

PROPOSED LIFE CYCLE OF *ASCAROPHIS MARINA* (NEMATODA: CYSTIDICOLIDAE) IN ARGENTINE WATERS

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ABSTRACT: The life history of *Ascarophis marina* in Argentina is proposed on the basis of morphometric features of larval and adult specimens, as well as of trophic relationships among invertebrate and vertebrate hosts in the same geographic area. Adult and larval specimens of *A. marina* were found in the alimentary tract of the teleost fishes *Parona signata* (Carangidae) and *Urophycis brasiliensis* (Gadidae). Third-stage larvae occurred in the shrimps *Peisos petrunkevitchi* (Sergestidae) and *Artemesia longinaris* (Penaeidae).

During a parasitological survey of marine vertebrates and invertebrates along the coast of Mar del Plata, Argentina, cystidicolid specimens with very similar features were found to be parasitizing both fishes and crustaceans. Adults and larvae from *Parona signata* (Carangidae) and *Urophycis brasiliensis* (Gadidae) were identified as *Ascarophis marina* (Szidat, 1961). What appeared to be L3 of the same species occurred in *Artemesia longinaris* (Argentine stiletto shrimp) and *Peisos petrunkevitchi* (shrimp) (Crustacea: Decapoda).

The specific goals of the present study were to describe the L3 of this nematode, to compare the third-stage and adult specimens using statistical analysis, and to suggest the possible life cycle of *A. marina*.

MATERIAL AND METHODS

Specimens examined were collected from 2 shrimp (*A. longinaris* [500] and *P. petrunkevitchi* [100]) and 2 fish (*P. signata* [100] and *U. brasiliensis* [10]). Shrimp and fishes were caught by commercial trawlers from Mar del Plata port (38°00'S, 57°30'W), Buenos Aires Province, Argentina. Shrimp were fixed in 10% formalin and dissected under a stereomicroscope. Most of the fish were examined for nematodes immediately after capture; others were kept in a freezer (-18°C) for 1–2 mo before examination. Live nematodes from fish were fixed in AFA, cleared in acetic acid or lactophenol, and stored in 70% ethanol. Larvae were cleared in 50% ethanol-glycerine. Measurements are expressed in millimeters, and the range is followed by the mean in parentheses. Similarities in parameters were analyzed using a univariate analysis of variance (ANOVA). A probability level of 0.05 was considered significant. The normal distribution of the data was verified using the variance-mean relation [$S^2/\bar{x} < 1$ in a normal distribution, (Cansela Da Fonseca, 1966)]. The homogeneity of variance was calculated using Hartley's *F*-max test ($F\text{-max} = S^2\text{max}/S^2\text{min}$), comparing *F*-max value with the tabulated one for a probability of 0.05 and $k = 6$ samples (Gravetter and Wallnau, 1992). For ANOVA, we tested 20 L3 specimens from shrimp, 10 L3 specimens from fishes, and 14 adults (7 males and 7 females). The characters used were: total length (L), maximum width (W), buccal capsule length (BC), total esophageal length (EL), muscular esophagus length (ME), tail length (T) distance of the nerve ring from anterior extremity (NA), distance of the excretory pore from anterior extremity (EA), and the ratios, EL/ME, ME/BC, EA/BC, and NA/BC. Prevalence and mean intensity of infection were calculated for the different hosts. The ecological terms follow the definitions of Margolis et al. (1982) and Bush et al. (1997).

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DESCRIPTION

Ascarophis marina (Szidat, 1961)

(Figs. 1–8)

L3 from invertebrate: (on the basis of 15 specimens from *A. longinaris* and 15 specimens from *P. petrunkevitchi*, with measurements on 10 from each).

Larva, small, filiform, in oval thin-wall capsule. Cuticle with fine annules. Oral opening dorsoventrally elongated (Fig. 1); pseudolabia 2, lateral, well-developed with a prominent conical structure on anterior margin. Cephalic papillae 4; lateral amphids 2. Buccal capsule long. Esophagus divided into muscular anterior and glandular posterior sections. Nerve ring surrounding anterior section of muscular esophagus. Excretory pore posterior to nerve ring. Tail ending as caudal knot; 0.003–0.006 (0.005) long. Rectal cells 3, distinct (Figs. 2–3). Quantitative data in Table I. Precocious development of the genital primordium, without eggs in 2 specimens (Fig. 4).

L3 from vertebrate: (on the basis of 5 specimens from *P. signata* and 5 specimens from *U. brasiliensis*). Small, filiform nematodes unencapsulated in host stomach and intestine. Cuticle with fine annules from anterior end to tail, oral opening dorsoventrally elongated, surrounded by 2 well-developed lateral pseudolabia (Fig. 5); cephalic papillae 4, slender, located ventrolaterally and dorsolaterally; amphids 2. Buccal capsules well developed; esophagus divided into muscular anterior and glandular posterior sections; larger than in invertebrate stage; nerve ring surrounds the anterior section of muscular esophagus (Fig. 6). Excretory pore posterior to nerve ring. Tail ending as caudal knot. Rectal cells distinct. See quantitative data in Table I. Three specimens found in *P. signata* with postequatorial vulva 4.59 (3.66–5.82) from posterior end (Figs. 7, 8). These specimens were larger than the L3 and appear to be the L4 stages.

Adults from fishes: (on the basis of 14 specimens from *P. signata* and *U. brasiliensis*, 7 males and 7 females). Description of the adults was given in an anterior paper (Ivanov et al., 1997). The morphology was similar to L3 specimens, except for the development of male and female genitalia and their larger size. Quantitative data are given in Table I.

Taxonomic summary

Hosts: Definitive host *Urophycis brasiliensis* (Kaup, 1958) (Gadidae) and *Parona signata* (Jenyns, 1842) (Carangidae). Intermediate hosts *Artemesia longinaris* Batte, 1888 and *Peisos petrunkevitchi* Burkenroad, 1945 (Crustacea: Decapoda).

Site of infection: Adults and L3 from vertebrates, stomach; L3 from invertebrates, encapsulated in cephalothorax.

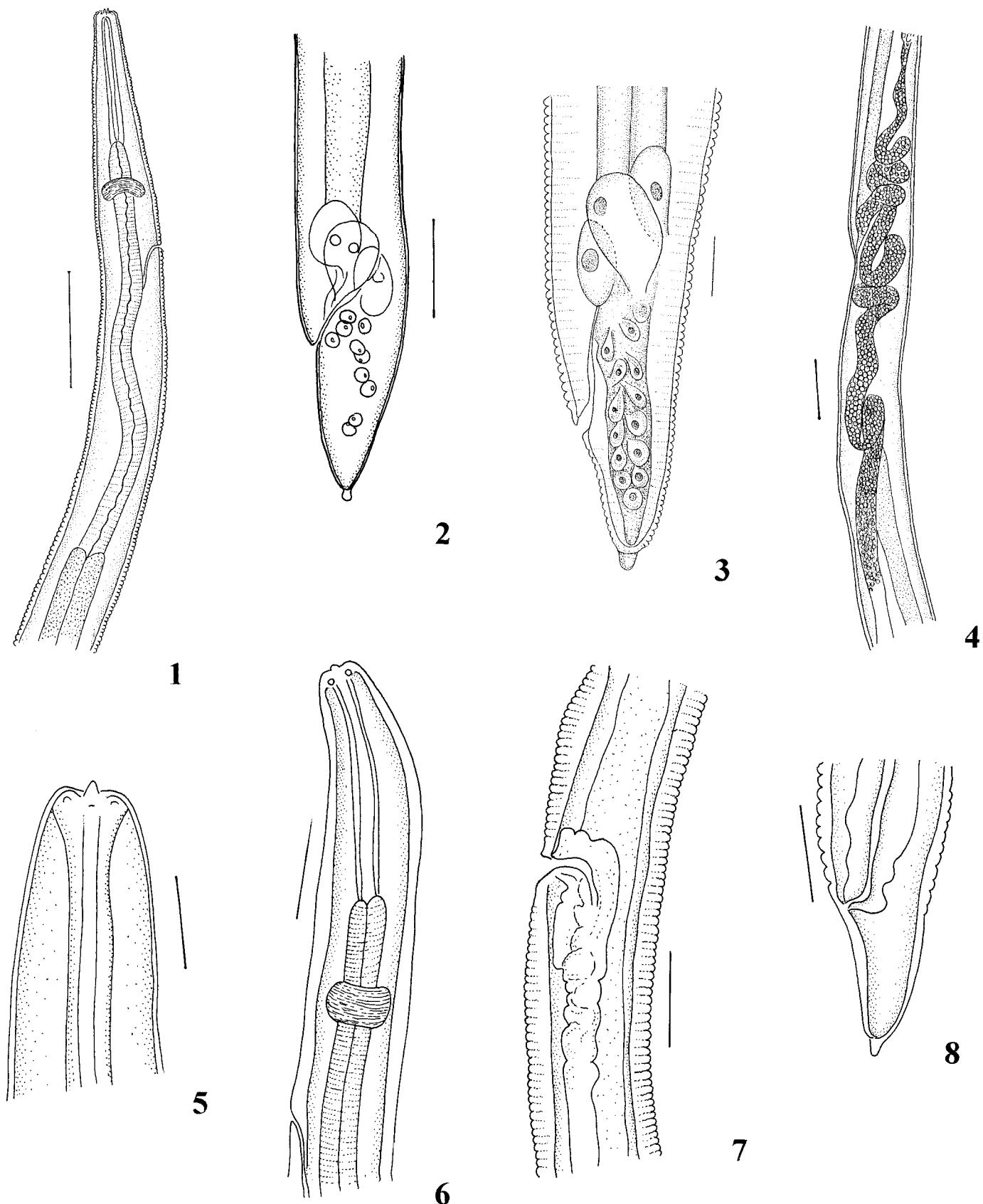
Locality: Mar del Plata, Buenos Aires Province, Argentina (38°00'S, 57°30'W).

Voucher specimens: Deposited in the Museo de La Plata, La Plata, Argentina, Helminthological Collection (MPHC).

Prevalence and intensity: Prevalence (mean intensity): adults, 35% (38) in *P. signata* and 28% (10) in *U. brasiliensis*; L3, 37% (2) in *P. petrunkevitchi* and 9% (2) in *A. longinaris*.

Remarks

Table I summarizes the morphometric data and ANOVA analysis on L3 and adults of *A. marina* from their invertebrate and fish hosts. The



FIGURES 1–8. L3 of *A. marina* in *Ar. longinaris* and *P. petrunkevitchii*. **1.** Anterior part, median view, buccal capsule, nerve ring, and excretory pore. **2.** Posterior end, lateral view in *P. petrunkevitchii*. **3.** Posterior end, lateral view in *Ar. longinaris*. **4.** Larval stage in *P. petrunkevitchii* with internal structures labeled. **5.** Anterior part, median view, buccal capsule, nerve ring, and excretory pore. **6.** Posterior end, lateral view in *P. petrunkevitchii*. **7.** Posterior end, lateral view in *Ar. longinaris*. **8.** Larval stage in *P. petrunkevitchii* with internal structures labeled.

TABLE I. Comparison of measurements of *Ascarophis marina* (larval stage, juveniles, and adults) from different hosts.*

Variable*	L3Al (n = 10)	L3Pp (n = 10)	L3Ps (n = 5)	L3Ub (n = 5)	AdM (n = 7)	AdF (n = 7)	F
L	10.85 (8.58–14.80)	10.56 (7.65–14.19)	14.51 (8.10–19.0)	9.35 (8.25–10.90)	16.52 (10.20–22.60)	38.30 (32.83–44.20)	61.4
W	0.06 (0.05–0.07)	0.06 (0.05–0.07)	0.07 (0.05–0.08)	0.06 (0.04–0.08)	0.07 (0.05–0.09)	0.16 (0.09–0.16)	14.23
BC	0.11 (0.08–0.13)	0.12 (0.10–0.13)	0.11 (0.10–0.12)	0.11 (0.10–0.12)	0.12 (0.11–0.13)	0.13 (0.12–0.14)	1.13†
NA	0.16 (0.15–0.18)	0.16 (0.13–0.17)	0.16 (0.14–0.17)	0.15 (0.13–0.16)	0.17 (0.16–0.18)	0.18 (0.17–0.19)	2.06†
EA	0.20 (0.18–0.23)	0.24 (0.20–0.26)	0.23 (0.21–0.24)	0.22 (0.21–0.22)	0.23 (0.21–0.27)	0.24 (0.22–0.27)	2.61†
EL	1.93 (1.74–2.25)	2.12 (1.59–2.61)	2.49 (2.2–2.85)	1.97 (1.56–2.21)	2.91 (2.04–4.21)	3.78 (2.05–4.91)	9.62
ME	0.35 (0.31–0.41)	0.39 (0.31–0.50)	0.39 (0.34–0.45)	0.31 (0.21–0.42)	0.40 (0.34–0.49)	0.46 (0.34–0.61)	2.45†
T	0.05 (0.04–0.07)	0.07 (0.04–0.08)	0.08 (0.07–0.09)	0.07 (0.06–0.08)	0.12 (0.11–0.13)	0.09 (0.09–0.10)	16.40
EL/ME	5.51	5.40	6.38	6.35	7.27	8.21	
ME/BC	3.18	3.36	3.45	2.82	3.44	3.83	
EA/BC	1.8	1.6	2.09	2	1.91	1.84	
NA/BC	1.45	1.3	1.45	1.36	1.41	1.38	

* L3A1 = L3 in *Artemesia longinaris*; L3Pp = L3 in *Pelos petrunkevitchi*; L3Ps = L3 in *Parona signata*; L3Ub = L3 in *Urophycis brasiliensis*; AdM = male adults in *P. signata* and *U. brasiliensis*; AdF = female adults in *P. signata* and *U. brasiliensis*; I and I; F = F ratio; L = total length; W = maximum width; BC = buccal capsule length; NA = distance of the nerve ring from anterior extremity; EA = distance of the excretory pore from anterior extremity; EL = total esophagus length; ME = muscular esophagus length; T = tail length.

† No statistically significant difference.

variance/mean relation (S^2/\bar{x}) was in all cases <1 and the F -max test values for variance homogeneity were ever less than 29.5 (critical value for $\alpha = 0.05$ and $k = 6$ samples). Characters such as the buccal capsule length (BC), muscular esophagus length (ME), distance of the nerve ring from anterior extremity (NA), and distance of the excretory pore from anterior extremity (EA) showed no significant differences among the samples. Also, the EL/ME and ME/BC ratios were similar in the 6 parasite groups. On the other hand, the total length (L), maximum width (W), total esophageal length (EL), and tail length (T) were significantly different, with the female specimens longer than the males.

In the south Atlantic, 2 other species of *Ascarophis* have been reported, *A. nototeniae* Johnston and Mawson, 1945 and *A. brasiliensis* Magahlaes Pinto et al., 1984. L3 specimens of *A. marina* differ from adults of *A. nototeniae* mainly by having smaller buccal capsule length, and a smaller distance of the nerve ring and excretory pore from anterior extremity. L3 could be distinguished from *A. brasiliensis* by their longer esophagus, and muscular esophagus lengths.

Pelos petrunkevitchi seems to be a better intermediate host for *A. marina* because prevalence of infection in this shrimp (37%) was higher than in *A. longinaris* (9%). The prevalence of *A. marina* in the vertebrate hosts was 35% in *P. signata* and 28% in *U. brasiliensis*. The nematodes showed some seasonality in the invertebrate and vertebrate hosts, with high peaks in spring and summer, and lowest values during the winter. The prevalence of *A. marina* in *P. signata* was highest in spring (99.43%) and summer (72.7%) (September through March). Similarly, the larval forms found in *A. longinaris* showed the highest prevalence in spring (9.88 %) and summer (10.9%). In *P. petrunkevitchi* the highest prevalence was found from January to May (50–56%).

DISCUSSION

The data presented suggest that the L3 specimens found in shrimp are similar to those in fish and are presumed to be conspecific. These larvae develop into adults in fishes, as reported by Ivanov et al. (1997). Therefore, the life cycle of *A. marina* in Argentine waters could follow the scheme suggested by Us-penskaya (1953) for *A. filiformis* and *A. morrhuae* in the Barent Sea, which in both species involves decapods as intermediate hosts. Such an assumption is based on the morphological comparison between L3 and adult specimens and results of the ANOVA test. Experimental infections will be necessary to confirm this proposed life cycle.

Because the fishes *P. signata* and *U. brasiliensis* rely on *A. longinaris* and *P. petrunkevitchi* as an important component of their diet (Ivanov, 1996), it seems reasonable that these crustaceans serve as intermediate hosts of *A. marina* in coastal waters of Mar del Plata. Furthermore, these nematodes showed a similar pattern of seasonality in vertebrate and invertebrate hosts.

In addition to the data presented in the present study, no other species of *Ascarophis* have been reported in the area of Mar del Plata. Moreover, the distributional range of hosts (Cousseau

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genital development, lateral view. Scale lines in millimeters: 1: 0.1; 2, 4: 0.05; 3: 0.015. L3 of *A. marina* in *U. brasiliensis*. 5. Detail of anterior end, pseudolabia, and papillae. 6. Anterior part, median view. Scale lines in millimeters: 5: 0.05; 6: 0.02. L4 of *A. marina* in *Parona signata*. 7. Female vulva, lateral view. 8. Female posterior end, lateral view. Scale lines: 0.05 mm.

and Denegri, 1995) for *Ascarophis nototheniae* and *A. brasiliensis* excludes waters between 34° and 55°S, where Mar del Plata is situated.

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