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Effect of Different Levels of Dietary L-Carnitine on Growth Performance, Food Efficiency and Body Composition of Pikeperch (*Sander lucioperca*) Fingerlings

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Abstract: The Effects of different levels of dietary L-Carnitine on growth performance, food efficiency and body composition of pikeperch (*Sander lucioperca*) were investigated in 42 days feeding trial. Fingerlings of pikeperch were fed without L-Carnitine added to control diet (treatment A), 1000 mg/kg L-Carnitine added to control diet (treatment B) and 2000 mg/kg L-Carnitine added to control diet (treatment C) in 1 m^3 concrete tanks. 200 fingerlings (1.63 g, mean weight) were stocked in each tank and fed up to 6 meals per day. Higher increment in body weight (5.92±0.37 g), specific growth rate (3.75±0.17) and food efficiency (98.04±4.56) were obtained with treatment C (2% L-Carnitine added to control diet). This treatment also showed significant ($P<0.05$) decrease in food conversion ratio (1.02±0.05) in comparison with other treatments. Highest survival rate was observed in treatment (B) with a (84.88±0.92%) rate and greatest cannibalism (1.25±0.09) was found in treatment (A), respectively. Also significant differences ($P<0.05$) was observed on crude protein and crude lipid content among the treatments. Highest crude protein (64.53±0.84) and lowest crude lipid (21.59 ±0.23) contents of pikeperch whole body was observed in treatment (C). The amount of fat in treatment (B) decreased but did not show any significant differences with treatment (C) ($P>0.05$). According the obtained results of this research, use of L-Carnitine through addition in the diet, during pikeperch fingerlings growth, is recommended and it seems that 2000 mg/kg L-Carnitine addition the diet will have best results on growth performance and suitable body composition.

Key words: *Sander lucioperca* · L-Carnitine · Protein · Lipid · Food Efficiency

INTRODUCTION

L-carnitine (β-hydroxy -γ-N -trimethylaminobutyric acid) widely is water-soluble quaternary amine and plays an important role in lipid β-oxidation to facilitate the importation of activated long-chain fatty acids into mitochondria and the accompanying intermediate compounds out of the mitochondrial matrix [1, 2]. It has also been suggested that L-carnitine supplementation may stimulate the protein-sparing action by increasing the energy derived from lipids [3] Due to its role in lipid metabolism in fish; dietary L-carnitine supplementation has been found to enhance protein synthesis and promote growth performance [4].

Increased fatty acid oxidation via L-carnitine mediation is accompanied with decrease in essential amino acids catabolism. The advantage of dietary L-carnitine supplementation for growth performance is related to optimum dietary utilization as well as inhibition from lysine and methionine catabolism [5]. During the past decade, some reports have indicated that L-carnitine improved the performance of species such as hybrid striped bass [6], beluga sturgeon [7], black sea bream [8], but has no effect on the growth performance of rainbow

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trout fry and fingerlings [9, 10], Atlantic salmon, [11], European sea bass [12] and tilapia [13].

Pikeperch (*Sander lucioperca*), is one of the most commercially important species of indigenous ichthyofauna of Caspian Sea [14] and is a valuable species for aquaculture due to its rapid growth, flesh quality and high commercial value [15]. Due to importance of pikeperch as an aquaculture species and little information on effect of L-carnitine on pike perch, this study aims to investigate the effect of different levels of dietary L-carnitine on growth performance, food efficiency, survival rate and body composition of pikeperch (*Sander lucioperca*) fingerlings.

MATERIALS AND METHODS

Experiment was performed with three treatments in triplicates, in 9 veniro tanks of 1000 L volume (with dimensions of 0.8 x 0.8 m) with completely randomized design [16]. Each veniro contained a volume of 400 L of water with proper aeration. The water inlet with a flow rate of 5-6 L/min, created a slight circulation current in the tank. The initial stocking rate was 2 fish/l, (same size and from same stock) [17]. Light intensity was 50 lux for 24 h in rearing saloon [15]. The pond cultured pikeperch fingerlings (initial mean weight of 1.63 g were transported to veniro tanks and acclimated over a 10-day. After acclimation period, the veniro were assigned as 3 treatments (3 replicats per treatment) receiving the control diet (Table 1), (France) with pellet size of 1.1 mm for first half of rearing period and 2.5 mm for second half of rearing period (group A), or control diet supplemented with 1000 (group B) and 2000 mg L-carnitine/kg diet (group C) for 6 weeks. The fingerlings were hand-fed from 8:00 to 20:00 with formulated. Feeding rate was 4-5 % of actual fish biomass for all groups. Water Physicochemical parameters were monitored during of experiment (Table 2). Every 7 days 20 fish from each veniro (60 individuals/treatment) were randomly captured and their length were measured individually, with 0.1 mm accuracy. Fingerlings were weighted collectively from day 0 to day 14 and individually from day 21 to day 42 with 0.01 mg accuracy. The number of sampled fingerlings was taken into account for survival calculation. Body weight index (BWI), average daily growth (ADG), specific growth rate (SGR), condition factor (CF), food conversion ratio (FCR), food efficiency (FE), survival rate (SR) and cannibalism were assessed as follows [18].

Table 1: Formulation and proximate composition of experimental diets (% of Dry Matter)

Pellet size (mm)	1.5-2.2
Ingredient (%)	
Fish meal	48
Fish oil	11
Concentrate*	9
Maize gluten	8
Soya cake	7
Field peas	6
wheat	6
wheat gluten	5
Chemical composition	
Crude protein (%)	45.61
Crude lipid (%)	17.45
Fiber (%)	1.52
Ash (%)	10.53
Gross energy	22.1

*Concentrate (pea proteins, krill meal, hydrolyzed fish, vitamins and minerals, antioxidant (ethoxyquin), lecithin, yeas extract).

Weight gain (g) = $(W_f - W_i)$
Average daily growth (ADG, %) = $100\ (W_f - W_i)\ T^{-1}$
Specific growth rate (SGR, % day^{-1}) = $100\ (LnW_f - LnW_i)?T^{-1}$
Condition factor (CF) = $100 \times W_f \times (L_f)^{-3}$
Feed conversion ratio (FCR) = $W_{TFS} \times AWG^{-1}$
Feed efficiency (FE) = $(FB - IB)TFC^{-1}$
Counted mortality (%) =$100\ (Nd+Nc)\ Ni^{-1}$
Survival rate =$100\ (N_f - N_i)$
Cannibalism (%) =$100\ (Nc+Nm)\ Ni^{-1}$

Table 2: Physical and chemical parameters during the experiment

	Duration of experiment (weeks)					
Physicochemical parameters	1	2	3	4	5	6
Water temperature (°C)	24.62 ± 0.43	24.72 ± 0.86	25.63 ± 0.96	26.23 ± 1.12	26.35 ± 0.59	26.32 ± 0.73
Dissolved O_2 (mg/L)	7.43 ± 0.68	7.29 ± 0.62	7.70 ± 0.22	6.86 ± 0.27	6.71 ± 0.69	7.01 ± 1.31
pH	8.16 ± 0.19	8.37 ± 0.22	7.92 ± 0.08	8.13 ± 0.15	8.26 ± 0.30	8.28 ± 0.24

Data are presented as mean and standard deviations

Abbreviations in above-mentioned equations are as follow: where W_i and W_f are the initial and final body weights (g), W_{TFS} is the weight of the total feed supplied (g)T = duration of experiment (days), L = final body length (cm), Ni= initial number of fingerlings, Nf= final number of fingerlings, Nd=number of dead fish without signs of cannibalism, Nc = number of dead fish due to cannibalism, Nm=number of missing fish at the counting (end of experiment), C = Cannibalism, ÄT=duration of the experiment (days), TFC the total food consumption (g), IB the initial biomass (g), FB the final biomass (g).

Fish samples were analyzed for proximate composition according to AOAC [19]. Ten fish from each veniro of experimental treatments were taken for chemical analysis of body composition. Sampled fish were killed and the viscera were removed and the carcasses were stored at -18°C for chemical analyses. Samples' moisture (at 105°C for 24h), crude protein (Kjeldahl apparatus, Gerhardt, Königswinter, Germany. Nitrogen* 6.25), crude fat (extraction with petroleum ether by Soxhlet apparatus, Behr, Düsseldorf, Germany) and ash (incineration at 550°C for 6 h) were determined according to AOAC [19].

Statistical Analysis: Results are given as mean ± standard deviations. Data normality was tested by Shapiro-Wilk's test. Data were subjected to one-way ANOVA and significant difference among treatments was determined by Duncan's test the values of P<0.05 were considered significantly different. All analyses were performed using statistical software SPSS (version, 16).

RESULTS

The results of growth performance of pikeperch fingerlings were fed by control diet (treatment A), 1000 mg/kg L-carnitine added to control diet (treatment B) and 2000 mg/kg L-carnitine added to control diet (treatment C) comparisons are as presented in Table 3. Average fish fingerlings weights were 5.93, 6.14 and 7.52 g for treatments A, B and C, respectively, at the end of experiment. Daily specific growth rates was significantly (P<0.05) higher in treatment C than others at the end of experiment. The final condition factor at the end of feeding trial, ranged from 0.72 for treatment A to 0.76 for treatment B (Control + 1000 mg/kg L-carnitine), respectively. There was no significant difference on CF of fingerlings fed different diet at the end of experiment. Food conversion ratio was significantly (P<0.05) higher in treatment A. Treatment C showed the lowest FCR (1.02) during experiment. Highest food efficiency was found in fingerlings fed Control + 2000 mg/kg L-carnitine (treatment C). The highest Survival rate was observed in treatment B but not significant different between treatment. The body compositions of Pikeperch (*S. lucioperca*) maintained on various treatments is presented in Table 4. Significantly (P<0.05) higher content of carcass total lipid was observed in fish fed diets (A) compared with those fed diets treatments. Higher and lowest content of carcass crude protein and crude lipid were observed in fish fed diets (c) compared with other treatments. In treatment (B) also the amount of fat decreased, but it was not a

Table 3: Effects of different diets on growth indices of pike perch fingerlings at the end of experiment

	Treatments		
Parameters	Control (A)	Control + 1000 mg/kg L-carnitine (B)	Control + 2000 mg/kg L-carnitine (C)
Initial weight (g)	1.63 ± 0.26 [a]	1.54 ± 0.47 [a]	1.60 ± 0.51 [a]
Final weight (g)	5.93 ± 0.66 [b]	6.14 ± 0.42 [b]	7.52 ± 0.69[a]
Weight gain (g)	4.30 ± 0.28 [b]	4.60 ± 0.46 [b]	5.92 ± 0.37 [a]
ADG (%)	11.04 ± 1.49 [b]	10.73 ± 1.65 [b]	12.89 ± 1.63 [a]
SGR (% per day)	3.09 ± 0.05 [b]	3.16 ± 0.14 [b]	3.75 ± 0.17 [a]
Initial length (mm)	5.98 ± 0.52 [a]	6.03 ± 0.31 [a]	6.11 ± 0.13 [a]
Final length (mm)	9.38 ± 0.67 [a]	9.29 ± 0.91 [a]	10.02 ± 0.25 [a]
Initial CF	0.74 ± 0.07 [a]	0.70 ± 0.09 [a]	0.70 ± 0.08 [a]
Final CF	0.72 ± 0.09 [a]	0.76 ± 0.14 [a]	0.74 ± 0.18 [a]
FCR	1.31 ± 0.08 [a]	1.28 ± 0.11 [a]	1.02 ± 0.05 [a]
FE	76.33 ± 4.65 [b]	78.16 ± 5.24 [b]	98.04 ± 4.56 [a]
Survival rate (%)	83.50 ± 1.26 [a]	84.88 ± 0.92 [a]	84.21 ± 1.34 [a]
Cannibalism (%)	1.25± 0.09 [a]	1.14 ± 0.11 [a]	1.03 ± 0.14 [a]

Values (mean ± SD) of the same line with different letters indicate significant differences (P<0.05)

Table 4: Mean and standard deviation of Body composition of pikeperch fingerlings fed diets with various dietary treatments

Duration of experiment (Day)	Treatments	Body Composition (%) Moisture	Crude protein	Crude lipid	Crude ash
1	A	76.45 ± 0.55[a]	59.23 ± 0.76[a]	20.84 ± 1.31[a]	11.23 ± 0.49[a]
	B	76.43 ± 0.74[a]	58.41 ± 0.82[a]	21.32 ± 0.78[a]	10.49 ± 0.71[a]
	C	75.76 ± 0.75[a]	58.67 ± 0.79[a]	21.36 ± 0.83[a]	10.67± 0.68[a]
42	A	74.15 ± 0.71[a]	60.96 ± 0.94[b]	25.39 ± 0.52[a]	10.79 ± 1.17[a]
	B	76.54 ± 1.02[a]	62.48 ± 1.06[ab]	23.63 ± 0.47[ab]	11.09 ± 0.65[a]
	C	75.67 ± 0.68[a]	64.53 ± 0.84[a]	21.59 ± 0.23[b]	11.33 ±0.83[a]

significant (P>0.05) change with treatment (A). Also there was no significant difference in Ash among all treatment (P>0.05).

DISCUSSION

The present study demonstrated that the addition of L-carnitine in different levels to diet improve the growth performance of pikeperch (*sander lucioperca*) and best results found in treatment C (2000 mg/kg L-carnitine). No significant differences were observed with fish fed on other treatment. The increased growth of pike perch (*sander lucioperca*) (Table 3) as a result of L-carnitine application might be due to the increased energy availability due to a raise in fatty acid oxidation. The obtained results are in close agreement whit Torreele *et al*. [5] on African catfish, Santulli and D`Amelio [12] on European sea bass, Chatzifotis *et al*. [20] on red seabream, Jayaprakas *et al.* [21] on Tilapia (*Oreochromis mossambicus*), Keshavanath and Renuka [22] on Ruoho big carp Hindi, Becker *et al.* [23] on Hybrid tilapia (*Oreochromis niloticus* × *Oreochromis aureus*), Twibell and Brown [6] on striped bass, Mohseni *et al.* [7] on beluga sturgeon and Ma *et al.* [8] on black sea bream, Jalali Haji-abadi *et al*. [24] on rainbow trout.

In contrast, several other studies reported that L-carnitine did not increased the growth in fish, for instance see results obtained for Channel catfish [25], Rainbow trout [9, 10], Atlantic salmon [26]. Cichlid ornamental (*Pelvicachromis pulcher*) [11], Hybrid tilapia (*Oreochromis niloticus* × *Oreochromis aureus*) [27].

The body composition analyses showed that Addition of L-carnitine increased the crude protein and reduced the crude lipid content in pikeperch (Table 4). This might be due to the fact that supplementation of L-carnitine results in an acceleration of fatty acid oxidation and improved nitrogen retention [26]. Also L-carnitine is most concentrated in tissues that use fatty acids as their primary dietary fuel, such as skeletal and cardiac muscle. In this regard, L-carnitine plays an important role in energy production by chaperoning activated fatty acids (acyl-CoA) into the matrix for and accompanying intermediate compounds out of the mitochondrial matrix to prevent their accumulation. Carnitine-acylcarnitine translocase is responsible for the transport of carnitine and its esters across the inner mitochondrial membrane. It can help to prevent the accumulation of fatty acids in tissues and thus the ratio of muscle to fat in the body rises [28]. Furthermore, OzõRio [4] indicated that 1000 mg kg^{-1} L-carnitine supplementation in the diet of African catfish raised the concentration of some amino acids in muscle tissue and concluded that L-carnitine increased fatty acid oxidation and decreased amino acid combustion for energy. Our results are in close agreement with Santulli and D`Amelio [12] on European sea bass, Torreele *et al.* [5] on African catfish, Burtle and Liu [25] on channel catfish fingerling, Ji *et al.* [26] on Atlantic salmon and Ma *et al.* [8] on black sea bream.

This difference may be related to the fish size [29], dietary composition and feed processing [30], possible species effects, fish developmental stage, environmental conditions and the section body used for analysis, whole body (present study; [9]) versus fish fillet [24, 26]. According the results of this research, use of L-carnitine through addition in the diet, during pikeperch fingerlings growth, is recommended and it seems that 2000 mg/kg L-carnitine addition the diet will have the best results on growth performance and suitable body composition.

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