POPULATION PARAMETERS OF POECILINE IN STREAMS OF MATO GROSSO DO SUL STATE, BRAZIL*

Angélica MENDONÇA¹; Milza Celi Fedatto ABELHA²; Valéria Flávia BATISTA-SILVA²; Elaine Antoniassi Luiz KASHIWAQUI²; Dayani BAILLY²; Carlos Alexandre FERNANDES²

ABSTRACT

This study aimed to describe and compare the population structure of Phalloceros harpagos in Perobão and Água Boa streams, Mato Grosso do Sul State, through quantitative population parameters. Samplings were conducted using electrofishing in the streams' upper, intermediate and lower stretches (sites 1, 2 and 3, respectively), between March and November 2008. Standard length, weight and sex were used to determine the sex ratio, standard length classes' frequency and populations' condition at each stream. A total of 686 and 3,404 specimens of P. harpagos were captured at Água Boa and Perobão streams, respectively. Only the population of the upper stretch of the Água Boa Stream was considered in the analysis due to the inexpressive capture in intermediate and lower stretches. Males with 1.4-1.8 and 1.8-2.2 cm standard length were the most captured in both streams. Females reached greater lengths, and the most frequent were those with 2.2-2.6 and 1.8-2.2 cm in the Água Boa and Perobão, respectively. Females predominated significantly (2.5:1) in the Água Boa Stream and at Site 1 (1.3:1) of Perobão Stream. The sex ratio was the same in all other sites. The Kolmogorov-Smirnov test indicated significant difference in the distribution of individuals within size classes for both streams. The application of covariance analysis or of the separate slope model on data from the linearization of the length-weight relationship indicated the best condition of the P. harpagos population in Água Boa Stream and of that present in the upper stretch of Perobão Stream.

Keywords: Cyprinodontiformes; Phalloceros; sex ratio; condition; sexual dimorphism

PARÂMETROS POPULACIONAIS DE POECILÍNEO EM RIACHOS SUL-MATO-GROSSENSES, BRASIL

RESUMO

Este trabalho objetivou descrever e comparar a estrutura das populações de Phalloceros harpagos nos riachos Perobão e Água Boa, MS, através de parâmetros populacionais quantitativos. As amostragens foram realizadas utilizando-se pesca elétrica nos trechos superior, intermediário e inferior dos riachos (Pontos 1, 2 e 3, respectivamente), entre março e novembro de 2008. Informações relativas ao comprimento padrão, peso e sexo foram utilizadas para a determinação da proporção sexual, frequência das classes de comprimento padrão e condição das populações de cada riacho. Foram capturados 686 espécimes de P. harpagos no Água Boa e 3.404 no Perobão. Somente a população do trecho superior do Água Boa foi considerada nas análises devido a inexpressiva captura nos trechos intermediário e inferior. Machos com comprimento padrão entre 1,4-1,8 e 1,8-2,2 cm foram os mais capturados em ambos os riachos. As fêmeas apresentaram maiores comprimentos, sendo mais frequentes aquelas com 2,2-2,6 e 1,8-2,2 cm no riacho Água Boa e Perobão, respectivamente. As fêmeas predominaram significativamente (2,5:1) no Água Boa e no Ponto 1 (1,3:1) do Perobão. A proporção sexual foi equitativa nos demais pontos. O teste de Kolmogorov-Smirnov indicou diferença significativa na distribuição dos indivíduos dentro das classes de tamanho para ambos os riachos. A aplicação da análise de covariância ou do modelo com inclinação separada sobre dados provenientes da linearização da relação peso-comprimento indicaram a melhor condição da população de *P. harpagos* no riacho Água Boa e daquela presente no trecho superior do Perobão.

Palavras chave: Cyprinodontiformes; Phalloceros; proporção sexual; condição; dimorfismo sexual

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 ¹ Pós-graduação em Entomologia e Conservação da Biodiversidade, Universidade Federal da Grande Dourados, Faculdade de Ciências Biológicas e Ambientais. Rodovia Dourados-Itahum, km 12 - Cidade Universitária - CEP: 79804-970 - Dourados - MS - Brasil. e-mail: angel_bio1@yahoo.com.br (autor para correspondência)

² Universidade Estadual de Mato Grosso do Sul, Grupo de Pesquisa em Ciências Ambientais e Educação (GEAMBE). BR 163, Km 20,2 - Bairro Universitário - CEP: 79980-000 - Mundo Novo - MS - Brasil. e-mail: mcfabelha@yahoo.com.br; vfb_silva@uems.br; elainealk@yahoo.com.br; dayanibailly@gmail.com; fxande@gmail.com

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INTRODUCTION

Phalloceros harpagos (Lucinda, 2008) popularly known as guarú or barrigudinho, is a cyprinodontiform fish species that belongs to the family Poeciliidae, subfamily Poeciliinae, whose distribution is restricted to South America (LUCINDA, 2008). In a recent taxonomic proposal to poecilines, new species for the genus *Phalloceros* were described, and the occurrence of *P. harpagos* was recorded for the Upper Paraná River basin (LUCINDA, 2008) (site of this study) and not *P. caudimaculatus*, as pointed out in previous surveys on fish fauna in this basin (CASTRO and MENEZES, 1998; AGOSTINHO *et al.*, 2004; LANGEANI *et al.*, 2007).

Poecilines are small fish that have internal fertilization and peculiar characteristics like ovoviviparity and sexual dimorphism (CASATTI *et al.*, 2001). The latter is shown by the larger size of females that in various species reach twice the males' size and by the first ray of males' anal fin turned into a gonopodium (VAZZOLER, 1996; BISAZZA and PILASTRO, 1997; CASATTI *et al.*, 2001).

Another characteristic common to members of this subfamily is the occurrence in streams (ARAÚJO et al., 2009) and, in the particular case of P. harpagos, it preferably inhabits stretches of backwaters (ARANHA and CARAMASCHI, 1997; CASATTI et al., 2001; ABILHOA et al., 2008). This species feeds on food from the water surface (CASATTI et al., 2001; ABILHOA et al., 2008) and presents diet composed predominantly of allochthonous plant matter and insects (CASTRO and CASATTI, 1997). However, the food spectrum may also include algae (DEUS and PETRERE JR., 2003) and detritus (OLIVEIRA and BENNEMANN, 2005).

The feeding plasticity combined with the reproductive behavior (i.e., ovoviviparity) are factors associated with high population abundance of *P. harpagos* in several surveys of fish populations from streams of the Upper Paraná River basin (CASATTI *et al.*, 2001; CASATTI, 2005; OLIVEIRA and BENNEMANN, 2005; WOLFF *et al.*, 2007). Despite its numerical representativeness in this basin, *P. harpagos* populations in Mato Grosso do Sul State were

little studied, especially regarding the estimation of its population parameters.

Quantitative of population parameters structure as size distribution, sex ratio and condition, are useful tools in the knowledge of fish ecology by revealing the relationship between species and environmental conditions the (WOOTTON, 1999). Particularly, the study of the condition assumes that heavier fish of a certain size are at better condition and thus, this attribute is often used as an indication of population welfare (VAZZOLER, 1996; POPE and KRUSE, 2007). Fish at relatively better condition present higher growth rate, higher reproductive and survival potential, summarizing the response of populations to changes in habitat (CAMARA et al., 2011).

It is noteworthy that recent fish surveys developed by the University of Mato Grosso do Sul State in four streams at Upper Paraná River basin showed an expressive numerical abundance of *P. harpagos* in two of these streams: Perobão and Água Boa (unpublished data). Thus, in order to extend the knowledge of *P. harpagos'* ecology in this region, this study describes and compares the distribution in length, sex ratio and the condition of this species in Perobão and Água Boa streams, Mato Grosso do Sul State, Brazil. Given the high abundance of *P. harpagos* in the Perobão Stream, this attribute was also examined over the spatial scale at this stream.

MATERIAL AND METHODS

Study area

Perobão and Água Boa are first-order streams, with 5.3 and 6.0 km long, respectively. They are located in the southern region of Mato Grosso do Sul State and are part of the Iguatemi River basin, a tributary of the right bank of the Paraná River (Figure 1). Both have their headwaters in the municipality of Japorã and run to rural areas until flowing into the right bank of Iguatemi River.

Another aspect in common are the kind of human impacts imposed, highlighting the lack of riparian vegetation and siltation. The replacement of native vegetation for agriculture and pasture is part of the history of regional occupation (SÚAREZ and PETRERE, 2007). As personal observation, we highlight some environmental features, as follow. Much of the riparian vegetation was reduced and it is generally formed by sparse or absent trees with pasture grasses as the most abundant component. The local soil is sandy and is easily eroded when improperly used in farming activities in a way that gully erosions are common in the landscape, which intensifies the input of sediments to the streams. The rural population frequently uses the streams as water supply for livestock. This habit turns in another aggravating factor in the silting process due to landslide caused by animal trampling on the banks.

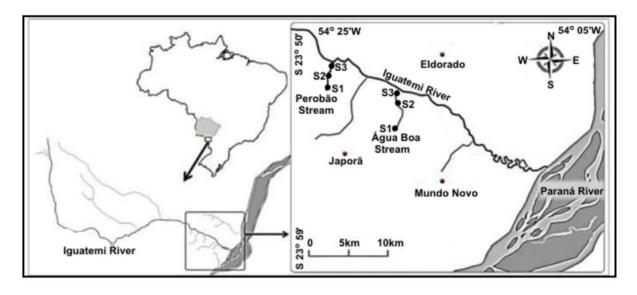


Figure 1. Location of Água Boa and Perobão streams, sampling sites and municipalities of Japorã, Mundo Novo and Eldorado, Iguatemi River basin, Mato Grosso do Sul State, Brazil. S1, S2 and S3: sampling sites.

Considering the accessibility and environmental heterogeneity along the longitudinal axis of the streams, three sampling sites were established, which encompassed their upper, middle and lower reaches, respectively called Site 1, Site 2 and Site 3. The location and characteristics of each site are detailed in Table 1.

Samplings

Four samplings were conducted at each sampling site, corresponding to the months of March/June/September/November 2008. Fish were collected using electrofishing (portable generator TOYAMA 1600, 220V, DC). The sampling effort was standardized throughout streams and months. The standardization protocol is described as follow. The extent of each sampled stretch was established according to (1998): FITZPATRICK et al. five width measurements (ranging between 2 to 6 m and 2 to 10 m for Água Boa and Perobão streams, respectively) were taken from each stream (with

emphasis on environmental heterogeneity, including habitats with fast flows, rapids and pools) and then calculated the arithmetic mean of these measurements, with the result multiplied by twenty. Thus, the minimum and maximum extent of the stretches sampled was 50 m and 94 m, respectively. At the end of each stretch, blocking nets (10.0 x 2.0 - 5.0 mm mesh) were assembled to prevent fish to escape. A single electrofishing pass was used to remove the fishes in each stretch.

Fish caught were placed in plastic bags, identified as for the stream, stretch sampled, date and fixed in 10% formalin and subsequently preserved in 70% alcohol. Each specimen was identified according to GRAÇA and PAVANELLI (2007) and specimens of *P. harpagos* were separated from the others. In the laboratory of the University of Mato Grosso do Sul State/Mundo Novo, the specimens were measured (cm, standard length = SL), weighed (g, total weight = TW) and sexed as males if they possessed any

evidence of a gonopodium (under stereomicroscope observation) and as females in the case of the

absence of gonopodium, but were larger than the smallest male (0.7 cm).

Stream	Sampling sites	Location and characteristics				
Água Boa						
	Site 1	23°52'42.24''S; 54°21'55.37''W. Backwater; clayey bed; marginal vegetatic consists of scattered trees, shrubs and grasses for grazing; abundant aquat grasses, width between 2.0 and 6.0 m and depth between 12.0 and 15.0 cm.				
	Site 2	23°50′16.65″S; 54°20′55.54″W. Backwaters alternate with rapids and pools; rocky and sandy bed; marginal vegetation composed of small clusters of trees and pasture grasses; use in recreational activities; width between 2.2 and 5.5 m and depth from 30.0 to 90.0 cm.				
	Site 3	23°50′3.33′′S; 54°20′58.53′′W. Rapid water flow; sandy bed; marginal vegetation consists of scattered trees, shrubs and grasses for grazing; natural course changed to create a fish farming, width between 2.2 and 2.7 m and depth from 30.0 to 63.0 cm.				
Perobão						
	Site 1	23°49′25.26″S; 54°26′43.30″W. Backwater; clayey bed; slightly steep banks; marginal vegetation consists mainly of pasture grasses; intense transit of cattle, width ranging from 2.0 to 10.0 m and depth from 5.0 to 15.0 cm.				
	Site 2	23°48′59.35′′S; 54°26′40.06′′W. Pools and rapids, with a prevalence of the latter; predominantly rocky and sandy bed, steep banks with narrow band of riparian vegetation and pasture grasses, width between 1.0 and 4.5 m and depth from 10.0 to 50.0 cm.				
	Site 3	23°48′05.50′′S; 54°26′26.00′′W. Alternating backwaters, rapids and pools; rocky and sandy bed, slightly sinuous, entrenched (with steep banks) and limited downstream by a waterfall; scarce marginal vegetation with scattered trees, and pasture grasses; width varying from 2.0 to 3.6 m and depth between 26.0 and 45.0 cm.				

Data analysis

To evaluate the structure in length, populations of each stream were divided into standard length classes of 0.4 cm interval. Possible differences in sex ratio were evaluated using the chi-square test ($\chi^2_{0.05}$ = 3.84, d.f = 1). The Kolmogorov-Smirnov test was applied to evaluate possible differences in the distribution of individuals within size classes, with significance level at α = 0.05. This evaluation was performed for the total captured in each stream, as for the specimens present along the longitudinal axis of the Perobão Stream. It is noteworthy that P. harpagos was found almost exclusively in the Site 1 of the Água Boa Stream, with a record of only three specimens in the other sampling sites, so they were not considered in the analysis.

For assessing fish condition between streams and along the Perobão Stream, we used the results from the linearization of the length-weight relationship of the specimens given by the equation: $TW = aSL^b$ through its natural logarithm: lnTW = lna + blnSL, where TW is the total weight in grams, SL is the standard length in cm, *a* is the intercept and *b* is the regression coefficient (POPE and KRUSE, 2007).

The condition was evaluated by analysis of covariance (ANCOVA) (GARCÍA-BERTHOU, 2001; POPE and KRUSE, 2007). The total weight of individuals was the dependent variable, the streams/sites of Perobão were the categorical variables and the individuals´ standard length was the continuous variable. When data did not meet the homogeneity of slopes assumption, the

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populations were compared using the separate slope model considering the Johnson-Neyman procedure (HUITEMA, 1980). This analysis identifies values of standard length that may be associated with significant differences in total weights between stream/sites, and consequently, allows for the identification of regions of non-significance and significance (HUITEMA, 1980). When significant differences were detected, the population with the highest mean of the natural logarithm of weight adjusted to a certain standard length was considered at best condition. Statistical analyses were carried out by the software StatisticaTM (STATSOFT, 2007) and the level of significance was set at $\alpha = 0.05$.

RESULTS

It was captured a total of 686 (199 males and 487 females) specimens of *P. harpagos* in the Água Boa Stream and 3,404 (1,660 males and 1,744 females) in the Perobão Stream. Of this total in Perobão, 680 were captured at Site 1 (294 males and 386 females), 93 at Site 2 (49 males and 44 females) and 2,631 at Site 3 (1,317 males and 1,314 females). The minimum and maximum values of standard length (SL) for males and females corresponded, respectively, to 1.1-2.4 cm and 0.7-3.6 cm in the Água Boa Stream and 0.8-2.9 cm and 0.8-3.4 cm in the Perobão Stream. Only two specimens smaller than 0.7 cm were caught and

they were excluded from the analysis since there was no indication of gonopodium development, preventing sex identification.

With respect to sex ratio, females predominated significantly ($\chi 2 = 121.7$) in the Água Boa Stream with ratio of 2.5:1. In Perobão, the population was not significantly different from 1:1 for the total catch ($\chi 2 = 2.1$), for the Site 2 ($\chi 2 = 0.3$) and Site 3 ($\chi 2 = 0.003$). There was a significant predominance of females ($\chi 2 = 6.2$) in the Site 1 with ratio of 1.3:1.

The Kolmogorov-Smirnov test identified differences in significant size frequency distributions for both sexes for the total captured in each stream (D \Im Agua Boa = 0.40, p<0.01; D \Im Perobão = 0.15; p < 0.01; $D_{+Agua Boa}^{\circ} = 0.11$, p < 0.01; $D_{+Perobão}^{\circ} = 0.11$ 0.07; p<0.01) and for Site 1 and Site 3 of Perobão Stream (D_d_{Site1} = 0.17, *p*<0.01; D_d_{Site3} = 0.17; *p*<0.01; $D^{\circ}_{+ \text{Site1}} = 0.09$, p<0.01; $D^{\circ}_{+ \text{Site3}} = 0.85$; p<0.01). No significant differences in size frequency distributions were observed for males and females in Site 2 of Perobão Stream (D $_{OSite 2}$ = 0.17, p<0.15; D_{+Site2}° = 0.16; *p*>0.20). Males showed a higher relative frequency in standard length classes of 1.4-1.8 cm in both streams (Figures 2 and 3). Females predominated in the classes of 2.2-2.6 cm and 1.8-2.2 cm in the Água Boa and Perobão streams, respectively, and reached greater amplitude of standard length in both streams (Figures 2 and 3).

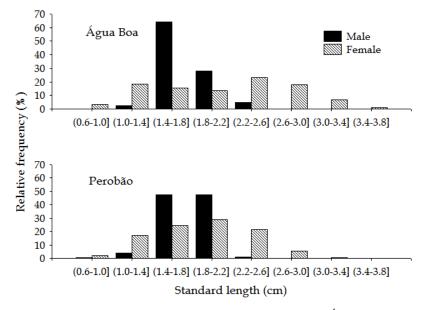


Figure 2. Size distribution of male and female *Phalloceros harpagos* at Água Boa and Perobão streams, Iguatemi River basin, State of Mato Grosso do Sul, Brazil.

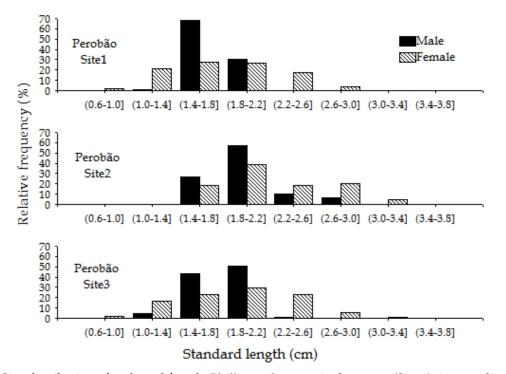


Figure 3. Size distribution of male and female *Phalloceros harpagos* in the upper (Site 1), intermediate (Site 2) and lower (Site 3) stretches of the Perobão Stream, Iguatemi River basin, State of Mato Grosso do Sul, Brazil.

In the analysis of the condition between populations of the two streams, the homogeneity of slopes was not met, precluding the calculation of adjusted means for a single standard length (Table 2; Figure 4). Thus, the application of the SSM and the Johnson-Neyman procedure revealed that the largest specimens and those with intermediate length had better condition in the Água Boa Stream, whereas the smaller specimens showed the best condition in the Perobão Stream (Table 3).

Table 2. Results of slope homogeneity tests, ANCOVA and separate slope model applied to the data resulting from the logarithmic transformation of the length-weight relationship of *Phalloceros harpagos* present in the Água Boa and Perobão streams, Mato Grosso do Sul State, Brazil. F = F statistics, p = p statistics; S1 = Site 1, S2 = Site 2, S3 = Site 3.

Factors/Statistic	Homogeneity of slopes		ANCOVA		Separate Slope Model	
	F	р	F	р	F	p
Água Boa and Perobão	31.10	< 0.001	-	-	48.66	< 0.001
S1 and S2 and S3 Perobão	0.03	0.86	86.3	< 0.001	-	-

Otherwise, the homogeneity of slopes was detected in the analysis of fish condition among the different stretches of the Perobão Stream (Table 2; Figure 5). The application of ANCOVA indicated a significant difference between the means of the total weight adjusted to the standard length of 0.588. The adjusted means reveled best condition for the fish of the Site 1 (mean $\ln TW = -1.96 \pm 0.006$), intermediate condition for the fish of the Site 3 (mean = $\ln TW$ -2.04 ± 0.003) and worst condition for the fish of the Site 2 (mean = $\ln TW$ -2.09 ± 0.016). The Tukey's test indicated that all means differed from each other (*p*<0.001).

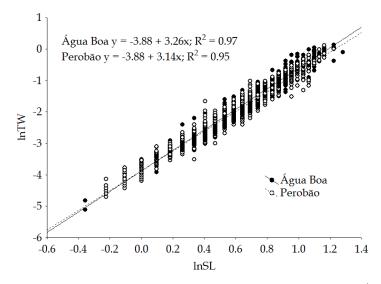


Figure 4. Length-weight relationship linear regression of *Phalloceros harpagos* in the Água Boa and Perobão streams, Iguatemi River basin, State of Mato Grosso do Sul, Brazil.

Table 3. Mean values of the natural logarithm (ln) of the total weight of specimens for minor standard length, intermediate standard length and major standard length.

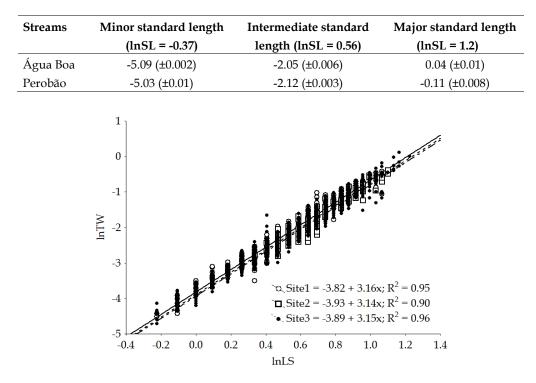


Figure 5. Length-weight relationship linear regression of *Phalloceros harpagos* between sites of the Perobão Stream, Iguatemi River basin, State of Mato Grosso do Sul, Brazil.

DISCUSSION

The significant numerical representativeness observed for *P. harpagos* in Água Boa and Perobão

streams can be explained by three factors: (i) it is a typical stream species, with preference for stretches of shallow and slow-flowing waters (MAZZONI *et al.*, 2011), which is provided by

both streams, mainly by Perobão; (ii) tolerance of *P. harpagos* to environmental changes (CASTRO and CASATTI, 1997; MAZZONI *et al.*, 2011), once the environments studied were impacted, mainly by the removal of riparian vegetation, siltation and transit of animals; (iii) ovoviviparity, which provides a greater chance of survival for the juveniles (VAZZOLER, 1996).

Considering the population attributes, the maximum and minimum standard lengths found for *P. harpagos* in both streams were consistent with the range of values reported for populations of this species in several Brazilian streams (ARANHA and CARAMASCHI, 1999; SHIBATTA and CHEIDA, 2003; WOLFF *et al.*, 2007; ARAÚJO *et al.*, 2009; ZANATTA *et al.*, 2013).

For sex ratio, the literature indicates significant predominance of females in other populations of P. harpagos (ARANHA and CARAMASCHI, 1999; WOLFF et al., 2007; ARAÚJO et al., 2009; ZANATTA et al., 2013) as it was detected for Água Boa Stream and Site 1 of Perobão. Nevertheless, an equitable proportion between the sexes, as found for the total population of Perobão, was also described by ZANATTA et al. (2013) in a stream of the Paraná State. Despite the classic relationship of fish sex ratio is 1:1 (WOOTTON, 1999), variations among populations of the same species have been described for fish (VAZZOLER, 1996, ARANHA and CARAMASCHI, 1999; ARAÚJO et al., 2009). The causes of these variations are not clearly known (ARAÚJO et al., 2009) and the literature points to the influence of a set of factors. One is the environmental quality, so that females tend to be more frequent when conditions are more favorable, especially under abundant food resources (NIKOLSKY, 1978; VAZZOLER, 1996).

Unbalanced sex ratio could still occur when males and females are subjected to different predation and growth rates (WINEMILLER, 1993; ARANHA and CARAMASCHI, 1999; ARAÚJO *et al.*, 2009). The argument is that the sex ratio could be related to the size difference between the sexes: small males are more susceptible to predation by other fish than females (ARAÚJO *et al.*, 2009). Moreover, differential rates of growth between the sexes could cause changes in this ratio, since sex with faster growth would pass more rapidly through smaller stages, which are the most vulnerable to predation, and thus have gradually increased frequency in the population (ARAÚJO *et al.*, 2009).

Unlike the sex ratio that can vary between populations of *P. harpagos*, the length distribution seems to follow a pattern. The smaller amplitude of variation of length classes in males and the predominance of females in larger size classes were also described in other studies on this species (ARANHA and CARAMASCHI, 1999; CASATTI *et al.*, 2001; ARAÚJO *et al.*, 2009; WOLFF *et al.*, 2007; ZANATTA *et al.*, 2013).

Among fish, there is a general trend that adult females are larger than males (NIKOLSKI, 1978; LOWE-McCONNELL, 1999), which is relevant to reproductive success (GROSS, 2005). The larger body size of females contributes significantly to the increased fecundity of the species, because it allows a greater number of eggs and/or embryos to be carried (VAZZOLER, 1996; WOOTTON, 1999). In addition to that, studies have shown that the smaller body size of males of poeciliids is an advantageous feature, since there is a negative and significant relationship between copulatory success and males' length (BISAZZA and MARIN, 1995; BISAZZA and PILASTRO, 1997). The remarkable maneuverability of small males gives greater number of successful thrusts compared to larger males, resulting in high insemination rates (BISAZZA and MARIN, 1995; BISAZZA and PILASTRO, 1997).

Furthermore, a larger number of females in the Água Boa Stream achieved larger classes. This suggests the occurrence of more favorable conditions in Água Boa, especially the relative abundance of food resources. In this regard, VAZZOLER (1996) and WOOTTON (1999) emphasized that the energy and nutrients provided by the feeding activity are among the main factors that determine variations in length of fish of the same species that occupy different environments.

Another population parameter pointed as a way to predict the environmental quality is the condition (STEVENSON and WOODS, 2006). In this context, the best condition of *P. harpagos* in the Água Boa Stream was consistent with environmental characteristics favorable to

ecological requirements of the species. The upper reach of this stream presents shallow and large backwater, clayey riverbed colonized by aquatic grasses whose roots and stems allow the accumulation of detritus and periphyton, offering an environment potentially rich in food resource and shelter for the juveniles and adults. Stream sections structured in this way have been described as sites commonly inhabited by P. harpagos (ARANHA and CARAMASCHI, 1997; CASATTI et al., 2001; ABILHOA et al., 2008). Among the different sections sampled in the Perobão stream, the best condition and the higher proportion of females were found for specimens at the Site 1, which similarly to Água Boa, is the stretch with large and shallow backwater area with clayey bed.

It is worth noting that the type of habitat used by a fish species is related to their morphophysiological characteristics, such as shape, size, mouth position and fins size (WOOTTON, 1999). In the case of *P. harpagos*, its small size, upper mouth and small fins, qualify the species as highly specialized to live in slow-flowing waters (MAZZONI *et al.*, 2011). These authors reported a significant species preference (detected by Ivlev Electivity Index) for shallow habitats (22-32 cm), with low water velocity (0 to 0.5 cm sec⁻¹) and clay/sandy bottom.

This selectivity of *P. harpagos* justifies the low number of catches and comparatively worse condition of *P. harpagos* in the Site 2 of Perobão Stream, whose environmental features include rapids and, predominantly, rocky bottom. The Site 3 also provided a microhabitat with rapids, but also shallow backwaters and pools, which could explain the species high capture. In turn, the intermediate condition of specimens at this site is an indication that the population density may be negatively influencing the fish's condition. Admittedly, increasing population density is a factor that reduces the availability of food and space, and consequently, the condition of species (WOOTTON, 1999).

CONCLUSIONS

The capture of *P. harpagos* in different sites of the Perobão Stream reflects the presence of appropriate structural components for the species along its course. However, the sex ratio statistically equal to 1:1 and the lowest condition suggest that the population is under an unfavorable environmental scenario. It is noteworthy that, if this scenario persists, the population could decline. In contrast, the sex ratio with predominance of females and most favorable condition in the Água Boa Stream reflect milder environmental constraints for the species. Meantime, the fact that the population is distributed almost exclusively in the Site 1 evidences that drastic changes in this stretch compromise the occurrence of *P. harpagos* in the Água Boa Stream.

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