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Cover Page Footnote
The Arkansas Game and Fish Commission (AG&F) issued Scientific Collecting Permits to CTM and HWR, and the Missouri Department of Conservation issued Wildlife Collector's Permit #17434 to Dr. Thomas J. Fayton (U.S. Fish & Wildlife Service, PA). We thank Drs. Scott L. Gardner and Gabor Racz (HWML) and R. Tumlison (HSU) for expert curatorial assistance, and T. J. Fayton for a pre-review of the ms. Several members of the Hope AG&F office, particularly Eric Brinkman (District Supervisor-Fisheries), assisted with collecting in Hempstead County. This material is based upon work supported by the National Science Foundation under Grant Number DEB 1253129 to MAB.

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Additional Records of Acanthocephalan Parasites from Arkansas Fishes, with New Records from Missouri Fishes

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Running Title: Acanthocephalans of Arkansas and Missouri Fishes

Abstract

Over the last decade, our research consortium has provided information on acanthocephalan parasites of Arkansas vertebrates, including records from some of the state's fishes. Here, we continue to provide data on new geographic and new host records of acanthocephalans from Arkansas fishes. In addition, for the first time, we report records of acanthocephalans for some Missouri fishes. We document 2 new state records as well as 10 new host records for some fish acanthocephalans.

Introduction

Acanthocephalans, as adults, are highly specialized, dioecious parasites of the intestinal tract of all groups of vertebrates, including a great variety of taxa from freshwater fishes. Recently, McAllister *et al.* (2016b) provided an overview of the acanthocephalans of Arkansas vertebrates which included information on those from some of the state's fishes. Here, we continue to document new host and geographic records for acanthocephalans from fishes of the state. In addition, there is little available on acanthocephalans from Missouri fishes (Banks and Ashley 2000, Day *et al.* 2014), so we report here, for the first time, on some acanthocephalans from fishes from the Ozark Faunal Region of the state (see Pflieger 1997).

Methods

Between March 2016 and June 2017, we collected fishes with backpack electroshockers (DC current), dipnets and seines. Fish were placed in habitat water and necropsied within 24 hr. We followed accepted guidelines for the use of fish in research (Use of Fishes in Research Committee 2014). Specimens were overdosed with a concentrated tricaine methanesulfonate solution and measured for total length (TL). A mid–ventral incision from cloaca up to the level of the stomach was made to expose the gastrointestinal tract as well as other internal viscera (including gallbladder and liver) which was removed and placed in a Petri dish containing 0.9% w/v saline. Acanthocephalans found were transferred to Petri dishes containing distilled water overnight to completely evert their proboscides. They were then fixed in 70–95% v/v DNA-grade ethanol, stained with acetocarmine and mounted entire with Canada balsam. Voucher specimens were deposited in the Harold W. Manter Laboratory of Parasitology Collection (HWML), Division of Parasitology, University of Nebraska-Lincoln. Host voucher specimens were deposited in the Henderson State University Museum (HSU), Arkadelphia, Arkansas. We follow Amin's (2013) classification of the Acanthocephala.

Our annotated list of data for fishes harboring acanthocephalans is as follows: host and TL (when available), collection site (latitude and longitude, WGS 84), collection date, HWML accession number (when available), prevalence, intensity, and remarks.

Results and Discussion

The following taxa of acanthocephalans were found in Arkansas and Missouri fishes from their river drainages/basin and counties as follows:

EOACANTHOCEPHALA: NEOECHINORHYNCHIDA: NEOECHINORHYNCHIDAE

***Neoechinorhynchus cylindratus* (Van Cleave, 1911) Van Cleave, 1919**

Hosts and locality: 2 (147, 152 TL mm) *Micropterus dolomieu* (Smallmouth Bass), Little Indian Creek (Meramec River Basin), ***Franklin Co.,*** Missouri (38° 12' 48.5958"N, 90° 54' 08.6862"W), 10 Jun. 2017. HWML 139434; 1 *M. dolomieu* (127 mm TL), Panther

Creek (Meramec River Basin), ***Webster Co.,*** Missouri (37° 28' 46.182"N, 92° 46' 57.7596"W), 12 Jun. 2017.

Prevalence and intensity: 2/2 (100%) Little Indian Creek, 1–2 worms; 1/1 (100%) Panther Creek, 1 worm.

Remarks: The first intermediate host in the life cycle is an ostracod, *Cypria* sp. (Ward 1940). Although *N. cylindratus* has been reported from a variety of fishes (see McAllister *et al.* 2016b), including *M. dolomieu* (Hoffman 1999), this is the first time it has been reported from Smallmouth Bass in Missouri. Banks and Ashley (2000) reported *N. cylindratus* was the most common helminth from Largemouth Bass, *M. salmoides* in Smithville Reservoir, northwest Missouri.

***Paulisentis missouriensis* (Keppner, 1974)**

Host and locality: 1 (95 mm TL) *Campostoma pullum* (Central Stoneroller), Panther Creek (Meramec River Basin), ***Webster Co.,*** Missouri (37° 28' 46.182"N, 92° 46' 57.7596"W), 12 Jun. 2017.

Prevalence and intensity: 1/1 (100%); 1 worm (retained for further study).

Remarks: In the life cycle, larval *P. missouriensis* develop in copepods (Keppner 1974). While this acanthocephalan has been previously reported in Missouri from Creek Chubs, *Semotilus atromaculatus* (Keppner 1974), this is the first report of its occurrence in *C. pullum*. McAllister *et al.* (2015) reported a *Paulisentis* sp. that could not be differentiated from *P. missouriensis* or *P. fractus* Van Cleave and Bangham, 1949 from *Campostoma anomalum* (Central Stoneroller) from Benton Co., Arkansas. It is quite possible that both of these acanthocephalans are synonymous and *P. fractus* would have priority; however, additional study is needed to arrive at that conclusion, including molecular analyses.

PALEOACANTHOCEPHALA: ECHINORHYNCHIDA: ECHINORHYNCHIDAE

***Acanthocephalus* sp.**

Host and locality: 1 (93 mm TL) *Fundulus catenatus* (Northern Studfish), Caddo River at Caddo Gap (Ouachita River drainage), ***Montgomery Co.,*** Arkansas (34° 23' 55.752"N, 93° 37' 17.3712"W), 21 Apr. 2016.

Prevalence and intensity: 1/3 (33%), 1 worm.

Remarks: McAllister *et al.* (2016a) reported cystacanths of an unknown acanthocephalan commonly in liver tissues of 17 of 25 (68%) *F. catenatus* from Crooked Creek, Marion Co., Arkansas. Therefore, we document the first adult acanthocephalan from this host.

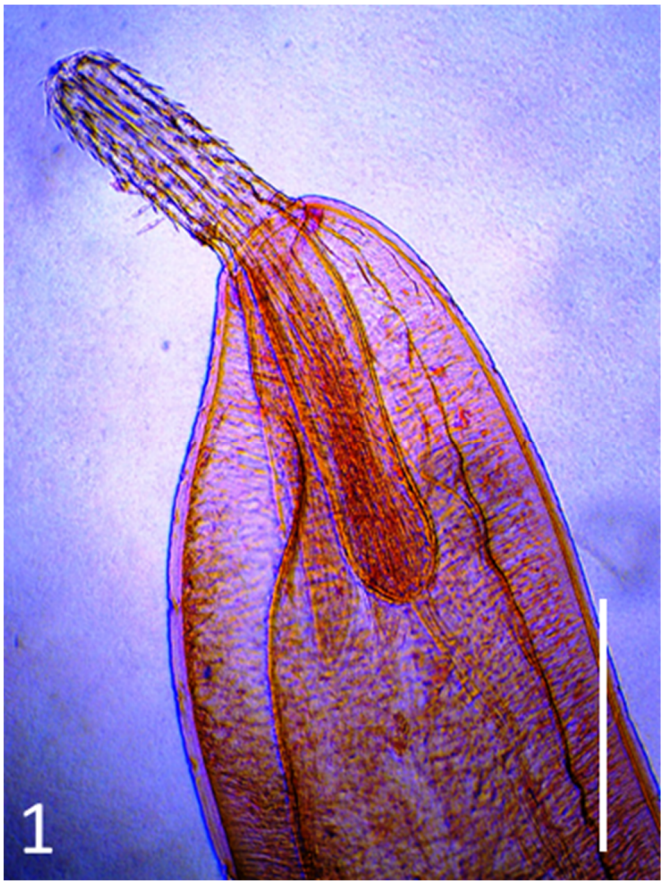

Figure 1. *Acanthocephalus dirus* (HWML 139439) from *Ambloplites constellatus*. Scale bar = 500 µm.

***Acanthocephalus dirus* (Van Cleave, 1931) Van Cleave and Townsend, 1936 (Fig. 1)**

Host and locality: 1 (191 mm TL) *Ambloplites constellatus* (Ozark Bass), Little Red River (White River drainage), ***Van Buren Co.,*** Arkansas (35° 39' 08.8992"N, 92° 19' 12.3774"W), 10 July 2016. HWML 139439.

Prevalence and intensity: 1/1 (100%), 1 worm.

Remarks: This acanthocephalan develops as larvae in isopod intermediate hosts, *Asellus* and *Lirceus* (Seidenberg 1973, Muzzall and Rabalais 1975). *Acanthocephalus dirus* has the widest distribution and diversity of hosts compared to its North American congeners (Amin 1985). McAllister *et al.* (2016b) reported this acanthocephalan from 2 darters, 1 sunfish, and 1 shiner from Arkansas for the first time. We document a new host record for *A. dirus*.

***Acanthocephalus tahlequahensis* Oetinger and Buckner, 1976 (Fig. 2)**

Hosts and localities: 1 (62 mm TL) *Etheostoma caeruleum* (Rainbow Darter), Smyrna Creek (White

River drainage), ***Madison Co.,*** Arkansas (35° 39' 04.8522"N, 92° 55' 13.6122"W), 31 Mar. 2016; 2 (60, 65 mm TL) *Etheostoma flabellare* (Fantail Darter), Panther Creek (Meramec River Basin), ***Webster Co.,*** Missouri (37° 28' 46.182"N, 92° 46' 57.7596"W), 10 Jun. 2017. HWML 139435; 1 (200 mm TL) *Hypentelium nigricans* (Northern Hogsucker), Little Indian Creek (Meramec River Basin), ***Franklin Co.,*** Missouri (38° 12' 48.5958"N, 90° 54' 08.6862"W); 1 (90 mm TL) *Cottus bairdi* (Mottled Sculpin), Bennett Spring (Osage River Basin), ***Laclede Co.,*** Missouri (37° 42' 6.5082"N. 92° 50' 24.8418"W), 11 Jun. 2017.

Prevalence and intensity: 1/1 (100%) *E. caeruleum*; 2/7 (29%) *E. flabellare*; 1/1 (100%) *H. nigricans*; 1/1 (100%) *C. bairdi*. All hosts with 1 worm each.

Remarks: Although the life cycle has not yet been demonstrated for this acanthocephalan, like other members of the genus, larval *A. tahlequahensis* are thought to occur in isopods (Hoffman 1999). This acanthocephalan was originally described from Oklahoma in the Illinois River drainage from 2 darters and 2 cyprinids (Oetinger and Buckner 1976). McAllister *et al.* (2014b, 2015a, 2016b) extended the host (family) range of *A. tahlequahensis* by reporting it in Arkansas from fishes of the families Centrarchidae, Cottidae (*C. carolinae*, Banded Sculpin) and Ictaluridae. We document additional hosts for *A. tahlequahensis*, including the first time from a catostomid fish, as well as a new state record for Missouri.

POMPHORHYNCHIDAE

***Pomphorhynchus bulbocolli* Linkins *in* Van Cleave, 1919 (Fig. 3)**

Hosts: 2 (57, 95 mm TL) *Aphredoderus sayanus* (Pirate Perch), Spring Mill at Big Spring (White River drainage), ***Independence Co.,*** (35° 49' 41.6856"N, 91° 43' 27.3822"W), 23 Apr. 2016; 2 (58, 62 mm TL) and one (63 mm TL) *Etheostoma spectabile* "complex" (Ozark Darter n. sp.), same locality, 23 Apr. 2016 & 8 Jun. 2017. HWML 139436; 1 (110 mm TL) *Luxilus zonatus* (Bleeding Shiner), same locality, 8 Jun. 2017. HWML 139437–139438.

Prevalence and intensity: 2/11 (18%) *A. sayanus*, 1 worm each; 2/6 (33%) and 1/11 (9%) *E. spectabile* complex, 2 worms each, 1 worm; 1/1 (100%) *L. zonatus*, 1 worm.

Remarks: In the life cycle, larvae occur in amphipods (Jensen 1953). We (McAllister and Robison *unpubl.*) have observed that the *E. spectabile* complex darter noted above with *P. bulbocolli* also contained *Hyalella azteca* amphipods in their gut. This acanthocephalan is widely-distributed in North American freshwater fishes (Amin 1987, Hoffman 1999) and show little host specificity. However, McAllister *et al.* (2016b) was the first to report *P. bulbocolli* from Arkansas fishes (2 cyprinids) and we add 3 new host records. In addition, McAllister *et al.* (2015b) reported *P. bulbocolli* from a Midland watersnake (*Nerodia sipedon pleuralis*) from the same site noted herein that was considered an artifact of its piscivorous diet.

Figures 2–3. (2) *Acanthocephalus tahlequahensis* (HWML 139435) from *Etheostoma flabellare*. Scale bar = 250 µm. (3) *Pomphorhynchus bulbocolli* (HWML 139437) from *Luxilus zonatus*. Note distal bulb (arrows). Scale bar = 500 µm.

ILLIOSENTIDAE

***Leptorhynchoides* sp. (Fig. 4)**

Host and locality: 5 (81.0 ± 5.6, 72–86 mm TL) *Lepomis marginatus* (Dollar Sunfish), Beard Lake (Red River drainage), ***Hempstead Co.,*** Arkansas (33° 41' 46.8996"N, 93° 56' 33.6336"W), 30 Jun. 2017. HWML 139433.

Prevalence and intensity: 5/7 (71%), 3.6 ± 2.2, 1–7 worms.

Remarks: Leptorhynchoides acanthidion Steinauer and Nickol, 2014 was reported from *L. marginatus* from Louisiana (Steinauer and Nickol 2014). Unfortunately,

because ours were juvenile specimens, we were not able to provide a specific identity. However, this finding represents the first report of an acanthocephalan parasite from this host in Arkansas.

***Leptorhynchoides aphredoderi* Buckner and Buckner, 1976 (Fig. 5)**

Host and locality: 3 (41, 47, 52 mm TL) *A. sayanus*, Locust Bayou at US 278 (Ouachita River drainage), ***Calhoun Co.,*** Arkansas (33° 33' 27.5034"W, 92° 40' 32.9124"N), 10 Jul. 2016. HWML 139441.

Prevalence and intensity: 3/5 (60%), 1, 2, and 3 worms.

Remarks: Larval *L. aphredoderi* occur in amphipods and larvae less than 30 days old and become encysted in mesenteries of fish (Hoffman 1999). McAllister *et al.* (2014a) reported a *Leptorhynchoides* sp. similar to *L. aphredoderi* from *A. sayanus* from the Rolling Fork River, Sevier Co., Arkansas. However, we document the first definitive report of this acanthocephalan from the state (see McAllister *et al.* 2016b). Buckner and Buckner (1976) originally described this acanthocephalan from *A. sayanus*, Redbreast Sunfish (*Lepomis auritus*) and Spotted Sunfish (*Lepomis punctatus*) from Louisiana. In addition, Buckner (1991) reported *L. aphredoderi* from Warmouth (*Lepomis gulosus*) and *A. sayanus* from Alabama. We document the first report of *L. aphredoderi* from west of the Mississippi River.

Figures 4–5. (4) *Leptorhynchoides* sp. (HWML 139433) from *Lepomis marginatus*. Scale bar = 500 µm. (5) *Leptorhynchoides aphredoderi* (HWML 139441) from *Aphredoderus sayanus*. Scale bar = 250 µm.

Compared to what we know about the acanthocephalans of Arkansas fishes, information on those from Missouri fishes is mostly lacking (Hoffman 1999). Missouri supports more than 200 species of fishes (Pflieger 1997) and many have never been surveyed for acanthocephalans or even helminths in general. Here we have attempted to augment that with some new records but additional work is badly needed.

In summary, we document 10 new host records for acanthocephalans from fishes from the Ouachita, Red, and White river drainages in Arkansas and the Meramec and Osage river basins in Missouri. We also document a new state record for an acanthocephalan found in 3 fish from Missouri and report *L. aphredoderi* from Arkansas for the first time. Additional surveys will undoubtedly increase our knowledge of these parasites in Arkansas and Missouri.

Acknowledgments

The Arkansas Game and Fish Commission (AG&F) issued Scientific Collecting Permits to CTM and HWR, and the Missouri Department of Conservation issued Wildlife Collector's Permit #17434 to Dr. Thomas J. Fayton (U.S. Fish & Wildlife Service, PA). We thank Drs. Scott L. Gardner and Gabor Racz (HWML) and R. Tumlison (HSU) for expert curatorial assistance, and T. J. Fayton for a pre-review of the ms. Several members of the Hope AG&F office, particularly Eric Brinkman (District Supervisor-Fisheries), assisted with collecting in Hempstead County. This material is based upon work supported by the National Science Foundation under Grant Number DEB 1253129 to MAB.

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