



## Research Article

# TWO NEW RECORDS OF FREE-LIVING MARINE NEMATODES (NEMATODA: ENOPLIDA: IRONIDAE AND MONHYSTERIDA: XYALIDAE) FROM INDIA

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## ABSTRACT

Two species of free-living marine nematodes were collected from the intertidal areas of East Coast of India namely, *Conilia sinensis* Chen and Guo, 2015 and *Corononema parvum* Nicholas and Stewart, 1995, both species are new records for the Indian fauna. Previously, the genus *Conilia* Gerlach, 1956 was known from Santos Beach, South Japan, East China Sea and Korea. The genus *Corononema* Nicholas and Stewart, 1995 was known only from Australia and Thailand and very recently reported from off the coast of Vietnam. Both the genera are being reported for the first time from India. The species are described and photomicrographed.

**Keywords:** *Conilia sinensis*, *Corononema parvum*, Distribution, East Coast of India, Morphology.

## INTRODUCTION

Free-living marine nematodes are ubiquitous and numerous organisms, accounting for four out of every five aquatic multicellular organisms that live at the planets surface (Bongers and Ferris, 1999). The members of this phylum occur in very high abundance within an environment (Lambsead, 2004; Danovaro *et al.*, 2008) and thought to be hyper diverse (Appeltans *et al.*, 2012). The order Enoplida Filipjev, 1929, comprises 7 suborders, amongst which the suborder Ironina Siddiqi, 1983, consist of one superfamily Ironoidea de Man, 1876, which further consists of 3 families; Ironidae de Man, 1876, Leptosomatidae Filipjev, 1916 and Oxystominidae Chitwood, 1935. Family Ironidae is further divided into two subfamilies; Ironinae de Man, 1876 and Thalassironinae Andrassy, 1976. The genus *Conilia* Gerlach, 1956 belongs to the subfamily Thalassironinae. Genus *Conilia* consists of 3 valid species (Website Nemys: World Database of Nematodes; Ahyong *et al.*, 2023), *C. divina* Gerlach, 1956 was described from sandy beach in the Santos area; *C. monospiculata* Aryuthaka, 1989 from Amakusa, South Japan and *C. sinensis* Chen and Guo, 2015

from Dongshan Island, East China Sea. Recently, *C. sinensis* is reported from the west coast of Korea (Lee *et al.*, 2023).

The order Monhysterida comprises three superfamilies: Siphonolaimoidea, Monhysteroidea and Sphaerolaimoidea. Superfamily Siphonolaimoidea, includes three families, Siphonolaimidae, Fusivermidae and Linhomoidae, while Monhysteroidea contain a single family, Monhystoridae, whereas Sphaerolaimoidea comprises two families, Sphaerolaimidae and Xyalidae (Fonseca and Bezerra, 2014). Subfamily Coronematinae under the family Xyalidae, was erected by Nicholas and Stewart, 1995. The genus *Corononema* include only three valid species from Australia, Thailand and off the coast of Vietnam namely, *C. parvum* Nicholas and Stewart, 1995, *C. thai* Nicholas and Stewart, 1995 and *C. vulgare* Phuong *et al.*, 2023 respectively.

The present study was undertaken to report species of free-living marine nematodes from the Indian coastal regions that were not recorded previously. *Conilia sinensis* Chen and Guo, 2015 and *Corononema parvum* Nicholas

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and Stewart, 1995 forms new country records. Both the species are described and photomicrographed.

## MATERIALS AND METHODS

Sediment samples were collected from Digha ( $21^{\circ}37.636' N$ ,  $87^{\circ}32.216' E$ ) in the East Medinipur district of West Bengal (samples collected in April 2023), Sagar Island ( $21^{\circ}38.061' N$ ,  $88^{\circ}04.095' E$ ) in the South 24 Parganas district of West Bengal (samples collected in July 2023) and Karaikal medu ( $10^{\circ}56'42.98'' N$ ,  $79^{\circ}51'10.81'' E$ ) in the Karaikal district of Puducherry (samples collected in September 2021). The sediment samples were collected with the help of hand corer. The samples were fixed with 4% Formalin in filtered sea water. Following fixation, the samples were stained using Rose Bengal stain (Williams and Williams, 1974). The fixed samples were brought to the laboratory and were sieved using 2 mm Coarse sieve to remove the larger particles followed by sieving in 53  $\mu m$  mesh sieve to retain the nematodes. The sieved residue was examined under SZX-16 Olympus Microscope and nematode specimens were picked out in Glycerin-Alcohol and kept in desiccator containing anhydrous Calcium Chloride for dehydration. After proper dehydration specimens were mounted on a drop of glycerin on glass slides and sealed by paraffin wax ring. Specimens were identified under BX-53 DIC Olympus research microscope and photomicrographed with DP27 camera. The specimens have been deposited in the National Zoological Collections of Nemathelminthes section, Zoological Survey of India, Kolkata.

## Abbreviations

*a*= Body Length/ maximum body diameter

*b*= Body length/ pharynx length

*c*= Body length/ tail length

*c'*= Tail length/anal body diameter

*V%*= Distance from anterior end to vulva  $\times 100/$  Body Length

*abd*= Anal body diameter

*vbd*= Vulval body diameter

*cbd* = corresponding body diameter

## RESULTS AND DISCUSSION

*Conilia sinensis* Chen and Guo, 2015

Phylum : Nematoda Cobb, 1932

Class : Enoplea Inglis, 1983

Subclass : Enoplia Pearse, 1942

Order : Enoplida Filipjev, 1929

Suborder : Ironina Siddiqi, 1983

Superfamily : Ironoidea de Man, 1876

Family : Ironidae de Man, 1876

Subfamily : Thalassironinae Andrassy, 1976

Genus : *Conilia* Gerlach, 1956

*Conilia sinensis* Chen and Guo, 2015

(Figures 1-2 & Table 1).

## Materials examined

Slide reference number ZSI-HQ/NZC/ *Conilia sinensis* / WN.4080, 1 ♂, 2 ♀; Digha, East Medinipur district, West Bengal;  $21^{\circ}37.636' N$ ,  $87^{\circ}32.216' E$ ; 29 April. 2023; WN.4210, 3 ♂, 1 ♀; Sagar Island, South 24 Parganas, West Bengal;  $21^{\circ}38.061' N$ ,  $88^{\circ}04.095' E$ ; 25 July. 2023.

## Description

Measurements – See Table 1

### Male

Body shape slender and cylindrical (Figure 1c), measures 1507-1663  $\mu m$ ; maximum body diameter 15.74-18.63  $\mu m$ . Head end blunt; set off from body by a strip (Figure 1a, 2b). Well-developed lip region with swelling on both sides; wrinkles present on the outside wall. Six papilliform inner labial sensilla, six outer labial setae. Four cephalic setae arranged in a ring, stout and blunt. Buccal cavity with two parts; a cup-shaped anterior part and a posterior tube, strong and cuticularized (Figure 1a). The border of cup-shaped anterior part of buccal cavity consists of three solid curved claw-like teeth of almost equal size (Figure 1a, 2a). A row of small cuticular denticles present at the anterior edge of the cup-shaped part. Amphids not observed. Anterior part of the pharynx with peribuccal swelling, posterior part without any obvious bulb (Figure 1b). Nerve ring present. Tail conico- cylindrical, ventrally bent with one ventro-median swelling in the middle of the tail (Figure 1d, 2f). Small terminal spinneret. Three caudal glands observed. Smooth cuticle. Somatic setae observed in few specimens. Diorchic reproductive system; two testes arranged in tandem to the right of intestine. Spicule single and elongated; transversely or slightly obliquely striated (Figure 1d, 2c). Telamons paired; with thickened antero-ventral and postero-dorsal ribs, distally bent with two strong hook-like structures and also with rounded proximal projection, slightly curved, thin strip like gubernaculum present. Single precloacal supplement as simple ventral swelling (Figure 2e).

### Female

General characteristics similar to males, body slightly plumper than males; measures 1406-1627  $\mu m$ , maximum body diameter 19.95-25.78  $\mu m$ . Tail straight and without any setae or swelling (Figure 2g). Didelphic reproductive system with two reflexed, equally developed ovaries.

## Remarks

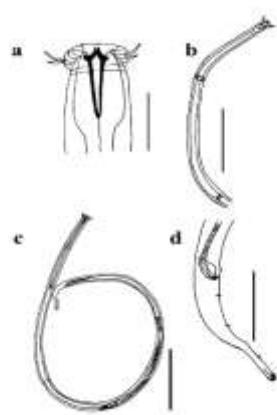
The genus *Conilia* is characterized by having elongated cuticularized tubular buccal cavity, three claw-like teeth at

the anterior end of the buccal cavity and the species under this genus, can be distinguished from the other genera of the family Ironidae by the male copulatory apparatus (Chen and Guo, 2015). *Conilia sinensis* is distinguished from the other species in having a relatively longer body length; lips well developed, the number, shape and the length of spicule; shape of telamon and the number of supplements (Chen and Guo, 2015). The present specimens of *C. sinensis* conforms well with the original specimens in the copulatory apparatus present in males, absence of

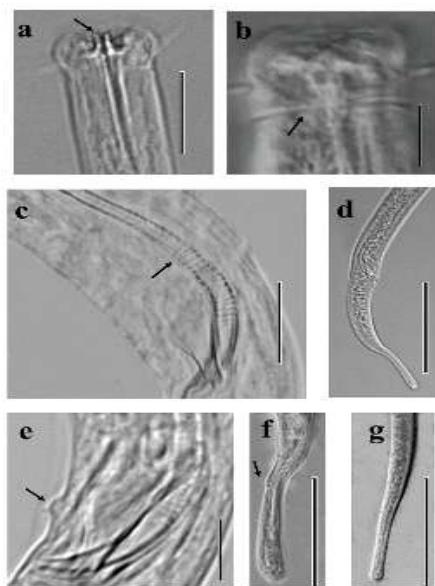
amphideal fovea, presence of single precloacal supplement, single ventral swelling in the middle of the ventrally bent tail in males. However, the present specimens differ from the original description in smaller body length (1507-1663  $\mu\text{m}$  vs 1956-2399  $\mu\text{m}$  in males; 1406-1627  $\mu\text{m}$  vs 1883-2080  $\mu\text{m}$  in females), shorter head diameter (12.87-14.63  $\mu\text{m}$  vs 25-26  $\mu\text{m}$  in males; 14.17-16.34  $\mu\text{m}$  vs 29-33  $\mu\text{m}$  in females) and shorter pharynx (252-315  $\mu\text{m}$  vs 380-423  $\mu\text{m}$  in males; 282-296  $\mu\text{m}$  vs 397-444  $\mu\text{m}$  in females).

**Table 1.** Measurements of *Conilia sinensis* Chen and Guo, 2015. All measurements are in  $\mu\text{m}$  and brackets indicate the mean values.

| Characters                            | Sagar Island population |        | Digha population |                        |
|---------------------------------------|-------------------------|--------|------------------|------------------------|
|                                       | Males                   | Female | Male             | Females                |
| n                                     | 3                       | 1      | 1                | 2                      |
| Body length                           | 1507-1663.3 (1582.59)   | 1486.3 | 1611.24          | 1406.66-1627 (1516.83) |
| Head diameter                         | 12.87-14.63 (13.65)     | 16.19  | 14.18            | 14.17-16.34 (15.25)    |
| Length of outer labial cephalic setae | 10.01- 12.36 (11.2)     | 10.86  | 10.76            | 10.35-11.28 (10.82)    |
| Length of cephalic setae              | 5.85-10.2 (7.45)        | 7.81   | 5.05             | 5.59-7.51 (6.55)       |
| Total length of buccal cavity         | 22.41-27.22 (25.31)     | 25.66  | 38.9             | 28.92-38.67 (33.79)    |
| Nerve ring from the anterior end      | 136.77-152.33 (142.73)  | 132.5  | 110.2            | 120.3-150.04 (135.17)  |
| Nerve ring cbd                        | 16.45-19.25 (18.2)      | 24.87  | 18.2             | 17.3-18.35 (17.83)     |
| Pharynx length                        | 286.97-315.19 (302.72)  | 282.4  | 252.4            | 208-296 (252)          |
| Pharynx cbd                           | 16.37-19.03 (17.67)     | 21.5   | 16.52            | 21.4-21.66 (21.53)     |
| Maximum body diameter                 | 15.74-18.09 (17.10)     | 20.42  | 18.63            | 19.95-25.78 (22.87)    |
| abd                                   | 15.54-17.63 (16.41)     | 18.8   | 13               | 13.5-16.6 (15.05)      |
| Tail length                           | 96.25- 111.65 (103.13)  | 116.5  | 104              | 98-104.5 (101.25)      |
| c'                                    | 5.45-7.18 (6.32)        | 6.19   | 8                | 5.90-7.74 (6.82)       |
| Spicule length as chord               | 46.82-53.4 (49.50)      | -      | 55.63            | -                      |
| Spicule length as arc                 | 58.4-62.33 (60.15)      | -      | 69.5             | -                      |
| Supplement                            | 1                       | -      | 1                | -                      |
| Length of telamon                     | 17.7- 21.48 (19.64)     | -      | 19.66            | -                      |
| Length of gubernaculum                | 20.3- 24.87 (21.79)     | -      | 17.5             | -                      |
| V' (from the ant end)                 | -                       | 957.14 | -                | (942)                  |
| vbd                                   | -                       | 20.5   | -                | 19-23 (21)             |
| V (%)                                 | -                       | 64.39  | -                | 58.02-66.82 (62.42)    |
| a                                     | 90.24- 95.74 (92.64)    | 72.78  | 86.49            | 54.56-81.55 (68.05)    |
| b                                     | 5.15-5.27 (5.22)        | 5.26   | 6.38             | 5.49-6.76 (6.12)       |
| c                                     | 13.49- 16.38 (15.42)    | 12.75  | 15.49            | 14.35-15.56 (14.96)    |



**Figure 1.** *Conilia sinensis* Chen and Guo, 2015. (WN.4210/ *Conilia sinensis*) a. Male, teeth, buccal cavity and strip.b. Male, anterior end. c. Male, whole body. d. Male, spicule and tail region. Scale bars: a= 20  $\mu\text{m}$ ; b= 100  $\mu\text{m}$ ; c= 200  $\mu\text{m}$ ; d= 50  $\mu\text{m}$ .



**Figure 2.** *Conilia sinensis* Chen and Guo, 2015 (WN.4080/ WN4210/ *Conilia sinensis*) (LM Photographs). **a**. Male, teeth. **b**. Male, head region with strip **c**. Male, spicule and telamon. **d**. Male, posterior end. **e**. Male, precloacal supplement. **f**. Male, tail with ventral swelling. **g**. Female, tail. Scale bars: a and c= 20  $\mu$ m; b and e= 5  $\mu$ m; d= 100  $\mu$ m; f and g= 50  $\mu$ m.

#### ***Corononema parvum* Nicholas and Stewart, 1995**

Class : Chromadorea Inglis,, 1983

Order : Monhysterida Filipjev, 1929

Suborder : Monhysterina De Coninck and Schuurmans Stekhoven, 1933

Superfamily : Sphaerolaimoidea Filipjev, 1918

Family: Xyalidae Chitwood, 1951

Subfamily : Coronematinae Nicholas and Stewart, 1995

Genus : *Corononema* Nicholas and Stewart, 1995

*Corononema parvum* Nicholas and Stewart, 1995

Figures 3-4; Table 2.

#### **Materials examined**

Slide reference number ZSI-HQ/NZC/ *Corononema parvum* / WN.3415, 2 ♂, 1 Juvenile; WN.3691, 2 ♂; WN.3692, 3 ♀; Puducherry, Karaikal medu, Karaikal district; 10°56'42.98" N, 79°51'10.81" E; 21 September. 2021.

#### **Description**

Measurements – See Table 2

#### **Male**

Medium-sized nematodes, 926.68-1036.02  $\mu$ m; strongly annulated cuticle (Figure 4d). Longitudinal ridges throughout the body, weakly developed in buccal cavity

and cervical region, prominent towards midbody. Six incised leaf-like lips (Figure 3b, 4a). Inner labial setae not visible under light microscope. Six outer labial setae; four cephalic setae faintly visible posterior to the groove (Figure 4c); four strong cervical setae, at the base of non-annulated buccal region on first annule. Numerous body setae present throughout body. Deep groove with cuticle surrounds the buccal region just below the lips. Two cuticular rings lie just within the mouth (Figure 3b, 4b). Buccal cavity wide, deep with almost parallel-sided walls (Figure 3b, 4a). Amphid fovea (Figure 3b, 4b) is over base of buccal cavity. Cylindrical pharynx (Figure 3a, 4g). Nerve ring present. Two outstretched testes; spicules cephalated, rectangular curvature with simple pointed tips (Figure 3c). Gubernaculum surrounds spicule tip (Figure 3c, 4h).

#### **Female**

Closely resemble males except in the reproductive system. Single anterior gonad, vulva without operculum; few oocytes visible (Figure 4i).

#### **Remarks**

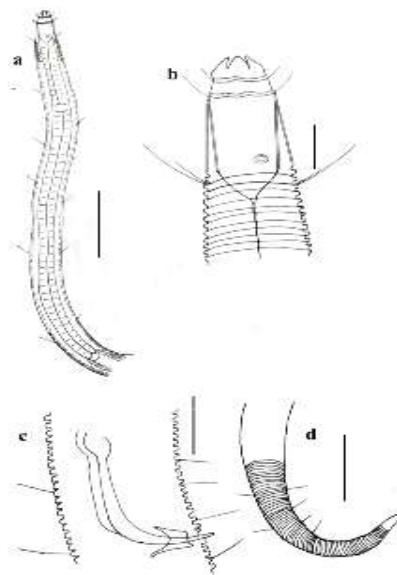
The genus *Corononema* Nicholas and Stewart, 1995 is mainly characterized by strongly annulated cuticle, high and incised lips, deep groove around the head just posterior to the insertion of six inner labial setae, in front of outer labial and cephalic setae, lip region sets off slightly. *C. parvum* differs from other species in the head shape and size and type of annulations. The morphometrics of *C. parvum* males and females from Puducherry conforms well with the specimens of the original description as regards to the body shape, body length, the presence of two cuticular

rings, the deep groove in the mouth region. However, the outer labial setae of the present specimens are slightly less than the type specimens (2.02-3.28  $\mu\text{m}$  vs 4-6  $\mu\text{m}$ ) and also differs in the presence of smaller amphid than the type

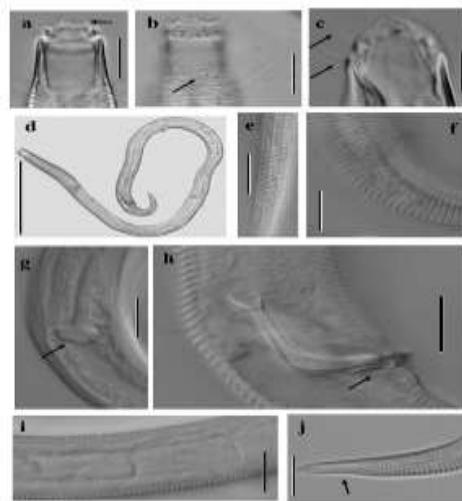
specimens (1.56-3.64  $\mu\text{m}$  vs 4-4.8  $\mu\text{m}$ ). The tail is slightly longer in size in the present specimens than the type specimens (75-101.1  $\mu\text{m}$  vs 72-87  $\mu\text{m}$ ).

**Table 2.** Measurements of *Corononema parvum* Nicholas and Stewart, 1995. All measurements are in  $\mu\text{m}$  and brackets indicate the mean values.

| Characters            | Males                   | Females                  | Juvenile    |
|-----------------------|-------------------------|--------------------------|-------------|
| n                     | 4                       | 3                        | 1           |
| Body Length           | 926.68-1036.02 (988.48) | 1051.75-1158.44(1087.74) | 862.89      |
| Maximum body diameter | 23.37-26.24 (24.55)     | 25.35-29.65 (28.16)      | 22.04       |
| Lip height            | 1.95-5.1 (3.65)         | 1.9-3.94 (2.71)          | 2.56        |
| Outer labial setae    | 2.02-3.28 (2.7)         | 2.9-3.2 (3.06)           | 2.76        |
| Cervical setae        | 12.11-13.47(12.83)      | 11.8-13.1 (12.6)         | 9.36        |
| Body setae            | 12.36-21.14 (15.31)     | 12.25-22.79 (17.05)      | 14.84       |
| Amphid                | 2-2.69 (2.29)           | 1.56-3.64 (2.78)         | 1.64        |
| Buccal cavity length  | 7.08-10.4 (8.84)        | 7.09-7.59 (7.31)         | 10.26       |
| Buccal cavity width   | 8.12-10.34 (8.9)        | 8.2-8.88 (8.56)          | 9.01        |
| Mouth to nerve ring   | 121-131 (127.25)        | 123.9-131 (126.9)        | 85          |
| Pharynx length        | 251.84-361 (301.29)     | 377.22-411.96 (395.5)    | 241         |
| Mouth to anus         | 819-897.31 (841.82)     | 814-830 (823.5)          | 801.2       |
| Width at anus         | 19.16-19.8 (19.38)      | 19.2-21.2 (20.1)         | 14          |
| Tail length           | 75-93.73 (84.83)        | 89.15-101.1 (95.93)      | 61.2        |
| Spicule arc           | 24.64-32.59 (28.62)     | -                        | -           |
| a                     | 38.46-41.69 (40.29)     | 35.60-41.48 (38.75)      | 39.15108893 |
| b                     | 2.86-3.67 (3.33)        | 2.64-2.81 (2.74)         | 3.57        |
| c                     | 9.88-13.81 (11.76)      | 10.78-11.81 (11.35)      | 14.0995098  |
| c'                    | 3.78-4.84 (4.38)        | 4.43-5.08 (4.76)         | 4.1         |
| V %                   | -                       | 83.00-85.45 (84.51)      | 80.08       |



**Figure 3.** *Corononema parvum* Nicholas and Stewart, 1995. (WN.3415/ *Corononema parvum*) **a.** Male, head and pharyngeal region. **b.** Male, head with lips, buccal cavity, cuticular rings, setae and amphid. **c.** Male, spicule and gubernaculum. **d.** Female, tail. Scale bars: a and d= 50  $\mu\text{m}$ ; b= 5  $\mu\text{m}$ ; c= 10  $\mu\text{m}$ .



**Figure 4.** *Corononema parvum* Nicholas and Stewart, 1995 (WN.3415/ WN.3692/ *Corononema parvum*) (LM photographs). **a.** Male, lips and buccal cavity. **b.** Male, amphid and cuticular rings. **c.** Male, outer labial setae and cephalic setae. **d.** Male, whole body. **e.** Male, longitudinal ridges in the mid-body region. **f.** Male, longitudinal ridges in the posterior region. **g.** Male, posterior pharynx and cardia. **h.** Male, spicule and gubernaculum. **i.** Female, oocytes. **j.** Male, tail tip. Scale bars: a, b, c, g, i and j= 5  $\mu$ m; d= 200  $\mu$ m; e and f= 50; h= 10  $\mu$ m.

## CONCLUSION

The two species *Conilia sinensis* Chen and Guo, 2015 and *Corononema parvum* Nicholas and Stewart, 1995 are reported for the first time from India. *C. sinensis* was collected from West Bengal and *C. parvum* from Puducherry. *C. sinensis* was originally described from East China Sea and recorded from Korea, whereas, *C. parvum* was originally described from Australia.

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