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EFFECT OF DIETARY LUPIN (*Lupinus angustifolius*) WHOLE-SEED MEAL PROTEIN ON THE GROWTH OF TILAPIA (*Oreochromis niloticus*) REARED IN TANKS

T.M. Srour

Department of Animal and Fish Production, Faculty of Agriculture (Saba Basha), University of Alexandria

SUMMARY

A factorial feeding experiment (2 × 5) was conducted in indoor circular 1 m^3 fiberglass tanks to evaluate the effect of different levels of un-cooked and cooked lupin whole-seed meal protein, at the expense of fish meal (FM) protein, on the growth of Nile tilapia, Oreochromis niloticus, fingerlings (3 g/fish). Nine isocaloric and isonitogenous diets (25 % protein) were formulated where FM protein was replaced partially by 0, 15, 30, 45 and 60 % of un-cooked or cooked whole lupin seed meal protein. Tanks were stocked with 15 fish/tank in duplicate groups/treatment. Fish were fed daily at a rate of 3 % of fish live body weight for 14 weeks. The results indicated that fish receiving diets containing uncooked lupin protein were better in growth performance, feed and nutrient utilization than those fed cooked lupin protein. Growth performance, feed intake, feed conversion ratio and energy utilization began to deteriorate significantly at 45 % lupin protein. Protein efficiency ratio and protein productive values declined significantly subsequent to 30 % lupin protein level. Fish fed diets containing 0, 15 and 30 % lupin protein gained preferable FCR. Dry matter and hepatosomatic index were not affected by lupin protein levels or lupin treatments whil, ash and gross energy contents were affected by lupin protein levels but not lupin treatments. Meanwhile, crude protein and ether extract were affected by both lupin protein levels and lupin treatments. The economic evaluation was in favor of diets containing 15, 30 and 60 % un-cooked in addition to 30 % cooked lupin protein. However, the results of this study indicate that up to and including 30 % lupin protein could replace FM protein in Nile tilapia diets without any retardation in growth.

INTRODUCTION

In order to produce fish economically, formulated feeds are required. Protein requirement for fish in general is higher than that of livestock. Fish meal (FM) has been a prominent protein source used in most fish feed formulations. Otherwise, FM is usually used with high level in fish diets (Tacon and Jackson, 1985). However, due to the high cost of FM, there has been interest in, at least partially, replacing this ingredient with less expensive, available and good nutritional value plant protein sources in aquaculture feeds (Hardy, 1996). Most modern, nutrient-dense, aquaculture diets use some of plant protein ingredients (Palmegiano, *et al*., 2006). Lupin (*Lupinus* spp.) was shown to provide some potential as a useful feed ingredient in fish diets and has been used in commercial diets in some countries (Burel *et al.*, 1998; Glencross *et al*., 2002; 2003; 2006a and 2006b).

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In comparison to other plant protein sources, the potential of lupin is equaled perhaps only by soybean meals, which are presently widely accepted and used in the aquaculture feeds sector. Although the occurrence of alkaloids in soybean are not detected, lupin contains lower amounts of trypsin inhibitor, oligosaccharides and phytate compared to those detected in soybean (Petterson, *et al*., 1997 and Sitja`-Bobadillaa, *et al*., 2005). In many instances, the digestibility of lupin protein has been significantly superior to that of many other plant protein and/or animal protein sources (Hughes, 1988; Gomes *et al*., 1995; Burel *et al*., 2000; Smith *et al*., 2000 and Booth *et al*., 2001). The highly digestible protein and lipid components of lupin meals constitute almost the entire digestible energy value of this seed source (Allan *et al*., 1998; Burel *et al*., 2000; Kissil and Lupatsch, 2000). Gomes *et al*. (1995) found that the digestibility (%) of dry matter, protein and energy for FM and *L. angustifolius* whole seed meal were 78.0, 86.6 and 69.7 for FM and 63.3, 85.5 and 61.2 for lupin, respectively.

Additionally, lupin seeds have been used as a key feed ingredient in diet formulations for terrestrial livestock species (Petterson, 2000). Petterson *et al*. (1998) demonstrated that in low-specification diets for tilapia, the partial replacement of FM and/or soybean could be effectively achieved with *L. angustifolius* kernel meal. Meanwhile, performance of milkfish fed diets containing incremented levels of *L. angustifolius* kernel meal from 0 % to 100 % as FM replacer, apparently improved with increasing lupin in the diet. The best performance of milkfish fed the experimental diets was observed with those fed the 100 % *L. angustifolius* kernel meal diet (Petterson, 2000). On the other hand, Refstie, *et al*. (2006) stated that the tested lupin kernel meals and protein concentrates did not alter the intestinal function in Atlantic salmon when included at 30 % of the diet. Also in a study on rainbow trout by Glencross, *et al*. (2004 and 2006a) concluded that lupin is a highly useful feed ingredient for aquaculture rations.

There is a considerable variability in reports on maximum inclusion level of lupin meals in diets for aquaculture species with values ranging from 20 % to 70 % (De la Higuera *et al*., 1988; Robaina *et al*., 1995; Burel *et al*., 1998; Saraç *et al*., 1998 and Williams, 1998). As with other protein sources, the maximum inclusion is likely to be a function of the protein content of source used, the protein requirements of the animal and the level of feed attractants and ingestants included in the diet. Otherwise, alkaloids are one of the anti-nutritional factors found in lupin, which are typically low. Present levels of alkaloids in *L. angustifolius* are usually less than 200 mg/kg (Harris and Jago, 1984). Robaina *et al*. (1995) demonstrated that heat-treated and soaked lupin seeds in water for 24 h remove excess alkaloids.

There are few reports, however, on the nutritional or biological value of lupin when fed to fish species (Glencross *et al*., 2002). Also, there is no clear study examining the influence of inclusion levels of this ingredient whether un-cooked or cooked in the diets of local fish in Egypt. Therefore, this study examines the influence of incremental inclusion of un-cooked or cooked lupin protein at the expense of fish meal protein in the diet of Nile tilapia (*Oreochromis niloticus*) with a simple economic evaluation.

MATERIALS AND METHODS

The experiment was carried out at the Laboratory of Fish Nutrition, Faculty of Agriculture (Saba Basha), Alexandria University.

Fish and culture facilities

The feeding trial was conducted in twenty circular fiberglass tanks (each of 1 m^3) kept an in-indoor laboratory. Nile tilapia, *Oreochromis niloticus,* fingerlings were obtained from the Experimental Fish Farm of Agriculture Faculty (Saba Basha) (the 10^{th} Village, Abbis, Alexandria). Fish were randomly allocated to each tank in duplicates (15 fish/tank) per treatment, and adapated to the experimental conditions for 7 days. **Average** individual fish weight at the start of the study was 3 g. Subsequently, fish from each replicate were weighed at fortnightly intervals during the experimental period and the daily amount of feeds were readjusted as percentage of live body weight. About 20 fish were frozen for initial proximate body chemical analysis. The indoor laboratory was covered with light transparent fiberglass sheets to allow natural light The experimental tanks were cleaned every morning before the first feeding, and about one fourth of the water was replaced by a mixture of fresh dechlorinated tap and ground water. Water level was maintained at 0.8 m^3 throughout the experimental period, water temperature was checked daily, and ranged between 25 and 27^{o}C. Dissolved oxygen was kept close to saturation level by continuous aeration.

Experimental diets

Ingredients were bought from the local market and the dry ingredients were first ground to fine particles. The calculated amount of lupin (*Lupinus angustifolius*) whole-seed were divided into two divisions. The first division was ground to small particles in size, and the second was cooked in a pressure cooker for 15 min (Jayaram and Shetty, 1981) then kept in a plastic dispenser where water was changed three times a day for three days to remove the alkaloid substances then dried and finally ground to small particles. Afterward, both of cooked and un-cooked powdered lupins were incorporated separately into the diets, where, un-cooked or cooked lupin protein whole-seed meal replaced FM protein at a level of 0, 15, 30, 45 and 60 % (Table 2). All diets were sufficient in essential vitamins and trace minerals (NRC, 1993). Diet ingredients were thoroughly mixed in a plastic container. The oil was added, a few drops at a time, during mixing. Warm water (45^{o}C) was slowly added under continuous mixing until the diets began to clump. The diets were passed through commercial meat mincer 3 times, and dried for 24 hrs at 80^o C in a drying oven. Dried diets were stored in a freezer at -20^o C throughout the experimental period. All diets were formulated to be isonitrogenous (25 % protein, according to Jauncey and Ross (1982) and NRC (1993) and isocaloric. The diets were fed to the experimental fish two times a day (09,00 and 15,00 hr) at a rate of 3 % of live body weight on feed dry weight basis for 14 weeks (6 days a week).

Samples collection and analysis

At the termination of the experiments, fish were collected, weighed and counted per each replicate in each treatment. The samples from each experimental unit were oven dried at 60-80^{o}C for 48 hrs, and then ground to minute particles. Finally, fish samples and the experimental diets were submitted to proximate chemical analyses following the AOAC (1999) standard procedures. The nutrition equations were used according to Hepher (1988). All data were analyzed for statistical significance by using analysis of variance and examined by linear regression modeling (Stat View 5.0 computing package). The least significant difference (LSD) multiple comparison test was used to identify significant differences among treatment means (Steel and Torrie, 1980).

RESULTS AND DISCUSSION

The chemical analysis of fish meal (FM) in comparison with un-cooked and cooked lupin is presented in Table 1. The results revealed that crude protein, ether extract and ash contents were higher in FM while the contents of dry matter were similar to un-cooked and cooked lupin. The chemical composition of un-cooked and cooked lupin was almost paralle while numerically, un-cooked lupin contained higher ash and nitrogen free extract than cooked lupin. Otherwise, cooked lupin contained higher amount of dry matter, crude protein, ether extract, crude fiber and gross energy. Likewise, Petterson *et al*. (1997) found that Lupin seeds are typified by higher protein content (31 to 42 %) than most other grain legumes. There is considerable variation in the protein content between the various species and between cultivars and even within cultivars as a result of growing season and soil type.

Table 1. Chemical composition of fish meal and lupin (un-cooked and cooked) as percentage of dry matter basis

Ingredients	Composition (%)						Gross energy*
	DM	CP	EE	Ash	CF	NFE	
Fish meal	89.5	60	15	24	1	-	480.00
Un-cooked lupin	88.2	40	11.5	3.4	12.6	32.5	467.74
Cooked lupin	88.8	41	12.5	2.7	12.8	31.0	476.65

DM = Dry matter; CP = Crude protein; EE = Ether extract; CF = Crude fiber and NFE = Nitrogen free extract.

*Gross energy (Kcal/100g DM), calculated on the basis of 5.64, 4.11 and 9.44 Kcal GE/g protein, NFE and lipid, respectively (NRC, 1993).

Results of formulation and chemical composition of diets are shown in Table 2. The results of chemical composition revealed that all diets were almost isocaloric (about 426.21 Kcal/100 g dry matter) and isonitogenous (24.94 % crude protein). The mean value of protein to energy ratio (P:E ratio) was 58.51 mg protein/Kcal gross energy. As the level of lupin whether uncooked or cooked in different diets increase, nitrogen free extract and crude fiber were slightly incremented while ash contents were decreased.

The effect of replacing FM protein by different levels of un-cooked or cooked lupin protein on protein in the diet and on growth performance is presented in Table 3 and the linear regression in Table 4. Fish receiving diets containing uncooked lupin protein had significantly ($P<0.05$) better growth performance than those fed cooked lupin protein . Results of final weight (g/fish), gain (g/fish), average daily gain (ADG, mg/fish/day), specific growth rate (SGR, %) and daily growth coefficient (DGC, %/day) followed a similar pattern for fish receiving diets containing 0, 15 and 30 % lupin protein but revealed significantly ($P < 0.05$) higher values of growth performance compared to 45 and 60 % lupin protein replacements of FM protein.

Non significant differences were obtained when lupin protein was incremented up to 30 %, however, the difference between 45 and 60 of lupin protein levels was significant. Relationships between growth and the inclusion level of either cooked or un-cooked *L. albus* whole-seed meal were poor (De la Higuera *et al*., 1988). This supports the present result where no significant effect was noted due to cooking of lupins. An assimilated study by Viola *et al*. (1988) examined different inclusion levels of whole-seed *L. angustifolius* at the expense of fishmeal, for 41 days in diets for common carp (*Cyprinus carpio*) of 225 g. The results indicated that there were no significant differences in growth parameters at 45 % inclusion level. Also, the results obtained in the present study are in partial agreement with those of Burel *et al*. (1998) who examined the inclusion of (*Lupinus albus)* kernel meal in diets for rainbow trout at 30 %, 50 % and 70 % in diets. They found that *L. albus* kernel meal could be included in the diet of rainbow trout up to a level of 50 % with no loss in growth rate. The inclusion of *L. albus* kernel meal at 70 % however, resulted in poorer growth. It was suggested that the loss in growth performance of fish fed the diets containing 70% of *L. albus* was attributed to low feed intakes of this diet. Since the high levels of *L. albus* kernel meal inclusion resulted in a loss of palatability due to high alkaloid contents (Petterson *et al*., 1997). Furthermore, in a study for 42 day on rainbow trout (35.8 g/fish) by Glencross, *et al*. (2004) the growth of fish was significantly reduced at 50 % lupin inclusion level, but linear regression modeling suggested a decline in final weight, gain and daily growth coefficient at each inclusion level.

Table 2. Ingredients and chemical composition of the experimental diets containing various levels of cooked and uncooked lupin fed to Nile tilapia

Items	Diets No.								
	1	2	3	4	5	6	7	8	9
Ingredients (%)									
Fish meal	18	15.3	12.6	9.9	7.20	15.3	12.6	9.9	7.20
Lupin meal	-	4.05	8.1	12.15	16.2	3.95	7.90	11.85	15.81
Soy bean meal	23	23	23	23	23	23	23	23	23
Yellow corn	28	28	28	28	28	28	28	28	28
Wheat bran	14	14	14	14	14	14	14	14	14
Rice particles	14.8	10.5	9.4	8.15	7.2	10.6	9.6	8.45	7.59
Vegetable oil	0.2	3.15	2.9	2.8	2.40	3.15	2.9	2.8	2.40
Vit. & Min.[1]	2	2	2	2	2	2	2	2	2
Proximate composition (%)									
Dry matter	88.7	89.24	89.02	88.83	88.70	89.11	88.93	88.75	88.61
On dry matter basis (%)									
Crude protein	25.05	25.01	24.95	24.87	24.80	25.03	25.03	24.88	24.83
Ether extract	7.73	7.64	7.56	7.67	7.66	7.64	7.65	7.66	7.67
NFE	51.32	51.58	51.89	52.04	52.32	51.58	51.86	52.1	52.40
Crude fiber	4.6	5.09	5.61	6.11	6.61	5.09	5.60	6.10	6.60
Ash	11.3	10.68	9.99	9.31	8.61	10.66	9.86	9.26	8.5
Gross energy[2]	425.18	425.17	425.35	426.56	427.22	425.29	426.53	426.77	427.81
P:E ratio[3]	58.92	58.82	58.66	58.30	58.05	58.85	58.68	58.30	58.04

Diets 1, 2, 3, 4 and 5 contained 0, 15, 30, 45 and 60 % un-cooked lupin protein, respectively.
Diets 6, 7, 8 and 9 were 15, 30, 45 and 60 % cooked lupin protein, respectively.
[1]Meveco premix, Vit. & Min., every 1.5 kg contains Vit. A 125 million IU, D_3 3 million IU, E 15 g, K_3 2.5 g, B_1 1.5 g, B_2 5 g, B_6 2 g, Pantothanic acid 10 g, B_{12} 0.01g, Nicotenic acid 30 g, , Folic acid 1.2 g, Fe 30 g, Mn 60 g, Cu 10 g, I1 g, Cobalt 0.25 g, Se 10 g and Zn 55 g. [2] Gross energy (Kcal/100g DM), calculated on the basis of 5.64, 4.11 and 9.44 Kcal GE/g protein, NFE and lipid, respectively (NRC, 1993).[3]Protein to energy ratio (mg/Kcal).

Table 3. Growth performance of Nile tilapia (*O. niloticus*) fed different levels of cooked and un-cooked lupin protein

Item	Final weight	Gain[1]	ADG[2]	SGR[3]
		Lupin treatment (T)		
Uncooked	50.23[a]	47.23[a]	481.949[a]	2.869[a]
Cooked	48.88[b]	45.88[b]	468.163[b]	2.838[b]
		Lupin level, % (L)		
0	55.000[a]	52.000[a]	530.638[a]	2.968[a]
15	54.750[a]	51.750[a]	528.060[a]	2.965[a]
30	53.250[a]	50.250[a]	512.755[a]	2.933[a]
45	43.975[b]	40.975[b]	418.112[b]	2.740[b]
60	40.800[c]	37.800[c]	385.715[c]	2.663[c]
Diet		**Interaction T × L**		
1	55.00[a]	52.00[ab]	530.638[ab]	2.968[ab]
2	55.60[a]	52.60[a]	536.735[a]	2.980[a]
3	53.40[a]	50.40[bc]	514.285[bc]	2.935[b]
4	45.05[a]	42.05[d]	429.085[d]	2.765[c]
5	42.20b[c]	39.20[e]	400.000[e]	2.700[d]
6	53.90[a]	50.90b[c]	519.385[bc]	2.950[ab]
7	53.10[a]	50.10[c]	511.225[c]	2.930[b]
8	42.90b[c]	39.90[e]	407.140[e]	2.715[d]
9	39.40c	36.40[f]	371.430[f]	2.625[e]

Means in the same column within each item having different superscripts are significantly different ($P < 0.05$).
Diets 1, 2, 3, 4 and 5 contained 0, 15, 30, 45 and 60 % un-cooked lupin protein, respectively. Diets 6, 7, 8 and 9 were 15, 30, 45 and 60 % cooked lupin protein, respectively.
[1]Gain (g/fish) = Final wt., g. – Initial wt., g.
[2]Average daily gain (mg/fish/day) = (Final wt. – Initial wt.) / period (days).
[3]Specific growth rate (%) = 100 (ln final weight–ln initial weight) / time (days).

Table 4. Linear regression of cooked and uncooked lupin protein relationships between dietary inclusion levels (x) and growth performance parameters (y)

Parameter	Equation	
	Uncooked lupin	Cooked lupin
Final weight	y = 57.1198–0.2197x	y = 57.3600–0.2827x
Weight gain	y = 54.4198–0.2397x	y = 54.3600–0.2827x
Average daily gain	y = 555.335–2.4462x	y = 554.694–2.8844x
Specific growth rate	y = 3.01798–0.0050x	y = 3.02300–0.0062x

The effect of replacing FM protein by different levels of un-cooked or cooked lupin protein in the diet of Nile tilapia on feed and nutrient utilization is shown in Table 5 and the linear regression molding is presented in Table 6. Fish fed cooked lupin surpassed those fed uncooked lupin in feed intake. On the contrary, fish receiving diets containing un-cooked lupin surpassed those fed cooked lupin in feed conversion ratio (FCR), protein efficiency ratio (PER), protein productive value

(PPV, %) and energy utilization (EU, %). These results are in accordance with the findings of Van Barneveld (1993).

Moreover, as with many other protein sources, cooking or autoclaving of lupin meals reduces the nutritional value of their protein content (De la Higuera *et al*., 1988; Vandepeer *et al*., 1999) and increases feed palatability because of alkaloid removal (Robaina *et al*., 1995). These may support our results of feed intake, PER and PPV. Feed intake, FCR and EU were not significantly ($P > 0.05$) different among fish fed diets containing 0, 15 and 30 % lupin protein instead of FM protein. Burel *et al*. (1998); Farhangi and Carter (2001) and Glencross, *et al*. (2004) deemed that the level of inclusion of the yellow lupin did not significantly affect feed intake. It was suggested that the high levels of *L. albus* kernel meal inclusion into fish diets resulted in a weak feed intake due to a loss of palatability caused by high alkaloid contents (Petterson *et al*., 1997). These excess alkaloids could be removed from Lupin seeds by heat treatment and soaking in water for 24 h (Robaina *et al*., 1995). Perhaps this may explain the increased feed intake in fish receiving diets containing cooked lupin compared with those receiving un-cooked lupin. In the same manner, fish receiving the control diet and those fed 15 % lupin protein in their diet had significantly higher ($P < 0.05$) values of PER and PPV than other groups followed by that fed 30 % lupin protein with nonsignificant difference between fish fed the control diet and that fed 30 % lupin protein instead of FM protein in their diet.

Meantime, no significant differences in FCR, PER, PPV and EU were found between fish receiving diets containing 45 and 60% lupin protein instead of FM protein while, a significant ($P<0.05$) diet response was seen in feed intake. Similar results were observed by Morales *et al*. (1994) and Burel *et al*. (1998).

Because of the variability of feed intake which had considerable impact on the FCR values (De la Higuera *et al*., 1988), fish fed diets containing 0, 15 and 30 % lupin protein had preferable FCR. Accordingly, Glencross *et al*. (2004) demonstrated that feed conversion ratio deteriorated significantly at the 50% lupin level, with a linear regression suggesting a decline in FCR at each inclusion level. Retention efficiency of nitrogen was unaffected by level of inclusion though a significant decline in the efficiency of energy retention was observed with increasing inclusion of yellow lupin. A significant ($P<0.05$) interaction was found between lupin protein levels (0, 15, 30, 45 and 60 %) and lupin treatments (un-cooked and cooked) on feed intake and FCR, while it was nonsignificant ($P > 0.05$) for other parameters.

Carcass proximate analysis results of the experimental fish fed diets containing different levels of un-cooked or cooked lupin protein instead of FM protein are summarized in Table 7 and the linear regression molding is shown in Table 8. Changes in dry matter of fish carcass in relation to lupin protein levels or lupin treatments were not significant ($P > 0.05$). Furthermore, body ash and gross energy contents in fish were not affected by lupin treatments (un-cooked and cooked). Meanwhile, fish fed un-cooked lupin had significantly higher percentage of protein and ether extract in their body than fish fed cooked lupin. Moreover, fish fed the control diet had significantly ($P<0.05$) higher carcass protein contents than other fish groups receiving different lupin protein levels in their diets instead of FM protein. Also, the difference in protein contents of fish received 30 and 45 % lupin protein was nonsignificant while, the protein contents of this group differ significantly ($P < 0.05$)from those receiving 45 and 60 % lupin protein. Fish receiving 60 % lupin protein had significantly ($P<0.05$) lower protein contents. However, there was a significant increase in ether extract and gross energy with increasing lupin protein instead of FM protein in the diet. This agrees with the results reported by Morales *et al*. (1994) In which, fish fed diets containing 45 and 60 % lupin protein exhibited a significantly higher contents of ether extract and gross energy. Nevertheless, the ash content of the groups receiving 0 and 15 % lupin protein instead of FM protein was significantly (P<0.05) higher than that of other groups, while the difference within them was nonsignificant. Conversely, Glencross *et al*. (2004) stated that the composition of rainbow trout fed the experimental diets was not significantly affected by the inclusion of yellow lupin kernel meal. Hepatosomatic index values were not affected by either lupin levels or lupin treatments. This result is inconsistent with the finding of Glencross *et al*. (2004) who found an increase in the relative size of gastrointestinal tract with increasing levels of lupin inclusion in the diet of rainbow trout.

Table 5. Feed and nutrient utilization of Nile tilapia (*O. niloticus*) fed different levels of cooked and uncooked lupin protein

Item	Feed intake,g	FCR[1]	Feed utilization		Energy utilization[4]
			PER[2]	PPV[3]	
		Lupin treatment (T)			
Uncooked	92.73[b]	1.979[a]	2.039[a]	28.495[a]	16.547[a]
Cooked	94.70[a]	2.090[b]	1.938[b]	27.015[b]	15.798[b]
		Lupin level, % (L)			
0	99.000[a]	1.908[a]	2.100[ab]	29.658[ab]	16.782[a]
15	96.875[a]	1.875[a]	2.140[a]	30.048[a]	17.340[a]
30	98.700[a]	1.965[a]	2.038[b]	28.383[b]	16.595[a]
45	90.500[b]	2.213[b]	1.823[c]	25.310[c]	15.100[b]
60	83.500[c]	2.213[b]	1.843[c]	25.378[c]	15.045[b]
Diet		**Interaction T × L**			
1	99.000[a]	1.908[c]	2.100[b]	29.658[b]	16.782[b]
2	94.25[bc]	1.795[d]	2.235[a]	31.410[a]	17.965[a]
3	100.40[a]	1.995[c]	2.010[bc]	28.170[b]	16.355b[c]
4	89.50[d]	2.130[b]	1.890[d]	26.300[c]	15.610[c]
5	82.50[e]	2.105[b]	1.920[cd]	26.445[c]	15.690[c]
6	99.50[a]	1.955[c]	2.045[b]	28.685[b]	16.715[b]
7	97.00[ab]	1.935[c]	2.065[b]	28.595[b]	16.835[b]
8	91.50[cd]	2.295[a]	1.755[e]	24.320[d]	14.590[d]
9	84.50[e]	2.320[a]	1.765[e]	24.310[d]	14.400[d]

Means in the same column within each item having different superscript are significantly different (P < 0.05).

Diets 1, 2, 3, 4 and 5 contained 0, 15, 30, 45 and 60 % un-cooked lupin protein, respectively. Diets 6, 7, 8 and 9 were 15, 30, 45 and 60 % cooked lupin protein, respectively.

[1]Feed conversion ratio: total dry diet fed (g)/total wet weight gain (g).

[2]Protein efficiency ratio: wet weight gain (g)/amount of protein fed (g).

[3]Protein productive value (%): $(P-P_0)\ 100/P_i$ where P is protein content in fish carcass at the end of the experiment, P_0 is the protein content in fish carcass at the start of the experiment and P_i is the protein in feed intake.

[4]Energy utilization (%): $(E-E_0)\ 100/E_i$ where E is the energy in fish carcass (Kcal) at the end of the experiment, E_0 is the energy in fish carcass (Kcal) at the start of the experiment, and E_i is the energy in feed intake (Kcal).

Table 6. Linear regression of cooked and uncooked lupin protein relationships between dietary inclusion levels (x) and feed & nutrient utilization parameters (y)

Parameter	Equation	
	Uncooked lupin	Cooked lupin
Feed intake, g	y = 99.4800–0.2250x	y = 102.900–0.275292x
Feed conversion ratio	y = 1.81800+0.0054x	y = 1.87200+0.007266x
Protein efficiency ratio	y = 2.19599–0.0052x	y = 2.11400–0.005866x
Protein productive value	y = 30.4988–0.0835x	y = 29.8300–0.093833x
Energy utilization	y = 17.5880–0.0347x	y = 17.0400–0.041433x

Table 7. Proximate body composition and hepatosomatic index of Nile tilapia (*O. niloticus*) fed different levels of cooked and un-cooked lupin

Item	Dry matter	On dry matter basis (%)			Gross energy[1]	HSI[2]
		Crude protein	Ether extract	Ash		
Lupin treatment (T)						
Uncooked	26.23[a]	53.089[a]	24.561[a]	22.771[a]	527.307[a]	3.014[a]
Cooked	26.32[a]	52.879[b]	24.140[b]	22.560[a]	530.093[a]	2.776[a]
Lupin level, % (L)						
0	26.275[a]	53.728[a]	22.598[d]	23.675[a]	516.345[d]	2.315[a]
15	26.300[a]	53.228[b]	23.573[c]	23.200[a]	522.725[c]	3.061[a]
30	26.100[a]	53.155[b]	24.370[b]	22.475[b]	529.848[b]	2.823[a]
45	26.35[a]	52.523[c]	25.478[a]	22.000[b]	536.735[a]	2.900[a]
60	26.250[a]	52.288[d]	25.735[a]	21.978[b]	537.840[a]	3.375[a]
Diet		**Interaction T × L**				
1	26.275[a]	53.728[a]	22.598[d]	23.675[a]	516.345[d]	2.315[a]
2	26.30[a]	53.325[b]	23.125[d]	23.550[ab]	519.050[cd]	3.373[a]
3	26.20[a]	53.210[bc]	24.040[c]	22.750[bc]	527.045[bc]	2.708[a]
4	26.30[a]	52.710[d]	25.290[ab]	22.000[cd]	536.020[a]	3.125[a]
5	26.20[a]	52.450[e]	25.400[ab]	22.150[cd]	535.595[a]	3.550[a]
6	26.30[a]	53.130[bc]	24.020[c]	22.850[abc]	526.400[bc]	2.750[a]
7	26.00[a]	53.100[c]	24.700[bc]	22.200[cd]	532.650[ab]	2.938[a]
8	26.40[a]	52.335[e]	25.665[a]	22.000[cd]	537.450[a]	2.675[a]
9	26.30[a]	52.125[f]	26.070[a]	21.805[d]	540.085[a]	3.200[a]

Means in the same column within each item having different superscript are significantly different ($P < 0.05$).

Diets 1, 2, 3, 4 and 5 contained 0, 15, 30, 45 and 60 % un-cooked lupin protein, respectively. Diets 6, 7, 8 and 9 were 15, 30, 45 and 60 % cooked lupin protein, respectively.

[1]Gross energy (Kcal/100 g dry matter), calculated on the basis of 5.64, 4.11 and 9.44 Kcal GE/g protein, NFE and lipid, respectively (NRC, 1993).

[2]Hepatosomatic index: (liver weight / total body weight) ×100

Table 8. Linear regression of cooked and uncooked lupin protein relationships between dietary inclusion levels (x) and carcass composition parameters (y)

Parameter	Equation	
	Uncooked lupin	Cooked lupin
Dry matter	Not Significant	Not Significant
Crude protein	y = 53.7300–0.0214x	y = 53.6700–0.0264x
Ether extract	y = 22.6850+0.0485x	y = 22.7440+0.0606x
Ash	y = 23.5830–0.0271x	y = 23.5860–0.0342x
Gross energy	y = 517.196–0.3369x	y = 517.401–0.4231x
Gonado-somatic index	Not Significant	Not Significant

Results of the simple economic evaluation of replacing lupin protein for FM protein in the diets of Nile tilapia are presented in Table 9. Mathematically, as lupin protein incremented in fish diets as FM replacer, the feed cost decreased. However, when biological values (e.g. feed conversion ratio) were taken into consideration, diets contained 15, 30 and 60 % un-cooked lupin protein as well as 30 % cooked lupin protein had lowest cost of kg fish gain and highest change in feed cost per kg fish gain. Diets contained 15 and 60 % un-cooked in addition to 30 % cooked lupin protein were superior economically as compared to other diets. While on the contrary, diet contained 45 and 60 % cooked lupin protein exhibited lower change in feed cost per kg fish gain.

Table 9. Cost (L.E.) of feed required for production of one kg gain of Nile tilapia (*O. niloticus*) fed different levels of un-cooked and cooked lupin whole seed meal protein

Diets	Feed cost (LE/ton feed)	Amount of feed/kg gain	Cost of kg fish gain (LE)	Change in feed cost/kg gain (%) compared to control
1*	2611.00	1.908	4.98	0
2	2559.96	1.795	4.60	-7.63
3	2560.38	1.955	5.01	+0.60
4	2454.40	1.995	4.90	-1.61
5	2455.35	1.935	4.75	-4.62
6	2351.45	2.130	5.01	+0.60
7	2352.88	2.295	5.40	+8.43
8	2243.40	2.105	4.72	-5.22
9	2245.46	2.320	5.21	+4.62

* Used as a base for calculation

Diets 1, 2, 3, 4 and 5 contained 0, 15, 30, 45 and 60 % un-cooked lupin protein, respectively.

Diets 6, 7, 8 and 9 were 15, 30, 45 and 60 % cooked lupin protein, respectively.

Cost in LE/ton: Fish meal: 7000, Soybean meal: 2600, Yellow corn: 1150, Un-cooked lupin: 2500, Cooked lupin 2550, Wheat bran: 840, Rice particles: 1000, Vegetable oil: 2700 and Min. & Vit.: 8000.

CONCLUSION

From the previous results, it may be concluded that un-cooked lupin protein is a useful feed ingredient for Nile tilapia. Furthermore, up to 30 % un-cooked lupin protein can replace FM protein without any reduction in growth, but feed intake problems were encountered with higher replacement of the fishmeal. Economically, 15, 30 and 60 % un-cooked in addition to 30 % cooked lupin protein were the best.

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تأثير التغذية على بروتين مسحوق بذور الترمس على نمو البلطى النيلى المربى فى تنكات

طارق محمد أحمد سرور

قسم الإنتاج الحيوانى والسمكى. كلية الزراعة، سابا باشا، جامعة الإسكندرية

أجريت تجربة تغذية عاملية (2 × 5) فى تنكات من الألياف الصناعية (سعة 1 م^3 /تانك من الماء) بداخل المعمل لتقدير إمكانية إحلال مستويات مختلفة من بروتين مسحوق بذور الترمس المطبوخ أو غير المطبوخ محل بروتين مسحوق السمك فى أصبعيات أسماك البلطى النيلى (3 جم/سمكة). تم تحضير 9 علائق متساوية فى المحتوى من الطاقة والبروتين الذى كانت نسبته فى العليقة حوالى 25 ٪ حيث حل بروتين مسحوق بذور الترمس المطبوخ وغير المطبوخ كل منهما منفصلا محل 0 و15 و30 و45 و60 ٪ من بروتين مسحوق السمك. خزنت التنكات بعدد 15 سمكة/تانك وكان معدل التغذية اليومى 3 ٪ من وزن الجسم الحى واستمرت التجربة لمدة 14 أسبوع. وقد أشارت النتائج الى أن الأسماك التى غذيت على بروتين مسحوق بذور الترمس غير المطبوخ أحرزت معدلات نمو أعلى من تلك التى تناولت بروتين مسحوق بذور الترمس المطبوخة. هذا وقد بدأ كل من كفاءة النمو وكمية الغذاء المستهلك ومعدل التحويل الغذائى والإستفادة من الطاقة فى الإنخفاض عند مستوى إحلال 45 ٪. وقد تساوت الأسماك التى غذيت على العليقة المعيارية مع تلك التى غذيت على مستوى إحلال 15 ٪ من ناحية ومع 30 ٪ من ناحية أخرى فى كفاءة الإستفادة من البروتين و قيمة البروتين المنتجة فى الأسماك. وقد كانت كفاءة التحويل الغذائى الأفضل عند مستويات الإحلال صفر و15 و30 ٪ بروتين مسحوق بذور الترمس. لم يتأثر كل من محتوى جسم الأسماك من المادة الجافة ولم يتأثر معامل وزن الكبد التقريبى بالنسبة لجسم السمكة بمستويات إحلال بروتين مسحوق بذور الترمس سواء كان مطبوخ أو غير مطبوخ. وقد تأثر المحتوى من الرماد والطاقة بمستويات الإحلال ولم يتأثر بطبخ أو عدم طبخ بروتين مسحوق بذور الترمس. فى حين تأثر محتوى أجسام السمكة من البروتين والدهون بكل من مستويات إحلال بروتين مسحوق بذور الترمس وطبخ أو عدم طبخ الترمس. وقد أظهر التحليل الإقتصادى للعلائق تفوق العلائق ذات مستويات الإحلال 15 و30 و60 ٪ من بروتين مسحوق بذور الترمس غير المطبوخ وتلاهما 30 ٪ بروتين مسحوق بذور الترمس المطبوخ. هذا وقد أظهرت نتائج هذه التجربة أنه يمكن إحلال بروتين مسحوق بذور الترمس غير المطبوخ بنسبة تصل الى 30٪ من بروتين مسحوق السمك فى علائق البلطى النيلى بدون إعاقة النمو.