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Effects of dietary lipid levels on growth performance and blood parameters of the African catfish (*Clarias gariepinus*).

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ABSTRACT

The African catfish (*Clarias gariepinus*) fed on diets containing different lipid levels (5%, 10%, 15% and 20%) at a feeding rate of 3% of the live body weight. Fish were reared in glass aquaria at a stocking rate of 10 fish at each aquarium. Each dietary lipid level was presented in triplicates. The trial lasted for 90 days from the start. Ten fish were distributed in 100-L glass aquaria. Fish fed daily for 90 days in triplicate form.

Results revealed that fish growth parameters as a final weight, weight gain, and specific growth rate were generally increased by increasing dietary lipid levels up to 10%, after which the growth rate was markedly decreased. Differences in feed intake among different treatments varied significantly ($P<0.05$). Feed conversion ratio (FCR) was affected by dietary levels of lipids. No significant changes in survival rate were observed due to the variation in dietary lipid levels. The highest values of the erythrocyte count (RBCS), hemoglobin content (Hb) and packed cell volume (PCV) were obtained in fish group fed on the diet containing 15% and 20% lipid. Furthermore, mean corpuscular value (MCV), mean corpuscular hemoglobin (MCH) and mean corpuscular hemoglobin concentration (MCHC) were changed in blood of fish fed on different levels of lipid. Glucose, total lipids, and total protein were significantly affected by increasing the lipid levels. Plasma total lipids and total protein were increased in fish with increasing lipid levels in the diets, Also, plasma AST and ALT activities increased significantly with increasing the level of lipids in fish diets, reaching the highest value in fish group fed diet with 20% lipids. The hepato-somatic index and gonado–somatic index of African catfish were significantly increased with increasing lipid level in fish diets. The present study demonstrated that the optimum dietary lipid level was 10% for African catfish and there was no observed beneficial effect when fish was fed a diet containing 15% or 20% lipid levels.

Keywords: African catfish, dietary lipids, growth performance, hematological parameters.

INTRODUCTION

Dietary lipids play an important role in commercial diets as a source of energy and essential fatty acids for the growth and development of fishes (NRC, 1993). Moreover, it could also spare dietary protein from use as energy and limit ammonia production (Arzel *et al.*, 1994; Chou and Shiau, 1996; Kim and Lee, 2005).

The low inclusion of dietary lipids could lead to the consumption of protein for energy requirements. On the other hand, high dietary lipid content might decrease feed consumption and reduce growth (Ahamad, 2004; Pei *et al.*, 2004; El-Marakby, 2006; Abbass, 2007). Therefore, it is important, from nutritional and economical points of view, to improve protein utilization for tissue synthesis rather than for energy purposes. The increase in dietary lipid improved protein utilization in brown trout (Gabaudan *et al.*, 1989) as in many other fish (Lee and Putnam, 1973; Alliot *et al*, 1979; Cho and Kaushik, 1990).

The African catfish (*Clarias gariepinus*) due to its tolerance to a wide range of temperatures, low oxygen and high salinity levels, fast fecundity and growth rate is therefore considered as a candidate of freshwater fish for aquaculture (Hecht *et al.*, 1996; Otémé *et al.*, 1996). Moreover, this fish has high nutritive value, good taste, and fewer bones. The optimal dietary lipid levels have to be carefully evaluated and determined for this fish. Therefore, this study was conducted to determine the effect of different lipid levels (5-20%) on growth performance of the African catfish (*C. gariepinus*).

MATERIALS AND METHODS

Fish rearing conditions:

This study was carried out at the Central Laboratory for Aquaculture Research, Abbassa, Abou-Hammad, Sharkia Governorate, Egypt. The African catfish (*C. garpinus*) was obtained from a private hatchery, at Abbassa, Abou-Hammad, Sharkia Governorate. Fish were kept in an indoor tank separately for two weeks for acclimatization. The average range of fish weight was 52.2 to 53.55g. Fish were fed on a commercial diet during the preliminary period. Fifty fish were stored in a refrigerator at - 5°C for proximate chemical analysis. Fish were graded and randomly distributed into glass aquaria at a rate of 10 fish per 100-L aquarium (75 x 40 x 50 cm, L x W x H) contained well-aerated and dechlorinated tap water, which was stored overnight in cylindrical fiberglass tanks. Aquaria were supplied with compressed air from air pumps via air stones. The water temperature was 21°C for first month and was adjusted at 26.0±2.0°C by thermostatically controlled column heaters. Fish in each aquarium were hand-fed at a rate of 3% of live body weight twice daily for 90 days. Fish faeces and uneaten feed were removed daily by siphoning and the half of water volume from each aquarium was removed and replaced by well-aerated tap water. Fish

in each aquarium was weighed biweekly and subsequently the amount of the given feed was readjusted accordingly. At the end of the experiment, fish were harvested, counted and weighed. Five fish from each treatment were stored in a refrigerator at - 5° for proximate chemical analysis. The different parameters of fish growth and feed utilization were calculated as described by Ahmad *et al.* (2004). Each lipid level tested was represented in three aquaria (triplicates) i.e. 12 aquaria each contained 10 experimental fish.

Experimental diets:

Four isonitrogenous diets (35% crude protein) were formulated to contain graded levels of fish oils and corn oil mixture (1:1) to obtain total crude lipid levels of 5%, 10%, 15% or 20%. Feed ingredients were ground into fine powder in a Thomas-Wiley Laboratory Mill (Model 4, USA). After sieving, the fine powder ingredients were thoroughly mixed with the oils mixture, and 40% water was added to produce clump dough. The dough was then pelleted through a die of Spaghetti Machine (La Parmigiana, Model D45 LE, Italy) and dried at room temperature. After drying, the diets were broken up and sieved into proper pellet size (2 mm diameter). Diets were transferred to dark plastic bags and stored in a refrigerator at -5°C until used. Composition of the experimental diets and its chemical analysis are represented in Table (1).

Table (1). Ingredients and nutrients composition of the experimental diets.

Items	Lipid levels			
	5%	10%	15%	20%
Fish meal (72% CP)	20.5	20.5	20.5	20.5
Soybean meal (44% CP)	40.0	40.0	40.0	42.0
Wheat bran (16.4% CP)	5.0	5.0	5.0	5.0
Yellow corn (8.5% CP)	27.0	22.0	17.0	10.0
Fish and oil (1:1)	5.0	10.0	15.0	20.0
Vitamin & Mineral Premix [a]	1.5	1.5	1.5	1.5
Carboxymethyl cellulose	1.0	1.0	1.0	1.0
Total	100	100	100	100
Proximate analysis (%)				
Dry matter	92.03	91.75	92.15	92.11
Crude protein	35.00	34.98	35.13	34.91
Ether extract	9.52	14.33	19.22	24.12
Crude fiber	4.89	4.82	5.00	5.13
Nitrogen free extract [b]	42.99	38.36	33.30	28.37
Ash	7.60	7.51	7.35	7.47

[a] Same as Lee *et al.* (2000).

[b] Nitrogen free extract was calculated by difference (100-crude protein-crude lipid-crude ash-crude fiber).

Proximate chemical analysis:

Fish, feed and feces were ground and analyzed in triplicate samples according the standard methods of AOAC (1990). Moisture content was determined by oven drying at 65°C for 24 hours and 105°C till a constant weight was obtained. Crude protein (N x 6.25) was determined by micro-Kjeldahl method. Total crude lipid was determined by Soxthlet extractor using petroleum ether for 16 hours. Ash by combustion at 550°C . Crude fiber was estimated according to Goering and Van Soest (1970). Nitrogen-Free Extract (NFE) calculated by difference. Gross energy of experimental diets was estimated according to NRC (1993).

Blood Analysis:

At the end of experiment, blood samples were taken from caudal vein of an anaesthetized fish by sterile syringe using EDTA solution as an anticoagulant. These blood samples were used for determining erythrocyte count (Dacie and Lewis 1984), hemoglobin content (Van Kampen and Zijlstra, 1961), and haematocrite value (Britton, 1963). Plasma was obtained by centrifugation at 3000 rpm for 15 min and unhaemolyzed plasma was stored in deep freezer for further biochemical analyses. Glucose was determined, using glucose kits supplied by Boebring Mannheium kit, according to Trinder (1969). Total protein content was determined colorimetrically according to Henry (1964). Total lipids contents were determined colorimetrically according to Joseph *et al.* (1972).

Statistical analysis:

The obtained data were statistically analyzed by completely randomized design according to Snedecor and Cochran,(1994). Differences between treatments were statistical tested by Duncan's multiple range test (Duncan, 1955).

RESULTS

The growth performance:

The growth performance parameters were affected by lipid levels as shown in Table (2). The best growth performance of African catfish was attained when fish were fed on diet contained 10% total lipid. The least growth was found in fish fed the diet contained 20% lipid. Fish growth parameters as a finial weight, weight gain, and specific growth rate were generally increased by increasing dietary lipid levels up to 10% after which the growth rate was markedly decreased. Differences in feed intake among different treatments differed significantly ($P<0.05$). Feed conversion ratio (FCR) was affected by dietary levels of lipids. The best (lowest) FCR was achieved when fish fed 10% (2.79) and the worst value was obtained using the diet containing 20% (4.35). While there was no difference ($P<0.05$) with regard to the survival rates among treatments, the dietary level containing 20% lipid came significantly different ($P<0.05$) compared to the 10% and 15% lipid levels with regard to SGR and FCR.

Table (2): Growth performance parameters of African catfish as affected with different dietary lipid levels.

Items	Dietary lipid levels			
	5%	10%	15%	20%
Initial weight (g/fish)	53.55±0.98 ^{a}	52.55±1.28 ^{a}	52.55±1.36 ^{a}	52.2±1.55 ^{a}
Final weight (g/fish)	73.5±2.02 ^{b}	78.5±1.46 ^{a}	75.00±2.12 ^{a}	70.00±1.28 ^{b}
Weight gain (g/fish)	19.95±0.38 ^{b}	25.9±0.64 ^{a}	22.4±0.42 ^{a}	17.8±0.56 ^{b}
SGR (%/day)	0.352±0.02 ^{a,b}	0.446±0.02 ^{a}	0.395±0.03 ^{a}	0.325±0.03 ^{b}
Feed intake (g feed/fish)	78.00±3.5 b ^{a}	72.50±6.5 ^{a}	74.81±5.4 ^{a}	77.50±7.2 ^{b}
FCR	3.909±0.35 ^{c}	2.799±0.24 ^{a}	3.339±0.32 ^{b}	4.353±0.36 ^{c}
Survival rate (%)	90±2.5 ^{a}	100±0.0 ^{a}	100±0.0 ^{a}	100±0.0 ^{a}

The same letter in the row is not significantly different at $p<0.05$.

Hematological parameters:

The erythrocyte count (RBCS), hemoglobin content (Hb) and packed cell volume (PCV) in the blood of African catfish were significantly affected with deferent levels of lipid in the tested diets (Table 3). The highest values of these parameters were obtained in fish group fed a diet with 15% and 20% lipid levels. Furthermore, blood indices; mean corpuscular value (MCV), mean corpuscular hemoglobin (MCH) and mean corpuscular hemoglobin concentration (MCHC) were changed in blood of fish fed on different dietary levels of lipid according to the changes occurred in the blood parameters.

Results of the chemical composition of plasma are shown in Table (4). Glucose was significantly affected by increasing the lipid levels ($P<0.05$). Also, the total lipids were increased in fish fed 10, 15 and 20% lipid diet (25.649, 26.93 & 31.8 g/l, respectively). Also, plasma total protein was increased significantly with increasing lipid levels in the diets. Moreover, the obtained results showed that plasma AST and ALT activities increased significantly with increasing the level of lipids in the fish diet, reaching the highest value in fish group fed diet with 20% lipids. The hepato-somatic index and gonado–somatic index of African catfish (Table 5) were significantly increased with increasing lipid level in fish diets ($P<0.05$).

Table (3): Changes in erythrocyte count (RBCs), hemoglobin (Hb) content, packed cell volume (PCV), mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH) and mean corpuscular hemoglobin concentration (MCHC) in the blood of African catfish fed different dietary lipid levels.

Items	Dietary lipid levels			
	5%	10%	15%	20%
RBCs ($x10^6$/ml)	2.33±0.11 [b]	2.56±0.07 [b]	3.15±0.14 [a]	2.62±0.05 [b]
Hb (g/100ml)	6.54±0.34 [d]	7.49±0.43 [c]	9.11±0.49 [a]	8.18±0.41 [b]
PCV (%)	23.6±0.60 [b]	24.0±1.06 [b]	28.2±1.17 [a]	23.3±0.96 [b]
MCV	93.9±1.38 [c]	102.6±2.65 [b]	1o7.9±4.13 [a]	73.6±2.35 [d]
MCH (pg)	29.3±0.75 [b]	28.1±0.92 [b]	31.4±1.02 [a]	29.3±1.01 [b]
MCHC (%)	31.4±1.05 [b]	29.7±1.08 [c]	39.3±1.22 [a]	29.9±0.81 [c]

The same letter in the row is not significantly different at $p<0.05$.

Table (4): Changes of glucose, total protein, total lipid, aspartate aminotransferas (AST) and alanine aminotransferase (ALT) in plasma of African catfish fed different dietary lipid levels.

Items	Dietary lipid levels			
	5%	10%	15%	20%
Glucose (mg/dl)	73.4±2.09 [d]	108.4±4.51 [c]	280.7±14.63 [a]	249.5±18.58 [b]
Total protein (mg/dl)	4.38±0.38 [b]	4.51±0.32 [b]	5.61±0.19 [a]	5.16±0.39 [a]
Total lipid (g/l)	21.44±0.79 [c]	25.65±1.32 [b]	26.92±1.33 [b]	31.82±2.43 [a]
AST (IU/l)	14.90±1.24 [c]	14.86±0.55 [c]	30.52±1.35 [b]	32.20±1.352 [a]
ALT(IU/l)	6.70±1.10 [b]	6.57±0.58 [b]	7.808±0.63 [a]	7.81±0.46 [a]

The same letter in the row is not significantly different at $p<0.05$.

Table (5): Change of hepatosomatic index (HSI) and gonadosomatic index (GSI) of African catfish fed different dietary lipid levels.

Items	Dietary lipid levels			
	5%	10%	15%	20%
HSI (%)	0.736±0.075 [b]	0.789±0.045 [b]	0.798±0.074 [b]	1.648±0.133 [a]
GSI (%) ♂	0.74±0.03 [b]	0.817±0.046 [a]	0.955±0.104 [a]	0.870±0.057 [a]
GSI (%) ♀	12.617±1.79 [c]	12.405±2.383 [c]	14.945±1.342 [b]	20.54±0.377 [a]

The same letter in the row is not significantly different at $p<0.05$.

DISCUSSION

The growth performance of the African catfish was improved with the gradual increase in dietary lipid levels from 5% to 10%, while the least fish growth values were achieved in fish fed on diet containing 20% lipid. The lowest fish growth obtained when fish fed diet containing excess lipid may be due to the imbalance of protein/energy ratio and/or reduced feed consumption. These results are in agreement with that reported by Lee *et al.*, (2000) who showed a significant decrease from 7% to 16% in weight gain. Almost the same trend was found by Oku and Ogata, (2000) who reported 10.5 % to 20% growth rate decrease as well as what has been reported by Alam *et al.*, (2003) reporting a reduction of 10% to 20% of growth rate. Also, Watanabe *et al.* (2001) found similar results with turbo, *Scophalmus maximus* and mutton snapper, *Lutjanus analis*. Cho and Watanabe (1988) showed that the increase of energy retention and the formation of new tissues promote a progressive increase in fish weight. The weight gain of young fish is usually a reliable indicator of the nutritional adequacy of diet. Anwar and Jafri (1995) reported that feed conversion, protein efficiency ratio and muscle protein deposition were higher and comparable in fish fed 7 and 9% lipid diet. Also, Martino *et al.* (2002) reported that the diet with 6% lipid gave the poorest performance, while fish fed the highest lipid level (18%) showed the best nutritional performance. Feed conversion ratio and daily feed consumption showed a marked decrease ($P<0.05$) (improvement) in inverse proportion to lipid levels in the diet.

The utility of hematology in assessing fish health and as an aid to fish disease diagnosis has been questioned. In this study, RBCs, Hb and PCV were were significantly affected by different lipid levels. Klinger *et al.* (1996) reported that RBCs are the cells most affected by dietary manipulations. As a particular fish acclimates to a lower temperature, more highly unsaturated fatty acids are incorporated into the phospholipids protein of the membrane. This is true for blood cells (Lie *et al.*, 1989) and other cells (Dey *et al.*, 1993).

The difference in lipid requirement among the different studies are existed due to the differences in many factors such as diet formulation and composition, feeding strategy, fish size, water quality and culture system (Lin *et al.*, 1997). The calculated blood indices i.e. MCH, MCHC and MCV have a particular importance in describing anemia in most animals (Coles, 1986). The disturbances occurred in these indices are related to those occurred in RBCs, Hb and PCV due to the exaggerated disturbances that occurred in both metabolic and hemopoietic activities of fish. The obtained results showed that the plasma AST and ALT activities increased significantly with increasing the lipid levels. The same trend was recorded in glucose, total protein and total lipids. These results are in agreement with Gallagher (1999) who demonstrated that liver ALT activity increased in the liver of sunshine bass as protein level increase or when lipid replaced dextrin in the diet. The increase in enzyme activity corresponded to an increase in liver lipid content. However, if too much lipid is provided in the

diet weight gain may actually decreased (Watanabe, 1982). Furthermore, it has been well demonstrated that too much dietary lipid may promote an undesirable increase in body lipid content causing many health problems (Klinger *et al.*, 1996).

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