

Larval nematodes in stomach wall granulomas of smelt *Osmerus eperlanus* from the German North Sea coast

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ABSTRACT: Occurrence of stomach wall granulomas in European smelt was studied at 6 locations along the German North Sea coast. Identification of larval nematodes inhabiting these granulomas is provided for the first time. Three species, isolated by pepsin-HCl digestion, are involved: *Hysterothylacium* cf. *cornutum*, *Cosmocephalus obvelatus* and *Paracuarina tridentata*. 72% of all stomachs examined were affected. The ratio of number of granulomas to number of the 3 larval species free in the mesentery was 1:1.3. Differences in prevalences and intensities were significant among all locations. Granuloma abundance was highest in samples from the Elbe estuary decreasing in the other locations relative to their distance from the Elbe. There was no relationship between the number of larvae encapsulated on the stomach wall and the number of larval *P. decipiens* in the musculature ($r = 0.3$). Host condition factor could not be related to number of granulomas. Smelt appears to be an important transmitter of spiruroid nematode larvae to marine birds in this region.

INTRODUCTION

The European smelt *Osmerus eperlanus* L. is widely distributed in the estuaries of the larger rivers in northern Europe and in neighbouring coastal areas. Data on the general biology of the species in these locations are available in Lillelund (1961) and Möller (1984). The parasitology of the European smelt has been fairly extensively investigated both in the North Sea (Kahl 1936, Jarling 1982, Möller 1984, Möller & Klatt 1990, Kerstan 1991, Lick 1991) and in the Baltic (Voigt 1975, 1981, 1989).

Most published studies on smelt parasites, however, have concentrated on *Pseudoterranova decipiens* as part of the efforts to understand the biology of this conspicuous species which occurs as adults in seals. While much has been done to identify and quantify the species of nematode larvae either in the musculature or body cavity of smelt, no similar information exists on those larvae encapsulated within stomach wall granu-

lomas, in spite of their value for assessing variations in nematode infestations since they represent the most recent acquisitions (Grabda 1978).

Möller & Anders (1986) first documented the occurrence of numerous whitish granulomas on the stomach wall of smelt, although the nature of their contents was not determined. Previously, Jarling (1982) referred briefly to *Thynnascaris* sp. and 'spiruroid' larvae in the intestinal wall of the same host from the Elbe River. Apparently, the full complement of the nematode species of *Osmerus eperlanus* from the North Sea has not been fully investigated nor is there any information on the contents of the stomach granulomas, or on their significance and regional distribution.

The present study was undertaken as part of a wider investigation into the biology and disease occurrence of fishes from the Wadden Sea. The aim was to obtain data on the identity of nematode larvae inhabiting stomach wall granulomas of smelt, and their host relations and occurrence pattern in the locality. Such data would provide additional information on regional differences in parasite occurrence of the species and might prove of value as biological indicators of local smelt populations.

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MATERIALS AND METHODS

Between July and September 1988, 576 specimens of European smelt ranging in total length from 15 to 20 cm were collected from several locations on the lower reaches of 4 rivers which empty into the North Sea (Ems, Weser, Elbe, Eider) and from Süderauë and Heverstrom on the northern part of the German Wadden Sea (Fig. 1). The fish samples originated from trawl

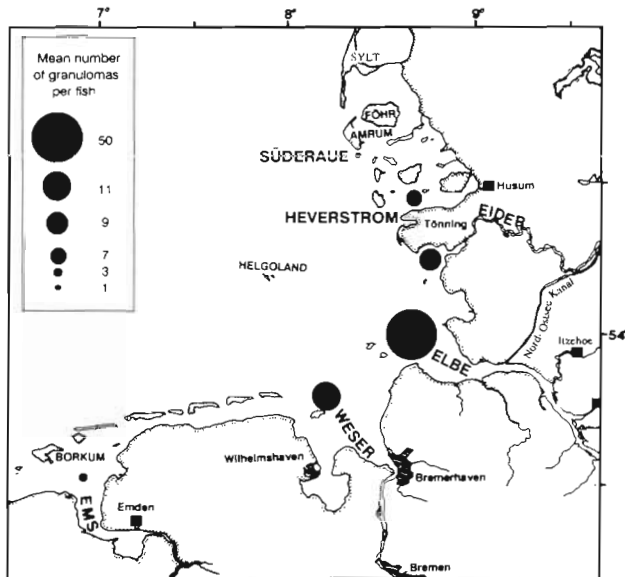


Fig. 1. *Osmerus eperlanus*. German North Sea coast showing locations and geographical variations in abundance of stomach wall granulomas in European smelt (15 to 20 cm). Symbols show mean number of granulomas per fish (including non-infected individuals)

catches. For each fish sample, data on standard length as well as total and gutted weight were recorded. Condition factor was calculated as $K = (\text{gutted weight in g}) \times 1000 / (\text{length in cm})$. Age was determined by otolith reading.

Stomachs dissected from each fish were cut open, cleaned and the wall examined under stereomicroscopy (50 \times) for the presence of granulomas. Relationships between granuloma counts and number of nematode species in the musculature were determined by simultaneous filleting and examination over a candling table.

Larvae were recovered from the granulomas, sometimes still alive, through pepsin-HCL digestion during incubation at 35 °C (Smith & Wootten 1975), washed and subsequently transferred to a mixture of ethanol and glycerine. For each location subsamples of infected specimens were pooled together and digested to determine the proportions of each granuloma-inhabiting larval species.

Due to the wide differences in intensities, the counts

were transformed to stabilize the variance using the equation suggested by Bishop & Margolis (1955)

$$y = \log_{10}(x + 1)$$

where y = transformed value of original observation x . Subsequent statistical analyses were performed on the transformed values. Quantitative terms used in the paper conform to the definitions of Margolis et al. (1982).

RESULTS

Three nematode larval species, *Cosmocephalus obvelatus*, *Paracuarina tridentata* and *Hysterothylacium* sp. cf. *cornutum*, were recovered by pepsin-HCl digestion from stomach wall granulomas (Figs. 2 & 3). On 1 additional occasion, a single *Spinitectus* sp. larva was found encapsulated on the stomach. Larvae of *C. obvelatus* and *P. tridentata* corresponded in morphology and size to parasites described by Plötz (1982a, b) from fishes and birds near Helgoland, Germany.

The following description of *Hysterothylacium* larvae is based on examination of 20 specimens from both stomach granulomas and mesentery. Measurements are in micrometers unless otherwise stated:

Body 1.75 to 1.85 mm long by 70 to 80 wide at greatest width; ratio of greatest width to length 1:10 to 15. Cuticle lacks lateral alae. Oesophagus 165 to 170 long, 2 to 2.5 wide, constituting 8 to 10% of body length.

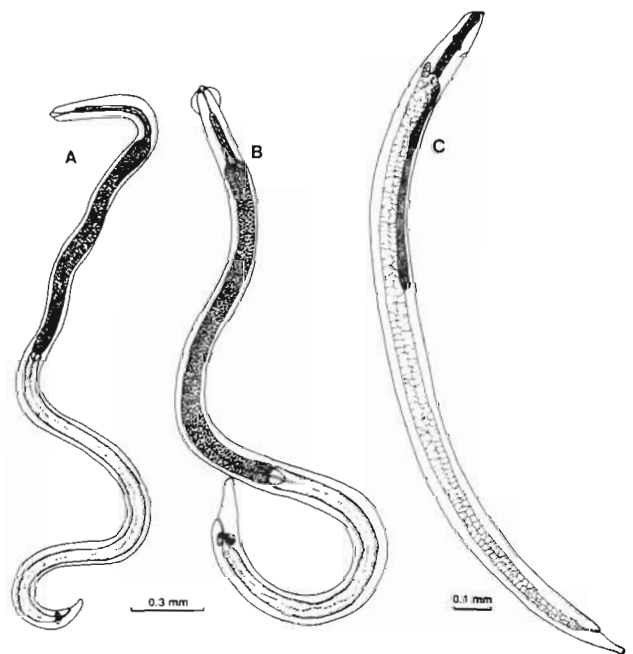


Fig. 2. Nematode larvae from stomach wall granulomas of *Osmerus eperlanus*. (A) *Paracuarina tridentata*. (B) *Cosmocephalus obvelatus*. (C) *Hysterothylacium* cf. *cornutum*

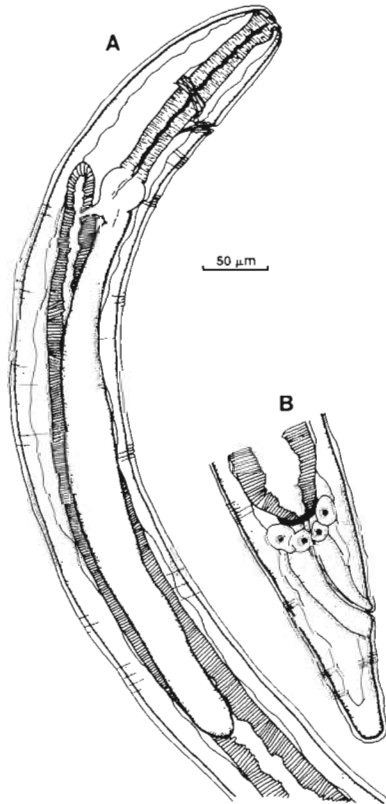


Fig. 3. *Hysterothylacium* cf. *cornutum*. Details of (A) head region and (B) tail

Ventriculus 25 to 30 in diameter; ventricular appendage 510 to 530 long by 2 to 3 wide. Ratio of ventricular appendage length to body length 1:3.5 to 4.0; and to the oesophagus length 1:0.30 to 0.35. Intestinal caecum 20 to 25 long by 10 to 15 wide; ratio of caecal to ventricular

appendage lengths 1:18 to 22; ratio of caecal to oesophagus lengths 1:7 to 10. Distance of nerve ring from anterior end 120 to 125 (or within anterior 60 to 70 % of the oesophagus), breadth 10 to 12. Excretory pore opens at about the nerve ring level or slightly posterior thereto. Tail bluntly rounded without spines, 65 to 72 long or 3 to 4 % of the body length.

The relationship between lengths of intestinal and ventricular caeca are close to those of *Hysterothylacium cornutum* as described by Petter (1969). Berland (1961) found adults of the species in *Thunnus thynnus* from Norwegian waters.

Granulomas on the stomach wall were detected in smelt from all locations sampled. Prevalence and intensity varied greatly among the localities (Table 1). The highest occurrence of infection was found in fish sampled from the Elbe and the lowest in Süderaue. A paired comparison *t*-test showed significant differences ($p < 0.001$) in granuloma counts between the various locations. A negative correlation ($r = 0.76$) could be established between the mean number of granulomas in the stomach wall in the different localities and their distance from the Elbe estuary (Fig. 4). In all locations, the coefficient of dispersion indicates a highly aggregated distribution. Frequency distributions of granuloma counts according to localities based on original untransformed data are shown in Fig. 5.

When granuloma counts on stomach walls were analysed according to host sex, no significant difference could be established. This justified the combination of data from male and female fish for the purposes of subsequent analyses. Within the 15 to 20 cm (total length) class of fish examined, analysis of variance

Table 1. *Osmerus eperlanus*. Occurrence of nematode larvae in stomach wall of European smelt at several locations along the German North Sea coast

Location	Stomach wall					Mesenteries				
	No. examined	No. infected (Prevalence %)	Mean intensity (SD)	Coefficient of dispersion	Range	No. examined	No. infected (Prevalence %)	Mean intensity (SD)	Coefficient of dispersion	Range
Ems	65	43 (66.2)	4.7 (9.9)	20.5	1-54	65	51 (78.5)	6.3 (11.8)	22.2	1-68
Weser	65	46 (70.8)	14.7 (50.4)	173.1	1-344	65	60 (92.3)	19.9 (63.6)	202.9	1-494
Elbe	83	82 (98.8)	48.9 (70.3)	101.2	1-450	65	64 (98.5)	77.7 (117.2)	176.8	1-750
Eider	231	196 (84.8)	9.9 (15.7)	24.8	1-116	231	178 (77.1)	9.2 (10.1)	11.0	1-100
Heverstrom	90	57 (63.3)	10.3 (22.6)	49.4	1-140	90	67 (74.4)	7.4 (7.5)	7.6	1-40
Süderaue	42	15 (35.7)	3.5 (4.1)	4.8	1-16	42	19 (45.2)	3.5 (3.7)	3.8	1-16

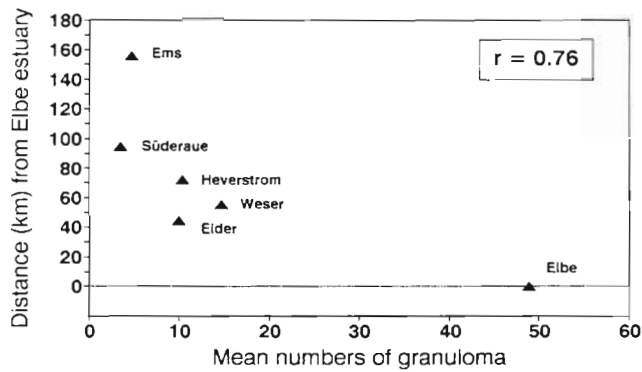


Fig. 4. *Osmerus eperlanus*. Relation between mean number of granuloma in smelt for 6 geographical locations and their distances from the Elbe estuary

showed no significant differences in infestation with either the length or age of host.

The numbers of granulomas on the stomach wall and

of inhabitant larvae of the 3 species free in the mesentery were highly correlated ($r = 0.93$). The ratio of stomach granulomas to larvae free in the mesentery was 1:1.3 (Fig. 6). There seems to be no relation ($r = 0.3$) between the number of larval *Pseudoterranova decipiens* in the musculature and granulomas in the stomach wall (Fig. 7).

Histologically, granulomas could be located in both the mucosa and the muscularis of the stomach wall. Encapsulated nematode larvae are surrounded by granulomatous and fibrotic tissue and varying degrees of cellular infiltration which involve a considerable portion of stomach musculature (Fig. 8). Although the condition factors of smelt differed in the various locations, these differences were not significant and no correlation could be established between granuloma load and condition factor of hosts ($r = 0.3$).

Distribution of granulomas on different parts of the stomach wall showed a concentration (76.1%) on the

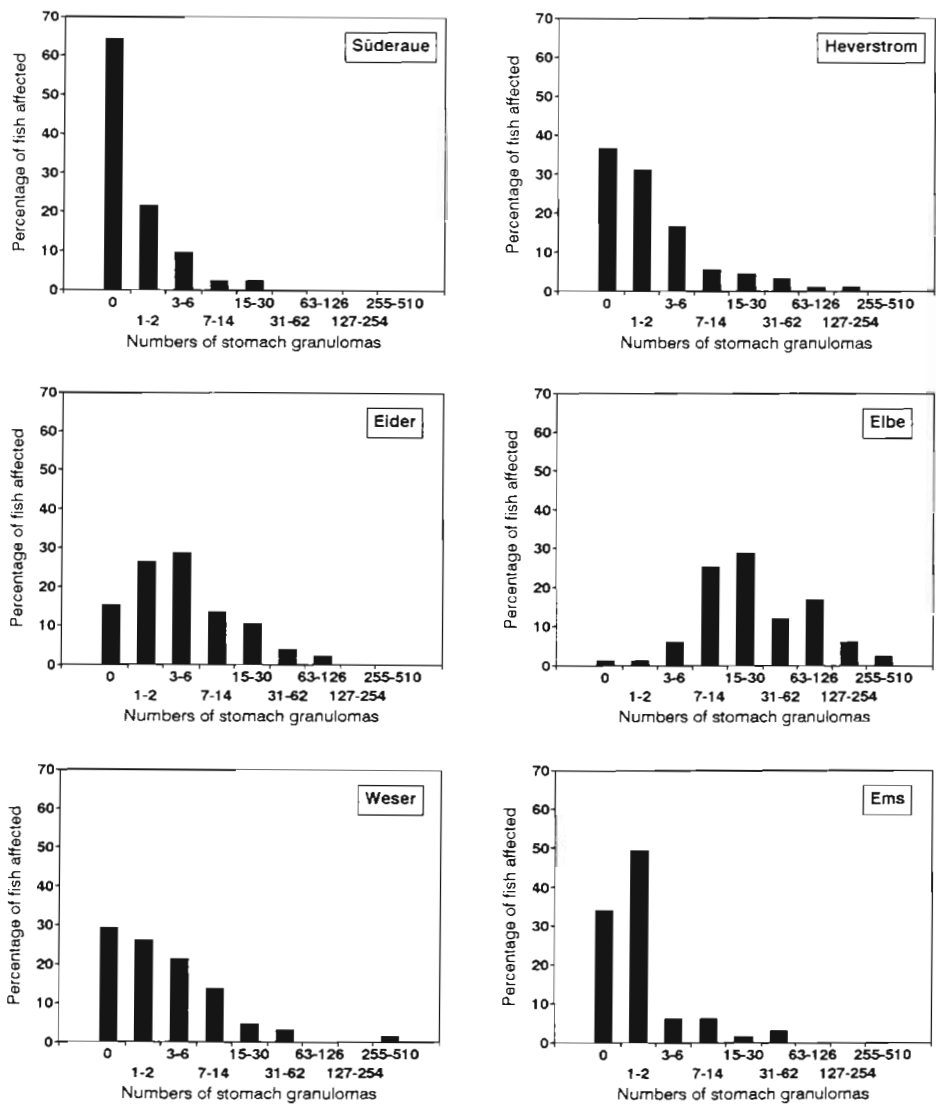


Fig. 5. *Osmerus eperlanus*. Relative frequency distribution of stomach wall granulomas of smelt from various locations along the German North Sea coast

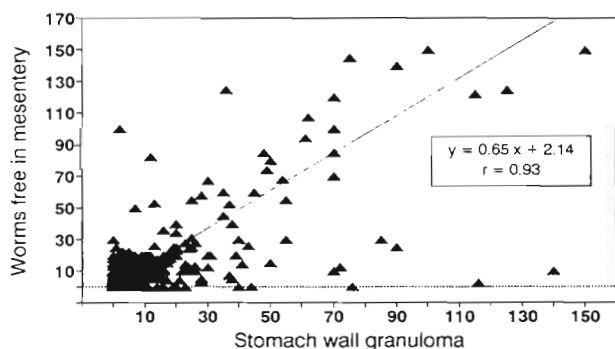


Fig. 6. Relationship between number of larval *Hysterothylacium cornutum*, *Cosmocephalus obvelatus*, *Paracuarina tridentata* free in the mesenteries and number of stomach wall granulomas in 576 smelt *Osmerus eperlanus*

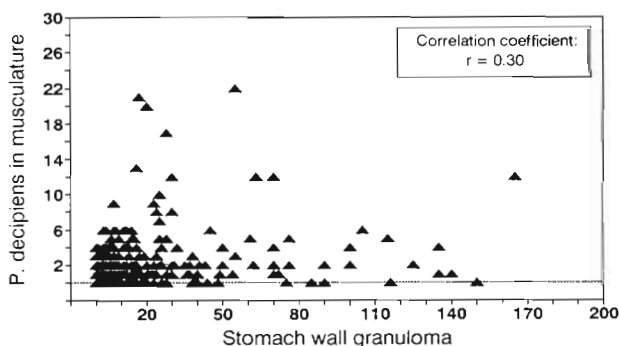


Fig. 7. *Pseudoterranova decipiens*, *Osmerus eperlanus*. Relationship between numbers of *P. decipiens* in the musculature and number of stomach wall granulomas in 576 smelt

pyloric end of this organ. The midsection of the stomach wall was affected by 23.1% of the granulomas whereas only 0.8% were located at the oesophageal end.

DISCUSSION

The present study has established that the European smelt can be infected in high numbers by 3 additional species of larval nematodes in both stomach wall granulomas and mesentery. There has been no report of these larvae in smelt from the Baltic despite extensive investigations into the parasitology of the host in this region (Voigt 1989). It is possible that they do not occur in smelt in this locality or that they have been missed in studies which did not include examination of the stomach wall.

Plötz (1982a, b) studied the biology of the spiruroid nematodes *Cosmocephalus obvelatus* and *Paracuarina tridentata* of Laridae in the North Sea and was able to experimentally infect several species of fish in which the larvae attained the L3 stage. Field observations,

however, did not lead to any indication of the fish species responsible for the transmission to Laridae in nature. It could be concluded from the present study that smelt is an important transmitter of spiruroid nematodes to marine bird species in the area of the Wadden Sea considering their high occurrence both in the mesentery and on stomach granulomas. In a parallel study, Lick (1991) isolated larvae of the 3 species from stomach granulomas in 10 out of 41 fish species examined in the Elbe estuary and confirmed *Osmerus eperlanus* and *Acerina cernua* as the most important intermediate hosts in the locality. Much lower infection rates with the L3 larvae of *C. obvelatus* have been recorded for the closely related rainbow smelt *O. mordax* in Lake Ontario (Wong & Anderson 1982).

Differences in infection rates of first invertebrate intermediate hosts might explain the strong regional variations in granuloma and nematode occurrence in smelt. The inverse relationship of mean granuloma counts with distance from the Elbe would imply that this location serves as a focus for the dissemination of the 3 larval nematode species involved in this study.

The absence of other larval species in granulomas might imply a more rapid migration through the stomach wall. The number of granulomas evidently does not influence the passage of *Pseudoterranova decipiens* into the musculature.

Similar quantitative studies of granuloma occurrence on stomach wall of smelt from other areas of its occurrence are not available for comparison. The condition satisfies most criteria for use as biological tags of local smelt populations: prevalences could be high and differ significantly with localities; the condition is easily recognizable on a specific site on the host; the health of the host is apparently not impaired; and the granulomas persist for some time even after destruction of the larvae.

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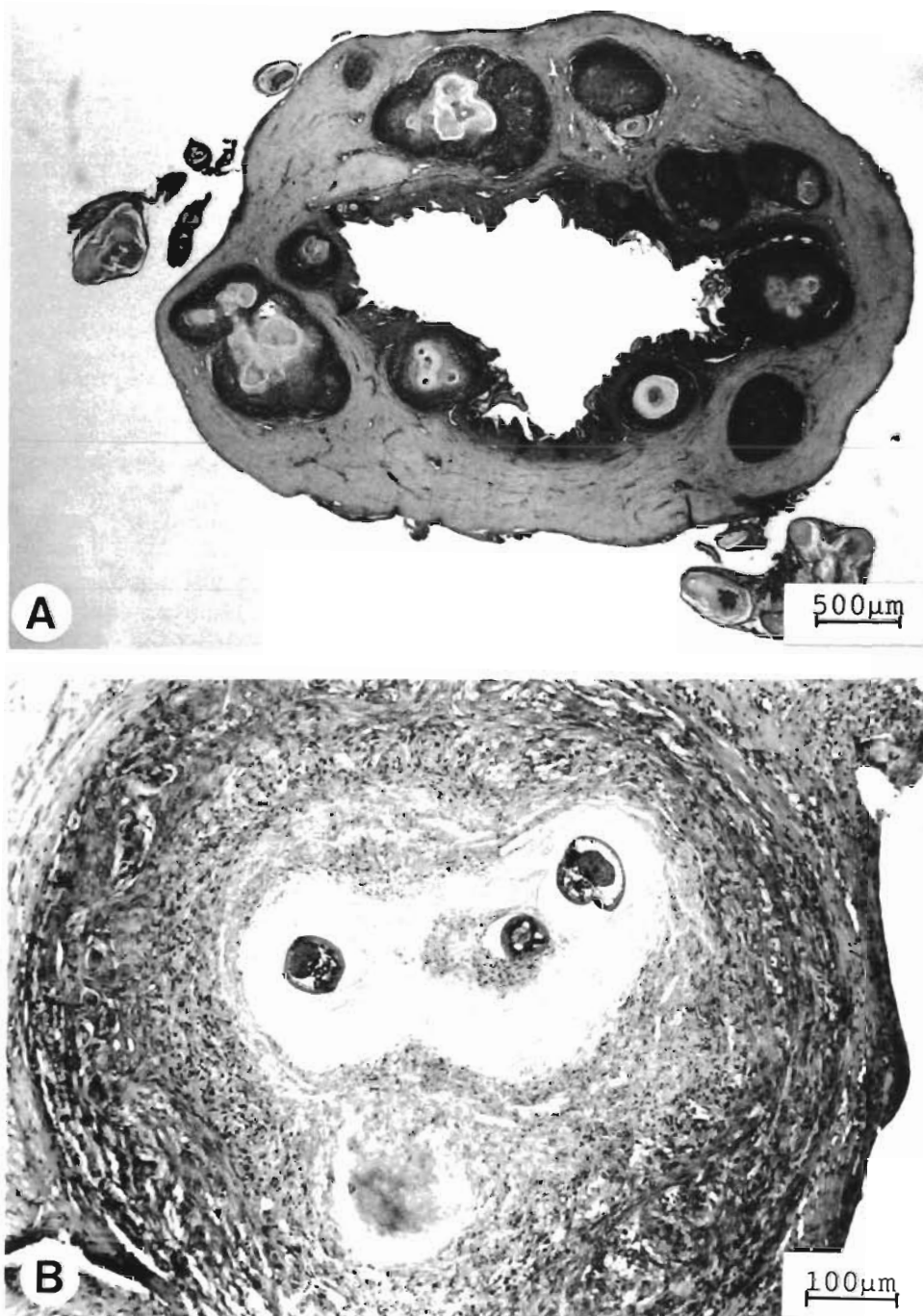


Fig. 8. *Osmerus eperlanus*. (A) Histological section through infected stomach wall of *Osmerus eperlanus*. (B) Cellular infiltration around encapsulated nematode larvae

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