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Full Length Research Paper

Description of a new species of insect parasitic nematode, *Leidynema* (Schwenck, in Travassos 1929) (Thelastomatidae) from host *Periplaneta americana* of Meerut region - India

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Three different types of insect parasitic nematodes namely - Leidynema, Thelastoma and Hammerschmidtiella, were collected from hind gut of host Periplaneta americana. The new species - Leidynema meerutensis sp. nov., is characterized with its females having a typical corpus broader at posterior and an enlarged blind intestinal diverticulum, females with unequally disposed lateral alae with posteriorly directed terminal spine, much enlarged excretory sac/bulb and a short attenuated tail. The male Leidynema meerutensis sp. nov. have four pairs of caudal papillae and a truncated tail with a short spine projection.

Key words: New species - *Leidynema meerutensis*, Thelastomatidae, *Periplaneta americana*, unequal lateral alae, Blind Gut diverticulum, four pairs of caudal papillae in males, truncated tail, Meerut region -India.

INTRODUCTION

The animal parasitic nematodes belonging to Order - Oxyurida, constitute two super-families - Oxyuroidea (nematode parasites of vertebrates) and Thelastomatoidea (the nematode parasites of invertebrates essentially arthropods) of order oxyurida. The thelastomatides are essentially the parasites thriving in invertebrate hosts, mostly the arthropods. They are parasitic (or commensal) in the gut of most saprophytic insect and other arthropod hosts. They feed upon host's gut contents like its micro-flora and body fluid (Jex et al., 2005). The super-family Thelastomatidae has been organized into five different families (Adamson and Van Waerebeke, 1992) - Thelastomatidae, Protrelloididae, Hystrignathidae,

Travassosinematidae and Pseudonymidae. Family Thelastomatidae is the largest family and have more than 35 genera.

Genus, Leidynema was first described by Schwenck (in Travassos, 1929), with L. appendiculatum Leidy, 1850 as its type species. At present, eight different species of Leidynema have already been described from different regions of world; namely: L. appendiculatum Leidy, 1850 (Chitwood, 1932); L. delatorrei Chitwood, 1932; L. periplaneti Farooqui, 1967; L. portentosae Van Waerebeke, 1978; L. Schwenckei Farooqui, 1967; L. socialis Leidy, 1850 (Adamson et Van Waerebeke, 1992); L. orientalis Singh and Malti, 2004; and L. saltense Achinelly and Camino, 2008. L.

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stylopygi (Biswas and Chakravarty, 1963) has been synonymised to *L. appendiculatum* (Kloss, 1966; Farooqui, 1967). *Leidynema meerutensis* sp. nov. is a new species of *Leidynema* from household cockroaches (*Periplaneta americana*) from Meerut region of India (Plate 1-4).

MATERIALS AND METHODS

Different household insects including house hold cockroaches (P. americana) were collected from different regions of district Meerut. The collected host insects were anaesthetized and dissected and nematodes were recovered from their hindgut. The collected nematodes were heat killed and fixed in Triethanolamine formalin (TAF) fixative. Nematodes were then dehydrated by slow dehydration method in alcohol glycerin mixture. The fixed nematodes were put into alcoholglycerin mixture and kept in glass dessicator with anhydrous CaCl2 for 3-5 days to allow slow dehydration. The dehydrated nematodes were mounted on glass slides in anhydrous glycerin with glass wool and sealed. Outline structures were drawn with the help of camera Lucida and photographs were taken using Motic image 2000 DMB 1 microscope. Morphometric measurements were recorded using stage and ocular micrometers. Comparative studies of morphology and morphometric measurements of different specimens of collected of nematode parasites were done using De Mann's formulae and parameters (De Man, 1884). The measurements and parameters are expressed in mean, range and standard deviation from the mean.

For scanning electron microscopy - the heat killed nematodes were transferred in primary fixative - Karnovasky's fixative for overnight and then transferred in 2.5% gluteraldehyde. The fixed nematodes were washed with 0.1 M phosphate buffer (pH - 7.2) at 4°C and gradually dehydrated in different grades of acetone. The dehydrated specimens were dried by critical point drying method using liquid CO₂. Dried specimens were mounted on aluminum stubs and gold coated. Scanning photography was done using LEO435 VP scanning electron microscope and measurements were recorded with LEO-32 annotation programme Tables 1 and 2.

Generic diagnosis

Female

Mouth surrounded with eight large sub-median labiopapillae and a pair of small amphids. Cephalic extremity formed by two annules. Oesophageal corpus divided into anterior narrow and relatively broader posterior portions of nearly equal length, a short distinct isthmus and a valvular spherical end bulb. Intestine have a large blind intestinal diverticulum called cardium. Lateral alae are present and terminate into terminal spine. Excretory pore is present at the posterior to base of oesophagus. Vulva is present at or near mid-body. Vagina anteriorly directed and opens into a common uterus. Ovaries are two in number and directed opposite - didelphic and amphidelphic. Eggs are large, elongate, ellipsoidal, crescent shaped and triangular in cross-section. Tail attenuated to long filliform.

Male

Cephalic extremity is formed by a single expanded annulus. Lateral alae may be present or absent. Oesophagus is simple and without any posterior swellings. Intestine is simple and without diverticulum. Caudal extremity abruptly truncated, with or without short terminal spine (spine like process on its ventral side) or provided with several protrubences. Caudal papillae 3-5 pairs, consisting of one pair of large sub-ventral pre-anal, one to three pairs tiny sub-ventral and one pair sub-lateral

post-anal papillae. Spicule is present.

Description

Female

Body cylindrical, tapering at both ends, 2.5 - 3.15 mm in length and with maximum body width of 0.28 - 0.35 mm; head with 2nd annulus much wider and with close-set of 5-6 annuli and then annuli expand abruptly in both, its length and width. First annule with 8 pairs of labial papillae have surrounding mouth. Cuticle is closely annulated throughout the body length. Lateral alae are prominent and each alae terminate into a spine-like projection at the posterior. These lateral alae and its spinelike projections are equally disposed in all species of Leidynema described so far (namely: Leidynema appendiculatum; however, in the present species - L. meerutensis sp. nov., it is much unequally disposed). Oesophagus is 0.37- 0.45 mm long (1/7th) occupying anterior 14-15% of the body length and consists of a 0.29 - 0.31 mm long corpus, 0.020-0.025 mm long distinct isthmus and an end bulb of 0.11 x 0.10 mm dimension. Corpus is demarcated into two distinct regions, the anterior half is narrow and its posterior part is cylindrical and broader. Intestine prominently enlarged at the anterior end and is provided with a posteriorly directed much enlarged characteristic oblong, blind intestinal diverticulum. Nerve ring is at the anterior 1/20th of the body and is located at 0.135 - 0.150 mm from anterior end (NR% - 5.04%). Excretory pore at 0.55 - 0.65 mm from the anterior end occupying anterior 1/5th of the body length (Ex% - 20.74%). The excretory bulb/sac is much enlarged and of the size approximately similar to the oesophageal end-bulb. Ovaries are paired and two in number and are divergent to each other - didelphic, amphidelphic. Vulva is transverse in orientation and slightly anterior to middle of the body at 1.27 - 1.56 mm from anterior end (V% = 48%). Vagina is sclerotized, muscular and anteriorly directed opening into a common uterus. Eggs are elliptical in shape with dimension of 0.125 x 0.050 mm and are laid singly. Tail attenuated and relatively much shorter comprising 1/6th of body length. A pair of phasmid is visible at the anterior of the tail (visible in SEM photomicrograph).

Male

Small body with length 0.80 - 1.15 mm and width 0.06-0.09 mm, curved at the posterior end upon fixation. Cuticle is annulated throughout the body length. Lateral alae present and continue to the tail. Oesophagus 0.18 - 0.20 mm long occupying anterior $1/5^{th}$ of the body and having a corpus of uniform diameter with length 0.11 - 0.13 mm, a short isthmus 0.02 mm and an end-bulb with dimension 0.03 - 0.04 mm × 0.03 - 0.04 mm. Nerve ring situated at anterior $1/10^{th}$ at 0.08 - 0.10 mm from the anterior end (NR - 9.53%) and excretory pore occupy anterior $1/3^{rd}$ of body at 0.27 - 0.35 mm from anterior end (Ex% - 33.12%). Testis is single and reflexed at the tip. Spicule is prominent with its length of 0.033 mm. Caudal extremity in male is abruptly truncated with a short terminal spine like structure, tail 0.015 - 0.017 mm in length from the anus. Caudal papillae four (four pairs) pairs in number and are symmetrically disposed - one pair sub-ventral pre-anal, two pairs ventral post-anal and one pair small sub-dorsal post-anal papillae.

Body dimensions (Mean ± SD in mm)

Holotype female

Total Length = 2.950; Width = 0.310; a (L/W) = 9.516; Oesophagus = 0.430; b (L/E) = 6.860; Tail = 0.480; c (L/T) = 6.146; Nerve ring = 0.150; NR% = 5.08%; Excretory pore = 0.650; Ex% = 22.03%; Vulva = 1.400; V% = 47.46%; Egg = 0.125 mm x 0.040 mm.

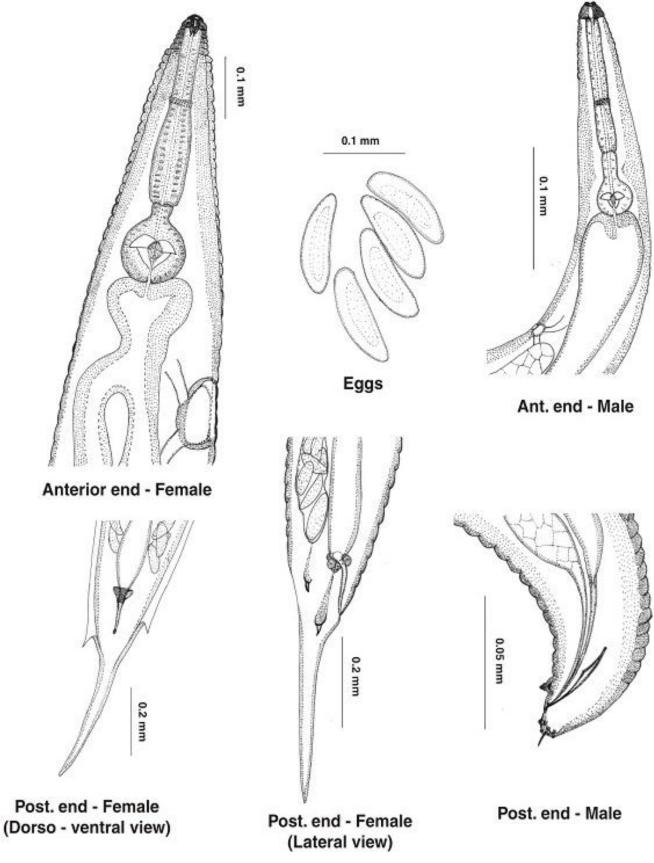
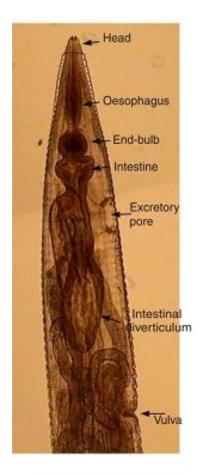
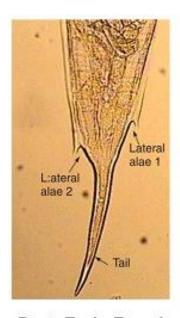


Plate 1. Morphological details of Leidynema meerutensis sp.



Ant. end - Female



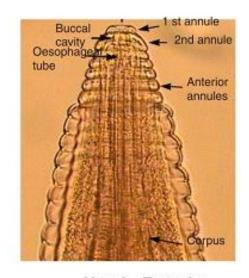
Post. End - Female



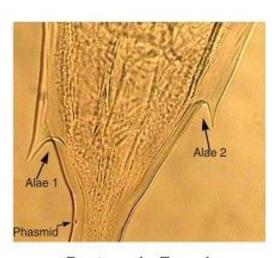
Eggs



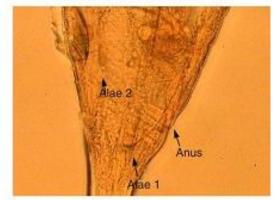
Female - W. M.



Head - Female

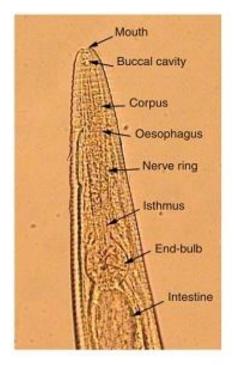


Post. end - Female (Dorso-ventral view)

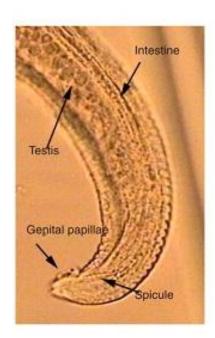


Post. end - Female (Lateral view)

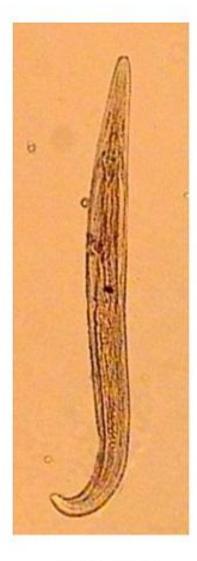
Plate 2. Photomicrographic details of *Leidynema meerutensis* n. sp. (female).



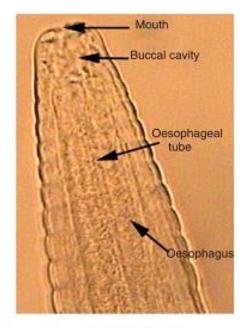
Ant. end - Male



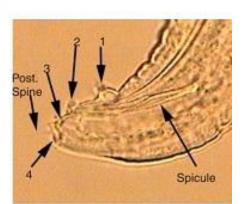
Post end with tail (Male)



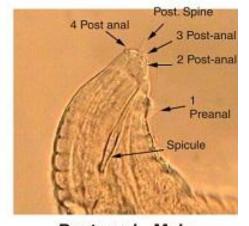
Male - W. M.



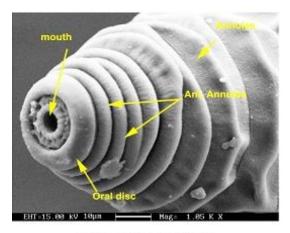
Head - Male



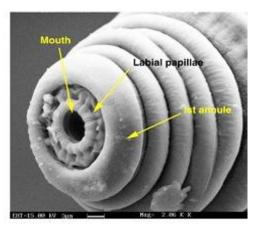
Post. end - Male (Lateral view)



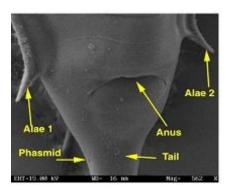
Post. end - Male



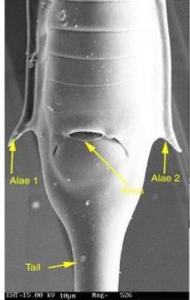
Ant. end - Female



Head - Female with oral papillae



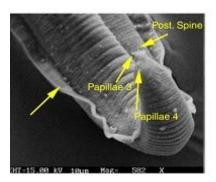
Post. end - Female (Unequal Alae Spine)



Post. end - Female (Equal Alae Spine)



Female W.M.



Post. end with tail (Male)

Plate 4. SEM photomicrographic details of Leidynema meerutensis n. sp.

Paratype Females (n = 9)

Total Length = 3.00 ± 0.25 ; Width = 0.31 ± 0.0206 ; a (L/W) = 9.46 ± 0.353 ; Oesophagus = 0.426 ± 0.0245 ; b (L/E) = 7.023 ± 0.305 ; Tail = 0.513 ± 0.042 ; c (L/T) = 5.847 ± 0.264 ; Nerve ring = 0.151 ± 0.008 ; NR% = 5.04%; Excretory pore = 0.622 ± 0.0363 ; Ex% = 20.74%; Vulva = 1.44 ± 0.093 ; V% = 48.00%; Egg = 0.125 mm × 0.035 mm.

Paratype Males (n = 10)

Total Length = 0.918 ± 0.133 ; Width = 0.074 ± 0.0107 ; a (L/W) = 12.48 ± 1.376 ; Oesophagus = 0.192 ± 0.0095 ; b (L/E) = 4.77 ± 0.579 ; Tail = 0.0155 ± 0.0008 ; c (L/T) = 59.016 ± 5.722 ; Nerve ring = 0.0875 ± 0.0092 ; NR% = 9.53%; Excretory pore = 0.304 ± 0.028 ; Ex% = 33.12%; Caudal papillae 4 pairs - 1 pair pre-anal, 2 pairs post-anal sub-

Table 1. Comparative morphometric measurements of different species of *Leidynema* (Female).

	Described species of Leidynema							Leidynema	ynema meerutensissp. nov.		
Parameter	L. Deltorrei (Chitwood, 1932)	<i>L.Periplaneti</i> (Farooqui, 1967)	<i>L.Portentosae</i> (Van Waerebeke, 1978)	<i>L.Schwenki</i> (Farooqui, 1967)	<i>L.socialis</i> (Adamson et Van Waerebeke,1992)	L.Appendiculata (Chitwood, 1932)	L.saltense (Achinelly and Camino, 2008)	<i>L.orientalis</i> (Singh and Malti, 2004)	Range	Mean	SD
Length (L)	3.4 - 3.78	1.99 - 2.6	1.85-2.44	1.50-2.80	2.11-4.65	2.168-3.213	3.990	2.60-2.75	2.50-3.15	3.000	0.260
width (W)	0.335-0.450	0.23-0.38	0.130-0.208	0.39-0.41	0.095	0.206-0.284	0.325	0.25-0.28	0.28-0.35	0.317	0.021
a=L/W	8.4 - 10.14	6.84-8.65	11.73-14.23	3.84-6.82	22.21-48.94	10.52-11.31	12.276	9.82-10.40	8.92-9.00	9.465	0.353
Esophagus (E)	0.506 - 0.570	0.37-0.43	0.46-0.598	0.31-0.38	-	0.368-0.446	0.432	0.38-0.40	0.37-0.45	0.427	0.024
b=L/E	6.63 - 6.71	5.37-6.05	4.02-4.08	4.83-7.36	-	5.88-7.20	9.236	6.84-6.87	6.75-7.00	7.024	0.306
Tail (T)	-	0.45-0.54	0.51-0.84	0.34-0.42	1/5 × L	0.491-0.756	-	0.58-0.65	0.43-0.56	0.513	0.042
c=L/T	-	4.42-4.64	2.90-3.62	4.41-6.66	5.000	4.25-4.41	-	4.23-4.48	5.62-5.81	5.847	0.265
Excretory pore	0.840- 0.920	0.59-0.62	0.650	0.46-0.64	-	0.510-0.628	0.460	0.55-0.58	0.55-0.65	0.622	0.036
Ex %	24.33 - 24.70%	23.84 - 29.64%	26.63%	22.8-30.6%	-	19.54-23.52%	11.52%	21.09-21.15%	20.63-22.00%	20.74%	1.03%
Vulva	1.4 -1.63	0.53-1.4	1.10-1.22	1.2-1.38	-	1.020-1.427	1.947	1.5-1.8	1.27-1.56	1.440	0.094
V%	41.17 - 43.12%	53.84%	50.00 - 59.45%	49.28-80.0%	-	44.41-47.04%	48.80%	57.69-65.45%	49.52-50.8%	48.00%	1.55%
Nerve ring	0.000	0.13-0.19	0.208	0.11-0.14	-	0.137-0.156	-	0.12-0.14	0.135-0.150	0.151	0.008
NR%	0.00%	6.53-7.30%	8.52%	5.0-7.33%	-	4.85-6.31%	-	4.61-5.09%	4.76-5.40%	5.04%	0.24%
Egg (lxb)	0.100 - 0.108 × 0.040 - 0.046	0.110 × 0.050	0.095-0.117 × 0.035-0.038	0.122x0.051	0.069-0.038	0.092-0.104 × 0.034-0.036	0.062 × 0.038	0.065-0.068 × 0.028-0.034	0.110-125 × 0.040-0.050	0.11 × 0.04	0.000

ventral, 1 pair post-anal sub-dorsal. Anal Spicule = 0.0348 ± 0.0006 mm.

Taxonomic summery

Type host

Periplaneta americana L. (Orthoptera, Blattidae)

Habitat/site of collection

Hind gut of the host.

Type locality

Meerut region, U. P. (India).

Type species

Leidynema appendiculatum (Leidy, 1850) Chitwood (1932).

Holotype

Leidynema meerutensis sp. nov. deposited at Dept. of Zoology, MCM, C.C.S. University Meerut (U.P. India).

Paratypes

Paratype females labeled as *L. meerutensis* 1-9 and paratype males labeled as *L. meerutensis* 10-19 are deposited at Department of Zoology, MCM, C.C.S. University Meerut (U.P. India).

Etymology

The present species name, *Leidynema meerutensis* is based on type locality of host collection.

Species diagnosis

The new species is characterized by the unequal distribution of lateral alae and terminal spine and short tail in females and 4 pair anal papillae in males. Also, the females have unusually large excretory sac/bulb opening in to excretory pore (not been reported so far). These three features make the species unique and distinct from all other eight species described so far.

RESULTS AND DISCUSSION

Genus *Leidynema* Schwenck, (in Travassos, 1929) with the type species *L. appendiculatum* Leidy 1850 (Chitwood 1932), is characterized by its gut diverticulum. *Leidynema meerutensis* sp. nov. shares the similarities in generic characters with

Table 2. Comparative morphometric measurements of different species Leidynema (Male).

	Different species of Leidynema								L.meerut	rutensis sp. nov.	
Parameter	L. Deltorrei (Chitwood, 1932)	<i>L.Periplaneti</i> (Farooqui, 1967)	<i>L.Portentosae</i> (Van Waerebeke, 1978)	<i>L.Schwenki</i> (Farooqui, 1967)	L.socialis (Adamson et Van Waerebeke,1992)	L.Appendiculata (Chitwood, 1932)	<i>L.orientalis</i> (Singh and Malti, 2004)	L.saltense (Achinelly and Camino, 2008)	Range	mean	S.D.
Length (L)	0.900	0.76	0.81-1.06	0.810	1.27-1.69	0.579-1.119	0.82-0.85	0.656	0.80-1.15	0.918	0.133
width (W)	0.060	0.090	0.06-0.082	0.090	0.064	0.058-0.097	0.09-0.12	0.083	0.06-0.09	0.074	0.011
a=L/W	15.000	8.440	12.92-13.50	9.000	19.84-26.40	9.98-11.53	4.25-9.11	7.900	12.77-13.33	12.482	1.376
Esophagus (E)	0.189	0.120	0.270-0.328	0.150	-	0.157-0.206	0.086-0.11	0.190	0.18-0.21	0.192	0.009
b=L/E	4.760	6.330	3.00-3.23	5.400	-	3.68-5.43	7.72-9.53	3.452	4.44-5.47	4.774	0.579
Corpus	0.145	0.060	0.185-0.229	0.102	-	0.102-0.123	0.05-0.06	-	0.11-0.13	0.121	0.007
Ishthmus	0.014	0.030	0.065-0.071	0.010	-	0.021-0.024	0.005-0.007	-	0.020-0.025	0.021	0.002
End bulb(lxb)	0.030 × 0.028	0.03 × 0.03	0.011016 × 0.36- 0.44	0.041 × 0.039	-	0.034-0.041-0.029- 0.038	0.025-0.027 × 0.024- 0.028	-	0.03-00.04 × 0.03-0.04	0.03 × 0.03	0.000
Buccal cavity	-	0.015	-	0.011	-	0.056-0.097	0.012	-	0.010	0.010	0.010
Tail (T)	-	0.020	-	0.020	1/8 of BL	0.009-0.012	0.009-0.012	0.060	0.015-0.017	0.016	0.001
c=L/T	-	38.000	-	40.500	-	64.33-93.25	70.83-91.11	10.933	53.33-67.64	59.016	5.722
Excretory pore	-	0.160	0.432	-	-	0.418	0.11-0.12	0.186	0.28-0.35	0.304	0.028
Ex%	-	21.05%	Aprox 40%	-	-	37.35%	13.41- 14.11%	28.35%	30.43- 35.00%	33.12 %	2.66 %
Spicules	0.037	0.030	0.042	0.030	0.063	0.0315-0.0328	0.049-0.059	0.054	0.033-0.035	0.035	0.001
Nerve ring	-	0.050	0.082	0.090	-	0.108-0.127	0.062-0.065	-	0.08-0.10	0.088	0.009
NR%	-	6.57%	7.73-10.12%	11.11%	-	11.34-18.65%	7.56-7.64%	-	8.69- 10.00%	9.53%	0.59 %
Caudal Papillae	3 pairs	5 pairs	3 pairs	5 pairs	5 pairs	3 pairs	5 pairs	4 pairs	4 pairs	4 pairs	0

other eight known species of *Leidynema - L. appendiculatum* Leidy, 1850; *L. delatorrei* Chitwood, 1932; *L. periplaneti* Farooqui, 1967; *L. portentosae* Van Waerebeke, 1978; *L. Schwencki* Farooqui, 1967; *L. socialis* Leidy, 1850 (Adamson et Van Waerebeke, 1992), and *L. saltense* Achinelly and Camino, 2008 in its shape of oesophagus and intestine, vulva near to mid-body in females, and a single spicule, and abruptly truncated caudal extremity with short terminal spine in males.

The present species, *Leidynema meerutensis* sp. nov. is similar to type species *L. appendiculatum* in general body form, but it is different from the type species *L. appendiculatum* as:

- (i) The female *Leidynema meerutensis* sp. nov. is relatively longer with more body width (a (L/W) = 9.46 ± 0.353) compared to *L. appendiculatum* (a = 11.146 ± 0.88).
- (ii) Tail in *Leidynema meerutensis* sp. nov. is attenuated and shorter (c (L/T) = 5.847 ± 0.264)

- relative to the filliform and elongated tail in *L.* appendiculatum (c (L/T) = 4.301 ± 0.235).
- (iii) Vulva is relatively anterior in position in L. appendiculatum (V% = 44%) compared to Leidynema meerutensis sp. nov. (V% = 48%).
- (iv) Lateral alae in females *L. appendiculatum* and all other species are equally disposed, but in *Leidynema meerutensis* sp. nov.; it is unequally disposed with one side lateral alae ending much shorter to the other.

(v) The male *Leidynema meerutensis* sp. nov. have four (4) pairs of caudal papillae compared to only three (3) pairs of caudal papillae present in *L. appendiculatum*.

The present species *Leidynema meerutensis* sp. nov. is different from all other known species of *Leidynema* in terms of its unequal disposition of lateral alae and number (4 pairs) and disposition of caudal papillae which is three (3) pairs in *L. deltorei, L. pontentosae* and *L.appendiculatum* while five (5) pairs in L. *Schwencki, L. socialis, L. orientalis* and *L. periplaneti* except *L. saltense* which also has four pairs of caudal papillae but differs in its disposition and tail shape. *Leidynema meerutensis* sp. nov. also differs from other species of *Leidynema*, as:

In *L. periplaneti*, the males lack lateral alae. Vulva in females is much posterior to mid-body (V > 60%).

In *L. portentosae*, there are only three pairs of thick papillae in males and it also possesses many ventral cuticular protuberances. The spicule is much longer and thinner. In females, vulva is much posterior (V = 55%).

In *L. Schwencki*, there are two (2) pairs of pre-anal papillae and three (3) pairs of post anal papillae.

L. delatorrei have 3 pairs of papillae and much anterior vulva (V = 41 - 43%) and lateral alae do not end into spine.

L. orientalis have five (5) pairs of caudal papillae and equal lateral alae.

In *L. saltense*, males have four (4) pairs of genital papillae but with different disposition and different tail shape. The females have much shorter oesophagus (b = 9.236) and excretory pore is much anterior (Ex% = 11.52%).

Thus, *L. meerutensis* sp. nov. is a new species of *Leidynema* and characterized as "females with distinct intestinal diverticulum, lateral alae unequally disposed and short tail; and males with four (4) pairs of caudal papillae and with truncated tail with spine".

Key to different species of Leidynema

1. Males	with three pairs	of papillae and n	nany ventra
cuticular	protuberances	in posterio	r region
	` L	. <i>portentosae</i> Van	Waerebeke
1978			
- N	lales without prot	uberances in post	erior region
		. 2	
2 (1) Ca	udal papillae 5	pairs in males. F	emales with
equally	di	sposed	latera
alae			3
- Caudal	papillae 4 pairs i	n males. Females	with equally
or	unequally	disposed	latera
oloo		-	C

elongat socialis	ed and Leidy, 18	> 0.060 50 (Adar e 5 pairs mm	mm in mson et \ in male	length. /an Waeı s. Spicul in	rebeke, le short	ule much <i>L.</i> 1992). er and < length 4
4(3)		alae			in	females.
-	Lateral a		ent in bo	oth male		nales
5 (4) with	In fema	les latera spine	al alae is I		рі	terminal rojection.
length projection 6(4) males was	and is on Females with 5 pa Females	oqui, 196 hales, late not poir with oe hirs of pa L. perip with oe	7. eral ala eted at <i>L. d</i> esophagu apillae <i>laneti</i> Fa	e extend the term elatorrei (is longer rooqui, 19 s shortei	ds throminal sp Chitwood (1/5 th) 	ough the pine like d, 1932. of body), of body), papillae7
7(6) Fe			pairs	nterior (V i	n	l%). Anal males
- '	iculatum Females papilla	s with vu ae	50 Ilva muc 5	h posteri	or (V% in	> 55%). males Orientalis
8(2) oesopha dispose - oesopha	agus mu d <i>L.</i> Excretor	2004 y pore m ch shorte saltense y pore m ger (b	nuch ante er (b = 9 (Achine) uch post = 7.024	erior (Ex% 9.23), late ly and Ca erior (Ex%) lateral	% = 11.5 eral alag amino, 2 % = 20.7 alae (52%) and e equally

Conflict of Interests

The author(s) have not declared any conflict of interests.

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