Annals of Parasitology 2021, 67(1), 123–128
doi: 10.17420/ap6701.320

Short note

Ergasilus colomesus (Copepoda: Ergasilidae) parasitizing gills of *Colomesus asellus* (Tetraodontiformes: Tetraodontidae) in the western Brazilian Amazon

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ABSTRACT. Records about *Ergasilus colomesus* and its interaction with the host fish are scarce. There is only a report describing this species from specimens collected from the gills of *Colomelus asellus*, in the State of Amazonas, Brazil. In this sense, this is the first record of *E. colomesus* parasitizing *C. asellus* for the southwest of the Brazilian Amazon. Fish were collected in the Môa river, located in the municipality of Cruzeiro do Sul, State of Acre, Brazil, during drought and flood. The fish captured were analyzed, and the parasites found were measured, fixed, and identified. We calculated the parasite indices and evaluated whether the seasonal periods (drought and flood) influence the levels of parasite infestation in their hosts. The prevalence and infestation of *E. colomesus* in *C. asellus* were higher in the drought, indicating that the infestation of this copepod probably occurred during this period. The lower number of individuals found during the flood indicates that this is the period when *E. colomesus* detaches from its host to release of eggs into the environment. In this sense, this study expanded the distribution record of *E. colomesus* parasitizing *C. asellus* to southwestern Amazonia.

Keywords: ectoparasite, seasonality, interspecific relationship, fish population, freshwater

Introduction

The family of copepods Ergasilidae [1] includes 26 genera and approximately 260 species, mainly inhabiting freshwater environments [2]. The genus *Ergasilus* Nordmann, 1832 has a wide distribution [1], with more than 180 species [3], occurring in both freshwater and seawater. Adult females of ergasilids parasitize gills, fins, nasal cavities, tissues, and urinary bladder of host fish [2–5]. Most taxonomic descriptions of ergasilids are based on the morphology of ovigerous females found in hosts [6], rarely describing free-living males and pre-metamorphic adult females, both planktonic forms [2].

The *Colomesus asellus* Mueller and Troschel, 1849 (Tetraodontiformes) is a small fish with great ornamental potential. It is widely distributed in freshwater environments, occurring mainly in Amazonian rivers [7]. Its parasitic fauna is poorly studied [8], where *Ergasilus colomesus* is one of the only parasites registered for this species [9]. *E.*

colomesus is a species recorded exclusively parasitizing the gills of *C. asellus*, on the Amazon River in Manaus, State of Amazon (Brazil), in 1983 [9]. Since then, this copepod has not yet been reported.

In this sense, this study aimed to describe the second report and a new location of occurrence of *E. colomesus* parasitizing *C. asellus*, in addition to evaluating for the first time, the influence of drought and flood on the parasite infestation in the host.

Materials and Methods

Study area

The fish were collected in the Môa River, located in the municipality of Cruzeiro do Sul, State of Acre, western Brazilian Amazon (7°37'14.9"S 72°47' 38.4"W) with authorization from the Brazilian Institute of the Environment and Renewable Natural Resources (ICMBio) – No. 59642-2/2019 (Fig. 1). The collections were performed during the flood (January 2019) and drought (August 2019). The region's climate is equatorial, hot, and humid, with two well-defined seasons: a drought season, which usually occurs from June to November, characterized by low rainfall (monthly average of 125.5 mm) and higher temperatures (32 to 40ºC). The flood period, which occurs from December to May and is characterized by intense rainfall (monthly average of 231 mm) and lower temperatures (27 to 30ºC) [10].

Sampling

The fish were caught using a 9 m wide and 2.4 m high trawl with 13 mm mesh on the wings. After being collected, the fish were kept in boxes with an oxygenator and transported to the Laboratory of Aquatic Ecology of the Federal University of Acre (UFAC). The species were identified through morphological characteristics described by Tyler [11], and then, each fish was measured (total length – cm), weighed (g), and necropsied. The viscera and gills were removed for the parasitic fauna analysis using a stereomicroscope (with 100× magnification). However, only the gills had copepod ectoparasites. The parasites found were fixed and preserved in 70% ethanol. The parasite identification was based on

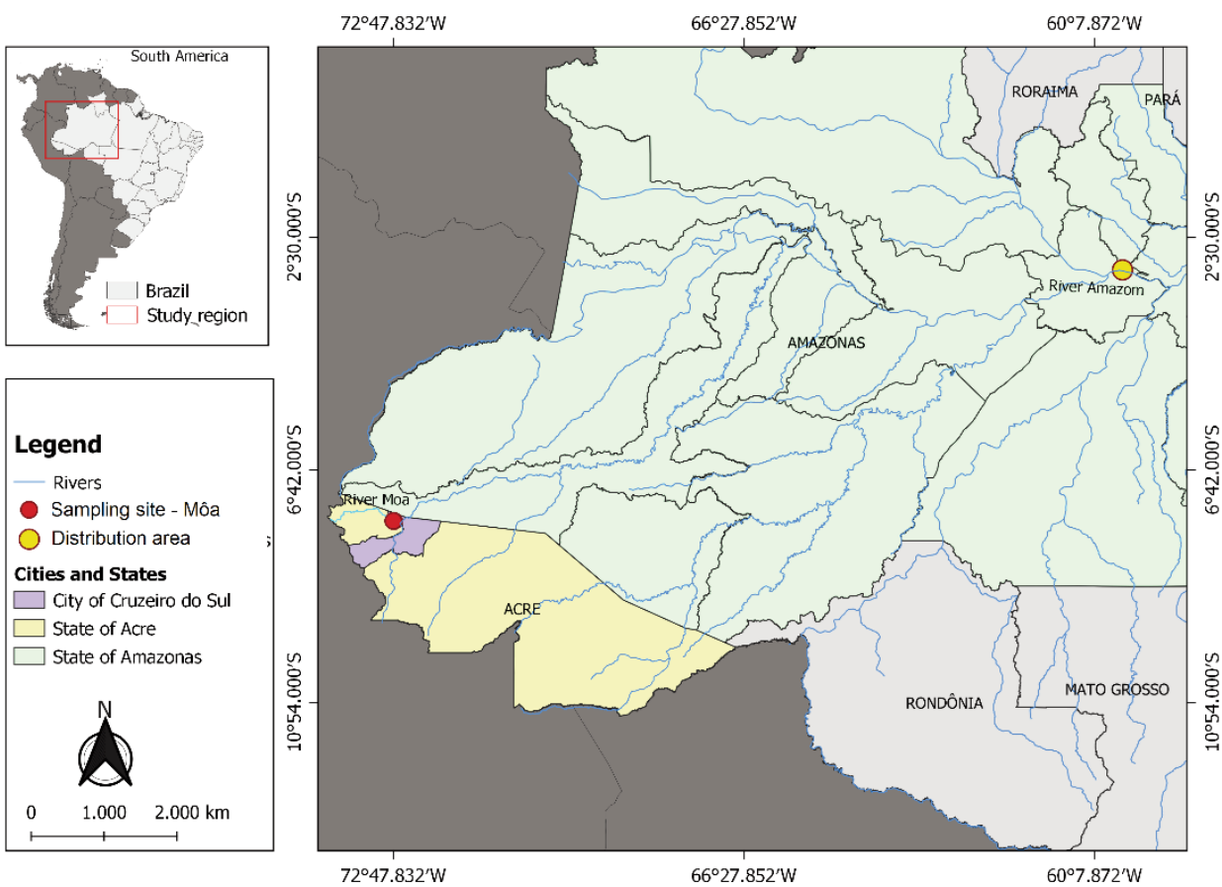

Figure 1. Collection site of *Colomesus asellus* in the Central and Southeast regions of the Amazon

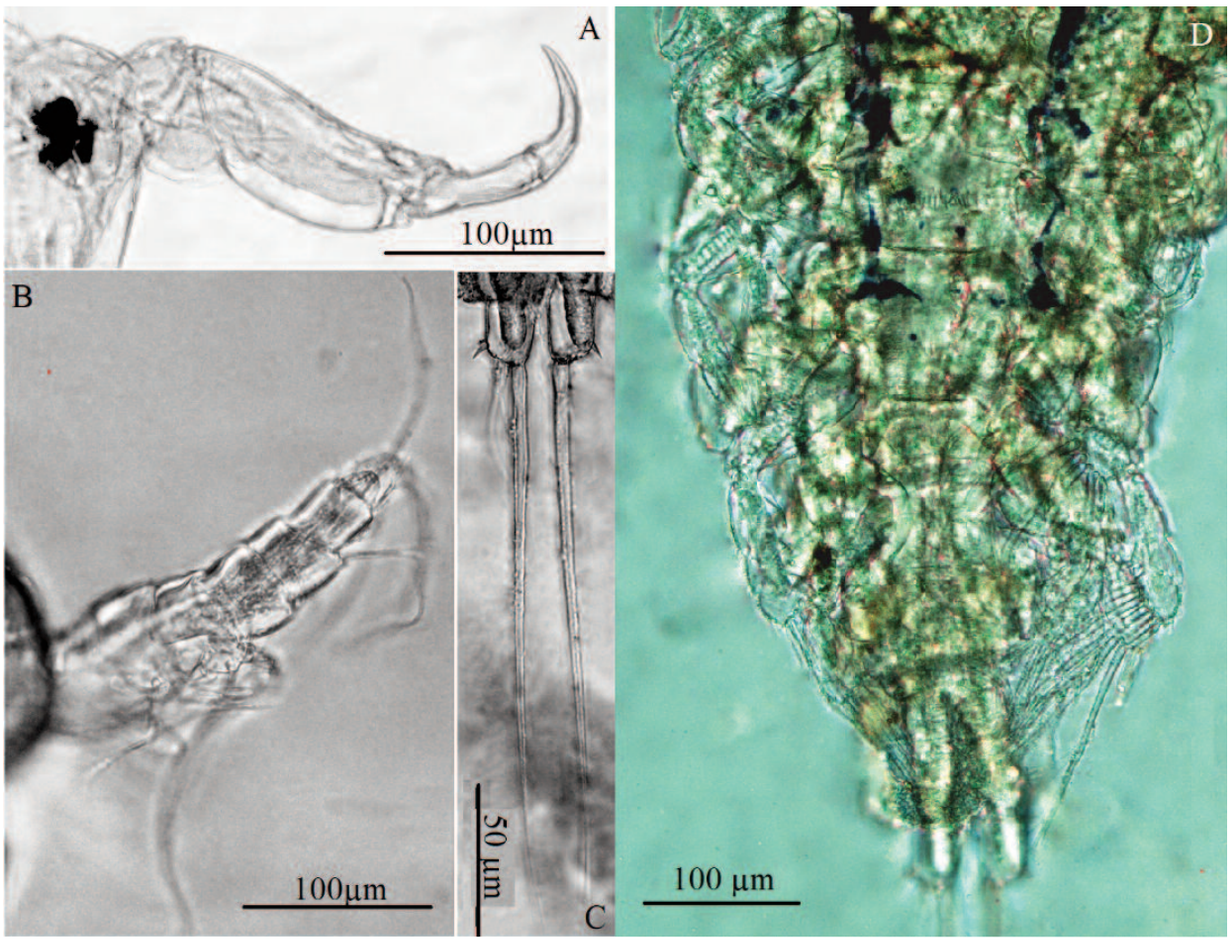

Figure 2. A – cephalic region of *Ergasilus colomesus* Thatcher and Boeger, 1983, with the ocular area and second antennae (prehensile), dorsal view, composed image (5 stacks); B – first antennae, dorsal view; C – uropods, ventral view; D – abdomen with appendices, ventral view, composed image (5 stacks)

morphological studies by Thatcher and Boeger [9].

Images were captured using a microscope Zeiss Axio Scope.A1 equipped with an Axiocam 503 color 60N-C 2/3" 0.63×. Measurements are in micrometers (µm). The composed images were taken with five manual stacks. The specimens were deposited in the invertebrate's collection of the Laboratory of Animal Biology, Federal University of Acre, Cruzeiro do Sul, Acre, Brazil (CBLA-In), curator E. O. Machado. Terminology follows Thatcher and Boeger [9].

Data analysis

The prevalence (%), mean abundance and mean intensity were calculated to assess the levels of parasite infestation Bush et al. [12]. The Shapiro-Wilk normality test was performed to assess data normality, where the data were non-normal. The Mann-Whitney (*U*) test was applied to test for differences in total abundance, mean abundance, and mean intensity of parasites between the dry and flood periods. The analyses were performed using the STATA software (Stata Corp., College Station, United States).

Results

Host: *Colomesus asellus* Mueller and Troschel, 1849
Site: Gills filaments
Locality: Amazon River, Manaus, State of Amazonas; Môa river, Cruzeiro do Sul, State of Acre, Brazil

The comparison of morphological characteristics was based on ten females of *E. colomesus*. The identification of *E. colomesus* was confirmed after exams on structures of taxonomic relevance, comparing them with Thatcher and Boeger [9] and further descriptions of another species. The study shows some remarks about morphology with taxonomic relevance. Thatcher and Boeger [9] states: "The copepod *E. colomesus* diagnosis presents the following morphological characteristics": "(1)

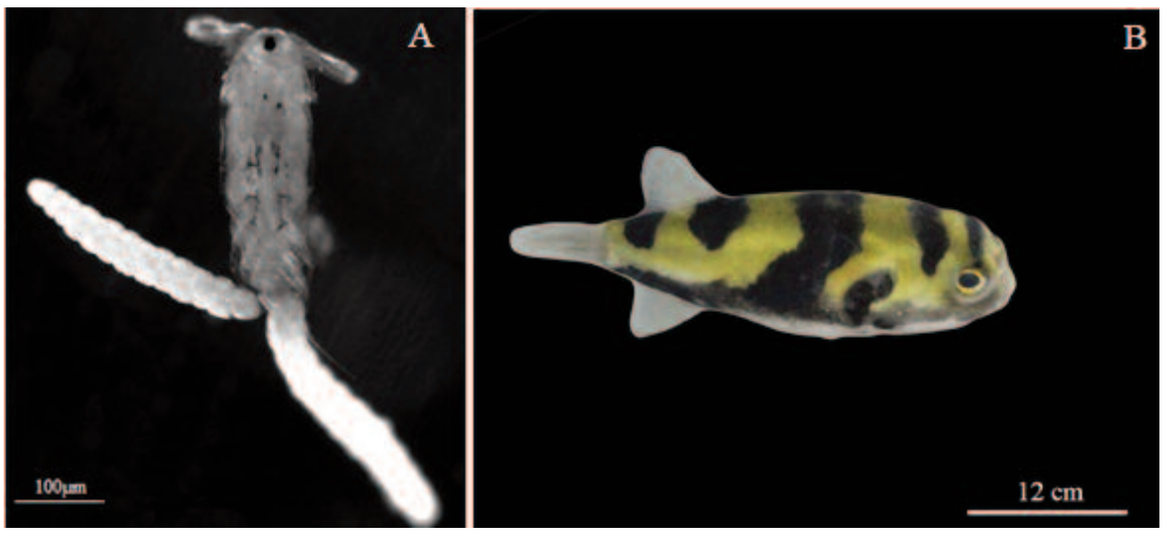

Figure 3. A – full body with eggs, dorsal view; B – *Colomeus asellus*

inflated segment 1 on the prehensile antenna; (2) two-segmented endopod 1; (3) two-segmented exopod 4; (4) three-segmented endopod 4; and (5) a sparsely pilose long seta, three short simple setae, and four spinules on the extremity of each uropod". The specimens examined were compatible with the original description and showed high similarity to the drawings in the original description. The first and second antennae presented the same proportions, shapes, setae, and spines (Fig. 2A,B). The prehensile second antennae were composed of four segments, with the same prolateral spines in the third segment with minor variation, and the terminal claw with the same curvature (Fig. 2A). Prominent eyespots, head fused with inflated thoracic segments with a small medial constriction, five free segments, and the abdominal segments with the same proportions and spinules (Fig. 3A). The uropods were slightly different, with longer subterminal lateral spines (Fig. 2C). All legs had the same shape and proportion, legs 3 and 4 with similar small spinules and plumose setae, but with slightly smaller lateral stout spines on endopod 3 and exopod 2 and 4 segments (Fig. 3A). Egg sac had a similar description with approximately 25 eggs each (Fig. 3A). The main difference resides in coloration. Both fresh and preserved (ethanol 70%) showed narrow black-bluish parallel lines with small lateral ramifications on free thoracic segments (Fig. 2D, 3A). This description differs significantly from Thatcher and Boeger [9] description: "Body pigmentation in ventrally situated longitudinal bands, color blue smalt in the head region and campanula, posteriorly". The original drawing shows strong and wide longitudinal bands ranging from cephalic regions to the free thoracic segments. Thatcher and Boeger [9] propose coloration "to be of some reliability in distinguishing the Amazonian species of *Ergasilus*. Moreover, these colors remain unchanged in alcohol, glycerin jelly, and balsam, for at least 15 years. Coloration could, therefore, be used to greater advantage if collectors preserved their material in alcohol". Besides the proposal being consistent, this trait can be useful but easily variable among populations, which is probably more useful below the species level.

We collected 37 specimens of *C. asellus* (Fig. 3B), all females, 16 specimens in the dry season (length: 5.5 ± 0.6 cm and weight: 5.0 ± 1.2 g) and 21 in the flood season (length: 6.4 ± 0.7 cm and weight: 6.0 ± 1.0 g).

The individuals of *C. asellus* were parasitized only by specimens of *E. colomesus*. A total of 35 specimens were collected from gill filaments of hosts, with 30 specimens collected during the drought and only five specimens during the flood. The predominance of parasites in hosts was significantly different between the seasonal periods ($U = 3.27$; $p = 0.01$).

The parasite prevalence was 37.5% in the drought and 23.8% in the flood period. The mean parasite intensity in the drought (MI = 5.0 ± 0.86) was higher than in the flood (MI = 1 ± 0.08) ($t = 2.75$; $p = 0.002$). The mean abundance of parasites in the drought (MA = 1.87 ± 0.04) was also higher than in the flood (MA = 0.23 ± 0.02) ($t = 4.15$; $p = 0.001$). The range of intensity of *E. colomesus* by *C. asellus* was 1 in the flood period and 4-7 in the drought.

Discussion

This study expanded the distribution of *E. colomesus* parasitizing *C. asellus* to southwestern Amazon, 36 years after its first and only description in the State of Amazonas, in Brazil [13] (Fig. 3).

Most *Ergasilus* species are distributed in freshwater environments, such as the Amazon River basin [13]. Some species show specific behavior regarding the type of host, such as the relationship observed between *E. colomesus* and *C. asellus*, which has hitherto not been reported for other fish species. This specificity in the relationship between Ergasilidae and host fish has been observed in *Erga - silus coatiarus* Araujo and Varela, 1998, specifically for cichlid species [14].

In *C. asellus*, prevalence and infestation of *E. colomesus* were higher in the drought, indicating that the infestation of this copepod probably occurred during this period. Species of the genus *Ergasilus* have a monogenean life cycle and only females are parasites because males die after copulation [15]. Thus, after copulation, females attach themselves to the fish gills, where they remain until egg maturation. After that, they release from the hosts, laying the eggs in the environment, initiating the new life cycle [16]. Therefore, the highest levels of parasite infestation observed during the drought is a strong indication that *E. colomesus* reproduces during this period in that region. The lower number of individuals found during the flood indicates that this is the period when *E. colomesus* detaches from its host to release of eggs into the environment. However, seasonal studies considering a shorter time interval are crucial to establish the infestation pattern of this parasite, as seasonal changes represent a combination of biotic and abiotic factors that influence the parasite's success in finding its host [5,17].

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Acknowledgements

We thank the Coordenação de Aperfeiçoamento de Pessoal de Nível Superior (CAPES) for granting a PhD scholarship to Oliveira, MSB. To the Laboratory of Animal Biology of the Federal University of Acre, in the figure of Professor Dr. Tiago Lucena da Silva for assisting in the analysis of the specimens.

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Received 31 July 2020
Accepted 23 February 2021