relatively high as approximately 2-3% of the red blood cells contained elongate-shaped and partially recurved thin banana-shaped gamonts (Fig. 2). Interestingly, McAllister et al. (2017) reported a *Hepatozoon* sp. from *N. s. pleuralis* from Missouri with very similar morphologies (their Figs. 2A-C) but with a much lower parasitemia of less than 0.5%. In addition, Smith et al. (1994) reported *Hepatozoon sipedon* Smith, Dessler, and Martin, 1994 from *N. s. sipedon* from Ontario, Canada, with a parasitemia of only 0.2%. All hemogregarines from snakes are members of the genus *Hepatozoon* according to Smith (1996). However, without complete life cycle data including developmental stages from hematophagous invertebrates, specific identification of these hematozoans is difficult (Jacobson 2007; Smith and Dessler 1997). Although the Midland watersnake has been previously reported as a host, albeit in Missouri, we document a new distributional record for the parasite.

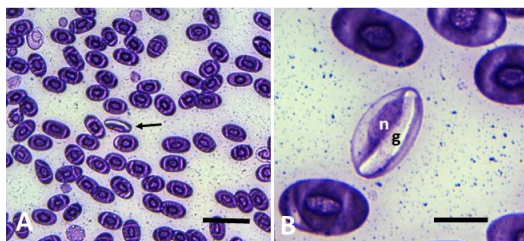


Figure 2. *Hepatozoon* sp. from *Nerodia sipedon pleuralis*. (A) View showing infected red blood cell (rbc, arrow). Scale bar = 25 µm. (B) Closer microscopic view showing nucleus (n) of infected rbc with banana-shaped gamont (g). Scale bar = 10 µm.

TREMATODA: DIGENEA: PLAGIORCHIIDA: RENIFERIDAE

***Dasymetra conferta* Nicoll, 1911** – The same specimen above harbored 15 specimens (Fig. 3) in its oral cavity and esophagus/lungs. Nicoll (1911) originally described this trematode from northern diamond-backed watersnake, *Nerodia rhombifer rhombifer* (Hallowell) from an unknown North American locality. This digenean has been previously reported in various

natricines, including plain-bellied watersnake *N. e. erythrogaster* (Forster), broad-banded watersnake, *N. fasciata confluens* (Blanchard) *N. r. rhombifer*, and *N. s. pleuralis* from Alabama, Missouri, Oklahoma, and Texas (see Ernst and Ernst 2006; McAllister and Bursey 2012). We document a new geographic record for *D. conferta*.



Figure 3. *Dasymetra conferta* from *Nerodia sipedon pleuralis*. (A) Whole mount showing testes (T). Scale bar = 500 µm. (B) Higher magnification of anterior region of same specimen showing oral sucker (OS) and pharynx (PH). Scale bar = 125 µm.

***Renifer aniarum* (Leidy, 1891)** – a 460 mm SVL *N. s. pleuralis* collected on 27 May 2025 was found to harbor two *R. aniarum* (HWML 119232) in its oral cavity. Another *N. s. pleuralis* (660 mm SVL) collected on 21 June 2024 from the same site had four specimens of *R. aniarum* (Fig. 4) in its oral cavity. McAllister et al. (2008) provided a summary of the North American hosts of *R. aniarum* that included 23 species/subspecies of snakes, including 12 species/subspecies of natricines. This digenean has been previously reported from *N. s. pleuralis* from Alabama, Arkansas (Washington County), North Carolina, and Texas (Detterline et al. 1984; McAllister and Bursey 2008; McAllister et al. 2025) and

N. s. sipedon from Michigan and Pennsylvania (McAllister and Bursey 2008). It has been also been reported previously from Lonoke County (Arkansas) in a yellow-bellied watersnake, *Nerodia erythrogaster transversa* (Hallowell) (McAllister and Bursey 2008). Interestingly, introduced *R. aniarum* have been reported from grass snakes, *Natrix natrix* (L.) from southern Italy, which is thought to have originated from translocated American bullfrogs, *Rana catesbeiana* (Santoro et al. 2011). The life cycle involves first intermediate host snails (pulmonates) which possess cercariae that penetrate second intermediate host anuran tadpoles, which then preyed on by snakes (Schell 1985). This is the second time the parasite has been reported from a Midland watersnake from Arkansas, but the first record from Polk County.

***Paralechriorchis natricis* (Holl and Allison, 1935) Byrd and Denton, 1938** – the same 460 mm SVL specimen above harbored three specimens (HWML 119231) of *P. natricis* in its oral cavity. The original name *Zeugorchis natricis*, proposed by Holl and Allison (1935), was for trematodes found in the uterus of a *N. s. sipedon* collected from Wolf Creek, Grove City, Mercer County, Pennsylvania. Byrd and Denton (1938a) later stated “we found it impossible to distinguish between *Z. natricis* Holl and Allison, 1935, and *Lechriorchis secundus* Canavan, 1937. Due to this striking similarity of body plan and almost identical size range for the various internal anatomy we feel justified in considering *L. secundus* as a synonym of *Z. natricis*. Since *Z. natricis* possesses the characters of the genus *Paralechriorchis* we transfer it to that genus. The species becomes *Paralechriorchis natricis* (Holl and Allison, 1935).” In addition, Goodman (1951, 1952), in an unpublished dissertation, reported the parasite with the name *Dasymetra natricis*, but we do not accept it. This parasite has been previously reported from eastern copperhead, *Agkistrodon contortrix* (L.), *N. erythrogaster*, and a *Nerodia* sp. from Illinois and Indiana (Ernst and Ernst 2006). We document *P. natricis* from Arkansas, and from *N. s. pleuralis*, for the first time.



Figure 4. *Renifer aniarum* from *Nerodia sipedon pleuralis*. (A) Whole mount showing testes (T). Scale bar = 500 μ m. (B) View of upper region of same specimen showing cirrus sac (CS), oesophagus (OE), oral sucker (OS), pharynx (PH), and vitellaria (VT). Scale bar = 250 μ m.

PLAGIORCHIDA: PLAGIORCHIIDAE

***Styplodora magna* Byrd and Denton, 1938** – eight specimens (Fig. 5) were taken from the gallbladder of a single snake (670 mm SVL) collected on 30 August 2025. This species has similar species with their synonymies needing clarification, but fortunately, in the current case, the large difference in egg size was sufficient for differentiation (VV Tkach, pers. comm.). In fact, Ernst and Ernst (2006) list it in synonymy as *Ochetosoma magnum*. Byrd and Denton (1938b) originally described this digenean from the gallbladder of *N. s. sipedon* from Georgia and Mississippi. McAllister et al. (2020, 2021, 2023) previously reported *S. magna* from northern cottonmouths, *Agkistrodon piscivorus* (Lacépède) from Arkansas and Oklahoma. In addition, McAllister et al. (2023) provided a summary of the ophidian hosts and localities of *S. magna*, to include several species of North American watersnakes (*Nerodia*), *A. piscivorus*, North American racer, *Coluber constrictor* L., and eastern ratsnake, *Pantherophis alleghaniensis* (Holbrook) from Florida, Louisiana, and North Carolina. We document the first time this digenean has

been reported from *N. s. pleuralis*.

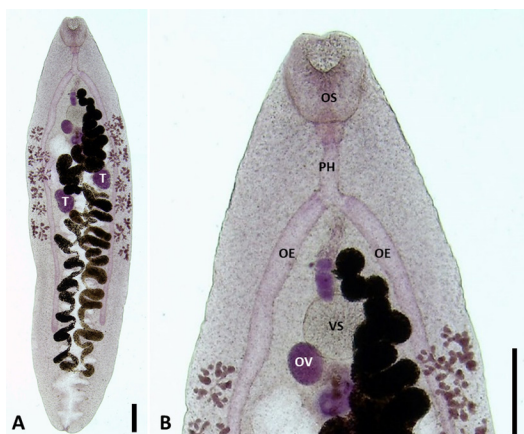


Fig. 5. *Styphlodora magna* from *Nerodia sipedon pleuralis*. (A) Whole mount showing testes. Scale bar = 1.0 mm. (B) Higher magnification of anterior region of same specimen showing oesophagus (OE), oral sucker (OS), ovary (OV), pharynx (PH), and ventral sucker (VS). Scale bar = 500 μ m.

CESTODA: CYCLOPHYLLIDEA: MESOCESTOIDIDAE

Mesocestoides sp. – the 460 mm SVL host was infected in its intestine with a single tetrathyridial specimen (HWML 119230) of *Mesocestoides* sp. The life cycle is an ongoing enigma, yet to be solved, but may involve at least three hosts, an arthropod first intermediate host, a vertebrate second intermediate host, and a vertebrate definitive host (Rausch 1994). However, McAllister et al. (2018) recently suggested that these tapeworms may develop through a simple two-host life cycle. This tapeworm has been reported from a variety of amphibians and reptiles, including several snakes from the Nearctic realm (Bursey et al. 2012; McAllister et al. 2013). In Arkansas, *Mesocestoides* sp. has been reported from various herpetofauna, including six species of salamanders, five species of frogs and toads, three species of lizards, and three species of snakes (McAllister et al. 2013, 2014a, b). We are unaware of reports of *Mesocestoides* sp. from any species of North American watersnake, although the genus has been documented in three species of *Natrix* watersnakes from Myanmar and Italy (see summary in McAllister et al. 1991).

NEMATODA: ASCARIDIDA: ASCARIDIDAE

Ophidascaris ashi Walton, 1927 – a single *O. ashi* (HWML 119229) was found in the stomach of the same *N. s. pleuralis* above. This nematode (as *O. labiatopapillosa*) has been reported from several amphibians as adult worms and encysted larval stages from Florida, and from at least six species of snakes from Alabama, Florida, Louisiana, and Texas, including *N. s. sipedon* from Michigan (see Baker 1987). Sprent (1988) described *O. ashi* as a new species name for *O. labiatopapillosa* in North American colubrids. The life cycle of *Ophidascaris* spp. is indirect with various snakes as definitive hosts (Sprent 1988). We document *O. ashi* from Arkansas, and from *N. s. pleuralis*, for the first time.

In summary, we provide new records for some helminths from a small sample of *N. s. pleuralis* collected from western Arkansas. Surveys on additional Midland watersnakes from other parts of its range, where they have not yet been examined, will likely yield new host and distributional records for helminths.

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