

## Characterization of a new plant parasitic nematode isolated from rhizospheric soil of rice-plants

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### ABSTRACT

Survey for soil and plant parasitic nematodes associated with paddy (*Oryza sativa* L.) in Purba Bardhaman district of West Bengal, India revealed the presence of a new plant parasitic species belonging to the genus *Tylenchus*. The new species *Tylenchus scythecaudus* sp. nov. comes close to *Tylenchus aquilonios* Wu, 1969, *Tylenchus cylindricaudus* Wu, 1969, *Tylenchus hazanensis* Wu, 1969, *Tylenchus helenae* Szczygiel, 1969, *Tylenchus quartus* Szczygiel, 1969 in having continuous lip region and filliform tail with pointed terminus, those differs from others. The present species differs from all existing species of *Tylenchus* in having different values of a, b, c, V, short gradually tapering tail with scythe-shaped tail terminus and short post-uterine sac. *Tylenchus scythecaudus* sp. nov. is described and illustrated here.

**KEY WORDS:** NEMATODE . NEW SPECIES. PADDY. *TYLENCHUS*. WEST BENGAL

### INTRODUCTION

West Bengal is contributing a significant part in India as far as rice production is concerned. Purba Bardhaman district is known as the 'Grainery of West Bengal' as it produces maximum amount of rice within the state. Nematodes are tiny creatures causing huge damages to

the crop. Species belonging to the order Tylenchida are plant parasitic in nature. A survey for soil and plant parasitic nematodes associated with paddy in Purba Bardhaman district of West Bengal was conducted in view to find out the plant parasitic nematodes associated with this crop. The study encountered a new species of plant parasitic nematode *Tylenchus scythecaudus* sp.

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nov from the rhizospheric region of paddy which is an added information of Tylenchid nematode species of this important crop.

The genus *Tylenchus* was proposed by Bastian (1865) originally and then it was re-established by Filipjev in 1936. The species of this genus are characterized by having small to medium sized body about 0.4–1.3 mm in length, ventrally curved after fixation, moderately thick cuticle around 1–2  $\mu\text{m}$ , distinctly annulated. Lateral fields having four incisures. Dorso-sublateral, postmedian phasmids present just behind the vulva. Annulated cephalic region continuous with the body. Stylet length varies between 8–21  $\mu\text{m}$ , conus is more than one-third of the total stylet length. Median oesophageal bulb oval shaped which is present anterior to the mid-oesophageal position. Cardia is distinct. In most of them excretory pore is present opposite to the basal bulb. Vulva, a transverse slit like, reproductive organ occupying 60–70% of the total body length. Post-uterine sac generally a body width or often a less long. Round to oval, offset spermatheca is present. Ovary outstretched. Ventrally arcuate tail, sometimes hooked, gradually tapering to a pointed or minutely rounded terminus. Review of literatures of all existing species of *Tylenchus* revealed that *Tylenchus scythecaudus* sp. nov. encountered during the survey distinctly differed from others. The species is illustrated here with morphometric details, line diagrams and photomicrographs.

## MATERIALS AND METHODS

### Soil sample collection

The rhizospheric soil samples were collected from paddy field of Kalna II block of Purba Bardhaman district of West Bengal, India.

*Processing of the soil Sample:* Nematodes were extracted from the samples following 'Cobb's Sieving Technique' (Cobb, 1918). For this at first 250g soil sample was mixed with water with gentle stirring by hand in a medium sized bucket. The mixing was done in such a way that there was no soil lumps present. The mixture was allowed to rest for few seconds (15–20s) so that the nematodes can float on the upper surface of the water and then sieved. In the final step of extraction, decanting was done by following 'Modified Baerman's Funnel Method' (Christie & Perry, 1951)

### Killing and fixation

The nematode samples were killed as well as fixed in hot F.A (Formaldehyde-Acetic acid) solution.

### Post fixation process

The nematode samples were further processed by 'Seinhorst's Slow Dehydration Method' (Seinhorst, 1959) in

which fixed nematodes were picked in cavity block and allowed them to dehydrate in Glycerin alcohol solution for thirty days. Desiccators were used to keep the cavity blocks. After that the specimens were mounted on slides in glycerin (anhydrous) and sealed. Ocular micrometer of Olympus Research Microscope (Model No. BX 41, with drawing tube attachment) was used for taking the measurements. Dimensions were tabulated following De Man's Formula (De Man, 1884). Diagrams were drawn using camera lucida. Photomicrographs were captured using Leica Research Microscope (Model No. Leica DM 1000). The species was identified following the keys, made by Siddiqi (2000)

## RESULTS AND DISCUSSION

### Systematics

The following classification is given by Siddiqi (2000)

*Tylenchus scythecaudus* sp. nov. (Figs. 1–10)

### Measurements

All measurements are provided in Table 1.

*The following abbreviations are used in the text and table* a= body length/maximum body diameter, b= body length/oesophageal length, c= body length/tail length, c'= tail length/body width, V= (position of vulva from anterior end/body length from)  $\times 100$ , V'= (position of vulva from head tip/distance of anus from head end)  $\times 100$ , G<sub>1</sub>= (length of anterior female gonad /body length)  $\times 100$ , G<sub>2</sub>= (length of posterior female gonad /body length)  $\times 100$ , m= length of conus/total stylet length, O= (distance between orifice of dorsal oesophageal gland and stylet/stylet length)  $\times 100$ , VL/VB= distance of vulva to posterior end of the body/vulval body width.

### Descriptions

#### Female

Body slender, small about 0.450–0.472 (0.461  $\pm$  0.008) mm long, uniformly tapering; ventrally curved at vulval region, tail end dorsally bended, scythe shaped (Fig.1). Cuticle with transverse annulations; lateral fields with four incisures. weakly developed deirids, located near end of oesophagus. Lip amalgamated, continuous with the body i.e. not set off. Stylet is well developed about 9–10 (9.6  $\pm$  0.380)  $\mu\text{m}$ , conus about 37% of stylet length; basal knobs prominent. Orifice of dorsal oesophageal gland nuclei present in very close proximity to the stylet base. Median bulb well developed, more or less oval in shape (4.2 $\times$ 3.4)  $\mu\text{m}$ , located at the position of 38% of total oesophageal length from anterior extremity. The terminal oesophageal bulb about 17.2–20.2

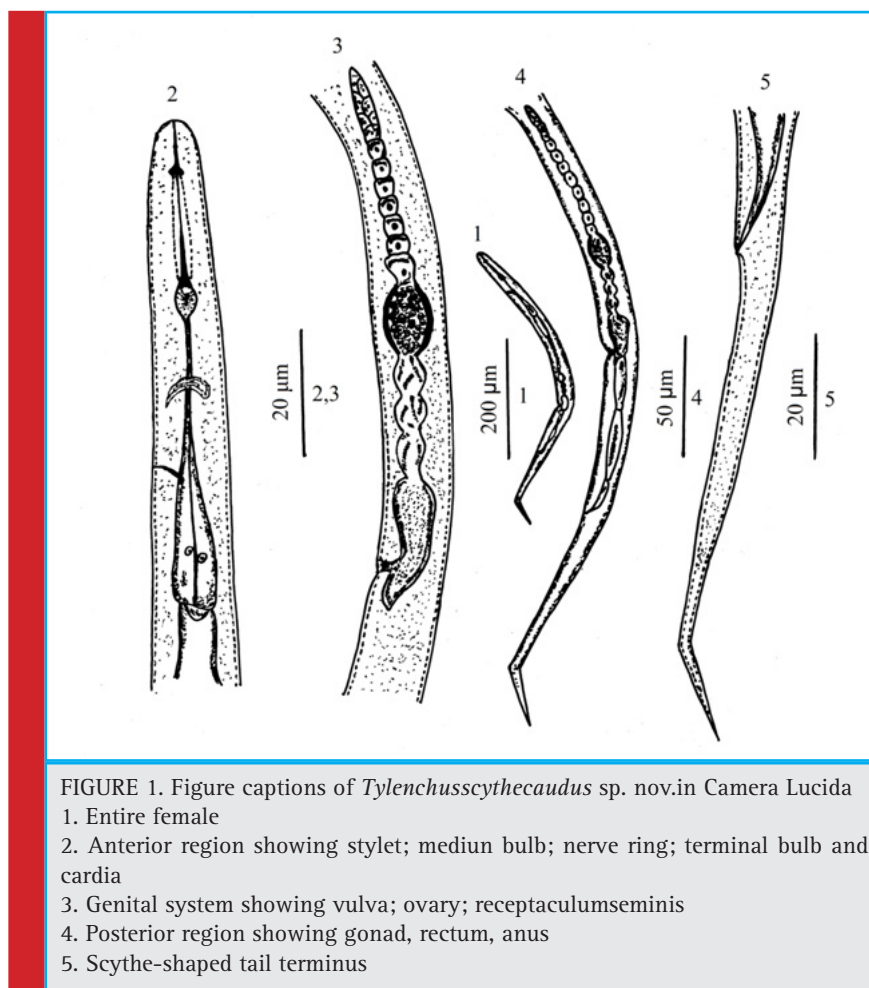


FIGURE 1. Figure captions of *Tylenchusscythecaudus* sp. nov. in Camera Lucida  
 1. Entire female  
 2. Anterior region showing stylet; median bulb; nerve ring; terminal bulb and cardia  
 3. Genital system showing vulva; ovary; receptaculum seminis  
 4. Posterior region showing gonad, rectum, anus  
 5. Scythe-shaped tail terminus

( $18.05 \pm 1.252$ )  $\mu\text{m}$ , sac like, occupying about 20% of total oesophageal length; two oesophageal gland nuclei prominent. Excretory pore present at the beginning of terminal oesophageal bulb.

Hemizonid present at 76.3% of oesophageal length from head end and anterior to excretory pore, cardia small about  $1.2\text{--}2.2$  ( $1.55 \pm 0.3728$ )  $\mu\text{m}$ , cone shaped, about 0.46 times of lip region width and occupying 9.6% of corresponding body width. Reproductive system prodelphic. Vagina thin walled, present at right angle to the body, occupying about half of corresponding body width. Anterior genital branch about  $140.5\text{--}145$  ( $142.88 \pm 1.710$ )  $\mu\text{m}$  in length, occupying 98% of total gonadal length. The post-uterine sac rudimentary with pointed tip, only about 0.23 times of corresponding body width. *Receptaculum seminis* (spermatheca) distinct, well set off and filled with sperms. Ovary with a single row of oocytes, except terminal region. Rectum about  $17\text{--}18.5$  ( $17.56 \pm 0.513$ )  $\mu\text{m}$ , 2.3 times anal body width long. Tail filiform,  $85\text{--}90.5$  ( $87.6 \pm 2.22$ )  $\mu\text{m}$  long with pointed terminus, 0.77 times of vulva-anal distance; tail terminus scythe-shaped and bended dorsally.

Male: Male not found. Type habitat and locality Specimens were collected from the rhizospheric soil of *Oryza sativa* L. by the first author in 20<sup>th</sup> October 2016 from Kalna II block ( $23.168391^\circ$  N,  $88.245036^\circ$  E) of Purba Bardhaman district, West Bengal, India. Type materials: Specimens were deposited to the National Zoological Collections of Zoological Survey of India, Kolkata, West Bengal, India under the Registration No. WN 1948 (Slide with Holotype female and two Paratype females).

**Etymology** :The new species is named *Tylenchus scythecaudus* as it bears scythe shaped tail terminus.

**Differential diagnosis and relationships**

*Tylenchus scythecaudus* sp. nov. differs from all existing species of *Tylenchus* in having short, gradually tapering tail with scythe-shaped tail terminus and short post-uterine sac.

*Tylenchus scythecaudus* sp. nov. comes close to *T. aquilonios* Wu, 1969, *T. cylindricaudus* Wu, 1969, *T. hazanensis* Wu, 1969, *T. helenae* Szczygiel, 1969, *T. quartus* Szczygiel, 1969 in having continuous lip region and filiform tail with pointed terminus, those differs from others.

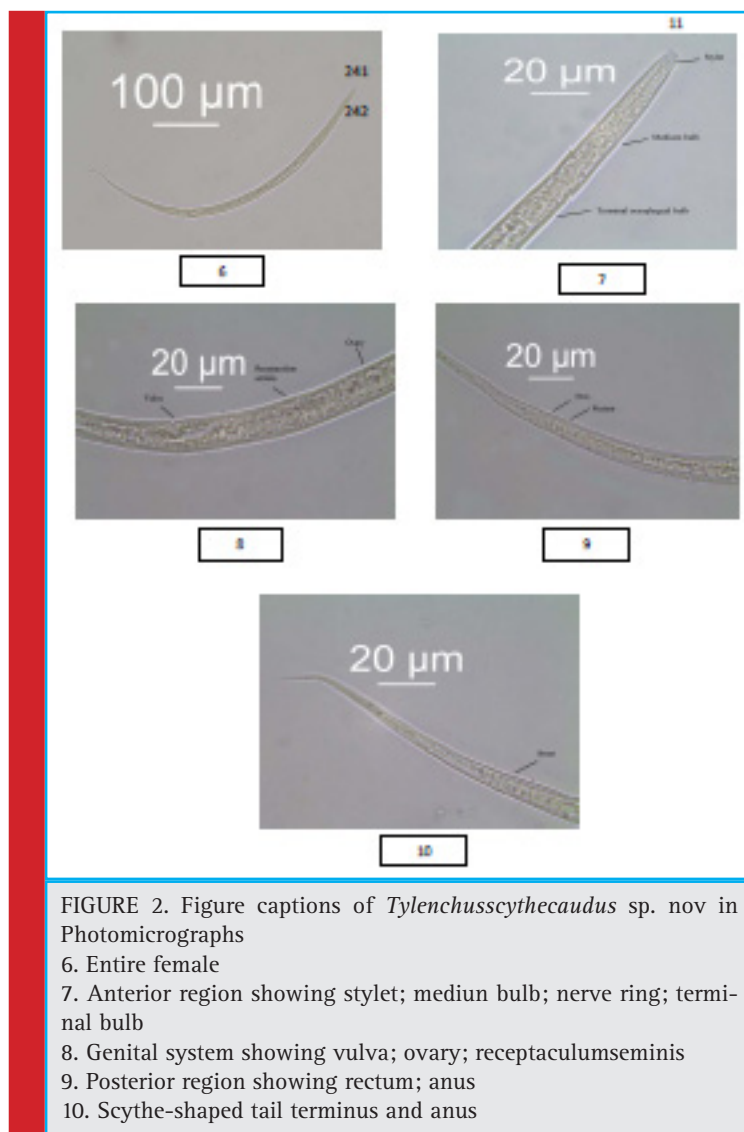


FIGURE 2. Figure captions of *Tylenchusscythecaudus* sp. nov in Photomicrographs  
 6. Entire female  
 7. Anterior region showing stylet; median bulb; nerve ring; terminal bulb  
 8. Genital system showing vulva; ovary; receptaculum seminis  
 9. Posterior region showing rectum; anus  
 10. Scythe-shaped tail terminus and anus

The species is similar to *Tylenchus aquilonios* Wu, 1969 in having continuous lip region, filliform tail but the present species differs from it in having small body size, smaller values of a, b, c, V, shorter oesophageal length, lesser vulval body width, shorter post-uterine branch, lesser vulva-anal distance and short tail with scythe-shaped terminus, (L= 0.803 mm, a= 35, b= 6.7, c= 6.9, V= 68, oesophageal length= 144-153 μm, post-uterine branch= 13-15 μm, body width at vulval region= 19-26 μm, vulva-anal distance= 115-149 μm, tail length and shape= 116 μm; tapering gradually to a pointed terminus in *Tylenchus aquilonios* Wu, 1969).

The present species is similar with *Tylenchus cylindricaudus* Wu, 1969 in having slender, gradually tapering body with continuous lip region, filliform tail with pointed terminus but differs from it in having smaller body size, lesser values of a, b, c, V, slightly shorter

stylet, short post-uterine branch, lesser body width at vulva, reduced vulva-anal distance, shorter tail and different shape of tail terminus (L= 0.95mm, a= 41, b= 6.9, c= 6.6, V= 66, stylet length= 11-13 μm, oesophageal length= 127-137 μm, post-uterine branch= 11-14 μm, body width at vulval region= 19-21 μm, vulva-anal distance= 183 μm, tail length= 147 μm, shape of tail terminus is straight filliform in *Tylenchus cylindricaudus* Wu, 1969). *T. scythecaudus* sp. nov. is similar to *Tylenchus hazanensis* Wu, 1969 in having rounded head with continuous lip region and filliform tail with needle like pointed terminus but varies from it in smaller body size, smaller values of a, b, c, V, smaller stylet, shorter oesophageal length, presence of annules at mid-tail region, lesser vulval body width, shorter post-uterine branch, lesser vulva-anal distance and short tail with scythe-shaped tail terminus (L= 0.2 mm, a= 38, b=

Table 1. Measurements of <i>Tylenchus scythecaudus</i> . sp. nov. (in $\mu\text{m}$ , except L in mm)		
Morphometric characters	Holotype female	Paratype females (n=4)
L	0.45	0.450-0.472 (0.461 $\pm$ 0.008)
A	31.2	31.2-31.7(31.4 $\pm$ 0.194)
B	5.2	5.2-5.3(5.24 $\pm$ 0.054)
c	5.2	5.2(0)
c'	11.6	11.4-12.5(11.8 $\pm$ 0.418)
V	56.7	55.6-56.8(56.44 $\pm$ 0.482)
V'	70	69.6-70.0(69.9 $\pm$ 0.173)
G1	31.4	30.7-31.4(31.06 $\pm$ 0.270)
G2	0.55	0.55-0.63(0.574 $\pm$ 0.033)
Width of lip	2.6	2.2-2.8(2.4 $\pm$ 0.189)
Total Stylet length	9.7	9-10(9.6 $\pm$ 0.380)
Median bulb from anterior end	32.3	38.5-40(32.96 $\pm$ 0.996)
Median bulb diameter	4.2 $\times$ 3.4	-
Oesophageal length	85.75	85.5-88.6 (86.67 $\pm$ 1.18)
Nerve ring from anterior end	44.5	44.2-48.4(45.4 $\pm$ 1.60)
Hemizonid from anterior end	65.5	65.3-67.8(66.34 $\pm$ 1.00)
Length of terminal bulb	17.15	17.2-20.2(18.05 $\pm$ 1.252)
Length of cardia	1.2	1.2-2.2(1.55 $\pm$ 0.3728)
Maximum body width	14.5	14.3-15(14.6 $\pm$ 0.264)
Vulva from anterior end	257.3	256-262.8(259.8 $\pm$ 3.05)
Body width at vulva	11.2	11-11.7(11.26 $\pm$ 0.219)
Anterior genital branch	142	140.5-145 (142.88 $\pm$ 1.710)
Posterior genital branch	2.5	2.5-3 (2.66 $\pm$ 0.207)
Body width at anus	7.4	7-7.9 (7.46 $\pm$ 0.320)
Rectum	17.3	17-18.5 (17.56 $\pm$ 0.513)
Tail length	85.8	85-90.5 (87.6 $\pm$ 2.22)
Vulva-anal distance	110.2	109.3-114.2 (111.9 $\pm$ 2.11)
m	37.4	33.3-38.7 (36.84 $\pm$ 2.100)
O	72	72 (0)
VL/VB	17.78	17.5-17.9 (17.7 $\pm$ 0.158)

6.8, c= 4.7, V= 62, stylet length= 15.5  $\mu\text{m}$ , oesophageal length= 149-165  $\mu\text{m}$ , mid-tail annulation absent, vulva-anal distance= 178  $\mu\text{m}$ , tail length= 224  $\mu\text{m}$  and tail terminus shape is straight filliform, needle like in case of *Tylenchus hazanensis* Wu, 1969).

The present species is again similar with *Tylenchus helenae* Szczygiel, 1969 in having small slender body, continuous lip region with body, filliform pointed tail, needle like tail terminus but differs from it in having larger body length, larger values of a, b, c, V, larger stylet, more anterior position of median bulb in oesophagus, smaller post-uterine sac, presence of spermatheca, distinct anus and scythe-shaped tail end (L= 0.43 mm, a= 37, b= 4.6, c= 3.6, V= 57.5, position of median bulb

in pharynx at 43%, size of post-uterine sac is large, more than one body diameter long, spermatheca absent, anus obscure, long filliform tail ending with a needle like terminus in *Tylenchus helenae* Szczygiel, 1969). *Tylenchus scythecaudus* sp. nov. shows similarities with *Tylenchus quartus* Szczygiel, 1969 in having similar body contour *viz.* delicate, slender, gradually tapering body with continuous lip region and filliform tail but the present species differs from it in having shorter body length, smaller value of a, c, V, but larger values of b, lesser value of G1, shorter stylet length, presence of distinct knobs, oval shaped median bulb, dorsally bended scythe-shaped tail (L= 0.56 mm, a= 39.5, b= 4.7, c= 5.3, V= 65.2, stylet length= 11.8  $\mu\text{m}$ , conspicuous basal

knobs, elongate fusiform median bulb, uniformly tapering tail with very sharp pointed terminus in *Tylenchus quartus* Szczygiel, 1969).

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*Compliance with Ethical Standard:* Ethical standard has been maintained, no toxic or harmful chemical is used during the experiment.

*Author contribution:* Author Paromita Roy carried out the work *viz.* collection of the specimen, slide preparation, and preparation of the manuscript. Author Viswa Venkat Gantait helped to identify the specimen. Author Soumendranath Chatterjee helped to prepare the manuscript. All the authors agreed and approved the final draft of manuscript.

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