

Trypanorhyncha cestode larvae of hygienic-sanitary importance parasitizing *Lobotes surinamensis* (Actinopterygii) obtained from fish markets in the state of Rio de Janeiro, Brazil

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Abstract

Lobotes surinamensis is an important fishery resource associated with artisanal fishing, occurring in tropical and subtropical regions throughout the world. Considering the hygienic-sanitary importance of Trypanorhyncha cestode larvae, 50 specimens of *L. surinamensis* were acquired from fish markets in the state of Rio de Janeiro, from May 2023 to December 2024. The fish were measured, necropsied, filleted, and their organs analyzed. Twenty-two larvae belonging to the following two species of Trypanorhyncha cestode plerocerci were found parasitizing the abdominal cavity and mesentery, *Callitetrarhynchus gracilis* and *C. speciosus*, with a prevalence of 10 and 20%, mean intensity of 2.20 and 1.70, and mean abundance of 0.22 and 0.34, respectively. The taxonomic identification of these parasites was based on morphological and morphometric characters using bright-field and scanning electron microscopes. This is the first report of these cestodes parasitizing specimens of *L. surinamensis* and is an important indicator for establishing fish hygiene control. In addition to repulsiveness, the presence of these cestode plerocerci is a concern due to the probable risk of allergic reactions, thus intensifying the hygienic-sanitary importance of monitoring these parasites to guarantee safe food for consumers.

Keywords: *Lobotes surinamensis*; Trypanorhyncha; hygienic-sanitary importance.

Practical Application: Larvae of cestodes with a disgusting appearance, which can cause rejection by consumers, generating economic losses and possibly causing human allergies.

1 INTRODUCTION

Lobotes surinamensis (Bloch, 1790) has a wide geographic distribution, inhabiting tropical and subtropical waters around the world (Froese & Pauly, 2023; Tiralongo et al., 2018). In Brazil, its incidence and consumption have increased over the years; it has become an important fishing resource associated with artisanal fishing and is consumed by riverside communities and the population residing along the Brazilian coast (Santos, 2023; Sazima et al., 2009; Silva et al., 2017). Due to the increase in its incidence, excess production began to be sold in large fish markets, thereby raising the risk of accidental exposure to fish parasites.

Trypanorhyncha cestodes have a global distribution, mainly in tropical and subtropical regions. The order has a great diversity of species, all parasitizing invertebrates and marine fish. The adults inhabit the gastrointestinal tract of elasmobranchs, while the larvae are found in intermediate hosts, such

as cephalopods and crustaceans, and in the coelomic cavity, visceral serosa, and musculature of teleosts (Palm, 2004).

Due to their disgusting appearance on fish, Trypanorhyncha larvae may cause rejection by consumers and restriction by health inspections from being commercialized, resulting in economic losses (Diniz et al., 2022; Leite et al., 2021; Menezes et al., 2023; Oliveira et al., 2019). Brazilian legislation has established that fish become unfit for consumption when they present massive muscular infection caused by parasites (Brasil, 2020). Although Trypanorhyncha larvae have no zoonotic potential, several studies have demonstrated immunological sensitization by antigens from these cestodes both in humans and in experimental animal models (Mattos et al., 2024; Pelayo et al., 2009; Vericimo et al., 2024).

The presence of Trypanorhyncha cestodes, such as *Callitetrarhynchus gracilis* (Rudolphi, 1819) and *Callitetrarhynchus speciosus* (Linton, 1897) Carvajal and Rego, 1985 has been

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reported in articles on parasite ecology, taxonomy, and hygienic-sanitary conditions worldwide (Abdelsalam et al., 2016; Charters et al., 2010; Mahmoud et al., 2015). In Brazil, recent studies concerning these parasites were mentioned by Leite et al. (2021) and Menezes et al. (2023), highlighting their importance in fish health inspections.

Therefore, the objective of this study was to identify the species of cestode larvae parasitizing *L. surinamensis* acquired from markets in the municipality of Rio de Janeiro, using morphological and morphometric analyses with optical and scanning electron microscopy of the specimens found, calculating their parasitic indices and reporting their infection sites, in addition to emphasizing their hygienic-sanitary importance for collective human health.

1.1 Relevance of the work

The presence of Trypanorhyncha metacestodes poses a threat to food hygiene due to their appearance and disgusting aspect, which may be rejected by consumers, resulting in economic losses. In addition, the ingestion of raw fish meat can cause accidental infections, which according to some studies can be potential causes of allergies in humans, thus reinforcing the importance of hygiene and sanitary measures and monitoring these parasites. Therefore, it is necessary to intensify inspections of fish-based foods and implement health education programs in order to guarantee safe food for consumers.

2 MATERIAL AND METHODS

A total of 50 specimens of *L. surinamensis* (40–65 cm total length), caught by professional fishermen, were purchased from fish markets in Copacabana and Sepetiba Bay, municipalities of Rio de Janeiro, state of Rio de Janeiro, Brazil, between May 2023 and December 2024. The fish were transported on ice to the Fish Inspection and Technology Laboratory (Laboratório de Inspeção e Tecnologia de Pescado) of the Faculdade de Veterinária, Universidade Federal Fluminense, RJ, where they were identified according to Menezes and Figueiredo (1980) and submitted to necropsy to examine the internal organs and musculature. Any cestode blastocysts were removed from the musculature and organs for further investigation and transferred to Petri dishes containing a physiological solution of 0.65% NaCl (sodium chloride). The plerocerci were removed from the blastocysts under a stereomicroscope with the sharp needles to release the larvae, which were placed in Petri dishes with distilled water, and then put in the refrigerator for at least 24 h to permit relaxation of the scolices and extroversion of the tentacles. Some of these helminths were fixed in alcohol formalin acetic acid (AFA), preserved in 70° GL ethanol, stained with Langeron's carmine, clarified in beechwood creosote, and preserved as whole mounts in Canada balsam according to Knoff and Gomes (2012). The taxonomic classification of the order Trypanorhyncha followed Palm (2004), and the larvae of Trypanorhyncha cestodes were identified according to Carvajal and Rego (1985), Kuraïem et al. (2016), Leite et al. (2021), and Menezes et al. (2018, 2023). The measurements were obtained by bright-field microscopy using an Olympus BX-41 microscope,

and samples were analyzed by bright-field microscopy using a Zeiss Axiophot microscope equipped with Nomarski's differential interference contrast (DIC) apparatus. The images were obtained using a Canon digital camera (Power Shot A640) in the Vertebrate Parasite Helminth Laboratory (Laboratório de Helmintos Parasitos de Vertebrados), Oswaldo Cruz Institute (IOC), Fiocruz, RJ.

Some specimens were prepared for scanning electron microscopy (SEM) according to Torres et al. (2013). The samples preserved in ethanol were dehydrated in an ethanol series (70–100° GL), CO₂ (carbon dioxide) critical point dried, coated in gold, and then examined and photographed using a SEM (Jeol JSM-6390LV), with an acceleration voltage of 15 kV. The measurements were reported in millimeters (mm) with averages and pattern deviations in parentheses, unless otherwise indicated.

The parasitic indices of prevalence, mean intensity, and mean abundance were calculated according to Bush et al. (1997). Representative specimens of each parasite species were deposited in the Helminthological Collection of the Oswaldo Cruz Institute (CHIOC, Coleção Helmintológica do Instituto Oswaldo Cruz), Fiocruz, Rio de Janeiro, RJ, Brazil.

3 RESULTS

Of the total 50 fish examined, 12 (24%) were found to be parasitized by 28 Trypanorhyncha cestode larvae belonging to two species, in the abdominal cavity and mesentery. Most of the cestode larvae were alive and showed moderate motility. Figure 1 shows one of these fish with the abdominal cavity open, revealing cestode blastocysts parasitizing the mesentery. The taxonomic identification of each species observed and the description of their main morphological characteristics are presented below. Table 1 indicates the morphometric data of the helminths found.

Lacistorhynchoidea Guiart, 1927, Lacistorhynchidae Guiart, 1927, Lacistorhynchinae Guiart, 1927, *Callitetrarhynchus* Pintner, 1931, *Callitetrarhynchus gracilis* (Rudolphi, 1819) Pintner, 1931. Figure 2.

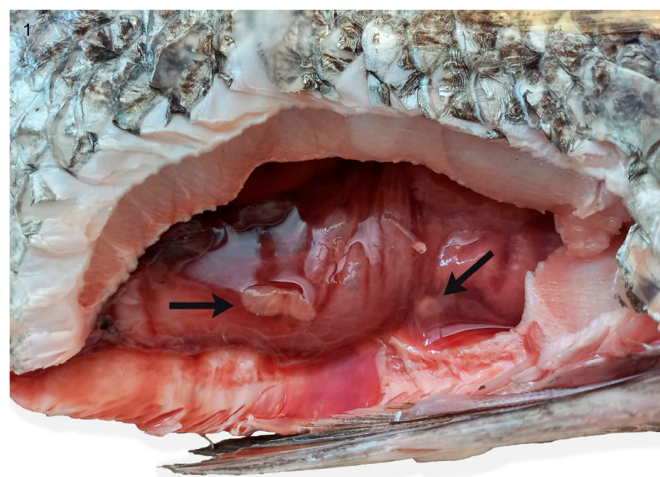


Figure 1. *Lobotes surinamensis* with the abdominal cavity open, showing cestode blastocysts (arrows) in the mesentery. Scale bar: 4 cm.

Table 1. Morphometric data of *Callitetrarhynchus gracilis* and *Callitetrarhynchus speciosus* collected from *Lobotes surinamensis*, Rio de Janeiro state, Brazil.

	<i>Callitetrarhynchus gracilis</i>	<i>Callitetrarhynchus speciosus</i>
Scolex (L)	1.6–3.6 (2.3 ± 0.78)	5.7–9.5 (7.6 ± 1.38)
Scolex (W)	0.1–0.2 (0.1 ± 0.03)	0.2–0.3 (0.3 ± 0.03)
Appendix (L)	0.7–1.8 (1.0 ± 0.47)	2.9–6.6 (4.8 ± 1.12)
Appendix (W)	0.1–0.2 (0.1 ± 0.01)	0.2–0.4 (0.3 ± 0.04)
Pars bothrialis (L)	0.2–0.4 (0.3 ± 0.08)	0.3–0.5 (0.4 ± 0.04)
Pars bothrialis (W)	0.11–0.15 (0.11 ± 0.02)	0.11–0.24 (0.13 ± 0.04)
Pars vaginalis (L)	1.4–2.0 (1.7 ± 0.20)	2.1–4.2 (2.8 ± 0.69)
Pars vaginalis (W)	0.12–0.19 (0.15 ± 0.02)	0.15–0.27 (0.19 ± 0.02)
Pars bulbosa (L)	0.4–0.5 (0.4 ± 0.04)	0.5–0.8 (0.6 ± 0.09)
Pars bulbosa (W)	0.1–0.2 (0.1 ± 0.02)	0.1–0.5 (0.2 ± 0.09)
Bulbs (L)	0.4–0.5 (0.3 ± 0.04)	0.4–0.7 (0.6 ± 0.10)
Bulbs (W)	0.05–0.08 (0.06 ± 0.01)	0.08–0.12 (0.09 ± 0.01)

Measurements are in millimeters, means and standard deviations in parentheses. L: length; W: width.

The main characteristics observed were based on 11 specimens obtained (eight whole mounts and three by SEM). Plerocerci with blastocyst. Elongated scolex, thin, with appendix, acraspedote. Two patelliform bothria with weakly notched posterior margins; long pars vaginalis; regularly sinuous tentacle sheath, less sinuous in the region of the pars bothrialis. The retractor muscles originate in the anterior third of the bulbs. Pars postbulbosa absent. Metabasal armature poeciloacanthous, atypical and heteromorphous; the hooks were hollow, in ascending half-spirals of eight principal hooks, beginning at the internal surface. Hooks 1(1') were large and uncinete, hooks 2(2') were uncinete and long, hooks 3(3') became falciform, large, and with large bases, hooks 4(4') and 5(5') falciform; hooks 6(6') became spiniform and were located near the external surface, the satellite hooks 7(7') were larger than hooks 8(8'), both with slender and uncinete shape. A simple chainette was present.

The parasitic indices were prevalence of 10%, mean intensity of 2.20, and mean abundance of 0.22. A voucher specimen was deposited in the CHIOC under the number 40470.

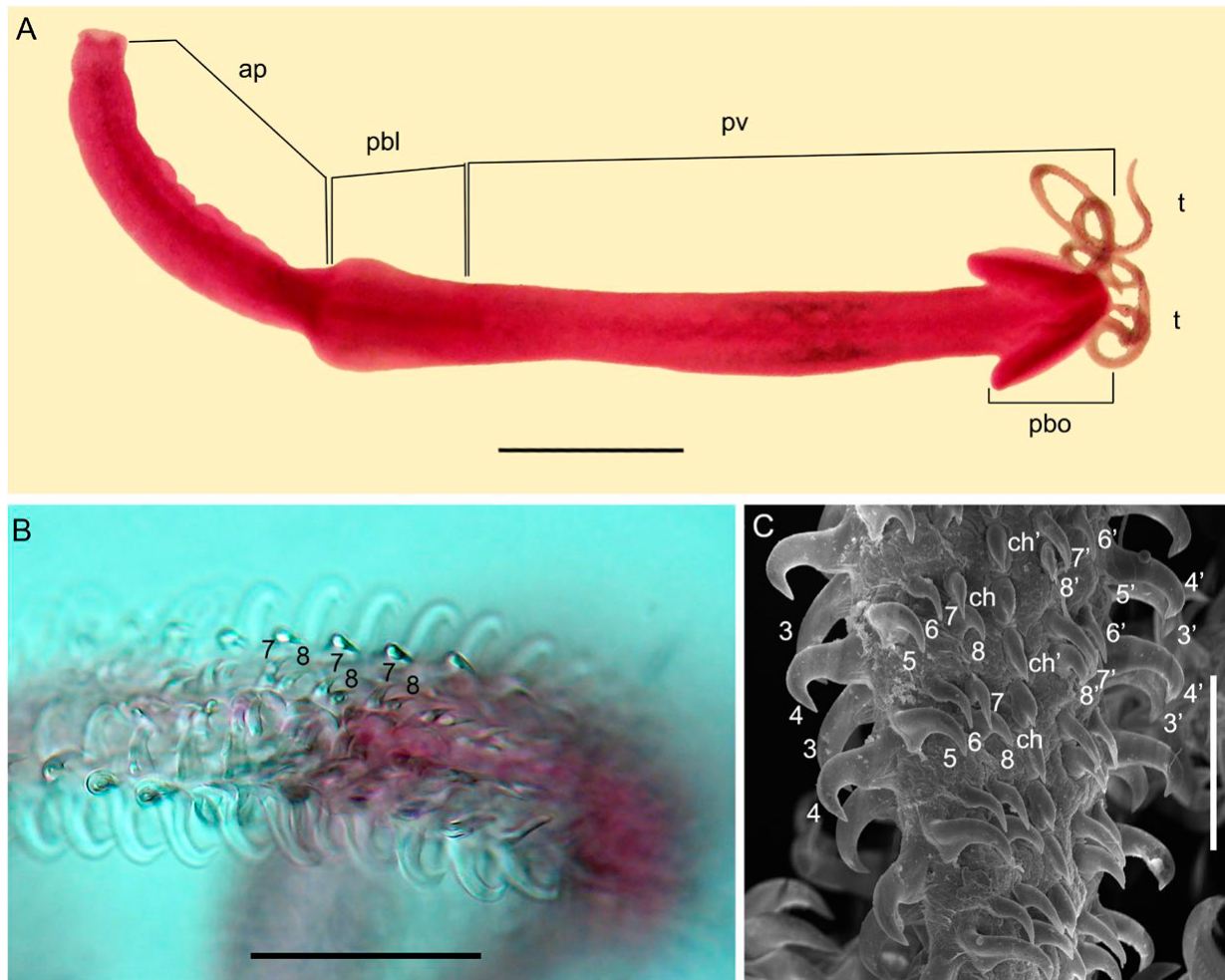


Figure 2. *Callitetrarhynchus gracilis* obtained from *Lobotes surinamensis*. (A) Entire plerocercus, tentacles partially extroverted, pars bothrialis, pars vaginalis, pars bulbosa, and appendix. (B) Details of the external surface hooks' arrangement of the metabasal armature tentacle, showing hooks 7 and 8. (C) Details of the external face hooks' arrangement of the metabasal armature tentacle, showing hooks 3(3'), 4(4'), 5(5'), 6(6'), 7(7'), and 8(8') and the chainette by scanning electron microscopy. t: tentacles partially extroverted; pbo: pars bothrialis; pv: pars vaginalis; pbl: pars bulbosa; ap: appendix. ch: chainette; ch': chainette. Scale bar: A = 0.5 mm; B = 0.05 mm; C = 30 μ m.

Callitetrarhynchus speciosus (Linton, 1897) Carvajal and Rego, 1985. Figure 3.

The main characteristics observed were based on 17 specimens (15 whole mounts and two by SEM). Plerocerci with blastocyst. Scolex elongated, thin, appendix acraspedote.

Two patelliform bothria with weakly notched posterior margins. Long pars vaginalis, tentacle sheath regularly sinuous but less sinuous in the region of the pars bothrialis. Bulbs elongated. Retractor muscles originating in the anterior third of the bulbs. Pars postbulbosa absent. The metabasal armature was

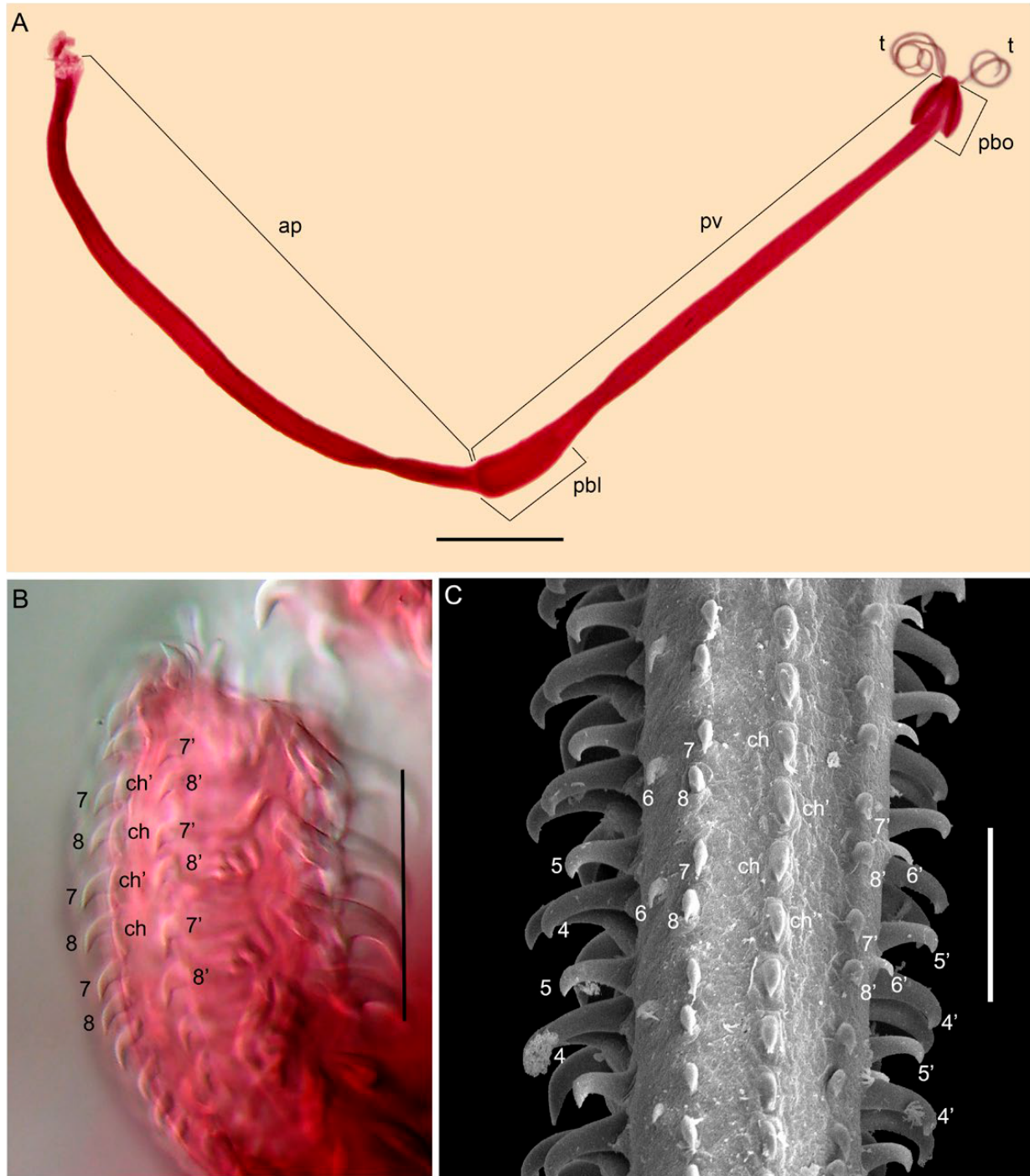


Figure 3. *Callitetrarhynchus speciosus* plerocercus obtained from *Lobotes surinamensis*. (A) Entire plerocercus, tentacles partially extroverted, pars bothrialis, pars vaginalis, pars bulbosa, and appendix. (B) Details of the external face hooks' arrangement of the metabasal armature tentacle, showing 7 (7') and 8 (8'), and the chainette by differential interference contrast. (C) Details of the external face hooks' arrangement of the metabasal armature tentacle, showing hooks 4 (4'), 5 (5'), 6 (6'), 7 (7'), and 8 (8') and the chainette by scanning electron microscopy. t: tentacles partially extroverted; pbo: pars bothrialis; pv: pars vaginalis; pbl: pars bulbosa; ap: appendix; ch: chainette; ch': chainette. Scale bar: A = 1 mm; B = 0.5 mm; C = 30 μ m.

poeciloacanthus, atypical, and heteromorphous; the hooks were hollow in ascending half-spirals of eight principal hooks, beginning on the internal surface. Hooks 1(1') were large and uncinat, hooks 2(2') were uncinat and long, hooks 3(3') became falciform, large and with large bases, hooks 4(4') and 5(5') were falciform, hooks 6(6') became spiniform and were located near the external surface, and the satellite hooks 7(7') and 8(8') were of the same size with a slender uncinat shape. A simple chainette was present.

The parasitic indices were prevalence of 20%, mean intensity of 1.70, and mean abundance of 0.34. A voucher specimen was deposited in the CHIOC under the number 40471.

In the present study, parasitic associations with *C. gracilis* and *C. speciosus* were found for three *L. surinamensis*. The species *C. gracilis* occurred as simple infections in two hosts, and *C. speciosus*, in seven hosts.

4 DISCUSSION

The species of *C. gracilis* and *C. speciosus* obtained were conformed with the morphological and morphometric data described by Carvajal and Rego (1985), Kuraïem et al. (2016), Leite et al. (2021), Menezes et al. (2018, 2023), Oliveira et al. (2019), and Palm (2004).

The specimens of *C. gracilis* and *C. speciosus*, obtained in the present investigation, revealed plerocerci with measurements compatible with those of the smallest specimens of these two species. According to Menezes et al. (2018, 2023) and Palm (2004), specimens of these two species of cestodes exhibit a variation in size within different hosts, associated with a life cycle with a trophic web of four hosts. The authors suggested that small-form plerocerci generally occurs in smaller fish species, and large-form plerocerci, in larger fish. However, the hosts in the present study were medium-sized fish and were parasitized by small-sized plerocerci, indicating that environmental factors could be influencing the ecology of the trophic chain of the host's life cycle, where they may be feeding more on crustaceans than on smaller fish, in the afore mentioned collection sites (Copacabana and Sepetiba Bay).

Based on the results of the present study, *C. gracilis* and *C. speciosus* had a prevalence of 10 and 20%, with mean intensities of 2.20 and 1.70, respectively. Ferreira et al. (2006) highlighted the fact that variations in prevalence and mean intensity of parasites of the order Trypanorhyncha are eventually dependent on the age and feeding habits of the fish, as well as on the host's immune response. Furthermore, the characteristics of local ecosystems and their food webs can determine not only the richness but also the abundance of larval helminths and explain the differences in the assemblages of parasitic species of teleost fish, as proposed by Diniz et al. (2022). The present study suggested that *L. surinamensis* varies according to the food web, which determines variability in the richness and abundance of helminth larvae.

In the present study, the *C. gracilis* and *C. speciosus* cestode larvae were alive and showed motility, occurring as a simple infection or in association, so in addition to their repugnant aspect, there could be a risk of accidental infection causing allergic reactions in humans, as warned by Verícimo et al. (2024).

5 CONCLUSIONS

The presence of Trypanorhyncha cestodes in *L. surinamensis* can adversely affect food hygiene, due to their appearance and disgusting aspect, and can be rejected by consumers causing economic losses. In addition, the ingestion of raw fish meat can cause accidental infections, which according to some studies can be potential causes of allergies in humans, thus reinforcing the importance of hygiene and sanitary measures and the monitoring of these parasites.

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