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# Comparison of scale and otolith age readings for trahira, *Hoplias malabaricus* (Bloch, 1794), from Paraná River, Argentina

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# Summary

The aim of this work was to compare age determinations and precision using two deposition structures of trahira Hoplias malabaricus (Bloch, 1794): the scales, which are most frequently used, and otoliths (lapilli). The length-age relationships were obtained with both structures and compared with results from previous studies. The 163 sets of trahira otoliths (lapilli) and scales were 17-46 cm standard length (SL) from Cayastá (Santa Fe) and Islas Lechiguanas (Entre Ríos), Paraná River, Argentina. Three independent readings of each structure were conducted. An age bias plot was performed to compare age estimations from scales and otoliths. To assess the precision of age determinations using both structures, the percent agreement among readers for both structures and the coefficient of variation were calculated (%CV). The age-length relationships were plotted and fitted with the von Bertalanffy growth function for both structures and compared with previous works. Age readings recorded for scales were lower than those recorded for otoliths for ages above or equal to 3 years. Percent agreement among readers was higher than 80% for otoliths and less than 65% for scales. The %CV obtained for scales was 20% for young fish and 17% for adults. For otoliths the %CV was 7% for young fish and 3% for adults. The %CV obtained for scales was over the recommended limit (>7.6%). The von Bertalanffy parameters for scales were  $L_{\rm inf} = 45.80$  mm; k = 0.29;  $t_0 = -1.34$  and for otoliths were  $L_{\rm inf} = 40.76$  mm; k = 0.39;  $t_0 = -1.05$ . Precision of age estimations assessed from the percent agreement and the coefficient of variation indicates that the scales of the trahira are inappropriate to estimate age in population studies for juvenile and adult specimens.

### Introduction

Accurate age estimates are fundamental for an appropriate fisheries management. Age determination allows calculation of growth rate, mortality rate and productivity (Campana, 2001). Fish retain a record of growth in several anatomical structures such as: scales, otoliths and bones (Chambers and Miller, 1995). The process of estimating fish age incorporates two major sources of error (Campana, 2001) (i) a process error associated with the structure being examined; not all bony structures in fish form a complete growth sequence throughout the lifetime of the animal, nor do all axes within

a given structure show a complete growth record (Beamish, 1979), and (ii) error due to the element of subjectivity required for all age estimations.

Historically, the major part of previously published studies of age and growth of species from South America fisheries were made with scales. In the case of trahira there were no published otolith studies (Ramírez, 1963; Paiva, 1974; Barbieri, 1989; Dománico et al., 1993; Dománico, 1998; Grosman et al., 2004; Tordecilla-Petro et al., 2005; Balboni et al., 2011).

The trahira (Hoplias malabaricus) (Bloch, 1794) is an ichthyophagous species with a wide geographical distribution of  $11^{\circ}N - 35^{\circ}S$ ;  $85^{\circ} - 35^{\circ}W$  (Fowler, 1950; Reis et al., 2003). In South America (Teresa et al., 2010; Carvalho et al., 2011; Pedroza et al., 2012; Volcan et al., 2012), the species inhabits most of the hydrographic basins except those to the west of the Andes and in Patagonia. The species is frequently caught for human consumption in freshwater lagoons of Argentina (Grosman et al., 2004), Colombia (Bentancur-Vásquez et al., 2004) and Uruguay (Crossa, 1994; Amestoy, 2001). In Argentina, the trahira occurs in lotic environments (lower and mid Paraná River, lower Uruguay River and pampasean rivers and streams) where it is caught for commercial and recreational purposes (Espinach Ros and Sanchez, 2007).

The objective of the present work is to compare age determinations and precision using two deposition structures of trahira: scales, which are most frequently used, and otoliths (lapilli). Length-age relationships obtained with both structures and compared with results obtained in previous studies are also analyzed.

# Materials and methods

Specimens were caught in the Paraná River (Argentina) in the framework of Proyecto de Evaluación del Recurso Sábalo en el rio Paraná, between Cayastá (Santa Fe province) and Islas Lechiguanas (Entre Ríos province), Argentina. Fish were captured between March 2007 and August 2009, whereby 163 sets of otoliths (lapilli) and scales from 17 to 46 cm standard length (SL) trahira were analyzed. Young fish were defined as smaller than 23.16 cm SL for males and 21.53 cm SL for females (pers. comm.).

Two otoliths and 10 to 20 scales were taken from each specimen. Selected for this analysis were lapilli otoliths that

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present some advantages with respect to the *sagittae* otoliths, which are too small, fragile and difficult to find. The *asteriscii* otoliths are the largest but are also fragile and cannot withstand the sanding procedure.

Scale readings followed the criterion outlined by Ramírez (1963): ten or more scales per fish were removed from the left flank (between dorsal and pectoral fin), rinsed in water with a soft brush and mounted between glass slides with the external side up. They were then placed under a stereoscopic microscope (10X) illuminated from below for the estimation of the number of annuli. Annuli were defined as the regions with crowding of circuli and/or 'cutting over' or breaks in the circuli (Bagenal and Tesch, 1978; Jerald, 1983; Fig. 1a). The otoliths were processed and read following the procedure of Espinach Ros (2008). One otolith from each pair was sanded in transverse cross-section through the nucleus using a minimite with a 600-grit waterproof sandpaper. The sanded surface of the otolith was then polished smooth using 1200grit waterproof sandpaper mounted on the minimite. The polished surfaces were burned in an alcohol flame to produce distinct banding patterns. Otoliths were mounted in plasticine and a drop of transparent nail enamel was placed on the burnt surface to improve the contrast between growth zones. Annuli were read using a stereoscopic microscope (40X). An annulus was considered to consist of a wide, light, opaque zone and an adjacent narrow, dark, translucent, hyaline zone, as seen when the burned otolith was viewed under reflected light (Jerald, 1983; Fig. 1b). The first three authors conducted three independent readings of each structure.

Although no formal validation of the annual frequency of ring marking was conducted, the criterion by Howland et al. (2004) was used. A seasonal pattern with alternating translucent (winter growth from March to August) and opaque bands (September to February) was observed. Based on previous observations and the fact that otoliths have been validated in a great number of species (Beamish and McFarlane, 1987), otoliths were used as reference validated structures to compare scale estimations. Also, Silva and Stewart (2006) validated the annual growth mark in another species of this order

With the mean values of estimated age from otoliths and scales, an age bias plot (Campana, 2001) was produced to visualize the deviation of the age readings of the two ageing methods from the 1:1 equivalence line.

To assess the precision of age determinations using both structures, the percent agreement among readers for both structures and the coefficient of variation were calculated (% CV) according to Chang (1982).

The age-length relationships were plotted and fitted with the von Bertalanffy growth function:

$$L_t = L_{inf}(1 - e^{-k(t-t_0)})$$

where  $L_{\rm t}$  is the fish length (cm) at age t (years),  $L_{\rm inf}$  the asymptotic maximum fish length (cm), and  $t_0$  is the theoretical age (years) when the fish was at zero length.  $L_{\rm inf}$ , the growth coefficient k, and  $t_0$  were calculated iteratively. To compare the effect of both age structures on the growth functions, growth parameters were calculated for each structure and compared with data in the literature.

#### Results

Age bias plots of ages estimated from scales compared to ages estimated from otoliths revealed a large discrepancy in agreement between the two methods (Fig. 2). The age read-

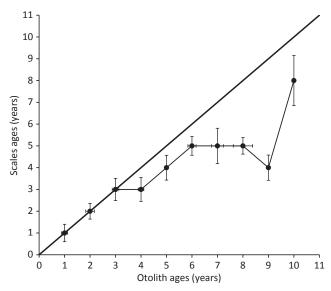
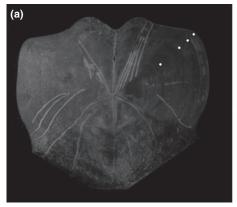


Fig. 2. Age bias plot comparing age readings for both methods. Each error bar = standard deviation around the mean age assigned for each age structure. Regression 1:1 (solid black line).



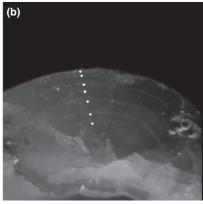


Fig. 1. Two ageing structure examples from the same *Hopliasmalabaricus* showing annual marks (white dots): (a) mounted scale (10X; 4 + marks) and (b) mounted otolith transverse cut (45X; 7 + marks)

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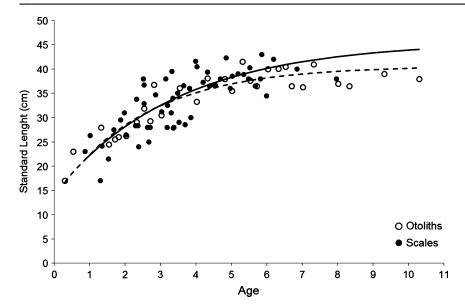


Fig. 3. Trahira length-at-age data, with fitted von Bertalanffy growth curves for scales (solid black line) and otoliths (dashed line). See Table 1 for curve equation parameters

ings recorded for scales were lower than those recorded for otoliths for ages above or equal to 3 years. The maximum estimated age using scales was 8 years, while the maximum for otolith was 10 years.

The percent agreement among scale readers was 63% for young fish and 48% for adults; in otoliths this was 83% for young fish and 82% for adults. The %CV obtained for scales was 20% for young fish and 17% for adults; for otoliths the %CV was 7% for young fish and 3% for adults.

The comparison of age-length function between both structures is shown in Fig. 3. The calculated growth parameters

Table 1 Von Bertalanffy parameters obtained with scales and otoliths per each set of *Hoplias malabaricus*.  $L_{\text{inf}}$ : asymptotic standard length; K: growth rate;  $t_0$ : theoretical age at zero length; SE: standard error.

Structure	$L_{\mathrm{inf}}$	SE	k	SE	$t_0$	SE
Scales	45.80	4.73	0.29	0.11	-1.34 $-1.05$	0.76
Otoliths	40.76	1.56	0.39	0.08		0.48

for *H. malabaricus* varied considerably between ageing structures (Table 1). The tendency for scales to underage resulted in underestimates of the growth rate and overestimates of the asymptotic length. Other studies on trahira growth were mostly made with scales (Table 2). The mean growth rate reported in previous works is  $0.31 \pm 0.17$  and the mean asymptotic length (estimated with total length) reported is  $542 \pm 148$  mm. These values are similar to those found in the present work for scales, 0.29 and 550 mm, respectively.

## Discussion

The comparison of estimated age in scales and otoliths of *H. malabaricus* showed considerable bias, caused by relative underestimation of age in scales. Our results show that the age estimation from scales tend to sub-estimate the age for adults fishes when compared with otoliths; in young fishes this tendency is not observed.

Compared to otoliths, scales had the lowest levels of precision. Analysis of the percent agreement between readers for young fish and adult specimens showed 80% agreement in

Table 2 Von Bertalanffy equation parameters estimated for different trahira populations.  $LT_{inf}$  asymptotic total length; K; growth rate;  $t_0$ : theoretical age at zero length.

Environment	Structure/Methodology	$LT_{inf}\left( mm\right)$	K	$t_0$	Source
Paraná River	Otoliths	492.02	0.40	-1.10	This work
Paraná River	Scales	550.96	0.29	-1.39	This work
Yalca shallow lake (summer group)	Scales	486.54	0.57	0.54	Balboni et al., 2011
Yalca shallow lake (spring group)	Scales	499.69	0.47	0.13	Balboni et al., 2011
Chascomús	Scales	536.75	0.37	0.02	Ramírez, 1963
Indio Muerto	Scales	564.38	0.51	0.03	Grosman et al., 2004
Monte	Scales	817.59	0.08	-0.76	Dománico, 1998
Lobos	Scales	792.06	0.07	-1.02	Dománico et al., 1993
Ciénaga Grande de Lorica	ELEFAN	481.00	0.29	-0.50	Tordecilla-Petro et al., 2005
Amanari, Maranguape Reservoir, females	Scales	388.01	0.26	-1.29	Paiva, 1974
Amanari, Maranguape Reservoir, males	Scales	452.34	0.19	-1.68	Paiva, 1974
Monjolinho Reservoir	Scales	405.00	0.24	0.10	Barbieri, 1989

the otolith readings. However, in scale readings this was below 50% for adult specimens. The coefficient of variation (%CV) obtained from scale reading was over the recommended limit of 7.6% (Campana, 2001) in young fish and adults; for otoliths, the %CV was lower than the recommended limit both in young fish and adults. Our ageing precision results were similar to those of earlier studies. Welch et al. (1993) observed a percent agreement among scale readers for Morone saxatilis of 53%. Erickson (1983) reported percent of accurate agreement for Stizostedion vitreum ranging from 78 to 93% among otolith readers and from 52 to 90% for scale readers. Libby (1985) and Boxrucker (1986) calculated the coefficient of variation among readers for the age determination of Alosa pseudoharengus and Pomoxis annularis showing higher agreement among otolith readers than among scale readers. In agreement with our observations, Sharp and Bernard (1988) observed a reduction in the ageing accuracy determination with the increase in the length of fish scales and otoliths for the trout Salvelinus namaycush.

Although the scales showed some practical advantages such as rapid extraction and no need of specialized expertise or sacrifice of specimens, results show that age determined from scales is slant compared with age determined from otoliths, the former being more reliable for older ages. The technique used in the present work for the preparation of the otolith readings requires a shorter period of time than the technique used for scale readings, although care must be taken to prevent over-burning.

Previously reported *H. malabaricus* results show very dissimilar values for von Bertalanffy parameters. For example, Dománico et al. (1993) reported a relatively very low growth rate value of 0.07, while Balboni et al. (2011) report a value of 0.57. Given that the majority of past studies of trahira employed the use of scales, it is probable that the growth rates and asymptotic length were incorrectly estimated. Thus, caution should be exercised in relying on scale-based data for management decisions and in some cases the re-examination of populations may be necessary (Howland et al., 2004).

Precision of age estimations assessed from the percent agreement and the coefficient of variation indicates that the scales of the trahira, *Hoplias malabaricus*, would be inappropriate to estimate the age in population studies for either young or adult specimens.

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