

A new species of the genus *Laimaphelenchus* Fuchs, 1937 (Nemata : Aphelenchina)

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Summary — *Laimaphelenchus cocuccii* n. sp., associated with dead branches of *Pinus elliottii* Engelm. from Córdoba, Argentina, is described. It is characterized by the strongly developed anterior vulva lip, overlapping the vulvar slit, and a posterior uterine sac of medium length. Close to *L. deconincki* Elmiligy & Geraert, 1971 in general aspect, it differs by the greater length of the post-uterine sac and the shape of the vaginal musculature. Close to *L. penardi* (Steiner, 1914) Filipjev & Schuurmans Stekhoven, 1941 in body length and vulva position, it differs by the length of the post-uterine sac, length and position of the tail stub and the degree of development of the muscles surrounding the vagina. The anterior region of the new species, as seen with SEM, shows a smooth labial area without labial disc. The combination of these two characters is unique among the Aphelenchoidea known up to now.

Résumé — Une nouvelle espèce du genre *Laimaphelenchus* Fuchs, 1937 (Nemata : Aphelenchina) — *Laimaphelenchus cocuccii* n. sp., associé à des branches mortes de *Pinus elliottii* Engelm. prélevées à Córdoba, Argentine, est décrit. Il se caractérise par la présence d'une lèvre vulvaire antérieure très développée, recouvrant la fente vulvaire, et par un sac postvulvaire de longueur moyenne. Proche de *L. deconincki* Elmiligy & Geraert, 1971 par son aspect général, il s'en différencie par la longueur du sac postvulvaire et par la forme des muscles entourant le vagin. Proche de *L. penardi* (Steiner, 1914) Filipjev & Schuurmans Stekhoven, 1941 par la longueur du corps et la position de la vulve, il s'en différencie par la longueur du sac postvulvaire, la longueur et la position du pédoncule caudal, et par le développement des muscles entourant le vagin.

Key-words : *Laimaphelenchus*.

A nematological analysis of dead branches of *Pinus* spp. from the province of Córdoba, Argentina, revealed the presence of *Aphelenchoides* spp. and of an undescribed species of the genus *Laimaphelenchus* Fuchs, 1937. The latter is described and illustrated hereunder.

Material and methods

The population studied was found in dead branches of *Pinus elliottii* Engelm. The branches were cut, transversally and longitudinally, into pieces of 3 cm in diameter or in length, and 1 cm in thickness. The pieces were placed on a screen (1 mm openings) suspended over a dish containing water slightly covering the wood pieces. During 7 days the nematodes (adults and juveniles) that reached the water were recovered. The specimens were killed and fixed in hot fixative (Netscher & Seinhorst, 1969) and processed to glycerin. Measurements were made on nematodes mounted in glycerin, drawings were made with a camera lucida. Nematodes prepared for scanning electron microscopy (Jeol SM-U3) were fixed as mentioned above, dehydrated in

an ethanol series (from 50 % to absolute) and critical-point dried in CO₂. They were coated with 300 Å gold and examined at 15 kV. Juvenile stages (J 2, J 3 and J 4) were determined using a technique for conglomerate analysis (Späth, 1980) taking into account the measurements; the reproductive system of each stage was observed.

*Laimaphelenchus cocuccii** n. sp.

MEASUREMENTS

See Table 1

Holotype female : L = 0.66 mm; body diameter just anterior to vulva = 17 µm; a = 38.8; oesophagus length = 169 µm; b' = 3.9; distance between anterior end and oesophago-intestinal junction = 71 µm; b = 9.3; stylet = 11 µm; MB = 36; distance from anterior

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* This species is named in honor of Professor Dr. Alfredo E. Cocucci (Cátedra de Morfología Vegetal, Facultad de Ciencias Exactas, Físicas y Naturales, Universidad Nacional de Córdoba) in acknowledgment of his outstanding work as a teacher, and of his contribution to the development of biological sciences in Argentina.

Table 1. Morphometrics of *Laimaphelenchus cocuccii* n. sp. (in μm , except L.)

	Females	♂2	♂3	♂4
n	50	4	21	70
L (mm)	0.57-0.74 (0.66 ± 0.03)	0.22-0.27 (0.24 ± 0.02)	0.34-0.45 (0.39 ± 0.03)	0.44-0.59 (0.51 ± 0.03)
Body diam.	16.0-19.5 (18 ± 0.76)	9.0-11.0 (10.5 ± 1.0)	12.0-15.0 (14.0 ± 0.74)	13.5-17.5 (15.0 ± 0.02)
a	34.0-40.0 (37 ± 1.58)	20.0-25.0 (22.7 ± 2.21)	24.0-33.0 (28.2 ± 2.27)	29.0-41.0 (33.4 ± 1.98)
Oesophagus length	140-194 (166 ± 11.38)	99-116 (107 ± 8.5)	122-150 (137 ± 7.94)	133-177 (155 ± 10.27)
b'	3.6-4.4 (4.0 ± 0.19)	2.1-2.4 (2.27 ± 0.14)	2.5-3.4 (2.85 ± 0.24)	2.9-3.8 (3.31 ± 0.22)
Oes. int. junct. (n = 40)	64-76 (70 ± 3.10)	46-50 (48 ± 2.08)	55-64 (58 ± 2.37)	61-71 (65.5 ± 2.40)
L oes. overlap	76-123 (96 ± 10.55)	41-54 (49 ± 7.0)	54-80 (65 ± 7.72)	57-93 (76 ± 7.91)
b (n = 40)	8.6-10.4 (9.5 ± 0.42)	4.5-5.4 (5.03 ± 0.47)	6.1-7.8 (6.76 ± 0.48)	7.0-8.9 (7.88 ± 0.38)
Tail length	28-41 (34 ± 3.01)	15-18 (16 ± 1.25)	19-27 (23 ± 2.28)	21-33 (28 ± 2.56)
c	17.1-22.9 (19.6 ± 1.46)	13.8-15.0 (14.6 ± 0.56)	14.8-20.0 (16.7 ± 1.36)	15.8-21.9 (18.2 ± 1.35)
Stylet	10-12 (11 ± 0.42)	8.0-8.5 (8.3 ± 0.28)	9-10 (9.5 ± 0.26)	9.0-10.5 (10.0 ± 0.30)
MB	31-41 (36 ± 2.01)	38-40 * (39.3 ± 1.15)	33-39 (36.5 ± 1.77)	32-39 (35.8 ± 1.76)
Excr. pore	86-105 (96 ± 4.6)	60-62 (61 ± 1.0)	72-85 (79 ± 3.9)	75-95 (87 ± 5.0)
Anal body diam.	9.5-12.0 (11 ± 0.47)	7.5-8.0 (7.75 ± 0.28)	9.0-10.5 (8.0 ± 0.50)	9.5-11.5 (10.5 ± 0.53)
c'	2.7-3.6 (3.2 ± 0.23)	2.0-2.3 (2.1 ± 0.14)	2.1-2.7 (2.4 ± 0.20)	2.1-3.3 (2.7 ± 0.24)
V	66-69 (67 ± 0.94)	—	—	—
Dist. ant. end-vulva	379-500 (444 ± 26.1)	—	—	—
Dist. vulva-anus	147-201 (179 ± 11.95)	—	—	—
L post-uterine sac	40-63 (54 ± 4.68)	—	—	—

end to excretory pore = 100 μm ; tail length = 35 μm ; $c = 18.8$; $c' = 3.1$; anal body diameter = 11 μm ; $V = 68$; distance from anterior end to vulva = 453 μm ; vulva-anus distance = 172 μm ; length of post-uterine sac = 47 μm .

DESCRIPTION

Female : Habitus open C- or J- shape when killed by gentle heat. Body thin, elongate, with anterior end rounded and slightly flattened; posterior end conoid. Cuticle with fine transverse annulation about 1 μm apart at mid-body. Lateral field marked by three inconspicuous lines, the inner one less apparent than the outer ones; about one-fifth of body diameter, not areolated. The external lines continue to close to the tail end; the internal one generally ends at level of the anus. Labial area hemispherical, smooth, anterior margin flattened; height 2.5-3.5 μm ($\bar{x} = 3$; $SD = 0.14$), width 6-7 μm ($\bar{x} = 6.5$; $SD = 0.28$), separated from the neck by a shallow constriction. SEM shows six labial sectors of equal width, amalgamated, separated by pairs of well-marked ribs; oral aperture surrounded by six small pores; labial disc absent. The amphidial apertures could not be seen. Cephalids at level of middle of retracted stylet, invisible in the majority of specimens. Cephalic framework hexaradiate, well developed. Stylet thin, anterior part conical, shaft tubular, elongate with three small basal swellings. Median bulb ovoid with crescentic valves in the middle, located 55-56 μm ($\bar{x} = 60$; $SD = 2.56$) from anterior end; its length 14.5-16.5 μm ($\bar{x} = 15.5$; $SD = 0.50$), width 11.0-12.5 μm ($\bar{x} = 12$; $SD = 0.33$). Hemizonid not seen. The nerve ring is located posterior to the oesophago-intestinal junction, at 76-89 μm ($\bar{x} = 83$; $SD = 3.27$) from the head end. Oesophageal glands dorsally overlapping the intestine for 76-123 μm ($\bar{x} = 96$; $SD = 10.5$). Intestine a straight tube; rectum straight or sigmoid. Reproductive system mono-prodelphic with post-uterine sac. Anterior branch outstretched; ovary with oocytes in one row; oviduct straight or tortuous; spermatheca empty, elongate; uterus elongate; vagina straight, surrounded by strongly developed, refringent muscular bundles. Post-uterine sac non-functional, undifferentiated. Anterior vulva lip well developed, overlying the vulvar slit, of equal thickness over almost all its length; its contour becomes narrower toward its posterior end and has a semicircular tongue-like shape. Tail curved, spirally coiled, subcylindrical, ending in a stub with four tubercles bearing ten finger-like projections; organic matter often adheres to these, making observation difficult.

Juveniles : External morphology similar to that of adult females. In the J2 the genital primordium is represented by a single oval cell 8 μm long. In the J3 a structure 25-32 μm long can be observed; it is composed of a well delimited cell, followed by a tubular mass, which is transversely, longitudinally and obliquely segmented into indistinctly delimited cells of variable size;

these will form the post-uterine sac. In the J4 the genital primordium has already differentiated into vagina, anterior genital branch and post-uterine sac.

TYPE LOCALITY AND HABITAT

Manfredi, Department of Rio Segundo, province of Córdoba, Argentina; inside dead branches of *Pinus elliottii* Engelm.

TYPE SPECIMENS

Holotype female catalogue number RAC 86, 30 female and 74 juvenile paratypes are in the collection of the Laboratorio de Nematología, Centro de Zoología Aplicada, Universidad Nacional de Córdoba. Five female and five juvenile paratypes are deposited in each of the following institutions : Muséum national d'Histoire naturelle, Laboratoire des Vers, Paris, France; Laboratorium voor Nematologie, Landbouwniversiteit, Wageningen, Netherlands; four female and five juvenile paratypes in the Instituut voor Dierkunde, Rijksuniversiteit Gent, Belgium; five female and six juvenile paratypes in the USDA Nematode Collection, Beltsville, Maryland, USA.

DIAGNOSIS

Laimaphelenchus cocuccii n. sp. belongs to the group of species characterized by the anterior vulva lip being strongly developed, overlapping the vulvar slit and the posterior vulva lip; lateral fields with three lines and post-uterine sac of medium length. It resembles *L. deconincki* Elmiligy & Geraert, 1971 in general aspect, but differs from it by the greater length of the post-uterine sac (40-63 *vs* 36-42 μm) and the shape of the muscles surrounding the vagina (round *vs* more angular). It also resembles *L. penardi* (Steiner, 1914) Filipjev & Schuurmans Stekhoven, 1941 but differs by the much shorter post-uterine sac (40-63 *vs* 72-173 μm), by the length and position of the tail stub (short, not abruptly separated from the remainder of the tail *vs* long, separated by a marked constriction) and the much stronger development of the vaginal musculature.

REMARKS

Observations on the vaginal region of the holotype of *L. deconincki* and of *L. cocuccii* n. sp. show that what has been called a "thick cuticularized tube" (Elmiligy & Geraert, 1971) or a "vaginal sclerotization" (Baujard, 1981) should probably be considered a strongly developed vaginal musculature.

The measurements of *L. cocuccii* n. sp. show low variation coefficients ($CV < 5$: body width anterior to vulva, stylet length, distance head end to oesophago-intestinal junction, distance from anterior end to excretory pore, anal body diameter, a, b, b', V; $5 < CV < 10$: L, length of oesophagus, distance from anterior end to vulva, vulva-anus distance, length of post-uterine sac, c, c', MB; $CV > 10$: length of oesophageal overlap). Thus

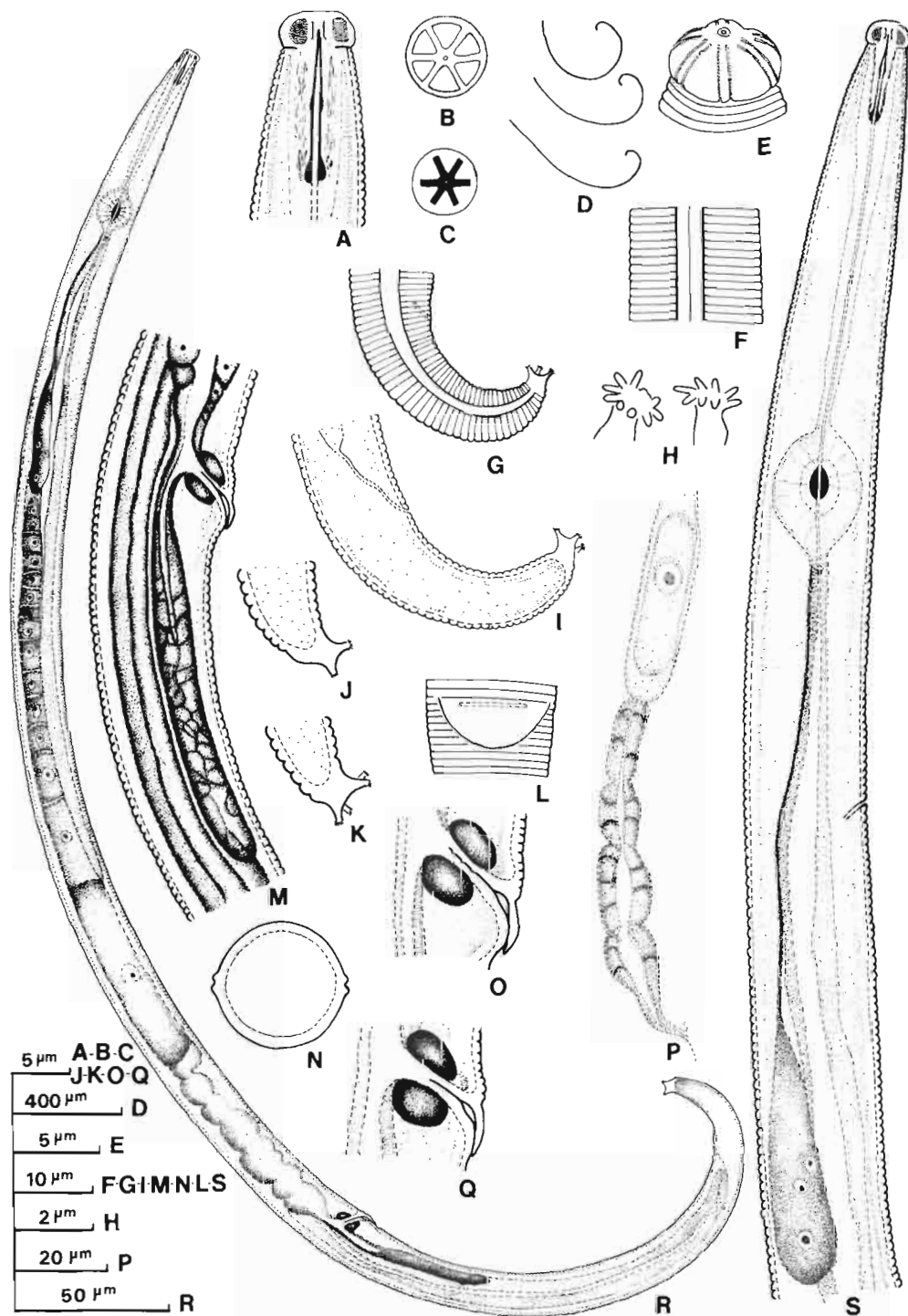


Fig. 1. *Laimaphelenchus cocuccii* n. sp., female. A : Anterior region; B : Lip region (en face view); C : Cross section through the basal plate; D : Habitus; E : Anterior region (lateral view, based upon SEM pictures); F : Cuticle (lateral field); G, I : Tails (lateral view); H : Tail tubercles; J-K : Tail tips (lateral view); L : Vulvar region (ventral view); M : Post-uterine sac; N : Cross section at middle of body; O, Q : Vulvar regions (lateral view); P : Terminal region of anterior genital branch; R : Whole specimen; S : Oesophageal region.

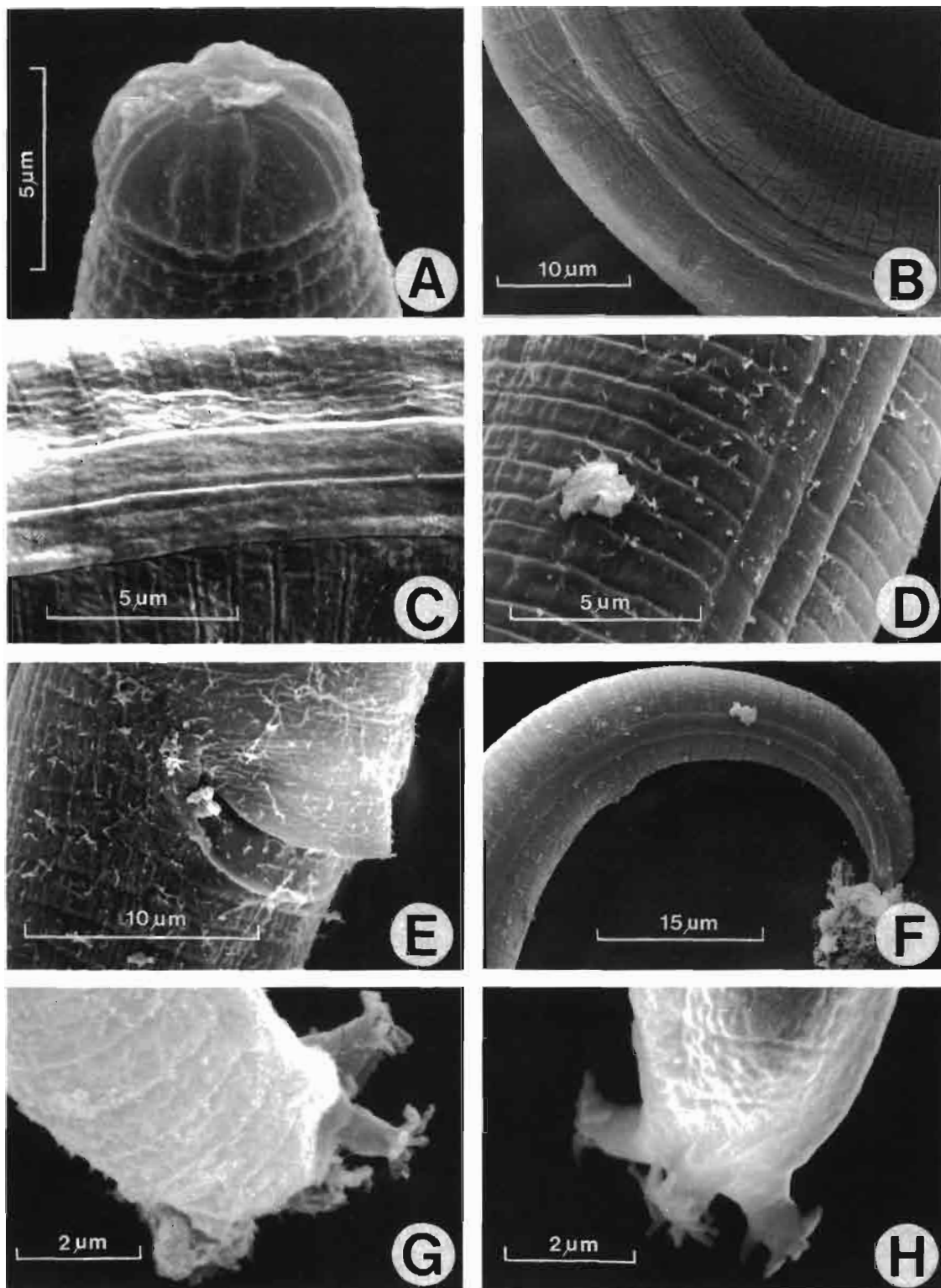


Fig. 2. *Laimaphelenchus cocuccii* n. sp., female, SEM photographs. A : Anterior region (lateral view); B, D : Cuticle with lateral field; E : Vulvar region (ventrolateral view); F : Tail (lateral view); G, H : Tubercles on tail tip.

it can be said that the morphometric characteristics of the species are well defined.

SEM observations have shown that *L. cocuccii* n. sp. is distinguished from all other species of the families Aphelenchoididae, Aphelenchidae and Paraphelenchidae by the combination of absence of a labial disc and of transverse striations in the labial area (Hooper & Clark, 1980; Raski & Valenzuela, 1987; Nickle *et al.*, 1981; Giblin-Davis *et al.*, 1989).

Baujard (1985) established a list of the valid species belonging to the genus *Laimaphelenchus*. In the same year Hirling published a paper describing *L. praepenardi* and synonymizing *L. deconincki* with *L. penardi*. The diagnostic characters of *L. praepenardi* do not justify establishing it as a separate species. Significant morphological differences were not given, the description is incomplete and the illustrations insufficient. The author points out that the most important difference between *L. penardi* and *L. praepenardi* is the mode of reproduction : parthenogenetic *vs* amphimictic. However, males of *L. penardi* have been described (Baujard, 1981). The major morphometric difference appears to be the length of the post-uterine sac; however, the author himself admitted that it was sometimes difficult to distinguish females of *L. penardi* from non-gravid females of *L. praepenardi*. Therefore we regard *L. praepenardi* a junior synonym of *L. penardi*.

The synonymization of *L. penardi* and *L. deconincki* is rejected, since there is one evident morphological character which separates these species, viz. the vaginal musculature : small, weakly developed *vs* large, strongly developed.

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