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ECOLOGY

Differences in ecomorphology and trophic niche segregation of two sympatric heptapterids (Teleostei: Siluriformes)

Thiago Deruza Garcia[1*], Bárbara Angélio Quirino[2], Leonardo Antunes Pessoa[2], Ana Lúcia Paz Cardozo[2] and Erivelto Goulart[2,3]

[1]Programa de Pós-graduação em Ciências Biológicas, Universidade Estadual de Londrina, Rodovia Celso Garcia Cid, PR-445, Km 380, 86057-970, Londrina, Paraná, Brazil. [2]Programa de Pós-graduação em Ecologia de Ambientes Aquáticos Continentais, Universidade Estadual de Maringá, Maringá, Paraná, Brazil. [3]Núcleo de Pesquisas em Limnologia, Ictiologia e Aquicultura, Universidade Estadual de Maringá, Maringá, Paraná, Brazil. *Author for correspondence. E-mail: thiago.deruza@hotmail.com

ABSTRACT. Morphological similarity, resource sharing, and differences in habitat use by species are factors that favor their coexistence. The objective of this study was to test possible differences in ecomorphology and diet composition of two Heptapterids (*Imparfinis mirini* and *Cetopsorhamdia iherengi*) to identify patterns related to resource use. Samplings were carried out in ten streams in Southern Brazil and 123 individuals were caught. A total of 21 ecomorphological indices were calculated for each individual and the volumetric and occurrence methods were used to quantify stomach contents. Both species presented significant differences in some ecomorphological traits, mainly related to foraging behavior. Even though both species were considered insectivorous, the consumed feeding resources differed between them. *Cetopsorhamdia iheringi* diet was dominated by Simuliidae and terrestrial Coleoptera larvae, while *Imparfinis mirini*, presented a diet dominated mainly by Trichoptera larvae, Ephemeroptera nymph, and *Annelida*. Although ecomorphological patterns cannot be used as an absolute factor to explain diet variations, they provide relevant information about how species share resources. These mechanisms allow us to obtain important subsidies for the conservation and management of freshwater ecosystems since they provide an effective understanding on the interactions that occur between the species.

Keywords: fish; coexistence; feeding specialization; morphological traits; resource partitioning.

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Introduction

High species diversity, trophic plasticity and the broad ecological niche allow the occupation of all trophic levels in aquatic environments by fish (Motta, Norton, & Luczkovich, 1995; Wootton, 1999; Abelha, Agostinho, & Goulart, 2001), which are the main organisms used in the ecomorphological studies (Moreira & Zuanon, 2002; Ramírez, Davenport, & Mojica, 2015). Ecomorphology can be applied to describe patterns along niche breadth, using body morphology to explain the way in which organisms are using resources in the environment (Fugi, Agostinho, & Hahn, 2001; Oliveira et al., 2010; Pagotto, Goulart, Oliveira, & Yamamura, 2011; Sampaio, Pagotto, & Goulart, 2013; Prado, Goulart, & Pagotto, 2016).

Sympatric species with similar morphology, such as phylogenetic related species, are expected to present strong competitive potential (Wootton, 1999). However, the differentiation in the ecological niche as distinct use of habitats and resources allows their stable coexistence in the environment (Peres-Neto, 2004; Herder & Freyhof, 2006; Van Zwol, Neff, & Wilson, 2012). These may also be related to adaptations in morphological traits, resulting in different ways of exploring the available resources (Labropoulou & Eleftheriou, 1997; Russo, Pulcini, O'Leary, Cataudella, & Mariani, 2008; Nandi & Saikia, 2015). Thus, considering that morphology and diet are strongly correlated, the ecomorphology is an important tool to understand the exploited niche space and trophic segregation among coexisting species (Prado et al., 2016; Pease, Mendoza-Carranza, & Winemiller, 2018; Baldasso, Wolff, Neves, & Delariva, 2019).

The Heptapteridae fishes are small to medium-sized species and represent one of the largest radiations of Neotropical catfish, being endemic to this region (Bockmann & Guazzelli, 2003). Such family stands out within the Siluriformes because they occupy a wide range of habitats, from small streams to large rivers, of clear or dark waters, cold and with medium - to fast - flowing waters (Ota, Deprá, Graça, & Pavanelli, 2018). Due to similar morphology, related habits are expected for heptapterids (Bockmann & Guazzelli, 2003). The closely related species *Imparfinis mirini* Haseman 1991 and *Cetopsorhamdia iheringi* Schubart, Gomes 1959 are abundant species in streams of the Pirapó River Basin (Upper Paraná River basin) (Ota et al., 2018). Both species have an elongated body with a terminal mouth, inhabit the bottom under substrates and their diets are composed of aquatic insects (Casatti, 2002; Bonato, Delariva, & Silva, 2012; Ota et al., 2018).

Although closely related species often have similar feeding habits, niche partitioning has been inferred as a valuable mechanism to facilitate their coexistence in the Neotropical region (Prado et al., 2016; Pease et al., 2018). This mechanism involves feeding on distinct items and consequently reduction of niche overlap to avoid competitive exclusion (Schoener, 1974; Ross, 1986; Portella, Lobón-Cerviá, Manna, Bergallo, & Mazzoni, 2017). In this context, considering that dissimilarities about morphology and habitat use can cause a small niche overlap (Hutchinson, 1959; Kokkoris, Jansen, Loreau, & Troumbis, 2002; Fowler, Lessard, & Sanders, 2014), we hypothesized that *Imparfinis mirini* and *Cetopsorhamdia iheringi* exhibit ecomorphological differences that are linked to differences in habitat use and food behavior. Thus, it is also expected a low niche overlap between these species, allowing the coexistence between them.

Material and methods

Study area

The study was performed in ten streams (1st to 3rd order), belonging to the Pirapó River Basin located in the Northern region of Paraná State, Southern Brazil (between 22°30’ and 23°30’S; 51°15’ and 52° 15’W). The Pirapó River Basin is one of the main tributaries at the left margin of the Paranapanema River, belonging to the Upper Paraná River Basin (Cunico, Ferreira, Agostinho, Beaumord, & Fernandes, 2012) (Figure 1). The dominant landscape in the basin is composed of agricultural activities and urban development, with the city of Maringá being the most important urban center in the region, with approximately 423,666 people (Cunico et al., 2012; Instituto Brasileiro de Geografia e Estatística [IBGE], 2019). The rainfall levels recorded annually are over 1,000 mm, with average annual temperatures between 16 and 20°C, with January being the hottest and most humid month and July the coldest and driest (Passos, 2007).

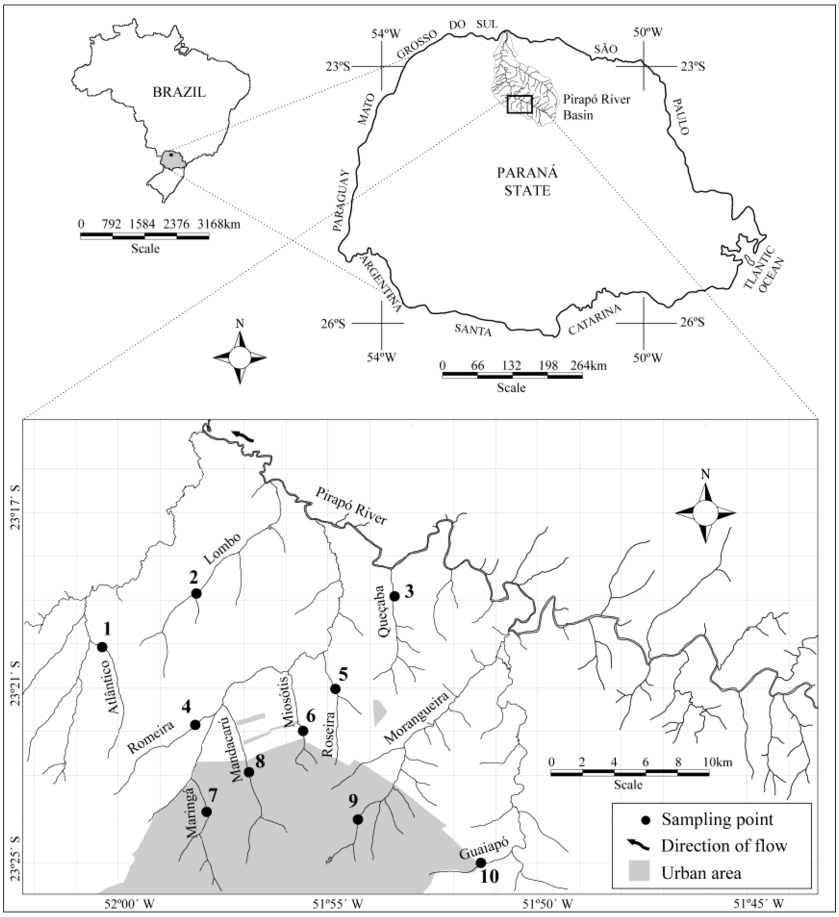

Figure 1. Streams of the Pirapó River Basin, in the municipalities of Maringá and Sarandi, Paraná, Brazil.

Sampling design

Sampling was carried out in April and May 2017 (dry season), using electrofishing in an area of approximately 30 meters. To maximize the sampling effort, each stream was blocked downstream with a 5 mm mesh net. The captured specimens were anesthetized with 100 mg L^{-1} Benzocaine solution until loss of equilibrium and remained immersed in the solution for at least 10 minutes after stopping opercular movements. Subsequently, they were fixed in 10% formalin and labeled for posterior analysis and measurements. Sampled fish were taken to the *Núcleo de Pesquisa em Limnologia, Ictiologia e Aquicultura* (Nupélia), of *Universidade Estadual de Maringá* (UEM), and specimens were deposited in the Ichthyological collection of Nupelia/UEM. Vouchers: *Cetopsorhamdia iheringi* (NUP 20045, 20057, 20097, 20101, 20109 and 20119) and *Imparfinis mirini* (NUP 20075, 20089, 20095, 20105, 20109 and 20123). The catches were taken under the *Instituto Chico Mendes de Conservação da Biodiversidade* (ICMBIO) License nº. 25560-1.

Morphological measures

Twenty-two morphological measurements were performed related to the body, fins, head, and mouth of each individual. These morphological measures are involved in habitat use and feeding (Gatz Jr., 1979; Watson & Balon, 1984; Wikramanayake, 1990; Oliveira et al., 2010). Morphological measurements were taken on the left side of each individual, using a digital caliper (accuracy of 0.01 mm). Measurements of fins and eye areas were obtained from the outline drawings of the structures, which were later digitized to calculate the internal area using the software ImageJ (Rueden et al., 2017). Only adult individuals were measured to eliminate the effects of ontogenetic change caused by allometric growth. Possible variations generated by differences in individual length were solved with the use of ecomorphological indices, since these indices minimize the effect of individual length, thus negating the likelihood of the analyzes being dominated by the individual's length (Winemiller, 1991; Prado et al., 2016). For each species, the mean of the linear morphological measure and area were calculated so that the ecomorphological indices were later obtained. After the measurements, 21 indices were calculated to determine the ecological attributes (Table 1) (Wikramanayake, 1990; Breda, Oliveira, & Goulart, 2005; Oliveira et al., 2010; Prado et al., 2016).

Table 1. Linear morphometric variables and areas used in calculating the ecomorphological proportions and their respective ecological meanings. Modified from Prado et al. (2016)

Indices	Formulas	Meanings
Body compression index	$BCI = MBH\ MBW^{-1}$	High values indicate lateral compression of fish, which is expected in fish that occupy low water velocity habitats (Gatz Jr., 1979; Watson & Balon, 1984).
Body depression index	$BDI= BMH\ MBH^{-1}$	Low values are associated with fish that exploit habitats with a rapid flow of water because the depressed body helps the fish to remain in the water column without having to swim (Watson & Balon, 1984).
Relative length of the caudal peduncle	$RLCP = CPL\ SL^{-1}$	Fish with long caudal peduncles are good swimmers. However, fish adapted to rapid water flows, but not necessarily nectonics, also present long caudal peduncles (Watson & Balon, 1984).
Relative height of the caudal peduncle	$RHCP = CPH\ MBH^{-1}$	Low values indicate high maneuverability (Winemiller, 1991; Willis, Winemiller, & Lopez-Fernandez, 2005).
Relative width of the caudal peduncle	$RWCP = CPW\ MBW^{-1}$	High values indicate continuous swimmers (Winemiller, 1991; Willis et al., 2005).
Relative length of the head	$RLH = HL\ SL^{-1}$	Higher relative head length values are found in fish that feed on larger prey, thus higher rates are expected for piscivorous species (Watson & Balon, 1984; Winemiller 1991; Willis et al., 2005).
Relative height of the head	$RHH = HH\ MBH^{-1}$	Higher relative values of head height are found in fish that feed on larger prey, thus higher rates are expected for piscivorous species (Winemiller, 1991; Willis et al., 2005).
Relative width of the head	$RWH = HW\ MBW^{-1}$	Higher relative values of head width are found in fish that feed on larger prey, thus higher rates are expected for piscivorous species (Winemiller, 1991; Willis et al., 2005).
Relative height of the mouth	$RHM = MH\ MBH^{-1}$	The relative height of the mouth allows inferring about the relative size of the prey (Gatz Jr., 1979).
Relative width of the mouth	$RWM = MW\ MBW^{-1}$	Index related to mouth size, suggesting relatively large prey for high indexes (Gatz Jr., 1979, Winemiller, 1991).
Vertical position of the eye	$VPE = EHRH\ HH^{-1}$	This index is associated with the foraging position of the species in the water column. High values indicate benthic fish (dorsally localized eyes), while low values indicate nectonic fish (lateral eyes) (Gatz Jr., 1979).
Relative area of the eye	$RAE = EA\ (SL)^{-2}$	This index is related to food detection and provides information on the visual acuity of the species. It can indicate the position of the species in the water column, as species that inhabit deeper areas have smaller eyes (Gatz Jr., 1979).

Indices	Formulas	Meanings
Relative area of the dorsal fin	$RADF = DFA\ (SL)^{-2}$	Larger relative areas present greater stabilization capacity in yaws (Breda et al., 2005)
Relative area of the caudal fin	$RACFi = CFA\ (SL)^{-2}$	Large caudal areas are important for acceleration (Oliveira et al., 2010).
Ratio-aspect of the caudal fin	$RACF = (CFH)^2\ CFA^{-1}$	Fishes with high aspect ratio caudal fins are more active and continuous swimmers in which there is a tendency to bifurcate of caudal and reduce their area (Gatz Jr., 1979)
Relative area of the anal fin	$RAAFi = AFA\ (SL)^{-2}$	Larger relative areas imply greater maneuverability and stabilization of movement (Breda et al., 2005)
Ratio-aspect of the anal fin	$RAAF = (AFL)^2\ AFA^{-1}$	Larger aspect-ratios imply a greater ability to perform faster progressive and retrograde movements (Breda et al., 2005)
Relative area of the pectoral fin	$RAPeFi = PFAr\ (SL)^{-2}$	Relatively larger pectoral fin areas are found in slow-swimming species, which use it for maneuverability (some Characidae) and may also be high among fishes that inhabit high current regions such as Siluriformes. Smaller areas are found in pelagic fish (Watson & Balon, 1984).
Reason-aspect of the pectoral fin	$RAPeF = (PFLe)^2\ PFAr^{-1}$	A high ratio indicates long and narrow pectoral fin, which is expected in fish that swim continuously and reach high speed, and consequently prefer pelagic regions (Oliveira et al., 2010).
Relative area of the pelvic fin	$RAPFi = PFA\ (SL)^{-2}$	Relatively larger pelvic fin areas are found in benthic fish and smaller areas in pelagic fish (Breda et al., 2005)
Ratio-aspect of the pelvic fin	$RAPF = (PFL)^2\ PFA^{-1}$	The largest values of pelvic fin aspect ratio are found in pelagic fish and are related to the ability to balance. The lowest values are associated with fish that prefer rocky habitats to support the body to the substrate (Gatz Jr., 1979).

Diet analysis

In the laboratory, stomachs were removed and only those with content were used in the diet analysis. To determine the diet composition, stomach contents were analyzed under a stereomicroscope and food resources were identified to the lowest practical taxonomic level using standard taxonomic keys. To quantify the diet, food items were quantified by the frequency of occurrence and by volumetric method, in which the volume of each food resource was measured using a millimeter Petri dish, obtained in mm^3 and later transformed into mL (Hellawell & Abel, 1971; Hyslop, 1980).

Data analysis

Differences in ecomorphology between species were evaluated by a multivariate permutation analysis of variance (PERMANOVA; Anderson, 2001). The pseudo-F statistic resulting from this analysis was tested by the Monte Carlo method using 999 randomizations. The ordering of the species according to their ecomorphological characteristics was evaluated by a Principal Component Analysis (PCA) performed using the correlation matrix of the ecomorphological proportions. This analysis indicates the grouping pattern of individuals with similar morphological traits. The axes were selected according to the Broken-Stick criterion, in which those with eigenvalues greater than the axes generated by the model were used for interpretation (McCune & Mefford, 1999).

Differences in diet between species were also evaluated with PERMANOVA. The pseudo-F statistic resulting from this analysis was tested by the Monte Carlo method using 999 randomizations. The ordering of the species according to their diet composition was visualized using a principal coordinate analysis (PCoA) based on a Bray-Curtis dissimilarity matrix (Bray & Curtis, 1957). Niche overlap between species was calculated using the Pianka index:

$$O_{jk} = \left(\sum_{i=1}^{n} |P_{ij} - P_{ik}|\right) / \sum_{i=1}^{n} P_{ij}^2 . \sum_{i=1}^{n} P_{ik}^2,$$

where Ojk = measure of Pianka food overlap between species j and species k; pij = proportion of food item i in the total of items used by species j; pik = proportion of food item i in the total of items used by species k, n = total number of food items.

P_{xi} and P_{yi} are the proportions (volume) of the food resource i used by species x and y, and n is the total number of feeding resources. The results of interspecific niche overlap were arbitrarily considered: high (> 0.6), intermediate (0.4-0.6) or low (<0.4) (Grossman, 1986). All analyses were performed in R (R Core Team,

2017) using the Vegan package 1.17 (Oksanen et al., 2018), except for the niche overlap analysis which was performed in EcoSim version 6.18 (Gotelli & Entsminger, 2001). Graphs were built in the software Statistica 12.5 (Statsoft Inc., 2011).

Results

A total of 123 individuals were measured, but only 58 stomachs of *Imparfinis mirini* and 29 stomachs of *Cetopsorhamdia iheringi* contained food and were analyzed.

The PERMANOVA showed significant difference in ecomorphological attributes between the species (pseudo-$F_{(1.121)} = 15.18$; $p > 0.001$). The PCA revealed the significance of the first two axes according to the Broken-Stick criterion. Together, axes 1 and 2 (Table 2) explained 38.68% and were used to characterize the morphological diversity between species (Figure 2). When analyzing the first axis of PCA, it is observed that *C. iheringi* is located on the negative side presenting lower values for relative area of the pectoral fin (RAPeFi), relative area of the anal fin (RAAFi), relative area of the pelvic fin (RAPFi), reason-aspect of the pectoral fin, relative length of the head (RLH) and relative length of the caudal peduncle (RLCP). *Imparfinis mirini* is located on the positive side presenting higher values for relative height of the mouth (RHM), relative width of the mouth (RWM) and ratio-aspect of the anal fin (RAAF).

Table 2. Eigenvalues and percentage of variation for each axis as well as the index's loadings. The indexes that were more important for interpretation are marked in bold.

Ecomorphological proportions	Abbreviations	Axis 1	Axis 2
Body compression index	BCI	-0.125	0.326
Body depression index	BDI	-0.257	-0.269
Relative length of the caudal peduncle	RLCP	-0.667	-0.022
Relative height of the caudal peduncle	RHCP	-0.266	-0.191
Relative width of the caudal peduncle	RWCP	-0.377	-0.013
Relative length of the head	RLH	-0.721	0.077
Relative height of the head	RHH	0.304	0.105
Relative width of the head	RWH	0.059	0.117
Relative height of the mouth	RHM	0.740	0.351
Relative width of the mouth	RWM	0.735	0.306
Vertical position of the eye	VPE	-0.304	-0.303
Relative area of the eye	RAE	0.541	0.341
Relative area of the dorsal fin	RADF	0.083	0.538
Relative area of the caudal fin	RACFi	-0.373	0.527
Relative area of the anal fin	RAAFi	-0.355	0.762
Relative area of the pectoral fin	RAPeFi	-0.280	0.843
Relative area of the pelvic fin	RAPFi	0.035	0.707
Ratio-aspect of the caudal fin	RACF	-0.670	-0.324
Ratio-aspect of the anal fin	RAAF	0.718	-0.473
Reason-aspect of the pectoral fin	RAPeF	0.105	-0.763
Ratio-aspect of the pelvic fin	RAPF	-0.169	0.048
Eigenvalues		4.178	3.945
Eigenvalues predicted by broken-stick		3.645	2.645
Proportion of the variances (%)		0.199	0.188

When evaluating the dietary composition of *C. iheringi* and *I. mirini*, it is observed that both species consumed predominantly aquatic food resources and Trichoptera larvae, Ephemeroptera nymph and Chironomidae larvae occurred in most of the analyzed stomachs (Figure 3A and B). However, some preferences were observed. Out of 11 food resources recorded in the diet of *C. iheringi*, Simuliidae larvae was highly consumed (22.6% of volume and 27.6% of occurrence) and it was an exclusive food resource to this species. Although the allochthonous feeding resources were less representative (Figure 3C and D), terrestrial Coleoptera larvae were the second most consumed by *C. iheringi* (21.84% of volume and 27.6% of occurrence), followed by the Trichoptera larvae (18, 2% of volume and 41.4% of occurrence). Out of 17 feeding resources consumed by of *I. mirini*, Trichoptera larvae dominated the diet, composing 54.9% of total volume and occurring in 76.8% of the analyzed stomachs, followed *by* Ephemeroptera nymph (9.9 % of the volume and 30.3% of occurrence), and *Annelida* (8.6% of the volume and 5.2% of occurrence). This latter and six other resources were consumed exclusively by *I. mirini* (Figure 3A and 3B).

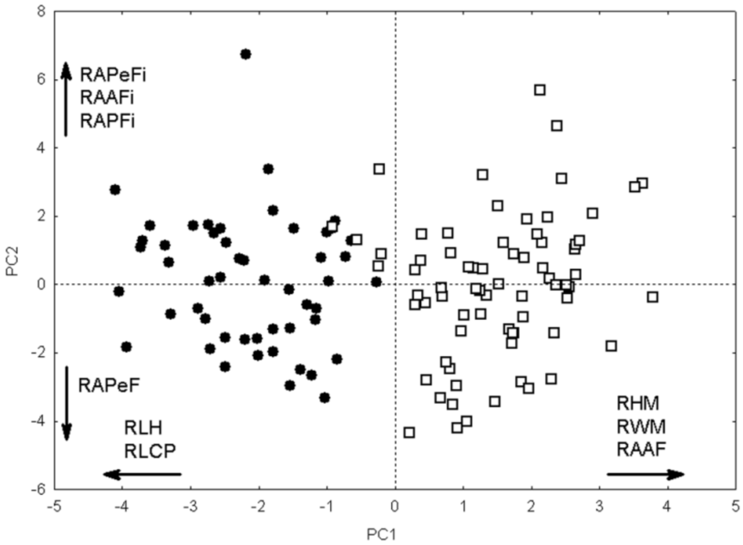

Figure 2. Ordination diagram of the first two PCA axes for the 22 ecomorphological attributes of Heptapteridae: (●) *Cetopsorhamdia iheringi* and (□) *Imparfinis mirini*. RHM = Relative height of the mouth, RWM = Relative width of the mouth, RAAF = Ratio-aspect of the anal fin, RLH = Relative length of the head, RACF = Ratio-aspect of the caudal fin, RAPeF = Reason-aspect of the pectoral fin, RAAFi = Relative area of the anal fin, RAPFi = Relative area of the pelvic fin and, RAPeFi = Relative area of the pectoral fin. Total variation explained by axes 1 and 2 = 37%.

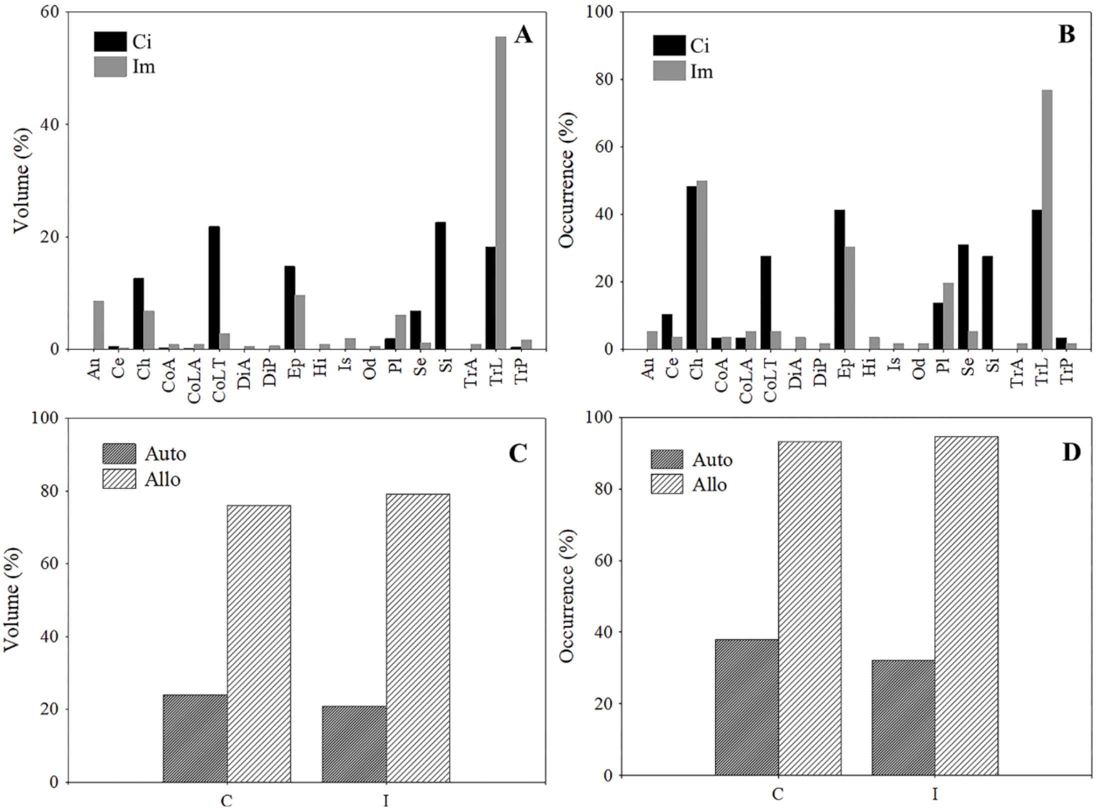

Figure 3. Composition and contribution of autochthonous (Auto) and allochthonous (Allo) items in the diet of *Cetopsorhamdia iheringi* (Ci) and *Imparfinis mirini* (Im), captured in April and May 2017, in streams of the Pirapó River Basin. A and C = % volume, B and D= % occurrence. An= Annelida; Ce= Ceratopogonidae; Ch= Chironomidae larvae; CoA= adult Coleoptera; CoLA= aquatic Coleoptera larvae; CoLT= terrestrial Coleoptera larvae; DiA= adult Diptera; DiP= Diptera pupae; Ep= Ephemeroptera; Hi= Hirudinea; Is= Isoptera; Od= Odonata; Pl= plants; Se= sediment; Si= Simuliidae; TrA= adult Trichoptera; TrL= Trichoptera larvae; TrP= Trichoptera pupae;Si= Simuliidae; CoLT= terrestrial Coleoptera larvae; TrL= Trichoptera larvae; Ep= Ephemeroptera; Ch= Chironomidae larvae; Se= sediment; Pl= plants; Ce= Ceratopogonidae; TrP= Trichoptera pupae; CoA= adult Coleoptera; CoLA= aquatic Coleoptera larvae; An= Annelida; Is= Isoptera; TrA= adult Trichoptera; Hi= Hirudinea; DiP= Diptera pupae; DiA= adult Diptera; Od= Odonata.

PERMANOVA also showed a significant difference in diet composition between the two species (pseudo-$F_{(1.83)} = 4.31$; $p < 0.05$) (Figure 4) and ordination analysis evidenced separation between the species, mainly on axis 1: *C. iheringi* located on the negative values and *I. mirini* located in the positive values. Together, axes 1 and 2 explained 30.2% of the variation (Figure 4). The niche overlap was considered intermediate (0.54).

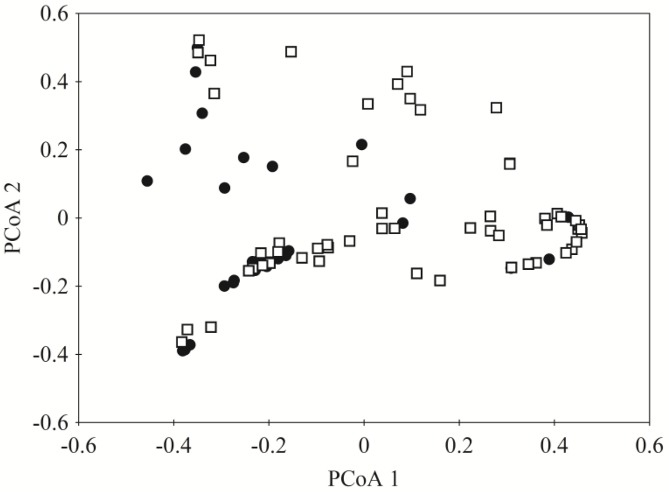

Figure 4. Ordination diagram of the first two PCoA axes (32.7%) for the diet of two species of Heptapteridae: (●) *Cetopsorhamdia iheringi* and (□) *Imparfinis mirini*. Axis 1 explained 30.3% of data variation.

Discussion

The results showed that despite *Imparfinis mirini* and *Cetopsorhamdia iheringi* are phylogenetically close and occupy the same stratum in the environment and are considered insectivorous species, they present differences in morphology and the use of food resources. Also, there is a low overlap, indicating low competition for food resources. Ecomorphological differences, mainly related to foraging, swimming, and habitat occupation favor the coexistence between these species. Correlations between diet and ecomorphology in fish are constantly observed (Neves, Delariva, & Wolff, 2015; Pease et al., 2018; Baldasso et al., 2019), which supports the idea that morphology acts as one of the mechanisms responsible for the variation of diet. In fact, sympatric species tend to differ in feeding preferences, segregating resources to avoid competitive exclusion (Hardin, 1960; Schoener, 1974).

In an attempt to explain the preference of fish for a particular resource, some factors may be directly related, such as abundance and nutritional quality, as well as their morphology and behavior. However, when we observe the diet of benthic fish, these factors in the predator-prey relationship are not clear, since the heterogeneity and complexity of habitat in the benthic region can act as a physical barrier during the foraging of benthic, preventing many preys from being consumed by fish (Hill & Grossman, 1987; Gillette, 2012, Guglielmetti, Silva, Higuti, & Fugi, 2019).' Several studies have shown that fish belonging to benthic invertivorous guild have a very specific diet and that the use of a determinate resource is mainly attributed to trophic morphology and predation foraging (Rolla, Esteves, & Silva, 2009; Brejão, Gerhard, & Zuanon, 2013; Neves et al., 2015). In our study, morphological characteristics such as head, mouth, and fins sizes allowed both species to feed on benthic organisms, while the terminal mouth and the laterally compressed body possibly allowed the expansion of the consumption of other resources available in the environment (Pease et al., 2018), as the allochthonous items present in the diet of both species such as terrestrial Coleoptera larvae, plants, adult Coleoptera, Isoptera, adult Trichoptera, and adult Diptera.

Despite the similarities, our results showed differences in morphology between the species that are probably related to different feeding preferences. The indices that influenced *Cetopsorhamdia iheringi* dispersal in ordination are associated with fish that prefer benthic and high water flow regions, presenting slow swimming and movement stability (Watson & Balon, 1984; Breda et al., 2005). In addition, the smaller head length indicates that *C. iheringi* feeds on smaller prey (Pouilly, Lino, Bretenoux, & Rosales, 2003; Oliveira et al., 2010), as is the case of Simuliidae larvae that attach to the substrates or plant surfaces (Kiel, 2001) and was found in abundance and exclusivity in their diet. However, the indices that influenced the dispersion of *Imparfinis mirini* in the ordination are associated with fish that have faster progressive and retrograde movements (Breda et al., 2005) and consequently act more efficiently in the exploration of complex structurally habitats (Prado et al., 2016). In addition, *I. mirini* presented a greater length and width of the mouth, suggesting that species with these characteristics feed on relatively large prey (Gatz Jr., 1979; Winemiller, 1991), such as Trichoptera larvae consumption, item of great abundance in the diet this species. This high abundance in the diet can be explained because this aquatic larvae has low mobility, which makes its escape capacity lower and the larvae became more vulnerable to predation (Tófoli, Alves, Higuti, Cunico, & Hahn, 2013). In addition, since it is a relatively large invertebrate, it makes the larvae more visible to the predator (Tófoli et al., 2013).

The niche overlap value indicates a low potential for competitive interaction (Zaret & Rand, 1971). Although we have not measured food availability in this study, several studies point out that diet divergences may reduce competition when resources are limited (Hutchinson, 1957; Colloca, Carpentieri, Balestri, & Ardizzone, 2010; Guo et al., 2014; Carniatto, Fugi, & Thomaz, 2017). Besides, it should be considered that the variety and abundance of resources consumed by the species must be mediated by their morphological characteristics (Prado et al., 2016), where one species is able to explore resources that other is not (Casatti, 2002; Freitas, Montag, & Barthem, 2017; Pagotto et al., 2011). In this case, a strong competitive potential would not be expected between *C. iheringi* and *I. mirini*, since both species occupy different morphological niches.

Another important ecological factor that can promote coexistence between species is the preference for some resources (Novakowski, Fugi, & Hahn, 2004; Tófoli et al., 2013; Walker, Kluender, Inebnit, & Adams, 2013; Bison et al., 2015). The differential use of resources, even partially, is an essential component of the trophic interactions within a community (Schoener, 1974; Portella et al., 2017; Gračan, Zavodnik, Krstinić, Dragičević, & Lazar, 2016). Trophic varieties observed in species analyzed here highlight interspecific differences in the use of food items. This specialization in different resources probably allows segregation of the niche (Prado et al., 2016; Portella et al., 2017). Trichoptera was the most abundant item in *I. mirini* diet and the third most abundant in *C. iheringi* diet, which reflects the great abundance of these invertebrates in the Pirapó basin streams (Guglielmetti et al., 2019) and makes them easily predated mainly by *I. mirini*, which forages the substrate since it uses the head barbs to find and capture the prey (Casatti & Castro, 2006). This feature may also explain the large number of unique items found in *I. mirini* diet. On the other hand, the high abundance of terrestrial Coleoptera suggests that *C. iheringi* uses both benthic and pelagic region, consuming drifting resources (Guglielmetti et al., 2019). Thus, the prey origin and the different use of fine-scale habitat contribute to the trophic segregation between these species (Barros, Zuanon, & Deus, 2017; Richardson, Lyons, Roby, Cushing, & Lerczak, 2018).

Conclusion

In short, our results showed ecomorphological differences between the sympatric species, and that such variations may be determinant in diet differences. Although both species are insectivorous, they present particular preferences, resulting in the low trophic overlap. The ecomorphological and diet relationship of species enable us to understand how they perform their ecological niches (Peres-Neto, 1999; Severo-Neto, Teresa, & Froehlich, 2015). Thus, these factors seem to be essential to coexistence between these species in these environments. Studying the ecology of these species provides subsidies for the management and conservation of these ecosystems that are under constant anthropogenic effects.

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References

Abelha, M. C. F., Agostinho, A. A., & Goulart, E. (2001). Plasticidade trófica em peixes de água doce. *Acta Scientiarium Biological Sciences*, *23*, 425-434. doi: 10.4025/actascibiolsci.v23i0.2696

Anderson, M. J. (2001). A new method for non-parametric multivariate analysis of variance. *Austral Ecology*, *26*(1), 32-46. doi: 10.1111/j.1442-9993.2001.01070.pp.x

Baldasso, M. C., Wolff, L. L., Neves, M. P., & Delariva, R. L. (2019). Ecomorphological variations and food supply drive trophic relationships in the fish fauna of a pristine neotropical stream. *Environmental Biology of Fishes*, *102*, 783-800. doi: 10.1007/s10641-019-00871-w

Barros, G., Zuanon, J., & Deus, C. (2017). Effects of species co-occurrence on the trophic-niche breadth of characids in Amazon forest streams. *Journal of Fish Biology*, *90*(1), 326-340. doi: 10.1111/jfb.13183

Bison, M., Ibanez, S., Redjadj, C., Boyer, F., Coissac, E., Miquel, C., ... Loison, A. (2015). Upscaling the niche variation hypothesis from the intra- to the inter-specific level. *Oecologia*, *179*(3), 835-842. doi: 10.1007/s00442-015-3390-7

Bockmann, F. A., & Guazzelli, G. M. (2003). Family Heptapteridae (Heptapterids). In R. E. Reis, S. O. Kullander & C. J. Ferraris JR (Eds.), *Checklist of the freshwater fishes of south and central America* (p. 406-431). Porto Alegre: Edipucrs.

Bonato, K. O., Delariva, R. L., & Silva, J. C. (2012). Diet and trophic guilds of fish assemblages in two streams with different anthropic impacts in the northwest of Paraná, Brazil. *Zoologia*, *29*(1), 27-38. doi: 10.1590/S1984-46702012000100004

Bray, J. R., & Curtis, J. T. (1957). An ordination of the upland forest communities of southern Wisconsin. *Ecological Monographs*, *27*(4), 325-349. doi: 10.2307/1942268

Breda, L., Oliveira, E. F., & Goulart, E. (2005). Ecomorfologia de locomoção de peixes com enfoque para espécies neotropicais. *Acta Scientiarum Biological Sciences*, *27*(4), 371-381. doi: 10.4025/actascibiolsci.v27i4.1271

Brejão, G. L., Gerhard, P., & Zuanon, J. (2013). Functional trophic composition of the ichthyofauna of forest streams in eastern Brazilian Amazon. *Neotropical Ichthyology*, *11*(2), 361-373. doi: 10.1590/S1679-62252013005000006

Carniatto, N., Fugi, R., & Thomaz, S. M. (2017). Highly segregated trophic niche of two congeneric fish species in Neotropical floodplain lakes. *Journal of Fish Biology*, *90*(3), 1118-1125. doi: 10.1111/jfb.13236

Casatti, L. (2002). Alimentação dos peixes em um riacho do Parque Estadual Morro do Diabo, bacia do Alto Rio Paraná, sudeste do Brasil. *Biota Neotropica*, *2*(2), 1-14. doi: 10.1590/S1676-06032002000200012

Casatti, L., & Castro, R. M. C. (2006). Testing the ecomorphological hypothesis in a headwater riffles fish assemblage of the rio São Francisco, southeastern Brazil. *Neotropical Ichthyology*, *4*(2), 203-214. doi: 10.1590/S1679-62252006000200006

Colloca, F., Carpentieri, P., Balestri, E., & Ardizzone, G. (2010). Food resource partitioning in a Mediterranean demersal fish assemblage: the effect of body size and niche width. *Marine Biology*, *157*, 565-574. doi: 10.1007/s00227-009-1342-7

Cunico, A. M., Ferreira, E. A., Agostinho, A. A., Beaumord, A. C., & Fernandes, R. (2012). The effects of local and regional environmental factors on the structure of fish assemblages in the Pirapó Basin, Southern Brazil. *Landscape and Urban Planning*, *105*(3), 336-344. doi: 10.1016/j.landurbplan.2012.01.002

Fowler, D., Lessard, J. P., & Sanders, N. J. (2014). Niche filtering rather than partitioning shapes the structure of temperate forest ant communities. *Journal of Animal Ecology*, *83*(4), 943-952. doi: 10.1111/1365-2656.12188

Freitas, T. M. S., Montag, L. F. A., & Barthem, R. B. (2017). Distribution, feeding and ecomorphology of four species of Auchenipteridae (Teleostei: Siluriformes) in Eastern Amazonia, Brazil. *Iheringia - Série Zoologia*, *107*, e2017008. doi: 10.1590/1678-4766e2017008

Fugi, R., Agostinho, A. A., & Hahn, N. S. (2001). Trophic morphology of five benthic-feeding fish species of a tropical floodplain. *Revista Brasileira de Biologia*, *61*(1), 27-33. doi: 10.1590/S0034-71082001000100005

Gatz Jr., A. J. (1979). Community organization in fishes as indicated by morphological features. *Ecology*, *60*(4),711-718. doi: 10.2307/1936608

Gillette, D. P. (2012). Effects of terrestrial invertebrate reduction on three stream fishes in experimental mesocosms. *Freshwater Science*, *31*(3), 835-847. doi: 10.1899/11-101.1

Gotelli, N. J., & Entsminger, G. L. (2001). *EcoSim: Null models software for ecology. Version 7.0.* Jericho, VT: Acquired Intelligence & Kesey-Bear.

Gračan, R., Zavodnik, D., Krstinić, P., Dragičević, B, & Lazar, B. (2016). Feeding ecology and trophic segregation of two sympatric mesopredatory sharks in the heavily exploited coastal ecosystem of the Adriatic Sea. *Journal of Fish Biology*, *90*(1), 167-184. doi: 10.1111/jfb.13158

Grossman, G. D. (1986). Food resources partitioning in a rocky intertidal fish assemblage. *Journal of Zoology*, *1*(2), 317-355. doi: 10.1111/j.1096-3642.1986.tb00642.x

Guglielmetti, R., Silva, M. R., Higuti, J., & Fugi, R. (2019). Diet of benthivorous fish and prey availability in streams of the Pirapó River basin-PR. *Acta Limnologica Brasiliensia*, *31*, e7. doi: 10.1590/s2179-975x4518

Guo, Z., Liu, J., Lek, S., Li, Z., Zhu, F., Tang, J., & Cucherousset, J. (2014). Trophic niche differences between two congeneric goby species: evidence for ontogenetic diet shift and habitat use. *Aquatic Biology*, *20*(1), 23-33. doi: 10.3354/ab00530

Hardin, G. (1960). The competitive exclusion principle. *Science*, *131*(3409), 1292-1297. doi: 10.1126/science.131.3409.1292

Hellawell, J. M., & Abel, R. (1971). A rapid volumetric method for the analysis of the food of fishes. *Journal of Fish Biology*, *3*(1), 29-37. doi: 10.1111/j.1095-8649.1971.tb05903.x

Herder, F., & Freyhof, J. (2006). Resource partitioning in a tropical stream fish assemblage. *Journal of Fish Biology*, *69*(2), 571-589. doi: 10.1111/j.1095-8649.2006.01126.x

Hill, J., & Grossman, G. D. (1987). Effects of subcutaneous marking on stream fishes. *Copeia*, *1987*(2), 492-495. doi: 10.2307/1445790

Hutchinson, G. E. (1957). Concluding remarks. *Cold Spring Harbor Symposia on Quantitative Biology*, *22*, 415-427. doi: 10.1101/sqb.1957.022.01.039

Hutchinson, G. E. (1959). Homage to Santa Rosalia or why are there so many kinds of animals? *The American Naturalist*, *93*(870), 145-159. doi: 10.1086/282070

Hyslop, E. J. (1980). Stomach contents analysis-a review of methods and their applications. *Journal of Fish Biology, 17*(4), 411-429. doi: 10.1111/j.1095-8649.1980.tb02775.x

Instituto Brasileiro de Geografia e Estatística [IBGE]. (2019). *Censo populacional. Paraná, 2019*. Retrieved from https://www.ibge.gov.br/cidades-e-estados/pr/maringa.html?

Kiel, E. (2001). Behavioural response of blackfly larvae (Simuliidae, Diptera) to different current velocities. *Limnologica-Ecology and Management of Inland Waters*, *31*(3), 179-183. doi: 10.1016/S0075-9511(01)80018-9

Kokkoris, G. D., Jansen, V. A. A., Loreau, M., & Troumbis, A. Y. (2002). Variability in interaction strength and implications for biodiversity. *Journal of Animal Ecology*, *71*(2), 362-371. doi: 10.1046/j.1365-2656.2002.00604.x

Labropoulou, M., & Eleftheriou, A. (1997). The foraging ecology of two pairs of congeneric demersal fish species: importance of morphological characteristics in prey selection. *Journal of Fish Biology*, *50*(2), 324-340. doi: 10.1111/j.1095-8649.1997.tb01361.x

McCune, B., & Mefford, M. J. (1999). *PC-ORD: multivariate analysis of ecological data, version 4.* Gleneden Beach, OR: MjM Software Design.

Moreira, S. S., & Zuanon, J. (2002). Dieta de *Retroculus lapidifer* (Perciformes: Cichlidae), um peixe reofílico do Rio Araguaia, estado do Tocantins, Brasil. *Acta Amazonica*, *32*(4), 691-705. doi: 1809-43922002324705

Motta, P. J., Norton, S. F., & Luczkovich, J. J. (1995). Perspectives on the ecomorphology of bony fishes. *Environmental Biology of Fishes*, *44*, 11-20. doi: 10.1007/BF00005904

Nandi, S., & Saikia, S. K. (2015). Size-selective feeding on phytoplankton by two morpho-groups of the small freshwater fish *Amblypharyngodon mola*. *Journal of Fish Biology*, *87*(2), 215-230. doi: 10.1111/jfb.12706

Neves, M. P., Delariva, R. L., & Wolff, L. L. (2015). Diet and ecomorphological relationships of an endemic, species-poor fish assemblage in a stream in the Iguaçu National Park. *Neotropical Ichthyology*, *13*(1), 245-254. doi: 10.1590/1982-0224-20140124

Novakowski, G. C., Fugi, R., & Hahn, N. S. (2004). Diet and dental development of three species of *Roeboides* (Characiformes: Characidae). *Neotropical Ichthyology, 2*(3), 157-162. doi: 10.1590/S1679-62252004000300008

Oksanen, J., Blanchet, F. G., Friendly, M., Kindt, R., Legendre, P., McGlinn, D, … Wagner, H. (2018). Package 'vegan': community ecology package. R package version 2, 4-6.

Oliveira, E. F., Goulart, E., Breda, L., Vera, C. V. M., Paiva, L. R. S., & Vismara, M. R. (2010). Ecomorphological patterns of the fish assemblage in a tropical floodplain: effects of trophic, spatial and phylogenetic structures. *Neotropical Ichthyology*, *8*(3), 569-586. doi: 10.1590/S1679-62252010000300002

Ota, R. R., Deprá, G. C., Graça, W. J., & Pavanelli, C. S. (2018) Peixes da planície de inundação do alto rio Paraná e áreas adjacentes: revised, annotated and updated. *Neotropical Ichthyology*, *16*(2), e170094. doi: 10.1590/1982-0224-20170094

Pagotto, J. P. A., Goulart, E., Oliveira, E. F., & Yamamura, C. B. (2011). Trophic ecomorphology of Siluriformes (Pisces, Osteichthyes) from a tropical stream. *Brazilian Journal of Biology*, *71*(2), 469-479. doi: 10.1590/S1519-69842011000300017

Passos, M. M. (2007). *A raia divisória: eco-história da raia divisória*. Maringá, PR: Eduem.

Pease, A. A., Mendoza-Carranza, M., & Winemiller, K. O. (2018). Feeding ecology and ecomorphology of cichlid assemblages in a large Mesoamerican river delta. *Environmental Biology of Fishes*, *101*, 867-879. doi: 10.1007/s10641-018-0743-1

Peres-Neto, P. R. (1999). Alguns métodos e estudos em ecomorfologia de peixes de riachos. In E. P. Caramaschi, R. Mazzoni & P. R. Peres-Neto (Eds.), *Ecologia de peixes de riachos: série Oecologia Brasiliensis* (p. 157-182). Rio de Janeiro: PPGE-UFRJ.

Peres-Neto, P. R. (2004). Patterns in the co-occurrence of fish species in streams: the role of site suitability, morphology and phylogeny versus species interactions. *Oecologia*, *140*(2), 352-360. doi: 10.1007/s00442-004-1578-3

Portella, T., Lobón-Cerviá, J., Manna, L. R., Bergallo, H. G., & Mazzoni, R. (2017). Eco-morphological attributes and feeding habits in coexisting characins. *Journal of Fish Biology*, *90*(1), 129-146. doi: 10.1111/jfb.13162

Pouilly, M., Lino, F., Bretenoux, J.-G., & Rosales, C. (2003). Dietary-morphological relationships in a fish assemblage of the Bolivian Amazonian floodplain. *Journal of Fish Biology*, *62*(5), 1137-1158. doi: 10.1046/j.1095-8649.2003.00108.x

Prado, A. V. R., Goulart, E., & Pagotto, J. P. A. (2016). Ecomorphology and use of food resources: inter- and intraspecific relationships of fish fauna associated with macrophyte stands. *Neotropical Ichthyology*, *14*(4), e-150140. doi: 10.1590/1982-0224-20150140

R Core Team. (2017). *R: A language and environment for statistical computing [Computer software manual - Internet]*. Vienna, AU: R Foundation for statistical computing.

Ramírez, F., Davenport, T. L., & Mojica, J. I. (2015). Dietary-morphological relationships of nineteen fish species from an Amazonian terra firme black water stream in Colombia. *Limnologica-Ecology and Management of Inland Waters*, *52*, 89-102. doi: 10.1016/j.limno.2015.04.002

Richardson, A. G. P., Lyons, D. E., Roby, D. D., Cushing, D. A., & Lerczak, J. A. (2018). Three-dimensional foraging habitat use and niche partitioning in two sympatric seabird species, *Phalacrocorax auritus* and *P. penicillatus*. *Marine Ecology Progress Series*, *586*, 251-264. doi: 10.3354/meps12407

Rolla, A. P. P. R., Esteves, K. E., & Silva, A. O. A. (2009). Feeding ecology of a stream fish assemblage in an Atlantic Forest remnant (Serra do Japi, SP, Brazil). *Neotropical Ichthyology*, *7*(1), 65-76. doi: 10.1590/S1679-62252009000100009

Ross, S. T. (1986). Resource partitioning in fish assemblages: a review of field studies. *Copeia*, *1986*(2), 352-388. doi: 10.2307/1444996

Rueden, C. T., Schindelin, J., Hiner, M. C., DeZonia, B. E., Walter, A. E., Arena, E. T., & Eliceiri, K. W. (2017). ImageJ2: ImageJ for the next generation of scientific image data. *BMC Bioinformatics*, *18*(1), 529. doi: 10.1186/s12859-017-1934-z

Russo, T., Pulcini, D., O'Leary, Á., Cataudella, S., & Mariani, S. (2008). Relationship between body shape and trophic niche segregation in two closely related sympatric fishes. *Journal of Fish Biology*, *73*(4), 809-828. doi: 10.1111/j.1095-8649.2008.01964.x

Sampaio, A. L. A., Pagotto, J. P. A., & Goulart, E. (2013). Relationships between morphology, diet and spatial distribution: testing the effects of intra and interspecific morphological variations on the patterns of resource use in two Neotropical Cichlids. *Neotropical Ichthyology*, *11*(2), 351-360. doi: 10.1590/S1679-62252013005000001

Severo-Neto, F., Teresa, F. B., & Froehlich, O. (2015). Ecomorphology and diet reflect the spatial segregation between two Siluriformes species inhabiting a stream of the Bodoquena Plateau, in Central Brazil. *Iheringia, Série Zoologia*, *105*(1), 62-68. doi: 10.1590/1678-4766201510516268

Schoener, T. W. (1974). Resource partitioning in ecological communities. *Science*, *185*(4145), 27-39. doi: 10.1126/science.185.4145.27

Statsoft Inc. (2011). *Statistica: data analysis software system, version 12.5.*

Tófoli, R. M., Alves, G. H. Z., Higuti, J., Cunico, A. M., & Hahn, N. S. (2013). Diet and feeding selectivity of a benthivorous fish in streams: responses to the effects of urbanization. *Journal of Fish Biology*, *83*(1), 39-51. doi: 10.1111/jfb.12145

Van Zwol, J. A., Neff, B. D., & Wilson, C. C. (2012). The effect of nonnative salmonids on social dominance and growth of juvenile Atlantic salmon. *Transactions of the American Fisheries Society*, *141*(4), 907-918. doi: 10.1080/00028487.2012.675899

Walker, R. H., Kluender, E. R., Inebnit, T. E., & Adams, S. R. (2013). Differences in diet and feeding ecology of similar-sized spotted (*Lepisosteus oculatus*) and shortnose (*Lepisosteus platostomus*) gars during flooding of a south-eastern US river. *Ecology of Freshwater Fish*, *22*(4), 617-625. doi: 10.1111/eff.12066

Watson, D. J., & Balon, E. K. (1984). Ecomorphological analysis of fish taxocenes in rainforest streams of northern Borneo. *Journal of Fish Biology*, *25*(3), 371-384. doi: 10.1111/j.1095-8649.1984.tb04885.x

Willis, S. C., Winemiller, K. O., & Lopez-Fernandez, H. (2005). Habitat structural complexity and morphological diversity of fish assemblages in a Neotropical floodplain river. *Oecologia*, *142*(2), 284-295. doi: 10.1007/s00442-004-1723-z

Winemiller, K. O. (1991). Ecomorphological diversification in lowland freshwater fish assemblages from five biotic regions. *Ecological Monographs*, *61*(4), 343-365. doi: 10.2307/2937046

Wikramanayake, E. D. (1990). Ecomorphology and biogeography of a tropical stream fish assemblage: evolution of assemblage structure. *Ecology*, *71*(5), 1756-1764. doi: 10.2307/1937583

Wootton, R. J. (1999). *Ecology of teleost fishes.* London, UK: Chapman & Hall.

Zaret, T. M., & Rand, A. S. (1971). Competition in tropical stream fishes: support for the competitive exclusion principle. *Ecology*, *52*(2), 336-342. doi: 10.2307/1934593