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Sunflower protein concentrate and crambe protein concentrate in diets for silver catfish *Rhamdia quelen* (Quoy and Gaimard, 1824): use as sustainable ingredients

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ABSTRACT

The objective of this study was to evaluate growth and metabolic parameters of silver catfish fed with protein concentrates of sunflower meal (SMPC) and crambe meal (CrMPC). The study evaluated two levels of substitution, where 25 or 50% of animal protein was replaced with plant-based protein. A total of 300 silver catfish (14 ± 0.26 g) were used in five treatments and three replications, in fifteen 280-liter experimental units. The results were submitted to analysis of variance and the means of the control diet was compared to the remaining treatments by Dunnett's test at 5% significance level. At the end of the trial, no differences were observed for the variables final weight and daily weight gain. However, minor feed conversion was observed in the groups Control and SMPC-25%. Metabolic parameters were analyzed in the plasma and liver, where no significant differences were found for any of the blood parameters analyzed. In the analyzed liver parameters (ammonia, protein, amino acids and ALAT), the liver protein content was lower in fish consuming SMPC-50%, CrMPC-50% and 25% CrMPC diets. The amino acids content was higher in fish receiving the SMPC-25% diet. It can be concluded that sunflower meal protein concentrate is better utilized by fish and more efficient metabolically than crambe meal. This study demonstrated that a newly developed protein concentrate SMPC and CrMPC can effectively replace 25% and 50% the animal protein in a diet free of FM.

Key words: Crambe abyssinica, fish feeding, Helianthus annus, jundiá, liver metabolism, plant-based protein concentrates.

INTRODUCTION

Fish diet formulation aims to obtain highly digestible, nutritionally balanced, economically viable and low environmental impact products

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(Shiau 2002). This concern is mainly related to protein sources, which is the main ingredient in fish nutrition (Cabral et al. 2011). Normally, the main protein source used in fish nutrition is of animal origin, such as fish meal (Santigosa et al. 2011), due to its high nutritional value and balance in essential amino acids (Gatlin et al. 2007, Larsen et al. 2012).



However, increasing demand, high price and supply fluctuations, makes it a priority to find an alternative replacement to animal protein sources (Larsen et al. 2012). Soybean meal (SBM) is by far the most widely used vegetable protein source in fish diets, followed by sources from other oleaginous plants such as sunflower meal and crambe meal. Leguminous seeds are also good dietary protein sources, particularly if they are locally produced, contributing to the sustainability and cost-effectiveness of fish-farming (Gatlin et al. 2007, Kaushik and Hemre 2010).

The use of plant-based protein sources in fish feeds has expanded considerably in recent years to meet the demand for feeds and sustain the development of worldwide aquaculture production (Tacon and Metian 2015). This protein sources in aquaculture is still challenging, since information on the bioavailability of nutrients are controversial (Cabral et al. 2011) and restraints. The low palatability (Gatlin et al. 2007), imbalance in the amino acid profile (Santigosa et al. 2008) and intrinsic antinutritional factors (Gatlin et al. 2007, Mérida et al. 2010) often restrict its use.

Many studies have focused on the use of plantbased protein sources as sustainable replacements of animal protein sources in fish diets (Gatlin et al. 2007, Tacon and Metian 2008, Hardy 2010, Cabral et al. 2011, Lovatto et al. 2015). Among the potential protein sources, sunflower meal (Helianthus annuus) and crambe meal (Crambe abyssinica) are mentioned. Studies using sunflower meal in fish feed, in different proportions, showed that substitution levels are around 15% for different species (Olvera-Novoa et al. 2002, Lozano et al. 2007, Mérida et al. 2010) low content lysine and methionine, high fiber content and the presence of antinutritional factors being the limiting aspects. Studies on crambe meal are scarce and there are no definite guidelines for its use in fish feed. The main limiting factors are its high levels of erucic acid, the presence of glucosinolates (Fundação MS 2010) and antinutritional factors for fish.

The use of plant-based protein concentrates is promising as an alternative to the use of *in-natura* plant-based (Deng et al. 2006). Concentrates enable the obtainment of protein sources with low fiber content (Salze et al. 2010, Larsen et al. 2012), which are free of antinutritional factors and have a better amino acid profile, favoring the digestion (Salze et al. 2010).

The increase in omnivorous fish production is a trend, because they are better adapted to plant-based ingredients (Naylor et al. 2009). Silver catfish (*Rhamdia quelen*) is an omnivorous fish species, native to southern Brazil (Baldisserotto 2004), which accepts artificial diets with plant-based protein sources. Out of the possible protein sources, soybean meal is the one most commonly used (Coldebella and Radünz Neto 2002, Refstie et al. 2010). However, further studies should focus on the use of different plant-based protein sources, which are underutilized and considered by-products.

This study aims to evaluate the growth and metabolic parameters of silver catfish fed diets containing sunflower meal and crambe meal protein concentrates as substitutes for different levels (25 or 50%) of animal protein.

MATERIALS AND METHODS

OBTAINING THE PLANT-BASED PROTEIN CONCENTRATES AND INGREDIENTS

Pelleted sunflower meal with hulls (Giovelli Ltda, from Cerro Largo, RS) was milled and later sifted in a granulometer, with 600 µm mesh sieves, to remove excess fibers. The crambe cake (FMS Brilhante variety - MS Foundation for Agricultural Technology Research, Maracaju/MS) was defatted with hexane (2:1 v/w) to obtain the crambe meal, which was, in turn, used to obtain the protein concentrate. Protein concentrates from sunflower

and crambe meal were obtained in our laboratory using the methodology described by Smith et al. (1946), with modifications purpose of Lovatto et al. (2017).

Protein enrichment methods with the following changes: 1) The protein was dispersed in an aqueous medium by processing it three times in a blender (LIQ789, Cadence, Brazil) at maximum speed for 3 minutes at room temperature; the meal was blended in water at a ratio of 1:10 - each time. 2) The ground sample was sieved a 140 µm, and the remaining solid fraction (i.e., the fraction retained in the sieve) was discarded. The liquid fraction was then used for protein extraction. 3) Protein solubilization by isoelectric pH was carried out by increasing the pH of the liquid sample to 9.0 with 1 N NaOH. To precipitate the protein, the pH of the liquid was then reduced to 4.5 with 1 N HCl. The solution was conditioned under refrigeration (8°C) over night to promote decantation of the dispersed protein fraction, followed by discarding the supernatant and drying the concentrated protein fraction in an air recirculation oven at 50°C for approximately 24 hours (Lovatto et al. 2017).

The soybean protein concentrate (IMCOSOY 62[®]) was acquired of IMCOPA, Paraná, Brazil. The pork meat meal was acquired in Fasa Group, Cruzeiro do Sul, Rio Grande do Sul, Brazil. In southern Brazil, pork meat meal is a widely used product and used as a source of animal protein due to the large production of pigs, easily found in local trade, when compared to fish meal (FM).

EXPERIMENTAL DIETS

Four experimental diets and a control diet (Table I) were used. Two levels (25 and 50%) of partial substitution of animal protein, replaced by plant-based protein concentrates of sunflower meal (SMPC) and crambe meal (CrMPC) were evaluated. Five isoproteic and isoenergetic diets were formulated to the requirements of 37% crude

protein, in accordance with Meyer and Fracalossi (2004) with 3.200 kcal ME kg⁻¹ (Jobling 1983) and amino acids in accordance with Montes Girao and Fracalossi (2006). Pork meat meal (Fasa, Brazil) and soy protein concentrate (Imcosoy 62®, Brazil) were used as protein base in the diet control. The chemical composition and amino acid profile are described in Table I.

For diets preparation, the ingredients were weighed and mixed until complete homogenization of the diets. Subsequently water was added and the pellets of the high density and size of 4mm were pelletized and later dried in an oven with forced air circulation at 50°C for 24 hours.

GROWTH TRIAL AND SAMPLING

This study was carried out at the experimental fish farm and laboratory of the Federal University of Santa Maria (Universidade Federal de Santa Maria- UFSM) in southern Brazil. All procedures involving animals were carried out in compliance with the guidelines approved by the Committee on Research Ethics and Animal Welfare of said university, protocol number 23081.004071/2011-95. The fish were acquired from Fish Culture Station of the University of Passo Fundo, Rio Grande do Sul, Brazil.

A total of 300 silver catfish, with initial mean weight of 14 ± 0.26 g, were distributed into fifteen 280-litter polypropylene tanks (corresponding to 20 animals per tank), with individual water inlets and outlets, connected to a water recirculating system consisting of two biological filters with gravel, backwash system and controlled temperature. The fish were acclimated for a two week period, were fed to control feed and for seven weeks the fish were fed the experimental diets.

In the early and late experimental period (seven weeks of treatment), biometrics was performed to collect data. Animals fasted for 24 h, were anesthetized using Eugenol 20 mg.L⁻¹ in the water

TABLE I Formulation, chemical composition and calculated amino acids of the experimental diets containing protein concentrate of sunflower meal and crambe meal in different proportions in diet (g $\,\mathrm{kg}^{\text{-1}}$).

	Diets code 1						
Ingredients (g kg ⁻¹)	CONTROL	SMPC-50%	CrMPC- 50%	SMPC-25%	CrMPC-25%		
	Diet formulation (g kg ⁻¹)						
Corn	195	177.7	200	230	220		
Corn starch	24	67	15	30	16.5		
DRM^2	37	30	45	60	65		
IMCOSOY 62®3	260	260.9	257.7	250	250		
SMPC*4	-	176	-	88	-		
CrMPC*5	-	-	179.3	-	89.7		
PMM^6	300	149.9	149.9	224.9	224.9		
Soy oil	35	106.4	45	65	35		
Vitamins and minerals ⁷	30	30	30	30	30		
Dicalcium phosphate	36.4	-	4	-	2.5		
MSG ⁸	2.5	2	2.5	2	2.5		
BHT ⁹	0.1	0.1	0.1	0.1	0.1		
Limestone	22	-	18	15	20		
Inert ¹⁰	58	-	53.5	5	43.8		
L-lysine	-	-	0.8	-	-		
DL- methionine	3.2	2.5	2.3	1.7	5.1		
		Diets chemi	cal composition (g	kg ⁻¹)			
Dry matter	955.5	970.6	961.6	948.8	948.2		
Crude protein*	370.7	368.5	371.1	371.4	371.5		
Mineral matter	42.8	34.9	46.5	45.0	47.8		
Calcium	37.8	11.0	18.6	21.4	23.9		
Phosphorus	19.9	8.2	9.7	11.5	12.2		
Phosphorus NDF ¹¹	62.8	94.8	102.2	86.8	88.6		
Lipids	103.2	154.5	105.3	124.9	100.3		
Digestible energy ¹² (MJ kg ⁻¹)	13.41	13.40	13.40	13.41	13.40		
Soluble detergent carbohydrates ¹³	376.7	317.9	336.5	320.7	340.0		
	Calculated amino acids ¹⁴ (g kg ⁻¹ of the diets)						
Lysine	20.4	18.9	16.7	20.4	20.4		
Methionine+cystine	13.7	13.7	13.7	13.7	13.7		
Threonine	12.8	10.2	10.2	13.0	13.8		
Tryptophan	1.3	0.7	0.7	1.8	1.0		
Valine 1	16.3	12.8	13.0	16.9	17.3		
Isoleucine	12.7	10.2	10.3	14.6	11.4		
Leucine	23.7	18.8	19.0	26.5	24.5		
Phenylalanine+tyrosine	14.8	12.3	12.4	17.7	15.7		
Histidine	9.2	7.4	7.5	11.1	9.7		
Arginine	21.9	16.4	16.6	27.2	21.8		

^{*}Crude protein of protein concentrates: 51.36% (sunflower) and 50.42% (crambe);

⁷Vitamins and minerals (kg/product): folic acid: 299.88 mg, ascorbic acid: 15000.12 mg, pantothenic acid: 3000.10 mg, biotin: 0.06 mg, vitamin B3: 9000.32 mg, vitamin B4: 103.500 mg, vitamin A: 1000.000 UI, vitamin B1: 1500.38 mg, vitamin B2: 1500.00 mg, vitamin B6: 1500.38 mg, vitamin D3: 240000.00 UI, vitamin E: 10000.00 mg, vitamin K3: 400.00 mg, vitamin B8: 9999.92 mg, iron: 6416.80 mg, manganese: 8000.40 mg, copper: 1000.00 mg, zinc: 13999.50 mg, iodine: 45.36 mg, cobalt: 60.06 mg, selenium: 60.30 mg, magnesium: 5.10 mg, chlorine: 23000.00 mg, sulfur: 100 mg;

¹Diets: SMPC-50% and SMPC-25%: sunflower meal protein concentrate, replacing 50% or 25% of the pork meat and bone meal; CrMPC-50% and CrMPC-25%: crambe meal protein concentrate, replacing 50% or 25% of the protein meat and bone meal swine;

²Defatted rice mean:

³Soy protein concentrate 60% (CPS60[®]), Imcopa, Brazil;

⁴Sunflower meal protein concentrate;

⁵Crambe meal protein concentrate;

⁶Pork meat and bone meal;

⁸Glutamate monosodium;

⁹Butyl-hydroxy-toluene (antioxidant);

¹⁰ Sand;

¹¹Neutral detergent fiber (Van Soest et al. 1991);

 $^{^{12}}$ Digestible energy calculated according to Jobling (1983) = [(crude protein * 23.61 MJ / kg * 0.9) + (Fat * 39.82 MJ / kg * 0.85) + (Neutral detergent soluble Carbohydrate * 17.21 MJ / kg * 0.50);

¹³Soluble detergent carbohydrates = 100 – (Moisture + Crude protein + Mineal matter + Ethereal extract + Neutral detergent fiber);

¹⁴Amino acids analyzed the ingredients (Laboratory of Analysis Mycotoxicologic (LAMIC - CCR / UFSM, Brazil) and calculated in the diets.

(Cunha et al. 2010) and slaughtered by cervical puncture. The following data were collected: Final weight (g): final weight obtained at the end of the period; AFC: Apparent feed conversion = [(Total feed intake)/(final biomass - initial biomass)]; DWG: daily weight gain (g) = [(final weight - initial weight)/day]; survival (%) = [(Total number of fish harvested/total number of fish stocked) x 100] and hepatosomatic index (HSI) (%): [(weight of the liver/weight of the whole fish) x 100].

WATER QUALITY RECIRCULATION SYSTEM

During the experimental period, the physical/chemical variables of water quality were measured. During the trial, temperature was kept at 24.9 \pm 1.5°C, oxygen concentration 6.7 \pm 0.4 mg L⁻¹, total ammonia 0.15 \pm 0.06 mg L⁻¹, nitrite 0.16 \pm 0.1 mg L⁻¹, pH 7.3 \pm 0.2, Alkalinity 48.8 \pm 13.7 mg CaCO₃ L⁻¹ and hardness of 56.4 \pm 34.6 mg CaCO₃ L⁻¹. The levels of water quality remained ideal for temperate climate fish (Baldisserotto 2004).

PLASMA BIOCHEMISTRY AND HEPATIC METABOLISM ASSAY

In the late experimental period, after fasting for 24 h, nine fish were captured per treatment. Blood was quickly collected from the caudal vein using heparinized syringes and the fish slaughtered by spinal cord excision behind the operculum and eviscerated to remove the liver. Thereafter, livers were quickly placed on ice and frozen at -20°C for biochemical parameters analysis. Plasma aliquots were separated after blood centrifugation at room temperature for 10 min at 1200 x g for posterior determination of plasmatic metabolic parameters: glucose, total proteins, triglycerides, cholesterol and albumin, using commercial kits (Doles® Reagents and Laboratory Equipment Ltda. Goiania, Goiás, Brazil).

Liver glycogen levels were determined according to Bidinotto et al. (1997). The liver tissue was weighed (50 mg), and KOH and ethanol (1 and

3 mL, respectively) were added for hydrolysis and glycogen precipitation. For hepatic protein analysis, the tissues were heated at 100° C with KOH and centrifuged at 1000 x g for 10 min. Supernatant was used to estimate the total protein level according to the method described by Bradford (1976), using bovine albumin serum as standard.

To measure hepatic amino acids, ammonia and transaminases, liver samples were mechanically disrupted by adding trichloroacetic acid 10% and the homogenate was centrifuged at 1000 x g for 10 min. The neutral supernatant extract was used for amino acid colorimetric determination according to Spies (1957), using ninhydrin 1.5% in isopropyl alcohol as the color reagent. Hepatic ammonia was measured by colorimetry according to Verdouw et al. (1978).

This neutral extract was used to measure the hepatic transaminases concentration, but it was necessary to dilute the crude extract in homogenization buffer for the protein and alanine aminotransferase (ALAT) (EC 2.6.1.2). The enzymes were determined using colorimetric procedures following the protocols described in the kits (Doles®). ALAT concentration was expressed as UI. mg-1 hepatic tissue.

STATISTICAL ANALYSIS

The experimental design was completely randomized. The results were submitted to analysis of variance (one-way ANOVA). The means of the control diet were compared to the means of the other treatments by the Dunnett's test at the 5% level of significance, using SPSS 8.0 software.

RESULTS

GROWTH PARAMETERS, SURVIVAL AND HEPATOSOMATIC INDEX

No differences (P>0.05) were observed in the final weight and daily weight gain (DWG) (Table II)

TABLE II

Growth parameters	s, survival and hepa	tosomatic index of	silver catfish juvenil	es of different exper	imental groups fed		
		different expe	rimental diets.				
*7 * 11 1	Diets ²						
Variables ¹	CONTROL	CMIDC 500/	C.MDC 500/	CMDC 250/	C MDC 250/		

Variables ¹	Diets ²					
	CONTROL	SMPC-50%	CrMPC-50%	SMPC-25%	CrMPC-25%	
Final weight (g)	44.72±3.77	47.10±3.84	45.48±7.41	48.24±0.32	46.98±7.82	
AFC	1.13 ± 0.12	1.43±0.21*	1.57±0.14*	1.17 ± 0.08	1.42±0.05*	
DWG (g)	0.59 ± 0.07	0.64 ± 0.07	0.61 ± 0.05	0.66 ± 0.02	0.62 ± 0.07	
Survival (%)	100	100	100	100	100	
HSI (%)	1.43 ± 0.10	1.64 ± 0.20	1.56±0.20	1.50 ± 0.10	1.46 ± 0.20	

Values represented as mean \pm standard deviation. The means with (*) differ significantly from the control diet by Dunnett's test (P<0.05).

DWG: daily weight gain, n-60;

HSI: hepatosomatic index, n-9;

of fish fed diets containing 25 or 50% SMPC and CrMPC when compared to the fish that received the control diet. The best (P<0.05) feed conversion (FC) was observed in the Control and SMPC-25% treatments. Lower efficiency in FC was observed for fish receiving the SMPC-50%, CrMPC-50% and CrPC-25% diets (Table II). No difference (P>0.05) was observed for survival, in all treatments survival was 100 %.

The fish submitted to the experimental diets showed no difference (P>0.05) regarding the hepatosomatic index (HSI) (Table II).

PLASMA BIOCHEMISTRY ASSAY

Plasmatic levels of glucose, total proteins, albumin, cholesterol and triglycerides (Table III) did not differ (P>0.05) of fish fed diets containing 25 or 50% SMPC and CrMPC when compared to the control treatment.

HEPATIC METABOLISM ASSAY

There was no significant difference (P>0.05) in hepatic ammonia levels among tested diets (Table IV). Lower content (P<0.05) of hepatic protein was observed in fish from diets SMPC-50%,

CrMPC-50% and CrMPC-25% when compared to the control diet. The hepatic protein content of the fish fed the SMPC-25% diet did not differ (P>0.05) from the control diet (Table IV). There was an increase (P<0.05) in free amino acids content in the liver of fish fed the SMPC-25% diet (Table IV). There was no significant difference (P>0.05) in ALAT hepatic activity for fish fed diets containing the plant-based protein concentrates when compared to the control diet (Table IV).

DISCUSSION

Diets containing plant-based protein concentrates (SMPC- 25% and 50% and CrMPC- 25% and 50%) provided similar growth rates to those observed in the control diet. These results confirm the efficiency of the concentrates as protein sources in the diet of silver catfish (*R. quelen*). The FC of the fish submitted to the SMPC-25% treatment may be related to the better protein quality of the diets. Furthermore, the values found for FC are similar to those observed in other studies (Piedras et al. 2004, Freitas et al. 2011). Increased HSI is generally observed for fish fed diets containing high levels of carbohydrates (Debnath et al. 2007) in association

¹AFC: Apparent feed conversion, n-60;

²Diets: SMPC-50% and SMPC-25%: sunflower meal protein concentrate, replacing 50% or 25% of the protein pork meat and bone meal; CrMPC-50% and CrMPC-25%: crambe meal protein concentrate, replacing 50% or 25% of the protein pork meat and bone meal.

.....Diets¹..... Plasma levels CONTROL **SMPC-50%** CrMPC-50% **SMPC-25%** CrMPC-25% Glucose (mg.dL⁻¹) 50.12±6.40 49.40±7.90 52.67±8.00 56.10±11.50 47.26±9.60 Total proteins (g.L⁻¹) 4.13 ± 0.60 3.96 ± 0.30 4.03 ± 0.70 4.20 ± 0.50 4.34 ± 0.40 Albumin (g.L⁻¹) 0.82 ± 0.20 0.79 ± 0.20 0.74 ± 0.10 0.86 ± 0.10 0.67 ± 0.20 Cholesterol (mg.dL⁻¹) 130.71 ± 8.60 152.39 ± 26.10 122.11 ± 20.20 139.52 ± 24.40 119.72 ± 32.20 Triglycerides (mg.dL⁻¹) 703.19 ± 260.80 694.08±312.30 610.25±131.00 730.14 ± 149.60 580.63±184.40

TABLE III

Plasma biochemistry values for silver catfish of different experimental groups fed different experimental diets.

Values represented as mean \pm standard deviation, n-9.

¹Diets: SMPC-50% and SMPC-25%: sunflower meal protein concentrate, replacing 50% or 25% of the protein pork meat and bone meal; CrMPC-50% and CrMPC-25%: crambe meal protein concentrate, replacing 50% or 25% of the protein pork meat and bone meal.

TABLE IV

Hepatic biochemistry values for silver catfish of different experimental groups fed different experimental diets.

	1 0 1					
	Diets ¹					
	CONTROL	SMPC-50%	CrMPC-50%	SMPC-25%	CrMPC-25%	
Glycogen (%)	9.56±4.00	6.18±2.11*	4.42±1.47*	9.82±1.74	5.41±2.13*	
Protein (mg.g ⁻¹)	0.038 ± 0.006	0.031 ± 0.002	$0.025\pm0.002*$	0.032 ± 0.008	$0.029 \pm 0.001 *$	
Ammonia (mM.g ⁻¹)	12.09 ± 5.22	9.84 ± 2.57	11.02 ± 2.11	13.089 ± 3.75	12.89 ± 5.57	
Amino Acids (mg.g ⁻¹)	35.21 ± 12.31	45.36 ± 14.00	46.87 ± 10.45	87.73±23.29*	52.96 ± 14.25	
ALAT (UI.mg ⁻¹)	7.19 ± 2.70	3.96 ± 0.54	4.81 ± 1.06	4.44±0.42	5.57±1.84	

Values represented as mean \pm standard deviation, n-9. The means with (*) differ significantly from the control diet by Dunnett's test (P<0.05).

¹Diets: SMPC-50% and SMPC-25%: sunflower meal protein concentrate, replacing 50% or 25% of the protein pork meat and bone meal; CrMPC-50% and CrMPC-25%: crambe meal protein concentrate, replacing 50% or 25% of the protein pork meat and bone meal.

to the increase in hepatic glycogen content, which was not observed in our study.

Borges et al. (2004) observed blood reference parameters in silver catfish, which were similar to those found in the present study, and no differences were observed among the treatments tested. This demonstrates that plasmatic levels were not altered by the use of plant protein concentrates in the diet of silver catfish juveniles.

The maintenance of normal levels of total circulating proteins is indicative of protein catabolism, which means that the protein in the diet is being used and metabolized, because when blood protein levels are low, liver protein synthesis is impaired (Lieberman et al. 2007). In

addition, serum albumin serves as an indicator of dietary protein quality (Lehninger et al. 2004) and the results may suggest that plant-based protein concentrates have provided the necessary protein for fish development.

The fish that received the diets containing CrMPC-25% and 50% also had lower levels of hepatic protein, when compared to the fish on the control diet. This fact may have occurred due to the glycogen mobilization in fish being slow and the gluconeogenic pathway by synthesizing glucose through carbon skeletons (Halver and Hardy 2002) being preferred, since the fish fed those same diets (CrMPC-25% and CrMPC-50%) also had lower rates of hepatic glycogen. According to Bombardelli

et al. (2003), in a fasting state fish first mobilize the protein pool and pool of circulating amino acids before mobilizing hepatic glycogen, suggesting that the use of glycogen stock is performed when dietary proteins are not catabolized correctly.

In our study, there was no increase in hepatic ammonia. Lund et al. (2011) have shown that ammonia excretion is high in fish diets containing plant-based protein sources. This trend was not evidenced in our study, thus, it is suggested that the protein concentrates used in the diets presented adequate nutritional value. Larsen et al. (2012) found higher levels of ammonia for Oncorhynchus mykiss using plant-based protein sources, suggesting less efficient use of dietary proteins, unlike our results. Vieira et al. (2005) reported that the increase in free amino acid content in the liver is related to the higher synthesis of dietary protein. In fish, it is known that tissue amino acid levels are affected by both the amount and quality of the dietary protein (Yamamoto et al. 2000). The same authors have found a high correlation between the protein and amino acids contained in the diet, with those contained in the tissues (blood, liver and muscle).

Authors report that fish fed diets containing plant-based protein sources have lower ALAT activity in the liver, indicating that the protein transamination was not suppressed (Gómez-Requeni et al. 2004, Hansen et al. 2007).

The study of the metabolism within fish liver is crucial to elucidate and understand how well silver catfish diets adapt to different protein sources. Since the use of plant-based protein sources tends to increase linearly in feeding omnivorous fish (Naylor et al. 2009), it is likely that the dietary source will directly affect the endogenous protein/amino acid metabolism (Larsen et al. 2012).

In our study it was observed that among the protein concentrates obtained from different coproducts, the SMPC was more metabolically efficient and better utilized by the fish than the

CrMPC. The fish that received the SMPC-25% diet had the best metabolic efficiency of the ingredients. Further studies are needed to confirm the use of these proteins concentrates in the diet of silver catfish. From the production of protein concentrates on an industrial scale, up to carrying out a study with a longer experimental period, aiming at growth and fattening of the fish.

Is essential for the future development of global feed aquaculture that continued to develop new obtention feed products that are ethical, economical and sustainable. The FM based feeds become more expensive, due to increases, and then the protein demand will need to be met using largely plant-based alternatives and processing byproducts from other industries (Bell et al. 2016).

This study demonstrated that a newly developed protein concentrate SMPC and CrMPC can effectively replace 25% and 50% the animal protein in a diet free of FM. However, the results indicate that SMPC was more metabolically efficient and better utilized by the fish than CrMPC. Furthermore, data demonstrate that it is possible to use only 15% of animal protein, in diets whit SMPC for silver catfish, without prejudice to the growth and hepatic metabolism of fish.

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