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Requirements of n-3 Highly Unsaturated Fatty Acids in Beluga (*Huso huso*) Juvenile and their Effects on Growth, Carcass Quality and Fatty Acids Composition

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ABSTRACT

An 8-week feeding experiment was carried out on beluga (*Huso huso)* juvenile to evaluate the effects of different levels of fish oil containing n-3 highly unsaturated fatty acids (n-3 HUFAs) on fish growth and fatty acid composition. The requirements of beluga juvenile for n-3 HUFAs were studied by feeding fish diets containing six different levels of n-3 HUFAs ranging from 1.56 to 17.25% (% of total fatty acids). Weight gain, feed conversion ratio, condition factor, specific growth rate and protein efficiency ratio were not significantly different among dietary treatments, nor was the body composition (including: moisture, protein, lipid and ash) of beluga juvenile (P> 0.05). There were no significant differences among plasma protein, glucose, cholesterol and triglyceride contents of dietary treatments (P> 0.05). However, haematocrit values were significantly lower in diets 1 and 2 (P< 0.05). The fatty acid composition of fish showed a pronounced change from the initial carcass with fatty acid composition changes in experimental diets. The fatty acid composition of the beluga carcass fed on diets containing various levels of n-3 HUFAs reflected the dietary fatty acid composition. The n-3 HUFAs contents of the lipids of fish increased with an increase in dietary n-3 HUFAs levels. Results suggested that minimum levels of n-3 HUFAs in diet have no effect on growth and n-3 HUFAs are not a restrictive factor on growth in beluga.

Keywords: Canola oil, DHA, EPA, Fish oil, Oil.

INTRODUCTION

The great sturgeon, *Huso huso*, is an important and commercial species found along the coasts of the southern Caspian Sea. Beluga is one of the most important species of sturgeon for aquaculture in Iran due to its desirable characteristics such as fast growth, easy and fast domestication, life acceptance in captivity, and great adaptation to artificial diet (Vaciliva *et al*., 2000) However, no commercial feeds are produced for this species and information on its nutritional requirements is insufficient. The presence of n-3 highly unsaturated fatty acids (HUFAs), particularly eicosapentaenoic (EPA, C20:5n-3) and docosahexaenoic acid (DHA, C22:6n-3) provides many health benefits. These substances are only found in fish and sea foods and play an important role in the development and functioning of the nervous system (brain), photoreception (vision), and the reproductive system (Sargent *et al*., 2001; Sidhu, 2003; Pirestani *et al*., 2010). Therefore, seafood is a very significant component in human nutrition. Fishery products are the principal sources of n−3 HUFAs in the human diet, but global

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fisheries have declined. This fact makes clear that these natural sources of n−3 HUFAs are restricted and aquaculture production will face a deficiency. Food grade fisheries that supply fish oil and fish meal have reached their limit of sustainability (Pike and Barlow, 2003; Shepherd *et al*., 2005). The aquaculture industry utilizes 68.2 and 88.5 % of the global production of fish meal and fish oil, respectively (FAO 2008) and within the next decade fish oil production may not meet the amounts required for aquaculture (Kaushik, 2004; Tacon, 2005). The general agreement is that alternative protein and oil sources are needed to supplement or replace fish meal and fish oil in aquafeeds, thus contributing to long-term sustainability of the aquaculture industry (Hardy, 2008). Vegetable oils constitute promising candidates for fish oil replacement in fish diet, having steadily increasing production, with high availability and better commercial value. Some vegetable oils for instance soybean oil and canola oil are considered possible alternative lipid sources for salmonids and freshwater and marine fish because they are rich in polyunsaturated fatty acids (PUFA), especially linoleic (18:2n−6) and oleic acid (18:1n−9), but devoid of n-3 HUFAs, (Caballero *et al*., 2002; Montero *et al*., 2005; Mourente and Bell, 2006). There is also an increasing interest in the role of fatty acids in metabolism as the use of vegetable proteins and oils in fish diets will reduce n-3 HUFAs contents and change the fatty acid balance in fish tissues (Bell *et al*., 1996). The replacement of fish oil by vegetable oils has a profound impact on the fatty acid composition of fish tissues with an increase in 18:2n−6 and 18:3n−3 and a decline in DHA and EPA (Greene and Selivonchick, 1990; Caballero *et al*., 2002). Various fish show differences in their apparent ability to convert linoleic to EPA and DHA. Early studies indicated that the rainbow trout, *Oncorhyuchus mykiss* (Walbaum), was able to convert linolenic acid to EPA and DHA (Owen *et al*., 1975) while the same conversions by ayu, *Plecoglossus altivelis* and Japanese eel *Anguilla japonica* (Kanazawa *et al*., 1979) were only 36% and 20% of that of trout, respectively. Several studies conducted on freshwater fish showed that vegetable oils can successfully replace fish oil in fish feeds without affecting survival and growth (Wonnacott *et al*., 2004; Subhadra *et al*., 2006). Caballero *et al*. (2002) found that that in rainbow trout up to 80–90% of vegetable oils (e.g., soybean, rapeseed, olive, and palm oils) can be used without compromising their growth. Salhi *et al*. (1999) indicated that the increase in dietary polar lipids may improve lipid transport from the enterocytes to the blood by enhancing the chylomicron synthesis.

There is little information on lipid utilization and fatty acid composition in sturgeon particularly beluga. Deng *et al*. (1998) reported that White sturgeon, *Acipenser transmontanus,* requires both n-3 and n-6, fatty acids. Similarly, Sener *et al*. (2005) suggest that sturgeon *Acipenser gueldenstaedtii* requires both n-3 and n-6 fatty acids and observed that total replacement of the fish oil by sunflower oils did not cause a significant difference in growth performance whereas replacement of soybean oil with fish oil resulted in a significant difference in growth performance.

The aim of the this study was to investigate the n-3 essential fatty acid requirement for the growth of beluga juvenile and the effect of dietary n−3 HUFAs levels on body composition to ensure the production of sturgeon with good growth and also rich in EPA and DHA. Thus, we attempted to replace n-3 HUFAs rich fish oil by vegetable oils in low fish meal diets for beluga juvenile.

MATERIALS AND METHODS

Experimental Diets

Ingredients and nutrient composition of the experimental diets are given in Table 1. Six experimental diets were formulated to be isoproteic and isolipidic containing six levels of n-3 HUFAs ranging from 1.56 to

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Table 1. Formulation (g 100 g^{-1} diet) and chemical composition (% DM) of the experimental diets.

	Experimental diets					
	1	2	3	4	5	6
Ingredients (g 100 g^{-1})						
Kilka fish meal	10.0	10.0	10.0	10.0	10.0	10.0
Corn gluten	17.0	17.0	17.0	17.0	17.0	17.0
Casein	18.7	18.7	18.7	18.7	18.7	18.7
Gelatin	14.0	14.0	14.0	14.0	14.0	14.0
Yeast	6.3	6.3	6.3	6.3	6.3	6.3
Soybean meal	6.0	6.0	6.0	6.0	6.0	6.0
Dry milk	5.0	5.0	5.0	5.0	5.0	5.0
Wheat meal	3.0	3.0	3.0	3.0	3.0	3.0
Vitamin mixture [a]	1.5	1.5	1.5	1.5	1.5	1.5
Mineral mixture [b]	1.5	1.5	1.5	1.5	1.5	1.5
L-methionine	1.5	1.5	1.5	1.5	1.5	1.5
L-lysine	1.5	1.5	1.5	1.5	1.5	1.5
Canola oil	14	13	12	11	9.5	5.0
Kilka fish oil	0.0	1.0	2.0	3.0	4.5	9.0
Chemical composition						
Dry matter (%)	92.2	90.8	91.6	92	91.7	90.6
Crud protein(% DM)	50.4	50.2	51.0	50.8	51.1	50.2
Crude fat (% DM)	16.4	16.4	16.2	16.3	16.0	15.8
Ash	4.6	4.3	4.3	4.0	4.1	4

[a] Supplied (IU or mg kg^{-1} diet): Vitamin A: 1800 IU; Vitamin D3: 1200 IU; Vitamin E: 120 mg; Vitamin B_{12}: 24 mg; Riboflavin: 15 mg; Niacin: 90 mg; D-pantothenic acid: 27 mg; Menadione: 3 mg; Folic acid: 4.8 mg; Pyridoxine: 9 mg; Thiamine: 9 mg; D-biotin: 0.48 mg; Choline chloride: 360 mg; Cobalamin: 24 mg; Ascorbic acid: 156 mg; Nicotinic acid: 90 mg; Inositol 72, Antioxidant: 15mg .

[b] Supplied (mg kg^{-1} diet): Zn: 18 mg; I: 0.6 mg; Mg: 7.8 mg; Co: 0.15 mg; Se: 0.15 mg; Cu: 1.8 mg, Fe: 12 mg.

17.25% (% of total fatty acids). Canola oil was used as the only lipid source in the first diet (Diet 1) and for preparing other diets, canola oil was partly replaced by fish oil. The compositions of fatty acids in experimental diets are shown in Table 2. Major protein sources such as (corn gluten, casein and gelatin) were used as a way to reduce n-3 HUFAs content in the basal diet, in order to obtain a minimal level of n-3 HUFAs close to 1.56% in Diet 1.

Fish and Feeding Trial

Beluga (*Huso huso*) juveniles were obtained from the Shahid Marjani Sturgeon Hatchery Center (Gorgan, Iran) and kept at the aquaculture laboratory at the Gorgan University of Agricultural Sciences and Natural Resources. They were acclimated to laboratory conditions and fed a commercial fish feed for 4 weeks before starting the experiment. Juveniles with 48.4±1.98 g weight (Mean±SE) were randomly distributed to eighteen 420 l circular fiberglass tanks (350 l water volume) with 15 fish to each tank. Water in each tank was daily changed at a rate of 80% of the volume and supplemental aeration was provided to maintain dissolved oxygen near saturation. Three replicate groups of fish were hand-fed to apparent satiation twice a day (0800 and 1700 hour for 6 days per week) for 8 week. Photoperiod was left at natural conditions during the feeding trail.

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Table 2. Composition of dietary fatty acids used in the experiments (% of total fatty acids).

Fatty acids	Experimental diets					
	1	2	3	4	5	6
∑Saturates	10.50	11.94	12.47	13.24	15.00	19.32
C14:0	0.59	1.03	1.34	1.68	2.13	3.51
C16:0	6.92	7.52	7.46	7.66	8.58	10.58
C18:0	2.52	2.84	3.07	3.22	3.61	4.53
C20:0	0.40	0.49	0.54	0.61	0.6	0.66
∑Monoenes	54.82	52.66	51.34	51.17	47.39	39.93
C14:1n5	0.10	0.17	0.26	0.32	0.41	0.88
C16:1n7	1.90	2.03	2.19	2.35	2.58	3.24
C18:1n7	2.38	2.34	2.32	2.23	2.18	2.03
C18:1n9	49.33	46.89	45.23	43.82	40.53	31.67
C20:1n9	1.04	1.19	1.27	1.40	1.62	2.05
∑n-6	18.66	17.87	16.95	16.15	14.86	10.80
C18:2n6	18.31	17.40	16.37	15.50	14.03	9.74
C20:2n6	0.00	0.10	0.18	0.23	0.30	0.45
C20:4n6	0.29	0.35	0.38	0.42	0.49	0.58
∑n-3	8.74	9.99	11.34	13.10	15.34	21.71
C18:3n3	7.17	6.72	6.43	6.19	5.73	4.46
C20:3n3	0.00	0.00	0.00	0.05	0.08	0.12
C20:4n3	0.07	0.12	0.18	0.25	0.38	0.62
C20:5n3	0.65	1.39	2.07	2.79	3.75	6.57
C22:5n3	0.11	0.17	0.26	0.38	0.50	0.81
C22:6n3	0.71	1.55	2.36	3.38	4.84	9.08
∑n-3 HUFA[a]	1.56	3.26	4.90	6.89	9.60	17.25
DHA/EPA	1.09	1.11	1.14	1.21	1.29	1.38

[a] Highly unsaturated fatty acids (C≥ 20).

Water quality analysis

Throughout the experiment, the water quality parameters were measured daily in each experimental tank. Water temperature, dissolved oxygen (DO) and pH were measured by a water checker (Horiba u 10, Japan). Average water temperature, DO and pH in all treatments were 25.48±1.8ºC, 6.0±0.3 mg L^{-1} and 8.35±0.1, respectively.

Sampling

At the beginning of the growth trial, 9 fish from an initial pool of fish were sampled and frozen for the analysis of their carcass composition. At the end of the growth trial, fish were fasted for 24 hours before slaughter. All fish were weighed individually and then three fish per each tank were anaesthetized with clove powder (300 mg l^{-1}) and killed for comparative carcass analysis and blood collection. Samples were immediately frozen and kept at −80°C individually until they were analyzed for fatty acid profile. Blood samples were collected by cutting peduncle and using heparinized micro-capillary tubes for measuring haematocrit (%) and by using a 1.5 tube ml for biochemical analysis. Heparinized microhaematocrit capillary tubes were centrifuged at 3500×g for 10 minutes in a clinical centrifuge for haematocrit evaluation. Tubes 1.5 ml in volume were centrifuged (3500×g for 5 minutes) and stored to measure protein, glucose, triglyceride, and cholesterol. Total body weight, standard length and feed rates were adjusted for each tank.

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Analytical Methods

Carcass and feed proximate composition analyses were conducted according to AOAC (1995) procedures as follows. Samples of carcass were dried to a constant weight at 105°C to determine moisture. Protein was determined by measuring nitrogen (N×6.25) using the Kjeldahl method. Lipids were extracted by ether using Soxhlet extraction, and ash was determined by combustion at 550°C. All analyses were performed in triplicate profiles.

Lipid and fatty acid composition were determined in diets and carcass. Lipid extraction was according to Folch *et al*. (1957) and fatty acids were transformed to methyl esters following Berry, Cevallos *et al*. (1965). Fatty acids were separated by gas chromatography using a Fisons Instruments 8000 series equipped with a DB-FFAP (30 m-0.25 mm ID, 0.25 mm film) capillary column and using a Fisons Instruments 8000 AS autosampler (Fisons Instruments, Milan, Italy). Nitrogen was used as the carrier gas. Injector and detector temperatures were set at 250°C. The column temperature was programmed initially at 183°C for 10 minutes and then to increase at a rate of 4°C min^{-1} to a final temperature of 220°C. Peaks were identified by comparing relative retention times with standards (Sigma-Aldrich, Buchs SG, Schweiz). Furthermore, individual FAME (fatty acid methyl esters) concentrations were calculated as a percentage of the total identifiable fatty acids.

Protein, glucose, triglyceride, and cholesterol contents in plasma were also measured by using commercial clinical investigation kits (Pars Azmun, Iran).

Statistical Analysis

The data was subjected to one-way analysis of variance (ANOVA) using the SPSS program Version 11.5 (SPSS, Chicago, IL, USA). Significant differences ($P< 0.05$) among means were determined by Duncan's multiple range test. All data in the text are presented as Mean±SE.

RESULTS

Growth Performance

All diets were well accepted. Growth performances of beluga fed with diets containing different n-3 HUFAs levels for 8 week are presented in Table 3. Survival was not affected by dietary n-3 HUFAs level. Weight gain, CF, FCR, SGR and PER were not significantly ($P> 0.05$) affected by

Table 3. Average (Mean±SE) weight gain (WG), condition factor (CF), feed conversion ratio (FCR), specific growth rate (SGR), protein efficiency ratio (PER) and survival of beluga juvenile after 60 days of rearing at different dietary n-3 HUFA levels.

	Diet no					
	1	2	3	4	5	6
WG[a] (g)	103.70±5.48	104.91±3.97	108.98±5.37	112.1±6.24	108.44±5.28	109.70±4.47
CF[b]	0.35±0.01	0.35±0.00	0.34±0.01	0.35±0.02	0.34±0.01	0.34±0.01
FCR[c]	0.82±0.01	0.82±0.00	0.81±0.01	0.80±0.02	0.81±0.01	0.81±0.01
SGR[d]	1.90±.0.03	1.91±.0.04	1.96±.0.05	1.99±.0.04	1.96±.0.05	1.96±.0.04
PER[e]	2.43±0.04	2.43±0.02	2.40±0.04	2.45±0.06	2.39±0.04	2.45±0.04
Survival[f] (%)	100	100	97	97	100	100

[a] Final weight-Initial weight; [b] Condition factor=100×(Final weight (g)/(Fork length (cm))^3, [c] Feed conversion ratio=dry feed weight/wet weight gain; [d] Specific growth rate = 100×(ln final weight-ln initial weight /Day); [e] Protein efficiency ratio= 100×Wet weight/Protein intake, [f] 100×(Initial fish number-Dead fish number)/(Initial fish number).

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dietary n-3 HUFAs content.

Body Composition and Fatty Acids

No significant differences in the percentage of moisture, protein and lipid were found among treatments (Table 4). Total fatty acid compositions of beluga carcass fed with different lipids are given in Table 5. Fatty acid compositions of fish showed pronounced changes from the initial companion which was related to the change of fatty acid composition in experimental diets. The composition of many fatty acids in beluga carcass was significantly ($P< 0.05$) affected by dietary treatments. Diets with high amounts of canola oil, resulted in carcasses with more total monounsaturated fatty acids levels especially oleic acid (18:1n-9). On the other hand, in the groups fed with diets containing more Kilka fish oil, the total n-3 fatty acids were higher.

Linolenic acid was negatively correlated to the dietary fish oil concentration and the level of this fatty acid in the beluga carcass was less than that in the dietary lipids, especially when the fish consumed diets 1, 2 and 3 (Figure 1). In contrast to linolenic acid, EPA and DHA were found to be positively related to the dietary fish oil concentration and the levels of these fatty

Table 4. Body composition of beluga juvenile fed with experimental diets.

Body composition	Diet no Initial	1	2	3	4	5	6
Moisture	75.28±0.08	76.13±0.5	76.05±0.24	75.67±0.54	76.25±0.43	75.71±0.29	75.00±0.38
Protein	15.85±0.14	14.81±0.26	14.89±0.18	15.10±0.11	14.80±0.22	15.25±0.34	15.33±0.23
Fat	6.36±0.23	6.40±0.3	6.62±0.19	6.81±0.27	6.75±0.39	6.90±.08	7.00±0.36
Ash	2.50±0.05	2.65±0.02	2.30±0.02	2.42±0.01	2.14±0.08	2.14±0.04	2.67±0.03

Table 5. Fatty acid composition of beluga juvenile carcass (area %) fed with the experimental diets.

Fatty acids	Experimental diets 1	2	3	4	5	6	initial
∑ Saturates	14.63±0.28^{a}	14.69±0.74^{a}	16.10±0.52^{ab}	16.86±0.58^{b}	17.91±0.54^{bc}	19.26±0.80^{c}	21.52±0.83
C14:0	1.91±0.45^{a}	2.03±.21^{a}	2.16±.05^{a}	2.32±0.10^{a}	2.56±0.12^{ab}	3.20±0.19^{b}	3.66±0.32
C16:0	10.68±0.43^{a}	10.60±0.61^{a}	11.71±0.62^{ab}	12.3±0.85^{ab}	13.06±0.50^{b}	13.50±0.60^{b}	15.52±0.68
C18:0	1.90±0.16^{a}	1.94±0.20^{a}	2.07±0.13^{a}	2.10±0.0^{a}	2.13±0.11^{a}	2.35±0.30^{a}	2.13±0.40
C20:0	0.10±0.00^{a}	0.10±0.00^{ab}	0.08±0.00^{ab}	0.10±0.00^{ab}	0.13±0.00^{bc}	0.16±0.00^{c}	0.16±0.00
∑Monoenes	49.24±1.31^{d}	48.35±0.61^{cd}	46.36±0.71^{bcd}	45.29±1.20^{bc}	44.09±0.69^{b}	40.08±0.66^{a}	37.23±0.81
C14:1n5	0.02±0.00^{a}	0.10±0.00^{ab}	0.12±0.07^{ab}	0.11±0.00^{ab}	0.15±0.00^{ab}	0.24±0.11^{b}	0.10±0.00
C16:1n7	2.80±0.11^{a}	3.20±0.14^{a}	3.14±0.44^{a}	3.37±0.21^{ab}	3.92±0.40^{ab}	4.57±.56^{b}	4.96±0.21
C18:1n7	2.89±0.10^{a}	2.70±0.10^{a}	2.72±0.21^{a}	2.65±0.04^{a}	2.57±0.02^{a}	2.42±0.06^{a}	4.24±0.23
C18:1n9	41.67±1.20^{e}	40.58±0.84^{de}	38.67±0.54^{cd}	37.43±1.01^{bc}	35.70±0.69^{b}	31.27±0.57^{a}	26.24±0.80
C20:1n9	1.82±0.21^{a}	1.74±0.15^{a}	1.70±0.04^{a}	1.71±0.03^{a}	1.70±0.32^{a}	1.56±0.00^{a}	1.66±0.01
∑ n-6	15.45±0.67^{c}	15.10±0.53^{bc}	14.66±0.69^{bc}	14.09±0.56^{bc}	13.50±0.20^{b}	10.92±0.40^{a}	10.14±0.50
C18:2n6	14.35±0.46^{c}	13.97±0.31^{bc}	13.46±0.76^{bc}	12.92±0.42^{bc}	12.27±0.20^{b}	9.67±0.62^{a}	9.37±0.50
C20:2n6	0.71±0.16^{a}	0.69±0.07^{a}	0.70±0.00^{a}	0.64±0.00^{a}	0.68±0.00^{a}	0.63±0.09^{a}	0.00±0.00
C20:4n6	0.39±0.00^{a}	0.43±0.00^{a}	0.46±0.18^{a}	0.50±0.09^{a}	0.54±0.04^{a}	0.61±0.11^{a}	0.74±0.24
∑ n-3	11.38±0.58^{a}	12.35±0.83^{ab}	13.93±0.41^{bc}	17.51±0.30^{cd}	19.03±0.24^{d}	22.86±0.66^{e}	23.25±0.79
C18:3n3	5.01±0.40^{a}	4.82±0.23^{a}	4.90±0.15^{a}	4.96±0.35^{a}	4.72±0.06^{a}	3.93±0.21^{a}	3.23±0.50
C20:4n3	0.10±0.04^{a}	0.13±0.00^{a}	0.22±0.01^{a}	0.25±0.06^{a}	0.31±0.04^{a}	0.45±0.09^{a}	1.42±0.05
C20:5n3	2.42±0.12^{a}	2.81±0.15^{a}	3.32±0.31^{ab}	3.90±0.33^{bc}	4.57±0.21^{c}	6.18±0.45^{d}	8.03±0.57
C22:5n3	0.32±0.10^{a}	0.30±0.02^{a}	0.37±0.05^{a}	0.46±0.01^{ab}	0.72±0.10^{bc}	1.00±0.24^{c}	0.71±0.05
C22:6n3	3.53±0.21^{a}	4.26±0.36^{ab}	5.10±0.28^{bc}	5.97±0.34^{cd}	6.97±0.41^{d}	9.56±0.48^{e}	9.02±0.49
∑n-3 HUFA	6.38±0.32^{a}	7.52±0.61^{ab}	9.04±0.10^{bc}	10.61±0.69^{cd}	12.62±0.14^{d}	17.23±0.44^{e}	19.22±0.77
DHA/EPA	1.46±0.01^{a}	1.51±0.05^{a}	1.52±0.06^{a}	1.53±0.06^{a}	1.54±0.03^{a}	1.55±0.03^{a}	1.12±0.01

Different superscript letters within each row represent significant differences ($P< 0.05$). Statistics not performed on the initial sample.

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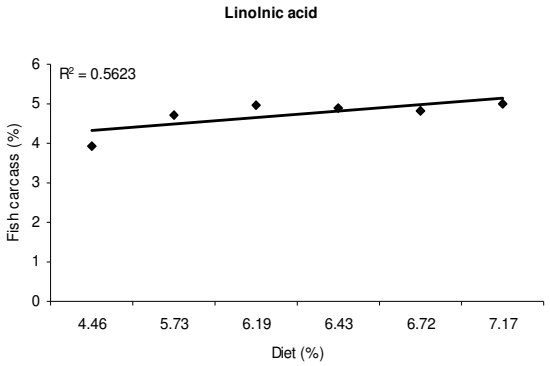

Figure 1. Relationships between dietary fatty acid concentrations and carcass fatty acid concentrations of linoleic acid.

acids in the beluga carcass were higher than that in the dietary lipids, especially when the fish consumed diets 1, 2 and 3 (Figure 2).

Haematocrit and Plasma Biochemical Parameters

The effects of dietary treatments on the biochemical blood parameters and haematocrit value are given in Table 6. There were no significant differences in plasma protein, glucose, cholesterol and triglyceride concentrations among dietary treatments. These results indicated that biochemical blood parameters were not affected by n-3 HUFAs content. The lowest haematocrit values were related to diets 1 and 2 and were significantly different from other treatments ($P< 0.05$).

DISCUSSION

This is the first study in which vegetable oil has been replaced by fish oil in low fish meal diets for beluga juvenile. Results of growth performance indicated that the level of n−3 HUFAs was not a restrictive factor, but most fatty acids in carcass were significantly affected by dietary treatments. This finding agrees with a previous study where total replacement of dietary fish oil with vegetable oils in low fish meal diets for white sturgeon, *Acipenser transmontanus,* did not cause any significant difference in

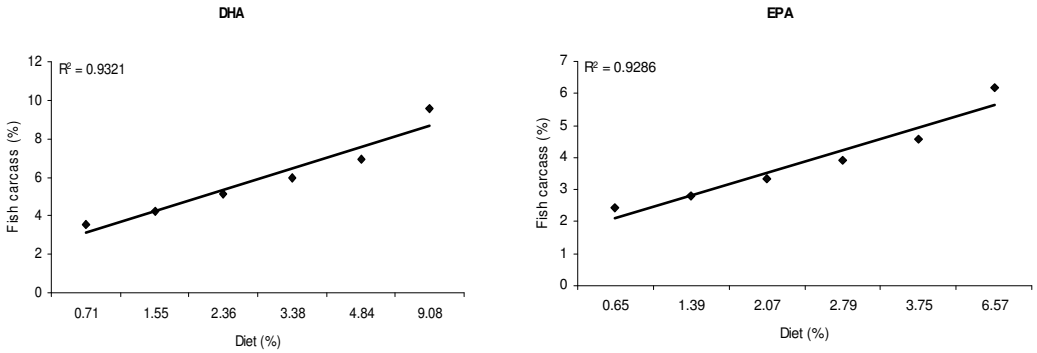

Figure 2. Relationships between dietary fatty acid concentrations and carcass fatty acid concentrations of DHA and EPA.

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Table 6. Haematocrit and plasma biochemical parameters of beluga juvenile fed with experimental diets.

Diets	Plasma protein ($g L^{-1}$)	Plasma glucose ($g L^{-1}$)	Plasma triglyceride ($g L^{-1}$)	Plasma cholesterol ($g L^{-1}$)	Haematocrit (%)
1	10.6±1.09	0.36±0.03	6.60±0.84	0.17±0.01	27.0±0.58[a]
2	10.30±1.10	0.34±0.01	6.12±0.43	0.17±0.01	27.1±0.81[a]
3	10.68±1.36	036±0.00	6.72±0.61	0.18±0.01	29.6±0.90[b]
4	12.04±0.13	0.37±0.00	5.64±0.92	0.19±0.01	30.6±0.52[b]
5	13.10±0.94	0.37±0.02	5.67±0.78	0.20±0.01	31.7±0.29[b]
6	11.06±1.97	0.37±0.02	5.56±0.66	0.22±0.00	31.6±0.77[b]

Different superscript letters within each column represent significant differences (P<0.05).

growth performance (Ruping *et al*., 1993).

Subhadra *et al*. (2006) indicated that fish growth efficacy can be affected by diet composition and feeding trial duration. For example, Llorens *et al*. (2007) suggested that the dietary soybean oil levels (0%, 24%, 48% and 72%) did not influence the growth performance of gilthead sea bream *Sparus aurata L.* until 211 days. However, at the end of the trial (day 309), fish feeding the 72% soybean oil diet weighed the lowest. Weight changes depending on trail duration may have occurred in our study if experimental duration was longer.

The relative ratio of fish versus vegetable oil, in diets 1 to 6, showed a gradual decrease of the typical fatty acids of vegetable oils (18:1n-9, 18:2n-6 and 18:3n-3) and an increase in marine type fatty acids (22:6n-3, 22:5n-3 and C20:5n3).According to fatty acid profile, fish tend to reflect diet composition. As reported in several species (Bell *et al*., 1991; Ruping *et al*., 1993; Sargent *et al*., 1995; Sener *et al*., 2005; Rinchard *et al*., 2007), fatty acid concentrations of beluga are influenced by the fatty acid composition of the dietary lipids.

In the groups feeding feed containing more n-3 fish oil, total n-3 fatty acids in the carcass were higher than those in the groups feeding feed containing more vegetable oil (Tables 2 and 5). However, the 22:6n-3 and C20:5n3 fatty acids were higher than the feed values in the carcass of fish fed with diets containing canola oil. These results indicated that the beluga juvenile has the ability to elongate 18:3n-3 to 20:5n-3 and 22:6n-3 fatty acids because 18:3n-3 was lower in the carcass than diets. On the contrary, 20:5n-3 and 22:6n-3 in the carcass of fish were higher than in the diets. These suggest that 22:6n-3 and 20:5n3 are essential fatty acids for this fish. Similar results were reported for the white sturgeon (Ruping *et al*., 1993; Deng *et al*., 1998) and in Russian Sturgeon, *Acipenser gueldenstaedtii* juveniles (Sener *et al*., 2005).

In all groups, moisture, protein and lipid showed no significant differences. This can be attributed to the isoproteic and isolipidic experimental diets. A similar phenomenon was observed in another study for the white sturgeon (Ruping *et al*., 1993).

Diets containing vegetable oils are rich in oleic acid, linoleic acid and linolenic acid and these fatty acids tend to reduce plasma cholesterol concentration (Dietschy, 1998; Fernandez and West, 2005). Our study results demonstrated that increasing fish oil in experimental diets, led to nonsignificant increases in plasma cholesterol concentration. Peng *et al*. (2008) announced that replacement of fish oil with soybean oil in juvenile black seabream, *Acanthopagrus schlegeli,* at levels of 60 to 100% had no significant effect on plasma cholesterol concentration, but a diet containing only fish oil showed a significant difference, while plasma triglyceride and plasma protein showed no significant differences between the experimental treatments.

According to these results (in an 8 week period), the beluga can utilize canola oil in diets well and it is possible to substitute fish oil by vegetable oil such as canola oil,

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without adverse effects on growth and feed utilization.

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نیازمندی به اسیدهای چرب چندغیراشباعی (n-3 HUFAs) در جیره غذایی فیل ماهی جوان *Huso huso* و اثرات آن روی برخی شاخص‌های رشد، کیفیت لاشه وترکیبات اسید چرب لاشه

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چکیده

مطالعه تغذیه‌ای به‌مدت ۸ هفته برای تعیین اثرات سطوح مختلف اسیدهای چرب چندغیراشباعی (n-۳ HUFAs) روی رشد و ترکیبات اسیدهای چرب انجام شد. شش جیره غذایی حاوی سطوح مختلف n-3 HUFAs (۱/۵۶ تا ٪ ۱۷/۲۵ از اسید چرب کل)، برای تعیین نیازمندی فیل ماهی به این اسیدهای چرب مورد استفاده قرار گرفتند. در وزن به‌دست آمده، ضریب تبدیل غذایی، فاکتور وضعیت، نرخ رشد ویژه و شاخص کارایی پروتئین، همچنین در ترکیبات لاشه فیل ماهی تفاوت معنی‌داری در بین تیمارهای مختلف تغذیه‌ای مشاهده نشد (P> ۰/۰۵). تفاوت معنی‌داری بین پروتئین، گلوکز، کلسترول و تری گلیسرید پلاسمای خون در تیمارهای آزمایشی وجود نداشت (P> ۰/۰۵). در صورتی که درصد هماتوکریت به‌طور معنی‌داری در جیره‌های غذایی ۱و۲ کمتر بود (P< ۰/۰۵). همراه با تغییرات ترکیبات اسیدچرب جیره‌های غذایی، ترکیبات اسید چرب لاشه فیل ماهیان جوان، نسبت به نمونه‌های اولیه تغییر نشان داد. با افزایش سطوح n-3 HUFAs در جیره غذایی، سطوح n-3 HUFAs لاشه فیل ماهیان جوان نیز افزایش یافت. نتایج به‌دست آمده پیشنهاد می‌کند که حداقل سطوح n-۳ HUFAs در جیره غذایی فیل ماهی جوان اثر منفی در رشد ایجاد نمی کند و n-3 HUFAs عامل محدود کننده در رشد این ماهی نیست.

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