First record of an epibiont protozoan *Epistylis* sp. (Ciliophora, Peritrichia) attached to *Ergasilus chelangulatus* (Ergasilidae) in Brazil

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Abstract
In the present paper, we described the first record of an epibiont protozoan *Epistylis* sp. Ehrenberg, 1830 (Ciliophora, Peritrichia) attached to *Ergasilus chelangulatus* Thatcher and Brasil-Sato, 2008, parasite of *Pimelodus maculatus* Lacépède, 1803 in Brazil, with electron microscope observations. Fish were collected in Veados River, state of São Paulo and the crustacean *Ergasilus chelangulatus* being registered for the first time in this river, expanding its geographical distribution in Brazil.

Keywords: *Epistylis*, *Ergasilus chelangulatus*, Veados River, Brazil, epibiosis.

Introduction

The genus *Epistylis* Ehrenberg (1830) includes approximately 100 described species of sessile ciliates, most of which are believed to live as epibionts on crustaceans, insects, rotifers, and aquatic plants in marine and freshwater environments (Utz, 2007). In addition, *Epistylis* spp. can also be found on the skin and fins of fish and on the carapaces of turtles (Crites, 1977).

Epibiosis is a facultative association of two organisms, the epibiont and the basibiont. The term “epibiont” includes organisms that, during the sessile phase of their life cycle, are attached to the surface of a living substratum, whereas the “basibiont” lodges and constitutes a support for the epibiont (Fernandez-Leborans and Tato-Porto, 2000).

Any host–parasite relationship is dynamic, shifting along a continuum precisely definable only at an instant in time and space. Much as in a waltz, the “partners” are constantly adapting to each other’s ‘moves’ in response to the presence, or potential presence, of one an other (Horwitz and Wilcox, 2005). Epibionts having crustaceans as hosts must be adapted to leave their host and to re-colonise the same or another substrate, after each moult (Utz and Coats, 2005). Some authors have found *Epistylis* attached only to adult copepods. According to Bozkurt and Genc (2009), *Epistylis* could be absent from the younger copepodid stages because moulting effectively removes epibionts. In addition, the greater body surface area of adults may make them more attractive to the epibionts.

According to Utz (2007), living hosts often represent a suitable attachment site for peritrichs and other sessile organisms, because the water currents passing by the host can supply the epibionts with food particles and can facilitate the removal of waste material. Mechanisms of recognition of a living substrate or of specific hosts by epibionts are still unknown. However, it is believed that chemical, mechanical, or even electrical cues may be involved in the recognition of the substrate for colonisation, as well as in the process of formation of free-living stages when the host moults or dies (Utz and Coats, 2005).

Crustaceans and molluscs commonly serve as substrata for sessile peritrich ciliates, and several previous studies have focussed on these relationships. Studies of epibiosis between *Epistylis* spp. and parasitic crustaceans are scarce.
Epistyliis sp. in Ergasilus chelangulatus

This topic has, primarily been investigated by Van As and Viljoen (1984), who have described *E. branchiophila* Perty-Stein, 1859, *E. epibrnimiana*, and *E. cyprinaceae* Van As and Viljoen, 1984 on *Lernaea cyprinaceae* Linnaeus (1758). Shafir and Oldewage (1992) have recorded *Epistyliis* sp. on *Argulus americanus* Wilson (1902); *A. mississippiensis* Wilson (1916); *A. lepidostei* Kellicott (1877); and *A. flavescens* Wilson, 1916 from Illinois, U.S.A. These authors have observed rust-coloured lesions characteristic of crustacean shell disease. Silva-Souza and Rosim (2005) have recorded *Epistyliis* sp. on lernaeid crustaceans from Córrego do Feijão, São Paulo, Brazil. Abdallah et al. (2011) recorded *Epistyliis* sp. on copepoda *Amplexibranchius bryconis* Thatcher and Paredes (1985) from Peixe’s River, the state of São Paulo, Brazil.

**Material and Methods**

We examined 10 specimens of the fish *Pimelodus maculatus* Lacépède (1803) collected from the Veados River (23° 15' 20.7"S 48° 37' 17"W), in the state of São Paulo, Brazil, from 25 to 27 July 2011 (Figure 1). The gills, skin, nostrils, and oral cavity were examined in search of parasitic crustaceans. The crustaceans found were fixed in 70°GL alcohol, were subsequently mounted between a slide and a coverslip using Gray & Wess, and were identified according to Thatcher and Brasil-Sato (2008). The sessile ciliates were mounted in Gray & Wess between a slide and a coverslip, since this mounting medium was very satisfactory for this group of parasites. Micrographs were taken with a Leica DM5000 microscope equipped with differential interference contrast (DIC) optics. Other specimens were prepared for scanning electron microscopy using standard techniques (Dedavid et al., 2007) and analysed with a FEI Company Quanta 200 scanning electron microscope in the Centre of Electron Microscopy of the Instituto de Biociências do Campus de Botucatu-UNESP. Voucher specimens of the Crustacea were deposited in the Coleção Helmintológica do Instituto de Biociências de Botucatu (CHIBB), in the state of São Paulo, Brazil, with the number CHIBB 7053.

**Results and Discussion**

In this study, we found the parasitic copepod, *Ergasilus chelangulatus* Thatcher and Brasil-Sato, 2008 (Figures 2 and 3) a member of the family Ergasilidae, order Poecilostomatoida parasitising the gills of *P. maculatus*. This species has been described by Thatcher and Brasil-Sato (2008) parasitising the gills of *P. maculatus* from the upper São Francisco River. The specimens collected here presented the measurements very similar to those found by Thatcher and Brasil-Sato (2008). Members of this family can produce epithelial hyperplasia, metaplasia, and lamellar fusion in the gill filaments of their hosts. Hence, they may cause the death of the fish by reducing respiratory efficiency and by paving the way for secondary bacterial invaders (Thatcher, 1991).

![Figure 1. Location of the reservoir Jurumirim no Alto Paranapanema River, SP and sampling area in the Veados River (arrow). (Source: satellite image of GoogleEarth - DigitalGlobe).](image-url)
Figure 2. *Ergasilus chelangulatus* Thatcher & Brasil-Sato, 2008. a, b. specimens with a large portion of its body covered by *Epistylis* sp.; c. legs of a specimen of *E. chelangulatus* covered by *Epistylis* sp.; d. head of a specimen of *E. chelangulatus* covered by *Epistylis* sp.

Figure 3. Scanning Electron Microscope (SEM) of *Ergasilus chelangulatus* Thatcher & Brasil-Sato, 2008, female. a. Dorsal view of basibiont with *Epistylis* sp. attached; b, c. Thorax with *Epistylis* sp. attached; d. Detail of insertion of *Epistylis* sp. on thorax of basibiont.
In the present study, all fishes were parasitised by *E. chelangulatus* (100%) and 20% of the copepods collected were in turn parasitised by *Epistylis* sp. on at least some part of the body. In some cases, this protozoan occurred over a large portion of the copepod’s body and on the copepod’s legs.

In general, epibiosis has been viewed as a commensal relationship. However, several studies have shown that epibionts affect their hosts in different ways, including decreased survival and fecundity, lesions and diseases, disturbance of locomotion, decreased competitive ability, increased susceptibility to predation, increased energy demands, and faster sinking rates (Bozkurt and Genc, 2009). The last-mentioned observations may thus imply that the presence of the ciliate protozoan could quite possibly have negative effects on the population of the copepod.

It is the first record of the ciliate *Epistylis* sp. attached to this crustacean in Brazil. After being described by Thatcher and Brazil-Sato (2008), this is the first record of *E. chelangulatus* parasitizing *P. maculatus* outside their type locality. This work has expanded the known geographic distribution of these two parasitic species.

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**References**


