

Research Note

Morphology and Diagnosis of Fourth-Stage Larvae of *Tridentoifundibulum gobi* (Nematoda, Strongyloidea) Parasitizing Horses, *Equus caballus*

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ABSTRACT: The fourth-stage larva of *Tridentoifundibulum gobi*, one of the rare species of Cyathostominae, is identified and described. The larva was identified as *T. gobi* as a result of finding molting forms showing characteristics of both larvae and adults. The buccal capsule is large and spherical; its width exceeds its depth. The buccal capsule walls are thick, gradually narrowed to the apical edge and abruptly narrowed to the posterior edge. The esophageal funnel has 3 large, triangular teeth projecting into the buccal cavity similar to those of *Triodontophorus* spp. and *Gyalocephalus capitatus*. The fourth-stage larva of *T. gobi* can be distinguished by its buccal capsule having a smaller diameter than the esophagus. In addition, the buccal capsule of *T. gobi* fourth-stage larvae is wider than deep but is as deep or deeper than wide in the fourth-stage larvae of *Triodontophorus* spp., and the buccal teeth of *T. gobi* fourth-stage larvae are much more steeply pointed and have smoother anterior edges than those of *G. capitatus*.

KEY WORDS: Nematoda, Strongyloidea, Cyathostominae, *Tridentoifundibulum gobi*, fourth-stage larva, morphology, systematics, horse, *Equus caballus*.

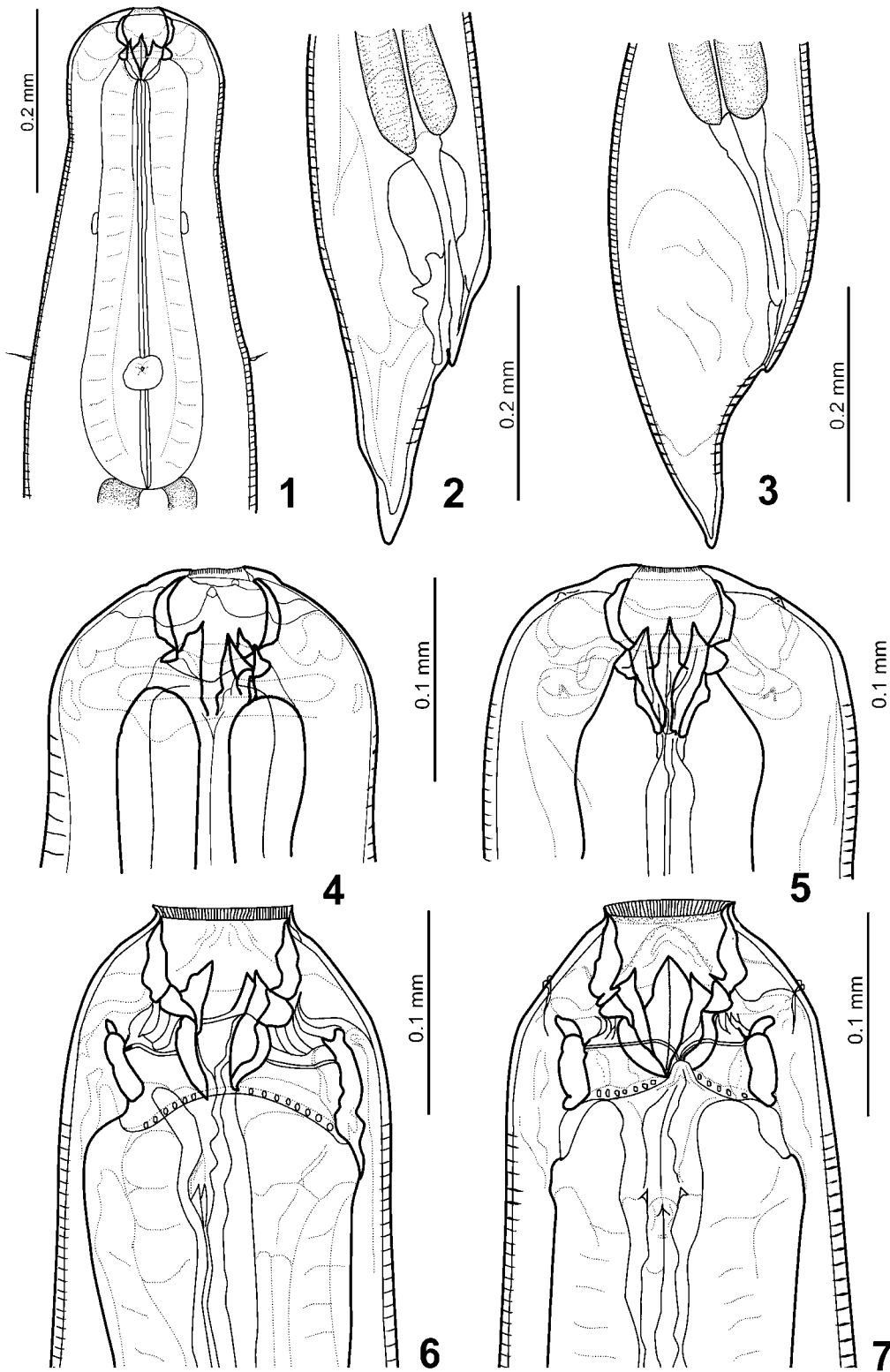
The majority of Cyathostominae parasitizing horses are cosmopolitan, and few have a limited distribution or show specificity to species of equids (Lichtenfels et al., 1998). *Tridentoifundibulum gobi* Tshojo in Popova, 1958, has been reported only 4 times. Described from a collection in Mongolia and reported later from Kazakhstan (Ivashkin and Dvojninos, 1984), *T. gobi* was considered to be endemic to Central Asia. However, *T. gobi* was discovered in North America (Louisiana, U.S.A.), where it was described as a new species (*Cylicostephanus torbertae* Lichtenfels and Klei, 1988), and reported recently in horses in Scotland (Lichtenfels et al., 2001). The adult stage of this rare species is distinguished by an

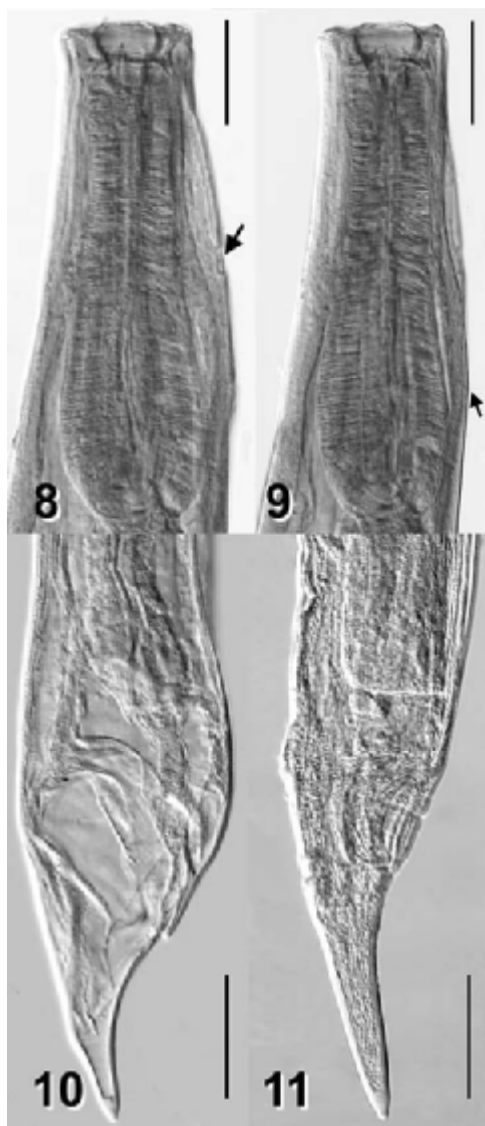
invagination on the dorsal side of the lining of the esophagus where 3 small teeth are present. The fourth-stage larva (L4) of *T. gobi* remained unidentified until now despite its earlier description (Dvojninos and Kharchenko, 1990b).

Specimens studied included L4 from horses from West Kazakhstan. The larvae examined are kept in the collection of the Parasitology Department in the Schmalhausen Institute of Zoology, National Academy of Sciences of Ukraine, Kiev, Ukraine. The material was collected by artificial digestion. Specimens were cleared for microscopy in a solution of phenol–glycerol (80% phenol in 20% glycerine by volume) or in phenol–alcohol (80% phenol in 20% absolute ethanol by volume). For microscopy an Olympus BX-51 was used. Drawings were made with the aid of a camera lucida (Figs. 1–7). Photomicrographs were made with a digital camera, and the halftone plates (Figs. 8–18) were prepared using Adobe Photoshop (Adobe Systems Incorporated, San Jose, California, U.S.A.). All measurements are given in micrometers unless otherwise indicated.

The buccal capsule is large and spherical; its width exceeds its depth. The buccal capsule walls are thick, gradually narrowed to the apical edge and abruptly narrowed to the posterior edge. The esophageal funnel ring is of moderate height and considerably shorter than the buccal capsule depth. The esophageal funnel has 3 triangular teeth projecting into the buccal cavity. The dorsal tooth reaches the center of the buccal cavity. Subventral teeth are shorter than the dorsal tooth. The esophagus is of a greater diameter than the buccal capsule. In molting L4s, the adult buccal capsule and the 3 small teeth in the esophageal funnel, characteristic of *T. gobi*, can be seen. Measurements of females and males are given separately (Tables 1, 2). With the exception of tail length and distance between anterior end and excretory pore, measurements of males correspond to those of females.

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Figures 8–11. L4 *Tridentoifundibulum gobi*. 8. Esophageal region, lateral view, showing excretory pore (arrow). 9. Esophageal region, dorsoventral view, showing cervical papilla (arrow). 10. Male tail, lateral view. 11. Female tail, lateral view. Scale bar = 100 μ m.

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Figures 1–7. L4 *Tridentoifundibulum gobi*. 1. Esophageal region, ventral view. 2. Female tail, lateral view. 3. Male tail, lateral view. 4. Buccal capsule, lateral view. 5. Buccal capsule, dorsoventral view. 6. Anterior end of molting larva, lateral view. 7. Anterior end of molting larva, dorsoventral view.

Table 1. Measurements of female fourth-stage larvae of *Tridentoifundibulum gobi*.

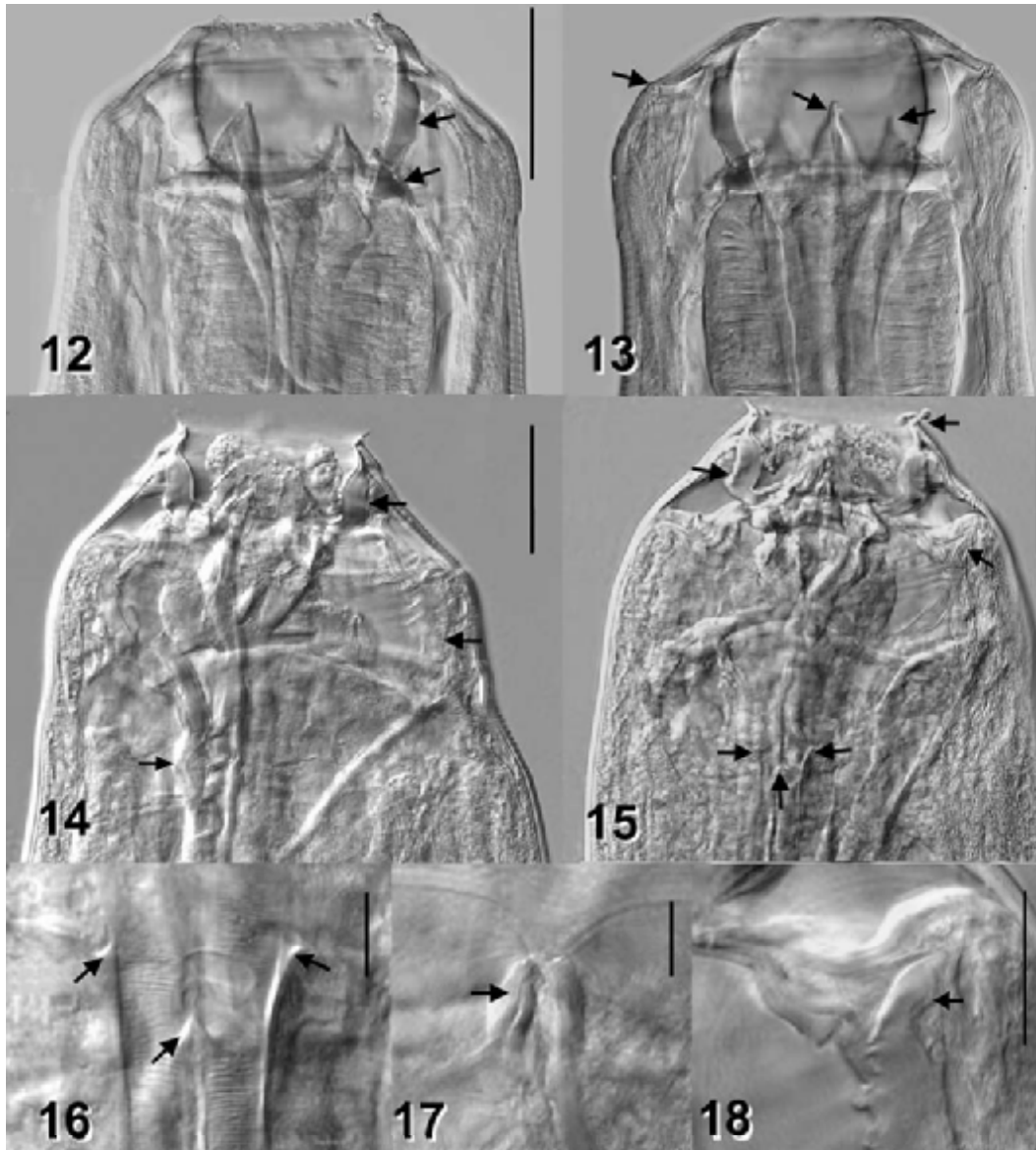
Character	n	Range	Mean \pm SD
Length (mm)	7	4.3–6.8	5.2 \pm 0.83
Depth of buccal capsule	10	30–41	34 \pm 3.0
Width of buccal capsule	10	61–77	69 \pm 5.1
Height of esophageal funnel ring	10	11–17	13 \pm 1.9
Length of esophagus	10	426–476	455 \pm 18.6
Distance from anterior end to excretory pore	4	297–392	344 \pm 40.5
Distance from anterior end to cervical papillae	3	336–381	355 \pm 23.3
Distance from anterior end to nerve ring	4	218–252	238 \pm 14.1
Length of tail	7	177–234	220 \pm 20.6

The L4 of *T. gobi* is similar to the L4s of *Triodontophorus* spp. (see Ihle and Oordt [1923] and Dvojnos and Kharchenko [1994]) and *Gyalocephalus capitatus* Looss, 1900 (of Dvojnos and Kharchenko, 1990a, 1994), because of the 3 large buccal teeth. The L4 of *T. gobi* can be distinguished by its buccal capsule having a smaller diameter than the esophagus. In addition, the buccal capsule of *T. gobi* L4 is wider than deep, but it is as deep or deeper than wide in the L4s of *Triodontophorus* spp. The buccal teeth of the *T. gobi* L4 are much more steeply pointed and have smoother anterior edges than those of the *G. capitatus* L4.

Boulenger (1921) was the first to describe L4s of horse cyathostomins. In his work, he identified *Cylicocycclus insigne* (Boulenger, 1917) L4s with adult forms. Later, Ihle and Oordt (1923) described 6 L4 types. Müller (1950) described 21 types of cyathostomin parasitic larvae from the gut walls of the horse but did not identify them to species.

Table 2. Measurements of male fourth-stage larvae of *Tridentoifundibulum gobi*.

Character	n	Range	Mean \pm SD
Length (mm)	3	4.7–6.1	5.2 \pm 0.76
Depth of buccal capsule	3	33–38	35 \pm 2.6
Width of buccal capsule	3	65–76	69 \pm 5.7
Height of esophageal funnel ring	3	11–12	12 \pm 0.6
Length of esophagus	3	431–454	444 \pm 11.7
Distance from anterior end to excretory pore	2	274–297	286 \pm 15.8
Distance from anterior end to cervical papillae	3	319–364	338 \pm 23.3
Distance from anterior end to nerve ring	2	235–241	238 \pm 4.0
Length of tail	2	174–195	185 \pm 14.8



Figures 12–18. L4 *Tridentoifundibulum gobi*, photomicrographs. **12.** Buccal capsule, lateral view, showing larval buccal capsule (upper arrow), esophageal funnel ring (lower arrow), dorsal tooth at left, and subventral teeth at right. **13.** Buccal capsule, dorsoventral view, showing dorsal tooth (middle arrow), flanked by 2 subventral teeth (right subventral tooth marked by right arrow) and left lateral papilla (left arrow). **14.** Anterior end of molting larva, lateral view, showing walls of larval buccal capsule (upper arrow), adult buccal capsule (middle arrow), and dorsal invagination of lining of esophagus (lower arrow) that contains adult esophageal teeth. **15.** Anterior end of molting larva, dorsoventral view, showing larval radial crown (top arrow), larval buccal capsule wall (left, upper arrow), support of external leaf-crown (ELC) (right, upper arrow), and 3 esophageal teeth in dorsal invagination (lower arrows). **16.** Higher magnification of 3 esophageal teeth of adult stage (arrows), in molting specimen. **17.** Adult-stage dorsal gutter (arrow), in molting specimen. **18.** Adult-stage support (arrow) for ELC, in molting specimen. **12–15.** Scale bar = 100 μm . **16–18.** Scale bar = 10 μm .

Baruš (1962) identified *Cylicocycylus nassatus* (Looss, 1900) and *Cylicocycylus elongatus* (Looss, 1900) L4s. Ogbourne (1978) identified the L4 of *Coronocycylus coronatus* (Looss, 1900), and Reinemeyer and Herd (1986) identified *Cylicostephanus longibursatus* (Yorke and Macfie, 1918) L4s. At present, together with *T. gobi*, 24 cyathostomin L4s have been identified to species (Dvojnjos and Kharchenko, 1987, 1990a, 1990b; Kharchenko and Dvojnjos, 1989).

The structure of L4s is much simpler than that of adults, and the buccal morphology is sharply different from that of mature worms. Identification of L4s has usually been based on molting specimens, which have buccal capsule features of both larvae and adults. In the future, DNA characteristics can be used to match larval and adult stages. The morphology of L4s is currently being tested for use in the classification of the Cyathostominae by the authors of this study.

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