Body condition and energy density of juvenile streaked prochilod
*Prochilodus lineatus* (Valenciennes, 1837) in a Neotropical floodplain

Daiany de Fátima Corbetta¹, Ana Lúcia Veronezzi² and Evanilde Benedito³

Variations in energetic density (*E*D) and the relative condition factor (*K*n) of juveniles of *Prochilodus lineatus* were investigated to identify whether these two variables respond similarly to environmental factors. We hypothesized that fluviometric levels in different sub-basins of the Upper Paraná River floodplain positively influence the *E*D and *K*n of juvenile fish. Temporally, the values of *E*D and *K*n were linked more directly to the dynamics of the flood pulses on the plain. Spatially, the lowest values of *E*D and *K*n were observed in the environment that was directly affected by the operation of dams, the sub-basin Paraná. Although the energy density and condition factor did not show similar results for juveniles in some of the analyses, the evaluation of both parameters provided a complementary tool and additional information that enabled a more accurate investigation of the temporal and spatial dynamic processes in this Neotropical floodplain. We conclude that water level variations in different sub-basins of the Upper Paraná River floodplain considerably affect the relative condition factor and energy status of *P. lineatus*. We suggest that the impacts of this modification should be mitigated or avoided in order to maintain fish stocks and promote ecosystem integrity.

**Keywords:** Condition Factor, Food web, Habitat, Impact, Paraná basin.

**Introduction**

Variations in water levels are the driving force behind the functioning and productivity of floodplains (Junk, 1989). Studies conducted in the Upper Paraná River floodplain have highlighted the key role of flood pulses in the interpretation of local processes (Thomaz et al., 2007; Agostinho et al., 2008; Leandrini et al., 2008; Roberto et al., 2009). The Paraná River basin is one of the largest watersheds in the world controlled by dams that affect floodplain functioning (Petere Júnior, 1996; Neiff, 2001; Agostinho et al., 2008). The dams result in diversity loss and reduced catches of commercial fish, which are becoming serious issues for the locals. Therefore, it is important to obtain bio-ecological information to achieve the best management program for the Paraná River (Agostinho et al., 2008).

Although the studied area in this floodplain is the last dam-free stretch, it is under the influence of impoundments along the main channel of the basin. It also provides an important habitat for several commercial fishes, such as *Pseudoplatystoma corruscans* (Spix & Agassiz, 1829), *Hemisorubim platyrhynchos* (Valenciennes, 1840) and *Salminus brasiliensis* (Cuvier, 1816) including

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Energy density of juvenile streaked prochilod

*Prochilodus lineatus* (Valenciennes, 1837) (Oliveira et al., 2015), also known as curimbatá, sábaló, streaked prochilod or locally as the curímba (Bayo & Yuan, 1996; Graça & Pavanelly, 2007; Froese & Pauly, 2015). *Prochilodus lineatus* is one of the most abundant and widely distributed fish in the various river basins of South America (Godoy, 1975; Agostinho et al., 1993; Silivasundar et al., 2001; Agostinho et al., 2003; Hahn et al., 2004; Espinola et al., 2010). In the Upper Paraná River floodplain, *P. lineatus* is important for recreational, commercial and subsistence fishing (Agostinho et al., 2003). *Prochilodus lineatus* is dentrivorous with a diet largely consisting of sediment and detritus (Bayo & Cordioli de Yuan, 1996; Hahn et al., 1997) and is an important prey for piscivores (Luz-Agostinho et al., 2008; Bozza & Hahn, 2010).

Due to its contribution to the natural food chain, *P. lineatus* plays an important role in energy flow and nutrient cycling in ecosystem and community dynamics. Therefore, condition factor and the energy density of fish have been used as indicators of their physiological status (Dourado & Benedito-Cecilio, 2005; Monteiro et al., 2007). The energy density allows the determination of whether consumed energy is allocated for growth, metabolic demands, waste loss and reproductive needs (Adams et al., 1982; Cui & Wootten, 1988; Adams & Breck, 1990). The condition factor quantifies the welfare of the fish, which can be indirectly related to stores of lipids and energy and allows a comparison of the condition of different specimens from the same sample, independent of length (Le Cren, 1951; Herbinger & Friars, 1991; Santos et al., 2004; Froese, 2006).

In ecology, knowledge of the variations of energy flows has been recognized as essential in understanding the organization of ecosystems (Lindeman, 1942), and thus, energy accumulation and allocation patterns are critical for the ecological success of organisms (Saldanã & Venables, 1983). The comprehension of energy density in Neotropical fish is one of the main issues in ecosystem ecology, especially when involving juveniles in natural environments. In this study, we investigated the variations of energy density and condition factor in *P. lineatus*. We hypothesized that fluviometric levels in different sub-basins of the Upper Paraná River floodplain positively influence the condition factor and energy density of juveniles of this species of dentrivorous fish. High water levels are predicted to contribute to a higher energy density and condition factor due to an allochthonous input into floodplain ecosystems, and this will be reflected in a correlation between the condition factor and energy density.

**Material and Methods**

The study area included a stretch of the Upper Paraná River floodplain located between Porto Primavera Dam and the beginning of the Itaipu Reservoir. This is the last long, undammed stretch of the Paraná River (approximately 200 km) (Thomaz et al., 2004). The floodplain of the Upper Paraná River is influenced by reservoirs and other projects located along the Paraná River, which is the main channel of the basin, and is joined by several tributaries, especially the Baia and Ivinhema. The plain also contains floodplain lakes, which play a vital role as spawning grounds and nurseries for migratory species. Due to impoundments, these environments have disappeared or were negatively influenced by the channel of the Paraná River.

**Sampling.** Samplings were performed quarterly from June 2010 to March 2011. The Paraná River floodplain consists of three sub-basins: the Ivinhema sub-basin (22°47’59.64”S; 53°32’21.30”W), which has no dams; the Paraná sub-basin (22°45’39.96”S; 53°15’7.44”W), which has a series of dams on its main channel, the Paraná River; and the Baia sub-basin (22°43’23.16”S; 53°17’25.50”W), which is affected by the Paraná River. Three to five sites were sampled at each sub-basin (Fig. 1).

*Prochilodus lineatus* were sampled using gill nets with different mesh sizes (from 2.4 to 8.0 cm). Gills nets were exposed for 24 h and were inspected every 8 h. All 272 collected specimens were anesthetized and killed (with Eugenol). Specimens of *P. lineatus* collected from the floodplain of the Upper Paraná River were deposited in the Museu de Peixes of NUPELIA (Núcleo de Pesquisas em Limnologia, Ictiologia e Aquicultura, Universidade Estadual de Maringá) (NUP 11092 - http://peixe.nupelia.uem.br/). The following biometric characteristics of the collected specimens were measured: standard length ($L_s$, cm), total weight ($W_t$, g), sex, and stage of gonadal development according to the criteria proposed and adapted by Vazzoler (1996). Only immature specimens were considered in this study to avoid the effects of the gonadal developmental stage on the energetic density and condition factors (Dourado & Benedito-Cecilio, 2005). According to Barbieri et al. (2000) and Suzuki et al. (2004), the male length were between 21.3 to 24.9 cm and females were between 24.0 to 25.0 cm.

Muscle samples were obtained from the dorsal region to above the lateral line of each specimen. Collected samples were washed with distilled water and wrapped in aluminum foil, labeled and frozen. Subsequently, samples were dried in a forced-air oven (60 °C, 48 h) and were ground to a fine powder with a ball mill.

The energy density ($E_{d,3}$) of these samples (measured as kilocalories per gram dry weight; kcal/g DW) was determined using an adiabatic bomb calorimeter (Parr 1261) at the Energy Ecology Laboratory of NUPELIA.

Spatial and temporal means of the limnological variables used in this study were obtained from the Long-Term Ecological Research/Brazilian National Council for Development (PELD/CNPq), the Laboratory of Basic Limnology of NUPELIA, at the same sampling times as those of the fish (Table 1).
Table 1. Mean values ± SD of limnological variables for sub-basins and months sampled quarterly from June (Jun) 2010 to March (Mar) 2011: Ivi = Ivinhema, Bai = Baia, Par = Paraná, Alk = alkalinity (mEq/L), Chl = chlorophyll (µg/L), Con = conductivity (µS/cm), DO = dissolved oxygen (mg/L), TSM = total suspended matter (µg/L), OSM = organic suspended matter (mg/L), MSM = mineral suspended matter (mg/L), NH4 = ammonia (µg/L), NO3 = nitrate (µg/L), TP = total phosphorus (µg/L), Tem = temperature (ºC), Turbidity = Tub (NTU). Different letters indicate significant differences (p < 0.05).

<table>
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<th>Months</th>
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<td>Jun</td>
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<td>169±103</td>
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</tr>
<tr>
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Study Design. Daily water levels were obtained from the Advanced Field Station of NUPELIA, in the municipality of Porto Rico, Paraná State. During samplings undertaken in 2010, the water level of the Paraná River was below 3.5 m, above which is considered typical flooding conditions (Agostinho et al., 2004b; Thomaz et al., 2004). However, the water level rose above 3.5 m during the final sampling in March of 2011. Therefore, this final sampling was regarded as the beginning of flooding, while the others are considered from drought periods. It is noteworthy that the values of energy density and condition factors are the result of food resources obtained before the period in which the specimens were sampled. Thus, the results were considered in the analyses from the previous fluviometric data samplings (Fig. 2).

Data analysis. No significant differences were detected between the sexes, and therefore the data from all analyses were pooled. The length–weight relationship was estimated for pooled immature males and females, using the expression \( W_t = a \times L_s^b \), where \( W_t \) = total weight, \( L_s \) = standard length, \( a \) = intercept of the regression and \( b \) = slope (Le Cren, 1951; Froese, 2006). The parameters \( a \) and \( b \) were high after logarithmic transformation of the data for weight and length and were, therefore, subjected to a subsequent adjustment of a straight line to the points for the Least-Squares Method (Vanzolini, 1993). Subsequently, the relative condition factor \( K_r \) was calculated as the ratio between the observed and expected weight for a given standard length \( K_r = W_t/P_e \); \( P_e \) = calculated weight derived from length-weight relationship (Le Cren, 1951).

To analyze temporal and spatial variations in the energy density \( E_d \) and relative condition factor \( K_r \), the sampled fish were grouped into two classes of standard length: Class 1 consisted of sizes between 13 and 16.4 cm and Class 2, between 16.5 and 20 cm. The range of \( L_s \) was 13.0 to 20.0 cm. The samples were grouped into classes to avoid interference of somatic growth on the analysis of the \( K_r \) (Garcia-Berthou, 2001) and \( E_d \) (Silva et al., 1999; Espinola et al., 2010). Significant differences in \( E_d \) values were calculated among the classes \( (H = 20.8, d.f. = 1, P < 0.0001) \), with the energy density of the fish increasing with fish length.

The assumptions of normality and homoscedasticity were not met; therefore, a nonparametric one-way analysis of variance (Kruskal-Wallis) was performed for each class of standard length, considering the values of \( E_d \) and \( K_r \) to identify significant differences in these
values between months and the sub-basins sampled. Each site was considered as a replicate of a sub-basin (Block et al., 2001). The significance level was set at α = 0.05. P values were subjected to Bonferroni correction after several statistical tests were performed simultaneously on a single data set (Zar, 1999).

Temporal and site variations on the limnological variables were analyzed through a one-way ANOVA without replication after log (x+1) transformation, followed by Tukey’s test (Sokal & Rohlf, 1995). The level of significance adopted was 0.05.

For limnological variables, a Principal Component Analysis (PCA) was performed to characterize the sampling months and sub-basins. We used log (x+1) transformation for the standardization of values in the PCA matrix, and significant axes for interpretation were selected using the broken-stick criterion. The scores of the retained axes were correlated (Spearman rank test) with the mean energy density and the mean relative condition factor for each sampling month and sub-basin. To measure how environmental variables influence the energy density and the condition factor, a multiple regression analysis between the original variable values was carried out in the selected Axis 1 of PCA for the medium energy density and condition factor.

The Spearman rank test \( r_s \) (Zar, 1999) was used to identify a potential correlation between the relative condition factor \( (K_n) \) and energy density \( (E_D) \), and between these parameters and standard length and total weight.

**Results**

Significant differences were not observed between the sexes of the 272 immature individuals of *P. lineatus* sampled in this study; therefore, the results presented are grouped for sex. The weight-length relationship with the pooled data of the immature *P. lineatus* males and females was \( a = 0.05 \) and \( b = 2.81 \). The equation of the fitted curve was \( \log W_t = -1.31 + 2.81 \log L_s \) \( (P < 0.01, R^2 = 0.86) \) (Fig. 3).

The quarterly distribution of mean values of \( E_D \) and \( K_n \), grouped into size classes, showed significant differences only for \( E_D \) and \( K_n \) in Classes 2 [Fig. 3; (a): \( H = 7.18, \text{d.f.} = 2, P = 0.028; \) (b): \( H = 39.41, \text{d.f.} = 3, P < 0.01; \) (c): \( H = 1.56, \text{d.f.} = 2, P = 0.48; \) (d): \( H = 17.12, \text{d.f.} = 3, P < 0.01\)]. Thus, in the second class size, energy density values were higher in June than in September and December, whereas for the relative condition factor, smaller sizes were observed in March than in June and September (Fig. 4).

For the sub-basins, a significant difference was observed only in Class 1 for \( K_n \) [Fig. 4; (a): \( H = 12.66, \text{d.f.} = 2, P < 0.01; \) (b): \( H = 12.15, \text{d.f.} = 2, P < 0.01; \) (c): \( H = 3.25, \text{d.f.} = 2, P = 0.20; \) (d): \( H = 19.25, \text{d.f.} = 2, P < 0.01\)]. The highest values of \( E_D \) and Kn were observed in the Ivinhema sub-basin, and the Paraná sub-basin showed the lowest values (Fig. 5).

![Fig. 3. Linear regression (a) and weight-length relationship (b) of juvenile *Prochilodus lineatus* in the floodplain of the Upper Paraná River.](image-url)
(F = 258.19, P < 0.001), dissolved oxygen (F = 10.82, P < 0.001), organic suspended matter (F = 3.29, P = 0.031), ammonia (F = 4.72, P = 0.007) and phosphate (F = 7.05, P < 0.001) (Table 1). The first three PCA axes retained by the broken-stick criterion accounted for 66% of the total variance. In the first PC1 axis (% total variance = 29.8%), the highest values were observed for conductivity, suspended organic matter, total suspended matter, and chlorophyll (eigenvector coefficients = -0.38, -0.35, -0.34 and 0.36, respectively). For PC2 (% total variance = 21.2%), higher values were identified for dissolved oxygen and temperature (-0.42 and 0.36 respectively), whereas for PC3 (% total variance = 14.1%), the highest values were observed for ammonia, total phosphorus and dissolved oxygen (-0.43, -0.41 and 0.08, respectively).

Fig. 4. Median values of energy density (E\textsubscript{D}) [(a), (b)] and the relative condition factor (K\textsubscript{n}) [(c), (d)], by standard length, in the floodplain of the Upper Paraná River, for each month sampled. Lower bars indicate the 25% percentile, upper bars indicate the 75% percentile. Different upper-case letters (ABC) indicate statistically significant differences (Kruskal-Wallis, P < 0.025).

A significant correlation for period (months) was not verified for the PCA axis. For sub-basins, a significant correlation was observed only for condition factor (K\textsubscript{n}), where it was related with the limnological variables from axis 3 in the Paraná sub-basin (rs = -0.82, P < 0.05) and with those from axis 1 in the Baia sub-basin (rs = 0.53, P < 0.05).

The multiple regression analyses for E\textsubscript{D} (adjusted R\textsuperscript{2} = -0.052, P = 0.75) and K\textsubscript{n} (adjusted R\textsuperscript{2} = 0.028, P = 0.28), using the variable axis 1, indicated that the regression model obtained was not significant (R\textsuperscript{2} adjusted = 0.871, P = 0.055).

A significant correlation was found between energy density (E\textsubscript{D}) and relative condition factor (K\textsubscript{n}) (r\textsubscript{s} = 0.24, P < 0.05), standard length (r\textsubscript{s} = 0.21, P < 0.05) and total weight (0.29, P < 0.05) (Fig. 6).
Fig. 6. Spearman rank correlations (rs) between the values of energy density ($E_D$) versus relative condition factor ($K_n$) (a), length standard ($L_s$) (b) and weight total ($W_t$) (c) of *Prochilodus lineatus* in the floodplain of the Upper Paraná River. The line represents the trend of significant Spearman correlations.

Discussion

Energy density and condition factor have been used as tools to investigate the physiological status of fish in prevailing environmental conditions. On a temporal and spatial scale, the variation in energy density and relative condition factor were mainly related to the limnological effects. However, the energy density and physiological condition factor are parameters resulting from environmental conditions prior to sampling, which do not allow a direct interpretation. This is reflected in the absence of a seasonal correlation between environmental variables and energy density values or condition factor.

In periodically flooded environments, such as the river floodplain system of the Upper Paraná River, the input of allochthonous material into the water increases as the river level rises (Pagioro & Thomaz, 1999). When the flood recedes, there remains a greater availability of organic debris from the decomposition of flooded terrestrial vegetation transported by the flood and retained in the roots and other parts of macrophytes or from river bottom sediment (Resende *et al.*, 1996). Nevertheless, this process leads to an improvement in feeding conditions and available food sources for *P. lineatus* juveniles. However, the production of periphyton in the floodplains was reported as higher in receding waters and during drought periods (Taniguchi *et al.*, 2005; Algarte *et al.*, 2006; Carapurnarla *et al.*, 2014) or in the Winter (Camargo & Ferragut, 2014), which also favors the bioavailability for this species.

The gradual reduction mainly in the values of energy density during the first three samplings, in class 2, can be related to the lack of inputs to the water column, which consequently generates nutritionally and energetically poor sediment and detritus and results in lower values of organic and total suspended matter found in December. The ongoing degradation of materials deposited from a previous flood gradually restricts the dietary conditions of the species that feed on detritus (Bowen, 1983; Chimney & Pietro, 2006).

The highest values of ammonia, total phosphorus and turbidity were also recorded in December. According to Abujanra *et al.* (2009), the highest nutrient availability in the water column in the Upper Paraná River floodplain was in low water. In the December 2010 sampling of our study, the water level remained low and the available nutrients may have resulted from the prolonged process of decomposition and release from the same sediment (Lee & Bukaveckas, 2002; Yahdjian *et al.*, 2006; Schönbrunner *et al.*, 2012). Thus, local processes such as the resuspension of bottom sediments caused by the wind (Evans, 1994; Rodrigues *et al.*, 2002) and disturbance from fish action, may contribute to the increase in nutrients in the water column (Breukelaar *et al.*, 1994).

The availability of periphyton was unclear during the sampling periods, and despite the availability of nutrients,
Energy density of juvenile streaked prochilod

turbidity might have influenced periphyton biomass due to a lower penetration of radiation into the water column in December. Camargo & Ferragut (2014) found that periphyton biomass and algal density in a shallow tropical reservoir were negatively correlated with macrophyte cover and phytoplankton chlorophyll-a, where the availability of nutrients in the water appeared to be less important to community organization. Therefore, studies on the association between detritivorous populations and the periphyton community are necessary.

Although an improved body condition of juveniles during the flood period was observed by Gomes & Agostinho (1997), this was not observed in the present study in which a non-significant increase in energy density values was observed in March. According to Bevelhimer & Breck (2009), fish energy content depends on the energy values of the component tissues, and fish in better condition generally have higher lipid levels, which translates into higher energy density. An improvement in the energetic condition of the food availability may have allowed more energy to be released and directed towards metabolism and growth, resulting in an increase in muscle-energy density. Low condition factor values observed during this period might be due to several factors, such as the sampling period occurring at the beginning of the flood period when the food conditions were not sufficiently improved to permit energy reserves for weight increases. It is also possible that the nutritional improvement of the food had not yet led to a weight gain by the beginning of the flood period. Thus, the condition factor values appear to be more resilient to fluctuations in environmental conditions than the energy values. Sacramento et al. (2016), who studied juveniles of *P. lineatus*, confirmed that the turnover of diets in tissues of this species is longer than 100 days. Therefore, our results require confirmation by additional studies that include sampling flood periods, because the deficiencies in sampling during a particular period might have limited the observation of this pattern.

The graphical depiction of the seasonal analysis revealed different patterns for condition factor values and energy density. However, the energy density variation patterns and condition factor were similar by spatial analysis. Spatially, the variation in energy density and condition factor were related to the characteristics of each sub-basin and the degree of conservation. The change caused by damming might explain the significant high values of muscle energy density and condition factor in Ivinhema and the significantly lower values in the Paraná sub-basin. Before the formation of the Porto Primavera Reservoir in the main channel of the Paraná River, the body condition of fish found in the Paraná, Baia and Ivinhema sub-basins varied similarly. However, there was a reduction in the condition factor of fish after completion of this impoundment, especially in the Paraná sub-basin, which was directly affected by the reservoir (Abujanra et al., 2009; Roberto et al., 2009).

Furthermore, the impoundment caused a decrease in the diversity of organisms (Agostinho et al., 2004a; Karling et al., 2013) and a reduction in migratory fish stocks that are important to the fisheries industry (Agostinho et al., 2003; Agostinho et al., 2007), including *P. lineatus* (Schork et al., 2012). The study area has been impacted by a cascade of upstream reservoirs, which promote the retention of nutrients and sediments (Gimenes et al., 2004; Agostinho et al., 2008) and directly affect the feeding grounds of *P. lineatus*. This results in the lowest values of chlorophyll, turbidity, and total phosphorus, and the highest values of conductivity, organic suspended matter, total suspended matter and nitrate in the Paraná sub-basin. Similar values have been confirmed for chlorophyll, turbidity, and total phosphorus by Roberto et al. (2009) after 20 years of studying the main channel of the Paraná River. The highest values of conductivity, organic suspended matter, total suspended matter and nitrate observed can result from anthropogenic input from land use including agriculture and cattle farming, in addition to urban runoff. The availability of food resources, as well as their energy and condition factor for the fish fauna, are directly related to the dynamics of the sub-basins.

Fish energy density can have a large effect on growth rate, survival and reproduction. For the same level of consumption, fish in good condition had a greater rate of growth in length than fish in poor condition (Bevelhimer & Breck, 2009). Fish in poor condition allocate more of their new tissue to improving conditions and consequently grow less in length than fish in good condition (Bajer & Hayward, 2006). Therefore, the absence of impoundments influencing the Ivinhema sub-basin within the “Várzeas do Rio Ivinhema” State Park (MS) protects part of the basin from human impact and contributes to a better condition of this species fish, which was reflected by the observed values of energy density and condition factor, contributing to the integrity of the sub-basin.

The low correlation between the condition factor and energy density is explained by the fact that the increase in weight is dependent on a sufficient amount of intake and time energy allocation to reflect in tissue and energy gain. This balance results in a significant increase in condition factor, and thus the increase in energy density may not reflect in improved body condition through the condition factor (Enrich-Prast & Esteves, 2005; Espinola et al., 2012). The parameters of condition factor and energy density present different patterns of variation, suggesting that their values could indicate different physiological conditions. A significant correlation between these two parameters was not observed in the analysis of other species (Santos et al., 2006; Garcia & Benedito, 2010), suggesting the importance of jointly verifying the results from these two parameters, as complementary tools. Although both parameters are directly related to food, they do not always respond similarly to environmental
variations. Further, the low correlation found between energy density and fish length or weight may have been a reflection of the small size range of individuals. Smaller fish have a lower energy density compared to older and larger fish (Bevelhimer & Breck, 2009).

Therefore, although the analysis of juvenile fish minimizes the effect of reproducing the energy density in the muscle, the use of size classes is suitable for this parameter, because there is a variation in energy during growth. Although the energy density and the condition factor were not similar for juveniles in some of the analyses, the results from including both parameters provides additional information, enabling a more accurate investigation of the dynamic temporal and spatial processes in Neotropical floodplains, and human interference on the populations of fishes. Even though the specimens from the Paraná sub-basin showed the lowest values of Kn and ED (promoted from the reservoir cascade), the increase in fluviometric levels in different sub-basins of the Upper Paraná River floodplain positively influenced the body condition and energy density of juveniles belonging to detritivorous fish species. This is due to the allochthonous input into the floodplain ecosystem. Because this effect is not immediate and is posteriorly reflected, new studies are required to identify the turnover ratio of energy in tissue and growth of organisms in nature. We suggest that the impacts of this modification should be mitigated or avoided in order to maintain fish stocks and promote ecosystem integrity.

Acknowledgements

The authors thank Cnpq/Capes/UEM and the Post-Graduate Course in Biologia Comparada (PGB-UEM) for financial and logistic support. We also thank all our colleagues for help in the laboratory, and Jaime L. Pereira for preparing the map.

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