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Retraction

The editorial board of Brazilian Journal of Veterinary Parasitology communicates the formal publication of Retraction for extracting the article:

Pinheiro RHS, Bezerra AM, Giese EG. Morphological identification of *Skrjabinisakis* Mozgovoi, 1951 (Nematoda: Anisakidae) in *Kogia sima* (Cetacea: Kogiidae) from Brazilian waters. *Rev Bras Parasitol Vet* 2023; 32(4): e013423. https://doi.org/10.1590/S1984-29612023064.

The article is being retracted because the aforementioned article, apparently, it was produced with biological samples, in theory, used without authorization from the teams responsible for dealing with the stranding of the cetacean.

Profa. Dra. Rosangela Zacarias Machado

Editor-chief

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Original Article

[Retraction] Morphological identification of *Skrjabinisakis* Mozgovoi, 1951 (Nematoda: Anisakidae) in *Kogia sima* (Cetacea: Kogiidae) from Brazilian waters

Identificação morfológica de *Skrjabinisakis* Mozgovoi, 1951 (Nematoda: Anisakidae) em *Kogia sima* (Cetacea: Kogiidae) em águas brasileiras

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Abstract

New morphological, morphometric and scanning electron microscopy data of a nematode of the family Anisakidae, recovered from a specimen of *Kogia sima*, a cetacean that died off the northern coast of Brazil, are presented in this paper. Morphological features such as the violin-shaped ventricle and short and equal spicules, as well as the distribution of post-cloacal papillae and specificity for the definitive host (Kogiidae cetaceans) demonstrate similarity to *Skrjabinisakis paggiae*. This research records *Kogia sima* and *S. paggiae* on the estuarine coast of Pará, northern Brazil.

Keywords: Nematode, parasite, dwarf sperm whale, Amazon estuary.

Resumo

São apresentados, neste trabalho, novos dados morfológicos, morfométricos e de microscopia eletrônica de varredura de um nematoide da família Anisakidae, recuperado de *Kogia sima,* um cetáceo que veio a óbito na costa norte do Brasil. Características morfológicas, como o ventrículo em forma de violino, espículos curtos e iguais, além da distribuição de papilas pós-cloacais e especificidade pelo hospedeiro definitivo (cetáceos Kogiidae) demonstram similaridade a *Skrjabinisakis paggiae.* Esta pesquisa registra *Kogia sima* e *S. paggiae* na costa estuarina paraense, norte do Brasil.

Palavras-chave: Nematoide, parasito, cachalote anão, estuário amazônico.

Introduction

The systematics and nomenclature of Anisakidae Railliet & Henry, 1912 nematodes belonging to the genus *Anisakis* Dujardin, 1845 was controversial and confused until the application of genetic and molecular methodologies that over the past 20 years have led to an apparently stable and widely accepted taxonomy (Mattiucci & Nascetti, 2006, 2008). Anisakid nematodes are parasites of aquatic organisms such as fish, marine mammals, and fish-eating birds and are distributed throughout much of the world (Anderson, 2000). They are grouped into two subfamilies and several genera, Contracaecinae Mozgovoi & Shakhmatova, 1971 (*Contracaecum* Railliet & Henry, 1912 and *Phocascaris* Höst, 1932) and Anisakinae Railliet & Henry, 1912 (*Anisakis*, *Pseudoterranova* Mozgovoi, 1951, *Skrjabinisakis*, *Peritrachelius* Diesing, 1851, *Sulcascaris* Hartwich, 1957, *Pulchrascaris* Vicente & Santos, 1972, *Euterranova* Moravec & Justine, 2020, *Neoterranova* Moravec & Justine, 2020), forming a group of biodiverse representatives with complex phylogenetic relationships, and containing several *taxon inquirendum* (Nemys, 2023).

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The genus *Anisakis* had been grouped into two subgenera, *Anisakis* and *Skrjabinisakis*, based on the shape and length of the ventriculus and male spicules. Few authors have used these subgeneric names (Takano & Sata, 2022). Safonova et al. (2021) proposed the use of *Skrjabinisakis* as a genus name rather than a subgenus for *Anisakis physeteris* (Baylis, 1923), *A. brevispiculata* (Dollfus, 1966) and *A. paggiae* Mattiucci, Nascetti, Dailey, Webb, Barros, Cianchi & Bullini, 2005, and the resurrected generic status of *Peritrachelius* for *A. typica*. Takano & Sata (2022), in their studies based on molecular data, do not assign *A. typica* to *Peritrachelius*, since the species was nested in *Anisakis s.s.*, with similar phylogenetic relationship for *A. simplex s.s.* and *A. typica* observed by cox1 sequences. Mostafa et al. (2020) characterizes *A. simplex s.s.*, *A. pegreffii* and *A. typica* as non-monophyletic groups, therefore, the use of cox1 sequences may be inadequate for the reconstruction of relationships among *Anisakis* species.

Morphologically, species in the genus *Skrjabinisakis* are characterized by presenting a short, straight ventriculus whose length is equal or nearly equal to width, vulva located in the anterior fourth or third of the body, spicules short, with length not exceeding 0.67 (Safonova et al., 2021). Four *Anisakis* species have been reclassified as *Skrjabinisakis: S. physeteris* (type species) (= *Anisakis physeteris* and *Anisakis skrjabini* (Mozgovoy, 1949)); *S. schupakovi* (Mozgovoy, 1951); *S. brevispiculata* and *S. paggiae* (Safonova et al., 2021; Chero et al., 2023). Species of *Anisakis* and *Skrjabinisakis* have a complex life cycle. In the parasite's adult stage it lives in marine mammals, mainly cetaceans (ziphiids, delphinids, sperm whales, or a wide array of odontocetes and mysticetes), while planktonic or semi-planktonic crustaceans act as first intermediate hosts of the parasite, and fish and squid represent intermediate/paratenic hosts (Mattiucci & Nascetti, 2008; Kuhn et al., 2016; Lopriano et al., 2022). Cetaceans of the families Physeteridae (*Physeter macrocephalus* Linnaeus, 1758) and Kogiidae (*Kogia sima* (Owen, 1866) and *K. breviceps* (de Blainville, 1838)) are the main definitive hosts for the species *S. physeteris*, *S. brevispiculata* and *S. paggiae* (Mattiucci et al., 2001, 2005; Mattiucci & Nascetti, 2006).

Parasite infections are important bioindicators of marine mammal ecology and health because they provide information on habitat use, diet, social behavior, and population dynamics (Mattiucci & Nascetti, 2008). They also enable inferences regarding parasite/host evolutionary relationships, as well as biogeographic and phylogenetic relationships throughout history (Anderson, 1993; Barriga & Raga, 1994; Leidenberger et al., 2007; Marcogliese & Pietrock, 2011; Lehnert et al., 2014).

Although the Brazilian literature includes several studies on the parasitic fauna in terrestrial mammals in Brazil (Vicente et al., 1997; Vieira et al., 2008; Pinto et al., 2011), these data are scarce for the Amazon region when related to helminthological studies in aquatic mammals. With that in mind, this study taxonomically describes the anisakid parasites, recovered post-mortem from a female pygmy sperm whale, found and stranded on the northern coast of Pará State, Brazilian Amazon.

Materials and Methods

Nematodes were recovered during necropsy of a female dwarf sperm whale (*Kogia sima*) stranded on Humaitá beach (0°55'37"S; 48°17'12"W), municipality of Colares, state of Pará, northern Brazil, on 4 October 2018. Nematodes were fixed with 10% formaldehyde, stored at room temperature and examined using light microscopy and scanning electron microscopy following procedures described by Pinheiro et al. (2018). For light microscopy 18 females, 1 male and 6 fourth-stage larvae were examined and, for scanning electron microscopy 5 females and 1 male were observed. All measurements are presented in micrometers, unless otherwise indicated. The taxonomic classification of nematodes was in accordance with Safonova et al. (2021) and Takano & Sata (2022).

Results

A total of 42 nematodes (32 females, 4 males and 6 fourth-stage larvae - L4) were recovered from the intestine of a specimen of *Kogia sima*. The morphological and morphometric characteristics of the nematodes recovered from dwarf sperm whale are presented below, Figures 1, 2, 3 and in Table 1.

Anisakidae Railliet & Henry, 1912

Skrjabinisakis Mozgovoi, 1951

Skrjabinisakis paggiae (Mattiucci, Nascetti, Dailey, Webb, Barros, Cianchi & Bullini, 2005)

Robust, medium-sized nematode, morphology of the anterior region similar in both sexes. Transverse cuticular striae present throughout body except for lips. Cephalic extremity long and rounded, triangular mouth opening,

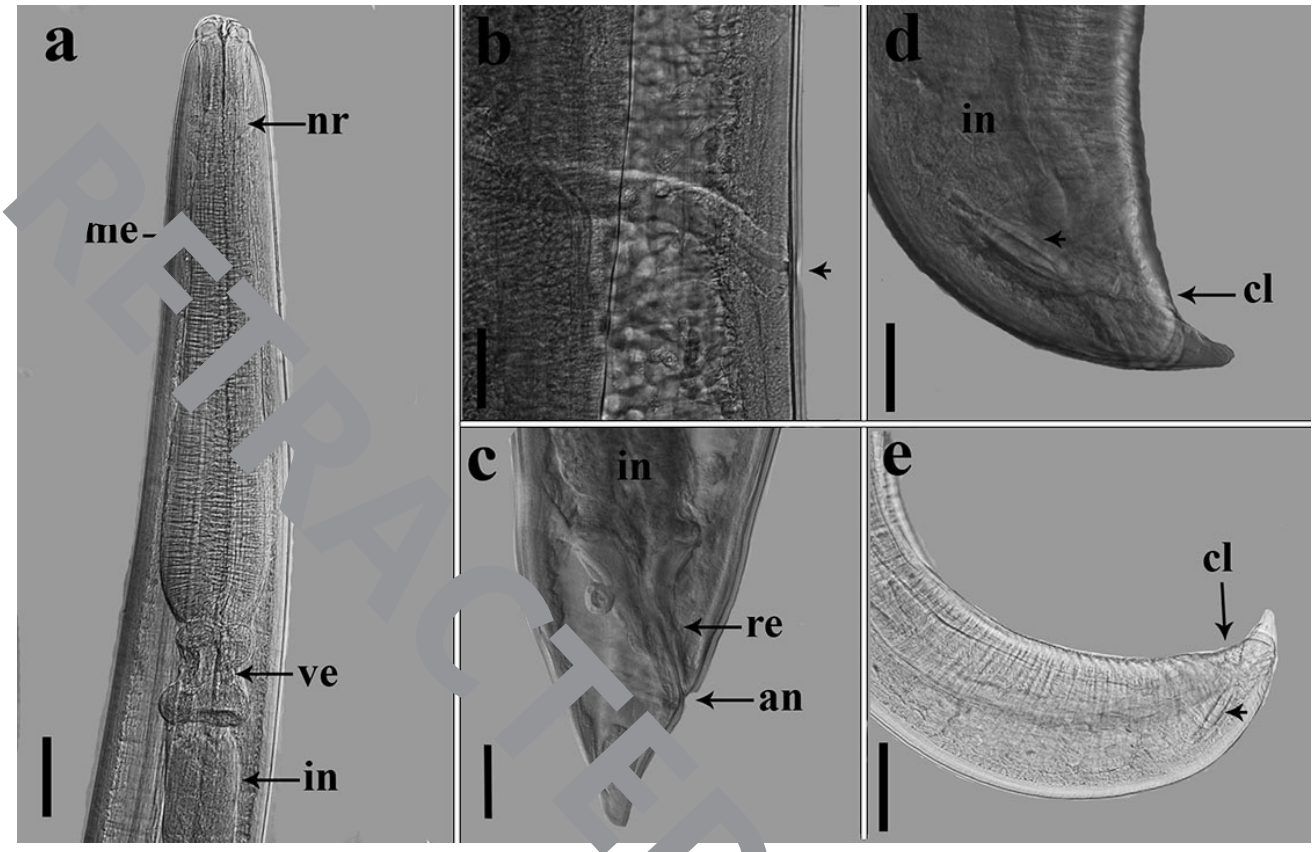

Figure 1. Light microscopy of *S. paggiae* identified in the present study. **(a)** lateral view of cephalic region showing muscular esophagus (me); nerve ring (nr); ventriculus (ve) and intestine (in). Bar = 100µm. **(b)** Lateral view of vulva (arrowhead). Bar = 100µm. **(c)** Posterior end of female lateral view: intestine (in), rectum (re) and anus (an). Bar = 100µm. **(d)** Detail posterior end of male, lateral view intestine (in), cloacal opening (cl) and spicules (arrowheads). Bar = 100µm. **(e)** Posterior end of male, lateral view cloacal opening (cl) and spicules (arrowheads). Bar = 200µm.

Table 1. Morphological and morphometric comparison of *S. paggiae* specimens collected from Kogiidae.

Character	Morphological and morphometric comparison of *Skrjabinisakis paggiae*							
Hosts	*K. sima* Atlantic coast of Brazil (State of Pará)				*K. breviceps* and *K. sima*, Atlantic coast of Florida		*K. sima* Atlantic coast of Brazil (State of Ceará)	
Locality								
Sex	Male	Females with eggs	Females without eggs	L4	Male	Female	Male	Female
Length (mm)	21	19–28	18–25	15–21	23–40	29–50	32–35	28–42
Width	343	600–800	571–800	443–643	–	–	–	–
Nerve ring	371	314–429	286–386	257–343	–	–	–	–
Esophagus (mm)^{L}	2	1.9–2.3	1.6–2	1.4–1.8	2–5	2.6–2.8	2.15–2.3	2.7
Esophagus^{W}	429	329–386	271–386	257–343	400–450	300–400	360–390	290–320
Ventriculus shape	violin				violin		violin	
Ventriculus^{L}	343	243–443	314–400	257–386	350–400	410–450	370–400	390–420
Ventriculus^{w}	200	200–229	157–271	143–229	240–270	300–350	240–250	280–330
Vulva (mm)	–	6–8	6–7	–	–	–	–	–
Spicules (mm)	0.18	–	–	–	0.17–0.22	–	0.18–0.19	–
Tail	267	147–233	137–207	77–110	150–170	–	–	–
Reference	Present study				Mattiucci et al. (2005)		Di Azevedo et al. (2015)	

L: length, W: width.

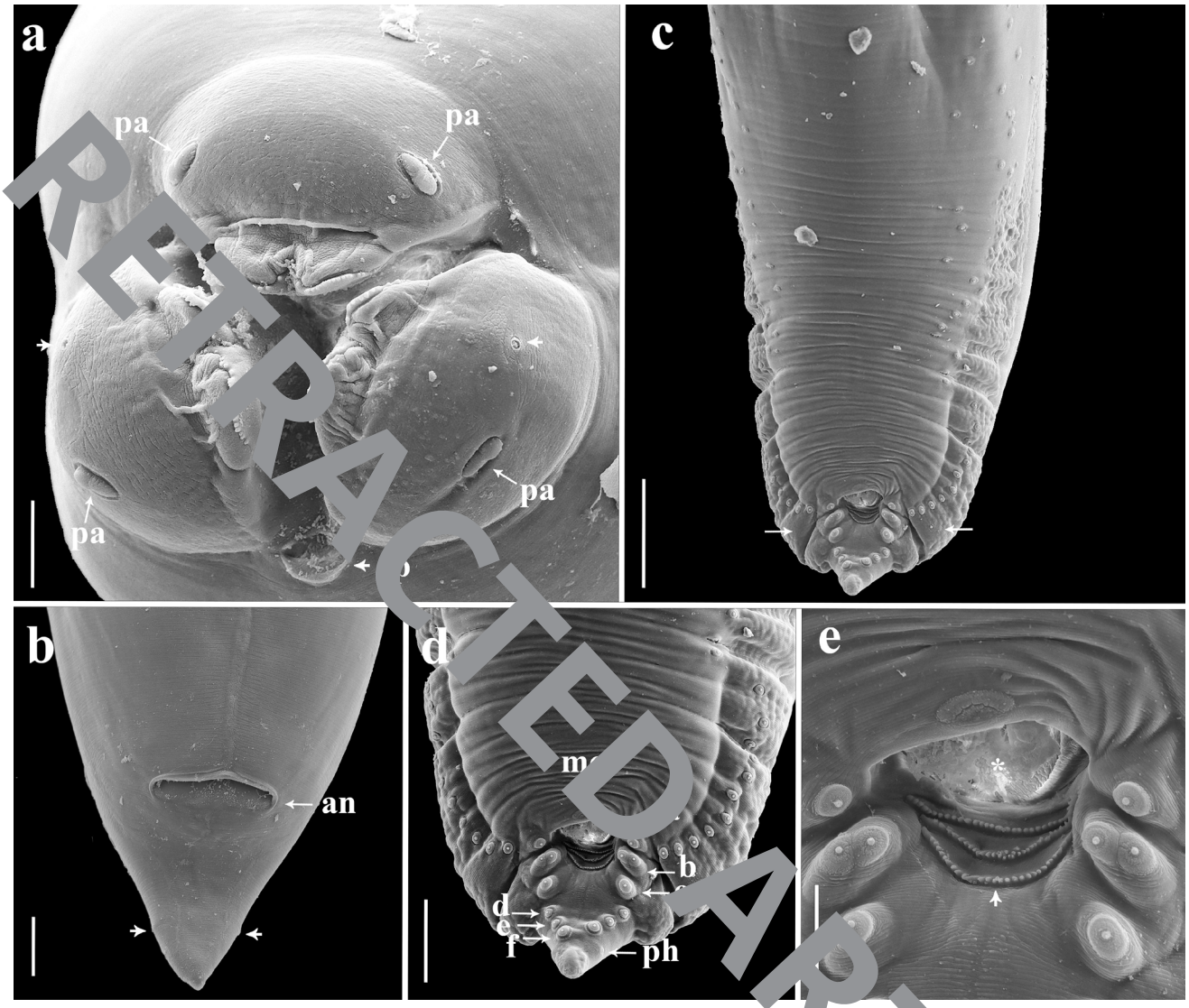

Figure 2. Scanning electron microscopy of *S. paggiae* identified in the present study. **(a)** Apical view of the anterior end showing three lips (two ventrolateral lips, one dorsolateral lip) with dentigerous ridges, lips papillae (pa), amphids (arrowheads) and excretory pore (ep). Bar = 20µm. **(b)** Posterior end of female, ventral view anus (an), on pair of phasmids lateral (arrowheads). Bar = 20µm. **(c)** Posterior end of male, ventral view, 2 of the lateral cuticular dilatations (arrow). Bar = 50µm. **(d)** Detail of caudal papillae sessile: 1 median papilla (me); 1 pair of single proximal papillae (a) lateral to cloaca; 1 pair of double paracloacal papillae (b); 4 pairs of distal papillae (c, d, e, f), of which (a, c, f) are equal in size and larger than (d, e), (d, e, f) are very close each other and one pair of very small papilla-like phasmids (ph), situated more laterally and posterior to last pair of distal papillae. Bar = 50µm. **(e)** Detail of plectanes (arrowheads) in cloacal (*). Bar = 20µm.

surrounded by three lips (one dorsal with two papillae present and each ventro-lateral lip having one papilla and one amphid) (Figures 1a and 2a). Projection with dentigerous ridges on surface inner in lips (Figure 2a). Interlabia absent. Esophagus muscular, flask-shaped, opening into the ventriculus (Figure 1a). Excretory pore at the base of the dorsal lip (Figure 2a). Ventriculus short, robust, violin-shaped (Figure 1a), with a distinct constriction in the middle, connected to the intestine by a slightly oblique junction (in dorsal view).

Males (based on 1 specimen): body 21 mm, maximum width at ventricule/intestinal junction 343. Nerve ring 371, measured to the anterior end. Muscular esophagus 2 mm long, 429 wide; the esophagus represents 9.5% of the total body length. Ventriculus 343 in length, 200 in width at level of constriction. Length of entire esophagus and ventriculus representing 11% of body length. Caudal papillae sessile, proximal and disposed in single row (Figure 2c): 1 median papilla; 1 pair of single proximal papillae (a) lateral to cloaca; 1 pair of double paracloacal papillae (b); 4 pairs of distal papillae (c, d, e, f), of which (a, c, f) are equal in size and larger than (d, e), and (d, e, f) are very close to each other (Figure 2d). The papillae all have the same circular structure surrounded by wrinkled

Figure 3. Light microscopy of fourth-stage larvae of *S. paggiae* identified in the present study. **(a)** lateral view of cephalic region showing nerve ring (nr), muscular esophagus (me); ventriculus (ve) and intestine (in). Bar = 200µm. **(b)** Posterior portion, portion of the intestine (in), rectum (re), three unicellular rectal (*) glands, anus (an), the tail without mucron. Bar = 50µm.

cuticle and provided with a small central knob. One pair of extremely small papilla-like phasmids situated more laterally and posterior to last pair of distal papillae (Figure 2d). Spicules short, stout, slightly equal and sclerotized 0.18 mm long, representing 0.85% of total body length (Figure 1d). Gubernaculum Absent. Tail rounded, 267 in length; distal extremity of tail rounded (Figure 1e) and 2 cuticular dilatations structures (Figure 2c). Three narrow denticulate caudal plates (plectanes) are present (Figure 2e).

Females with eggs (based on 6 specimens): body 23 mm (19–28 mm), maximum width at the height of the vulvar region 671 (600–800). Nerve ring 362 (314–429), measured to the anterior end. Muscular esophagus 2 mm (1.9–2.3 mm) long, 357 (329–386) wide; the esophagus represents 9% (8–10%) of body length. Ventriculus 357 (243–443) in length, 217 (200–229) in width at level of constriction. Length of entire esophagus and ventriculus representing 11% (9–12%) of body length. Vulva situated at 7 mm (6–8 mm) from anterior end, at about 30% (22–33%) of body length; vulval lips not elevated (Figure 1b). Muscular vagina directed posteriorly, uterus eggs elliptical or round shape, shell single, thin, smooth and transparent and embryo morulate. The eggs are 54 (47–67) long by 36 (30–47) wide. Rectum is a short hyaline tube; 3 small, unicellular rectal glands are present measuring 204 (180–233). Anus with prominent upper lip. Tail is conical, 176 (147–233) in length (Figures 1c and 2a).

Females without eggs (based on 6 specimens): body 21 mm (18–25 mm), maximum width at the height of the vulvar region 676 (571–800). Nerve ring 336 (286–386), measured to the anterior end. from anterior extremity. Muscular esophagus 2 mm (1.6–2 mm) long, 329 (271–386) wide; the esophagus represents 9% (8–10%) of body length. Ventriculus 343 (314–400) in length, 198 (157–271) in width at level of constriction. Length of entire esophagus and ventriculus represents 11% (10–12%) of body length. Vulva situated at 6 mm (6–7 mm) from anterior end, at about 30% (29–33%) of body length; vulval lips not elevated. Muscular vagina directed posteriorly, uterus without eggs. Rectum is a short hyaline tube; 3 small, unicellular rectal glands are present measuring 183 (167–200). Anus with prominent upper lip. Tail conical with a length of 166 (137–207).

Fourth-stage larvae (based on 6 specimens): body 18 mm (15–21 mm), maximum width at ventriculus/intestinal junction 512 (443–643). Mouth has three lips: one dorsal showing two double papillae and each subventral

lip presenting a single double papilla. Excretory pore opening between two subventral lips (Figure 3a). Nerve ring 288 (257–343), measured to the anterior end. Muscular esophagus 1.6 mm (1.4–1.8 mm) long, 286 (257–343) wide; the esophagus represents 9.5% of the total body length. Ventriculus 317 (257–386) in length, 186 (143–229) in width at level of constriction. Length of entire esophagus and ventriculus represents 11% of body length. Rectum is a short hyaline tube; 3 small, unicellular rectal glands are present measuring 113 (103–123) (Figure 3b). Tail is conical, 93 (77–110) in length, mucron absent.

Discussion

The nematode parasites from *Kogia sima* collected in the municipality of Colares, State of Pará, have similar characteristics to those of other species of the family Anisakidae and genus *Skrjabinisakis*. Based on the morphological features of the violin-shaped ventriculus, short and equal spicules and the distribution of caudal papillae in the male, the anisakid adult and L4 specimens were identified as *S. paggiae.* Although there are few articles related to parasitic helminths of cetaceans in Brazil, different researchers have sporadically attempted to learn about the biodiversity of whale parasites present along the Brazilian coast (see Yamaguti, 1959, 1963, 1971; Travassos, 1965; Travassos et al., 1969; Vicente et al., 1997; Muniz-Pereira et al., 1999). Muniz-Pereira et al. (1999) presented the first checklist of helminths in cetaceans in Brazil. Later Vieira et al. (2008) and Muniz-Pereira et al. (2009), seeking a more comprehensive and integrated description of the parasitic helminth fauna of vertebrates (host, locality and geographical distribution), included records of parasitism in cetaceans present in the Helminthology Collection of the Instituto Oswaldo Cruz (CHIOC).

The species in this study (*Skrjabinisakis paggiae*), is recorded worldwide as a parasite of whales of the families Kogiidae (i.e., dwarf sperm whale, *K. sima*, and pygmy sperm whale, *K. breviceps*), which serve as definitive hosts, mainly in the mid- and southern Atlantic Ocean (Valentini et al., 2006; Mattiucci & Nascetti, 2008; Quiazon, 2016; Shamsi, 2021; Cipriani et al., 2022). The association observed between Anisakis species and various cetacean taxa may reflect a coevolutionary history between these endoparasites and their hosts that has been estimated as older than 50 million years ago, driven by common trophic adaptations (Mattiucci & Nascetti, 2008; Klimpel & Palm, 2011; Li et al. 2018).

When proposing the description of *Skrjabinisakis paggiae* the authors associated the morphology of the ventricle (the adult ventriculus is short, never sigmoid and broader than long) and the spicules (the male spicules are short, stout and of similar length). These characteristics are observed in *Skrjabinisakis physeteris* and *S. brevispiculata,* which are parasites of cetaceans of the families Physeteridae and Kogiidae (Mattiucci & Nascetti, 2008). *Skrjabinisakis paggiae* and *S. brevispiculata* share the pygmy sperm whale as definitive host (Paggi et al., 1998; Mattiucci et al., 1986, 1997, 2001, 2005; Mattiucci & Nascetti, 2006, 2008; Safonova et al., 2021). This host preference allows anisakids to be used as biological indicators of their distribution and ultimate host abundance, since they closely follow the trophic relationships of their successive hosts (Mattiucci & Nascetti, 2008; Kuhn et al., 2011).

For Brazilian waters, Luque et al. (2010) presented a checklist that adds data to previous diverse publications but does not mention the occurrence of *S. paggiae*, only that of *A. insignis*, *A. simplex*, *S. physeteris* and *P. typica*, in addition to *Anisakis* sp., *Contracaecum* sp. and *Pseudoterranova* sp. along the Brazilian coast in different hosts. The same authors recorded *S. physeteris*, *Anisakis* sp. and *Pseudoterranova* sp. in Rio de Janeiro State, Ceará State and Pernambuco State (Fernando de Noronha Archipelago), parasitizing the stomach of pygmy sperm whales.

This is the first record of *S. paggiae* on the northern coast of Brazil (brackish water), although not its first record for *K. sima*. Among anisakid parasites infecting stranded cetaceans, especially Kogiidae whales, Di Azevedo et al. (2017) recorded *P. typica*, *A. ziphidarum*, *S. brevispiculata* and *S. paggiae*, parasitizing *K. sima* on the northeast coast (marine environment) of the country. *Skrjabinisakis paggiae* was recorded by Di Azevedo et al. (2015) in dwarf sperm whales stranded on Barra das Moitas beach, municipality of Amontada in Ceará state, northeastern Brazil, with details obtained by scanning electron microscopy for this parasite. Di Azevedo et al. (2017) recorded parasitism by *Skrjabinisakis paggiae* infecting *K. breviceps* in the country.

Although in this study the specimens observed were recovered from the intestine, factors such as time of host death and deterioration can cause parasites to migrate between organs, thus modifying the usual site of infection, which for *Skrjabinisakis paggiae* is the stomach. *Skrjabinisakis paggiae* was described from the stomach of *K. breviceps* on the Atlantic coast of Florida (United States) based on three males and four females collected and on one male *K. sima* from the same locality (Mattiucci et al., 2005). Subsequently, its known geographic distribution was expanded

to include the Philippine archipelago, western Atlantic Ocean, Caribbean Sea, Gulf of Mexico, Atlantic coast of Brazil and Australian waters (Quiazon et al., 2013; Di Azevedo et al., 2015, 2017; Shamsi et al., 2019).

In this work, observations by light microscopy and scanning electron microscopy highlight important features for future taxonomic studies for *Skrjabinisakis paggiae*, such as anterior end morphology (denticles that may be related to fixation on host tissue, double papillae in the dorsal lip), location and morphology of the vulva (6–8 mm), in addition to the distribution of the postcloacal papillae (1 median papilla; 1 pair of adcloacal papillae; 1 pair of double paracloacal papillae; 4 pairs of distal papillae) in males, confirming the data observed in SEM by Di Azevedo et al. (2015) for *Skrjabinisakis paggiae* and in light microscopy by Mattiucci et al. (2005). The spicule size (0.18 mm) of this study showed equivalence to that in Mattiucci et al. (2009) (0.17–0.22 mm); Di Azevedo et al. (2015) (0.18–0.19 mm) and Shamsi et al. (2019) (0.18–0.25 mm) for *S. paggiae*. Additional morphometric comparisons between *S. paggiae* are presented in Table 1.

Due to the type of preservation of the material (in 10% formaldehyde) we do not present molecular data, although Di Azevedo et al. (2015) have provided molecular records based on *cox*2 mtDNA sequences, for the occurrence of an *A. paggiae*-like species, with a genetic distance between the *A. paggiae*-like specimen and *S. paggiae* of (0.06), which is the same as between *A. simplex* and *A. pegreffii*, defined as distinct species (see Mattiucci et al., 1997, 2014) which are possibly being misinterpreted because they are different genera. According to phylogenic analyses, the branching order so far proposed for the *Anisakis/Skrjabinisakis* taxa showed that nematodes from the sperm whale and pygmy sperm whales (i.e., *S. physeteris*, *S. brevispiculata* and *S. paggiae*) always occupy a well-supported basal lineage (Mattiucci & Nascetti, 2008). Mattiucci et al. (2022) when presenting the phylogeny of some Anisakids, based on *cox*2 mtDNA sequences, divided the species into three clades: Clade (I) comprises species of the *A. simplex* (*s.s.*) complex, A. *pegreffii*, *A. berlandi*; Clade (II) *A. ziphidarum* and *A. nascettii* and Clade (III) *S. physeteris*, *S. brevispiculata*, and *S. paggiae*, the last clade also being confirmed with the morphological data for the three species.

Conclusion

This research records *S. paggiae* parasitizing *Kogia sima* in brackish water of northern Brazil, adding clarifying taxonomic features in SEM for *S. paggiae*.

Acknowledgements

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Ethics declaration

The material was collected during the necropsy of the specimen and later deposited in the didactic collection of the Zoology Museum at UFRA and made available for study, which justifies the waiver of authorization to use the material.

Conflict of interest

The authors declare that they have no conflict of interest.

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