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Fisheries and Aquaculture Journal

Mini-Review

Freshwater Angelfish *Pterophyllum scalare* (Lichtenstein; Pisces: Cichlidae) Phenotypic Plasticity during Fasting Periods

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ABSTRACT

One of the most important species for the ornamental fish trade, the freshwater angelfish (*P. scalare*), is a cichlid from the Amazon basin. As ornamental fish market requires live animals, these fish are transported for trading inside bags, and in some cases, they experience long days of fasting. Thus, the present experiment was carried out to elucidate the effects of fasting on the phenotypic plasticity of organs with increased activity during such periods (e.g., the liver and intestine) and the likelihood of survival after such events, concerning the development of juveniles of the Amazon Cichlid *P. scalare*.

Keywords: P. scalare; Angelfish; Amazon basin

INTRODUCTION

The juveniles (*P. scalare*) were subjected to different fasting periods: 0, 3, 6, 9, 12 and 15 days of fasting, respectivily T0, T3, T6, T9, T12 and T15. After the respective fasting periods, the fish from all treatments were fed for 30 days. Treatment 0 (T0) was assumed to be the positive control, and so, the fish subjected to this treatment did not go through any fasting period. A total of 224 juveniles, with no significant difference (P>0.05) for weight (1.09 mg ± 0.12) and length (26.51 mm ± 0.71), were used in the experiment. From these, 20 specimens were killed (desensitized using clove oil and ice) the day before the onset of the experiment to obtain the initial body indexes (length; weight; liver weight; intestine weight). Also, from this group of 20 fish, liver and intestine samples (n=6) were collected for histological analysis on these organs before the fasting period. These data were used as initial body indexes for T0. For T0, 24 juveniles were distributed randomly into three different tanks, eight fish per tank. For the other treatments (T3, T6, T9, T12 and T15), 180 juveniles were distributed randomly into 15 different tanks, totaling 12 fish per tank. Thus, 18 tanks (20 L, each) (six treatments with three repetitions each) were used in this experiment; each one represents an experimental unit. At the end of each treatment's fasting period four animals from each tank were killed to obtain liver and intestine weight values. The eating routine of *Pterophyllum scalare* comprises of a wide food range; they feed on adolescent fishes, prawns, and worms. They feed on various sorts of worms, for example, Tubifex worms, vermicomposting worms, and bloodworms, the tubifex diet are high in protein and supplements for the angelfish bringing about high critical development and live food diet gave better regenerative execution. Besides, the mosquito hatchlings are additionally an extremely nutritious live feed food. Anglerfish are circulated all through the world and incorporate both free-swimming (pelagic) species and seabed-staying (benthic) species. Most types of angler fish live on the sea depths and have strange body structures. They have little gill openings situated at or behind the foundation of the pectoral blades, which are appendage like in structure; a few animal groups additionally have appendage like pelvic balances.

LITERATURE REVIEW

The liver and the initial portion of the intestine were fixed in formalin for further histological analyses. Thus, at the beginning of the feeding period, all treatments had eight fish per tank. In

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Received: September 13, 2021; **Accepted:** September 27, 2021; **Published:** October 04, 2021

Citation: Motta de JHS, Glória LS, Souza de AB, Polese MF, Bosisio F, Vidal MV (2021) Freshwater Angelfish Pterophyllum scalare (Lichtenstein; Pisces: Cichlidae) Phenotypic Plasticity during Fasting Periods. Fish Aquac J. S4:002.

the final of the experimental period of each treatment, three fish from each tank were killed to obtain liver and intestine weight (data for Viscerosomatic Index-VSI and Hepatosomatic Index-HSI); the liver and the initial portion of the intestine were fixed in formalin for further histological analyses. Deaths were recorded daily to obtain values for the fasting effect on the estimated survival rate. It is important to mention that fish that were euthanized at the end of each fasting period (T3-third day; T6-sixth day; T9-ninth day; T12-twelfth day; T15-fifteenth day) were not counted as mortality since they did not die through the influence of the treatments.

The discussion can be seen in Table 1. There was a significant difference (P<0.05) in treatments 12 and 15 days of fasting for liver and intestine weight. The fish subjected to treatments 6,9,12 and 15 days of fasting showed different vacuolization in hepatocytes from the ones subjected to 0 and 3 days of fasting (P<0.05) (Figure 1a). The effect of different periods of fasting on the villi can be seen in Figure 1b. These data show the regression of villi, which tended to decrease the length from 6 days of fasting onwards. However, after 30 days of feeding, none of the intestinal villi of the juveniles in any of the treatments presented any significant difference in length (P>0.05). No difference (P>0,05) was observed between treatments for survival rate (mean ± standard error: T0-96% ± 0,04; T3-96,77% ± 0,03; T6-96,77% ± 0,03; T9-96,77% ± 0,03; T12-90,90% ± 0,05; T15-81,10% ± 0,06). Although, the increase in the number of deaths from the eighth day of fasting may indicate that periods with more than 15 days of fasting could be fatal to this species during this life stage.

Table 1: Phenotypic plasticity of juveniles (*P. scalare*) during periods of fasting and feeding (mean ± standard error). IW: intestine weight; LW: liver weight; 0 DF-zero days of feeding; 30 DF-thirty days of feeding. Different letters in the same column mean a significant difference among treatments.

Treatment (fasting days)	**IW (mg)**		**LW (mg)**	
	0 DA	30 DA	0 DA	30 DA
T0	57.35 ± 6.4 A	97.25 ± 6.4 A	36.96 ± 4.3 A	65.39 ± 4.3 A
T3	42.13 ± 9.0 AB	92.72 ± 6.4 AB	16.45 ± 6.0 AB	57.13 ± 4.3 AB
T6	36.20 ± 9.0 AB	91.03 ± 6.4 AB	11.47 ± 6.0 AB	50.96 ± 4.3 AB
T9	36.32 ± 9.0 AB	91.81 ± 6.4 AB	13.40 ± 6.0 AB	49.16 ± 4.3 AB
T12	32.73 ± 9.0 BC	68.14 ± 6.4 BC	10.00 ± 6.0 B	45.14 ± 4.3 B
T15	33.75 ± 9.0 C	46.43 ± 6.4 C	9.33 ± 6.0 B	24.81 ± 4.3 C

Animals that go through periods of fasting tend to create mechanisms to adapt to this adversity. For juveniles of *P. scalare*, the reduction in intestinal villus length may also be considered a strategy for dealing with seasonal food shortage in this species' habitat. The intestine is an organ that expends a large amount of energy and is less commonly used during fasting periods. Thus, the reduction in the intestine mass may lead to a reduction of maintenance energy and rearrangement of energy to organs of more significant importance during fasting [1].

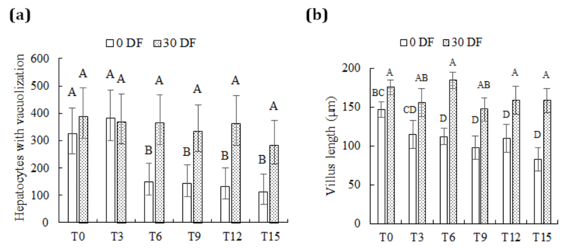

Figure 1: (a) The degree of vacuolization of the hepatocytes of juveniles (*P. scalare*) subjected to different periods of fasting; (b) Intestinal villi length of juveniles (*P. scalare*) subjected to periods of fasting and feeding. 0 DF: zero days of feeding; 30 DF: thirty days of feeding. Different letters ("A" and "B") mean significant difference (P<0.05).

The influence of feeding on intestine development has been reported by some authors. The act of feeding and the need to digest food for energy supply induces the intestine development, which may occur with more intensity in the early stages of life (larval and juvenile stages). It may even involve the penetration of villi towards the lumen of the intestine, thereby increasing the organ's absorption surface. However, fasting seems to have the opposite effect, with regression for the intestinal villi observed. Similar results were observed in birds, reptiles, fish and mammals. For fish, Day reported that a reduction in microvilli size occurred soon after the onset of the fasting period and highlighted the relevance of this phenotypic adaptation to this environmental difficulty [2-6].

DISCUSSION

The decrease in liver weight and hepatocytes vacuolization over the fasting period indicates that juvenile (*P. scalare*) uses the energy reserves contained in this organ during these periods. Qian indicate that the use of metabolic pathways such as fat digestion and absorption, and glycolysis/gluconeogenesis during fasting periods. As in the present experiment, Gaucher observed no decrease in liver weight in *Hyphessobrycon luetkenii* subjected to nine days of fasting, but when these fish were subjected to 16 days of fasting, a significant difference in liver weight was observed [7,8].

The reduction of the HSI in the relation of fasting indicates that this variable is directly related to the use of the fish's energy reserves. This hypothesis has also been put forward for other species. Quero suggested, regarding Locariichthys platymetopon, that there was a direct relationship between HSI and the accumulation of energy reserves. These authors suggest that the increase in HSI is a consequence of a strategy used by this species to overcome periods of stress during winter. Power demonstrated that there was a rapid decrease in HSI values among juvenile sea bream (Sparus aurata) during fasting and correlated this variable with the use of liver storage energy [9,10].

The results of the rapid reduction of HSI values, followed by stability, may indicate that for juveniles freshwater angelfish, this energy reserve is used in short periods of fasting, but if the fasting persists, another energy source should be used. Regarding this other energy source, the intestinal mass loss may indicate that visceral fat is one of the energy sources when fasting periods become too long.

Other authors have already reported a decrease in visceral fat in an increased fasting period. Gaucher reported a decline in mean visceral fat values in *Hyphessobrycon luetkenii* subjected to nine and sixteen days of fasting, but these authors mentioned that despite the decline in mean values, no significant difference was observed between the fish subjected to fasting and those that were not. Vidal reported a significant difference in the levels of visceral fat in *Jenynsia multidentata* on fasting for seven days [8,11].

For the freshwater angelfish (P. *scalare*) juveniles subjected to the treatment T3,T6,T9 and T12, followed by 30 DF, the normalization of HSI showed the importance for the fish metabolism the maintenance of liver energy storage. It is important to note that after the feeding period, the vacuolization of hepatocytes was observed again. However, with the data of the present experiment, it is not possible to observe whether this is a natural condition of the species, or if this condition is caused by inadequate feeding in captivity. Also, even after 30 DF, the fish subjected to T15 treatment had low HSI values, demonstrating the severity of the fasting duration's damage.

CONCLUSION

According to our findings, it can be assumed that juvenile freshwater angelfishes have strategies (e.g. a decrease in liver and intestine weight) to resist 15 days of fasting. Also, further experiments should be carried out to understand what the natural condition of the species' liver is and how captive feeding is affecting these animals.

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