



A new species of *Paraheligionella* (Nematoda, Heligionellidae), parasite of *Sylvilagus floridanus* (Leporidae) from Costa Rica

Una especie nueva de *Paraheligionella* (Nematoda, Heligionellidae), parásita de *Sylvilagus floridanus* (Leporidae) en Costa Rica

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Abstract. *Paraheligionella lamothei* n. sp. (Heligionellidae: Heligionellinae) is described from the small intestine of a cottontail, *Sylvilagus floridanus* (Allen, 1890) (Leporidae), from Costa Rica. New morphological data on the synlophe and caudal bursa of *Paraheligionella romerolagi* (Gibbons and Kumar, 1980), the most similar species, are also provided. The new species differs from *P. romerolagi*, parasitic of *Romerolagus diazi* (Ferrari-Pérez, 1893) from Mexico, mainly by the characters of the caudal bursa and the synlophe of the female within the posterior region of body. Males of *P. lamothei* n. sp. possess a caudal bursa not bell-shaped, with a pattern of type 2-2-1 for right lobe and 2-3 with a tendency to type 2-2-1 for left lobe, whereas males of *P. romerolagi* possess a bell-shaped caudal bursa, with a pattern of type 2-2-1 with a tendency to type 4-1 for both lobes. In *P. lamothei* n. sp. rays 3 are slightly longer than rays 2 and the genital cone is poorly developed, whereas in *P. romerolagi* rays 3 are much longer than rays 2 and the genital cone is large and bulbous. Females of *P. lamothei* n. sp. possess, at ovejector level, ventral and latero-ventral ridges hypertrophied and dorsal ridges reduced, whereas at the same level, females of *P. romerolagi* possess lateral ridges hypertrophied and dorsal and ventral ridges reduced. Additionally, hypertrophied cuticular ridges posterior to the vulva are present in *P. lamothei* n. sp., but absent in *P. romerolagi*.

Key words: *Paraheligionella lamothei* n. sp., *Paraheligionella romerolagi*, Trichostrongylina, Heligmosomoidea, lagomorphs, Neotropical region.

Resumen. Se describe *Paraheligionella lamothei* n. sp., parásita del intestino delgado de *Sylvilagus floridanus* (Allen, 1890) (Leporidae) de Costa Rica. Se aportan además datos nuevos morfológicos sobre el synlophe y la bolsa caudal de *Paraheligionella romerolagi* (Gibbons y Kumar, 1980), la especie morfológicamente más parecida. La especie nueva se diferencia de *P. romerolagi*, parásita de *Romerolagus diazi* (Ferrari-Pérez, 1893) de México, principalmente por caracteres de la bursa caudal y el synlophe de la hembra en la región posterior del cuerpo. Los machos de *P. lamothei* n. sp. poseen una bursa caudal sin forma de campana, con un patrón de tipo 2-2-1 para el lóbulo derecho y 2-3 con tendencia a tipo 2-2-1 para el lóbulo izquierdo, mientras que los machos de *P. romerolagi* poseen una bursa en forma de campana, con un patrón de tipo 2-2-1 con tendencia a tipo 4-1 para ambos lóbulos. En *P. lamothei* n. sp. los rayos 3 son ligeramente más largos que los rayos 2 y el cono genital está poco desarrollado, mientras que en *P. romerolagi* los rayos 3 son mucho más largos que los rayos 2 y el cono genital es grande y bulboso. Las hembras de *P. lamothei* n. sp. poseen, a la altura del oviector, las crestas cuticulares ventrales y latero-ventrales hipertrofiadas y las dorsales y laterales reducidas, mientras que a la misma altura, las hembras de *P. romerolagi* poseen las crestas laterales hipertrofiadas y las dorsales y ventrales reducidas. Adicionalmente, las crestas cuticulares hipertrofiadas posteriores a la vulva en *P. lamothei* n. sp., están ausentes en *P. romerolagi*.

Palabras clave: *Paraheligionella lamothei* n. sp., *Paraheligionella romerolagi*, Trichostrongylina, Heligmosomoidea, lagomorfos, región neotropical.

Introduction

The genus *Paraheligionella* Durette-Desset, 1971 was established to include those species of Heligionellidae with no more than 14 cuticular ridges and a hypertrophy of the ridges adjacent to the lateral fields. Species originally included are *Paraheligionella interrogans* (Lent and Freitas, 1938), the type species, and *Paraheligionella cubaensis* (Pérez-Vigueras, 1943). A third species, *Paraheligionella romerolagi* (Gibbons and Kumar, 1980), originally described as *Boreostrongylus romerolagi* Gibbons and Kumar, 1980, was transferred to *Paraheligionella* by Durette-Desset and Santos (2000). The earliest description of *P. romerolagi* was provided by Bravo-Hollis (1950), under the name of *Longistriata dubia* (Travassos, 1921) Travassos and Darriba, 1929. Gibbons and Kumar (1980), upon re-examination of that material, considered the specimens studied by Bravo-Hollis as identical to *B. romerolagi*. During a survey of eukaryotic parasites of vertebrates inhabiting the Área de Conservación Guanacaste, Costa Rica, an undescribed species of *Paraheligionella* was found in the small intestine of *Sylvilagus floridanus* (Allen, 1890) (Leporidae). This species is herein described and illustrated. Syntypes of *P. romerolagi* were re-examined for comparative purposes, and new morphological data on the synlophe and caudal bursa of this species are also provided.

Materials and methods

The specimen of *S. floridanus* was captured in the Área de Conservación Guanacaste, Costa Rica, in May, 2003, and the nematodes recovered were preserved in 70% ethanol. Syntypes of *P. romerolagi* [labelled as *L. dubia sensu* Bravo-Hollis, 1950, *nec* (Travassos, 1921) Travassos and Darriba, 1929] were borrowed from the Colección Nacional de Helmintos, México (CNHE N° 1919). Only 1 male and 1 female among the borrowed material were available for sectioning. Some serial sections broken in 1 point were, however, informative and consequently were included among the figures. The synlophe of both species was studied following the method of Durette-Desset (1985) and the nomenclature used for the study of the caudal bursa is that of Durette-Desset and Chabaud (1981). The cuticular ridges are numbered from left to right as 1 to 6 for the dorsal ridges and as 1' to 8' for the ventral ridges. Left and right hypertrophied ridges are counted among ventral and dorsal ridges, respectively. Measurements (holotype/allotype followed by range and mean of paratypes in parentheses) are in micrometers except where stated otherwise. The parasite classification

used above the family group level is that of Durette-Desset and Chabaud (1993) and the nomenclature of the hosts at the species level follows Wilson and Reeder (2005).

Description

Paraheligionella lamothei n. sp. (Figs. 1-19)

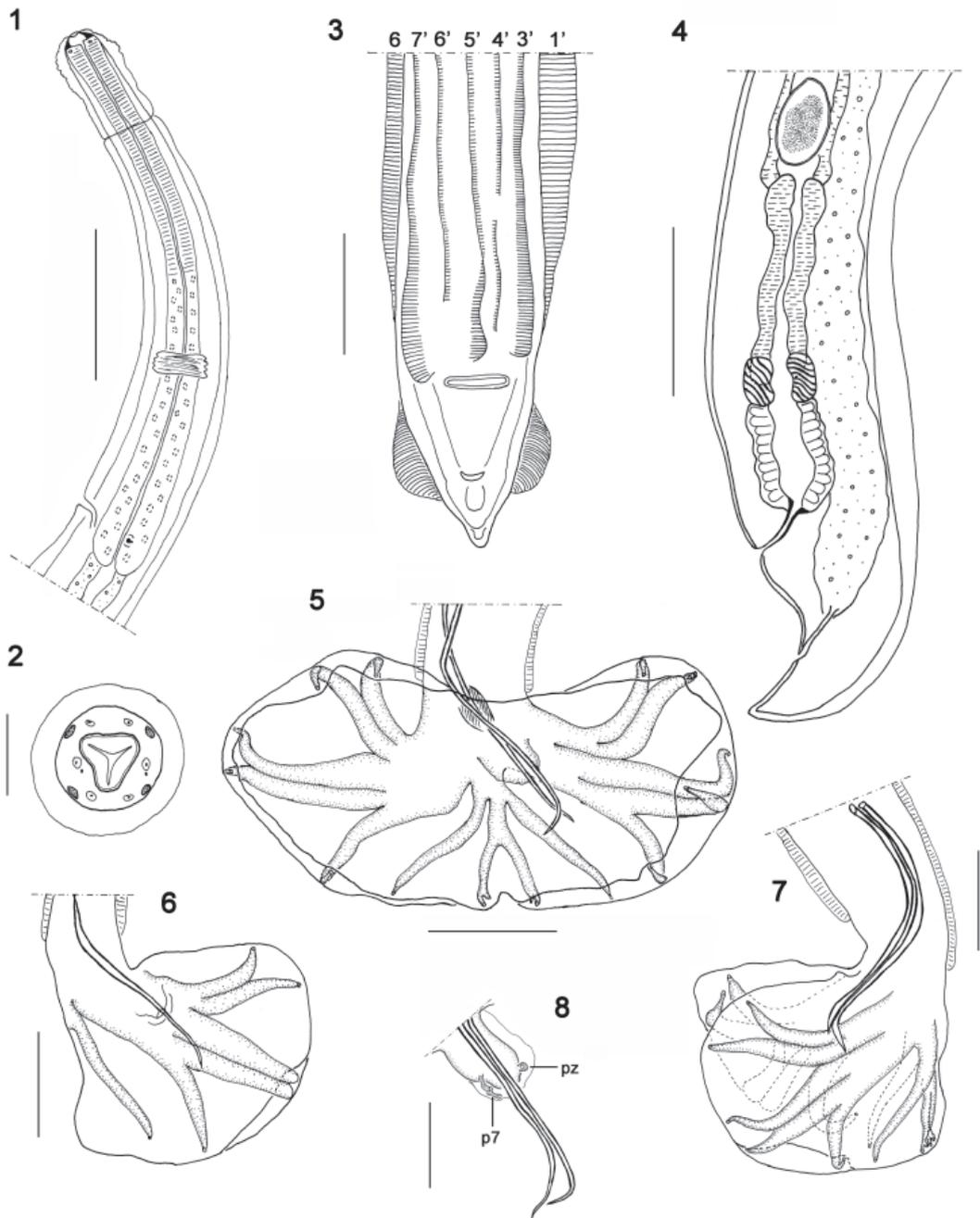
General. Heligmosomoidea: Heligionellidae. Small nematodes, curved ventrally. Body loosely coiled in male, strongly coiled in female, following 4-5 spirals. Excretory pore situated between 80 and 97% of esophagus length. Deirids situated at same level as excretory pore or slightly anterior or posterior to it (Fig. 1). Uterus taking up 15-17% of body length.

Head. Cephalic vesicle well-developed. In apical view, triangular oral opening with round corners surrounded by small buccal ring. Six externo-labial papillae, 2 amphids and 4 cephalic papillae present (Fig. 2).

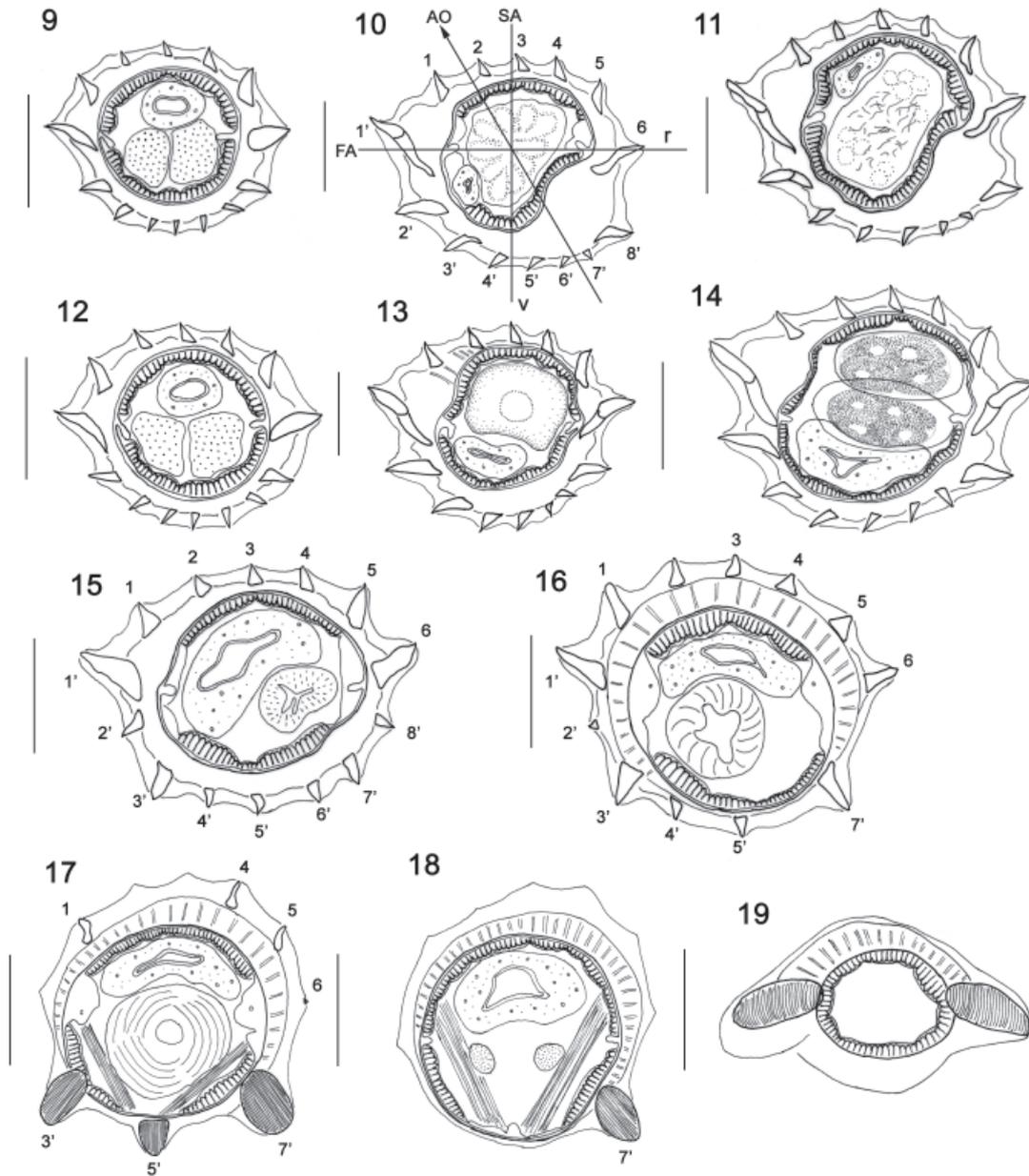
Synlophe (studied in 1 male and 1 female paratypes). In both sexes, body bearing continuous uninterrupted cuticular ridges with chitinous struts, all appearing just posterior to cephalic vesicle, except dorsal ridge n° 3 appearing at about mid-length of esophagus and ventral ridge n° 5' appearing at distal end of esophagus. Ridges disappearing just anterior to caudal bursa in male and at level of anus in female (Fig. 3). Number of ridges: 14 (1 lateral left, 1 lateral right, 5 dorsal, 7 ventral) in both sexes present all along body except in ovejector region in female. Size of ridges: lateral ridges (1' and 6') hypertrophied. Ridges of ventral, ventral right quadrant smallest. At mid-body, gradient of size of ridges latero-median, from right to left on dorsal side, from left to right on ventral side. In both sexes, single axis of orientation directed from right ventral quadrant to left dorsal quadrant, passing between ridges 1 and 2 on left side and between ridges 6' and 7' on right side, inclined at 30° to sagittal axis (Figs. 10, 13).

Synlophe modified at level of ovejector (Figs. 3, 15-18). From level of infundibulum (Fig. 15), progressive reduction of dorsal and lateral ridges and hypertrophy of ventral (n° 5') and ventro lateral ones (n° 3' and n° 7') reaching maximum development at prevulvar level (Fig. 17). No ridges at level of vulva, except ridge 7' in some specimens (Figs. 3, 18). Between vulva and anus presence of 2 alae with thick chitinous strut (Figs. 3, 19).

Males (Holotype and 5 paratypes). 6.60 (5.95-6.95; 6.42) mm long and 100 (100-150; 130) wide at mid-body; cephalic vesicle 68 (68-75; 71) long and 40 (37-40; 39) wide; nerve ring, excretory pore and deirids situated at 195 (170-210; 187), 330 (290-340; 325) and 322 (295-340; 324) from apex, respectively; esophagus, 350 (342-



Figures 1-8. *Paraheligmonebella lamothei* n. sp. 1, male, anterior extremity, left lateral view. 2, female, head, apical view. 3, female, posterior extremity, arrangement of cuticular ridges, ventral view. 4, female, details of ovejector, left lateral view. 5-7, male, caudal bursa. 5, ventral view. 6, right lobe, latero-dorsal view. 7, left lobe, latero-dorsal view. 8, male, genital cone, right lateral view. Abbreviations: pz, papilla zero; p7, papillae 7. Scale-bars: all figures 100 μ m, except 2 (20 μ m) and 8 (50 μ m).



Figures 9-19. *Paraheligmone lamothei* n. sp. Transverse sections of body. 9-11, male, 6.75 mm long. 9, at esophago-intestinal junction. 10, at mid-body (3.35 mm from apex). 11, at 1.5 mm anterior to caudal bursa. 12-19, female, 8.25 mm long. 12, at esophago-intestinal junction. 13, at mid-body (3.75 mm from apex). 14, within median part of uterus. 15, within distal infundibulum. 16, at level of sphincter. 17, at level of vestibule. 18, at level of vulva. 19, at level of anus. Abbreviations: r, right side; v, ventral side; AO, axis of orientation; FA, frontal axis; SA, sagittal axis. All figures orientated as figure 10. Scale-bars: all figures 50 μm.

370; 354) long, representing 5.3% (4.9-5.9; 5.5%) of body length.

Caudal bursa (n=3): Subsymmetrical, with pattern of type 2-2-1 on right lobe and of type 2-3 with tendency to type 2-2-1 on left lobe (Figs. 5-7). Rays 2 slightly shorter than rays 3, both parallel and joined for more than half of their length. Rays 4 and 5 of similar length, diverging at posterior third of their length (Figs. 5, 7). Rays 6 diverging from common trunk 2-6 at same level than ray 3 in right lobe and slightly distally to ray 3 in left lobe. Rays 8 arising symmetrically from base of dorsal ray, slightly longer than it. Dorsal ray divided into 2 branches at about mid-length, each branch giving rise to 2 small branches, rays 9 (external branches) slightly longer than rays 10 (internal branches) (Fig. 5).

Spicules 285 (265-310; 284) long, each ending in sharp tip. (Figs. 5, 7-8). Spicules taking up 4.3 % (3.9-4.8; 4.4)% of body length. Genital cone 45 (40-50; 47.6) long and 40 (32-40; 37.8) wide at its base. Single sessile papilla zero on ventral lip and pedunculated papillae 7 on dorsal lip (Fig. 8). Gubernaculum not seen or barely visible in some paratypes, 30-32 long (n=3) (Fig. 5).

Females (Allotype and 7 paratypes). 8.2 (6.9-8.6; 8.0) mm long; 110 (100-120; 113) μ m wide at mid-body, cephalic vesicle 70 (70-75; 72) long and 40 (33-45; 40) wide; nerve ring, excretory pore and deirids situated at 190 (155-230; 198), 335 (305-345; 326) and 315 (305-350; 327) from apex, respectively; esophagus 400 (340-400; 378) long, 4.9% (4.3-4.9; 5.0%) of body length. Monodelphic. Vulva situated at 100 (95-135; 110) from apex; vagina vera 35 (30-40; 36) vestibule 65 (55-90; 68) long, sphincter 25 (30-35; 31) long and 42 (30-40; 38) wide, infundibulum 130 (80-125; 107); uterus 1.28 (1.04-1.49; 1.31) mm long; containing 78 (62-90; 73) eggs at blastomere stage, 50-60 (52-70; 58.6) long and 28-30 (28-38; 31.8) wide; tail 30 (30-55; 36) long; uterus taking up 15.6 % (15.1-17.3; 16%) of body length.

Taxonomic summary

Type-host: *Sylvilagus floridanus* (Allen, 1890) (Leporidae).

Site of infection: small intestine.

Type-locality: Sector El Hacha, Los Almendros, Guanacaste, Costa Rica. 11°00'00''N, 85°32'0.9''W.

Prevalence and intensity of infection: 1 out of 3 examined hosts harboured 6 males and 8 females.

Type specimens: holotype, allotype, and 4 paratypes deposited in the Colección Nacional de Helminthos, Costa Rica: holotype CHCR N° 284, allotype CHCR N° 285, 1 male paratype CHCR N° 286, 3 female paratypes CHCR

N° 287. Other paratypes: 1 male in the Helminthological Collection of the Muséum national d'Histoire naturelle, Paris, France, MNHN 406 MQ; 1 male, 2 females in the Helminthological Collection of the Museo de La Plata, La Plata, Argentina, CHMLP-5639; 1 male, 1 female in the Colección Nacional de Helminthos, México, D.F., Mexico, CNHE-6029; 1 male, 1 female in the United States National Parasite Collection, USNPC-100623.

Etymology: in honor of Prof. Rafael Lamothe Argumedo, in recognition of his contributions to the knowledge of the biodiversity of helminths of wildlife of the Americas.

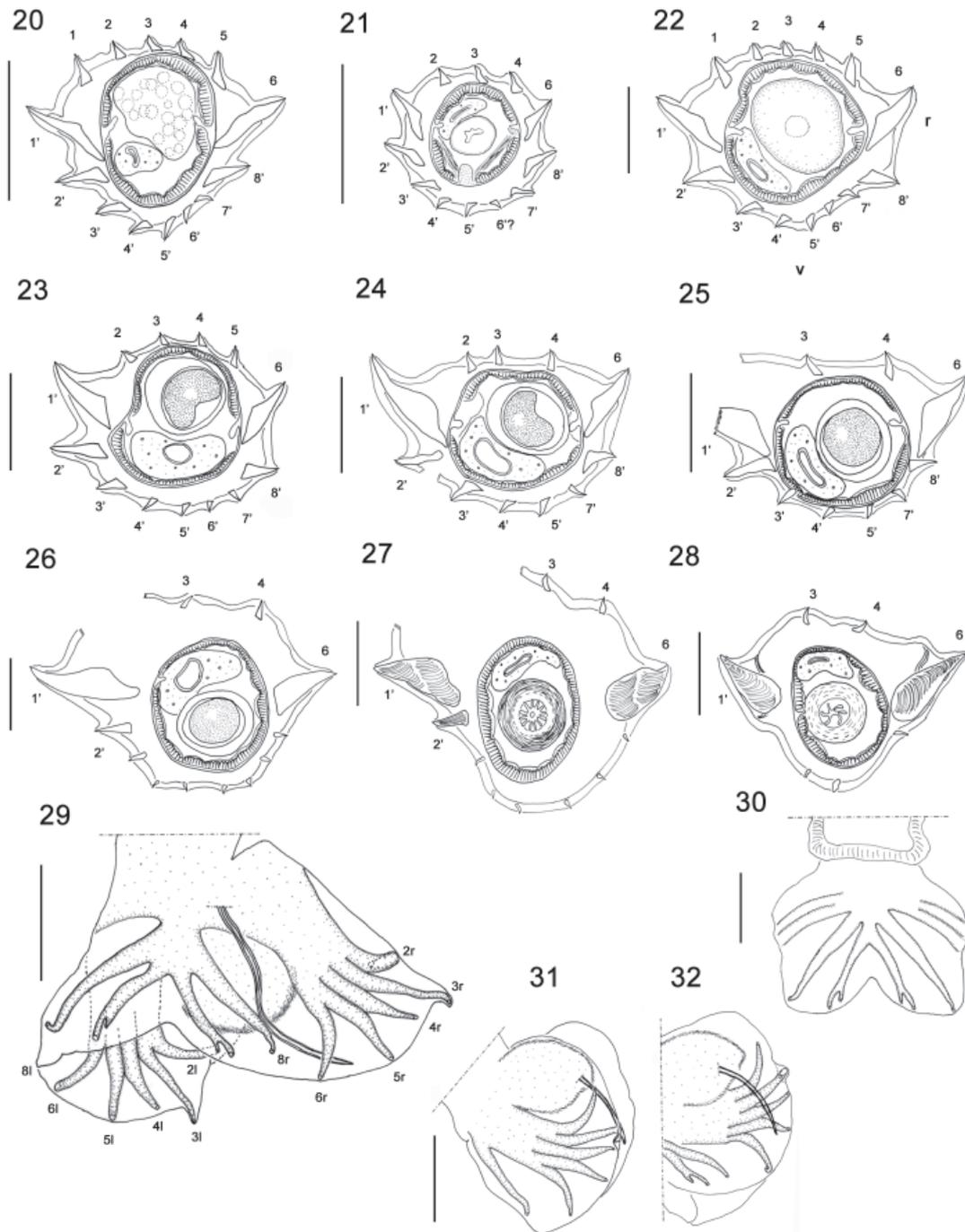
New morphological data on *Paraheligionella romerolagi* (Gibbons and Kumar, 1980) (Figs. 20-32)

(= *Boreostrongylus romerolagi* Gibbons and Kumar, 1980)

(= *Longistriata dubia sensu* Bravo-Hollis, 1950 *nec* (Travassos, 1921) Travassos and Darriba, 1929).

Synopse. At mid-body in both sexes: 14 ridges (hypertrophied lateral ones, 5 dorsal, 7 ventral) (Figs. 20, 22). Single axis of orientation directed from right ventral quadrant to left dorsal quadrant, passing between ridges 1 and 2 on left side and between ridges 6' and 7' on right side, inclined at about 30° to sagittal axis. In male, at 220 μ m anterior to caudal bursa: 12 ridges (2 lateral ones, 3 dorsal, 7 ventral); lateral ridges (1' and 6) still well developed but not hypertrophied; ridge 6' orientated as 7' and double axis of orientation passing between ridges 1' and 2 on left side and between 5' and 6' on right side (Fig. 21). In female, at level of proximal portion of uterus ridge 1 has disappeared (Fig. 23). From level of distal portion of uterus, dorsal ridges disappear progressively: first ridge 5, then ridge 2, and rays 3 and 4 become more separated. At this level ventral ridge 6' disappeared (Figs. 24-25). Just at level of distal infundibulum, presence of ventral inflation supported by 8 ridges (Fig. 26) then ventral ridges reduce to 7 at level of sphincter (Fig. 27). At level of proximal vestibule, body diameter decreasing and ventral ridges reduce to 3 (Fig. 28). All along body, lateral ridges (1' and 6) hypertrophied and dorsally directed (Figs. 24-28). At vulvar level: body diameter still decreasing markedly. In specimen sectioned, posterior end is ventrally bent at this level and sectioning of the body becomes difficult. Hypertrophied lateral ridges disappear slightly posterior to the vulva, and no new ridges are visible on the caudal region.

Male caudal bursa (2 specimens mounted). symmetrical, bell-shaped. Pattern of caudal bursa of type 2-2-1 with a tendency to type 4-1 due to rays 6 arising slightly first from common trunk of rays 2 to 6. Rays 2 and 3 on the one hand, 4 and 5 on the other hand strongly divergent. Divergence



Figures 20-33. *Paraheligmone romerolagi* (Gibbons and Kumar, 1980). 20-29, transverse sections of body. 20-21, male, 3.85 mm long. 20, at mid-body (1.70 mm from apex). 21, at 220 μ m anterior to caudal bursa. 22-28, female, 6.6 mm long. 22, at mid-body (3.40 mm from apex). 23, within proximal uterus (600 μ m anterior to posterior extremity). 24, at level of proximal infundibulum (ca.250 μ m anterior to vulva). 25, at level of middle infundibulum. 26, at level of distal infundibulum. 27, at level of sphincter (ca. 130 μ m anterior to vulva). 28, at level of proximal vestibule. 29-32, male, caudal bursa. 29, entire bursa, right latero-dorsal view. 30, dorsal lobe, dorsal view. 31, right lobe, latero-dorsal view. 32, left lobe, latero-ventral view. Abbreviations: r, right side; v, ventral side; 2l, left ray 2; 2r, right ray 2. All figures orientated as figure 22. Scale-bars: all figures 50 μ m.

of rays 2-3 proximal to those of rays 4-5. Rays 3 very long, forming a protuberance on bursal margin (Figs. 29, 31-32). Rays 8 arising symmetrically from base of dorsal ray, about same length than dorsal ray. Dorsal ray divided into 2 branches at varying levels within proximal half. Each branch divided into 2 sub-branches: external (rays 9) longer than internal (rays 10) (Figs. 29-30). Gubernaculum not observed. Genital cone large and bulbous, ca. 60 long and 50 wide. Papillae on genital cone not observed.

Material studied: 2 males, 2 females, syntypes CNHE-1919.

Host: *Romerolagus diazi* (Ferrari-Pérez, 1893) (Leporidae).

Site of infection: small intestine.

Locality: Cerro Pelado, 33 km. S, 5 km. NW, México, D.F., Mexico.

Remarks

The specimens studied from *S. floridanus* possess the main characters of the genus *Paraheligionella*, characterised by a synlophe with 14 or fewer cuticular ridges and hypertrophy of the ridges adjacent to the lateral fields. Three species are described in the genus, parasitic in rodents (Echimyidae and Capromyidae) and lagomorphs in the Neotropical region: *P. cubaensis*, parasitic in *Capromys pilorides* (Say, 1822) (Capromyidae) from Cuba, *P. interrogans*, parasitic in *Thrichomys apereoides* (Lund, 1839) (Echimyidae) (= *Cercomys cunicularius*) from Brazil and *P. romerolagi*, parasitic in *R. diazi* (Leporidae) from Mexico.

The new species is readily distinguished from *P. cubaensis* by the structure of the synlophe and the pattern of the caudal bursa: in *P. cubaensis*, the right ventral and left dorsal ridges are reduced or poorly developed and the caudal bursa has a pattern of type 1-4 (Pérez-Vigueras, 1943, Baruš and Ryšavý, 1967, Durette-Desset, 1972). It is also distinguished from *P. interrogans* by the structure of the synlophe and the pattern of the caudal bursa: in *P. interrogans* the synlophe possess 13 cuticular ridges, with dorsal ridges discontinuous, whereas the caudal bursa has a pattern of type 2-3 and the dorsal ray divided at its base (Durette-Desset, 1968).

The Costa Rican specimens closely resemble *P. romerolagi*, especially by having the synlophe at mid-body. Both species have 14 ridges with a similar gradient of size, and a single axis of orientation passing between ridges 1 and 2, with similar degree of inclination. The bursal pattern of *P. romerolagi* was not clearly illustrated in the previous descriptions because the bursa was not spread out. The examination of syntypes of *P. romerolagi*

provided some important differences between both species: in *P. lamothei*, rays follow a pattern of type 2-2-1 (rays 2-3 grouped on the one hand, 4-5 grouped on the other hand, and ray 6 diverging separately at about same level than ray 3) or 2-3 with tendency to type 2-2-1 (rays 2-3 grouped and rays 4-6 having a short common trunk). In *P. romerolagi* the pattern is a type 2-2-1 rather atypical, with rays 6 diverging the most proximally from the common trunk 2 to 6. In *P. lamothei* n. sp. rays 2-3 on the one hand, and 4-5 on the other hand are joined over at least half of their length, diverging distally, whereas in *P. romerolagi* the divergence of the rays 2-3 and 4-5 is much proximal. Additional differences are the shape of the bursa, bell-shaped in *P. romerolagi* and not bell-shaped in *P. lamothei* n. sp.; the length of rays 3, much longer in *P. romerolagi* than in *P. lamothei* n. sp.; the development of the genital cone, poorly developed in *P. lamothei* n. sp., large and bulbous in *P. romerolagi*; and the spicule tips, separated in *P. lamothei* n. sp. and joined in *P. romerolagi*.

Additional differences between both species were observed in the posterior extremity of females and in the synlophe at ovejector level. The earliest description of *P. romerolagi* was provided by Bravo-Hollis (1950), under the name of *L. dubia*. This author, in her description of females, mentioned and figured the presence of 2 alae-like cuticular thickenings lateral to the vulva. The re-examination of Bravo-Hollis' specimens showed that such cuticular thickenings are actually the lateral ridges 1' and 6, hypertrophied and dorsally directed. Thus, the main difference is that, in *P. lamothei*, from the level of infundibulum the lateral ridges become progressively reduced and the ventral and latero-ventral ridges hypertrophy; whereas, in *P. romerolagi*, the lateral ridges progressively hypertrophy, as the dorsal and ventral ones become reduced. Additionally, the females of *P. romerolagi* differ from the specimens from *S. floridanus* by the absence of hypertrophied ridges posterior to vulva.

The differences mentioned herein are considered enough to propose that the specimens from *S. floridanus* from Costa Rica as a new species.

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