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Original Article

Diet of characin, *Brycon falcatus* (Muller and Troschel, 1844) in the Amazon Basin: a case study on an attractant for fish in the Teles Pires River

By L. S. de Matos[1,2], J. O. S. Silva[3], P. S. M. de Andrade[3] and L. N. Carvalho[1,2]

[1]*Programa de Pós-Graduação em Ciências Ambientais, Universidade Federal de Mato Grosso-UFMT, Campus Universitário de Sinop, Sinop, MT, Brazil;* [2]*Programa de Pós-Graduação em Ecologia e Conservação da Biodiversidade, UFMT, Campus Universitário de Cuiabá, Cuiabá, MT, Brazil;* [3]*Instituto de Ciências Agrárias, UFMT, Campus Universitário de Sinop, Sinop, MT, Brazil*

Summary

The use of fresh soybeans as an attractant in fishing has been altering the diet of the characiform *Brycon falcatus*, which are thriving at fishing spots. This has subsequently affected the amount of abdominal fat in the species from the Tapajós River basin, Southern Amazon. In the Teles Pires River basin region, one of the most common capture techniques employed by fishermen is the use of attractant feeds (soybeans, corn, cassava) at certain locations to attract mostly omnivorous fish. Tourists also feed fish chips and breadcrumbs. In the experimental design, rivers were mapped according to attractant availability to develop an attractant-density classification system comprising four rivers with different attractant densities (low, medium, and high), plus a river serving as control (no attractants). Monthly collections were carried out during the 2012 and 2013 dry seasons. For diet analyses, methods of frequency of occurrence and relative volume were used to calculate the feeding index (*IAi*) of 97 specimens. For analyses of abdominal fat, an index was calculated by dividing the wet weight of the abdominal fat by the total wet weight of each individual. Standard length ranged from 15.0 to 48.5 cm, and weight between 0.18 and 4.40 kg. Composition and diversity of diet items changed with the increasing density of attractants. In the river with a high attractant density, fresh soybeans and corn were dominant diet items; in the control river, the diet was natural (e.g. seeds, fruits, leaves, and insects) and therefore similar to those described for *B. falcatus* in undisturbed environments. Fish collected from rivers with high attractant densities were in better condition than those from the river with no attractants. Although use of attractants is prohibited by State legislation, there are no inspections. This study clearly shows an alteration in the natural diet and abdominal fat of *B. falcatus* resulting from an imbalanced, high-calorie feed via an attractant. It is recommended that these areas be monitored for the preservation of *B. falcatus*.

Introduction

Brycon falcatus (Muller and Troschel, 1844) is a migratory fish, popularly known as matrinxã. *Brycon* species generally live in small schools and have an omnivorous diet that includes fruits, seeds, flowers, insects, fish and even small vertebrates (Goulding, 1980; Lilyestrom and Taphorn, 1983; Correa et al., 2007). Studies of the *Brycon falcatus* diet include specimens from the Mesay River in the Colombian Amazon (Blanco-Parra and Rodriguez-Bejarano, 2006) and the Tocantins River (Albrecht et al., 2009). In the Negro River, *B. falcatus* exhibited frugivorous (during the rainy season) and insectivorous (during the dry season) feeding habits, with some preference for aquatic insects (Borges, 1986). In the middle of the Teles Pires River, *B. falcatus* was classified as omnivore, with material of allochthonous origin, such as plants and flowers, the most frequent diet items (Godoi, 2004). According to Goulding (1980), flooded forests and riparian forests are the main sources of energy for this and other fish species in the Amazon.

The main economic activities conducted in southern Amazonia, particularly in the Teles Pires River basin, are logging, gold mining, ranching and agriculture (mainly corn and soybeans) (Barthem and Goulding, 1997). In the Teles Pires River basin, one of the current most frequently used fishing techniques is to supply a food supplement (SFS). ‘Food supplement’ is regionally known as attractant or ‘cevas’ (Matos and Carvalho, 2015), such as soybeans, corn, or cassava placed in a certain area to attract mainly omnivorous fish. However, tourists also employ SFS to observe fish (Bessa and Gonçalves-de-Freitas, 2014). Public viewing of bonitos, *B. hilarii* (Sabino et al., 2005) is also enhanced by offering snack foods and breadcrumbs as attractants (Sabino et al., 2005). In coral reefs in Australia, tourists feed the fish, a practice that results locally in an increase in liver fat of stationary species, which can be fatal. In coral reefs of Northeast Brazil, food supplied by tourists has caused changes in the composition of the fish assemblage (Ilarri et al., 2008). In French Polynesia, feeding sharks results in their becoming easy prey, and altering their role as cleaners (Clua, 2010). In the Cayman Islands, the use of artificial foods has changed the behavior and spatial distribution and increased the densities of stingrays (Corcoran et al., 2013). Even a small human-induced perturbation can affect the behavior or population biology of a species locally and influence the function of the species within its community (Geffroy et al., 2015). In this context, the use of feeds might cause harm to animals and ecosystems.

Due to their large supply in the region, fresh soybeans (*Glycine max*) are most frequently used in the Teles Pires River basin as a fish attractant (Matos and Carvalho, 2015). However, soybeans have several anti-nutritional factors (Francis et al., 2001), which may even cause mortality if used for extended periods (Makkar and Becker, 1997; Sabino et al., 2005). Thus, the aim of this study was to compare the diet of *B. falcatus* in the Teles Pires River basin collected from sites with and without such supplemental feeding practices as well as to examine the accumulation of abdominal fat in these fish.

Materials and methods

Study area

The Teles Pires River basin is located in the northern part of the State of Mato Grosso in the Brazilian Legal Amazon and is a major tributary of the Tapajós River. The Teles Pires River (also known as São Manuel) is a clear-water river. The main economic activities in the area around this basin have been logging, gold mining, ranching and agriculture (especially corn and soybeans), which have had a severe effect on the water resources (Barthem and Goulding, 1997).

The experimental design of our study considered the quantity of attractant used at various sampling sites in the Teles Pires River basin (Fig. 1). Collections took place in the (a) Verde River (municipality of Sorriso), which is classified as having a low density of attractant, with approximately one type of attractant used for each 1000 m of river; (b) Celeste River (municipality of Vera), classified with a medium level use of attractant (one 'attractant' for every 500 m of river; (c) Teles Pires River (municipality of Sinop), classified as a high level of attractant use with about one attractant for every 100 m of river; (d) Tapaiuna River (municipality of Nova Canaã do Norte), where currently no attractants are being used but with a location relatively close to the attractant system; and (e) Cristalino River (municipality of Novo Mundo), which served as the control site, and within a Conservation Unit.

The Cristalino River flows principally within the Cristalino State Park ('Parque Estadual do Cristalino' – PEC). The area surrounding the Cristalino basin is legally protected against deforestation and human settlement. The Cristalino River is a typical headwater river with black water (dystrophic) (Godoi, 2004) and therefore served as the control. Additional to the diet results of fish from the Cristalino River, for comparison we used data from the literature on the *Brycon* diet in non-impacted environments.

Fish collection and biometrics

Fish were collected monthly during the dry season (May – October) of 2012 and 2013. Various capture techniques were used, including gillnets (mesh size 12 cm between knots), 60-meter longline with 30 No. 7 hooks, 'anzol de galho' (hooks tied to a branch with a longline), and a fishing rod with artificial bait. After capture, the fish were anesthetized with Eugenol and submerged in ice (American Veterinary Medical Association, 2001; Vidal et al., 2008). Fish were then placed in labeled plastic bags and packed again in ice. Total length (TL), standard length (SL, nearest 0.1 cm), total weight (TW, to 0.01 g), degree of stomach fullness, and abdominal fat weight were recorded in the laboratory of the Federal University of Mato Grosso (Biological Collection Southern Amazon 'Acervo Biológico Amazônia Meridional' – ABAM). The stomachs were removed, fixed in 10%

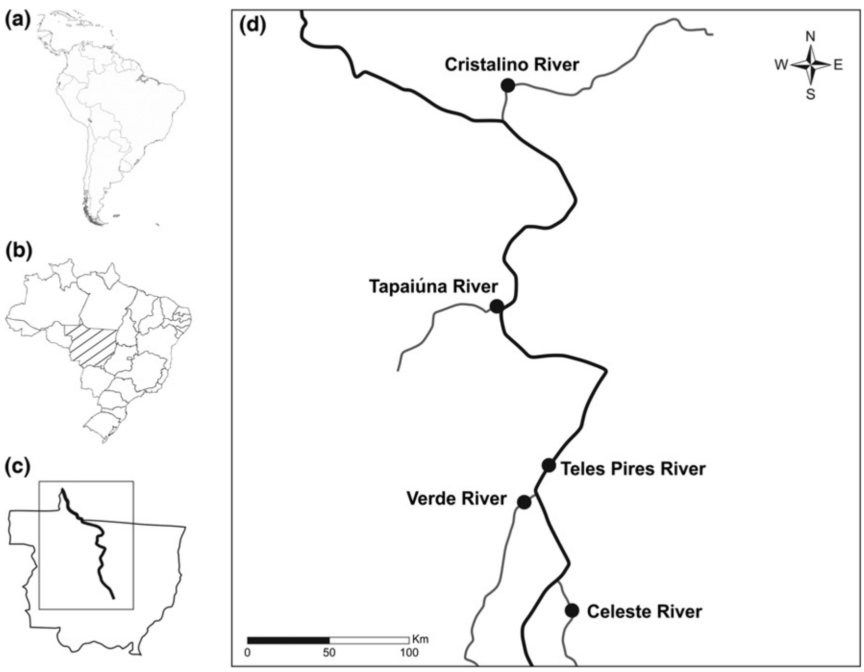

Fig. 1. A. Map of South America. B. Map of Brazil. C. Map of Mato Grosso state highlighting Teles Pires River basin. D. Collection points for *Brycon falcatus,* Teles Pires River basin, sampled in dry season, 2012 and 2013: Celeste River (12°24′56,00″ S; 55°31′28,00″W), Verde River (11°4′1,99″S; 55°34′17,00″W), Teles Pires River (11°34′48,00″S; 55°39′5,00″ W), Tapaiúna River (10°41′29,28″S; 55°56′51,11″W) and Cristalino River (9°32′47,00″S; 55°47′38,00″W). State of Mato Grosso, Brasil

formalin and stored in 70% ethanol for further analyses. Voucher specimens were deposited at INPA and UNICAMP (n° ZUEC 9190).

Diet analysis

Stomach contents were removed, placed in Petri dishes and observed under a stereomicroscope; an optical microscope was used when necessary. The degree of stomach fullness was determined by visual assessment of the contents, classified according to Santos (1986) by category: grade 0 = empty stomach; grade 1 = 25% fullness; grade 2 = 50% fullness; grade 3 = 75% fullness; and grade 4 = 100% full stomachs.

Relative volume was also identified for each fish as the proportion to the total volume of the stomach contents (Hyslop, 1980). Food items were identified to the lowest taxonomic category possible. The frequency of occurrence of each item was calculated. In addition, the relative volume was also calculated (amount of each item to the total volume). By combining these two parameters, we obtained the feeding index (IA_i), which indicated the importance of each item in the diet (Kawakami and Vazzoler, 1980) according to the equation: $IA_i = F_i.V_i/\sum(F_i.\ V_i)$, where IA_i = the feeding index; F_i = the frequency of occurrence (%) of item i and V_i is the ratio of the item i volume to the total contents.

Analysis of abdominal fat

The abdominal fat weight was determined, divided by the total weight of each fish and expressed as a percentage.

Statistical analysis

Qualitative (item occurrence) and quantitative (item percentage) data of the *B. falcatus* diet in each river were compared using NMDS (Non-Metric Multidimensional Scaling) to test whether there were differences in the diet composition between rivers, also giving the percentage consumed of each diet item in each river (statistical software R version 3.0.2). The data generated were then subjected to MANOVA to determine if there were significant differences among diets.

Abdominal fat data from *B. falcatus* in the Teles Pires basin rivers were expressed as the mean and standard deviation and subjected to analysis of variance (ANOVA) followed by Tukey's post-hoc test (R Statistical Software version 3.0.2), considering a significance level of 5% ($P < 0.05$).

Results

A total of 97 *Brycon falcatus* were captured in the Teles Pires River basin; the means and ranges of total and standard length as well as their weights are presented in Table 1.

In the Cristalino River the diet of captured *B. falcatus* specimens consisted of 16 items (Table 2). Most important were leaves (IA_i = 9.53), followed by crustaceans (Decapoda) (IA_i = 8.48) and fish (IA_i = 6.31). Also incorporated were seeds (IA_i = 2.93), flowers (IA_i = 0.58), and fruits (IA_i = 1.16), and included minor items such as scales, bivalves, vertebrates, insects of the orders Coleoptera and Hymenoptera, and detritus. After analyzing the entire composition of the diet and the feeding index, we classified the diet of *B. falcatus* in the Cristalino River as omnivorous with a tendency to carnivory.

The diet of *B. falcatus* in the Tapaiuna River consisted of 13 items (Table 2). Most important was fruit (IA_i = 27.05), followed by seeds (IA_i = 4.16) and scales (IA_i = 3.93). The diet also consisted of flowers (IA_i = 2.78), crustaceans (Decapoda) (IA_i = 1.39), leaves (IA_i = 0.35), and minor items, such as insects of the orders Orthoptera, Hymenoptera, Lepidoptera, Coleoptera, and Arachnids and detritus. *B. falcatus* in the Tapaiúna River was classified as an omnivore with a tendency to frugivory.

The diet of the samples collected from the Verde River consisted of 12 items (Table 2). Most important was seeds (IA_i = 42.12), followed by flowers (IA_i = 4.98) and soybeans (IA_i = 4.79). Leaves were also present (IA_i = 2.81), together with minor items such as scales, insects of the orders Hymenoptera and Hemiptera, Coleoptera, and Hemiptera, and detritus. In the Verde River, *B. falcatus* exhibited an omnivorous diet with a tendency to frugivory due to the importance of fresh soybeans.

In the Celeste River the diet of *B. falcatus* consisted of 11 items (Table 2). Most important were fresh soybeans (IA_i = 62.65), followed by corn (IA_i = 21.47) and fish (IA_i = 0.39). The diet also included minor items, such as leaves, fruits, seeds, scales and insects of the order Hymenoptera. Considering soybeans and corn to be fruits, the diet of *B. falcatus* in the Celeste River was classified as omnivorous with a

Table 1
Means and ranges of total length (TL), standard length (SL) and weight, *Brycon falcatus* specimens, Teles Pires River basin, dry season of 2012 and 2013

River	n	TL (cm)	SL (cm)	Weight (Kg)
Cristalino	36	45.3^{ab} (28.0–54.0)	35.0^{ac} (15.0–43.5)	1.70^{a} (0.18–3.01)
Tapaiúna	10	34.5^{ab} (28.5–39.5)	27.4^{b} (22.5–31.0)	0.67^{b} (0.35–0.95)
Verde	10	38.4^{a} (23.5–51.0)	30.5^{ab} (19.0–41.0)	1.09^{ab} (0.21–2.83)
Celeste	14	42.3^{ab} (23.5–57.5)	33.7^{abc} (19.0–47.5)	1.54^{abc} (0.21–4.34)
Teles Pires	27	42.8^{b} (23.5–57.5)	34.4^{ac} (19.0–48.5)	1.66^{ad} (0.21–4.40)
P		<0.01	<0.001	<0.001
SD		16.66	6.96	0.89
CV		44.52	20.32	55.76

Rivers followed by the same letter did not differ significantly ($P > 0.05$) by ANOVA. SD, standard deviation; CV, Coefficient of variation.

Table 2
Frequency of occurrence (F), relative volume (RV), and Index of Alimentary Importance (IA_i) according to Kawakami and Vazzoler (1980) for food items consumed by *Brycon falcatus*, Teles Pires River basin, sampled in the dry season of 2012 and 2013

	Cristalino River n = 36			Tapaiuna River n = 10			Verde River n = 10			Celeste River n = 14			Teles Pires River n = 27		
Items	F	RV	IA_i	F	RV	IA_i	F	RV	IA_i	F	RV	IA_i	F	RV	IA_i
Bivalves	2.86	1.71	0.14	–	–	–	–	–	–	–	–	–	–	–	–
Order Decapoda	20.00	14.86	8.48	10.00	5.45	1.39	–	–	–	–	–	–	–	–	–
Order Orthoptera	–	–	–	10.00	3.41	0.87	–	–	–	–	–	–	–	–	–
Order Coleoptera	–	–	–	10.00	3.64	0.92	10.00	1.25	0.38	–	–	–	–	–	–
Order Lepidoptera	–	–	–	10.00	0.91	0.23	–	–	–	–	–	—	–	–	–
Order Hymenoptera	8.57	0.24	0.02	10.00	3.41	0.87	10.00	0.38	0.11	21.43	0.72	0.36	3.70	1.37	0.09
Order Hemiptera	2.86	0.14	0.01	–	–	–	20.00	1.25	0.38	–	–	–	–	–	–
Class Aranae	–	–	–	10.00	3.64	0.92	–	–	–	–	–	–	–	–	–
Vertebrates	2.86	1.07	0.09	–	–	–	–	–	–	–	–	–	–	–	–
Fish	22.86	9.67	6.31	–	–	–	–	–	–	7.14	2.32	0.39	–	–	–
Scales	8.57	0.30	0.07	40.00	3.86	3.93	10.00	0.63	0.19	7.14	0.52	0.09	14.81	1.44	0.37
Leaves	34.29	9.74	9.53	10.00	1.36	0.35	30.00	3.06	2.81	7.14	0.15	0.03	14.81	1.36	0.35
Flowers	5.71	3.57	0.58	20.00	5.45	2.77	20.00	8.13	4.98	–	–	–	11.11	1.00	0.19
Seeds	17.14	6.00	2.93	20.00	8.18	4.16	40.00	34.38	42.12	21.43	0.70	0.35	7.41	0.63	0.08
Fruits	5.71	7.09	1.15	40.00	26.59	27.05	–	–	–	14.29	1.03	0.34	–	–	–
Corn	–	–	–	–	–	–	-	–	–	35.71	25.75	21.47	25.93	14.06	6.26
Soybean	–	–	–	–	–	–	20.00	7.81	4.79	50.00	53.66	62.65	74.07	69.36	88.25
Plant fragments	14.29	7.03	2.86	–	–	–	10.00	6.25	1.91	28.57	2.53	1.69	7.41	1.06	0.13
Detritus	8.57	8.71	2.13	10.00	2.73	0.69	20.00	4.69	2.87	–	–	–	3.70	0.14	0.01
Other	77.14	29.86	65.69	70.00	31.36	55.84	40.00	32.19	39.44	42.86	12.63	12.64	25.93	9.59	4.27

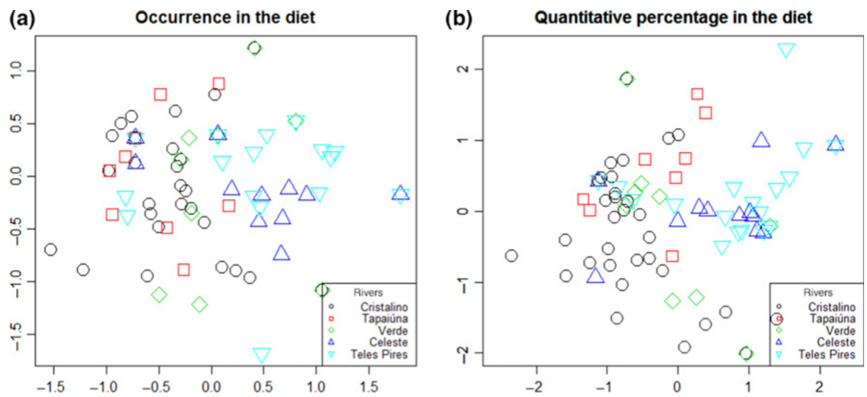

Fig. 2. NMDS analysis of (a) item occurrence in *Brycon falcatus* diet, Teles Pires River basin, sampled in dry season, 2012 and 2013; (b) NMDS analysis of the quantitative percentage of each item in the diet of *Brycon falcatus*, Teles Pires River basin, dry season, 2012 and 2013

tendency to frugivory. However, these items are not part of the natural diet of this fish, as they are supplied as an attractant (or food supplement for capture).

In the Teles Pires River, the *B. falcatus* diet consisted of 11 items (Table 2). Most important were fresh soybeans (IA_i = 88.25), followed by corn (IA_i = 6.26). The diet also included minor items, such as leaves, seeds, flowers, scales and insects of the order Hymenoptera. Based on the composition and dietary importance of these items, the diet of *B. falcatus* in the Teles Pires River was similar to that of the Celeste River, i.e. omnivorous with a tendency to frugivory. However, fruits with greater importance in the diet were those given as a food supplements for capture and are not native to riparian forests of the Teles Pires River.

Data regarding the occurrence of items in the diet of *B. falcatus* in each river were subjected to an NMDS analysis (Fig. 2a). This analysis indicated a significant difference in the diet between rivers, as shown by the clustering of points in each river, explaining 58.11% of the variance in our model. In addition, MANOVA statistical analysis showed that the diets differed from each other ($P < 0.001$). The percentage of each item in the diet of *B. falcatus* in each river of the Teles Pires basin was also subjected to NMDS analysis (Fig. 2b), which also showed a significant difference ($P < 0.001$) between diets when considering the clustering of points in each river and explained 53.95% of the variance in our model. Moreover, the diets were also significantly different according to the MANOVA statistical analysis.

The percentage of abdominal fat was significantly different between *B. falcatus* in the Teles Pires River and the Cristalino River, but did not differ significantly in the other rivers ($p > 0.05$) (Fig. 3).

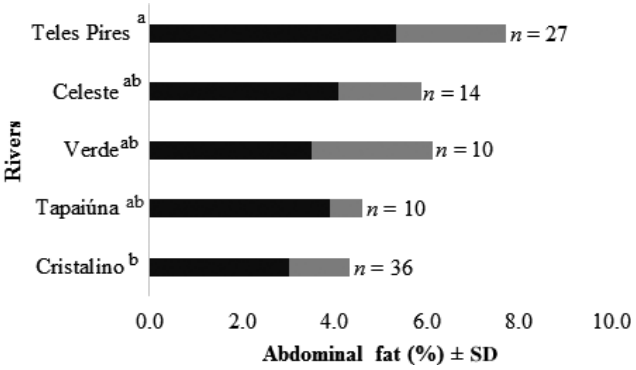

Fig. 3. Abdominal fat percentage relative to wet gross weight, *Brycon falcatus*, Teles Pires River basin, sampled in dry season, 2012 and 2013. Black bars = percentage of abdominal fat; grey bars = standard deviation of abdominal fat. CV = 47.08%. SD, standard deviation. Rivers followed by the same letter did not differ significantly (P>0.05) by ANOVA

Discussion

Because of its natural diet *B. falcatus* can be described as omnivorous with a tendency towards frugivory (Borges, 1986; Godoi, 2004; Blanco-Parra and Bejarano-Rodríguez, 2006; Albrecht et al., 2009). However, the present study showed that the fish diet was altered in rivers where attractants were used, whereby the composition and diversity of items changed with an increasing usage of attractants. The natural fish diet in the control river was the most diverse, with this diversity of items decreasing in those rivers where attractants are used. This change was also observed in a study performed in streams in southeastern Brazil, in which the diet of the benthivorous species *Imparfinis mirini* exhibited a lower diversity of items in streams with a high degree of urbanization (Tófoli et al., 2013). The human impact on food consumed by fish was also analyzed in a stream in southeastern Brazil, indicating a change in the diversity of items in the fish assemblage (Oliveira and Bennemann, 2005). In the Teles Pires River basin, using *B. falcatus* as an indicator, this type of anthropogenic impact comes from the continuous supply of soybeans as attractants by fishermen.

In addition to the change in the diversity of items, there were also changes in the importance of autochthonous and allochthonous items in the *B. falcatus* diet. The natural diet of *Brycon* features abundant use of allochthonous foods, demonstrating the importance of riparian forests (Borges, 1986; Godoi, 2004; Blanco-Parra and Bejarano-Rodríguez, 2006; Gomiero et al., 2008; Albrecht et al., 2009; Reys et al., 2009; Botero-Botero and Ramírez-Castro, 2011). However, in the Celeste and Teles Pires rivers the most important allochthonous items in the diet were fresh soybeans and corn. The ability to capture food is determined by behavioral traits. When an animal no longer needs to actively hunt for its food, it becomes less efficient (Orams, 2002); food supplied by humans is an easy option, requiring little effort. If this supply is frequent, foraging skills are gradually lost and the dependence on supplied feed increases (Orams, 2002). This supply by fishermen in the Teles Pires River basin is generating a 'dietary comfort zone' for *Brycon falcatus* around the attractant areas. This behavior was also observed by Sabino et al. (2005) in the bonito, where *B. hilarii* became accustomed to food supplied by tourists and as a consequence, stopped migrating and consequently developed large amounts of abdominal fat. In Australia, a study of the bird *Anthochaera carunculata* suggested that the additional food supply causes animals to remain year-round in places normally abandoned during seasonal migration (Paton et al., 1983). To confirm whether the same phenomenon might occur with *B. falcatus*, it will be necessary to mark individuals conditioned to this '*dietary comfort zone*' and monitor whether they remain in these areas during the migration season.

The balance between energy and protein may become deficient if the diet is unbalanced, potentially enhancing body fat accumulation (Lovell, 1991). With the excess consumption of protein by *B. falcatus*, only part of that protein is used for tissue formation and growth; the remainder will be converted into energy or accumulate as body fat (Bromley, 1980; Millward, 1989). The accumulation of abdominal fat may be associated with the reproductive cycle (Vazzoler, 1996) or with an unbalanced diet. Based on unpublished data on the reproductive biology of *B. falcatus* and the comparison of specimens collected in Teles Pires and Cristalino rivers, a macroscopic analysis of the gonads showed that approximately 80% of the samples collected from the Cristalino River and 60% collected in the Teles Pires River were undergoing gonadal development whereas approx. 30% of the specimens in Teles Pires River were undergoing gonadal regression. A higher accumulation of abdominal fat due to the reproductive stage is expected in specimens from the Cristalino River; however, we observed that specimens collected from the Teles Pires River exhibited a greater fat accumulation. Thus, the abdominal fat accumulation in our fish is not due to the reproductive cycle but likely due to the high crude protein supply in the fresh soybean diet, supporting an increase in abdominal fat.

The introduction of fresh soybeans in the *B. falcatus* diet has other implications, such as the contamination of fish by pesticides used in soybean cultivation. There may also be a lower genetic variability in fish reproducing near the attractant, which requires further investigation. An unbalanced diet can compromise the immune system, possibly increasing susceptibility to parasitic infestation. Despite the legal ban of attractants, there is no enforcement. This study shows some of the effects of attractants; the monitoring of these river areas should continue to help promote adequate conservation measures for *B. falcatus*.

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Author's address: Liliane Stedile de Matos, Universidade Federal de Mato Grosso-UFMT, Campus Universitário de Sinop, Av. Alexandre Ferronato, 1200, St Industrial, CEP 78557-267. Sinop, MT, Brazil.
E-mail: lilistedile@hotmail.com