Description of two new species of ectoparasitic *Trichodina* Ehrenberg, 1830 (Ciliophora: Trichodinidae) from freshwater fishes in the river Ganges, India

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Abstract Two new species of the genus *Trichodina* Ehrenberg, 1838, *T. silondiata* sp. nov. and *T. pangasi* sp. nov. from the gills of freshwater fish *Silonia silondia* (Hamilton 1822) and *Pangasius pangasius* (Hamilton-Buchanan) respectively from the river Ganges of West Bengal are described here. Wet smears of gills and skins were prepared in the field, air dried and impregnated with Klein's dry silver method. In case of *S. silondia* (Hamilton 1822) 24 out of 146 host fishes were parasitized on the gills. Infestation rate in case of *P. pangasius* (Hamilton-Buchanan) was not significant. From a total of 86 examined host fish, only seven were parasitized on the gills. The mean diameters of the body of the specimens of *T. silondiata* sp. nov. and *T. pangasi* sp. nov. were 32.7–60.6 (46.4 ± 6.3) μm and 38.9–54.1 (44.9 ± 3.0) μm respectively. Taxonomic and morphometric data for these

ectoparasitic trichodinids based on wet silver nitrate impregnated specimens are presented.

Keywords Ectoparasite · Protozoa · Fish · River Ganges · *Trichodina silondiata* sp. nov. · *Trichodina pangasi* sp. nov. · India

Introduction

Trichodinid ciliates are well-known ectoparasites of molluscs, fishes and amphibians. Family Trichodinidae Claus, 1874 comprises ten genera including the largest and most widely distributed genus *Trichodina* Ehrenberg, 1838 having more than 250 species. Since the studies of Lom (1960, 1970), numerous species have been described from various parts of the world including India (Mitra and Halder 2004, 2005; Mitra and Bandyopadhyay 2006, 2009, 2012), Bangladesh (Asmat et al. 2003, 2006), China (Gong et al. 2006; Xu et al. 1999; Zhao and Tang 2011), North America (Wellborn 1967), Cuba and Russia (Arthur and Lom 1984a, b), South Africa (Basson and Van As 1991) Japan (Imai et al. 1991), Germany (Dobberstein and Palm 2000) and Egypt (Al-Rasheid et al. 2000).

The present paper is based on the outcome of the biodiversity survey of ectoparasitic trichodinid ciliates of freshwater fishes in the river Ganges in the area confined to the district of Murshidabad of West Bengal, India with taxonomic and morphometric descriptions of the two new species of the genus *Trichodina* Ehrenberg, 1830.

Materials and methods

During a biodiversity survey of ectoparasitic trichodinid ciliophorans in the freshwater fishes of the river Ganges in

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the Murshidabad district of West Bengal, host fishes were collected by fishermen between February 2011 and December 2011. Gill, fin and skin smears were made on grease free slides at the river side. Slides having presence of trichodinids were impregnated using Klein's (1958) dry silver impregnation technique. Examinations of prepared slides were made under an Olympus research microscope (Model CH 20i) at 1,000 $\times$ magnification with an oil immersion lens and photographs were taken with an Olympus digital camera. All measurements are in micrometers and follow the uniform specific characteristics as proposed by Lom (1958); Wellborn (1967) and Arthur and Lom (1984a). In each case minimum and maximum values are given, followed in parentheses by arithmetic mean and standard deviation. In the case of denticles and radial pins, the mode is given instead of the arithmetic mean. The span of the denticle is measured from the tip of the blade to the tip of the ray. Body diameter is measured as the adhesive disc plus border membrane. The description of denticle elements follows the guidelines of Van As and Basson (1989). Sequence and method of the description of denticle elements follows the recommendations of Van As and Basson (1992).

Results and discussion

Trichodina silondiat sp. nov. (Figs. 1–4, 7, 8; Table 1)

This trichodinid falls within the upper range of dimensions as a medium sized ciliophoran measuring 32.7–60.6 (46.4 $\pm$ 6.3) μ m. The concave adhesive disc is surrounded by a finely striated border membrane. Denticular apparatus consists of some uniquely shaped denticles.

Each blade is fleshy and triangular. It is very difficult to differentiate between the distal margin and the anterior margin of the blade. Blades keep themselves away from inner margin of the border membrane. Distal margin of the blades falls downwards with a smooth curve, takes a sharp turn to form a clear apex that just touches the $y + 1$ axis (Figs. 7, 8) in most of the cases. Anterior blade apophysis is present. Sharp tangent point lies below the distal margin. Posterior margin of the blade forms a moderately deep semilunar curve, the deepest point of which lies at the same level as apex. Posterior blade apophysis is absent. Central part of the denticle is broad, triangular. The posterior tip of the central part is bluntly rounded and reaches almost midway between the y and $y-1$ axis (Figs. 7, 8), fits tightly with the next denticle. The section of the central part above and below x axis is similar. Ray connection is short and broad. Ray is uniformly thick along its length, and directed to the geometrical centre of the adhesive disc in most of the cases.

There are 6–8 (7.1 ± 0.8) radial pins per denticle. Adoral ciliary spiral takes a turn of 390–400°.

Taxonomic summary

Type host: *Silonia silondia* (Hamilton 1822)

Type locality: River Ganges (24.16912°N and 88.32502°E), Berhampore, Murshidabad district, West Bengal, India.

Holotype: One slide no. CP1/2011 obtained from the gills of *Silonia silondia* (Hamilton 1822) collected at Berhampore of Murshidabad, West Bengal, India is deposited in the Department of Zoology, Ranaghat College, P.O. Ranaghat, Nadia, 741201, West Bengal, India.

Paratype: On the above numbered slide as well as on other slides (CP4/2011, CP/2011, CP16/2011) are deposited in the Department of Zoology, Ranaghat College, P.O. Ranaghat, Nadia, 741201, West Bengal, India.

Prevalence: 24/146 (16.43 %)

Etymology: The name of the new species has been given after the species name of the host fish, *Silonia silondia* (Hamilton 1822).

Discussion

The new species was compared with other fresh water and marine trichodinids and found three similar species with which a meaningful comparison could be made. These are: *Trichodina caspialosae* Lom 1962 from the gills of *Alosa braschnikowi milotica* in Rumanian Black Sea coast and *T. (lucioercae) nigra* Lom 1970 from the gills of *Stizostedion lucioerca* in the Hungary.

T. silondiat sp. nov. resembles *T. caspialosae* (Lom 1962) in the structure of its blade. In the new species the blades are broad, rectangular and occupying most of the areas between y -axis and anterior margin runs almost parallel to the y -axis. In the new species obtained from the freshwater fish *Silonia silondia* (Hamilton 1822) prominent anterior blade hypophysis is present which is absent in *T. caspialosae* (Figs. 9, 10). Central parts of the denticles of the new species are more robust, conical than that of *T. caspialosae*. Connection between the central part and ray of the denticle is more prominent in *T. caspialosae* than the new species. Rays of the denticles in *T. silondiat* sp. nov. are robust, with almost same width and pointing towards the centre of the denticulate ring. In few specimens of *T. caspialosae*, rays are shorter than the blades which end in comparatively sharp ends (Fig. 10). Biometrical data also differ in these two species.

The trichodinid species, reported from *Silonia silondia* (Hamilton 1822) also resembles *T. (lucioercae) nigra* Lom 1970 but may be easily differentiated by its denticle

Fig. 1 Adoral ciliary spiral of *Trichodina silondiata* sp. nov. **Figs. 2–6** Photomicrographs of silver nitrate impregnated adhesive discs of *Trichodina* spp. (**2–4**) *T. silondiata* sp. nov. (**5, 6**) *T. pangasi* sp. nov.

structure. The blades of the new species are broad and almost rectangular, which have triangular outline in *T. (luciopercae) nigra* (Fig. 11). Blades of the new species have prominent anterior hypophysis, which are almost lacking in the specimens reported by Lom (1970). Tangent points in the new species are mostly pointed, which are flat *T. (luciopercae) nigra* (Fig. 11). Connection between the blade and the central part in the new species are not prominent like that of *T. (luciopercae) nigra*. Rays of the new species obtained from the freshwater fish in the river

Ganges have prominent ray apophysis which are absent in *T. (luciopercae) nigra*. Rays of the denticle of the new species have same width along the entire length and tapers to blunt ends, while those of *T. (luciopercae) nigra* are gradually tapers to pointed ends. Morphometric data between the two also differ significantly (Table 1).

From the discussion above, it clearly appears that *Silonia silondia* (Hamilton 1822) harbors a new species of trichodinid ciliophoran and is proposed in this paper as *Trichodina silondiata* sp. nov.

Table 1 Morphometric comparisons of *Trichodina silondiatra* sp. nov. obtained in the present study with other closely relates species

Species	<i>T. silondiatra</i> sp. nov.	<i>T. caspialosae</i>	<i>T. (luciopercae) nigra</i>
Host	<i>Silonia silondia</i> (Hamilton 1822)	<i>Alosa braschnikowi nilotica</i>	<i>Steizostedion lucioperca</i>
Locality	River Ganges Berhampore, West Bengal	Black Sea, Rumania	River Tisza, Hungary
Location	Gills	Gills	Gills
Reference (s)	Present study	Lom (1962)	Lom (1970)
Diameter of			
Body	32.7–60.6 (46.4 ± 6.3)	26–33 (30)	41–55 (45–52)
Adhesive disc	25.2–44.6 (33.7 ± 6.6)	18–23 (21)	33–44 (37–40)
Dimension of body			
Denticulate ring	17.1–32.3 (22.5 ± 5.5)	11–14 (13)	19–26 (21–33)
Central area	6.8–13.4 (6.7 ± 1.7)	–	–
Width of the border membrane	3.5–6.2 (4.8 ± 1.1)	2.2–2.5	3.5–5.0
Number of denticles	21–23 (21.9 ± 0.9)	17–24 (21–22)	20–23 (20)
Radial pins/denticle	6–8 (7.1 ± 0.8)	–	7–9
Dimension of denticle			
Span	8.9–10.3 (11.5 ± 1.9)	–	–
Length	4.4–9.2 (6.0 ± 1.3)	2.7	8–9
Dimension of denticle components			
Length of the ray	3.2–7.3 (4.5 ± 0.8)	1.6	5–5.5
Length of the blade	3.4–6.5 (4.9 ± 1.0)	3.6	4.5–5.0
Width of the central part	1.4–3.2 (1.9 ± 0.3)	1.1	2.1–3.0
Adoral ciliary spiral (°)	390	370	–

Figs. 7–11 Diagrammatic drawings of the denticles of trichodinids. (7, 8) *Trichodina silondiatra* sp. nov. from *Silonia silondia* (Hamilton 1822) in the present study; (9, 10) *T. caspialosae* Lom 1962 from the gills of *Alosa braschnikowi nilotica*, redrawn from (Lom 1962); 11 *T. (luciopercae) nigra* Lom 1970 from the gills of *Steizostedion lucioperca*, redrawn from Lom (1970)

Trichodina pangasi sp. nov. (Figs. 5, 6, 12, 13; Table 2)

This new species is a medium sized ciliophoran with a body dimension of 38.9–54.1 (44.9 ± 3.0) μm. Blade is

broad and slightly angular. Distal surface of the blade is truncated and runs parallel to the border membrane that keeps a wide distance from the adhesive disc. Tangent point is flat, situated almost at the same level as distal surface. Blade fills most of the space between y-axis.

Table 2 Morphometric comparison of *Trichodina pangasi* sp. nov obtained in the present study with *Trichodina siluri* Lom 1970

Species	<i>Trichodina pangasi</i> sp. nov.	<i>Trichodina siluri</i>	
Host	<i>Pangasius pangasius</i>	<i>Silurus glanis</i>	<i>Silurus glanis</i>
Locality	River Ganges Berhampore, West Bengal India	South Bohemia	River Tisza Hungary
Location	Gills	Gills	Gills
Reference (s)	Present paper	Lom (1970)	Lom (1970)
Diameter of			
Body	38.9–54.1 (44.9 ± 3.0)	56 (47–60)	54 (42–60)
Adhesive disc	33.1–44.7 (34.3 ± 2.8)	43 (22–46)	40 (36–46)
Dimension of body			
Denticulate ring	17.4–24.6 (19.4 ± 1.8)	26 (22–30)	26 (23–29)
Central area	7.5–9.7 (7.2 ± 0.5)	–	–
Width of the border membrane	3.7–4.4 (4.5 ± 0.4)	4	3–5
Number of denticles	24–26 (25.4 ± 1.3)	25 (24–27)	25 (23–26)
Radial pins/denticle	4–8 (5.1 ± 0.6)	8	7–9
Dimensions of denticle			
Span	9.7–12.7 (10.5 ± 0.4)	–	–
Length of the denticle	4.4–6.8 (5.5 ± 0.9)	8	8–9
Denticle components			
Length of the ray	4.8–5.9 (4.8 ± 0.3)	6–7	6.5
Length of the blade	3.8–5.5 (3.9 ± 0.5)	5.5	5.5
Width of the central part	1.6–2.4 (1.3 ± 0.5)	2.5	2
Adoral ciliary spiral (°)	390–400	–	–

Figs. 12–15 Diagrammatic drawings of the denticles of trichodinids. (14, 15) *Trichodina pangasi* sp. nov. from *Pangasius pangasius*; (16, 17) *Trichodina siluri* Lom 1970, redrawn from Lom (1970)

Anterior blade surface is almost straight and runs parallel to the posterior blade surface as well as to the y-axis. Apex of the blade is not prominent. Inconspicuous apex (when present in some matured specimens) almost touches y-axis. Blade apophysis present. Posterior surface forms shallow, near perfect semilunar curve with deepest point at the same level of blade apophysis. Blade connection is broad. Posterior projection of the blade is not present. Central part is moderately sized, with triangular and flat rounded tip fitting

tightly into preceding denticle. Central part extends half-way to y-1 axis (Figs. 12, 13). Section of central part above and below x-axis is almost similar in shape. Ray connection is short and broad. Ray apophysis is present, situated very high and extending in an anterior distal direction. Rays are of uniform thickening along their length and flat rounded tips are pointed towards the y-1 direction.

Number of radial pins per denticle is 4–8 (5.1 ± 0.6). Adoral ciliary spiral takes a turn of 390–400°.

Taxonomic summary

Type host : *Pangasius pangasius* (Hamilton-Buchanan)
Type locality : River Ganges (24.16912°N and 88.32502°E), Berhampore, Murshidabad, West Bengal, India.

Holotype : One slide no. PP7/2011 obtained from the gills of *Pangasius pangasius* (Hamilton-Buchanan) collected at Berhampore of Nadia, West Bengal, India is deposited in the Department of Zoology, Ranaghat College, P.O. Ranaghat, Nadia, 741201, West Bengal, India.

Paratype : On the above numbered slide as well as on other slides (P27/2011, PP4/2011, PP14/2011) are deposited in the Department of Zoology, Ranaghat College, P.O. Ranaghat, Nadia, 741201, West Bengal, India.

Prevalence : 07/86 (3.77 %)

Etymology : The species name of the new species is given after the genus name of the host fish, *Pangansius pangansius* (Hamilton-Buchanan).

Discussion

Populations of trichodinid ciliophorans obtained from freshwater fish *Pangasius pangasius* (Hamilton-Buchanan), with a mixed infestation of a species of *Tripartiella* Lom 1958 were compared with other freshwater and marine trichodinids and *T. siluri* Lom 1970 was found to have a minor degree of resemblance with the populations harboring gills of the freshwater fish, *Pangasius pangasius*.

The shape of the adhesive disc obtained from *P. pangasius* is unique. The adhesive disc remains confined to the inner side of the body. As a result, the adhesive disc keeps a significant wide gap with the border membrane, expressing the orientation of the radial pins very beautifully. But in case of *T. siluri* (Lom 1970) (Figs. 14, 15) the gap is minimum between the border membrane and the adhesive disc. Structures of blades in both the species are somewhat identical, but in the described species apex and blade apophysis are present in majority of the specimens; these are absent in *T. siluri*. Tangent points in the new species are sharp, which are flat in *T. siluri*. Central part in the specimens under discussion is moderately sized and angular in shape; but in case of *T. siluri* it is sharply angular and conical. In case of the populations of trichodinids obtained from *P. pangasius*, rays are having very prominent ray apophysis which are totally absent in the specimens of *T. siluri* reported by Lom (1970). Morphometric data also varies significantly (Table 2).

With these distinct and important differences in denticle structures it is justified to give a new species status to the ciliate isolated from *Pangansius pangansius* (Hamilton-Buchanan) and we propose this trichodinid ciliophoran as, *Trichodina pangasi* sp. nov.

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