

Further characterisation of *Schistonchus caprifici* (Gasparrini, 1864) Cobb, 1927 from *Ficus palmata* with a tabular key for the globally known species

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Summary. A population of *Schistonchus caprifici* was recovered from syconia of *Ficus palmata* grown in New Delhi (India). The population was identified based on morphological and molecular characteristics. The morphological and morphometric features of the *Schistonchus* population from *Ficus palmata* were found to be consistent with the original and subsequent descriptions of *S. caprifici*. This comprehensive morphological study of *S. caprifici* from India has added additional information on morphological and morphometric characters, such as lip shape, stylet, vulva position from anterior end, spermatheca, anal/cloacal body diameter, vulval structure, and ovary/testis length. In addition, some morphological descriptions including spicule shape and gubernaculum in males, vulval lips and vaginal lines, post-uterine sac, tale shape, and terminus are amended. The identification of this population was further confirmed by using sequences of the D2-D3 region of 28S rDNA (LSU), 18S rDNA (SSU) and *COI* of mtDNA, and its phylogenetic relationship with the *S. caprifici* from Turkey, Spain, Italy, and other species from around the world was established. A tabular key for the species under the genus *Schistonchus* is provided.

Key words: fig nematode, large subunit rDNA (LSU), mitochondrial DNA subunit I (mt*COI*), molecular phylogeny, morphology, small subunit rDNA (SSU), tabular key.

During a survey to determine the diversity of nematodes, high density of the fig nematode, *Schistonchus caprifici* (Gasparrini, 1864) Cobb, 1927 (Rhabditida; Aphelenchoidea), was observed in syconia of *Ficus palmata* Frossk. tree at Pusa campus, Indian Council of Agricultural Research-Indian Agricultural Research Institute (ICAR-IARI), New Delhi, India. *S. caprifici* was described for the first time in 1864 as a mutualistic associate of wasp, *Blastophaga psenes* (L.) from *Ficus carica* L. in Italy. This species was further described and characterised by Cobb (1927) from Algeria, Thorne (1961) from Turkey, Vovlas *et al.* (1992) from Italy, and Kolaei *et al.* (2016) from Iran. Gulcu *et al.* (2008) recorded genetic variability of *S. caprifici* from Turkey by using the D2-D3 region of 28S rDNA and cytochrome oxidase I (*COI*) of mitochondrial DNA. De Luca *et al.* (2010) characterised Italian and Spanish populations using 18S, D2-D3 of 28S, and *COI* of mt DNA. Fard and Zare (2020) characterised it by using D2-D3

molecular markers from Iran. Thus so far, *S. caprifici*, has been recorded from Algeria, Turkey, Spain, Italy, Iran and India. In India, *S. caprifici* was recovered from the fig wasp (*B. psenes*), and fig fruit (*Ficus carica*) and morphologically characterised based on a limited number of specimens and characters (Bajaj & Tomar, 2014), but no molecular sequence data is available for the species. Morphological and morphometric data of *S. caprifici* available in published literature appear to be incomplete and incongruent, although several attempts have been made to redescribe the species based on a few specimens from Turkey, Italy, Iran and India. Therefore, the present study focused on a population of *Schistonchus* recovered from syconia (enclosed globular infructescence or fruit – phase B and C) of *Ficus palmata*, a new host plant for the nematode, to further characterise the Indian population of *S. caprifici* based on morphology, morphometrics and molecular data.

MATERIAL AND METHODS

Nematode material. Nematodes were freshly collected from the syconia (phase B & C) of *Ficus palmata* from ICAR-IARI, New Delhi, India (28°39'25.9" N, 77°11'5.9" E; elevation 167 m a.s.l.). Syconia were cut open separately in beakers containing clear tap water; nematode species and associated wasps were observed after 1 h in the suspension. Nematodes were concentrated in a small amount of water, killed by a hot-water bath (Hooper, 1986) and fixed in TAF fixative (triethanolamine: 2 ml, formalin: 7 ml, distilled water: 91 ml) and processed to anhydrous glycerin by the glycerol-ethanol method (Seinhorst, 1959).

Morphological and morphometric study. Heat and TAF fixed mature male and female specimens of nematodes were hand-picked under a stereo zoom binocular microscope Olympus SZX-ILLK200 (Olympus Optical Co. Ltd, Japan), placed in anhydrous glycerin on clean glass slides, and sealed by wax-ring method (De Maeseneer & D'Herde, 1963) for morphological studies. The morphological characters described by earlier studies and some additional parameters were included for further characterisation of the species. De Manian ratios (De Man, 1884) and other measurements (De Grisse, 1964) were obtained using an Olympus BX50 compound microscope (Olympus Optical Co. Ltd, Japan) fitted with an ocular micrometre and a drawing tube. The photomicrographs were taken with a compound microscope Zeiss, AX10 Axiocam M2m (Carl Zeiss Meditec AG, Germany) fitted with the differential interference contrast (DIC) optics.

Molecular techniques. For the molecular characterisation, a single female nematode was transferred into a 0.2 ml micro centrifuge tube containing 25 µl of sterile water. An equal amount of lysis buffer containing 0.2 M NaCl, 0.2 M Tris-HCl (pH 8.0), 1% (v/v) β-mercaptoethanol, and 2 µl proteinase K (stock – 10 mg ml⁻¹) was added. The lysis was conducted in a thermocycler at 65°C for 2 h with an intermittent flicking, followed by incubation at 95°C for 10 min. Two µl of the lysate was used as DNA template in PCR reaction and excess lysate was stored at –20°C. For amplification, three molecular markers were used. The PCR amplification was done by using forward and reverse primer pairs; for D2-D3 expansion segments of the large subunit 28S rDNA gene (LSU) – D2A (5'-ACA AGT ACC GTG AGG GAA AGT-3') and D3B (5'-TGC GAA GGA ACC AGC

TAC TA-3') primer pair (Nunn, 1992); for 18S rDNA gene (SSU) SSUF07 (5'-AAA GAT TAA GCC ATG CAT G-3') and SSUR26 (5'-CAT TCT TGG CAA ATG CTT TCG-3') primer pair (Floyd *et al.*, 2002); and for cytochrome oxidase subunit I (COI) gene COI-F1 (5'-CCT ACA TGA TTG GTG GTT TTG GTA ATT G-3') and COI-R2 (5'-GTA GCA GCA GTA AAA TAA GCA CG-3') primer pair (Kanzaki & Futai, 2002). The PCR reactions were set up by using 2 µl of lysed DNA in a 0.5 ml micro centrifuge tube containing 25µl GoTaq® Green master mix (cat. no. M712, Promega, USA); 1 µl of each forward and reverse primer (10 µM); and nuclease-free water to a final volume of 50 µl. The thermal cycling program consisted of denaturation at 95°C for 5 min, followed by 35 cycles of denaturation at 94°C for 30 s, annealing at 55°C for 45 s, and extension at 72°C for 2 min. A final extension was performed at 72°C for 10 min. PCR products were cleaned using Wizard® SV Gel and PCR Clean-Up System (Promega, USA) according to the manufacturer's protocol. The PCR-amplified products were electrophoresed on 1.2% agarose gel and after confirming band size, purified PCR products were sent for sequencing (Sanger sequencing by ABI 3730xl at AgriGenome Labs, India). The obtained raw sequences were manually checked and modified to ensure quality, aligned and a consensus sequence was created for each marker gene using Mega X (Kumar *et al.*, 2018). The newly obtained D2-D3, SSU and COI sequences were used for a BLAST search to retrieve available sequences of other *Schistonchus* species from the NCBI GenBank. Phylogenetic analysis of the D2-D3 expansion segments of the 28S rDNA, SSU and cytochrome oxidase subunit I (COI) sequences were carried out. In brief, the outgroup taxa for each dataset were selected based on previously published data. The new and previously published/closely related sequences from NCBI GenBank for each gene were aligned (separately) using ClustalW in MEGA X (Kumar *et al.*, 2018) with default parameters. The appropriate model to generate phylogenetic tree was evaluated and evolutionary history was inferred using the Maximum Likelihood (ML) method with 1,000 bootstraps (Hasegawa *et al.*, 1985; Saitou & Nei, 1987).

RESULTS

Survey. In the present study, only one species of fig nematode, *Schistonchus caprifici* was dominant and was detected in syconia of *Ficus palmata* at

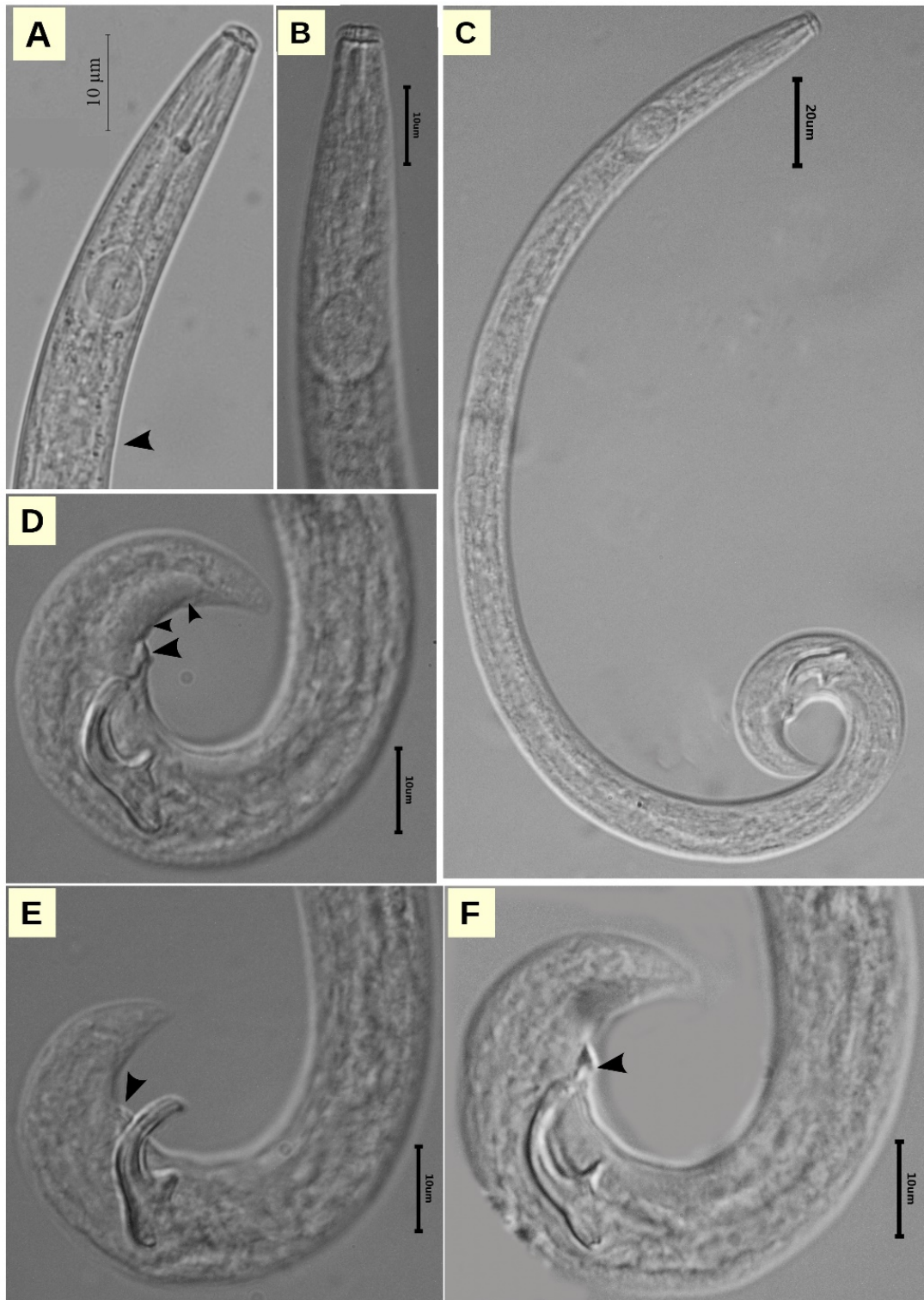


Fig. 1. Photomicrographs of *Schistonchus caprifici*. A: Anterior end of male with EP (arrowhead); B: Anterior end of male; C: Entire male; D-F: Posterior ends of males showing by arrowheads rostrate spicules, gubernaculum and caudal papillae. Scale bars: A, B, D-F = 10 μm, C = 20 μm.

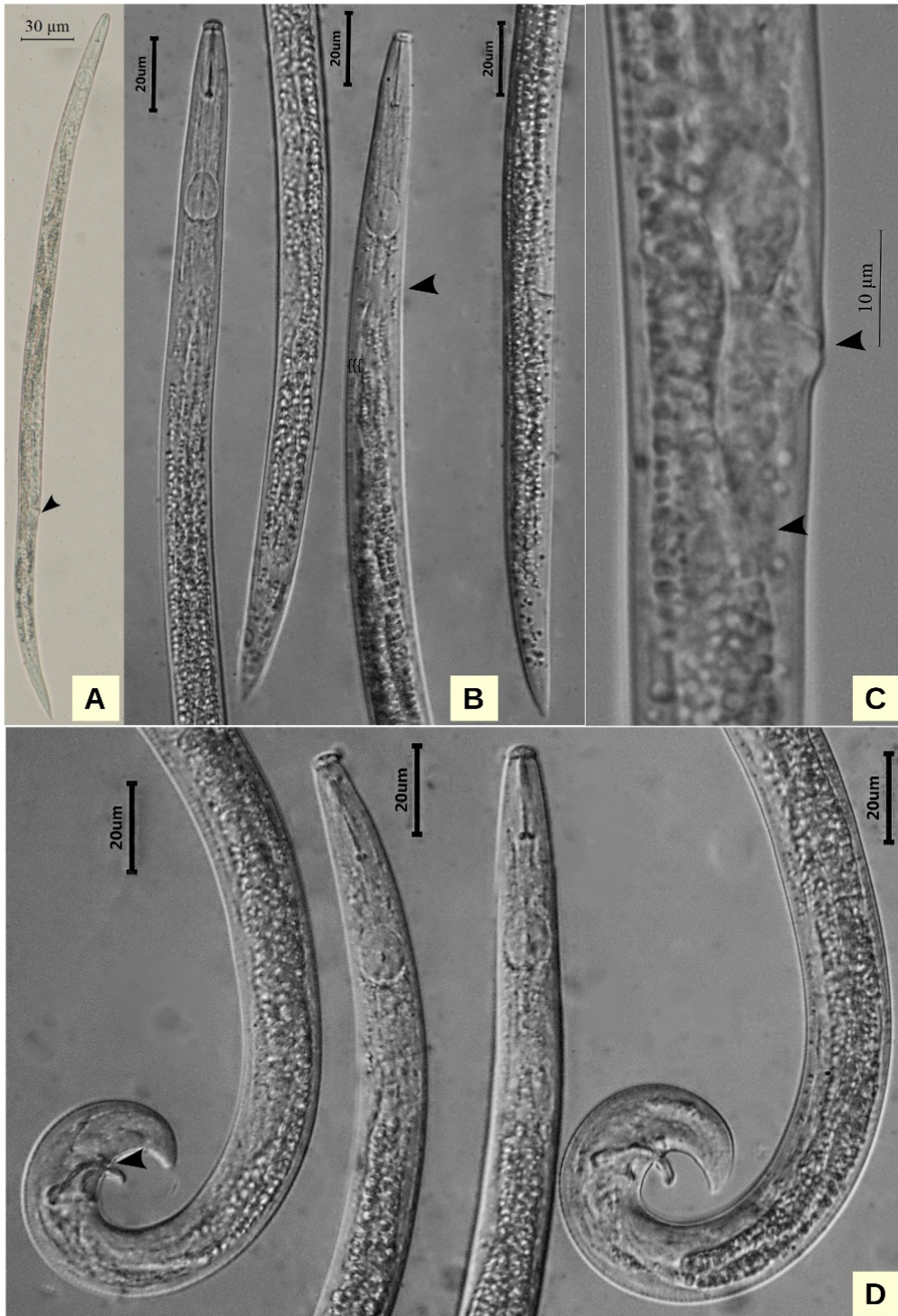


Fig. 2. Photomicrographs of *Schistonchus caprifici*. A: Entire female; B: Anterior and posterior end of reproductive females (arrowhead showing excretory pore); C: Vulva (vagina ventrally inclined) and post-vulval uterine sac of reproductive female (arrowhead pointing to terminal end of sac); D: Male anterior and posterior ends. Scale bars: A = 30 µm, B & D = 20 µm, C = 10 µm.

Table 1. Measurements and characters of *Schistonchus caprifici* from *Ficus palmata* from India compared with previously published measurements and characters.

Populations	Indian (Delhi) population present study		Indian (Hisar) population Bajaj & Tomar, 2014		Italian population Vovlas <i>et al.</i> , 1992	
Host	<i>Ficus palmata</i>		<i>Ficus carica</i>		<i>Ficus carica sylvestris</i>	
Character	Female n = 30	Male n = 30	Female n = 12	Male n = 12	Female n = 10	Male n = 10
L	488.8 ± 49.5 (402-575)	452.3 ± 61.6 (351-570)	466.1 ± 45.11 (359-527)	330.9 ± 20.8 (299-361)	496 ± 11.7 (435-557)	384.9 ± 9.3 (354-450)
a	33.3 ± 4.3 (26.8-44.2)	27.2 ± 4.7 (20.1-38.1)	29.6 ± 2.84 (24.5-34.0)	21.4 ± 1.72 (18.4-23.8)	38.4 ± 0.7 (33-42)	23.8 ± 0.4 (22-26)
B	4.5 ± 1.0 (2.9-8.2)	4.9 ± 1.0 (3.1-6.9)	8.3 ± 0.77 (6.5-9.4)	6.9 ± 0.47 (6.3-7.6)	3.3 ± 0.1 (3.0-3.6)	3.5 ± 0.1 (3.1-3.9)
c	13.6 ± 2.8 (10.5-25)	25.7 ± 5.2 (18.5-35.6)	13.0 ± 0.97 (10.6-14.2)	16.2 ± 1.45 (14.1-19.3)	13.6 ± 0.4 (12-16)	12.4 ± 0.3 (11-14)
c'	4.1 ± 0.6 (2.6-5.3)	0.9 ± 0.1 (0.7-1.2)	3.9 ± 0.49 (3.0-4.8)	1.4 ± 0.09 (1.2-1.5)	–	–
V/T	68.4 ± 7.8 (55.5-84.1)	55.2 ± 7.9 (42.8-69.8)	70.6 ± 0.65 (69.6-71.8)	–	70.3 ± 0.4 (69-73)	43.1 ± 1.3 (36-48)
Ant. end to base metacarpus	60.8 ± 6.0 (52-71)	53.9 ± 6.6 (41-75)	–	–	52 ± 0.7 (48-56)	48.3 ± 0.63 (45-52)
Ant. end to base pharyngeal gland	111.8 ± 17.1 (70-144)	95.4 ± 13.4 (68-119)	77.6 ± 4.82 (71-87)	61.0 ± 6.21 (54-69)	–	–
Max. body diam.	14.7 ± 0.9 (13-16)	16.7 ± 1.0 (14-18)	–	–	12.8 ± 0.2 (12-14)	16.1 ± 0.3 (14-17)
Lip diam.	7.2 ± 2.1 (5-12)	6.4 ± 0.5 (6-7)	–	–	–	–
Lip height	2.8 ± 0.6 (2-4)	2.9 ± 0.7 (2-4)	–	–	–	–
Conus stylet	9.4 ± 1.4 (6-12)	9.5 ± 2.0 (6-13)	–	–	–	–
Stylet shaft	12.3 ± 1.5 (9-15)	11.3 ± 1.7 (9-15)	–	–	–	–
Stylet length	21.7 ± 1.0 (19-24)	20.8 ± 1.5 (19-24)	19 (–)	19 (–)	22.9 ± 0.3 (21-24)	23.1 ± 0.3 (22-24)
Knobs diam.	3 ± 0.2 (2-3.5)	2.9 ± 0.3 (2-3)	–	–	–	–
Knobs height	2 ± 0.2 (0.8-2)	2.0 ± 0.2 (1.5-2.5)	–	–	–	–
Excretory pore from anterior end	72.2 ± 5.6 (64-86)	64.0 ± 7.2 (52-84)	66.9 ± 5.16 (60-76)	59.8 ± 5.80 (51-71)	–	–
Vulval position from ant. end	331.1 ± 21.5 (300-375)	–	–	–	–	–
PUS	14.6 ± 1.7 (11-17)	–	–	–	8.4 ± 0.3 (7.0-10.0)	–
Length spermatheca	14.2 ± 0.7 (13-15)	–	–	–	–	–
Width spermatheca	13.3 ± 0.6 (12-14)	–	–	–	–	–
Tail length	36.6 ± 4.4 (23-44)	17.9 ± 2.1 (13-21)	35.8 ± 2.75 (31-40)	20.4 ± 0.81 (19-21)	37.3 ± 1.5 (30-44)	31.1 ± 0.6 (29-36)
Anal/cloacal body diam.	9 ± 0.9 (8-11)	19.1 ± 1.0 (17-21)	–	–	–	–
Vulval width	16.5 ± 1.6 (13-20)	–	–	–	–	–
Spicule length	–	20 ± 1.9 (18-29)	–	22	–	20.8 ± 0.6 (18-23)
Ovary/Testis length	313.2 ± 6.5 (295-321)	245.6 ± 14 (218-266)	–	–	–	–
S-E pore	Close to pharyngo-intestinal junction or at the level of nerve ring	Close to nerve ring	Base of median pharyngeal bulb	Base of median pharyngeal bulb	*Much below pharyngo-intestinal junction	At the level of pharyngo-intestinal junction
Vulval lip	Lower lip swell out	–	Prominent	–	*Lower lip swell	–
Vagina	Posteriorly inclined	–	Straight or slightly anteriorly inclined	–	*Posteriorly inclined	–

Table 1 (continued). Measurements and characters of *Schistonchus caprifici* from *Ficus palmata* from India compared with previously published measurements and characters.

PUS	Equal to greater than VBW	–	–	–	*Less than VBW	–
Tail tip	Conoid, tapering towards tip but tip rounded	Short conoid, tip round	Conoid, tapering to conical process	Short conoid, round	*Conoid, sharply pointed tip	*Short conoid, tip pointed
Spicule	–	Condylus high, rounded and dorsal limb curved and its tip hook-like	–	Condylus well-developed, truncate, dorsal limb curved and its tip curved	–	*Condylus high, rounded and dorsal limb curved and its tip smooth
Gubernaculum	–	Triangular sclerotized str. at the mouth of cloacal aperture	–	Faintly visible	–	*Triangular
Spermatheca	Spheroid to elongate-ovoid	–	Rounded to elongate ovoid	–	–	–

Note: All measurements are in μm and in the form: mean \pm SD (range); *described based on illustrations only; – means either not applicable or available.

ICAR-IARI, New Delhi, India. The associated fig wasp species *Blastophaga* sp. indet. was identified with the help of Entomologist Dr Nithya in the Division of Entomology, ICAR-IARI.

Morphological characterisation of *Schistonchus caprifici* (Gasparrini, 1864) Cobb, 1927. Comparative measurements and morphological characteristics of males and females of *S. caprifici* from edible fig, *F. palmata* in India are presented in Tables 1, 2 (Supplemental material) and Figures 1, 2.

Male from figs. Body cylindrical, ventrally curved, and J-shaped when killed by gentle heat. Lateral lines not clearly marked; head slightly offset with round cap nearly one third high as wide at base without annulations. Cuticle with fine striations, lip diameter wider than lip height. Excretory pore 64 (52-84) μm from anterior end, close to base of nerve ring. Deirids not visible. Stylet medium in size and robust 19-24 μm in length and have rounded stylet knobs. Conus forming 45-54% of total stylet length, conus 9.5 (9-10) μm , and shaft length 11.3 (9-15) μm , stylet knobs height 2.0 (1.5-2.5) μm and diameter 2.9 (2-3) μm . Median pharyngeal bulb appeared rounded to oval, 53.9 (41-75) μm away from anterior end. Basal pharyngeal lobe 95.4 (68-119) μm long, maximum body diameter 16.7 (14-18) μm . Pharyngo-intestinal junction near to the base of the nerve ring. Testis outstretched or reflexed – 245.6 (218-266) μm in length from cloaca to anterior distal end of testis. Testis situated at 55.2% (42.8-69.8%) of body length from the anterior end. Cloacal body width 19.1 (17-21) μm . Spicules paired, strong, separate, rose-thorn shaped, arcuate. Condylus well developed, very stout, and conoid and rounded. Rostrum conical and rounded. Dorsal line of lamina smoothly symmetrically curved, its tip hook-shaped. Junction

of rostrum and calomus smooth. Spicule tip appears broad, no cucullus. Prominent gubernaculum, a short-sclerotised triangle shaped structure located towards cloacal aperture. Three pairs of subventral caudal papillae observed, one pair adcloacal (P3), one just posterior to mid-tail length (P3a) and one near tail-tip (P4). Tail conoid, 0.7-1.2 anal body widths long, tip rounded without mucro. Bursa or bursal flap absent.

Reproductive female from figs. Body almost straight, slightly ventrally arcuate when relaxed by gentle heat, tapering at both ends. Lip region wide, offset, without striations almost similar to male. Cephalic framework moderately to strongly sclerotised, lip height 2.8 (2-4) μm and diameter 7.2 (5-12) μm . Stylet 21.7 (19-24) μm long, weakly sclerotised stylet with small rounded knobs. Conus forming 45-59% of total stylet length. Length of conus 8.6 (9-11) μm and shaft 13.1 (8-15) μm . Knobs height 1.97 (1-2) μm and diameter 3.0 (2-3.5) μm . Secretory-excretory (S-E) pore at 72.2 (64-86) μm away from anterior end, posterior to median pharyngeal bulb, nearly close to the pharyngo-intestinal junction or at the level of nerve ring, lateral field observed but lateral lines not clearly marked or indistinct. Median pharyngeal bulb is rounded to oval; 60.8 (52-71) μm in length from anterior end, basal pharyngeal lobe 111.8 (70-144) μm long from anterior end. Maximum body diameter 14.7 (13-16) μm . Deirids not visible. Vulval position 331.1 (300-375) μm in length away from anterior end and vulva located at 55.5-84.1% of total body length. Vulval lips prominent, lower lip bulged out, vulval body width (VBW) 16.5 (13-20) μm . Vulval flap absent. Vagina straight or mostly posteriorly inclined. Ovary outstretched or reflexed, may reach up to middle of basal

pharyngeal lobe, ovary length 313.2 (295-321) μm . Monodelphic gonad, oocytes arranged in a single row. Spermatheca spheroid to elongated ovoid not offset 13-15 \times 12-14 μm . The post-vulval uterine

sac equal or greater than the width of the vulval body width, 14.6 (11-17) μm in length. Tail straight, conoid, round, 2.7-5.4 anal body widths long and tapering towards end, tip rounded.

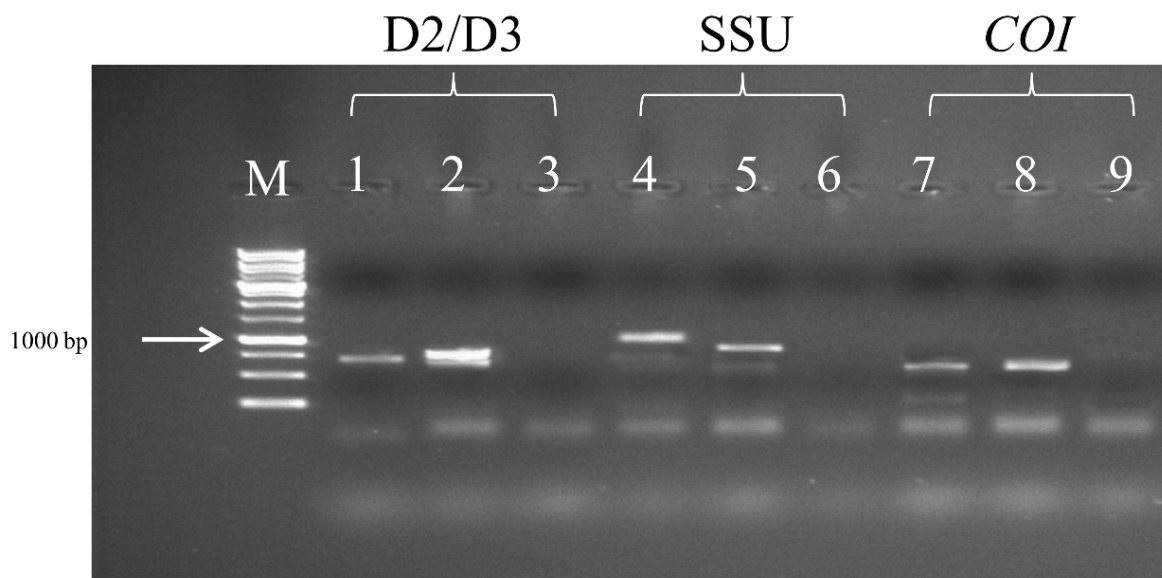


Fig. 3. PCR amplification of D2-D3, SSU and *COI* for *S. caprifici* (M-1 kb ladder, 1, 4, 7: Sample; 2, 5, 8: + ve control for respective markers; 3, 6, 9: – ve control for respective markers).

Entomophilic female from figs. Not found.

Host and locality. Male and reproductive females of *S. caprifici* from *Ficus palmata* growing at ICAR-IARI, New Delhi (28°39'25.9" N, 77°11'5.9" E; elevation 167 m a.s.l.) and figs inhabited by fig wasp species, *i.e.* *Blastophaga* sp. indet. were recovered from it.

Molecular characterisation and phylogenetic relationships. The amplified sequence fragments of D2-D3, SSU and *COI* were of 762, 817 and 687 bp size, respectively (Fig. 3). Phylogenetic relationship of *S. caprifici* with closely related nematode species were inferred by maximum likelihood method. The D2-D3 sequence was deposited in GenBank NCBI (accession number OK054582). BLAST search revealed that the D2-D3 sequence was 99.88-100% similar to D2-D3 sequences of other *S. caprifici* species. To reconstruct the D2-D3 phylogenetic tree, several previously sequenced populations as well as sequences generated by Gulcu *et al.* (2008) were used. The two most divergent Turkish sequences (EU287674 and EU287689) were also included (Gulcu *et al.*, 2008). The newly generated sequences had 100% identity with the sequences from the Turkey, Spain and Italy populations (*e.g.*, EU287643, EU287665, FN564936, EU287646). Similarly, 18S rDNA (SSU) and *COI* sequences were deposited in

GenBank NCBI with accession numbers OK047719 and OK067404. BLAST search for both the sequences revealed that they had 99.59% identity with the 18S rDNA (SSU) sequences and 94.08 to 99.85% with *COI* sequences of other *S. caprifici* isolates deposited in GenBank NCBI, respectively.

The ML phylogenetic tree of the available *Schistonchus* species based on D2-D3 sequences (Fig. 4) showed that the newly generated sequences clustered with *S. caprifici*. The closest sequence neighbours were *S. caprifici* species from Turkey (EU287645, EU287646, EU287654, EU287665, EU287670, EU287676, EU287689). Similarly, the phylogenetic tree reconstructed by using the 18S rDNA and *COI* sequences indicated that the Delhi population of *Schistonchus* sp. matched closely with the *S. caprifici* from Turkey, Italy and Spain (*e.g.*, GU190764, GU190763, FN564938, FN564940) for SSU (Fig. 5). The sequences showed similar evolutionary trend across all the tree reconstruction algorithms. Substantial sequence divergence for the three tested molecular phylogenetic markers distinguished *S. caprifici* from the other studied *Schistonchus* species. To the best of our knowledge, this is the first study and molecular phylogenetic characterisation of this species from India.

DISCUSSION

The aphelenchid nematode, *Schistonchus caprifici* has its association with edible fig fruit (*Ficus carica*) and other hosts including insects (Vovlas *et al.*, 1992; Hunt, 1993; Vovlas & Larriza, 1996; Bajaj & Tomar, 2014; Kolaei *et al.*, 2016; Namjoo *et al.*, 2019; Fard & Zare, 2020). The genus *Schistonchus* comprises 14

species, of which five species (*S. hispida* Kumari & Reddy, 1984, *S. racemosa* Reddy & Rao, 1986, *S. osmani* Anand, 2002, *S. flagellobenghalensis* Bajaj & Tomar, 2014, *S. mucroracemosus* Bajaj & Tomar, 2014) have been described from the states of Haryana, Andhra Pradesh, Karnataka and Uttar Pradesh of India. *S. caprifici* was found in fig wasp (*Blastophaga psenes*), and the fig fruit (*Ficus carica*)

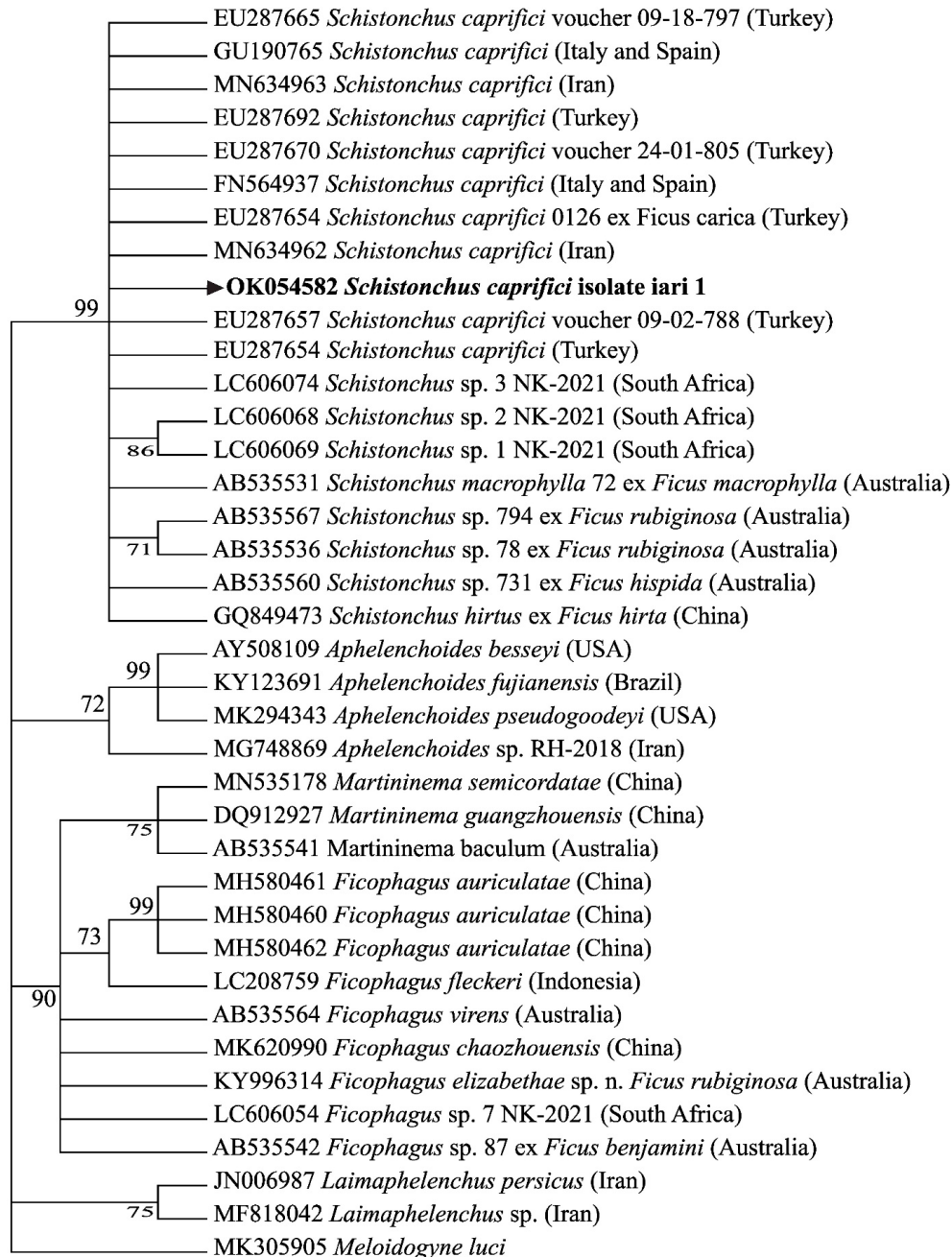


Fig. 4. Molecular phylogenetic analysis of *Schistonchus caprifici* using the D2-D3 (LSU) marker. The evolutionary history was inferred by Maximum Likelihood method using Kimura two-parameter model (K2P) with gamma-distributed rate variation across sites (K2P + G).

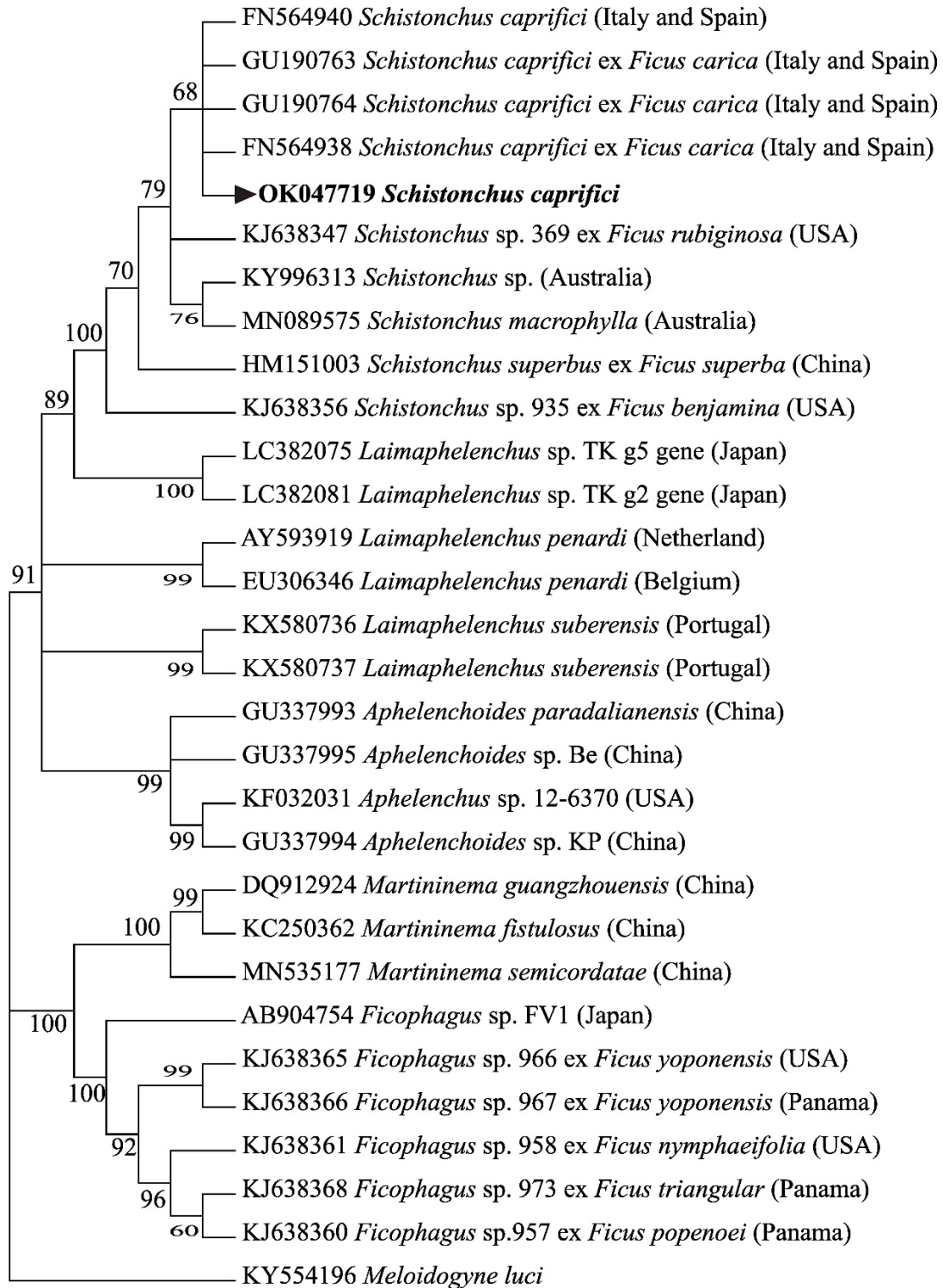


Fig. 5. Molecular phylogenetic analysis of *Schistonchus caprifici* using the 18S rDNA (SSU) marker. The evolutionary history was inferred by Maximum Likelihood method using Kimura two-parameter model (K2P) with gamma-distributed rate variation across sites (K2P + G).

from Haryana (Bajaj & Tomar, 2014). We recovered *S. caprifici* from the sycones of the Punjab fig (*Ficus palmata*) from Pusa Campus, ICAR-IARI, New Delhi, India and confirmed its identification and described it further. Gasparini (1864) described *S. caprifici* from *B. psenes*, an Agaonidae insect pollinator for commercial figs, and Cobb (1927) redescribed it from an Algerian population. Later more detailed descriptions were given by Thorne (1961) and Vovlas *et al.* (1992) from the sycones of *Ficus carica*. However, the *S. caprifici* showed variations in its morphology and morphometrics. The present study is a more detailed investigation on *S. caprifici* population recovered from the *Ficus palmata* and compared with the previous descriptions of Thorne (1961), Vovlas *et al.* (1992), Kolaei *et al.* (2016), and Bajaj and Tomar (2014). The morphological and morphometric observations are consistent with the description of Indian populations by Bajaj and Tomar (2014) and also with the previous descriptions with some deviations in certain parameters. Morphometrics of males differs from Italian populations in having larger testis length (55 vs 43 μm) and shorter tail length (17.9 vs 31.1 μm). Females are relatively thicker (body diam. 14.7 vs 12.8 μm) than that of the Italian population. This population also differs from the female population of Iran (Kolaei *et al.*, 2016) in having shorter conus length (9.4 vs 12.6 μm), and longer PUS (14.6 vs 9.8 μm). According to Bajaj and Tomar (2014) description of *S. caprifici*, the species has a spicule with a well-developed and truncated condylus, a faintly visible gubernaculum in the male, and a slightly anteriorly inclined or straight vagina with a tail that tapers to a conical process in the female. Later Kolaei *et al.* (2016) described a gubernaculum-like triangular structure below the spicules in male and sharp conical and mucronate tail in female. Our morphological observations are almost similar to the illustration of males and females of Thorne's (1961) description but they differ in having the larger PUS (equal to greater than one VBW vs less than one VBW), and female tail shape (tapers towards the end with rounded tip vs sharply pointed tip). The illustration of female in Thorne's (1961) description shows a small PUS and sharply pointed tail. We observed a large number of specimens of *S. caprifici*, but the similar tail shape was not seen. Based on our observations, this species has distinctive characters of the genus *Schistonchus*, *i.e.*, strongly developed stylet with prominent knobs, a sclerotised triangular shape gubernaculum located at the mouth of cloacal aperture, spicule with high conoid and rounded

condylus, short conoid rostrum, and a hook-like ventrally curved tip in male, and straight slender body, vagina downwardly inclined with lower lip swell outside, PUS equal to greater than one VBW, tail tapers towards the end, terminus smooth rounded and no conical process or mucro in female. This study has also included additional morphometric parameters for further characterisation of the species. In addition, the present descriptions are based on 30 specimens of male and female each and, thus these morphometric variations are more reliable for species delimitation and a better understanding of the species. A tabular key of *Schistonchus* species described across the world is provided as a ready reference for easy comparison and diagnosis of the species.

In addition to morphology and morphometrics, molecular data were used to assess phylogenetic relations of *S. caprifici* from India with that of Turkey, Italy and Spain. The molecular characters of this population were studied using partial sequences of LSU, SSU and *COI* for the first time in India. In comparison with the previously available data, no remarkable genetic distance was found between the currently studied isolate and other *S. caprifici* isolates from Turkey, Spain and Italy.

In India, more than 5,600 ha are under fig cultivation mostly confined in Maharashtra, Gujarat, Karnataka, Uttar Pradesh and Tamil Nadu and the occurrence of fig nematodes has not yet been fully investigated to understand its damage potential. In India *S. caprifici* are encountered in large numbers in edible fig fruits from Haryana and Delhi. The present study indicates its association with edible Punjab fig (*F. palmata*), an important fruit tree as well as shrub grown in India. It is evident from previous studies that *S. caprifici* being plant parasites can cause histological alterations to infected figs (Vovlas *et al.*, 1992; Vovlas & Larriza, 1996), and thus it might cause substantial damage in the commercial cultivation of edible figs in India.

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SUPPLEMENTAL MATERIAL

Table 2. Tabular key for the globally known species of *Schistonchus* (Cobb, 1927) Fuchs, 1937. – [http://russjnematology.com/Articles/rjn311/Paper8_Waghmare-\(Table2\)_FINAL-on-line-\(SUPPL\).pdf](http://russjnematology.com/Articles/rjn311/Paper8_Waghmare-(Table2)_FINAL-on-line-(SUPPL).pdf).

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C.D. Waghmare, M.R. Khan and V.S. Somvanshi. Характеристика *Schistonchus caprifici* (Gasparrini, 1864) Cobb, 1927 из *Ficus palmata* с табличным ключом для всемирно известных видов. **Резюме.** Популяция *Schistonchus caprifici* (Gasparrini, 1864) Cobb, 1927 была выделена из сикониев *Ficus palmata*, выращенного в Нью-Дели (Индия). Популяцию идентифицировали на основании морфологических и молекулярных признаков. Установлено, что морфологические и морфометрические признаки популяции *Schistonchus* из *Ficus palmata* соответствуют первоначальному и последующим описаниям *S. caprifici*. Это всестороннее морфологическое исследование *S. caprifici* из Индии выявило дополнительную информацию о морфологических и морфометрических признаках, таких как форма губ, стилет, положение вульвы относительно переднего конца тела, сперматека, анальный/клоакальный диаметр тела, структура вульвы и длина яичника/семенника. Наряду с некоторыми другими морфологическими описаниями, изменения внесены в следующие признаки: форма спикул и губернакулума у самцов, половые губы и вагинальные линии, задний маточный мешок, форма хвоста и терминуса. Идентификация этой популяции была дополнительно подтверждена с использованием последовательностей D2-D3 участка 28S рРНК (LSU), 18S рРНК (SSU) и *COI* мтДНК, и установлено ее филогенетическое родство с *S. caprifici* из Турции, Испании, Италии и другими видами со всего мира. Табличный ключ-определитель для видов рода *Schistonchus* также представлен.
