

INCORPORATION OF LUPIN SEED MEAL AS PLANT PROTEIN SOURCE IN GILT HEAD SEA BREAM (*Sparus aurata*) DIETS.

Yones, A. M.

National Institute of Oceanography and Fisheries, Shakhshouk Aquatic Research Station at El Fayoum, Egypt.

ABSTRACT

The extruded lupin seed meal (*Lupinus albus*) was used as alternative protein source to partially replace fish meal in feeding of *Sparus aurata*. Four experimental diets were formulated to contain 40.68% \pm 0.12 CP. Diet 1, without lupin seed meal and considered to be the control, where L10, L20 and L30, lupin seed meal was incorporated at 10,20 and 30%, respectively. Sixty fingerlings of *Sparus aurata* with an initial body weight of 5.45 \pm 0.05g were distributed at random in twelve fiber glass tanks (each of 1m³). The fish were fed 3% of their total body weight and the diets were offered at two times/day. Growth performance of fish showed a significance differences ($P < 0.05$) between treatments. The highest performance was recorded with the fish fed 20% lupin seed meal (L20), followed with lesser extent by the control and L10 diets, respectively. However, the least performance was recorded with 30% incorporation level of lupin seed meal (L30). The carcass composition of fish were not affected by the incorporation level of lupin. The digestibility coefficients of the experimental diets showed a good utilization for protein in all tested diets. However, the carbohydrate digestibility coefficients recorded inferior results. On the other hand, the blood characteristics of fish (Hematocrite, Hemoglobin and plasma protein content) not differently by lupin seed level in the diets. The results of the present trial concluded that, the lupin seed meal can be utilized at level of 20% in feeding of *Sparus aurata*, without adversely effects on growth performance, digestibility coefficient and blood characteristics.

Keywords: Fish- Growth performance- Carcass composition- Amino acid- Fatty acid- Digestibility coefficient- *Lupinus albus* – *Sparus aurata*.

INTRODUCTION

Replacement of fish meal with protein rich plant ingredients in fish diets has been the objectives of numerous recent nutritional studies. However, fish meal and plant protein sources differ in a number of ways including protein quantity, amino acid profile, energy density and mineral content. Plant proteins also contain one or more of anti-nutritional factors (ANFs), which may have adverse effects on both nutritional value and palatability (Kaushik, 1989)

The supply of different protein sources (meat meal, fish meal and particularly soybean) was revised. Among these, cultivated lupin- species (*Lupinus L.*) have good potential due to their high protein concentration (30-40%), promising oil content (6- 12%), and their similarity in composition to that of soybean. So lupins could substitute for soybean. Moreover, it can be grown in different soils, different environmental conditions and under conditions not suitable for soybean (Gladstones, 1970).

Lupins as grain legumes, are attracting attention throughout the world as a potential providers of high quality protein and fat for future (Lopez Bellido and Fuentes, 1986). A factor which could limit the acceptance of lupin seeds for consumption is undoubtedly the presence of toxic alkaloids. The alkaloids concentration may, however, be reduced by subjecting the seed to heat treatment and rinsing with water. The oil content of lupin is too low to warrant extraction, but it makes a valuable contribution to the metabolisable energy value of the seed (Hansen and Zochanska, 1974). Lupin seeds appear to be free of the major anti-nutritional factors, trypsin inhibitors and haemagglutinins (Hill, 1977 and Hudson, 1979) and are fed without heat processing. Newer, "sweet" varieties are virtually free of alkaloids (< 0. 1g/kg, Batterham *et al.*, 1986).

Several researches investigated the use of lupin seed meal in fish feeding. Robaina *et al.*, (1995) have shown that lupin meal was a suitable ingredient in diets for gilthead sea bream. Workers with rainbow trout De la Higuera *et al.*, 1988 and Hughes, 1988 & 1993) have reported that incorporation of between (30 and 40%) dietary lupin meal did not affect on growth performance. Yet De la Higuera *et al.* (1988) noted a decrease in feed intake at 30% lupin incorporation. In fact, the palatability of lupin may be affected because it contains quinalizadine alkaloids (Hill and Pastuszewska, 1993). However, over the past decade, plant breeders have developed some strains of white lupin (*Lupinus albus*) with lower content of alkaloids (Roemer, 1983). Also, an advantage of lupin is that not require heat treatment as shown by De la Higuera *et al.* (1988) and Hassanen (1998), because it does not contain haemagglutinins (lectins) or trypsin inhibitors.

The present study was carried out to determine the nutritional value of extruded lupin (*L. albus*) in feeding of gilthead sea bream (*Sparas aurata*), besides, growth performance nutrient utilization, apparent digestibility coefficient and blood characteristics of sea bream (*Sparus aurata*).

MATERIALS AND METHODS

Fish and experimental condition

This study was conducted using the research facilities of the experimental station at Shakshouk, Fayaum Governorate, south west of Egypt. Gilthead sea bream (*Sparas aurata*) fingerlings were obtained from the Mediterranean sea coast at Damietta in the northern east of Egypt. They were transported in convenient tanks with aeration system to the experimental station.

The experimental fish were randomly distributed and stocked at density of 60 fish with an initial average weight of 5.45 ± 0.05 g per fiber glass tanks (1m^3), containing filtered sea water and provided with aeration. The water was delivered at a rate of 300 L/tank/hr.

The system contained two water pumps and upstream sandy filter units at a point between the water source and tanks. Each pump drew the water from Qarun lake and forced it through the body of the tanks and storage units. The fish were fed the experimental diets for two weeks as an

acclimatization period. The experimental period lasted 120 days (from May to August 2002). Physicochemical characteristics of water tanks were examined every week according to APHA (1992).

Diets and digestibility measurements.

Lupin albus L. Was prepared to be used as a plant ingredient for diets of *Sparus aurata*. They provided by Agriculture Research Center. After crushing, seeds were separated from its hulls. The rough flours obtained was extruded by extruding system had the following characteristics: temperature 127°C, pressure of 38 bar and specific energy of 89.8 w/kg. The chemical composition of lupin for protein and amino acids content is presented in Table (1). While, the fatty acid profiles were shown in table (2).

Table (1): Chemical composition and essential amino acids of lupin seed meal (*Lupinus albus*) and soybean meal (*Glycine max*) (% dry matter basis)

Chemical composition:	Lupin seed meal	Soybean meal
Dry matter	92.8	91.4
Crude protein	40.0	49.7
Crude fat	10.0	1.4
Nitrogen free extract	32.6	34.5
Crude Fiber	12.8	5.1
Ash	4.6	9.3
Total alkaloid	0.22	-
Essential amino acids:		
Arginine	5.12	3.67
Histidine	1.34	1.22
Isoleucine	1.64	2.14
Leucine	3.12	3.63
Lysine	1.82	3.08
Methionine	0.34	0.68
Cystine	0.69	0.75
Phenylalanine	1.51	2.44
Tyrosine	2.12	1.78
Tryptophan	0.43	0.69
Valine	1.82	2.55

Table (2). Fatty acids composition of % of total seed mixture.

Saturated fatty acids:	Lupin seed meal	Soybean meal
Palmitic acid (C16: o)	7.18	0.98
Stearic acid (C18: o)	2.11	3.88
Arachidonic acid (C20:o)	0.7	0.25
Behenic acid (C 22:o)	4.38	1.9
Unsaturated fatty acids:		
Oleic acid (C18:1)	48.78	36.2
Linolic acid (C18:2)	19.91	36.3
Linolenic acid (C18:3)	12.64	49.7
Erucic acid (C 22: 1)	2.23	0.2

Four isonitrogenous diets were formulated to containing average crude protein of 40.68% \pm 0.12. Each diet had three replacets. The diets were given at 3% of body weight (BW) per day at two times day (at 10.00 and 16.00 h) in two equal portions. The diets formulation is presented in Table (3). The first diet without extruded lupin was considered as a control diet (C). However diets 2 (L,10),3 (L20) and 4 (L30) containing extruded lupin with an incorporation levels of 10, 20 and 30%, respectively. All the diets were made into dry sinking pellet, using California pelleting machine with 3mm diameter.

During the last week of the experiment, fishes were fed on the experimental diets after adding 0.5% chromic oxide to study the apparent digestibility coefficients (ADC) of nutrients. During the collection period (10 days) for each diet, fecal samples were collected using the filtration system developed by Choubert *et al.*, (1982) and frozen daily. After freeze-drying the Feces were analyzed. The ADC values of nutrients were calculated using the formulation of Maynard and Loosli, (1969).

Chemical analysis of ingredients, feces and carcass composition.

Analysis of ingredients, diets, freeze dried feces and carcass samples were analyzed according to AOAC (1995). Chromic oxide in diets and feces was determined according to Bolin *et al.*, (1952). For amino acids analysis, the ingredients and diets were hydrolyzed with 6 N HCl at 110 c° for 24h for the chromatographic separation and analysis of the amino acids using a high performance liquid chromatograph (HPLC) as described by Gardner and Miller (1980). Tryptophan was determined calorimetrically in alkaline hydrolysate according the method described by Blauth *et al.* (1963).

For fatty acids, total lipids were extracted according to Folech *et al.* (1957). Fatty acid methyl esters were prepared by acid - catalyzed transmethylation of total lipids (Shantha and Ackman, 1990). They were analyzed in a varian 3400 gas chromatograph (equipped with a DB. Wax fused capillary colum (30 mx 0.25 mm i.d. film thickness, 0.25 Mm, Jw, USA) using helium as carrier gas (1.2 ml / min) and thermal gradient from 180 to 240 °c/ min. injector and flam ionization detector temperature were 260 and 250 °c, respectively. Data were recorded on a spectra physics 4270 integrator. Identification of individual fatty acid was made by comparison with known standard mixture).

Total alkaloids of lupins were determined gravimetrically according to the methods described by Priddis (1983), the dominant alkaloids in extracts obtained from defated mixture of the particular varieties were identified by thin layer chromatography. Alkaloid extracts were separated on silica gel plates (Merck) using chloroform: cyclohexane: diethylamine (6: 4: 1) as the solvent system. The alkaloid standards used were Lupanine perchlorate isolupanine and 13- hydroxylupanine (Koch- Light ltd Colnbrook, UK). The concentrated alkaloids were dissolved in 1ml of methanol Merck silica gel plates (60F- 25420 \times 20) and cyclohexane: diethylamine (1:10) were used.

The gross energy content of the diets was calculated as 5.5, 9.19 and 4.1 Kcal/ g of protein, lipid and carbohydrate, respectively according to Jobling (1981).

The metabolizable energy contents of the experimental diets were calculated as 4.5, 8.5 and 3.49 Kcal/g of protein, lipid and carbohydrate, respectively according to Jauncy (1982b).

Blood analysis

Blood samples were drawn from the caudal vein with heparinized syringes. Hematocrite percentage and hemoglobin concentration were determined according to Harding and Høglund (1983). Plasma protein, glucose, triglyceride and phospholipid were determined according to Shimeno *et al.* (1981).

Statistical analysis

The analysis of variance (ANOVA) was employed to test the effect of lupin incorporation on various growth parameters, digestibility coefficients and blood characteristics according to Snedecor and Cochran (1987). Duncan multiple range test was used to detect the significant differences between the means of treatments (Duncan, 1955).

RESULTS AND DISCUSSION

Results of the chemical composition of lupin seed meal (*L. albus*) compared with soybean meal (*Glycine max*) are shown in Table (1). The Lupin showed a good protein content (40.0%) and high content of fiber (12.8%) compared with soybean meal. This strain of lupin also showed a lower content of total alkaloid (0.22%). Similar values of amino acids were found in the two crops, with few exception.

As presented in Table (2), the fatty acid contents in *Lupinus albus* are comparable to that of soybean, except that the Oleic acid and Erucic acid were higher in *Lupinus albus*. However, the linolic and linolenic acids showed higher values in soybean compared with *L. albus*.

Data presented in Table (3) showed that the crude protein in the experimental diets ranged from 40.5 to 40.8% and these values within the recommended values (40%) of this fish species (Sabour and Luquet, 1973).

The amino acid contents of the experimental diets were presented in Table (4). All diets having the essential amino acids required for rearing *Sparus aurata* as recorded by Jauncy (1988).

In Table (5), the physicochemical characteristics of tanks water showed good values for temperature, PH, dissolved oxygen, salinity and unozied ammonia. The recorded values are within the optimum ranges for rearing *Sparus aurata*. According to Porter *et al.* (1986).

As given in Table (6), the growth performance of fish fed the experimental diets showed significant differences ($P < 0.05$) among diets. The highest final body weight and gain were obtained with fish received diet L20, which contain 20% extruded lupin seed meal as an incorporation level followed (at a decreasing order) by the control diet and that contain 10% extruded lupin seed meal. However, the incorporation of 30% lupin meal showed less final body weight and gain in weight compared with the other

diets. The highest growth rate (GR) and specific growth rate (SGR), were recorded with diet L20, followed by the control and L10, respectively without significance difference among them. The (GR) rate and (SGR) were obtained with 30% incorporation of lupin (L30).

Table (3). Formulation and chemical composition of the experimental diets.

Ingredients %	Control	L10%	L20%	L30%
Fish meal	40	30	20	15
Lupin seed meal	-	10	20	30
Poultry -by-product meal	15	20	25	25
Gluten meal	10	10	10	10
Maize	20	15	10	5
Wheat bran	10	10	10	10
Fish oil	3	3	3	3
Vitamin/ Mineral Mix ¹	2	2	2	2
Chemical composition (%DM basis)				
Dry matter	92.5	91.4	29.0	92.2
Crude protein	40.8	40.65	40.5	40.8
Ether extract	15.39	15.2	16.24	15.4
Nitrogen free ext.	25.71	26.79	25.89	25.2
Fiber	4.78	5.07	6.09	8.51
Ash	13.32	12.29	11.28	10.09
Calcium	1.5	1.40	1.35	1.30
Phosphorus	0.8	0.86	0.75	0.70
Gross energy kcal/kg diets	4098.1	4717.2	4766.7	4678.6
ME Kcal/Kg diet	4041.3	4053.3	4106.4	4024.4
P/E ratio, mg Kcal	115.14	116.04	117.69	114.67

1- Vitamin mineral premix, each Kg contain : vitamin A, 2500 IU, Vitamin C, 6.0g (ascorbate polyphosphate), Vitamin D3, 2400 IU, α tocopheryl acetate 0.2 g , thiamin, 0.01g (thiamin cl), riboflavin 0.02g , pyridoxine 0.01g, (pyridoxine Cl), Ca- pantothenate 0.04g, niacin 15g, folic acid, 5mg vitamin k. 0.04g, Mn,35mg, (MnSO₄), Zn, 90mg (Zn₂SO₄), cu, 12mg (CuSO₄), I, 2mg (KI), Se,0.2mg (Na₂SeO₃), Cl,1.25g KCl). The ingredients and vitamin mineral premix were obtained from Zoocontrol company, 6 October Giza - Egypt.

Table (4). Essential amino acid composition of the experimental diets (g/100g diet).

Amino acid	Diets				
	Requirement*	Control(C)	L10	L20	L30
Arginine	2.11	3.03	3.14	3.31	3.54
Histidine	0.89	1.2	0.91	0.97	1.03
Isoleucine	1.37	1.4	1.5	1.59	1.65
Leucine	2.5	3.16	3.30	3.44	3.53
Lysine	2.90	3.2	3.12	3.05	2.98
Methionine+ cyst	1.28	1.39	1.32	3.24	3.25
Phenylalanine + Tyrosine	2.25	3.37	3.25	3.24	3.25
Threonine	1.58	1.73	1.62	1.65	1.46
Tryptophane	0.29	0.54	0.52	0.49	0.46
Valine	1.63	1.84	1.93	1.94	1.73

* According to Jauncy (1988).

Table (5). Average values of the physicochemical characteristics of water in the experimental tanks throughout the experimental period.

Parameters	Diets			
	Control	L,10	L20	L30
Temperature °C	24.5	24.8	24.7	24.6
PH	7.6±0.2	7.5± 0.3	7.6±0.1	7.5±0.2
Dissolved oxygen (mg/l)	6.5±0.1	6.4±0.2	6.5±0.1	6.3±0.1
Salinity ‰	35.1 ±0.1	35.2 ±0.2	35.1 ±0.1	35.1 ±0.1
Unionized ammonia (mg/L)	0.034± 0.002	0.032±0.001	0.032±0.002	0.034±0.001

Table (6): Growth performance of *Sparus aurata* fed the experimental diets.

Parameters	Diets				SE±
	Control	L,10	L20	L30	
Initial avg. weight (g/fish)	5.5 ^a	5.4 ^a	5.4 ^a	5.5 ^a	0.05
Final avg. weight (g/fish)	57.4 ^{ab}	55.8 ^b	60.2 ^a	45.8 ^c	2.33
Gain (g/fish)	51.9 ^{ab}	50.4 ^b	54.8 ^a	40.3 ^c	0.06
Feed consumed (g/ fish)	92.0	90.0	85.0	80.0	-
Growth rate (g/day)	0.43 ^a	0.42 ^a	0.45 ^a	0.33 ^b	0.04
Specific growth rate ¹	1.95 ^a	1.95 ^a	2.0 ^a	1.76 ^b	0.09
Feed conversion ²	1.77 ^a	1.78 ^a	1.55 ^a	1.98 ^b	0.15
Feed efficiency ³	0.56 ^a	0.56 ^a	0.64 ^a	0.50 ^b	0.04
Protein efficiency ratio ⁴	1.38 ^a	1.37 ^a	1.59 ^a	1.23 ^b	0.12

Means in the same row with different superscript letters are significantly different (P<0.05). S.E. standard error of the mean.

- 1- Specific growth rate = 100 X (Ln final weight- Ln initial weight)/ 120.
- 2- Feed conversion= (feed given per fish)/ (weight gain per fish).
- 3- Feed efficiency= (weight gain per fish)/ (feed given per fish).
- 4- Protein efficiency ratio = (weight gain per fish)/ (protein intake).

As can be seen from Table (6), the feed intake was similar in the control and L10 diets. However, L20 and L30 showed a decrease in feed intake. The slight tendency toward a lower feed intake as dietary lupin meal content increased could be due to a progressively more difficult adaptation of fish to the organoleptic properties of such diets. The same finding was recorded in salmon trout by De la Higuera *et al.*, (1988). The decrease in feed intake of fish diets was increased where lupin level increased, probably due to the high content of fiber (L30) in the present study, the lupin used was a sweet variety with a lower content of alkaloids and it was extruded. The beneficial effects of extrusion treatment of raw materials on their digestive utilization has already been reported (Kaushik, 1989 and Bangaula *et al.*, 1993).

As presented in Table (6), the feed conversion, feed efficiency and protein efficiency ratio showed significant differences among treatments (p< 0.05). The best results were found with the fish fed at 20% incorporation of extruded lupin seed meal (L20). On the other hand, the least efficiency was recorded with 30% incorporation level (L30). These results were in agreement with the results of Gomes and Kaushik (1989) and Bangaula *et al.* (1993).

Date of the apparent digestibility coefficient (ADC) of nutrients are showed in Table (7). The results showed no significant differences due to the incorporation of extruded Lupin up to 20%. However, the incorporation of 30% extruded lupin gave less digestibility coefficients than that of the control, L10 and L20, respectively concerning dry matter, protein and fat digestibilities. These results are in agreement with the results of Burel *et al.*, (1998) in rainbow trout and Hassanen (1998) in *Sparus aurata*. As can be seen from Table (7), the results indicated good utilization of all extruded lupin tested, except that L30 level and the values were similar to those presented in the literature for fish meal and soybean meal (Pffeffer, 1982). The results of ADC (protein, lipid and carbohydrate) in the present trial were in agreement with the results of Yones (1989) in *Sparus aurata*. However, the less digestibility of carbohydrate in lupin diets may be a result of insufficient amylase enzymes in the intestine of *Sparus aurata* to metabolize dietary carbohydrate. Similar finding was recorded in salmon by Saponnhof and Plantikov (1983).

Difference in carcass composition, HSI and VSI between the control and the three incorporation levels were not statistically significant (Table 8). These results are in agreement with the results recorded for *Sparus aurata* by Pereira and Oliva- Teles.(2002).

Table (7). Apparent digestibility coefficients of the experimental diets.

Nutrients	Diets				SE±
	Control	L,10	L20	L30	
Dry mater	85.3 ^a	84.5 ^a	85.6 ^a	80.0 ^b	2.25
Protein	94.0 ^a	93.5 ^a	94.5 ^a	90.0 ^b	1.76
Fat	93.0 ^a	92.5 ^a	93.2 ^a	89.0 ^a	1.7
Carbohydrate	76.0 ^a	71.0 ^c	72.0 ^b	68.0 ^d	2.86

Means in the same row with different superscript letters are significantly different (P<0.05)

Table (8). Final body composition, HSI and VSI for *Sparus aurata* fed the experimental diets (%dry mater basis).

Items	Initial	Diets			
		Control	L,10	L20	L30
Drymatter	23.0	24.5 ^a	25.0 ^a	25.2 ^a	25.4 ^a
Protein	75.0	74.0 ^a	73.8 ^a	73.5 ^a	73.2 ^a
Lipid	17.0	18.5 ^a	18.8 ^a	19.8 ^a	20.0 ^a
HSI ¹	108	2.4 ^a	2.2 ^a	5.5 ^a	2.8 ^a
VSI ²	2.1	4.3 ^a	4.5 ^a	4.8 ^a	4.6 ^a

Means in the same raw with different super script letters are significantly different (P<0.05).

1- Hepatosomatic index = (liver weight/ body weight) × 100.

2- Viscerasomatic index= (viscera weight/ body weight) × 100.

The results of the present trial suggest a maximum level of 20% incorporation from extruded lupin seed meal can be used in *Sparus aurata* diets. These results were in agreement with the results of Robaina *et al.*, (1995) in the same species and within the maximum range (20- 40%) in trout diet which reported in previous studies (De la Higuera *et al.*, 1988, Hughes,

1988, Gomes and Kaushik, 1989, Moyano *et al.*, 1992, Bangaula *et al.*, 1993 and Gauveia *et al.*, 1993).

As presented in (Table, 9), the blood contents were not affected by the incorporation levels. Insignificant results were found among treatments. Similar results were recorded in *Sparus aurata* by Hassanen *et al.*, (1992), *Diplodus argenteus* (Filho *et al.*, 1992), coho salmon (*Oncorhynchus kisutch*) Higgs *et al.* (1978) and trout (*Salmo gairdneri*) Alexis *et al.*, (1985). In the same trend, the plasma contents showed a significant differences among treatments. These results were in agreement with the results recorded in tilapia (*Oreochromis niloticus*) by Shemino *et al.* (1993).

Table (9).Hematocrite, hemoglobin and some plasme contents of *Sparus aurtata* fed the experimental diets.

Parameter	Diets			
	Control	L,10	L20	L30
Hematocrite(%)	41.0 ^a	41.4 ^a	41.8 ^a	41.0 ^a
Hemoglobin (g/100ml)	7.5 ^a	7.2 ^a	7.3 ^a	7.1 ^a
Protein (g/100ml)	4.1 ^a	4.3 ^a	4.2 ^a	4.0 ^a
Glucose (mg/100ml)	55.5 ^a	45.8 ^a	55.2 ^a	54.7 ^a
Trigly ceride (mg/100ml)	242.0 ^a	243.0 ^a	242.5 ^a	241.6 ^a
Phospholipid(mg/100ml)	576.0 ^a	576.2 ^a	575.5 ^a	574.5 ^a

Means in the same row with different superscript letters are significantly different (P<0.05).

In conclusion: the results of the present trial indicated that the partial replacement of fish meal by extruded lupin seed meal is very interesting in terms of growth performance. The main finding of this study is that extruded lupin seed meal up to 20% could be used, without adversely effect on growth performance, digestibility coefficient and blood characteristics of *Sparus aurata*.

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إدخال مسحوق الترمس كمصدر للبروتين النباتي في علائق أسماك الدنيس.

عبد المنعم عبد الصادق مهدي يونس

المعهد القومي لعلوم البحار والمصايد - محطة بحوث الأسماك بشكشوك - الفيوم - مصر.

تم استخدام مسحوق الترمس كمصدر للبروتين النباتي ليحل جزئيا محل مسحوق السمك في تغذية أسماك الدنيس حيث تم تكوين ٤ علائق تحتوى علي بروتين خام (٤٠,٦٨ ± ٠,١٢%) حيث كانت العليقة رقم ١ هي عليقة المقارنة والتي لا تحتوى علي مسحوق الترمس بينما العلائق L30, L20, L10 تم استخدام مسحوق الترمس فيها بنسب ١٠, ٢٠, ٣٠% علي التوالي. استخدمت ٦٠ أصبعية من أصبقيات الدنيس ذات الوزن الأولي (٥,٤٥ ± ٠,٠٥ جم) حيث وزعت عشوائيا علي أحواض من الفيبرجلاس سعة الحوض ٣م^٣ وتم تغذية الأسماك بمعدل ٣% من وزن الجسم قدمت علي مرتين يوميا. أظهرت معدلات أداء النمو نتائج معنوية علي مستوى (٠,٠٥%) بين المعاملات حيث أعطت العليقة المحتوية علي ٢٠% مسحوق ترمس أعلى أداء، تلتها وبتدرج أقل عليقة المقارنة والعليقة المحتوية علي ١٠% ترمس في حين أعطت العليقة المحتوية علي ٣٠% معدلات أداء منخفضة. لم يتأثر تركيب جسم الأسماك بمستوى مسحوق الترمس المستخدم في العليقة وكذلك صفات الدم من الهيماتوكريت، الهيموجلوبين ومحتوى البلازما. وأظهرت معاملات الهضم للعلائق استعادة جيدة لمسحوق الترمس المستخدم في العلائق وخاصة معاملات الهضم للبروتين و الدهون، في حين انخفضت معاملات الهضم للكربوهيدرات. خلصت نتائج الدراسة إلي أنه يمكن استخدام مسحوق الترمس حتى مستوى ٢٠% من العليقة بدون تأثيرات معاكسة علي أداء الأسماك، صفات الدم ومعاملات الهضم بالنسبة لأسماك الدنيس.