

# FEED MANAGEMENT OF PACU JUVENILES CULTIVATED IN NET CAGES

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## ABSTRACT

The aim of this work was to evaluate the feed management (feed percentage and feeding frequency) of pacu (*Piaractus mesopotamicus*) juveniles bred in net cages. To evaluate the feed percentage 2,000 juveniles (12.12±0.51 g) were randomly distributed in 20 cages of 1 m<sup>3</sup> (0.8 m<sup>3</sup> of useful volume). The fish were fed four times a day (8:00 a.m.; 11:00 a.m.; 2:00 p.m.; and 5:00 p.m.) with 4, 7, 10, 13, and 16% of feed. To evaluate feeding frequency, another 2,000 juveniles (9.56±0.56 g) were randomly distributed in 20 cages of 0.8 m<sup>3</sup> of useful volume. The fish were fed one, two, three and four times a day. Due to the feed percentage, significant effects (P<0.05) were observed for the parameters of weight gain, final length, final biomass, specific growth rate, and apparent feed conversion. The performance parameters, centesimal composition, and blood biochemistry were not influenced (P>0.05) by the feeding frequency. Considering the weight gain and apparent feed conversion of pacu juveniles cultivated in net cages, 8% of feed is recommended regardless of the feeding frequency adopted.

**Key words:** intensive aquaculture; feeding frequency; native fishes; blood parameters; feed percentage.

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## MANEJO ALIMENTAR DE JUVENIS DE PACU CRIADOS EM TANQUES REDE

### RESUMO

O objetivo do presente trabalho foi avaliar o manejo alimentar (porcentagem de arraçoamento e frequência alimentar) de juvenis de pacu (*Piaractus mesopotamicus*) criados em tanques rede. Para avaliar a porcentagem de arraçoamento foram utilizados 2.000 juvenis (12,12±0,51 g), distribuídos em 20 tanques rede de 0,8 m<sup>3</sup> de volume útil, alimentados quatro vezes ao dia (8, 11, 14 e 17 horas), com 4, 7, 10, 13 e 16% de arraçoamento. Para avaliar a frequência alimentar, foram utilizados 2.000 juvenis (9,56±0,56 g), distribuídos em 20 tanques rede com 0,8 m<sup>3</sup> de volume útil, alimentados uma, duas, três e quatro vezes ao dia. Observou-se efeito significativo (P<0,05) para os parâmetros de ganho de peso, comprimento final, biomassa final, e conversão alimentar aparente, em função do percentual de arraçoamento. Não foram observadas influências (P>0,05) da frequência alimentar sobre os parâmetros zootécnicos, composição centesimal e bioquímicos do sangue. Considerando o ganho de peso e conversão alimentar aparente de juvenis de pacu criados em tanques rede, recomenda-se 8% de arraçoamento, independentemente da frequência alimentar adotada.

**Palavras-chave:** aquicultura intensiva; frequência alimentar; peixes nativos; parâmetros sanguíneos; porcentagem de arraçoamento.

## INTRODUCTION

Determining of the feed percentage and feeding frequency for different species of fish is fundamental for the production of fish with satisfactory economic return to the producer. Supplying more feed than is consumed by fish may result in food waste, negatively influencing water quality (MEURER *et al.*, 2005). The optimum feeding rate is the one that provides the best apparent feed conversion, together with the best weight gain of the animals. On the other hand, when the rate is not well defined, it can cause uneven growth, raise production costs, and impair water quality.

However, the feeding rate varies with the age of the fish and with the temperature of the water (SANTIAGO *et al.*, 1987; DENG *et al.*, 2003; HAYASHI *et al.*, 2004;

SANTOS *et al.*, 2013). Moreover, it is necessary to make constant adjustments in the amount of feed to be offered to the animals (SALARO *et al.*, 2008), which is a fundamental practice in intensive production systems (SCORVO FILHO *et al.*, 2010).

The feeding frequency influences the development of the animals and was studied in different species of fish (LEE *et al.*, 2000a; LEE *et al.*, 2000b; DWYER *et al.*, 2002; BITTENCOURT *et al.*, 2013; SOUZA *et al.*, 2014, SANTOS *et al.*, 2015); observing an increase in weight gain when fed more than once a day. The objective of the present work was to evaluate the feeding frequency and feed percentage of juvenile pacu (*Piaractus mesopotamicus*) raised in net cages.

## METHODS

The trials were conducted at the Fisheries and Ecology of Native Species Research Station at the Binational Itaipu Reservoir. The entire experimental procedure was evaluated and approved by the Committee of Ethics in the Use of Animals of the West Paraná State University, according to certificate N° 23/2016 CEUA.

### Experimental design - feed percentage - experiment I

To evaluate the feed percentage, 2000 juveniles ( $12.12 \pm 0.51$  g) were randomly distributed in 20 net cages with  $0.8 \text{ m}^3$  of useful volume, holding 100 fish per tank. The net cages consisted of a 15 mm, 1.1 wire, stainless steel screen arranged in lines, respecting a distance of 1.5 m between the net cages. The fish were fed at 8:00 a.m.; 11:00 a.m.; 2:00 p.m.; and 5:00 p.m., according to feed offer rates of 4, 7, 10, 13 and 16% of live weight, for a period of 60 days (03/21/2015 to 5/19/2015).

### Experimental design - feeding frequency - experiment II

To evaluate the feeding frequency, 2,000 juveniles were used, with a mean initial weight of  $9.67 \pm 0.55$  g, distributed randomly in 20,  $0.8 \text{ m}^3$  net cages (100 fish per tank). The net cages consisted of a 15 mm, 1.1 wire, stainless steel screen arranged in lines, respecting a distance of 1.5 m between the net cages. The fish were submitted to different feeding frequencies/schedules (Table 1), for a period of 114 days (10/21/2015 to 02/11/2016). Biometrics (weighing

and counting) of 100% of the fish were performed biweekly for the correction of the feed that was provided based on 7% of live weight. The amount of feed was given according to the frequencies adopted, once daily (100% of the feed at once), twice daily (50% at each feeding), three times a day (33, 33% at each feeding), and four times a day (25% at each feeding).

Extruded commercial feeds of 2 mm and 32% crude protein were used. The chemical compositions of the feeds are shown in Table 2.

The water quality variables such as temperature, dissolved oxygen and pH were monitored daily in the morning using the HORITA® U50 multiparameter probe. The averages for Experiment I were  $28.4 \pm 1.09$  °C,  $7.66 \pm 0.93$  mg L<sup>-1</sup> and  $7.68 \pm 0.24$  for temperature, dissolved oxygen and pH, respectively. For Experiment II,  $27.10 \pm 4.77$  °C,  $6.19 \pm 1.08$  mg L<sup>-1</sup> and  $6.90 \pm 0.49$  for temperature, dissolved oxygen and pH, respectively.

### Blood collection, hematological and biochemical analysis - experiments I and II

At the end of the 60-day (Experiment I) and 114-day (Experiment II) experimental periods, the fish were fasted for 12 hours to empty their digestive tracts, then were anesthetized with benzocaine ( $60 \text{ mg L}^{-1}$ ) (GOMES *et al.*, 2001). Two fish from each tank (experiments I and II) (ten fish per treatment) were submitted to blood collection (1.5 mL) by caudal puncture with the aid of a syringe. For each aliquot of blood collected, the counts of the number of erythrocytes were performed in a Neubauer chamber (COLLIER, 1944). For the determination of hemoglobin and hematocrit, the methodology of COLLIER (1944) and GOLDENFARB *et al.* (1971) were followed, respectively. Subsequently, the absolute hematimetric indices (WINTROBE, 1934) were calculated as mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH) and mean corpuscular hemoglobin concentration (MCHC).

For the biochemical analyses in experiment II, the following parameters were evaluated: serum, for dosage of protein (WEICHSELBAUM, 1946), triglycerides (TRINDER, 1969; BUCOLO and DAVID, 1973), cholesterol (TRINDER, 1969; ALLAIN *et al.*, 1974), and plasma (with fluoride) for glucose dosing (BERGMEYER, 1986). To determine the analyses, which were performed with the use of specific Gold Analisa Diagnóstica® kits, and the reading was made in a spectrophotometer, according to the instructions of the manufacturer. The colorimetric enzymatic

**Table 1.** Feed frequency / feeding time of fish.

Feeding frequency	Feeding times / quantity of feed provided			
	8:00	11:00	14:00	17:00
1X/day	X	-	-	-
2X/day	X	-	-	X
3X/day	X	X	-	X
3X/day	X	-	X	X
4X/day	X	X	X	X

**Table 2.** Chemical composition of feeds.

Parameters	Experiments I	Experiments II
Dry matter (%)	91.77	90.55
Crude protein (%)	31.90	32.06
Lipids (%)	6.18	5.85
Mineral matter (%)	9.29	10.05
Energy (Kcal kg <sup>-1</sup> )	4539.02	4436.11

**Table 3.** Mathematical modeling of statistical analysis.

Response Variables	Regression Parameters*			R <sup>2</sup>	Mathematical Model
	A	B	C		
WG (g)**	29.51±3.73	6.11±0.84	-0.245±0.041	0.962	Y = A + B.x + C.x <sup>2</sup>
CF (cm)**	11.75±0.48	0.42±0.11	-0.202±0.005	0.877	Y = A + B.x + C.x <sup>2</sup>
BioF (g)**	3979.56±210.94	666.28±47.29	-27.19±2.33	0.989	Y = A + B.x + C.x <sup>2</sup>
AFC***	0.74±0.045	0.31±0.004	0	0.999	Y = A + B.x

WG = weight gain; CF = final length; BioF = final biomass; AFC = apparent feed conversion; \*Values expressed as mean ± standard error; \*\*Best mathematical adjustment by quadratic regression; \*\*\*Best mathematical fit by linear regression.

method was used for the dosages of cholesterol, triglycerides and glucose, and the colorimetric-biuret was used for protein analysis.

### Zootechnical performance - experiments I and II

All fish were weighed and measured individually. Upon obtaining the data the following parameters were calculated: weight gain (WG = final weight - initial weight), final biomass (BioF = final weight \* number of individuals), final length (FL), condition factor (CF = (final weight / final length<sup>3</sup>) \* 100), survival (SUR = (number of fish at the end / number of fish initial) \* 100), apparent feed conversion (AFC = feed intake / weight gain), and specific growth rate (SGR = 100 \* [(ln average final weight - ln average initial weight) / time]).

### Centesimal composition - experiments I and II

Five fish from each tank (20 fish per treatment) were separated, euthanized with 250 mL L<sup>-1</sup> of benzocaine (GOMES *et al.*, 2001), identified, and frozen (-18 °C) for further analysis of centesimal composition. The analysis of centesimal composition followed the methodology recommended by AOAC (2000). The following analyses were performed: Dry matter, at 105°C in a greenhouse (ASL102, Solab, Piracicaba/SP, Brazil) for 8 hours; Mineral matter, at 550 °C by muffle incineration (0318m25T, Quimis, Diadema / SP, Brazil); Lipids, by Soxhlet extractor (TE-044-5, Tecnal, Piracicaba/SP, Brazil); Determination of total nitrogen by the Kjeldahl method (MA036, Marconi, Piracicaba/SP, Brazil), and nitrogen conversion factor for protein 6.25.

### Statistical analyzes - feed percentage - experiment I

All data were submitted to analysis of variance (ANOVA) at significance levels of 1 and 5%, checking the assumptions of normality and homogeneity. In the case of statistical differences, the regression test was applied for the elaboration of models in the matters of weight gain, final length, final biomass and apparent feed conversion. A completely randomized experimental design (CRD) was used, using the statistical program STATISTICA 8.0 (Copyright® StatSoft). The results were represented by the mean ± standard deviation.

### Variable behavioral responses test - experiment I

In order to evaluate the behavior of the varying responses to changes in the feed percentage, the data of weight gain, final length and final biomass were analyzed, respectively, by quadratic regression analysis (Table 3), and for the feed conversion data was analyzed by linear regression (Table 3). To obtain the curves and identify adjustable parameters, the software OriginPro version 8 SR0 (OriginLab®) was used. The applicability of the mathematical models was evaluated by the most appropriate determination coefficient, R<sup>2</sup>.

### Statistical analyses - feeding frequency - Experiment II

All data were submitted to analysis of variance (ANOVA) at the 5% level of significance, and the normality and homogeneity assumptions were checked, following statistical program STATISTIC 8.0 (Copyright® StatSoft) protocol, GLM. The results were represented as the mean ± standard deviation.

## RESULTS

### Feed percentage - experiment I

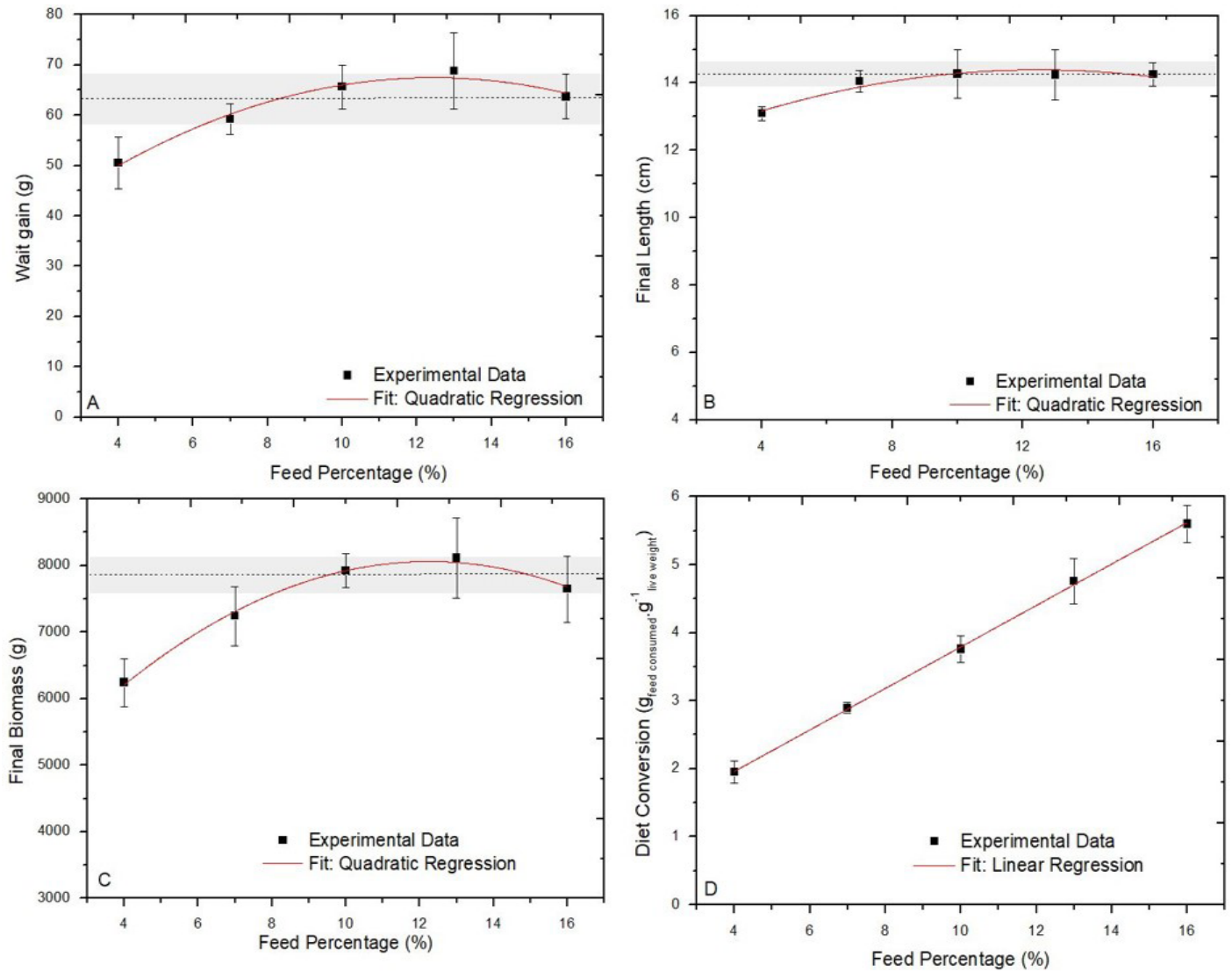
Quadratic effects were observed for weight gain (Figure 1A), final length (Figure 1B) and final biomass (Figure 1C), and there was a linear effect observed for apparent feed conversion (Figure 1D).

The influence of the feeding rates on the condition factor, survival and centesimal composition of the carcass of pacu juveniles were not observed (P > 0.05) (Table 4).

Regarding hematimetric hematological parameters (Table 5), no influences were observed (P > 0.05) among the different feed percentages.

### Feeding frequency - experiment II

No influence of feeding frequency was observed on performance, centesimal composition (Table 6), and hematological and biochemical parameters (Table 7).



**Figure 1.** Effects of feed percentage about: A = weight gain; B = final length; C = final biomass and D = feed conversion.

**Table 4.** Performance and centesimal composition (means ± standard deviation) of pacu juveniles submitted to different feeding rates.

Parameters	Feed Percentages (%)*					P-value**
	4	7	10	13	16	
Productive performance						
IW (g)	11.99±0.79	12.02±0.63	12.21±0.38	12.23±0.47	12.17±0.52	0.9592
CF	2.24±0.11	2.21±0.14	2.26±0.17	2.33±0.19	2.28±0.06	0.8110
SUR (%)	100	100	100	100	100	0.8691
Centesimal Composition						
HUM (%)	67.73±1.06	67.50±0.87	66.83±0.23	66.13±1.70	65.73±2.56	0.3171
CP (%)	16.75±0.46	16.17±1.37	16.47±1.10	16.65±1.66	18.30±1.42	0.2010
LP (%)	11.12±0.36	11.53±0.78	12.76±0.37	12.39±0.94	13.26±1.66	0.0552
MM (%)	4.55±0.40	4.58±0.56	4.57±0.53	4.50±0.16	4.43±0.17	0.9831

IW = initial weight; CF = condition factor; SUR = survival; HUM = humidity; CP = crude protein; LP = lipids; MM = mineral matter; \*Mean values ± standard deviation; \*\*Values of p-values obtained by ANOVA, with a 95% confidence level.

**Table 5.** Haematological and hematimetric parameters (means  $\pm$  standard deviation) of pacu juveniles submitted to different feeding rates.

Parameters	Feed Percentages (%)*					P-value**
	4	7	10	13	16	
Hematology						
VG (%)	33.69 $\pm$ 2.58	32.75 $\pm$ 2.89	35.67 $\pm$ 1.74	36.06 $\pm$ 1.55	36.00 $\pm$ 1.63	0.812
ERY ( $10^6 \mu\text{L}^{-1}$ )	1.91 $\pm$ 0.16	1.95 $\pm$ 0.19	1.94 $\pm$ 0.22	1.95 $\pm$ 0.202	2.01 $\pm$ 0.51	0.975
HE (g dL <sup>-1</sup> )	7.63 $\pm$ 0.87	7.54 $\pm$ 0.94	8.30 $\pm$ 0.58	8.1 $\pm$ 0.71	8.30 $\pm$ 1.11	0.234
Hematimetrics						
MCH (g dL <sup>-1</sup> )	2.53 $\pm$ 0.32	2.62 $\pm$ 0.42	2.34 $\pm$ 0.24	2.40 $\pm$ 0.20	2.41 $\pm$ 0.48	0.354
MCV (fL)	177.11 $\pm$ 13.90	169.07 $\pm$ 21.47	170.37 $\pm$ 52.64	186.71 $\pm$ 17.08	191.79 $\pm$ 78.46	0.785
MCHC (g dL <sup>-1</sup> )	22.70 $\pm$ 2.54	23.08 $\pm$ 2.57	23.28 $\pm$ 1.47	22.52 $\pm$ 1.85	23.18 $\pm$ 3.81	0.341

VG = hematocrit; ERY = erythrocytes; HE = hemoglobin; MCH = mean corpuscular hemoglobin; MCV = mean corpuscular volume; MCHC = mean corpuscular hemoglobin concentration; \*Mean values  $\pm$  standard deviation; \*\* P-values obtained by ANOVA, with a 95% confidence level.

**Table 6.** Performance and centesimal composition (means  $\pm$  standard deviation) of pacu juveniles submitted to different feeding frequencies.

Parameters	Feeding Frequencies*					P-value**
	1X/day	2X/day	3X/day	3X/day	4X/day	
Performance						
IW (g)	9.55 $\pm$ 0.89	9.60 $\pm$ 0.84	9.75 $\pm$ 0.48	9.75 $\pm$ 0.40	9.68 $\pm$ 0.30	0.987
WG (g)	199.12 $\pm$ 22.76	206.49 $\pm$ 11.64	185.58 $\pm$ 28.40	194.77 $\pm$ 38.28	178.88 $\pm$ 23.80	0.611
BioF (kg)	18.10 $\pm$ 1.60	18.85 $\pm$ 1.62	15.92 $\pm$ 2.82	14.65 $\pm$ 3.73	16.58 $\pm$ 0.82	0.139
FL (cm)	21.37 $\pm$ 1.43	21.69 $\pm$ 2.36	20.28 $\pm$ 1.03	20.96 $\pm$ 0.93	20.07 $\pm$ 0.99	0.276
SUR (%)	87.25 $\pm$ 9.07	87.50 $\pm$ 9.71	81.25 $\pm$ 4.79	72.00 $\pm$ 15.87	88.75 $\pm$ 8.38	0.168
AFC	1.66 $\pm$ 0.04	1.69 $\pm$ 0.06	1.79 $\pm$ 0.16	2.05 $\pm$ 0.53	1.72 $\pm$ 0.15	0.268
CF	2.05 $\pm$ 0.28	2.04 $\pm$ 0.26	2.21 $\pm$ 0.10	2.10 $\pm$ 0.21	2.20 $\pm$ 0.06	0.641
SGR	2.70 $\pm$ 0.11	2.73 $\pm$ 0.06	2.62 $\pm$ 0.15	2.66 $\pm$ 0.14	2.60 $\pm$ 0.14	0.556
Centesimal Composition						
HUM (%)	63.84 $\pm$ 3.03	63.32 $\pm$ 3.71	64.68 $\pm$ 4.65	62.61 $\pm$ 1.50	64.74 $\pm$ 2.69	0.872
CP (%)	18.03 $\pm$ 2.18	18.25 $\pm$ 1.73	18.85 $\pm$ 1.91	18.88 $\pm$ 1.75	18.26 $\pm$ 3.37	0.974
LP (%)	15.96 $\pm$ 1.32	17.14 $\pm$ 4.95	15.43 $\pm$ 3.33	17.34 $\pm$ 1.85	14.81 $\pm$ 1.54	0.700
MM (%)	2.91 $\pm$ 0.52	3.65 $\pm$ 0.86	3.62 $\pm$ 0.44	3.34 $\pm$ 0.49	3.58 $\pm$ 0.48	0.384

IW = initial weight; WG = weight gain; BioF = final biomass; FL = final length; SUR = Survival; AFC = apparent feed conversion; CF = condition factor; SGR = specific growth rate; HUM = humidity; CP = crude protein; LP = lipids; MM = mineral matter; \*Mean values  $\pm$  standard deviation; \*\*P-values obtained by ANOVA, with a 95% confidence level.

**Table 7.** Hematological, hematimetric and biochemical parameters (means  $\pm$  standard deviation) of pacu juveniles submitted to different feeding frequencies.

Parameters	Daily Feeding Frequency*					P-value**
	1X	2X	3X	3X	4X	
Hematological						
VG (%)	35.25 $\pm$ 3.52	36.88 $\pm$ 4.17	33.88 $\pm$ 1.45	36.12 $\pm$ 2.72	36.00 $\pm$ 3.08	0.716
HE (g dL <sup>-1</sup> )	3.75 $\pm$ 0.15	3.63 $\pm$ 0.36	3.49 $\pm$ 0.32	3.54 $\pm$ 0.17	3.70 $\pm$ 0.54	0.795
ERY ( $10^6 \mu\text{L}^{-1}$ )	171.08 $\pm$ 27.26	185.87 $\pm$ 10.75	176.02 $\pm$ 29.74	181.16 $\pm$ 15.56	185.46 $\pm$ 18.41	0.842
Hematimetric						
MCV (fL)	2.08 $\pm$ 0.21	1.99 $\pm$ 0.25	1.97 $\pm$ 0.36	2.00 $\pm$ 0.18	1.97 $\pm$ 0.37	0.977
MCH (g dL <sup>-1</sup> )	0.22 $\pm$ 0.04	0.20 $\pm$ 0.03	0.20 $\pm$ 0.02	0.20 $\pm$ 0.03	0.20 $\pm$ 0.03	0.658
MCHC (g dL <sup>-1</sup> )	10.71 $\pm$ 1.06	9.86 $\pm$ 0.39	10.31 $\pm$ 0.97	9.85 $\pm$ 1.02	10.34 $\pm$ 1.64	0.771
Biochemical						
GL (mg dL <sup>-1</sup> )	79.61 $\pm$ 17.25	76.89 $\pm$ 16.66	71.13 $\pm$ 26.95	78.47 $\pm$ 10.07	65.71 $\pm$ 8.53	0.934
CL (mg dL <sup>-1</sup> )	139.35 $\pm$ 33.34	148.74 $\pm$ 15.58	153.50 $\pm$ 10.43	152.99 $\pm$ 6.34	145.71 $\pm$ 20.74	0.839
AB (g dL <sup>-1</sup> )	0.80 $\pm$ 0.07	0.76 $\pm$ 0.09	0.83 $\pm$ 0.08	0.75 $\pm$ 0.04	0.78 $\pm$ 0.08	0.606
PR (g dL <sup>-1</sup> )	3.30 $\pm$ 0.10	3.26 $\pm$ 0.12	3.28 $\pm$ 0.28	3.36 $\pm$ 0.10	3.03 $\pm$ 0.26	0.186
TR (mg dL <sup>-1</sup> )	318.67 $\pm$ 107.56	363.30 $\pm$ 151.38	299.53 $\pm$ 17.75	325.02 $\pm$ 49.16	347.18 $\pm$ 63.47	0.872

VG = hematocrit; HE = hemoglobin; ERY = erythrocytes; MCV = mean corpuscular volume; MCH = mean corpuscular hemoglobin; MCHC = mean corpuscular hemoglobin concentration; GL = glucose; CL = cholesterol; AB = albumin; PR = protein; TR = triglyceride; \*Mean values  $\pm$  standard deviation; \*\*P-values obtained by ANOVA, with a 95% confidence level.



## DISCUSSION

### Feed percentage - experiment I

The linear effect observed by the apparent feed conversion showed that the feeding rate (4%) did not allow the necessary intake of nutrients for the expression of the maximum pacu growth, however, it did result in a better utilization of dietary nutrients by the fish, resulting in a lower index of apparent feed conversion. At higher feed rates, remains, non-ingestion and/or under-utilization of fish feed resulted in nutrient waste in the aquatic environment, impairing apparent feed conversion. The quadratic effect observed for weight gain, shows, through the regression equation, that the best result occurs for 12.46% of feed, but it raises apparent food conversion too much (4.80), due to the linear effect; implying higher costs with feed use. However, the intersection of the quadratic fit with the dotted line (Figure 1A) occurs close to the 8% feed, reducing feed conversion by 30.79%.

Quadratic effects for weight gain, final length and final biomass, and linear effect for apparent feed conversion were reported by MEURER *et al.* (2005) for Nile tilapia submitted to 1, 4, 7, 10, 13 and 16% of feed. Those fish that receive a higher feed percentage, had their growth impaired mainly due to the overfeeding, which culminates in elevated feed conversion indices. The leftovers of the rations are directly related to satiety, which for pacu juveniles cultivated in net cages, reported by HILBIG *et al.* (2012) have shown that a 70% restriction of feed in relation to satiety, improves apparent feed conversion and does not impair pacu performance.

For Nile tilapia fingerlings, SANTOS *et al.* (2015) show that the feeding rate that presented the best weight gain was 9 and 12% of feed, however, these rates present high indices of apparent feed conversion, due to excess feed, similar responses were observed for those pacu that were fed with rates of 10, 13 and 16% culminating with higher apparent feed conversion rates. For tambaqui, CHAGAS *et al.* (2005) did not observe influence of 5, 7.5, and 10% of feed on final weight. However, the best apparent feed conversion was observed for those fed with 5% of live weight. These influences may be related to their growth phase (DENG *et al.*, 2003) and water temperature (SANTIAGO *et al.*, 1987) that directly influence establishing an ideal rate of fish feeding.

When fish are fed at rates above their capacity to ingest, feed leftovers may occur, worsening feed conversion rates (MARQUES *et al.*, 2004; MEURER *et al.*, 2005) because the feed contacting the water drains and solubilizes the nutrients. High feed rates may reduce growth, as well as increase production costs, feed waste, and compromise crop water quality (SALARO *et al.*, 2008). According to MEURER *et al.* (2005), the problem with water quality is related to excess feed, and consequently reduction of oxygen levels dissolved during the earlier morning hours, due to the excess of eutrophication. This can culminate in reduced performance, susceptibility to diseases, and mortality; a factor not observed for the pacu.

There was no influence of the feeding rate on the condition factor, showing a good nutritional and physiological status of the fish.

According to NG *et al.* (2000) and MIHELAKAKIS *et al.* (2002), the amount of food supplied not only affects the conversion efficiency of food to muscle mass, but also the condition factor, which is widely used to evaluate the nutritional status of the fish, since it is a good indicator of their physiological condition.

Among the parameters of centesimal composition, no influence of the feeding rate was observed (Table 4) when the whole fish was submitted for analysis, which shows that the different feeding rates do not interfere in their chemical composition. Levels of body fat of the common carp carcass, are related to higher feeding rates according to SHIMENO *et al.* (1997).

The different feed percentages (4, 7, 10, 13 and 16) did not influence hematological parameters and hematimetric indices (Table 5), showing that the different rates do not interfere with animal health and well-being. HILBIG *et al.* (2012) worked with different feeding rates (70, 80, 90 and 100) for pacu with an initial size of  $84.75 \pm 4.52$  g and did not observe differences in the centesimal, hematological and hematimetric indices parameters. This demonstrates that the different feeding rates were sufficient to meet pacu requirements without compromising blood parameters.

### Feeding frequency - experiment II

It is possible to obtain similar growth regardless of the number of daily feeding, from one to four times a day, similar to the statement by LUZ and PORTELLA (2005); where they report that feeding frequency does not affect the performance of the animals. However, increasing feeding frequency allows for greater visual contact between the producer and the fish, allowing better monitoring of the animals' health status (CARNEIRO and MIKOS, 2005).

Recommendations of three daily feedings for pacu juveniles (65.9 grams) cultivated in 5m<sup>3</sup> net cages were reported by DIETERICH *et al.* (2013). Due to higher weight gain, the authors worked with a density of 40 fish m<sup>-3</sup>, compared to 125 fish m<sup>-3</sup> of the current work. This indicates that at low stocking densities fish have more space for swimming and present more growth, which directly influences the demand and availability of food. On the other hand, FIOD *et al.* (2010) reported that a feeding frequency of two times a day can be considered efficient for pacu juveniles (24 g) in the growth phase, due to greater weight gain and better specific growth rates, cultivated in controlled environments. However, in the current work only one daily feeding was enough for this stage of cultivation, because greater feeding frequencies did not improve the final weight. In contrast, the greater feeding frequency allowed for more contact with the animals, however the increased operating costs must be evaluated.

The fish feeding frequency is influenced by the developmental stage (DENG *et al.*, 2003), species, temperature and water quality (HAYASHI *et al.*, 2004; BARBIERI and BONDIOLI, 2015), the storage density is also added to these parameters. HAYASHI *et al.* (2004), for lambari fingerlings (0.34 g), and BITTENCOURT *et al.* (2013), for common carp (4.88g), recommend four daily feedings, however, the weights of the animals were lower than the current work, besides being developed in a controlled environment (laboratory). SANTOS *et al.* (2015)

evaluating the feeding frequency of Nile tilapia fingerlings (0.57g) in an excavated tank did not observe any influence of four and six daily feedings. This demonstrates that when cultivated in outdoor environments the food frequency has little influence on growth, possibly through supplementation through the natural productivity of the environment. SOUZA *et al.* (2014), evaluating the feeding frequency of two, four, six and eight times a day for tambaqui juveniles (16 g), show no influence on zootechnical parameters such as weight gain, apparent feed conversion, specific growth rate, and survival.

However, for this same species, CORRÊA *et al.* (2009), evaluating food frequencies (one, two and three times a day) with 100 g tambaqui cultivated in net cages, observed significant effects for weight gain and specific growth rates of the animals when fed with three daily servings of feed.

For matrinxã juveniles FRASCA-SCORVO *et al.* (2007) found that only one daily feeding was efficient for daily weight gain and apparent feed conversion rate. CANTON *et al.* (2007), for jundiá juveniles (8 to 45 g), recommend a frequency of at least two daily feedings, to provide better zootechnical performances of the fish. According to SAMPAIO *et al.* (2007) the ideal food frequency is a species-specific factor, and differences can occur within the same species depending on their stage of development.

The centesimal composition of the fish carcass expresses the quality and quantity of the nutrients via feed, however, the feeding frequencies adopted did not influence the centesimal composition (Table 6). This demonstrates that there is no nutrient deficiency at the level of affecting the carcass. HILBIG *et al.* (2012) and DIETERICH *et al.* (2013) also did not observe any interference of the feeding frequency on the centesimal composition of pacu.

The hematological and biochemical characteristics of the blood were not influenced ( $P < 0.05$ ) by the feeding frequency (Table 7). DIETERICH *et al.* (2013), for pacu, and SOUZA *et al.* (2014), for tambaqui, did not observe any effect of food frequency on biochemical parameters of the blood, similar to those observed for pacu in the current study.

Feeding management can directly influence economic returns, since feeding below physiological needs may result in low growth and increased growing time. On the other hand, the excess feed supplied, besides burdening production with the loss of feed, impairs the water quality of the crop by the solubilization of the nutrients. According to SANTOS *et al.* (2013) the feeding of fish should be done daily, always at the same time, aiming to condition the fish to seek food at predetermined times. However, it is important to avoid providing feed when the dissolved oxygen concentrations of the water are low.

## CONCLUSIONS

Considering the weight gain and apparent feed conversion of pacu juveniles raised in net cages, 8% of feed it is recommended, regardless of the feeding frequency adopted.

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