

DESCRIPTION OF TWO NEW SPECIES - *HELICOTYLENCHUS SIDDIQII* N.SP. AND *PARATYLENCHUS (PARATYLENCHUS) KARACHIENSIS* N. SP. (NEMATODA: TYLENCHIDA) FROM SINDH, PAKISTAN

Zarina Begum and Shaheen Akhter

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

ABSTRACT

Two new species, *Helicotylenchus siddiqii* n.sp. and *Paratylenchus (Paratylenchus) karachiensis* n.sp. are isolated from chilli plants cultivated in Sindh, Pakistan and described and illustrated here. *Helicotylenchus siddiqii* n. sp. comes close to *H. abunaami* Siddiqi, 1972; *H. craigi*, Knobloch and Laughlin, 1973; *H. densibullatus*, Siddiqi, 1972 and *H. gerti*, Marias *et al.*, 2005 on the basis of head rounded with three indistinct annules, stylet length and basal knobs anteriorly flattened and concave, mucronate tail and male absent on many of the morphological characters. *Paratylenchus (P.) karachiensis* n. sp. comes close to *P. (P.) bukowinensis* Micoletzky, 1922, *P. (P.) curvatus* Van der Linde, 1938, *P. (P.) dianthus* Jenkins and Taylor, 1956, *P. (P.) halophilus* Wouts, 1966, *P. (P.) projectus* Jenkins and Taylor, 1956, *P. (P.) rotundicephalus* Bajaj, 1987 on the base of stylet length, four lateral incisures and vulval flap. These species are new to science for the world.

Key-words: Chilli plants, *Helicotylenchus siddiqi* sp. n., *Paratylenchus (Paratylenchus) karachiensis* sp. n., Sindh, Pakistan.

INTRODUCTION

Chilli is economically important and valuable crop. New Mexico is thought to be the native home of chillies. In Kunri, a small town of Umer Kot district of Sindh is the home of Red Chillies. It contributes around 85% of Pakistan Red Chilli production and is known as one of the largest production centers for Red Chillies in Asia. Various plant parasitic nematodes cause damage to chilli plants all over the world (Adamou *et al.*, 2013) and in Pakistan several plant parasitic nematodes have been found associated with this valuable crop and reduce its cash value (Zarina, 1996; Khan *et al.*, 2000, 2006; Anwar and McKenry, 2010; Anwar *et al.*, 2013). Fifty two species of nematodes have been reported from chillies of Sindh recently by Zarina *et al.* (2015). The genus *Heicotylenchus* included *Helicotylenchus digonicus*, *H. dihystra*, *H. indicus*, *H. mucronatus*, *H. multicinctus*, *H. pseudorobustus* and *Paratylenchus* species (*Pratylenchus penetrans*, etc.). *Helicotylenchus siddiqii* n. sp. and *Paratylenchus (P.) karachiensis* n. sp. were isolated around chilli plants and described for the first time. These species are described in detail in this paper.

MATERIALS AND METHODS

During a survey of nematodes belonging to the genus *Helicotylenchus* and *Paratylenchus (Paratylenchus)* were collected around the roots of chilli (*Capsicum annuum* L.) from Malir, Karachi. Nematodes were processed by the following procedure given by Siddiqi (2000) and Southy (1986). Specimens were separated from soil by modified Baermann funnel method (Schindler, 1961), and processed by following procedure given by Siddiqi (2000) and Southy (1986). Processed specimens were mounted in absolute glycerin. Measurements and observations were made by using an ocular micrometer. Drawings were made by means of drawing tube.

***Helicotylenchus siddiqii* n. sp.**
(Table 1) (Fig. 1, A-H); (Plate 1 A-F)

Measurements: (Table 1)

Description Female: Body spirally curved when relaxed by gentle heat. Cuticle coarsely striated annules 1.05- 1.58µm wide at mid body. Lateral field occupying 1/4th of body width, marked by four smooth incisures, inner incisures not fused in posterior part of tail. Head rounded, with inconspicuous annules 5.67-6.93 µm high, 2.52-3.78 µm wide, continuous with body contour, cephalic framework well developed. Stylet well developed 22.68- 23.94 µm long with anteriorly flattened and concave knobs 3.15- 4.41 µm long and 1.26-1.89µm wide, conus 11.43- 12.6 µm long and 51.35 - 52.77% of the total stylet length. Conus slightly longer than shaft, m ratio 49.53-

55.55, and O ratio 26.31-30.42. Dorsal oesophageal gland 6.3-6.93 μm long behind stylet base. Procorpus elongated with median bulb round to ovate 10.08-12.6 x 6.3-7.56 μm with well developed with muscular vulvalur apparatus. Isthmus encircled by nerve ring in the middle. Oesophageal gland overlapping the intestine ventro-laterally, oesophageal length 113.40 - 176.0 μm from anterior end. Hemizonid 2-3 annules anterior to Excretory pore. Ex. pore posterior to nerve ring 87.0- 100.80 μm from head, close to level with oesophageal intestinal junction. Vulva with transverse slit extending about half into the body at right angle to the body axis, gonad paired and outstretched. Ovaries with a single row of oocytes, anterior ovary 155.55- 199 μm long slightly longer than the posterior ovary, 105- 130.66 μm long, spermatheca round to oval slightly setoff, usually filled with sperms. Intestine not extending over rectum. Phasmids distinct 2 annules posterior, 3-5 annules anterior to level of anus. Tail cylindrical from anus to tip dorsally, convex conoid and variable tip shaped, 10-13 annules with broad annulated ventral projection.

Table 1. Morphometrics of *Helicotylenchussiddiqii* n.sp. All measurements in μm except body length.

Characters	Holotype female	Paratype females	Paratype juveniles
n	1♀	14♀♀	5jj
L	0.446	(0.43-0.59)0.493±0.39	(0.331-0.376)0.35±0.016
a	25.28	(23.64-29.78)25.421±2.08	(19.03-21.36) 20.17±0.80
b	4.75	(5.01-6.32)5.297±0.39	(3.69-4.3)4.04±0.313
b'	3.83	(3.5-4.62) 4.151±0.26	(3.04-3.52)3.19±0.192
c	33.71	(33.71-46.66) 40.895 ±4.12	(26.28-28.55)26.99±0.94
c'	1.31	(1.05-1.375) 1.194 ±0.10	(1.18-1.29)1.22±0.046
stylet	23.31	(21.42-23.94) 23.19 ±0.78	(21.42-22.68)21.92±0.53
stylet conus	11.97	(10.61-11.97) 11.43 ± 0.48	(10.5-11.3)10.94±0.40
m	51.35	(49.53-55.55) 50.26 ±2.44	(49.01-50.79)49.64±0.48
O	29.73	(26.31-30.42) 27.79± 1.56	(27.28-31.43)29.56±1.83
V	55.51	(45.77-57.5)51.73±0.33	-
DGO	6.93	(5.67-6.3) 6.057±0.31	(6.3-6.93)6.55±0.34
Body annules (μm)	1.26	(1.05-1.58) 1.31±0.21	-
Lip H. (μm)	6.3	(5.67-6.93)6.43±0.44	(5-5.7)5.35±0.4
Lip .W (μm)	3.15	(2.52-3.78) 3.10 ± 0.49	(2-2.2)2.1±0.1
Lateral incisures	4	4	4
Metacarpus-lip (μm)	69.3	(69.3-77.49) 74.83 ± 2.17	-
Metacarpus L.	10.71	(10.08-12.6) 11.45± 0.88	(9.2-10.9)10.22±0.93
Metacarpus W.	6.3	(6.3-7.56) 6.57 ± 0.69	(5.6-6.9)6.05±0.49
Oesophageal gland-lip	116.55	(113.40-176) 121.23 ± 11.09	(103.95-118.44)111±5.64
Excretory pore	89.41	(87-103.95) 95.83± 6.19	(85.05-91.98)87.95±3.07
ABW	10.08	(10.08-13.23) 11.15 ± 0.193	(10.71-11.34)10.84±0.28
Tail annuls	12	(10-13)	(10-12)

Type habitat and Locality: Specimens were collected from soil around the roots of (*Capsicum annuum* L.) chilies from Malir, Mammon Goth Malir, and New Karachi, Pakistan.

Type Specimens: Holotype slide No NNRC-68/105 and paratypes on slide No NNRC 18/70-73 (13 females and 5 juveniles) deposited in the National Nematode Collection of NNRC, University of Karachi, Pakistan.

Diagnosis and Relationship: *Helicotylenchus siddiqii* n. sp. comes close to *H. abunaami* Siddiqi, 1972; *H. craigi*, Knobloch and Laughlin, 1973; *H. densibullatus*, Siddiqi, 1972 and *H. gerti*, Marias *et al.*, 2005 on the basis of head rounded with three indistinct annules, stylet length less than 24 μm , basal knobs anteriorly flattened and concave, mucronate tail and male absent on many of the morphological characters.

It differs from *H. abunaami* by having smaller body length, longer stylet length, larger c and V value (L= 0.52-0.63mm; stylet = 21.0-22.0 μm , V= 59.0-65.0, c = 23.0- 44.0 in *H. abunaami* after Siddiqi, 1972).

It can be differentiated from *H. craigi* by having smaller body size, larger c value and V value and phasmids anterior to anus. (L= 0.52 - 0.65mm, c= 0.23 - 0.33, V= 56.0 - 61.0, head with 4 annules, phasmids located anterior to anus *H. craigi* after Knobloch & Laughlin, 1973).

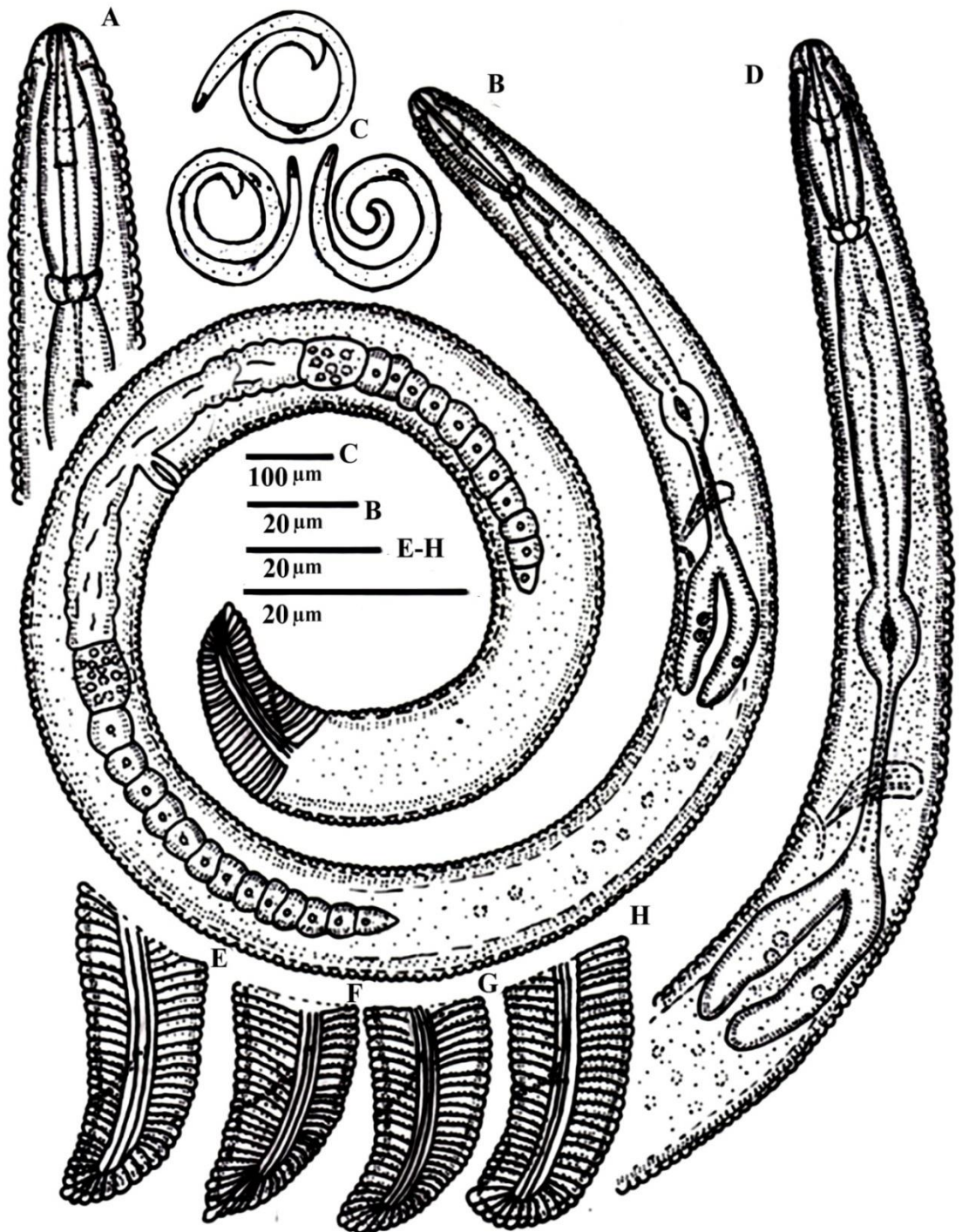


Fig. 1 (A-H). *Helicotylenchus siddiqii* n.sp. A. Head region; B. Entire body; C. Posture of adults; D. Oesophageal region; E-H. Tail end.

Male: Not found.

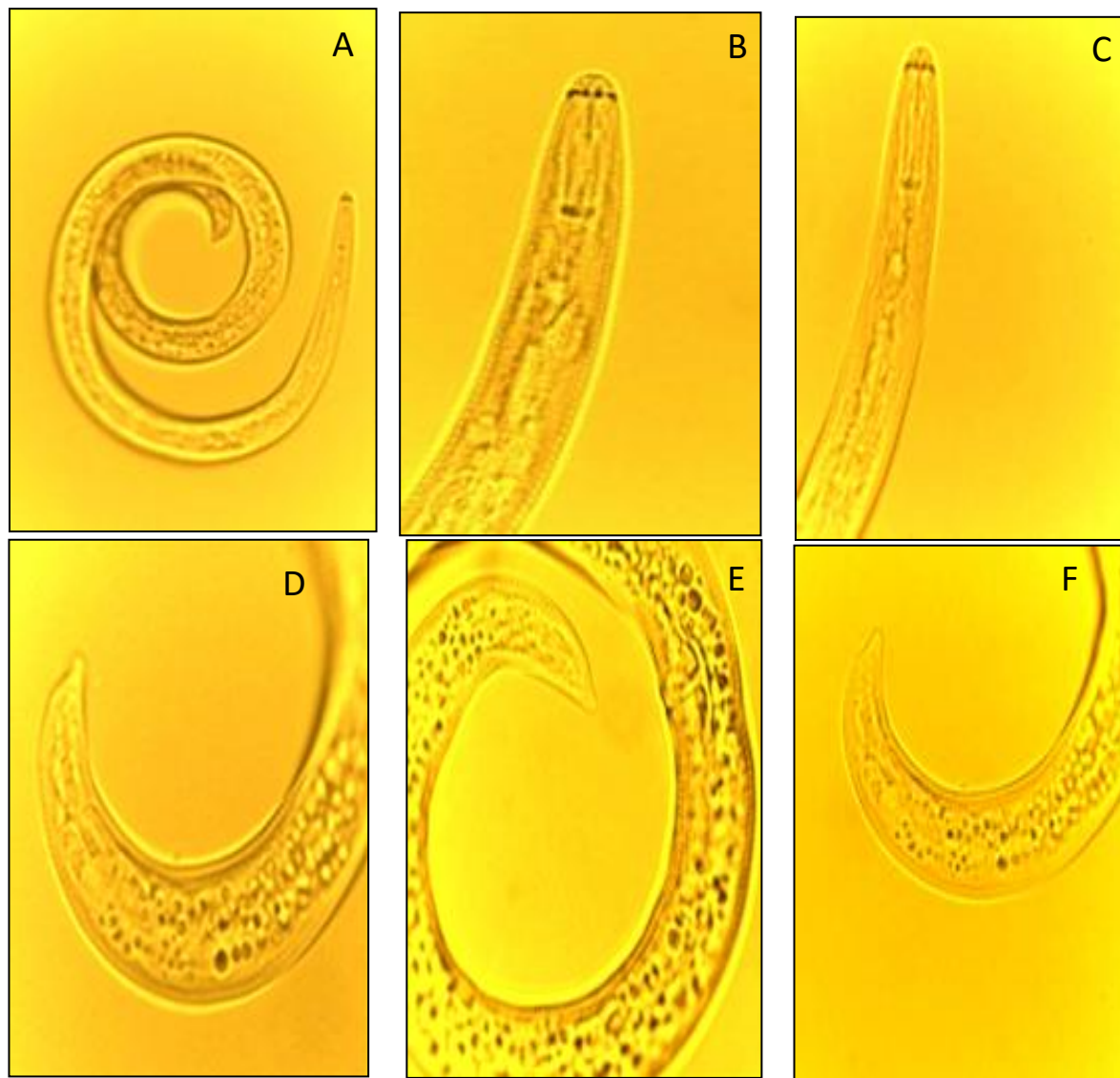


Plate 1. (A-F): *Helicotylenchus siddiqii*.n.sp. A. Entire female; B. Head region; C. Oesophageal region; D & F. Tail; E. Vulval region .

It can be distinguished from *H. densibullatus* by having larger body size, V value, stylet length and smaller c value (L = 0.43- 0.59mm, c = 41.0-51.0, V = 62.0-65.0, Stylet = 22.0 - 23.0 μ m in *H. densibullatus* after Siddiqi, 1972).

It can be differentiated from *H. gerti* by having smaller body size, c value, longer stylet, larger a, c, m, V values and Ex. pore length. (L= 0.527 - 0.694mm, stylet = 22.0 - 24.0 μ m, a = 24.9 - 288.7, c = 32.7 -44.6, c' = 1.1 - 1.8, m = 43.0 - 48.0, V = 63.0 - 66.0, Ex. pore = 99.0 - 100.0 μ m *H. gerti* after Marias et al., 2005).

***Paratylenchus (Paratylenchus) karachiensis* n.sp.**

(Table 2) (Fig. 2, A-D)

Measurement: Table 2

Holotype female: L = 0.306 mm; a = 25.49; b = 55; c=15.25; c' = 1.67; V =82.50; Stylet = 26.4 μ m

Females Description : Body cylindrical, tapering at both extremities, ventrally curved, slightly to close 'C' shaped posteriorly when relaxed by gentle heat (Fig.15 C), maximum body width 36.75-38.0 μ m , cuticle annulated, annules about 1-3 μ m wide at mid body. Lateral field 4 μ m wide with four incisures. The cephalic region

truncate, conoid continuous with the body contour 4.5-5.0 μm wide, 7.0-10 μm high with 3-4 annules. Cephalic frame work weakly sclerotized, stylet slender 25.6-27.2 μm long conus longer than shaft, knobs rounded 3.75 -4.0 μm across (Fig. 15 B). Orifice of dorsal oesophageal gland 12.0-13.0 μm behind the spear base. Median oesophageal bulb with elongated bulb with elongated valvular apparatus, narrow isthmus, crossed by nerve ring, basal bulb pyriform, 17-18 μm wide. Oesophago-intestinal valve small oblong, Hemizonid 2-3 annules long, 0-1 annules anterior to Excretory pore 60-65.6 μm from anterior end of the body and located opposite to the basal bulb region. Vulval flap present, inconspicuous, spermatheca not distinct. Female reproductive system prodelphic, ovary outstretched, comprised of a single row of oocytes. Anus indistinct, tail 19.2-25.6 μm long, conoid with round to pointed terminus.

Table 2. Comparative morphometric data of species of *Paratylenchus* (*Paratylenchus*) sp. having small stylet.

Morphological characters	<i>P. (P.) karachiensis</i> n.sp.	<i>P. (P.) bukowinensis</i>	<i>P. (P.) curvatus</i>	<i>P. (P.) dianthus</i>	<i>P. (P.) halophilus</i>	<i>P. (P.) Projectus</i>	<i>P. (P.) rotundicephalus</i>
L	(0.305-0.341) 0.323 $\pm$ 0.15	0.390	0.330	(0.32-0.44) 0.37	0.320-0.390	(0.289-0.475) 0.373	(0.306-0.376) 0.345 $\pm$ 16.8
a	(22.45-25.49) 23.97 $\pm$ 3.35	20.5	23.8	(18.0-25.8) 21.7	19-25	(15.6-20.8) 18.7	(23-27) 2.4 $\pm$ 1.1
b	(4.25-4.58) 4.42 $\pm$ 0.55	4.3	3.7	(3.5-4.8) 4.2	4.1-4.6	(3.5-4.9) 4.0	(3.8-4.6) 4.15 $\pm$ 0.27
c	(15.25-20.22) 17.74 $\pm$ 2.24	16.8	16.4	12.7	11-14	(11.3-19.5) 15.4	(12-14) 12.9 $\pm$ 0.7
c'	(1.7-2.2) 1.96 $\pm$ 3.2	-	-	-	-	-	(2.7-3.6) 3.14 $\pm$ 0.34
V	(78.18-86.09) 82.1 $\pm$ 9.5	84	80.6-87	(80.5-87.1) 84.0	76-80	(82.6-86.9) 84.3	(81-84) 82.1 $\pm$ 1.1
Stylet	(25.6-27.2) 26.4 $\pm$ 2.04	24	22-25	(20.6-28.9) 25.8	26-30	(24.8-37.1) 31.8	(26-27) 26.7 $\pm$ 0.8
Ex. pore	(60.6-66.0) 62.8 $\pm$ 1.23	-	-	-	-	-	66-77

Male: Not found.

Type habitat and locality: Specimens were collected from soil around the roots of chili (*Capsicum annum* L.) from Malir, Karachi, and Sindh, Pakistan.

Diagnosis and relationship: *Paratylenchus* (*P.*) *karachiensis* n. sp. comes close to *P. (P.) bukowinensis* Micoletzky, 1922, *P. (P.) curvatus* Van der Linde, 1938, *P. (P.) dianthus* Jenkins and Taylor, 1956, *P. (P.) halophilus* Wouts, 1966, *P. (P.) projectus* Jenkins and Taylor, 1956, *P. (P.) rotundicephalus* Bajaj, 1987 on the base of stylet length, four lateral incisures and vulval flap.

It differs from *P. (P.) bukowinensis* by having smaller body size, 'a' value and longer stylet length (L = 0.390 mm; a = 20.5, stylet = 24 μm *P. (P.) bukowinensis* Micoletzky, 1922).

It can be differs from *P. (P.) curvatus* by longer stylet length, oesophagus length, smaller 'V' value (L = 0.330 mm, b=3.7, V=80.0 -87.0, stylet =22.25 μm in females of *P. (P.) curvatus* after Van der Linde, 1938).

It can be differentiated from *P. (P.) dianthus* by having smaller body length, greater 'c' value longer stylet length and absence of male (L=0.32-0.44 mm, c =11-13, stylet = 20.6-28.9 μm male present in *P. (P.) dianthus* after Jenkins and Taylor, 1956).

It can be distinguished from *P. (P.) halophilus* by having a smaller body length, greater 'C', V value and smaller stylet length, male absent (L = 0.320-0.390 mm; c = 11-14, V=76-80, stylet =26-30 μm male present in *P. (P.) halophilus* after Wouts, 1966).

It differs from *P. (P.) projectus* by having smaller body length, b, c, v, values and stylet length and greater 'a' value ($L = 0.289 - 0.475$ mm, $a = 15.6 - 20.8$, $b = 3.5 - 4.9$, $c = 11.3 - 19.5$, $v = 82.6 - 86.9$, stylet = $24.8 - 37.1$ μ m, in *P. (P.) projectus* after Jenkins, 1956).

It can be distinguished from *P. (P.) rotundicephalus* by having smaller body length, a, c, c' and larger 'V' values, absence of male ($L = 0.306 - 0.376$ mm, $a = 23 - 27$, $c = 12 - 14$, $c' = 2.7 - 3.6$, $V = 81 - 84$, male present in *P. (P.) rotundicephalus* after Bajaj, 1987).

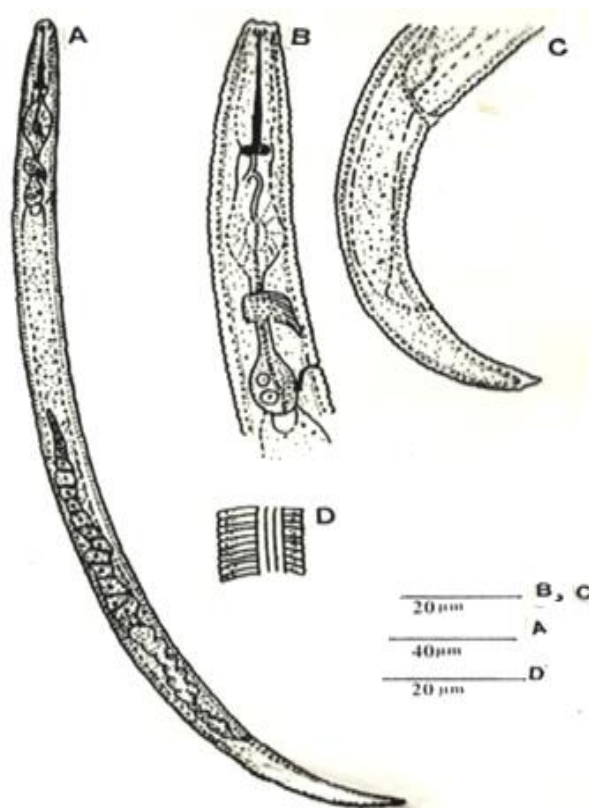


Fig. 2 (A-D). *Paratylenchus (P.) karachiensis* n.sp. A. Entire Female ; B. Oesophageal region; C. Female tail ; D. Lateral view at mid body.

Acknowledgement

We sincerely acknowledge and thank Prof. Dr. D. Khan and Prof. Dr. M. Javed Zaki for critical review of manuscript and valuable suggestions.

REFERENCES

- Adamou, H., B. Adamou, A.H. Omarou, S.R.D. Seyni and E. A. Toudou (2013). Characterization of plant-parasitic nematode communities associated with tomato, eggplant and pepper in the suburban area of Niamey (Niger). *Int. J. Agricult. And crop Sciences* 5(20): 2488-2494.
- Anwar, S. A. and M. V. McKenry (2010). Incidence and reproduction of *Meloidogyne incognita* on vegetable crop genotypes. *Pak. J. Zool.*, 42: 135-141.
- Anwar, S.A., M.M. Mehdi, M.V. McKenry and A. Qadir (2013). Survey of plant protection nematodes with four vegetable as crops cultivated within tunnels. *Pak. J. Zool.* 45 (3): 595-603.
- Bajaj, H. K. (1987). On the species of *Paratylenchus*, Micoletzky (Nematoda: Criconematina) from Haryana, India. *Indian Journal of Nematology*, 17:318-326.
- Brown, K. F. (1962). *A survey of some plant parasitic nematodes problem in Pakistan*. Shell Int. Chem. Co. Ltd. pp.9.
- Jenkins, W. R. (1956). *Paratylenchus projectus* new species (Nematoda, Criconematidae) with a key to the species of *Paratylenchus*. *J. Washington Academy of Science*, 46: 296-298.

- Jenkins, W. R. and D. P. Taylor (1956). *Paratylenchus dianthus* n. sp. (Nematoda, Criconematidae) a parasite of carnation. *Proceedings of Helminthological Society of Washington*, 23:124-127.
- Khan, A., K. A. Khanzada, S. S. Shaukat and S. Shahzad (2006). Nematodes associated with chili in Sindh. *Pak. J. Nematol.*, 24: 145-150.
- Khan, A., S. S. Shaukat and I. Ahmad (2000). Nematode communities associated with chillies in lower Sindh, Pakistan. *Pak. J. Biol. Sci.*, 3: 457- 458.
- Knobloch, N. A. and C. W. Laughlin (1973). A collection of plant parasitic nematodes (Nematoda) from Mexico with description of three new species. *Nematologica*, 19: 205-217.
- Marais, M. T., Mekete and L. R. Tiedt (2005). Description of *Helicotylenchus gerti* sp. n. and new records of some species of *Helicotylenchus* (Steiner, 1945) from Ethiopia (Nematoda: Hoplolaimidae). *Journal of Nematode Morphological and Systematics*, 8: 39-42
- Micoletzky, H. (1922). Die freilebenden Erd-Nematodes mit besonderer Berücksichtigung der Steiermark und der Bukowina, zugleich mit einer Revision Beschreibung und Bestimmungsschlüsse. *Archiv für Naturgeschichte Berlin*, 87: 321-650.
- Schindler, A. F. (1961). A simple substitute for Baermann funnel. *Plant Disease Reporter*, 45: 747-748.
- Siddiqi, M. R. (1972). On the genus *Helicotylenchus* Steiner, 1945 (Nematoda: Tylenchida), with the description of nine species. *Nematologica*, 18: 74-91.
- Siddiqi, M. R. (2000). *Tylenchida, Parasites of Plants and Insects*. (2nd Edition) Commonwealth Agricultural Bureaux, Slough, United Kingdom 833 pp.
- Southy, J.F. (Ed.) (1986). *Laboratory Methods for Work with plant and soil nematodes*. Technical Bulletin, Ministry of Agriculture, Fisheries and Food Reference Book, 402. Her Majesty's Stationary Office, London.
- Van der Linde, W. J. (1938). A contribution to the study of nematodes. Entomopathogenic Memorandum Department Agriculture and Forest Union South Africa, 2: 1-40.
- Wouts, W. M. (1966). *Paratylenchus halophilus* (Nematoda: Criconematidae), a new species from New Zealand. *New Zealand Journal of Science*, 9: 281-286.
- Zarina, B. (1996). *Studies on Plant Parasitic Nematodes of Ornamental and Vegetable Plants with Special Reference to Root-knot Nematodes*. Ph. D. Thesis Department Botany, University of Karachi, Karachi, Pakistan, pp. 299
- Zarina, B., S. Akhtar, D. Khan and M. Javed Zaki (2015). The nematodes of chilli plants of Sindh: Abundance, diversity and the assemblage. *FUUAST J, Biol.* 5(1): 71-92.

(Accepted for publication June 2016)