Evaluation of broken faba beans meal, *Vicia faba (L.)* as a partial or total replacement of fishmeal in practical diets of Nile tilapia, *Oreochromis niloticus* (L.) fingerlings.

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ABSTRACT:

The main objectives of this study was to evaluate the effect of partial and total replacement of fish meal protein by faba bean meal (FBM) protein in practical diets on growth performance, feed utilization and body composition of Nile tilapia, *Oreochromis niloticus* (L.). Fish of an average initial weight of 2.0 ± 0.01 g were stocked in 15 glass aquariums (80 l each) at a rate of 15 fish per aquarium. Fishmeal protein (15% of the diet) was used as the sole source of animal protein in the control diet. Percent replacement of fishmeal by faba bean on the basis of crude protein were as follows: 0 % (control diet A), 25 % (diet B), 50 % (diet C), 75 % (diet D) and 100 % (diet E). Diets were fed to fish at a rate of 6 %, then gradually reduced to 3 % of the total fish biomass daily, for a period of 16 weeks. The results of this study revealed that, the fish fed diet B (25 % FBM) had the best average body weight, specific growth rate (SGR), weight gain %, feed conversion ratio (FCR), and protein efficiency ratio (PER), but the lowest was obtained with fish fed diet E (100 % FBM). The same parameters of fish fed diet C (50 % FBM) and diet D (75 % FBM) were not significantly different ($P > 0.05$) from those of fish fed the control diet A. Proximate composition of whole body moisture and ash contents were not significantly different ($P > 0.05$) among all experimental diets and control diet. Whole body protein contents for fish fed diets B, C and D were superior to the control diet. Incorporation of faba bean meal in the diets increased significantly whole body fat content. Incorporation of faba bean meal in the diets significantly increased apparent digestibility coefficient of crude protein crude fat and energy. Therefore, these findings suggest that up to 75 % of fish meal protein can be replaced by faba bean meal protein in Nile tilapia diets without any adverse effects on growth performance and feed utilization, body composition and digestibility.

Key words: Nile tilapia, faba bean meal, growth, body composition, digestibility.

INTRODUCTION

The principal protein source in fish feeds is most often fishmeal. However, the rising cost and uncertain availability of fish meal has necessitated studies at either lowering the fish meal content or substituting fish meal with the more economical plant based proteins such as legumes (De Silva et al., 1988), lupine seed meal (De la Higuera et al., 1988), soybean meal (El-Saidy and Gaber, 1997; Eric et al., 2000; Carter and Hauler, 2000), cottonseed meal (El-Sayed, 1990; El-Saidy, 1999; El-Saidy and Gaber (2004) and cluster bean meal (El-Saidy et al., 2005) which were reported to be suitable as partial replacements for fish meal.

Cultivated faba bean is used as human food in developing countries and as animal feed, mainly for pigs, horses, poultry and pigeons in industrialized countries. It can be used as a vegetable, either green or dried, fresh or canned. It is a common breakfast food in the Middle East, Mediterranean region, China and Ethiopia (Bond et al., 1985). Feeding value of faba bean is high, and is considered in some areas to be superior to field peas or other legumes. It is one of the most important winter crops for human consumption in the Middle East. Faba bean has been considered as a meat extender or substitute and as a skim-milk substitute. Sometimes grown for green manure, but more generally for stock feed. Large-seeded cultivars are used as vegetable. Roasted seeds are eaten like peanuts in India" (Duke, 1981). The nutritional value of faba bean, *(Vicia Faba L.)* tested by Gropp et al. (1979) was found to be similar to that of soybean meal, while its cost per kg was approximately half. Utilizable protein, protein digestibility and biological value of faba bean are reported to vary from 14.8-15.5 %, 82-92 5 and 45-55 %, respectively (Hulse, (1994). Bond et al. (1985) reported that,the amino acid content of faba bean except for methionine is reasonably well balanced.

In Egypt**,** the faba bean, *Vicia Faba L.* known locally as 'Field bean' is widely grown, as a legumes plant and the young tender pods is popular vegetable for humans particularly in the rural areas.There is less information available on the use of broken faba bean meal as a protein source in aquatic animals' feeds. Therefore, a study has carried out to determine the feasibility of this faba bean brocken seeds meal as a possible replacement for fishmeal protein in practical diets for Nile tilapia, *Oreochromis niloticus* (L.) fingerlings.

MATERIALS AND METHODS

Experimental diets

Five isonitrogenous experimental diets containing approximately 32.6% crude protein were formulated. Diet A (control), with 15% fishmeal protein was formulated to be a high-quality commercial tilapia fish diet. The other four diets (diets B, C, D and E) contained 25, 50, 75 and 100% faba bean meal (FBM) protein replacement of fishmeal protein (Table 1). Faba bean meal was heated in boiled water under pressure (0.5 psi) for 30 min. at 80 C to reduce antitrypsin activity and tannin level according to Liener (1980).

In preparing the diets, dry ingredients were first ground to a small particle size (approximately 250 μm) in a Wiley mill. Ingredients were thoroughly mixed and then thoroughly added water to obtain a 30 % moisture level. Diets were passed through a mincer with die into 0.5-mm diameter spaghetti- like strands and were dried under the sun for 8 h. After drying the diets were broken up and sieved into appropriate pellet sizes. The proximate compositions of feed ingredients are given in Table 2 and the proximate compositions of each of the diets are given in Table 1. The calculated essential amino acids (EAA) are presented in Table 3. These values were calculated based on their ingredients levels presented in Table 1, according to NRC (1993). Percentage protein of the diets was determined by micro-kjeldahl, percentage fat was determined by ether extract method, and moisture was determined by drying (100 C) until constant weight (AOAC, 1995). Metabolizible energy (ME) was estimated from the diet ingredient according to NRC (1993).

Experimental system and animals

The feeding trial was conducted in 15 glass aquaria each containing 80-liter of dechlorinated tap water. About one third of water volume in each aquarium was daily replaced by aerated fresh water after cleaning and removing the accumulated excreta. All aquaria were supplied with compressed air for oxygen requirements. A photoperiod of 12-h light, 12-h dark (08.00 to 20.00 h) was used. Fluorescent ceiling lights supplied the illumination.

Water temperature and dissolved oxygen were measured every other day using a YSI Model 58 oxygen meter. Total ammonia and nitrite were measured twice weekly using a DREL, 2000 spectrophotometer. Total alkalinity and chloride were monitored twice weekly using the titration method; pH was monitored twice weekly using an electronic pH meter (pH pen; Fisher Scientific, Cincinnati, OH). During the 16-week feeding trial, the water-quality parameters averaged (± SD): water temperature, 26.8 ± 0.8 C: dissolved oxygen, 5.3 ± 0.7 mg /l: total ammonia, 0.20 ± 0.12 mg /l: nitrite, 0.08 ± 0.03 mg / l: total alkalinity, 175 ± 42 mg /l: chlorides, 571 ± 152 mg / l: pH, 8.3 ± 0.3.

A set of 225 fish of Nile tilapia, *Oreochromis niloticus* (L.) fingerlings average initial weight of 2.0 ± 0.01 g were collected from the stock of fish research laboratory in Shebin El-Kom, Faculty of Agriculture, Minufiya University and were used for the feeding trial. Fifteen fish were randomly stocked into each aquarium with three replications per treatment. After stocking, to minimize stress of handling, fish from each aquarium were weighed every 2 weeks and at the end of the feeding trial. All fish were fed initially 6 % of the total body weight daily and gradually decreased to 3 % daily. Fish were fed twice a day (0800 and 1600 h) 6 days per week for 16 weeks.

At the beginning of the study, 15 fish were sampled and frozen at –18 C for analysis of whole body composition. At the end of the experiment a random sample of five individual fish were randomly sampled from each aquaria. They were pooled, ground, stored in polyethylene bags and freeze for subsequent body crude protein, lipid, moisture and ash contents determined according to AOAC methods (AOAC, 1995).

Growth performance and feed conversion were measured in terms of final individual fish weight (g), survival rate (%), specific growth rate (SGR, % day $^{-1}$), feed conversion ratio (FCR), protein efficiency ratio (PER), and food intake (% body weight). Growth response parameters were calculated as follows: SGR (% day^{-1}) = ({In W_t - In W_i}/ T) x 100, where W_t is the weight of fish at time t, W_i is the weight of fish at time 0, and T is the rearing period in days: FCR = total dry feed fed (g) / total wet weight gain (g): PER = wet weight gain (g) / amount of protein fed (g): Feed intake = total dry feed fed (g/fish) (Richardson, et al., 1985).

Apparent nutrient digestibility:

After two-month feeding of experimental diets, feces were collected from each aquarium once daily every morning prior to feeding for a one-month period. The feces were collected on filter paper for drying as described previously by El-Saidy and Gaber (2002). The chemical analyses were conducted according to AOAC (1995). Chromic oxide is determined according to the procedure described by Furukawa and Tusukahare (1966). Apparent nutrient digestibility was calculated using the formula of Maynard and Loosli (1969).

$$\text{Apparent nutrient digestibility (\%)} = 100 - \left(100 \times \frac{\text{\% Cr2 O3 in feed}}{\text{\% Cr2 O3 in feces}} \times \frac{\text{\% Nutrient in feces}}{\text{\% Nutrient in feed}}\right)$$

Statistical analysis

Data were analyzed by analysis of variance (ANOVA) using the SAS ANOVA procedure (Statistical analysis system, 1988). Duncan's multiple range test (Duncan 1955) was used to compare differences among individual means. Treatment effect were considered significant at $P \leq 0.05$. All percentage and ratio were transformed to arcsin values prior to analysis (Zar, 1984).

Table 1: Composition and proximate analysis of the practical diets fed to Nile tilapia (*Oreochromis niloticus*).

	Diets				
	A (Control) (100 % FM)	**B** (25 % FBM)	**C** (50 % FBM)	**D** (75 % FBM)	**E** (100 % FBM)
Ingredient (%)					
Menhaden fish meal (60%CP)	25.0	18.75	12.5	6.25	0.0
Faba bean meal (37.5% CP)	0.0	10.0	20.0	30.0	40.0
Soy bean meal (44 % CP)	30.0	30.0	30.0	30.0	30.0
Wheat bran	22.75	6.75	11.5	16.0	19.0
Corn meal	12.75	23.0	14.5	6.25	0.0
Fish oil	5.0	5.0	5.0	5.0	5.0
Vit. & Min.Premix[1]	1.3	1.3	1.3	1.3	1.3
L-Methionine	0.0	1.0	1.0	1.0	1.0
L-Lysine	0.0	0.5	0.5	0.5	0.5
Sodium diphosphate	1.2	1.2	1.2	1.2	1.2
Molasses	1.5	1.5	1.5	1.5	1.5
Chromic oxide	0.5	0.5	0.5	0.5	0.5
Total (%)	100	100	100	100	100
Dry matter	93.5	94.6	93.6	92.8	92.9
Crude protein	32.6	32.6	32.6	32.6	32.6
Crude fat	13.8	13.9	14.0	13.9	13.7
Ash	7.1	7.5	7.8	6.6	6.3
Crude fiber	4.5	4.1	4.2	4.3	4.4
NFE	35.5	36.5	35.0	35.4	35.9
Met. Energy.[3]	3966	4015	3964	3971	3966

[1] Premix supplied the following vitamins and minerals(mg or IU)/ kg of diet, vit. A, 8000 I.U.; vit. D_3, 4000 I.U.; vit. E 50 I.U.; vit. K_3, 19 I.U.; vit. B_2, 25 mg; vit. B_3, 69 mg; Nicotinic acid, 125 mg; Thiamin, 10 mg; Folic acid, 7 mg; Biotin, 7 mg; vit. B_{12}, 75 mg; Cholin, 400 mg; vit. C, 200 mg; Manganese, 350 mg; Zinc, 325 mg; Iron, 30 mg; Iodine, 0.4 mg; Cobalt 2 mg; Copper, 7 mg; Selenium, 0.7 mg and 0.7 mg B.H.T. according to Lovell, 1989).

[2] Values represent the mean of three sample replicates.

[3] Based on the physiological fuel values of 4, 4, and 9 kcal ME / g Carbohydrate, Protein and Lipid, respectively.

Table 2. Proximate composition of feed ingredients (%).

Proximate composition	Ingredients				
	Fish meal	Soybean	Faba bean[1]	Wheat bran	Corn meal
Moisture	8.6	10.9	6.8	12.0	12.0
Protein	60.0	44.0	37.5	15.0	8.0
Ether extract	12.5	4.3	9.3	2.7	3.9
Ash	13.5	6.1	7.8	4.0	1.3
Fiber	1.8	6.3	11.6	9.4	3.5
NFE	3.6	28.4	27.0	56.9	71.3

[1]Faba bean had wide variation of protein content (20-41%) according to Chaven et al., (1989).

Table 3: Calculated levels of amino acids in different experimental diets and amino acid requirement of tilapia.

Amino acid	Requirement for tilapia [1]	Diets				
		A	B	C	D	E
Arginine	1.60	1.93	2.18	2.40	2.62	2.84
Histidine	0.65	0.75	0.76	0.77	0.77	0.78
Isoleucine	1.18	1.60	1.43	1.41	1.38	1.34
Leucine	1.29	2.20	2.16	2.13	2.11	2.08
Lysine	1.95	1.96	1.97	1.96	1.95	1.59
Methionine	1.02	1.61	1.57	1.53	1.50	1.46
Phenylalanine	1.43	1.46	1.45	1.45	1.43	1.37
Threonine	1.43	1.47	1.46	1.45	1.44	1.39
Valine	1.06	1.42	1.41	1.43	1.44	1.45
Tryptophan	0.32	0.38	0.47	0.56	0.65	0.74

[1]Source: Santiago and Lovell 1988.

RESULTS AND DISCUSSION

The results of average final body weight , specific growth rate (SGR), weight gain %, feed conversion ratio (FCR) and protein efficiency ratio (PER) are presented in Table 4. Fish fed diet B which contained 25 % faba bean meal protein showed the best average final body weight, specific growth rate (SGR), weight gain %, feed conversion ratio (FCR), and protein efficiency ratio (PER). However, the same parameters for diets A (control), C and D did not differ significantly from those of fish fed diet B (25% FBM). Diet E which contained 100 % protein from faba bean meal resulted in the lowest significantly average final body weight, specific growth rate (SGR), weight gain %, feed conversion ratio (FCR), protein efficiency ratio (PER). The unsuitability of substitution at this level was further reflected by the lack of response of the fish to the test diet at feeding times. The survival rate at the end of the feeding trail was high and recorded 100 % for all groups of fish fed the experimental diets. In the present study fish fed diet B (25% FBM) had faster growth rate and better food utilization than fish fed diet E (100 % FBM). Fish fed diets C

(50% FBM) and D (75 %FBM) did not differ significantly (P > 0.05) from those of fish fed control diet A (100 % fish meal protein)**.** The present study exhibited that faba bean meal protein can replace fishmeal protein up to 75 % in practical diets of Nile tilapia. This is in agreement with the results of Jackson et al. (1982), who fed *S. mossambicus* (13.9 g) isocaloric, isonitrogenous diets with varying levels of plant protein from cottonseed meal for 9 weeks. The best feed conversion ratio (FCR) and specific growth rate (SGR) were obtained at 50 % cottonseed meal. However, the fish grew at a reasonable rate even at a 100 % cottonseed meal inclusion level. Similar results have been reported for *Tilapia zillii* (El-Sayed, 1987), Nile tilapia (El-Sayed, 1990), Carp Hasan et al.(1997), Nile tilapia El-Saidy and Gaber, (2001) and Carp El-Saidy et al. (2005).

In the present study, however, tilapia which has an accelerated growth in much higher water temperatures have shown very significant growth depression with faba bean meal protein over 75 % of dietary protein (diets E). This growth depression was further elaborated by the lack

Table 4: Growth performance and nutrient utilization of Nile tilapia fed the experimental diets after 16 week. Values are mean ± SD of three replicates. Means in the same row bearing different superscript letters differ significantly at 0.05 level.

Parameters	Diets				
	A (control) (0 % FBM)	**B** (25 % FBM)	**C** (50 % FBM)	**D** (75 % FBM)	**E** (100 % FBM)
Initial wt. (g/fish)	2.0 ± 0.01	2.0 ± 0.01	2.0 ± 0.03	2.0 ± 0.03	2.0 ± 0.04
Final wt.(g / fish)	16.27 ± 0.20^{b}	18.87 ± 0.21^{a}	14.46 ± 0.23^{bc}	14.40 ± 0.35^{bc}	11.76 ± 0.29^{d}
Gain in wt.(g / fish)	14.27 ± 0.13^{b}	16.87 ± 0.14^{a}	12.46 ± 0.21^{bc}	12.40 ± 0.20^{bc}	9.76 ± 0.29^{d}
Weight gain (%)	713.50 ± 12.2^{b}	843.50 ± 7.6^{a}	623.0 ± 4.0^{bc}	620.0 ± 9.40^{bc}	488.0 ± 15.9^{d}
SGR (% day^{-1})	1.69 ± 0.01^{b}	1.82 ± 0.01^{a}	1.59 ± 0.01^{bc}	1.59 ± 0.01^{bc}	1.43 ± 0.02^{d}
FCR	2.2 ± 0.06^{a}	2.2 ± 0.01^{a}	2.4 ± 0.01^{ab}	2.1 ± 0.06^{a}	2.6 ± 0.1^{c}
PER	1.38 ± 0.03^{a}	1.43± 0.01^{a}	1.27 ± 0.01^{b}	1.43 ± 0.04^{a}	1.16 ± 0.04^{c}
FER	0.45 ± 0.01^{a}	0.46 ± 0.01^{a}	0.42 ± 0.01^{ab}	0.47 ± 0.02^{a}	0.38 ± 0.01^{c}
Feed intake (g/fish)	31.7 ± 0.48^{c}	36.31 ± 0.06^{d}	30.02 ± 0.34^{b}	26.56 ± 0.31^{a}	25.76 ± 0.12^{a}
Survival rate (%)	100	100	100	100	100

of response of the fish to the test diets at feeding times. This is in agreement with the results of Hashim et al., (1994) who reported that winged bean seed meal can not be used as a sole protein source for *O. niloticus* fry. Fish fed winged bean seed meal based diets exhibited poor growth performance, feed conversion ratio (FCR) and SGR. El-Saidy and Gaber (2001) and El-Saidy et al (2005) reported the same results with Nile tilapia and common carp, respectively.

The good growth, excellent condition and high survival in Nile tilapia fed diets containing faba bean meal protein up to 75 % in the present study demonstrated that faba bean meal protein had no adverse effect on fish performance at this level (75 %) of faba bean meal protein. However, increasing levels of faba bean meal protein to 100 % in the diets exhibited the adverse effect.

Table 5: Chemical analysis whole body composition of fish at the start and end of the experiment. Values are mean ± stander deviation. Values in the same row with same superscripts are not significantly different (P > 0.05).

Components	Initial	Diets				
		A (control) (0% FBM)	B (25% FBM)	C (50% FBM)	D (75% FBM)	E (100% FBM)
Moisture	79.0 ± 0.7	76.9 ± 0.4	75.9 ± 1.1	76.9 ± 0.3	75.8 ± 0.8	76.5 ± 0.7
Crude protein	61.3 ± 2.2	71.2 ± 0.3^{a}	72.4 ± 0.2^{a}	73.2 ± 0.5^{a}	71.2 ± 0.2^{a}	65.6 ± 0.4^{b}
Crude lipid	15.3 ± 1.4	11.8 ± 0.4^{b}	8.7 ± 0.3^{c}	12.5 ± 0.2^{a}	12.5 ± 0.03^{a}	13.9 ± 0.5^{a}
Crude ash	14.1 ± 1.3	15.59 ± 1.2	16.9 ± 1.3	13.5 ± 0.3	15.5 ± 0.3	18.2 ± 0.5

In the present study, the proximate composition of the experimental fish at the start and the termination of the feeding trial is shown in Table 5. Moisture and ash contents were not significantly different (P > 0.05) among all experimental diets and control diet. The highest value of protein contents was obtained on fish fed 50% FBM protein (diet C) and the lowest was obtained on fish fed 100% faba bean meal protein (diet E) and fish fed the control diet A. There was no significant differences (P > 0.05) in protein content between fish fed 75 % faba bean meal diet D and control diet A. Incorporation of faba bean in the diets increased significantly whole body fat content. The highest value of lipid content was recorded with fish fed 100 % faba bean meal protein (diet E) and the lowest was recorded with fish fed 25 % faba bean meal protein (diet B). The results of El-Saidy et al. (2005) exhibited the same trend.

Table 6: Apparent nutrient digestibility coefficients for Nile tilapia fingerlings fed diets with partial and total replacement of fishmeal by broken faba bean meal. Values in the same column with same superscripts are not significantly different (P > 0.05).

Diets	FM protein replacement (%)	Digestibility coefficients		
		Crude protein	Crude fat	Energy
A	0 (control)	84.9 ± 0.2^{c}	64.0 ± 1.4^{b}	59.0 ± 1.3^{c}
B	25	87.7 ± 0.8^{b}	51.3 ± 5.4^{c}	60.9 ± 1.6^{c}
C	50	90.4 ± 0.8^{a}	64.6 ± 2.5^{b}	65.0 ± 0.3^{b}
D	75	87.7 ± 0.8^{b}	72.2 ± 2.5^{a}	69.2 ± 1.2^{a}
E	100	85.2 ± 0.8^{c}	77.6 ± 1.4^{a}	65.1 ± 0.3^{c}

The results of apparent nutrients digestibility coefficients for Nile tilapia fingerlings fed diets with partial and total replacement of fish meal by broken faba bean meal are presented in Table 6. Faba bean meal significantly influenced apparent digestibility of crude protein crude fat and energy. Crude protein digestibility was significantly highest for diet C that contained 50 % faba bean meal. The apparent digestibility coefficients of crude protein and energy for all faba bean meal test diets were higher significantly compared with the control diet A (100% FM). In case of fat digestibility, the highest values was recorded with fish fed diet E (100% FBM) but, the lowest was recorded with

fish fed diet B (25% FBM). The values of apparent digestibility coefficient of fat for all faba bean meal diets, except diet B were superior to the control diet A (100% FM). Hulse (1994) reported that faba bean had higher protein digestibility, utilizable protein and biological value which ranged from 82-92%, 14.8-15.5 and 45-55%, respectively.

In conclusion, the present study revealed that faba bean meal protein can replace up to 75 % of fish meal protein in practical diets of Nile tilapia without any adverse effects on growth performance, feed utilization and body composition analysis of fish. In addition, broken faba bean meal used in the present study is available at much lower prices (1.5 LE/kg) than fishmeal (5.75 LE/kg) in many tropical and sub-tropical regions where tilapia culture is well established. Further research should be conducted under the field conditions.

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