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Description of *Amphibelondira sindhicus* N. Sp. with Observation on *Belondira paraclava* Jairajpuri, 1964 (Family Belondiridae) from Sindh, Pakistan

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ABSTRACT

A new and a known species belonging to the family Belondiridae are described from Sindh, Pakistan. *Amphibelondira sindhicus* n. sp., is characterized by having reduced anterior genital branch, more slender body, smaller neck length and expanded part of pharynx, smaller spicule and clavate tail with distinct radial striae. *Belondira paraclava* Jairajpur, 1964 is reported for the first time from Pakistan during the present studies on cotton.

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Authors' Contribution
AAN conducted the survey, collected samples and processed the samples. NK helped in taxonomy of the species and provided technical assistance in writing results and discussion. SF helped in identification of the species.

Key words
***Amphibelondira sindhicus* n. sp., *Belondira paraclava*, Cotton, Sindh, Pakistan**

INTRODUCTION

Rahman *et al*. (1986) proposed the genus *Amphibelondira* with *A. bhutanensis* as its type species from Bhutan in the family Belondiridae. Another new species *A. bongersi* was described by Shaheen and Ahmed (2005) from Costa Rica. More recently, another new species *A. wasimi* n. sp. have been described by Roy *et al*. (2017). The present paper proposed a new species *Amphibelondira sindhicus* making a total of four valid species under this genus.

During the course of present survey of cotton (*Gossypium hirsutum* L.) soil samples collected from five districts of Sindh, mainly from Umerkot, Tando Allahyar, Sanghar, Mirpurkas and Matiari yielded several populations belonging to the genus *Amphibelondira* Rahman *et al*. (1986) and *Belondira* Jairajpuri (1964). On detailed study, they were found to represent a new species and a new record species described, redescribed and illustrated herein.

MATERIALS AND METHODS

Nematodes were extracted from the soil by Cobb's decanting and sieving method (Cobb, 1918) followed by modified Baermann's funnel technique (Baermann, 1917). The nematodes were killed on a hot plate at 60-70º C. The killed nematodes were instantly fixed in TAF for 24 hours (Courtney *et al*., 1955). The samples were then washed thrice with distil water, 2 ml of 1.25% glycerine was added to the cavity and placed it in incubator at 55º C for 5-6 days. Nematodes were mounted in pure glycerin on a glass-slide. Measurements were taken with an ocular micrometer and illustrations were made with the help of a drawing tube attached to the compound microscope. Photomicrographs were taken with a Nikon DS-Fi1 camera attached to the same microscope.

Amphibelondira sindhicus n. sp.

(Figs. 1 and 2 A-F)

***Measurements*:** See Table I

Description

Female

Body almost straight to slightly curved ventrally after fixation, tapering towards anterior end. Cuticle finely striated. Cuticle 0.98 µm thick at mid body and 9 µm on tail. Lateral chords about one fourth as wide as body width at mid body. Body pores indistinct. Lip region offset by slight depression, lips amalgamated amphid stirrup shaped, their aperture about as wide as lip region width. Odontostyle 1.6-1.8 times lip region widths long its aperture about one fourth of its length. Guiding ring single, located at twice lip region width from anterior end. Odontophore slightly longer then odontostyle length. Nerve ring at 80-90 µm from anterior end of body.

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Table I. Measurement of *Amphibelondira sindhicus* n. sp. (All measurements in µm except L).

Morphological characters	Holotype ♀	Paratype females 3 ♀♀	Paratype male 1 ♂
L (mm)	1.23	1.21±0.01 (1.20-1.23)	0.99
a	61.6	59.7±1.75 (57.4-61.6)	52.3
b	6.4	6.3±0.08 (6.2-6.4)	5.0
c	56	55.2±0.52 (54.8-56)	45.1
c`	1.46	1.41±0.03 (1.37-1.46)	1.46
V	51.0	51.2 ±0.2 (51-51.5)	-
Lip region width	3	2.8 ±0.23(2.5-3.5)	5
Lip region height	5	5.1 ±0.62 (4.5-6)	4
Amphid aperture	3	2.6 ±0.23 (2.5-3.0)	3
Odontostyle length	5	4.83 ±0.23(4.5-5)	5
Odontophore length	8	7.6 ±0.47(7-8)	8
Guiding ring from anterior end	7	6.6 ±0.47 (6-7)	7
Nerve ring from anterior end	80	85 ± 4.08 (80-90)	74
Neck length	190	190 ±0.47 (190-191)	198
Expanded part of pharynx	70	68.3±1.69 (66-70)	68
Cardia length	6	5.83 ±0.62 (5-6.5)	9
Body width at neck base	20	21±0.81(20-22)	19
Body width at anus/cloaca	15	15.3 ±0.47 (15-16)	15
Vaginal depth	10	9.6 ±0.47 (9-10)	-
Vulva from anterior end	630	624± 4.3 (620-630)	-
Prerectum length	76	73.6 ±2.6 (70-76)	-
Rectum length	20	19±0.81 (18-20)	-
Tail length	22	21.8 ±0.23 (21.5-22)	-
Spicule length	-	-	28
Lateral guiding pieces	-	-	6

Basal expanded portion of pharynx occupying about 34-36% of pharyngeal length expanding gradually enclosed in a thick sheath of sinistrally spiral muscles. Cardia 5-6.5 µm long, oval shaped, occupying 25-29% of corresponding body width. pharyngeal gland nuclei distinct. Reproductive system amphidelphic, anterior gonad reduced, posterior gonad longer then anterior genital branch. Ovaries reflexed, anterior ovary reaching up to vulva. Oocytes arranged in a single row except near tip region. Vulva transverse, vagina extending inward occupying about 50% of the corresponding body width. A distinct spincter at oviduct-uterus junction in posterior genital system. Prerectum 4.6-4.7 anal body width long. Rectum 1.2-1.25 anal body width long. Tail short, rounded, 1.3-1.4 anal body width long, with a hyaline part occupying 37-40% of total tail length. A pair of caudal pores on each side.

Male

Body almost similar in shaped with female except for posterior region being slightly more curved ventrad. Cardia elongated, 0.4 times long of corresponding body width. Supplements consisting only of an adanal pair, no ventromedian supplements. Spicule dorylaimoid, 1.8 anal widths long. Lateral guiding pieces present 6-8 µm long.

Type habitat and locality

Specimens were collected from the rhizospheric region of cotton (*Gossypium hirsutum* L.) from Umerkot, Sindh, Pakistan.

Type specimens

Holotype female, three paratype females and a paratype male deposited in the National Nematode Collection of National Nematological Research Center, University of Karachi, Karachi Pakistan.

Etymology

The species name refers to the location of samples collected from province Sindh.

Diagnosis and relationship

Amphibelondira sindhicus n. sp., is distinguished from other species of the genus *Amphibelondira* Rehman *et al*. (1986) by having reduced anterior genital branch, more slender body, smaller neck length and expanded part

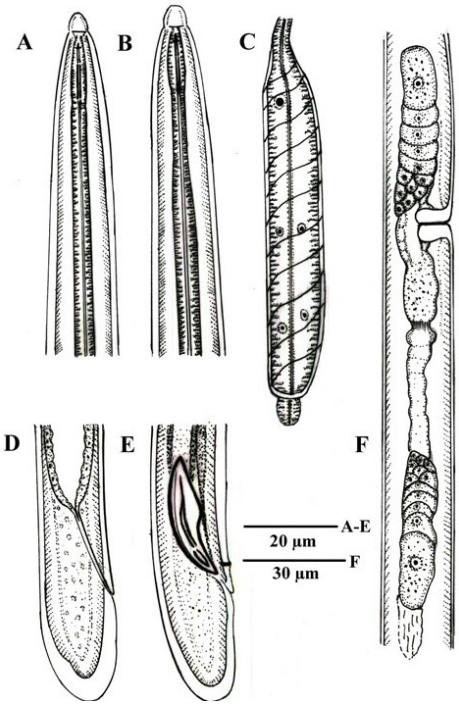

Fig. 1. (A-F). *Amphibelondira sindhicus* n. sp. A. Anterior region of female; B. Anterior region of male; C. Expanded part of pharynx; D. Female tail; E. Male tail; F. Vulval region.

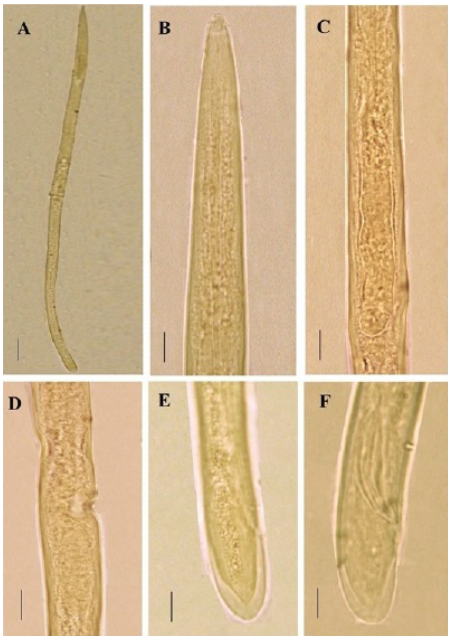

Fig. 2. (A-F). *Amphibelondira sindhicus* n. sp., A. Whole body of female; B. Anterior region; C. Expanded part of pharynx; D. Vulval region; E. Female tail; F. Male tail (Scale : A=10µm; B-F=100µm).

of pharynx and smaller spicules. It resembles all the three described species by having similar body shape, slightly offset lip region, gradually expanded pharynx, male with an adanal pair of supplements, no ventromedians, and bluntly rounded tail in both the sexes. But differs from *A. bhutanensis* Rehman *et al*. (1986) in having short body and neck length (1.20-1.23 vs 1.38 mm; 190-191 vs 240 µm, respectively). Values of a, b, c and c`, shorter odontostyle and odontophore (a= 57.4-61.6 vs 36; b= 6.2-6.4 vs 5.7; c= 54.8-56 vs 76.6; c`= 1.3-1.4 vs 0.6; odontostyle= 4.5-5 vs 14 µm; odontophore= 7-8 vs 16 µm) and shorter spicule (28 vs 44 µm). The present species differs from *A. bongersi* Shaheen and Ahmed (2005) in having short neck length (190-191 vs 198-225 µm), values of a, b, c` shorter odontostyle and odontophore (a= 57.4-61.6 vs 34.5-40.5; b= 5.2-6.4 vs 4.9-5.5; c`= 1.37-1.46 vs 0.86-1.05; odonostyle= 4.5-5 vs 12.5 µm; odontophore= 7-8 vs 20.5-23.5 µm), and spicule present vs absent. The new species also differs from *A. wasimi* Roy *et al*. (2017) from shorted body size (L=1.2-1.23 vs 2.25mm), values a, b, c, shorter odontostyle and odontophore (a= 57.4-61.6 vs 51; b= 6.2-6.4 vs 4.3; c= 54.8-56 vs 70.83; odontostyle= 4.5-5 vs 15.8 µm; odontophore= 7-8 vs 23.7 µm), shorter spicule (28 vs 34.3 µm); shorter rectum and tail length (rectum= 18-20 vs 41.6 µm; tail length= 21-22 vs 31.8 µm, respectively).

***Belondira paraclava* Jairajpuri, 1964**

(Fig. 3 A-E; Fig. 4 A-G)

***Measurements*:** See Table II

Description

Female

Body almost straight or slightly curved ventrally after fixation. Cuticle finely striated. Lateral chords about one third body width at mid body. Pores are indistinct. Lip region continuous with body. Cephelic framework faintly sclerotized. Amphid stirrup shaped, their aperture about 0.66 times as wide as lip region width. Odontostyle with distinct lumen, about 0.8-1.0 times lip region width long. Guiding ring obscure. Odontophore linear 1.3-1.4 times the odontostyle length. Anterior part of pharynx slender, enlarging gradually, basal expansion occupying 36-38% of total neck length and enclosed in a spiral muscular sheath. Nerve ring encircling the anterior part of pharynx at 35-36% of neck length from anterior end. Cardia spherical to cylindroid. Genital system mono-opisthodelphic, anterior branch represented by a short uterine sac 1.2-1.6 times the corresponding body width long, posterior branch well developed, ovary reflexed, with oocytes arranged in a single row except near tip. Vagina extending inwards about half of the corresponding body width with almost straight

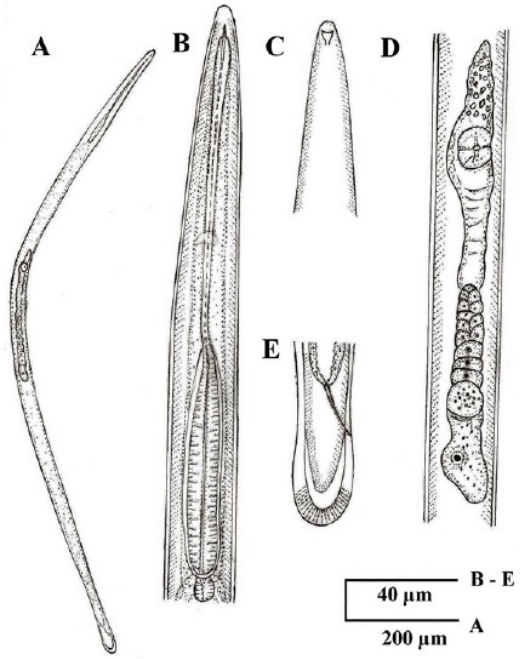

Fig. 3. (A-E). *Belondira paraclava* Jairajpuri, 1964. A. Whole body; B. Pharyngeal region; C. Anterior region showing amphid; D. Vulval region; E. Tail region.

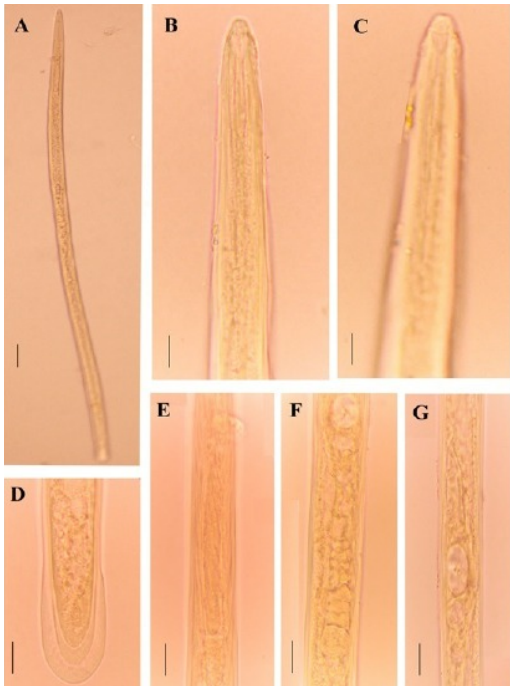

Fig. 4. (A-G). *Belondira paraclava* Jairajpuri, 1964. A. Whole body; B. Pharyngeal region; C. Anterior region showing amphid; D. Tail region; E. Expanded part of pharynx; F. Female gonad; G. Post uterine sac (Scale: A=10µm; B-G= 100µm).

Table II. Measurements of *Belondira paraclava* Jairajpuri, 1964. All measurements are in µm (except L).

Morphological characters	Female (n = 8)	
	Mean±SD	Range
L (mm)	1.16±0.02	1.12-1.21
a	45.2±2.23	42.5-48.8
b	5.7±0.33	5.2-6.1
c	41.1±1.71	38.5-43.5
c`	1.30±0.11	1.12-1.5
V	37.7±0.89	36.7-39.5
Lip region width	6±0	6
Lip region height	4±0.53	3-5
Amphid aperture	4.2±0.45	4-5
Odontostyle length	5.1±0.34	5-6
Odontophore length	7.2±0.45	7-8
Guiding ring from anterior end	7.8±0.83	7-9
Nerve ring from anterior end	73.4±3.49	70-80
Neck length	203.5±10.4	192.5-224
Expanded part of pharynx	76.3±5.1	70-85
Cardia length	8.1±0.98	7-9
Body width at mid body	25.4±1.38	24-24.5
Body width at neck base	24.7±1.57	22-27
Body width at anus	20.4±0.60	20-21.6
Vaginal depth	15.7±1.24	14-18
Vulva from anterior end	423.0±18.2	395-444
Prerectum length	42.8±3.35	40-50
Rectum length	20.2±1.27	18-22
Tail length	27.2±2.55	22.5-30.4
PUS (Post uterine sac)	44±5.7	30-44

walls; vulva a transverse slit. Prerectum 2-2.3, rectum 0.9-1.0 anal body width long. Tail clavate with cuticle expanded and marked with radial striation, caudal pores two pairs.

Male

Not found.

Remarks

Jairajpuri (1964) described *Belondira paraclava* from Saharanpur, India. The present population from Sanghar, Sindh agrees well with the original one as well as the description given by Naz and Ahmed (2009) except for having slightly shorter expanded part of pharynx 70-85 vs 90-115 µm and cardia 7-9 vs 10-14 µm. *B. paraclava* is a new record species from Pakistan.

CONCLUSION

One new species of soil nematode viz., *Amphibelondira sindhicus* n. sp., along with one new record species viz., *Belondira paraclava* (Jairajpuri,1964) identified from this survey. This study will help further for the management of the cotton soil nematodes.

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Statement of conflict of interest

The authors declare that, there is no conflict of interests regarding the publication of this article.

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