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Report of rotifer-ciliate-gastropod hyperepibiosis found on snail (Mollusca) from Goa, India

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AbstractHere we report a rotifer-ciliate-gastropod hyperepibiosis in freshwater canal, Goa, India. A ro-
tifer Philodina cf. megalotrocha Ehrenberg, 1832 was found attached on the colony of Epistylis
plicatilis Ehrenberg, 1838 which was epibiont on gastropod Pomacea bridgesi (Reeve, 1856).
This is first report of a member of Epistylis as an epibiont on Pomacea bridgesi.

Raport o hiperepibiozie wrotków, orzęsków i ślimaków znalezionych na ślimaku (Mollusca) z Goa w Indiach

Słowa kluczowe hiperepibioza, orzęski, wrotki, ślimak, słodkowodne, Indie

Streszczenie Artykuł dotyczy doniesienia o hiperepibiozie wrotków, orzęsków i ślimaków w kanale słodkowodnym w Goa w Indiach. Wrotek *Philodina* cf. *megalotrocha* Ehrenberg, 1832 został znaleziony w kolonii *Epistylis plicatilis* Ehrenberg, 1838, która była epibiontem na ślimaku *Pomacea bridgesi* (Reeve, 1856). Jest to pierwsze stwierdzenie *Epistylis plicatilis* jako epibionta na *Pomacea bridgesi*.

Introduction

Epibiosis is a facultative and interspecific association where one organism lives on the surface (epibiont) of another organism (basibiont) (Wahl, 1989). A number of peritrich ciliates are epibionts on freshwater invertebrates (e.g., Precht, 1935; Jankowski, 2007; Fernandez-Leborans, Tato-Porto, 2000; Green, 1974; Clamp, 1973; Clamp, Chatterjee, Fernandez-Leborans, 2016; Chatterjee, Fernandez-Leborans, Ramteke, Ingole, 2013).

Hyperepibiosis, in turn is the form of relation between organisms when an epibiont lodges on another epibiont harbouring on to a host (eg. Fernandez-Leborans, 2003b, 2013; Mestre et al., 2019). Ciliate-bryozoan-crustacean hyperepibiosis of the brachyuran decapods Goneplax rhomboids (Linnaeus,1758) colonized by the bryozoan Triticella flava Dalyell, 1884, collected in the north west Mediterranean coast hasbeen studied by Fernandez-Leborans (2003b). Ciliate hyperepibiosis on the hydrozoan Dicoryne conferta (Alder, 1856), attached to the shell occupied by hermit crab Pagurus bernhardus (Linnaeus, 1758) from the west coast of Scotland was studied by Fernandez-Leborans (2013). Ciliate hyperepibiosis was also noted on the ostracod Ankylocytheres inuosa (Rioja, 1942) attached to a crayfish Procambarus clarkia (Girard, 1852) in the Iberian Peninsulaand Balearic Islands SW Europe (Mestre et al., 2019). Suctorian ciliate Tokophrya fasciculata (Lopez-Ochoterena, 1964) was observed on stalks of suctorian T. quadripartita (Claparede & Lachmann 1859) and peritrich Epistylis plicatilis Ehrenberg, 1838, in turn colonising the shells of freshwater snails collected from Mexico (Lopez-Ochoterena, 1964). Recently, Chatterjee, Dovgal, Vieira, Dutta and Nanajkar (2020) reported Ciliates-bryozoancrustacean hyperepibiosis on crab (Decapoda: Brachyura) from the west coast of India, Arabian Sea. They reported ciliates Paracineta saifulae (Mereschkowsky 1877) and Cothurnia ceramicola Kahl 1933, that were found as epibionts on bryozoan Triticella pedicellata (Alder 1857), attached to the crab Atergatis sp.

Peritrich ciliates of the genus *Epistylis* Ehrenberg, 1830, are characterized by formation of colonies of zooids supported by non-contractile stalk and possession of a peristomial lip that folds outward when the oral area (peristome) is expanded ("epistyliform" peristome) (Clamp et al., 2016). The ciliates of genus *Epistylis* are often found attached on different substrates such as macrophytes, stones, and as unspecific epibionts of freshwater invertebrates, bugs, beetles, decapods, copepod crustaceans and gastropod molluscs (Jankowski, 2007). *Epistylis* were earlier reported as epibiont on gastropod shell (Sartini et al., 2018; Lopes-Ochoterena, 1964; Dias, D'Ávila, Agosto, 2006; Dias, Wieloch, D'Agosto, 2008; Utz, 2007).

In the present study, colonies of a species of *Epistylis* were discovered on a gastropod *Pomacea bridgesi* from Goa, India and identified as *E. plicatilis* Ehrenberg, 1832. This is first report of the member of *Epistylis* as an epibiont on this host.

Several species of rotifers have been found as epizoic on various aquatic invertebrates eg. on sponges (Berzins, 1950; Bołtruszko, 201; Bołtruszko, Ejsmont-Karabin, 2013), on crustaceans (Ahlstrom, 1940; Hollowday, 1949; Chengalath, Fernando, Koste, 1973; May, 1989; Cook, Chubb, Veltkamp, 1998; De Smet, Verolet, 2016; Dražina, Korša, Špoljar, Maguire, Klobučar, 2018), on molluscs (Boltruszko, 2011, Ejsmont-Karabin, Karpowicz, 2019).

However, the cases of rotifer epibiosis on peritrichs are poorly known e.g., unidentified rotifers along with nematodes, and occasional copepods were found on colonies of peritrich ciliate *Ophrydium versatile* (Müller, 1786), which was a basis to seen the peritrich colony as the consortia (Duval, Margulis, 1995).

In this study we report a peritrich-rotifer hyperepibiosis on freshwater gastropod mollusc. Here we presented the characteristics of rotifer *Philodina* cf. *megalotrocha* Ehrenberg, 1832 found on colonial ciliate *Epistylis plicatilis* Ehrenberg, 1838 attached on the gastropod *Pomacea bridgesi* (Reeve, 1856) from a freshwater canal of Goa, India.

Material and methods

Snail *Pomacea bridgesi* (Gastropoda, Mollusca) was collected by 3rd and 4th authors (Mandar Nanajkar and Aniket Desai) from road side canal (freshwater) of Goa (Lat. 15°28'14.9"N and Long. 73°48'36.5"E), India. Microscopic observations and photography was carried out for snail and its attachments, using Olympus-SZX10 stereoscopic microscope and upright Microscope Olympus-BX63 with a Camera DP74 and Cell Sense Software. Specimens on the host are kept in the 4th author's (Mandar Nanajkar) collection in National Institute of Oceanography Goa, India (CSIR-NIO).

Ciliate epibiont Epistylis plicatilis on snail Pomacea bridgesi

Phylum: Ciliophora Doflein, 1901 Subphylum: Intramacronucleata Lynn, 1996 Class: Oligohymenophorea de Puytorac et al., 1974 Subclass: Peritrichia Stein, 1859 Order: Sessilida Kahl, 1933 Family Epistylidae Kahl, 1935 Genus *Epistylis* Ehrenberg, 1832

Epistylis plicatilis Ehrenberg, 1838

Brief Description. Freshwater, colonial peritrich species with dichotomously branched uncontractible stalk. Zooids look like an elongated funnel. The peristomial lip some wider than the body, peristomial disk is somewhat elevated above the peristome. Macronucleus horseshoe-shaped, oriented transversally and positioned under peristome (Figure 1A).

Dimensions (based on measurement of 6 living zooids, in µm). Body length 91–123, maximal body width 31–45, peristomial lip width 39–51, peristomal disc width 27–43, stalk diameter near base of zooid 7–10, macronucleus length 34–49, macronucleus diameter 7–11.

Remarks. *Epistylis plicatilis* is a common ciliate found in freshwater reported from different parts of India (eg., Rajabunizal, Ramanibai, 2013; Agrawal, Sanjay Thiske, Mondal, 2014; Kour, Gupta, 2014; Bakare, Nalawade, 2019; Bakare, Supekar, Nalawade, 2019; Gupta, Kour, Devi, 2019)

Rotifer epibiont on ciliate *Epistylis* colony which is attached to freshwater snail *Pomacea bridgesi*

Phylum: Rotifera Cuvier, 1798 Class: Eurotatoria De Ridder, 1957 Subclass: Bdelloidea Hudson, 1884 Order: Philodinida Melone & Ricci, 1995 Family: Philodinidae Ehrenberg, 1838 Genus: *Philodina* Ehrenberg, 1830

Philodina cf. megalotrocha Ehrenberg, 1832

Material Examined. One rotifer individual found on ciliate *Epistylis plicatilis* colony attached to freshwater snail *Pomacea bridgesi*. **Brief Description.** *Philodina megalotrocha* has wide corona, which is much wider than cingulum. The sulcus is wide as well. One of the prominent diagnostic characteristics is the shape of trunk and rump. The trunk is plump and abruptly tapering to rump, which is much narrower than trunk; the dental formula is 2/2.

Remarks. There are six species of genus *Philodina* Ehrenberg, 1830 viz. *Philodina brevipes* Murray, 1902, *Philodina citrina* Ehrenberg, 1830, *Philodina flaviceps* Bryce, 1906, *Philodina indica* Murray, 1906, *Philodina megalotrocha* Ehrenberg, 1832, *Philodina roseola* Ehrenberg, 1832, *Philodina squamosa* Murray, 1906 and *Philodina vorax* (Janson, 1893), which were recorded from India. Four species viz. *Philodina brevipes*, *Philodina indica, Philodina squamosa* and *Philodina vorax* represented with only single report from India (Murray, 1906).

Philodina megalotrocha has been reported earlier in India from Eastern Himalayas (Murray, 1906), Andhra Pradesh (Kameswar, 1977) and Haryana (Chopra, Tyor, Kumari, 2014). Present report is the first report of this species from Goa state of India.



Figure 1. A. Epibiont colony attached on snail *Pomacea bridgesi* (Gastropoda, Mollusca); B. Epibiont colony of *Epistylis plicatilis*; C. Epibiont colony of ciliate *Epistylis plicatilis* and one rotifer *Philodina megalotrocha* (arrow indicating rotifer); D. *Epistylis plicatilis*, arrow indicating C shaped macronucleus; E. Two *Epistylis plicatilis* zooides and stalk; F. Magnified view of rotifer *Philodina megalotrocha* (arrow indicating rotifer)

Discussion

The *Epistylis plicatilis* is a ubiquitous freshwater species, which is reported from a wide variety of aquatic substrates, including aquatic plants, algae, crustaceans, insects and insect larvae, snails, and inanimate objects (Roux, 1901; Daday, 1910; André, 1912; Haempel, 1918; Kahl, 1935; Henebry, Ridgeway, 1979; Jankowski, 2007; Clamp et al., 2016; Keiser, 1921), also found in activated sludge of treatment facilities (Konstantynenko, Dovgal, 2009).

Epistylis plicatilis was frequently reported as epibiont on snail shell. For example, the species was found on shell of *Physa acuta* Draparnaud, 1805 in a eutrophic stream of Brazil (Sartini et al 2018), on *Lymnaea atenuata* Say, 1829 and *Physa osculan* Haldeman, 1841 in a lake from Mexico (Lopes-Ochoterena, 1964). Dias et al (2006) reported *Epistylis* sp. on *Pomacea lineate* (Spix, 1827). Dias et al. (2008) reported *Epistylis plicatilis* on *Pomacea fingulina* (Spix, 1827). Utz (2007) reported this species on *Pomacea canaliculata* (Lamarck, 1891) in southern Brazil.

Rotifer *Philodina megalotrocha* has been considered to be a cosmopolitan species (Donner, 1965; Koste, Shiel, 1986). It is found in variety of habitats; epibenthic typically found among aquatic plants in running waters, pool, ponds, lakes (Donner, 1965). *Philodina megalotrocha* can attach to the lorica of other rotifers and to other aquatic organisms (Hamdan, 2010). *Philodina megalotrocha* also has been recorded as living on bryozoans (May, 1989). It should be mentioned that Mossallam, Amer and Abou-Ei-Naga (2013) reported relationship between *Philodina* sp. and snail *Biomphalaria alexandrina*.

Dias, Cabral, Siqueira-Castro, Silva-Neto and D'Agosto (2010) reported high infestation of peritrich ciliate *Cachesium polypinum* on *Pomacea figulina* collected from a stream receiving domestic sewage. Which is similar to the present study, as collection point is situated in urban area influenced by domestic sewage. Increase in bacterial density (serves as primary food for ciliates) is caused by nutrient enrichment by discharge of untreated domestic sewage in to water bodies (Prime, 1998).

The colonies of sessile peritrichs are often colonized by other ciliate species. For example, suctorians *Tokophrya quadripartata* Claparede & Lachmann, 1879 has been reported attached to the stalk of colony of *Epistylis plicatilis* (Edmonson, 1906; Small, 1973; Dovgal, 2013). *Tokophrya fasciculata* (Lopez-Ochoterena, 1964) was observed at stalks of suctorian *T. quadripartita* and peritrich *Epistylis plicatilis*, in turn colonising the shells of freshwater snails *Lymnaea attenuata* and *Physa osculans* (Lopez-Ochoterena, 1964). *Urnula epistylidis* Claparede & Lachmann, 1859, *Mistarcon parasiticus* (Nozawa, 1939), *Trichophrya epistylidis* Claparede & Lachmann, 1859 were also registered on *E. plicatilis* (Jankowski, 2007; Dovgal, 2013). It is obvious that these carnivorous or parasitic species feed by zooids and telotrochs of *Epistylis*.

However, there are cases wherein filter–feeding peritrich species also inhabit the colonies of other peritrichs (Jankowski, 2007). For example, small colonial species *Epistylis epibioticum* Banina, 1983 colonizing the larger colonies of peritrich *Carchesium batorligetiense* Stiller, 1953, *E. balatonica* Stiller, 1931 and *E. entzii* Stiller, 1935 (Jankowski, 2007).

It is possible that the epibiosis is favoured by a higher food supply. It was found (Konstantynenko, Dovgal, 2009) that the nutrition rates per zooid in colonial peritrich species (*E. plicatilis, E. bimarginata* Nenninger, 1948 and *Opercularia phryganeae* Kahl, 1935) were higher than the solitary species (*Vorticella convallaria* (Linnaeus, 1758)) and *V. striata* f. *octava* Dujadrin, 1841). It is the Konstantynenko and Dovgal's opinion that team-work of peristomes of all zooids in colony increases the efficiency of food particle accumulation in comparison to

solitary species of similar size. In such a case smaller peritrich species also can take advantage of the water flow produced by larger species for intensifying their own feeding.

It is quite possible that multicellular filter-feeders such as bdelloid rotifers can attach to colonial peritrichs and may stimulate their feeding at the expense of host species.

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