BROWN SEAWEED MEAL TO NILE TILAPIA FINGERLINGS

ALGA MARINHA MARROM PARA ALEVINOS DE TILÁPIA DO NILO

Costa, M.M.[1A]; Oliveira, S.T.L.[1B]; Balen, R.E.[2]*; Bueno Junior, G.[3A]; Baldan, L.T.[2A]; Silva, L.C.R.[3B] and Santos, L.D.[3C]

[1]Laboratório de Microbiologia e Imunologia Animal. Universidade Federal do Vale do São Francisco, UNIVASF. Petrolina, PE. Brazil. [A]mateus.costa@univasf.edu.br; [B]mmatiuzzi@univasf.edu.br
[2]Zoologia. Universidade Federal do Paraná, UFPR. Centro Politécnico. Setor de Ciências Biológicas. Curitiba, PR. Brazil. *rebalen@yahoo.com.br; [A]baldanlt@ufpr.br
[3]Aquicultura e Desenvolvimento Sustentável. Universidade Federal do Paraná, UFPR. Palotina, PR. Brazil. [A]gilsonjbueno@yahoo.com.br; [B]lcrsilva@ufpr.br; [C]liliansantos@ufpr.br

ADDITIONAL KEYWORDS

Ascophyllum nodosum. Feed additive. Fish nutrition. *Oreochromis niloticus*.

PALAVRAS CHAVE ADICIONAIS

Ascophyllum nodosum. Aditivo alimentar. Nutrição de peixes. *Oreochromis niloticus*.

SUMMARY

This work evaluated the effects of increasing levels of the brown seaweed *Ascophyllum nodosum* meal (BSM) in the feed given to Nile tilapia *Oreochromis niloticus* fingerlings. The completely randomized design experiment lasted 42 days. Nile tilapia fingerlings (n=75) with an average age of 30 days and average weight of 0.43 ± 0.02 g were assigned to 25 plastic tanks and submitted to five treatments in five repetitions. The treatments were increasing levels of BSM (5, 10, 15, and 20 g kg^{-1} of feed) and a control feed (no BSM). Each experimental unit was made up of a 36 L plastic tank with three fingerlings. The parameters studied were final average weight, percentage of weight gain, apparent feed conversion ratio, total length and carcass yield with and without head. The final average weight, percentage of weight gain, and total length were not affected by the treatments (p>0.05). The apparent feed conversion ratio improved linearly (p<0.01) and carcass yield increased linearly (p<0.01) as the BSM in feeds increased. BSM is an additive with potential use in Nile tilapia feed at the level of 20 g kg^{-1} because it does not result in growth loss and improves the feed conversion ratio and carcass yield in Nile tilapia during the fingerling period.

RESUMO

Este trabalho avaliou os efeitos de níveis crescentes da farinha de alga marrom *Ascophyllum nodosum* (FAM) nas rações para alevinos de tilápia do Nilo *Oreochromis niloticus*. O experimento, com delineamento inteiramente casualizado, durou 42 dias. Foram utilizados alevinos de tilápia do Nilo (n= 75) com idade média de 30 dias e peso médio de 0,43 ± 0,02 g, distribuídos em 25 tanques plásticos submetidos à cinco tratamentos e cinco repetições. Os tratamentos foram níveis crescentes de farinha de alga marrom (5, 10, 15 e 20 g kg^{-1} de ração) e uma ração controle (sem FAM). Cada unidade experimental foi composta por um tanque plástico de 36 L com três alevinos. Foram estudados os parâmetros médios de peso final, percentagem de ganho de peso, taxa de conversão alimentar aparente, comprimento total e rendimento de carcaça, com e sem cabeça. As médias de peso final, percentagem de ganho de peso e comprimento total não foram influenciadas pelos tratamentos (p>0,05). Os valores da taxa de conversão alimentar aparente melhoraram linearmente (p<0,01) e os valores de rendimento de carcaça aumentaram linearmente (p<0,01) à medida que se aumentaram os níveis de FAM nas rações. A farinha de alga marrom é um aditivo com potencial utilização em rações de tilápia do Nilo no nível de 20 g kg^{-1}, pois não diminui o crescimento e promove melhora da taxa de conversão alimentar e do rendimento de carcaça de alevinos de tilápia do Nilo.

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INTRODUCTION

Brazil's large territory, extensive hydrologic basin, and weather make it suitable for raising tropical fish species, such as Nile tilapia, *Oreochromis niloticus*, while producing large amounts of feed raw materials. The Brazilian semi-arid northeast is one of the regions with excellent conditions for breeding tilapia, despite its low rainfall. As such, fish culture is a potential economic activity in that area.

The presence of some rivers, particularly the São Francisco River and its dams, irrigation canals and lakes, affords suitable conditions for setting up fish production systems. The proximity to sites of production of raw materials for the manufacture of feeds (west of Bahia State) is a positive factor for the development of fish culture in the region (Meurer *et al*., 2010).

From the social viewpoint, fish culture may become an important income source for the local population, particularly for riverside populations such as small-scale fishers, who can hold regular jobs in both large firms and work in small production associations or cooperatives. The culture of species such as the Nile tilapia is an important alternative in the region, as the culture technology for this has been fully mastered (Meurer *et al*., 2009).

Brown seaweed (*Ascophyllum nodosum*) is commonly found on the North Atlantic shore (Branden *et al*., 2007). It is commercially used in the manufacture of agricultural and animal rearing products (Fleurence, 1999). According to Nakagawa *et al*. (1997), *A. nodosum* may also be a good additive to fish feeds.

Brown seaweed meal was obtained as a very fine powder with a light cream color. According to the manufacturer, it is composed of 87 % dry matter, 6 % crude protein, 6 % crude fiber, 22 % mineral matter, 3 % ether extract, and 50 % carbohydrates; however, composition may vary as a function of fresh seaweed composition (Sharp, 1986).

The dietary addition of *A. nodosum* to feeds results in the improvement of animal performance, immunity and meat quality; however, most reports of this have been in terrestrial animals (Allen *et al*., 2001; Turner *et al*., 2002; Leupp *et al*., 2005; Archer *et al*., 2007; Branden *et al*., 2007; Kannan *et al*., 2007a; Kannan *et al*., 2007b; Gardiner *et al*., 2008). Despite the evidence regarding the use of *A. nodosum* in terrestrial animals, its use in fish has been little studied.

The Nile tilapia is an important species for fish culture in the world and the most commonly cultured fresh water specie in Brazil (El-Sayed, 2006). Due to the importance of this fish species and the benefits that *A. nodosum* may confer upon Nile tilapia culture, the present study evaluated the effects of brown seaweed meal inclusion in Nile tilapia fingerling feed on the performance, survival rate and carcass yield.

MATERIAL AND METHODS

The present study was carried out in the Aquaculture Laboratory of the Zootechnics Collegiate of Universidade Federal do Vale do São Francisco (UNIVASF), Agrarian Science Campus at Petrolina, PE, Brazil to test the use of increasing levels of brown seaweed meal (BSM) *A. nodosum* in feeds for sexually reversed Nile tilapia fingerlings of the chiltralada breed.

A lot of 300 sexually reversed Nile tilapia fingerlings in the initial phase of development and aged 30 days, from the Bebedouro Fish Farm run by the 3rd Regional Superintendence of CODEVASF at Petrolina, PE, were placed in a 1000 L tank with constant aeration and water circulation. The fishes received feed with 30 % of digestible protein (DP) and 3000 kcal kg^{-1} of digestible energy (DE) without BSM for one week.

It were took 75 fingerlings with an average weight of 0.43 ± 0.02 g from the initial lot and assigned them to 25 plastic tanks with 36 L water in a completely random design with five treatments and five repetitions. Each experimental unit was made up of a plastic tank with three fingerlings. The experimetal period was 42 days.

The water of the plastic tanks was constantly aerated with microporous stones and a mini air compressor. The experimental management involved daily morning (7:00 h) and afternoon (16:30 h) siphoning with the removal of 40 % water. Feces and the unconsumed feed were also removed. The internal tank walls were cleaned weekly to prevent the appearance of periphyton.

The treatments used in the present work were five feeds, four with increasing amounts of BSM (5, 10, 15, and 20 g kg^{-1} feed) and a control feed (without the addition of BSM). Five feed formulations with 30 % of DP and 3000 kcal kg^{-1} of DE were prepared (Boscolo *et al*., 2002; Meurer *et al*., 2003). The feed ingredients were ground and sieved through a 1 mm mesh, except BSM, which was already a fine powder.

Feed formulations are given in **table I**. After mixing, the feeds were pelletized in an experimental electric pelletizer and the pellets were dried in a forced air oven at 56 °C for 24 h. After drying, the pellets were crushed, and sorted in fractions by diameters

Table I. *Percent composition of feeds.* (Composição percentual das dietas).

	Ascophyllum nodosum (g kg^{-1} feed)				
	0.00	5.00	10.00	15.00	20.00
Ingredients					
Soybean meal	70.79	70.89	71.19	71.29	71.39
Maize	16.70	15.80	14.80	13.90	13.00
Soybean oil	5.00	5.30	5.50	5.80	6.10
Dicalcic phosphate	2.80	2.80	2.80	2.80	2.80
Calcitic lime	0.20	0.20	0.20	0.20	0.20
Ascophyllum nodosum[1]	0.00	0.50	1.00	1.50	2.00
Mineral and vitamin supplement[2]	4.00	4.00	4.00	4.00	4.00
Salt (NaCl)	0.50	0.50	0.50	0.50	0.50
Butyl hydroxyl toluene (BHT)	0.01	0.01	0.01	0.01	0.01
Total	100.00	100.00	100.00	100.00	100.00
Chemical composition[3] (%)					
Linoleic acid	3.52	3.65	3.78	3.91	4.04
Starch	19.95	19.40	18.85	18.30	17.74
Calcium	1.00	1.00	1.00	1.00	1.00
Digestible energy (kcal kg^{-1})	3000.00	3000.00	3000.00	3000.00	3000.00
Crude fiber	4.51	4.54	4.56	4.58	4.60
Phosphorus	0.97	0.97	0.97	0.97	0.97
Fat	6.62	6.87	7.12	7.36	7.61
Lysine	2.01	2.01	2.01	2.02	2.02
Methionine + cystine	0.96	0.96	0.96	0.96	0.95
Digestible protein	30.00	30.00	30.00	30.00	30.00
Crude protein	33.54	33.56	33.58	33.61	33.63

[1]From Valeagro Comércio e Distribuição de Produtos Agropecuários Ltda.; [2]Warranty levels per product kilogram: Vit. A, 1 200 000 UI; Vit. D3, 200 000 UI; Vit. E, 12 000 mg; Vit. K3, 2400 mg; Vit. B1, 4800 mg; Vit. B2, 4800 mg; Vit. B6, 4000 mg; Vit. B12, 4800 mg; Folic acid, 1200 mg; Pantotenate Ca, 12 000 mg; Vit. C, 48 000 mg; Biotin, 48 mg; Coline, 65 000 mg; Niacin, 24 000 mg; Fe, 10 000 mg; Cu, 6000 mg; Mn, 4000 mg; Zn, 6000 mg; I, 20 mg; Co, 2 mg; Se, 20 mg. [3]According to Rostagno *et al*. (2000).

appropriate for fingerling bite.

The fingerlings were fed three times a day at 8:00, 12:00 and 17:00 h at 10 % of their live weight. Every week, the experimental units were weighed for correction of diet amounts.

A BSM sample was sent to the Animal Nutrition Laboratory of UNIVASF for chemical analysis (Silva and Queiroz, 2002). The percent moisture, mineral matter, ether extract, crude protein and crude energy (kcal kg^{-1}) values found (as feed basis) were 9.64, 22.34, 2.49, 7.70, and 2826.99, respectively. The BSM values agree with those reported by Sharp (1986) and are close to the manufacturer's specifications.

The water oxygen concentration (oxymeter model Oxy-Check HI 9147, HANNA Instruments) and temperature were measured daily before siphoning. Water pH (pHmeter model Combo HI 98130, HANNA Instruments) and electric conductivity (conductometer model CA 150p TECNOPON Special Equipment Ltd.) were measured weekly before siphoning.

The mean values of morning and afternoon water temperature were, respectively, 28.6 ± 0.5 ºC and 29.6 ± 0.6 ºC, and of pH, dissolved oxygen and electric conductivity were 7.4 ± 0.4, 5.7 ± 0.9 mg L^{-1}, and 109.7 ± 7.4 µSm cm^{-1}, respectively. These parameters did not vary with treatments (p>0.05). The mean values of physical-chemical parameters of the plastic tank water noted above (pH, dissolved oxygen, electric conductivity, and morning and afternoon temperature) remained within the recommended values for the species (El-Sayed, 2006).

The fishes of the experimental units were individually weighed and measured weekly for correction of feed amounts and evaluation of growth. At the end of the experimental period (42 days), the fingerlings of all experimental units were counted, weighed and measured individually to determine the average survival rate, average final weight, percentage of weight gain, apparent feed conversion ratio, average length, head length, height, and breadth. Later, the fingerlings were stunned with cold water (2 ºC), slaughtered and gutted for evaluation of percent carcass yield with and without heads.

The studied parameters were submitted to variance analysis and, when the differences were significant at the 0.05 level, to regression testing with SAEG (Statistical and Genetic Analysis System) software (UFV, 2000).

RESULTS AND DISCUSSION

The survival ratio during the experimental period was 100 % in all experimental units. The mean initial weight, final weight, and weight gain values of Nile tilapia fingerlings submitted to feeds with increasing levels of BSM are given in **table II**. The initial weight of fingerlings used in the experiment was statistically similar (p>0.05) across treatments. Likewise, the mean final weight and weight gain did not differ significantly (p>0.05) among treatments.

The mean apparent feed conversion

Table II. *Mean final values of performance in Nile tilapia fingerlings submitted to feeds with increasing levels of brown seaweed* (A. nodosum) *meal during 42 days.* (Valores médios finais do desempenho para alevinos de tilápia do Nilo submetidos a dietas com níveis crescentes de farinha de alga marinha marrom *(A. nodosum)* durante 42 dias).

BSM	Weight (g) Initial	Final	Gain
0	0.42	3.76	3.34
5	0.43	4.27	3.85
10	0.43	4.26	3.83
15	0.43	4.29	3.86
20	0.42	5.19	4.71
CV	4.49	21.64	23.44

BSM= Brown seaweed meal (*A. nodosum*) g kg^{-1} feed; CV= Coefficient of variation.

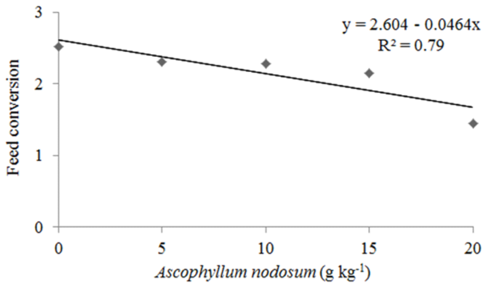

Figure 1. *Apparent feed conversion ratio of Nile tilapia* (O. niloticus) *submitted to feeds with increasing levels of brown seaweed* (A. nodosum) *meal during 42 days.* (Conversão alimentar aparente da tilápia do Nilo *(O. niloticus)* submetidos a rações com níveis crescentes de farinha de alga marinha marrom *(A. nodosum)* durante 42 dias).

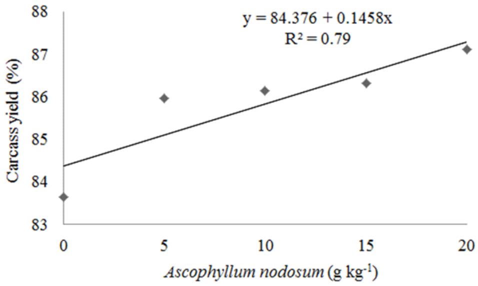

Figure 2. *Carcass yield of Nile tilapia fingerlings submitted to feeds with increasing levels of brown seaweed* (A. nodosum) *meal during 42 days.* (Rendimento de carcaça de alevinos de tilápia do Nilo submetidos a dietas com níveis crescentes de farinha de alga marinha marrom *(A. nodosum)* durante 42 dias).

(FCa) in Nile tilapia fingerlings showed a significant ($p<0.01$) linear improvement as a function of the increased levels of BSM in feeds **(figure 1)**.

The final standard length, head length, height and body width of Nile tilapia fingerlings submitted to feeds with increasing levels of BSM did not vary significantly ($p>0.05$) among treatments **(table III)**.

The final carcass yields, with and without heads, of Nile tilapia fingerlings increased linearly and significantly ($p<0.01$) with the increasing BSM levels (**figures 2** and **3**).

The effect of BSM on the final weight found in the present work corroborates the

Table III. *Mean final values (cm) of body parameters of Nile tilapia submitted to feeds with increasing levels of brown seaweed* (A. nodosum) *meal during 42 days.* (Valores finais médios (cm) de parâmetros corporais da tilápia do Nilo submetidas a dietas com níveis crescentes de farinha de alga marinha marrom *(A. nodosum)* durante 42 dias).

BSM[1]	SL	HL	BH	BW
0	5.03	1.73	1.74	0.93
5	5.37	1.85	1.84	0.97
10	5.25	1.83	1.80	1.02
15	5.33	1.83	1.82	0.94
20	5.19	1.82	1.77	0.92
CV	7.35	7.98	8.94	12.07

BSM= Brown seaweed meal (*A. nodosum*) g kg^{-1} feed; SL= Standard length; HL= Head length; BH= Body height; BW= Body width. CV= Coefficient of variation.

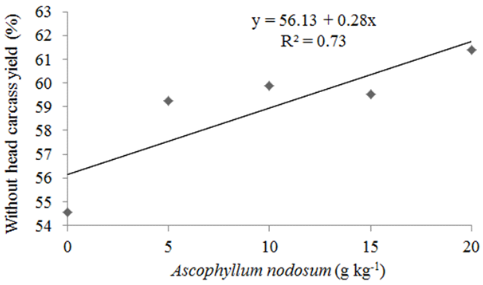

Figure 3. *Without head carcass yield of Nile tilapia fingerlings submitted to feeds with increasing levels of brown seaweed* (A. nodosum) *meal during 42 days.* (Rendimento de carcaça sem cabeça de alevinos de tilápia do Nilo submetidos a dietas com níveis crescentes de farinha de alga marrom *(A. nodosum)* durante 42 dias).

results reported by Nakagawa *et al*. (1997) for red sea bream (*Pargus major*) fingerlings. The weight of fingerlings administered feeds containing 2 % BSM did not vary significantly compared with the control treatment; however, the 5 % BSM treatment resulted in a larger final weight. The data in the present work differ from those of Alves Filho *et al.* (2011), who observed a decrease in weight gain in fingerlings of Nile tilapia (*O. niloticus*) at brown seaweed (*A. nodosum*) level of 3 %, and Davies *et al*. (1997), in juvenile grey mullet (*Chelon labrosus*) at red seaweed (*Porphyra purpurea*) levels of 0, 16.5, and 33 %.

The FCa of Nile tilapia given increasing levels of BSM during the experimental period differs from the results obtained by Nakagawa *et al*. (1997) for red sea bream fingerlings. Likewise, Davies *et al.* (1997) found poorer FCa values for juvenile grey mullet given feeds with high levels of red seaweed meal compared with control treatment results.

The standard length results for fingerlings also agree with the results reported by Nakagawa *et al*. (1997), in which the length of red sea bream administered feeds containing 2 % BSM was statistically similar to that of control fish. The carcass yields with and without head differed from those reported by Nakagawa *et al*. (1997), which did not reveal any difference in percent muscle ratio. Moreover, Alves Filho *et al.* (2011) asserted no significant effect of BSM levels on carcass weight without viscera. Nevertheless, the present results corroborate those obtained by Mustafa *et al*. (1995), who demonstrated that BSM resulted in increased muscle protein deposition in red sea bream. Despite the difference in parameter values, a larger muscle and/or protein deposition is generally related to better carcass yields.

The FCa is related to the live weight gain afforded by ingested food. In this experiment, the feed was pelletized with 10 % moisture, which is substantially lower than the 75 % body water of fish (Meurer *et al*., 2007). Therefore, the FCa may be influenced by the type of biomolecule or tissue deposited in the fish body. If the deposition occurs preferentially as lean tissue, the dry feed is converted to tissue with high water concentration, which improves the FCa. When deposition occurs as fat, as it does not have water, the FCa values are lower.

The FCa and carcass yield results obtained are consistent with each other, as the fat deposition in Nile tilapia generally occurs in viscera, which are removed for carcass analysis. Thus, it can be expected that a longer experimental period will result in better FCa values, and significantly better weight and weight gain values with BSM treatments.

The better FCa and carcass yield afforded by the treatments may be related to the BSM composition. Fike *et al*. (2005) stated that the bioavailability of microminerals, vitamins, antioxidants, and other metabolites present in *A. nodosum* may affect animal performance positively. Kumar *et al*. (2008) reported that microalgae have bioactive compounds, such as phycocolloids, minerals, vitamins, carotenoids, and n3 fatty acids, thus playing both a nutritional and medicinal role.

Another important factor pointed out in the literature is the immune stimulating role of BSM. According to Berteau and Mulloy (2003), brown seaweed has a large amount of sulphated fucans, which are oligosaccharides that act on the immune system. Saker *et al*. (2004) claim that the extract of *A. nodosum* has a positive effect on the immune system of ruminants. Among the few studies on fish, those of Gabrielsen and Austreng (1998) revealed that alginate from *A. nodosum*, used as an agglutinant in Atlantic salmon feed, had an immune stimulant effect with significantly increased levels of lysozyme in plasma. In addition, Nakagawa *et al*. (1997) reported that *A. nodosum* improved the physiological condition of fish.

Despite these encouraging results,

further research is necessary to determine the levels for the maximal development of the species, given the linear improvement in FCa and carcass yield with the addition of BSM. However, one should not expect an indication of much higher levels for Nile tilapia in this phase, as, according to Davies *et al.* (1997), the use of high levels of seaweeds in fish feed composition may lead to reduced performance. This may be related to the concentration of non-amilaceous carbohydrates in BSM. According to Meurer and Hayashi (2003), non-amilaceous carbohydrates have a negative effect on fish performance, while small amounts may result in the improvement of some performance parameters.

Another important factor to evaluate for BSM is its role as an additive, either as a food complement, an agglutinant, an immune stimulant agent, or a prebiotic. Greater attention should be paid to the study of BSM as a prebiotic, which is a non-digestible food component that affects the host positively by selectively stimulating the proliferation or the activity of desirable bacteria in the colon and possibly inhibiting the multiplication of pathogens and guaranteeing further benefits to the animal's health (Saad, 2006). According to Budiño *et al.* (2005), there is some evidence that prebiotics may result in anatomical changes in the digestive tract and stimulate the immune system (Silva and Nörnberg, 2003).

The evaluation of fish growth in experimental units by means of intermediate weighing demonstrated that the treatments with BSM stood out after the fifth week and the results became pronounced in the last week, which may be indicative that the tested ingredient may be effective over longer periods in the species. The BSM has good potential as a feed additive for Nile tilapia fingerlings. Nevertheless, an investigation of its application during other phases of the Nile tilapia growth, over longer treatment periods, as well as under sanitary challenging to verify the effect of BSM on immune responses and production performance, is important for its commercial use in Nile tilapia cultures.

CONCLUSIONS

The addition of BSM at the level of 20 g kg^{-1} feed does not result in growth loss and improves the feed conversion ratio and carcass yield in Nile tilapia during the fingerling period.

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