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Richard L. Buckner Livingston University

David F. Oetinger Houghton College

Shareen C. Buckner Livingston University

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## **BIOLOGICAL AND MEDICAL SCIENCES**

## PAULISENTIS MISSOURIENSIS KEPPNER, 1974 (ACANTHOCEPHALA) IN CREEK CHUBS, SEMOTILUS ATROMACULATUS, OF SOUTHEASTERN NEBRASKA

Richard L. Buckner<sup>\*</sup>, David F. Oetinger<sup>†</sup>, and Shareen C. Buckner<sup>\*</sup>

School of Life Sciences University of Nebraska-Lincoln Lincoln, Nebraska 68588-0118

Occurrence of the acanthocephalan *Paulisentis missouriensis* Keppner, 1974, in *Semotilus atromaculatus* of Richardson County, Nebraska, was monitored from September 1974 through July 1975. Six hundred nineteen fish were examined of which 78% were infected with 1 to 32 worms. A relatively high prevalence of infection, 72% to 81%, was noted throughout the study. There was no significant difference in the mean density of parasites between the months. Overall mean density was 4.0 (2.9 to 4.6). Fish of all lengths (35 mm to 215 mm standard length) were infected. Both prevalence and mean density increased as fish age/length increased. There were consistently fewer male worms than female worms. The number of males to females best fit a 2:3 ratio as verified by a Chi-square test. Worms appeared to live slightly more than one year with recruitment occurring in early summer and old worms disappearing from the population late the following summer.

#### † † †

The acanthocephalan genus *Paulisentis* comprises two species, *P. fractus* Van Cleave and Bangham, 1949, and *P. missouriensis* Keppner, 1974. Both species were described from the creek chub, *Semotilus atromaculatus*, of North America. Life cycles and larval development have been described for *P. fractus* by Cable and Dill (1967) and for *P. missouriensis* by Keppner (1974). The present study reports on the occurrence of *P. missouriensis* in creek chubs of southeastern Nebraska.

#### MATERIALS AND METHODS

In September and November 1974 and January, April, May, and July 1975 at least 39 Semotilus atromaculatus

were collected each month by seine from Easly, Fourmile, Honey, and Spring creeks (tributaries of the Missouri River) in Richardson County, Nebraska, and examined for Paulisentis missouriensis. Fish were kept alive until necropsied, within 24 hr of capture. The standard length of each fish was recorded and fish were grouped into age/length-classes according to scale annuli, as described by Dinsmore (1962). Worms were placed in tap water to evaginate their proboscides and then fixed in AFA. Number of worms, numbers of males and females, and trunk length (measured by microprojection) of each worm were recorded for each infected fish. Prevalence, intensity, and mean density, as defined by Margolis et al. (1982), were determined for each month's collection and each host's length-class. The reproductive status of each female was determined by microscopic examination for the presence of eggs. These categories were used in evaluation of the females: 0 = no eggs present, 1 = few eggs present, 2 = moderate number of eggs present, 3 = many eggs present, and 4 = pseudocoelom packed with fully formed eggs. The number of females in each category was recorded for each collection month. Representative worms were stained in Mayer's carmalum, mounted in Canada balsam, and deposited as voucher specimens in the University of Nebraska State Museum, Harold W. Manter Laboratory, Lincoln, Nebraska (HWML 21,630).

## **RESULTS AND DISCUSSION**

A total of 619 S. atromaculatus was examined for P. missouriensis of which 480 (79%) were infected with 1 to 32 worms. Data are summarized in Table I.

The prevalence of *P. missouriensis* remained high throughout the study, ranging from 72% to 81%. The mean density

<sup>\*</sup>Present address: Division of Natural Sciences and Mathematics, Livingston University, Livingston, Alabama 35470.

<sup>&</sup>lt;sup>†</sup>Present address: Department of Biology, Houghton College, Houghton, New York 14744.

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TABLE I. Prevalence, intensity, and mean density  $\pm$  95% confidence interval (CI) for *Paulisentis missouriensis* in *Semotilus atromaculatus* by collection month, 1974–1975.

Month	No. Fish Examined	Prevalence	Intensity	Mean Density ± CI	
Sept.	39	72%	1-28	$2.9 \pm 1.2$	
Nov.	240	78%	1-31	$4.6 \pm 0.8$	
Jan.	40	75%	1-10	$3.4 \pm 1.3$	
Apr.	109	81%	1-22	$4.1 \pm 0.8$	
May	98	81%	1-29	$3.2 \pm 0.9$	
July	93	72%	1-32	$3.6 \pm 1.1$	

ranged from 2.9 to 4.6 with an overall mean density of 4.0. There was no significant difference, as determined by single factor analysis of variance, between mean densities of infection for the collection months (calculated F = 1.68, critical  $F_{0.05} = 2.21$ ).

Fish in all length-classes were infected (Table II). The smallest infected fish was 35 mm long, and the largest was 215 mm long. There was a positive correlation between prevalence of the parasite and the length-class of the host (r = 0.884), i.e., prevalence increased as host length (age) increased. The correlation was significant by a Student's *t*-test (calculated t = 3.286, critical  $t_{0.05} = 3.182$ ).

TABLE II. Prevalence, intensity, and mean density  $\pm 95\%$  confidence interval (CI) for *Paulisentis missouriensis* in *Semotilus atromaculatus* by host's age/length-class.

Fish Length- Class	Standard Length (mm)	No. Fish Examined	Prevalence	Intensity	Mean Density ± CI
0	20-39	7	43%	1-2	$0.6 \pm 0.7$
I	40-89	345	75%	1-24	$3.3 \pm 0.5$
II	90-114	151	79%	1-29	$4.5 \pm 0.9$
III	115-134	76	82%	1-32	$5.3 \pm 1.5$
IV+	>135	40	90%	1-29	$5.4 \pm 1.9$

Single factor analysis of variance indicated a significant difference between the mean densities of infection for the various length-classes (calculated F = 4.42, critical  $F_{0.05}$  = 2.37). The mean density ranged from 0.6 in length-class 0 to 5.4 in lengthclass IV+. The mean densities and their confidence limits are compared in Figure 1. Mean density increases as host length increases; however, there is overlap of the confidence limits for all length-classes except class 0. Mean densities were compared using Duncan's new multiple range test. There was no significant difference between the mean densities of length-classes II, III, and IV+. Mean density of length-class 0 differed significantly from that of class I and both length-classes 0 and I differed from the other three length-classes.

Keppner (1974) found infections of *P. missouriensis* in chubs from 39 to 144 mm in total length. In contrast to the present data, he noted that fish less than 60 mm long had a greater prevalence of infection (91%) than fish more than 60 mm (80%). In addition, Cable and Dill (1967) did not find *P. fractus* in chubs greater than 60 mm in length.

Numbers of male and female worms collected during each sample period were compared (Table III). Males and females were present throughout the study and exhibited similar fluctuations in numbers. Males were consistently less numerous than females. For most collections the ratio of males to females appeared to be closest to 2:3. Chi-square tests for goodness



FIGURE 1. Mean densities and confidence limits (stippled bars) of *Paulisentis missouriensis* in different age/length-classes of *Semotilus atromaculatus*.

TABLE III. Observed numbers of male and female *Paulisentis missouriensis* by collection month and Chi-square tests for goodness of fit to expected ratios of 1:1 and 2:3 (male: female).

Month	No. Males	No. Females	X <sup>2</sup> for expected 1:1	X <sup>2</sup> for expected 2:3
Seni	54	60	0.316	2.579
Nov.	437	666	47.542†	0.066
Ian.	55	82	5.320*	0.005
Apr.	179	271	$18.808^{\dagger}$	0.009
May	116	197	20.960†	0.119
July	127	207	$19.160^{+}$	0.075
Total	968	1,483	$108.21^\dagger$	0.020

\*Rejection of null hypothesis at the 5% significance level ( $X^2_{0.05} = 3.841$ ).

†Rejection of null hypothesis at the 1% significance level ( $X^{2}_{0.01} = 6.635$ ).

of fit were performed comparing observed frequencies of males to females with expected frequencies of 1:1 and 2:3 (Table III). Observed frequencies of males to females best fit a 2:3 ratio for all months except September.

Male and female acanthocephalans were grouped into length-classes (at increments of 1 mm), and the distribution of the classes was compared through the collection months (Fig. 2). From September through May, worms exhibited a gradual increase in length. In July both large and small worms were found. The scarcity of young worms during any sample month except July indicates that recruitment occurs in early summer. Disappearance of large, older worms in the fall indicates that worms live slightly more than one year.

Assuming that recruitment begins in summer, the mean size of males and females was determined for each sample month, the sample months were ranked from 1 to 6 beginning with July, and coefficients of correlation were calculated (Table IV). The correlation of worm sizes to months was positive and significant (Table IV).

Comparison of the monthly reproductive status of female worms (Table V) provides additional evidence that recruitment begins during the summer. The greatest percentage of

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FIGURE 2. Percentage distributions of male (open bar) and female (solid bar) *Paulisentis missouriensis* by age/lengthclasses from September 1974 through July 1975.



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TABLE IV. Mean trunk length (mm) of *Paulisentis missouri*ensis males and females by month, coefficients of correlation (r), and Student's t.

Months								
Mean Length	July	Sept.	Nov.	Jan.	Apr.	May	r	t*
Males	3.1	3.5	3.6	3.7	4.3	4.7	0.965	7.397
Females	4.0	4.6	4.8	4.9	5.9	6.9	0.949	6.015

\*Critical t 0.05(2)4 = 2.776.

TABLE V. The percentage of female *Paulisentis missouriensis* occurring in each reproductive category during each sample month.

Reproductive		Months							
Category*	Sept.	Nov.	Jan.	Apr.	May	July			
0	8%	35%	50%	5%	0%	68%			
1	53%	30%	38%	6%	3%	0%			
2	27%	26%	11%	37%	4%	3%			
3	11%	9%	0%	43%	32%	10%			
4	0%	0%	0%	<b>9</b> %	61%	19%			

\*0 = no eggs, 1 = few eggs, 2 = moderate number of eggs, 3 = many eggs, and 4 = pseudocoelom packed with fully formed eggs.

young worms (those without eggs or with few eggs) occurred from July through January, whereas older, mature worms (those with many eggs or a pseudocoelom packed with fully formed eggs) predominated during April and May.

### CONCLUSIONS

Paulisentis missouriensis is a common parasite of Semotilus atromaculatus in the creeks sampled in southeastern Nebraska. It occurs in a large portion of the chubs (72% to 81%) throughout the year. Fish were infected with 1 to 32 worms. The prevalence and mean density of infection increases as fish length (age) increases. There are fewer males than females, 2:3 being the most common ratio. Worms live slightly longer than 1 yr with recruitment occurring during the summer.

#### ACKNOWLEDGMENTS

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