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Original Article

Acuariidae (Nematoda) in Procellariiformes (Aves) on the southern coast of Rio Grande do Sul, Brazil

Acuariidae (Nematoda) em Procellariiformes (Aves) no litoral sul do Rio Grande do Sul, Brasil

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Abstract

Acuariidae nematodes are normally found in the digestive tract of aquatic birds, including Procellariiformes. Were examined *Calonectris borealis* (n = 4), *Diomedea exulans* (n = 1), *Macronectes giganteus* (n = 8), *Thalassarche chlororhynchos* (n = 5), *Thalassarche melanophrys* (n = 15), *Procellaria aequinoctialis* (n = 4), *Puffinus gravis* (n = 2) and *Puffinus puffinus* (n = 6), collected on the southern coast of RS, Brazil. A total of 16 birds (35.5%) were parasitized by two species of Acuariidae. *Stegophorus diomedeae* and *Seuratia shipleyi* were identified, with prevalences of 26.1% and 21.7%, respectively. Few studies on nematodes in Procellariiformes have been conducted. Here, the acuariids *Seuratia shipleyi* in *Calonectris borealis* and *Procellaria aequinoctialis* and *Stegophorus diomedeae* in *Diomedea exulans*, *Procellaria aequinoctialis* and *Thalassarche chlororhynchos* were reported for the first time.

Keywords: Nematoda, albatrosses, petrels, parasites, prevalence, infection rates.

Resumo

Os nematoides Acuariidae são normalmente encontrados no trato digestivo de aves aquáticas, incluindo os Procellariiformes. Foram examinadas *Calonectris borealis* (n=4), *Diomedea exulans* (n=1), *Macronectes giganteus* (n=8), *Thalassarche chlororhynchos* (n=5), *Thalassarche melanophrys* (n=15), *Procellaria aequinoctialis* (n=4), *Puffinus gravis* (n=2) e *Puffinus puffinus* (n=6), coletados na costa sul do Rio Grande do Sul, Brasil. Dentre os exemplares examinados, 16 (35,5%) estavam parasitados por duas espécies de Acuariidae. Foram identificados *Stegophorus diomedeae* e *Seuratia shipleyi,* com prevalência de 26,1% e 21,7%, respectivamente. Existem poucos estudos relacionados a nematoides em Procellariiformes, e aqui registra-se pela primeira vez os Acuariidae *Seuratia shipleyi* em *Calonectris borealis* e *Procellaria aequinoctialis*, e *Stegophorus diomedeae* em *Diomedea exulans*, *Procellaria aequinoctialis* e *Thalassarche chlororhynchos.*

Palavras-chave: Nematoda, albatrozes, petréis, parasitos, prevalência, intensidade média.

Introduction

Acuariidae include midsized nematodes that inhabit the upper digestive tract of aquatic birds (Procellariiformes, Pelecaniformes, Ciconiiformes, Anseriformes, Charadriiformes and Coraciiformes), although a few are associated with terrestrial birds such as Falconiformes, Galliformes and Passeriformes (ANDERSON, 2000). The species of this subfamily are characterized by the presence of cephalic cords as fixation structures (CHABAUD, 2009).

Over the life cycle of acuariids, it has been observed that larvae in the infective stage are encapsulated in the intestine or mesentery of paratenic hosts (frog or fish) to reach the final host. However, some nematodes undergo all their developmental stages within the gizzard of the definitive host, where the adults are found, while others first develop to the subadult stage in the proventriculus before migrating to the gizzard. Some esophageal parasites develop entirely in the esophagus, while others reach

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the subadult stage in the proventriculus before parasitizing the esophagus. Proventricular nematodes probably undergo their entire development in this organ, and some of these species occur in seabirds. On the migration routes used by these birds, ingestion of shellfish that are intermediate hosts can contribute towards the diversity of acuariids that infect birds (ANDERSON, 2000).

Acuariidae comprises Acuariinae, Schistorophinae and Seuratiinae. The subfamily Seuratiinae comprises seven genera, among which are *Stegophorus* and *Seuratia*, commonly found parasitizing seabirds in different regions (Table 1).

Procellariiformes are widely distributed seabirds, but with greater diversity of species in the southern hemisphere. This order consists of 99 species divided into four families: Procellariidae, Hydrobatidae, Pelecanoididae and Diomedeidae (SICK, 1997; IBAMA, 2006). These are predatory birds at the top of the food chain and they generally require productive waters to ensure their survival and reproduction (CURY et al., 2011). Their diet consists of squid, krill, fish, carcasses and chicks of other birds. These birds also often follow fishing boats in search of debris that has been in the sea for a long time (BARBIERI, 2008; CUBAS et al., 2014.). Presence of seabirds of the order Procellariiformes is indicative of the presence of fish shoals and hence of ecosystem health. Presence/absence of prey is reflected in changes to these birds' diet (CUBAS et al., 2014).

The coast of Rio Grande do Sul, in southern Brazil, is an important feeding site for several species of seabirds. One factor responsible for this large concentration of birds is high biological productivity, especially in winter, when a coastal branch of the Falkland current enters, bearing cold water rich in organic matter from the south (PEREZ, 2012).

Considering the ecological importance of these birds, the conservation condition of these species and the lack of information about their biology and helminth fauna, the aim of the present study was to report on occurrences of the acuariid nematodes *Stegophorus diomedeae* and *Seuratia shipleyi* as parasites of Procellariiformes on the southern coast of Rio Grande do Sul, and the infection rates in these birds.

Materials and Methods

A total of 45 specimens of Procellariiformes were examined, collected from the southern coast of Brazil between Farol de Mostardas (31° 14' S; 50° 54' W) and Barra do Chui (33° 44' S; 53° 22' W). Most of them (n = 32) were received from the Centro de Recuperação de Animais Marinhos (CRAM-FURG), where they had died during the rehabilitation process. Nine hosts were found dead on beaches and four individuals were caught through longline fisheries. The host species examined were *Calonectris borealis* (n=4), *Diomedea exulans* (n=1), *Macronectes giganteus* (n=8), *Thalassarche chlororhynchos* (n=5), *Thalassarche melanophrys* (n=15), *Procellaria aequinoctialis* (n=4), *Puffinus gravis* (n=2) and *Puffinus puffinus* (n=6).

The esophagus, proventriculus, ventricle, small intestine, large intestine, cecum and cloaca of the birds were examined separately during the necropsy. The nematodes were fixed in AFA (70° GL ethanol, 37% formalin and glacial acetic acid, in the proportions 95:3:2) and preserved in 70° GL ethanol that was 10% glycerinated (AMATO & AMATO, 2010). After clarification in Amann's lactophenol, the nematodes were mounted on semi-permanent

Table 1. Species of Seuratiinae (Nematoda: Acuariidae) recorded in birds, according to host collection site, with the respective bibliographical references.

Seuratiinae	Host	Host collection location	Reference
Seuratia shipleyi	*Thalassarche melanophrys* (Procellariiformes)	Rio de Janeiro (Brazil)	Mendonça & Rodrigues (1968)
	Puffinus gravis (Procellariiformes)	Gough Island (South Atlantic)	Hoberg & Ryan (1989)
	Phoebetria palpebrata (Procellariiformes)	Antarctica	Barbosa & Palácios (2009)
Stegophorus diomedeae	*Thalassarche melanophrys* (Procellariiformes)	Rio de Janeiro (Brazil)	Rodrigues & Mendonça (1967)
	Puffinus gravis (Procellariiformes)	Florida (USA)	Foster et al. (1996)
Stegophorus stellaepolaris	*Puffinus gravis* (Procellariiformes)	Florida (USA)	Foster et al. (1996)
Stegophorus macronectes	*Pygoscelis papua* (Sphenisciformes)	Antarctica	Barbosa & Palácios (2009)
	Pygoscelis adeliae (Sphenisciformes)		
	Eudyptes chrysocome (Sphenisciformes)		
	Eudyptes chrysolophus (Sphenisciformes)		
	Macronectes giganteus (Procellariiformes)		
	Chionis albus (Charadriiformes)		
	Pygoscelis antarcticus (Sphenisciformes)	Antarctica	Vidal et al. (2015)
Stegophorus arctowski	*Macronectes giganteus* (Procellariiformes)	Antarctica	Barbosa & Palácios (2009)
Stegophorus heardi	*Oceanites oceanicus* (Procellariiformes)	Antarctica	Barbosa & Palácios (2009)
	Pelecanoides georgicus (Procellariiformes)		
	Pelecanoides urinatrix (Procellariiformes)		
	Catharacta lonnbergi (Charadriiformes)		
Stegophorus stercorarii	*Cerorhinca monocerata* (Charadriiformes)	Japan	Yoshimo et al. (2015)
	Synthliboramphus antiquus (Charadriiformes)		

slides and identified in accordance with Mendonça & Rodrigues (1968), Rodrigues & Mendonça (1967) and Chabaud (2009).

The parasitological indexes, i.e. prevalence, mean intensity of infection (MII), mean abundance (MA) and range of infection (R), were calculated in accordance with Bush et al. (1997). Photomicrographs were taken using an Olympus BX 41 microscope coupled to a digital camera system. Representative specimens of nematodes were deposited in the "Coleção de Helmintos do Laboratório de Parasitologia de Animais Silvestres da Universidade Federal de Pelotas (LAPASIL/UFPel)" Rio Grande do Sul, Brazil, under no CHLAPASIL 682-684.

Results

Out of the total of 45 hosts examined, 16 (35.5%) were parasitized by acuariid helminths. *Seuratia shipleyi* Stossich, 1900, and *Stegophorus diomedeae* Johnston and Mawson, 1942 (Figure 1), were identified with prevalences of 26.1% and 21.7%, respectively (Table 2).

Seuratia shipleyi had greatest mean intensity of infection and mean abundance in *P. gravis* (MII = 39 helminths/host and MA = 19.5), while *Stegophorus diomedeae* showed highest rates in *D. exulans* (MII = 28 helminths/host and MA = 28). However, only

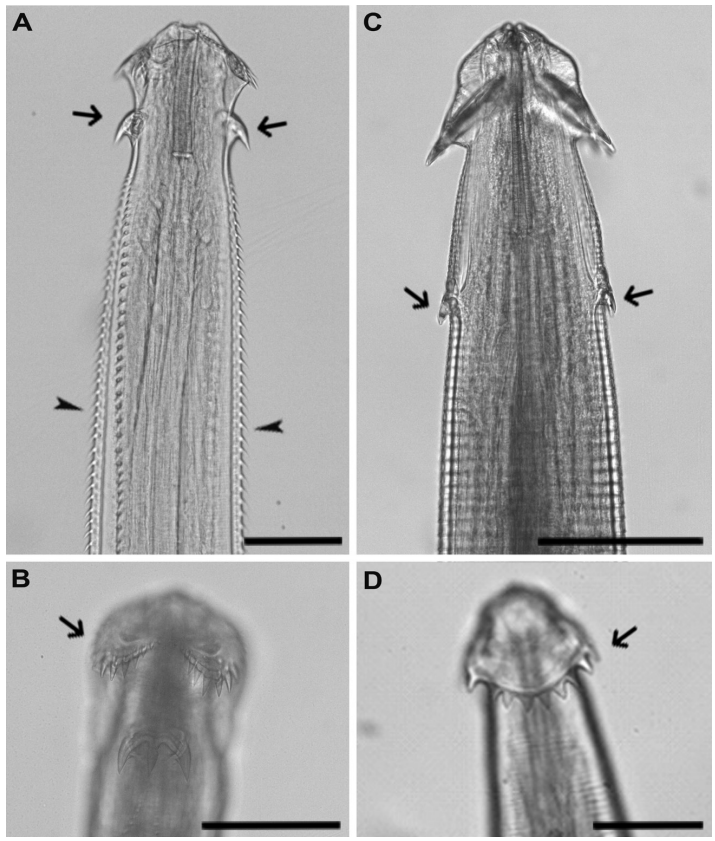

Figure 1. Seuratiinae (Acuariidae: Nematoda) parasite of Procellariiformes (Aves) on the southern coast of Rio Grande do Sul, Brazil. (**A-B)** *Seuratia shipleyi* parasite of *Puffinus puffinus*, (**A)** Ventral view, the arrows show the cervical papillae and the arrowheads show the row of spines (Bar = 142,5 µm); (**B)** Lateral view, the arrow show the hood (Bar = 150 µm); (**C- D)** *Stegophorus diomedeae* parasite of *Diomedea exulans*, (**C)** Ventral view, the arrows show the cervical papillae (Bar = 132,5 µm); (**D)** Lateral view, the arrow show the hood (Bar = 85 µm).

Table 2. Prevalence (P%), mean intensity of infection (MII), mean abundance (AM) and range of infection (R) of *Seuratia shipleyi* and *Stegophorus diomedeae* (Nematoda: Acuariidae) in Procellariiformes (n = 45), Rio Grande do Sul, Brazil.

Host	*Seuratia shipleyi*		MA	R	*Stegophorus diomedeae*		MA	R
	P%	MII			P%	MII		
Procellariiformes (n = 45)	26.1	7.8	2.0	1-37	21.7	5.2	1.1	1-24
Calonectris borealis (n = 4)	100.0	1.0	7.5	1-22	-	-	0	-
Diomedea exulans (n = 1)	-	-	0	-	100.0	28.0	28.0	24
Macronectes giganteus (n = 8)	-	-	0	-	25.0	2.0	0.5	2
Thalassarche chlororhynchos (n = 5)	-	-	0	-	20.0	4.0	0.8	4
Thalassarche melanophrys (n = 15)	13.3	1.5	0.6	1-2	6.6	2.0	0.1	2
Procellaria aequinoctialis (n = 4)	25.0	1.0	0.2	1	25.0	1.0	0.2	1
Puffinus gravis (n = 2)	50.0	39.0	19.5	2-37	50.0	7.0	3.5	7
Puffinus puffinus (n = 6)	50.0	7.0	3.5	3-14	50.0	2.3	1.2	1-4

a few hosts belonging to this species were examined. In contrast, in *T. melanophrys* (n = 15), the MII and MA for both acuariid species showed low values (Table 2).

Coinfection between the two acuariids was observed in four individuals (8.9% of the hosts): three specimens of *Puffinus puffinus* and one of *P. aequinoctialis*. In *P. puffinus*, it was observed that in the birds coinfected with these two species, *S. shipleyi* had higher mean intensity than *S. diomedeae*.

Discussion and Conclusion

Most of the studies that have reported Acuariidae in Procellariiformes have dealt with records of these helminths in different hosts and have generally only presented the prevalence index (HOBERG & RYAN, 1989; FOSTER et al., 1996). Similarly, there have been few studies on occurrences of coinfection between acuariid helminths (MENDONÇA & RODRIGUES, 1968; RODRIGUES & MENDONÇA, 1967).

Mendonça and Rodrigues (1968) and Rodrigues and Mendonça (1967) observed *S. shipleyi* and *S. diomedeae* in the proventriculus of a specimen of *T. melanophrys*, thus indicating that coinfection with these acuariids was present.

Hoberg & Ryan (1989) examined the gastrointestinal tract of *P. gravis* and identified *S. shipleyi* in the proventriculus of five birds (25%). (FOSTER et al. 1996) examined 15 specimens of *P. gravis*, from Florida, and reported *S. shipleyi* in the proventriculus and *Stegophorus stellaepolaris* and *S. diomedeae* in the ventricle, with prevalences of 60%, 20% and 93%, respectively. However, they did not report any data on coinfection.

Spotorno et al. (2005) examined 51 specimens of Procellariiformes that were found dead on Cassino beach (32° 22' S; 52° 18' W), in Rio Grande do Sul. They reported that 47% were infected with nematodes, and observed injuries in the proventriculus caused by *Seuratia* spp. in *Macronectes giganteus*, they reported ulcers, epithelial and gastric gland injuries and cell necrosis in the proventriculus. In *Macronectes halli*, presence of *Seuratia* spp. was associated with granuloma in the mucosa of the proventriculus. These authors commented that the damage caused by these nematodes might, in some cases, cause infection by bacteria, thus indirectly leading these birds' death. Barbosa & Palacios (2009) listed the parasites from seabirds and cited *Seuratia* and *Stegophorus* parasitizing various Charadriiformes, Procellariiformes and Sphenisciformes, but did not report the infection rates.

La Sala et al. (2012) examined 103 specimens of *Larus atlanticus* (Charadriiformes) in Argentina between 2005 and 2006. They found the acuariids *Pectinospirura argentata*, *Sciadiocara* sp. and *Skrjabinoclava andersoni* parasitizing the proventriculus of these hosts, with prevalence and mean intensity of 80.3% and 23.7 helminths/infected host, and 89.2% and 50.8 helminths/infected host, in 2005 and 2006, respectively. Mild inflammation in the proventriculus was caused by these nematodes because of their mucosal adherence and associated ulcerations.

This is the first record of parasitological indexes for both of these species of Acuariidae, *Seuratia shipleyi* and *Stegophorus diomedeae*, in Procellariiformes in Rio Grande do Sul, southern Brazil. It was observed that these helminths have low host specificity, since they occur not only in various species of Procellariiformes but also in Charadriiformes and Sphenisciformes. Regarding knowledge of helminth diversity in Procellariiformes, *S. shipleyi* is reported for the first time in *Calonectris borealis*, *Procellaria aequinoctialis, Puffinus gravis* and likewise, *Stegophorus diomedeae* in *Diomedea exulans*, *P. aequinoctialis*, *Thalassarche chlororhynchos*, *Macronectes giganteus* and *Puffinus puffinus.*

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