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Communication

[*Comunicação*]

Feed cost reduction with total replacement of fish meal by soybean meal for Nile tilapia reared in biofloc system

[*Redução de custos dietéticos com substituição total da farinha de peixe por farelo de soja para tilápias-do-nilo criadas em sistema de bioflocos*]

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Feed is one of the most significant costs in fish farming. The feed can demand up to 60% of the production cost, in the case of Nile tilapia (*Oreochromis niloticus*) reared in an earthen pond. (Trombeta *et al*., 2017). In addition, fishmeal has been commonly used as the main source of protein in aquatic diets due to its desirable nutritional characteristics. However, its high demand and fluctuation in availability stimulated research aiming at the quest for an ingredient (or a combination of more than one) to supply the protein demand for the feed of aquatic organisms (Food outlook, 2019; Hua *et al*., 2019). Thus, soy and its derivatives are one of the replacements for aquatic feeds as the primary source of protein. This can be justified by the high availability of soybeans, adequate nutritional profile that makes it possible to use as a source of protein in fish feed, and its by-products can be successfully used, such as soybean meal (Hua *et al*., 2019; Montoya-Camacho *et al*., 2019).

Another possibility to reduce feeding costs would be the implementation of the biofloc technology (BFT) system, in which the nitrogen in the medium is used by microorganisms, allowing their growth, forming the bioflocs. These can be consumed by the cultivated species, being a microbial protein source, contributing to the good performance of the animals acting as a food supplement (Avnimelech 2015; Ebeling *et al*., 2006).

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In this perspective, the use of diets with soybean meal as the main source of protein in BFT is interesting, but further investigation is needed to relate both replacement/reduction in the use of fishmeal in BFT. Thus, the main goal of this work was to evaluate the growth performance and feed cost per kilogram of Nile tilapia reared in BFT, fed a commercial diet containing total replacement of fish meal (FM) by soybean meal (SM).

The work was carried out at the Aquaculture Laboratory of the Catarinense Federal Institute of Education, Science and Technology, Campus Araquari. All procedures performed in this study were approved by the Ethics Committee on the Use of Animals under the protocol number "263/2018".

The feeds were produced by NUTRICOL®, located at São Ludgero, Santa Catarina, Brazil. To prepare the feeds, some of the dry ingredients (corn, soybean, and wheat bran) were previously ground (2000 µm) in order to obtain particles with a diameter of less than 0.42 mm. Later, they were mixed with the other macro and micro ingredients and homogenized in a horizontal paddle mixer for 4 minutes. The resulting mixture went through a second grinding step (800 µm), followed by extrusion at 105 ° C in an extruder (FERRAZ®, Ribeirão Preto, SP, Brazil) with a capacity of 3,000 $kg.h^{-1}$ to obtain extrudates of 3 mm in diameter. The diet composition (Tab. 1) used in this work was the same as tested by Stockhausen *et al.* (2022).

Fish were maintained under constant aeration at a temperature of approximately 27 °C. On the day juveniles were transferred, the water in the nursery tank measured 27.4 °C temperature, 6.1 mg L^{-1} dissolved oxygen (DO), 558 mg L^{-1} total suspended solids (TSS), 101.2 mg L^{-1} alkalinity, 0.27 mg L^{-1} ammonia, 4.2 mg L^{-1} nitrite, and 7.01 pH. Biofloc was formed in the tank after 30 days. Biofloc was at heterotrophic stage; thus, sugar was added at a 50% ratio of the feed amount.

Table 1. Composition of control and experimental diet without fish meal (FM)

Ingredients (%)	Control	Without FM
Soy meal	35.00	46.50
Fishmeal (FM)	13.00	0.00
Corn meal	14.00	12.00
Ground beans	9.38	12.00
Wheat flour	9.00	4.12
Meat and bone flour	13.50	13.50
Blood meal	4.00	6.46
Fish oil	0.00	0.86
Soy oil	1.00	1.13
NaCl	0.30	0.30
Calcitic Limestone	0.00	2.21
Premix[1]	0.40	0.40
DL-Methionine	0.09	0.09
Essential (Functional) Oil[2]	0.10	0.10
Antifungal[3]	0.10	0.10
Adsorber[4]	0.10	0.10
Antioxidant[5]	0.03	0.03

[1]Premix = Vitamin A (min.) 800,000 IU; Vitamin D3 (min.) 410,000 IU; Vitamin E (min.) 15,000 IU; Vitamin K3 (min.) 505mg; Vitamin B1 (min.) 1,395.9mg; Vitamin B2 (min.) 2,000mg; Vitamin B6 (min.) 1,862mg; Vitamin B12 (min.) 2,500mg; Vitamin C (min.) 125g; Niacin (min.) 3,781mg; Pantothenic acid (min.) 4,018mg; Folic Acid (min.) 198mg; Biotin (min.) 100mg; Choline (min.) 86.68g; Copper (min.) 750 mg; Iron (min.) 8,310 mg; Manganese (min.) 1,320mg; Cobalt (min.) 24 mg; Iodine (min.) 264.8mg; Zinc (min.) 15.05g; Selenium (min.) 47.55mg; Inositol (min.) 25 g. [2]Castor oil and Canola oil. [3]Propionic acid. [4]Bentonite, Sepiolite, Calcium Propionate, Sodium Chloride, and other ingredients. [5]B.H.T. (Butylhydroxytoluene), Propyl gallate. (Stockhausen *et al.*, 2022).

Five days before acclimating, "mature" water from the nursery tank was used, and the experimental units were supplied with 125 L of water from the tank and 125 L of sterile water. Throughout the BFT experiment, carbon:nitrogen (C:N) ratio was kept at 10:1 (Avnimelech, 2015; Ebeling *et al*., 2006), resulting in an initial solids concentration of 229.0mg ± 14.5 mg.L^{-1}. Seven days after fish stocking, fertilization was maintained at 10:1 (C:N) to neutralize 40% of the feed nitrogen and to keep the level of ammonia below 1.0 mg L^{-1}. Calcium hydroxide was added when alkalinity went below 80 mg.L^{-1} $CaCO^3$, and if necessary a dose of 10% of the daily feed was added.

The experimental units consisted of two treatments: control, consisting of a commercial diet formulated with FM and regular ingredients, and a commercial diet, consisting of total replacement of FM by soybean meal, the tank position was randomized and also conducted in triplicate. Each tank was stocked with 25 Nile tilapias (average weight 18.3 g), maintaining an initial density of 100 fish m^{-3} and initial biomass of 457.5g m^{-3}. Rearing time was five weeks.

The animals were fed 3 times a day (09:00; 11:00 and 15:30 h) with an offer of 3 to 6% of the total biomass. Water quality parameters during the entire experimental period were: temperature of 28.10 ± 0.36 °C, dissolved oxygen of 5.80 ± 0.26 mg L^{-1}, pH of 7.01 ± 0.11, ammonia of 2.36 ± 1.85 NH_3 mg L^{-1}, nitrite of 1.70 ± 1.17 mg L^{-1}, nitrate of 0.63 ± 0.81 mg L^{-1}, alkalinity 147.75 ± 50.81 mg L^{-1}), turbidity of 17.51 ± 0.29 cm, total suspended solids of 562.12 ± 98.33 and Floc volume of 14.02 ± 2.01 ml. Temperature and dissolved oxygen (YSI PRO20 Oximeter) were monitored twice daily, the amount of floc (Inhoff cylinder) was measured daily and TSS (Stantard…, 1995) were checked twice a week. While the other parameters were measured weekly.

Average final weight, daily weight gain, specific growth rate, feed conversion rate, protein efficiency rate, survivorship, yield and feed cost per kg of fish (considering only the feed cost, represented by the sum of all the ingredients, according to the equation below) were all determined at the end of the experiments.

$$\text{Cost per kilogram of fish (R\$/kg)} = \frac{\text{(diet offered x price per kg)}}{\text{produced biomass}}$$

All data were first subjected to Shapiro-Wilk normality test and then Bartlett's analysis to verify the homogeneity of variance. Subsequently, they were submitted to the *t* test (Stata®□ Statistical Software). All analyses were conducted with a 5% significance level.

The feed cost per kg of fish was higher in the control than in the treatment without fishmeal (Table 2). While there was no statistical difference between treatments for the variables of final average weight, average daily weight gain, specific growth rate, feed conversion rate, protein efficiency rate, yield and survivorship.

Table 2. Growth performance of Nile tilapia reared in BFT, fed with a commercial diet with total replacement of fishmeal (FM) by soy meal

	BFT	
Variables	Control	Without FM
Mean final weight (g)	67.36±3.01	67.76±4.67
Daily weight gain (g.day^{-1})	0.88±0.05	0.89±0.07
Specific growth rate (%.day^{-1})	1.20±0.03	1.20±0.04
Feed Conversion rate	1.40±0.05	1.43±0.04
Protein efficiency rate	0.32±0.02	0.32±0.02
Survivorship (%)	100.00±0.00	99.33±0.89
Yield (Kg.m^{-3})	13.30±0.72	13.55±0.93
Feed cost per kg of fish (R$.kg^{-1})	1.57±0.04*	1.48±0.04

*Statistical difference by *t*-test (P<0.05).

Reducing the cost of the feed by replacing and/or reducing the use of fishmeal, without compromising performance, is a current issue. The result of this experiment corroborated with Qbal *et al.* (2021), in which the experimental diet (without FM) also did not affect the growth performance of tilapia (*O. niloticus*) in clean water, in addition to other variables such as nutrient utilization and carcass composition. However, these authors used an experimental diet that consisted of the association of FM with two other plant-based protein sources (soy meal + canola meal).

In another study, up to 50% of fishmeal in the diet was replaced by soybean meal, without significant performance impairment. However, this experiment was carried out with Nile tilapia fingerlings, with an initial average weight of 2.09g (Obirikorang *et al*., 2020), in contrast to this work, in which juveniles with an initial average weight of 18.3 g were used. Normally younger fish are nutritionally more demanding than juveniles, probably their metabolism does not produce all the digestive enzymes to break down some protein sources, especially those of plant origin, justifying the difference in results.

In addition, both studies mentioned above were carried out in clear water, unlike this study performed in BFT. Therefore, the possibility of total replacement of fishmeal may be because it is combined with the adequate nutritional balance of the diet with the rearing system used. Because the BFT offers a complement to the diet by the presence of flakes, being consumed by the tilapia and able to supply possible deficiencies and differences between the offered diets (Avnimelech, 2015). Making it possible for this system to achieve higher yields than conventional production systems for Nile tilapia (Jatobá *et al.*, 2019).

Despite the limitation of studies about FM replacement for tilapia in BFT, Mansour and Esteban (2017), tested two diets with different protein contents (20 and 30%), but both were formulated without FM and with plant-based protein sources, including soybean meal. The diets were evaluated in clear water and BFT. The fish reared in BFT showed better results for growth performance and immunological status, even being fed a diet without FM. These authors in addition to allowing the use of protein levels at 20%, evidencing the system's ability to contribute to the nutrition of the animal through the flakes, being a source of protein and always available food. These authors showed that BFT can contribute to the diet in biofloc flakes, being a source of crude protein and natural food, allowing the use of protein levels in 20%.

The feed cost per kg of fish produced was higher in the diet containing fishmeal. This result corroborates with Qbal *et al*. (2021) that when calculating an economic analysis, taking into account the price per kilogram of the three protein sources (FM, soybean meal and canola meal) and the feed conversion rate according to each treatment, it was possible to find the minimum cost result utilizing the three sources with smallest proportion of FM. Obirikorang *et al*., (2020) reduced approximately 43% of feed costs even without significant difference in some growth performance variables (feed conversion rate and protein efficiency). In this experiment, feed cost reduction was 10.6% per kilogram of fish, below the one presented by Obirikorang *et al.* (2020), but it is attractive to producers especially because it does not compromise growth performance.

For a native species of Mexico, *Dormitator latifrons*, the increase in the percentage of soybean meal in the diet reduced the feed cost and making it possible to completely replace fishmeal, providing adequate performance combined with cost reduction in a recirculation system (Badillo-Zapata *et al*., 2021). This data corroborate our study, as we also found a reduction in feed costs without interfering with fish growth performance. Thus, this cost reduction (due to the substitution of ingredients) can provide a better profit margin to farmers, making the activity more profitable.

In addition to the effects related to the growth performance of the species and feed cost, the replacement of fishmeal for soybean is also related to other variables. As for example, in an experiment with Nile tilapia, in a recirculation system, the total replacement of FM improved the growth performance and nitrogen retention, in addition, their health status was not affected even after an experimental infection with *Aeromonas hydrophila* (Stockhausen *et al.*, 2022).

This work and the others cited demonstrate the applicability of using soy meal as a substitute for fish meal in different culture systems, maintaining growth performance and animal health, in addition to reducing the feed cost. Considering the results obtained in the present study, it is possible to reduce feed costs per kilogram of fish produced by replacing fishmeal with soybean meal in diets offered to juvenile Nile tilapia (*O. niloticus*) reared in BFT, without compromising the growth performance of the animals.

Keywords: plant-based protein, feed cost, biofloc, growth, nutrition

RESUMO

O objetivo deste estudo foi avaliar o desempenho zootécnico e o custo dietético por quilograma de tilápias-do-nilo (Oreochromis niloticus) *cultivadas em sistema de bioflocos, sendo alimentadas com uma dieta prática contendo total substituição da farinha de peixe (FP) por farelo de soja (FS). Foram usados*

150 juvenis de tilápia-do-nilo, distribuídos em seis tanques com 250L úteis. Os tratamentos consistiram em: dieta controle, sendo composta por uma dieta comercial formulada com FP, e outra dieta teste, com substituição total do FP pelo farelo de soja. Os parâmetros zootécnicos e o custo dietético por quilograma de peixe produzido foram mensurados. O desempenho zootécnico dos peixes não foi afetado significativamente pela substituição, enquanto o custo por kg de peixe foi maior no tratamento controle (com FP). É possível reduzir custos dietéticos substituindo a FP pelo farelo de soja sem afetar os parâmetros zootécnicos para tilápia-do-nilo cultivada em sistema de bioflocos.

Palavras-chave: proteína vegetal, custo de alimentação, bioflocos, crescimento, nutrição

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