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ANIMAL SCIENCE

Ractopamine supplementation in the diet of pintado amazônico during the final growth phase

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Abstract: The objective of this unprecedented research was to evaluate performance traits, carcass yield, meat quality, and hematological parameters of pintado amazônico fish (*Pseudoplatystoma reticulatum* × *Leiarius marmoratus*) fed diets containing ractopamine (20 mg kg^{-1}) for different periods, during the final growth phase. The following treatments were evaluated: 45 days without ractopamine supplementation (control diet); 30 days of control diet, followed by 15 days of ractopamine supplementation; 15 days of control diet, followed by 30 days of ractopamine supplementation; and 45 days of ractopamine supplementation. Performance traits, carcass yield and hematological parameters were not significantly influenced by the ractopamine supplementation periods. The pH, L* color and a* color parameters of the filet post-slaughter and the L* color of the filet post-thawing were significantly influenced by the period of supplementation with the additive in the diet. Cooking loss was significantly lower in the fillet of fish that received ractopamine. Ractopamine supplementation at the level of 20 mg kg^{-1} in the diet of pintado amazônico for 15, 30, or 45 days does not alter their production performance, carcass yield, or hematological parameters, but changes the qualitative traits of their filet.

Key words: Beta-adrenergic agonist, *Learius marmoratus*, nutrition, *Pseudoplatystoma reticulatum*.

INTRODUCTION

Pintado amazônico (*Pseudoplatystoma reticulatum* × *Leiarius marmoratus*) has been increasingly cultured on a commercial scale in South American countries, mainly in Brazil (Souza et al. 2017). However, farmers have been known to report large amounts of body fat in this fish species, especially in the abdominal region. Fat may affect the taste, appearance, and final quality of fish meat (Pezzato et al. 2004).

In an effort to minimize the fat content and improve carcass quality, additives have been included in the diets of some species, e.g., swine. For this purpose, farmers have used ractopamine, a β-adrenergic agonist whose results have been demonstrated in several meta-analytic studies (Apple et al. 2007, Kiefer & Sanches 2009, Pompeu et al. 2017), including positive results in performance and decreased fat deposition in the carcass without compromising meat quality.

Despite the low number of studies, evidence suggests that, as in swine, ractopamine can also reduce fat in the carcass of fish. Positive results for muscle and mesenteric fat reduction, and weight gain have been reported in channel catfish (Mustin & Lovell 1993). Recent studies also indicate a decrease in the fat content of pacu (*Piaractus mesopotamicus*) filets (Oliveira et al. 2014).

Due to the great genetic variability between the different fish species, it has not yet been possible to establish the ideal level and period of supplementation with ractopamine to cause a reduction in body fat and improve production performance indices. For swine, however, studies show a positive effect with 5 mg kg^{-1} of the product during 28 days. In fish, inclusion levels range from 4 mg kg^{-1} (Neto et al. 2017) to 100 mg kg^{-1} (Mustin & Lovell 1993), with positive results in fish with supplementation up to 20 mg of ractopamine kg^{-1} (Mustin & Lovell 1993, Vanderberg et al. 1998, Oliveira et al. 2014).

Response to dietary ractopamine supplementation may be altered by the duration of the supply (Vanderberg et al. 1998). Ractopamine supplementation in fish diets has usually been used for a period between 28 (Mustin & Lovell 1993) and 60 days (Bicudo et al. 2012). Research results show a rapid response for growth and feed eficiency during the first seven to ten days (Mills 2002) and can mantain good response for up to five weeks of supplementation (Armstrong et al. 2004). However, due to its high cost, ractopamine has been supplemented in periods of less than four weeks (Oliveira et al. 2013). In addition, after 28 days of supplementation, slow reduction of the responses may occur due to the phenomenon called down-regulation or ß-adrenergic receptor desensitization (Moody et al. 2000).

There are no studies investigating ractopamine in diets for pintado amazônico and the elevated fat content in the carcass of this fish at slaughter. Therefore, the present study was undertaken to examine performance traits, carcass yield, meat quality, and hematological parameters of pintado amazônico fish fed diets containing ractopamine during the final growth phase.

MATERIALS AND METHODS

The experiment was conducted in Campo Grande - MS, Brazil (20°49'96" S and 54°61'46" W). The fish used in the study belong to the surubim group, originating from the cross between cachara females (*Pseudoplatystoma reticulatum*) and jundiá males (*Leiarius marmoratus*). The fish were acquired from a commercial fish farm located in Terenos - MT, Brazil (20°25'57" S and 55°17'11" W), and housed in 4-m^3 net cages until reaching a weight close to slaughter weight. Research on animals was conducted according to the institutional committee on animal use (approval no. 905/2017).

Eighty fish were distributed into eight 4-m^3 net cages (ten animals per cage), in a 150-m^2 excavated pond with 10% daily water exchange. The net cages were interspersed and were 1.5 m apart between the vertices. Prior to the start of the experiment, the fish were allowed an acclimation period of 15 days during which they received a commercial feed twice daily (9:00 and 17:00) without ractopamine inclusion. The experiment was conducted during the months of March and April, totaling 45 days of experimental period.

The experiment was set up as a completely randomized design with four treatments (days of ractopamine supplementation) and two replicates (net cages) with ten fish per experimental unit. The following treatments were evaluated: 45 days without ractopamine supplementation (control diet); 30 days of control diet, followed by 15 days of ractopamine supplementation (20 mg kg^{-1}); 15 days of control diet, followed by 30 days of ractopamine supplementation (20 mg kg^{-1}); e 45 days of ractopamine supplementation (20 mg kg^{-1}). In all treatments, the fish were fed twice a day (9:00 and 17:00).

The level of 20 mg of ractopamine kg^{-1} defined in the experiment with pintado amazônico was based on the studies with fish evaluating ractopamine supplementation between 0 and 10 mg kg^{-1} (Haji-Abadi et al. 2010), between 0 and 40 mg kg^{-1} (Vanderberg et al. 1998), between 0 and 45 mg kg^{-1} (Oliveira et al. 2014), and between 0 and 100 mg kg^{-1} (Mustin & Lovell 1993). Fatty acid reduction was observed at 10 mg kg^{-1} (Vanderberg et al. 1998) and 11.25 mg kg^{-1} (Oliveira et al. 2014). Moreover, in the work of Mustin & Lovell (1993) with channel catfish, the authors observed an improvement in production performance and fat reduction (muscle and mesenteric) of fish supplemented with 20 mg of ractopamine kg^{-1} for 28 days (no differences were found between the levels of 20 and 100 mg of ractopamine kg^{-1}). In addition, we chose to evaluate the maximum permited level for pigs in Brazil, which is 20 mg kg^{-1} (Brasil 2008). The period of ractopamine supplementation in the experiment was defined considering that the supplementation in many studies with fish has ranged from 28 (Musting & Lovell 1993) to 60 days (Bicudo et al. 2012).

The average initial weight of the fish was 1.43±0.352 kg and their average length was 47±3.36 cm, in a total initial biomass of 3.56 kg m^{-3}. The maximum biomass per net cage was estimated at 5.62 kg m^{-3}. This value was calculated by dividing the maximum biomass of the excavated pond (180 kg) by the number of net cages and by the size in m^2. The average final biomass of each cage (4.44 kg m^{-3}) was lower than the calculated maximum value. Each net cage was used in this experiment only with the purpose of separating the different periods of supplementation with the additive.

Ractopamine was incorporated into the commercial diet using starch as an excipient, which was also added to control diet. The fish were fed twice daily (morning and afternoon) with an extruded feed for carnivorous fish. The nutritional composition of the experimental diets (Table I) was analyzed by NIRS (near-infrared reflectance spectroscopy).

The amount of feed supplied to the fish was based on the total biomass of each net cage (approximately 1.5% of the biomass), water temperature, and animal behavior at the time of feeding. These observations helped to maintain the feed supply homogeneous across the treatments.

Throughout the experimental period, the water quality traits were analyzed daily in two areas of the excavated nursery (water inlet and outlet). Water temperature, dissolved oxygen and pH were evaluated using a multiparameter (YSI ProPlus). A blower was used as a source of aeration whenever the dissolved oxygen level

Table I. Analyzed composition of the diets fed to the pintado amazônico fish.

Analyzed composition	Diet with ractopamine	Basal diet
Crude protein (%)	41.0	41.3
Ether extract (%)	11.1	12.3
Crude fiber (%)	3.8	3.6
Mineral matter (%)	7.6	7.5
Dry matter (%)	92.8	95.6
Starch (%)	29.1	31.4
Ractopamine (mg kg^{-1})	20.0	-

was less than 2 mg L^{-1}. In addition to these analyzes, water samples were taken weekly from the tank to measure total ammonia nitrogen and nitrite (Alfakit).

Biometric measurements were performed at the start and end of the experimental period to obtain the growth results. Body weight (g) and standard length (from the anterior extremity of the head to the start of the caudal fin, in cm) were measured. Weight gain (g) was calculated based on the initial and final weights.

During the last biometric measurement, blood was also harvested by puncturing the caudal vein, in Vacutainer® tubes. The fish were anesthetized with Eugenol (Biodinâmica Química e Farmacêutica Ltd) at a concentration of 50 mg L^{-1} until reaching the stage of surgical anesthesia, as recommended by Ross & Ross (1999). Subsequently, the harvested blood was centrifuged (Excelsa baby II, Model 206-R; 15 min, 3,500 rpm) and the serum was harvested to determine the concentrations of glucose, cholesterol, and triglycerides. The method used for serum extraction followed the procedures performed by Drumond et al. (2018).

At the end of the experimental period, all animals were slaughtered for analyses of body yield and meat quality traits. Yields (of eviscerated fish; back filet with skin; belly filet with skin; and total filet), abdominal fat, and visceral fat were evaluated. The body yields were calculated by dividing the weight of the measured trait by the final live weight of the fish and multiplying the result by 100 (results expressed as %).

The meat quality traits (pH, L* color, a* color, b* color, cooking loss, and shear force) were evaluated by following the protocol proposed by Fantini et al. (2015) for surubim. After the filet attributes were analyzed post-slaughter (fresh filet), the filets (identified after slaughter) were frozen at a temperature of –16 °C for seven days. After this period, they were placed in a cold chamber to thaw at 0 ± 2 °C for 24 h for post-freezing evaluations.

Intramuscular pH was measured using a digital pH meter (HI 99163, Hanna, with a specific electrode for meats) at three points in each filet sample, considering the average of the results obtained in the three measurements. These analyses were performed on filets post-slaughter and post-freezing.

Parallel to the analysis of pH, the filet color was measured using a colorimeter (CR-400, Konica Minolta) adopting the CIELAB system, which defines the L* [lightness, ranging from 0 (black) to 100 (white)], a* [green (–60) to red (+60)] and b* [blue (–60) to yellow (+60)] parameters. These analyses were performed on filets post-slaughter and post-freezing.

Cooking loss was determined as the difference in weight of the samples before and after cooking. For this test, the samples were weighed raw and then cooked in a conventional electric oven at a temperature of 170 °C. The temperature was monitored by a thermometer inserted into the geometric center of the samples. Upon reaching 42 °C, the samples were turned over and kept in the oven until reaching a final temperature of 72 °C. After cooking, the samples were weighed again to calculate the amount of fluids lost during cooking. These analyses were carried out on filets post-freezing.

For the analysis of shear force, after cooking, the filets were wrapped in polyethylene film and chilled at 5 °C for 24 h. This variable was determined using a texturometer (CT3, Brookfield) (25-kg capacity, shearing speed of 3.3 mm s^{-1}) equipped with a Warner Bratzler blade. These measurements were performed on the right filet of each species post-freezing, collecting the maximum possible number of samples (2 cm × 1 cm; at least 4 subsamples per animal). The average value of the subsamples

of each filet was considered the shear force of each filet sample.

Centesimal composition analyses were performed on the fresh filet (without pre-drying) to determine the following components: dry matter (DM), organic matter (OM), mineral matter (MM), crude protein (CP), and ether extract (EE). Samples were ground, homogenized, and analyzed for the concentrations of DM, OM, MM, CP, and EE in accordance with the methodology proposed by the AOAC (2000). The centesimal composition of the filets was analyzed in triplicate, using the filet of three fish from each replicate.

The data were evaluated statistically by analysis of variance followed by Student's t test for mean comparison. The ANOVA - Analysis of Variance model was:

$$Y_{ij} = \mu + \tau_j + \varepsilon_{ij},\ i = 1,...,4;\ j = 1 \text{ or } 2$$

in which Y_{ij} is an observation in treatment i and replication j; μ is the overall mean; τ_j is the effect of treatment i; ε_{ij} is the random error.

RESULTS

The mean values of water temperature, dissolved oxygen and pH were 25.5 °C, 3.5 mg L^{-1} and 7.5, respectively. Concentrations of 0.52 mg L^{-1} of total ammonia nitrogen and 0.0 mg L^{-1} of nitrite were observed. The ractopamine supplementation (20 mg kg^{-1}) periods did not influence (P>0.05) the performance traits or carcass yield of pintado amazônico (Table II).

The filet pH post-slaughter was lowest (P<0.05) in the fish that did not receive the additive, whereas the highest pH value (P<0.05) was found in those supplemented for 30 days. There was no significant difference for pH in the post-freezing filet across the different periods of supplementation with ractopamine (Table III).

Ractopamine supplementation for 45 days provided a higher filet L* color post-slaughter (P<0.05) than the other treatments. However, the highest (P<0.05) L* color value of the filet post-freezing was observed in the animals which had not received the additive (Table III).

The a* color component of the filet post-slaughter was highest (P<0.05) in the fish fed the diet containing ractopamine for 30 days. The a* color of the fish post-freezing, in turn, was not influenced by the different periods during which the fish consumed the additive. No difference was detected for the b* color values of the filets post-slaughter or post-freezing (Table III).

Cooking loss was lower (P<0.05) in the fish supplemented with ractopamine (regardless of the period of supply) than in control fish. No differences were found for shear force (P>0.05) between the supplementation periods (Table III).

As regards the centesimal composition of the filets, the period of supplementation with ractopamine affected (P<0.05) only the percentage of dry matter. The lowest dry matter percentage was observed in the fish which received ractopamine for 30 days (25.11%), whereas the highest dry matter content was obtained when the fish consumed ractopamine in the periods of 0 (26.63%) and 15 days (26.60%). The other chemical components (mineral matter, organic matter, ether extract, and crude protein) were not influenced by the supplementation period (Table IV).

The blood glucose, triglycerides, and cholesterol levels in the fish were not affected (P>0.05) by the ractopamine supplementation periods (Table V).

Table II. Production performance and carcass yield of pintado amazônico supplemented with ractopamine (20 mg kg^{-1}) for four different periods.

Variable	Period of ractopamine inclusion in the diets (days)				P-value[1]	CV
	0	15	30	45		
Final weight (g)	1809.95	1802.05	1740.25	1763.10	0.61	3.16
Weight gain (g)	365.36	351.75	314.00	356.88	0.74	14.14
AFC	2.50	2.33	3.03	2.42	0.40	15.34
EFY (%)	92.19	91.45	91.29	91.95	0.82	1.16
YBAFS (%)	40.21	40.11	40.83	40.00	0.40	1.16
YBEFS (%)	14.28	13.21	14.18	14.33	0.12	2.75
YWFS (%)	54.49	53.33	55.02	54.34	0.08	0.82
AF (%)	1.59	1.48	1.38	1.72	0.80	18.38
VF (%)	0.06	0.09	0.12	0.08	0.31	24.10
Viscera weight (g)	84.40	84.22	81.39	82.91	0.85	4.60

Period of ractopamine inclusion in the diets: 0: 45 days without ractopamine supplementation (control diet); 15: 30 days of control diet, followed by 15 days of ractopamine supplementation (20 mg kg^{-1}); 30: 15 days of control diet, followed by 30 days of ractopamine supplementation (20 mg kg^{-1}); and 45: 45 days of ractopamine supplementation (20 mg kg^{-1}). CV - coefficient of variation. AFC - apparent feed conversion; EFY - eviscerated fish yield; YBAFS - yield of back filet with skin; YBEFS - yield of belly filet with skin; YWFS - yield of whole filet with skin; AF - abdominal fat; VF - visceral fat. [1]Significant P-value in the analysis of variance (P<0.05).

Table III. Meat quality characteristics of pintado amazônico supplemented with ractopamine (20 mg kg^{-1}) for four different periods.

Variable	Period of ractopamine inclusion in the diets (days)				P-value[1]	CV
	0	15	30	45		
pH_{PS}	6.13c	6.36b	6.58a	6.29b	0.0014*	0.60
L^*_{PS}	47.35b	47.85b	46.96b	49.89a	0.0127*	1.00
a^*_{PS}	0.80b	0.68b	2.68a	1.91ab	0.0287*	29.22
b^*_{PS}	1.34	1.46	2.67	2.22	0.1217	24.35
pH_{PT}	5.99	6.02	6.09	6.06	0.0509	0.40
L^*_{PT}	52.83a	52.20ab	51.13b	50.97b	0.0497*	0.94
a^*_{PT}	1.81	1.84	2.74	1.36	0.1398	23.23
b^*_{PT}	6.75	7.21	7.12	6.42	0.5401	8.20
CL (g)	60.00a	51.70b	50.00b	50.33b	0.0137*	3.36
SF (kg)	0.690	0.701	0.676	0.639	0.4716	5.57

Period of ractopamine inclusion in the diets: 0: 45 days without ractopamine supplementation (control diet); 15: 30 days of control diet, followed by 15 days of ractopamine supplementation (20 mg kg^{-1}); 30: 15 days of control diet, followed by 30 days of ractopamine supplementation (20 mg kg^{-1}); and 45: 45 days of ractopamine supplementation (20 mg kg^{-1}). CV - coefficient of variation. pH_{PS} - pH value post-slaughter; L^*_{PS} - filet lightness post-slaughter; a^*_{PS} - filet a* color post-slaughter; b^*_{PS} - filet b* color post-slaughter; pH_{PT} - pH value post-thawing; L^*_{PT} - filet lightness post-thawing; a^*_{PT} - filet a* color post-thawing; b^*_{PT} - filet b* color post-thawing; CL - water loss from cooking post-thawing; SF - shear force post-thawing. [1*]Significant P-value in the analysis of variance (P<0.05).

Table IV. Apparent centesimal composition of filets of pintado amazônico supplemented with ractopamine (20 mg kg^{-1}) for four different periods.

Variable	Period of ractopamine inclusion in the diets (days)				P-value[1]	CV
	0	15	30	45		
Dry matter (%)	26.63a	26.60a	25.11c	25.79b	0.01*	0.81
Mineral matter (%)	3.61	3.57	3.91	3.61	0.83	9.11
Organic matter (%)	96.38	96.42	96.08	96.38	0.83	0.34
Ether extract (%)	4.56	4.57	4.34	3.33	0.62	20.20
Crude protein (%)	16.70	18.83	17.74	18.71	0.20	4.37

Period of ractopamine inclusion in the diets: 0: 45 days without ractopamine supplementation (control diet); 15: 30 days of control diet, followed by 15 days of ractopamine supplementation (20 mg kg^{-1}); 30: 15 days of control diet, followed by 30 days of ractopamine supplementation (20 mg kg^{-1}); and 45: 45 days of ractopamine supplementation (20 mg kg^{-1}). CV - coefficient of variation. 1*Significant P-value in the analysis of variance (P<0.05).

Table V. Blood glucose, triglycerides and cholesterol levels of pintado amazônico supplemented with ractopamine for four different periods.

Variable	Period of ractopamine inclusion in the diets				P-value[1]	CV
	0	15	30	45		
Glucose (mg dL^{-1})	84.37	72.75	91.61	81.47	0.30	10.23
Triglycerides (mg dL^{-1})	414.42	438.17	385.74	423.72	0.84	14.32
Cholesterol (mg dL^{-1})	140.00	122.40	138.27	137.50	0.45	8.22

Period of ractopamine inclusion in the diets: 0: 45 days without ractopamine supplementation (control diet); 15: 30 days of control diet, followed by 15 days of ractopamine supplementation (20 mg kg^{-1}); 30: 15 days of control diet, followed by 30 days of ractopamine supplementation (20 mg kg^{-1}); and 45: 45 days of ractopamine supplementation (20 mg kg^{-1}). CV - coefficient of variation. 1*Significant P-value in the analysis of variance (P<0.05).

DISCUSSION

Water quality characteristics remained stable and adequate for the development of the pintado amazônico throughout the experiment. According to the review presented here about the use of ractopamine in fish, this is the first study to investigate the use of this additive in diets for pintado amazônico (*L. marmoratus* × *P. reticulatum*) during the final growth phase.

The results for characteristics of performance and carcass yield obtained in the presente study are similar to those found by Neto et al. (2017) in Nile tilapia at the end of the growth phase with supplementation of 0, 4, 8, 12 and 16 mg of ractopamine kg^{-1} for 31 days. Similarly, Devens et al. (2012), did not find positive results for the performance traits of Hungarian common carp (*Cyprinus carpio*) with initial weight (18.64 ± 1.25 g) after supplementation with 7, 14 and 21 mg of ractopmaine kg^{-1} for 56 days; and Guimarães et al. (2017) in tambaqui (*Colossoma macropomum*) with weight in the final phase of growth (1.00 ± 0.04 kg) also found no improvement in performance with 30 days supplementation with 0, 2.5, 10 and 20 mg of ractopamine kg^{-1}. In contrast, Mustin & Lovell (1993) observed a 17% increase on the weight gain in channel catfish (*Ictalurus punctatus*) with weight in the initial phase (156 g) after supplementation with 20 mg of ractopamine kg^{-1} in relation to

the control diet (without ractopamine) for 28 days (they did not find differences between the supplementations of 20 and 100 mg kg^{-1}). Thus, it can be inferred that the effects of ractopamine can vary according to the rearing phase and the species, given that 20 mg kg^{-1} ractopamine supplementation was sufficient to improve the production performance of channel catfish, but not to improve the production performance of pintado amazônico, as well as the levels of 16, 20 and 21 of ractopamine kg^{-1} did not improve performance of Nile tilapia, tambaqui and Hungarian common carp, respectively.

The level of ractopamine used in the present project was based on the works mentioned above, but it did not result in improvement of the performance of pintado amazônico. These findings may indicate that diferent fish species have distinct physiological and metabolic responses to ractopamine supplementation. There are few studies with ractopamine in tropical freshwater fish of importance for South America and, therefore, more work with fish is necessary, since the data indicate that the dose and/or period of supplementation were not sufficient to act on performance and carcass yield traits of pintado amazônico.

Neto et al. (2017) tested the ractopamine doses of 4, 8, 12, and 16 mg kg^{-1} in Nile tilapia in the final growth phase and observed no influence on filet yield, which has been corroborated in the present study with pintado amazônico. The eviscerated-fish and filet yields are found here are similar to those found by Souza et al. (2017) in the same species, in diets not supplemented with the additive. For this trait to improve in ractopamine-supplemented fish, a higher dose or a longer period are possibly necessary.

Positive results for muscle and mesenteric fat reduction in fish were reported by Mustin & Lovell (1993), who worked with channel catfish (*Ictalurus punctatus*) juveniles with an initial weight of 156 g supplemented with diets containing 20 and 100 mg kg^{-1} ractopamine in relation to non-supplemented fish (there was no significant difference between the levels of supplementation). However, there is a trend for fat deposition to increase along with animal weight and age. This fact raises the hypothesis that ractopamine quantities larger than 20 mg kg^{-1} cause the amount of body fat in pintado amazônico during the final growth phase to decrease. Another suggested hypothesis is that some part of ractopamine may be lost by solubilization in the water when the feed is supplied, which may indicate a need for higher supplementation doses.

Although ractopamine did not lead to improvements in the percentage of visceral fat, the obtained values (lower than 2%) were much lower than the 3.28±1.49% and 4.81±1.76% reported by Souza et al. (2017) for diets containing 32% and 40% crude protein, which resulted in final weights of 1,016.63±250.59 and 1,152.22±338.50 kg, respectively. It should be stressed that variations in protein level, or even variations in the calorie:protein ratio of the diets, may lead to an increase or reduction of visceral fat and, in situations of nutritional imbalance that causes increase in visceral fat, ractopamine might provide a better metabolic action. Visceral fat levels observed by Souza et al. (2017) in pintado amazônico were similar to those obtained by Guimarães et al. (2017) for tambaqui (above 3%), and these authors observed 13.4% reduction of visceral fat in fish supplemented with 20 mg of ractopamine kg^{-1} compared to non-supplemented fish. Differences in abdominal fat in the magnitude of 13.4% would only be significant with more than five repetitions per treatment.

In this study, the differences found in the pH values of the filet post-slaughter did not remain post-freezing, suggesting that this trait

did not affect final filet quality. Both pH values were close to the 6.17±0.23 and 6.39±0.10 found by Fantini et al. (2015) in surubim fish reared in net cages and in nurseries, respectively.

The higher L* color of the filet post-slaughter found in the fish supplemented with ractopamine for 45 days may be interesting from the sales perspective, as it indicates that filets from these fish are lighter in color, when fresh. However, in the filets post-freezing, a higher L* color value was noted in those which did not consume the additive.

The post-slaughter filet of the fish fed the diet containing ractopamine for 30 days showed a redder color, as higher a* values mean redder meat (Fantini et al. 2015). This variable may be associated with the amount of myoglobin present in the muscles (Maia & Ogawa 1999). After thawing, no difference was detected for the a* color values.

The filet b* color values both post-slaughter and post-freezing found in this experiment were lower than those reported by Fantini et al. (2015) in surubim. This difference indicates that even though the fish belong to the same genus and the species used in the crosses are similar, their meat quality may be different, which is often reported by consumers.

Considering the present results for cooking loss, it can be stated that ractopamine supplementation may result in a juicier filet, which is a desirable feature to the final consumer. By contrast, ractopamine supplementation did not alter shear force, whose values were similar to those obtained by Fantini et al. (2015) in a study with another surubim species.

The lowest dry matter value observed in the filets of fish fed ractopamine for 30 days (25.11%) and the highest value found for non-supplemented fish (26.63%) and that were supplemented with ractopamine for 15 days (26.60%) disagree with the observations of Neto et al. (2017), who found no significant differences in the dry matter of the filets of Nile tilapia fed diets containing ractopamine in the final growth phase (average 23.60% dry matter). The different dry matter values between the present study and the study of Neto et al. (2017) indicate that responses to ractopamine supplementation vary greatly between species.

Similar results for blood biochemical parameters evaluated in pintado amazônico, supplemented with ractopamine and non-supplemented, contrast with those reported by Bicudo et al. (2012) in pacu, in which they observed blood glucose reduction with ractopamine supplementation (10, 20 e 40 mg kg-1) for 60 days; by Vanderberg et al. (2008) in rainbow trout (*Oncorhynchus mykiss*) in which they observed an increase in glucose concentration with ractopamine supplementation (20 mg kg-1 for 56 days; and 5, 10, 20 and 40 mg kg-1 for 84 days), and decrease in the concentration of non-esterified fatty acids with 10 mg of ractopamine kg-1 in the diet for 28 days (supplementation with 5 and 20 mg kg-1 for 28 days increased the concentration of esterified fatty acids); by Haji-Abadi et al. (2010) with rainbow trout, in which they noticed an increase of triglycerides when supplemented with 10 mg kg-1 in the diet for 56 days; and by Guimarães et al. (2017) with tambaqui in which they observed decreased triacylglycerol concentration in fish supplemented with 5 mg of ractopamine kg-1 in the diet for 30 days compared to non-supplemented fish and to fish supplemented with 0, 2.5, 10 and 20 mg of ractopamine kg-1. The contrasting results in these studies indicate different responses for biochemical parameters to ractopamine supplementation in different species, period and dose of supplementation with ractopamine.

Ractopamine supplementation at 20 mg kg^{-1} in the diet of pintado amazônico for 15, 30,

or 45 days does not change their performance, carcass yield traits, or blood traits. Ractopamine slightly affects filet color and pH. Ractopamine supplementation for 30 and 45 days increases filet moisture.

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