Journal of Nematology 42(2):128–138. 2010.

Some new and known species of genera *Tripylina* Brzeski and *Trischistoma* Cobb, 1913 (Nematoda) with a Discussion on their Relationships

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Abstract: The paper deals with description of three new and two known species of Tripylidae Örley, 1880. Two new and a known species of the genus *Tripylina* Brzeski, 1963 while one new and one known species of *Trischistoma* are described. *Tripylina ymyensis* n. sp. is characterised by robust body; long, slender outer labial setae; stoma with thick cuticularised wall bearing a prominent dorsal tooth and two equal-sized posteriorly-placed subventral teeth; large ovoid coelomocytes with conspicuous, globular inclusions; simple vagina without cuticularised pieces and a long, curved tail with a U- or S-shaped turn, uniformly tapering into a narrow terminus. *Tripylina valiathani* n. sp. is characterised by moderately large body; plump, leaf-shaped outer labial setae; stoma with thick cuticularised dorsal wall bearing a prominent dorsal tooth and two small anteriorly-placed subventral denticles; large fusiform coelomocytes with tapering ends having fine granular inclusions; cuticularised vagina, a distinct prerectum and a stumpy, laterally curved tail with a blunt terminus. *T. stramenti* (Yeates, 1972) Tsalolikhin, 1983 has been reported for the first time from India. *Trischistoma minor* n. sp. is characterised by medium-sized body; with dense crystalloids and granular aggregates; low, flattened lip region; plump outer labial setae; slenderer post-labial cephalic setae lying closely posterior to outer labials; a post-uterine sac of one body diameter and a dorsally curved tail with ventral terminal turn. *T. pellucidum* Cobb, 1913 forms the first report from India. The relationships of *Tripylina* and *Trischistoma* have been discussed.

Key words: Tripylina ymyensis n. sp, *T. valiathani* n. sp, *T. stramenti*, *Trischistoma minor* n. sp, *T. pellucidum,* description.

The family Tripylidae Örley, 1880 is a predominantly aquatic group of nematodes, with species found on land, continental waters and occasionally in sea. The group is comprised of large-sized predators that are *k*-strategists in the environment. Intestinal contents indicate that their food consists primarily of small micro fauna that often include nematodes (Yeates, 1971). The representative species tend to have long life cycles (Yeates, 1967) and life spans and are very sensitive to slight changes in the environment (Bongers, 1990). Örley (1880) placed *Tripyla* Bastian, 1865 under Tripylidae as its type genus. Cobb (1913) described the genus *Trischistoma* and proposed (1920) the Order Triplonchia. The latter was amended to Triplonchida by Siddiqi (1983). Brzeski (1963) erected a new genus *Tripylina* under Tripylidae and considered *Trischistoma* as *genus inquirendum*. Later he (1964) revised the genera *Tripyla*, *Paratripyla* and *Tripylina*. In his opinion *Paratripyla* differs from *Tripylina* in having a larger amphid, paired gonads and in dissimilar position of dorsal pharyngeal gland opening. Andrássy (1964) raised superfamily Tripyloidea to the status of suborder (Tripylina) and further transferred Oxystominidae and Lauratonematidae from Enoploidea to Tripylina. However, he (1985) considered *Trischistoma* as a valid genus and described *T. gracile*. Genus *Tripylina* has been discussed and reviewed by several workers (Tsalolikhin, 1983; Andrássy, 1985, 2007; Brzeski and Winiszewska-Ślipińska, 1993; Zullini, 2006). Loof (1973) added a new genus *Tripyloides* under family Tripylidae. Brzeski and Winiszewska-Ślipińska (1993) discussed the morphology of Tripylidae and also rediagnosed the family. He included *Tripylella* and *Tripylina* but removed *Abunema* Khera, 1971 and *Trischistoma* from the family and also added three new species to genus *Tripyla*. Andrássy (2006) redefined *Tripylella* and *Trischistoma* along with the keys to their species. Later he (2007) proposed two new sub families under Tripylidae *viz.*, Trischistomatinae for *Trischistoma*; and Tobriliinae for *Tobrilia* in addition to the existing subfamily Tripylinae de Man, 1876. He accepted six species under genus *Tripylina*. Lately Zhao (2009) reviewed the genus *Tripylina* with six valid species *Tripylina arenicola* (de Man, 1880) Brzeski, 1963, *T. longa* Brzeski and Winiszewska-Slipinska, 1993; *T. macroseta* (Vinciguerra and La Fauci, 1978) Tsalolikhin, 1983; *T. sheri* Brzeski, 1963; *T. stramenti* (Yeates, 1972) Tsalolikhin, 1983 and *T. ursulae* (Argo and Heyns, 1973) Tsalolikhin, 1983 along with five newly added species from New Zealand mainly differentiated on the basis of small subunit rDNA sequences.

During the screening of preserved samples from different localities two new and one known species of *Tripylina* and one new and one known species of *Trischistoma* were found that are reported hereunder.

Materials and Methods

Collected soil samples were processed by Cobb's (1918) sieving and decantation and modified Baerman's funnel techniques. The nematodes extracted were heat-killed and fixed in Formalin-glycerol fixative, dehydrated by slow evaporation method (Seinhorst, 1959) and mounted in anhydrous glycerine. The measurements were taken by ocular micrometer and drawings made using drawing tube attached to Olympus BX-51 DIC Microscope. LM photographs were taken by Olympus digital camera DP-11. For Scanning Electron Microscopy (SEM), the specimens were fixed in 2% glutaraldehyde, post-fixed in 2% osmium tetroxide, dehydrated in alcohol series, critical point dried using CO_2, mounted on stubs, coated with 10 nm gold before viewing under Hitachi S-3200H Scanning Electron microscope.

Received for publication March 13, 2010.
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This paper was edited by Kris Lambert.

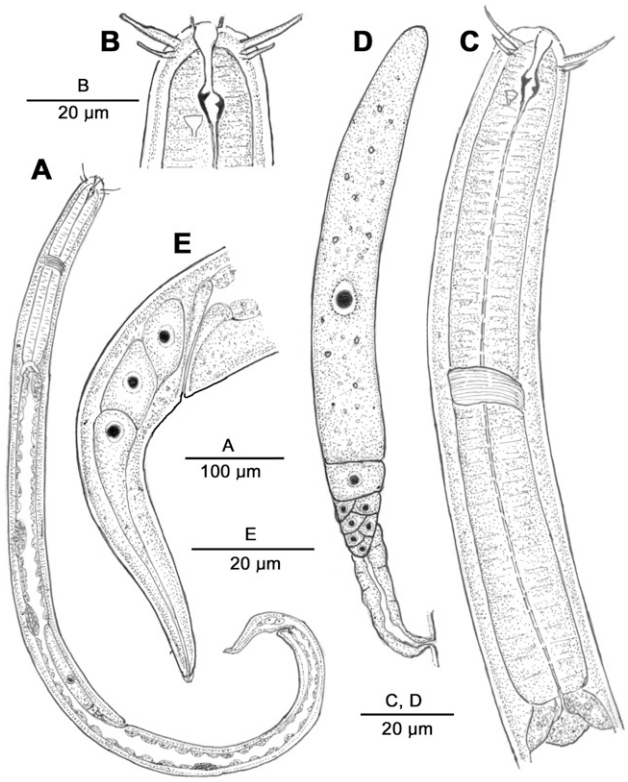

Fig. 1. *Tripylina ymyensis* n. sp. female. A) Entire. B) Anterior region. C) Pharyngeal region. D) Reproductive system. E) Caudal region.

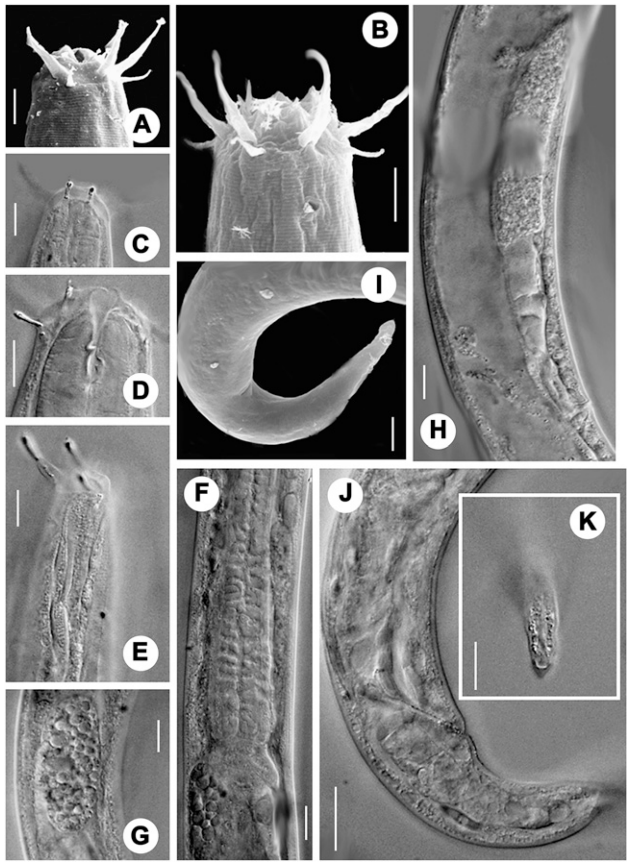

Fig. 2. *Tripylina ymyensis* n. sp. female. A,B) SEM micrograph of *En face* view. C-E) Anterior end. F) Pharyngeo-intestinal junction. G) Coelomocyte. H) Female genital system. I, J) Posterior body region. K) Tail terminus. (Scale bar = 10 µm).

Description

*Tripylina ymyensis** n. sp.
(Figs 1, 2)

Measurments of holotype and paratypes are given in Table 1.

Female (n = 9): Body moderately large, robust, J-shaped upon fixation with posterior end tapering and strongly curved. Outer cuticle transversely striated with striations indiscernible in LM. Inner cuticle smooth. Total cuticular thickness 0.5-1.5 µm at different body regions. Somatic setae sparse, paired as well as unpaired, about 0.5-1.0 µm long, 23-24 in number scattered all over body with 6 confined to pharyngeal region. Subdorsal setae in the mid-body region more conspicuous than those located on subventral or lateral sides. Crystalloids usually fine or obscure. Lip region dome-shaped and continuous with the adjoining body. Inner labial sensilla slender and raised 1.0-1.5 µm above cuticular surface; outer labial sensilla slender, regularly tapering from base to tip, 14-18 µm long and relatively cylindroid. Cephalic sensilla, 4-6 µm long, thinner than outer labials, located in the same circlet close to outer labial. Amphids calyciform, located at 20-22 µm from anterior end. Amphidial fovea small, slit-like, 2.5-3.0 µm wide or 1/10-1/14 of corresponding body diameter (Fig. 2B). Cheilostom arched, weakly sclerotized. Rest of stoma divided into two parts. Anterior triangular or funnel-shaped part is followed by posterior cylindrical part bearing small pockets for teeth. Both dorsal and subventral walls are cuticularised bearing prominent teeth. Dorsal tooth is conspicuous, triangular, borne in a socket on a ledge at 13-16 µm from anterior end. Sub-ventral teeth of equal size, located posterior to dorsal tooth in a relatively larger chamber at 15-18 µm from anterior end (Fig. 1B, C, 2D). Pharynx cylindroid, highly muscular, gradually expanding at base occupying 18-22% of body length. Body at pharyngeal end 30-36 µm wide. Nerve ring located at 45-49% of pharyngeal length from anterior end. Excretory pore can not be observed. Cardia with elongate pericardial cells of 13-17 x 5-7 µm dimension (Fig. 1C, 2F). About 5-6 large (30-40 x 13-16 µm), ovoid, coelomocytes containing conspicuously large, round globules, are present between the pharyngeal base and rectum (Fig. 2F, G). Of these, two coelomocytes lie close to cardia on middorsal and midventral sides opposite each other. Distance between pharyngeal base and vulva 450-485 µm or 2.0-2.3 times pharyngeal length. Intestine thin-walled with wide and convoluted lumen, compressed in the region of gonad. Anterior intestinal cells of similar size with smooth outline

* The species name is based on its type locality, **Y**uan **M**ing **Y**uan park, China

TABLE 1. Measurements (μm) of populations of *Tripylina ymyensis* n. sp., *T. valiathani* n. sp., *T. stramenti* (Yeates, 1972) Tsalolikhin, 1983 with mean ± standard deviation (range).

Characters	*Tripylina. ymyensis* n. sp.		*Tripylina valiathani* n. sp.		*Tripylina stramenti*
	Holotype female	Paratype females (n= 8)	Holotype female	Paratype females (n= 6)	Females (n= 9)
Body length	1020	1030.2±4.4 (1015-1056)	923	934.7±33.8 (904-992)	1097±55.1 (1034-1178)
Body diameter	38	39.5±0.9 (38-42)	32	32.7±1.9 (31-39)	39.7±1.6 (37-41)
a	26.5	26.9±0.8 (25.6-28.7)	28.8	28.5±0.7 (27.5-29.6)	27.6±1.1 (25.8-28.7)
b	4.7	5.0±0.1 (4.9-5.1)	5.4	5.2±0.2 (4.8-5.5)	5.0±0.9 (4.8-5.9)
c	17.8	16.7±1.1 (15.7-18.8)	24.2	23.7±1.5 (21.4-25.8)	15.6±3.8 (12.2-22.2)
c'	4.7	4.9±0.2 (4.7-5.2)	1.7	1.7±0.1 (1.5-1.9)	2.6±0.3 (2.2-3.1)
V	66.5	66.5±0.7 (65.7-67.3)	66.4	67.1±0.8 (66.1-67.9)	64.8±1.8 (62.3-67.4)
Lip height	5	5±0 (5-5)	3	2.7±0.4 (2-3)	6.1±1.2 (4-7)
Lip diameter	20	20.1±0.5 (18-21)	14	12.2±0.4 (12-17)	22.2±1.4 (20-24)
Stoma length	17	15.5±2.4 (15-20)	14	13.5±0.5 (13-19)	19.5±1.5 (17-26)
Stoma diameter	2	2±0 (2-2)	5	5±0.7 (4-6)	4.2±0.4 (4-5)
Pharynx	215	215.9±5.3 (205-225)	168	180.2±15.8 (164-204)	220.2±7.4 (213-230)
Nerve ring	92	91.2±4.4 (87-97)	76	81.2±3.2 (76-85)	102.5±7.2 (100-115)
Rectum length	17	15.9±0.6 (15-19)	20	20.5±2.1 (18-24)	21±1.4 (19-23)
V-A distance	286	281.5±7.4 (278-290)	272	267.7±7.8 (255-276)	312±22.9 (284-340)
Anal Body Diameter	12	11.7±1.1 (11-15)	22	20.5±0.5 (17-22)	27±1.2 (25-28)
Tail	57	58.2±0.5 (55-60)	38	39.5±3.2 (35-43)	73±12.7 (53-87)

whereas posterior intestinal cells irregular and vacuolated. Rectum 15-18 μm long or 1.0-1.3 times anal body diameter. Reproductive system mono-prodelphic without post-uterine sac. Ovary reflexed laterally with tapering distal end. Oocytes arranged compactly in germinal zone and a single large growing oocyte at proximal end (Fig. 2H). Vagina anteriorly directed, 5 μm long or 1/5-1/7 of corresponding body diameter; vaginal pieces obscure or absent. Vulva a small slit. Vulva-anus distance 278 μm long, equal to 4.8 times of tail length. Anus a crescent-shaped slit (Fig. 2I). Tail ventrally curved with a terminal turn or S-shaped, uniformly tapering into a narrow terminus (Fig. 2K). Three conspicuous caudal glands arranged in tandem, open through common duct at tail tip. Seven caudal setae present including two lateral pairs. The terminal seta 2.5-3.0 times corresponding tail diameter anterior to tail tip.

Male: Not found

Diagnosis: Tripylina ymyensis n. sp. is characterised by robust body; long, slender outer labial setae; stoma with thick cuticularised wall bearing a prominent dorsal tooth and two equal-sized posteriorly-placed subventral teeth; large ovoid coelomocytes with conspicuous, globular inclusions; simple vagina without cuticularised pieces and a long, curved tail with a U- or S-shaped turn, uniformly tapering into a narrow terminus.

Relationship: Tripylina ymyensis n. sp. differs from *T. arenicola* (de Man, 1880) Brzeski, 1963 in having greater 'ć' value (4.7-5.2 *vs* 1.9-4.5), thickened (*vs* simple) stomal walls, dorsal tooth equal to (*vs* larger than) subventral teeth, mid-ventral cervical seta 3 body diameters (*vs* 2 body diameters) from anterior end and sclerotized pieces absent (*vs* present in vagina in *T. arenicola apud* Andrássy, 1985 and Zhao, 2009); from *T. ursulae* (Argo and Heyns, 1973) Tsalolikhin, 1983 in having greater 'ć' value, mid-ventral cervical seta 2.5 body diameters (*vs* one body diameter) from anterior end and post-uterine sac absent (*vs* present in *T. ursulae apud* Zhao, 2009); from *T. macroseta* (Vinciguerra and La Fauci, 1978) Tsalolikhin, 1983 in having larger body (1015-1056 μm *vs* 800-940 μm), greater 'a' (25.6-28.7 *vs* 20-24) and 'ć' (4.7-5.2 *vs* 3.2) values, thickened (*vs* simple) stomal walls, subventral teeth prominent (*vs* indistinct) and posterior (*vs* anterior) to dorsal tooth, sclerotization present (*vs* absent in vulval region in *T. macroseta apud* Andrássy, 1985 and Zhao, 2009); from *T. sheri* Brzeski,

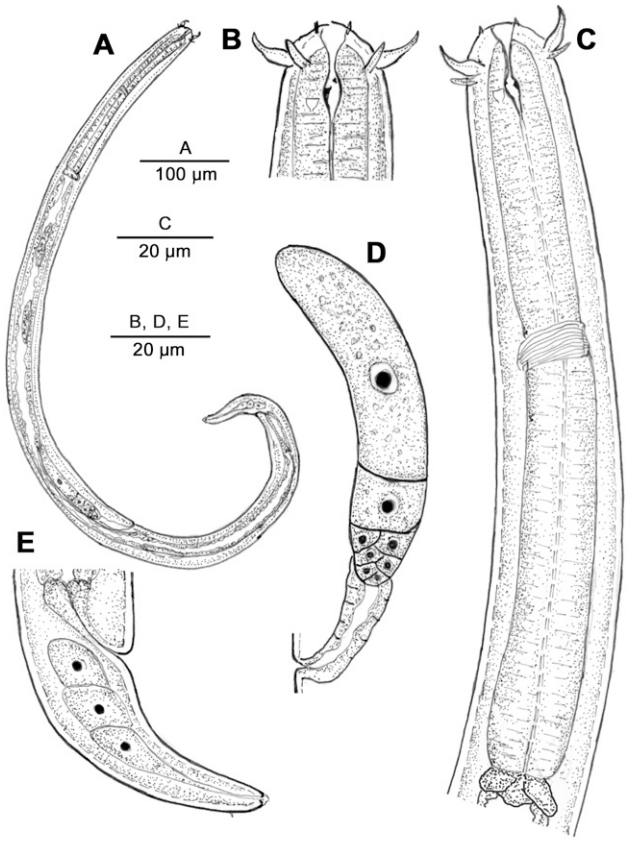

Fig. 3. *Tripylina valiathani* n. sp. female A) Entire female. B) Anterior region. C) Pharyngeal region. D) Reproductive system. E) Caudal region.

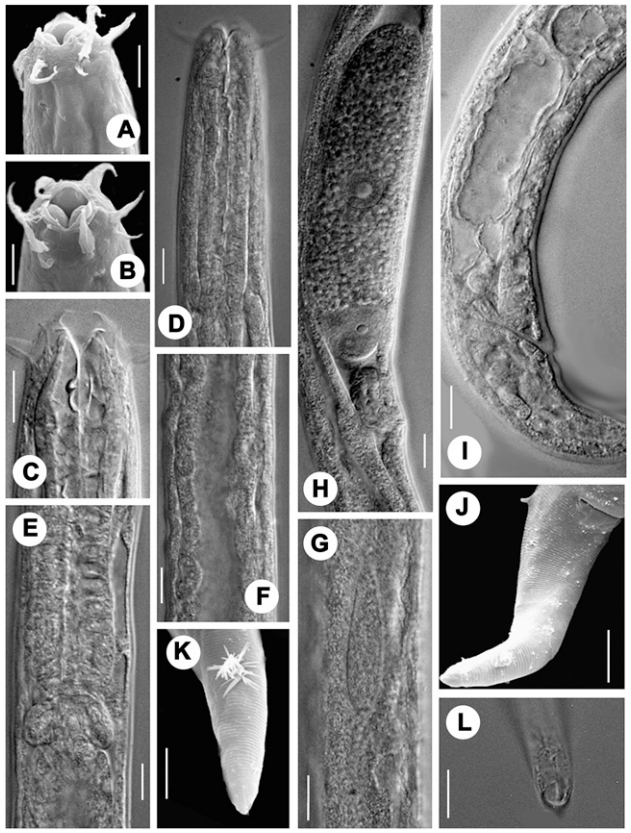

Fig. 4. *Tripylina valiathani* n. sp. female. A, B) SEM micrograph of En face view. C) Anterior end. D) Anterior pharyngeal region. E) Pharyngeo-intestinal junction., F) Intestinal region. G) Coelomocyte. H) Reproductive system. I) Anal region. J, K) Caudal region. L) Tail terminus.

1963 in having greater 'ć' value (4.7-5.2 *vs* 1.9-4.5), subventral teeth large (*vs* small) and posterior (*vs* anterior) to dorsal tooth, sclerotized pieces in vagina absent (*vs* comma-shaped in *T. sheri apud* Andrássy, 1985 and Zhao, 2009).

Type habitat and locality: Wet sandy loam soil collected from the shore of a water body at Yuan Ming Yuan Park, Beijing, China.

Type designation: Holotype female, seven paratype females on the slides *Tripylina ymyensis* n. sp. No.B/1-4 deposited at Nematode Collection of Department of Zoology, Aligarh Muslim University, Aligarh. One paratype female on slide *Tripylina ymyensis* n. sp. No.B-12, deposited at the Wageningen Nematode Collection (WaNeCo), P.O. Box 9102, 6700 HC Wageningen, The Netherlands.

Tripylina valiathani* n. sp.

(Figs 3, 4)

Measurments of holotype and paratypes are given in Table 1.

Female (n= 7): Body moderately large, robust, C-shaped upon fixation with posterior end tapering and strongly curved. Outer cuticle transversely striated with striations indiscernible in LM. Inner cuticle smooth. Total cuticular thickness 1.0-1.5 µm at different body regions. Somatic setae sparse, very small and inconspicuous, about 0.5 µm long, 14-16 in number scattered all over body with 2-3 confined to pharyngeal region. Crystalloids not conspicuous. Lip region elevated, conical and continuous with the adjoining body. Inner labial sensilla slender and slightly raised 0.5 µm above cuticular surface; outer labial sensilla 10-12 µm long, plump and leaf-shaped with wide base and pointed apex (Fig. 3B, C; 4A, B). Cephalic setae 4-5 µm long, slenderer and placed posterior to the base of outer labials apparently forming a third whorl. Amphids calyciform, located at 15-17 µm or 1.2-1.3 times labial diameter from anterior end. Amphidial fovea small, elliptical, 3-4 µm wide or 1/10-1/12 of corresponding body diameter (Fig. 4A). Cheilostom arched, weakly sclerotized. Rest of stoma divided into two parts. Anterior triangular or funnel-shaped part is followed by posterior cylindrical part bearing pocket for teeth. Dorsal tooth prominent, at 13-14 µm from anterior end whereas subventrals smaller, at 11-12 µm from anterior end. Dorsal wall more cuticularised than subventral walls (Fig. 4C). Pharynx highly muscular, 18-20% of body length, narrowing slightly at level of nerve ring but broadened posteriorly (Fig. 4D).

* The species name is given in honour of the renowned scientist Dr. M. S. Valiathan who has been very supportive for my research endeavours.

Nerve ring located at 41-46% of pharyngeal length. Body at pharyngeal end 28-30 µm wide. Excretory pore can not be observed. Cardia conoid with ovoid pericardial cells of 12-15 x 7-9 µm dimension (Fig. 3C, 4E). About 5-6 large (22-28 x 6-11 µm), elongate fusiform coelomocytes with tapering ends containing fine granules, are present between the pharyngeal base and rectum. Out of these one coelomocyte lies close to cardia. Intestine heterocytous, intestinal wall of uneven thickness having large granular cells bordering wide lumen. Intestine compressed in the region of gonad. A distinct prerectum present measuring 1.7-2.3 times corresponding body diameter (Fig. 4I). Rectum about 1.0-1.3 anal body diameter. Reproductive system monoprodelphic without posterior uterine sac, occupying 17-22% of body length. Ovary reflexed with rounded germinal end filled compactly with proliferating germ cells. A large developing oocyte placed at proximal end. Vagina directed anteriorly, cuticularised, 3-5 µm or $1/6^{th}$ to $1/10^{th}$ of corresponding body diameter. Vulva a small transverse slit. Vulva-anus distance 6.4-7.2 times tail length. Anus a crescent-shaped slit (Fig. 4J). Tail plump, bent ventrally followed by a dorsal curvature and a lateral turn, tapering to a thick and blunt terminus (Fig. 4J). Caudal glands large, three in tandem opening through a terminal spinneret. Caudal setae 5 in number including one subdorsal and one lateral pairs. The terminal seta 2.5-3.0 times tail tip diameter anterior to tail terminus.

Male: Not found

Diagnosis: Tripylina valiathani n. sp. is characterised by moderately large body; plump, leaf-shaped outer labial setae; stoma with thick cuticularised dorsal wall bearing a prominent dorsal tooth and two small anteriorly-placed subventral denticles; large fusiform coelomocytes with tapering ends having fine granular inclusions; cuticularised vagina, a distinct prerectum and a stumpy, laterally curved tail with a blunt terminus.

Relationship: Among the smaller species with anteriorly placed subventral teeth, *Tripylina valiathani* n. sp. differs from *T. sheri* Brzeski, 1963 in having relatively smaller body (904-992 µm *vs* 780-1380 µm), relatively greater 'ć' value (1.5-1.9 *vs* 1.9-4.5), dorsal wall thickened than (*vs* similar to) subventral walls, prerectum present (*vs* absent) and smaller tail (35.0-43.0 *vs* 48.1-70.9 in *T. sheri apud* Andrássy, 1985 and Zhao, 2009); from *T. macroseta* (Vinciguerra and La Fauci, 1978) Tsalolikhin, 1983 in having greater 'a' (27.5-29.6 *vs* 20-24) and smaller 'ć' (1.5-1.9 *vs* 3.2) values, thickened (*vs* simple) stomal walls, subventral teeth prominent (*vs* indistinct), vaginal wall sclerotised (*vs* not sclerotized) and prerectum present (*vs* absent in *T. macroseta apud* Andrássy, 1985 and Zhao, 2009); from *T. tamaki* Zhao, 2009 in having greater 'a' (27.5-29.6 *vs* 20-23) and 'c' (21.4-25.8 *vs* 14.5-15.6) values, smaller 'ć' (1.5-1.9 *vs* 2.9-3.2) and 'b' (4.8-5.5 *vs* 5.8-6.5) values, smaller (4-5 µm *vs* 6-7 µm) cephalic setae and prerectum present (*vs* absent in *T. tamaki apud* Zhao, 2009); from *T. stramenti* (Yeates, 1972) Tsalolikhin, 1983 in having smaller body (0.90-0.99 mm *vs* 1.0-1.7 mm), relatively greater 'c' (21.4-25.8 *vs* 12.2-22.2), smaller 'ć' (1.5-1.9 *vs* 2.2-3.5) values, leaf-shaped (*vs* tapering) outer labial setae, cephalic setae appearing in a separate (*vs* same) whorl of outer labials, vagina with sclerotized pieces (*vs* simple, without sclerotized pieces) and a distinct prerectum present (*vs* absent in *T. stramenti apud* Yeates, 1972 and Zhao, 2009); from *T. ymyensis* n. sp. in having greater 'c' (21.4-25.8 *vs* 15.7-18.8), smaller 'ć' (1.5-1.9 *vs* 4.7-5.2) values, leaf-shaped (*vs* slender, uniformly tapering) outer labial setae, cephalic setae appearing in a separate (*vs* same) whorl of outer labials, dorsal tooth anterior (*vs* posterior) to subventral denticles, subventral denticles smaller than (*vs* equal to) dorsal tooth, coelomocytes fusiform with pointed ends (*vs* ovoid or egg-shaped) with fine (*vs* conspicuous and globular) granules, larger rectum (18-24 *vs* 15-19 µm) and a distinct prerectum present (*vs* absent in *T. ymyensis* n. sp.)

Type habitat and locality: Soil collected from from Kishtwar, Srinagar, Jammu and Kashmir, India.

Type designation: Holotype female, five paratype females on the slides *Tripylina valiathani* n. sp. No.JK/1-3 deposited at Nematode Collection of Department of Zoology, Aligarh Muslim University, Aligarh. One paratype female on slide *Tripylina valiathani* n. sp. No.JK/4, deposited at the Wageningen Nematode Collection (WaNeCo), P.O. Box 9102, 6700 HC Wageningen, The Netherlands.

Tripylina stramenti (Yeates, 1972) Tsalolikhin, 1983
(Figs 5, 6)

Measurments of females are given in Table 1.

Female (n = 9): Body large, robust, C-shaped upon fixation with posterior end tapering and curved. Cuticle transversely striated with striations indiscernible in LM, 0.9-1.5 µm thick at different body regions. Somatic setae sparse, very small and inconspicuous to be observed. In few specimens a midventral seta in anterior half of pharynx was observed with variable positions. Crystalloids fine and compactly arranged in pharyngeal region. Lip region continuous with the adjoining body. Inner labial sensilla slightly raised above cuticular surface; outer labial sensilla 13-16 µm long with tapering distal ends. Cephalic setae 5-7 µm long, slenderer and placed close to the base of outer labials. Amphids calyciform, located at 19-20 µm or 1.2-1.3 times labial diameters from anterior end. Amphidial fovea small, elliptical, 6-7 µm wide or 1/5-1/6 of corresponding body diameter (Fig. 5B, C). Cheilostom arched, weakly sclerotized. Rest of stoma divided into two parts. Anterior triangular or funnel-shaped part is followed by posterior cylindrical part bearing pocket for teeth. Dorsal tooth prominent, at 17-21 µm from anterior end whereas subventrals smaller, at 15-17 µm from anterior end. Dorsal wall cuticularised (Fig. 6B). Pharynx cylindroid, highly muscular, 20-21% of body length with

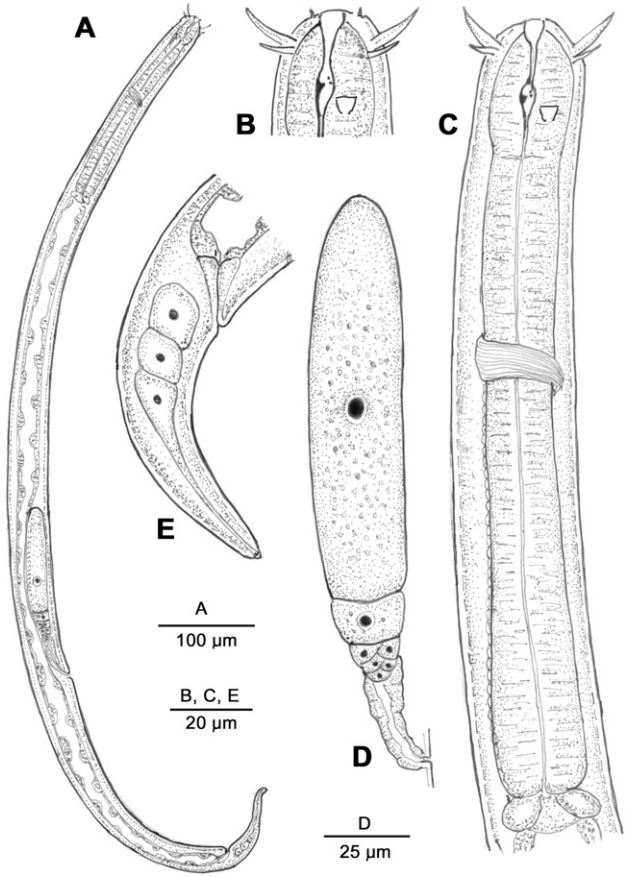

Fig. 5. *Tripylina stramenti* Cobb, 1913 female. A) Entire. B) Anterior end. C) Pharyngeal region. D) Reproductive system. G) Caudal region.

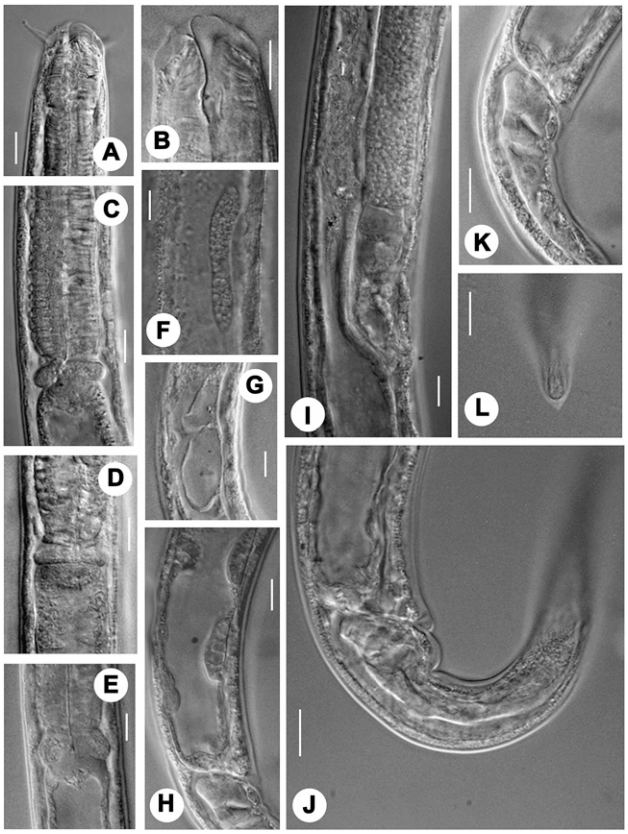

Fig. 6. *Tripylina stramenti* Cobb, 1913 female. A, B) Anterior region. C-E) Pharyngeo-intestinal junction. F) Coelomocyte. G, H) Intestinal region. I) Reproductive system. J, K) Anal region. (Scale bar = 10 µm).

an anterior peribuccal swelling. Nerve ring located at 46-50% of pharyngeal length . Body at pharyngeal end 34-40 µm wide. Excretory pore can not be observed. Cardia conoid with round to ovoid pericardial cells of 12-15 x 7-9 µm dimension (Fig. 6C, D, E). About 6-7 large (35-42 x 6-9 µm), elongate, cylindroid coelomocytes containing fine granules, are present between the pharyngeal base and rectum. Out of these, two coelomocytes lie close to cardia on middorsal and midventral side opposite each other. Intestine heterocytous and compressed in the region of gonad. Intestinal wall of irregular thickness bordering wide intestinal lumen. Rectum about 0.7-0.9 times corresponding anal body diameter. Reproductive system mono-prodelphic without posterior uterine sac, occupying 20-25% of body length. Ovary laterally reflexed having small germinal end filled compactly with proliferating germ cells. A large developing oocyte occupying greater volume of ovary. Vagina directed anteriorly, not cuticularised, 5-7 µm long or 1/5 to 1/6 of corresponding body diameter. Vulva a small slit. Vulva-anus distance 3.9-5.3 times tail length. Anus a crescent-shaped slit.

Tail uniformly tapering, ventrally curved followed by a dorsal curvature. Caudal glands large, three in tandem opening through a terminal spinneret. Caudal setae cannot be observed.

Male: Not found

Habitat and locality: Soil samples collected from flower bed in the lawn of Department of Zoology, Aligarh Muslim University, Aligarh.

Voucher specimens: Eight females on slide *Tripylina stramenti*/No. ALG 1-3 deposited in the Nematode Collection, Department of Zoology, Aligarh Muslim University, Aligarh. One female on slide *Tripylina stramenti*/No. ALG 4, deposited at the Wageningen Nematode Collection (WaNeCo), P.O. Box 9102, 6700 HC Wageningen, The Netherlands.

Remarks: The present population shows the first record of *T. stramenti* (Yeates, 1972) Tsalolikhin, 1983 from India. Earlier the species has been reported from New Zealand and West Africa. The present specimens resemble *T. stramenti* in all morphometric and morphological characteristics except the smaller body size and the slightly larger outer labial sensilla. However, the sensilla length (13-16 µm) conforms well to the population (13-15 µm) of *T. stramenti* described from West Africa (Andrássy, 2008). No males could be found in the present population similar to the observations of Yeates (1971), Andrássy (2008) and Zhao (2009), indicating the parthenogenetic mode of reproduction in the species. Ovoviviparity or intra-uterine segmentation was not observed. Earlier *Abunema indicum* Khera, 1971 has been reported from India from soil at the bank of a stagnant pond but was later synonymised with *T. arenicola*

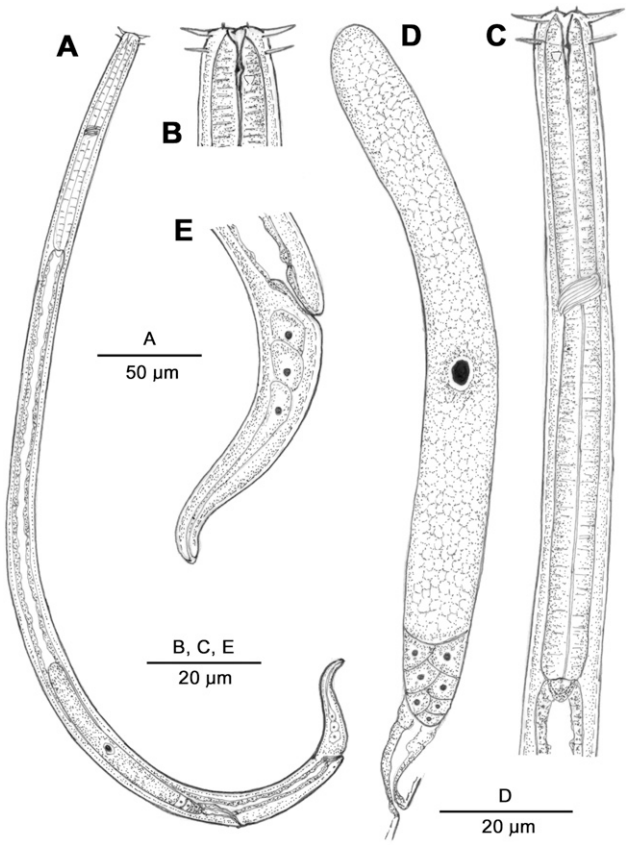

FIG. 7. *Trischistoma minor* n. sp. female. A) Entire. B) Anterior end. C) Pharyngeal region. D) Reproductive system. G) Caudal region.

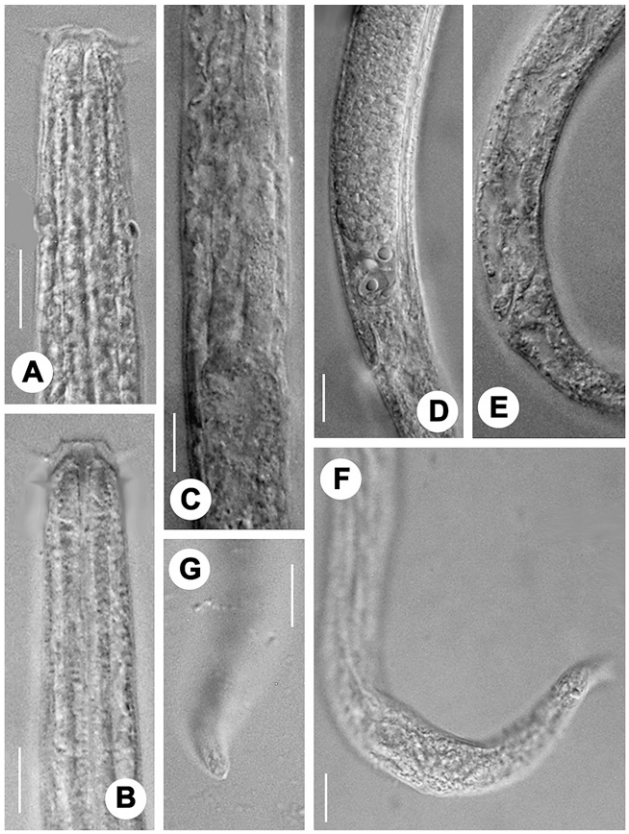

FIG. 8. *Trischistoma minor* n. sp. female. A, B) Anterior body region. C) Posterior pharyngeal region. D) Reproductive system. E, F) Posterior body region. G) Tail terminus. (Scale bar = 10 µm).

by Andrássy, 1985. The present specimens differ from the population described by Khera, 1971 in having a relatively greater 'a' value (12.2-22.2 *vs* 12-14), three lips (*vs* six indistinct lips), calyciform (*vs* ovoid) amphidial fovea, longer stoma (17-26 µm *vs* 12-14 µm), larger rectum (19-23 µm *vs* 10 µm) and absence (*vs* presence) of intra-uterine egg/s. However, the features of *Abunema indicum* as reported by Khera, 1971, *viz.*, six lips, only six setose sensilla, ovoid amphidial fovea as well as the cylindroid oral aperture do not conform to characteristics of *Tripylina* hence the species *Abunema indica* (=*indicum*) is considered *species inquirenda*.

Trischistoma minor* **n. sp.**

(Figs. 7, 8)

Measurments of holotype and paratypes are given in Table 2.

Female (n= 9): Body small to medium-sized, C-shaped upon fixation with posterior end tapering and curved. Outer and inner cuticle smooth, cuticular thickness 0.5 µm in different regions of body. Somatic setae very sparse almost indiscernible due to densely packed crystalloids and granulation present in pseudocoelom (Fig. 8A, C, E). Lip region low, flattened, continuous with the adjoining body. Inner labial sensilla raised above cuticular surface; outer labial sensilla 5-6 µm long, plump. The submedian cephalic sensilla 2-3 µm long, slender, placed post-labially about 3-4 µm posterior to outer labials (Fig. 7B, C; 8A, B). Amphids calyciform, amphidial fovea small, elliptical, located at 7-10 µm from anterior end, occupying 1/3 to 1/4 of corresponding body diameter. Stoma triangular or slightly widened at anterior end with posterior narrowing. Cheilostom simple, sclerotized. Dorsal wall of stoma not thickened, inconspicuously dilated to hold a minute dorsal tooth at 11-12 µm from anterior end. Subventral denticles indiscernible. Pharynx cylindroid, muscular, 17-21% of body length. Nerve ring located at 47-59 % of pharyngeal length from anterior end. Body at pharyngeal end is 12-16 µm wide. Excretory pore can not be observed. Cardia 3-9 µm long, without distinct pericardial cells. Distance between pharynx base and vulva 367-436 µm long, 2.9 times longer than pharynx. Intestine tubular with walls of irregular thickness bordering wide intestinal lumen, not compressed in the region of gonad. Rectum almost equal to anal body diameter. Reproductive system mono-prodelphic occupying 41-44% of body length. Ovary laterally reflexed, located at right side of intestine. Germinal end with compactly arranged germ cells while a large developing oocyte present at the proximal end of ovary. Vagina directed forward,

* The species name indicates its smallest body size among the congeners.

TABLE 2. Morphometric characteristics of *Trischistoma minor* n. sp. and *T. pellucidum* Cobb, 1913. Measurements are in μm and in the form: mean ± standard deviation (range).

Character	*T. minor*		*T. pellucidum*
	Holotype female	Paratype females (n = 8)	Females (n = 11)
Body length	550	521.5±97.5 (424-619)	686.6±33.5 (641-748)
Body diameter	17	15.0±2 (13-17)	17.7±0.6 (17-19)
a	32.3	34.5±1.9 (30.3-36.4)	38.9±2.4 (35.7-43.5)
b	4.0	3.9±0.09 (3.7-4.2)	4.8±0.2 (4.3-5.2)
c	11.9	11.5±0.9 (10.6-12.4)	12.3±1.0 (10.3-14.5)
c'	4.1	5.1±1.1 (4.0-6.3)	4.6±0.4 (4.1-5.8)
V	79.8	79.9±0.1 (79.5-80.1)	80.0±1.8 (76.4-83.6)
Lip height	2	2±0 (2-2)	3.2±0.2 (3-4)
Lip diameter	9	8.5±0.5 (8-9)	9.0±0.5 (9-11)
Stoma length	7	6.5±0.5 (6-8)	7.0±1.9 (6-10)
Stoma diameter	2	2±0 (2-2)	2±0 (2-2)
Pharyngeal length	135	134.5±9.5 (125-144)	141.9±11.5 (125-166)
Nerve ring	68	59±4 (65-73)	57.3±5.5 (50-63)
Anal Body Diameter	11	10.5±0.5 (10-11)	12.0±0.6 (11-13)
Rectum length	9	8±0 (8-9)	10.2±1.4 (7-12)
Vulva-anus distance	75	75±2.6 72-78	89.4±5.8 70-100
Tail length	46	45±5 (40-50)	56.0±5.5 (50-70)

4-5 μm long, 1/4 of corresponding body width. Vulva-anus distance about 1.3-2 times of tail length. Anus a crescent-shaped slit. Tail uniformly tapering, ventrally curved with a terminal dorsal turn. Three caudal glands arranged in tandem with terminal opening.

Male: Not found

Diagnosis: Trischistoma minor n. sp. is characterised by medium-sized body; with dense crystalloids and granular aggregates; low, flattened lip region; plump outer labial setae; slenderer post-labial cephalic setae lying closely posterior to outer labials and a dorsally curved tail with ventral terminal turn.

Relationship: Trischistoma minor n. sp. resembles *T. pellucidum* Cobb, 1913 and *T. gracile* Andrássy, 1985 in lacking a post-uterine sac but differs from *T. pellucidum* in having relatively smaller body (0.4-0.6 mm *vs* 0.6-0.8 mm), relatively smaller 'a' (30.3-36.4 *vs* 36-43) and 'b' (3.7-4.2 *vs* 4.4-4.6 values) and distance between the outer labials and cephalic setae (less *vs* equal to or more than cephalic seta length in *T. pellucidum* Cobb, 1913); from *T. gracile* Andrássy, 1985 in having relatively smaller body (0.4-0.6 mm *vs* 1.0-1.1 mm), smaller 'a' (30.3-36.4 *vs* 49-53) and 'b' (3.7-4.2 *vs* 4.9-5.3) values, relatively smaller 'c' (10.6-12.4 *vs* 12.2-13.3), shorter distance between the outer labials and cephalic setae (3-4 μm *vs* 6.5-7.0 μm in *T. gracile* Andrássy, 1985); from *T. equatoriale* Andrássy, 2006 in having smaller body (0.4-0.6 mm *vs* 1.3-1.6 mm), smaller 'a' (30.3-36.4 *vs* 38-44) and 'b' (3.7-4.2 *vs* 5.4-5.6) values, relatively smaller 'c' (10.6-12.4 *vs* 12.2-13.3), shorter distance (3-4 μm *vs* 6-7 μm) between the outer labials and cephalic setae, simple (*vs* sclerotized) vulval lips and post-uterine sac absent (*vs* present in *T. equatoriale* Andrássy, 2006); from *T. monohystera* (de Man, 1880) Schuurmans Stekhoven, 1951 in having smaller body (0.4-0.6 mm *vs* 1.4-1.8 mm), smaller 'a' (30.3-36.4 *vs* 45-70) and 'b' (3.7-4.2 *vs* 5.0-5.6) values, and post-uterine sac absent (*vs* present in *T. monohystera* (de Man, 1880) Schuurmans Stekhoven, 1951 *apud* Andrássy, 1985).

Type habitat and locality: Sandy loam soil collected from a ditch at Keoladeo National Park, Rajasthan.

Type designation: Holotype female, seven paratype females on the slides *Trischistoma minor* n. sp. No.KNP/7-9 deposited at Nematode Collection of Department of Zoology, Aligarh Muslim University, Aligarh. One paratype female on slide *Trischistoma minor* n. sp. No. KNP /6, deposited at the Wageningen Nematode Collection (WaNeCo), P.O. Box 9102, 6700 HC Wageningen, The Netherlands.

Trischistoma pellucidum Cobb, 1913
(Figs. 9, 10)

Measurments of females are given in Table 2.

Female (n= 11): Body medium-sized, uniformly wide except tapering behind the vulva, dorsally curved, J-shaped upon fixation. Outer and inner cuticle smooth, cuticular thickness 0.5 μm in different regions of body. Body with two pairs of somatic setae at level of nerve ring. Body structures obscured by densely packed crystalloids and granulation present in pseudocoelom (Fig.10B, D, J). Lip region round or conical, continuous with the adjoining body. Inner labial sensilla raised above cuticular surface; outer labial sensilla 5-7 μm long, on outer lip margins. submedian cephalic sensilla relatively slender, 4-5 μm long, slenderer and shorter than outer labials and located 5-8 μm posterior to latter (Fig. 10A, B). Amphids 3-4 μm wide, calyciform, located at 1.5 lip diameter from anterior end. Stoma funnel-shaped, slightly sclerotised with posterior narrowing. Cheilostom and gymnostom fused, dorsal metastegostomal wall with inconspicuous tooth, located at one labial diameter from anterior end. Subventral denticles almost indiscernible. Pharynx cylindroid, muscular, 17-23% of body length. Nerve ring located at 32-49% of pharyngeal length. Secretory-excretory pore can not be observed. Body width at pharyngeal end 12-16 μm. Cardia conoid, 5 μm with conspicuous nuclei. A pair of coelomocytes present at base of pharynx. Distance between pharynx base and vulva 320-350 μm or 2.3-2.5 times as long as pharynx. Intestine granular with wide

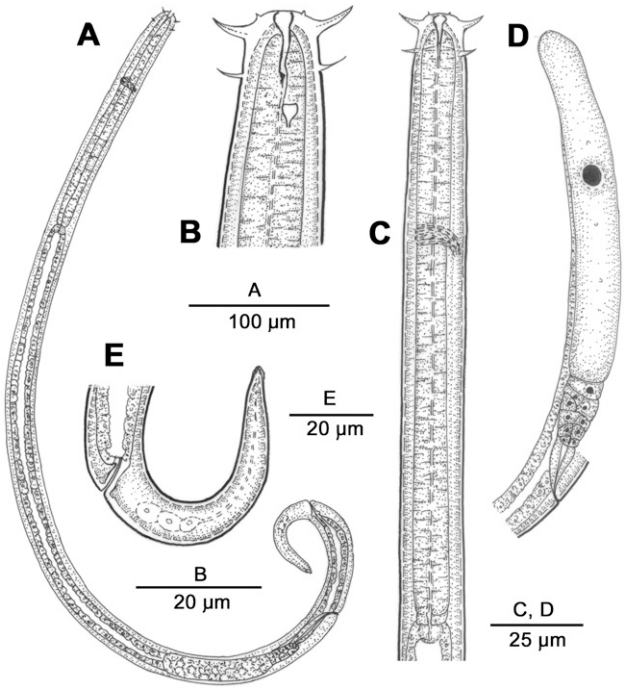

FIG. 9. *Trischistoma pellucidum* Cobb, 1913 female. A) Entire. B) Anterior end. C) Pharyngeal region. D) Reproductive system. G) Caudal region.

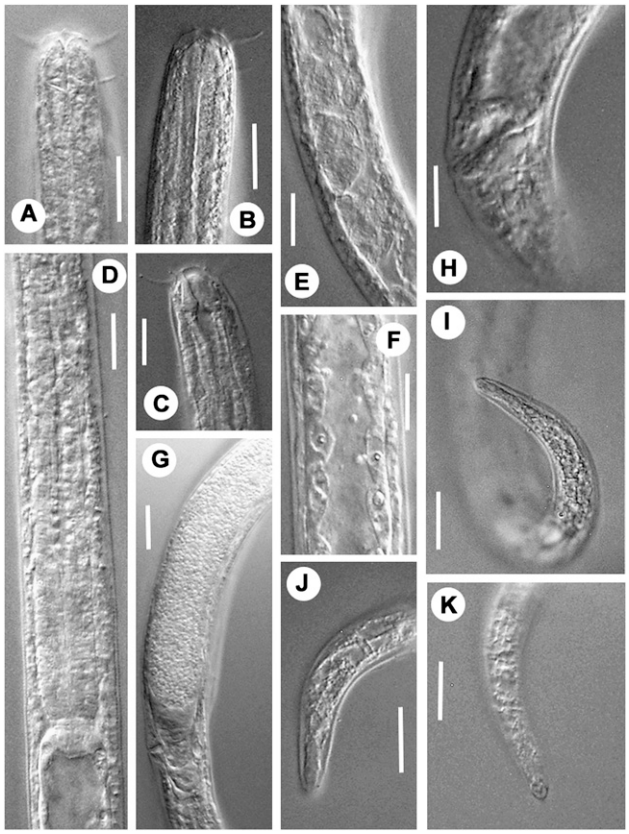

FIG. 10. *Trischistoma pellucidum* Cobb, 1913 female. A-C) Anterior body region. D) Posterior pharyngeal region. E, F) Intestinal region. G) Reproductive system. H) Anal region. I-K) Posterior body region. (Scale bar = 10 µm).

lumen not compressed in the region of gonad. Intestinal cells prominent with conspicuous granules. Rectum thin-walled, less than one anal body diameter long. Rectum almost equal to anal body diameter. Anus a crescent-shaped slit. Reproductive system monodelphic, prodelphic occupying 22% of body length without a post-uterine sac. Ovary laterally reflexed, on right side of intestine, never extending beyond vulva. Mature oocytes of 94x18 µm dimension observed at proximal end of ovary. Vagina anteriorly directed, about 1/3 vulval body diameter long. Vulva equatorial, a transverse slit. Vulva-anus distance 70-100 µm. Tail elongate conoid, regularly tapering, dorsally bent with a terminal ventral turn. Three caudal glands arranged in tandem opening terminally through small, weak spinneret. Tail uniformly tapering, ventrally curved.

Male: Not found

Habitat and locality: Sample containing *Trischistoma pellucidum* Cobb, 1913 collected from duck pond, Department of Zoology, Aligarh Muslim University, Aligarh, Uttar Pradesh, India.

Voucher specimens: Ten females on slide *Trischistoma pellucidum* No.NOA/5 1-6 deposited in Nematode Collection, Department of Zoology, Aligarh Muslim University, Aligarh, India. One female on slide *Trischistoma pellucidum* No.NOA/7, deposited at the Wageningen Nematode Collection (WaNeCo), P.O. Box 9102, 6700 HC Wageningen, The Netherlands.

Remarks: The present specimens of *Trischistoma pellucidum* Cobb, 1913 form the first report of the genus from India. The specimens agree well in all aspects with *T. pellucidum apud* Cobb, 1913. However, some minor differences were observed with the population described by Andrássy, 1968 *viz.*, greater 'ć' value (4.1-5.8 *vs* 3.6-3.9), 'V' value (76.4-83.6 *vs* 73-74) and longer sub cephalic setae (4-5 *vs* 3-3.5). This species has also been reported from Mexico, Jamaica and Paraguay (Andrássy, 1985).

DISCUSSION

The species of both genera *Tripylina* Brzeski, 1963 and *Trischistoma* Cobb, 1913 are largely continental and soil dwelling species reported mainly from litter and moss or soil around roots of plants. Showing a wide distribution, the species have been reported from tropical and subtropical regions of all the continents except Australia and Antarctica (Fig. 11). Genus *Tripylina* represents medium to large-sized, usually plump species of about 0.7-1.8 mm. Other features of the genus include smooth or finely striated cuticle (striations usually indiscernible in LM) with sparse somatic setae. Labial sensilla in 6+10 configuration; six slightly raised labial sensilla in inner circlet, six longer outer labials and four shorter submedian cephalic sensilla in outer circlet. Stoma having anterior triangular or funnel-shaped part followed by posterior cylindrical part bearing a small pocket for one prominent dorsal tooth and two subventral teeth/denticles. Pharynx cylindroid with pharyngeal-intestinal valve comprised of three glandular

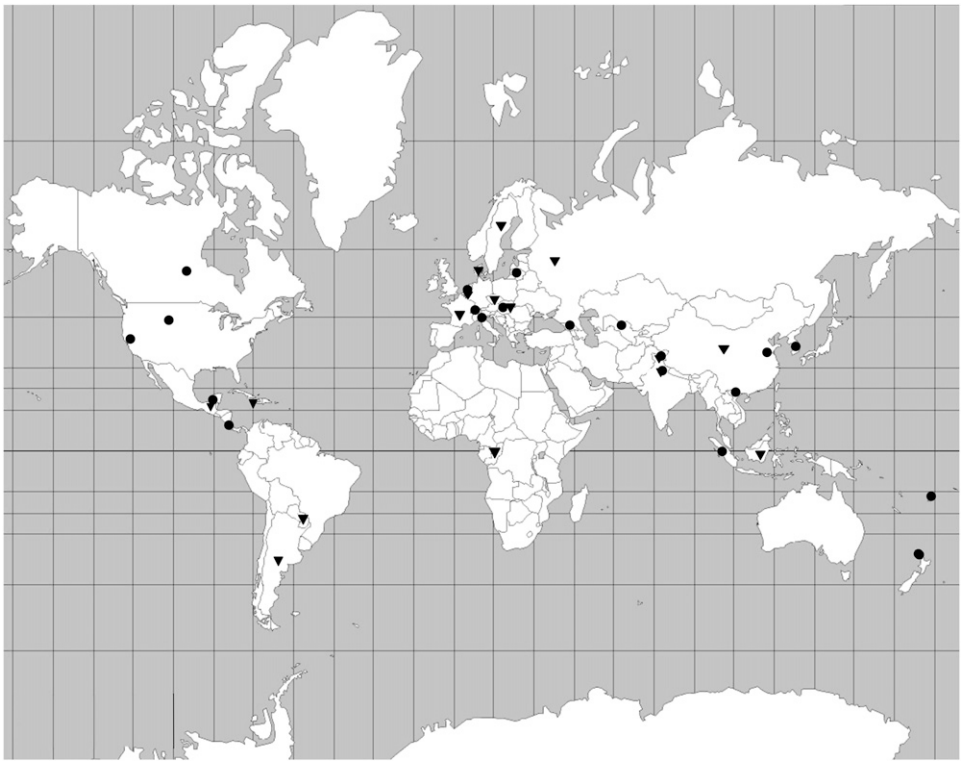

Fig. 11. World map showing distribution of species of *Tripylina* Brzeski, 1963 and *Trischistoma* Cobb, 1913. circle = *Tripylina* spp.; triangle = *Trischistoma* spp.

pericardial cells. Intestine compressed in the region of gonad. Female reproductive system mono-prodelphic with reflexed ovary and no post-vulval uterine sac. Vagina simple or with sclerotization. Tail in both sexes strongly bent or twisted. Zhao (2009) grouped the species of *Tripylina* based on anterior or posterior location of subventral teeth with respect to dorsal one. Two species *viz.*, *T. arenicola* and *T. ursulae* fall apart from rest in having posteriorly placed subventrals as also found in present *T. ymyensis* n. sp. population. However, there is disparity regarding the position of teeth in Zhao's description of *T. ursulae* (Page No. 21) and its diagnosis and relationship, presumably due to oversight. The six previously described species of *Tripylina* along with two presently described ones can be well differentiated on the basis of gross morphological differences. The five lately-added species from New Zealand were separated on the basis of small subunit ribosomal DNA sequences (Zhao, 2009). These species with nearly similar morphometrics and close alignment in the phylogenetic tree, have been identified on the basis of position of cervical setae, the sole differentiating morphological character. It appears to be a rare coincidence as the cervical setae in the present populations were not found to be consistent in number or position. Generally the species showing genetic differences show their manifestation in morphological characters unless they are sibbling species. Therefore, the difference may exist presumably in the characters not commonly accounted for as differentiating ones. In this regard, the shape and appearance of coelomocytes based on their inclusions, can be good differentiating characters.

Genus *Trischistoma* Cobb, 1913 is represented by small to large-sized (0.4-2.2 mm), slender species usually with dorsal curvature of body. The cuticle is thin and smooth. The arrangement of labial sensilla is in 6+6+4 configuration with four submedians placed in the third circlet much posterior to outerlabials. Stoma having narrow walls without a distinct pocket but minute denticles. Female reproductive system mono-prodelphic with or without post-uterine sac. Tail in both sexes bent. Despite the dissimilarity in the body curvature (ventral *vs* dorsal), configuration of lip sensilla (6+10 *vs* 6+6+4), cardial glandular cells (conspicuous *vs* obscure), the genera *Tripylina* and *Trischistoma* demonstrate similarity in various characteristics *viz.*, the smooth and thin cuticle, stoma with anterior funnel-shaped part and posterior narrow part with three small teeth/denticles, six longer outer labials and four shorter submedian setae, female reproductive system mono-prodelphic with posteriorly placed vulva and a twisted or variously bent tail. Hence their placement (Zullini, 2006; Andrássy, 2007) in the same family Tripylidae seems to be justified. The evidence of their close relationship can be found in *Tripylina valiathani* n. sp. where a transitional position of the submedian cephalic sensilla has been observed. The cephalic sensilla instead of forming a whorl with outer labials, are shifted slightly posteriad. A farther placement of these cephalic sensilla has been witnessed in *Trischistoma*. The close grouping of the two genera was

further supported by the small subunit ribosomal DNA sequences (Zhao and Buckley, 2009 and Megen et al., 2009) that showed *Tripylina* and *Trischistoma* to be closely related but much distant from the supposedly confamilial taxa *Tripyla* and *Tripylella*. Earlier the studies of Meldal et al. (2007) showed a closer position of *Trischistoma monohystera* and *Ironus dentifurcatus* but no conclusions can be drawn in view of the inadequate number of representatives. The Beyesian analysis (Zhao and Buckley, 2009) as well as fast likelihood methods (Megen et al., 2009) revealed the heterogeneity of Tripylidae taxon thus putting a question mark on its credence as the genera *Tripylina* and *Trischistoma* aligned under Enoplida while *Tipyla* and *Tripyllella* grouped under Triplonchida. *Trischistoma* showed a much closer relationship with *Trefusia* de Man, 1893 having similarities in the morphology of the labial region, the amphids, the digestive system and the male copulatory apparatus (Megen et al., 2009).

Acknowledgments. The financial support granted by TWAS, Italy; CAS, China; DST, Delhi and MEF, Delhi during the tenure of the study has been gratefully acknowledged.

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