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The effect of mango waste meal in the protein:carbohydrate ratio on performance and body composition of pacamã fish (*Lophiosilurus alexandri*)

[*Efeito do farelo residual da manga na relação proteína:carboidrato sobre o desempenho e a composição corporal do pacamã* Lophiosilurus alexandri]

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ABSTRACT

Avaliou-se a inclusão do farelo residual de manga sem casca como fonte de carboidrato na relação proteína:carboidrato (PB:CHO) sobre o desempenho e a composição química da carcaça em juvenis de pacamãs. Foram utilizados 150 peixes (11,31±0,96g) estocados em 16 caixas de 500L, alimentados três vezes ao dia (10% peso vivo), em um sistema de recirculação de água com biofiltro. Os tratamentos consistiram em quatro dietas experimentais com níveis decrescentes da relação entre proteína bruta e carboidrato (1,40; 0,94; 0,56 e 0,29), com quatro repetições cada tratamento. Ao final de 60 dias, foram avaliados o desempenho zootécnico (ganho de peso médio final, taxa de crescimento específico, consumo total de ração aparente, conversão alimentar aparente, rendimento de carcaça, sobrevivência) e a composição físico-química da carcaça. As relações de proteína:carboidrato afetaram todas as variáveis de desempenho (P<0,05), com exceção da sobrevivência (P>0,05). Os valores da composição química da carcaça foram alterados, com exceção da matéria mineral, do pH e da umidade. A farinha de manga pode ser utilizada na proporção de até 15% na ração de pacamã, estabelecendo uma relação 1,40 PB:CHO, sem prejudicar o desempenho zootécnico e a composição química da carcaça.

Palavras-chave: nutrição, inclusão, carcaça

RESUMO

We evaluated the inclusion of peeled-mango waste meal as a source of carbohydrate in the protein:carbohydrate ratio (CP:CH) on performance and chemical composition of pacamã (Lophiosilurus alexandri) juveniles. One hundred and fifty fish (11.31±0.96g) were stocked in sixteen 500 L tanks, fed three times daily (10% of live weight), in a system with water recirculation with biofilter. The treatments consisted of four experimental diets with decreasing levels of the ratio between crude protein and carbohydrate (1.40, 0.94, 0.56 and 0.29), with four replications per treatment. At the end of 60 days, we evaluated animal performance (final average weight gain, specific growth rate, total apparent feed intake, carcass yield, survival) and physicochemical composition of the carcass. The protein:carbohydrate ratios affected all performance variables (P<0.05), except for survival (P>0.05). The carcass chemical composition variables were modified, except for mineral matter, pH and moisture. Mango meal can be used at the proportion of up to 15% in the diet for pacamã, establishing a CP:CHO ratio of 1.40 without impairing animal performance and the carcass chemical composition.

Keywords: nutrition, inclusion, carcass

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INTRODUCTION

Mango is cultivated in several regions of Brazil, especially in the northeast, because it presents excellent conditions for its development and production, especially in the Vale do São Francisco region. The mango fruit is widely available in this region, with a large volume of exports: in 2010, 108.38 thousand tons were exported, representing 90% of all national production (Coelho, 2002).

In Brazil, the deficiency of modern and appropriate techniques in handling, transportation and stocking of fruits, associated with the high perishability and lack of trained staff and infrastructure for their processing and preservation, have generated a loss of around 30% of the production, and in some cases it exceeds 50% (Argaiz *et al*., 1993), and often they are disposed of in the environment with no previous treatment, causing significant economic damages, in addition to environmental problems. However, fruits unsuitable for human consumption have nutritive potential and can be used as a source of carbohydrates in the formulation of diets for fish.

According to Monteiro (2009), mango contains 10.97 to 14.36g/100g carbohydrates; 11.77 to 22.5mg/100g vitamin C; and an energy content of 50.02 to 58.12kcal in g/100 g in its pulp and skin, respectively. Utilizing mango waste meal to feed Nile tilapia at 0, 5, 10 and 15%, Lima *et al*. (2011) did not observe differences in their performance and also in the apparent digestibility coefficients of dry matter, crude protein and gross energy, which enabled the inclusion of up to 15% of the mango waste meal in diets for tilapia.

To intensify production, it is necessary to introduce technology in fish management, especially utilizing balanced diets. Thus, it is essential that we conduct studies on nutrition that contribute to the production of species-specific diets. According to Kaushik (1989), the expenses with feeding in fish farming account for up to 70% of the total production costs, so the use of effective diet-processing techniques and application of appropriate feeding strategies are paramount to reducing the final cost (Kubitza,1998).

According to Andriguetto *et al*. (1985), the fact that protein is the most important macronutrient for the growth and the most expensive ingredient in diets leads scientific research to seek the lowest amount of this ingredient that the organism needs in order to reciprocate with better production, and because carbohydrate is the ingredient with the lowest cost in the formulation of diets (Silveira *et al*., 2010), it is necessary to know the ideal proportions of these components, in diet formulation, so as to maximize the profitability of intensive fish farming.

The species utilized in this study was pacamã (*Lophiosilurus alexandri*), a fish of the family Pseudopimelodidae (order Siluriformes), native to the São Francisco river basin. This fish is highly marketable because its meat is devoid of intramuscular spine, and its taste is highly appreciated by the consumers. It is a carnivore species and has sedentary behavior, with multiple spawning and preference for lentic environments in sandy or stony areas (Shibata, 2003).

Given the importance of determining the ratio between protein and carbohydrate in diets for fish, along with the marketability and performance potential of this species for intensive fish farming in the semi-arid region, the aim of this study was to determine the ideal proportion between protein and carbohydrate in the feeding of pacamã juveniles using mango waste meal as source of carbohydrate, evaluating animal performance and physicochemical carcass composition under different diets, thereby proposing the ideal protein level in the composition of their diet.

MATERIAL AND METHODS

The experiment was conducted in the facilities of the Laboratory of Aquiculture of Campus Ciências Agrárias of Universidade Federal do Vale do São Francisco, during 60 days. One hundred and fifty pacamã juveniles from Companhia de Desenvolvimento dos Vales do São Francisco e do Parnaíba - CODEVASF-PE were utilized. The fish were housed at a density of seven fish per tank, with initial live weight (ILW) of 11.31±0.96g. They were distributed into 16 circular polyethylene water tanks (500L) in a water recirculation system with mechanic

and biological filters. The animals went through an acclimation period of seven days, fed a diet formulated to have 40% crude protein and 3,000kcal gross energy (National…, 2011). Mango waste meal was used as the carbohydrate source at the proportion of 25%.

The water temperature, pH, dissolved oxygen and electrical conductivity were measured; the latter was measured with the Hanna Oxy-chek portable device. The experimental units were siphoned in the morning and afternoon, before feeding, with exchange of approximately 10% of the total water so as to remove feces and feed leftovers.

The experimental design was entirely random, with four treatments and four replications. The data were subjected to variance analysis, and the test of means used were Tukey's. The treatments consisted of four experimental diets with decreasing levels of the ratio between crude protein and carbohydrate (CP:CH - 1.40, 0.94, 0.56 and 0.29). The CP:CH ratios were calculated based on the proportion of nitrogen-free extracts (NFE) over the proportion of crude protein (CP).

Table 1. Chemical and calculated composition of the experimental diets

INGREDIENTS	CP:CH ratios			
	1.40	0.94	0.56	0.29
Fish meal	64.1	51.6	39.1	26.6
Soybean meal	10	7.5	5	2.5
Mango meal	15	30	45	60
Dicalcium phosphate	0.84	0.84	0.84	0.84
Soybean oil	8	8	8	8
Min. and vit. mix[1]	2	2	2	2
Vitamin C[2]	0.05	0.05	0.05	0.05
BHT[3]	0.01	0.01	0.01	0.01
Total (%)	100	100	100	100
Calculated values[4]				
CP (%)	40.16	34.40	25.63	16,24
NFE (%)	28.68	36.63	46.01	56,33
GE:CP (Kcal. g^{-1})	10.61	12.37	16.57	35.83
GE (Kcal/kg)	4263	4255	4248	5826
EE (%)	16.03	15.26	14.17	12.58
CF (%)	1.78	1.99	2.19	2.40
MM (%)	10.81	8.27	8.26	6.35
OM (%)	2.54	3.45	3.74	6.08
DM (%)	97.46	96.55	96.26	93.22

1. Min. and vit. mix (Supremais, Campinas/SP, Brazil): Composition per kg: vit. A = 1,200,000 IU; vit. D3 = 200,000 IU; vit. E = 12,000mg; vit. K3 = 2,400mg; vit. B1 = 4,800mg; vit. B2 = 4,800mg; vit. B6 = 4,000mg; vit. B12 = 4,800mg; folicacid = 1,200mg; calciumpantothenate = 12,000mg; biotin= 48mg; choline = 65,000mg; nicotinicacid = 24,000mg; Fe = 10,000 g; Cu = 600mg; Mn = 4,000mg; Zn = 6,000mg; I = 20mg; Co = 2mg; and Se = 20mg.
2. Vit. C (Basf, São Paulo/SP, Brazil): calcium salt, ascorbic acid-2-monophosphate, 42% active ingredient.
3. Butylatedhydroxytoluene.
4. CP = crude protein; NFE = nitrogen=free extract; GE:CP = gross energy:crude protein ratio; GE = gross energy; EE = ether extract; CF = crude fiber; MM = mineral matter; OM = organic matter; DM = dry matter; CP:CH = crude protein:carbohydrate ratio.

To produce the mango waste meal we utilized ripe mangos from the Tommy Atkins variety, originating from discarded fruits from Centro de Abastecimento e Logística da Bahia – CEASA-BA, located in the city of Juazeiro/BA, Brazil. The fruits were washed, peeled, cut in small pieces and had their seeds removed. Afterwards, they were dried in a forced-ventilation oven at 55ºC for 86 hours. Subsequently, they were ground in knife mills with 1mm sieves and stored in a freezer at –18ºC. All diet ingredients were ground in a knife mill with 0.5mm sieve, and then gathered until the mixture appeared homogeneous. To pelletize the diets, the mixture

of ingredients was moistened with water at 50ºC and then the diets were processed in a meat grinder and dried in a forced-ventilation oven at 65ºC for about 24 hours. The pellets were broken with the aid of a manual mill, and separated into different diameters in sieves of different pores to have appropriate sizes for the mouths of the animals according to their growth.

The fish were fed three times daily, at 08h00, 12h00 and 17h00. The amount of feed supplied daily was 10% of the body weight, which was corrected every 15 days, when biometry was performed. Four biometric analyses were performed, totaling 60 days of feeding. The productive performance of the pacamã juveniles in each experimental plot was analyzed by the final average weight gain (FAWG), specific growth rate (SGR), carcass yield (CY), total apparent feed intake (TAFI), apparent feed conversion (AFC) and survival (S).

At the end of the experimental period all animals were weighed to measure the growth data, and subsequently stunned with water and ice. The fish were sacrificed by medullar section, a procedure approved by the Ethics Committee on Human and Animal Studies (CEEHA – UNIVASF, protocol no. 0022/26091).

The analysis of the body composition of fish was performed by dehydrating the carcasses in a forced-ventilation oven (55 ºC) for 72 hours and grinding them according to the technique described by the AOAC (Official…, 1990), shown in Tab. 1. Moisture (M) was determined with the method based on the evaporation of the water present in the sample in a vacuum-oven at 70ºC and 660mmHg until constant weight. Crude protein (CP) was determined by the Kjeldahl method to determine the total nitrogen. This method is based on the nitrogen content of the organic matter, including the protein nitrogen itself and other non-protein nitrogen compounds such as amines, amino acids, among others. In this case, the result was expressed as crude or total protein, using the 6.25 factor for the calculation. Ether extract (EE) was determined by using chloroform and methanol as solvent. Mineral matter (MM) was determined by incinerating the dry matter in a muffle furnace at 550ºC, until constant weight. To determine the pH, the potentiometric method described in the Lanara manual (Brasil, 1981) was used. Initially, the pH was measured using a buffer solution ph 7 at 20 ºC. Next, the sample was homogenized with 10 mL distilled water, and then the electrode of the pH meter was introduced and reading was performed directly on the sample.

The data were subjected to variance analysis at 5% of significance. The statistical software Assistat 7.5 beta was used (Silva, 2010).

RESULTS AND DISCUSSION

Regarding water-quality parameters, the dissolved oxygen levels were 5.4±1.8 standard mg/L, electric conductivity mS 0.13±0.09; pH 7.77±1.8 and temperature of 26±3.5ºC. These results are considered acceptable for rearing fish (Boyd, 1990). The mean performance values are described in Tab. 2. All the analyzed variables, except for survival, showed significant differences ($P<0.05$).

Table 2. Mean values plus standard deviation of the performance variables and carcass characteristics of pacamã juveniles fed different CP:CH ratios

Performance	CP:CH ratios				
	1.40	0.94	0.56	0.29	CV (%)
IWG (g)*	12.4±2.98	10.45±1.93	12.11±2.08	10.27±2.24	20.72
FAWG (g)	46.5±0.88a	17.93±2.07b	16.32±1.34b	3.17±0.84c	18.01
SGR (%/day)	0.96±0.11a	0.46±0.12b	0.39±0.10b	0.01±0.04c	12.64
TAFI (g)	78.3±1.42a	47.01±0.94b	54.47±1.38b	46.19±1.33b	23.45
AFC	1.68±0.03c	2.62±0.05b	3.34±0.01b	14.57±0.02a	13.65
CY(%)	86.50±1.15ab	86.41±1.14ab	83.80±2.71b	89.58±2.51a	2.33
S (%)*	100±0.00	100±0.00	100±0.00	100±0.00	0.00

IAW = initial average weight; FAWG = final average weight gain; SGR = specific growth rate; TAFI= total apparent feed intake; AFC = apparent feed conversion; CY= carcass yield; S = survival.
*Mean values followed by the same letter in the row do not differ according to the Tukey test ($P>0.05$).
** Values on a natural-matter basis.

The peeled-mango waste meal without seed exhibited the following results for the chemical composition on a dry-matter basis: dry matter, 91.10%; crude protein, 4.74%; crude fiber, 4.99%; ether extract, 6.09%; mineral matter, 3.14%; and gross energy, 3724 kcal kg^{-1}. Evaluating the chemical composition of mango waste meal, but with the seed and skin, Vieira *et al.* (2008) found similar results to the meal evaluated in the present study for dry matter (92.2%), crude protein (3.9%), crude fiber (14.6%), ether extract (4.4%) and mineral matter (2.1%). However, they found different values for crude fiber, due to the inclusion of the mango seed and skin, which might have affected the nutritional values.

Lower final average weight gain was observed on the animals fed the CP:CH ratio of 0.29, in a diet containing 50% of mango meal as source of carbohydrate. Low concentrations of mango meal seem to be more efficient in the performance of the pacamã. Lima *et al.* (2011) included levels 0, 5, 10 and 15% mango waste meal in diets for trout and did not observe significant differences in weight gain. Melo *et al.* (2012) evaluated the inclusion of peeled-mango waste meal replacing corn and concluded that substitution of 100% of corn by mango does not impair the tilapia performance. According to Hilton *et al*. (1987), a tolerable level of carbohydrate in diets for fish is a level which does not compromise growth or results in increased mortality, whereas an optimal carbohydrate level in diets for fish is what enables glucose to be totally oxidized to produce energy, and thereby spare protein for the use of its energy in muscle growth.

For the total apparent feed intake (TAFI) of each treatment, intake was observed to increase along with the protein concentration of the diet, and consequently with the CP:CH ratio. There were no significant differences between the ratios of 0.94, 0.56 and 0.29, but they presented lower results than the 1.40 CP:CH. Aksnes (1995) verified that feed intake decreased as the carbohydrate level of the diet for Atlantic salmon was elevated, which corroborates the results of the present study, indicating that the energy from the carbohydrate more readily available in the diet might have influenced the satiety of carnivore fish, limiting their appetite. Sampaio *et al*. (2000) obtained results for feed intake that followed the same trend, and the higher TAFI followed the higher CP levels of the diet.

Regarding the apparent feed conversion (AFC) of the protein:carbohydrate ratios, it was observed that 1.40, as well as other parameters studied in this experiment, was the one to present the best result, whereas 0.56 and 0.29 provided the worst performances. Teixeira *et al.* (2009) found similar results to those of the present study, with the highest CP level (57.63%) and a CP:CH ratio of 2.13, achieving an AFC of 1.03, also verifying exponential improvement of this important animal-performance parameter as the studied ratio was increased. Sampaio *et al*. (2000) found at the highest CP:CH studied, 1.58, the best result (1.20) for AFC, and corroborating the present study, they verified that as this ratio was elevated, AFC improved. Although carbohydrate is largely used as a source of energy in diets for domestic animals, some fish seem to have lower ability to utilize it. Several possibilities have been discussed to explain the mechanism of fish intolerance to carbohydrates. It is also suggested that omnivore fish are capable of utilizing higher carbohydrate concentrations in the diet than the carnivores, due to the greater activity of amylase in the digestive tract (Hidalgo *et al*., 1999). Therefore, the ability to use different forms of carbohydrate varies according to the species.

Increasing values were found for SGR, as the CP:CH ratios are elevated, atthe ratio of 1.40, which caused an SGR of 0.96, whereas the ratio of 0.29 caused an SGR of 0.01, clearly demonstrating the significant differences between the treatments. Sánchez *et al.* (2009) studied the influences of the protein levels (28, 32 and 36%) on the growth of catfish (*Leiarius marmoratus*) fingerlings and did not find statistical difference between the treatments, unlike the results obtained herein. Teixeira *et al.* (2009) used diets with different protein levels for golden dorado (*Salminus brasiliensis*) fingerlings weighing 0.75g for 29 days and found similar results to those obtained in the present study, with the highest level of CP of 57.63% and a CP:CH ratio of 2.13, having an SGR of 2.8.

For carcass yield (CY), the CP:CH ratio of 1.40 showed similar statistical result to the ratio of 0.29, differing from the 0.56, which displayed the worst result, although this rate increased as

the ratio was elevated in absolute values. The best yield was obtained with the ratio containing the lowest value for the CP:CH ratios, which occurred due to the mobilization of the energy reserves from visceral fat, decreasing the proportion between viscera and carcass. Reidel (2007) observed non-significant differences with increase in CP for carcass yield, which disagrees with our results. Burkert *et al.* (2008) worked with the processing yield of *Pseudoplatystoma* sp. fillets, testing three types of diet with variation in CP and did not observe differences between the treatments.

Survival was not affected by the different CP:CH ratios (P>0.05). The elevated survival rates indicate that the diets containing mango meal as a carbohydrate source did not have a harmful effect on the health of fish. The survival rate in this study was 100%, corroborating the studies of Azaza *et al*. (2008), who observed significant difference in the survival rate of animals fed increasing levels of date meal in contrast to decreasing soybean levels in diets for tilapia.
These values show that the efficiency of mango waste meal as a carbohydrate source in diets depends on the species and its eating habits. In a study conducted by Aksnes (1995) with the carnivore Atlantic salmon, *Salmo salar* L., increasing levels of carbohydrates were tested in substitution of protein in the animal diets, utilizing the CP:CH ratios of 25.08, 5.31, 2.75 and 1.66. The author obtained highest growth with the ratio of 5.3, which was much higher than that used in our study with pacamã. Figueiredo (2011) evaluated different CP:CH ratios utilizing corn meal as a carbohydrate source for pacamã juveniles and verified better performance results, as well as other parameters studied in this experiment, for the highest CP:CH ratio of 1.24.

The carcass chemical composition values were changed (P<0.05), except for mineral matter (Tab. 3). Crude protein and ether extract were lower in the diet with a CP:CH ratio of 0.29. The literature has few reports of the types of carbohydrates and the chemical composition of fish meat; most of them are related to energy, protein and lipid in the diet.

Table 3. Mean values plus standard deviation of the carcass characteristics of pacamã juveniles fed different CP:CH ratios.

Physicochemical carcass composition**	CP:CH ratios				
	1.40	0.94	0.56	0.29	CV (%)
M* (g. 100g^{-1})	76.78±1.15	76.63±2.01	76.71±1.74	77.01±1.65	1.78
pH*	6.0±0.10	6.1±0.30	6.0±0.20	6.2±0.10	0.90
CP (%)	61.79±0.63a	59.94±0.58a	51.36±0.78a	47.96±0.57b	7.53
EE (%)	26.78±0.36a	23.56±0.39a	22.74±073a	17.87±0.86b	6.87
MM* (%)	12.13±0.04	15.74±0.07	14.87±0.63	17.10±1.74	9.74

M = moisture; pH – potential hydrogen; CP = crude protein; EE = ether extract; MM = mineral matter.
*Mean values followed by the same letter in the row do not differ according to the Tukey test (P>0.05).
** Values on a natural-matter basis.

The diet with a CP:CH ratio of 0.29 contained 16.24% CP, in which there was a deficit for this nutrient, according to the requirements for this species (National…, 2011), and this reduced the protein synthesis and ether extract in the carcass of the animals.

Signor *et al.* (2007) did not observe significant differences for crude protein, ether extract and mineral matter in the carcass when they used poultry offal meal in the feeding of *Leporinus macrocephalus*.

The concentrations of carbohydrates in the diet modify the chemical profile of pacamã meat. In trout meat they reduced the fat and energy and increased protein, and ash contents were elevated with higher carbohydrate (Veiverberg *et al*., 2010). Fabregat *et al*. (2011) evaluated substitution of fish meal by soybean meal in the diet and verified reduction in the crude protein contents of the carcass of *Prochilodus lineatus* juveniles.

The Brazilian legislation considers that fish whose external-meat pH is above or equal 6.8, or

whose internal-meat pHi s above or equal 6.5, are unsuitable for consumption (Brasil, 1997). The pH values remained within the Brazilian norms for commercialization and consumption; the different CP:CH ratios did not affect the pH of the pacamã meat.

The chemical composition of the carcass is reflected by the nutrients in the diet, and so their imbalance may affect it.

CONCLUSIONS

Increase in the CP:CH ratio improves performance of pacamã fish. The CP:CH ratio of 0.29 reduces the protein and lipid deposition in the carcass. Among the ratios studied in this experiment, the CP:CH ratio of 1.40 is recommended for the best performance of pacamã; however, higher ratios may be tested to verify their effectiveness. Pacamã has desirable animal performance characteristics for commercial cultivation.

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