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Exogenous enzymes on the feeding of pirarucu *Arapaima gigas* Schinz. 1822 (Osteoglossiformes. Arapaimidae)

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ABSTRACT: The objective of this work was to verify the effect of exogenous digestive enzymes protease, lipase and amylase on the feeding of pirarucu. In a completely randomized design, we tested three levels of inclusion for both exogenous enzymes against a control commercial feed lacking any enzymatic enrichment. Inclusion levels were as follows: (Control) = no enzymes; (T1) = 0.1% Amylase; (T2) = 0.2% Amylase; (T3) = 0.4% Amylase; (T4) = Protease 0.1%; (T5) = Protease 0.2%; (T6) = Protease 0.4%; (T7) = Lipase 0.1%; (T8) = Lipase 0.2%; (T9) = Lipase 0.4% . After the end of the experiment it was possible to find a significant difference ($p < 0.05$) in relation to the Control treatment only in the zootechnical data of the treatments that added protease and lipase to the pirarucu feed (T4, T5, T6, T7, T8, T9). The use of exogenous digestive enzyme protease and lipase are indicating on the pirarucu feeding, as it positively influences its growth.

Key words: amylase; carnivorous fish; lipase; nutrition; protease

Enzimas exógenas na alimentação do pirarucu *Arapaima gigas* Schinz, 1822 (Osteoglossiformes, Arapaimidae)

RESUMO: O objetivo deste trabalho foi verificar o efeito das enzimas digestivas exógenas protease, lipase e amilase na alimentação do pirarucu. Em um delineamento inteiramente casualizado foram testados três níveis de inclusão para ambas as enzimas exógenas frente a uma ração comercial controle ausente de qualquer enriquecimento enzimático. Os níveis de inclusão foram os seguintes: (Controle) = sem adição enzimas; (T1) = Amilase 0,1%; (T2) = Amilase 0,2%; (T3) = Amilase 0,4%; (T4) = Protease 0,1%; (T5) = Protease 0,2%; (T6) = Protease 0,4%; (T7) = Lipase 0,1%; (T8) = Lipase 0,2%; (T9) = Lipase 0,4%. Após o término do experimento foi possível encontrar diferença significativa ($p<0,05$), em relação ao tratamento Controle, nos dados zootécnicos dos tratamentos que adicionaram protease e lipase a alimentação do pirarucu (T4, T5, T6, T7, T8, T9). As enzimas digestivas exógenas protease e lipase são indicadas na alimentação do pirarucu, influenciando positivamente no seu crescimento.

Palavras-chave: amilase; peixe carnívoro; lipase; nutrição; protease

Introduction

The pirarucu, *Arapaima gigas*, native to the Amazon basin, is a large-sized fish of carnivorous feeding habit, being able to reach 3 m in length and 200 kg of weight in the nature, geographically distributed in the Amazon region and occurring in the floodplain areas of the rivers Araguaia-Tocantins, Solimões-Amazonas and its affluents, Amazon river in the Peruvian Andes and tributaries from the Essequibo and Rupununi rivers of Guyana (Andrade-Porto et al., 2015). The absence of intramuscular spines, the average fillet yield of 57%, the light-colored, lean, tender and high quality meat made this fish a species of high demand and market value (Mesquita, 2017), but due to its carnivorous feeding habit, it is not unrelated to the nutritional issue that this type of fish presents. Its protein requirement is high (between 40 and 45% CP); however, it has good feed conversion when the management is adequate (Marinho et al., 2015). It also presents gregarious association and absence of cannibalism, a fact that allows its storage in high densities (Embrapa, 2015).

According to the Brazilian Institute of Geography and Statistics (IBGE, 2016), omnivorous species dominate the Brazilian aquaculture scenario, and the carnivorous ones that appear best placed in the productive ranking are the surubim and its hybrids (3.1%) and the pirarucu (1.7%). Although the species have an interesting commercial value, much of this low result in relation to the omnivores can be explained by the fact that carnivorous cultivation requires, beyond the greater difficulties in relation to the feeding training, a higher productive cost with a feed that requires high levels of crude protein (Moreira et al., 2013). That is, a decrease in production costs for carnivores could lead to a popularization and consequent increase in the supply of these fish in the national market.

Ingredients from animal origin (using fishmeal as an example) are traditionally used in the feed for carnivorous species because they are protein sources that meet the requirements of these species for factors related to the palatability, digestibility and essential amino acids requirement. Due to it being one of the most expensive ingredients used in fish feed, the partial substitution of this animal protein source by a vegetable one is always a subject for researchers that may bring greater profitability and sustainability to the fish production chain, considering that reducing costs with artificial feeding assists the process of reducing production costs, thus making this activity more commercially attractive (Hill et al., 2015).

Regarding piscivorous fish species such as the pirarucu, Yamamoto et al. (2017) have replaced, in increasing levels, fishmeal with wheat flour, which is considered a low cost vegetable ingredient, although it does not have a balanced profile of essential amino acids. The substitution of fishmeal by vegetal origin sources stumbles, for example, upon antinutritional characteristics present in soybean, a vegetable ingredient rich in protein and widely used in commercial fish feed. Soybean, in its whole, is a hemoglutinant (trigger of red blood cells agglutination processes) and antitrypsic (characteristic that inhibits the action of trypsin) (Oba-Yoshioka et al., 2015). With this increase in the sources of vegetable protein in feed, it is automatically incorporated higher amounts of starch in the feeding of carnivorous species, making it mandatory the monitoring of the liver ability in producing enzymes and storing glycogen (Souza et al., 2014).

One of the tools for making it possible to improve the feed digestibility and better utilization of nutrients is through the inclusion of exogenous enzymes in the diet of non-ruminants. Its effectiveness has been demonstrated, specially, for less digestible cereals in relation to food of better digestibility (Wallace, 2015). In addition to the previously cited objectives, enzymatic supplementation favors the reduction of nitrogen and phosphorus levels in the fish feces and increases the action of endogenous enzymes, contributing to the increase of amino acids available in the body for natural protein synthesis (Gomes et al. 2016).

Unlike herbivorous and omnivorous fish, the carnivores have limited physiological resources for the use of carbohydrates that seek in fulfilling an energetic function in commercial diets. The inclusion of non-protein energy sources reduces the need of protein use sources to fulfill energy functions, shifting these protein sources to the growth of animals, in a metabolic phenomenon also known as “protein sparing effect” (Gonçalves, 2014).

The need in substituting protein from animal sources to vegetable ones as an ingredient in the feed formulation is a priority for ensuring the aquaculture sustainability and the inclusion of exogenous digestive enzymes can help in the nutrient utilization processes of enriched feed with protein sources from vegetable origin. Therefore, the objective of this work was to verify the effect of the exogenous digestive enzymes protease, lipase and amylase in the feeding of the pirarucu.

Materials and Methods

This work was carried out at the facilities of the Aquaculture Research Coordination (CPAQ) of the National Institute of Amazon Researches (INPA), Manaus, Amazonas, Brazil, under Protocol No. 0820180039 approved by the Committee on Ethics in the Use of Animal (CEUA) of the Federal University of Western Pará - UFOPA.

For the feeding of the pirarucu juveniles, the following additions of exogenous protease, lipase and amylase were tested: (Control) = no enzymes addition; (T1) = 0.1% Amylase; (T2) = 0.2% amylase; (T3) = 0.4% amylase; (T4) = 0.1% protease; (T5) = 0.2% protease; (T6) = 0.4% protease; (T7) = 0.1% lipase; (T8) = 0.2% lipase; (T9) = 0.4% lipase. The protease and exogenous amylase were obtained from the fungus *Aspergillus oryzae*, with lipase being obtained from the *Aspergillus niger*.

The experiment was conducted in a completely randomized design with 10 treatments (including the control group) and 4 replicates, totaling 40 experimental units. The experimental units were formed by plastic tanks of 310 L (250 L of useful volume) with an approximate discharge of 5 L h^{-1}.

The fish were initially weighed, measured and homogeneously distributed in each of the experimental units. In each of those units, 10 pirarucu juveniles of 6.6 ± 0.5 g and 10.0 ± 0.1 cm of total length were stocked, being fed until apparent satiety, 5 times per day (08:00 a.m., 11:00 a.m.; 14:00 p.m., 17:00 p.m. and 19:00 p.m.) for a period of 37 days.

The feed used in the experiment was commercial and specific for carnivorous fish, with 45% of crude protein and 3,000 kcal of raw energy kg^{-1} of feed, extruded and triturated (Table 1). In order to compare with the data provided by the commercial manufacturer, a bromatological analysis was carried out at the Fish Nutrition Laboratory of the Aquaculture Research Coordination/CPAQ from the National Institute of Amazon Researches (INPA).

The water quality was monitored once a day (17:00 p.m.) by evaluating the following physicochemical parameters: total ammonia (mg L^{-1}), nitrite (mg L^{-1}), pH, temperature (ºC), dissolved oxygen (mg L^{-1}) and conductivity (µS cm^{-1}) of the water.

At the end of the experiment, the fish were weighed and measured for verifying the treatments effect on their performance. From the obtained results, the following zootechnical performance indexes were calculated: Weight gain (WG) = final weight –initial weight; Mean feed consumption at the end of the experiment (MFFi) = Σ Mean daily consumption (per experimental tank); Apparent feed conversion (AFC) = MFFi / (Final mean weight – Initial mean weight); Specific growth rate (SGR) = 100*(Ln Mean final weight – Ln Mean initial weight)/time (days) and survival (S) = 100*(fish No. at the end of the experiment/fish no. at the beginning of the experiment).

In order to guarantee the fish homogeneity at the beginning of the experiment, they were weighed (g) and shortly after that, the data analyzed by means of the "F" Test at 5% probability. The data of the treatments final biometry were subjected to an analysis of variance and the means compared by the Tukey test at 5% of probability (Bhujel, 2009).

Results and Discussion

Exogenous enzymes in the zootechnical performance of pirarucu juveniles

The inclusion of exogenous protease and lipase in the feed for pirarucu juveniles showed significant and positive differences in relation to the control treatment ($p < 0.05$) (Table 2). Recent and similar results were also achieved by Yildrim & Turan (2010), Zamini et al. (2014) and Adeoye et al. (2016), being this latter work important in determining the importance of phytase for the mitigation of antinutritional factors related to phytate, an agent capable of reducing the bioavailability of proteins, amino acids and minerals from the ingredients present in the diet.

In general, the exogenous enzymes use in non-ruminant diets allows the inclusion of ingredients that the animals would not normally take advantage of, improving the apparent digestibility and reducing the amount of certain nutrients in their excreta, which in turn ends up contributing to the quality maintenance of the environment. Exogenous enzyme complexes are widely used in various agricultural sectors, since the feeds are formulated with different ingredients, requiring enzymes that have the degradation capacity of the chemical compounds present in these feeds (Gomes et al., 2016).

Diógenes et al. (2018), evaluating the exogenous supplementation effect of enzymes, concluded that the enzymatic complexes Synergen® and Natugrain® increased apparent digestibility, the lipase and protease enzymatic activity and were also important in enhancing the richness and diversity of the intestinal microbiota, being then

Table 1. Centesimal composition of the commercial feed for carnivorous fish with 45% protein and 3000 kcal of raw energy kg^{-1} extruded.

Information	Humidity (%)	Data at 100% of dry mass					
		CP (%)	EE (%)	CH_2O (%)	Ashes (%)	CF (%)
CPAQ[1]	8.9	46.5	10.5	30.6	10.0	2.4
Manufacturer[2]	13.0	45.0	14.5	20.5	14.0	6.0

[1] Bromatological analysis carried out at the Fish Nutrition Laboratory of the Aquaculture Research Coordination/CPAQ from the National Institute of Amazon Researches/INPA; [2] Values supplied by the feed manufacturer; CP = Crude protein; EE = ethereal extract (fat); CH_2O = Carbohydrate; CF = Crude fiber.

Table 2. Initial and final weight, weight gain, specific growth rate, apparent feed conversion and survival of pirarucu *Arapaima gigas*) juveniles (maintained under experimental conditions and fed with supplementation of exogenous amylase, protease and lipase.

ZI	Control	0.1% Amylase	0.2% Amylase	0.3% Amylase	0.1% Protease	0.2% Protease	0.3% Protease	0.1% Lipase	0.2% Lipase	0.3% Lipase
IW (g)	6.0 ± 0.4	6.8 ± 0.5	6.9 ± 0.4	6.5 ± 0.6	6.0 ± 0.5	6.3 ± 0.7	6.2 ± 0.3	6.4 ± 0.8	6.3 ± 1.0	6.6 ± 0.9
FW (g)	16.7 ± 2.1^{a}	21.5 ± 5.0^{a}	22.8 ± 3.7^{a}	22.3 ± 2.1^{a}	25.9 ± 4.5^{b}	25.6 ± 2.5^{b}	26.0 ± 5.6^{b}	26.6 ± 3.4^{b}	24.3 ± 5.3^{b}	26.5 ± 2.4^{b}
WG (g)	10.7 ± 2.0^{a}	14.7 ± 5.2^{a}	15.9 ± 4.0^{a}	15.8 ± 1.9^{a}	19.9 ± 4.7^{b}	19.3 ± 2.3^{b}	19.8 ± 5.7^{b}	20.2 ± 3.9^{b}	18.0 ± 5.1^{b}	19.9 ± 2.7^{b}
SGR (%)	2.8 ± 0.3^{a}	3.1 ± 0.7^{a}	3.2 ± 0.5^{a}	3.3 ± 0.2^{a}	3.9 ± 0.5^{b}	3.8 ± 0.3^{b}	3.9 ± 0.6^{b}	3.9 ± 0.5^{b}	3.6 ± 0.9^{b}	3.7 ± 0.4^{b}
AFC	1.8 ± 0.3^{a}	1.4 ± 0.5^{ab}	1.1 ± 0.2^{ab}	1.2 ± 0.1^{ab}	0.8 ± 0.1^{b}	0.8 ± 0.1^{b}	0.8 ± 0.1^{b}	0.8 ± 0.3^{b}	0.9 ± 0.1^{b}	0.8 ± 0.1^{b}
SR (%)	82.5 ± 9.5^{a}	95.0 ± 5.8^{b}	82.5 ± 9.6^{a}	77.5 ± 17.0^{a}	63.3 ± 15.3^{ab}	80.0 ± 17.3^{a}	66.7 ± 1.5^{ab}	85.0 ± 12.9^{a}	62.5 ± 22.1^{ab}	77.5 ± 9.5^{a}

[1] Means followed by the same letter, in the lines, do not differ between themselves by the Tukey "t" test at 5% of probability (p > 0,05); [2] Percent values were transformed by the arcsine before the means comparison; (IW) = initial weight; (FW) = final weight; (WG) = weight gain; (SGR) = specific growth rate, shown in %; (AFC) = apparent feed conversion; (SR) = survival rate, shown in %.

recommended the use of these enzymatic complexes together with the artificial feeding indicated for *Scophthalmus maximus* juveniles with carnivorous food habits.

However, there are natural enzymatic complexes that allow a zootechnical performance as good as those commercially applied. Khati et al. (2015) decided to test the papain present in the papaya leaf (*Carica papaya*) and in the milky solution present in fruit that are not yet ripe as an enzymatic source in the diet of *Labeo rohita* fingerlings. After 90 days of experimentation, the treatment that used 10 g of papain per kilo of feed was more efficient, showing more than twice as much weight gain when in comparison with the control treatment. This growth is justified by the fact that papain has the power to hydrolyze lipids, carbohydrates and proteins in a wide range of pH and temperature, in addition to minimizing antinutritional factors that larvae and fingerlings have difficulty in coping. Artificial microparticulate diets have 60 to 90% of dry matter, while zooplankton, a natural food during this growth phase, has only 10%. That is, food supplementation with natural enzymatic complexes tends to facilitate the digestion of the feed offered during this growth phase.

The use of balanced feed in concomitance with additives translates into the only viable path in search of an increasingly intensive and competitive aquaculture production. With each passing day, fish cultivations tend to intensify production, generating an ever-increasing amount of biomass in an increasingly smaller area or volume. Therefore, it is imperative that the commercial feed of both carnivores and omnivores are able to meet the nutritional requirements of the species, taking into account the ingredients that form a balanced feed, their respective nutrients and the apparent digestibility of the manufactured commercial diet (Gomes et al., 2016).

However, the inclusion of exogenous amylase did not influence the performance of the pirarucu in a way that the experimental units presented significant differences in comparison with the control treatment ($p > 0.05$) (Table 2).

Carbohydrates, along with lipids, are the major non-protein energy sources for fish diets; both are also cheaper than protein ingredients and more available for diets formulation (Moreira et al., 2013). The use of starch as an energy source will depend directly on the ability of the animal in synthesizing amylase, the enzyme responsible for the starch digestion. Carnivorous species fed for long periods with feed containing high levels of carbohydrates develop a restrictive condition in relation to the growth and high accumulation of glycogen in the liver, being able to cause losses in situations of stress generated by cultivation in intensive systems (Santos et al., 2013). This tendency in not being able to efficiently use exogenous amylase to improve zootechnical results may be associated with the carnivorous food habit of the pirarucu, since that in carnivorous fish species the enzymatic profile and microbiota are more restricted than in omnivores (Liu et al. , 2016).

Moro et al. (2015), determining the ideal relationship between starch and lipids and their influence on the digestive enzymatic profile of dourado (*Salminus brasiliensis*), found that a starch:lipid ratio greater than 1:1 impaired the fish performance, despite the greater inclusion of carbohydrates having resulted in a better utilization of the available protein and energy.

In a study conducted by Gioda et al. (2017), the carnivorous species traíra (*Hoplias malabaricus*) presented low enzymatic activity in the digestive tract for amylase and maltase. The results of this study postulate that this enzyme activity depends directly on the natural diet of the animal and their ability in metabolizing carbohydrates.

Song et al. (2018), carrying out a similar work, subjected the carnivorous species *Micropterus salmoides* to isoprotein and isolipidic diets with four different sources of starch: wheat starch, cassava starch, pea starch and an amylose-enriched corn starch. Despite presenting the best zootechnical results for the diet with pea starch, no significant difference was found in the centesimal muscle composition of the animals. Regarding the enzymatic activity, *M. salmoides* demonstrated adaptive power to the different sources of starch, thus making it possible to affirm that the starch catabolism seems to be regulated by its composition.

In the last decade, the increase in the price of fishmeal has increasingly promoted the inclusion of corn gluten and wheat gluten in the feeding of carnivorous species, being the use of the latter also favored by the absence of antinutritional factors present in the chemical composition of most vegetables and that cause interference in its nutrients use. Other factors that should be taken into account when adding starch sources in diets are related to the humidity content, pesticide residues, seed toxicity and the presence of mycotoxins (Ribeiro et al., 2010; Lewandowski et al. 2017).

Water quality

The concentrations of total ammonia ($NH_3 + NH_4$) and nitrite (NO_2^-), and the measurements of temperature, pH, dissolved oxygen and water conductivity in the experimental tanks did not show any variations ($p > 0.05$) which could point the influence of these environmental parameters on the performance of pirarucu juveniles (Table 3).

The observed pH during the experimental period was between 4.8 and 5.1, lower than the one observed by Drumond et al. (2010) and Tavares-Dias et al. (2010) in breeding sites located in Amazônia municipalities, respectively Manaus and Manacapuru.

The potential of hydrogen, commonly known as pH, translates into a numerical and logarithmic scale that determines the acidity or basicity of a liquid, and for aquaculture, it is important to determine the chemical conditions of the growing environment. If pH levels are too low or too high, damage to aquatic animals occurs in both cases, and at low levels causes irreversible damage to the gills, skin and eyes, and at levels above normal may lead to a greater diffusion of Na^+ and Cl^- ions, causing disturbances on the natural mechanisms of osmoregulation (Lekang, 2013).

Table 3. Mean values ± standard deviation of the water quality parameters during the experimental period of pirarucu juveniles (*Arapaima gigas*).

Parameters	Control	Amylase 0.1%	Amylase 0.2%	Amylase 0.3%	Protease 0.1%	Protease 0.2%	Protease 0.3%	Lipase 0.1%	Lipase 0.2%	Lipase 0.3%
pH	5.0 ± 0.1	4.9 ± 0.1	4.9 ± 0.2	4.8 ± 0.1	4.9 ± 0.2	5.1 ± 0.1	4.9 ± 0.2	4.9 ± 0.1	5.1 ± 0.2	5.1 ± 0.3
Temp.	26.7 ± 0.4	27.0 ± 0.3	26.8 ± 0.3	26.9 ± 0.3	26.5 ± 0.3	26.6 ± 0.2	27.0 ± 0.1	26.8 ± 0.1	26.8 ± 0.4	26.7 ± 0.3
Cond.	34.7 ± 5.7	44.7 ± 18.4	45.9 ± 15.5	31.9 ± 4.2	31.9 ± 3.1	39.8 ± 6.9	36.4 ± 8.9	40.5 ± 8.4	32.8 ± 1.1	44.8 ± 10.7
DO	5.3 ± 0.3	5.0 ± 0.3	4.9 ± 0.6	5.0 ± 0.2	5.4 ± 0.2	4.9 ± 0.1	5.2 ± 0.2	4.9 ± 0.3	5.1 ± 0.1	5.2 ± 0.2
TA	0.3 ± 0.06	0.3 ± 0.1	0.4 ± 0.1	0.3 ± 0.05	0.4 ± 0.1	0.4 ± 0.1	0.3 ± 0.1	0.3 ± 0.1	0.3 ± 0.08	0.3 ± 0.2
NIT	$4.1^{-3} \pm 10^{-3}$	$4.1^{-3} \pm 2.1^{-3}$	$5.1^{-3} \pm 3.1^{-3}$	$4.1^{-3} \pm 2.1^{-3}$	$4.1^{-3} \pm 2.1^{-3}$	$5.1^{-3} \pm 4.1^{-3}$	$3.1^{-3} \pm 2.1^{-3}$	$4.1^{-3} \pm 4.1^{-3}$	$3.1^{-3} \pm 10^{-3}$	$5.10^{-3} \pm 3.1^{-3}$

(pH) = potential of hydrogen; (Temp.) = temperature, in Celsius degrees; (Cond.) = electrical conductivity, in S cm^{-2}; (DO) = dissolved oxygen; (TA) = total ammonia, in mg L^{-1} of NH_3 + NH_4; (NIT) = nitrite, in mg L^{-1}.

It is also worth noting that environments with high pH levels tend to present a higher amount of non-ionized ammonia, a more toxic chemical form among those generated by the protein catabolism of most aquatic organisms (Campos et al., 2012; Hurtado et al. 2018).

Another important limnological parameter in experimental studies is the electrical conductivity of the water, measured in µS cm^{-2}. The conductivity is the ability of the aqueous medium to transmit electric currents, and one of the most important factors for characterizing a water destined to the cultivation of aquatic organisms, also directly related to the presence of ions, salts and total dissolved solids (Rocha et al., 2017). The electrical conductivity observed during the experiment was limited to the interval between 31.9 and 44.8 µS cm^{-2}, characterizing that the used water is within acceptable parameters for the cultivation of amazonian fish (Barroncas et al., 2015) and within the limits established by Tavares-Dias et al. (2010) for a semi-intensive cultivation of pirarucu.

The temperature at which the experimental units were subjected fluctuated from 26.5 to 27.0 °C, not showing significant statistical difference between them, and being slightly below of what was used by Hoshino et al. (2017), but within the interval measured by Oliveira et al. (2012). According to Hill & Lawson (2015), the optimal temperature for pirarucu growth vary from 24.0 to 31.0 °C, not tolerating temperatures below 16 °C. As it is a pecilothermic animal, its body temperature will vary as a result of the changes in the environment temperatures, possessing a tolerance or performance curve related to some parameters, such as: fecundity, growth, metabolic rate or swimming capacity. That is, the closer to the optimum temperature is the aquatic environment found, the more efficient the metabolic activities associated with the homeostasis process in aquatic organisms are, including in this group the pirarucu (Schulte, 2011).

Breathing is an essential process for maintaining any vertebrate that requires aerobic metabolic reactions, which includes fish and their basic conditions for growth, swimming, reproduction, immune system, among other factors. Fluctuations and/or low levels of DO during long periods may hinder the degradation of the organic matter present in the aquatic environment, compromising many of the physiological functions of aquatic organisms, generating difficulties for the development of the basic cellular biochemical processes for maintaining the homeostasis of these animals (Cech Jr & Brauner, 2011).

The concentration of dissolved oxygen found in this experiment (around 5.0 mg L^{-1}) is the recommended for most species of cultivable aquatic organisms (Lekang, 2013). However, the pirarucu, which tolerates very well reduced concentrations of oxygen and has air breathing, obtains most of the O_2 through its swim bladder and is naturally found under hypoxic conditions (Hill & Lawson, 2015). Analyzing these information, it can be considered that daily variations in the concentration of dissolved oxygen do not influence significantly in an environment destined for the cultivation of the species.

The concentrations of total ammonia and nitrite measured during the experimental period were similar to those found by Cavero et al. (2003) and Tavares-Dias et al. (2010). The determination of total ammonia and nitrite levels are important in aquaculture due to excretion capacity reduction and for producing an asphyxiation state in fish, respectively. The total ammonia in the culture water is the result of the nitrogenous compounds that form the proteins and, therefore, are crucial for the fruitful growth of aquatic organisms. A very high amount of total ammonia in the culture water will make it difficult for the fish to eliminate these compounds, thus ending up accumulating them in their tissues. However, nitrite, a chemical compound generated by the incomplete nitrification of total ammonia to nitrate, is toxic for the fish due to its combination with hemoglobin, originating something known as methemoglobin, a substance unable to keep performing the plasmatic tasks of its predecessor responsibility, making it impossible to continue the O_2 transport service through the blood vessels with the same efficiency (Hurtado et al., 2018).

Conclusion

The exogenous digestive enzymes protease and lipase, when added to feed, improve the weight gain, growth rate and feed conversion of pirarucu juveniles.

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