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Effects of replacement of corn flour with different forms of *Boscia senegalensis* leaf meal on growth, feed efficiency and survival of *Oreochromis niloticus* FRY

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Abstract

This study investigated the effects of replacement of corn flour with different forms of *Boscia senegalensis* leaf meal on the growth performance of *Oreochromis niloticus*. Five isonitrogenous (40% CP) and isolipidic (7% CL) were formulated to contain 7% corn flour meal and replace 18% corn flour meal with different forms of vaporized (M1), raw (M2), toasted (M3) and fermented (M4) *Boscia senegalensis* leaf meal. The control diet (M5) contained 25% corn flour meal without *Boscia senegalensis* leaf meal. Each aquarium (50L) stocked fifteen 0.4 g tilapia fry was assigned randomly to one of the three replicates of five treatment diets. Fishes were fed three times a day (08:00 A.M, 13:00 P.M and 17:00 P.M) for 12 weeks. The feeding rate was 10 % of the biomass during the first two weeks, then reduced to 8 % for the second two weeks, further reduced to 6 % for the third two weeks and finally reduced to 4 % for the last two weeks. The weight gain percentage of fish fed with diet containing toasted leaf meal is significantly higher than that of fish fed control diet. However, the weight gain percentage of fish fed with different types of leaf meal showed no significant difference.

Keywords: *Oreochromis niloticus*, *Boscia senegalensis*, growth, feed efficiency, survival

1. Introduction

The global warming, ocean pollution and the over-exploitation of marine fisheries resources are negatively impacting on capture fisheries production. Faced with these issues, aquaculture is challenged to redouble its efforts in order to reduce the impact especially that of over-exploitation by reducing the fishing efforts. The shortage of global fishmeal supply, combined with increased demand and competition for its use in other food producing sectors has further increased fishmeal price. In the long run, it is clear that many developing countries will not be able to rely on fishmeal as a major source of protein in aquaculture feeds. With this foresight, many researchers are working tirelessly in searching for locally available ingredients that could be used in fish feed whether cultivated or grown wild in order to reduce feed cost.

In Senegal, the development of aquaculture is facing a lot of challenges especially with regard to accessibility of high-quality feeds to fish farmers. As a result, there is a slight increase in aquaculture production, 1095 tonnes in 2014 compared to 2082 tonnes in 2016 [1]. The slow growth of aquaculture in Senegal, which is common in other parts of the world, is caused by high cost of aquaculture feeds due to high price of fishmeal and fish oil [2].

As a result, several attempts have been made to partially or totally replace fishmeal with cheaper and locally available plant protein sources while seeking to maintain the nutritional qualities of cultured fish [3-5].

Boscia senegalensis leaf is used as vegetable in the human diet and also as insecticides to control insects [6]. The main objective of this study was to investigate whether *Boscia senegalensis* leaf meal could replace corn flour in tilapia diet without negative effects on the growth and other parameters. Despite the use of *Boscia senegalensis* seeds in human diet, leaf has antinutritional factors, hence the idea of different transformation of the leaf in order to minimize the risk.

Tilapia is an important aquaculture species due to its hardiness and high yield potentials. In 2015, Tilapia ranked third in terms of global production after Cyprinids and Salmonids [7].

The specific objective of this research was to study the effects of replacing 18 % corn flour

with different forms of *Boscia senegalensis* leaf meal (raw, fermented, toasted and vaporized) on the growth performance, feed efficiency and survival of 0.2 g *Oreochromis niloticus* fry.

2. Materials and methods

2.1 Experimental Feeds

Five iso-nitrogenous (40 % CP) and iso-lipidic (7 % CL) practical diets were prepared. Four of the diets (M1, M2, M3 and M4) had different forms of *B. senegalensis* leaf meal (vaporized, raw, toasted and fermented) respectively while diet five (M5) had no *B. senegalensis* leaf meal. The ingredient composition of the experimental diets is shown in Table 1. The main ingredients used in the experimental feeds were obtained locally. The ingredients were first ground into fine powder. The *B. senegalensis* leaf meal was divided into four parts. One part was kept raw until the time of feed making and was used in the raw form. The other three parts were transformed into three forms (fermented, toasted and vaporized). During the feed preparation, the ingredients were measured according to the formulation and mixed thoroughly. Oil was added and further mixed. Thereafter, hot water was added and mixed to obtain dough. The dough obtained from each treatment was passed through a moulinex to produce spaghetti-like filaments that were dried for 30 to 60 minutes in a dryer. The dried filaments were ground into powder in a mortar before being used for experiment.

Table 1: Formulations of the experimental diets (as fed basis)

Ingredients (%)	Treatments				
	M1	M2	M3	M4	M5
Fish meal	44	44	44	44	44
Raw Boscia meal	0	18	0	0	0
Toasted Boscia meal	0	0	18	0	0
Fermented Boscia meal	0	0	0	18	0
Steamed Boscia meal	18	0	0	0	0
Fish oil	2	2	2	2	2
Fonio meal	6	6	6	6	6
Yeast	4	4	4	4	4
<i>Sterculia</i> Gum	2	2	2	2	2
Corn meal	7	7	7	7	25
Sesame meal	15	15	15	15	15
Vitamins	1	1	1	1	1
Minerals	1	1	1	1	1
Total	100	100	100	100	100

^a Vit A 250000 UI; Vit D₃ 250000 UI; Vit E 5000 mg ;Vit B₁ 100 mg ; Vit B₂ 400 mg ; Niacine 1000 mg ; Pantothenate Ca 2000 mg ; Vit B₆ 300 mg ; Vit K₃ 1000 g ; Vit C 5000 mg ; Biotine 15 mg ; Choline 100 g ; BHT 1000 mg;

^bPhosphorus 7%; Calcium 17%; Sodium 1.5%; Potassium 4.6%; Magnesium 7.5%; Manganese 738 mg; Zinc 3000 mg; Iron 4000 mg; Copper 750 mg; Iodine 5 mg; Cobalt 208 mg; Calcium and ground attapulgit q_s 1000 g ; Fluoride 1.5%.

2.2 Growth trial

The experiment was done at the aquaculture station of the University Institute of Fisheries and Aquaculture (IUPA). The fry was obtained by semi-artificial propagation at the same station. Fry of 23 days old of *O. niloticus* were used and the average weight was 0.2 g. The experiment lasted for 12 weeks. A total of 225 fry were divided among the five treatments and each treatment was triplicated. The stocking density was 15 fish per tank and the capacity per tank was 50 liters. During the trial, the tanks were filled to two-thirds of their volume and were covered in order to prevent the fish from jumping out. The tanks were cleaned daily to remove the

faeces. The water was sourced from tap water which was dechlorinated before use. The system was equipped with an air distributor that provided oxygen to the tanks. The experiment was conducted in an isolated culture system.

At the start of the experiment, the fishes were group weighed. They were fed three times per day (8:00 A.M, 1:00 P.M and 5:00 P.M) at 10 % of their body weight for the first two weeks, then 8 % for the second two weeks then 6 % for the thier two weeks and 4% thereafter until the end of the experiment. Water temperature in each tank was taken every day: morning (7:30) and evening (16:30) before feeding.

2.3 Sampling

Sampling was conducted after every two weeks by taken individual weight. At the termination of the experiment, all the fishes were counted, the weight and the body length of each fish were taken for determination of mean weight gain (MWG), relative mean weight gain (RWG %), specific growth rate (SGR), feed conversion ratio (FCR), protein efficiency ratio (PER) and survival rate (SR).

2.4 Formulae used for calculations

Growth performance parameters such as mean weight gain (MWG), relative mean weight gains (RMWG), specific growth rate (SGR) and feed efficiency parameters like feed conversion ratio (FCR), protein efficiency ratio (PER) and survival rate (SR) were measured using the following formulae:

- MWG (g) = final average weight - initial average weight
- SGR = [(ln (final weight) - ln (initial weight)) / duration of the experiment] * 100
- FCR = total dry feed fed / weight gain
- PER = weight gain × 100/ protein intake
- SR = (number of final fish / number of initial fish) * 100

2.5. Chemical analysis of the fish

At the end of the experiment, the fish were sacrificed, muscles collected from each treatment and sent to a local laboratory for analysis in order to determine the biochemical composition Crude Protein (CP), Crude Lipid (CL) and Ash). The analysis was based on the procedure of the Association of Official Analytical Chemists (AOAC). Samples of fish muscles were dried to constant weight at 105 °C for 24 h to determine moisture and consequently dry matter values were determined. Crude protein (total Nitrogen x 6.25) was determined by using micro-Kjeldahl method (Kjeltec System 1002 Distilling Unit, Tecator, Hoeganaes, Sweden). Crude lipid was extracted by Soxhlet method, ash was determined by incineration of samples in muffle furnace at 550 °C for 6 h.

2.6 Statistical Analysis

The data were analysed using the Statistical Analysis System Software (SAS-PC) [8] subjected to an analysis of variance (ANOVA). TUKEY test was used to compare the differences among the treatments. The differences were considered significant at P <0.05. The results were presented as means of the three replicates.

3. Results

Temperature varied between 25 °C and 29.4 °C with an average of 27.25 °C during the experiment. No pathological signs and symptoms were observed and all the diets were accepted by the fish. The culture environment was conducive for tilapia growth. Survival ranged between 93 – 100 % and

there were no statistical differences among the treatments. At the end of the experiment, different growth parameters (final weight, weight gain, relative mean weight gain and specific growth rate) and feed efficiency parameters (feed conversion ratio and protein efficiency ratio) were evaluated in order to determine the performances of the fish after subjecting them to different diets. The results are presented in Table 2.

With regard to the final weight, mean weight gain, relative mean weight gain and specific growth rate, M3 diet had the best results but there were no significant differences between it and the rest of the diets except M5 diet. There were no significant differences between M5 diet and M1, M2, and M4 diets with respect to the above mentioned parameters. With regard to the feed conversion ratio and protein efficiency ratio, there were no significant differences among M3, M1, M2 and M4 diets but they were significantly different from M5 diet.

Table 2: Growth Performance, Feed Efficiency and Survival Rate (N=3).

Parameters	Treatments				
	M1	M2	M3	M4	M5
IMWg/fish	0.2	0.2	0.2	0.2	0.2
FMW g/fish	10.56 ^{ab}	9.58 ^{ab}	11.49 ^a	11.06 ^{ab}	8.63 ^b
MWG g/fish	10.36 ^{ab}	9.38 ^{ab}	11.29 ^a	10.86 ^{ab}	8.43 ^b
RMWG (%)	5282 ^{ab}	4790 ^{ab}	5743 ^a	5529 ^{ab}	4314 ^b
SGR (%/d)	6.56 ^{ab}	6.41 ^{ab}	6.72 ^a	6.66 ^{ab}	6.23 ^b
FCR	0.93 ^a	0.94 ^a	0.91 ^a	0.93 ^a	1.4 ^b
PER	0.43 ^a	0.43 ^a	0.44 ^a	0.43 ^a	0.39 ^b
SR (%)	100	93	100	100	100

Values are means of the triplicates; values within the same row without a common superscript are significantly different ($p < 0.05$).

IMW = initial mean weight; **FMW** = final mean weight; **MWG** = mean weight gain; **RMWG (%)** = relative mean weight gain; **SGR (%/d)** = specific growth rate; **FCR** = feed conversion ratio; **PER** = protein efficiency ratio; **SR** = survival rate.

The results of the body composition (protein, lipid, ash) of the initial fish and after subjecting them to the different diets are shown in Table 3. The results showed that when the fishes were fed on different experimental diets, their body protein content significantly increased while their body lipid and ash levels were greatly decreased compared to the initial levels. There were no major differences among the experimental diets with regard to the protein, lipid and ash contents.

Table 3: The body composition of experimental fish.

Samples	Initial	M1	M2	M3	M4	M5
Protein (%)	68.01	88.43	87.51	87.12	87.30	85.43
Lipid (%)	14.01	3.14	2.91	2.91	2.73	3.74
Ash (%)	15.01	7.93	6.76	8.19	7.60	7.89

4. Discussion

The range of temperature (25 and 29.4 °C) during the experiment was conducive for tilapia culture. The values are similar to those reported by Towers^[9], (24 to 28 °C) ;^[10] (26 °C) and^[11] (27.1-29.6 °C) for good growth of *Oreochromis niloticus*. Survival ranged from 93 – 100% and substitution of 18 % corn flour with *Boscia senegalensis* leaf meal had no negative effect on the survival of 0.2 g *Oreochromis niloticus* fry.

The results revealed that substitution of 18 % corn flour with toasted *Boscia senegalensis* leaf meal had positive effects on growth parameters. Meanwhile, the substitution of 18 % corn

flour with raw or vaporized or fermented *Boscia senegalensis* leaf meal had no negative effects on the growth parameters. These results are in line with other studies on other leaf meals. Adewolu^[12] stated that sweet potato leaf meal can be used as a protein source in the *Tilapia zilli* diet up to 15 % without compromising growth. Although there were no significant differences among *Boscia senegalensis* leaf diets, valuewise fish fed treated (vaporized, toasted and fermented) ones were slightly higher than those fed on untreated (raw) one. The difference may be due to the high level of antinutritional factors (tannins, polyphenols and pythic acids) in the raw *B. senegalensis* leaf meal^[13].

In this experiment, the lowest growth performance was recorded in diet without *B. senegalensis* meal. The weight gain values recorded (8.43 to 11.29 g) are greater than 6.42 g obtained by^[4] with 10 % of cassava leaf meal inclusion in *Clarias gariepinus* diets and (6.68 g) obtained by^[14] with 10 % inclusion of *Leucaena leucocephala* leaf meal in *Clarias gariepinus* diets.

The fish fed on M3 diet recorded the best SGR (6.72 % / d) and the lowest SGR (6.23 % / d) was observed from fish fed on the control diet. The lower value in this study is higher than 2.42 % obtained by^[4] when 10 % cassava leaf meal was included in *Clarias* diets. The difference could be due to rearing conditions and the duration of the experiments.

With regard to feed efficiency (FCR and PER), the 18 % substitution of corn flour with *Boscia senegalensis* leaf meal had significant positive influences especially the transformed ones. This is supported by Holli *et al.*^[13] who claimed that treatment of leaf meals improved protein digestibility. The results obtained in this study on feed conversion ratio are good for all the diets. This is in line with those of Fasuyi^[15] who considered cassava leaf meal to be a good substitute for conventional protein sources of animal origin (fishmeal) which are currently not only very expensive but also competing with human consumption. The highest value (1.4) in the present research is better than the lowest value (1.94) reported by^[4] when fish were fed on diets containing 10 % cassava leaf meal. On the other hand, the best value (0.91) is higher than that of^[16] (0.43) on the partial replacement of soy flour by Moringa leaf flour. They suggested that Moringa leaf meal can be used as a staple for carp in place of soybean meal at 20 g / kg with no negative effect on growth and digestibility.

The best protein efficiency ratio (PER) (0.44) obtained at the end of the current experiment was lower than that obtained by^[5] (0.80) in the diet of *Oreochromis niloticus* and^[3] (3.96) when 25 % moringa leaf flour was used in the diet of *Clarias gariepinus*.

The results of the bromatological analysis of the flesh of the fish (protein, lipid, ash and dry matter) are recorded in Table 3. This analysis determined the composition of fish body at the beginning and end of the experiment. The protein content increased from 68.01 % for the initial fish up to 93.41 % after subjecting the fish to the test diets. These results are consistent with those of Hontiveros *et al.*^[17] who found that the carcass protein was higher in fish fed die tat 15 and 30% replacement of soybean by protein concentrate of water hyacinth leaf meal in Nile tilapia. On the contrary, the lipid content decreased from 14.01 % for the initial fish up to 2.73 % after subjecting them to experimental diets. These results are in line with those of Sarr *et al.*^[18] who claimed that the fat content of the initial fish was higher than that of the fish subjected to the different test diets. The result of the ash

content of the fish body also decreased from 15.01 % for the initial fish up to 6.76 % for the experimental fish. These results are consistent with those of Madalla *et al.* ^[19] reported that the ash content, however, declined significantly with increased moringa leaf meal (MLM) inclusion in tilapia diet.

Conclusion

The results of the research indicate that substitution of 18 % corn flour with *Boscia senegalensis* leaf meal has no negative effects on growth, feed efficiency and survival of *Oreochromis niloticus* fry. Transformation by toasting even has positive influence on growth and feed efficiency. The study reveals the possibility of incorporating *Boscia senegalensis* leaf meal in the diet of *Oreochromis niloticus* fry.

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