

FREE-LIVING DORYLAIMID NEMATODES FROM NATURE RESERVES IN COSTA RICA. THE GENUS *PACHYDORYLAIMUS* SIDDIQI, 1983

A. Esquivel,¹* P. Guerrero,² R. Peña-Santiago² and T. Powers³

¹Laboratorio de Nematología, Escuela de Ciencias Agrarias, Universidad Nacional, Costa Rica, Apartado 83-3000, Heredia, Costa Rica; ²Departamento de Biología Animal, Vegetal y Ecología, Universidad de Jaén, Campus "Las Lagunillas" s/n, Edificio B3, 23071-Jaén, Spain; and ³Department of Plant Pathology, University of Nebraska, 406 Plant Sciences Hall, Lincoln, NE 68583, U.S.A. *Corresponding author: aesquive@una.ac.cr.

ABSTRACT

Esquivel, A., P. Guerrero., R. Peña-Santiago and T. Powers. 2007. Free-living dorylaimid nematodes from nature reserves in Costa Rica. The genus *Pachydorylaimus* Siddiqi, 1983. *Nematropica* 37:317-333.

A taxonomic study of three species, two known and one new, belonging to the genus *Pachydorylaimus* Siddiqi, 1983 from natural areas in Costa Rica is presented. *Pachydorylaimus holovachovi* sp. n. is described and can be distinguished by its body 0.90-1.00 mm long, lip region 14-17 μ m wide, odontostyle 13-15 μ m long, odontophore morphology with differentiated hyaline spindle-shaped area surrounding its junction to pharyngeal lining but lacking distinct basal flanges, total stylet length 35-44 μ m, *pars refringens vaginae* absent, $V = 43-47$, female tail straight and tapering to an acute tip (57-78 μ m, $c = 12.5-15.8$, $c' = 2.0-3.0$), and males unknown. Costa Rican populations of *P. notabenus* and *P. schizodontus* are compared with their respective original descriptions. Descriptions, measurements and illustrations (light microscope pictures) are provided for the three species; a line illustration of *P. holovachovi* sp. n. is presented as well as SEM photographs of *P. holovachovi* and *P. notabenus*. Finally, a key to identification of *Pachydorylaimus* is given together with a compendium of their main measurements and ratios. *Pachydorylaimus notabenus* is a new record for Costa Rican nematode fauna.

Key words: Costa Rica, nematode diversity, neotropical forest, *Pachydorylaimus*, taxonomy.

RESUMEN

Esquivel, A., P. Guerrero., R. Peña-Santiago y T. Powers. 2007. Nematodos dorylaimidos de vida libre de áreas protegidas de Costa Rica. El género *Pachydorylaimus* Siddiqi, 1983. *Nematropica* 37:317-333.

Se presenta un estudio sobre tres especies, dos conocidas y una nueva, pertenecientes al género *Pachydorylaimus* Siddiqi, 1983 recolectadas en áreas protegidas de Costa Rica. *Pachydorylaimus holovachovi* sp. n. puede distinguirse por su cuerpo 0.90-1.00 mm de largo, región labial 14-17 μ m de ancho, odontostilo 13-15 μ m de largo, odontoforo con un área diferenciada hialina en forma de huso rodeando su unión con el revestimiento de la faringe, aunque sin rebordes basales definidos, longitud total del estilote igual a 35-44 μ m, *pars refringens vaginae* ausente, $V = 43-47$, cola de la hembra recta estrechándose hasta un extremo afilado (57-78 μ m, $c = 12.5-15.8$, $c' = 2.0-3.0$), y machos desconocidos. Las poblaciones costarricenses de *P. notabenus* y *P. schizodontus* se comparan con sus respectivas poblaciones originales. Se proporcionan descripciones, medidas e ilustraciones (fotografías tomadas con microscopio óptico) para las tres especies; además, se incluyen dibujos de *P. holovachovi* y fotografías SEM de *P. holovachovi* y *P. notabenus*. Finalmente, se presenta una clave para la identificación del género *Pachydorylaimus*, junto con un compendio de sus principales medidas e índices. *Pachydorylaimus notabenus* constituye una nueva cita para la nematofauna de Costa Rica.

Palabras clave: bosques neotropicales, Costa Rica, diversidad de nematodos, *Pachydorylaimus*, taxonomía.

INTRODUCTION

Free-living dorylaimid nematodes have received surprisingly little attention in spite of their relatively high levels of diversity in neotropical forests. Nevertheless, there are relevant contributions (Andrássy, 1967, 1997; Zullini, 1973; Siddiqi, 1982, 1983; Loof, 1964) that indicate taxonomic studies in these regions are ideal for an in-depth evaluation of the origin, extent, and significance of nematode diversity. Costa Rica is well-known as a "hotspot" of biological diversity (Janzen, 1983; INBio, 2007; WCMC, 2007). The geographic history, complexity of the physical environment, and the remarkable range of bioclimates contributes to flora and fauna that are estimated to include 4% of the total global biodiversity (Obando, 2002). The study of Costa Rica's nematode fauna has increased in recent years as a result of international cooperative projects, reflected by the growth of the Nematode Collection at Universidad Nacional which currently contains over 20,000 specimens, including more than 8000 dorylaims collected from a variety of habitats (i.e., soil, litter, fresh water sediments, mosses, and bromeliads). Only a small part of the collection has been studied taxonomically (Bongers *et al.*, 2003; Esquivel, 2003). Loof and Zullini (2000), Ahmad and Shaheen (2004), and Shaheen and Ahmad (2004), have provided the first contributions on Costa Rican dorylaims.

The genus *Pachydorylaimus* Siddiqi, 1983 is remarkable, in part, due to its restriction to Neotropical forests across a broad range of climatic zones. It was originally discovered in Colombian rain forests by Siddiqi (1983), who described four new species. Later, Andrássy (1997) found two other species at high altitudes in Ecuador, and Loof and Zullini (2000) described the first species from Costa Rica.

In this study, a new species is described and we provided additional information on

known species of *Pachydorylaimus* from preserved material in the Nematode Collection of Universidad Nacional of Costa Rica.

MATERIALS AND METHODS

Samples of 200-500 g soil were collected in different protected areas of Costa Rica, soaked in tap-water for 2 hours and processed by Cobb's modified decanting and sieving method (s'Jacob and van Bezooijen, 1984). The extracted nematodes were fixed with hot 4% formaldehyde and transferred to pure glycerol by a modification of Seinhorst's method (Seinhorst, 1959). Permanent mounts of adult nematodes were constructed on Cobb slides using the paraffin wax ring method (de Maeseneer and d'Herde, 1963). Standard measurements and ratios were taken for each specimen (Tables 1-3) and each specimen was photographed with a Nikon DS digital camera on a Nikon Eclipse 80i microscope. All the material included in this study is deposited in the nematode reference collection at the Universidad Nacional, Heredia, Costa Rica. Drawings were made with a camera lucida, scanned and digitally redrawn with a Wacom® tablet and Adobe® Photoshop® software. SEM studies were made on specimens dehydrated in a graded ethanol series, critical point dried, coated with gold and observed with a Jeol JSM-5800 microscope.

RESULTS

Pachydorylaimus holovachovi sp. n. (Figs. 1, 2 and 4E-G)

Material examined: One female from La Selva Biological Station, two females from Braulio Carrillo National Park, one female from Corcovado National Park and one female from Forest Reserve Pacuare River.

Measurements: See Table 1.

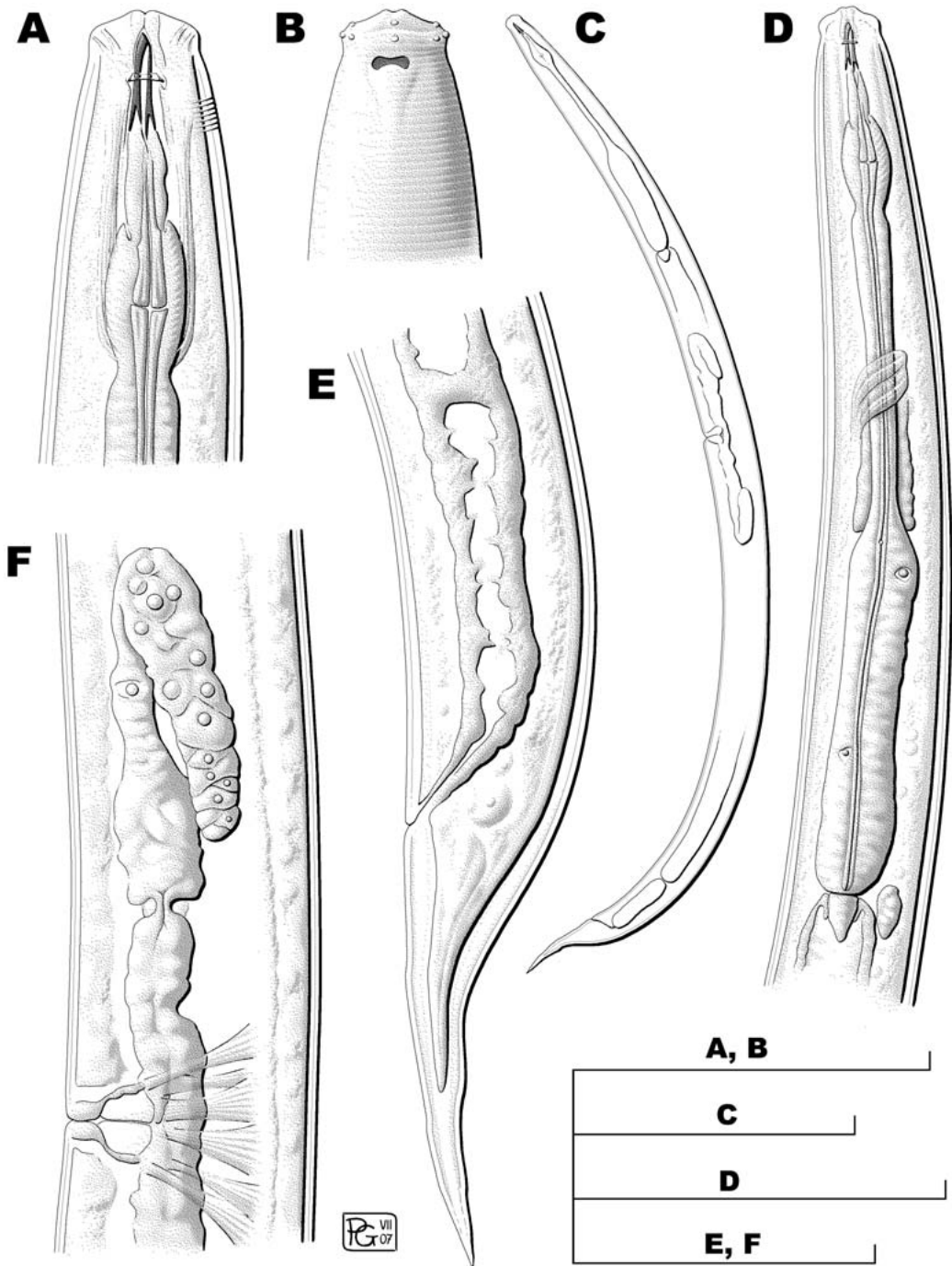


Fig. 1. *Pachydorylainus holovachovi* sp. n. (female). A) Anterior body region. B) Reconstruction of superficial appearance of cephalic body region. C) Entire body. D) Pharyngeal region. E) Posterior body region. F) Anterior genital branch. (Scale bars: A-B, E-F = 50 µm; C = 250 µm; D = 100 µm.)

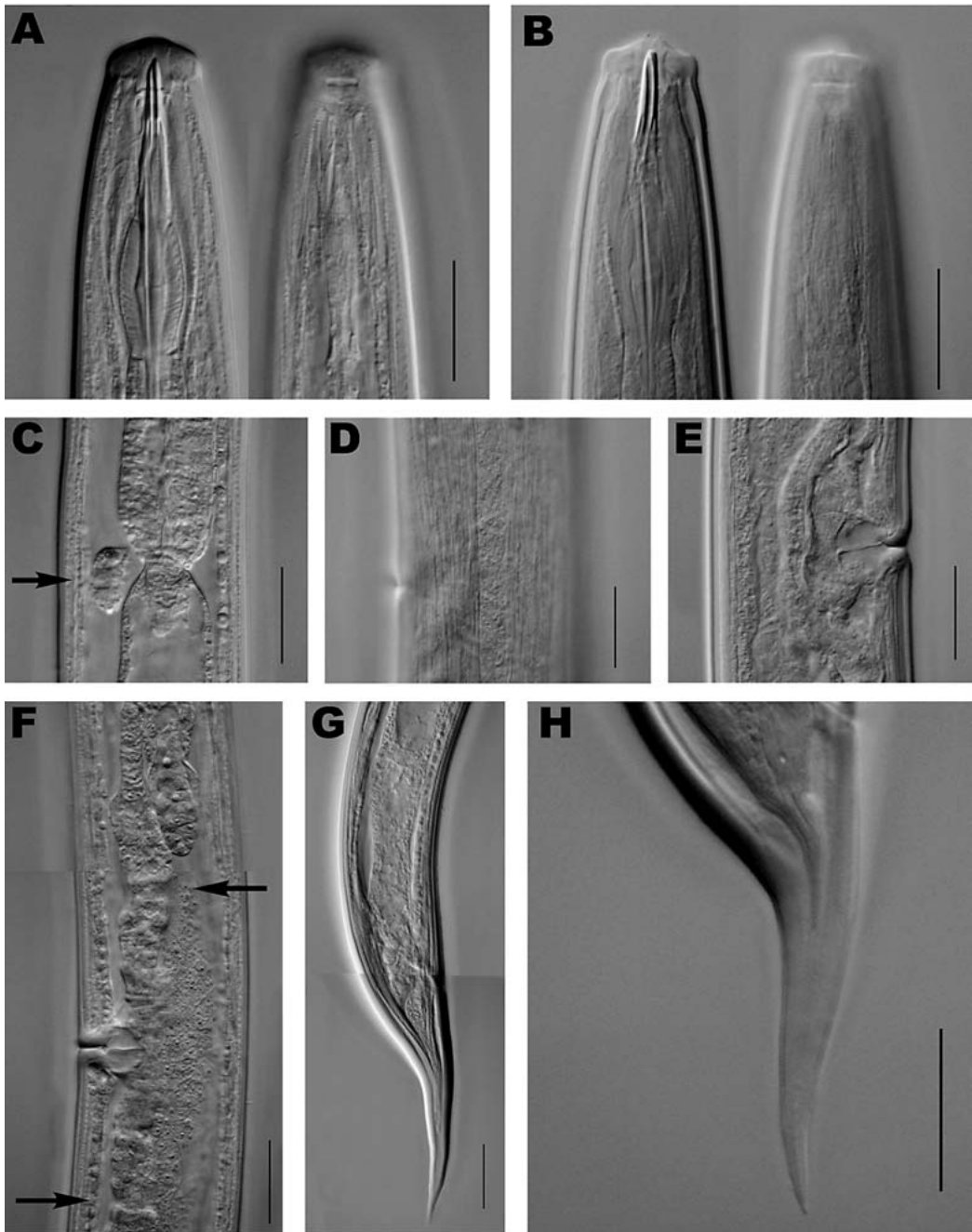


Fig. 2. *Pachydorylaimus holovachovi* sp. n. (female). A-B) Anterior body region. C) Pharyngo-intestinal junction area showing dorsal cell mass (arrow). D) Lateral chord. E) Vulval region. F) Genital system. Note location of sphincters between uterus and oviduct (arrows). G) Posterior body region. H) Tail. (Scale bars: 20 μ m.)

Table 1. Measurements and ratios of *Pachydorylaimus holovachovi* sp. n. All measurements are in μm , except L in mm.

Population character	La Selva	Braulio Carrillo	Osa	Pacuare	Total range
----- Females -----					
N	1	2	1	1	5
L	0.96	0.97, 1.00	0.90	0.98	0.90-1.00
a	23.4	19.9, 22.0	22.6	21.6	19.9-23.4
b	4.0	4.2-4.3	4.2	4.0	4.0-4.3
c	12.6	15.6, 15.6	15.8	12.5	12.5-15.8
c'	3.0	2.0, 2.2	2.5	2.7	2.0-3.0
V	44	45, 47	47	45	44-47
Lip region diam	16	16, 17	14	15	14-17
Odontostyle length	13	14, 15	13	14	13-15
Odontophore length	26	24, 24	23	30	23-30
Odontostyle + Odontophore	38	38, 39	36	44	36-44
Neck length	243	230, 234	217	242	217-243
Pharyngeal expansion length	99	99, 100	97	89	89-100
Diam. at neck base	39	39, 43	32	41	32-43
at midbody	41	44, 47	40	45	40-47
at anus	26	28, 32	23	28	23-32
Prerectum length	50	43, 43	?	33	33-50
Rectum length	19	18, 18	18	19	18-19
Tail length	77	62, 65	57	78	57-78

?Structure not clear enough to be measured in the specimen.

Female: Moderately slender nematodes, 0.90-1.00 mm long. Habitus slightly ventrad curved adopting a "comma" shape to almost straight upon fixation. Body cylindrical, distinctly tapering towards posterior end, and less so towards anterior end. Outer cuticle layer with faint transverse striations at the anterior end of the body; inner layer without visible transverse striations; cuticle thickness 1.0-2.0 μm in anterior region, 2.0-3.0 μm at midbody and 3.0 μm on tail. Lateral chord 6.5-14.0 μm wide or occupying 14-34% of the corresponding body diameter at midbody; its aspect granular, with no special differentiation. Lip practically continuous to slightly offset by depression, distinctly elevated at perioral

region; it is about 2.4-3.0 times as wide as high, and about one-third of body diameter at neck base. Lips amalgamated; labial and cephalic papillae hardly protruding. Amphid fovea cup-like; opening oval, at level of the cephalic depression and occupying 5-7 μm or 35-41% of the corresponding body diameter. Cheilostom cylindrical, without special differentiations, about 1.5 times as long as wide. Odontostyle strong and robust, with massive walls which are distinctly furcated at the base; it is about 4.0-5.6 times as long as wide and 0.8-0.9 times the lip region width; furcated portion about one-fourth of total length; its aperture small, about one-fifth of total length. Odontophore almost rod like, clearly thick-

walled at the posterior portion, 1.6-2.0 times the odontostyle length; its junction to pharyngeal lining surrounded by differentiated fusiform hyaline area with more or less distinct radial lines. Total stylet length 35-44 μm , measured from anterior tip of odontostyle to odontophore base. Protractor muscles well developed. Guiding ring weak, but apparently double. Pharynx consisting of a slender but muscular anterior portion that expands gradually; basal expansion occupying 37-44% of its length, 3.6-4.9 times as long as wide or 2.0-3.0 times longer than body diameter at neck base. Cardia tongue-shaped, longer than wide, 11-14 \times 8.0-9.5 μm . A dorsal cell mass is present at the pharyngo-intestinal junction of some specimens. Genital system didelphic-amphidelphic. Both branches equally and moderately developed: anterior branch 96-138 μm and posterior one 90-112 μm long. Ovary small, reflexed two-thirds halfway to vulva; oocytes few in number. Oviduct consisting of a slender portion and a poorly developed *pars dilatata*, 44 μm long. Sphincter very faint. Uterus a simple tube measuring 31 μm . Vagina extending inwards about 14.5 μm or 30-36% of the corresponding body diameter; *pars proximalis* 8.0-9.5 μm long, with practically straight walls and surrounded by scarcely developed musculature; *pars refringens* absent; *pars distalis* measuring 3.0-5.0 μm , including inside a very weakly sclerotized area. Prerectum 1.0-2.0 times the anal body diameter. Rectum shorter than anal body diameter. Tail conical elongate, ventrally almost straight, suddenly tapering at its anterior portion, then more gradually to an acute tip; hyaline portion 28-44 μm long, occupying about 40-60% of tail length. Caudal pores not visible. Male: Unknown.

Diagnosis: *Pachydorylaimus holovachovi* sp. n. can be distinguished by its body 0.90-1.00 mm long, lip region almost continuous to slightly offset by depression and 14-

17 μm wide, odontostyle 13-15 μm long, odontophore 23-30 μm long or 1.6-2.0 times the odontostyle with differentiated hyaline spindle-shaped area surrounding its junction to pharyngeal lining and lacking distinct basal flanges and sclerotization, total stylet length 35-44 μm , neck 230-243 μm long, pharyngeal expansion 89-100 μm or 37-44% of total neck length, female genital system amphidelphic, *pars refringens vaginae* absent, vulva a pre-equatorial (V = 43-47) transverse slit, female tail straight and tapering to an acute tip (57-78 μm , c = 12.5-15.8, c' = 2.0-3.0), and males unknown.

Relationships: This species differs from others belonging to the genus *Pachydorylaimus* in the morphology of odontophore base, in which a differentiated sclerotized flanged area is lacking. In having a large body (about 1 mm long), it resembles *P. aequatorialis* Andr ssy, 1997 and *P. schizodontus* Loof & Zullini 2000, but differs from them in having a narrower lip region (14-17 μm vs 19-21 μm), shorter odontophore (23-30 μm vs 28-33 μm), and shorter tail (57-78 μm vs 105-198 μm , c = 5.3-10.1, c' = 3.6-7.8).

Type locality and habitat: Cordillera Volc nica Central Conservation Area. Soil from tropical rain forest at the La Selva Biological Station, 76 meters above sea level SSO path, 400 mark.

Other localities and habitats: Cordillera Volc nica Central Conservation Area, soil from tropical cloudy forest at the Braulio Carrillo National Park, Las Palmas path, 2450 meters above sea level; Osa Conservation Area, soil from tropical rain forest at the Corcovado National Park, 350 meters above sea level and La Amistad Caribe Conservation Area, soil from forest plantation at the Forest Reserve Pacuare River, 400 meters above sea level.

Type material: Holotype female on slide 803 s-8 in the nematode collection at the Laboratorio de Nematolog a, Escuela de

Ciencias Agrarias, Universidad Nacional, Heredia, Costa Rica.

Etymology: This species is named in honour of Dr. Oleksandr Holovachov (Ivan Franco National University of L'viv, Ukraine) in recognition of his valuable work in the nematode diversity project.

Remarks: *Pachydorylaimus holovachovi* sp. n. differs from the remaining species of the genus in the nature of odontophore, which lacks the typical differentiation with flanges having refractive margins. This is a major difference from a taxonomic point of view, which might support the proposal of a separate generic taxon; however, we consider that only a fraction of the diversity of the group is currently known, and such a decision should be adopted after a general revision of the genus. For instance, *P. andreasi* Andr ssy, 1997 is characterized by having 'faint flanges' and resembles *P. holovachovi* sp. n. in this respect.

Pachydorylaimus notabenus Siddiqi, 1983
(Figs. 3 and 4A-D)

Material examined: Several specimens collected in Costa Rica's protected areas. Four females from La Selva Biological Station, two females from Forest Reserve Pacuare, two females from Pittier Biological Station, two females from Corcovado National Park, one female from Palo Verde National Park and one female from Chirrip  National Park.

Measurements: See Table 2.

Female: Moderately slender or slightly robust nematodes of small size, 0.52-0.77 mm long. Habitus regularly ventrad curved upon fixation, adopting an open "C" shape. Body cylindrical, distinctly tapering towards posterior end, and less so towards anterior end. Outer cuticle layer smooth; inner layer with transverse striations, more visible in anterior body region; cuticle thickness 1.0-1.5 μm in anterior region, 1.0-

2.0 μm at midbody and 1.5-3.0 μm on tail. Lateral chord 6.5-9.5 μm wide or occupying 20-31% of the corresponding body diameter at midbody; its aspect granular, with no special differentiation. Lip region truncate, distinctly expanded and offset by slight constriction; it is about 2.8-3.4 times as wide as high, and two-fifths to one-half of body diameter at neck base. Lips amalgamated; lateral lips with inner refractive masses in some specimens. Labial and cephalic papillae hardly protruding. Amphid fovea cup-like; opening oval, at level of the cephalic constriction and occupying 3.0-5.5 μm or 23-32% of the corresponding body diameter. SEM pictures show a lip region expanded and truncate; oral opening cross-shaped, biradial, and surrounded by a small circular smooth area; abundant radial or more irregular striations running from oral area to lip region margin; both inner and outer labial papillae near the lip region margin; and amphid aperture a short and oval transverse slit, narrower at its middle. Cheilostom cylindrical and thick-walled, almost as long as wide; its anterior end distinctly widened. Odontostyle peculiarly short and robust, with massive walls which are distinctly furcated at the base; it is about 3-4 times as long as wide and 0.6-0.8 times the lip region width; furcated portion up to one-third of total length; its aperture small, one-fifth to one-fourth of total length. Odontophore linear, but with small knobs at its base, 2.0-2.6 times the odontostyle; its junction to pharyngeal lining surrounded by a differentiated area whose margins are sclerotized and resembles the flange-like structures of other dorylaimid genera. Total stylet length, from anterior tip of odontostyle to posterior end of flange-like structures, 28-35 μm . Protractor muscles very well developed. Guiding ring weak, but apparently double. Pharynx consisting of a slender (but muscular) anterior portion that expands gradually; basal

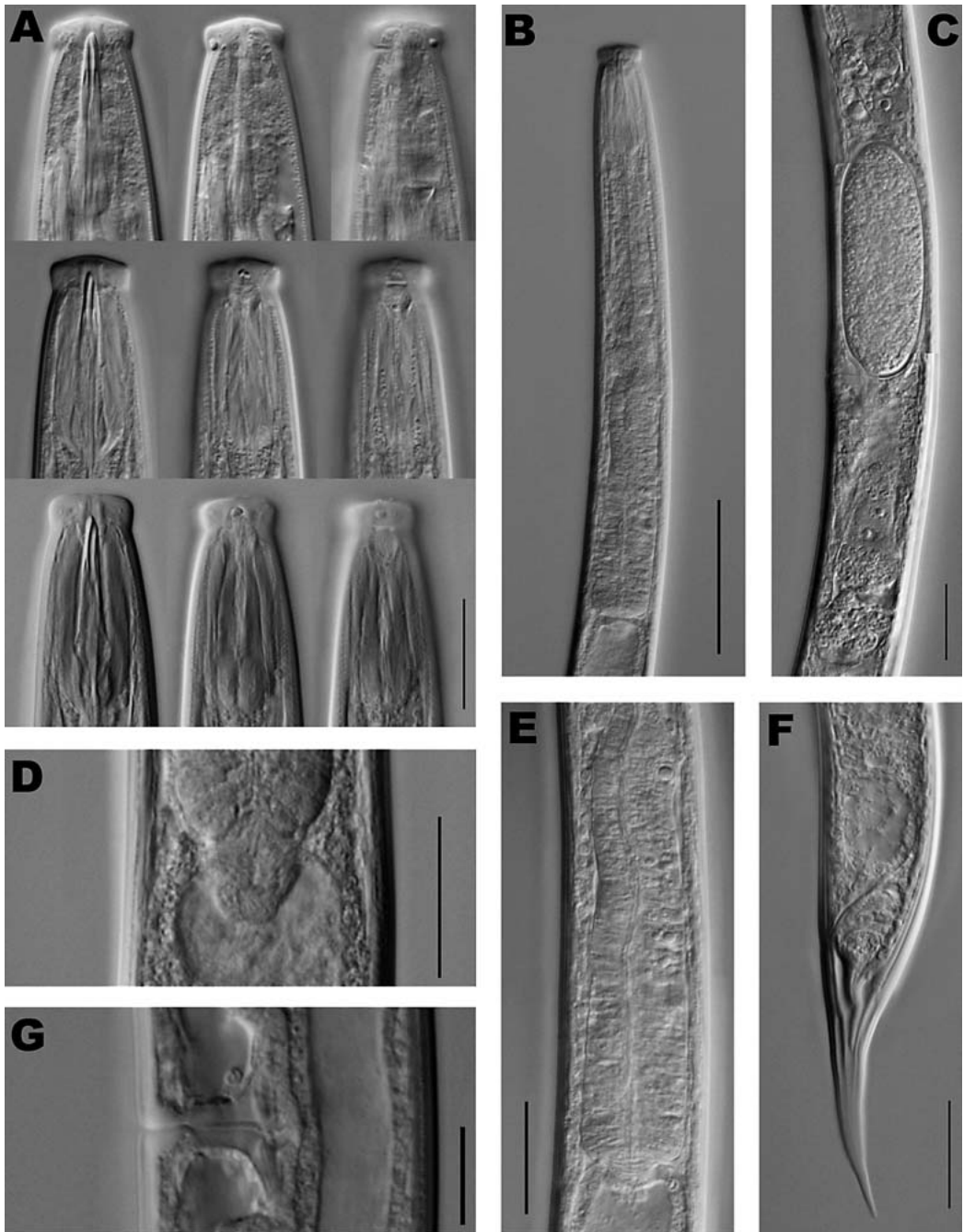


Fig. 3. *Pachydorylaimus notabenus* Siddiqi, 1983 (female). A) Anterior body region. B) Pharyngeal region. C) Genital system showing egg. D) Pharyngo-intestinal junction area. E) Expanded part of pharynx. F) Posterior body region. (Scale bars: A, C-F = 20 µm; B = 50 µm.)

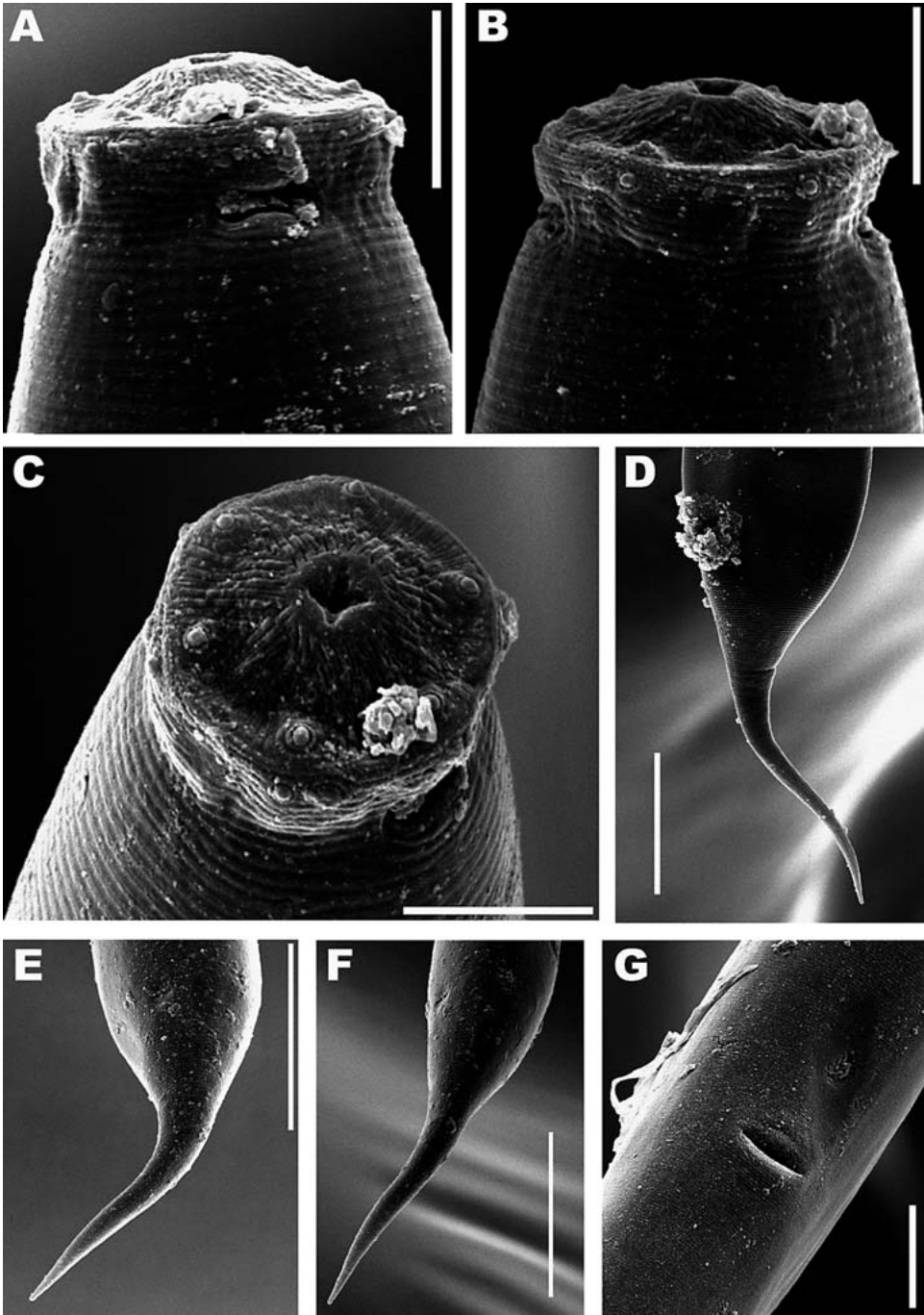


Fig. 4. A-D: SEM microphotographs of *Pachydorylaimus notabenus* Siddiqi, 1983. A-B) Cephalic region, lateral view. C) Cephalic region, *en face* view. D) Posterior body region. E-G: SEM microphotographs of *Pachydorylaimus holovachovi* sp. n. E-F) Posterior body region. G) Vulval region. (Scale bars: A-C = 8 μ m; D, G = 20 μ m; E-F = 30 μ m.)

Table 2. Measurements and ratios *Pachydorylaimus notabenus* Siddiqi, 1983 found in Costa Rica's protected areas. All measurements are in μm , except L in mm.

Population	La Selva	Pacuare	Pittier	Corcovado	Palo Verde	Chirripó	Total range
Character	----- Females -----						
N	4	3	2	2	1	1	13
L	0.52-0.63	0.65-0.67	0.66, 0.75	0.69, 0.72	0.62	0.77	0.52-0.77
a	19.0-21.0	18.6-20.4	19.0, 21.3	21.0, 23.0	19.3	20.7	18.6-23.0
b	2.7-3.6	3.4-3.6	3.1, 4.4	3.6, 4.4	3.2	3.6	2.7-4.4
c	7.8-11.0	8.9-12.3	10.7, 11.2	11.3, 11.8	9.3	11.6	7.8-12.3
c'	3.0-3.4	3.0-4.0	3.2, 3.2	2.9, 3.4	3.0	2.8	2.8-4.0
V	53-60	51-55	51, 52	51, 53	52	53	51-60
Lip region diam	13-16	13-15	15, 16	13, 13	14	17	13-17
Odontostyle length	8.5-9.5	9.5-10.0	9.5, 10.0	8.0, 9.0	10.0	9.5	8.0-10
Odontophore length	19-22	19-23	23, 24	19, 20	20	25	19-25
Odontostyle + Odontophore	30-33	29-33	34, 35	29, 29	30	35	29-35
Neck length	146-202	181-188	172, 211	158, 199	195	212	146-212
Pharyngeal expansion length	64-95	77-85	85, 88	74, 89	?	93	64-95
Diam. at neck base	28-32	27-35	29, 35	25, 33	36	35	27-36
at midbody	27-32	31-36	31, 40	30, 34	32	37	27-40
at anus	17-20	17-22	18, 22	18, 21	22	23	17-23
Prerectum length	29-35	33	25, 30	39	?	48	25-48
Rectum length	14-16	15	16, 17	13, 14	14	17	13-17
Tail length	49-69	51-73	59, 70	61, 61	66	66	49-73

?Structure not clear enough to be measured in the specimen.

expansion occupying 40-51% of its length, 4.5-5.3 times as long as wide or 2.4-3.0 times longer than body diameter at neck base. Cardia conoid, wider than long, 6.0-12.0 \times 4.5-10.5 μm . Genital system didelphic-amphidelphic. Genital branches short, often asymmetric in length. Ovary small, reflexed up to halfway to vulva; oocytes few in number. Oviduct and uterus poorly differentiated, distinct sphincter apparently lacking. Uterine eggs 19-27 \times 49-64 μm . Vagina extending inwards 9-13 μm or 29-40% of the corresponding body diameter; *pars proximalis* 5-8 μm long, with practically straight walls and surrounded by scarcely

developed musculature; *pars refringens* absent; *pars distalis* measuring 2.5-8.0 μm . Prerectum 1.4-2.0 times the anal body diameter. Rectum shorter than anal body diameter. Tail conical elongated, tapering to an acute tip; ventrally straight, but somewhat dorsad bent at its posterior region; hyaline portion 26-55 μm long, occupying one- to two-thirds of tail length. Caudal pores not visible. Male: Unknown.

Diagnosis: Costa Rican specimens of *P. notabenus* can be distinguished by the body length 0.52-0.77 mm long, lip region expanded and offset by constriction and 13-17 μm wide, odontostyle 8.0-10.0 μm

long and 0.6-0.8 times as long as lip region diameter with furcated base occupying up to one-third of total length, odontophore 19-25 μm long or 2.0-2.6 times the odontostyle with differentiated flanged-like area surrounding its junction to pharyngeal lining, total stylet length 28-35 μm , neck 146-212 μm long, pharyngeal expansion 64-95 μm or 40-51% of total neck length, female genital system amphidelphic, *pars refringens vaginae* absent, vulva a post-equatorial ($V = 51-60$) transverse slit, female tail conical elongated and dorsally bent in its posterior region (49-73 μm , $c = 7.8-12.9$, $c' = 2.8-3.4$), and males unknown.

Distribution and habitat: *P. notabenus* is widely distributed in Costa Rica. The nematode was found in soil samples from forest plantations, secondary and primary forest in dry and wet areas as well as in the Pacific and Caribbean regions from 0-1900 meters above sea level.

Remarks: Material examined fits well Siddiqi's original description, with some notable differences. In general, the range of measurements and ratios is extended, for instance body length (*vs* $L = 0.49-0.56$), lip region width (*vs* 11-14 μm), and odontostyle (*vs* 7.5-9.5 μm), odontophore (*vs* 17-21 μm), and tail (*vs* 42-54 μm) lengths. Other differences concern odontostyle nature, more robust and with longer furcated base in Costa Rican specimens; and more differentiated area surrounding the odontophore base; both odontostyle and odontophore resemble those of *P. furcatus* Siddiqi, 1983. Moreover, female tail usually appears distinctly dorsad bent at its posterior region. We regard these differences as intraspecific variability.

Pachydorylaimus schizodontus Loof & Zullini, 2000 (Fig. 5)

Material examined: Holotype collected in Tapantí National park, three females from

Pittier Biological Reserve and one female from Arenal-Monteverde protected zone.

Measurements: See Table 3.

Female: Moderately slender nematodes of small size, 0.97-1.06 mm long. Habitus adopting an open "C" or "comma" shape upon fixation. Body cylindrical, distinctly tapering towards posterior end, but less so towards the anterior end. Outer cuticle with very faint transverse striations, inner layer without visible striations; cuticle thickness 1.5 μm in anterior region, 1.0-1.5 μm at midbody and 3.0-4.0 μm at tail. Lateral chord 11-17 μm wide or occupying 30-32% of the corresponding body diameter at midbody; its aspect granular, with no special differentiation. Lip region offset by a more or less marked depression; it is about 3.0-3.9 times as wide as high, and about one-half of body diameter at neck base. Lips amalgamated; labial and cephalic papillae hardly protruding. Amphid fovea cup-like; opening a narrow slit, at level of the cephalic depression and occupying 6-8 μm or 29-36% of the corresponding body diameter. Cheilostom cylindrical, without special differentiations, 1.0-1.5 times long as wide. Odontostyle strong and robust, with massive walls which are distinctly furcated at the base; it is about 4.0-4.6 times as long as wide and about 0.6 times the lip region width; furcated portion about one-fourth of total length; its aperture small, about one-fifth of total length. Odontophore rod-like, 2.0-2.7 times the odontostyle; its junction to pharyngeal lining surrounded by a differentiated area whose margins are sclerotized and resembles the flange-like structures of other dorylaimid genera. Total stylet length 43-46 μm , measured from anterior tip of odontostyle to odontophore base. Protractor muscles developed. Guiding ring weak, double. Pharynx consisting of a slender anterior portion that expands gradually; basal expansion occupying 39-45% of its length, 4.1-4.8 times as long as wide or 2.5-

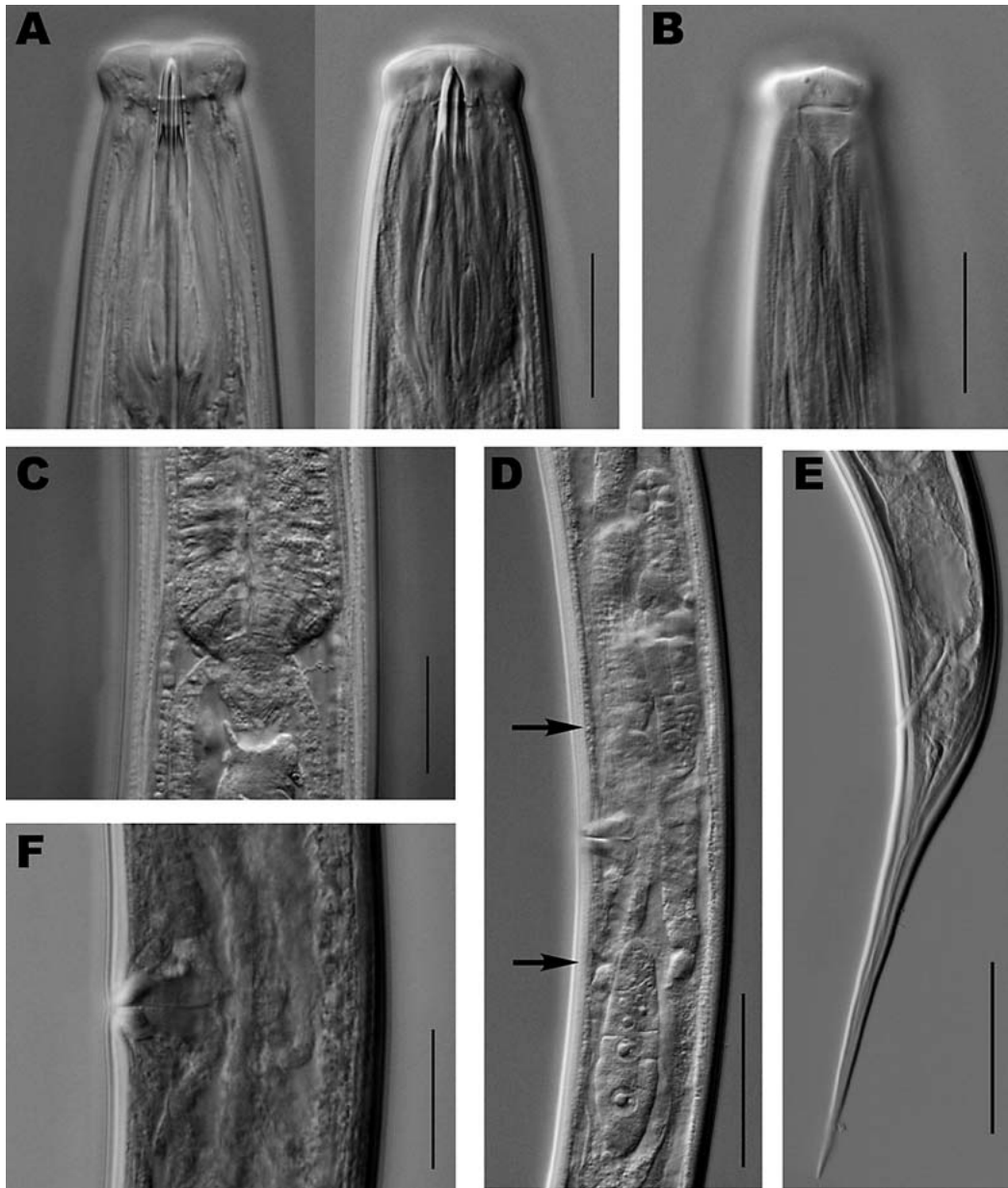


Fig. 5. *Paheydorylaimus schizodontus* Loof & Zullini, 2000. A) Variability of anterior body region. B) Surface of anterior body region. C) Pharyngo-intestinal junction area. D) Genital system. Note location of sphincters between uterus and oviduct (arrows). E) Posterior body region. F) Vulval region. (Scale bars: A-C, F = 20 μ m; D-E = 50 μ m.)

2.7 times longer than body diameter at neck base. Cardia tongue-shaped, longer than wide 12-15 \times 9-11 μ m. Genital system

didelphic-amphidelphic. Both branches equally and moderately developed: anterior branch 72-145 μ m and posterior one 84-148

Table 3. Measurements and ratios of *Pachydorylaimus schizodontus*. All measurements are in μm , except L in mm.

Population	Tapantí ^z	Pittier	Monte Verde	Total range
Character	----- Females -----			
N	1	3	1	5
L	1.02	0.97-1.06	1.03	0.97-1.06
a	19.0	21.8-26.5	22.5	19.0-26.5
b	4.0	4.0-4.2	4.1	4.0-4.2
c	8.8	9.2-10.1	7.5	7.5-10.1
c'	4.8	3.6-3.8	4.8	3.6-4.8
V	50	51-52	50	50-52
Lip region diam	21	21-22	21	21-22
Odontostyle length	14	12-14	13	12-14
Odontophore length	29	30-33	32	29-33
Odontostyle + Odontophore	43	44-46	45	43-46
Neck length	255	243-255	251	243-255
Pharyngeal expansion length	115	97-110	111	97-115
Diam. at neck base	46	36-40	43	36-46
at midbody	53	40-47	46	40-53
at anus	24	28-29	29	24-29
Prerectum length	37	39-66	44	37-66
Rectum length	22	22-25	24	22-25
Tail length	115	105	139	105-139

^zHolotype measurements included.

long. Ovary small, reflexed, one-half to two-thirds halfway to vulva; oocytes few in number. Genital tract barely differentiated: oviduct with relatively long slender part and a small *pars dilatata*, sphincter present but weakly marked, and uterus a simple tube shorter than body diameter. Vagina extending inwards 14-18 μm or 34-38% of the corresponding body diameter; *pars proximalis* 8.5-11.0 μm long; *pars refringens* with two trapezoidal close pieces. Prerectum 1.4-2.2 times the anal body diameter. Rectum shorter than anal body diameter. Tail conical elongate, ventrally straight, tapering more abruptly at its anterior portion, then more gradually to an acute terminus; hyaline portion 35-55 μm long, occupying

about 30-52% of tail length. Caudal pores not visible.

Male: Unknown.

Diagnosis: *Pachydorylaimus schizodontus* can be distinguished by its body 0.97-1.00 mm long, lip region offset by a depression and 21-22 μm wide, odontostyle 12-14 μm long, odontophore 29-33 μm long or 2.0-3.4 times the odontostyle and flanged, total stylet length 43-46 μm , neck 243-255 μm long, pharyngeal expansion 97-115 μm or 39-45% of total neck length, female genital system amphidelphic, *pars refringens vaginae* present, vulva a post-equatorial (V = 50-52) transverse slit, female tail conical elongate (105-139 μm , c = 7.5-10, c' = 3.6-4.8), and males unknown.

Distribution and habitat: In addition to type material, this species has been collected in La Amistad Pacifico Conservation Area, soil from secondary forest at the Pit-tier Biological Reserve, Canasta path, 1820 meters above sea level; and Arenal Conservation Area, soil from cloud forest at the Monteverde Forest Reserve, Chomogo path, 1400 meters above sea level.

Remarks: The new material studied here fits very well with the original description by Loof and Zullini (2000), but the range of measurements and ratios is extended and some new morphological details are provided.

On the Genus *Pachydorylaimus* Siddiqi, 1983

Diagnosis (amended): Plump and small nematodes, up to 1.2 mm long. Cuticle bearing radial elements. Lip region expanded, offset by more or less marked depression; papillae not distinctly protruding. Amphid opening a oval slit. Odontostyle short and robust, thick-walled, with small aperture and prominently furcated in its posterior part. Odontophore linear, with its posterior portion flanged-like, i.e., surrounded by a differentiated area, with sclerotized margins; in some cases lacking this differentiation. Protractor muscles well developed. Pharynx expanding gradually; basal expansion occupying two-fifths to one-half of total neck length; DN close to pharyngeal enlargement. Female genital system didelphic. Genital tract poorly differentiated. *Pars refringens vaginae* absent or present. Vulva transverse. Tail similar in sexes, conical elongate to almost filiform, with long hyaline terminal portion. Male with dorylaimid spicules and few spaced ventromedian supplements.

Relationships: This genus is characterized by a series of interesting apomorphies: expanded lip region, odontostyle nature, odontophore basal differentiation, and tail

with long hyaline portion. Its lip region resembles that of some discolaims by having very marginally located outer labial papillae, but without their distinctive offset structure. The odontostyle is very similar to that described in the genera *Metadorylaimus* Jairajpuri & Goodey, 1966 and *Neometadorylaimus* Jairajpuri & Ahmad, 1992, and the relationships with these genera should be a topic of further studies. The morphology of the odontophore might be an autapomorphy of *Pachydorylaimus* since similar but not comparable structures have been described in the genera *Xiphinema* Cobb, 1913 and *Enchodelus* Thorne, 1939.

Siddiqi (1983) classified his (new) genus under Qudsianematinae, and compared it with *Eudorylaimus* Andr ssy, 1959, from which it is easily distinguished. It also resembles *Epidorylaimus* Andr ssy, 1986 in having a conically elongated to almost filiform tail, similar in both sexes, but both odontostyle and odontophore are completely different, and also the general body pattern is not comparable. Jairajpuri (1965) described the genus *Qudsianema*, characterized by the presence of an odontophore with basal flanges, but the identity of this taxon has been a matter of controversy (Jairajpuri and Ahmad, 1992) and it is not comparable in its general morphology to *Pachydorylaimus*.

Accordingly, the taxonomic position of *Pachydorylaimus* should be analyzed in more detail, perhaps with the support of an independent molecular phylogenetic hypothesis.

List of species:

P. aequatorialis Andr ssy, 1997

P. andreasii Andr ssy, 1997

P. furcatus Siddiqi, 1983

P. holovachovi sp. n.

P. longicaudatus Siddiqi, 1983

P. notabenus Siddiqi, 1983

P. pachyvulvus Siddiqi, 1983

P. schizodontus Loof and Zullini, 2000

Table 4. Measurements and ratios of species belonging to the genus *Pachydorylaimus* (measurements in μm , except L in mm).

Species	n	L	a	b	C	V/T	c'	lrw	odont.	odon-top.	neck	ph. exp.	abw	prerec.	tail	spicul.	ve. sup.	Geog. dis.
<i>aequatorialis</i>																		
	4 ♀ ♀	0.90-1.06	20-24	3.9-4.3	5.3-6.3	43-51	6.5-7.8	19-20	15-16	28-30	224-263	123-137 52-53%	27 ^x		173-198	—	—	Ecuador (2)
	♂	1.13	34	5.3	6.3	—	7.5	?	?	?	213 ^y	?	24 ^x		180	46	5	
<i>andreasii</i>																		
	♀	0.76	19	4.1	9.2	50	4.2	15	13	26	187	82	20 ^y	?	83	—	—	Ecuador (2)
<i>furcatus</i>																		
	10 ♀ ♀	0.54-0.78	19-25	2.7-3.3	6-9	50-53	4.2-5.0	18 ^y	9-11	22-30	200-250	100-118 46-52%	19.5 ^y		90-94	—	—	Colombia (1)
	♂	0.74	23	3.2	8	41	4.2	?	11	25	231 ^y	?	22 ^x	?	92	?	2	
<i>holovachovi</i>																		
	5 ♀ ♀	0.90-1.00	20-23.5	4.0-4.3	12.5-15.8	43-47	2.0-3.0	14-17	12.5-15	23-30	217-243	89-100 41-43%	23-32	33-50	57-78	—	—	Costa Rica
<i>longicaudatus</i>																		
	6 ♀ ♀	0.54-0.66	23-26	3.0-3.6	5.2-6.1	46-50	6-8	11-12	7.5-10	16-17	175-190	44-50%	13.5 ^y		94-110	—	—	Colombia (1)
<i>notabenus</i>																		
	13 ♀ ♀	0.52-0.77	19-23	2.7-4.4	8-12	51-60	2.8-4.0	12.5-16.5	8.0-10.0	19-25	146-212	64-95 40-46%	17-23	?	49-73	—	—	Colombia (1) Costa Rica
<i>pachyvulvus</i>																		
	10 ♀ ♀	0.62-0.77	19-23	3.0-3.6	10-13	52-54	2.1-3.3	16 ^y	11-12.5	19-22	203 ^y	90-95 38-45%	22 ^x	?	56-61	—	—	Colombia (1)
<i>schizodontus</i>																		
	5 ♀ ♀	0.97-1.06	19, 26.5	4.0-4.2	7.5, 10.1	50, 52	3.6, 4.8	21-22	12, 14	29-33	243, 255	97-115 39-50%	24-29	37-66	105-139	—	—	Costa Rica (3)

(1) Siddiqi, 1983; (2) Andrassy, 1997, (3) Loof and Zullini, 2000.

^yMeasurements calculated from the drawings.^xData not given in the original description.

KEY TO SPECIES IDENTIFICATION

- 1a—Odontophore typically dorylaimid, its junction to pharyngeal lining surrounded by a spindle-shape area, lacking basal flange-like differentiation and without sclerotized or refractive margins *holovachovi*
- 1b—Odontophore with basal differentiation consisting of a thickened flange-like portion with sclerotized or refractive margins 2
- 2a—Longer tail, at least six times the anal body diameter 3
- 2b—Shorter tail, up to five times the anal body diameter 4
- 3a—Longer body, L = 0.9-1.1 mm long; wider lip region, 19-20 μm ; longer odontostyle, 15-16 μm ; shorter neck, b = 3.9-5.3; *pars refringens vaginae* well developed; female tail longer, 173-198 μm *aequatorialis*
- 3b—Shorter body, up to 0.8 mm long; narrower lip region, 11-12 μm ; shorter odontostyle, 7.5-10.0; longer neck, b < 3.7; *pars refringens vaginae* absent; female tail shorter, 94-110 μm *longicaudatus*
- 4a—Odontostyle longer, 11-15 μm 5
- 4b—Odontostyle shorter, up to 11 μm 7
- 5a—Larger general size: body about 1 mm long, neck more than 250 μm long, tail more than 100 μm long *schizodontus*
- 5b—Smaller general size: body shorter than 1 mm long, neck less than 250 μm long, tail less than 100 μm long 6
- 6a—Perioral liplets present; shorter odontophore, 19-22 μm ; *pars refringens vaginae* present; shorter female tail (56-66 μm , c' = 2.1-3.3) *pachyvultus*
- 6b—Perioral liplets absent; longer odontophore, 26 μm ; *pars refringens vaginae* absent; longer female tail (83 μm , c' = 4.2) *andreasii*
- 7a—Lip region offset by depression and wider, 18 μm ; longer pharyngeal expansion, 100-118 μm ; longer tail (90-94 μm , c = 6-9, c' = 4.2-5.0) *furcatus*
- 7b—Lip region offset by constriction and narrower, 12.5-16.5 μm ; shorter pharyngeal expansion, 64-95 μm ; shorter tail (49-73 μm , c = 8-12, c' = 2.8-4.0) *notabenus*

A compendium of these species, including their main measurements and ratios, is provided in Table 4.

ACKNOWLEDGMENTS

The corresponding author wish to express the gratefulness to CR-USA foundation, National Biodiversity Institute of Costa Rica, Nematology Department of Jaén University, Spain and Agronomy School at Universidad Nacional of Costa Rica for providing financial, facilities and logistic support to conclude this research. Coauthor T. Powers acknowledges support from NSF (DEB-0640807).

LITERATURE CITED

- Ahmad, W., and A. Shaheen. 2004. Five new and two known species of the family Dorylaimidae (Nematoda: Dorylaimida) from Costa Rica. *Nematology* 6:567-584.
- Andrássy, I. 1967. Nematoden aus Chile, Argentinien und Brasilien, gesammelt von Prof. Dr. H. Franz. *Opuscula Zoology Budapest* 7:3-33.
- Andrássy, I. 1997. Nematodes from Ecuador. A new genus, four new and known species (Dorylaimida). *Opuscula Zoology Budapest* 29-30:12-16.
- Bongers, T., A. Esquivel, and H. Arias. 2003. Preliminary results of the Costa Rican nematode inven-

- tory. *Journal of Nematode Morphology and Systematics* 6:91-94.
- de Maeseneer, J. and J. d'Herde 1963. Méthodes utilisées pour l'étude des anguillules libres du sol. *Revue de l'Agriculture Bruxelles* 16:441-447.
- Esquivel, A. 2003. Nematode Fauna of Costa Rican Protected Areas. *Nematopica* 33:131-145.
- Jairajpuri, M. S, and W. Ahmad. 1992. *Dorylaimida: Free-living, Predaceous and Plant-Parasitic Nematodes*. Brill, Leiden. 458 pp.
- INBio. 2007. Biodiversity in Costa Rica. National Biodiversity Institute. Santo Domingo de Heredia, Costa Rica. Online. http://www.inbio.ac.cr/es/biod/bio_biodiver.htm.
- Janzen, D. H. 1983. *Costa Rican Natural History*. D. H. Janzen, ed. London. The University of Chicago. 816 pp.
- Loof, P. A. A. 1964. Free-living and plant parasitic nematodes from Venezuela. *Nematologica* 10:201-300.
- Loof, P. A. A., and A. Zullini. 2000. Freelifing nematodes from natural reserves in Costa Rica. 1: *Dorylaimina*. *Nematology* 2:605-633.
- Obando, V. 2002. Biodiversidad en Costa Rica. Estado del conocimiento y gestión. Santo Domingo de Heredia, Costa Rica. Instituto Nacional de Biodiversidad. 250 pp.
- Seinhorst, J. W. 1959. A rapid method for the transfer of nematodes from fixative to anhydrous glycerine. *Nematologica* 4:67-69.
- Shaheen, A. and W. Ahmad. 2004. Three new and known species of *Dorylaimida* (Nematoda) from Costa Rica. *International Journal of Nematology* 14:177-185.
- s'Jacob, J. J. and J. V. Bezooijen. 1984. A manual for practical work in nematology. Agricultural University Wageningen, Department of Nematology, Wageningen, The Netherlands. 77 pp.
- Siddiqi, R. M. 1982. Seven new genera of dorylaimid nematodes from Colombian rain forest. *Systematic Parasitology* 4:69-87.
- Siddiqi, R. M. 1983. Four new species of *Pachydorylainus* gen. n and *Tylenchodorus tylosus* gen. n., sp. n (Dorylaimida: Qudsianematidae) from Colombia rain forest. *Revue de Nematologie* 6:207-215.
- UNEP-WCMC. 2007. World Atlas of Biodiversity. United Nations Environment Programme. World Conservation Monitoring Centre. Online. <http://www.unep-wcmc.org/>
- Zullini, A. 1973. Some soil and freshwater nematodes from Chiapas (Mexico). *Accademia Nazionale Dei Lincei. Quaderno* 171:55-96.

Received:

3/X/2007

Accepted for publication:

7/XI/2007

Recibido:

Aceptado para publicación:

