Plasticity in the shape and growth pattern of *asteriscus* otolith of black prochilodus *Prochilodus nigricans* (Teleostei: Characiformes: Prochilodontidae) freshwater Neotropical migratory fish

Rosa Maria Rodrigues da Costa¹, Nidia Noemí Fabré², Sidinéia Aparecida Amadio¹ and Víctor Manuel Tuset³

Using morphometric measurements and wavelets functions, the *asteriscus* otoliths of curimatã, *Prochilodus nigricans* were analysed to identify the variation in shape and growth increment of individuals from Solimões, Japurá and Negro rivers of the Amazon basin, Brazil. The morphometric and morphological analyses did not reveal evidences of population segregation among rivers, but variations were found in the estimation of otolith growth increment. Also, the otolith shape showed a high variability between individuals, identifying four morphotypes. Morphotype 1 shows a more oval shape with a posterior zone clearly rounded; Morphotype 2 shows the posterior zone rounded, but the anterior end is more elongated; Morphotype 3 shows a completely different shape, elliptic-pentagonal and Morphotype 4 shows the posterior zone rounded, but the anterior end is more elongated and it is the pattern with antirostrum and rostrum more pointed and a deep notch. Therefore, the otolith shape exhibited a phenotypic plasticity that it was not associated with the metabolism of otolith growth. Whereas the otolith shape indicated a homogeneity in the sound perception through Amazon basin, the growth rates revealed an adaptive mechanism to environmental conditions or migratory process of this species.

Key words: Amazon Basin, Growth increments, Morphotypes, Otolith shape, Phenotypes.

Introduction

Species distribution depends on the comprehensive knowledge of evolutionary history, dispersal ability and ecological aspects that determine the ability of the species to perpetuate in contemporary environmental, climatic or other conditions such as habitat preference, life history and behavior (McDowall, 2000). The phylogeography of freshwater fish species, widely distributed in large rivers, reflects the geomorphological processes that contributed to the current physical geography and hydrology (Bermingham, Martin, 1998; Sivasundar et al., 2001). The dispersion of fish depends on the direct connectivity between river basins as well as their interconnection history that reflects the underlying
Morphology of asteriscus otolith in Prochilodus nigricans

geology (Lundberg, 1993; Bermingham, Martin, 1998). In the Amazon, the development of a complex geological structure, along with the effect of global climate fluctuations in the Cenozoic, are the basic factors which, according to the refuge hypothesis, caused ecological vicariant populations of vertebrates (Jegu, Santos, 1993; Haffer, 2008). The Prochilodontidae family comprises one of the most important migratory fish groups of commercial value of South America (Goulding, 1981; Bayley, Petere Jr, 1989; Castro, Vari, 2004). This family includes 21 species divided into three genera (Ichthyoelephas, Prochilodus and Semaprochilodus), which occur mostly in the rivers of South America (Reis et al., 2003). The Prochilodontidae fishes migrate at the beginning of the raising water period to spawn, after which juveniles and adults disperse into the flooded areas by the muddy water rivers, rich in nutrients, to feed and grow. They leave the floodplains towards upriver at the onset of the receding water period to disperse in lakes or rivers of muddy or black water. The use of different environments during their life cycle can provide an interesting approach to evaluate the relative contribution and interaction of geomorphological processes due to environmental condition. In this sense, Sivasundar et al. (2001) applied nested clad analysis to relate recurrent gene flow and historical events that could shape the population structure of the four species of Prochilodus occurring in river basins of South America: Magdalena River bocachico Prochilodus magdalenae Stein-dachner, 1879, Orinoco River coporo Prochilodus mariae Eigenmann, 1922, Amazonas River curimatá Prochilodus nigricans Spix & Agassiz, 1829, and Paraná River corimbata Prochilodus lineatus (Valenciennes, 1837). They found a high genetic variability within populations from Paraná River and a clear separation between species, but no strong association among phylogenetic relationships, haplotypes and geographical distribution. In ostariophysi fishes, asterisci and lapilli are the most developed otoliths (Adams, 1940). The sacculus and lagena are involved in sound detection (Popper, 1980) and appear to have evolved in close association with the specialization of hearing (Popper, Coombs, 1982), whereas utriculus has the function to maintain balance (Mugiya,Takahashi, 1985; Lombarte, Popper, 2004). The sacculus otoliths certainly constitute an important tool for taxonomic and phylogenetic diagnosis for most teleost fishes (Nolf, 1993; Lombarte, Castellón, 1991; Stransky, MacLellan, 2005; Schulz-Mirbach, Reichenbacher, 2006; Tuset et al., 2008; Tuset et al., 2016b), as well as for the diagnosis of intraspecific variation (Tombari et al., 2011; Curin-Osorio et al., 2012) But moreover, the morphology fish otoliths from different geographical areas can express different stages of their life story and represents a phenotypic measure of the environmental conditions (Hlssen et al., 1981; Begg, Brown, 2000). Reichenbacher et al. (2009) showed that the saccula otolith morphology and morphometry contributed to the detection of genetic differentiation in isolated populations of Aphanius dispar (Rüppell, 1829), a freshwater species in southeastern Saudi Arabia. Regarding the same species, Teimori et al. (2012) showed that population from three basins of the Persian Gulf could be distinguished by six phenotypic characters based on fish morphometric and meristic measurements and otolith morphology and morphometry. Collins et al. (2013) evaluated the population structure of P. mariae from Apure River, a tributary of the Orinoco River, and identified breeding areas by microchemical analysis of the otoliths; however, the authors did not detect any genetic structure, indicating there is gene flow between the rivers which prevents population differentiation. In the same region, Pérez, Fabré (2013), using otoliths morphochemistry and skulls of Pseudoplatystoma metaense Buitrago-Suárez & Burr, 2007 differentiated nursing, breeding and feeding areas, within the tributaries of the Apure basin, but no segregation of population groups. This study emphasize the use of the asteriscus otolith to determine if the populations of Amazonas River curimatá P. nigricans distributed in three rivers of the Amazon basins (Reis et al., 2003) are connected. We selected this species due to socio-economic interest to be widely captured in the main rivers of the Amazon basin (Barthem, Fabré, 2004) for subsistence (Garcz et al., 2009) and commercial fishing (Batista, Petere Jr., 2003; Batista et al., 2012). Furthermore, this species performs trophic and reproductive migrations and dispersal movements according to the hydrological cycle of the rivers (Ribeiro, 1983; Barthem, Fabré, 2004), hence to establish the fishing units is very important for an adequate assessment and management of resource and for a better knowledge of connectivity of fish populations in larger rivers of the South America as the Amazon. For our proposal, we compared the otolith outline using wavelets functions (Parisí-Baradad et al., 2005) and the growth rate between growth annuli (Silva, Stewart, 2006).

Material and Methods

Sampling and data collection. In the years of 1996 and 1997 a total of 126 specimens of Amazonas River curimatá P. nigricans were captured in Solimões (n = 42), Japurá (n = 42) and Negro rivers (n = 42) (Fig. 1). Fish were measured in standard length (L, in mm), the head was cut off of the fish, the otoliths were removed, washed, dried and stored in labeled plastic vials.

The asteriscus otolith was used for this study because of its larger size and clear calcified deposits, which enabled the identification of growth rings (Pérez, Fabré, 2003; Silva, Stewart, 2006). The images of the left side of the asteriscus were taken using a digital video camera coupled to a microscope stereoscopic (Zeiss Magnifier CL1500 ECO) on a black background. Magnification of the microscope lens was adjusted to the size of the otolith using object-glass 1.6x. The rostrum of the otoliths was set on a horizontal line to

---

Neotropical Ichthyology, 16(4): e180051, 2018
minimize distortion errors in the normalization process of the images. A voucher specimen was deposited in the fish collection of the National Institute of Amazon Researches under the number INPA-ICT 039004.

**Otolith morphometry.** The following morphometric measurements were recorded from the asteriscus otolith using ImageJ software package (Schindelin et al., 2012): \(W_0\) (otolith maximum width in mm), \(L_0\) (otolith maximum length in mm), \(L_r\) (rostrum length in mm) and \(A_o\) (otolith area in mm^2). From this measurements, three ecomorphological indexes were calculated (Volpodo, Echeverría, 2003): the \(E\) index \((E = A_o/L_0)\) expresses the tendency in the shape of the asteriscus (circular or elongate); the \(R\) index \((R = L_r/L_0)\) defines the percentage of the length to the rostrum; and the \(C_o\) \((C_o = \text{perimeter of the otolith}/(2\sqrt{\text{area} \times \pi}))\) displays the complexity of the otolith and refers to the measure of the contour.

**Otolith contour.** Otolith shape analysis was based on wavelet functions. A total of 512 equidistant cartesian coordinates of the otolith were extracted, being the rostrum the origin of the contour (see more details in Parisi-Baradad et al., 2005; Parisi-Baradad et al., 2010). Image processing was performed by the image analysis software Age & Shape (v. 1.0, Infaimon SLVR, Spain). Since our purpose was to determine otolith patterns, we used the level 4 of wavelet transformed function defining with enough details the otolith silhouette for the identification of populations or morphotypes (see Sadighzadeh et al., 2014; Abaad et al., 2016).

**Otolith growth increments.** Whole otoliths were immersed in KOH (5%) for an hour, for decalcification. They were submerged in 70% alcohol for observation under black background with reflected light, using an increase of 1X. The images were captured with a camera attached to a stereomicroscope Leica LAS EZ, v. 2.1.0 (Silva, Stewart, 2006). The distance (mm) between the otolith nucleus and each growth mark was measured using ImageJ Program. Independent otolith reading was carried out six times at different times, in order to confirm visualization and interpretation of growth marks.

**Statistical analysis.** To determine if the otolith morphometry and shape varied with the origin of individuals, two analysis were performed. Firstly, the mean values of \(I_e\) index, \(I_r\) index and \(C_o\) were compared for each site by one-way analysis (ANOVA), with Tukey HSD test for post hoc comparison. The assumption of normality and homogeneity for each variable was analysed by Kolmogorov-Smirnov with correlation of the Lilliefors and Levene’s tests (González et al., 2011).

Secondly, a principal component analysis (PCA) based on the variance-covariance matrix was performed to reduce the wavelet functions without the loss of information in the otolith shape analysis (Sadighzadeh et al., 2014). Only the PCA variables that explained more than 1% of the variability were used for further analyses (Hammer et al., 2001). To test if local differences might be attributed to allometry, Pearson’s correlations between otolith length and the PC’s were calculated (Burke et al., 2008; Maderbacher et al., 2008) and the effect of fish size was removed using the residuals of the common within-group slopes of the linear regressions of each component (Stransky, MacLellan, 2005). Thus, a new PCA matrix was built. A canonical variate analysis (CVA) was computed on the reduced PCA matrix to summarise the ecomorphological differences among populations.
the variation between localities maximising their distances (Linde et al., 2004). Finally, the identification of the different otolith patterns was established from distance values using a cluster analysis with unweighted pair group method of arithmetic averages (UPGMA) algorithm based on the Euclidean distance (Tuset et al., 2014). Finally, significant differences between the means of groups formed was tested using a multiple variance analysis (MANOVA) and the sequential Bonferroni correction for multiple comparisons (Rice, 1989). Finally, they were classified using a jackknife procedure on the classifier matrix, whereby a concordance between classification success rates in the non-validated and procedure on the classifier matrix, whereby a concordance between classification success rates in the non-validated and validated analyses (obtained from the confusion matrix) indicates that group discrimination was not based on a one-case contribution (Yazdi, Adriaens, 2013).

To establish if there were statistical differences in the growth increments of otoliths between the different sites were used an ANCOVA test. All statistical analyses were performed in Statistica version 7.1 (StatSoft Inc.) and PAST (Palaeontological Statistics, version 1.81) (Hammer et al., 2001). Significance level was set at 0.05 for all statistical tests used.

**Results**

An ANOVA test did not show significant differences (ANOVA, $F_{(2,123)} = 2.033; P = 0.135$) in the fish size between river populations. Therefore, fish length did not influence in the following analyses. The descriptive statistics of all variables are summarized in (Tab. 1).

**Tab. 1. Summary of descriptive statistics of fish size and otolith asteriscus of Prochilodus nigricans** from the rivers Solimões, Japurá and Negro, Amazon Basin, Brazil. $L_{F}$, fish length; $L_{O}$, otolith length; $W_{O}$, otolith width; $R$, rostrum length; $A_{O}$, otolith area; $E$, $I$, $R$, $O$, complexity of the otolith.

<table>
<thead>
<tr>
<th>Variables</th>
<th>Solimões (n = 42)</th>
<th>Japurá (n = 42)</th>
<th>Negro (n = 42)</th>
</tr>
</thead>
<tbody>
<tr>
<td>$L_{F}$ (mm)</td>
<td>Min-Max 210-355</td>
<td>205-351</td>
<td>224-344</td>
</tr>
<tr>
<td>Mean ± SD</td>
<td>277.20 ± 43.00</td>
<td>267.40 ± 33.30</td>
<td>261.70 ± 28.50</td>
</tr>
<tr>
<td>$L_{O}$ (mm)</td>
<td>Min-Max 5.11-7.5</td>
<td>5.12-7.74</td>
<td>5.16-7.48</td>
</tr>
<tr>
<td>Mean ± SD</td>
<td>6.31 ± 0.73</td>
<td>6.40 ± 0.75</td>
<td>6.18 ± 0.62</td>
</tr>
<tr>
<td>$W_{O}$ (mm)</td>
<td>Min-Max 3.71-6.41</td>
<td>4.00-5.56</td>
<td>3.94-5.82</td>
</tr>
<tr>
<td>Mean ± SD</td>
<td>4.64 ± 0.60</td>
<td>4.74 ± 0.50</td>
<td>4.66 ± 0.46</td>
</tr>
<tr>
<td>$R_{O}$ (mm)</td>
<td>Min-Max 0.56-2.73</td>
<td>0.90-2.46</td>
<td>0.70-2.16</td>
</tr>
<tr>
<td>Mean ± SD</td>
<td>1.62 ± 0.51</td>
<td>1.50 ± 0.39</td>
<td>1.40 ± 0.35</td>
</tr>
<tr>
<td>$A_{O}$ (mm)</td>
<td>Min-Max 13.53-33.41</td>
<td>14.46-30.24</td>
<td>15.09-29.01</td>
</tr>
<tr>
<td>Mean ± SD</td>
<td>20.52 ± 4.77</td>
<td>21.43 ± 4.70</td>
<td>20.06 ± 3.85</td>
</tr>
<tr>
<td>$E_{O}$</td>
<td>Min-Max 62.89-86.20</td>
<td>66.28-82.72</td>
<td>68.77-85.06</td>
</tr>
<tr>
<td>Mean ± SD</td>
<td>73.51 ± 4.60</td>
<td>74.13 ± 3.63</td>
<td>75.51 ± 4.60</td>
</tr>
<tr>
<td>$I_{E}$</td>
<td>Min-Max 9.60-40.11</td>
<td>15.46-35.38</td>
<td>12.45-36.86</td>
</tr>
<tr>
<td>Mean ± SD</td>
<td>25.75 ± 7.55</td>
<td>23.48 ± 5.69</td>
<td>22.68 ± 5.36</td>
</tr>
<tr>
<td>$I_{R}$</td>
<td>Min-Max 1.27-1.78</td>
<td>1.37-1.85</td>
<td>1.38-1.73</td>
</tr>
<tr>
<td>Mean ± SD</td>
<td>1.49 ± 0.10</td>
<td>1.51 ± 0.09</td>
<td>1.53 ± 0.09</td>
</tr>
</tbody>
</table>

**Otolith morphological analysis.** The format of a typical asteriscus is oval shape, with rounded edges. Rostrum and antirostrum are located on the anterior part of the otolith, clearly differentiated with an acute and depth notch between. The position of sulcus is medial, heterosulcoid and ostial, with the a cauda curvature at the end. The morphometrical analysis did not revealed significant differences between river populations for ecomorphological indices ($I_{E}$, $F_{(2,123)} = 2.37, P = 0.09$; $I_{R}$, $F_{(2,123)} = 2.69, P = 0.07$; and $O$, $F_{(2,123)} = 1.87; P = 0.15$). For the analysis of otolith contour, the first 5 PC components explained 91.6% of total variance, where the first component reached 54.6% and the second accounted for 23.1% of variability. The MANOVA analysis did not reveal significant difference among localities in the otolith shape (Wilk’s lambda = 0.89, $F_{(19,234)} = 1.35$, $P = 0.20$) (Fig. 2).

The cluster analysis allowed to identify four otolith morphotypes: (Fig. 3 and Fig. 4) respectively. Morphotype 1 (42% of the cases): shows a more oval shape with a posterior zone clearly rounded. The antirostrum and rostrum are small with a narrow excisura and shallow notch; Morphotype 2 (50%) shows the posterior zone rounded, but the anterior part is more elongated. The antirostrostrum is not pointed, but blunt, and the notch is deeper than morpho 1; Morphotype 3 (16%) shows a completely different shape, elliptic-pentagonal. Antirostrostrum and rostrum pointed increasing the depth of notch and; Morphotype 4 (15%) shows the posterior zone rounded, but the anterior zone is more elongated. The antirostrum and rostrum are more pointed with the deepest notch and the cauda lesser curved.

Significant differences were found among the morphotypes (MANOVA, Wilks lambda = 0.0112, $F_{(15,317)} = 86.66$,}
between double peak blunt-pointed from pointed-pointed in the anterior zone (Figs. 4-5). The Fig. 4 shows four otolith morphotypes: a. Morphotype 1; b. Morphotype 2; c. Morphotype 3 and d. Morphotype 4. Moreover, the maximum height was in the posterior zone in the positive axis, and in the middle of the otolith in the negative value. The classification success was tested reached up 98.0%, indicating a clear differentiation between them. Specimens of Solimões River showed two morphotypes in the similar proportion: 53.3% for morpho 3 and 46.6% for morpho 4. In the Japura River, three morphotypes were found: morpho 1 (35.7%), 2 (33.3%) and 4 (40.0%). And finally, in Negro River predominated the morphotype 1 (40.4%) and 3 (33.3%).

Otolith growth increments. The descriptive statistics of all average radius of the increments otoliths of *Prochilodus nigricans* from the rivers Solimões, Japurá and Negro are summarized in (Tab. 2). The ANCOVA test showed significant differences to otolith growth increments of \( F_{(2,553)} = 95.7, P<0.001 \) among rivers (Fig. 6). The interaction between the rivers and the increments of growth was not significant \( F_{(2,553)} = 1.89, P = 0.15 \).

Tab. 2. Summary of descriptive statistics of average radius of the increments otolith of *Prochilodus nigricans* from the rivers Solimões, Japurá and Negro, Amazon Basin, Brazil. \( R_1 \), first ring; \( R_2 \), second ring; \( R_3 \), third ring; \( R_4 \), fourth ring; \( R_5 \), fifth ring; \( R_6 \), sixth ring; \( R_7 \), seventh ring of asteriscus otolith. (n) number of individuals; (Mean ± SD, standard deviation) of increment otolith each river.

<table>
<thead>
<tr>
<th>Radius</th>
<th>Solimões Mean ± SD</th>
<th>Japurá Mean ± SD</th>
<th>Negro Mean ± SD</th>
</tr>
</thead>
<tbody>
<tr>
<td>( R_1 )</td>
<td>1.09 ± 0.10</td>
<td>1.09 ± 0.11</td>
<td>1.06 ± 0.09</td>
</tr>
<tr>
<td>( R_2 )</td>
<td>1.66 ± 0.12</td>
<td>1.64 ± 0.10</td>
<td>1.56 ± 0.09</td>
</tr>
<tr>
<td>( R_3 )</td>
<td>2.08 ± 0.12</td>
<td>2.06 ± 0.14</td>
<td>1.97 ± 0.13</td>
</tr>
<tr>
<td>( R_4 )</td>
<td>2.46 ± 0.11</td>
<td>2.41 ± 0.17</td>
<td>2.30 ± 0.12</td>
</tr>
<tr>
<td>( R_5 )</td>
<td>2.81 ± 0.15</td>
<td>2.73 ± 0.18</td>
<td>2.63 ± 0.15</td>
</tr>
<tr>
<td>( R_6 )</td>
<td>3.06 ± 0.17</td>
<td>2.98 ± 0.15</td>
<td>2.90 ± 0.16</td>
</tr>
<tr>
<td>( R_7 )</td>
<td>3.58 ± 0.15</td>
<td>3.35 ± 0.00</td>
<td>0.00 ± 0.00</td>
</tr>
</tbody>
</table>

Discussion

In this study the analysis of the variation in the shape and growth increments of *asteriscus* otoliths of *Prochilodus nigricans* among specimens from the different rivers of the Amazon Basin was performed by a combination of methods. Although no differences among populations from the three rivers were detected from morphometric and morphological analyses, there were variations in the estimation of otolith growth increment. The specimens inhabiting Japura and Solimões rivers presented larger otoliths with faster growth (seven bands of seasonal growth), whereas specimens living in Negro River were smaller otoliths and lower growth showing (six growth marks). The otolith shape wavelet analysis also revealed a high variability among individuals, showing four
Morphology of *asteriscus* otolith in *Prochilodus nigricans*

Fig. 4. Four morphotype *asteriscus* otolith *Prochilodus nigricans* established by cluster analysis from the amplitudes of wavelets. **a.** morphotype 1; **b.** morphotype 2; **c.** morphotype 3 and **d.** morphotype 4 respectively, sampled of rivers Solimões, Japurá and Negro. Scale bars: 1 mm.

Different morphotypes. Some studies have suggested a relationship between *sagittae* otolith morphs and otolith rate (Secor, Dean, 1989; Reznick *et al*., 1989; Worthington *et al*., 1995; Tuset *et al*., 2004; Abaad *et al*., 2016). Although they were not linked to one specific river, slight differences were observed in their predominance, which may be related to adaptive responses (evolutionary) and environmental conditions (selective pressures) within of each rivers of the Amazon Basin (Swain, Foote, 1999; Hubert, Renno, 2006). Changes in otolith morphology (*sagittae, asteriscus or lapilli*) are the result of an interaction between environmental factors and genetic plasticity (Teimori *et al*., 2012; Vignon, 2015; Avigliano *et al*., 2016), which are complex of interpretation since they may be generated by a variety of processes and interactions such as ontogenetic, adaptations, biogeographic and phylogenetic processes (Mclachlan *et al*., 2011; Tuset *et al*., 2016b). Indeed, otolith morphotypes may reflect adaptations to optimize fish survival in the context of different sound environments (Gauldie, Crampton, 2002). Physiological variations reflect the somatic growth of fish and such events are recorded as daily or seasonal growth marks on the otoliths, as well as otolith shape. Thus, many elements of the surrounding environment where fish lives are incorporated into the otolith calcified matrix (Panella, 1971; Campana, Casselman, 1993; Campana, 1999; Cardinale *et al*., 2004) and otolith outline is used to identify stocks (Campana, Casselman, 1993; Begg, Brown, 2000; Stransky, 2005; Stransky *et al*., 2008). The knowledge on ecology of *P. nigricans* is very limited, but our results suggest that the individuals could spent parts of their life cycle in different habitats of the rivers, influencing in the annuli increments and otolith shapes, as it has been described in other fishes (Molony, Choat, 1990; Zhang *et al*., 2014; Romo-Curiel *et al*., 2015). However, these habitats should have to appear in the three rivers studied since all otolith morphotypes are present in them, although the morphotype 2 was only found in the Japurá River. In any case, we conclude that specimens of *P. nigricans* inhabiting the three rivers of Amazon basin are the same population. This is not abnormal in freshwater species, for instance, Mallen-Cooper, Stuart (2003) found variability in otolith growth of golden perch *Macquaria ambigua* (Richardson, 1845) and silver perch *Bidyanus bidyanus* (Mitchell, 1838) from Australian rivers spite of the specimens allow to same stock. In fact, a recent study revealed the existence of breeding areas in *P. lineatus* from in the Plata, and the results obtained indicate that the otolith microchemistry and morphometry, and scale morphometry are good markers of habitat and represent a potential tool for identification of...
streaked Prochilodus nursery areas (Avigliano et al., 2016). In general, prochilodontids have large population sizes in its distribution area and this feature (Turner et al., 2004), combined with high gene flow provided by their migratory habits, may contribute to high levels of variability in growth and shape of otolith. Both characteristics tend to minimize the effects of genetic drift in eroding intrapopulation genetic diversity (Machado, 2009). The lack of population segregation in the Amazon water system is probably due to the immense water connections that are established between the main channel of the river and lakes and lowland areas during the period of high water, favoring the genetic flow among individuals of different locations (Lowe-McConnell, 1987; Turner et al., 2004). The patterns of diversification at species level for groups such as genus Prochilodus and similar taxa, present comparisons of life history patterns and ecological characteristics that probably reflect the recent biogeographical history of the large river basins of South America and contribute to form our view of landscape evolution in neotropical lowland region (Sivasundar et al., 2001).

Acknowledgments

The authors thank Doctor Maria Mercedes Bittencourt (in memoriam), Project Coordinator, for providing the otoliths and data. Doctor José Celso Malta from the Fish Parasitology Laboratory, INPA for providing the camera used for the photos. CNPq for granting the doctoral scholarship.

References


Morphology of asteriscus otolith in Prochilodus nigricans


Reis RE, Kullander SO, Ferraris JC. Check list of the fresh water fishes of South and Central America. Porto Alegre: EDIPUCRS; 2003.


Morphology of asteriscus otolith in Prochilodus nigricans


Submitted April 17, 2018
Accepted July 02, 2018 by William Crampton