# A NEW HELIGMONELLID SPECIES (NEMATODA) FROM *OLIGORYZOMYS NIGRIPES* (RODENTIA: SIGMODONTINAE) IN THE ATLANTIC FOREST, BRAZIL

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ABSTRACT: Stilestrongylus lanfrediae n. sp. is described from the small intestine of Oligoryzomys nigripes (Cricetidae: Sigmodontinae) collected in the Atlantic Forest (Rio de Janeiro State, Teresópolis, Brazil). The new species shows some similarities to Stilestrongylus stilesi, Stilestrongylus freitasi, Stilestrongylus inexpectatus, Stilestrongylus moreli, and Stilestrongylus andalgala, but it can be distinguished from these species by the following combination of characters: 26 ridges in males and 25 in females at the mid-body, asymmetrical caudal bursa with a pattern of type 2-2-1, rays 6 markedly shorter than other lateral rays, rays 8 inserted asymmetrically on dorsal trunk and shorter than other species, and a proportion of spicule length in relation to body length (SpL/BL) of 25–29%. The new species is also distinguished from other species of Stilestrongylus by the asymmetry of the branches of the dorsal ray and by having the longest spicules.

*Stilestrongylus* Freitas, Lent, and Almeida, 1937 (Heligmonellidae: Nippostrongylinae) includes 23 Neotropical species, of which 22 are parasites of Sigmodontinae rodents and 1 of Hystricognathi rodents (Digiani and Durette-Desset, 2007). This genus is broadly distributed across the Neotropical region (Digiani and Durette-Desset, 2003).

The rice rat, *Oligorizomys nigripes* Olfers, 1818 (Cricetidae: Sigmdontinae), is frequently found in the Atlantic Forest in Brazil. These rodents are numerous in disturbed and secondary forests and can be agricultural pests in rice fields and storage barns (Mattevi and Andrades-Miranda, 2006).

During a parasitological survey of rodents in the Atlantic Forest, Brazil, numerous specimens of nematodes were collected that represent a new species. This new species of *Stilestrongylus* (Trichostrongylina: Nippostrongylinae) is described and illustrated herein.

# MATERIALS AND METHODS

Twenty specimens of *O. nigripes* (males and females) were captured in a forest fragment at Serra dos Órgãos, Teresópolis  $(22^{\circ}12'44''S, 42^{\circ}48'40''W)$ , Rio de Janeiro State, Brazil, between March and October 2005 using Sherman and Tomahawk live traps. Collection permits for rodents were issued by the Instituto Brasileiro de Meio Ambiente e Recursos Renováveis, Ministério do Meio Ambiente (IBAMA no. 068/ 2005). Animals were killed with CO<sub>2</sub> and dissected. Nematodes were collected from the small intestine, washed in physiologic solution, and fixed in hot AFA. Ten male and 10 female nematodes were cleared in lactophenol and examined using a Zeiss Standard 20 light microscope. Drawings were made with the aid of a camera lucida. Measurements are given in micrometers (unless otherwise stated) for holotype or allotype specimens, followed by range for paratypes, with the mean in parenthesis. Synlophe description follows Durette-Desset (1985), and

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terminology of the caudal bursa follows Durette-Desset and Chabaud (1981). The axis of synlophe orientation was characterized according to Durette-Dusset and Digiani (2005). Total number of ridges (dorsal/ventral) are given. Host nomenclature follows Musser and Carleton (2005).

# DESCRIPTION

# Stilestrongylus lanfrediae n. sp.

(Figs. 1–13)

General: Small nematodes tightly coiled in small intestine. Excretory pore within distal third of esophagus (Fig. 1). Deirids next to excretory pore (Fig. 1). Cephalic vesicle present. Triangular mouth opening in apical view. Two amphids, 6 internal labial papillae, 4 external labial papillae, and 4 submedian cephalic papillae connected to each other by weak arc-like cuticular structures (Fig. 2). Males and females with cuticle bearing longitudinal, uninterrupted ridges appearing posterior to cephalic vesicle and ending just anterior to caudal bursa in males, reaching posterior extremity in females. Synlophe with 23 ridges (10/13) in males and 25 (12/13) in females (Figs. 3, 4) at level of esophagealintestinal junction; 26 ridges (13/13) in males and 25 (12/13) in females (Figs. 5, 6) at mid-body; 23 (11/12) in males anterior to caudal bursa and 25 (12/13) in females anterior to vulva (Figs. 7, 8). At mid-body ridges slightly unequal in size with smaller ridges on ventral right and dorsal left quadrant (Figs. 5, 6). Double axis of orientation of ridges at mid-body in females: right axis inclined at 62° to sagittal axis and left axis at 72°. Single axis of orientation inclined at 55° to sagittal axis in males. Left ridge immediately ventral to axis of orientation larger than other ridges at esophageal-intestinal junction and at mid-body. Ridges of equal size just anterior to caudal bursa in males and just anterior to vulva in females. In males, most ridges perpendicular to body surface; in females, double axis of orientation with same inclination than at midbody.

Male (holotype and 10 paratypes, except otherwise stated): Length 4 [3.9-5.13 (4.42)] mm, 0.1 mm, n = 1, wide at mid-body. Cephalic vesicle 50 (60-70 [63]), n = 6, long and 20 [20-35 (29)], n = 8, wide. Excretory pore, deirids, and nerve ring situated 224 [170-290 (230)], n = 6, 210, n = 1, and 135 [100-158 (129)], respectively, from anterior end esophagus 333 [300-380 (340)] long (Fig. 1). Bell-shaped caudal bursa, difficult to distend out, with left lobe more developed than right lobe, both of type 2-2-1 (Figs. 9, 10). Rays 2 and 3 long, grouped in V-shape. Rays 4 slightly shorter than rays 5, both divergent at extremity. Rays 6 short, arising at about same level than rays 3, slightly distally. Rays 8 asymmetrical, left ray 8 thinner than right one and both shorter than dorsal ray. Rays 8 arising asymmetrically on dorsal trunk, left ray arising more distally than right one (Fig. 11). Dorsal ray asymmetrical, divided at about distal third into 2 branches, left branch thicker and slightly longer than right one, each branch divided at tip into 2 subequal branches: rays 9 (external) slightly longer than rays 10 (internal). Spicules alate, slightly unequal in length, 1.11 mm [0.90-1.23 (1.09)],

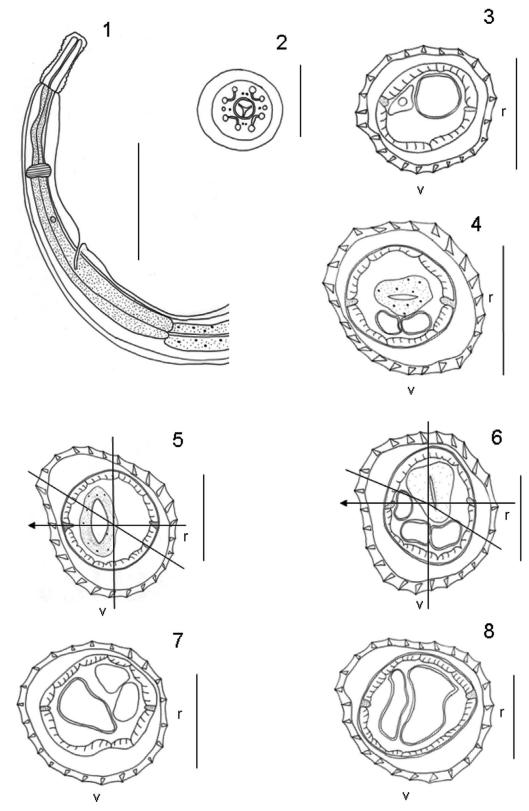
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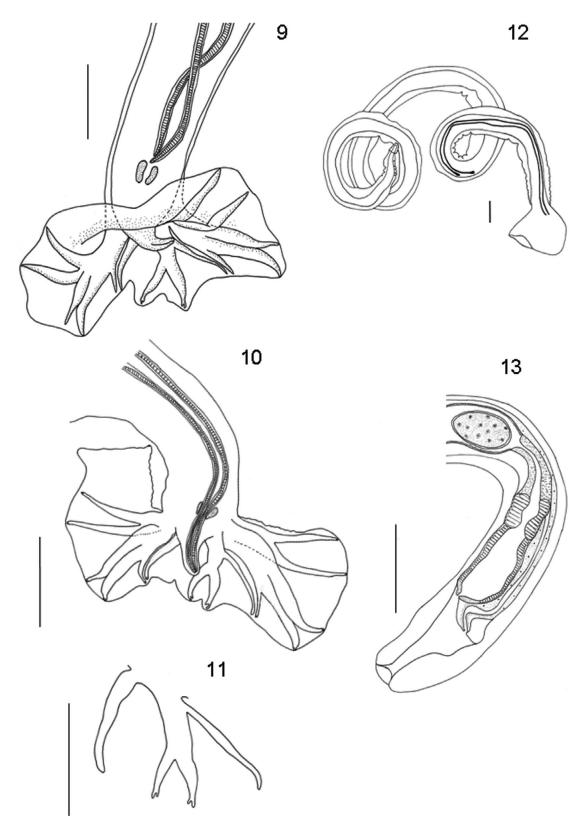
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FIGURES 1–8. *Stilestrongylus lanfrediae* n. sp. (1) Anterior extremity, right lateral view, male. Scale bar: 50  $\mu$ m. (2) Male, head, apical view. Scale bar: 25  $\mu$ m. (3) Section through anterior body, at posterior esophago-intestinal junction, male. Scale bar: 25  $\mu$ m. (4) Section through anterior body, at posterior esophago-intestinal junction, female. Scale bar: 25  $\mu$ m. (5) Transverse section of body, at mid-body, male. Scale bar: 25  $\mu$ m. (6) Transverse section of body, at mid-body, female. Scale bar: 25  $\mu$ m. (7) Transverse section of body, male, just anterior to caudal bursa. Scale bar: 25  $\mu$ m. (8) Transverse section of body, female, just anterior to vulva. Scale bar: 25  $\mu$ m. *Abbreviations:* r, right; v, ventral.



FIGURES 9–13. Stilestrongylus lanfrediae n. sp. (9) male, bell-shaped caudal bursa, ventral view, genital cone not represented. Scale bar: 100  $\mu$ m. (10) Male, caudal bursa after splitting of the ventral margin. Scale bar: 100  $\mu$ m. (11) Male, dorsal ray, ventral view. Scale bar: 100  $\mu$ m. (12) General view of male showing spicules proportion. Dorsal view. Scale bar: 100  $\mu$ m. (13) Female, posterior extremity, lateral view. Scale bar: 100  $\mu$ m.

n = 9 long, and 1.0 mm [0.80–1.06 (1.08)], n = 9, representing 27.7 (25–29%) spicule length in relation to body length (SpL/BL), ending in sharp tip (Fig. 12). Gubernaculum present, with 29 [17–30 (27)] long and 24 [24–30 (28)], n = 7, wide. Genital cone well developed with 68 [60–80 (68)], n = 7, long and 55 [48–70 (55)], n = 7, wide. Papillae on genital cone not observed.

Female (allotype and 10 paratypes, except otherwise stated): Length 4.93 [4.3–6.0 (4.4)] mm, 0.1 [0.09–0.14 (0.12)] mm wide at mid-body region. Cephalic vesicle present; 60 [50–70 (62)] long, 30 [20–30 (26)] wide. Distance from anterior end to excretory pore and nerve ring: 210 [190–320 (248)] and 130 [110–155 (138)], respectively. Derids not visible. Esophagus 320 [280–390 (351)] long. Monodelphic, posterior extremity strongly invaginated, with cuticular ridges reaching distal end. Vulva situated at 50 [35–58 (48)], n = 7, from caudal extremity, vagina vera 30 [20–50 (30)] long, vestibule 80 [50–100 (80)] long, sphincter 30 [30–40 (35)] long and 10 [10–15 (12.5)] wide and infundibulum 53 [53–79 (64)], n = 7, long, uterus 1,017 [800–1,248 (1,024)] long, number of eggs 15 [4–16 (10)], n = 4. Tail 35 [18–35 (28)], n = 7, long (Fig. 13). Eggs 0.07 [0.06–0.07 (0.065)] long, 0.03 [0.03–0.05 (0.036)] wide (Fig. 13).

#### **Taxonomic summary**

*Type host: Oligoryzomys nigripes* Olfers, 1818 (Cricetidae: Sigmodontinae). Common name, rice rats.

*Type locality:* Serra dos Órgãos, Teresópolis (22°12'44"S, 42°48'40"W), Rio de Janeiro State, Brazil.

Site of infection: Small intestine.

*Specimens deposited:* Holotype male (CHIOC no. 36925a), allotype female (CHIOC no. 36925b), and 20 paratypes (10 males and 10 females) (CHIOC no. 35536) were deposited in the Helminthological Collection of the Institute Oswaldo Cruz, Rio de Janeiro, Brazil.

*Etymology:* New species is named in honor of Dr. Reinalda Lanfredi for her contribution to helminthology.

## REMARKS

Durette-Desset (1971) defined Stilestrongylus Freitas, Lent, and Almeida, 1937 (Heligmonellidae: Nippostrongylinae), as including species with more than 24 cuticular subequal ridges in the synlophe at mid-body, a generally markedly asymmetrical caudal bursa, and a hypertrophied genital cone. In contrast, Hassalstrongylus (Durette-Desset, 1971) was defined as including species with 19 to 24 cuticular ridges, unequal in size, asymmetrical or subsymmetrical caudal bursa, and a genital cone that is not hypertrophied (Durette-Desset, 1971). Finally, it was remarked by Digiani and Durette-Desset (2007) that differentiation of Hassalstrongylus and Stilestrongylus has become somewhat problematic because of the description of several species in which the synlophe overlap among species of both genera. For these species it was not possible to decide, based only on the synlophe, whether they belong to one genus or another, and, in most cases, the generic attribution was based on the bursal characters. The specimens studied here belong to a group of species in which the number of ridges of synlophe show characters of both genera. However, the development of the genital cone and the dissymmetry of the caudal bursa are more similar to those found in species of Stilestrongylus. The dissymmetry of the caudal bursa in these specimens is determined not only by a different development of the lateral lobes, but also by the asymmetry of the dorsal ray branches and the difference in size, shape, and insertion of both rays 8. In particular, this latter character is present in most Stilestrongylus species, including the type species Stilestrongylus stilesi Freitas, Lent, and Almeida, 1937, whereas no species of Hassalstrongylus has an asymmetrical insertion of rays 8 on the dorsal ray.

These characters are thus considered sufficient to include the

new species in *Stilestrongylus*. The main characters of the new species are the dissymmetrical caudal bursa with a pattern of type 2-2-1, marked asymmetry of rays 8, small rays 6 and dorsal ray asymmetrical, a large number of ridges (26 in males and 25 in females at the mid-body), and the longest spicules in the genus (SpL more than 20% of BL).

The asymmetry of the dorsal ray branches seems to be a unique character among Stilestrongylus species, whereas the remaining characters are present in other species of the genus. Species having a pattern of type 2-2-1, asymmetrical insertion of rays 8, and SpL/BL greater than 20% include S. stilesi Freitas, Lent, and Almeida, 1937, in Holochilus balnearum (syn. H. chacarius), from Argentina; Stilestrongylus freitasi Durette-Desset, 1968, in Necromys lasiurus (syn. Zygodontomys lasiurus) and Cerradomys subflavus (syn. Oryzomys subflavus) from Brazil; Stilestrongylus inexpectatus Durette-Desset and Tchéprakoff, 1969, in Thrichomys apereoides (syn. Cercomys cunicularius) from Brazil; Stilestrongylus moreli Diaw, 1976, in Phyllotis boliviensis (syn. Auliscomys boliviensis) from Bolivia; and Stilestrongylus and algala Digiani and Durette-Desset, 2007, in Phyllotis sp. from Argentina. All these 5 species (with the exception of S. inexpectatus) have 26, or more, ridges in the synlophe and a marked asymmetry of both lobes of the caudal bursa.

The new species can be distinguished from *S. stilesi*, *S. freitasi*, and *S. inexpectatus* by having rays 2 and 3 separated in all their length. In addition, *S. lanfrediae* is differentiated from *S. stilesi*, *S. freitasi*, *S. inexpectatus*, *S. moreli*, and *S. andalgala* by having the shortest rays 6 and a distinct insertion, size and breadth of rays 8, and the highest SpL/BL.

Thus, the presence of particular characters of the caudal bursa and the SpL/BL allows us to consider the specimens from *O. nigripes* as a new species.

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## LITERATURE CITED

- DIGIANI, M. C., AND M. C. DURETTE-DESSET. 2003. Suttonema delta n.g., n. sp. (Nematoda: Trichostrongylina: Heligmosoidea) from Oxymycterus rufus (Rodentia: Sigmodontinae) in Argentina. Systematic Parasitology 55: 33–38.
- —, AND ——. 2007. Trichostrongylina (Nematoda) parasitic in *Phyllotis* sp. (Rodentia: Sigmodontinae) from Argentina, with description of three new species. Parasitology International **56**: 9–18.
- DURETTE-DESSET, M. C. 1971. Essai de classification des Nématodes Héligmosomes. Corrélation avec la paléobiogéographie des hôtes. Memoires du Museum National d'histoire Naturelle **49:** 1–126.
- ———. 1985. Trichostrongyloid nematodes and their vertebrate hosts: Reconstruction of the phylogeny of a parasitic group. Advances in Parasitology 24: 239–306.
- —, AND A. G. CHABAUD. 1981. Nouvel essai de classification des Nématodes Trichostrogyloidea. Annales de Parasitologie Humanie et Comparée 56: 297–312.
- —, AND M. C. DIGIANI. 2005. The axis of orientation of the synlophe in the Heligmosomoidea (Nematoda, Trichostrongylina): A new approach. Parasite **12:** 195–202.
- MATTEVI, M. S., AND J. ANDRADES-MIRANDA. 2006. Estudos genéticos

nos roedores da tribo Oryzomysi. *In* Mamíferos do Brasil: Genética, sistemática, ecologia e conservação, T. R. O. Freitas, E. Vieira, S. Pacheco, and A. Christoff (eds.). Suprema, São Paulo, Brazil, p. 107–137. MUSSER, G. G., AND M. D. CARLETON. 2005. Family Muridae. *In* Mammal species of the world: A taxonomic and geographic reference, D. E. Wilson, and D. A. M. Reeder (eds.). Johns Hopkins University Press, Baltimore, Maryland, p. 1039–1045.