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ORIGINAL ARTICLE

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On a new species of the genus *Cobboldina* (Nematoda: Atractidae) from Hippopotamus (*Hippopotamus amphibius* Linnaeus, 1758) captivated at the Alipore Zoological Garden, Kolkata, West Bengal, India

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Abstract The specimens of the genus *Cobboldina* Leiper, 1911 and family Atractidae (Railliet, 1917) Travassos, 1919 recovered from the faecal matter of Hippopotamus (*Hippopotamus amphibius* Linnaeus, 1758) from the Alipore Zoological Garden, Kolkata, India recognized as a new species after careful observation. The collected nematode differs from the only valid species *Cobboldina vivipara* Leiper, 1911, in the presence of gubernaculum (13.2–29.7 μm in length) and the number of caudal papillae (10 pairs) and named as *Cobboldina gubernacularia* sp. n., This is the second species of the genus *Cobboldina* recorded from the host *Hippopotamus amphibius* Linnaeus, 1758 remaining captive in the Alipore Zoological Garden, Kolkata, India.

Keywords Nematoda · Atractidae · *Cobboldina gubernacularia* sp. n. · Hippopotamus · Alipore Zoological Garden

Introduction

During a survey on gastro intestinal parasites based on faecal sampling from March, 2007 to February, 2010, a total of 150 specimens were recovered from the Hippopotamus (*Hippopotamus amphibious* Linnaeus, 1758) remaining captive at the Alipore Zoological Garden, Kolkata, West Bengal, India. Examination and study of these nematodes revealed that these nematodes belonged to the superfamily Cosmocercoidea Travassos 1925 of the family Atractidae (Railliet 1917) Travassos 1919 and appears to be new to science which is described here and named as *Cobboldina gubernacularia* sp. n.

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Materials and methods

The nematodes collected from the faeces of *Hippopotamus amphibius* Linnaeus, 1758 remaining captive in the Zoological Garden, Kolkata, India, were fixed by dipping them in hot 4 % FA (formalin: glacial acidic acid, 4:1) and subsequently stored in 70 % alcohol. Most of the specimens after fixation were dehydrated slowly (Seinhorst 1966; Mondal and Manna 2010) and were mounted in anhydrous glycerin and sealed with paraffin wax. Four male specimens after clearing in glycerin alcohol was hand sectioned using a razor blade and was mounted in glycerin jelly to examine the oral structures and caudal papillae. Specimens were observed under different magnification with Zeiss trinoculor research microscope. Figures were drawn with the aid of Camera Lucida. Images were captured using a Sanyo Digital Camera. All measurements are in micrometer unless otherwise stated. Total of 150 specimens (48 females, 30 males and 72 larvae) have been collected during the study period. The average measurements of five male, seven female and five larvae specimens were taken and calculated ± standard deviation. Range is mentioned in parenthesis.

Result

Description

C. gubernacularia n. sp. (Figs. 1, 2).

General

Worms are small filliform, cuticle faint transversely striated. Mouth with a cuticular sheath, prolonged laterally into two triangular flaps; cephalic region with a pair of lateral amphids and six pairs of papillae (Fig. 2j). Buccal cavity present. Oesophagus divided into—a short muscular anterior portion (corpus) and a large posterior glandular portion (isthmus) and a bulb (Fig. 2c). Males are with two similar but very unequal spicules, tip pointed, a small gubernaculum is present (Fig. 2h, i). Females are viviparous and monodelphic (Fig. 2e). Vulva is close to the anus (Fig. 2d). Tail of both sexes long and pointed (Fig. 2b, g).

Male (Holotype and four paratypes)

Body 4 ± 0.10 (3.975–4.175) mm long and 130 ± 14.25 (112.5–150) µm thick; head diameter 48.84 ± 3.89 (42.9–52.8); pharynx 25.74 ± 4.30 (19.8–29.7) µm long and 19.14 ± 1.47 (16.5–19.8) µm wide; anterior oesophagus 215 ± 10.45 (200–225) µm long and 41.5 ± 4.62 (36.25–47.5) µm wide, posterior oesophagus 300 ± 15.30 (275–312.5) µm long and 38 ± 1.11 (37.5–40) µm wide; oesophageal bulb 75 ± 8.83 (62.5–87.5) µm long and 60 ± 5.59 (50–62.5) µm wide; nerve ring, excretory pore and anus at 252.5 ± 25.61 (212.5–275) µm, 492.5 ± 6.84 (487.5–500) µm and 3252.5 ± 65.19 (3150–3237.5) µm respectively from anterior end; tail filamentous, ventrally curved, 762.5 ± 66.73 (662.5–850) µm long; spicules 277.2 ± 29.88 (231–303.6) µm long and smaller spicule 67.98 ± 5.99 (59.4–75.9) µm long; spicular ratio 1: 3.9–4.3; gubernaculum very small measuring 21.78 ± 5.99 (13.2–29.7) µm in length; caudal papillae 10 pairs, discernable (Fig. 2b, i, h).

Female (seven paratypes)

Body 4.74 ± 0.30 (4.33–5.1) mm long and 210.7143 ± 36.39 (150–250) µm wide; head diameter 50.44 ± 9.27 (36.3–66) µm; buccal cavity measuring 30.64 ± 3.67 (23.1–33) µm long and 18.62 ± 1.56 (16.5–19.8) µm wide; anterior oesophagus 222.32 ± 11.32 (200.1–237.5) µm long and 41.07 ± 6 (37.5–50) µm wide, posterior oesophagus 316 ± 29.28 (275–356.25) µm long and 32.14 ± 6.68 (25–37.5) µm wide, bulb length 79.46 ± 11.81 (62.5–100) µm long and 71.42 ± 15.66 (50–87.5) µm wide. Nerve ring, excretory pore and anus at 292.85 ± 14.17 (275–312.5) µm, 571.42 ± 55.76 (487.5–562.5) µm and

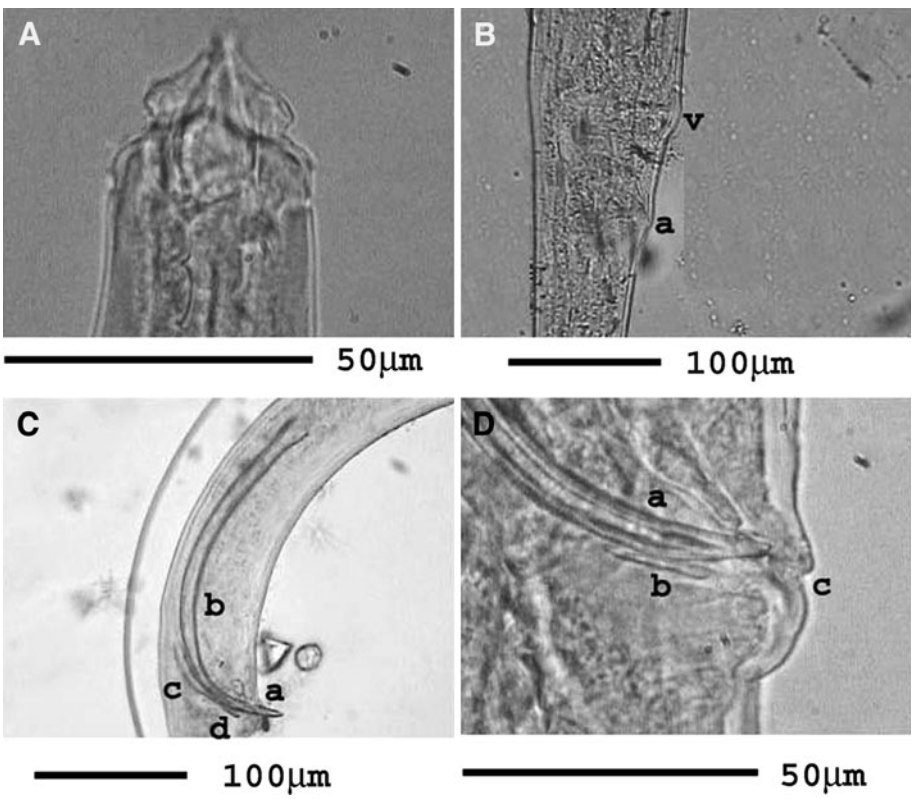

Fig. 1 Photomicrographs of *C. gubernacularia* n. sp. Male: **a** head region (*lateral view*); **c** cloacal region. *a* cloacal opening; *b* large spicules; *c* small spicule; *d* gubernaculum; **d** cloacal region (*magnified view*). *a* spicule; *b* gubernaculum; *d* cloacal opening. Female: **b** position of vulva (*v*) and anus (*a*)

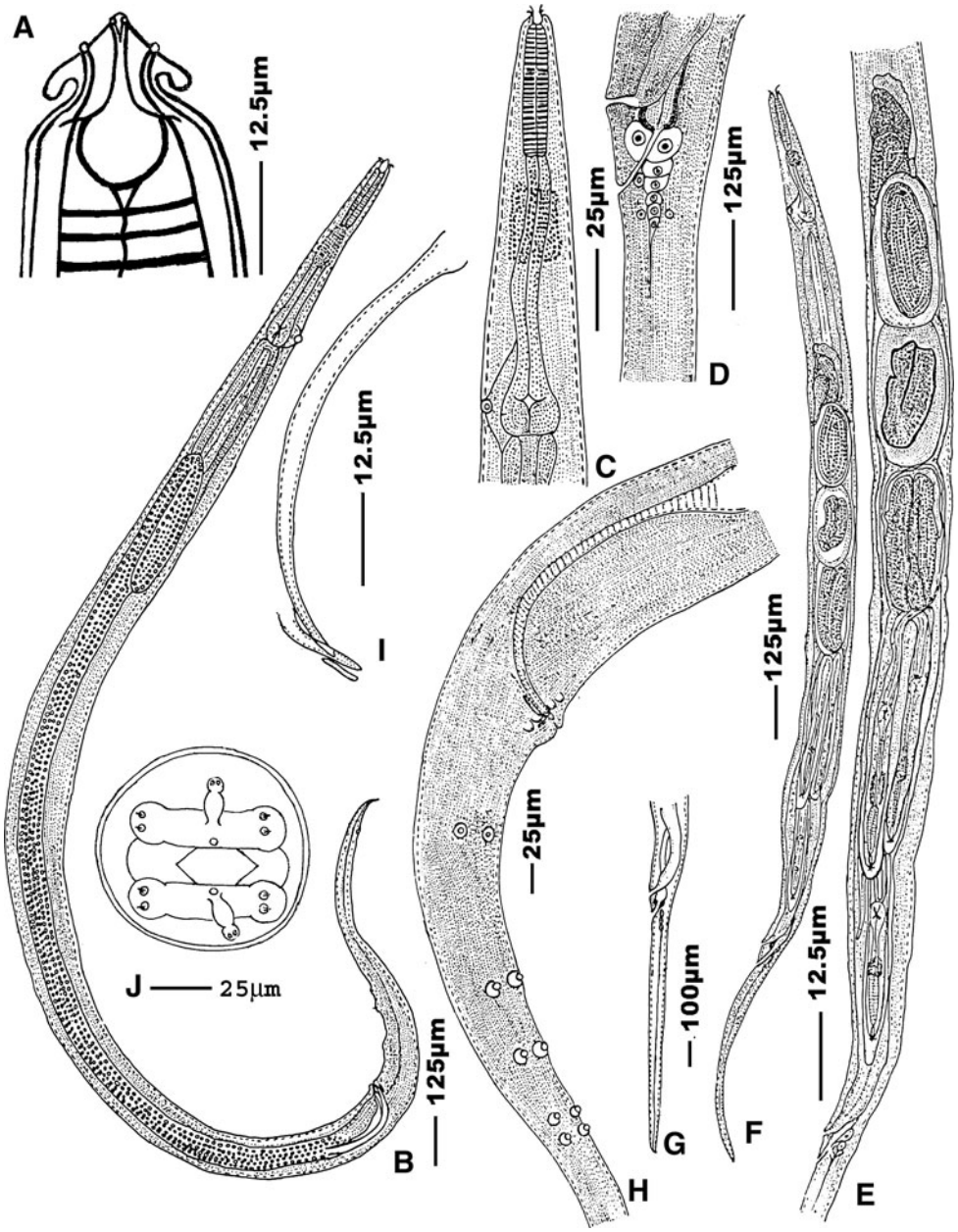

Fig. 2 Camera Lucida drawings of *C. gubernacularia* sp. n. Male: **a** head (*lateral view*); **b** whole body; **h** anal region showing positions of papillae; **I** spicules and gubernaculum; **j** en face view of head. Female: **c** anterior region; **d** vulva and anal region; **e** gonad and vulva and anal region. **f** whole body; **g** tail region

3796.42 ± 265.33 (3450–4100) µm respectively from anterior end; vulva post equatorial at 3615.78 ± 415.12 (2837.5–4000) µm distance from anterior end. Gonad 2380.35 ± 433.18 (1687.5–2825) µm monodelphic, uterus filled with more than two well developed larvae and one or two eggs; tail filamentous, 948.21 ± 71.59 (812.5–1025) µm long (Fig. 2d–g).

Larvae	The larvae measures 1,750–3137.5 µm long and 37.5–50.0 µm wide. Other characters are similar to general description
Type host	Hippopotamus (*Hippopotamus amphibius* Linnaeus 1758) in captivity in Alipore Zoological Garden, Kolkata, India
Location	Unknown, recovered from faecal matter of the host
Type locality	Alipore Zoological Garden, Kolkata, West Bengal, India
Date of collection	March, 2007 to February, 2010
Prevalence	Total 120 faecal samples were collected out of which 97 were positive (80.84 %)

Deposition of specimens	Holotype (♂) in one slide; and Paratypes (1♂, 2♀♀) in two separate slides, deposited at present to the nematode collection of Parasitology research unit, Department of Zoology, University of Calcutta, India. Accession Nos.: 000077N/10 (Holotype); 000078N/10; 000079N/10; 000080N/10.
Etymology	The new species described here is named as *C. gubernacularia* because of the presence of a gubernaculum in male

Discussion

Leiper (1910) erected the genus *Cobboldia* for nematode collected from the stomach of Hippopotamus from tropical Africa and described the new species *Cobboldia vivipara.* Later, Leiper (1911) replaced the generic name with *Cobboldina,* because *Cobboldia* had previously been used for a genus of insects. Further two species viz., *Cobboldina hyracis* Ezzat 1954 and *Cobboldina longicaudata* Köhler and Supperer 1960 from the host Procavia sp. (mammal) were described. But Petter (1962) and Ogden (1967), placed them under the atractid genus *Grassenema* Petter 1959. As such, till date there is only one valid species under the genus *Cobboldina* Leiper (1911).

The nematode specimen described here has the characteristic head, oesophagus, male and female reproductive organs similar to the genus *Cobboldina*. The presence of similar spicules and arrangements of the caudal papillae surrounding the cloaca brings the present species closer to *C. vivipara* (Table 1), but differs from the latter in having a gubernaculum, and 10 pairs of caudal papillae (where *C. vivipara* has nine pairs of caudal papillae). Thus for all these distinct characters, the present specimen described here is separated from the only valid species under the genus *Cobboldina* and hence it is considered as a new species and named as *C. gubernacularia* sp. n. as it possess gubernaculum in male specimens.

Table 1 Comparison of *C. gubernacularia* n. sp. with *C. vivipara* Leiper (1911); both from *Hippopotamus amphibius* Linnaeus, 1758

Morphometrics (In μm else otherwise Stated)	*C. gubernacularia* n. sp. from *Hippopotamus amphibius* Linnaeus, 1758		*C. vivipara* from *Hippopotamus amphibious* Linnaeus, 1758	
Characters	Male	Female	Male	Female
Length (mm)	3.975–4.175	4.33–5.1	3.46–4.62	3.97–4.69
Thickness	112.5–150	150–250	107–162	174–228
Head (L)	33–46.2	33–49.5	–	–
Head (B)	42.9–52.8	36.3–66	25-29	–
Oesophagus (L)	562.5–625	550–675	580–700	510–670
Anterior oesophagus (L)	200–225	200.1–237.5	170–210	170–220
Anterior oesophagus (B)	36.25–47.5	37.5–50	–	–
Posterior oesophagus (L)	275–312.5	275–356.25	–	–
Posterior oesophagus (B)	37.5–40	25–37.5	–	–
Oesophagesl bulb (L)	62.5–87.5	62.5–100	–	–
Oesophagesl bulb (B)	50–62.5	50–87.5	–	–
Nerve ring[a]	212.5–275	275–312.5	232–331	185–298
Excretory pore[a]	487.5–500	487.5–562.5	410–600	380–570
Tail	662.5–850	812.5–1025	680–920	800–1130
Large spicule	231–303.6	–	242–279	–
Small spicule	59.4–75.9	–	54–68	–
Gubernaculum	13.2–29.7	–	[b]	–
Caudal papillae (post cloacal)	10–pairs	–	9-pairs	–
Spicular ratio	1: 3.9–4.3	–	1:1.39–1.46	–
Vulva[a]	–	2837.5–4000	–	2981–3522
Distance from vulva to anus	–	62.5–106.25	–	63–79

[a] From anterior end

[b] Absent

Key to tile species of the genus *Cobboldina*

Gubernaculum absent. Presence of nine pairs of caudal papillae.

..…….. *Cobboldina vivipara* Leiper 1911.

Gubernaculum present. Presence of ten pairs of caudal papillae.

…….. *Cobboldina gubernacularia* sp. n.

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