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RESEARCH ARTICLE

Length-weight relationship, sex ratio, and diet of three fish species (Actinopterygii: Teleostei) in streams of the Pomba River basin, Paraíba do Sul river drainage, Southeastern Brazil

Igor David Costa[1,2], Natalia Neto Santos Nunes[3], Ilana Rosental Zalmon[4]

[1]*Departamento de Ciências Exatas, Biológicas e da Terra, Universidade Federal Fluminense. Avenida João Jasbick, 28470-000 Santo Antônio de Pádua, RJ, Brazil.*
[2]*Programa de Pós-graduação Profissional em Gestão e Regulação de Recursos Hídricos, Universidade Federal de Rondônia. Rua Rio Amazonas 351, 76900-726 Ji-Paraná, RO, Brazil.*
[3]*Programa de Pós-Graduação em Ciências Ambientais, Universidade Federal de Rondônia. Avenida Norte Sul 7300, 76940-000 Rolim de Moura, RO, Brazil.*
[4]*Laboratório de Ciências Ambientais, Universidade Estadual do Norte Fluminense. Avenida Alberto Lamego 2000, 28013-602 Campos dos Goytacazes, RJ, Brazil.*
Corresponding author: Igor David da Costa (igorbiologia@yahoo.com.br)

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ABSTRACT. This study aimed to estimate the parameters of length-weight relationship, sex ratio, and diet of three freshwater fish species that inhabit streams in the Pomba River basin (Rio de Janeiro, Brazil). Fishes were sampled with seine nets (2.0 × 1.20 m, 2 mm mesh size) and dip nets (0.46 × 0.33 m, 2 mm mesh size) from October to December 2018. Measurements were taken for total length (TL – 0.1 cm precision) and total weight (TW – 0.01 g precision). Growth model parameters were estimated. This study provides new length-weight relationship data for *Deuterodon intermedius* (Eigenmann, 1908), *Australoheros paraibae* Ottoni & Costa, 2008, and *Hypostomus punctatus* Valenciennes, 1840. All of them ingested a wide variety of food items, including plant material like algae, animal material such as terrestrial and aquatic insects, and undefined matter such as detritus or mud. The three species showed a higher number of females. Our study provides basic information for functional diversity studies, as well as for future investigations associated with the evaluation of anthropic impacts, either occasional or chronic, on the fish assemblage structure of the Pomba River basin.

KEY WORDS. Allometric coefficient, *Australoheros paraibae*, Atlantic Forest, *Deuterodon intermedius*, *Hypostomus punctatus*, ichthyofauna, reproduction.

INTRODUCTION

The drainage basin of the Paraíba do Sul River is a large ecosystem composed of several sub-hydrographic basins (Almeida and Souza 2008). However, most published studies regarding the biology and ecology of fish are concentrated in the Paraíba do Sul River channel (e.g., Araújo et al. 2001, Teixeira et al. 2005, Pinto et al. 2006, 2009, Teixeira et al. 2019, Neves et al. 2020, Lima et al. 2021). Many of the smaller tributaries in this hydrographic basin remain poorly studied, both regarding ichthyofauna assemblage composition (Bizerril 1999, Melo et al. 2006) and the biology of fish species (Oyakawa and Menezes 2011).

The Pomba River is one of the main tributaries of the Paraíba do Sul River (Soares et al. 2007), which drains an area covering 34 municipalities in the state of Minas Gerais and three in the state of Rio de Janeiro (Silva et al. 2018). Despite growing environmental degradation (e.g., toxic substance spills, deforestation, livestock rearing, and continuous contact with domestic and industrial effluents) (Ruas 2006, Gonçalves et al. 2007, Jacovine et al. 2008, Silva 2014, Berriel et al. 2018), the Pomba River basin is home to

a highly diversified ichthyofauna (Teixeira et al. 2005), with about 51 species of fish (Caramaschi et al. 1991, Berriel et al. 2018). However, information on fish biology in this basin is still scarce. Most studies in the Pomba river basin were carried out in the river channel, recording the species' distribution (e.g., Bizerril 1999, Pompeu and Vieira 2003, Teixeira et al. 2005, Menezes and Caramaschi 2007), length-weight relationship (LWR) (Freitas et al. 2017, Freitas and Salvador 2022), reproductive aspects (Mazzoni and Caramaschi 1997), genetic diversity (Fonseca et al. 2017), and chemical contamination (Azevedo et al. 2018, Linhares et al. 2021).

The LWR is an important tool to depict many aspects concerning the life cycle of fish (Le Cren 1951, Froese 2006), such as the identification of the reproductive cycle and feeding conditions of fish populations (Camara et al. 2011). LWR parameters of fish species are key factors for fitting weight-growth curves, and are necessary for managing fishery resources in the classic Dynamic Pool Model (Beverton and Holt 1957) and similar approaches (Froese 2006).

Data on the structure of assemblages, as well as information on feeding ecology (Ribeiro et al. 2021), reproductive aspects (Rossi-Wongtschowski et al. 2009), and LWR (Freitas et al. 2017) of fish species from neotropical small streams are scarce in the literature. Given the increase in the number of studies on functional diversity that need basic information (Ribeiro et al. 2021), this information is useful for future investigations on human impacts on fish assemblages (Ribeiro et al. 2021). Thus, the present study described reproductive aspects, diets, and LWR values of the Characidae *Deuterodon intermedius* (Eigenmann, 1908), the Cichlidae *Australoheros paraibae* Ottoni & Costa, 2008, and the Loricariidae *Hypostomus punctatus* Valenciennes, 1840, three native species of the Pomba River basin, state of Rio de Janeiro, Brazil.

MATERIAL AND METHODS

The Pomba River basin covers an area of 8,544 km^2 and its spring is located at 1,182 m in altitude in the Sapateiro Mountain Range, state of Minas Gerais, Brazil (IBGE 2011). Mean annual temperatures vary between 15 and 26 °C and mean accumulated rainfall is 1,300 mm (INMET 2013), which characterizes two very distinct seasons: dry (May to September) and rainy (October to March) (Silva et al. 2018).

Samplings were concentrated from October to December 2018 in ten small streams (Table 1). Three collectors worked for up to one hour in each intensive sampling survey, using seine nets (2.0 × 1.20 m, 2 mm mesh size) and dip nets (0.46 × 0.33 m, 2 mm mesh size) during the daytime along an 80-m long stretch of the streams, which had been previously blocked with a net (0.5 cm mesh size) to prevent fish from escaping. Individuals collected were euthanized in a clove oil solution (American Veterinary Medical Association 2001) and their biometric data, including total length (TL, cm) and total wet weight (TW, g), were determined using an ichthyometer (precision of 0.1 cm) and precision digital scale (0.01 g), respectively. Fish sampling was authorized by the national environmental authority, the Instituto Chico Mendes de Conservação da Biodiversidade (ICMBio, license code: 66046-1/2018). For species identification, we consulted the most currently accepted taxonomic literature and experts. Voucher specimens were deposited in the Museu Nacional do Rio de Janeiro: *D. intermedius* (MNRJ 53753), *A. paraibae* (MNRJ 53754) and *H. punctatus* (MNRJ 53755).

Table 1. Geographical coordinates of the ten stream streches sampled in the Pomba river basin, Brazil, from October to December 2018.

Sites	Latitude (S)	Longitude (W)
1	21°29'06"	42°14'48"
2	21°23'36"	42°10'30"
3	21°22'06"	42°09'18"
4	21°24'22"	42°13'10"
5	21°30'48"	42°16'42"
6	21°31'42"	42°18'48"
7	21°28'25"	42°01'55"
8	21°29'13"	42°01'03"
9	21°34'24"	42°11'19"
10	21°36'14"	42°09'08"

The specimens were later fixed in a 10% formalin solution, and preserved in 70% ethanol. All specimens were identified to species level. The abdominal cavities of individuals were opened for the macroscopic determination of sex (following Nuñez and Duponchelle 2009) and removal of the stomach. The contents of the stomach of each specimen were examined using a stereomicroscope (Stemi 305) and food items were identified to the lowest possible taxonomic level using Bicudo and Bicudo (1970) and Mugnai et al. (2010). Volumetric frequency (Vi%) was calculated to quantify the species' diet and the volume of each item was obtained by a percentage relative to the total value of all stomach contents. Volume was obtained using dishes gridded in cubic millimeters, later transformed into milliliters (Hellawell and Abel 1971).

Growth model parameters, $TW = aTL^b$, were estimated through linear regressions and the Levenberg-Marquardt

algorithm (Lourakis 2005), applying log transformed data: lnTW = ln a + blnTL, where TW and TL are total weight and length respectively, a is the intercept, and b is the allometric coefficient.

Prior to regression analyses, log-log plots of the length and weight pairs were calculated to identify outliers (Froese et al. 2014). Outliers attributed to data error were excluded from the analyses. Additionally, a 95% confidence interval (CI) for the b coefficient and the coefficient of determination (R^2) of LWR was estimated for each species. The absence of LWR records for the three species was checked in FishBase (Froese and Pauly 2022). Sex ratio was determined for the entire sampling and deviation from the 1:1 expected ratio was tested using the Chi-squared (χ^2) test. All analyses were performed with R version 4.1.1. (R Core Team 2021) and differences with $p < 0.05$ were considered statistically significant.

RESULTS

Length-weight relationship values were calculated for 239 specimens, 93 of which belonged to the species *D. intermedius*, 21 to *A. paraibae*, and 32 to *H. punctatus* (Table 2). All linear regressions were statistically significant ($p < 0.001$). The coefficient of determination (R^2) varied between 0.9810 and 0.9974, values of the intercept (a) varied from 0.0106 to 0.01392, and values of the allometric coefficient (b) varied from 2.8934 to 3.1943. New LWR were determined for *D. intermedius*, *A. paraibae* and *H. punctatus*.

Considering all specimens examined, these ingested a wide variety of food items, including plant material like algae, animal material such as terrestrial and aquatic insects, and undefined matter (detritus/mud). The predominant items in the diet of *D. intermedius* were Coleoptera (36%) and macrophytes (15%); for *A. paraibae* it was Diptera (larva) (45%), followed by Trichoptera (15%) and larvae of unidentified insects (15%); and for *H. punctatus* it was algae such as Chlorophyta (28%), Euglenaceae (22%), and Cyanophyta (20%) (Fig. 1).

For all species, a greater number of females than males was observed. Specimens of *D. intermedius* were represented by 130 females and 56 males (sex ratio = 2.32:1; $\chi^2 = 29.4$; df = 1; $p < 0.001$), *A. paraibae* had 15 females and 6 males (sex ratio = 2.50:1; $\chi^2 = 3.9$; df = 1; $p = 0.04$), and *H. punctatus* had a sex ratio of 1.60:1, with 20 females and 12 males ($\chi^2 = 10.1$; df = 1; $p < 0.001$).

DISCUSSION

Information on fish biology in streams is scarce for most species, with the largest number of species known from a scientific perspective those with the largest size and commercial importance (Alves and Pompeu 2001). There are no studies for the Pomba River basin on the distribution and biology of fish species in streams.

The LWR allometric coefficient (b) for all species was within the expected range of 2.5–3.5 (Froese 2006) and the parameters can be used safely within the indicated length range (Froese 1998). The species analyzed showed b values close to those provided by the FishBase platform, which are based on mathematical models. In addition to the data provided by the platform, *H. punctatus* presented an isometric coefficient (b = 3). However, *D. intermedius* (b = 3.1) and *A. paraibae* (b = 2.9) showed positive allometry and negative allometry, respectively, indicating results that are contrary to those described in FishBase. The relationship parameters can vary significantly according to season and habitat, and can be affected by gonadal maturation, sex, diet, biogenetic and population aspects, and preservation techniques (Bagenal and Tesch, 1978, Silva et al. 2005), highlighting the importance of local studies. We are aware that the LWR reported are only comparable to others with either similar or iden-

Table 2. Abundance (N), total body length and total wet weight, mean (M) ± standard deviation (SD) and LWR parameters of three fish species in the streams of the Pomba River basin, from October to December 2018. Min = minimum, Max = maximum, R^2 = coefficient of determination of the LWR, 95% CI = 95% confidence interval.

	Length (cm)				Weight (g)			Regression parameters		
	N	Min	Max	M ± SD	Min	Max	M ± SD	Intercept (95% CI)	Slope (95% CI)	R^2
Characiformes: Characidae										
Deuterodon intermedius	186	3.2	5.3	3.8 ± 0.5	0.4	0.7	0.5 ± 0.2	0.0106 (0.0032–0.0243)	3.1943 (2.9258–3.3665)	0.9821
Cichliformes: Cichlidae										
Australoheros paraibae	21	4.0	6.9	5.5 ± 1.4	6.3	11.0	9.6 ± 1.9	0.01392 (0.0049–0.0492)	2.9234 (2.6832–3.1023)	0.9974
Silurifomes: Loricariidae										
Hypostomus punctatus	32	10	26.7	16.0 ± 8.1	64.0	170.5	100.0 ± 60.6	0.01298 (0.0069–0.0380)	2.8934 (2.6535–3.0287)	0.9810

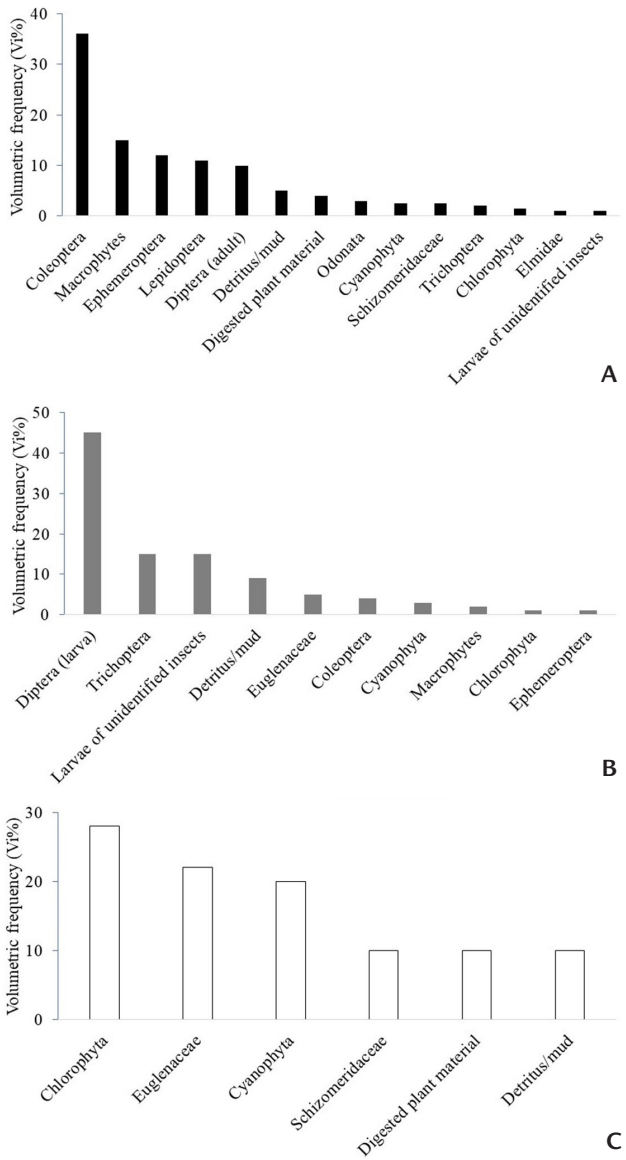

Figure 1. Volumetric frequency (Vi%) of food items identified in the diet of *D. intermedius* (A), *A. paraibae* (B) and *H. punctatus* (C) in streams in the Pomba River basin, Brazil, from October to December 2018.

tical sampling methodologies. Otherwise, the selectivity of the fishing gear that is used in different methods can yield misleading results (Froese 2006, Prchalová et al. 2020).

Information on the diet and reproduction of native species in the presence of introduced species is essential as a basis for ecosystem management and conservation plans (Súarez and Petrere-Jr 2005, Bellay et al. 2016). Insects and plant material were the main items consumed. Other studies (e.g., Vitule and Aranha 2002, Mazzoni et al. 2010, Ottoni et al. 2011, Silva and Cetra 2021) corroborate these results. In our study, *D. intermedius* consumed mainly allochthonous insects and plant material. Species of *Deuterodon* were initially described as herbivores (Géry and Junk 1977). Later, Sabino and Castro (1990), studying *D. iguape* in streams of the Atlantic forest, considered this species as omnivorous, with equal proportions of autochthonous and allochthonous diet items, inferring a possible specialization (insectivory) of smaller individuals, a difference which was associated with bowel length. In recent studies carried out in streams of the Sorocaba river basin in the state of São Paulo, Brazil, *D. intermedius* was considered an insectivorous species (with predominance of terrestrial insects) by Silva and Cetra (2021). Insectivores are the most representative trophic guild of the majority of tropical streams (Winemiller et al. 2018). As invertebrates decrease in abundance and diversity as a food source due to habitat degradation by urbanization, insectivorous species can be majorly impacted by changes in land use (deforestation) (Abelha et al. 2001), which is possibly what occurred in the Pomba river basin. Such an alteration in the environmental matrix adjacent to streams can promote changes in the diet of fish species (Barbour et al. 1999), as observed in the diet data of *D. intermedius* and *A. paraibae* in our study.

Information about the diet of *A. paraibae* is scarce and studies on *Australoheros* Říčan & Kullander, 2006 are mainly related to taxonomic aspects of the group (e.g., Ottoni and Cheffe 2009, Ottoni 2010, Ottoni et al. 2011, Ottoni and Katz 2017). Similar to our results, Ottoni and Katz (2017) in a literature review indicated aquatic insects (Hemiptera, Odonata and Coleoptera) and gastropods as the most representative items in the diet of *Australoheros* cf. *paraibae*. In the Doce River Valley, southeastern Brazil, Ottoni et al. (2011) described that species of *Australoheros* ingested mainly shrimp, followed by fish scales and aquatic insects (Chironomidae larvae, Odonata nymph). The predominance of autochthonous items of animal origin in the stomachs of this species is in accordance with the idea that autochthonous prey predominates in environments without vegetation cover (Silva et al. 2014). Thus, the consumption of such items can be explained by morphological and feeding characteristics of *Australoheros* species, with improved foraging based on exploration of the substrate (Ottoni et al. 2011). Cichlidae species present a versatile design of pharyngeal dentigerous plates used for grinding food (Kullander 2003), and the shape and orientation of their mouth, the development of the lips, and the capacity for jaw protrusion reflect where these

fishes capture their food (at the surface, bottom or middle of the water column) (Sampaio and Goulart 2011), these characteristics are also shared with *A. paraibae*.

The armored catfish *H. punctatus* presented a diet predominantly composed of plant material (algae), which is a recurrent food item in the diet of Loricariidae species (Cardone et al. 2006, Côrrea et al. 2021). The origin of food resources available to fishes may change substantially among streams and rivers. However, there is some autochthonous primary production in these systems, which several fish species take advantage (Costa and Rocha 2017). In streams exposed to direct sunlight, some species such as small catfishes (Loricariidae) are the main consumers of periphyton, which they eat as they graze on the surface of submerged tree trunks, macrophytes, and rocky substrates (Carvalho et al. 2007). Many studies have shown that Loricariidae species consume mainly plant items (Costa 1987, Casatti and Castro 2006, Mazzoni et al. 2010, Zandonà et al. 2021). They are highly specialized and have adaptations related to morphology, feeding habits, and digestive strategies, including processing of debris, sediment, algae, and periphyton (Lujan et al. 2011). *Hypostomus punctatus* have a ventral sucker mouth, typical of fish that feed on periphyton on rocks, and they inhabit and explore benthic microhabitats, grazing on algae and associated microfauna (Zandonà et al. 2021).

Dietary plasticity is a hallmark of Neotropical fish, and most species are able to switch from one food item to another since the available trophic resources might change (Manna and Rezende 2021). Such plasticity is modulated by abiotic conditions, such as microhabitat heterogeneity and resource availability (Evangelista et al. 2014), which can influence trophic variability and, consequently, individual differences in the consumption of food items. Thus, the analysis of the diet of local fish is important to understand the ecosystem services delivered by the species (Manna and Rezende 2021).

The reproductive biology of three species that are native to streams of the Atlantic forest was analyzed. The predominance of females is common in fish populations (Súarez et al. 2009), factors such as predation and variation in local environmental conditions such as temperature, growth rate, behavior or natural mortality (Vazzoler 1996) may be involved when differences in the 1:1 sex ratio are found (Garcia et al. 2004). The sex ratio is poorly discussed in the ecological context (Marcucci et al. 2005), and it may vary in different populations of the same species and at different times in the same population, however this is usually an adaptation which promotes the predominance of females and which is related to favorable conditions for the production of eggs, as, for example, during the colonization of a new environment (Nikolsky 1963).

Caramaschi and Brito (2021) describe that species of *Deuterodon* present an opportunistic strategy, classified as small fish that present rapid recruitment, early sexual maturation and reproductive activity during most of the year. Ruiz et al. (1992), point out that *Australoheros facetus* (Jenyns, 1842) spawns in the substrate with biparental care, this species carries out the strategy of rearing offspring by pairs, with territorial behavior and aggressive interactions, with a positive correlation between dominance and size for both sexes (Baduy et al. 2017). For *H. punctatus*, Menezes and Caramaschi (1994) reported that this species presents total spawning, and that the male guards and ventilates the brood for several weeks after hatching (Burgess 1989).

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