

Research Article

## Lysine effect on the characterization of fillet, by-products, residues, and morphometry of tambaqui *Colossoma macropomum* (Cuvier, 1818)

Ariany Rabello da Silva Liebl<sup>1</sup> , Marcelo dos Santos Nascimento<sup>2</sup> , Paulo Henrique Rocha Aride<sup>3</sup> 

Jackson Pantoja-Lima<sup>4</sup> , Márcia Regina Fragoso Machado Bussons<sup>5</sup> 

Wilson Massamitu Furuya<sup>6</sup> , Adriano Teixeira de Oliveira<sup>3</sup> 

<sup>1</sup>Postgraduate Program in Animal Science and Fishery Resources  
Federal University of Amazonas, Amazonas, Brazil

<sup>2</sup>Postgraduate Program in Aquaculture, Nilton Lins University, Amazonas, Brazil

<sup>3</sup>Federal Institute of Science and Technology of Amazonas  
Campus Center Manaus, Amazonas, Brazil

<sup>4</sup>Federal Institute of Science and Technology of Amazonas  
Campus Presidente Figueiredo, Amazonas, Brazil

<sup>5</sup>Institute of Technology and Education Galileo da Amazônia, Manaus, Brazil

<sup>6</sup>Department of Animal Science, University of Ponta Grossa, Paraná, Brazil

Corresponding author: Adriano Teixeira de Oliveira (adriano.oliveira@ifam.edu.br)

**ABSTRACT.** This study aimed to evaluate the yields of by-products and residues and the morphometry of juvenile tambaqui *Colossoma macropomum* undernutrition with different levels of lysine. Diets were elaborated containing 6.60, 9.72, 12.84, 15.96, 19.08, and 22.20 g kg<sup>-1</sup> of total lysine, and these were fed to fish distributed in 18 tanks (310 L each one) for 90 days. Morphometric analysis, measurements of whole fish, by-products, and residues were taken and compared to each other. All variables were validated using ANOVA, Tukey's test, and quadratic regression analysis ( $P < 0.05$ ). No differences were found for the morphometric variables ( $P > 0.05$ ). The ratio between morphometric data for the standard and total length showed significance at levels of 9.72 and 15.96 g kg<sup>-1</sup> ( $P = 0.03$ ). The gutted fish and fillets, the protein and lipid contents of the fillets, and the weights and lengths of the intestines did not change ( $P > 0.05$ ). Moisture was modified with lysine at levels of 22.20 g kg<sup>-1</sup> ( $P = 0.00$ ), and minerals, between 6.60 and 22.20 g kg<sup>-1</sup> ( $P = 0.01$ ). The increase in lysine levels in the diet led to an increase in fin weight ( $P = 0.00$ ). It was concluded that the addition of lysine in the diet did not directly influence the morphometry and yields of the fish in the juvenile phase, but it did affect the fin waste and the skin by-product.

**Keywords:** *Colossoma macropomum*; aquaculture; nutrition; amino acid; yield; native species; Amazon

### INTRODUCTION

The increase in fish productivity affects the production of by-products and residues of the species being cultivated and allows for fractions with low or no commercial value when the yield and chemical composition of these are known (Aride et al. 2017, 2020). Thus, the logistical organization of fish production requires knowledge that is not restricted to the number of edible products but includes information on

by-products and wastes from processing and analyzes the viability, productivity gains, and financial investments (Simões et al. 2007). Of the investments related to production, the diet is the greatest of all, and the percentage of amino acids is the one that requires the most expenditure (NRC 2011, Oliva-Teles 2012, Souza et al. 2019).

Among the amino acids, lysine, which is directed to protein synthesis, stands out since it is a limiting factor in fish diets made with plant-origin products (Ritcher et

al. 2020). Thus, the use of lysine tends to increase the yield of the main product of the fish, the fillet, of by-products such as the skin and the trunk, in addition to the filleting residues, such as the head and fins (Furuya et al. 2006, Gaylord & Barrows 2009, Ovie & Eze 2013, Hua et al. 2019).

Thus, the waste and by-products of the fish can be used as ingredients of animal feed, for the extraction of collagen, for the production of fertilizers, of chemicals, artisanal products, and other products for human nutrition, since fish residues are rich in protein and omega-3 (Cavalcante et al. 2005, Feltes et al. 2010, De Sousa et al. 2016).

The exploitation of the fish's residual portions raises the fishing industry's importance by helping to overcome the traditional demand and offering products obtained from the use of materials with high nutritional value. This process also favors more affordable prices for low-income populations, in addition to reducing waste that, in excess, tends to generate environmental and public health problems (Feltes et al. 2010).

The percentage of residues and the anatomical characteristics of the fish's body are among the factors for obtaining a greater yield of the product (Contreras-Guzmán 1994, Leonardth et al. 2006, Basso & Ferreira 2011). Considered crucial in the determination of the yield of the product and by-products are the morphometric characteristics. These characteristics indicate parameters for the percentage of prime cuts of the fish, the choice of the species to be produced, and the planning and standardization of portions of the industrialized fish, particularly the fillet (Mafra et al. 2016).

In addition to the anatomical aspects of the fish body, the yield of the fillet is linked to other conditions, including the weight of residues, sex of the animal, body chemical composition, as well as the human and machines' ability in the filleting process (Contreras-Guzmán 1994, Leonardth et al. 2006, Basso & Ferreira 2011). In this context, lysine is inserted in the metabolic process of stretching the myocytes that make up the fillet and thus acts on the hypertrophy and unfolding of the tissue and on the hyperplasia of the cells that, in fish, operate concomitantly and are controlled by factors such as species, phase of life and nutrition of the fish (Aguar et al. 2008, Valente et al. 2013, Michelato et al. 2016, Chu et al. 2018).

The tambaqui *Colossoma macropomum* (Cuvier, 1818), is an Amazonian fish consolidated in the Brazilian fishing industry due to high productivity, ease in rearing, hardiness, and having meat with texture and flavor that is highly appreciated by the consumer (Araújo-Lima & Gomes 2005, Souza & Inhamuns

2011, Rodrigues 2014, Aride et al. 2016, 2017, 2020, Buzollo et al. 2019, Lima et al. 2020, 2021, Nascimento et al. 2020, Pantoja-Lima et al. 2020, 2021, Liebl et al. 2021, Mattos et al. 2021, Pinto et al. 2021, Polese et al. 2022).

Although tambaqui has a good acceptance of the nutritional system commonly used for fish production, the diets provided disregard the intrinsic conditions of the species for product yield and the effect on the by-products and potentially usable residues. There is a lack of specific nutritional information for this species, such as the amino acid lysine. Given the above, this study aimed to verify the influence of the crystalline amino acid L-lysine on the yields of by-products, residues, and fillet characterization and morphometry when included in the diet of juvenile tambaqui.

## MATERIALS AND METHODS

### Ethical animal use

The experiment was developed following the ethical guidelines for animal experimentation provided by the National Council for the Control of Animal Experimentation (CONCEA) and approved by the Ethics Commission on the Use of Animals (ECUA) of the Federal University of Amazonas, under approval #005/2016. All experiments were conducted according to local and ARRIVE guidelines (Percie du Sert et al. 2020).

### Location of the experiment and experimental diets

Juvenile tambaqui *Colossoma macropomum* ( $n = 180$ ) from the Balbina fish farming station (Center for Training, Technology, and Production in Aquaculture-CTTPA; Presidente Figueiredo, Amazonas State, Brazil) were analyzed biometrically ( $33.88 \pm 2.47$  g and  $11.72 \pm 0.36$  cm) in the Aquatic Organisms Production Laboratory (LaPOAq) at the Nilton Lins University, Manaus, Amazonas, after acclimation in 18 polyethylene tanks, aerated, with a maximum volume of 310 L (10 fish per tank). The animals were fed a commercial diet (28% crude protein) until the start of the experiment.

After being formulated, the experimental diets (27% crude protein) with 6.60, 9.72, 12.84, 15.96, 19.08 and 22.20 g kg<sup>-1</sup> of total lysine, respectively, 0.00, 4.00, 8.00, 12.00, 16.00 and 20.00 g kg<sup>-1</sup> of L-lysine (Ajinomoto®, Chuo, Japan), were extruded at the Aquaculture Laboratory of the National Institute for Amazonian Research (INPA), Manaus, Amazonas State.

The ingredients for the diet (Table 1) were ground in a Wiley-type knife mill (Tecnal®, Piracicaba, Brazil)

**Table 1.** Diets with different lysine levels for juvenile tambaqui *Colossoma macropomum*. <sup>1</sup>Composition calculated according to Furuya et al. (2006) and Rostagno et al. (2011). BHT: butylated hydroxytoluene.

Ingredient (g kg <sup>-1</sup> ) <sup>1</sup>	Lysine (g kg <sup>-1</sup> )					
	6.60	9.72	12.84	15.96	19.08	22.20
Maize	356.2	360.2	364.2	368.2	372.2	376.2
Maize gluten 60	230.0	230.0	230.0	230.0	230.0	230.0
Broken rice grains	120.0	120.0	120.0	120.0	120.0	120.0
Wheat bran	120.0	120.0	120.0	120.0	120.0	120.0
Fish meal	60.0	60.0	60.0	60.0	60.0	60.0
Dicalcium phosphate	20.0	20.0	20.0	20.0	20.0	20.0
Soybean oil	10.0	10.0	10.0	10.0	10.0	10.0
Mineral and vitamin supplement	7.0	7.0	7.0	7.0	7.0	7.0
Calcitic limestone	5.0	5.0	5.0	5.0	5.0	5.0
Sodium bicarbonate	2.0	2.0	2.0	2.0	2.0	2.0
Antifungal additive	2.0	2.0	2.0	2.0	2.0	2.0
Antioxidant BHT	0.2	0.2	0.2	0.2	0.2	0.2
<b>Amino acids (g kg<sup>-1</sup>)<sup>1</sup></b>						
L-glutamic acid	50.00	42.00	34.00	26.00	18.00	10.00
DL-methionine	2.50	2.50	2.50	2.50	2.50	2.50
L-tryptophan	1.60	1.60	1.60	1.60	1.60	1.60
L-valine	1.00	1.00	1.00	1.00	1.00	1.00
L-arginine	2.50	2.50	2.50	2.50	2.50	2.50
L-histidine	2.50	2.50	2.50	2.50	2.50	2.50
L-isoleucine	2.50	2.50	2.50	2.50	2.50	2.50
L-phenylalanine	2.50	2.50	2.50	2.50	2.50	2.50
L-threonine	2.50	2.50	2.50	2.50	2.50	2.50
L-lysine HCl	0.00	4.00	8.00	12.00	16.00	20.00
<b>Total (g)</b>	<b>1000</b>	<b>1000</b>	<b>1000</b>	<b>1000</b>	<b>1000</b>	<b>1000</b>

and added to the crystalline nutrients DL-methionine, L-arginine, L-histidine, L-isoleucine, L-phenylalanine, and L-threonine, in a quantity of 2.50 g each. L-glutamic acid, L-tryptophan, and L-valine were added in quantities of 10 to 50 g; 1.6 and 1.0 g, respectively (Liebl 2019). After preparation for 90 days, the diets were provided four times a day until the apparent satiety of the experimental animals.

At the end of the experimental period, feeding was interrupted for 24 h, and the fish were sampled (n = 9 fish per diet), anesthetized with benzocaine (100 mg L<sup>-1</sup>), euthanized, washed in chlorinated water (5 ppm), individually packed in plastic bags and stored at -20°C. The fish were thawed at room temperature (20°C) (Simões et al. 2007, Maghelly et al. 2014). After the morphometric verification, extraction and weighing (scale accuracy of 0.001 g; Gehaka®, São Paulo, Brazil) of the fish's body parts were performed to verify yields.

### Morphometric analysis

The data for the morphometric analysis was obtained using a caliper and ichthyometer (Mitutoyo®, São Paulo, Brazil). The parameters analyzed were those of total length (TL), standard length (SL), trunk length (TRL), trunk height (TRH), head length (HL), head

height (HH), body height (BH), tail height (TH), lower caudal fin length (LCFL), upper caudal fin length (UCFL) and lumbar thickness (LT) (Pires et al. 2011).

Among the parameters analyzed, the ratios (cm cm<sup>-1</sup>) were also verified according to Adames et al. (2014): HL SL<sup>-1</sup>: head length/standard length; HL HH<sup>-1</sup>: head length/head height; SL TL<sup>-1</sup>: standard length/total length; TRH TRL<sup>-1</sup>: trunk height/trunk length.

### Fillet characterization: filleting yields and chemical composition

The percentage of gutted fish (carcass), fillet with skin, and fillet without skin were verified. For this, scissors, scalpel, cutting blades, tweezers, and pliers were used to filleting with the removal of the skin.

The quantities of the products were calculated using the following formulas:

- Gutted fish (GF) = (weight of fish without viscera / weight of whole fish) × 100
- Fillet with skin (FWS) = (weight of fillet with skin) / (weight of whole fish) × 100
- Skinless fillet (SF) = (weight of skinless fillet / weight of whole fish) × 100

For the centesimal verification of the crude protein and ether extract (%) (AOAC 2005), the fillets were ground, homogenized (Leonhardt et al. 2006), dried in an oven at 105°C and chopped for analysis with correction of the moisture content.

In the analysis of crude protein, the Micro-Kjehldal method was utilized and included steps of digestion and distillation of the matter, followed by titration with acid. Total lipids (ether extract) were verified using the Bligh and Dyer method, and neutral lipids, phospholipids, and glycolipids were analyzed using heat-free extraction. In order to verify the total minerals, the organic matter was incinerated in a muffle furnace at 550°C. The data obtained were inserted in the following equations:

- Ether extract (EE) = (lipid weight x 4) / (sample weight) × 100
- Integral protein in the sample (IP) = total protein / moisture correction factor
- Moisture correction factor (MCF) = 100 / dry matter
- Dry matter (DM) = (dry sample weight / moist sample weight) × 100
- Total minerals or ash (TM) = (ash weight × 100) / sample weight

### By-product and waste analysis

The whole fish (g) was weighed to determine the relationship between its weight and the weight of by-products and waste. The following by-products were verified: clean trunk and gross skin, and the residues were the head, ventral abdominal musculature, fins, gills, and viscera. The fish were processed using scissors, scalpel, cutting blades, tweezers, and anatomical pliers to remove the skin extracted in the cephalocaudal direction. For evisceration, a ventral-dorsal incision was made.

The relationship between the weight of the whole fish and the weight of the clean trunk as a percentage was calculated after the extraction processes of the head, skin, and dorsal, caudal, pelvic, pectoral, and ventral fins. The weight of the head was determined after the decapitation of the whole fish. A cut in the belly lateral extension until the end of the ribs was performed to calculate the ventral abdominal musculature as a percentage (Souza 2002). The fins and gill's weight was verified after the removal of these residual portions.

The products (%) were calculated according to Costa et al. (2014), using the following formulas:

- Headless carcass (HLC) = (headless fish weight / weight of whole fish) × 100

- Fillet with skin (FWS) = (weight of fillet with skin / weight of whole fish) × 100

- Skinless fillet (SF) = (weight of skinless fillet / weight of whole fish) × 100

By-products (%) and residues (%) of fish fillets were calculated according to Pires et al. (2011):

- Gross skin (GS) = (weight of skin / weight of whole fish) × 100

- Clean body (CB) = (decapitated fish / weight of whole fish) × 100

- Head (H) = (head weight with gills / weight of whole fish) × 100

- Ventral abdominal musculature (VAM) = (weight of ventral abdominal musculature / weight of whole fish) × 100

- Fins (F) = (weight of fins / weight of whole fish) × 100

- Gills (G) = (weight of gills / weight of whole fish) × 100

- Viscera (V) = (weight of viscera / weight of whole fish) × 100

### Water quality monitoring

Concentrations of dissolved oxygen (mg L<sup>-1</sup>), temperature (°C) and pH were checked daily using multiparameter apparatus (G-50, Horiba®, Kyoto, Japan), and nitrite content (mg L<sup>-1</sup>) was monitored weekly using specific colorimetric kits (Alfakit®, Florianópolis, Brazil). Water quality was maintained through siphoning and daily water replenishment (20% of the volume), according to Liebl (2019).

The mean values obtained for pH, water temperature, dissolved oxygen, and nitrite content were 5.74 ± 0.07; 26.25 ± 0.06°C; 4.95 ± 0.32 mg L<sup>-1</sup> and 0.013 ± 0.008, respectively. The average values obtained for pH, water temperature, dissolved oxygen, and nitrite content were within the recommended values for tambaqui cultivation (Araujo-Lima & Gomes 2005, Aride et al. 2007, Mendonça et al. 2012, Rebouças et al. 2014).

### Statistical analysis

The experimental design used was completely randomized (CRD), with six treatments and three repetitions. The treatments were diets containing the amino acid lysine at levels of 6.60, 9.72, 12.84, 15.96, 19.08, and 22.20 g kg<sup>-1</sup>. The values for yields, morphometry, and chemical composition were verified using the normality (Shapiro-Wilk normality test) and homogeneity (Bartlett test of homogeneity of variances) of the data. The information was submitted to va-

riance analysis (ANOVA,  $P < 0.05$ ) with Tukey's multiple comparison test. According to the best fit for the variable, the results were interpreted with linear, quadratic, quadratic with Plateau, or exponential regression models.

The data as percentages were standardized before being statistically analyzed, using the formula:  $\arcsin \sqrt{P\%/100}$  (Haddad & Vendramim 2000, Furuya et al. 2006). The R<sup>®</sup> software (R-project, Auckland, New Zealand), version 3.5.3, was used to confirm all the analyses performed statistically.

## RESULTS

### Morphometric parameters and morphometric ratios

In the analysis of morphometric parameters, no statistical significance was found between lysine levels for total length (TL), standard length (SL), trunk length (TRL), head length (HL), head height (HH), trunk height (TRH), and tail height (TH) ( $P > 0.05$ ). For lower tail thickness, upper tail thickness, and lumbar thickness, the significant difference between the treatments did not occur ( $P > 0.05$ ) (Table 2).

For morphometric ratios, the mean ratios of HL/SL, HL/HH, and TRH/TRL showed no variations (Table 3). However, a significant statistical difference was detected in the ratio between SL/TL ( $P = 0.03$ ) (Fig. 1).

### Fillet characterization: cutting yields and chemical composition

The gutted carcass, skinless fillet, and fillet with the skin showed no statistically significant difference between the different lysine levels added to the diets ( $P > 0.05$ ). In the regression analysis (quadratic effect with Plateau), plateau formation was verified for a fillet with skin with the addition of 1.37% L-lysine or 18.22 g kg<sup>-1</sup> of total lysine ( $y = 34.00273 + 4.532873x - 1.657618x^2$ ;  $R^2 = 0.13$ ) in the diet, with a production of 37.10% of the fillet ( $P = 0.00$ ).

The composition of the fillet, the crude protein, and the ether extract were also absent of any expression that statistically differentiated the means of the treatments ( $P > 0.05$ ). The highest total minerals or ash content was observed in the experimental treatment containing 15.96 g kg<sup>-1</sup> lysine. The mineral levels in the fillets of fish fed with 22.20 and 6.60 g kg<sup>-1</sup> lysine differed, and 22.20 g kg<sup>-1</sup> lysine was the level that presented the lowest percentage of minerals in the difference (Table 4).

### By-products and waste

The mean whole fish weight was  $61.32 \pm 5.48$  g. By-products and residues from filleting of *C. macropomum*

were verified under the influence of the different lysine levels, and no statistical significance was found ( $P > 0.05$ ) in the by-product clean trunk and the residual portions of the head, ventral abdominal muscle, gills, and viscera.

The percentage of the by-product gross skin expressed the difference between the levels of 12.84 and 22.20 g kg<sup>-1</sup> of total lysine ( $P = 0.03$ ) added in the diets of the juvenile tambaqui, with data corresponding to 13.63 and 8.27%, respectively, of the weight of the fish analyzed (Table 5).

From the data obtained in this study, it was possible to estimate the quadratic equation  $y = 1.47232 - 0.06487134x + 0.03726182x^2$  for the percentage of the juvenile tambaqui skin ( $P < 0.03$ ;  $R^2 = 0.39$ ), in which the curve evolved increasingly from the addition of 13.96 g kg<sup>-1</sup> of total lysine (0.87% L-lysine) generating skin production that was calculated as 1.44% of the weight of the fish (Fig. 2).

The increase in lysine levels made it possible to estimate the growth of the fins quadratically ( $P = 0.00$ ), generating the x and y values described in the algebraic formula  $y = 1.552856 - 0.019362x + 0.00799914x^2$  ( $R^2 = 0.7568$ ) with a positive response from the addition of 18.36 g kg<sup>-1</sup> of lysine and estimated yield of 1.54% (Fig. 3).

## DISCUSSION

### Morphometric parameters and morphometric ratio

The morphometry of the body structure of the fish is one of the tools used to optimize the fish industry, which standardizes the by-products and products, and is used as a tool in the process of genetic improvement of productive species (Santos et al. 2007, Adames et al. 2014).

Mourad et al. (2018) evaluated the morphometric growth of tambaqui *Colossoma macropomum*, pacu *Piaractus mesopotamicus* and tambacu and paqui hybrids (*P. mesopotamicus* × *C. macropomum*), estimated final measures to obtain genetic parameters and observed significant statistical difference in SL, HL, BH, and, BW in statistical models. The juvenile tambaqui, with an initial weight of  $54.59 \pm 11.01$  g, cultivated by Mourad et al. (2018), had the final weight estimated to be 1056.82 g, with SL calculated to be 30.47 cm, HL at 9.69 cm, and BW and WH of 15.35 and 4.10 cm, respectively.

The analysis of the measurements of the fish demonstrates the structural arrangement of the fillet and the yields (Carneiro et al. 2004, Maghelly et al. 2014). The absence of statistical significance observed in the

**Table 2.** Morphometric characterization of juvenile tambaqui *Colossoma macropomum* fed diets with increasing levels of lysine. Analysis of variance ( $P < 0.05$ ).

Morphometric variables (cm)	Lysine (g kg <sup>-1</sup> )						P
	6.60	9.72	12.84	15.96	19.08	22.20	
Total length	14.33 ± 1.0	14.47 ± 0.9	15.05 ± 0.9	15.60 ± 0.7	14.77 ± 1.3	15.40 ± 0.3	0.48
Standard length	11.30 ± 0.8	11.63 ± 0.7	11.75 ± 0.4	2.70 ± 0.6	11.90 ± 1.13	12.27 ± 0.2	0.28
Trunk length	7.50 ± 0.5	7.63 ± 0.4	7.40 ± 0.4	8.27 ± 0.5	7.40 ± 0.2	8.10 ± 0.1	0.10
Head length	3.90 ± 0.2	3.97 ± 0.2	4.25 ± 0.0	4.47 ± 0.1	4.13 ± 5.4	4.17 ± 0.1	0.53
Head height	3.60 ± 0.3	3.43 ± 0.3	3.60 ± 0.1	3.73 ± 0.2	3.30 ± 5.4	3.60 ± 0.1	0.14
Trunk height	6.00 ± 0.3	5.90 ± 0.4	6.05 ± 0.3	6.47 ± 0.3	6.40 ± 0.7	6.43 ± 0.0	0.32
Tail height	6.50 ± 0.4	7.10 ± 0.5	6.90 ± 0.5	7.40 ± 0.4	7.10 ± 0.7	7.53 ± 0.3	0.15
Lower caudal fin length	3.30 ± 0.3	3.47 ± 0.3	3.60 ± 0.1	3.67 ± 0.1	3.43 ± 0.2	3.77 ± 0.0	0.54
Upper caudal fin length	3.30 ± 0.0	3.63 ± 0.4	3.55 ± 0.0	3.53 ± 0.2	3.37 ± 0.1	3.70 ± 0.1	0.27
Lumbar thickness	4.00 ± 0.3	4.00 ± 0.3	4.55 ± 0.2	4.37 ± 0.5	4.63 ± 0.5	4.87 ± 0.2	0.18

**Table 3.** Morphometric ratios of juvenile tambaqui *Colossoma macropomum* fed diets with increasing levels of lysine. Analysis of variance ( $P < 0.05$ ). \*Significant values in the same row. Means followed by the same letter do not differ from each other by the Tukey test ( $P < 0.05$ ). HL:SL: head length/standard length; HL:HH: head length/head height; SL:TL: standard length/total length; TRH:TRL: trunk height/trunk length.

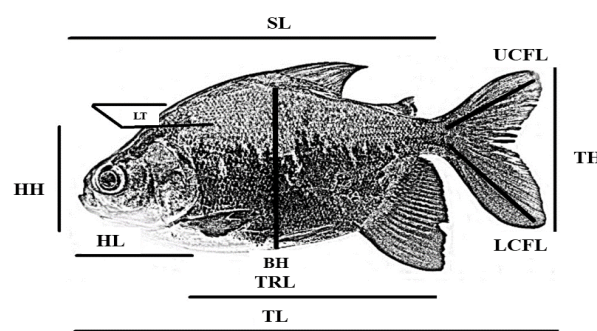
Morphometric ratios (cm cm <sup>-1</sup> )	Lysine (g kg <sup>-1</sup> )						P < 0.05
	6.60	9.72	12.84	15.96	19.08	22.20	
HL:SL	0.48 ± 0.01	0.45 ± 0.01	0.49 ± 0.01	0.45 ± 0.01	0.45 ± 0.03	0.44 ± 0.00	0.73
HL:HH	0.60 ± 0.07	0.58 ± 0.06	0.60 ± 0.02	0.58 ± 0.06	0.52 ± 0.02	0.56 ± 0.05	0.93
SL:TL	0.66 ± 0.01 <sup>ab</sup>	0.65 ± 0.01 <sup>ab</sup>	0.63 ± 0.01 <sup>a</sup>	0.65 ± 0.01 <sup>b</sup>	0.62 ± 0.01 <sup>ab</sup>	0.66 ± 0.01 <sup>ab</sup>	0.03*
TRH:TRL	1.67 ± 0.01	1.79 ± 0.02	1.62 ± 0.00	1.66 ± 0.03	1.72 ± 0.08	1.81 ± 0.00	0.18

morphometry suggests the same behavior for the yield data of juvenile tambaqui receiving L-lysine in their diet.

The morphometric ratio SL:TL describes the SL, which is the extension from the end of the head to the insertion of the caudal fin. The TL corresponds to the measurement from the end of the head to the end of the fish's tail (Fig. 1). The difference found for SL:TL ( $P = 0.03$ ) may be related to the influence of lysine inclusion in diets on this portion of the body. In the present study, only the length of the caudal fins in the lower and upper portions was verified (Fig. 1).

#### Fillet characterization: cutting yields and chemical composition

Michelato et al. (2016) found  $30.69 \pm 0.04\%$  of fillet with the skin at the observed level of  $15.1 \text{ g kg}^{-1}$  of lysine for the Nile tilapia *Oreochromis niloticus* (Linnaeus, 1758), in the final production phase, with the maximum yield estimated at  $14.6 \text{ g kg}^{-1}$ , corresponding to 5.8% of the protein in the diet. The percentage remained below the values for the tambaqui yield, for which the highest response was at the level of  $15.96 \text{ g kg}^{-1}$  lysine with 38.24% fillet (Table 4).

**Figure 1.** Morphometric measurements in tambaqui *Colossoma macropomum* in the juvenile phase. TL: total length, SL: standard length, TRL: trunk length, HL: head length, HH: head height, BH: body height, TH: tail height, LCFL: lower caudal fin length, UCFL: upper caudal fin length, and LT: lumbar thickness.

Tambaqui specimens caught in Amazonian rivers yield about 32.00 to 35.96% of fillet with skin and 25.7 to 29.08% skinless fillet (Souza & Inhamuns 2011). Only the treatment with 0.8% L-lysine remained below these values (22.91%), and the others were close to or above that described. The percentage yield of the fillet can vary between 25.4 to 42.0% according to the biological particularities of each species and factors such as nutrition (Adames et al. 2014).

**Table 4.** Fillet characterization and carcass yield of juvenile tambaqui *Colossoma macropomum* fed diets with increasing levels of lysine. Analysis of variance ( $P < 0.05$ ). \*Significant values in the same row. Means followed by the same lowercase letter do not differ statistically from each other by the Tukey test ( $P < 0.05$ ).

Variables (%)	Lysine (g kg <sup>-1</sup> )						P
	6.60	9.72	12.84	15.96	19.08	22.20	
<b>Product</b>							
Gutted carcass	89.37 ± 0.80	89.76 ± 1.18	90.12 ± 0.70	90.83 ± 1.29	82.28 ± 9.05	90.42 ± 1.70	0.29
Skinless fillet	24.66 ± 11.01	24.64 ± 4.03	22.91 ± 2.28	26.66 ± 2.93	25.75 ± 2.04	28.81 ± 1.93	0.27
<b>Fillet</b>							
with skin	34.92 ± 3.53	34.56 ± 5.74	36.54 ± 2.44	38.24 ± 1.44	35.75 ± 2.69	37.08 ± 2.00	0.71
<b>Chemical composition</b>							
Crude protein	18.26 ± 0.65	16.94 ± 0.43	14.28 ± 0.68	16.34 ± 1.94	18.13 ± 0.36	16.80 ± 0.05	0.05
Ether extract	2.53 ± 0.16	1.62 ± 0.21	2.25 ± 0.01	2.51 ± 0.73	2.08 ± 0.45	1.75 ± 0.06	0.06
Total minerals	1.28 ± 0.03 <sup>a</sup>	1.30 ± 0.01 <sup>abc</sup>	1.22 ± 0.01 <sup>ab</sup>	1.31 ± 0.01 <sup>abc</sup>	1.16 ± 0.01 <sup>ab</sup>	1.27 ± 0.01 <sup>c</sup>	0.01*
Moisture	76.16 ± 0.01 <sup>a</sup>	77.26 ± 0.01 <sup>a</sup>	77.75 ± 0.01 <sup>a</sup>	77.95 ± 0.11 <sup>a</sup>	77.49 ± 0.12 <sup>a</sup>	79.12 ± 0.04 <sup>b</sup>	0.00*

**Table 5.** Average yields of by-products and waste from juvenile tambaqui *Colossoma macropomum* fed diets with increasing levels of lysine. Analysis of variance ( $P < 0.05$ ). \*Significant values in the same row. Means followed by the same letter do not differ from each other by the Tukey test ( $P < 0.05$ ).

Variables	Lysine (g kg <sup>-1</sup> )						P
	6.60	9.72	12.84	15.96	19.08	22.20	
Whole fish (g)	58.22 ± 9.17	.	57.89 ± 5.03	70.47 ± 6.97	57.76 ± 10.87	65.82 ± 1.38	0.46
<b>By-products (%)</b>							
Clean trunk	23.01 ± 1.06	24.36 ± 4.88	24.54 ± 0.86	25.63 ± 1.65	23.44 ± 0.72	27.08 ± 2.00	0.66
Gross skin	10.26 ± 0.71 <sup>ab</sup>	9.92 ± 1.83 <sup>ab</sup>	13.63 ± 4.34 <sup>a</sup>	11.57 ± 1.69 <sup>ab</sup>	10.01 ± 0.49 <sup>ab</sup>	8.27 ± 0.81 <sup>b</sup>	0.03*
<b>Waste (%)</b>							
Head	21.29 ± 1.98	20.08 ± 1.44	20.83 ± 0.59	19.37 ± 1.12	20.15 ± 1.06	20.96 ± 0.90	0.59
<b>Abdominal</b>							
ventral muscle	3.16 ± 0.61	2.94 ± 1.00	3.38 ± 0.93	3.93 ± 0.85	3.21 ± 0.96	3.19 ± 0.63	0.77
Fins	1.628 ± 0.64 <sup>a</sup>	2.67 ± 1.23 <sup>b</sup>	2.85 ± 0.07 <sup>b</sup>	2.84 ± 0.29 <sup>b</sup>	2.76 ± 0.09 <sup>b</sup>	2.54 ± 0.25 <sup>b</sup>	0.00*
Gills	3.63 ± 0.37	3.38 ± 0.36	3.49 ± 0.85	3.89 ± 0.36	3.70 ± 0.64	3.59 ± 0.31	0.89
Viscera	8.37 ± 0.00	7.95 ± 0.01	7.37 ± 0.51	7.43 ± 0.01	7.18 ± 0.01	8.42 ± 0.01	0.12

Carneiro et al. (2004) observed maximum fillet yields of 34.74% for the gray jundiá, *Rhamdia quelen* (Quoy & Gaimard, 1824), while for the barbado *Pirinampus pirinampu* (Spix & Agassiz, 1829), the percentage of the fillet in relation to the weight of the whole fish found by Adames et al. (2014) was from 38.57 to 42.13%.

Lima et al. (2018) found a higher percentage in tambaqui with lower weights, and the yield of fillet with skin was up to 57.5 ± 8.7%, extrapolating the value of 35.96% observed in the study of Souza and Inhamuns (2011) (weights between 1071.2 and 1229.0 g).

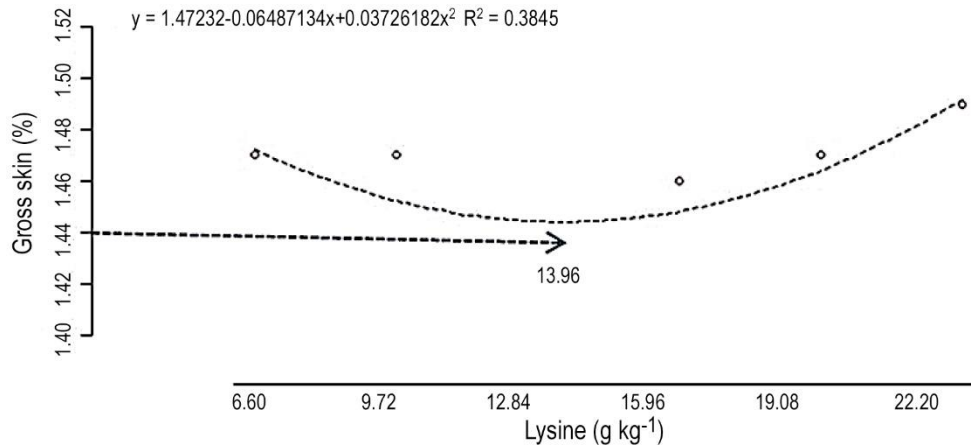
The chemical composition of the fish fillet nutritionally characterizes the portion for human consumption and is a determining factor in the definition of equipment, in industrial processes, and definition of

standards for commercialization of the fish (Leonhardt et al. 2006).

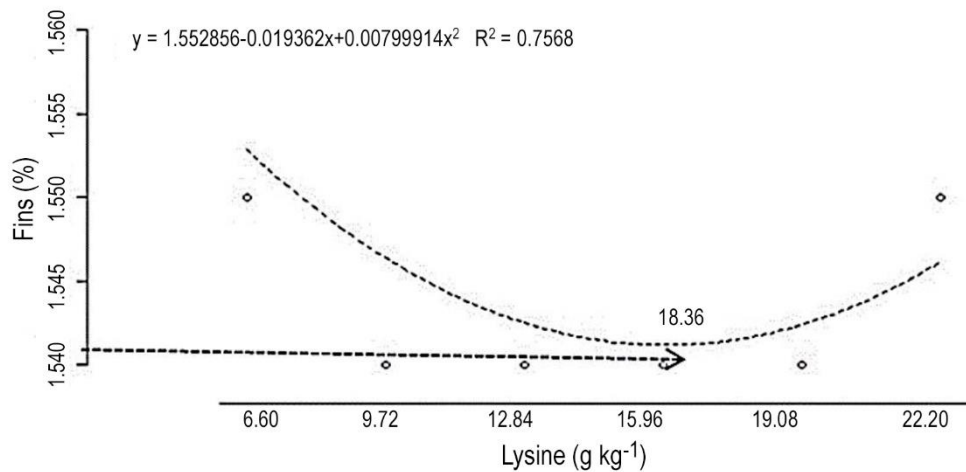
The contents of the chemical components of the fish range from 70.0 to 85.0% for moisture, 15.0 and 24.0% for protein, 1.0 and 2.0% for minerals, and can vary between species and within the same species, and is influenced by age, cultivation conditions, body portion and diet consumed (Arbeláez-Rojas et al. 2002).

Minerals in association with amino acids tend to increase the nutrient absorption in the intestine, providing transport through the mucous membranes and avoiding the formation of insoluble compounds with possible antinutritional factors of the diet (Barros et al. 2004).

In the fillets of fish fed with 22.20 g kg<sup>-1</sup> of total lysine, the moisture content found exceeded the others and presented a significant statistical difference (Table



**Figure 2.** Effect of quadratic regression on the percentage of gross skin yield of juvenile tambaqui *Colossoma macropomum* fed diets containing increasing levels of lysine.



**Figure 3.** Effect of the quadratic regression model on the percentage (arc sen  $\sqrt{P\%/100}$ ) 100 yield of fins of juvenile tambaqui *Colossoma macropomum* fed diets containing increasing levels of lysine.

4). However, the data are close to the values described by Oetterer et al. (2004) for red tilapia (79.20%) and Nile tilapia (78.43%). Notwithstanding, they are lower than the values found by Arbeláez-Rojas et al. (2002) for juvenile tambaqui ( $86.2 \pm 10.9$  g and  $15.1 \pm 0.5$  cm) in intensive and super-intensive cultivation systems.

Some authors describe an inversely proportional relationship between moisture and protein content in fish fillets (Oliveira et al. 2008, Moura et al. 2009). However, for the juvenile tambaqui, the increase in moisture observed in fish fillets with diets with 22.20 g kg<sup>-1</sup> of total lysine did not change the protein content. This absence of difference in protein content in tambaqui fillets may be related to the absence of significance between mean fillet yield and carcass yield.

The study by Furuya et al. (2006), involving Nile tilapia fed with lysine in the diet ( $5.72 \pm 0.10$  g), describes different results for gutted carcass from that found in the present study. The study describes positive linear behavior, attributed to lysine being an amino acid with a preference for deposition in muscle tissue. However, this trend did not occur for tambaqui. It is possible that the period necessary for balancing the relationship of lysine in the diet with the body amino acid profile under expression of carcass yield is longer than the experimental period used in the present study with tambaqui.

#### By-products and waste

The information obtained regarding the by-products favors innovations in the use of fish and offers options



in the processing of native species, which tend to be marketed in traditional ways as whole fish, gutted fish, or fillets (Bombardelli & Sanches 2008).

The skin yield percentages observed for tambaqui ranged from 6.29 to 6.89%. For pacu, the oscillation varies between 8.90 and 9.58%, and for matrinxã *Brycon amazonicus* (Spix & Agassiz, 1829), between 9.46 and 12.8% (Souza & Inhamuns 2011). However, in general, the percentage of skin for teleost fish is 7.5% (Contreras-Guzmán 1994, Simões et al. 2007).

Although the data obtained (Table 5) exceed the expected amount of epithelial tissue for bony fish, the juvenile tambaqui fed with 22.20 g kg<sup>-1</sup> of total lysine remained close to this estimate. However, the same did not occur for juvenile tambaqui from the other treatments, especially for fish that received diets containing 12.84 g kg<sup>-1</sup> of total lysine, which produced a greater amount of gross skin and differed statistically from fish fed with 22.20 g kg<sup>-1</sup> of total lysine. This trend, observed in the higher levels of lysine (15.96, 19.08, and 22.20 g kg<sup>-1</sup> of total lysine), in reducing the percentage of skin shows that the lysine that is not transformed into the skin may have been used for the maintenance of other biological processes or has been used to generate a greater amount of fillet.

For Nile tilapia in the adult phase, the study by Souza & Maranhão (2001), with fish weighing between 300 and 500 g, described values of 6.16 and 6.56% gross skin. The study by Souza et al. (2005) describes data for the percentage of skin between 6.30 to 6.71% (500 to 800 g), while the study by Silva et al. (2009) indicates between 5.77 to 6.31% (200 to 600 g). All these data were below the values of the skin by-product for tambaqui juveniles fed with lysine. Thus skin can be directed for use in other foods, manufacturing, or collagen extraction.

Adequate levels of lysine in the diet aids preventing fin damage and tend to improve the zootechnical performance and yields of the usable fish fractions (Takishita et al. 2009, Furuya et al. 2013). The values observed and estimated for fins in the present study are lower than the percentages of between 8.0 and 8.1 found for Nile tilapia (300 to 500 g) in the study of Souza & Maranhão (2001), and the values of 3.6 to 4.2% described by Silva et al. (2009) for Nile tilapia of weights between 300 and 500 g.

However, the information on the yield of tambaqui fins with higher percentages observed at 12.84 and 15.96 g kg<sup>-1</sup> of total lysine is in accordance with the response obtained in the morphometry ratio for SL:TL, which indicated a possible influence of lysine on the fins.

## CONCLUSION

The inclusion of lysine in diets of juvenile tambaqui *C. macropomum* did not significantly favor the percentage of by-products, waste, or the fish's morphometry. Thus, we cite the need for further investigations into the behavior of the nutrient on the potentially marketable portions of the Amazonian fish species and conditioning measures in cultivation and processing that allow greater use of fish and thus maximize productive gain.

## ACKNOWLEDGMENTS

A.T. Oliveira (Process 315713/2020-8) and W.M. Furuya are CNPq research fellows. This study was funded by the Amazonas State Research Support Foundation (FAPEAM, POSGRAD 2020/2021 - Resolution 006/2020).

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Received: February 22, 2021; Accepted: May 30, 2021