DESCRIPTION AND PREVALENCE OF *Thynnascaris* sp. LARVAE DOLLFUS, 1933 (NEMATODA: ANISAKIDAE) IN *Plagioscion squamosissimus* HECKEL, 1840 FROM VOLTA GRANDE RESERVOIR, STATE OF MINAS GERAIS, BRAZIL

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ABSTRACT

The present work studied helminth parasites of freshwater "corvina" *Plagioscion squamosissimus* from Volta Grande Reservoir, MG, Brazil. Sixty eight fishes with averages of 25.2 cm length and 180.9 g weight were collected with net, bimonthly from December 1995 thru December 1996. Parasites were carefully removed from their cysts that were present in the intestinal mesentery. Specimens were fixed in AFA 65ºC and preserved in alcohol 70% with 5% of glicerine. In camera lucida 21 nematodes were drawned after clarification with acetic acid or Amann lactophenol. Nematode larvae were identified as *Thynnascaris* sp. (Nematoda: Anisakidae). From examined fishes 30 out of 68 presented nematode larvae with a prevalence of 44.1%. The average number of parasites per host was 0.0 to 13.8 and mean intensity of 0.0 to 16.0. Statistical analysis according to Fisher's Exact Test showed that these infections were dependent on the pluviosity and air temperature

Key words: Nematoda, *Thynnascaris* larvae, *Plagioscion squamosissimus*, seasonality, prevalence.

RESUMO

Descrição e Prevalência de larvas de *Thynnascaris* sp. Dollfus, 1933 (Nematoda: Anisakidae) em *Plagioscion squamosissimus* Heckel, 1840 Proveniente do Reservatório de Volta Grande, Estado de Minas Gerais, Brasil

O presente trabalho teve por objetivo o estudo da helmintofauna da corvina de água doce *Plagioscion squamosissimus*, do reservatório de Volta Grande, MG, Brasil. Foram colhidos 68 peixes com comprimento médio de 25,2 cm e peso médio de 180,9 g, com rede de espera, bimestralmente no período de dezembro de 1995 a dezembro de 1996. Os parasitos foram cuidadosamente retirados de seus cistos que estavam aderidos ao mesentério intestinal. Os espécimes foram fixados em AFA a 65ºC e conservados em álcool 70ºGL contendo 5% de glicerina. Após diafanização com ácido acético ou lactofenol de Amann, 21 nematóides foram desenhados em câmara clara. As larvas foram identificadas como *Thynnascaris* sp. (Nematoda: Anisakidae). Das 68 corvinas examinadas, 30 estavam infectadas por esses parasitos com uma prevalência de 44,1%. O número médio de parasitos por hospedeiro foi

de 0 a 13,8 e a intensidade média, de 0 a 16. A análise estatística, de acordo com o Teste Exato de Fisher, mostrou que a sazonalidade foi dependente da pluviosidade e temperatura do ar.

Palavras-chave: Nematoda, larvas de *Thynnascaris*, *Plagioscion squamosissimus*, sazonalidade, prevalência.

INTRODUCTION

Anisakid nematodes are found parasitizing various species of Brazilian fishes. Still, one of the first work to be written on them was *Contracaecum spiculigerum* from *Pimelodella lateristriga* and *Acestrorhamphus* sp. (Travassos *et al*., 1928). The worm may infect piscivorous birds, mammals and fish. Commonly the larval stage is found in fish (Vidal-Martinez *et al*., 1994). *Contracaecum* was related from mesentery of the intestine and estomach of *Astyanax* sp. (Kloss, 1966); *C. fortalezae* from *Scomberomorus cavalla* and *S. maculatus* (Klein, 1973), and *Contracaecum* sp from *Serrasalmus altuvei* (Leão *et al*., 1991).

In marine environment *C. fortalezae* was reported from *Harengula clupeola* (Guimarães & Cristofaro, 1974) and *Contracaecum* sp. from *Macrodon ancylodon* (Vicente & Fernandes, 1978). Other species such as *C. epinepheli*, *C. seriolae*, *C. paralichthydis*, *C. legendrei*, *C. scomberomori* were found in Japan (Yamaguti, 1941a,b; Kagei *et al*., 1970; Moravec *et al*., 1985). Their presence in the cod liver was studied by Rokicki *et al*. (1993).

Prevalence studies in brazilian fishes showed 64.4% in *Hoplias malabaricus* (Fabio, 1982); 32.2% of fishes from Mogi Guassu River (Kohn & Fernandes, 1987); 100% in *Crenicichla lepidota* from Salto Osório Reservoir, Parana State (Kohn *et al*., 1988); 68.9% in *Salminus maxillosus*, *Serrasalmus nattereri*, *S. marginatus* and *Brycon hilarii* (Rego & Vicente, 1988). Valtonen *et al.* (1988) related *C. osculatum* showing 20% prevalence in *Salmo salar* and *Myoxocephalus scorpius*, 16% in *Lota lota* and 15% in *Gadus morhua*. Controversy about taxonomy of *Thynnascaris*, *Contracaecum* and *Hysterothylacium* remain unsettled. Records of *Thynnascaris* larvae (Wooten, 1978; Hurst, 1984; Arthur & Arai, 1980) and *Hysterothylacium* sp larvae (Vicente *et al*., 1985; Moravec *et al*., 1985; Moravec & Sei, 1988; Koie, 1993; Moravec & Nagasawa, 1998; González, 1998) were observed.

In the present work, the authors made a description of *Thynnascaris* sp. larvae Dollfus, 1933 (Nematoda: Anisakidae) from *Plagioscion squamosissimus* collected in the Volta Grande Reservoir, MG, Brazil. Prevalence and seasonality were evaluated.

MATERIAL AND METHODS

Sixty eight specimens of corvina, *Plagioscion squamosissimus* Heckel, 1840 (Sciaenidae) were collected bimonthly with the aid of a net, during a period of one year (12/1995 to 12/1996). This work was developed in the Volta Grande Reservoir, MG, Brazil which presents a flooded area of 195 km^2. Nematode larvae were carefully collected from the intestinal mesentery, dissecated from their cysts and fixed in AFA 65^{o}C for posterior preservation in alcohol 70% with 5% of glicerine. For the observation of the internal structures acetic acid or Amann lactophenol were employed. Some larvae were quicklly imersed in lugol solution. Twenty one specimens were measured (mm) and drawn with the help of a camera lucida and a light microscope. Prevalence (number of infected host/number of examined host) and mean intensity (total number of parasite/number of infected host) were calculated according to Bush *et al*. (1997). To determine if there were significant differences among the prevalence in each month, Fisher's Exact Test was performed according to Pimentel Gomes (1985).

RESULTS

Thynnascaris *sp. Dollfus, 1933 (Nematoda: Anisakidae)*

Elongated larvae with total length of 2,544 mm (2,169 to 2,782) and maximun width of 86.2 mm (80.6 to 104.8). Anterior end presenting two pairs of papillae with a tooth 3.4 mm (3.2 to 4.0) long. Cylindrical esophagus measuring 249.5 mm (168.1 to 276.6) length and 21.3 mm (19.1 to 23.4) width. Esophagean bulb slightly enlarged with 27.3

mm (19.1 to 40.4) width. Nerve ring 129.2 mm (95.7 to 146.8) and excretory pore 135.5 mm (85.1 to 148.9) from the anterior extremity. Very long ventricular appendix pointed to the posterior extremity measuring 765.5 mm (564.5 to 1,032.2) length. A small and enlarged intestinal caecum pointed to the anterior extremity measuring 28.5 mm (21.3 to 42.5) in length was observed. Conical tail with 71.1 mm (63.8 to 82.9) long. Caudal process lacking spines (Figs. 3 to 5).

Remarks

Many times the *Contracaecum* genus as a synonym of the *Hysterothylacium* (Bristow & Berland, 1991) was observed although these two genera are separated in some identification Keys. If the excretory pore is situated subventrally in the mouth cavity it belongs to *Contracaecum*, but if its situated near to the nerve ring it belongs to *Hysterothylacium* (Vicente *et al.*, 1985). To confirm these disagreement some authors identified *C. fortalezae* without considering the excretory pore (Klein, 1973; Vicente & Fernandes, 1978) or Yamaguti (1941a, b). Guimarães & Cristofaro (1974) described the excretory pore distant from the anterior end in *C. incurvum, C. histiophori, C. marinum, C. pagrosoni, C. gracile, C. saba* and *C. fortalezae*. For this reason, in the present paper the authors identified the nematode larvae by the priority law according to Hartwitch (1974).

The present description differs from the cited literature in the total length, width and other characters that are smaller than in the other specimens. Ventricular appendix presented increased length when compared to Kagei *et al*. (1970), Klein (1973), Guimarães & Cristofaro (1974), Vicente & Fernandes (1978) and Moravec *et al.* (1985), but decreased length when compared to Moravec *et al.* (1985) and Rokicki *et al.* (1993). Intestinal caecum showed shorter length than described by Moravec *et al*. (1985), Moravec & Sei (1988) and Hurst (1984). From the paper of Yamaguti (1941a, b) it was not possible to make any comparisons in spite of so different morphology and larval stages. Distance of the nerve ring and excretory pore from anterior extremity presented lower measurements than related by Moravec *et al*. (1985) and Moravec & Sei (1988) (Table 2). To confirm the species of parasite it will be necessary to collect definitive hosts that will encorage future study.

Prevalence

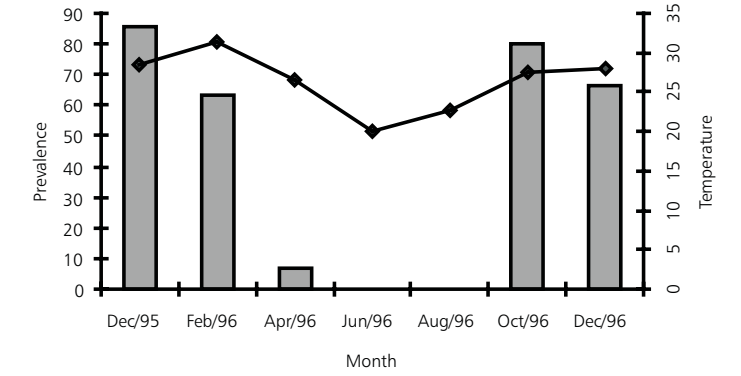
Thirty out of sixty eight examined fishes presented nematode larvae in the internal organs showing a prevalence of 44.1%. The total of 354 larvae were collected from the intestinal mesentery. December 1995 and October 1996 showed higher prevalence of nematodes, respectively 85.7% and 80.0% than observed in February, April and December 1996, respectively 63.6%, 6.7% and 66.7%. In June and August the presence of parasite (Table 1) was not related. These results showed strong relation between temperature and pluviosity in the reservoir region. When the plotted data are put together prevalence related to elevated temperature and pluviosity is evident (Figs. 1a, b; 2a, b). However, mean intensity was 13.8 and 10.6 in February and October 1996 and just one specimen of nematode larvae was collected in April 1996. An interesting fact that may be observed is the great number of infected fishes in December 1995, February, October and December 1996 as shown in Table 1. Statistical analysis according to Fisher's Exact Test showed that these infections were dependent on the pluviosity and air temperature.

DISCUSSION

The first record found in the literarature was *Contracaecum* sp and *Amplicaecum* sp larvae in brazilian fishes (Travassos *et al.*, 1928). Years later reports of *Contracaecum* sp. in the visceral mesentery in traíra (*Hoplias malabaricus*) showed a prevalence of 64.4% and mean intensity of 5.3 (Fabio, 1982). Other observed prevalences of nematode larvae in brazilian native fishes were 29.2% in *Salminus maxillosus* (Kohn & Fernandes, 1987); 100% (Kohn *et al*., 1988); 33.3% in *S. maxillosus*, 100% *Serrasalmus nattereri*, 100% *S. marginatus* and 100% *Brycon hilarii* (Rego & Vicente, 1988).

According to our findings a total prevalence of 44.1% was not higher than that of other studies, but when analized and considered per month, the present work showed up to 85.7% values. Lower prevalences (20%) was also reported of *C. osculatum* from the Baltic Sea (Valtonen *et al.*, 1988).

The authors demonstrated mean intensity of 6.0 that was thus different from the present study (16.0).



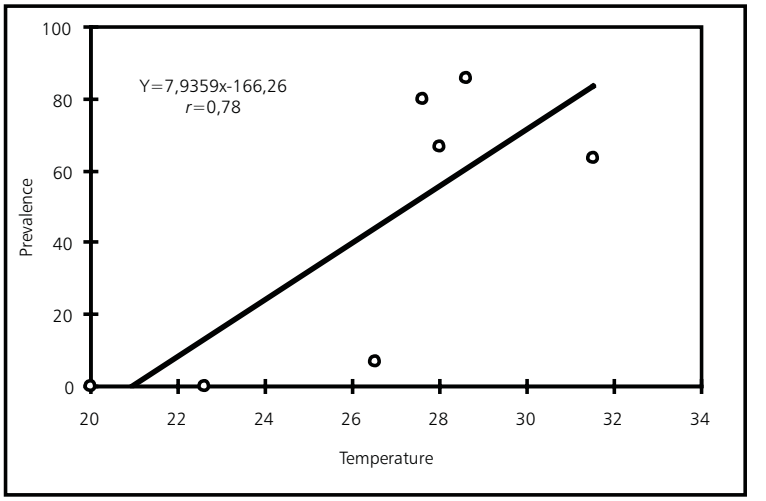


Fig. 1 — (a) Prevalence of *Thynnascaris* sp. larvae from *P. squamosissimus* related to temperature. Bar- prevalence (%); line- air temperature (ºC). (b) Positive correlation between the data.

TABLE 1

Mean values of total length and weight of fishes, number of parasites and prevalence in *P. squamosissimus*, during a period of December 1995 to December 1996. Volta Grande Reservoir, State of Minas Gerais, Brazil.

Month	CF/IF	FL (cm)	W (g)	TNP	MI (V)	P (%)
Dec/95	07/06	19.3	82.1	42.0	7.0 (3-11)	85.7
Feb/96	11/07	25.9	180.2	111.0	15.8 (2-65)	63.6
Apr/96	15/01	33.5	443.2	1.0	1.0	6.7
Jun/96	09/0	37.9	727.0	0.0	0.0	0.0
Aug/96	05/0	25.3	177.2	0.0	0.0	0.0
Oct/96	15/12	26.3	195.0	163.0	13.6 (1-23)	80.0
Dec/96	06/04	29.5	265.8	35.0	8.7 (1-17)	66.7

(CF/IF) collected fish/infected fish; (FL) fish length; (W) fish weight; (TNP) total number of collected parasite; (MI) mean intens and variation (V); (P) prevalence.

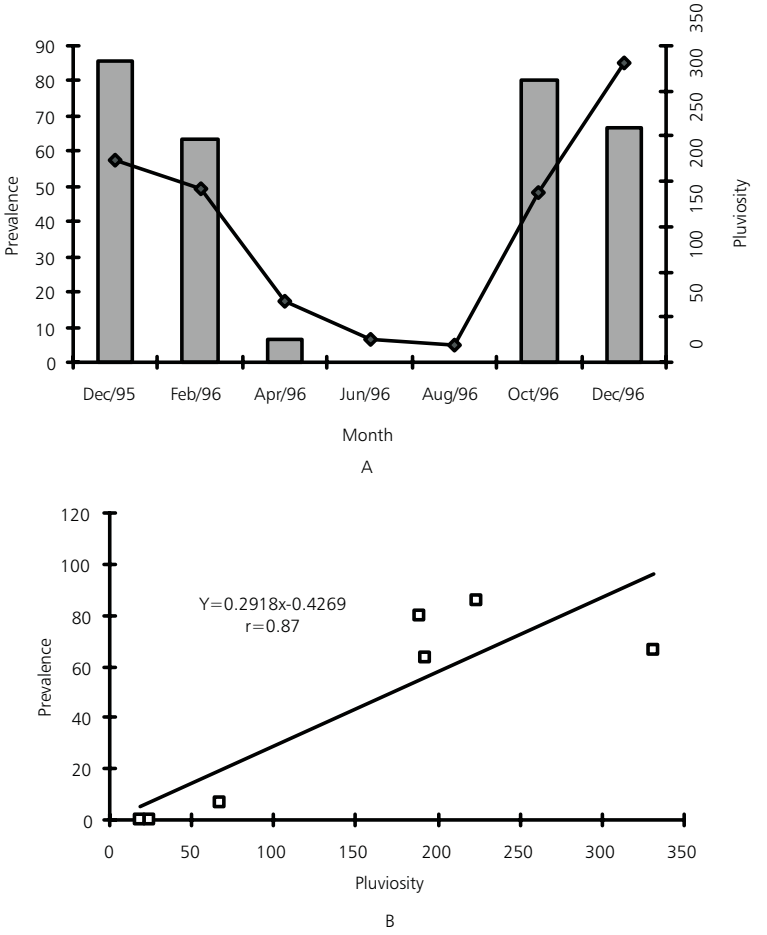


Fig. 2 — (a) Prevalence of *Thynnascaris* sp. larvae from *P. squamosissimus* related to pluviosity. Bar- prevalence (%); line- pluviosity (mm). (b) Positive correlation between the data.

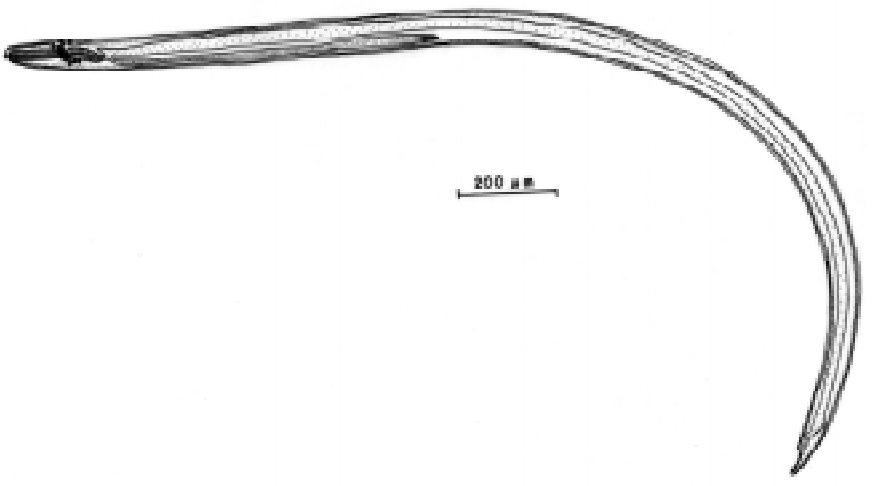


Fig. 3 — *Thynnascaris* sp larvae from *P. squamosissimus.* Note ventricular appendix length.

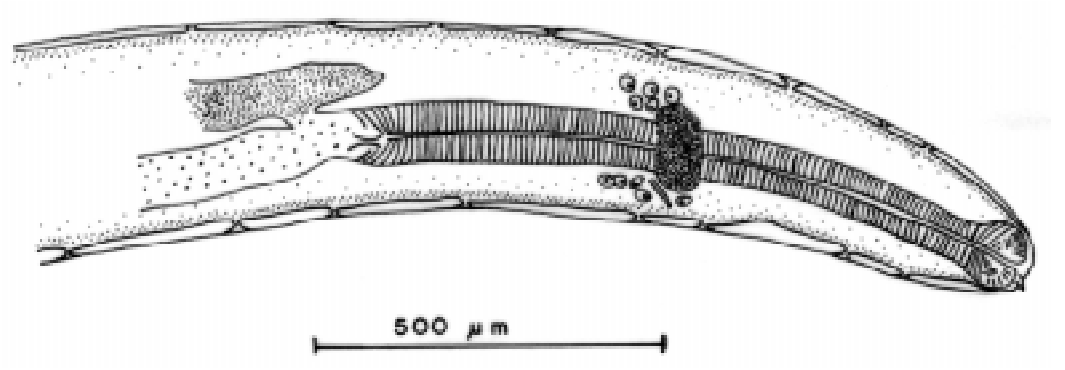


Fig. 4 — Esophagus, nerve ring and excretory pore of *Thynnascaris* sp. larvae in molt stage.

TABLE 2

Comparative values (mm) of anisakid nematodes larvae and their authors with the present work.

Characters	Contracaecum Kagei et al. (1970)	Contracaecum Rokicki et al. (1993)	Hysterothylacium Moravec et al. (1985)	Hysterothylacium Moravec & Sei (1988)	Thynnascaris Hurst (1984)	Thynnascaris present work
Intestinal caecum	180 (130-240)	761 (561-935)	*476 **530	503 (360-510)	(260-800)	28.5 (21.3-42.5)
Ventricular appendix	5,350 (3,060-7,900)	1,163 (891-1,540)	*408 **476	(4,760-5,690)	(280-720)	765.5 (564.5-1,032.2)
Nerve ring[a]	-	(264-361)	*326 **313	(218-261)	–	129.2 (95.7-146.8)
Excretory pore[a]	near to nerve ring	opens at the head	*353 **367	(231-326)	–	135.5 (85.1-148.9)
Length	16,100 (11,200-21,400)	18,377 (11,319-22,534)	*8,640 **9,810	(8,230-10,200)	(3,700-12,300)	2,544 (2,169-2,782)
Width	580 (410-820)	500 (385-693)	*163 **218	(258-340)	(100-290)	86.2 (80.6-104.8)
Tail[b]	190 (130-290)	190 (95-275)	*120 **190	(96-138)	(90-190)	71.1 (63.8-82.9)

* male measurement; ** female measurement.
[a] distance from the anterior end.
[b] distance of anus to the posterior end.

One of the higher prevalences was observed in *T. adunca* larvae (100%) (Wootten, 1978), *H. aduncum* (Bristow & Berland, 1991; González, 1998) which presented 66.1% and 79.0%, respectively, in the intestinal mesentery (mean intensity of 44.5 and 4.9, respectively). In the present work mean intensity was 16.0. Studies on seasonality of Anisakid larvae are scarce.

Wooten (1978) and Valtonen *et al*. (1988), respectively, observed a great distribution of *T. adunca* and *C. osculatum* related to lower length classes of the collected fishes in the North Sea and Baltic Sea. However, the present work is the first record on seasonality related to temperature and pluviosity. In *P. squamosissimus* it was not observed the relation between the weight or length of fish.

Nevertheless, higher prevalence of larvae was observed with increased temperature and pluviosity during the year (Figs. 1 and 2).

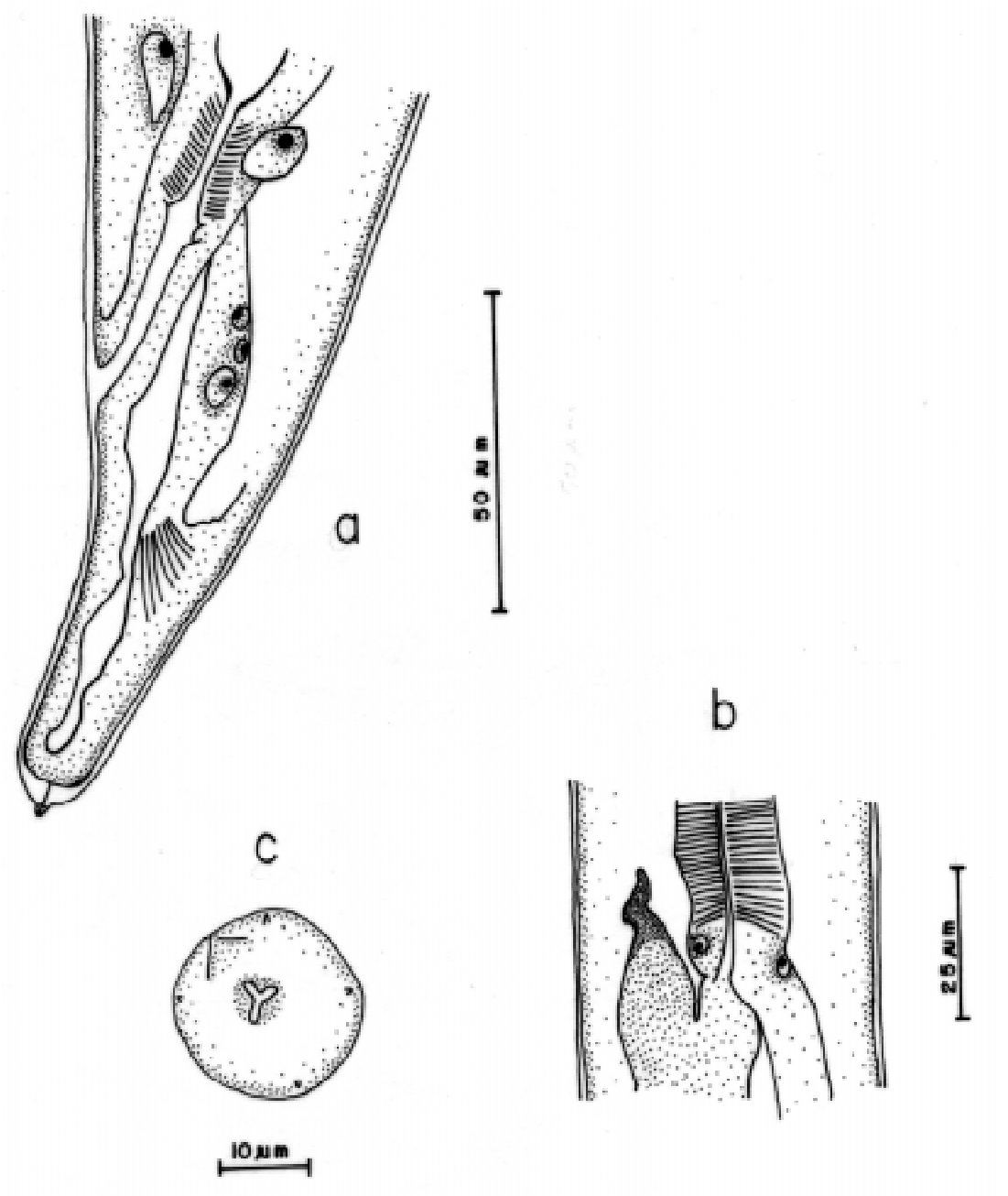


Fig. 5 — (a) Tail of larvae in molt stage; (b) intestinal caecum; (c) apical view of the anterior end of *Thynnascaris* sp. larvae.

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