

Two new species of *Paratrichodoros* Siddiqi, 1974 (Nematoda : Trichodoridae) with observation on *P. mirzai* (Siddiqi, 1960) Siddiqi, 1974 and *P. renifer* Siddiqi, 1974 from Pakistan

Kazi NASIRA and Mohammad A. MAQBOOL *

National Nematological Research Centre,
University of Karachi, Karachi-75270, Pakistan.

Accepted for publication 28 September 1993.

Summary – Two new *Paratrichodoros* species found in the rhizosphere of guava (*Psidium guajava*) are described and illustrated from Faisalabad, Pakistan. *Paratrichodoros psidiumi* n. sp., can easily be distinguished from all known *Paratrichodoros* species by smaller spicules, gubernaculum and body length, number of lateral body pores posterior to vulva. *P. faisalabadensis* n. sp., differs from species of the genus in combination of characteristics : shape of vagina and vaginal sclerotization, smaller spicules and gubernaculum. Additional morphometrics and morphological details are given for *Paratrichodoros mirzai* (Siddiqi, 1960) Siddiqi, 1974 having bivulval character and *P. renifer* Siddiqi, 1974.

Résumé – Deux nouvelles espèces de *Paratrichodoros* Siddiqi, 1974 (Nematoda : Trichodoridae) et observations sur *P. mirzai* (Siddiqi, 1960) Siddiqi, 1974 et *P. renifer* Siddiqi, 1974 provenant du Pakistan – Deux nouveaux *Paratrichodoros* provenant de la rhizosphère de goyavier (*Psidium guajava*), à Faisalabad, Pakistan, sont décrits et illustrés. *Paratrichodoros psidiumi* n. sp. peut être aisément distingué de toutes les espèces déjà décrites par la taille plus faible du corps, des spicules et du gubernaculum, et par le nombre de pores latéraux postérieurs à la vulve. *P. faisalabadensis* n. sp. diffère des autres espèces par la combinaison de caractères suivante : forme du vagin et de la sclérotisation vaginale, spicules et gubernaculum plus petits. Des précisions morphométriques et morphologiques sont données sur *Paratrichodoros mirzai* (Siddiqi, 1960) Siddiqi, 1974 – vulve double – et *P. renifer* Siddiqi, 1974.

Key-words : *Paratrichodoros*, nematode, morphology, taxonomy.

Trichodorid nematodes are fairly common in Pakistan yet up till now their characteristics have not been well documented. Nematodes of the family Trichodoridae are obligatory root ectoparasites of many crop plants and other vegetation. Their importance is determined not only by their own pathogenic effect but also by the ability of more than ten species of this family to transmit plant viruses (Hooper, 1973). The representatives of the family are widespread and their great number often results in specific damages to grown crops.

So far two species belonging to the genus *Trichodoros* Cobb, 1913 and two species to the genus *Paratrichodoros* Siddiqi, 1974 have been reported from Pakistan. The earliest record of the trichodorid was *Trichodoros obtusus* Cobb, 1913 reported by Akhtar (1962) on sugarcane. In the same year Siddiqi (1962) described a new species of stubby root nematode *Trichodoros pakistanensis* from soil samples collected around the roots of mulberry plants in Haripur Abbottabad region. Anwar and Sarwar (1981) reported *Paratrichodoros minor* (Colbran, 1956) Siddiqi, 1974 (= *T. christiei*) in 32 % of citrus samples from Punjab. According to them the ectoparasitic nematode *P. minor* was found usually in high number in roots and rhizosphere of declining trees rather than healthy ones. The presence of species of this genus suggests that they may play a vital role in the

transmission of citrus virus diseases which are gaining importance in the Punjab. Anwar *et al.* (1986) found *P. minor* in 30 % of sugar cane sample. Maqbool (1984) reported the association of *Paratrichodoros mirzai* and *Trichodoros obtusus* in sugarcane growing areas of Pakistan.

Specimens were killed by gentle heat, fixed in TAF and mounted in thin glass slides. They were placed in a tiny drop of glycerine and covered with a 19 mm cover slip supported by paraffin wax. Measurements were taken by an ocular micrometer, illustrations were made with the aid of a drawing tube and photomicrographs were taken with an automatic camera attached to the high-resolution compound microscope.

Paratrichodoros psidii sp. n. (Figs 1 & 5)

MEASUREMENTS

See Table 1.

DESCRIPTION

Female : General appearance typical of Trichodoridae. Body straight after fixation. Cuticle swollen, about 5 µm at lip region, slightly separated from the body

Table 1. Morphometric data of *Paratrichodorus psidii* n. sp. (all measurements are in μm except L.).

	Holotype (female)	Females (Paratypes)	Males (Paratypes)
n		10	6
L (mm)	0.44	0.40-0.52 (0.46 \pm 0.04)	0.43-0.51 (0.48 \pm 0.01)
Body diam	32.8	30.4-33.6 (31.9 \pm 1.34)	27-35 (30 \pm 3.0)
Pharynx	34.4	32-37.6 (30.9 \pm 1.97)	33.6-35.0 (34.3 \pm 0.26)
Onchiostyle	32	30.4-33.6 (31.9 \pm 1.34)	30-32 (30.6 \pm 0.89)
Ant. end to excret. pore	54.2	49.6-60 (53.4 \pm 3.7)	52-59 (56.2 \pm 2.92)
Ant. end to CP 1	-	-	30.4-35.6 (32.8 \pm 1.82)
Spicule	-	-	30-31 (30.6 \pm 0.54)
Gubernaculum	-	-	7-8 (7.1 \pm 0.44)
Cloaca to SPI	-	-	11-13 (12.0 \pm 0.74)
SP 1 to SP 2	-	-	21-22.5 (21.9 \pm 0.54)
SP 2 to SP 3	-	-	52.5-57 (54.5 \pm 1.80)
Ant. gen. br.	104	64-88 (77.2 \pm 8.9)	-
Post gen. br.	108	80-114 (98.2 \pm 11.8)	-
a	13.8	13-18 (1.52 \pm 2.2)	13.8-18.0 (16.2 \pm 1.71)
b	4.4	3.5-5.3 (4.3 \pm 0.6)	3.4-5.1 (4.3 \pm 0.46)
v	55	51-56 (53.9 \pm 1.49)	-
T	-	-	52-58 (55.2 \pm 2.7)
G 1	50.9%	40.9-47.6 (44 \pm 2.3)	-
G 2	52.9%	52.3-59.0 (55.9 \pm 2.35)	-
Ant. end to EP/	167.4	142.5-163	151.1-182.8
Pharynx length (%)		(152.6 \pm 8.4)	(165.6 \pm 12.1)
Onchiostyle length/	93.0	86.9-95	88.5-98.2
Pharynx length (%)		(90.9 \pm 2.81)	(93.1 \pm 2.01)
Cloaca to SP 1/Spicule length (%)	-	-	29.2-39.4 (34.6 \pm 3.7)
Cloaca to SP 2/Spicule length (%)	-	-	75.4-108.1 (96.9 \pm 12.8)
Cloaca to SP 3/Spicule length (%)	-	-	256.7-294.7 (279.4 \pm 15.01)

except at the head, vulva and tail. Amphidial pouch vase shaped; amphidial aperture sublateral. Excretory pore at 49.6-60 μm , i.e. about 1.6-1.78 onchiostyle lengths, from the anterior end of the body. Pharyngeal bulb occupying 33.6-36.2 % of the neck length. Three pharyngeal gland nuclei present. Nucleus of dorsal gland at about 30 % of the enlargement from its anterior end, almost of the same size as those of the subventral glands which are behind the middle of the pharyngeal bulb. All females with anterior dorsal overlap by the intestine. Intestinal overlap 7.2-8.8 μm . Nerve ring enveloping isthmus, 40-56 μm from anterior end. Female reproductive system didelphic amphidelphic, ovaries reflexed, spermatheca present, sperm scattered throughout the length of the uteri, but occasionally they appear accumulated in an elongate oval spermatheca distally. Sperm 2-3 μm in diameter with rounded nucleus. Vulva pore as in ventral view. Vagina, 8-12 μm long extending inwards over, i.e. 27.5-37.5 % of corresponding body width, vaginal refractive thickenings inconspicuous, appearing as two small dots in lateral view, shape of vagina rounded in lateral view. Two lateral body pores at 24-76 and 92-137 μm posterior to vulva. Anus terminal, one pair of terminal caudal pores, tail bluntly rounded or hemispheroidal.

Male: General appearance similar to female, posterior end straight. Two distinct ventro-median cervical papillae (CP) present at 30.4-35.6 (32.8) μm and 47.2-53 (50.4) μm from anterior end of body. The anterior one lies near the level of the onchiostyle base and the posterior one 14.4-16.8 (15.4) μm behind it. Excretory pore 4-9 (7.7) μm behind the posterior cervical papillae and 49.6-60 μm , i.e. about 1.6-2.0 onchiostyle lengths, from the anterior end of the body. All males with a dorsal intestinal overlap 4-6.4 μm . Lateral cervical pores absent. Testis single, outstretched producing 2.4-3.2 μm wide rounded sperm with rounded nucleus. Three precloacal ventromedian supplementary papillae present: the posterior one (SPI) and median one (SP 2) lying at 29.2-39.4 % and 75.4-108.1 % respectively of the spiculum length, anterior to the cloacal aperture. The anterior supplement (SP 3) lies out of reach of the retracted spicules at 76-92 μm anterior to cloacal aperture. Spicules almost straight, with slight ventral curvature in distal third, marked with fine transverse striation in the middle. Gubernaculum with distal half thickened. Suspensor muscles elongate, oval, not conspicuous. Tail with one pair of post cloacal subventral papillae and one pair of subterminal pores. Tail enveloped by a bursa. Bursa reaching from posterior to the second ventromedian preanal papilla to the tail terminus.

TYPE HABITAT AND LOCALITY

Soil around the roots of guava (*Psidium guajava*), Faisalabad, Pakistan.

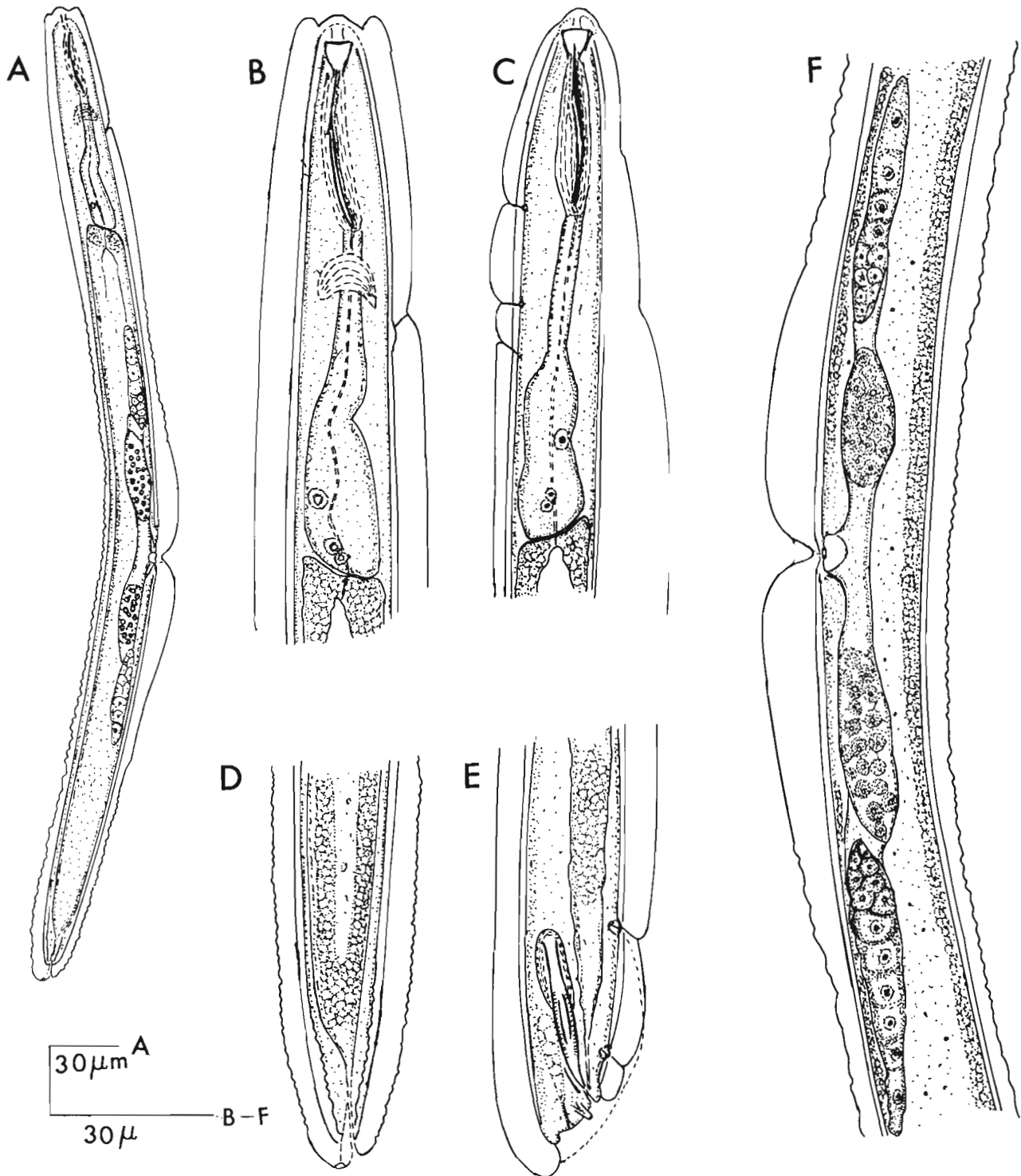


Fig. 1. *Paratrichodorus psidii* n. sp. A: Total view (holotype); B: Anterior part of female (holotype); C: Anterior part of male; D: Posterior part of female (holotype); E: Posterior part of male; F: Reproductive system (holotype).

OTHER LOCALITY AND HABITAT

Specimens have also been collected from soil around roots of citrus (*Citrus* sp.) at Faisalabad.

TYPE SPECIMENS

Holotype (female): Slide No.: NNRC-73/11 and *paratype* slides Nos. NNRC-73/12 - 27 (Eight females, five males) deposited in the National Nematode Collection of NNRC, University of Karachi, Karachi, Pakistan. Slide No. NNRC-73/28 (two females and one male) deposited in USDA Nematode collection, Beltsville, Maryland, USA.

DIAGNOSIS AND RELATIONSHIP

P. psidii n. sp. can be separated from all species in the genus by the combination of the following characters: Smaller spicules, gubernaculum and body length, number of lateral body pores posterior to vulva and shape of vulval sclerotizations in the female.

P. psidii n. sp. resembles *P. mirzai* (Siddiqi, 1960) Siddiqi, 1974 and *P. porosus* (Allen, 1957) Siddiqi, 1974. The new species comes close to *P. mirzai* in having males with same number and position of ventromedian supplements; in females with presence of caudal pores, more posterior vulva. It differs from *P. mirzai* in shape of vulva in ventral view, slightly shorter stylet, spicule and gubernaculum, and by the presence of two ventro-median cervical pore (vulva longitudinal slit, stylet 33-36 μm ; σ spicules = 32-34 μm ; gubernaculum = 11-13 μm ; three ventromedian cervical pore in *P. mirzai*). *P. psidii* n. sp. is also quite similar to *P. porosus* by its shape of vulva, in ratios "a" and "b". It differs from *P. porosus* in male having three ventromedian preanal papillae in contrast to two, number of ventromedian cervical pores is two as compared to one, smaller onchiostyle, spicules, and gubernaculum (onchiostyle = 43-50 μm ; spicules 36-39 μm ; gubernaculum = 12-13 μm in *P. porosus*).

***Paratrichodorus faisalabadensis* n. sp.**

(Figs 2 & 5)

MEASUREMENTS

See Table 2.

DESCRIPTION

Female: General appearance typical of Trichodoridae. Lip region with raised papillae. Amphids vase-shaped with aperture a sublateral lateral slit, cuticle only moderately swollen, 2.4-4 μm thick at midbody. Excretory pore at 56-76 (65.4) μm , i.e. 1.8-2.1 onchiostyle length, from the anterior end of the body. Pharyngeal bulb occupying 31-36.7% of the neck length. Dorsal nucleus and posterior ventro-sublateral nuclei prominent, the former one usually lies in the middle, the later ones in the posterior third of the pharyngeal bulb. Intestine of all females with anteriorly directed dorsal overlap of pharynx. Intestinal overlap 5.6-8.8 μm . Nerve

Table 2. Morphometric data of *Paratrichodorus faisalabadensis* n. sp. (all measurements are in μm except L.).

	Holotype (female)	Females (Paratypes)	Males (Paratypes)
n		5	5
L (mm)	0.65	0.54-0.65 (0.58 \pm 0.04)	0.52-0.66 (0.57 \pm 0.02)
Body diam	28.8	27-32 (29.2 \pm 1.34)	27.2-32 (29.2 \pm 2.2)
Pharynx	37.6	33.6-37.6 (36.4 \pm 1.29)	35.2-36.8 (35.9 \pm 0.65)
Onchiostyle	34	30-34.4 (33.3 \pm 1.05)	32.8-35.2 (34.3 \pm 1.33)
Ant. end to excret. pore	72	56-76 (65.4 \pm 6.24)	65.6-68.0 (66.6 \pm 1.0)
Ant. end to CP 1	-	-	34-39.2 (37 \pm 2.7)
Spicule	-	-	30-33.6 (31.9 \pm 1.81)
Gubernaculum	-	-	7.0-8.8 (7.9 \pm 0.9)
Cloaca to SPI	-	-	11.2-13 (11.85 \pm 1.03)
SP 1 to SP 2	-	-	23-26 (25 \pm 0.5)
SP 2 to SP 3	-	-	60-70 (63.75 \pm 5.0)
Ant. gen. br.	109.6	72-109.6 (95 \pm 12.1)	-
Post gen. br.	116.0	93.6-112 (108.4 \pm 7.32)	-
a	21	18.6-23.3 (19.7 \pm 1.82)	16.8-21.2 (19.4 \pm 1.92)
b	4.9	4.2-5.4 (4.8 \pm 0.55)	4.7-5.6 (5.1 \pm 0.20)
v	54	51-54 (52.7 \pm 1.42)	-
T	-	-	54-61.3 (57.1 \pm 3.26)
G 1	48.5	41.6-50.7 (46.4 \pm 3.37)	-
G 2	51.4	49.2-60.5 (53.3 \pm 3.3)	-
Ant. end to EP/	187.2	148.9-193.3 (165.6 \pm 14.8)	180-193.1 (186.6 \pm 5.3)
Pharynx length (%)			
Onchiostyle length/	91.4	89.3-95.3 (85.1 \pm 2.02)	90.1-95.4 (92.6 \pm 2.01)
Pharynx length (%)			
Cloaca to SP 1/Spicule length (%)	-	-	35.7-38.4 (37.2 \pm 1.27)
Cloaca to SP 2/Spicule length (%)	-	-	107.1-120.5 (113.5 \pm 5.92)
Cloaca to SP 3/Spicule length (%)	-	-	295.2-337.5 (310.6 \pm 17.7)

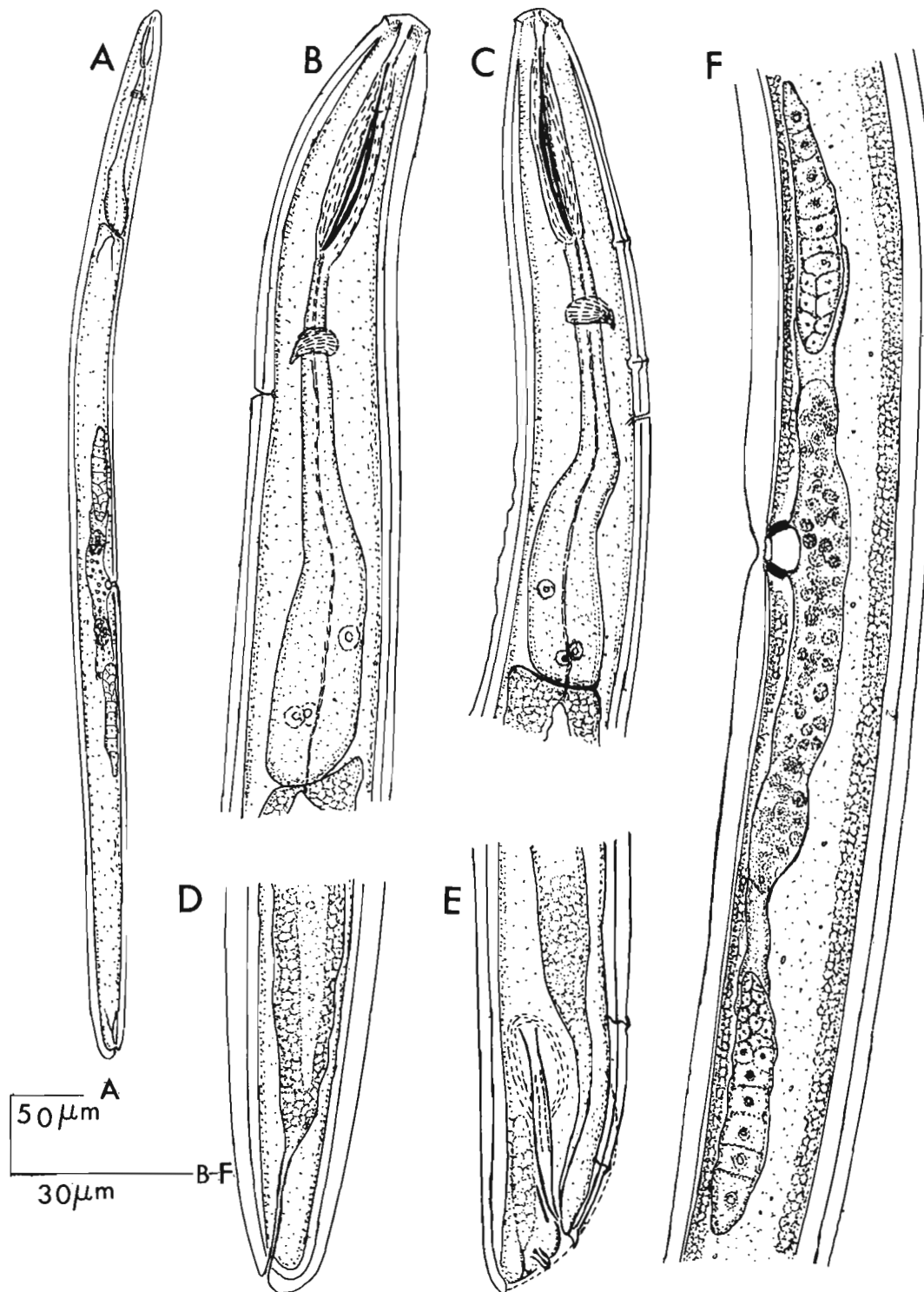


Fig. 2. *Paratrichodorus faisalabadensis* n. sp. A: Total view (holotype); B: Anterior part of female; C: Anterior part of male; D: Posterior part of female; E: Posterior part of male; F: Reproductive system.

ring at the level of the anterior part of the isthmus, 45.6-53.6 μm from the anterior end. Female reproductive system didelphic, amphidelphic. Ovaries reflexed, spermatheca present filled with rounded sperm. Vaginal sclerotization roundly triangular in lateral view with two short rods in optical section. Vagina 7.2-8.8 (7.9) μm , extending inwards over 23.6-29.7% (27.1) of the corresponding body width. Vulva opening a pore in ventral view. Sperm scattered throughout the length of the uteri. Anus subterminal. One pair of subventral subterminal caudal pores present.

Male: General appearance typical of the genus. Lip region with distinct labial papillae. Amphidial aperture sublateral. Cuticle slightly to clearly swollen after fixation 2.4-3.2 μm thick at mid body. Pharynx gradually widening posteriorly to a relative slender bulb with the dorsal gland nucleus situated at mid bulb to posterior half of bulb, at level of or just anterior to the posterior ventro-sublateral gland nuclei. Pharyngeal bulb with a short dorsal intestinal overlap. Nerve ring at level of anterior part of isthmus. Two ventromedian cervical papillae (CP) present at 34-39.2 (37) μm and 49.2-57.6 (52.8) μm from anterior end of body. The anterior one lies near the level of the onchiostyle base and the posterior one 15.5-18.4 (16.8) μm behind it. Excretory pore 11.2-17.4 (13.5) μm behind the posterior cervical papillae. Testis single outstretched, granular sperm cells with rounded nucleus. Three precloacal ventromedian supplements present. The posterior one (SP 1) and median one (SP 2) lying at 35.7-38.4% and 107-120.5% respectively, of the spiculum length anterior to the cloacal aperture, (SP 1) is within the reach of the retracted spicules; the anterior supplement (SP 3) lying out of reach of the retracted spicules, at 99.2-108 (100.2) μm anterior to the cloacal aperture. Spicules curved 30-33.6 (31) μm , gubernaculum 7-8.8 (7.9) μm thickened at its distal half. Anterior lip of cloaca with finger like projection. Tail with one pair of post cloacal subventral papillae and one pair of subterminal pores. Tail enveloped by a bursa. Bursa arises opposite middle of spicules to the tail terminus.

TYPE HABITAT AND LOCALITY

Specimens were collected from the rhizospheres of guava (*Psidium guajava*) from Faisalabad in 1993.

TYPE SPECIMENS

Holotype (female) : Slide No. NNRC-73/28 and **paratype** slide Nos. NNRC-73/28-39 (thirteen females, four males) deposited in the National Nematode collection of NNRC, University of Karachi, Karachi, Pakistan. Slide No. NNRC-73/40 (two females and one male) deposited in USDA Nematode collection, Beltsville, Maryland, USA.

DIAGNOSIS AND RELATIONSHIPS

Paratrichodorus faisalabadensis n. sp., can be separated from all species in the genus by the combination of the following characters : Shape of vagina and vaginal sclerotization, smaller spicules and gubernaculum.

P. faisalabadensis n. sp. resembles *P. mirzai* (Siddiqi, 1960) Siddiqi, 1974, *P. porosus* (Allen, 1957) Siddiqi, 1974 and *P. psidiumi* n. sp. The new species comes close to *P. mirzai* in body length, onchiostyle and spicule length, and in number of ventromedian, preanal papillae. It differs from *P. mirzai* in the male by the possession of two ventromedian cervical papillae *vs* three ventromedian cervical papillae, shorter gubernaculum 7-8.8 *vs* 11-13 μm ; in female, by a pore like vulva in ventral view *vs* longitudinal slit. It is also quite similar to *P. porosus* by having pore like vulva, in body length, in ratios "a" and "b". It differs from *P. porosus* in male having three preanal papillae in contrast to two, number of ventromedian cervical pores is two as compared to one, smaller onchiostyle, spicules, and gubernaculum. (onchiostyle = 43-50 μm ; spicules = 36-39 μm ; gubernaculum 12-13 μm in *P. porosus*). The new species also resembles *P. psidiumi* n. sp. in female, by the shape of vulva in lateral view, in body diameter, in onchiostyle length; in male, in spicules and gubernaculum length, in distance from cloaca to SP 1. It differs from *P. psidiumi* n. sp. in having slightly longer body; distance between oral aperture to excretory pore, in the shape of vaginal sclerotization, in the shape of spicules (L = 0.40-0.52 mm; oral aperture to excretory pore = 49-60 μm ; vagina rounded in lateral view; spicules almost straight with fine transverse striation in *P. psidiumi*).

***Paratrichodorus mirzai* (Siddiqi, 1960) Siddiqi, 1974 (Fig. 3)**

Measurements are given in Table 3.

These specimens – collected from soil around the roots of sugarcane (*Saccharum officinarum*) from Thatta, Sindh, Pakistan – are in close agreement with the description of Siddiqi (1962), except that they have slightly shorter spicules and onchiostyle, more over in one female specimen two vulva have been observed which is unusual observation in this species so far (Fig. 3 F). There are also some differences in position of ventromedian cervical papillae, CP 1 located at the level of onchiostyle base whereas it is slightly behind onchiostyle base (Siddiqi, 1962).

***Paratrichodorus renifer* Siddiqi, 1974 (Fig. 4)**

Measurements are given in Table 3.

These specimens – collected from soil around the roots of pine trees (*Pinus* spp.) at Murree and also found among the roots of grass (*Cynodon dactylon*) at Karachi,

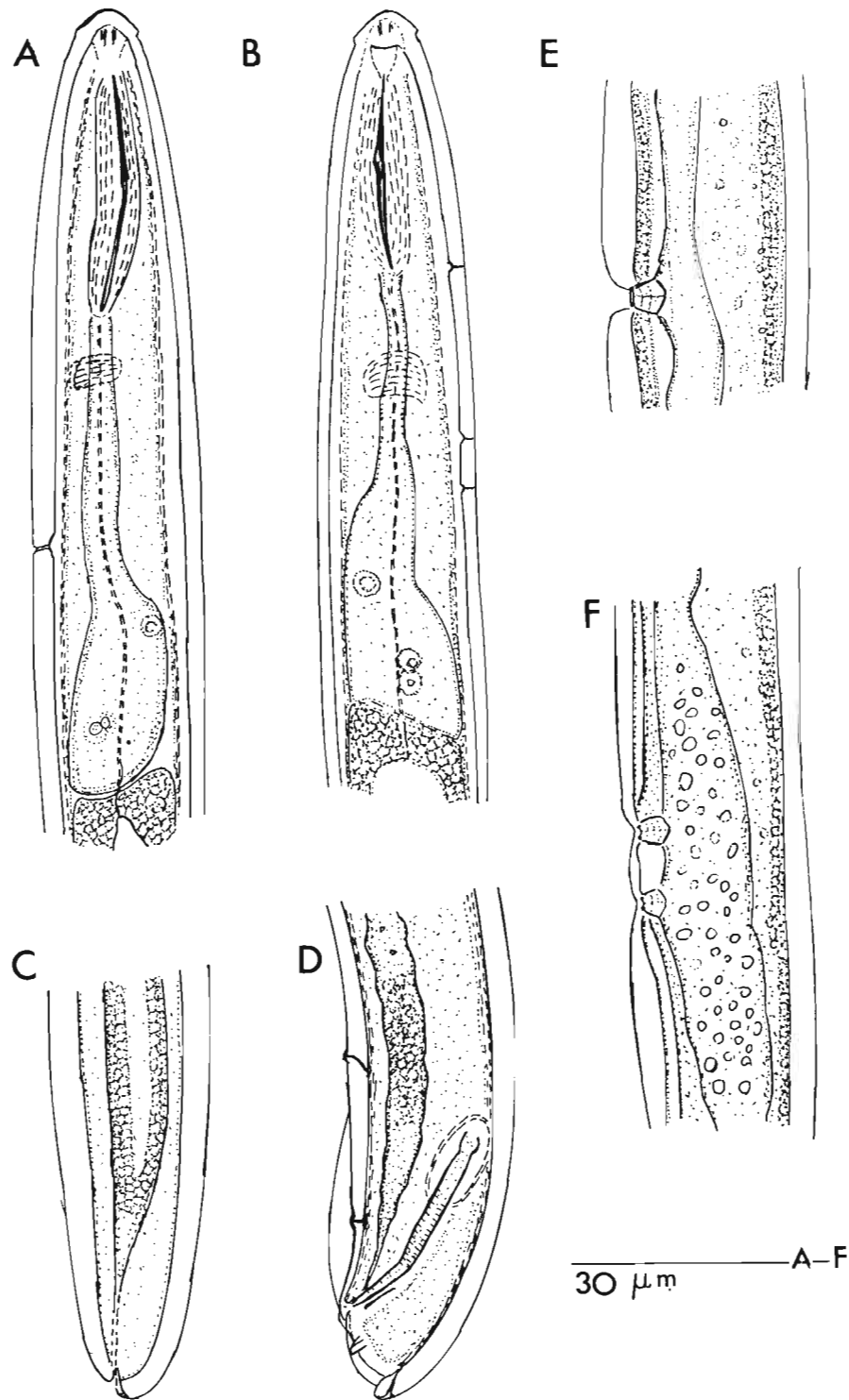


Fig. 3. *Paratrichodorus mirzai* (Siddiqi, 1960) Siddiqi, 1974. *A*: Anterior part of female; *B*: Anterior part of male; *C*: Posterior part of female; *D*: Posterior part of male; *E*: Vulva area; *F*: Bivulval area.

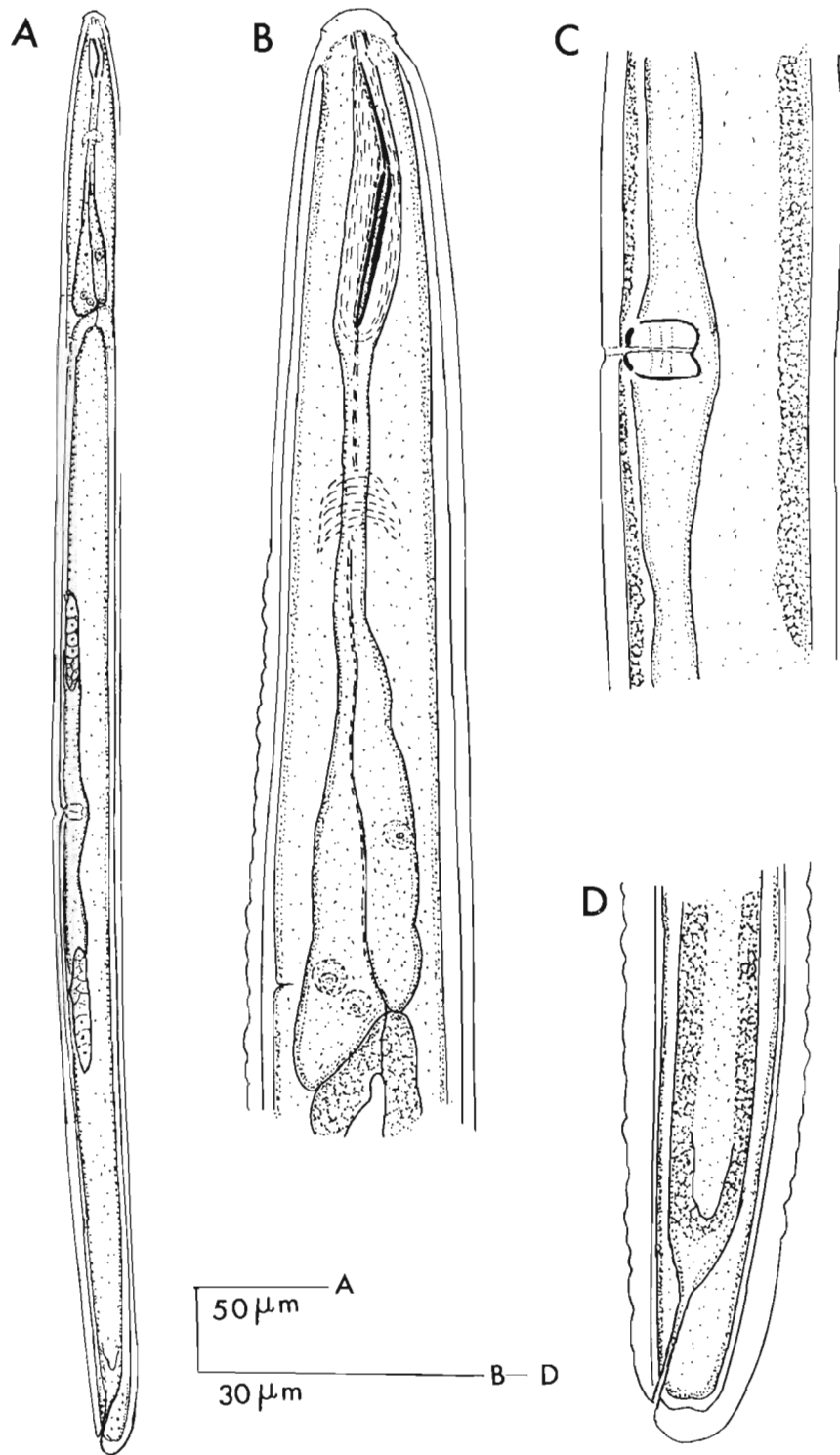


Fig. 4. *Paratrichodorus renifer* Siddiqi, 1974. A : Total view; B : Anterior part of female; C : Vulva area; D : Posterior part of female.

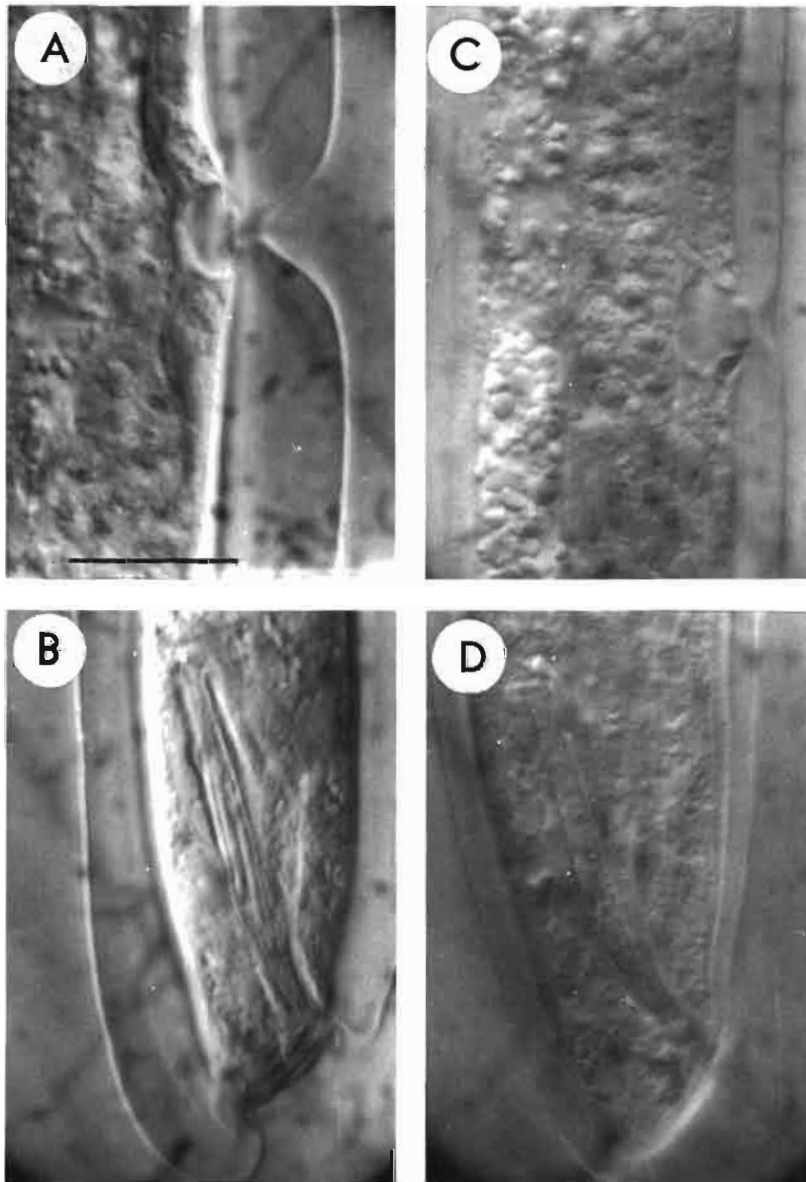


Fig. 5. *Paratrichodorus psidii* n. sp. *A*: Vulval area; *B*: Posterior part of male – *Paratrichodorus faisalabadensis* n. sp. *C*: Vulval area; *D*: Posterior part of male. (Bar on A – D = 15 μ m.)

Pakistan – agree with the original description of *P. renifer* given by Siddiqi (1974) except that the excretory pore 57.6 μ m from anterior end *versus* 83–90 μ m. Oesoph-

ageal gland overlaps the intestine ventrally versus the oesophageal enlargement with the flat base, offset from intestine.

Table 3. Biometrical data of *Paratrichodorus mirzai* (Siddiqi, 1960) Siddiqi, 1974 and *P. renifer* Siddiqi, 1974.

	<i>P. mirzai</i>				<i>P. renifer</i> Siddiqi, 1974	
	Pakistani specimens		(Siddiqi, 1962)		Pakistani specimens	(Siddiqi, 1974)
	♀ (n = 15)	♂ (n = 10)	♀ (n = 8)	♂ (n = 6)	♀ (n = 3)	♀ (n = 20)
L	0.59-0.66	0.58-	0.52-0.6	0.45-0.63	0.53-0.58	0.42-0.56
a	21.8-23.7	20-22.9	18-20	17-20	20.1-21.5	20-25
b	5.5-6.3	5.9-	5-5.9	4.8-5.8	4.7-5.2	4.8-6.9
v	53.2-56	-	54-58	-	55-56	55-59
T	-	57.2-60	-	56-70	-	-
Onchiostyle	31-32	30-32	33-37	33-36	30-31.2	31-34
excretory pore						
to front end	67.2-69	70-78.4	-	-	57.6-57.6	83-90
Spicules	-	29.6-32	-	32-34	-	-
Gubernaculum	-	8-10	-	11-13	-	-
Vulva shape	Longitudinal slit		Longitudinal slit	-	Transverse slit	Transverse slit

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