Research Article

The inclusion of soybean oil in the diets of silver catfish (*Rhamdia quelen*) in relation to growth quality and fillet acceptability

Rafael Lazzari¹, Tatiana Emanuelli², Daniel Maschio³, Cristiano C. Ferreira⁴ Eduardo K. Battisti³ & João Radünz-Neto³ ¹Departamento de Zootecnia e Ciências Biológicas, Universidade Federal de Santa Maria (UFSM) Aven. Independência, 3751, Palmeira das Missões, RS, Brazil ²Departamento de Tecnologia e Ciências dos Alimentos, Universidade Federal de Santa Maria (UFSM) Av. Roraima, 1000, Santa Maria, RS, Brazil ³Departamento de Zootecnia, Laboratório de Piscicultura, Universidade Federal de Santa Maria (UFSM) Av. Roraima, 1000, Santa Maria, RS, Brazil

⁴Departamento de Zootecnia, Laboratório de Ictiologia, Universidade Federal de Pelotas (UFPEL), Pelotas, RS, Brazil

Corresponding author: Rafael Lazzari (rlazzari@ufsm.br)

ABSTRACT. The aim of this study was to evaluate the inclusion of soybean oil in the diet of female silver catfish (*Rhamdia quelen*) and its effect on the growth, composition, lipid profile and fillet acceptability thereof. In this experiment, 144 fish $(237.75 \pm 22.35 \text{ g})$ were distributed among 18 tanks (280 L) for 80 days. The use of 10% soybean oil in the fishes' diet promoted higher weights (373.82 g) and better feed conversion ratios (1.5) for female silver catfish. The soybean oil levels tested did not interfere with fillet acceptability. Increasing soybean oil in the diet increased the quantity of unsaturated fatty acids in the fillets, which enhanced their nutritional quality. The use of 10% soybean oil is recommended for the female silver catfish diet.

Keywords: Rhamdia quelen, silver catfish, diet, fatty acids, lipid profile, nutritional quality, aquaculture.

Inclusión de aceite de soya y su relación con la calidad del crecimiento y aceptabilidad de los filetes del bagre plateado (*Rhamdia quelen*)

RESUMEN. El objetivo de este estudio fue el de evaluar la inclusión de aceite de soya en dietas, sobre el crecimiento, la composición, el perfil lipídico y la aceptabilidad de los filetes de hembras de bagre plateado (*Rhamdia quelen*). En este experimento realizado durante 80 días, se utilizaron un total de 144 peces (237,75 \pm 22,35 g) distribuidos en 18 tanques (280 L). Se observó que el uso de 10% de aceite de soya en la dieta promueve mayor peso (373,82 g) y una mejor relación de conversión alimenticia (1,5) para las hembras de bagre plateado. Los niveles de aceite de soya de las muestras no interfirieron en la aceptabilidad del filete. El aumento de aceite de soya en la dieta aumentó la cantidad de ácidos grasos insaturados en el filete, mejorando así su calidad nutricional. Se recomienda el uso de 10% de aceite de soya en la dieta para las hembras de bagre plateado.

Palabras clave: Rhamdia quelen, dieta, ácidos grasos, perfil lipídico, calidad nutricional, acuicultura.

INTRODUCTION

Lipids are the main energy and fatty acid source in a fish's diet, and they affect growth and nitrogen excretion. A lipid-deficient diet results in protein catabolism for energy production. However, excess energy can suppress appetite as well as reduce growth and feed efficiency (Kim *et al.*, 2004; Craig *et al.*, 2006).

Currently, in aquaculture, due to environmental and economic considerations, alternatives to marine ingredients are increasingly used, and fish oil is commonly replaced by vegetable oils (Olsen, 2011). The demand for fish oil will likely exceed available resources over the next decade (Tacon, 2005). Due to high cost, limited availability and difficulty of maintenance, this source has been replaced by vegeta-

Corresponding editor: Jesús Ponce

ble sources. Increasingly substituting fish oil in aquafeed not only modifies the fatty acid composition in feed, but simultaneously and progressively reduces cholesterol levels (Norambuena *et al.*, 2013). Prime examples include soybean, sunflower, rice, canola and linseed oils. Among these oils, soybean oil is more available in Brazil because it is produced at high levels. The increase in whole-body fatness may reduce fish fillet quality, which is a process that can be influenced by dietary lipid digestibility depending on the lipid's fatty acid profile (Caballero *et al.*, 2002).

Several studies on *Rhamdia quelen* have investigated the protein requirements, and only lipid sources, such as vegetable oils, cod liver oil and lard, were tested (Losekann *et al.*, 2008). For *Rhamdia quelen* females, the level of digestible energy above 2.700 kcal kg⁻¹ with diets containing vegetable ingredients not affect reproductive performance (Bombardelli *et al.*, 2015). Due to limited information on silver catfish nutrition, the aim of this study was to evaluate the growth, composition, lipid profile and fillet acceptability of females in this species that were fed diets containing soybean oil.

MATERIALS AND METHODS

This experiment was conducted for 80 days in a recirculating water system with 18 tanks (280 L). The system included thermostats for temperature control, biofilters for biological filtration and a backwash system for waste disposal (Radünz-Neto *et al.*, 1987). A total of one hundred forty four adult females (237.75 \pm 22.35 g - 8 fish/tank) were used. The water flow rate in each tank was approximately 2.8 L min⁻¹. Soybean oil treatments in the fishes' diets were tested at 0, 2, 4, 6, 8 and 10% (six treatments in triplicate) (Table 1). The fish were fed daily (09:00 h) until apparent satiation. The daily feed, residue and feces remains were removed through siphoning, and 10% of the water volume was replaced.

For analysis, water was collected at the entrance of the first biological filter prior to feeding. The dissolved oxygen and temperature were measured using an oximeter (YSI-YSI Inc., Yellow Springs, OH, USA). The total ammonia levels, nitrites, pH, alkalinity and hardness were determined using a colorimetric kit (Alfakit[®]). The observed water quality did not affect the results.

At the end of the experiment, five fish per experimental unit were subjected to fasting (24 h), anesthetized and slaughtered by hypothermia (water x ice 1:1). Initially, six animals per treatment were eviscerated for weighing and to determine the carcass

yield. The fish fillets were removed to collect samples, determine the yield as well as lipid profile and obtain samples for an acceptability evaluation using a sensory panel.

At the beginning and end of the experiment, the weights (g) and lengths (cm) of the fish were measured after the fish were anesthetized with triphenoxyethanol (0.03%). For this procedure, a digital balance (precision 0.001 g) and ichthyometer were used. From these data, the following values were calculated: condition factor = body weight (g)/body length³ (cm); feed conversion ratio = feed supplied (g)/weight gain (g); biomass = mean weight x total fish remaining at the end of treatment; and daily intake.

The fillets' moisture, ash and protein (using the Kjeldahl method, conversion factor = 6.25) were determined following the methods described in AOAC (1995). The fat was extracted and quantified using the Bligh & Dyer (1959) method. To determine the lipid profile, fat samples were extracted and methylated in accordance with Hartman & Lago (1973) and analyzed using gas chromatography (Hewlett-Packard chromatograph model HP 6890 equipped with a flame ionization detector-FID) using a capillary column DB-23 (Agilent-60 m x 0.25 mm x 0.25 mM).

The preference-ordering test (ABNT-NBR 13170, 1994) was used to ascertain the sensory differences between the fillet samples. In this procedure, a sensorial evaluation was conducted using 23 untrained judges who received six fresh fillet samples coded with random numbers and were asked to rank the samples according to preference in descending order relative to the following attributes: appearance, flavor and texture. The fillet samples from the female silver catfish were roasted in an electric oven at 250°C for 30 min. The preferred sample received the lowest score in the evaluation. The results of each review were submitted to the Friedman test (ABNT-NBR 13170, 1994).

To test normality of data, the Shapiro-Wilk test was used. When the data showed a normal distribution, the means were compared with the control diet using Dunnett's test. The variables that did not show normality were analyzed using a non-parametric ANOVA (Kruskal-Wallis). SAS software version 8.02 was used to test the data. The means of the final weight, biomass and feed conversion ratio were subjected to an analysis of covariance using the feed intake as the covariate, and the Pdiff test was used to compare the adjusted means.

RESULTS

Temperature (22.4 \pm 1.97°C), dissolved oxygen (4.74 \pm 0.43 mg L⁻¹), total ammonia (0.42 \pm 0.11 mg L⁻¹), nitrite (0.13 \pm 0.04 mg L⁻¹), pH (6.8 \pm 0.56), total alkalinity

Table 1. Composition of different diets (D) used in the experiment (%).

Ingredients	Diets (% soybean oil)						
Ingreatents	D1	D2	D3	D4	D5	D6	
Meat and bone meal	16	16	16	16	16	16	
Soybean meal	22	22	22	22	22	22	
Wheat bran	22	22	22	22	22	22	
Corn (grain)	25.9	25.9	25.9	25.9	25.9	25.9	
Soybean oil	0	2	4	6	8	10	
Inert	10	8	6	4	2	0	
Vitamins and minerals ^a	2	2	2	2	2	2	
Bicalcium phosphate	1	1	1	1	1	1	
Sodium chloride	1	1	1	1	1	1	
Antioxidant (BHT)	0.01	0.01	0.01	0.01	0.01	0.01	
	Composition analyzed						
Moisture	5,22	5,09	5,13	5,13	5,24	5,07	
Crude Protein	26,28	26,31	26,99	25,36	26,23	25,52	
Digestible energy (kcal kg ⁻¹) ^b	2491	2635	2784	2845	2974	3046	
Energy (kcal DE) / protein (g)	9,47	10,01	10,31	11,21	11,33	11,93	
Minerals	11,10	10,11	10,00	9,91	9,56	9,84	
Ether extract	4,09	6,09	8,56	10,31	12,17	13,68	
Crude fiber	3,53	3,13	3,59	3,52	3,65	3,34	
Calcium	2,27	2,30	2,15	2,25	2,02	2,22	
Phosphorus	1,43	1,41	1,41	1,37	1,38	1,45	

^aComposition of vitamin and mineral kg⁻¹ product (SUPRE MAIS ®) mixture: folic acid: 1200 mg, Ác. nicotinic: 24000 mg, Ác. pantothenic: 12000 mg, cobalt: 10 mg, copper: 3000 mg, choline chloride: 108 g, iron: 50000 mg, biotin 48 mg, iodine: 100 mg, manganese: 20000 mg, selenium 100 mg, vit. A: 1200000UI, vit. B1: 4800 mg, vit. B2: 4800 mg, vit. B6: 4800 mg, vit. B12: 4800 mcg, vit. C: 48 g vit. D3: 200000UI, vit. E: 12000 mg vit. K3: 2400 mg, and zinc: 3000 mg. ^bDigestible energy calculated, considering the following: lipid = 9 kcal g⁻¹, protein = 5 kcal g⁻¹, and carbohydrates = 4 kcal g⁻¹ with the digestibility values 85, 90 and 50%, respectively.

 $(36.3 \pm 8.98 \text{ mg CaCO}_3 \text{ L}^{-1})$, and hardness $(88.5 \pm 36.1 \text{ mg CaCO}_3 \text{ L}^{-1})$ were determined.

The female silver catfish weight was influenced by adding soybean oil to the diet (Table 2). The best weight (373.82 g) was obtained by females fed diets containing 10% soybean oil as the lipid source (Table 2). The three lower levels tested (0, 2 and 4% soybean oil) resulted in lower fish weights (P < 0.05).

The silver catfish feed intake and feed conversion decreased with an increasing level of dietary soybean oil (8 and 10%, Table 2). The daily feed intake ranged from 3 to 5% of body weight in this study.

The total biomass increased with increasing levels of dietary soybean oil, but the length and condition factor did not exhibit a significant effect (Table 2). Survival was lower with increasing levels of oil (P < 0.01). The female carcass yield was greater with 6% soybean oil in the diet (88.25%).

The fillet moisture was greater in fish fed the diet containing 6% soybean oil (79.31%) (P = 0.04). The

fillet lipid (P = 0.02) and protein (P = 0.009) quantities varied among the treatments (Table 3).

Dietary levels of soybean oil modified the fatty acid composition of the fillets (Table 3). The quantity of MUFA (40.87%) was higher in the fillets of fish fed a diet without soybean oil (Table 3). Fish fed the diet containing 8% oil exhibited higher levels of PUFA in the fillets (30.71%, P < 0.0001).

Higher quantities of omega-3 UFA were observed in fillets from fish fed a diet with 4% soybean oil (P < 0.01), but the fillets of fish fed diet with 8% of oil exhibited more omega 6 UFA (P < 0.0001). Fillets from female fish fed a diet containing 4% soybean oil exhibited a higher value for the ratio n-3/n-6 (0.21) (Table 3).

A paired comparison using the sensory panel revealed no significant differences in appearance, flavor or texture between the fillet samples (Table 4). The samples did not reach the critical values 37 and 38; note that this critical value was established at the predetermined minimum significance (P < 0.05).

Table 2. Performance parameters of female silver catfish fed soybean oil in the diet. W: final weight; FCR: feed conversion ratio; DI: daily intake; L: length; CF: condition factor; SU: survival; CY: carcass yield; FY: fillet yield; * P < 0.05; ** P < 0.01; sem: standard error of mean; NS: not significant (P > 0.05). aVariables with adjusted means, where different letters indicate a significant difference using the Pdiff test. ^bMeans marked with [#] indicate a significant difference from the control diet using the Dunnett test.

Soybean oil level (%)								
	0	2	4	6	8	10	sem	Р
W (g) ^a	317.35 ^b	323.16 ^b	333.03 ^b	346.70 ^{ab}	340.39 ^{ab}	373.82ª	22.62	*
FCR	3.16 ^a	2.77 ^{ab}	2.45 ^b	1.97 ^b	2.37 ^b	1.50 ^c	0.62	**
DI (%) ^b	3.22	2.80	3.07	2.76	3.06	2.57#	0.21	**
Biomass (g)	2549.4°	2652.8 ^{bc}	2918.9 ^b	3155.7 ^{ab}	3017.0 ^b	3471.5 ^a	330.41	**
L (cm)	32.03	31.74	31.70	32.02	31.59	31.86	0.78	NS
CF	1.04	0.98	1.03	0.98	1.03	1.06	0.04	NS
SU (%)	100	97.22	91.67	88.89#	91.66	86.11#	8.04	*
CY (%)	81.16	88.20#	84.77	88.25#	86.11	83.06	2.92	*
FY (%)	30.70	32.06	31.41	31.56	30.64	29.76	2.35	NS

Table 3. Chemical composition (%) and lipid profile (% of fatty acids in the total lipids) of female silver catfish fillets fed diets containing soybean oil. aMeans marked with # indicate a significant difference from the control diet using Dunnett's test; means with different letters indicate significant differences using ANOVA Kruskal-Wallis; * P < 0.05; ** P < 0.01; *** P < 0.001; ser: standard error of mean; and NS: not significant (P > 0.05). aLinear effect: Y = 26.06 – 0.28X, r² = 0.60, bMonounsaturated fatty acids, cPolyunsaturated fatty acids and dUnsaturated fatty acids/saturated fatty acids, linear effect: Y = 1.69 + 0.03X, r² = 0.70.

	Soybean oil level (%)							
	0	2	4	6	8	10	sem	Р
Chemical composition								
Moisture (%)	75.43	76.38	77.25	79.31	76.18	74.65	1.99	*
Ash (%)	1.08	1.18	1.20	0.94	1.11	1.15	0.10	NS
Lipid (%)	6.63	5.10	4.55	4.44	4.73	6.19	1.24	*
Protein (%)	20.82	20.41	19.45	16.71	20.60	20.57	1.20	**
	Lipid profile							
C14:0	1.55	1.38#	1.48	1.42	1.25#	1.40	0.09	**
C16:0 ^a	26.31	25.00	24.81	25.26	23.11	23.41	0.82	***
C18:0	8.80	9.11	8.95	8.69	8.98	8.40#	0.36	*
C16:1n-7c	5.93 ^a	5.14 ^{ab}	4.10 ^b	5.29 ^a	3.74 ^b	5.08 ^{ab}	1.10	**
C18:1n-9c	33.94 ^a	33.56 ^a	32.17 ^b	33.34 ^a	31.41 ^b	33.73 ^a	1.13	**
C20:1n-9	0.99 ^a	0.99ª	0.98 ^a	1.02 ^a	0.71 ^b	0.87^{a}	0.21	**
\sum MUFA ^b	40.87	39.71	37.25#	39.67	35.86#	39.69	1.54	***
C18:2n-6c	17.46 ^c	18.84 ^{bc}	20.48 ^b	19.57 ^{bc}	24.81 ^a	22.13 ^b	2.07	***
C18:3n-3	1.15 ^d	1.29 ^d	1.46 ^c	1.30 ^d	2.05 ^a	1.73 ^b	0.14	***
C20:4n-6	1.28 ^{bc}	1.58^{ab}	1.74 ^a	1.39 ^b	1.31 ^b	1.21°	0.33	**
C22:5n-3	0.57 ^b	0.64^{ab}	0.79^{a}	0.59 ^b	0.62^{b}	0.60^{b}	0.20	**
C22:6n-3	1.57 ^{bc}	2.04^{ab}	2.51 ^a	1.89 ^b	1.91 ^b	1.39°	0.57	**
$\sum PUFA^{c}$	22.05	24.44	27.01#	24.75	30.71#	27.07#	1.66	***
$\overline{\Sigma}n-3$	3.30	4.01	4.78#	3.79	4.58#	3.73	0.52	**
$\overline{\Sigma}n-6$	18.75 ^b	20.42 ^b	22.23 ^{ab}	20.96 ^b	26.13 ^a	23.33 ^{ab}	5.08	***
<u>n-3/n-6</u>	0.17	0.19	0.21#	0.18	0.17	0.16	0.02	**
UFA/SFA ^d	1.70	1.79	1.79	1.81	1.99	2.01	0.07	***

Table 4. Sensory ranking test scores based on the preference of female silver catfish fillets fed different levels of soybean oil. The results refer to the analysis performed by 23 tasters. The critical value for 23 tasters and 6 samples is 38 with a 5% significance (ABNT - NBR 13170, 1994).

Soybean oil (%)	Appearance	Flavor	Texture
0	76	83	85
2	78	77	77
4	89	85	82
6	89	100	92
8	103	92	91
10	90	88	98

DISCUSSION

The quality of water was maintained within the proper limits for fish farming (Poli & Arana, 2003; Zaniboni, 2003). The protein-sparing effect is the use of as much available dietary protein as possible for conversion into muscle protein instead of energy production (Ljubojević et al., 2015) This effect is well-known for catfish, including silver catfish (Meyer & Fracalossi, 2004; Salhi et al., 2004.). Adding 12% soybean oil to of surubim catfish juveniles (Pseudoplatystoma *coruscans*) diets, results in excellent performance of the fish with enhanced body protein deposition (Martino et al., 2002). In this study, the best results for weight in female silver catfish (10% soybean oil) reinforces the notion that increasing dietary lipid level has a sparing effect on protein (protein sparring effect) and enhances the use of this nutrient for weight gain.

Feed energy affects feed intake and lipid deposition in the carcass (Médale *et al.*, 1995). For fish fed higher levels of lipid, the voluntary feed intake decreases (Brauge *et al.*, 1994; Skalli *et al.*, 2004), which was observed in the female silver catfish in this study. The daily feed intake values observed in this experiment are similar to the African catfish (Fagbenro & Davies, 2001). Studies using malabar groupers (*Epinephelus malabaricus*) (Tuan & Williams, 2007) and surubim juveniles (*Pseudoplatystoma corruscans*) (Martino *et al.*, 2002) reported less feed intake where the levels of dietary energy increased.

For most species of farmed fish, using vegetable oils (soybean, canola, rice, and linseed) does not affect fish growth; however, it modifies body composition and the fillet lipid profile (Regost *et al.*, 2003a, 2003b). In a study on Atlantic salmon (*Salmo salar*), including rapeseed oil at up to 50% of the supplemented lipid significantly decreases the (n-3)/(n-6) PUFA ratio as well as EPA and DHA concentrations in the fish (Bell

et al., 2001). Although raised in a closed system, the female silver catfish herein exhibited higher levels of UFA (60-65%) compared with fish from rivers (53%) (Ramos *et al.*, 2008). Using soybean oil in aquatic diets may be limited by the lack PUFA and high n-6 levels (Rombenso *et al.*, 2015).

The lipid profile (fatty acid composition in a fillet) is important for assessing the nutritional quality of the fish. In this context, not only are the composition and availability of the FA linolenic acid (18:3n-3) and linoleic (18:2n-6) important, but the elongation and desaturation products are also important (Steffens, 1997). The capacity and mechanisms of desaturation/ elongation of precursors to the FA linoleic and linolenic are unknown to silver catfish (Vargas *et al.*, 2008). In work on silver catfish fingerlings, these authors showed that this species exhibits desaturation and elongation capacity to generate unsaturated FA.

The (n-3)/(n-6) ratio of silver catfish is affected by several factors (Weber *et al.*, 2008). According to the World Health Organization (WHO, 2008), the appropriate value for the n-3/n-6 ratio in the total diet should be less than 0.25. Considering the results obtained in the fillets of female *Rhamdia quelen*, 0.16 to 0.21, ingesting this fish almost completes the daily need for human consumption.

The American Heart Association (2014) suggests that healthy people should consume at least two servings of fish per week. In addition to modulating the metabolism of lipoproteins, dietary supplementation with fish (rich in n-3 FA) can treat obesity and metabolic syndrome (Raposo, 2010). Further, the fatty acids eicosa-pentaenoic and docosahexaenoic (n-3) as well as arachidonic acid (n-6) prevent the decrease in LDL receptor protein expression caused by cholesterol in fibroblast culture cells and HepG2 (Yu-Poth *et al.*, 2005).

The sensory properties are decisive in determining consumer interest and market demand (Kubota & Emanuelli, 2004). Training judges is critical for obtaining sensory profiles in a practical industry context (Labbe *et al.*, 2004). For the feed industry and its stringent time restrictions, the performance evaluation from a panel of judges is a difficult quality tool to apply (Silva *et al.*, 2012). Consumer acceptability of and preference for meat from a species is based on four criteria: smell, texture, color and flavor (Alasalvar *et al.*, 2001). These factors can be altered by nutritional factors and explain consumer satisfaction for a particular product.

Often the taste of wild fish (derived from natural rivers and lakes) is more accepted by consumers. This difference is related to feed composition in these environments, where phytoplankton abundance, which is a rich source of polyunsaturated fatty acids (PUFAs), provides a more palatable characteristic fat (Steffens, 1997). Adding vegetable oils, such as soybean, does not affect the sensory characteristics of "Seabass" fillets (Izquierdo *et al.*, 2003). However, levels of 11% soybean oil may decrease the sensory evaluation scores (Guillou *et al.*, 1995).

The present work demonstrates that silver catfish females fed diets with soybean oil can be associated with good nutritional quality and consumer acceptance.

Finally, female silver catfish, at least 10% soybean oil in the diet is recommended. Increasing the soybean oil in the female silver catfish diet increases the quantity of PUFA in the fillets.

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Received: 24 October 2014; Accepted: 7 September 2015