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Original Article

Zootechnical indices and digestibility in juveniles of tambaqui *Colossoma macropomum* fed a diet containing particulate maize

Índices zootécnicos e digestibilidade em juvenis de tambaqui *Colossoma macropomum* alimentados com dieta contendo milho particulado

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Abstract

Three experiments were done with different particle sizes of corn feed on its zootechnical performance, passing rate and apparent digestibility of juvenile tambaqui (*Colossoma macropomum*). In the first, 200 juveniles were used and distributed in 20 tanks (220 L), 10 fish per unit (four replicates). The experimental system used to record passage time was composed of five incubators with 200 L. In the second, 75 juveniles were used per 55 days (three replications). In the third, 75 juveniles were used and distributed in five incubators of 200 L. All experiments were performed randomly. Different linear behavior treatments were observed for apparent digestibility of crude protein; and the smaller particle size (150 µm) had better results digestibility. smaller particle size of the corn had better results, affected the growth performance of tambaqui and the apparent digestibility of crude protein and ether extract. Thus, is recommended that a particle size of 150 µm of corn be used for tambaqui.

Keywords: fish nutrition, aquaculture, Myleinae, bromatological analysis, intestinal tract.

Resumo

Três experimentos foram realizados com diferentes tamanhos de partícula de ração para o desempenho zootécnico, taxa de aprovação e digestibilidade aparente de tambaqui juvenil (*Colossoma macropomum*). No primeiro, foram utilizados 200 juvenis e distribuídos em 20 tanques (220 L), 10 peixes por unidade (quatro repetições). O sistema experimental utilizado para registrar o tempo de passagem foi composto por cinco incubadoras com 200 L. No segundo, 75 juvenis foram utilizados por 55 dias (três repetições). No terceiro, 75 juvenis foram utilizados e distribuídos em cinco incubadoras de 200 L. Todos os experimentos foram realizados aleatoriamente. Diferentes tratamentos de comportamento linear foram observados para digestibilidade aparente da proteína bruta; e o menor tamanho de partícula (150 µm) apresentou melhor digestibilidade dos resultados. menor tamanho de partícula do milho apresentou melhores resultados, afetou o desempenho de crescimento de tambaquis e a digestibilidade aparente da proteína bruta e extrato etéreo. Portanto, recomenda-se o uso de um tamanho de partícula de 150 µm de milho para o tambaqui.

Palavras-chave: nutrição de peixe, aquicultura, Myleinae, análise bromatológica, trato intestinal.

1. Introduction

Feed processing techniques are important since they contribute to improving the digestibility of the substance and thus reflect in better growth performance, health and, consequently, lower environmental impact (Aride et al., 2007, 2016, 2018, 2020; Lima et al., 2020; Nascimento et al., 2020; Leal et al. 2020). The digestibility of feed is one of the most important aspects in the evaluation of feed (Sadiku and Jauncey, 1995) due to the question of biological efficiency (Hanley, 1987). According to Hepher (1988), feed digestibility depends on three main factors: the diameter of the particles that make up the feed, as these become more or less susceptible to the action of digestive enzymes depending on the size of the particles; the activity of these enzymes; and the exposure time of food to the digestive system.

Food digestion efficiency can be influenced by, among other factors, better surface exposure to the digestive secretions, as well as by alterations in the time food takes

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to pass through the gastrointestinal tract (NRC, 1993). Since the grinding of the ingredients reduces the size of the particles, and thus increases the area exposed to the action of digestive enzymes during the animal's digestion process (Monticelli et al., 1996) it may also reduce the speed of the feed's passage through the gastrointestinal tract (Hayashi et al., 1999; Meurer et al., 2005). However, the studies relating the particle size of the ingredient in the feed with zootechnical performance, digestibility and passage rate through the intestines are still scarce in literature.

Therefore, the main reasons for adequate grinding of the ingredients can be summarized as follows: increase the exposure of the food to the action of digestive enzymes, improve digestion and absorption of nutrients and, consequently, improve feed conversion; meet the preference of different animal species and livestock categories of zootechnical interest, thus helping to maximize food intake and optimize productivity according to their particularities (Couto, 2008).

Fish farming, especially farming of the tambaqui (*Colossoma macropomum)* and other aquatic organisms, has significantly and ascendingly participated in the production of animal protein. Due to increased aquaculture production, it is important to make best use of ingredients that are considered fundamental in formulation of feeds. Since feed accounts for most of the operating costs, it is therefore an important aspect to study. The study of processing technologies becomes important in order to improve how the animals benefit from the ingredients in animal feed.

One technology that can be considered is the reduction or the determination of the particle size of the ingredient required for a specific species or developmental stage. This has already been tested and applied in poultry and swine farming, and can be an applicable technique for better results in fish farming. This aspect is crucial in fish feeds since a fine particle size is a prerequisite for good stability of extruded diets in water, which, as a result, increases fish feed efficiency (Kubitza, 1997). This is doubly important for omnivorous species and since it is essential to meet the nutritional needs of these fish effectively.

2. Materials and Methods

The growth performance experiments, body composition, passage rate and digestibility of feed in tambaqui were conducted in the aquaculture sector of the Support Unit for Research in Animal Science at Center for Agricultural Science and Technology/ Norte Fluminense State University (CCTA/UENF), situated in Campos de Goytacazes, Rio de Janeiro state, Brazil (21° 42'38.3" 41° 20'26.5"W).

2.1. Materials – Experiment 1

The experimental system for the development of growth performance and body composition (Exp. 1) consisted of 20 circular plastic tanks, each with a volume of 220 L, and a closed recirculation water input and output system. The filtering system of these tanks were performed by a mechanical filter (acrilon and expanded clay) and by a biological filter (plastic rings). The system also had a 36W ultraviolet filtering lamp for elimination of microorganisms.

The pump used in the system had a 4200 L.h^{-1} capacity, and the flow rate in the experimental tank was set for approximately 1.83 L.min^{-1}, so that in a 24-hour period, the entire volume of the tank was exchanged approximately 12 times. To maintain the temperature at approximately 28 °C, the water of the experimental system passed through a heat exchanger (Heliotek®).

2.2. Materials – Experiments 2 and 3

The experimental system for the ingestion rate and digestibility of feed, experiments two and three, respectively, were composed of five glass fiber incubators with a volume of 200 L. These had water input and output at the top, and a closed system for water recirculation. The collection and observation of feces were facilitated due to the fact that the incubator had a conical bottom and that this bottom was linked to a transparent container where feces accumulated. Control of the water flow was monitored using a hydraulic log between the incubator and the stool collector system (Guelph system).

The filtering system of these incubators assisted in maintaining the water quality, and was performed by mechanical filter (acrilon and expanded clay), bio filter (plastic rings), and had a 36 w lamp ultraviolet filter to eliminate microorganisms.

The boxes were equipped with continuous air flow from blowers, and vents were composed of cylindrical porous stones, placed 10 cm deep, preventing an increase in the leaching of feces due to air flow. Each 300 W incubator an automatic thermostat, regulated to maintain the water temperature at 28 °C.

2.3. Parameters

Water quality parameters for Experiment 1 were monitored once daily, however, in Experiments 2 and 3, dissolved oxygen (mg.L^{-1}), pH, temperature (°C) and ammonia (mg.L-1) were monitored twice a day, always before feeds. Oxygen levels were maintained with the aid of aerators and measured using a digital oxygen meter (YK Tlutron® 22DO ± 0.01), the pH was measured using a table digital pH meter (pH TEK®, PHS-3B ± 0.02), and the temperature using digital thermometer (oximeter). Ammonia in water was determined using the photometer, with ammonia parameter (Model HI 95 715, HANNA®).

2.4. Feed preparation

To obtain different particle sizes of maize, 40 kg of maize were used, which were milled in a hammer mill (Nogueira® 10 Hp) with 1 mm sieve, and subsequently ground to prevent clogging of the sieve and then dried in a ventilated oven at 65 °C for 24 h. For separation of different grain sizes, 200 g of ground maize was placed on a sieve shaker with intermittent pulse (MA 750, MARCONI®) for 10 minutes. The sieves were allocated in descending order from largest to smallest mesh grid (1000, 850, 710 500, 300, 150 m and bottom). Maize particle sizes used for preparation of experimental diets were 850, 710, 500, 300 and 150 μm.

A total of 10 kg feed, extruded in 2 mm pellets, was prepared for each particle size (treatment). The experimental diets (Table 1) were formulated according to the nutritional requirements of the species under study. The ingredients of the diets were weighed and homogenized in a type "Y" mixer in equal amounts for 12 minutes, totaling 10 kg of mixture for each treatment. For each extrusion, 300 grams were eliminated at the start of extrusion to prevent contamination of particle sizes with the prior mixture. For all experiments, five isoproteic (280.00 $g.kg^{-1}$ crude protein) and isocaloric (3,100 $kcal.kg^{-1}$ of digestible energy) experimental diets were used, differentiated only by the particle size of the maize.

2.5. Methodology

All animal measurements at the start and end of the experiments were carried out using a precision scale to 0.001g and an analog caliper rule measuring 6" x 150 mm (0.05 mm x 1/128"). In order to perform weighing and measuring, the fish were sedated with eugenol (5 mg. L^{-1}) in water (one part of eugenol oil to nine parts ethanol) to reduce animal stress during handling.

In Experiment 1, 200 juvenile tambaqui, aged approximately 50 days, were selected and standardized by size and weight. Then, 10 juveniles were randomly distributed to each of the 20 tanks. The experimental design was completely random, totaling 20 experimental units, with the following initial weights: T1 (150 μm) - 11.006 ± 0.037 g, T2 (300 μm) - 11.001 ± 0.023 g, T3 (500 μm) - 11.017 ± 0.024 g, T4 (710 μm) - 10.958 ± 0.020 g, T5 (850 μm) - 11.045 ± 0.020 g. Feeding took place three times a day (7:00, 13:00 and 17:00 hours), and the fish fed *at libitum* for 15 minutes. After the time limit, any excess was removed and stored in identified jars for subsequent correction of the amount of feed actually consumed by the fish. The monitoring of growth performance of the fish in the experiment lasted for 68 days.

In Experiment 2, 75 juvenile tambaqui, aged approximately 55 days, were selected and standardized by size and weight, and 15 juveniles were randomly distributed to each incubator, with the following initial weights: T1 (150 μm) - 12.003 ± 0.051 g, T2 (300 μm) - 11.980 ± 0.079 g, T3 (500 μm) - 12.123 ± 0.083 g, T4 (710 μm) - 12.109 ± 0.065 g, T5 (850 μm) - 11.899 ± 0.039 g. The beginning of the experiment started with an *at libitum* feed (7:00 am), in which the feed contained 1.5% chromium oxide. After the feed, any leftovers were removed and the fish were then observed every half hour until the verification of the fecal elimination stained with chromium oxide, which is indicative of the test food excretion.

In Experiment 3, 75 juvenile tambaqui, aged approximately 130 days, were selected and standardized by size and weight. Then 15 juveniles were distributed,, entirely at random, in each of the 5 digestibility incubators, with the following initial weights: T1 (150 μm) - 34,500 ± 2,551 g, T2 (300 μm) - 34.952 ± 2.752 g, T3 (500 μm) - 34.602 ± 2.823 g, T4 (710 μm) - 34.731 ± 2.825 g, T5 (850 μm) - 34.910 ± 2.698 g. The three repetitions of this experiment were made over time, namely, that is, each five days of a stool collection represented one repetition. The animals were fed at 7:00 am with 12 g of feed (satiation) after eating, the collection of feces was done throughout the day until 19:30 hours at intervals of 30 minutes, to prevent leaching of feces.

At the end of Experiment 1, all fish from each treatment were anesthetized with eugenol and sacrificed by thermal shock (2/3 to 1/3 of ice and water), the carcasses were triturated with an industrial processor, weighed and identified for later chemical analysis, the same was done with animals at the start of the experiment.

At the end of Experiment 3, the pots containing the stools which had been stored in the freezer were thawed and excess water was removed. All the feces from the same experimental unit were homogenized and packed into smaller pots (100 mL) and then identified.

The feed ingredients, the feed, animal carcasses and fish feces were dried in an oven at 65 °C for 48 hours and, subsequently, a final drying was performed at 105 °C for 24 h. After drying, the samples were weighed on a digital scale with an accuracy of 0.001 g, ground in a ball mill, identified and stored. After processing the sample, analysis was performed by adapting the methodology described by Silva and Queiroz (2002), for analysis of dry matter (DM), ethereal extract (EE), crude protein (CP) and mineral

Table 1. Experimental formula diet and amount of each ingredient in the different treatments.

Ingredients	Feed ($g. kg^{-1}$)	Crude Protein ($g. kg^{-1}$)	Quantity per treatment (g)
Corn	39.0	9.49	3900
Fish meal	30.0	54.86	3000
Soy Flour	16.5	44.71	1650
Wheat Flour	8.0	15.20	800
Soybean Oil	6.0	-	600
Premix*	0.5	-	50
Total	100	28.82	10000

*Premix fish full plus (amounts per kilogram of commercial product): vitamins A (1.000.000 UI); B_1 (1250 mg); B_2 (2500 mg); B_6 (1875 mg); B_{12} (3750 mcg); C (42000 mg); D_3 (500.000 UI); E (20.000 UI); K_3 (500 mg); folic acid (250 mg); panthoteic acid (5000 mg); biotin (125 mg); choline chloride (75000 mg); niacin (5000 mg); L-lisine aminoacid (20g); minerals Ca (240mg); P (78mg); Na (60g); Co (25 mg); Cu (2000 mg); Fe (13820 mg); I (100 mg); Mn (3750 mg); Se (75 mg); Zn (17500 mg); Fluoride (780mg); antioxidants (600 mg).

matter (MM). All analyses were performed in triplicate at the Zootechnical Laboratory (LZO) of the Center for Science and Agricultural Technology (CCTA/UENF).

With the result of the biometrics from Experiment 1, it is possible to calculate the increase in weight, total length, standard length, height and coefficient of length variation [CV = (standard deviation of the length/average length) x 100], the rate specific growth (TCE = [(ln weight time 1 - ln weight time 0) / time] x 100), the crude protein retention efficiency (ERPB or ANPU) = [(PBFC x PF) - (PBic x PI) x 100/CPB] and protein efficiency ratio (live weight gain/consumed crude protein). The parameters of the final production evaluated were as follows: survival (%), weight gain (GP = initial weight - final weight) and apparent feed conversion (CAA = feed intake/weight gain).

The passage rate of tambaqui submitted to different diets with different particle sizes of maize was evaluated in Experiment 2. In Experiment 3, the apparent digestibility of dry matter (CDAMS), ether extract (CDAEE), crude protein (CDAPB) and the amount of digestible protein in the food were evaluated. To calculate the apparent digestibility of dry matter, ether extract and crude protein, we used the following Formula 1:

$$CDA(\%) = 100 - \left(\frac{Xrecovered}{Xingested}\right) * 100 \qquad (1)$$

where: CDA = digestibility coefficient relative to the dry matter, Gross and ethereal extract protein; X = amount recovered (g.kg^{-1}) of MS, PB and EE recovered in feces; and X = amount ingested (g. kg^{-1}) of MS, PB and EE intake in the diet. And to calculate the digestible protein the following Formula 2 was used:

$$PD = CDAPB * PB \qquad (2)$$

where: PD = grams of digestible protein in the feed, CDAPB = apparent digestibility coefficient of PB; and PB (g. kg^{-1}) = Crude protein in the food.

Statistical analysis consisted initially in obtaining the means of the variables in the treatments (150, 300, 500, 710, 850 µm of particle size). Afterwards, regression equations between variables as a function of particle size equations were obtained, with the regression model chosen by analyzing the regression parameters. Analyses were performed by using the Statistical Analysis Systems and Genetic application (SAEG, version 9.2), and adopted a 5% level of significance.

3. Results

There was no significant difference between treatments (p> 0.05), and the observed standard deviations were low, indicating little change in parameters over the experiments. Table 2 shows the water quality values for Experiment 1, however, values in the other two experiments (2 and 3) were very close to those for Experiment 1, as shown in Table 2. This shows that the control in the experimental units was efficient and within recommended limits for juvenile tambaqui, thus not affecting the results of growth performance, gastrointestinal passage rate and digestibility of feeds when tested with maize of different particle sizes.

The average final weight, weight gain, feed conversion, total length, standard length, height, specific growth rate, protein efficiency ratio and crude protein retention efficiency of juvenile tambaqui were influenced by the different treatments (p <0.05), and the comparisons are presented in Table 3.

The different particle sizes of maize influenced the performance parameters of juvenile tambaqui. The average final length observed for the treatment with 150 µm was greater than in all other treatments (Table 3). By using regression analysis, it was found that there were statistical differences between the treatments (Figure 1 and 2). Juveniles who received treatment with a maize particle size of 150 µm had higher final weight and greater weight gain than those who received treatment with a particle size of 850 µm.

The survival rates reported in this study do not differ significantly (p>0.05), when the different treatments are considered. The survival rate of the tambaqui was 100% in all treatments, showing that the water recirculation system was efficient, and the physicochemical parameters of the water were within the comfort range for the species.

Through the regression analysis, significant difference was observed between treatments, as shown in Figure 3, whereas the feed conversion ranged from 0.982 ± 0.034 to 1.173 ± 0.036 between treatments, showing a tendency of poor conversion in treatments with larger particle sized maize.

With the help of the regression analysis, we noticed significant differences in feed intake between the different treatments. It was observed that the treatment with lower granulometry (150 µm) showed a total feed intake of 1976.9 g during the trial period, while treatment with the highest particle size is much lower at 1407.8 g for the same period (Table 3). Consumption per experimental unit for each treatment is shown in Figure 4.

Table 2. Physical-chemical parameters of the water during the trial period of 68 days.

Parameters	Maximum	Average	Minimum	*C.V. (%)
Temperature (°C)	28.86	28.40	27.30	3.9
Dissolved oxygen (mg.L^{-1})	6.0	5.4	4.7	5.5
pH	6.68	6.1	5.58	3.0
Total ammonia (mg.L^{-1})	0.15	0.11	0.08	18.8

*C.V. Coefficient of Variation.

Table 3. Average values of performance parameters on tambaqui fed diets with different particle size of corn in its composition.

Performance parameter	Particle size of corn (μm)					*C.V. (%)
	150	300	500	710	850	
Initial weight (g)	11.01	11.00	11.02	10.96	11.05	0.28
Final weight (g)	61.16	53.26	48.11	41.07	46.54	15.17
Weight gain (g)	50.15	42.26	37.09	30.11	35.50	19.4
Total feed consumption (g)	1976.9	1657.4	1576.5	1403.6	1407.8	14.66
Apparent feed conversion	0.99	0.98	1.07	1.17	1.00	7.66
Total final length (cm)	13.93	13.27	12.80	12.32	12.77	4.67
Standard final length (cm)	12.19	11.62	11.21	10.75	11.15	4.78
Final height(cm)	6.22	5.95	5.79	5.45	5.72	
Specific growth rate (%)	5.75	550	5.31	4.99	5.22	
Survival (%)	100	100	100	100	100	
Protein efficiency ratio	3.67	3.67	3.38	3.10	3.56	
PB Retention Efficiency (%)	82.71	83.46	76.89	73.68	78.41	

*C.V. Coefficient of Variation.

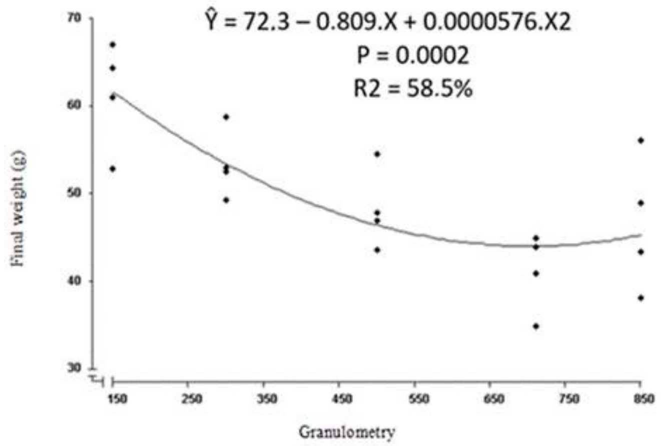

Figure 1. Regression Graph (Model Quadratic) for variable weight final after 68 days of experiment ($\hat{Y}$ = 72.3 – 0.809.X + 0.0000576X^2; (R^2 = 58.5%; *p*= 0.0002)).

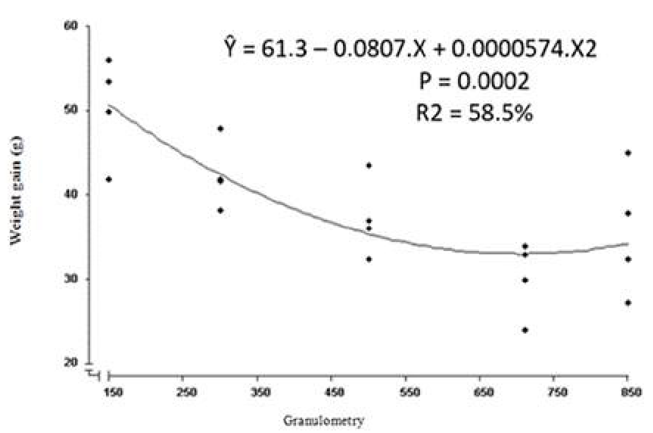

Figure 2. Regression Graph (Model Quadratic) for variable weight gain in the 68 days of experiment ($\hat{Y}$ = 61.3 – 0.0807.X + 0.0000574X^2 (R^2 = 58.5%; *p*= 0.0002)).

The best animal performance was recorded in the treatment that had smallest particle size of maize (150 µm) The curves of the specific growth rates (TCE), obtained by the regressions, showed a quadratic behavior (Figure 5), and 5.749 ± 0.094% was the best result.

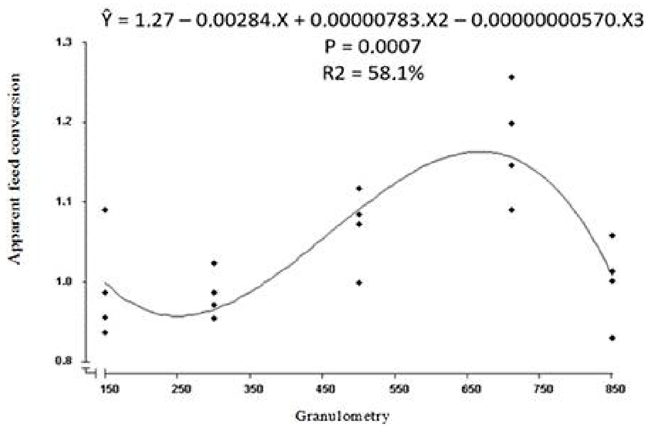

Figure 3. Regression Graph (Cubic Model) for apparent feed conversion variable after 68 days of experiment (Ŷ = 1.27 – 0.00284X + 0.00000783X^2 – 0.00000000570X^3; (R^2 = 58.,1%; *p*= 0.0007)).

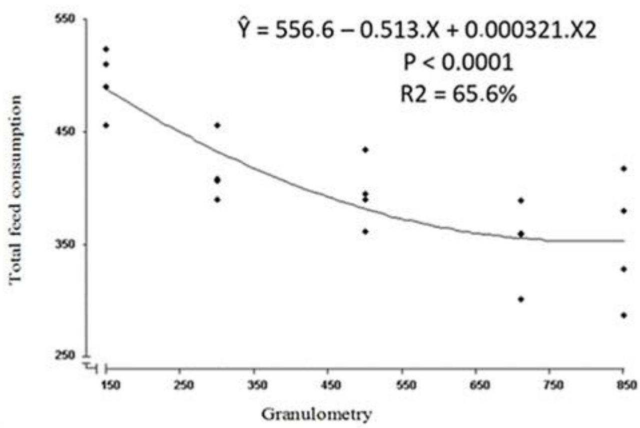

Figure 4. Regression Graph (Quadratic Model) for variable Total feed consumption in the 68 days of experiment (Ŷ = 556.6 – 0.513.X + 0.000321.X^2; (R^2 = 65.6%; *p*< 0.0001)).

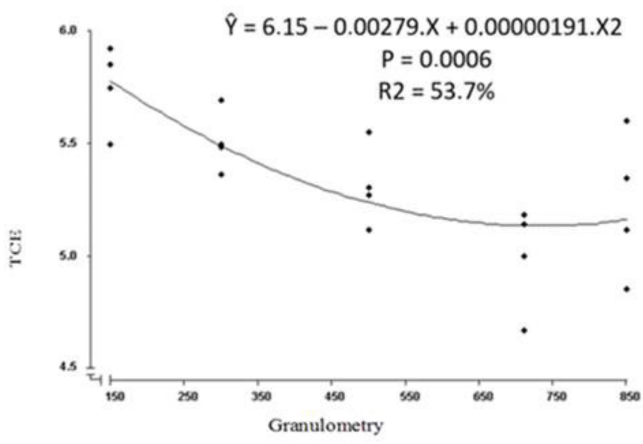

Figure 5. Regression Graph (Quadratic Model) for the variable specific growth rate (TCE) after 68 days of experiment (Ŷ = 6.15 – 0.00279.X + 0.00000191.X^2; (R^2 = 53.7%; *p*= 0.0006)).

The results of gastrointestinal passage rate of tambaqui fed diets with five different grain sizes of maize showed no differences ($p > 0.05$), and are presented in Table 4. The average dry matter (DM, crude protein (PB), ethereal extract (EE) and mineral matter of the five experimental models, where only the maize grain sizes varied, are presented in Table 5.

Table 4. Gastrointestinal passage time of tambaqui fed five diets with different particle size of corn.

Treatment	Granulometry (µm)	Gastrointestinal passage (h)
Incubator I	150	16.0a
Incubator II	300	16.0a
Incubator III	500	16.0a
Incubator VI	710	16.0a
Incubator V	850	16.5a

Averages followed by the same letter do not differ significantly from each other, at 5% probability per the Tukey test.

The average dry matter (DM), crude protein (PB), ethereal extract (EE) and mineral matter of feces of different treatments with five diets where only the maize grain sizes varied, are presented in Table 6. The averages of the apparent digestibility coefficients (CDA) of dry matter fractions (DM), crude protein (PB) and ethereal extract (EE) and the digestible protein of the five diets with different particle sizes of maize are presented in Table 7.

As shown in Figure 6, the values of apparent digestibility of crude protein (CDAPB) had linear behavior when the regression analysis was applied to the different treatments. And diets with the smallest particle size showed the best CDAPB.

4. Discussion

The behavior observed in growth performance variables analyzed in the present study are in accordance with Kubitza (1997), where it is suggested that ideally food should be ground to less than 0.5 mm. This similarity in

Table 5. Chemical composition of the experimental diets (dry matter).

Treatments	Dry Matter	Crude Protein	Ethereal extract	Mineral matter
T1 (150 µm)	96.77	28.47	8.47	10.15
T2 (300 µm)	96.83	28.49	7.94	9.30
T3 (500 µm)	96.78	28.66	8.59	9.75
T4 (710 µm)	97.08	28.60	8.22	9.69
T5 (850 µm)	96.95	29.04	7.21	9.66
*C.V. (%)	0.13	0.81	6.79	3.11

*C.V. Coefficient of variation.

Table 6. Chemical composition of feces (dry matter) collected in the experiment.

Treatments	Dry Matter	Crude Protein	Ethereal extract	Mineral matter
T1 (150 µm)	90.70	9.26	0.55	31.90
T2 (300 µm)	89.73	9.14	0.69	27.72
T3 (500 µm)	89.47	9.96	0.57	27.16
T4 (710 µm)	89.61	10.59	1.01	26.16
T5 (850 µm)	89.65	10.73	0.61	25.55
*C.V. (%)	0.55	7.35	27.51	9.00

*C.V. Coefficient of variation.

Table 7. The Apparent digestibility coefficients (CDA) of dry matter (CDAMS), crude protein (CDAPB) and Ethereal Extract (CDAEE) and digestible protein (PD) of the experimental diets.

Treatments	CDAMS (%)	CDAEE (%)	CDAPB (%)	PD (%)
T1 (150 µm)	62.6	94.16	67.45	19.20
T2 (300 µm)	73.3	91.27	67.90	19.34
T3 (500 µm)	75.5	93.30	65.22	18.69
T4 (710 µm)	76.9	87.63	62.97	18.01
T5 (850 µm)	75.2	90.99	63.04	18.31
*C.V.	7.94	2.76	3.58	3.04

*C.V. Coefficient of variation.

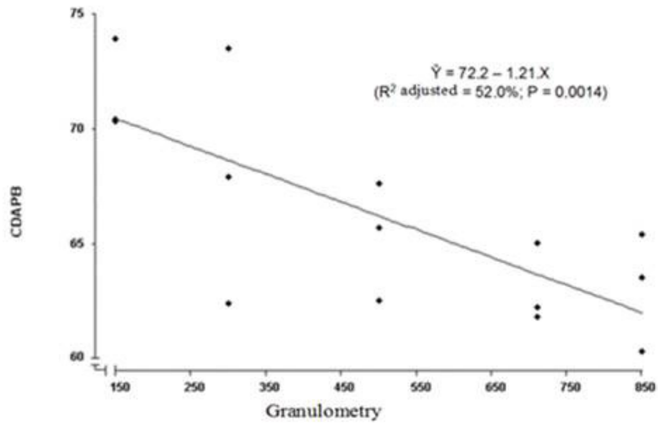

Figure 6. Regression Graph (linear model) for the variable coefficient: Apparent digestibility of crude protein for tambaqui fed diets with different particle size of corn ($\hat{Y}$ = 72.2 – 1.21.X; (R2 = 52.0%; *p*= 0.0014)).

findings can be seen in Figure 1, which shows the regression of final weight graph. The analysis shows a quadratic behavior, that is, in the range evaluated, the smaller the particle size of the maize, the greater the final weight of tambaqui. However, Kubitza refers to aquaculture in general, though in reality, one must evaluate and research specific species of fish and their establishment phase.

The improved performance of animals fed with the smallest particle size of foods can be explained by the studies of Zanotto et al. (1995), where they state that the food digestion efficiency can be influenced, among other factors, by exposing more of the feed's surface to the digestive secretions, as well as by the passage rate in the gastrointestinal tract. Therefore, smaller particle sizes may increase the feed's efficiency of the action of digestive enzymes and also improve the expansion of the starch in the extrusion process, improving the digestibility of both the ingredients and the feed.

In Nile tilapia (*Oreochromis niloticus*) evaluated different levels of grinding (500 µm and 1500 µm) in the growth phase, and they found that the lowest degree of grinding showed a better growth performance (Hayashi et al., 1999). The results of these authors corroborate this experiment, since the grinding of the feed, where a 150 µm sieve mesh was used, showed superior results for the performance of tambaqui.

However, and contrary to the report of the above authors, Pezzato et al. (2002) states that ingredients with a very low degree of grinding interferes with the selectivity of food by the animal, since in the larval stage, the fish can feed on finely ground food, however this becomes more difficult as it grows. This author worked with mash feeds and with the larval stage, different from our experiment that used extruded feed and animals with approximate initial weight of 11 grams.

For this experiment, when the weight gain variable was measured in isolation, it was noted that the higher weight gain values and final weight are found in the treatment with 150 µm particle size of maize. As for the apparent feed conversion (CAA), there was significant difference between treatments, and the feed conversion ranged from 0.982 ± 0.034 to 1.173 ± 0.036.

In working with the inclusion of exogenous enzymes such as amylase in juvenile tambaqui, found a feed conversion of 1.25 (Nunes et al., 2006). The CAA results found by Mendonça et al. (2012b), working with juvenile tambaqui supplemented with phytase in extruded feed were very close to those found in this experiment, with a range from 1.0 to 1.08 between the different treatments. This demonstrates that the diets responded well to the nutritional requirements for animal performance.

In an experiment to determine the size of the food particle for pacu and tambaqui, Cantelmo and Ribeiro (1994) observed that there is a positive correlation between fish performance, the particle size, mouth opening and fish standard length.

In regard to feed intake in our study, significant difference was observed between the different treatments. It was observed that the treatment with lower granulometry (150 µm) showed a feed consumption of 1976.9 g during the experimental period, while treatment with the largest particle size is much lower than a total feed intake during the trial period of 1407.8 g (Table 3). Given the aforementioned explanation, the species cultivation time may be reduced, since the animals under the 150 µm grain size feed treatment had a weight gain (Figure 2) 40% superior to the treatment with greater sized maize particles. Lemos et al. (2011) worked with coconut meal ground with the aid of 0.50 mm mesh sieves. When this was included in the juvenile tambaqui diet, they obtained a daily intake of 2.51%. This value highlights the efficiency of ground maize, as it allows the animal a better performance when compared to the other treatments.

When working with different protein sources and levels in feed for juvenile pacu 79-144 g, pointed out that the energy imbalance of the feed may limit the animal's consumption (Fernández et al., 2001). As such, the feeds in this experiment did not have energy imbalance, and even then, there was a significant difference in feed intake. Although they are of different species, we can emphasize the same food habit of the species, since the pacu is an omnivorous species and it also has easy adaptation to artificial feeding (Castagnolli and Zuim, 1985), similar to the tambaqui. In addition, both species make good use of plant-based ingredients, since in their natural habitats they would feed on fruits and nuts, among other items.

It was also observed that the best specific growth rate (TCE) was recorded in the treatment that had smaller maize particle size (150 µm) The TCE's curves obtained through regressions showed a quadratic behavior (Figure 5), and 5.749 ± 0.094% was the best result. These values are higher than those obtained by Coldebella and Radünz-Neto (2002), who recorded 3.5% and 4.23%, in tambaqui weighing between 4.0 and 6.0 g and nurtured for 120 days, while Salhi et al., (2004) states that, within 30 days of nurturing, specific growth rates ranging from 3.71 to 5.28% were obtained in young tambaqui weighing from 1.0 to 1.5 grams. In contrast, Vaz (2003), working with silver catfish (*Rhamdia quelen*) fingerlings weighing from 0.6 to 1.5 g and kept captive for 30 days, obtained specific growth rates ranging between 2.3% and 3.2%. Piedras et al. (2004), on the other hand, in a 33-day experiment, obtained TCE of 3.05 in 13 g tambaqui; while Lazzari et al. (2006) when working with 15 g juvenile silver catfish, found a specific growth rate of 3.6% and 3.8% by 60 days.

The high values of the TCE in this work can be explained through the high-quality ingredients in the diet formulation, and especially the smaller maize particle size. These factors may have favored the better gelatinization and expansion of starch during extrusion and also favored an increased feed intake and lower CCA, enabling better use of feed by animals.

Contrary to the present study, Mendonça (2007) had even lower TCE values (0.44%) and protein efficiency ratio (TEP) (1.65%) working with different photoperiod with juvenile tambaqui. These values correspond with the TCE values (0.43%) found by Burkert (2007) while working with different L-carnitine levels in feed for juvenile pacu. The protein efficiency ratio (TEP) uses values obtained in biometrics and chemical analysis and transforms the data for evaluation and quantification of the use of nitrogen supplied in the diet correlated with the animal protein production.

With the grinding of the feed ingredients below 500 µm, it can be inferred that, although the efficiency of digestion of foods improves with reduced particle size (Monticelli et al., 1996), there is a reduction in the passage rate through the digestive tract (NRC 1993; Hayashi et al., 1999). This may lead to restrictions on the animal's voluntary food intake, resulting in reductions in performance, such as reduced growth rates (Meurer et al., 2005). However, in this experiment, we observed the opposite of what is reported above, since the highest feed consumptions were in treatments with smaller particle size of maize and these obtained significant increases in the development variables of the tambaqui.

Conversely, for the feeds containing ingredients ground in larger mesh sieves, there is an increase in passage rates of the digestive tract and a lower ratio of contact surface area of the feed particles with digestive enzymes; these events decrease the efficiency of food digestion in animals (Meurer et al., 2005).

Evaluating the size of food particles for post-larvae common carp (*Cyprinus carpio*), concluded that the most appropriate is that of 125-500 µm (Hasan and Macintosh, 1992). In the present experiment, the diet compositions containing different particle sizes of maize did not significantly affect the passage rate through the tambaqui gastrointestinal system. It can be noted that although the length of the intestine is longer, 2.66 ± 0.52 times greater than standard length), the passage rate of vegetable materials through the intestine is relatively low, suggesting that in a 24-h cycle, the species can evacuate feces with nutrients from 3 to 4 meals.

In comparison with the data obtained by Dias-Koberstein et al. (2005), the gastrointestinal passage rate for tambaqui was greater than that for pacu (*Piaractus mesopotamicus*) at 27 °C (14 h), and lower at the temperature of 23 °C (36 h). This rate was lower than in juvenile African catfish (*Clarias gariepinus*), whose stomach emptying took place after 32 h when the temperature was 30°C (Hossain et al., 1998). The present experiment did not adversely affect the gastrointestinal passage on tambaqui fed diets with different particle size of maize, where there was no difference in water temperatures.

Silva et al. (2003), by working with tambaqui (*Colossoma macropomum*), found a passage time of 34 h for fish subjected to a temperature of 24 °C and 11 h for fish at a temperature of 28 °C, indicating that the passing food rate fell sharply as the temperature increased. Besides water temperature and crude fiber diet influencing the gastrointestinal passage rate, other authors also cite deprivation of food, intake of subsequent meals, food composition and animal physiology combination as other factors that influence the gastric evacuation rate (Salvanes et al., 1995).

In experiments with Nile tilapia, both concluded that high crude fiber levels in the diet decreased food passage rate in the gut and worsen the use of nutrients (Lanna et al., 2004).

The gastrointestinal passage rate between diets containing 2.50, 5.00, 7.50, 10.00 and 12.50% crude fiber decreased respectively from 13.32 h 12.45 h 11.49 h, 9.56 h and 11.22 h (Lanna et al., 2004). In the present experiment the same level of crude fiber in the diet was probably kept, since they had the same formulation, with only the particle size of the maize varying.

The rate found in this gastrointestinal passage experiment (16 h) was higher than that found by Zarate et al. (1999) when working with channel catfish, *Ictalurus punctatus* with an average weight of 70 g, temperature 28.0 ± 2, 0 °C. Their finding was 7 to 8 h. However, tambaqui, despite their long intestine, only required 8.8 hr to initiate the excretion after consuming diets containing 9.6% crude fiber (Silva et al., 2003). The longer times found in this experiment by Silva et al. (2003) may be due to low level of crude fiber in the diet because the ingredients used for formulation of rations in our study were of high quality.

The aforementioned data corroborate with those of other authors who claim that the passage of food through the stomach depends on temperature, postprandial time, size and type of food, weight, size and physiology of the animal, and stress levels (Salvanes et al., 1995).

In this experiment, all these factors were well controlled and only the particle size of the maize in the feed that differed between treatments, demonstrating that the experimental variables were well controlled and that the size of the maize particle did not affect the gastrointestinal passage rate of juvenile tambaqui.

Almeida et al. (2006) studied the protein digestibility of diets for tambaqui and could not perceive the reason for the low coefficient of digestibility in diets containing the lowest protein content, but the authors confirm that reducing the coefficient shown in the diet with 26.0% protein was because of this protein content level was above the needs required by the *Colossoma macropomum*.

On the other hand, values detected in this experiment were similar to those obtained by Almeida et al. (2006) with tambaqui and lower than those obtained by Pezzato et al. (2002). The CDAPB feed with 2.0 mm diameter for the pacu was similar (p <0.05) to 1.0 mm and higher than CDAPB of others (p <0.05). It was also noted that the feed's CDAPB food particles with diameters of 0.5 and 1.5 mm were similar (p <0.05) between themselves and between those of diameters of 1.0 and 2.5 mm. However, the 2.5 mm diameter and had the worst CDAPB, and although similar to the 0.5 and 1.5 mm, it was significantly (p <0.05) lower than that of 1.0 mm diameter (CDAPB of 85.01 ± 0.44) (Pezzato et al., 2002). Therefore, in this study, the best CDAPB values were observed in treatments with smaller particle sizes of maize, possibly due to increased exposure to nutrient uptake and enzymatic action in the gut.

Silva et al. (2007), working with tambaqui obtained an apparent digestibility of crude protein (CDAPB) of 83.57% for maize, which was similar to that found in wheat bran (82.87%). Pezzato et al. (2002) also found similarity between the protein fraction digestibility of maize and wheat bran, despite the divergence of the absolute values found, 91.66 and 91.13%, respectively. However, these values differ from those found by Furuya et al. (2001), which correspond to 87.12 and 78.21%, respectively. And, in the present study, they were even lower than those observed by the authors mentioned above, probably due to the fact that only the particle size of the maize was changed and all the other ingredients were kept at 1 mm particle size.

However, the PB digestibility coefficients of maize (95%), wheat bran (92%), soybean meal (96%) and fishmeal (92%) found by Cho and Bureau (1997) for the rainbow trout (*Oncorhynchus mykiss*) were high, yet similar to each other. The high-quality protein present in maize at the time of its preparation, even if in small doses, together with the milling process of the grains, exposes this protein to rapid digestion, which can justify the high values of apparent digestibility of maize protein in smaller particle sizes. However, the feed contained much of its protein derived from fish meal, which may affect the digestibility levels due to lack of information and standardization of fishmeal in the Brazilian market.

According to Pontes et al. (2010), fish meals available in the market exhibit great variation in quality, which can be attributed to freshness, type of material and processing conditions. Brazilian fish meal often has lower digestibility coefficients because they are formulated with harvest residues and may have high percentages of ash and low quality protein from the bone protein matrix, skin, scales and viscera (Boscolo et al., 2004), besides being a seasonal product that varies in its composition and changes the performance of animals fed with them (Oliveira Filho and Fracalossi, 2006).

These differences can be attributed partly to the crude fiber content of the experimental diets used to determine the digestibility of fractional protein, especially in diets containing wheat bran, found in literature. Several studies have shown that the variation of crude fiber levels in fish diets can change the digestibility, the gastrointestinal passage rate and the morphology of the digestive tract (Lanna et al., 2004). However, in our study, the crude fiber content in diets did not alter.

The ether extract's CDA determined in this study ranged from 87.63% to 94.16%, which are similar results to those obtained by Meurer et al. (2003) (92.0% to 93.6%) for Nile tilapia; and to those reported by Takeuchi et al. (1994), who obtained CDA of 87.7% with grass carp, using raw maize and 92.2% when extruded; and slightly higher with tilapia, 97.1% and 97.8%, respectively. These numbers were higher than the CDA values obtained by Hernández et al. (1994) for common carp, when they replaced raw maize with an extruded type, and the ether extract's CDA improved from 86.9% to 88.8%; and to those reported by Oliveira et al. (1998) with Nile tilapia using palm kernel cake (92.7% to 97.8%).

Pezzato et al. (2002), studied two different species with the same eating habits (omnivores), and found that the pacu showed better digestibility (p<0.05) when compared to the Nile tilapia, in all studied diameters. With the pacu, the ether extract's CDA, whose ingredients had a diameter of 1.0 mm (80.59 ± 0.58) was significantly better than the others (p<0.05). CDA treatments with diameters of 0.5; 1.5; 2.0 and 2.5 mm (78%) were similar (p<0.05). However, in this study the best digestibility value of the ether extract was in treatment with the smallest particle size of the maize in the feed.

The apparent digestibility (CDA) obtained with the feed whose particle size of the maize had different diameters of maize, ranging from 150 µm to 850 µm, demonstrated that these particles have the ability to digest what is otherwise considered small for some fish species. These diameters seem to define the acceptable limits for the grinding of raw material that will compose the diet of this species. It is important to note that the fish in this experiment had juvenile weight. Such responses corroborate the results of He and Wurtsbaugh (1993), where the diameter of the particle feed size has a significant effect on the digestion. According to these authors, the smallest surface area of these particles hinders its mechanical and chemical digestion.

In the experiment in question the treatments with smaller particle sizes of maize had better results, affected the growth performance of tambaqui and the apparent digestibility of crude protein. Ether extract obtained the best values as the particle size of the maize was reduced, probably due to it being an omnivorous species and therefore has a high capacity to process ingredients of plant origin such as maize, not to mention that the extrusion process may have further improved these feeds, improving gelatinization of starch and expansion. Therefore, for best results in the production of tambaqui, it is recommended that a particle size of 150 µm of maize be used in the feed. Since research regarding particle size of ingredients in fish feed are still scarce, we also recommend that further research be done, since only the particle size of maize was assessed in this research.

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