

## Digestibility and performance of juvenile Nile tilapia fed with diets containing forage palm

### Digestibilidade e desempenho de juvenis de tilápia nilótica alimentadas com dietas contendo palma forrageira

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#### Abstract

Supplying the nutritional needs of animals in an economical way is essential to the success of any type of production. The forage palm is a cactaceae well-adapted to semiarid regions and commonly used in the feeding of ruminants. However, studies assessing its use in fishes are scarce. We aimed to evaluate the digestibility of bran from five different forage palm cultivars (*Opuntia ficus*) in juvenile Nile tilapia and the development of juveniles when receiving increasing levels of the cultivar with the best apparent digestibility coefficient of crude protein (ADCCP) (75.2%). We used 90 masculinized juvenile Nile tilapia (with an average weight of  $6.02 \pm 0.65$  g) in the digestibility experiment to determine the apparent digestibility coefficients of dry matter (ADCDM), crude energy (ADCCP), and crude protein of the cultivars (Orelha de onça, Miúda, Gigante, Comum, and IPA 20). The experimental design was completely randomized (CRD) with five treatments and three repetitions. In the development experiment, 300 masculinized juvenile Nile tilapia (with average weight of  $1.39 \pm 0.12$  g) received feed with different inclusion levels (0, 72, 144, and 216 g Kg<sup>-1</sup>) of IPA 20 palm bran in a CRD with four treatments and five repetitions. We observed significant differences ( $P < 0.05$ ) for ADCDM and ADCCP between the treatments. The cultivars that presented the highest ADCDM were IPA 20 (41.75%), Orelha de onça (36.41%), and Gigante (32.59%). For ADCCP, the best cultivar was IPA 20 with 75.2%, followed by Orelha de onça (61.5%), and Miúda (56.1%). As for the performance, the inclusion of IPA 20 palm bran at a rate of 113 g/kg<sup>-1</sup> provided the best values in terms of weight gain and final weight in juvenile Nile tilapia.

**Key words:** Alternative food. *Opuntia ficus*. *Oreochromis niloticus*. Nutritional value.

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## Resumo

O suprimento das necessidades nutricionais de forma econômica é essencial para o sucesso de qualquer produção animal. A palma forrageira é uma cactácea bem adaptada as regiões semi-áridas, sendo comumente utilizada na alimentação de ruminantes, entretanto estudos com peixes são escassos. Objetivou-se avaliar a digestibilidade de farelos de cinco cultivares de palma forrageira (*Opuntia ficus*) para juvenis de tilápia nilótica e posteriormente o desempenho dos juvenis recebendo níveis crescentes da cultivar com melhor coeficiente de digestibilidade aparente da proteína bruta (CDAPB) (75,2%). No experimento de digestibilidade foram utilizados 90 juvenis de tilápia nilótica masculinizadas (com peso médio de  $6,02 \pm 0,65$  g), para determinar os coeficientes de digestibilidade aparente da matéria seca (CDAMS), energia bruta (CDAEB) e proteína bruta dos cultivares (Orelha de onça, Miúda, Gigante, Comum e IPA 20). O delineamento experimental foi inteiramente casualizado (DIC) com cinco tratamentos e três repetições. No experimento de desempenho 300 juvenis de tilápia nilótica masculinizados (peso médio de  $1,39 \pm 0,12$  g) receberam rações com diferentes níveis de inclusão (0, 72, 144 e 216 g Kg<sup>-1</sup>) do farelo de palma-cultivar IPA 20, em um DIC com quatro tratamentos e cinco repetições. Foram observadas diferenças significativas ( $P < 0,05$ ) para o CDAMS e o CDAPB entre os tratamentos. As cultivares que apresentaram maiores CDAMS foram a IPA 20 (41,75%), Orelha de onça (36,41%) e Gigante (32,59%). Para o CDAPB a melhor cultivar foi a IPA 20, com 75,2%, seguida da Orelha de onça (61,5%) e Miúda (56,1%). Quanto ao desempenho, a inclusão do farelo de palma-cultivar IPA 20 na proporção de 113 g kg<sup>-1</sup> promove os melhores valores de ganho de peso e de peso final para juvenis de tilápia nilótica.

**Palavras-chave:** Alimento alternativo. *Opuntia ficus*. *Oreochromis niloticus*. Valor nutritivo.

## Introduction

Among the groups of fish produced in Brazil, tilapia occupies the first position, with production of approximately 253,800 tons in 2011. This is due to important characteristics such as good market acceptance, relatively easy reproduction, the high survival rate of juveniles, suitability for different breeding systems (used in both extensive and intensive production systems), and suitability for different regions of the country (MPA, 2011). Another relevant characteristic concerns the acceptance of the species regarding different diet ingredients (FURUYA, 2010), enabling the inclusion of ingredients with seasonal and regional availability to reduce the highest cost of production, the feed.

For an ingredient to be included in the formulation of a feed in a safe and efficient way, knowledge on the digestibility and on the maximum level of inclusion of the ingredient by tilapia are needed to avoid negative effects on the animal's development. Being aware of this is not only important economically speaking, but also from an environmental point of view, since well-formulated

feeds (considering nutritional requirements and the use of all ingredients in the composition) reduce the waste generation per kilogram of consumed feed (CYRINO et al., 2010).

The forage palm is an alternative animal feed ingredient, particularly for goats, sheep, and cattle in Brazilian semiarid regions (GALVÃO JÚNIOR et al., 2014). The interest in the use of this cactus is due to its resistance to long periods of drought, common in that region (FARIAS et al., 2000), in addition to its nutritional characteristics such as a high energy value and the presence of polysaccharides and antioxidants (MEDINA-TORRES et al., 2011).

We aimed to assess the digestibility of bran from five forage palm cultivars in juvenile Nile tilapia and the development of fish fed with diets containing increasing levels of palm bran with the best digestibility of nutrients and energy.

## Material and Methods

Two experiments were performed at the Laboratory of Fish Nutrition and Feeding

(AQUANUT) of the Santa Cruz State University (UESC), in the city of Ilhéus, Bahia, Brazil. The first experiment investigated digestibility and the second assessed development.

### Digestibility

In the first test of digestibility, we acquired five palm cultivars (Orelha de onça, Miúda, Gigante, IPA 20, and Comum), used in the form of bran. We used 90 masculinized juvenile tilapia (with an average weight of  $6.02 \pm 0.65$  g), allocated into six feeding tanks, where they remained throughout the day; they were later transferred to six digestibility aquariums, where fecal samples were collected. All tanks were provided with independent water supplies in a closed water circulation system with a biological filter and continuous aeration using 1cv (Mod. CV-101R, VENTBRAS Ind. Eletro Metalúrgica Ltda, São Paulo, Brazil).

The cleaning of tanks and digestibility aquariums was carried out daily for the removal of leftover feed and feces. The water physical and chemical parameters such as temperature, pH, and dissolved oxygen were verified at two day intervals using a digital multiparameter probe (models YSY 63-10 FT and 55-12 FT, YSI Corporation, Owings Mills, MA, USA), with values of  $26.8 \pm 0.43^\circ\text{C}$ ,  $7.00 \pm 0.11$ ,  $7.2 \pm 1.43$  mg/L<sup>-1</sup>, and  $90 \pm 18\%$ , respectively, i.e. within the range recommended for this species (ROSS, 2000).

A reference diet containing no palm bran was formulated (Table 1) with the use of the computer program SUPERCAC<sup>®</sup>, considering the nutritional needs of Nile tilapia (FURUYA, 2010). The experimental diets consisted of replacing 30% of the reference diet with one of the palm cultivars (Table 2). To prepare the diets, ingredients were milled in a shredding mill (0.7 mm), mixed with microingredients and soybean oil, and moistened with 20% water at 40°C for subsequent pelletizing in a fish pellet machine with a frequency inverter and a 1.7 mm sieve. An indirect methodology was used including 0.1 g kg<sup>-1</sup> chromium oxide in the tested feeds. The pellets were dried in an oven at 55°C, and after being cooled they were packed and stored at 4°C.

**Table 1.** Food composition of the reference diet.

Ingredient	Quantity (g Kg <sup>-1</sup> )
Soybean meal - 45 g 100g <sup>-1</sup>	290.0
Fish meal - 55 g 100g <sup>-1</sup>	215.8
Wheat bran	210.0
Corn starch	138.1
Cornmeal	127.4
Vitamin-mineral complex <sup>1</sup>	10.0
Soybean oil	8.5
Butylhydroxytoluene	0.2
Chromium oxide	1.0
Calculated composition	
Crude protein (g Kg <sup>-1</sup> )	290.7
Digestible protein (g Kg <sup>-1</sup> )	260.0
Digestible energy (Kcal Kg <sup>-1</sup> )	3000
Ether extract (g Kg <sup>-1</sup> )	40.4
Crude fiber (g Kg <sup>-1</sup> )	40.0

**Table 2.** Bromatological characteristics of forage palm cultivars based on dry matter.

Cultivar	Dry matter (g Kg <sup>-1</sup> )	Crude energy (kcal/kg)	Crude protein (g Kg <sup>-1</sup> )	NDF <sup>1</sup> (g Kg <sup>-1</sup> )	ADF <sup>2</sup> (g Kg <sup>-1</sup> )
Orelha de onça	883.3	3.215	79.13	450.35	218.05
Miúda	892.2	3.396	142.45	343.08	123.63
Redonda	888.2	3.782	68.34	439.21	211.10
IPA 20	883.3	3.713	113.78	472.88	222.23
Comum	887.7	3.852	79.86	483.95	249.30

<sup>1</sup>Neutral detergent fiber

<sup>2</sup>Acid detergent fiber

For the collection of feces, we applied the modified Guelph method, in which fish went through a period of adjustment of four days for each experimental diet and another four days to collect fecal samples, so that every eight days it was possible to evaluate two experimental diets, totaling 24 days of the experiment. The fish were held during the day in feeding tanks to receive the diets until apparent satiety, five times a day. After the last feeding (5 p.m.), the fish were transferred to the digestibility aquariums (200 L), where they spent the night. The digestibility aquariums were cylindrical in shape with a conical bottom, with a collector (200 mL) attached to the bottom, where the feces accumulated by decantation. The external part of the collector was kept in contact with ice inside a styrofoam box to avoid leaching of the feces.

The next morning (by 7:00 a.m.), after the fish were returned to the feeding tanks, the excess water was removed from the collectors and the wet feces were subjected to drying in ventilated greenhouses (55°C) for 36 hours, for subsequent qualitative food analyses (AOAC, 2000).

The analysis of matter (DM) and gross energy (GE) were performed at the Laboratory of Food and Fish Nutrition (AQUANUT-UESC) and the crude protein (CP) tests were performed according to AOAC (2000) methodology. Amino acid (AA) analysis was performed by the CBO Laboratory Analysis (Campinas/SP) according to the methodology described by White et al. (1986). The apparent digestibility coefficients (ADC) of the diets were calculated (DE SILVA, 1989; BUREAU et al., 2002) using the following formula:

$$\text{ADC}(\%) = 100 - \left( \frac{\% \text{Cr}_2\text{O}_3 \text{ diet}}{\% \text{Cr}_2\text{O}_3 \text{ feces}} \right) \times \left( \frac{\% \text{nutrients or gross energy feces}}{\% \text{nutrients or gross energy diet}} \right)$$

$$\text{ADC}_n = \frac{\text{ADC}_{DT} - \text{ADC}_{DR} \times Y}{\text{ADC}_{DT} \times Z}$$

where the % Cr<sub>2</sub>O<sub>3</sub> diet is the percentage of chromium oxide in the diet; and % Cr<sub>2</sub>O<sub>3</sub> feces is the percentage of chromium oxide in the feces; ADC<sub>n</sub> is the apparent digestibility coefficient of nutrients or energy; ADC<sub>DT</sub> is the apparent digestibility coefficient of nutrients or energy in the diet tested; ADC<sub>DR</sub> is the apparent digestibility coefficient of nutrients or energy in the reference diet; Y = the proportion of the reference diet; and Z = the ratio of the test diet.

### Performance

From the results on the digestibility of the five cultivars of forage palm, we executed the development experiment with tilapia fed with IPA 20 forage palm bran for 60 days. Around 300 masculinized Nile tilapia were used (average weight

of 1.39 ± 0.12 g), distributed into 20 circular tanks (150 L). We used a closed water circulation system with continuous water flow (0.084 m<sup>3</sup> h<sup>-1</sup>) using a water pump of 3/4<sup>-1</sup>cv (Mod. CP-R6, Dancor S.A. Ind. Mecânica, Rio de Janeiro, Brazil). Cleaning of the tanks was carried out for the removal of decanted debris.

The experimental design was completely randomized with four treatments and five repetitions, with 15 fish distributed per experimental unit. Four isoproteic and isoenergetic diets were formulated according to the recommendations of Furuya (2010), using the computer program SUPERCRAC<sup>®</sup>. From the control treatment, without palm bran, three diets were formulated with increasing levels of inclusion (72, 144, and 216 g/kg<sup>-1</sup>) of IPA 20 palm bran (Table 3). During the trial period, the fish were fed four times a day until satiety to avoid leftovers.

**Table 3.** Composition of experimental diets.

Ingredient	Treatment			
	Palma 0	Palma 72	Palma 144	Palma 216
Soymeal - 45 g 100 g <sup>-1</sup>	292.7	296.4	306.9	320.0
Fishmeal - 55 g 100 g <sup>-1</sup>	118.0	120.0	120.0	120.0
Meat-bone flour - 40 g 100 g <sup>-1</sup>	122.2	120.0	120.0	104.8
Wheat Bran	217.2	150.0	108.0	70.0
Cornmeal	140.1	140.0	103.0	70.0
IPA 20 palm bran	0	72.0	144.0	216.0
Starch	86.6	75.3	56.0	50.0
Vitamin-mineral complex <sup>1</sup>	15.0	15.0	15.0	15.0
Soybean oil	8.0	11.1	25.9	31.0
Butylhydroxytoluene	0.2	0.2	0.2	0.2
Total	1000	1000	1000	1000
<i>Composição calculada</i>				
Crude protein (g Kg <sup>-1</sup> )	296.7	294.0	295.0	299.4
Digestible protein (g kg <sup>-1</sup> )	245.2	250.0	250.0	247.6
Digestible energy (Kcal Kg <sup>-1</sup> )	2.800	2.800	2.800	2.750
Ether extract (g Kg <sup>-1</sup> )	41.3	44.5	57.3	59.6
Crude fiber (g Kg <sup>-1</sup> )	43.6	43.5	47.1	51.2

<sup>1</sup>Composition (Kg<sup>-1</sup>): Mg - 2.600 mg; Zn - 14.000 mg; Fe - 10.000 mg; Cu - 1.400 mg; Co - 20 mg; I - 60 mg; Se - 60 mg; Vit. A - 1.000.000 UI; Vit. D3 - 400.00 UI; Vit. E - 10.000 mg; Vit. K3 - 500 mg; Vit. B1 - 2,500 mg; Vit. B2 - 2.500 mg; Vit. B6 - 2.500 mg; Vit. B12 - 3.000 mcg; Vit. C - 35.000 mg; Ac. Folic - 500 mg; Ac. Pantothenic acid - 5.000 mg; Niacin - 10.000 mg; Biotin - 80.000 mcg; Coline - 200.000 mg; Methionine - 130 g; Inositol - 5.000 mg; Etoxiquin - 15.000 mg.

After 60 days of the experiment, biometry was carried out on the fish, so that all fish were counted, weighed and measured to estimate survival calculations, final weight, and final total length, respectively. We calculated the total consumption controlling the feed consumed by weighing the diet containers and conducting the development analysis on a weekly basis. We also assessed:

Daily consumption of the diet = (total consumption/experimental period) x 100;

Daily weight gain (g) = (final weight - initial weight)/experimental period;

Weight gain (g) = (final weight - initial weight);

Feed conversion = consumption of the diet/weight gain;

Specific growth rate (% per day<sup>-1</sup>) = (ln initial weight - ln final weight/experimental period) x 100;

Protein efficiency ratio (%) = (weight gain/diet consumption x % CP in the diet) x 100;

Survival (%) = (dead individuals/alive individuals) x 100

#### *Statistical analysis*

We subjected the data of digestibility coefficients to analysis of variance, and, when we detected significant differences between treatments, we compared the averages using Tukey's test. We subjected the data obtained with the development of analysis of variance, and when we detected significant differences between the factors, we decomposed the quadratic sums in orthogonal polynomials up to the third degree. We adopted 5% probability in the two experiments and analyzed the data using the R Core Team statistical package (R, 2011).

## Results and Discussion

### Digestibility

There were no significant differences ( $P>0.05$ ) between treatments for apparent digestibility coefficient of the crude energy variable (ADCCE). However, the apparent digestibility coefficients of dry matter (ADCDM) and of crude protein (ADCCP) showed significant differences ( $P<0.05$ ) between treatments (Table 4). The cultivars that showed the highest ADCDM were IPA 20

(41.75%), Orelha de onça (36.41%), and Gigante (32.59%), although the latter two cultivars did not differ from the cultivar Comum (25.26%). The Miúda cultivar presented the smallest ADCDM (17.04%), presenting no differences compared to Comum. Oliveira et al. (2013), when working with Nile tilapia (around 110 g), found superior ADCDM with all palm cultivars tested, i.e. the cultivars Orelha de onça, Comum, Gigante, IPA 20, and Miúda at 54%, 53%, 50%, 48.3%, and 47.9%, respectively.

**Table 4.** Average values of apparent digestibility coefficients (%) of dry matter, crude protein and crude energy of brans of different palm cultivars for Nile tilapia (6 g).

Treatment	Apparent digestibility coefficient (%)		
	Dry matter	Crude protein	Crude energy
Orelha de onça	36.41 ± 1.92 <sup>ab</sup>	56.07 ± 7.71 <sup>ab</sup>	52.76 ± 10.02
Miúda	17.04 ± 0.21 <sup>c</sup>	22.07 ± 1.65 <sup>b</sup>	37.44 ± 19.0
Gigante	32.59 ± 8.0 <sup>ab</sup>	55.12 ± 22.4 <sup>ab</sup>	36.94 ± 11.62
Comum	25.26 ± 0.06 <sup>bc</sup>	61.50 ± 10.26 <sup>a</sup>	30.77 ± 8.85
IPA 20	41.75 ± 8.11 <sup>a</sup>	75.18 ± 6.22 <sup>a</sup>	50.34 ± 6.29
CV(%)	16.88	22.29	21.76

Averages followed by different letters in the columns differ among each other with a 5% level of significance obtained by the Tukey test.

The ADCDM found for different cultivars of palm were inferior to those found by Gonçalves et al. (2009) for some foods such as wheat bran, showing 70.37% and 59.29%, and cornmeal, showing 79.30% and 55.52%, respectively, using Nile tilapia (100 g).

A factor that probably contributed to the low apparent digestibility of dry matter and of crude protein and energy in most cultivars was the high presence of fiber in the bran. Lanna et al. (2004) observed reductions in dry matter and crude protein digestibility using feeds with crude fiber levels above 7.5% and reduced digestibility of ether extract using 10% of crude fiber, for Nile tilapia, in addition a the decrease in the retention time of the bolus in the digestive tract, as also noted by Meurer et al. (2003).

As for the ADCCP, the cultivars IPA 20, Comum, Orelha de onça, and Gigante presented the highest values: 75.18%, 61.50%, 56.07%, and 55.12%, respectively. Miúda presented the lowest ADCCP (26.77%) as well as Orelha de onça (56.07%) and Gigante (55.12%). Oliveira et al. (2013) observed a similar result, in which the digestibility of forage palm cultivars was evaluated with Nile tilapia (110 g), finding the highest ADCCP for IPA 20 (75.43%) and Comum (78.21%) cultivars.

Braga et al. (2010), when analyzing the digestibility of mesquite pod flour, cassava leaf, and cocoa bran with juvenile tilapia, found ADCDM values of 48.7%, 50.2%, and 43.9%, ADCCP values of 51.6%, 49.8, and 38.5%, and ADCCE values of 30.5%, 29.3%, and 23.1%, respectively. Comparing the palm cultivars with these other products, we

observed that palm cultivars showed inferior ADCDM values. The ADCCP was higher among most cultivars, except for the Miúda cultivar. For the ADCCE, values were superior for most palm cultivars, except for the Comum cultivar, which presented an ADCCE similar to those of mesquite pod flour and cassava leaf.

### Performance

No significant differences were observed for the following variables: survival, feed conversion, final length, protein efficiency ratio, and specific growth rate ( $P>0.05$ ), with average values of 89, 1.33, 108.04, 2.64, and 4.85, respectively. We observed a significant quadratic effect ( $P<0.05$ ) of including increasing levels of IPA 20 palm bran in diets on the final average weight, weight gain and daily weigh

gain (Table 5). To achieve the highest final average weight (Figure 1) and weight gain (Figure 2), we estimated that the inclusion of approximately 113 g  $\text{kg}^{-1}$  palm bran would provide the best values. With that, the highest weight gain for fishes submitted to treatments with inclusion may be related to a greater availability of glucose, decreasing the use of protein through the glycolytic pathway, as well as being a sign of better use of the starch contained in the diet. However, the inclusion of higher levels of palm bran did not lead to greater development. This effect may be related to the high amounts of fiber contained in this ingredient. High concentrations of fiber in fish diets change the bowel transit time (MEURER et al., 2003) and increase the viscosity of the bolus, having as consequence a lower apparent digestibility coefficient because of reduced enzymatic action (SCHWARZ et al., 2010).

**Table 5.** Average values of zootechnical development of juvenile tilapia submitted to diets with different levels of replacement with IPA 20 palm bran.

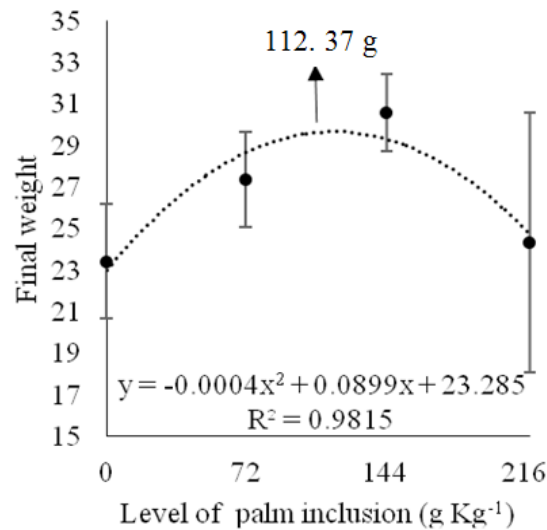
Variable	Inclusion level (g $\text{kg}^{-1}$ )			
	0	72	144	216
Final weight (g) <sup>1</sup>	23.41±2.75	27.37±2.31	30.59±1.87	24.31±6.25
Weight gain (g) <sup>2</sup>	22.02±2.75	25.97±2.31	29.19±4.77	22.92±6.25
Daily weight gain (g) <sup>3</sup>	0.37±0.04	0.43±0.04	0.49±0.08	0.38± 0.1
Feed consumption (g) <sup>NS</sup>	30.42±3.56	28.75±1.60	32.48±4.61	33.01±4.28
Daily consumption (g) <sup>NS</sup>	0.51±0.06	0.48±0.03	0.54±0.08	0.55±0.07
Feed conversion <sup>NS</sup>	1.39±0.19	1.11±0.13	1.29±0.25	1.53±0.44
Survival (%) <sup>NS</sup>	88.00±0.01	96.00±0.06	86.67±0.08	85.33±0.12
Final length (mm) <sup>NS</sup>	104.08 ± 5.21	109.16 ± 3.24	112.94 ± 7.32	105.98 ± 10.42
SGR (% $\text{dia}^{-1}$ ) <sup>4 NS</sup>	4.96±0.14	5.01±0.31	4.69±0.19	4.72±0.44
PER (g 100 <sup>-1</sup> ) <sup>5 NS</sup>	3.08±0.35	2.69±0.50	2.45±0.30	2.35±0.71

<sup>1,2,3</sup> Quadratic effect. <sup>NS</sup> Not significant ( $P>0.05$ ). <sup>4</sup>Specific growth ratio. <sup>5</sup>Protein efficiency ratio

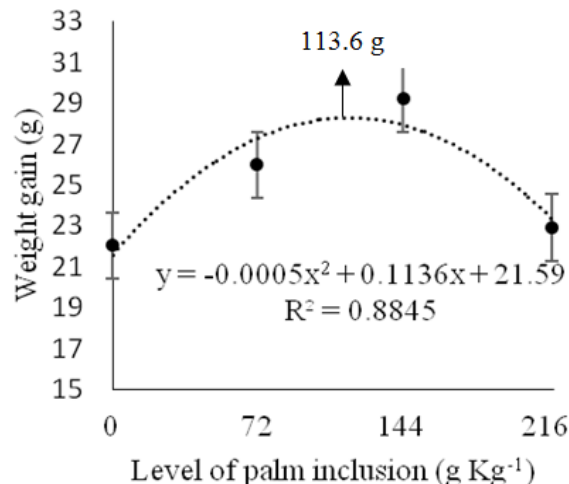
The highest final weight of fish fed up to the inclusion of 113 g  $\text{kg}^{-1}$  palm bran may be a result of the action of bioactive compounds present in this ingredient, such as flavonoids, carotenoids, ascorbic acid, and polysaccharides (MEDINA-TORRES et al., 2011). These compounds have multiple functions such as reducing lipid peroxidation because of their

antioxidant action (PEREIRA; CARDOSO, 2012), selecting beneficial intestinal microbiota thanks to prebiotic action (SCHWARZ et al., 2010), as well as acting as enzyme cofactors (SEALOCK; GOODLAND, 1951), which results in immune and productive gain.

**Figure 1.** Regression curve adjusted to the levels of inclusion of IPA 20 palm bran in the feed of juvenile Nile tilapia for final average weight.



**Figure 2.** Regression curve adjusted to the levels of inclusion of IPA 20 palm bran in the feed of juvenile Nile tilapia for weight gain.



The search for energy ingredients that promote tilapia development was tested by Boscolo et al. (2002), who observed the possibility of totally replacing corn with cassava leaf flour without affecting the development of juveniles of Nile tilapia. Souza et al. (2013) evaluated the development of tilapia fed mango bran instead of cornmeal and verified the possibility of replacing up to 33% without inhibiting performance.

## Conclusion

The palm cultivar best exploited by juvenile Nile tilapia was IPA 20, considering the apparent digestibility coefficients of crude protein and energy. The inclusion of 113 kg<sup>-1</sup> of IPA 20 palm bran in the diet of juvenile Nile tilapia provides the best values of final weight, weight gain, and daily weight gain.



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