Species of *Ligictaluridus* (Monogenoidea: Dactylogyridae) Parasitizing Large Catfishes (Siluriformes: Ictaluridae) from Arkansas, Oklahoma, and Texas

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Abstract: Three species of *Ligictaluridus* (Monogenoidea: Dactylogyridae) were found on large ictalurid catfishes in Arkansas, Oklahoma, and Texas. We provide the second report of *Ligictaluridus michaelalicea* from the gills of the Flathead Catfish (*Pylodictis olivaris*) but the first report from Lake Texoma, Oklahoma, and the Mississippi River, Arkansas. The species, originally described in 2018 from the upper Mississippi River in Iowa and Wisconsin, appears to be restricted to *P. olivaris*. *Ligictaluridus michaelalicea* was found in mixed infections with *L. mirabilis* and *L. pricei* on *P. olivaris*. We also document new distributional records for *L. mirabilis* and *L. pricei* on Blue Catfish (*Ictalurus furcatus*) from Lake Texoma, and Channel Catfish (*Ictalurus punctatus*) from Lake Texoma and the Verdigris River, Oklahoma. We suggest that previous records of *L. floridanus* from Arkansas, Oklahoma, and Texas be considered as *L. mirabilis*.

Introduction

Three species of large catfishes, Blue Catfish, *Ictalurus furcatus* (Lesueur), Channel Catfish, *Ictalurus punctatus* (Rafinesque), and Flathead Catfish, *Pylodictis olivaris* (Rafinesque) (Siluriformes: Ictaluridae), are native to the Mississippi, Mobile, and Rio Grande drainages of the United States and northeastern México (Page and Burr 2011). They are piscivorous predators found in large streams, rivers, and lakes, and have in recent decades been widely introduced along the Atlantic Slope and western drainages for sport and commercial fish management (Jackson 1999). General life history information for these species has been compiled in two International Ictalurid Symposia (Irwin et al. 1999; Michaletz and Travnichek 2011).

The parasites of these three species of catfish are fairly well known, including the Monogenoidea (Klassen and Beverley-Burton 1985; Hoffman 1999; Leis et al. 2018). To date, three species of monogenes of the genus *Ligictaluridus* Beverley-Burton, 1984, (Klassen and Beverley-Burton, 1985), have been reported from *I. furcatus*, *I. punctatus*, and *P. olivaris*, including *L. floridanus* (Mueller, 1936), *L. mirabilis* (Mueller, 1937), and *L. pricei* (Mueller, 1936) (Beverley-Burton 1984; Klassen and Beverley-Burton 1985; Hoffman 1999). Due to relatively few records, particularly from *I. furcatus* and for *P. olivaris*, previously undetected new species are possibly yet to be discovered, such as *L. michaelalicea*

Leis, Easy, MacLean, and Cone, 2018, recently described *P. olivaris* from the upper Mississippi River, Wisconsin and Iowa (Leis et al. 2018). *Ligictaluridus bychowskyi* (Price and Mura, 1969) has only been reported once, from *I. punctatus* in Louisiana (Price and Mura 1969), indicating need for further distributional studies. Also, there are questions remaining that concern the validity of early records of *L. floridanus* and *L. mirabilis*, often considered conspecific, until revisionary studies conducted by Klassen and Beverley-Burton (1985), particularly in the Mississippi River and western portion of the range of these catfishes (Leis et al. 2018). Here, we report on three species of *Ligictaluridus* found on the gills of *I. furcatus*, *I. punctatus*, and *P. olivaris* from Arkansas, Oklahoma, and Texas.

Methods

Host specimens were collected with a trawl in the Mississippi River, Arkansas, back-pack electrofisher from South Concho River, Texas, and with gill nets and boat-electrofisher from Lake Texoma and Verdigris River, Oklahoma, respectively. We followed accepted guidelines for the use of fish in research (AFS 2014). The fish were placed on ice and, if alive, were overdosed in a concentrated chloretone solution; all were subsequently preserved in 10% formalin (McAllister et al. 2015). Specimens were measured for total length (TL). Gills were removed from the fish and examined for ectoparasites under a stereomicroscope at 20–30×. Parasites, picked directly from the gills of their hosts with minuten nadeln, were mounted in Grey and Wess medium stained with Gomori's trichrome (Kritsky et al. 1978). Observations and measurements were made from digital images taken with an Accu-scope Ecelis HDS camera mounted on an Accu-scope LED series phase-contrast microscope. Measurements of haptoral sclerites, in micrometers (μm), and identifications of monogenes were based on Klassen and Beverley-Burton (1985), Klassen et al. (1985), and Leis et al. (2018). The curved male copulatory organ was measured as a straight line extending between the two most distant points of such structures. Ranges are followed by means and number of specimens measured in parentheses. Numbering of haptoral hooks follows Mizelle (1936). Prevalence, intensity, and range of infection were calculated according to Bush et al. (1997).

Voucher specimens of parasites were deposited in the Harold W. Manter Laboratory of Parasitology (HWML), University of Nebraska, Lincoln, Nebraska. Photovouchers of the hosts were deposited in the Henderson State University Museum (HSU), Arkadelphia, Arkansas.

Results

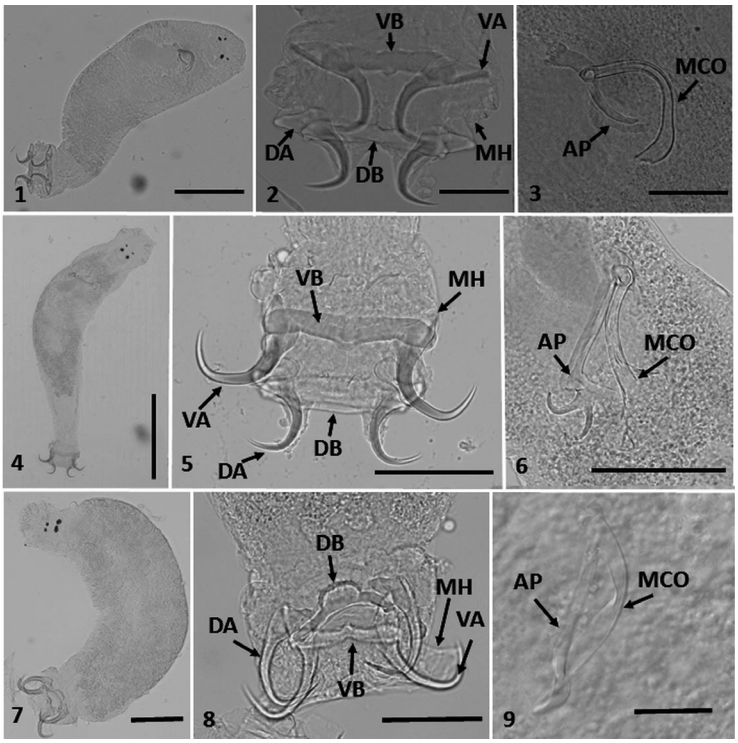
In this study, three species of Monogenoidea were found on the gills of *I. furcatus*, *I. punctatus*, and *P. olivaris*, including a recently described species that we provide morphological and mensural data on as follows:

Class Monogenoidea Bychowsky, 1937
Family Dactylogyridae Bychowsky, 1933
Genus *Ligictaluridus* Beverley-Burton, 1984
***Ligictaluridus michaelalicea* Leis, Easy, Maclean, and Cone, 2018**

***Host, localities, and dates collected*:** *Pylodictis olivaris* (one adult, 610 mm TL, host destroyed during necropsy, collected 21 February 2017), Lake Texoma in the vicinity of Willis Bridge, Red River Drainage, Marshall County, Oklahoma (33°52'31.9224"N, 96°50'01.2804"W). *Pylodictis olivaris* (2 juveniles, 118, 125 mm TL, hosts destroyed during necropsy, collected 16 October 2015), Mississippi River at Sans Souci Landing, Mississippi River Drainage, Mississippi County, Arkansas (35°39'21.387"N, 89°55'33.4956"W).

Prevalence, intensity, and material deposited: 2 of 3 (67%, 1 and 160 worms) overall; Lake Texoma in the vicinity of Willis Bridge, 1 of 1 (100%), 160 worms, HWML 139470 (1 specimen) and HWML 139471 (19 specimens); Mississippi River at Sans Souci Landing, 1 of 2 [HSU photovoucher, 125 mm TL] (50%), 1 worm, HWML 139472.

Site of infection: gills.



Figures 1-9. Monogeneans from ictalurids. Figs. 1-3. *Ligictaluridus michaelalicea* from *Pylodictis olivaris*, Lake Texoma, Oklahoma (HWML 139471). 1. Whole mount, ventral view. Scale bar = 200 µm. 2. Haptor showing dorsal anchor (DA), dorsal bar (DB), ventral anchor (VA), ventral bar (VB), marginal hook (MH). Scale bar = 50 µm. 3. Male copulatory organ (MCO) and accessory piece (AP). Scale bar = 50 µm. Figs. 4-6. *Ligictaluridus mirabilis* from *Ictalurus punctatus*, Lake Texoma, Oklahoma. 4. Whole mount, ventral view. Scale bar = 200 µm. 5. Haptor showing dorsal anchor (DA), dorsal bar (DB), ventral anchor (VA), ventral bar (VB), marginal hook (MH). Scale bar = 50 µm. 6. Male copulatory organ (MCO) and accessory piece (AP). Scale bar = 50 µm. Figs. 7-9. *Ligictaluridus pricei* from *Ictalurus punctatus*, Lake Texoma, Oklahoma. 7. Whole mount, ventral view. Scale bar = 100 µm. 8. Haptor showing dorsal anchor (DA), dorsal bar (DB), ventral anchor (VA), ventral bar (VB), marginal hook (MH). Scale bar = 50 µm. 9. Male copulatory organ (MCO) and accessory piece (AP). Scale bar = 15 µm.

***Description of specimens* (Figs. 1–3):** With characters of the genus *Ligictaluridus* as diagnosed by Beverley-Burton (1984) and Klassen and Beverley-Burton (1985). Body 490–1020 (752) (n = 20) long, greatest width 140–275 (214) (n = 20). Peduncle 0–100 (26) (n = 20) long, 66–137 (105) (n = 20) wide. Haptor 54–120 (97) (n = 20) long, 120–175 (152) (n = 20) wide. Cephalic lobes poorly developed, each lobe with a group of cephalic glands. Two pairs of eyes, anterior pair smaller and usually farther apart than posterior pair. Pharynx subcircular to ovate; 52–80 (68) (n = 20) long, 46–70 (60) (n = 20) wide. Gut smooth and confluent posteriorly. Two pairs of anchors; composed of solid base with short deep root, elongate superficial root, solid elongate, blade-like shaft curving to a sharp point; similar in shape, dorsal pair slightly smaller than ventral pair. Dorsal anchor 53–67 (60) (n = 20) long (total length measurement "a" of Klassen and Beverley-Burton (1985)-distance from tip of superficial root to curve of blade); greatest width of base 28–38 (34) (n = 20); length from curve of blade to notch between superficial and deep roots (measurement "b" of Klassen and Beverley-Burton (1985) 44–55

(51) (*n* = 20); superficial root 18–29 (22) (*n* = 20) long (measurement "d" of Klassen and Beverley-Burton (1985); distance from tip of blade to curve of blade 21–28 (24) (*n* = 20) long (measurement "e" of Klassen and Beverley-Burton (1985). Ventral anchor 62–76 (68) (*n* = 20) long (total length measurement "a" of Klassen and Beverley-Burton (1985); greatest width of base 33–44 (40) (*n* = 20); length from curve of blade to notch between superficial and deep roots (measurement "b" of Klassen and Beverley-Burton (1985) 47–65 (57) (*n* = 20); superficial root 21–30 (26) (*n* = 20) long (measurement "d" of Klassen and Beverley-Burton (1985); distance from tip of blade to curve of blade 23–30 (26) (*n* = 20) long (measurement "e" of Klassen and Beverley-Burton (1985). Dorsal bar broadly curved to straight with knobs on each end and broad median flange; 80–129 (100) (*n* = 20) long. Ventral bar broadly curved to straight with notched knobs on each end and flap-like median flange; 66–112 (89) (*n* =20) long. Fourteen marginal hooks (7 pairs), similar in size and shape. Each hook composed of solid base, solid slender shaft, sickle-shaped termination provided with opposable piece. Hook lengths: nos. 1, 13–21 (17) (*n* = 16); 2, 15–21 (19) (*n* = 19); 3, 17–23 (20) (*n* = 18); 4, 17–23 (20) (*n* = 15); 5, 17–23 (20) (*n* = 11); 6, 15–20 (18) (*n* = 8); 7, 14–19 (16) (*n* = 8). Copulatory complex composed of male copulatory organ and accessory piece. Male copulatory organ with proximal flap-like base bearing slender, curving tubular shaft, becoming bulbous distally; 61–108 (78) (*n* = 20) total length including base; shaft 49–98 (60) (*n* = 20) long. Accessory piece solid, slender, elongate, broadly curved uniramous rod, 29–54 (35) (*n* = 20) long; accompanied by elongate, lightly sclerotized membranous flap extending along and slightly past entire length. Testis ovoid, post ovarian. Two large prostatic reservoirs near male copulatory organ. Ovary ovoid, near middle of body. Vagina not observed. Vitellaria distributed from pharynx to peduncle.

Remarks: Our specimens of *L. michaelalicea* match the overall description of the monogenean provided by Leis et al. (2018). *Ligictaluridus michaelalicea* is easily distinguished from other species of *Ligictaluridus* by possessing a unique combination of the male copulatory organ becoming bulbous distally and a uniramous accessory piece. We extend the distributional range of the parasite about 900 km south of Lansing, Iowa, into the southern Mississippi River, Arkansas, and about 600 km upstream from the Mississippi River into the Red River, Oklahoma. This species has thus far been found only on *P. olivaris*.

***Ligictaluridus mirabilis* (Mueller, 1937) Klassen and Beverley-Burton, 1985**

Hosts, localities, dates collected, prevalence and intensity, range, specimens deposited: *Ictalurus furcatus,* Lake Texoma in the vicinity of Willis Bridge, Red River Drainage, Marshall County, Oklahoma (33°52'31.9224"N; 96°50'01.2804"W) collected 21 February 2017; 1 of 1 [host destroyed during necropsy; 850 mm TL] (100%, 6 worms) (HWML 139475, 2 slides). *Ictalurus punctatus*, Lake Texoma in the vicinity of Willis Bridge, Red River Drainage, Marshall County, Oklahoma (33°52'31.9224"N; 96°50'01.2804"W), collected 21 February 2017; 2 of 2 [hosts destroyed during necropsy; 310-375 mm TL] (100%, 4 and 72 worms) (HWML 139862, 3 slides). *Ictalurus punctatus*, Verdigris River at McClellan-Kerr lock and dam 17, Arkansas River Drainage, Cherokee County, Oklahoma (35°52'17.3712"N; 95°23'16.4184"W), collected 13 March 2017; 1 of 1 [adult host destroyed during necropsy; not measured] (100%, 12 worms) (HWML 139863, 1 slide). *Pylodictis olivaris*, Lake Texoma in the vicinity of Willis Bridge, Red River Drainage, Marshall County, Oklahoma (33°52'31.9224"N; 96°50'01.2804"W), collected 21 February 2017; 1 of 1 [host destroyed during necropsy; 610 mm TL] (100%, 40 worms) (HWML 139473, 5 slides). *Pylodictis olivaris*, South Concho River at Christoval, Colorado River Drainage, Tom Green County, Texas (31°11'16.386"N; 100°30'0.0792"W), collected 23 July 2018; 1 of 1 [HSU photovoucher; 137 mm TL] (100%, 8 worms**)** (HWML 139859, 2 slides).

Remarks: *Ligictaluridus mirabilis* has been reported mostly on species of *Ictalurus* and *P. olivaris* (Klassen and Beverley-Burton 1985;

Hoffman 1999; Leis et al. 2018); its type host is *P. olivaris* (Mueller 1937). *Ligictaluridus mirabilis* was found in mixed infections with *L. michaelalicia and L. pricei*. This is the first confirmed report of *L. mirabilis* from Oklahoma and Texas, although previous reports of *L. floridanus* from these states are likely based on *L. mirabilis* (see Discussion).

***Ligictaluridus pricei* (Mueller, 1936) Klassen and Beverley-Burton, 1985**

Hosts, localities, dates collected, prevalence and intensity, specimens deposited: *Ictalurus furcatus,* Lake Texoma in the vicinity of Willis Bridge, Red River Drainage, Marshall County, Oklahoma (33°52'31.9224"N; 96°50'01.2804"W), collected 21 February 2017; 1 of 1 [host destroyed during necropsy; 850 mm TL] (100%, 3 worms) (HWML 139476, 2 slides). *Ictalurus punctatus*, Lake Texoma in the vicinity of Willis Bridge, Red River Drainage, Marshall County, Oklahoma (33°52'31.9224"N; 96°50'01.2804"W), collected 21 February 2017; 2 of 2 [hosts destroyed during necropsy; 310, 375 mm TL] (100%, 3 and 24 worms) (HWML 139862, 4 slides). *Ictalurus punctatus*, Verdigris River at McClellan-Kerr lock and dam 17, Arkansas River Drainage, Cherokee County, Oklahoma (35°52'17.3712"N; 95°23'16.4184"W), collected 13 March 2017; 1 of 1 [host destroyed during necropsy; not measured] (100%, 5 worms) (HWML 139864, 1 slide). *Pylodictis olivaris*, Mississippi River at Sans Souci Landing, Mississippi River Drainage, Mississippi County, Arkansas (35°39'21.387"N; 89°55'33.4956"W), collected 16 October 2015; 1 of 2 [HSU photovoucher; 118–125 mm TL] (50%, 1 worm**)** (HWML 139474, 1 slide). *Pylodictis olivaris*, South Concho River at Christoval, Colorado River Drainage, Tom Green County, Texas (31°11'16.386"N; 100°30'0.0792"W), collected 23 July 2018; 1 of 1 [HSU photovoucher; 137 mm TL] (100%, 2 worms**)** (HWML 139860, 1 slide).

Remarks: *Ligictaluridus pricei* has been found on a wide variety of ictalurid catfishes, including *P. olivaris*, bullheads (*Ameiurus* spp.), madtoms (*Noturus* spp.), and *Ictalurus* spp., over a wide geographical range including Arkansas, Oklahoma, and Texas, and thus appears to be the least host specific species of *Ligictaluridus* (Klassen and Beverley-Burton 1985; Hoffman 1999; Leis et al. 2018). *Ligictaluridus pricei* was found in mixed infections with *L. michaelalicea* and *L. mirabilis*.

Discussion

In a cladistic analysis of *Ligictaluridus*, Klassen and Beverley-Burton (1987) hypothesized that *L. monticelli* (Cognetti de Martiis, 1924) and *L. pricei* are in a clade characterized by a slender, curved male copulatory organ. Because they share these characteristics, we hypothesize that *L. michaelalicea* and *L. bychowskyi*, which were not included in the analysis by Klassen and Beverley-Burton (1987), are also members of this clade, and these four species form a monophyletic group. We consider the bulbous distal terminus of the male copulatory organ of *L. michaelalicea* to be an autapomorphy derived within this clade. Because they share an elongate basal process directed opposite of the shaft of the male copulatory organ and a membrane running the length of the single-rod accessory piece (Price and Mura 1969; Fig. 3, present study), we hypothesize that *L. bychowskyi* and *L. michaelalicea* are sister species. Because *L. monticelli* and *L. pricei* both possess an accessory piece comprising two rod-like parts and an accompanying membrane (Cognetti de Martiis 1924; Klassen and Beverley-Burton 1985), we hypothesize that they are sister species. Leis et al. (2018) reported a segment of the 18S rRNA gene of *L. michaelalicea* was significantly similar (94%) to that of *L. pricei*, but cladistic analysis comparing data based on morphology and rRNA sequences cannot be performed because rRNA data are lacking for other species of *Ligictaluridus*.

We agree with Klassen and Beverley-Burton's (1987) analysis that *L. posthon* Klassen, Beverley-Burton, and Dechtier, 1985, possessing a male copulatory organ with an apomorphic enlarged diameter but lacking distal flairing, is in a clade by itself, and that *L. floridanus* and *L. mirabilis*, possessing an

enlarged diameter and distal flairing of the male copulatory organ, form another more recently derived monophyletic clade.

Species of *Ligictaluridus* are the only monogeneans that typically parasitize the gills of ictalurid catfishes (Klassen and Beverley-Burton 1985; Hoffman 1999). Some species, i.e., *L. bychowskyi*, *L. michaelalicea*, *L. monticelli*, and *L. posthon* have been found on only one host, whereas *L. floridanus*, *L. mirabilis*, and *L. pricei* have been found on several species (Klassen and Beverley-Burton 1985; Hoffman 1999; present study). Because of confusion over the identity of *L. floridanus* and *L. mirabilis* previous to the revisionary study by Klassen and Beverley-Burton (1985), further research is needed to clarify the host, prevalence and geographic distribution of these two species. Although reported several times from the Mississippi River (Hoffman 1999) previous to Klassen and Beverley-Burton (1985), *L. floridanus* has not been found in the drainage since (Leis et al. 2018; present study). We suspect that the natural range of *L. floridanus* is limited to east of the Mississippi drainage, as it was described from Florida (Mueller 1936) but it appears to have been introduced into northeastern México with *I. punctatus* (Rábago-Castro et al. 2014).

Further knowledge of parasites, including monogeneans, may be important in management of catfishes, especially under crowded culture conditions. For example, Rábago-Castro et al. (2014) reported that chronic exposure to *L. floridanus* reduced the mean weight of farmed Channel Catfish. In addition, mortality of *I. punctatus* due to heavy infections of *L. pricei* has been reported in culture ponds (Allison and Rogers 1970) and seems plausible under similar conditions with other species.

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