

## Original papers

# *Trichodina amblypharyngodoni* sp. n. and *Trichodina hoffmani* Wellborn, 1967 (Ciliophora: Trichodinidae) from the freshwater fishes in the Baikka Beel of Moulvibazar district in Sylhet division, Bangladesh

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**ABSTRACT.** During a survey on species diversity of trichodinid ciliates from freshwater fishes in the Baikka Beel of Moulvibazar district in Sylhet division from January to December 2015, two species of ciliates were identified, of which one is new to science. *Trichodina hoffmani* Wellborn, 1967 is collected from the gills of *Mystus tengara*, and *Trichodina amblypharyngodoni* sp. n. is described from *Amblypharyngodon mola* using the silver nitrate impregnation method. *T. amblypharyngodoni* sp. n. is characterized by having a large, darkly stained central area; rectangularly-rounded butterspoon-shaped blade with very narrow interblade space and blunt tangent point; indistinct anterior and posterior blade apophysis, but a rounded apex near the base of the central part of denticle; slender, sometimes slightly triangular central part with rounded tip; and filamentous, curved, anteriorly directed rays with inflated tip and indistinct central groove. Based on these characters and the unique shape and absence of variability of the denticles among the silver impregnated specimens of the present species, it may be said that to a lesser extent, it resembles *Trichodina prowazeki* Grupcheva and Lom, 1980.

**Key words:** Ciliophora, Trichodinidae, Bangladesh, *Trichodina amblypharyngodoni* sp.n., *Trichodina hoffmani*

## Introduction

The genus *Trichodina* Ehrenberg [1] is the most numerous within the family Trichodinidae [2]. Representatives of the family parasitize or are symbionts of a broad spectrum of aquatic invertebrate and vertebrate hosts [3], are best known ectoparasites of fish with most of the species reported from freshwater environments [4]. They infect fish skin, fins and gills [5]. Over 300 species of *Trichodina* have been established from different parts of the world, most by means of Klein's silver impregnation. In Bangladesh, Asmat et al. [6] made the first report of trichodinid ciliates. Since then scanty and infrequent information is available on the taxonomy of the particular group. As a result, a total of 36 species of trichodinid ciliates are reported from host fish species in different regions of Bangladesh representing the four genera *Trichodina* Ehrenberg [1], *Paratrachodina* Lom [7], *Tripartiella*

Lom [8] and *Trichodinella* Šramek-Hušek [9] from various species of freshwater and estuarine fish. The existing data on this matter can be found in papers of Asmat et al. [6,10,11], Bhouyain et al. [12], Asmat et al. [13–15], Asmat and Sultana [10], Habib and Asmat [17], Kibria et al. [18–20], Habib et al. [21], and Kibria and Asmat [22].

The objectives of the present survey was to ascertain the trichodinid ciliates of freshwater fishes from the Baikka Beel area of Moulvibazar district in Sylhet division as no previous study was done on fishes of this area and emphasizing species diversity, morphological variability with reference to existing taxonomic information.

## Materials and Methods

Live host fishes, *Amblypharyngodon mola*, Mola carplet (2.5–5.0×2.0–4.2 cm) and *Mystus tengara*, Tengara catfish (7.6–10.0×20–35 cm) were

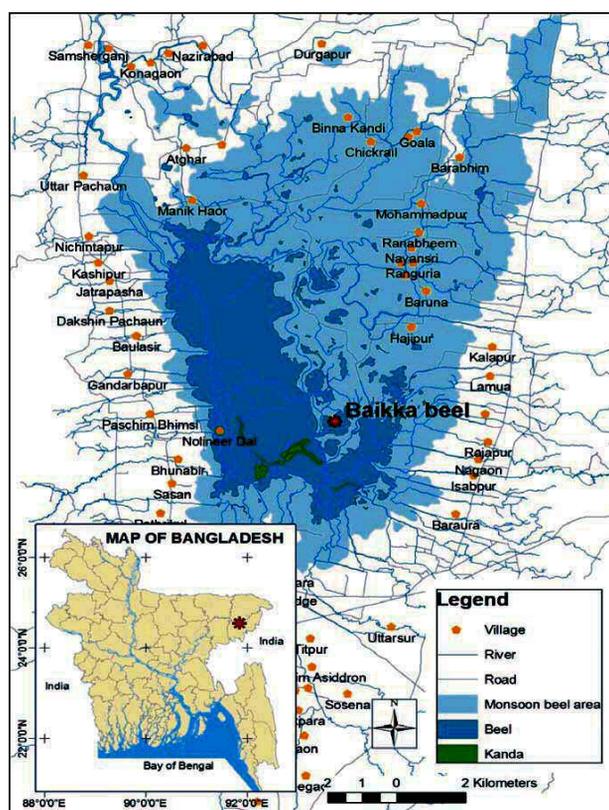


Fig. 1. Map of sampling localities at the Baikka Beel, Bangladesh (source: IPAC 2013, www.nishorgo.org)

collected from local markets and adjacent areas of the Baikka Beel with the help of local people and fishermen from January to December, 2015. The fish specimens were examined for trichodinids soon after died. Wet smears of the gills were prepared on clean grease free slides and allowed to air-dry at the sampling sites. Air-dried slides containing smears were transported to the laboratory to examine under a research microscope for sorting out slides with trichodinid ciliates. Slides positives for trichodinids were processed by using modified Klein's [23] silver impregnation technique as recommended by Lom [24]. Each trichodinid population was photographed by using a SONY cyber shot camera in order to have comprehensive morphological data analyses and measurement. Measurements of the parasites were made with an ocular micrometer and followed the recommendation of Lom [24], Wellborn [25], Arthur and Lom [26], Van As and Basson [3,27] and Basson and Van As [28].

## Results and Discussion

*Trichodina amblypharyngodoni* sp. n. (Figs 3, 4, 6; Table 1)

**Description (n=20):** Falls in range of large-sized ciliate. Disc-shaped body with diameter of 66.0–71.9 ( $69.2 \pm 1.8$ ), concave adhesive disc 60.3–64.9 ( $62.5 \pm 1.4$ ) in diameter, surrounded by wide border membrane 3.8–4.9 ( $4.2 \pm 0.3$ ). Central area impregnated, almost similar to rest of disc without granules about 23.8–28.0 ( $26.7 \pm 0.9$ ) in diameter. Denticulate ring 51.6–56.5 ( $54.3 \pm 1.6$ ) in diameter. Denticle stout, curved. Number of denticles 31–33 ( $32.1 \pm 0.6$ ), number of radial pins per denticle 5–7 ( $6.1 \pm 0.7$ ), span 17.8–20.2 ( $18.9 \pm 0.6$ ), length 5.7–6.5 ( $6.0 \pm 0.3$ ). Length of ray 7.7–8.9 ( $8.3 \pm 0.3$ ), blade 6.8–8.2 ( $7.4 \pm 0.4$ ), and width of central part 2.4–3.6 ( $3.0 \pm 0.3$ ). Adoral ciliary spiral about  $390\text{--}410^\circ$ .

**Denticle morphology:** Blade butter-spoon shaped with parallel anterior and posterior margin (Fig. 3). Distal margin of blade flat, lies close contact with inner side of border membrane, sometimes convex or conically rounded, and parallel to border membrane. Tangent point blunt, forms a line rather than point, and situated slightly

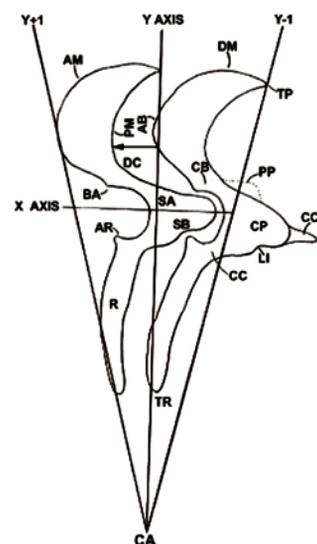


Fig. 2. Denticle structure and construction of X and Y axes as fixed references for description denticles after Van As and Basson [3].

Explanations: **AB**, apex of blade; **AM**, anterior margin of blade; **AR**, apophysis of ray; **B**, blade; **BA**, apophysis of blade; **CA**, central area of adhesive disc; **CB**, section connecting blade and central art; **CC**, section connecting part and ray; **CCP**, central conical part; **CP**, central part of blade; **DC**, deepest point of semi-lunar curve relative to apex; **DM**, distal margin of blade; **PM**, posterior margin of blade; **PP**, posterior projection; **R**, ray; **SA**, section of central part above x axis; **SB**, section of central part below x axis; **TP**, tangent point; **TR**, tip of ray.

Table 1. Morphometric comparison of *T. amblypharyngodoni* sp. n. with two populations of *T. prowazeki* (measurements in  $\mu\text{m}$ )

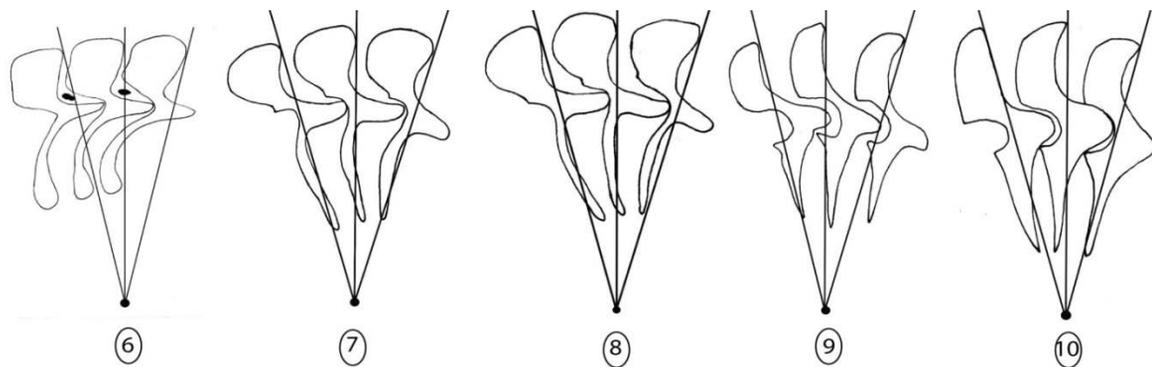
	<i>T. prowazeki</i> (n=25) [29]	<i>T. prowazeki</i> (n=25) [26,30]	<i>T. amblypharyngodoni</i> sp.n. (n=20)
Host	<i>Rutilus rutilus</i>	<i>Rutilus rutilus</i>	<i>Amblypharyngodon mola</i>
Locality	Bulgaria	Czech Republic	Baikka Beel, Bangladesh
Location	Gills	Gills	Gills
Diameter of			
body	62.0(57–67)	54.1–87.7(63.4 $\pm$ 8.3)	66.0–71.9(69.2 $\pm$ 1.8)
adhesive disc	46.0(42–30)	42.3–55.1(46.9 $\pm$ 2.8)	60.3–64.9(62.5 $\pm$ 1.4)
denticulate ring	30.0(26–30)	25.0–34.2(29.3 $\pm$ 2.0)	51.6–56.5(54.3 $\pm$ 1.6)
central area	–	–	23.8–28.0(26.7 $\pm$ 0.9)
Width of border membrane	3.8–5.8	3.6–6.6(5.6 $\pm$ 0.6)	3.8–4.9(4.2 $\pm$ 0.3)
Number of denticles	23(22–24)	23–26(24.3 $\pm$ 1.0)	31–33(32.1 $\pm$ 0.6)
radial pins/denticle	10(9–11)	9–11	5–7(6.1 $\pm$ 0.7)
Span of denticle	–	13.3–16.3(15.0 $\pm$ 0.7)	17.8–20.2(18.9 $\pm$ 0.6)
Length of denticle	6.3–8.6	8.2–12.2(10.7 $\pm$ 1.0)	5.7–6.5(6.0 $\pm$ 0.3)
ray	4.8–7.7	6.1–8.7(7.5 $\pm$ 0.7)	7.7–8.9(8.3 $\pm$ 0.3)
blade	5.3–6.3	4.6–6.6(5.4 $\pm$ 0.5)	6.8–8.2(7.4 $\pm$ 0.4)
Width of central part	1.5–2.4	1.5–2.6(2.0 $\pm$ 0.2)	2.4–3.6(3.0 $\pm$ 0.3)
Degree of adoral ciliature			390–410°

lower than distal margin. Anterior margin angularly slopes down to form an angular or slightly curved with depression at base of blade. Sometimes concave depression found on anterior margin, interblade space too narrow. Lower border of apical depression sometimes impregnates with silver. Apex rarely extends beyond  $y+1$  axis (Fig. 6). Anterior blade apophysis although present, not always prominent. Posterior margin of blade has same angle as anterior, hence different to mark any semilunar curve. Space between anterior and posterior blade attachment with blade connection

forms a kind of a semilunar curve with deepest point at below apex. Posterior blade apophysis absent. Blade connection thin. Central part strong, tubular, straight or slightly curved downwards with bluntly rounded, robust point of tip extending halfway to  $y-1$  axis and strongly interlinked into next denticle. Indentation in lower central part prominent. Ray connection short and thin. Ray apophysis prominent directed upward towards central part. Ray larger than blade with constriction just below ray apophysis. Post constricted part of ray gradually inflated and ends in bulbous tip having bright



Figs 3–5. Photomicrographs of silver impregnated adhesive discs of *Trichodina amblypharyngodoni* sp. n. (3–4), and *Trichodina hoffmani* (5). Scale bar 20  $\mu\text{m}$ .



Figs 6–10. Diagrammatic drawings of the denticles of *Trichodina* species: 6. *Trichodina amblypharyngodoni* sp. n. from the gills of *Amblypharyngodon mola* in Bangladesh; 7. *Trichodina prowazeki* from the gills of *Rutilus rutilus* in Bulgaria redrawn from [29]; 8. from the gills of *Rutilus rutilus* in Czech Republic redrawn from [26,30]; 9. *Trichodina hoffmani* from the gills of *Mystus tengara* in Bangladesh; 10. from the gills of the *Etheostoma edwini* in USA redrawn from [25].

argentophilic impregnation. Ray gently curved in posterior direction. Tip of ray almost touches y+1 axis. Lower half of ray remains almost parallel to y axis.

The present populations of *T. amblypharyngodoni* sp. n. were found to invade the gills of *A. mola* at various seasons and show very little variation in measurements and denticle morphology. However, the individuals with somewhat conically rounded distal margin and slightly angular central part could be found even in the same subpopulation. *T. amblypharyngodoni* sp. n. is characterized by having a large, darkly stained central area; rectangularly-rounded butterspoon-shaped blade with very narrow interblade space and blunt tangent point; indistinct anterior and posterior blade apophysis, but a rounded apex near the base of the central part of denticle; slender, sometimes slightly triangular central part with rounded tip; and filamentous, curved, anteriorly directed rays with inflated tip and indistinct central groove. Based on these characters and the unique shape and absence of variability of the denticles among the silver impregnated specimens of the present species, it may be said that to a lesser extent, it resembles *Trichodina prowazeki* Grupcheva and Lom [29].

*T. prowazeki* is found in only two European countries, viz., Bulgaria and Czech Republic, and Russian Federation. The species was first described by Grupcheva and Lom [29] from the gills of *Rutilus rutilus* in the Batak and Georgi Dimitrov dam lakes, Bulgaria. Subsequently, Arthur and Lom [26] reported its presence from the same host from northwestern Rybinsk Reservoir in Russian Federation, located on the upper reaches of the

Volga River System. Kepr [31] reported this species from the skin of *Rutilus rutilus* in Czech Republic.

The slender central part of blade, very narrow blade connection and, to some extent, the slender and anteriorly slanted, gradually tipped ray of *T. prowazeki* is accountable, but in close observation the details of the mentioned features are distinctly different between the two species. For example, in *T. prowazeki*; blade broad and rounded (vs broadly rectangular); blade shorter than ray (vs longer than ray); anterior blade apophysis distinct (vs indistinct, sometimes prominent); posterior blade margin almost straight, parallel to y-1 axis (vs same angle as anterior one) (Fig. 7); semilunar curve shallow (vs deep); central part narrow tube-like, (vs with truncated tip slightly triangular with smoothly rounded tip); ray straight, anteriorly directed though anteriorly directed but immediately curve down to the posterior direction); ray tip sharply pointed (vs filamentous ray ends in an inflated, rounded tip); and central groove distinct (vs indistinct). The body is also significantly smaller in the presently described trichodinid (Table 1). Forty out of 65 (61.53%) of the examined *A. mola* were found infected with *T. amblypharyngodoni* sp. n. The level of infection was low. This ciliate was found invading the gills of host fish in the month of June 2015.

**Type host:** *Amblypharyngodon mola* (Hamilton, 1822). **Type locality:** The Baikka Beel (24.3514°N, 91.6979°E) of Moulvibazar district in Sylhet division, Bangladesh. **Type location on host:** Gills. **Type specimens:** Holotype, slide CUZM-AM-BB-1 (25. 06. 2015); Paratypes, slide CUZM-AM-BB-2 (25. 06. 2015) is in the collection of the Museum of

Table 2. Morphometric comparison of *Trichodina hoffmani* obtained in the present study with *Trichodina hoffmani* Wellborn, 1967 [25] (measurements in  $\mu\text{m}$ )

	<i>T. hoffmani</i> [25]	<i>T. hoffmani</i> (n=20)
Host	<i>Etheostoma edwini</i>	<i>Mystus tengara</i>
Locality	Chattahoochee State Park, Houston County, Alabama, USA	Baikka Beel, Bangladesh.
Location	Fins and body	Gills
Diameter of		
body	61.0–90.0(70)	38.8–50.4(43.7 $\pm$ 3.8)
adhesive disc	28.0–36.0(33)	29.6–40.5(35.2 $\pm$ 3.3)
denticulate ring	15.5–21.0(18)	15.5–22.4(19.5 $\pm$ 2.3)
central area	–	5.3–11.2(8.2 $\pm$ 2.0)
Width of border membrane	2.5–4.0(3)	3–4.4(3.8 $\pm$ 0.4)
Number of denticles	21–23(22)	17–25(20.3 $\pm$ 2.7)
radial pins/denticle	–	5–7(6.2 $\pm$ 0.6)
Span of denticle	–	9.2–13.6(11.1 $\pm$ 1.5)
Length of denticle	5.0–7.0(6.4)	2.9–3.7(3.2 $\pm$ 0.3)
ray	3.0–4.5(3.8)	3.6–5.8(4.4 $\pm$ 0.8)
blade	3.0–4.0 (3.6)	3.1–5.1(4.0 $\pm$ 0.7)
Width of central part	1.0–1.5	2.2–3.1(2.7 $\pm$ 0.3)
Degree of adoral ciliature	390°	375–390°

Department of Zoology, University of Chittagong-4331, Bangladesh. **Etymology.** Named after the generic name of the host fish species, *Amblypharyngodon mola*.

***Trichodina hoffmani* Wellborn, 1967** (Figs 5, 9, 10; Table 2)

**Host:** *Mystus tengara* Hamilton, 1822. **Locality:** The Baikka Beel (24.3514°N, 91.6979°E) of Moulvibazar district in Sylhet division, Bangladesh. **Location on host:** Gills. **Prevalence:** Eight out of thirty specimens examined (26.66%). **Infection:** Low. **Voucher specimens:** Two slides (CUZM-MT-1-BB and CUZM-MT-2-BB) with sliver impregnated specimens prepared on 9 February 2015 have been deposited in the Museum of Department of Zoology, University of Chittagong 4331, Bangladesh.

**Description (n=20):** Large sized trichodinid 38.8–50.4 (43.7 $\pm$ 3.8) diameter. Adhesive disc rounded, rarely oblong, 29.6–40.5 (35.2 $\pm$ 3.3) diameter surrounded by finely striated border membrane 3.0–4.4 (3.8 $\pm$ 0.4) wide. Central area uniformly dark stained. Texture of central area similar to rest of adhesive disc, 5.3–11.2 (8.2 $\pm$ 2.0)

diameter. Denticulate ring 15.5–22.4 (19.5 $\pm$ 2.3), consisting of 17–25 (20.3 $\pm$ 2.7) denticles with 5–7 (6.2 $\pm$ 0.6) radial pins per denticle. Span and length of denticle 9.2–13.6 (11.1 $\pm$ 1.5) and 2.9–3.7 (3.2 $\pm$ 0.3) respectively. Length of ray 3.6–5.8 (4.4 $\pm$ 0.8), blade 3.1–5.1 (4.0  $\pm$  0.7), and width of central part 2.2–3.1 (2.7 $\pm$ 0.3). Adoral ciliary spiral about 375–390°.

**Denticle morphology:** Blade broad, stout and falcate-shaped, shorter than ray, filling major spaces between y and y+1 axes (Fig. 9). Distal margin loping down from tangent point, difficult to recognize, hence not parallel to border membrane, gap between distal margin of blade and inner side of border membrane large. Tangent point blunt or sometimes slightly sharp, almost on same level or slightly upper than distal margin. Anterior margin smoothly curves down and forms rounded apex, not touching y+1 axis. Apical depression not always distinct and never impregnates. Anterior blade apophysis invisible. Posterior margin forms shallow crescent like depression and leaving broad space between convex apex of one denticle and concave crescent of other. Deepest point of crescent present below center of apex. Posterior blade projection

absent. Central part robust, widely triangular and forwardly directed with roughly rounded tip (Fig. 5). Indentation of lower central part distinct. Section of central part above and below dissimilar, upper part broad convex. Tip of central part firmly interposed into corresponding denticle, extending more than half to y-1 axis. Ray stout, slightly slanted anteriorly, larger than blade, proximal part wider than distal part, central groove indistinct. Ray connection thick with prominent, thick, spike shaped and upwardly directed ray apophysis. Tip of ray blunt, rarely sharply pointed.

*Trichodina hoffmani* was established as a new species by Wellborn [25] from the fins and body of *Etheostoma edwini* at the Chattahoochee State Park, Houston County, Alabama, USA. In accordance with Wellborn [25], *T. hoffmani* is characterized by some special features, including large-sized trichodinid with cup shaped adhesive disc; the central area of adhesive disc is finely granular and dark stained while in silver impregnated; the tip of blade is truncated; the distal margin sloping down from the tangent point and difficult to recognize; the ray is larger than blade with broadened first half and the rest of ray narrow, slightly pointed; the ray apophysis and the indentation in lower central part is distinct; and the presence of indistinct central groove. Wellborn [25] established *T. hoffmani* from the fins and body of *Etheostoma edwini* from a stream 200 yards south of the entrance to the Chattahoochee State Park, Houston County, Alabama, USA. He also obtained the species from *Percina maculata*, Little Uchee Creek, Lee County, Alabama. Since the establishment of *T. hoffmani* as new species by Wellborn [25] this ciliate has not been recorded in elsewhere yet. However, during present study period *T. hoffmani* has been recorded as new country record in Bangladesh. Presently discussed specimens found from the gills of *M. tengara* have coincident morphological features of that originally described by Wellborn [25]. However, presently discussed specimen shows a range of variation in morphometric data. Body dimension of *T. hoffmani* that described by Wellborn [25] is larger than presently described specimens (Table 2) But, dimension of adhesive disc, denticular ring, central area, number of denticles, number of radial pins per denticles, and measurement of denticle component almost falls within the range of each range. Eight out of 30 (26.66%) of the examined *M. tengara* were found infected with *T. hoffmani*. The level of infection was low.

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