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## Two new records of the Genus *Neopsilenchus* Thorne & Malek, 1968 and *Tylenchus Bastian*, 1865 (Nematoda: Tylenchidae) from Iran

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### Abstract

*Neopsilenchus citri* (Jairajpuri, 1968) Bello, 1972 and *Tylenchus bhitaii* Maqbool & Shahina, 1987, the species belonging to the family Tylenchidae Örley, 1880 are reported from Iran for the first time. The morphological and morphometric characters of Iranian populations of the two species are discussed and illustrated. Iranian population of *N. citri* is characterised by having a 738.5-928 µm long body, rounded and unstriated head, lateral field with four lines, slender and weak stylet (12-13.5 µm), postvulval uterine sac less than one body width long, 10-12.5 µm and filiform tail with acute terminus (73.5-94 µm). Compared to the morphological characters and morphometric data given in original description, there were no differences between both the populations. The Iranian population of *T. bhitaii* is morphologically and morphometrically similar to the original population. It is characterised by having a body length of 600-668 µm, stylet length of 10-11 µm, cuticular annulations 1.0-1.5 µm width at mid body, for lines in lateral fields, postvulval uterine sac 10-17 µm and curved tail length 60-68 µm with rounded smooth terminus.

**Keywords:** Eastern Iran, Taxonomy, *Neopsilenchus citri*, *Tylenchus bhitaii*

### Introduction

Tylenchidae Örley, 1880<sup>[1]</sup> includes Boleodorinae Khan, 1964<sup>[2]</sup>, Duosulciinae Siddiqi, 1979<sup>[3]</sup>, Tanzaniinae Siddiqi, 2000<sup>[4]</sup>, Thadinae Siddiqi, 1986<sup>[5]</sup> and Tylenchinae<sup>[4]</sup>. Besides Boleodorinae and Tylenchinae, Geraert<sup>[6]</sup>, recognized three other subfamilies under Tylenchidae: Atylenchinae Skarbilovich, 1959<sup>[7]</sup>, Ecphyadophorinae Skarbilovich, 1959<sup>[7]</sup> and Tylodorinae Paramonov, 1967<sup>[8]</sup>. On the other hand, De Ley and Blaxter<sup>[9]</sup>, using molecular data to revise Nematoda phylogeny, did not recognize lower taxonomic levels under Tylenchidae (*sensu* De Ley & Blaxter<sup>[9]</sup>). The genus *Neopsilenchus* Thorne & Malek, 1968<sup>[10]</sup> belonging to the subfamily Boleodorinae was described by *N. magnidens* Thorne, 1949<sup>[11]</sup>, as type species<sup>[10]</sup>. The absence of knobs was the main differentiating character from *Basiria* Siddiqi, 1959<sup>[12]</sup> and the possession of a single genital tube in comparison to two from *Psilenchus* de Man, 1921<sup>[13]</sup>. The genus *Tylenchus* Bastian, 1865<sup>[14]</sup> belong to the subfamily Tylenchinae. Bastian<sup>[14]</sup> designated no type species. He placed the genus (as well as other free-living genera of the Nematoda) in the family Anguillulidae Gervais & Van Beneden, 1859<sup>[15]</sup>, and listed six species under it. The genus *Tylenchus* is most closely related to *Filenchus* Andrassy, 1954<sup>[16]</sup> by virtue of its delicate head sclerotization, short stylet and labial plate configuration. It differs in the heavier stylet with a cone of about half the stylet length (*vs.* cone 0.3-0.4 the total stylet length), the larger pit-like amphidial opening essentially confined to the labial plate in *Tylenchus* (*vs.* elongate, slit-like apertures extending 3-4 annuli beyond labial plate in most *Filenchus* species). Nevertheless by following this diagnosis Geraert & Raski<sup>[17]</sup> transferred several species considered by Andrassy<sup>[18]</sup> as *Tylenchus* to *Filenchus*. Brzeski<sup>[19]</sup>, Siddiqi<sup>[4]</sup> and Andrassy<sup>[20]</sup> followed this proposal. There was no information about the Tylenchidae fauna associated with the *Berberis vulgaris*, *Crocus sativus* and *Ziziphus zizyphus* fields in Iran. To fill this gap, this study aimed on determining the plant-parasitic nematodes of the family Tylenchidae on this crop in South Khorasan province, eastern Iran. During nematode surveys conducted in 2011–2014, we detected some species of Tylenchidae associated with root and soil samples. This list includes *B. impar*, *B. thylactus*, *B. typicus* and *B. volutus*<sup>[21]</sup>, *F. aquilonius*, *Irantylenchus vicinus* and *N. magnidens*<sup>[22]</sup>, and *F. pratensis*<sup>[23]</sup>. Additional data about the species of the family Tylenchidae in eastern Iran are

presented hereafter by identification of two new species *N. citri* from *Z. zizyphus* and *T. bhitaii* from *B. vulgaris*.

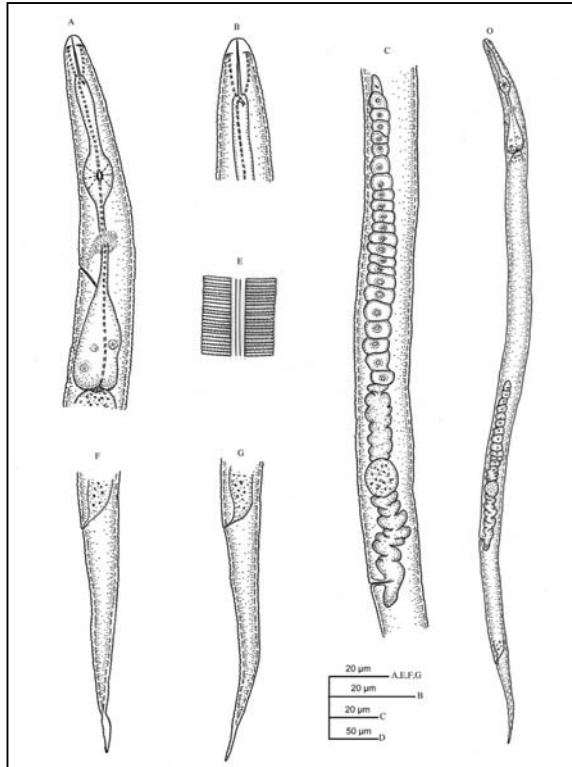
### Materials and methods

Surveys were conducted during 2011–2014 to determine the plant-parasitic nematodes of the family Tylenchidae associated with the *B. vulgaris*, *C. sativus* and *Z. zizyphus* fields in eastern Iran. A total number of 360 soil samples were collected from eastern Iran. To obtain a cleaner suspension of nematodes, the tray method [24] was used. Nematodes of interest were handpicked, heat-killed by adding boiling 4% formalin solution, transferred to anhydrous glycerin according to De Grisse [25], mounted in permanent slides, and examined using a light microscope. Drawings were made using a drawing tube attached to the microscope. Genera and species were identified based on morphological and morphometrical characters. The species *Neopsilenchus citri* [26, 27] and *Tylenchus bhitaii* [28] was identified by using a key to species given by Geraert [6].

### Results and discussion

During nematode surveys conducted in 2011–2014 in South Khorasan province, eastern Iran, two populations of two species belonging to the genus of *Neopsilenchus* and *Tylenchus* were collected. These populations were identified as *N. citri* and *T. bhitaii*. These two species are new records for Iranian nematofauna. Morphological descriptions of the two species as well as illustrations of females are provided.

*Neopsilenchus citri* (Jairajpuri, 1968) Bello, 1972: Figure 1



**Fig 1:** *Neopsilenchus citri* (Female). Part of Pharynx (A). Anterior end (B). Part of reproductive system (C). Entire body (D). Lateral fields (E), Tail (F, G)

**Measurements:** Table 1.

**Table 1:** Morphometric characters of *Neopsilenchus citri* and *Tylenchus bhitaii* collected from eastern Iran. [All measurements are in  $\mu\text{m}$  and in the form: mean $\pm$  S.D. (range).

characters	<i>Neopsilenchus citri</i>	<i>Tylenchus bhitaii</i>
n	12	8
L	811.7 $\pm$ 98.5 (738.5-928)	627.8 $\pm$ 23.4 (600-668)
a	34.9 $\pm$ 4.2 (29.5-40.3)	35.5 $\pm$ 1.1 (34.3-37.5)
b	6 $\pm$ 0.3 (5.5-6.4)	5.1 $\pm$ 0.2 (4.8-5.6)
c	9.5 $\pm$ 0.4 (9-10)	9.7 $\pm$ 0.5 (8.8-10.3)
c'	7.2 $\pm$ 1 (6.1-8.5)	5.5 $\pm$ 0.3 (5-5.9)
V	70.3 $\pm$ 1 (69-71.4)	71.1 $\pm$ 0.8 (70.1-72.5)
V'	78.5 $\pm$ 1.5 (76.8-80.3)	79.5 $\pm$ 0.8 (78.4-80.7)
Stylet length	12.3 $\pm$ 0.6 (12-13.5)	10.5 $\pm$ 0.5 (10-11)
Pharynx length	132.2 $\pm$ 12.4 (120-48)	122.3 $\pm$ 10.9 (109-138)
E-pore	100.3 $\pm$ 5.6 (96-110)	95.9 $\pm$ 10.7 (85.5-115)
Maximum body width	23.2 $\pm$ 1.3 (22-25)	17.6 $\pm$ 1 (16-19)
Head-Vulva	570.5 $\pm$ 61.2 (521.5-41)	447.1 $\pm$ 20.8 (423-480)
Vulva-Anus	156.6 $\pm$ 29.7 (130-193)	116 $\pm$ 5 (109-120)
Postvulval uterine sac length	11.2 $\pm$ 1.1 (10-12.5)	13.3 $\pm$ 2.5 (10-17)
Vulval body width	23.2 $\pm$ 1.3 (22-25)	17.6 $\pm$ 1 (16-19)
Anal body width	11.7 $\pm$ 0.5 (11-12.5)	11.6 $\pm$ 0.6 (11-12.5)
Tail length	84.6 $\pm$ 8.7 (73.5-94)	64.6 $\pm$ 3 (60-68)
T/VA	0.5 $\pm$ 0.08 (0.4-0.6)	0.5 $\pm$ 0.04 (0.5-0.6)

### Female

Body straight to slightly ventrally arcuate, 738.5-928  $\mu\text{m}$ . Lateral fields marked by four lines. Head bluntly rounded, unstriated. Amphidial apertures are transverse slits, located well below the base of the lateral lips. Stylet slender and weak, without basal knobs, 12-13.5  $\mu\text{m}$ . The orifice of the dorsal gland close to the base of stylet. Median bulb oval, 10-15  $\mu\text{m}$  long. Elongate, pyriform basal bulb, 26-29  $\mu\text{m}$  long. Excretory pore 96-110  $\mu\text{m}$  from anterior end. Spermatheca not offset, short or elongated, Posterior uterine sac less than one body width long, 10-12.5  $\mu\text{m}$ . Vagina short, Vulva-anus distance 130-193  $\mu\text{m}$ . Filiform tail with acute or finely rounded terminus, 73.5-94  $\mu\text{m}$  (it is cuticularized or irregularly swollen in one specimen).

### Male

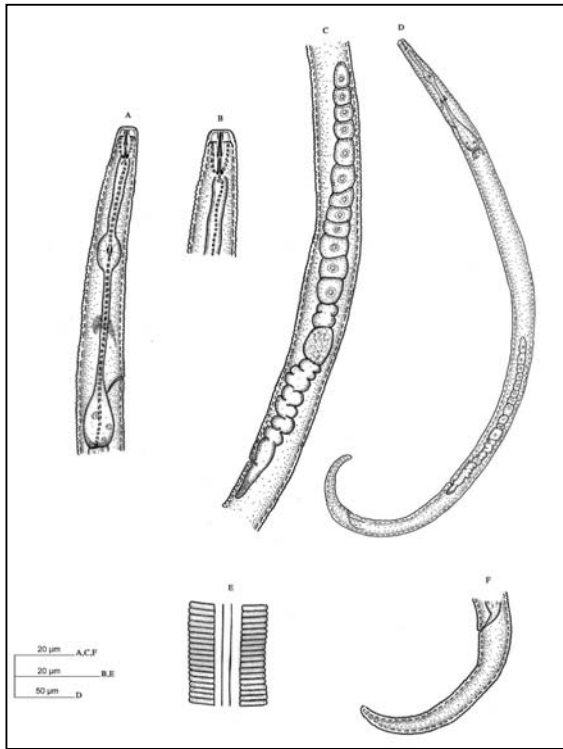
Not found.

### Remarks

According to the morphological characters and morphometric data given in the original description, there were no differences between the Iranian population of *N. citri* and the original description. *N. citri* is morphologically similar to *N. microdens* Ahmad & Khan, 1995 [29], but it differs in having longer stylet (12-13.5 vs 7-9  $\mu\text{m}$ ), shorter tail length (73.5-94 vs 106-112  $\mu\text{m}$ ), c ratio (9-10 vs 6.6-6.9), V ratio (69-71.4 vs 71-72), T/VA (0.4-0.6 vs 0.9-1.1) and the length of postvulval uterine sac (10-12.5 vs 13-15  $\mu\text{m}$ ).

### Type habitat and locality

Recovered from soil samples collected about the rhizosphere of *Z. zizyphus* in village of Mazhan, Southern Khorasan province, eastern Iran. GPS coordinates: N 32°33'59.36", E 59°00'34.6".

***Tylenchus bhitaii* Maqbool & Shahina, 1987: Figure 2.**

**Fig 2:** *Tylenchus bhitaii* (Female). Part of Pharynx (A). Anterior end (B). Part of reproductive system (C). Entire body (D). Lateral fields (E), Tail (F)

**Measurements:** Table 1.**Female**

Body strongly bent ventrally, 600-668  $\mu\text{m}$ . Cuticle finely annulated with 1.0-1.5  $\mu\text{m}$  wide annuli at mid body. Lateral fields 1/3 as wide as body, with four lines, marginal incisures weakly crenate. Head 6-7  $\mu\text{m}$  wide, continuous with body contour, with 4-5 fine annuli. Stylet 10-11  $\mu\text{m}$  long, knobs rounded. Median bulb oval, 10.5-13  $\mu\text{m}$  long. Isthmus long and slender, Basal bulb cylindrical-elongated, 17-20  $\mu\text{m}$  long. Excretory duct heavily sclerotized, 85.5-115  $\mu\text{m}$  from anterior end, opens at the base of the isthmus to the level of anterior half of terminal bulb. Gonad short. Spermatheca rounded. Vulval lips not protruding. Postvulval uterine sac 10-17  $\mu\text{m}$  long. Tail strongly curved ventrally, tapering slowly to a rounded smooth terminus, 60-68  $\mu\text{m}$ .

**Male**

Not found.

**Remarks**

According to the morphological characters and morphometric data given in the original description, there were no differences between the Iranian population of *T. bhitaii* and the original description. *T. bhitaii* is morphologically similar to *T. naranensis* Maqbool, Zarina, & Ghazala, 1987<sup>[30]</sup>, but it differs in having shorter body length (600-668 vs 730-870  $\mu\text{m}$ ), longer stylet (10-11 vs 9-10  $\mu\text{m}$ ), shorter tail length (60-68 vs 110-117  $\mu\text{m}$ ), c ratio (8.8-10.3 vs 6-7), c' ratio (5-5.9 vs 7.2-9.1), V ratio (70.1-72.5 vs 66-69), T/VA (0.5-0.6 vs 0.8-1) and cuticular annulations wide at mid body (1.0-1.5 vs 1.5-2  $\mu\text{m}$ ).

**Type habitat and locality**

Recovered from soil samples collected about the rhizosphere of *B. vulgaris* in village of Bureng, Southern Khorasan province, eastern Iran. GPS coordinates: N 33°00'56.2", E 059°45'42.5".

**Acknowledgement**

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