Short Communication



Endoparasites of spinner dolphin *Stenella longirostris* (Cetacea: Delphinidae) from the southern Gulf of Mexico

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ABSTRACT. The spinner dolphin (*Stenella longirostris*) is one of the most frequently sighted cetacean species in the southern Gulf of Mexico (SGM). In 2021, a mass stranding of *S. longirostris* (n = 3) occurred in a coastal location of the SGM. Necropsies were performed, and intestines, stomachs, mesenteries, kidneys, and lungs were collected for parasitological examination. The objective was to determine the species of parasitic helminths found in stranded spinner dolphins from the SGM. Helminths were collected and prepared using staining and/or clearing techniques for morphological analysis and taxonomic identification. A total of 1,014 helminths were recovered, belonging to seven taxa: 1 digenean (*Synthesium tursionis*), 2 cestodes (*Tetrabothrius* sp. and plerocercoids of *Clistobothrium* sp.), 3 nematodes (*Halocercus brasiliensis*, *Anisakis* sp., and *Pseudoterranova* sp.), and 1 acanthocephalan (*Bolbosoma vasculosum*). All three spinner dolphins were parasitized by *S. tursionis* and *Tetrabothrius* sp. Furthermore, all taxa found represent new records for this host species in the Gulf of Mexico. In addition, *H. brasiliensis*, *B. vasculosum*, and *Tetrabothrius* sp. are new records for Mexican waters.

Keywords: helminths; parasites; mass stranding; cetaceans; Yucatan; Mexico

Spinner dolphins (Delphinidae: Stenella longirostris) are found in tropical and subtropical marine waters worldwide, primarily in oceanic areas, although coastal populations can also be observed (Perrin 2008). In Mexican waters, this species inhabits the Pacific Ocean, the Gulf of Mexico, and the Caribbean Sea (Heckel et al. 2018), with sightings recorded in the southern Gulf of Mexico (SGM) (Antochiw-Alonzo & Manzano 2004), and reported strandings in the coastlines of Quintana Roo and Yucatan states (Heckel et al. 2018). Their diet consists of small mesopelagic fish from intermediate waters, benthic fish, mesopelagic squid, and shrimp (Jefferson et al. 2008, Perrin 2008). In relation to the parasitic diversity found in the spinner dolphin, there are reports in the Atlantic side from Puerto Rico (Colón-Llavina et al. 2009), Florida (Cavallero et al. 2011), and the northeast coast of Brazil (Carvalho et al. 2010, Iñiguez et al. 2011, Di Azevedo et al. 2017). In Mexico, Aguilar-Aguilar et al. (2001) reported gastrointestinal helminths from spinner dolphins stranded in La Paz Bay (Pacific Ocean). Currently, there are no parasitological studies in spinner dolphins from the Gulf of Mexico.

In marine mammals, parasitological information can provide a basis for assessing their impact on the ecology of these hosts (Lehnert et al. 2019) and for a better understanding of life history and parasite-host interactions (Fraija-Fernández et al. 2016). In this sense, the study's objective was to determine the species of parasitic helminths in spinner dolphins (*S. longirostris*) that were stranded in the SGM.

A mass stranding of spinner dolphins occurred on December 11, 2021, in Chuburna Puerto (21°15'24"N, 89°49'26"W), Yucatan. The stranding report of the spinner dolphins led to the implementation of the Attention Protocol for Stranding of Marine Mammals in Mexico (DOF 2014), with the support of the Yucatan Marine Mammal Research and Conservation Program (PICMMY, by its Spanish acronym) at the Autonomous University of Yucatan. Total body length and sex were recorded. The decomposition code of the stranded animals or carcasses was obtained according to Geraci & Lounsbury (2005).

A necropsy of the dolphins was performed; during the procedure, the intestines, stomach, mesenteries, lungs, and kidneys were collected and frozen (-20°C) for later parasitological analysis. Intestines were divided into anterior and posterior parts, further subdivided into 1 m segments, and opened to examine their walls and contents. Afterward, each portion of the intestine was flushed with tap water and filtered through a 0.2 mm mesh sieve. The stomach, lungs, and kidneys were opened and rinsed; their contents were filtered and thoroughly examined. The contents of each organ were laid in large Petri dishes with a 0.7% saline solution. These contents were inspected using an Olympus SZ51 stereomicroscope in search of parasites. All helminths found were collected, fixed, and preserved in 70% ethyl alcohol.

For the taxonomic study, platyhelminths and acanthocephalans were stained with hydrochloric carmine, and nematodes were turned transparent using various concentrations of glycerin and 70% ethyl alcohol (modified from Vidal-Martínez et al. 2001). Taxonomic identification of helminths was conducted through morphological analysis, aided by specialized literature from Yamaguti (1963), Hartwich (1974), Fernández et al. (1994), Khalil et al. (1994), Marigo et al. (2008), and Demarque et al. (2020). Parasite prevalence and mean intensity were obtained according to Bush et al. (1997) and calculated using Quantitative Parasitology (Qpweb) v.1.0.15 (Reiczigel et al. 2019). Voucher specimens were deposited in the National Helminth Collection (CNHE, by its Spanish acronym) of the National Autonomous University of Mexico.

The mass stranding consisted of three dead adult spinner dolphins (males), in body length from 204 ± 7.81 cm (mean \pm standard deviation), with a code 3 decomposition (carcass is decomposed, but internal organs remain intact). Helminths were found in the stomach, intestines, and lungs, whereas none were observed in the kidneys. A total of 1,014 worms belonging to 7 taxa, including 1 digenean adult of

Synthesium tursionis; 2 cestodes, 1 adult of Tetrabothrius sp., and 1 plerocercoid of Clistobothrium sp.; 3 nematodes, 1 adult of Halocercus brasiliensis, 2 immature Pseudoterranova sp., and Anisakis sp.; and the immature acanthocephalan Bolbosoma vasculosum were identified morphologically (Table 1, Fig. 1a-h). All three spinner dolphins were parasitized by S. tursionis and Tetrabothrius sp. Besides, Clistobothrium sp. were the most abundant (Table 1).

In this study, only three spinner dolphins from the SGM were analyzed, finding a helminth richness of seven species. However, it is similar to that reported in other studies with the same host and a larger number of samples, such as Dailey & Perrin (1973), who analyzed 19 dolphins and reported nine helminth species, and Aguilar-Aguilar et al. (2001), with 31 dolphins and seven species. Most parasites were found in the digestive tract (six taxa), with only one taxon in the lungs and none in the kidneys. In the latter case, there are few reports of kidney parasites in cetaceans, such as the nematode *Crassicauda anthonyi* in Cuvier's beaked whale, *Ziphius cavirostris* (Oliveira et al. 2011).

The helminth community found in spinner dolphins in the SGM included S. tursionis, a species of the Brachycladiidae family, which infects cetaceans and pinnipeds (Shiozaki et al. 2019). Synthesium tursionis is found in the intestines of odontocetes (Jones et al. 2005). Its life cycle is unknown; however, in digeneans, it begins with the ciliated larva or miracidium, which is ingested by its first intermediate host (mollusks). Subsequently, different larval stages, such as cercariae, can penetrate their definitive host or encyst in a second intermediate host, including invertebrates and/or fish, where they develop into metacercariae, which eventually reach their definitive host (García-Prieto et al. 2014). This digenean is a cosmopolitan species, commonly found in bottlenose dolphins (Tursiops truncatus) and other odontocete species (Shiozaki et al. 2019). It has been reported in the Mexican Pacific for spinner dolphins (Aguilar-Aguilar et al. 2001).

Tetrabothrius sp. could not be identified at the species level due to the poor condition of the samples. It is cosmopolitan and parasitizes seabirds and cetaceans (Khalil et al. 1994). Its life cycle is unknown, but the cestode cycle begins with larvae, which may be inside the egg or free-living. They then find an intermediate host, develop into various metacestodes, and infect the definitive host (vertebrate) (García-Prieto et al. 2014). Tetrabothrius (Tetrabothrius) forsteri has been recorded for spinner dolphins in the northeast coast of Brazil (Carvalho et al. 2010).

Table 1. Helminths registered in the spinner dolphins (*Stenella longirostris*) from the southern Gulf of Mexico. St: stomach, Pi: posterior intestine, Ai: anterior intestine, Lu: lungs. *Colección Nacional de Helmintos, Institute of Biology, National Autonomous University of Mexico. CI: confidence interval.

Helminth	Abundance	Prevalence (%)	Mean intensity (95% CI)	Site of infection	Number in CNHE*
Digenea					
Synthesium tursionis	25	100	8.7 (1-16)	St, Pi, Ai	11868
Cestoda					
Tetrabothrius sp.	62	100	20.7 (13-26.3)	Ai	11869
Clistobothrium sp.	920	33	920	St, Ai	11870
Nematoda					
Halocercus brasiliensis	3	33	3	Lu	11871
Anisakis sp.	1	33	1	St	12840
Pseudoterranova sp.	1	33	1	St	11872
Acanthocephala					
Bolbosoma vasculosum	2	66	1	Ai, Pi	11873

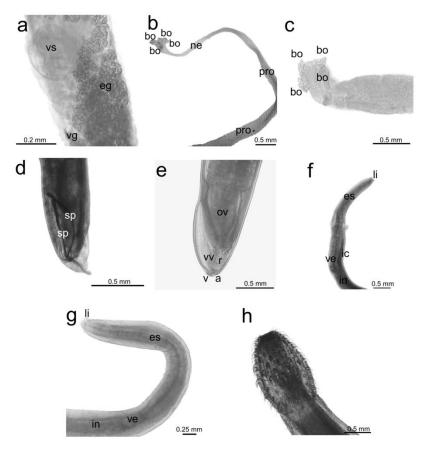


Figure 1. a) *Synthesium tursionis* with a large number of eggs and where the vitellogen glands reach almost to ventral sucker, b) *Tetrabothrius* sp. observing their square bothrydia, c) anterior part of the body of *Clistobothrium* sp. with its four bothrydia, d) male *Halocercus brasiliensis* with its spicules (right length 0.83 mm and left 0.73 mm), e) female showing the terminal region of the reproductive system, f) *Pseudoterranova* sp. with intestinal cecum, g) *Anisakis* sp. with simple ventricle, h) *Bolbosoma vasculosum*, with a cylindrical proboscis has 22 longitudinal hooks. a: anus; bo: bothridia; eg: eggs; es: esophagus; in: intestine; ic: intestinal cecum; li: lips; ne: neck; pro: proglottid; vg: vitellogenic glands; vs: ventral sucker; r: rectum; sp: spicule; vv: vagina; ve: ventricle; v: vulva.

The plerocercoids found were identified as Tetraphyllidea gen. sp. "large" type (Agusti et al. 2005, Aznar et al. 2007). However, they were later transferred to the genus Clistobothrium due to their phylogenetic affinities (Caira et al. 2020). Plerocercoids are found free in the lumen of the intestine, bile ducts, and within the anal crypts of cetaceans (Aznar et al. 2007). These types of larvae are primarily acquired by cetaceans in the high seas through the consumption of infested fish and cephalopods (Agusti et al. 2005, Aznar et al. 2007), indicating that at least the spinner dolphin infected with these larvae inhabited the deep waters of the SGM. The adult forms of these larvae are unknown; however, it has been suggested that cetaceans play a crucial role in their life cycle (Aznar et al. 2007, Caira et al. 2020). Adults of the genus *Clistobothrium* have an affinity for lamniform sharks (Caira et al. 2020), which predate spinner dolphins (Perrin 2008). These larvae have only been reported in spinner dolphins in the Mexican Pacific (Aguilar-Aguilar et al. 2001).

On the other hand, H. brasiliensis belongs to the Pseudaliidae family, which can cause severe lung infections in cetaceans (Demarque et al. 2020). It is a common nematode found in the bronchi and bronchioles of some cetaceans, where it can cause pneumonia and septicemia (Guimarães et al. 2015). Parasitic pneumonia has been reported in dolphins infected with H. brasiliensis, which may have directly contributed to the stranding and death of the animals in northeast Brazil (Guimarães et al. 2015). Its life cycle is unknown; however, Halocercus can be transmitted via the placenta, as observed in *T. truncatus* (Dailey et al. 1991) and the killer whale Orcinus orca (Reckendorf et al. 2018). This nematode has been previously recorded only in spinner dolphins in northeast Brazil (Carvalho et al. 2010).

In the case of anisakid nematodes (Anisakis sp. and Pseudoterranova sp.), they have an indirect life cycle, involving multiple hosts at different levels of marine food webs (Blažeković et al. 2015, Cipriani et al. 2022). Their initial intermediate hosts are planktonic or semiplanktonic crustaceans, their secondary intermediate/ paratenic hosts are fish and squid, and their definitive hosts are primarily marine mammals, mostly cetaceans (Mattiucci et al. 2018, Cipriani et al. 2024). Anisakids can infect their hosts with superficial lesions and deep ulcers in the gastric chambers (Blažeković et al. 2015), which leads to malnutrition and stress (Shamsi et al. 2019). Anisakis spp. have been reported in S. longirostris in Brazil (Carvalho et al. 2010), Costa Rica (Oliveira et al. 2011), and Puerto Rico (Colón-Llavina et al. 2009), and Anisakis typica in the Mexican Pacific (Aguilar-Aguilar et al. 2001), Brazil (Nadler et al. 2005, Iñiguez et al. 2011), and Florida (Cavallero et al. 2011). *Pseudoterranova* sp. has been reported in spinner dolphins off the northeastern coast of Brazil (Di Azevedo et al. 2017). Acanthocephalan of the genus *Bolbosoma*, such as *B. vasculosum*, use crustaceans as their first intermediate host, fish as their second intermediate host, and marine mammals as their definitive hosts (Yamaguti 1963). They have been recorded for spinner dolphins in the eastern Pacific (Dailey & Perrin 1973).

There are no studies currently on the feeding habits of spinner dolphins in the Gulf of Mexico. However, a study in the South Atlantic (off the coast of Brazil) reports that spinner dolphins feed on small prey, including oceanic squids from the families Enoploteuthidae, Ommastrephidae, and Cranchiidae, fish (Carangidae and Exocoetidae), and shrimp (Silva et al. 2007), which could act as intermediate/paratenic hosts. While spinner dolphins are preyed upon by sharks of Dalatiidae, Lamnidae, and Carcharhinidae (Silva et al. 2007), these fish can serve as definitive hosts for some helminths, such as Clistobothrium sp. (Caira et al. 2020). On the other hand, spinner dolphins can respond to general and local changes in the prey abundance (Benoit-Bird & Au 2003), which could be reflected in the diversity of helminths and their infection parameters. Additionally, parasitic infections are common in cetaceans. However, it has been suggested that severe infections could occur due to compromised immune function (Li et al. 2021).

Future parasitological studies, which include larger samples and molecular markers for species identification, particularly for larval stages and worms in poor condition, would provide a more comprehensive inventory of the parasitic fauna of spinner dolphins in the region. There are few parasitological studies on marine mammals of Mexico; therefore, this work contributes to their biological knowledge in the country and the region. This study represents the first parasitological report of spinner dolphins inhabiting the Gulf of Mexico, as well as the first geographic records in the North Atlantic of *H. brasiliensis* and *Pseudoterranova* sp., as well as *H. brasiliensis*, *B. vasculosum*, and *Tetrabothrius* sp. infecting this cetacean in Mexican waters.

Credit author contribution

A. Hernández-Olascoaga: concept, design, execution, analysis, interpretation, writing-original draft; S. Guillén-Hernández: analysis, interpretation, writing-review, and editing; R.E. Díaz-Gamboa: concept,

execution, analysis, funding, writing-review, and editing. All authors have read and accepted the published version of the manuscript.

Conflict of interest

The authors declare they have no conflicts of interest.

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