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The microhabitat, behavior and diet of *Centromochlus meridionalis*, a small catfish of Amazon streams (Teleostei: Auchenipteridae)

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Centromochlus meridionalis was recently described from streams of Brazilian Amazon. The marbled dark brown and black color pattern together with the use of submersed leaf litter accumulations for shelter differentiates this species among its congeners. We present information about the biology of *C. meridionalis* based on data gathered in streams and under captivity, as well as on a dietary analysis. Behavioral observations were made in captivity (aquaria). Dietary analysis was performed based on stomach contents of 38 specimens. The species was found in 8 out of 12 small streams sampled, where specimens were captured predominantly amidst submerged leaf litter. Nine types of behavioral acts were identified, of which "swimming near the substratum" and "charging the substratum for food" were the most frequent. Thirty (~79 %) out of the 38 stomachs had food, and the diet was composed of 27 types of food items. *Centromochlus meridionalis* can be considered a generalist microcarnivore, consuming predominantly authochtonous and allochtonous insects, and other terrestrial invertebrates, crustaceans and fish as well. The use of different water column strata during foraging and the diversity of food items consumed indicate that this small caffish utilizes several feeding tactics.

Introduction

The catfish family Auchenipteridae is endemic of the Neotropics and broadly distributed in South America, with 20 genera and approximately 90 species (Ferraris, 2003, 2007). Most of those species are active at night and at dusk, seeking refuge during the day in deep water or in cavities in logs and rocks (Rodriguez et al., 1990). Auchenipteridae are the only siluriforms that possess a copulatory

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structure and a reproductive pattern in which the male uses its modified anal fin to inseminate the female (Loir et al., 1989; Soares-Porto, 1998; Downing Meisner et al. 2000). Males of some species can also develop temporary structures, such as bony whiskers and hypertrophied fin spikes, during the reproductive period (Burgess, 1989; Akama & Ferraris, 2003). The Auchenipteridae is composed of two subfamilies: Auchenipterinae, which includes larger species that usually inhabit rivers, lakes and swamps, and Centromochlinae, including small species found in rivers as well as in small streams (Santos et al., 2004; Cabalzar et al., 2005).

Currently, the Centromochlinae includes 31 valid species, in the genera Tatia, Glanidium, Gelanoglanis and Centromochlus (Ferraris, 2007). Most of the literature on the subfamily involves the description of new species (Sarmento-Soares & Buckup, 2005; Sarmento-Soares & Martins-Pinheiro, 2008), a systematic review (Sarmento-Soares & Martins-Pinheiro, 2008) and lists of species (Ferraris, 2003, 2007), with relatively little information on the biology of the species and related to feeding habits (Ortêncio Filho et al., 2001; Cassemiro et al., 2005). *Centromochlus* comprises 14 valid species (Brindelli et al., 2015). Most of the information about the biology of the genus is scattered in studies related to ichthyofaunal surveys (Mees, 1974; Santos et al., 2004; Souza et al., 2012), phylogenetic relationships (Birindelli, 2010), and a report of accidents caused by the spines (Sazima et al., 2005). All species of the genus (except C. meridionalis) have crepuscular habits, foraging near surface.

Centromochlus meridionalis is a small species inhabiting streams of the Teles Pires River basin, Tapajós basin (Sarmento-Soares et al., 2013). Specimens were captured from underwater leaf litter accumulations in small 1st- and 2ndorder streams. In contrast, some congeners (e.g., C. heckelii) tend to occupy higher-order streams and large rivers (Sarmento-Soares et al., 2013). Centromochlus meridionalis is similar to species of Trachelyopterus (Auchenipterinae) with respect to general morphology and especially color pattern (Sarmento-Soares et al., 2013). The marbled dark brown and black body pattern may serve as cryptic coloration among the submersed leaves (FGC, pers. obs.). Therefore, the particular color pattern and the use of a microhabitat that is unusual for Centromochlus species (submerged leaf litter accumulations) indicate that this species may have different behavioral characteristics from the other members of the genus. In this study, we present information about the behavior of *C. meridionalis* based on information about its natural habitat, its behavioral repertoire in captivity and the results of a dietary analysis of preserved specimens.

Materials and methods

Specimens of *Centromochlus meridionalis* were collected in 1st- and 2nd-order streams (cf. Strahler, 1957) of the Teles Pires River basin (11°35'59"S 55°15'21.0" W) in June and July 2011, during the dry season in the region. The streams are located in Amazon Forest remnants in the municipality of Cláudia, Mato Grosso, southern Brazilian Amazon. This region is covered by vegetation classified as Transitional Amazon Forest (Ferreira et al., 1999).

The fish were collected actively during the day by two collectors that explored all microhabitats in a stretch of 150 meters with sieves and hand nets. All the leaf litter in the stretch were searched for fish. Minnow traps were set in the same stretch in the afternoon of the day before and left overnight in the stream. The fish were kept alive in plastic boxes with aeration and transported to the laboratory of the Acervo Biológico da Amazônia Meridional (ABAM) at Universidade Federal de Mato Grosso (UFMT), Sinop campus.

Voucher specimens were deposited in: ABAM, fish collection of Acervo Biológico da Amazônia Meridional, Sinop; INPA, Instituto Nacional de Pesquisas da Amazônia, Manaus; MBML, Museu de Biologia Prof. Mello Leitão, Santa Teresa; and MNRJ, Museu Nacional do Rio de Janeiro, Rio de Janeiro. All from Brazil: Mato Grosso state: Loanda stream, a small tributary of Roquete river: ABAM I-00316, 12, 23.8-42.8 mm SL; 11°25'33.1" S 55°16'39.3" W; F. G. Cabeceira, W. S. de Moraes & J. Dambroz 24 July 2010. - ABAM I-00317, 11, 31.7-43.7 mm SL; 11°25'42.7" S 55°16'34.6" W; F. G. Cabeceira, W. S. de Moraes & Amanda F. Mortati, 25 July 2010. – ABAM I-00318, 9, 9.4–48.5 mm SL; 11°35'59.1" S 55°15'21.0" W; F. G. Cabeceira, W. S. de Moraes & Márcia C. V. dos Santos, 9 July 2010. – ABAM I-00398, 2, 53.2–60.2 mm SL; 11°25'48.7" S 55°20'16.30"W; F. G. Cabeceira & E. Barbosa, 6 May 2011. - INPA 37893, 10, 40.4-47.9 mm SL; 11°25'33.1"S 55°16'39.3"W; F. G. Cabeceira & E. Barbosa, 8 May 2011. - MBML 5617, 3, 32.2-46.2 mm SL; 11°25'33.1" S 55°16'39.3" W; F. G. Cabeceira & E. Barbosa, 8 May 2011. – MNRJ 40701, 2, 51.8–52.3 mm SL; 11°25'33.1" S 55°16'39.3" W; F. G. Cabeceira, W. S. de Moraes & J. Dambroz, 24 July 2010.

Habitat. The characterization of the habitat was based on descriptive parameters of the stream environment and on information about the structure of the submersed leaf accumulations. Environmental parameters were measured according to the protocol proposed by Mendonça et al. (2005). The stream's width, depth, current speed, vegetation (canopy) cover, and limnological data (pH, temperature) were measured. In meandering streams leaf litter accumulations occur in depositional areas, forming discrete (i. e. discontinuous) habitats to the aquatic fauna.

Behavioral observations. All the observations were performed on two groups of fish, each consisting of four specimens and housed in separate tanks (80 and 50 liters), both containing submerged branches and leaves on a sand substrate. The fish were fed with granulated commercial aquarium fish food and living coleopteran (Tenebrionidae) larvae. The behavioral observations were performed only in the larger tank. During the observation sessions, a blind (a black cloth) separated the tank from the observer to avoid disturbing the fish. After 10 observation sessions, the fish group in the larger tank was changed and a similar set of observations was made with the second group.

Before conducting the quantitative behavioral measurements, 16 hours of preliminary observations were made, aiming to identify the main activity period and to record the behavioral repertoire. To quantify the observed behavioral acts, standardized observation sessions were conducted at night from 6:00 pm to 6:00 am, divided into 13 sessions of 20 minutes each and totaling 4.3 hours. Each group of C. meridionalis was observed for 10 non-consecutive nights, resulting in a total of 86 hours of observations for both groups. The observations were performed using all-occurrences sampling for a general characterization of the behavior and focal-animal sampling to count the number of times a given fish performed each behavior (Altman, 1974). The results are presented as frequencies of occurrence (%) in relation to the total behavioral acts recorded.

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Diet analysis. The diet was analyzed based on the stomach contents of 38 specimens (ABAM I-00316, ABAM I-00317, ABAM I-00318) preserved immediately after collecting in the field. The fish were measured (standard length, SL, in mm) and weighed (g), and the stomach contents were analyzed under a stereoscopic magnifier. The food items were identified to the lowest taxonomic level possible and later grouped into seven categories: fish, crustaceans, aquatic insects, terrestrial insects, other terrestrial arthropods, insect fragments and organic debris.

The fullness level of each stomach was estimated visually and categorized as follows: 0 % = empty stomach; 10 % = 0-10 % of the volume of the stomach filled with food; 25 % = 10-25 %; 50 % = 25-50 %; 75 % = 50-75 %; and 100 % = stomach totally full (adapted from Goulding et al., 1988). The relative volume (R.V.%) occupied by each item in the stomach was assessed considering the whole amount of food in the stomach as 100 %, according to Soares (1979). To correct the R.V.% values for the differences in the amount of food in each stomach, the values of relative abundance were then multiplied by the degree of stomach fullness. Finally, the R.V.% values were calculated by dividing the sum of values for each food category by the total volume of food consumed by the specimens analyzed. The frequency of occurrence (F.O.%) of the food items was also assessed as proposed by Hyslop (1980), according to the formula F.O.% = $(n^{\circ} \text{ of stomachs})$ with item i/n° of stomachs with food) \times 100. These two parameters (R.V.% and F.O.%) were used to calculate the Alimentary Index (A.I.) (Kawakami & Vazzoler, 1980), in which A.I. = $(R.V.\% \times F.O.\%)$ Σ R.V% × F.O.%) × 100.

Results

Microhabitat. Specimens of *C. meridionalis* (Fig. 1) were collected in 1st- and 2nd-order streams 1.8–3.2 m wide and 0.2–0.5 m deep, current speed 0.2–0.4 m \cdot s⁻¹, and discharge 0.1–0.3 m³ \cdot s⁻¹. Vegetation cover by riparian forest canopy averaged 88.6 %. The water was acidic (mean pH 4.9; min=4.2, max=5.4), and the mean temperature was 23.7 °C (min=22.8, max=24.5). The stream bed was composed primarily of sand (45 %; grains approximately 0.6–2.0 mm in diameter), fine sediment (20.4 %), coarse litter (17.6 %; leaves and small branches), fine litter (8 %; finely particulate



Fig. 1. *Centromochlus meridionalis* photographed alive in aquarium. **a**, female, 60.2 mm SL; and **b**, male, 53.2 mm SL (ABAM I-00398). Arrow indicates modified anal fin forming the intromittent organ. (Photographs L. N. Carvalho and F. G. Cabeceira).

plant matter) and logs (4.4 %; wood with diameter greater than 10 cm).

Centromochlus meridionalis was found in 8 of 12 streams sampled in the Teles Pires River basin, and co-occurred with 30 other fish species (FGC, pers. obs.). In a survey performed with standardized sampling effort during the dry season of 2010–2011, *C. meridionalis* was the second most abundant fish species in the studied streams, outnumbered only by *Moenkhausia phaeonota* (Characiformes: Characidae). An average of 9 individuals (range: 1–32) of *C. meridionalis* were collected per 150-m long stream stretch. The catfish was primarily captured in loosely submersed leaf litter accumulations. On most of those occasions, two or three specimens were captured in a single leaf litter pack, with a maximum of five individuals (in that case, three adults and two juveniles). Individuals were also captured under submerged logs greater than 10 cm in diameter.

Behavior in aquarium. Behavioral observations revealed peak activity between 8:00 pm and 3:00 am, with individuals resting after dawn. Nine distinct behavioral acts were identified and divided into four categories: resting, moving,

Table 1. Characterization and frequency of behavioral acts performed by individuals of *Centromochlus meridionalis* in captivity. Total number of behavioral acts = 4190.

Behavioral category Behavioral act	Description	Frequency
Resting		
Resting over sand	Remains exposed over sand substrate	5.9 %
Resting among leaves and logs	Rests between submerged leaves and logs	13.3 %
Settled on the substrate	Undulating body movements while stationary, sometimes dis- rupting sand particles and dead leaves	1.6 %
Movement		
Swimming near the substrate	Swimming close to the substrate (maximum 5 cm above it)	45.6 %
Swimming in midwater	Swimming in middle layer of water column (5 cm above substrate and 5 cm below surface)	7.7 %
Swimming close to the surface	Swimming in top 5 cm of water column	6.9 %
Feeding		
Swimming rapidly toward food on the substrate	During swimming close to the substrate and after detecting food, the individual assumes a head-down position and swims forward rapidly, biting the food item	17.5 %
Picking food at the surface	The individual swims rapidly towards food floating at the surface, engulf it and returns rapidly to midwater	1.4 %
Interaction		
Agonistic interactions	When two individuals moving around the tank approach each other closely (less than 2 cm), both suddenly move away to avoid physical contact	0.1 %



Fig. 2. Illustration of behavioral acts performed by *Centromochlus meridionalis*: **a**, resting between leaves; and **b**, feeding at the bottom in head-down posture. For a description of the behaviors see Table 1. (Illustrations by Cristiane C. de Freitas).

feeding and agonistic interactions (Table 1; Fig. 2). The most frequent behavior was swimming near the substrate (Frequency of Occurrence = 34 %), food searching in the substrate (17.5 %), resting among leaves and logs (13.3 %) and swimming among submerged leaves and logs (11.6 %).

Movements across the aquarium were performed by rapid swimming and sudden changes in direction. During swimming, the fish often remained close to the substrate but also explored the middle layer of the water column at times. Swimming near the surface was less frequent and always occurred for very short periods (a few seconds) before returning to lower layers. At times, the fish quickly swam to the surface and returned to the bottom. The greatest activity was observed during feeding. When food was offered, the fish swam frenetically close to the substrate throughout the tank, over open sand and between leaves and logs. After locating a food particle, the catfish quickly positioned head-down and bit the food, using rapid mouth movements (Fig. 2b). Only during the last observation sessions the fish began to search for food on the water surface, which was interpreted as conditioned feeding behavior.

Diet. The specimens analyzed for stomach content analysis measured 9.3-52 mm SL and weighed 0.1-4.2 g. Of the 38 stomachs analyzed, 8 (21.1 %) were empty, 8 (21.1 %) showed up to 10 % of fullness, 3 (7.9 %) were 25 % full, 5 (13.2 %) were

50 % full, 9 (23.7 %) were 75 % full, and 5 (13.2 %) were 100 % full. The food items were grouped in 13 categories (Table 2). In terms of volume, debris appear with R.V. = 25 %, Ephemeroptera with R.V. 16 %, Hymenoptera (ants) with R.V. 15 % and Trichoptera with R.V. = 15 %. The most frequent

Table 2. Values of Alimentary Index (A.I.%) for the items found in stomach contents of 30 specimens of *Centromochlus meridionalis*, with their respective Frequency of Occurrence (F.O.%) and Relative Volume (R.V.%).

Food item	F.O.%	R.V.%	A.I.%
Insect fragments	13	3	1.4
Terrestrial insects Hymenoptera (ants)	33	15	18.7
Terrestrial arthropods Spirostreptida (Diplopoda)	3	2	0.3
Aquatic insects (mostly larvae) Trichoptera Ephemeroptera Hemiptera Plecoptera Megaloptera Diptera Coleoptera	37 10 10 3 3 3 17	15 16 2 2 1 0.2 9	21.1 6 0.6 0.2 0.1 0.02 5.4
Fish Moenkhausia phaeonota	7	2	0.3
Crustaceans Decapoda (shrimps)	3	2	0.3
Crustaceans Decapoda (shrimps) Debris	3 47		2 25

items were debris with F.O.=47 %, Trichoptera with 37 %, and Hymenoptera (ants) with 33 %. In terms of Alimentary Index, the most important items were organic debris (A.I.=40 %), Trichoptera larvae (A.I.=21.1 %) and adult Hymenoptera (ants) (A.I.=18.7 %).

Discussion

Centromochlus meridionalis was found in streams with structural and limnological features similar to those of the small Central Amazon streams (Espírito-Santo et al., 2009). However, the substrate of the studied streams was covered by a smaller amount of coarse litter than that present in Central Amazon streams (Mendonça et al., 2005; Carvalho, 2008). This apparent scarcity of coarse litter suggests that C. meridionalis present a strong selection for this type of substrate, which is used as shelter during the day (FGC, pers. obs.). Considering the distinctive mottled color pattern of C. meridionalis, we hypothesize that use of coarse litter packs as resting sites is part of its primary defense system, by providing cover and camouflage (Keenleyside, 1979), which could be an effective strategy against visually-oriented predators. Available data for other species of Centromochlinae indicate that most of them generally inhabit larger streams and rivers (e.g., Py-Daniel et al., 2007; our personal observations), where they use various types of substratum for resting, such as rubble, cracks and small cavities in boulders and logs (e.g., Barreto & Rocha, 2005; Sarmento-Soares & Buckup, 2005; Py-Daniel et al., 2007). Therefore, the use of litter packs for shelter and foraging by C. meridionalis appears to represent unusual features for a member of this subfamily, more common in fishes of the subfamily Auchenipterinae.

The behavioral observations recorded in aquaria showed that *C. meridionalis* has nocturnal habits similar to the majority of species of Auchenipteridae (Burgess, 1989; Akama & Ferraris, 2003). In captivity, however, the individuals of this species did not show the dusk activity peak known for other Centromochlinae (e.g., Ferraris, 2003; Sazima et al., 2005). During the day or under artificial light, the fish hid under leaves and logs at the bottom of the tank. Thus, the observed behaviors show that the species uses predominantly the lower layer of the water column both during resting and activity periods. This behavioral pattern contrasts to the typical behavior known for most species of Centromochlinae, which usually swim close to the surface during foraging, searching for insects that fall into the water (Ferraris, 2003). However, such behavior was not commonly observed for *C. meridionalis* in the aquarium. The relatively short and stout body of *C. meridionalis* is also indicative of its nektobenthic habits, a trait more commonly observed in species of Auchenipterinae.

Centromochlus meridionalis showed a distinctive feeding behavior, foraging on the bottom by biting at food items from a head-down position. This feeding tactic may be associated with the main types of food items ingested, which included both terrestrial and aquatic insects possibly dragged by the current close to the bottom. Siluriforms show a variety of predatory tactics depending on the environmental conditions and prey types (Pohlmann et al, 2004). Centromochlus meridionalis may be considered a generalized microcarnivore because its diet primarily includes items of animal origin, such as autochthonous and allochthonous insects, other terrestrial invertebrates, crustaceans and (occasionally) small fish. Other species of Centromochlinae, e.g., Glanidium ribeiroi (Ortêncio Filho et al., 2001) and G. bockmanni (Sarmento-Soares & Buckup, 2005) feed primarily on autochthonous and allochthonous insects (Ferraris, 2003). Adult G. ribeiroi may also feed on fish and plant matter (Ortêncio Filho, 2001), whereas G. albescens is considered an omnivorous species (Araújo et al., 2009). Nevertheless, there is no detailed information about the feeding behavior of those catfishes, which denotes the need for more behavioral and naturalistic studies focusing on species of Centromochlinae.

Although a considerable amount of organic debris was found in the stomach contents of *C. meridionalis*, this finding does not necessarily indicate that the species is a detritivore. Debris may have been accidentally ingested along with other food items collected at the stream bottom. Approximately 20 % of the substrate in the streams where the species was collected consisted of fine sediments similar to the debris found in the stomach contents. Fine sediment accumulates in slow-flowing stretches and near the margins of streams, where most of the leaf litter accumulations inhabited by *C. meridionalis* is found. Moreover, they ingested a considerable amount of trichopteran larvae of the family Leptoceridae,

which live buried in the mud (Mugnai et al., 2010), which may help explain the debris found in the stomach and also the unusual head-down posture employed during foraging.

The use of different layers of the water column by C. meridionalis during foraging, as well as the diversity of food items ingested, indicate a generalized microcarnivore diet. The food items ingested by C. meridionalis included the characid Moenkhausia phaeonota and small shrimps. Although those items are not particularly representative of the diet of C. meridionalis, they suggest the use of varied foraging tactics. Moenkhausia phaeonota is a diurnal fish that is abundant in the studied streams. It swims rapidly in small midwater schools (sensu Casatti et al., 2001; Padial et al., 2009; FGC, pers. obs.). This characid species rests close to the bottom and along the stream margins at night, where it remains stationary or moves very slowly (FGC, pers. obs.). Due to those differences in activity time and microhabitat use, C. meridionalis most likely captures small fish while the prey rests at night.

Finally, it is necessary to remark that the streams inhabited by *C. meridionalis* are located in an area that is strongly threatened by humaninduced disturbances, mainly resulting from the deforestation for agricultural use. In this sense, the restricted geographic distribution of *C. meridionalis* emphasizes the extinction risk to the species and reinforces the need to establish conservation measures to protect the headwaters of the Teles Pires River in the upper Tapajós River Basin, a region known for its high number of endemic fish species.

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