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Dietary Lipid Requirement for the Kutum Fingerlings, *Rutilus frisii kutum* (Kamenskii 1901)

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Abstract: A growth trial was conducted to investigate the effects of dietary lipid levels on growth, feed utilization, condition factors, survival rate and carcass composition and to evaluate the lipid requirement of kutum fingerlings, *Rutilus frisii kutum*. Five isonitrogenous experimental diets were formulated to contain graded levels of lipid (50, 80, 120, 160 and 190 g kg^{-1}). After 2 weeks of the conditioning period, triplicate groups of 80 kutum (initial average weight of 0.50±0.07 g, mean±SD) were stocked in 250-l tanks and fed to apparent satiation thrice a day for 8 weeks. After feeding trial, the results showed that growth performance, feed efficiency and whole-body composition were significantly affected by dietary treatment. Growth performance generally improved with increasing dietary lipid from 50-120 g kg^{-1} lipid diets and slightly decreased thereafter with further increase in dietary lipid. Food conversion ratio was negatively correlated with the dietary lipid (R^2 = 0.897). Protein retention efficiency and energy retention efficiency, generally increased with increasing dietary lipid level but lipid retention efficiency were inversely correlated with dietary lipid concentration ($p<0.05$). The carcass composition of kutum was affected by dietary treatments except for ash and protein contents. The carcass lipid of fish in this study was obviously increased corresponding to dietary lipid levels. The optimal dietary lipid level, estimated by polynomial regression analysis for maximum growth of kutum fingerling was 13.55%.

Key words: Kutum *Rutilus frisii kutum*, lipid requirement, growth, food intake, body composition

INTRODUCTION

Dietary lipid is an important nutrient element for fish nutrition which supplies a source of energy and essential fatty acid (Sargent *et al*., 1989) and is also transporters of fat-soluble vitamins. Extreme lipid or carbohydrate (non-protein) energy sources in diet may have adverse influences on growth, reduce feed intake and inhibit the utilization of other nutrients and lead to increased fat deposition (Garling and Wilson, 1976; Winfree and Stickney, 1981). Use of appropriate dietary lipid level is prevented to employ the dietary protein for energy generation, resulting in higher specific dynamic action and will lead to a decrease in ammonia excretion and the feed costs (LeGrow and Beamish, 1986; Catacutan and Coloso, 1995). Protein sparing effect implies that inclusion of lipid and carbohydrate (non-protein) energy sources has been displayed to lower dietary protein employed for energy and elevate protein utilization for growth (El-Sayed and Teshima, 1992). Dietary lipid has been shown to provide a protein sparing effect in some fish (Santinha *et al*., 1999). The effects of dietary lipid level on growth rate, flesh quality, immune response, disease resistance and reproduction performance have been investigated in several species important in aquaculture (Luquet and Watanabe, 1986; Bromage *et al*., 1992; Regost *et al*., 2001; Chaiyapechara *et al*., 2003).

Fishing in Caspian sea concentrates mainly on two fish species: Kutum and Common carp, both being appropriate growth rate. But kutum, *Rutilus frisii kutum* (Kamenskii 1901) is a very popular food fish because of its good meat quality and good consumer acceptance especially in north of Iran. In recent decades, the natural reproduction of kutum has been limited and its artificial reproduction is successfully done in hatcheries (Abdoli, 1990).

In the restocking centers, larval rearing takes place in earth ponds and kutum larvae feed on zooplankton and formulated diet. The formulated diets are used in the form of paste and contain 28-35% protein and 8-10% lipid, based on the requirements for common carp (*C. carpio* L.) because nutritional requirements of this species are still unknown. Although, several studies have been

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conducted to appraise the nutritional parameters of kutum (Neverian *et al*., 2008; Neverian and Shabanipour, 2008), there is no accurate information on the dietary lipid requirement of kutum. Therefore, the present study was designed to evaluate the effect of lipid levels in a practical diet on growth rate, feed utilization and body composition of kutum figerlings, *Rutilus frisii kutum* (Kamenskii 1901).

MATERIALS AND METHODS

Experimental fish and rearing conditions: Kutum (*Rutilus frisii kutum*) fingerlings were obtained from Shahid Rajaee restocking center located in the southern coast of Caspian sea, Sari, Iran. After 2 week's acclimation, kutum fingerlings with initial body weight of 0.5±0.09 g were randomly distributed into 250-l fiberglass tanks with 80 fish per tank. Each dietary treatment had 3 replications.

Water quality parameters were checked periodically, pH was found between 7.8-8.0; water temperature ranged from 19.7-23.2°C and dissolved oxygen was not <7.2 mg L^{-1}. Fish were hand-fed with the experimental diets until apparent satiation 3 times daily (7 days a week).

Diet preparation: Five isonitrogenous diets with 42% Crude Protein (CP) were formulated to provide five dietary lipid levels at 50, 80, 120, 160 and 190 g kg^{-1} diet (dry basis). Gross energy content of each diet was calculated based on 23.4, 39.2 and 17.2 MJ kg^{-1} for protein, lipid and carbohydrate, respectively. Feedstuffs were homogenized through a 1.0 mm sieve, blended in the computed ratios, moistened with distilled water (25% v/w) and pelleted in a grinder. The formulation and proximate analysis of experimental diets are shown in Table 1.

Table 1: Formulation and proximate composition of diets used in experiment

	Dietary lipid level (g kg^{-1} diet)				
Ingredient (g)	50	80	120	160	190
Fish meal[a]	540.00	540.00	540.00	540.00	540.00
Wheat flour	300.00	300.00	300.00	300.00	285.00
Fish oil[b]	0.00	7.00	34.00	60.00	70.00
Soybean oil	7.50	25.50	38.50	52.50	64.00
Mineral mixture[c]	20.00	20.00	20.00	20.00	20.00
Vitamin mixture[d]	20.00	20.00	20.00	20.00	20.00
Cellulose[e]	112.50	87.50	47.50	7.50	1.00
Total	1000.00	1000.00	1000.00	1000.00	1000.00
Proximate composition (g kg^{-1} diet)					
Moisture	92.30	89.00	96.80	105.60	104.80
Crude protein	419.10	417.20	414.10	413.20	415.40
Lipid	53.70	88.10	119.30	168.90	188.20
Ash	94.80	93.90	94.80	102.80	96.70
NFE[f]	340.10	311.80	275.00	209.50	194.90
Gross energy (MJ kg^{-1})[g]	17.76	18.57	19.09	19.75	20.45
P/E ratio[h]	23.59	22.45	21.68	21.22	20.31

[a]Pars kelika Co., Mirood, Iran; [b]Herring oil; [c]Vitamin premix contained the following vitamins (each kg^{-1} diet): vitamin A, 10 000 IU; vitamin B_6 20 mg; vitamin B_{12}, 0.04 mg; biotin, 0.2 mg; choline chloride, 1200 mg; folic acid, 10 mg; inositol, 200 mg; niacin, 200 mg; pantothenic calcium, 100 mg. [d]Contained (g kg^{-1} mix): $MgSO_4.2H_20$ 127.5; KCl, 50.0; NaCl, 60.0; $CaHPO_4.2H_2O$, 727.8; Ca $(IO_3)_2.6H_2O$, 0.295; $CrCl_3.6H_2O$, 0.128. [e]Sigma, St. Louis, MO, USA.. [f]Nitrogen-Free Extract (NFE) = 1000-(protein+lipid+moisture+ash). [g]Based on 23.4 kJ g^{-1} protein, 39.2 kJ g^{-1} lipid and 17.2 kJ g^{-1} NFE. [h]Protein/Energy ratio (g MJ^{-1})

Sample fish and chemical analysis: At the end of the experiment, 20 fish from each tank were randomly sampled and euthanized by overdose of ethylene glycol monophenyl ether and kept frozen (-20°C) for determination of final whole body composition. The analysis of diets and fish samples followed standard AOAC (1984). Crude protein content (N×6.25) was determined according to the Kjeldahl method after acid digestion using an Auto Kjeldahl System (1030-Auto-analyzer, Tecator, Hoganos, Sweden). Crude lipid was determined by etherextraction using a Soxtec extraction System HT (Soxtec SystemHT6, Tecator, Sweden). Ash was determined by muffle furnace at 550°C for 24 h. Moisture was determined by oven-drying at 105°C for 24 h.

Calculations: The performance parameters, body composition and survival percentage were calculated as below:

$$\text{Weight gain} = [(W_F - W_I)/W_I]\times 100$$

$$\text{Specific growth rate} = [(\text{Ln } W_F - \text{Ln } W_I)/T]\times 100$$

$$\text{Feed conversion ratio} = \left[\frac{\text{Total feed intake (g)}}{\text{Total wet weight gain (g)}}\right]$$

$$\text{Protein efficiency ratio} = \left[\frac{\text{Wet weight gain (g)}}{\text{Total protein intake (g)}}\right]$$

$$\text{Feed efficiency ratio} = \left[\frac{\text{Wet weight gain (g)}}{\text{Dry feed fed (g)}}\right]$$

$$\text{Feed intake} = \left[\frac{\text{Total feed intake}}{\text{Number of fish}}\right]$$

$$\text{Protein retention efficiency} = \left[\frac{\text{(Final body protein-initial body protein)}}{\text{Total dietary protein fed}}\right]\times 100$$

$$\text{Condition factor} = \left[\frac{\text{Fish Wt (g)}}{\text{Fish length (cm)}^3}\right]\times 100$$

$$\text{Survival (\%)} = \left[\frac{\text{Final number of surviving fish}}{\text{Initial number of fish}}\right]\times 100$$

where, W_F and W_I are the mean final body weight and the mean initial body weight, T is the experimental duration in days.

Statistics: The data were subjected to one-way ANOVA and correlation analysis where appropriate. Multiple comparisons among means between individual treatments were made with Duncan's multiple range tests. The significance level was at $p<0.05$. All statistical analyses were performed using SPSS Base 16 for Window.

RESULTS AND DISCUSSION

The survival rates were generally high from 95.00-99.58% over the 8 weeks trial and it was affected by the differing dietary lipid level of the diets ($p<0.05$). The lowest percent survival was observed for fish fed the diet containing 50 g kg^{-1} lipid diets. Growth rate and feed efficiency were significantly affected by dietary lipid content of the diets (Table 2). Growth performance (final body weight and SGR) generally increased with increasing dietary lipid from 50-120 g kg^{-1} lipid diets. The highest growth performance ($p<0.05$) was achieved in kutum fed the diet containing 120 g kg^{-1} lipid. The FCR of the kutum was significantly affected by dietary lipid level ($p<0.05$). FCR was negatively correlated with the dietary lipid ($R^2 = 0.897$). The high levels of dietary lipid seemed to have significantly effect on FCR ($p<0.05$). When the dietary lipid level was higher than 120 g kg^{-1} lipid diets, this nutrient no showed significant effect on FCR ($p>0.05$). Protein efficiency and feed efficiency were improved as increasing of dietary lipid level. FER of fish fed on dietary lipid levels 120, 160 and 190 g kg^{-1} lipid was not different ($p>0.05$). FI and PI decreased significantly ($p<0.05$) with increasing dietary lipid level and lipid intake tended to decrease with the decline of dietary lipid level. Kutum fed the diet with 190 g kg^{-1} lipid had the highest PRE among the dietary treatments. However, the lowest values for both parameters were recorded in fish fed the diet with 50 g kg^{-1} lipid.

At the end of the experiment, moisture, lipid and energy contents in body composition of fish were significantly affected by dietary lipid levels ($p>0.05$) while crude protein in carcass was not affected by dietary treatments ($p<0.05$) (Table 3). Crude lipid and energy contents in carcass of kutum had increased, when dietary lipid was increased from 50 g kg^{-1} lipid diets to 190 g kg^{-1} lipid diets but moisture contents in carcass tended to decrease with the increase of dietary lipid level and same trend was observed for ash contents, meanwhile it was not significantly affected by dietary lipid level ($p<0.05$).

Dietary lipids are important nutrient affecting energy production in most of fish. They are well utilized by fish and essential for common growth and development.

Table 2: Growth response and feed efficiency of kutum fed the diets containing different lipid levels for 8 weeks

	Dietary lipid level				
Treatment	50	80	120	160	190
Initial weight (g)	0. 50±0.040	0.50±0.0900	0.49±0.1300	0.49±0.1400	0.50±0.1000
Weight gain (g)	1.26±0.0700^{ab}	1.16±0.1100^{a}	1.38±0.0700^{b}	1.22±0.0900^{ab}	1.67±0.1200^{d}
Weight gain (%)	158.3±15.6000^{a}	231.50±22.000^{b}	276.00±14.000^{c}	247.70±19.000^{bc}	238.70±24.000^{bc}
FCR	3.50±0.2200^{c}	2.33±0.2000^{b}	1.85±0.1000^{a}	1.85±0.1300^{a}	1.77±0.1100^{a}
SGR	2.34±0.1400^{ab}	2.30±0.1200^{a}	2.54±0.0700^{b}	2.39±0.1100^{ab}	2.39±0.1100^{ab}
PER	0.77±0.0700^{a}	1.02±0.0900^{b}	1.28±0.0700^{c}	1.29±0.0900^{c}	1.17±0.1200^{bc}
FER	0.28±0.0200^{a}	0.42±0.0400^{b}	0.54±0.0200^{c}	0.54±0.0300^{c}	0.56±0.0300^{c}
FI	2.70±0.1000^{d}	2.71±0.0100^{d}	2.55±0.0100^{c}	2.25±0.0400^{b}	2.07±0.0800^{a}
PI	1.135±0.449^{d}	1.138±0.004^{d}	1.071±0.004^{c}	0.945±0.019^{b}	0.872±0.036^{a}
PRE	22.46±2.2900^{a}	25.70±2.0600^{a}	27.95±5.6100^{ab}	27.13±5.8200^{a}	36.33±6.4100^{b}
EI	48.01±1.8900^{c}	55.42±0.2100^{e}	50.74±0.1900^{d}	42.97±0.8700^{b}	38.56±1.6200^{a}
Survival (%)	96.60±1.9000^{ab}	99.50±0.7000^{c}	95.00±1.2000^{a}	97.90±1.4000^{bc}	96.20±1.2000^{ab}

Mean values and Standard Deviation (±SD) are presented for each parameter (Three groups per treatment). Means in the same line with different letters are significantly different ($p<0.05$). FCR: Food Conversion Ratio; SGR: Specific Growth Rate; FER: Feed Efficiency Ratio; PER: Protein Efficiency Ratio; FI: Feed Intake; PI: Protein Intake; PRE: Protein Retention Efficiency; EI: Energy Intake

Table 3: Carcass composition of kutum fed the diets containing different lipid levels

Treatment	Protein	lipid	Ash	Moisture	Energy
Initial values	12.3±0.520	8.4±0.11	2.8±0.100	76.2±0.10	642.0±15.4
Final values					
50	14.9±0.320^{a}	9.9±0.05^{a}	2.54±0.11^{a}	74.4±0.35^{c}	740.1±8.50^{a}
80	15.3±0.230^{a}	10.9±0.19^{b}	2.52±0.24^{a}	73.0±0.31^{b}	786.3±12.2^{b}
120	15.3±0.590^{a}	12.0±0.46^{c}	2.50±0.16^{a}	72.1±0.72^{b}	833.1±13.1^{c}
160	14.9±0.490^{a}	12.8±0.39^{d}	2.48±0.24^{a}	72.4±0.60^{b}	852.0±17.4^{cd}
190	15.5±0.410^{a}	12.9±0.25^{d}	2.40±0.14^{a}	69.9±0.86^{a}	871.7±19.2^{d}

Initial body composition: bodies of 50 fish (means±SD) were used for statistical analysis. Final body composition: values are means of three groups per treatment. Values in the same column with different superscripts are significantly different ($p<0.05$)

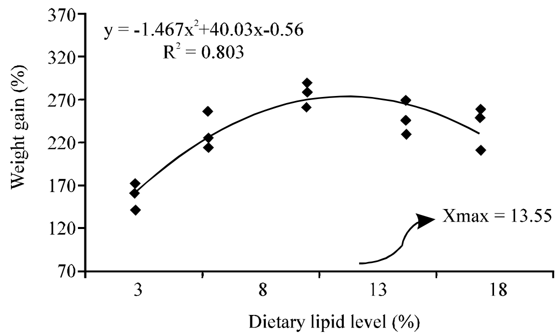

Fig. 1: Relationship between percent weight gain and dietary protein level for kutum fingerlings as described by second-polynomial regression

However, fish are known to utilize protein preferentially to lipid or carbohydrate as an energy source. The results of this trial based on feeding dietary lipid levels from 50-190 g kg^{-1} lipid diets indicate that survival rate varied 95.00-99.58%. The optimal dietary lipid level, estimated by polynomial regression analysis, for maximum growth of kutum fingerling was 13.55% (Fig. 1). The results showed that the increase in dietary lipid level from 50-120 g kg^{-1} lipid diets increased growth and slightly decreased thereafter with further increase in dietary lipid. In this study, the highest specific growth rate of kutum (500 mg initial weight) was 2.54% day^{-1} which is good score compared to other researches. For instance, Haghighi (2006) registered maximum SGR of 2.78% day^{-1} for kutum fry (200 mg initial weight) fed a diet containing 468 g kg^{-1} protein and 80 g kg^{-1} lipid.

Some fish especially carnivorous species (most marine fish and marine crustacean) can employ higher dietary lipid for optimum growth rate (Peres and Oliva-Teles, 1999; Lopez *et al*., 2006). On the other hand, some researchers have reported that high dietary lipid level may be depressed fish growth (El-Sayed and Garling, 1988). This reaction may be due to decrease in feed intake, excessive lipid deposition in visceral organs to decrease in the efficiency or activity of digestive enzymes (Tocher, 2003). Likewise, Baker and Davies (1997) reported that growth depression in high dietary lipid level could be due to increased malonaldehyde in oxidized lipid which is poisonous to fish. In the present study demonstrated that the increase in dietary lipid level was collaborated with a reduction in feed intake, probably because fish eat to satisfy their energy requirements.

Feed and protein efficiencies at the present study tended to improve with increasing level of dietary lipid. It implies that kutum fingerlings have a relatively good ability to utilize dietary lipids as energy sources. Appropriate quantities of lipid and/or carbohydrate sources (non-protein energy) in the fish diet can minimize catabolism of protein (Cho and Kaushik, 1990). Lipid as energy source has been shown to provide a protein sparing effect in some fish (Sargent *et al*., 1989; Skalli *et al*., 2004).

In the present study, the FCR of experimental diets varied 1.77-3.50. The lowest dietary lipid level (50 g kg^{-1} lipid diets) produced the highest FCR, probably due to the intake of insufficient nutrient levels to promote growth. The best FCR was observed in fish fed diet containing 190 g kg^{-1} lipid diets.

The carcass composition of kutum was affected by dietary treatments except for ash and protein contents (Table 3). This effect has previously been observed in other fish species and aquatic animals (Cho *et al*., 2001; Sa *et al*., 2006). Very high dietary lipid levels might result in excessive lipid aggregation in the visceral cavity, tissue and liver and might consequently increased the whole body lipid concentrations (Tocher, 2003). The carcass lipid of fish in this study was obviously increased corresponding to dietary lipid levels. A slight increase in body protein contents with the increase of dietary lipid level was also observed in kutum but increase in body protein content was not significantly different among the treatments.

CONCLUSION

In this study, whole body moisture was inversely related to whole body lipid content and a mutual relevance between lipid and moisture has been reported in many fish in general (Parazo, 1990). Based on the results of this study, the maximum growth of kutum fingerling was observed when fed the diet containing 120 g kg^{-1} lipid diets and the protein/energy ratio was approximately 21.68 g MJ^{-1}. From the polynomial regression analysis, the optimal dietary lipid requirement of kutum was estimated to be 13.55%.

REFERENCES

AOAC, 1984. Official Methods of Analysis. 12th Edn., AOAC, Washington DC, USA.

Abdoli, A., 1990. The Inland Water Fishes of Iran. Natural and Wild Life Museum of Iran, Tehran, pp: 198-200.

Baker, R.T.M. and S.J. Davies, 1997. Modulation of tissue α-tocopherol in African catfish, *Clarias gariepinus* (Burchell), fed oxidized oils and the compensatory effect of supplemental dietary vitamin E. Aquacult. Nutr., 3: 91-97.

Bromage, N., J. Jones, C. Randall, M. Thrush and B. Davies *et al*., 1992. Broodstock management fecundity, egg quality and the timing of egg production in the rainbow trout *Oncorhynchus mykiss*. Aquaculture, 100: 141-166.

Catacutan, M.R. and R.M. Coloso, 1995. Effect of dietary protein to energy ratios on growth, survival and body composition of juvenile Asian seabass, *Lates calcarifer*. Aquaculture, 131: 125-133.

Chaiyapechara, S., M.T. Casten, R.W. Hardy and F.M. Dong, 2003. Fish performance, fillet characteristics and health assessment index of rainbow trout (*Oncorhynchus mykiss*) fed diets containing adequate and high concentrations of lipid and vitamin E. Aquaculture, 219: 715-738.

Cho, C.Y. and S.J. Kaushik, 1990. Nutritional energetics in fish: Energy and protein utilization in rainbow trout (*Salmo gairdneri*). World Rev. Anim. Nutr., 61: 132-172.

Cho, H.S., J.Y. Jo and S.D. Kim, 2001. Effects of variable feed allowance with constant energy and ratio of energy to protein in a diet for constant protein input on the growth of common carp *Cyprinus carpio* L. Aquacult. Res., 32: 349-356.

El-Sayed, A.M. and D.L. Jr. Garling, 1988. Carbohydrate-to-lipid ratio in diets for *Tilapia zillii* fingerlings. Aquaculture, 73: 157-163.

El-Sayed, A.M. and S. Teshima, 1992. Protein and energy requirements of Nile tilapia, *Oreochromis niloticus*, fry. Aquaculture, 103: 55-63.

Garling, D.R. and R.P. Wilson, 1976. Optimum dietary protein to energy ratio for channel catfish fingerlings, *Ictalurus punctatus*. J. Nutr., 106: 1368-1375.

Haghighi, D.T., 2006. Embryonic development and nutritional requirements of kutum fry, *Rutilus frisii kutum*. Ph.D. Thesis, Putra University, Kuala Lumpur, Malaysia.

LeGrow, S.M. and F.W.H. Beamish, 1986. Influence of dietary protein and lipid on apparent heat increment of rainbow trout, *Salmo gairdneri*. Can. J. Fish. Aquat. Sci., 43: 19-25.

Lopez, L.M., A.L. Torres, E. Durazo, M. Drawbridge and D.P. Bureau, 2006. Effects of lipid on growth and feed utilization of white seabass (*Atractoscion nobilis*) fingerlings. Aquaculture, 253: 557-563.

Luquet, P. and T. Watanabe, 1986. Interaction nutrition-reproduction in fish. Fish Physiol. Biochem., 2: 121-129.

Neverian, H.A. and N. Shabanipour, 2008. Effect of dietary digestible energy level on growth indeces of kutum (*Rutilus frisii kutum*). Iran. J. Fish. Sci., 7: 205-214.

Neverian, H.A., A. Zahmatkesh, H.A. Zamani-Kyasajmahalleh and A. Ghanaatparast, 2008. Determination of the optimum level of vitamin premix for the diet of *Caspian frisii* Kutum (Advanced fry). Pajouh. Saza., 79: 166-174.

Parazo, M., 1990. Effect of dietary protein and energy level on growth, protein utilization and carcass composition of rabbit fish, *Siganus guttatus*. Aquaculture, 86: 41-49.

Peres, H. and A. Oliva-Teles, 1999. Effect of dietary lipid level on growth performance and feed utilization by European sea bass juveniles *Dicentrarchus labrax*. Aquaculture, 179: 325-334.

Regost, C., J. Arzel, M. Cardinal, J. Robin, M. Laroche and S.J. Kaushik, 2001. Dietary lipid level, hepatic lipogenesis and flesh quality in turbot (*Psetta maxima*). Aquaculture, 193: 291-309.

Sa, R., P. Pousao-Ferreira and A. Oliva-Teles, 2006. Effect of dietary protein and Lipid levels in growth and feed utilization of white sea bream (*Diplodus sagus*) juvenile. Aquacult. Nut., 12: 310-321.

Santinha, P.J.M., F. Medale, G. Corraze and E.F.S. Gomes, 1999. Effects of the dietary protein: Lipid ratio on growth and nutrient utilization in gilthead seabream (*Sparus aurata* L.). Aquacult. Nutr., 5: 147-156.

Sargent, J., R.J. Henderson and D.R. Tocher, 1989. Fish Nutrition: The Lipids. Academic Press, London, UK., pp: 154-218.

Skalli, A., M.C. Hidalgo, E. Abellan, M. Arizcun and G. Cardenete, 2004. Effects of the dietary protein/lipid ratio on growth and nutrient utilization in common dentex (*Dentex dentex* L.) at different growth stages. Aquaculture, 235: 1-11.

Tocher, D.R., 2003. Metabolism and functions of lipids and fatty acids in teleost fish. Rev. Fish. Sci., 11: 107-184.

Winfree, R.A. and R.R. Stickney, 1981. Effects of dietary protein and energy on growth, feed conversion efficiency and body composition of *Tilapia aurea*. J. Nutr., 111: 1001-1012.