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Placobdella cryptobranchii (Rhynchobdellida: Glossiphoniidae) on *Cryptobranchus alleganiensis bishopi* (Ozark Hellbender) in Arkansas and Missouri

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?1 ABSTRACT: *Placobdella cryptobranchii* is a rarely collected leech of the Ozark hellbender (*Cryptobranchus alleganiensis bishopi*) in northern Arkansas and southern Missouri, U.S.A. Between October 2002 and August 2005, 58 hellbenders were examined from Eleven Point River (Randolph Co., Arkansas and Oregon Co., Missouri), the north fork of the White River (Ozark Co., Missouri), and the Spring River (Fulton Co., Arkansas). Forty-one of the 58 hellbenders (70.7%) were infested with 1–140 leeches with a mean intensity (±SD) of 8.7 (±22.1) and a relative abundance (±SD) of 6.3 (±18.9). Contingency table analysis and *t*-tests revealed no significant differences in prevalence and mean intensity among various years and localities sampled. Leech size did not substantially change over the time period sampled. The dorsal pigmentation of live specimens of *P. cryptobranchii* is described for the first time.

KEY WORDS: Rhynchobdellida, Glossiphoniidae, *Placobdella cryptobranchii*, *Desserobdella*, Ozark Hellbender, *Cryptobranchus alleganiensis bishopi*, Arkansas, Missouri.

Leech association with hellbenders was first documented by Dundee and Dundee (1965) from the Spring River in Arkansas, U.S.A. Dundee and Dundee (1965) noted that the Ozark hellbender (*Cryptobranchus alleganiensis bishopi*) characteristically harbors numerous unidentified leeches, perhaps an undescribed species. None of the specimens collected by Dundee and Dundee (1965) are available for study. In their tome on the biology of hellbenders, Nickerson and Mays (1973) noted an undescribed species of leech on most *C. a. bishopi* collected from the north fork of the White River in southern Missouri, U.S.A., and documented host attachment and range of leech infestation from 56 *C. a. bishopi* captured on 24 September 1972. Johnson and Klemm (1977) described *Batracobdella cryptobranchii* based on some of the specimens collected by Nickerson and Mays (1973). *Batracobdella cryptobranchii* was subsequently transferred to the genus *Actinobdella* by Sawyer (1986), then to the genus *Desserobdella* by Barta and Sawyer (1990), and most recently to the genus *Placobdella* by Moser et al. (2006).

Placobdella cryptobranchii was known only from its type locality of the north fork of the White River (Ozark Co., Missouri) until Moser et al. (2006) reported it from 2 localities in Arkansas. The only known host of *P. cryptobranchii* is *C. a. bishopi*, and the leech has never been collected free living (Johnson and Klemm, 1977; Moser et al., 2006). This study describes the prevalence and intensity of *P. cryptobranchii* infestation on *C. a. bishopi* from northern Arkansas and southern Missouri and documents the dorsal pigmentation of live specimens for the first time.

MATERIALS AND METHODS

As part of a long-term monitoring program of *C. a. bishopi* by 2 of the authors (B.A.W., K.J.I.), hellbenders

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Table 1. Prevalence, intensity, and size of *Placobdella cryptobranchii* collected from Ozark hellbenders (*Cryptobranchus alleganiensis bishopi*) from the Eleven Point, north fork (White River), and Spring rivers in northern Arkansas and southern Missouri, October 2002–August 2005.

Date	Locale*	*n*	Prevalence (%)	Intensity		Leech length (mm)	
				Range	Mean ± SD	Range	Mean ± SD
October 2002	Eleven Point River, AR	12	50	1–5	2.8 ± 1.8	2.0–14.0	6.1 ± 3.7
	Eleven Point River, MO	5	100	1–9	4.0 ± 3.0	2.5–17.0	5.7 ± 3.4
	North fork (White) River, MO	16	69	1–20	4.1 ± 5.5	1.5–15.0	6.7 ± 3.4
November 2002	Spring River, AR	3	33	14–14	14.0 ± 0.0	3.0–14.0	5.4 ± 3.8
August 2003	Eleven Point River, AR	7	86	1–10	4.8 ± 3.2	3.0–12.0	4.7 ± 2.2
July/August 2004	Eleven Point River, MO	2	50	6–6	6.0 ± 0.0	2.0–5.5	3.6 ± 1.2
	North fork (White) River, MO	3	100	3–13	7.3 ± 5.1	2.0–11.0	5.8 ± 1.9
	Spring River, AR	1	100	140–140	140.0 ± 0.0	3.0–17.0	8.1 ± 3.1
September 2004	North fork (White) River, MO	3	67	2–7	4.5 ± 3.5	4.0–10.0	6.9 ± 2.0
December 2004	Spring River, AR	1	100	39–39	39.0 ± 0.0	2.5–12.0	5.5 ± 2.3
July/August 2005	Eleven Point River, AR	5	80	1–8	4.0 ± 3.2	2.0–11.0	5.8 ± 3.0

* AR, Arkansas; MO, Missouri.

were captured by hand via scuba in October–November 2002, August 2003, July–September, December 2004, and July–August 2005 from the Eleven Point River (Randolph Co., Arkansas; Oregon Co., Missouri), the north fork of the White River (Ozark Co., Missouri), and Spring River (Fulton Co., Arkansas). Owing to the sensitive status of *C. a. bishopi*, exact localities are not given. Host tag number, total length (TL), snout-vent length (SVL), sex, and mass of each hellbender were recorded. Leeches were removed, counted, and measured, and hellbenders were then released.

Prevalences were compared using contingency table analysis (Bhattacharyya and Johnson, 1977), and mean intensities and mean leech lengths were compared by 2-tailed *t*-tests using Microsoft Excel (Microsoft Corporation, Redmond, Washington, U.S.A.) and Welch 2 Sample *t*-tests using R (The R Foundation for Statistical Computing, www.r-project.org). Means are presented followed by standard deviations (SD). Significant differences are indicated where $P \leq 0.05$. Linear correlation analyses were conducted with Microsoft Excel and R. Because only 4 hellbenders were sampled from the Spring River, these individuals were excluded from the analyses.

Leech specimens were relaxed in 5–10% ethanol (added dropwise until the leech no longer reacted to a probe), fixed in 10% buffered formalin, preserved in 70% ethanol, and examined under a stereoscopic microscope. Additional specimens of *P. cryptobranchii* were preserved in 95% ethanol and deposited in the Invertebrate Zoology collections of the National Museum of Natural History, Smithsonian Institution (accession no. 2043004).

RESULTS

Fifty-eight hellbenders were captured, of which 37 were male, 19 were female, and 2 were undetermined juveniles. Captured *C. a. bishopi* varied from 151- to 407-mm SVL, 242- to 606-mm TL, and a mass of 88 to 1,848 g. Males were smaller in mass, SVL, and TL than females. Forty-one of 58 hellbenders (70.7%) were infested with 1–140 leeches, with a mean intensity of 8.7 (±22.1) and relative abundance of 6.3 (±18.9) (Table 1). Leeches ranged in size from 1.5- to 17-mm long and 0.75- to 6-mm wide (Table 1). Thirty-eight of the 53 (71.2%) hellbenders from the Eleven Point and north fork of the White rivers were infested with 1–20 leeches with a mean intensity of 4.3 (±3.8) and relative abundance of 3.1 (±3.8) (Table 1).

One hellbender was captured twice from the Spring River during 2004. On 27 July 2004, this hellbender was infested with 140 leeches, and on 1 December 2004, this hellbender was infested with 39 leeches. Welch 2 Sample *t*-test revealed that leech length in July was significantly longer than in December ($t = 5.78$, 81.3 d.f., $P < 0.005$). At both observations, this hellbender was the most heavily parasitized of those examined, suggesting that certain individuals may be prone to extremely heavy infestation. The single hellbender that was captured twice accounted for over half of the leeches collected in this study. This population was highly aggregated and clearly exhibited a negative binomial distribution, which is characteristic of parasitic infestations; the variance to mean ratio was 58.2.

Contingency table analysis and *t*-tests revealed no significant differences in prevalence and mean intensity among various years sampled for the Eleven Point and north fork of the White rivers; thus, for subsequent analyses, all sample periods were combined for each river. Likewise, no significant differences were detected in prevalence and mean intensity between the leech populations parasitizing hellbenders collected from the Eleven Point and the north fork of the White rivers.

Contingency table analysis and *t*-tests revealed no significant differences in prevalence and mean intensity between leeches parasitizing male and female hellbenders. Leeches parasitizing male hellbenders were significantly larger than those parasitizing females ($t = 3.10$, 151.9 d.f., $P < 0.005$) exhibiting mean lengths of 6.5 ($\pm$3.3) mm (range: 2–17) and 5.2 ($\pm$2.2) mm (range: 1.5–11), respectively.

No correlations were detected between SVL and number or size of leeches for either male or female hellbenders. Likewise no correlation was detected between leech number and leech size.

No substantial change in leech size was observed over the entire sampling period (July–December). Overall the mean leech length was 6.7 mm (Table 1).

Dorsal pigmentation

Dorsum rust, reddish brown with 2 paramedial rows of metameric white prominences; cephalic white patches extending beyond thin nuchal band; whitish caudal sucker with small amount of pigment at center; white patches (typically 3) between white cephalic and white caudal regions (Fig. 1). In ethanol, body pigmentation gradually fades to golden brown.

DISCUSSION

This study characterizes a population of *P. cryptobranchii* on Ozark hellbenders from northern Arkansas and southern Missouri. Prevalence, intensity, and leech size did not change from July through December, suggesting that the leech population remains stable throughout this time period.

That 1 recaptured hellbender was reinfested with 39 leeches between July and December suggests that leeches are being acquired during this time period. Attachment of *P. cryptobranchii* in December 2004 may indicate overwintering on the host. Moser et al. (2005) found seasonal host attachment in *Oligobdella biannulata*. Between late May and early July, *O. biannulata* leaves its salamander host to lay eggs and reattaches between late August and early October, overwintering on the host (Moser et al., 2005). *Placobdella cryptobranchii* may exhibit similar seasonal host attachment, but sampling should be conducted during other seasons to characterize seasonal changes in leech prevalence and intensity, thereby providing a better picture of the life history of this leech.

Johnson and Klemm (1977) found no evidence of dorsal color pattern or metameric markings on preserved specimens of *P. cryptobranchii*. In this study, it was also found that the dorsal pigmentation of *P. cryptobranchii* fades in ethanol. The dorsal pigmentation of *P. cryptobranchii* is similar to *Placobdella phalera* and *Placobdella michiganensis*, since all have whitish anal patches and a whitish nuchal band. Unlike *P. phalera*, *P. cryptobranchii* does not have a dark middorsal line or middorsal black-tipped papillae. *Placobdella cryptobranchii* also has fewer rows of metameric white prominences than *P. michiganensis*. The base color of *P. phalera* and *P. michiganensis* is gray, whereas the base color of *P. cryptobranchii* is rust, reddish brown.

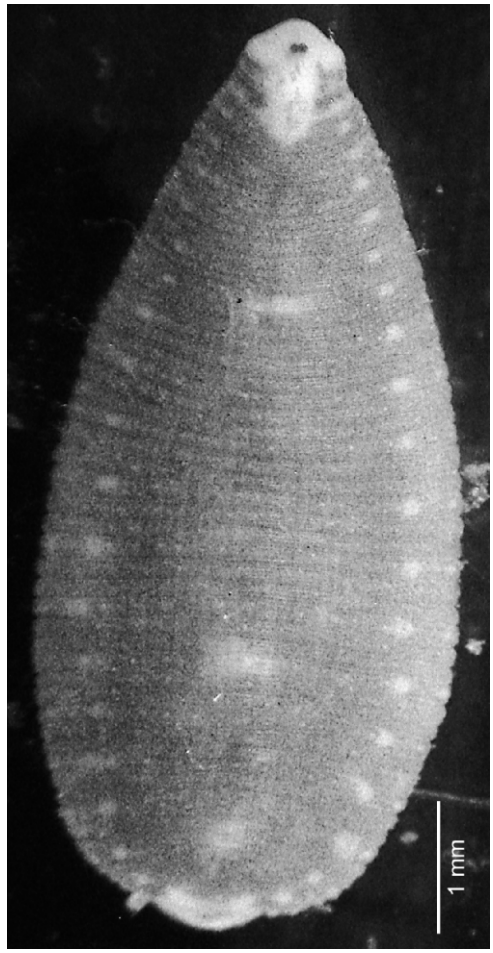

Figure 1. Dorsal surface of *Placobdella cryptobranchii*.

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