

**Estimation of the age and biometry of *Salminus brasiliensis* (Cuvier 1816)
captured in the Funil hydroelectric plants****Estimativa da idade e biometria de *Salminus brasiliensis* (Cuvier 1816)
capturada nas hidrelétricas de Funil**

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ABSTRACT

In order to better understand the ecology and biology of *Salminus brasiliensis*, and to provide basic information for the evaluation of fish stocks, this study verified the age and biometry *S. brasiliensis*. Sixty-six fish (aged 2-15 years) were captured at the Funil Hydroelectric Power Plant, Brazil, between September 2006 and August 2007. The biometric data recorded were weight, total and standard length, height, thickness and head length. Sex was determined by macroscopic evaluation of gonads. The gonads and liver were removed and weighed for calculation of gonadosomatic index and hepatosomatic index. The age of each fish was determined by analysis of growth rings in the scales, while the scale radius was measured from the focus to the end of the scale. Differences between seasonal biometric averages and reproductive indexes were verified using the Newman Keuls test at 5%. Results indicate that in the spring and summer seasons, smaller fish were caught compared to those caught during the winter and autumn. Females tend to have greater weight and morphometric measurements than males. The age and radius of the scale were positively correlated with the biometric variables. The biometric variables of *S. brasiliensis* can be used as an indicator of age in this species.

Keywords: Dourado, growth; fish; morphometry; ray of scale.

RESUMO

Com o objetivo de melhor compreender a ecologia e a biologia de *Salminus brasiliensis* e fornecer informações básicas para a avaliação dos estoques de peixes, este estudo verificou a idade e a biometria de *S. brasiliensis*. Sessenta e seis peixes (de 2 a 15 anos) foram capturados na Usina Hidrelétrica de Funil, Brasil, entre setembro de 2006 e agosto de 2007. Os dados biométricos registrados foram peso, comprimento total e padrão, altura, espessura e comprimento da cabeça. O sexo foi determinado por avaliação macroscópica das gônadas. As gônadas e o fígado foram removidos e pesados para o cálculo do índice gonadossomático e do índice hepato-somático. A idade de cada peixe foi determinada pela análise dos anéis de crescimento nas escalas, enquanto o raio da escala foi medido do foco até o final da escala. As diferenças entre as médias biométricas sazonais e os índices reprodutivos foram verificadas pelo teste de Newman Keuls a 5%. Os resultados indicam que, nas estações primavera e verão, peixes menores foram capturados em comparação com os capturados durante o inverno e o outono. As mulheres tendem a ter maior peso e medidas morfométricas do que os homens. A idade e o raio da escala foram positivamente correlacionados com as variáveis biométricas. As variáveis biométricas de *S. brasiliensis* podem ser usadas como um indicador de idade nesta espécie.

Palavras-chave: Dourado, crescimento; peixe; morfometria; raio de escala.

1 INTRODUCTION

The estimation of the age of the fish provides parameters necessary to evaluate the fish population dynamics (recruitment, growth, and mortality) and stock structure (Koch and Quist, 2007; Maceina et al., 2007; Methot and Wetzel, 2013). The data obtained can be used in the creation of age-structured population models (Maceina et al., 2007), which can guide the management and implementation of conservation programs (Spurgeon et al., 2015). Information used to determine age

in fish populations is usually derived from calcified structures, such as gills, pectoral fin rays, scales and sagittal otoliths (Buckmeier et al., 2018). These marks are usually associated with environmental factors (Felizardo et al., 2010).

Scales are a very useful biometric structure since they are easy to collect and do not negatively impact the health of the fish after removal. The scales have certain characteristics that can be used to estimate the age via counting the number of annuli, growth, spawning activity, migrations, diseases, environmental conditions related to temperature, and lack of food, among others (Rojo, 1991). A number of studies such as Araya et al. (2005) and Agostinho et al. (1999) have been carried out on correlation between the formation of rings and water temperature, fluctuations in water levels, photoperiod, transparency, water velocity in the habitat, and food availability.

In Brazil, one of the criteria adopted by the government to guide decisions related to species conservation is related to fish age estimates (Santana and Minte-Vera, 2017). Consequently, using scales for determination of age have been widely used in native fish in Brazil, such as *Hypophthalmus marginatus* (Cutrim and Batista, 2005), *Leporinus obtusidens* (Araya et al., 2005), *L. acutidens* (Araya et al., 2008) and *Salminus brasiliensis* (Dei Tos et al., 2009). The *S. brasiliensis* scale is of the elasmoid type, specifically of the cycloid type. According to Holden & Raitt (1974) this type of scale is subcircular, disk-shaped and thin and is found in most soft-rays fish in the fins, presenting on its outer surface concentric deposited sclera or striations following a regular pattern, from the center and origin of the scale, the foccus.

The estimation of the age of the fish, using the scaling methodology is widely employed in South America, due to its low sampling cost, ease of preparation of the samples, and sidestepping the use of expensive equipment (Isely and Grabowski, 2007; Santana and Minte-Vera, 2017).

Salminus brasiliensis commonly known as dourado, is a characin found in the rivers of Bolivia, Brazil, Paraguay and Uruguay (Gagne et al., 2017) and travel long distances (up to 1000 km) to reach reproductive areas (Petrere Junior, 1985). Despite its importance, this species is considered vulnerable according to the red list of species, being threatened with extinction in some basins (Barletta et al., 2010). Agostinho et al. (2007) noted that fish usually come under threat due to the construction of dams, which leads to the destruction of habitat, interruptions of migratory routes, blocking access to spawning and changes in water quality.

In order to better understand the ecology and biology of *S. brasiliensis* and to provide basic information for the evaluation of fish stocks, this study aimed to determine the age, a variation of biometric and seasonal parameters for each age and the correlation between age and the scales rays with the biometric parameters and reproductive indices of *S. Brasiliensis* in the Funil Hydroelectric Plant, Brazil, three years after the dam closed.

2 MATERIAL AND METHODS

2.1 STUDY AREA

The study was conducted in the upper course of the Rio Grande, between the municipalities of Lavras and Perdões, in the south of Minas Gerais (21 ° 08 'S and 44 ° 55'W), downstream of the Funil Hydroelectric Power Plant UHE Funil). The construction of the Funil HPP started in 2000 and started operating in 2003. The Funil HPP has a fish transposition system (FTS) of the lift type designed to allow for the migration of different species of fish upstream of the dam(Murgas et al., 2018).

2.2 SAMPLING

A total of 32 males and 32 females of *S. brasiliensis* were evaluated between 2006 and 2007. Individuals of *S. brasiliensis* were caught between September 2006 and August 2007 in the Rio Grande, downstream of the Funil HPP, with authorization from the State Forestry Institute (IEF), through the scientific fishing license category D-N. 063-07. Fish were collected at intervals of 15 days in the morning, using a rod and windlass, using as bait earthworms and bovine heart.

The highest pluviometric indexes observed during the period of capture of the animals were between November and February, with a peak of 17.9 mm in January. The mean temperature was 20.5°C (\pm 3.2) using a portable digital thermometer. After the capture, the animals were desensitized in benzocaine (80 mg L⁻¹) and kept immersed in ice during transport for the Animal Physiology and Pharmacology Laboratory, Veterinary Medicine Department at the Federal University of Lavras. Afterward, the specimens were taxonomically identified and the following morphometric parameters were measured with a pachymeter and an ichthyometer: (i) total length (TL - cm), (ii) standard length (SL - cm), (iii) height (H - cm), (iv) depth (DP - cm) and (v) head length (HL - cm). The total weight (TW - g) of each animal was also obtained through a digital balance. Width measurements were obtained in the region of the 1st radius of the dorsal fin, the SL, between the head end and the lower perimeter of the stem (caudal fin insertion), the HL between the anterior end of the head and the caudal part of the operculum, the body height was measured from the radius of 1° of the dorsal fin (Figure 1).

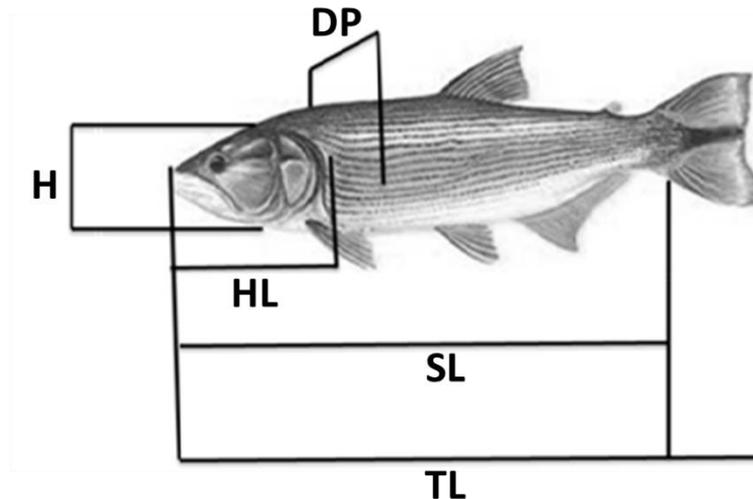


Figure 1: Morphometric measurements performed on a golden specimen of *S. brasiliensis*.

After obtaining the morphometric data, the animals were incised longitudinally and the walls resulting from this incision were folded, and the gonads removed, and macroscopically analyzed for sex determination and weighed to calculate the gonadosomatic index [$GSI = (gw/tw) \times 100$], where gw is the weight of the gonads (g) and tw is the total weight (g) of the fish (Vazzoler, 1996). Likewise, the liver was removed and weighed to obtain the hepatosomatic index, calculated as follows: $HSI (\%) = \text{hepatopancreas weight (g)} \times 100 / \text{bodyweight (g)}$.

Prior to the extraction of the scales, the fish were washed in running water to remove any dirt residues and lost fish scales from other fish. The scales were collected with forceps from the region below the dorsal fin and above the lateral line. All the scales collected for each specimen were inspected for deformities, scratches or any changes that made it difficult to count the annuli. Three scales (symmetrical) were selected from each animal, identified and stored in envelopes for later determination of age. Due to the hydrophilic characteristics of the scales, a hydration treatment by immersion in distilled water was performed prior to readings.

The scales of each specimen were placed on microscope slides and their readings were performed with a binocular loupe (Wild ST4, Swiss) at a magnification of 20x, allowing for the complete visualization of the scales. The reading was made by visualizing the zones where the striae are more spaced (fast growing areas, which usually occurs in summer), followed by areas where the scales are less spaced (slow growth zones - which occurs in winter) that form a mark on the scale (ring or annulus). Together these two zones correspond to an annual growth zone. These growth marks should be observed throughout the surface of the scale (Holden, 1974). As the age was read, the radius (mm) of the scale was also measured from the focus (center of the scale) to the edge of the scale (Figure 2).

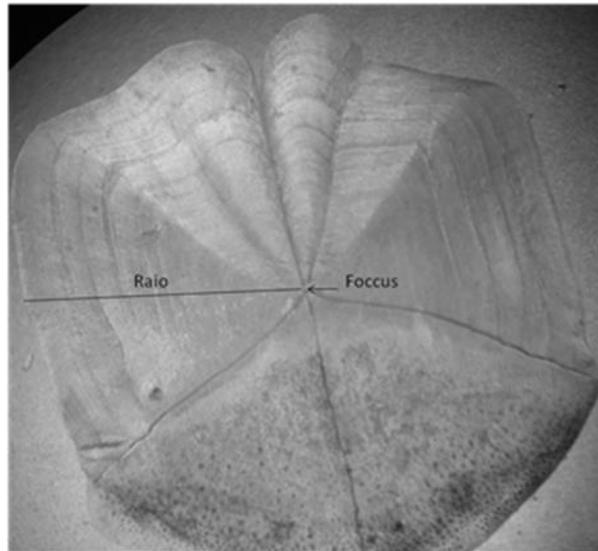


Figure 2: Measurement of the scale radius.

2.3 DATA ANALYSIS

For the study of the effects of sex and seasonal season on the biometrics variables of the fish collected, the data were submitted to analysis of variance using a statistical model with two cross-classification criteria, which included the effects of sex, season of the year and of the interaction between these factors. The means were compared using the Newman Keuls (SNK) test at a 5% probability level. A correlation of age and radius of the scales was made with the variables related to the biometric data and Pearson's correlation coefficient. Statistical analyzes were performed using R software version 3.0.2 (R Development Core Team, 2013).

3 RESULTS AND DISCUSSION

3.1 SEX RATIO

The sexual ratio found of 1:1 (Table 1) was within the expected range for this population. Not relevant to this study as the 1:1 ratio was within expectations for a normal population. Dala-Corte e Azevedo, (2010) suggest that for species with a ratio of 1:1 it implies that it does not present differences in the birth, mortality and growth rates, nor does it have sexual dimorphism.

Table 1. Specimens of *Salminus brasiliensis* captured downstream of the Funil HPP, grouped by season and sex.

Season	Sex		Total
	Male	Female	
Winter	6	9	15
Spring	6	8	14
Summer	10	7	17
Autumn	10	8	18
Total	32	32	64

3.2 AGE OF INDIVIDUALS

Although the Scales of *S. brasiliensis* was generally well defined, thus facilitating identification and counting, approximately 20% of the scales presented irregularities in the rings, making it impossible to identify the age of the fish. Irregularity of the growth rings is common due to the occurrence of losses and regeneration of the scales. Regenerated scales can be easily identified by the irregular shape of the striations and by the absence of concentric striations near the center of the scale (Holden, 1974). The 20% irregular scales are on the low side seeing that Araya et al. (2008) discarded 45% of the scales in a study of *L. acutidens*. Indicate possible reason(s) for your low count of irregular scales as compared to Araya et al. (2008).

The age of the individuals sampled varied from 2 to 15 years, and the age group with the highest frequency of capture were those of 6 (n=10), 7 (n=12) and 8 (n=11) years of age, decreasing in number for the lowest or highest observed age (Table 2). A similar finding (most individuals in the 9-year-old category and a decrease in animals collected at higher or lower ages) was observed by Felizardo et al. (2015), in a study evaluating the age of *L. obtusidens*. According to Dei Tos et al. (2010), freshwater fish in South America do not present high longevity, reaching a maximum of 15 years of age.

Table 2. Biometric data, reproductive indexes, radius of scale according to the ages (N) of *Salminus brasiliensis* captured downstream of the Funil hydroelectric plant

Age (N)*	Parameters								
	Weight(g)	TL (cm)	SL (cm)	Width (cm)	Height (cm)	HL (cm)	Ray (mm)	GSI (%)	HSI (%)
2 (3)	916.6±381.8	44.8±4.3	40.1±1.7	555.0±0.7	9.6±1.2	9.7±1.5	3.3± 1.1	0.2±0.1	0.4±0.1
3 (3)	1250.0±661.4	48.0±8.8	40.3±5.8	4.9±0.8	11.2±2.3	10.4±2.2	4.3± 1.5	0.3±0.3	0.5±0.0
4 (4)	1600.0±424.2	51.2±2.0	45.0±3.5	6.1±0.5	11.9±0.6	10.9±1	5.1±1.3	0.4±0.4	0.7±0.3
5 (8)	2590.2±1164.3	56.7±7.2	49.8±7.2	6.4±1.0	14.4±3.1	12.4±2.5	5.8±0.8	0.2±0.1	0.5±0.1
6 (10)	3284.1±827.7	63.6±4.3	55.2±3.8	6.9±1.5	15.7±2.1	13.7±1.4	6.2±0.8	0.1±0.1	0.5±0.1
7 (12)	3505.0±1014.1	62.6±5.3	55.2±3.5	7.0±0.5	15.9±2.4	14.3±1.9	6.6±0.4	0.2±0.1	0.5±0.1
8 (11)	3940.9±1182.1	64.0±5.8	55.9±6.1	7.0±0.5	16.7±3.5	14.8±2.6	6.8±0.9	0.2±0.1	0.6±0.2
9 (7)	5045.7±1106.7	66.0±8.8	60.8±5.0	8.3±0.8	19.5±2.6	16.0±1.4	8.1±0.3	0.4±0.1	0.5±0.0
11 (2)	5250.0±1060.6	72.0±7.0	63.5±4.9	7.7±0.4	20.5±0.7	15.5±0.7	9.5±0.7	0.2±0.1	0.7±0.2
12 (2)	6400.0±424.2	78.2±6.0	71.5±2.1	8.1±0.0	19.2±3.1	19.2±0.3	10.0±0.7	0.4±0.2	0.4±0.1

13 (1)	5600.0±0.0	75.0±0.0	66.0±0.0	7.3±0.0	18.5±0.0	14.5±0.0	10.6±0.0	0.1±0.0	0.9±0.0
15 (1)	8000.0±0.0	83.0±0.0	75.0±0.0	9.5±0.0	24.0±0.0	19.5±0.0	11±0.0	0.40±0.0	0.6±0.0

* N number of animals. TL: Total length, SL: Standard length, HL: Head Length, GSI: Gonadsomatic index, HIS: Hepatosomatic index

3.3 SIZE OF INDIVIDUALS

As there was no significant interaction ($P>0.05$) of the biometric data of males and females seasonally, the values obtained for each sex were grouped for analysis. In this way, it was observed that in the spring and summer season smaller fish were captured in relation to those captured during the winter and fall (Table 3). With an increase in the water current caused by the floods, the hook that was thrown on the river was dragged to the embarkment edge, where smaller fish were found, since in deeper places smaller animals become easy prey for larger fish. On the other hand, the increase in river flow may also stimulate fish to seek margins. The increase in the current was generally caused by the release of water from the spillways, opened during the flood season to control the level of the upstream dam.

3.4 WEIGHT OF INDIVIDUALS

Females had a greater weight and morphometric measurements than males (Table 3). The mean total length of females observed was 64.8cm, whereas in males it was 58.6cm. These results are similar to those obtained by Barbieri et al. (2001), evaluating *S. brasiliensis* in the Mogi-Guaçu river and by Araya et al. (2005), for fish collected in the Paraná River. Felizardo et al. (2010) evaluating *Megaleporinus obtusidens* (piapara) swimming bladder abnormalities collected in the same place of study, verified that the females were larger than males, although they were of the same age. This difference is due to the fact that females present a higher growth rate than males and, as a consequence, reach higher lengths for the same age (Felizardo et al., 2015).

Table 3. Seasonal biometric data of *S. brasiliensis* caught downstream of the Funil hydroelectric plant.

		Weight (g)	Age (years)	Ray (mm)	TL (cm)	SL (cm)	Width (cm)	Height(cm)	HL (cm)
Season*	Spring	4521.2±1429.9 ^a	8.3±2.8 ^a	7.0±0.7 ^a	66.8±8.0 ^a	60.3±6.4 ^a	7.4±1.5 ^a	17.8±2.6 ^a	16.0±2.1 ^a
	Summer	1997.9±1544.8 ^b	5.1±2.3 ^d	5.1±1.5 ^b	53.5±10.1 ^b	46.8±8.7 ^b	6.2±1.4 ^b	12.4±3.4 ^c	11.4±2.7 ^b
	Autumn	4128.6±1278.0 ^a	7.4±2.1 ^b	6.6±0.9 ^a	64.9±6.3 ^a	56.3±6.4 ^a	7.2±0.8 ^a	18.2±2.7 ^a	14.5±1.7 ^a
	Winter	3501.3±1250.7 ^a	6.9±2.3 ^c	6.6±0.9 ^a	62.5±7.4 ^a	55.7±6.9 ^a	7.0±0.7 ^a	15.3±2.7 ^b	14.3±2.6 ^a
Sex	Male	2975.1±1668.1	6.8±2.8	5.9±1.5	58.6±10.0	51.9±8.9	6.7±1.2	14.9±3.7	12.7±2.5
	Female	4027.8±1508.2	6.8±2.3	6.7±0.9	64.8±7.8	57.0±7.5	7.2±1.2	16.9±3.5	15.1±2.5

* Different overwritten letters in columns indicate difference at 5% probability. TL: Total length, SL: Standard length, Width, HL: Head length.

3.5 REPRODUCTION OF INDIVIDUALS

In the present study, we observed that the seasonal gonadosomatic index (GSI) of the females was higher ($P < .05$) than in the males, except in the summer, where males and females showed similar GSI ($P > .05$). The most discrete variation in the GSI of males during the seasons, when compared to females can be attributed to the differences between testes and ovaries, in terms of volume, mass and energy demand required for the production of gametes (Ribeiro et al., 2007). On the other hand, the similarity between GSI in the summer can be explained, due to the emptying of the gonads of the females after spawning, whereas the gonads of the males do not present seasonal variations. The fact that the GSI of the females has decreased significantly ($P < .05$) in summer in relation to spring, may indicate that the reproductive period of this species occurs preferentially in spring, where the highest GSI was observed (Figure 3).

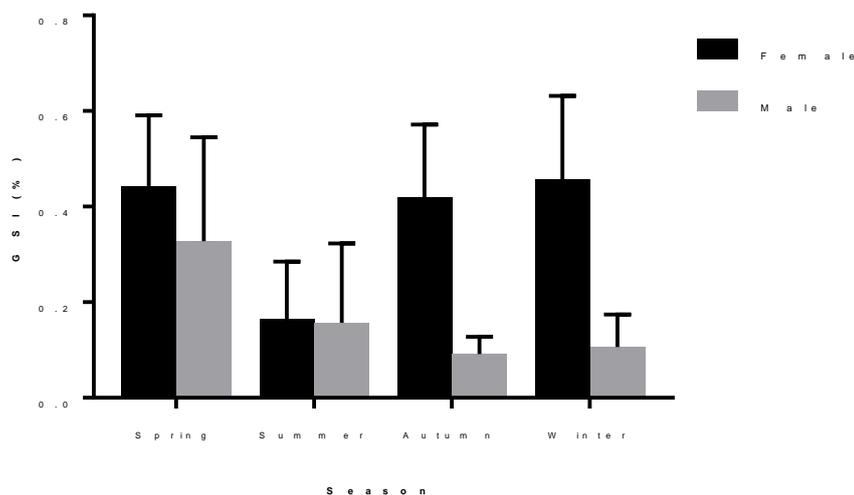


Figure 3: Gonadosomatic index (GSI) (%) seasonal of males and females of *S. brasiliensis*.

3.6 LIVER WEIGHT

The hepatosomatic index (HSI) for both sexes of *S. brasiliensis* can be used as indicator of the reproductive period, since the highest HSI for females and males was observed in spring and summer, differing significantly ($P < 0.05$) of the other stations (Figure 4). This observed result coincides with the reproductive period of the species. According to Querol et al. (2005), the HSI is related to the mobilization of energy reserves necessary for the (i) process of vitellogenesis, (ii) reproduction or (iii) preparation for the winter period. The importance of the HSI was also commented on by Costa et al. (2005), citing the changes caused by the liver, since this organ synthesizes and secretes the precursor of the proteins of the vitellogenin. The vitellogenin is removed from the bloodstream for the development of the oocytes, while the rapid accumulation of the yolk probably

happens by the decrease of the weight of the liver (Yoneda et al., 2001). On the other hand, during testicular and sperm maturation, the liver also participates as an energy supplier for the testicular reabsorption and reorganization process, as well as the transfer of liver substances to the metabolism involved in the production of gametes in the testes. Thus, it is expected that a variation in liver weight will occur during the seasons, reflecting the assimilation or use of energy reserves by the fish. In the present study, the lower HSI were observed in the months before the reproductive period, this fact may be related to the high energy cost for oocyte production, in addition to other costs, such as male demand and the high risk of predation during the cut and copula (Querol et al., 2005). Similar results were observed in *Loricariichthysplatymetopon* (Querol et al., 2005) and *Cynoscion acoupa* (Almeida et al., 2016), which indicated a decrease in HSI at spawning peaks.

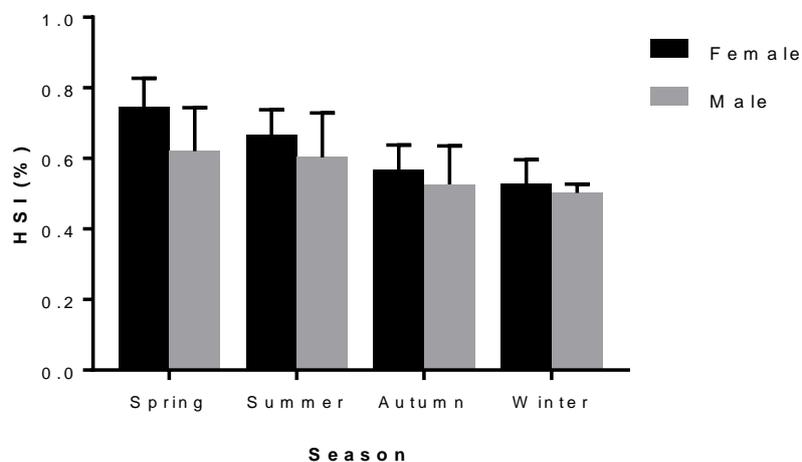


Figure 4: Seasonal Hepatosomatic (%) index of males and females of *S. brasiliensis*.

3.7 CORRELATION BETWEEN AGE AND RADIUS OF THE SCALE

A strong correlation ($r > 0.7$) was present for weight, total length and pattern, with the age of the animals in both sexes (Table 4).

Table 4. Pearson's correlation coefficient between age and radius of the scale with the biometric variables of males and females of *S. brasiliensis*

Variable	Variable	Correlation (r)	
		Male	Female
Age	Weight (g)	0.873	0.802
	Ray (mm)	0.819	0.658
	Width (cm)	0.699	0.560
	Height (cm)	0.819	0.612
	Head length (cm)	0.791	0.681
	Total length (cm)	0.858	0.790
	Standard length (cm)	0.820	0.838
	Gonadosomatic index (%)	-0.027*	-0.129*
	Hepatosomatic index (%)	0.231*	-0.127*
Ray	Weight (g)	0.775	0.681

Width (cm)	0.499	0.411*
Height (cm)	0.713	0.554
Head length (cm)	0.754	0.523
Total length (cm)	0.848	0.767
Standard length (cm)	0.890	0.790
Gonadosomatic index (%)	0.088*	0.285*
Hepatosomatic index (%)	-0.059*	0.048*

* There is no significant correlation with the Pearson correlation at 5% probability.

Similarly, we observed a strong correlation between the radius and the total and standard-length measurements in *S. brasiliensis*, and a moderate correlation ($r > 0.6$) between the radius scale and the total and standard length of females. The high correlation coefficient ($r = 0.74$) was also observed between the radius and the standard length independently of the sex of the fish. In this case a regression model was applied to describe the relation between the radius and the standard length (Figure 5). Radius growth showed a positive increase in relation to fish SL. As reported by Ibáñez and O'Higgins, (2011), the size of the fish has a high correlation with the size of the scales. In *Leporinus obtusidens*, a correlation above 80% between the radius of the scales and the standard length were reported by Felizardo et al. (2015), similar to that observed in the present study. As reported by Haimovici e Reis, (1984), the radius of the scales of *Umbrinacanosai* increase in proportion to the length, in addition, these same authors assure that the length of this fish can be used as an indicator of the age.

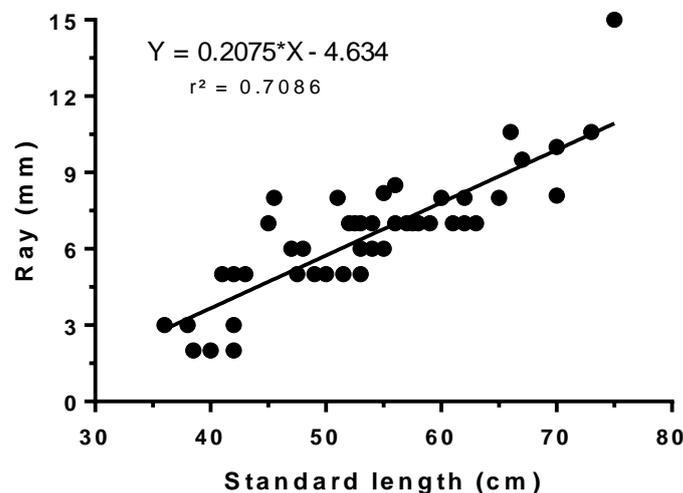


Figure 5: Radius (mm) vs Standard length (cm) of *S. brasiliensis*.

It was also verified that the age of *S. brasiliensis* does not influence ($P < 0.05$) in the GSI and HSI, suggesting that the age does not affect the reproductive performance of this species. However, the quality of the gametes should be verified in animals with advanced age (Felizardo et al., 2015), since the reproductive potential of the animals may be influenced by biotic and

abiotic factors (Jakobsen et al., 2009). The results observed in this study demonstrate that the age identification of *S. brasiliensis* can be estimated using the biometric length data of the animals.

4 CONCLUSION

Based on this work, we can conclude that the dourado captured downstream of the Funil Hydroelectric Power Plant had ages between 2 and 15 years; the age and radius of *S. brasiliensis* scales are highly correlated with biometric variables, thus indicating the possibility of using these variables as an indication of age in this species.

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