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Effect of black cumin oil (*Nigella sativa*) on the growth performance, body composition and fatty acid profile of rainbow trout (*Oncorhynchus mykiss*)

ÖZ M.[1]*; Dikel S.[2]; Durmus M.[3]

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Abstract

In this study, rainbow trout were fed for a total of 144 days with different feed mixes containing 0.00%, 0.10%, 0.40%, 0.70%, 1.00% and 1.30% black cumin oil. The effect of black cumin oil on growth performance, feed content and fatty acid composition of rainbow trout was researched. The starting weight of fish in the experimental group was approximately 60 g, which increased to 215.21±4.12g, 217.32±3.14g, 235.12±2.9g, 240.21±6.5g, 260.14±4.7g, and 258.89±1.6g, respectively and the differences in growth between the groups were observed to be statistically significant ($p<0.05$). Black cumin oil decreased the feed conversion rate of the fish. The addition of black cumin oil in the fish feed increased feed consumption and the daily feed consumed per fish for each group was as follows: 0.93g, 0.93g, 0.97g, 1.00g, 1.02g and 1.02g, respectively. At the end of the experiment the economic conversion rates of the fish ($ Kg^{-1}) were between 1.58 and 1.67, while the economic benefit index was between 0.60 and 0.72. The study showed that protein efficiency rates and specific growth rates of the experimental groups had increased. The addition of black cumin oil to the feed increased the proportions of raw protein, oil and raw cinder in the trout and decreased the proportion of moisture, while also increasing the amounts of linoleic, oleic and stearic acids.In conclusion, the addition of 1.00% black cumin oil in feeds increases growth performance in fish and decreases production costs.

Keywords: Black cumin oil, Rainbow trout, Growth performance, Body composition, Fatty acid profile

1-Department of Fisheries and Diseases, Faculty of Veterinary Medicine, Aksaray University, Aksaray, Turkey
2-Department of Aquaculture, Faculty of Fisheries, Cukurova University, 01330, Adana, Turkey.
3-Department of Seafood Processing Technology, Faculty of Fisheries, Cukurova University, 01330, Adana, Turkey.
*Corresponding author's Email: ozmustafa@aksaray.edu.tr

Introduction

Due to the rich nutritional content, fish is an important food source in both developed and developing countries. Fish oils contain essential fatty acids and poly unsaturated fatty acids (PUFA), which are important for human health. There is an increasing demand from consumers for healthy, high quality, natural and fresh fish products.

There is a limit to the amount of fish that can be obtained through commercial fishing in both fresh and salt-water. Therefore, it is imperative that aquaculture products, which fulfil an important protein need for human health, are obtained through cultivation. In this case, obtaining the highest volume of production with the lowest cost per unit of area and within the shortest time is a significant challenge. In achieving this goal, great strides have been made with the introduction of biotechnological methods (Melamed *et al*., 2002). However, in order to achieve the required efficiency levels, the correct feeding program must be implemented. Rainbow trout, part of the Salmonidae family (*Oncorhynchus mykiss*), is a commercially significant species consumed widely in Europe (Çakli *et al*., 2006). It is a species of fish with an exceptionally fast growth rate and high nutritional value, leading to widespread cultivation in a variety of countries.

Black cumin has been cultivated since ancient times and the nutritional content is as follows: 20.8% raw protein, 3.7% raw cinder, 7.0% moisture, 34.8% lipids and 33.7% carbohydrates (Atta, 2003). The pharmacological properties of black cumin according to the chemical composition are as follows: antibacterial, antifungal, antiviral, antiprotozoan, antihistaminic, antioxidant, non-inflammatory and immuno-stimulant. Black cumin is used in the treatment of a range of diseases, including asthma, hypertension, inflammation, coughing, bronchitis, headaches, eczema, influenza, fever and vertigo (Antınterim, 2010).

Volatile black cumin oil is active in a number of chemical reactions (Ali and Blunden, 2003). It has antihistaminic, anti-inflammatory and anti-infective properties, causes bronchial dilatation, is an inhibitor of protein kinase C which is known to be a triggering compound for crystallised nigellon histamine secretion, the essential oils regulate the immune system and allergic reactions, support the metabolism, decrease cholesterol and blood sugar, stimulate bone marrow to increase interferon production and contain cofactors essential for enzyme reactions for trace elements.

In cultivation, increasing the growth performance of the fish is one of the most important goals. A number of studies have thus been performed on the subject. Some of these studies examine the effect of natural immune-stimulant additives to fish feed. The addition of oils into feeds given to aquatic animals, especially fish, increases the growth performance of the species being cultivated, as well as feed consumption and protein utilisation rates (Bell *et al*., 2000; Montero *et al*., 2005). In our study, we aimed to examine the gains in growth and live weight in trout for household consumption using feed enriched with black cumin oil and to determine the effects of this feed on carcass composition and the fatty acid profile.

In recent years, a number of studies were performed in the area of aquaculture regarding probiotic and prebiotic substances to regulate the population of bacteria in the water, to decrease the pathological bacterial load, increase the water quality, increase live weight and the effects of benefits from feed on the immune system (Irianto and Austin, 2002; Korkut *et al*., 2003; Vine *et al*., 2004). In the light of the studies performed regarding probiotics and prebiotics, it has been illustrated that the usage of these substances in aquaculture has positive effects of benefiting from feed, live weight gain, survival rate, increase in growth performance and the immune system, regardless of the species and age of the species cultivated (Panigrahi *et al*., 2004; Salinas *et al*., 2005; El-Haroun *et al*., 2006; Kumar *et al*., 2006; Bagheri *et al*., 2008).

In this study, through the addition of black cumin oil in trout feed, it was aimed to achieve growth to market size in a shorter period of time or to achieve more live weight gain within the same time period. Due to the rich vitamin, iron, selenium and zinc content of black cumin oil, we aimed to achieve a higher rate of benefiting from the feed and to allow a faster and higher growth rate using the same feed.

Materials and methods

Fish and sampling

The feeding activities were performed for 144 days at a private trout farm (Öz Alabalık Üretim Tesisi, Adana, Turkey). 1,800 rainbow trout (*O. mykiss*) were used in the study. 18 concrete pools with dimensions of 4m x 1m x 1.20m and each containing 100 fish were used. At the beginning of the experiment, the mean live weights of the fish were 59.86±0.01g, 59.82±0.00 g, 59.77±0.00g, 59.82±0.00g, 59.66±0.01g and 59.82±0.00g, respectively. The water temperature and oxygen content of the pools used in the experiment were checked twice daily using an Oxyguard brand oxygen meter and the water temperatures measure between 1°C and 13°C, while the oxygen dissolved in the water was between 9.9 mg L^{-1} and 12.35 mg L^{-1}. Throughout the experimental period the fish were fed ad libitum twice a day.

Diet preparation

During the study, commercial trout feed obtained from a private company (Abalıoğlu, Denizli, Turkey) was used. Cold pressed black cumin oil was added to the fish feed using a spraying technique and the feed was mixed until homogenised. The amounts of black cumin oil added to the feed used in the study are 0.00%, 0.10%, 0.40%, 0.70%, 1.00% and 1.30%. Table 1 shows the nutritional values of the feed and Table 2 shows the fatty acid content of the experimental feed. Sun flower oil was added to the experimental feeds to unify the lipid levels of fish feed in the study. The amounts of sun flower oil added to fish feed were 1.30%, 1.00%, 0.70%, 0.40%, 0.10% and 0.00%, respectively.

The fatty acid profile of the black cumin oil produced using a cold press and added to the trout feed is given under Table 3.

Table 1: Nutritional values of the Abalıoğlu trout feed used in the study.

Proximate composition	Mean (%)
Crude protein	42
Lipid	22
Crude cellulose	3
Moisture	10
Crude ash	12
Total energy (kcal kg^{-1})	4350 KCal Kg^{-1}

Table 2: Fatty acid profile of the feed used in the experiment.

	Fatty acid profile of the feed					
Fatty acid	**Group 1**	**Group 2**	**Group 3**	**Group 4**	**Group 5**	**Group 6**
C14:0	3.58±0.06^{a}	2.81±0.12^{b}	2.84±0.06^{b}	2.91±0.04^{b}	2.74±0.04^{b}	2.80±0.02^{b}
C14:1	0.13±0.01^{a}	0.10±0.01^{b}	0.10±0.01^{b}	0.09±0.01^{b}	0.09±0.00^{b}	0.09±0.00^{b}
C16:0	12.92±0.17^{a}	10.94±0.30^{b}	10.74±0.19^{bc}	10.26±0.21^{cd}	10.36±0.25^{cd}	10.15±0.10^{d}
C16:1	4.5±0.45^{a}	4.17±0.21^{ab}	3.84±0.24^{b}	3.76±0.07^{b}	3.75±0.03^{b}	3.65±0.08^{b}
C17:0	0.35±0.03^{a}	0.35±0.02^{a}	0.29±0.01^{b}	0.28±0.01^{b}	0.28±0.02^{b}	0.28±0.01^{b}
C17:1	0.11±0.00^{a}	0.11±0.01^{a}	0.1±0.00^{b}	0.10±0.01^{b}	0.10±0.00^{b}	0.10±0.00^{b}
C18:0	2.93±0.07^{a}	2.43±0.31^{a}	2.80±0.01^{a}	2.86±0.02^{a}	2.90±0.00^{a}	2.92±0.04^{a}
C18:1 n9	32.60±0.70^{ab}	33.27±0.15^{a}	32.53±0.39^{ab}	32.70±0.52^{ab}	33.89±0.26^{a}	31.71±0.09^{b}
C18:2 n6	11.27±0.15^{d}	12.87±0.02^{c}	13.21±0.04^{bc}	13.92±0.01^{b}	14.84±0.04^{a}	14.81±0.24^{a}
C18:3 n3	3.39±0.04^{c}	4.11±0.24^{a}	3.79±0.06^{b}	3.75±0.02^{b}	3.68±0.03^{b}	3.80±0.09^{b}
C20:0	0.24±0.00^{a}	0.26±0.00^{a}	0.25±0.00^{a}	0.28±0.01^{a}	0.24±0.06^{a}	0.24±0.06^{a}
C20:1n9	4.59±0.00^{a}	3.73±0.23^{b}	4.25±0.18^{ab}	3.84±0.36^{b}	4.16±0.01^{ab}	3.67±0.40^{b}
C20:2	0.49±0.02^{ab}	0.54±0.08^{a}	0.40±0.04^{b}	0.38±0.01^{b}	0.38±0.03^{b}	0.38±0.03^{b}
C20:3n6	0.43±0.00^{a}	0.36±0.00^{b}	0.35±0.00^{b}	0.34±0.00^{b}	0.33±0.00^{b}	0.33±0.00^{b}
C20:4 n6	4.41±0.31^{a}	3.95±0.00^{ab}	3.84±0.13^{ab}	3.95±0.02^{ab}	3.68±0.15^{b}	3.72±0.17^{b}
C20:5 n3	4.53±0.02^{a}	3.10±0.15^{c}	3.53±0.04^{b}	3.38±0.27^{bc}	3.29±0.04^{bc}	3.14±0.02^{c}
C22:1n9	0.11±0.01^{a}	0.10±0.01^{b}	0.09±0.01^{b}	0.10±0.01^{b}	0.09±0.01^{b}	0.09±0.01^{b}
C23:0	0.13±0.01^{ab}	0.15±0.01^{a}	0.11±0.01^{c}	0.11±0.00^{c}	0.11±0.00^{c}	0.12±0.01^{bc}
C24:0	1.98±0.25^{a}	1.86±0.04^{ab}	1.73±0.02^{ab}	1.72±0.01^{ab}	1.67±0.01^{b}	1.63±0.09^{b}
C22:6 n3	5.17±0.22^{a}	5.44±0.10^{a}	5.33±0.08^{a}	5.41±0.20^{a}	5.34±0.32^{a}	5.47±0.38^{a}
SFA	22.15±0.60^{a}	18.81±0.76^{b}	18.77±0.09^{b}	18.43±0.31^{b}	18.32±0.29^{b}	18.16±0.01^{b}
MUFA	39.94±1.85^{a}	39.80±1.02^{a}	38.96±1.24^{a}	38.74±1.02^{a}	38.21±0.61^{a}	37.75±0.98^{a}
PUFA	29.52±0.66^{a}	30.12±0.21^{a}	30.32±0.36^{a}	31.01±0.23^{a}	31.40±0.01^{a}	31.53±0.38^{a}

Each value indicates the mean ± standard deviation (n=3). The means expressed using different letters in each row are significantly different (p<0.05).

Table 3: Fatty acids of the black cumin oil added to the fish feed.

Fatty acids of the black cumin oil		(%)
C14:0	Miristic acid	0.46±0.02
C14:1	Miristoleic acid	0.72±0.27
C16:0	Palmitic acid	12.47±0.08
C16:1	Palmitoleic acid	1.10±0.22
C18:0	Stearic acid	3.45±0.11
C18:1 n9	Oleic acid	27.37±0.82
C18:2 n6	Linoleic acid	49.72±1.44
C18:3 n3	Linolenic acid	0.355±0.03
C20:1	Ekosenoic acid	2.295±0.02

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Growth performance

Specific growth rate (SGR body weight day^{-1}); [(ln w1 ± ln w0) (t1-t0)±1] 100, where w1 and w0 are wet weight at times t1 and t0.

The economic conversion ratio (ECR, US$ kg^{-1}) = (feed cost (US$$^{-1}$) + black cumin oil cost (US$kg^{-1})) x feed conversion ratio (kg diet kg^{-1}fish),

Feed conversion ratio (FCR); (W_{final}-$W_{initial}$)/consumed feed, where W_{final} and $W_{initial}$ are live weights (g) of the fish at day initial (t) and final (T), respectively.

The economic profit index [EPI (US$ fish^{-1}) = final weight (kg fish^{-1}) x fish sale price (US$ kg^{-1})-ECR (US$ kg^{-1}) x weight increase (kg)] developed by Martinez-Llorens *et al*., (2007).

Protein efficiency ratio (PER)= Weight gain (g)/protein intake (g), (Skalli et al, 2004).

FAME analyses

Lipid extraction was made according to Bligh and Dyer method (1959). Methly esters were prepared by transmethylation using 2M KOH in methanol and *n*-heptane according to the method as described by Ichibara, Shibahara, Yamamoto and Nakayama (1996) with minor modification. 10 mg of extracted oil was dissolved in 2mL *n*-heptane followed by 4mL of 2M methanolic KOH. The tube was then vortexed for 2 min at room temperature. After centrifugation at 4000 rpm for 10 min, hexane layer was taken for GC analyses.

Proximate analysis

The fish samples were analysed in triplicate for proximate composition: lipid content of rainbow trout by the Bligh and Dyer (1959) method, moisture and the ash content of fish by AOAC (1990) method, total crude protein by Kjeldhal method (AOAC, 1984).

Gas chromatographic condition

The fatty acid composition was analysed by GC Clarus 500 with autosampler (Perkin Elmer, USA) equipped with a flame ionization detector and a fused silica capillary SGE column (30 m X 0.32 mm ID X 0.25 μm BP20 0.25 UM, USA). The oven temperature was held at 140 ^{o}C for 5 min, raised to 200 ^{o}C at the rate of 4 ^{o}C min^{-1} and held at 220^{o}C at rate 1 ^{o}C min^{-1}, while the injector and the detector temperature were set at 220 ^{o}C and 280 ^{o}C, respectively. The sample size was 1 μL and the carrier gas was controlled at 16 ps. The split used was 1:50. Fatty acids were identified by comparing the retention times of FAME with the standard 37 component FAME mixture. Two replicate GC analyses were performed and the results expressed in GC area % as a mean value and ±standard deviation.

Statistical analysis

The mean value and standard deviation were calculated from the data obtained from the three samples for each treatment. One way ANOVA was used to determine the significance of differences at $p<0.05$. All statistics were performed using SPSS 15.0 for

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Windows (SPSS Inc., Chicago, IL. USA).

Results

The growth parameters achieved from the study are shown in Table 4 and figure 1. The starting weight of fish in the experimental group was approximately 60 g, which increased to 215.21±4.12g, 217.32±3.14g, 235.12±2.9g, 240.21±6.5g, 260.14±4.7g, and 258.89±1.6g, respectively while the difference in growth between the groups was observed to be statistically significant (p<0.05). The mean weights were measured once every 15 days throughout the study and the live weight gain values are shown in

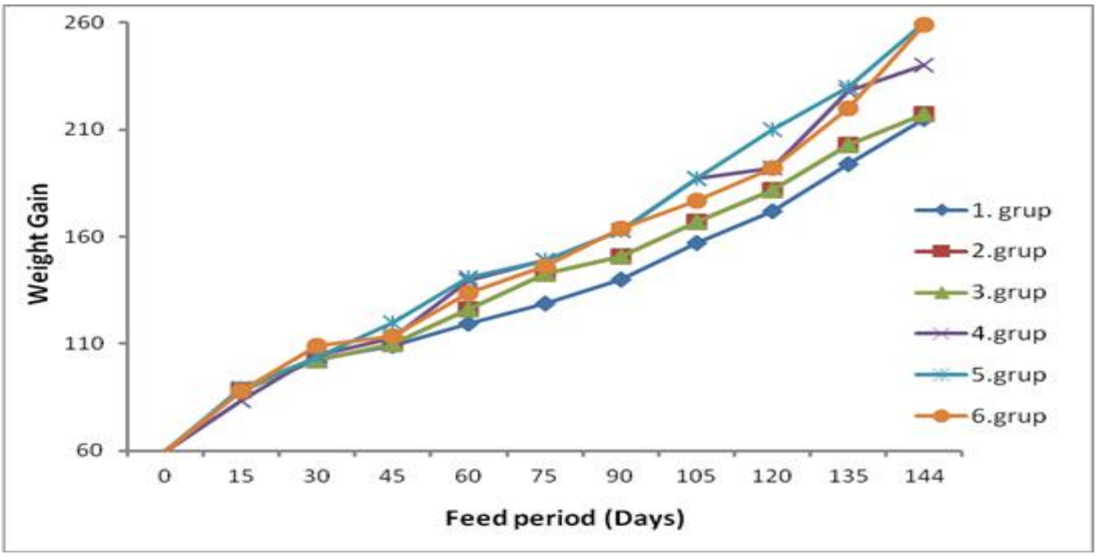

Figure 1: Live weight gain (g) for rainbow trout throughout the experiment.

Table 4: Growth parameters of trout after 144 days of feeding.

	Research Groups					
	1.Group	**2.Group**	**3.Group**	**4.Group**	**5.Group**	**6.Group**
IFW (g)	59.86±0.01^a	59.82±0.00^a	59.82±0.00^a	59.77±0.00^a	59.66±0.01^a	59.82±0.00^a
FW (g)	215.21±4.12^f	217.32±3.14^e	235.12±2.9^d	240.21±6.5^c	260.14±4.7^a	258.89±1.6^b
WG (g)	155.35	157.48	175.3	180.44	200.48	199.07
SGR (%)	0.89±0.01^e	0.90±0.01^d	0.95±0.01^c	0.97±0.01^b	1.02±0.01^a	1.01±0.01^a
FI (g)	133.33±0.01^e	133.33±0.01^e	140±0.00^d	143.75±0.04^c	155.30±0.01^a	154.50±0.01^b
DFI(g)	0.93±0.01^d	0.93±0.00^d	0.97±0.00^c	1.00±0.01^b	1.02±0.00^a	1.020.00^a
FCR	0.86±0.00^a	0.85±0.00^b	0.80±0.00^c	0.80±0.00^c	0.77±0.001^d	0.78±0.01^d
PER	2.77±0.00^d	2.81±0.01^c	2.98±0.01^b	2.98±0.01^b	3.07±0.01^a	3.07±0.01^a
SUR(%)	100	100	100	100	100	100
ECR($ Kg^{-1})	1.67±0.04^a	1.65±0.01^b	1.59±0.01^e	1.61±0.00^d	1.58±0.01^f	1.62±0.01^c
EPI	0.60±0.00^d	0.61±0.00^d	0.66±0.01^c	0.67±0.01^c	0.72±0.01^a	0.71±0.00^b

Each value indicates the Mean ± standard deviation. The means expressed using different letters in each row are significantly different (p<0.05).

IFW: Initial fish weight, FW: Final weight, WG: Weight gain, SGR: Specific growth rate, FI: Feed intake, DFI; Daily feed intake, FCR: Feed conversion rates, PER: Protein efficiency ratio, SUR: Survival, ECR: Economic Conversion rate, EPI: Economic profit index.

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Body composition

The composition of the feed including black cumin oil given to rainbow trout for 144 days was analysed in terms of nutrition and the results are given under Table 5. The crude protein content of the rainbow trout was 18.79%, 19.99%, 20.12%, 20.12%, 20.29%, 20.60% and 20.37%, respectively. The black cumin oil affected protein content and the differences between groups were statistically significant (p<0.05). In the study, the control group had the highest moisture content (74.57%) while Group 6 had the lowest moisture content (71.34%). The total amount of lipids was 4.56%, 4.63%, 5.24%, 5.30%, 5.60% and 5.89%, respectively. According to these results, the amount of black cumin oil in the feed increased the total amount of lipids in the fish. There were also statistically significant differences in the crude ash content observed in the study (p<0.05). The crude ash content varied between 1.94% and 2.27%, with the highest amount observed in Group 6 and the lowest observed in the control group.

Table 5: Basic nutritional values of the rainbow trout after the experiment.

	Body Composition			
Groups	**Crude Protein**	**Moisture**	**Lipid**	**Crude ash**
Group1	18.79±0.01^{f}	74.57±0.01^{a}	4.56±0.00 ^{f}	1.94±0.01 ^{f}
Group2	19.99±0.01 ^{e}	73.28±0.01^{b}	4.63±0.05 ^{e}	2.04±0.00 ^{e}
Group3	20.12±0.01 ^{d}	72.35±0.01^{c}	5.24±0.01 ^{d}	2.09±0.00 ^{d}
Group4	20.29±0.00 ^{c}	72.25±0.01^{d}	5.30±0.01 ^{c}	2.13±0.01 ^{c}
Group5	20.60±00 ^{a}	71.62±0.00^{e}	5.60±0.01 ^{b}	2.16±0.00 ^{b}
Group6	20.37±0.01 ^{b}	71.34±0.01^{f}	5.89±0.01 ^{a}	2.27±0.01 ^{a}

Each value indicates the mean standard deviation, n=3 (Each analysis was performed 3 times for each group from 12 mixed fish meat). The means expressed using different letters in each row are significantly different (p<0.05).

Fatty acid profile

The fatty acid composition of rainbow trout fed with feeds containing black cumin oil is given in Table 6.

The basic fatty acids for all groups in the study are as follows: miristic acid (C14:0, 2.44-3.12%), palmitic acid (16:0, 10.07-12.18%), palmitoleic acid (C16:1, 3.70-5.13%), stearic acid (18:0, 1.98-3.05%), vaccenik acid (C18:1n7, 3.03-4.22%), oleic acid (18:1n9, 19.62-31.08%), linoleic acid (C18:2n6, 11.14-13.97%), linolenic acid (C18:3n3, 2.68-3.45%), arachidic acid (C20:0, 3.14-3.88), ecosadienoic acid (C20:2 cis, 0.52-0.71%), EPA (20:5ω3, 1.83-2.10%), DHA (22:6ω3, 10.20-14.76%).

Table 6: Fatty Acid Composition of rainbow trout after the experiment.

Yağ Asitleri	Group1	Group2	Group3	Group4	Group5	Group6
C12:0	0.02±0.00$^{a.y}$	0.15±0.07 $^{a.x}$	0.01±0.00 $^{a.y}$	0.01±0.00$^{a.y}$	0.02±0.00 $^{a.y}$	0.01±0.00 $^{b.y}$
C14:0	2.49±0.21 $^{a.y}$	3.12±0.11 $^{ab.x}$	2.52±0.21 $^{a.y}$	2.46±0.25 $^{a.y}$	2.44±0.03 $^{a.y}$	2.49±0.17 $^{a.y}$
C16:0	10.07±0.32 $^{a.y}$	12.01±0.30 $^{a.x}$	10.85±0.07 $^{a.xy}$	12.18±1.33 $^{a.x}$	10.8±0.08 $^{ab.xy}$	10.84±0.22 $^{a.xy}$
C17:0	0.07±0.01 $^{bc.z}$	0.09±0.00 $^{ab.x}$	0.08±0.01 $^{a.yz}$	0.08±0.01 $^{ab.yz}$	0.07±0.00 $^{a.yz}$	0.08±0.00 $^{ab.xy}$
C18:0	2.77±0.22 $^{a.y}$	1.98±0.01 $^{a.z}$	2.80±0.04 $^{ab.xy}$	2.80±0.02 $^{a.xy}$	2.91±0.01 $^{a.xy}$	3.05±0.13 $^{a.x}$
C20:0	3.23±0.04 $^{ab.y}$	3.88±0.01 $^{abc.x}$	3.54±0.05 $^{a.xy}$	3.16±0.02 $^{a.y}$	3.47±0.06 $^{a.xy}$	3.14±0.54 $^{a.y}$
C23:0	0.17±0.02 $^{a.xy}$	0.19±0.01 $^{a.xy}$	0.16±0.01 $^{a.xy}$	0.20±0.01 $^{a.x}$	0.15±0.00 $^{a.y}$	0.15±0.03 $^{a.y}$
C24:0	0.34±0.03 $^{d.x}$	0.14±0.04 $^{c.x}$	0.50±0.01 $^{ab.x}$	0.30±0. 01 $^{a.x}$	0.46±0.00 $^{a.x}$	0.47±0.00 $^{a.x}$

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Table 6 continued:

SFA	19.13±1.16$^{a.y}$	21.54±1.21$^{abc.x}$	20.44±1.27$^{a.xy}$	18.17±1.14$^{a.y}$	20.30±1.23$^{a.xy}$	20.22±1.28$^{a.xy}$
C14:1	0.07±0.01$^{a.x}$	0.07±0.02$^{a.x}$	0.07±0.01$^{a.x}$	0.07±0.00$^{a.x}$	0.07±0.00$^{a.x}$	0.07±0.00$^{a.x}$
C16:1	4.31±0.04$^{a.xy}$	5.13±0.16$^{a.x}$	3.72±0.40$^{a.y}$	4.09±0.7$^{ab.xy}$	3.71±0.50$^{ab.y}$	3.70±0.08$^{a.y}$
C17:1	0.16±0.01$^{ab.xy}$	0.20±0.02$^{a.x}$	0.15±0.03$^{a.xy}$	0.16±0.2$^{ab.xy}$	0.13±0.00$^{bc.y}$	0.14±0.01$^{ab.y}$
C18:1n9	27.38±2.23$^{a.xy}$	19.62±0.01$^{c.z}$	29.53±0.57$^{a.xy}$	25.87±2.39$^{a.y}$	29.81±1.6$^{a.x}$	31.08±0.02$^{a.x}$
C18:1n7	4.22±0.34$^{a.x}$	3.03±0.01$^{a.x}$	3.23±0.24$^{ab.x}$	4.19±0.09$^{ab.x}$	3.73±0.98$^{a.x}$	3.81±0.38$^{a.x}$
C20:1	1.52±0.13$^{a.x}$	1.09±0.01$^{ab.x}$	0.97±0.01$^{c.x}$	1.01±0.01$^{a.x}$	1.08±0.01$^{ab.x}$	1.07±0.01$^{a.x}$
C22:1n9	0.22±0.04$^{a.x}$	0.16±0.00$^{a.y}$	0.13±0.00$^{a.y}$	0.12±0.03$^{a.y}$	0.14±0.01$^{a.y}$	0.12±0.00$^{a.y}$
MUFA	37.86±1.25$^{a.xy}$	29.29±1.1$^{a.z}$	37.7±1.5$^{a.xy}$	35.49±1.5$^{a.y}$	38.66±2.3$^{a.x}$	39.98±3.14$^{a.x}$
C18:2n6	11.14±1.23$^{a.z}$	13.97±0.72$^{a.x}$	11.86±0.45$^{a.yz}$	12.67±0.9$^{a.xyz}$	12.2±0.1$^{bc.xyz}$	13.1±0.04$^{ab.xy}$
C18:3n3	2.75±0.45$^{a.y}$	3.45±0.08$^{a.x}$	2.74±0.04$^{b.y}$	2.88±0.13$^{a.y}$	2.75±0.04$^{ab.y}$	2.68±0.00$^{ab.y}$
C20:2	0.57±0.03$^{a.yz}$	0.64±0.05$^{a.xy}$	0.55±0.02$^{bc.z}$	0.71±0.03$^{a.x}$	0.61±0.01$^{ab.yz}$	0.52±0.06$^{a.z}$
C20:3n6	0.47±0.02$^{a.x}$	0.43±0.04$^{a.x}$	0.32±0.00$^{c.x}$	0.35±0.03$^{a.x}$	0.38±0.01$^{a.x}$	0.33±0.00$^{b.x}$
C20:4n6	3.57±0.23$^{ab.yz}$	4.31±0.20$^{a.x}$	3.35±0.11$^{a.z}$	4.05±0.47$^{a.xy}$	3.18±0.00$^{ab.z}$	3.30±0.09$^{ab.z}$
C20:5n3	1.83±0.07$^{cd.z}$	2.09±0.01$^{bc.x}$	2.09±0.04$^{a.x}$	1.90±0.03$^{ab.yz}$	1.98±0.01$^{a.y}$	2.10±0.01$^{a.x}$
C22:2	0.27±0.03$^{a.z}$	0.39±0.02$^{a.x}$	0.32±0.02$^{a.y}$	0.06±0.00$^{a.w}$	0.04±0.00$^{b.w}$	0.05±0.01$^{a.w}$
C22:6n3	10.51±0.24$^{a.z}$	13.30±1.48$^{a.xy}$	11.49±1.16$^{a.xy}$	14.76±1.18$^{a.x}$	11.4±0.04$^{ab.xy}$	10.20±1.26$^{a.z}$
PUFA	32.09±2.02$^{a.y}$	38.56±2.78$^{a.x}$	32.70±1.72$^{a.y}$	37.37±2.06$^{a.x}$	32.66±1.61$^{a.y}$	32.33±1.48$^{a.y}$

Each value indicates the mean ± standard deviation, n=3 (Each analysis was performed 3 times for each group from 12 mixed fish meat). The means expressed using different letters in each row are significantly different ($p<0.05$).

Discussion

The feed conversion rates for the groups were as follows: 0.86, 0.85, 0.80, 0.80, 0.77 and 0.78. The addition of black cumin oil in the fish feed increased feed consumption and the daily feed consumed per fish in each group was as follows: 0.93g, 0.93g, 0.97g, 1.00g, 1.02g and 1.02g, respectively. The protein efficiency values were also observed to increase and were 2.77, 2.81, 2.98, 3.07 and 3.07, respectively.

The specific growth rates for the groups of fish receiving feeds with black cumin oil additives in the scope of the study were 0.89, 0.90, 0.95, 0.97, 1.02 and 1.02, respectively.

One of the indicators allowing the parametric evaluation of growth is Economic Conversion Rate (ECR), which is calculated as a multiple of feed prices and feed benefit rates and shows the economic reflection of feed benefit. Group 5 had the highest economic conversion rate (1.58\$ Kg^{-1}), while the control group had an economic conversion rate of 1.67 \$ Kg^{-1}. Higher economic conversion rate indicates higher costs. Therefore, Group 5 had the lowest costs and was the most profitable among the different groups. Furthermore, the Economic Profit Index (EPI) was calculated to show the level of benefits achieved by the addition of black cumin oil in fish feed, and it was observed that an addition of 1% of black cumin oil in the feed, as given to Group 5, yields an EPI of 0.72. An examination of these results show that an addition of 1% black cumin oil in trout feed increases the EPI.

Regarding rainbow trout, Tokur *et al*. (2006) reported the protein value as 22.96% and the lipid content as 2.71%. Furthermore, it has been reported that the nutritional values vary according to the feeding regime, feed composition, living area, harvest season, sex, size and environmental factor (Weatherup and McCracken, 1999; Rasmussen, 2001; Özden and Erkan, 2008).

Korkmaz and Kırkagac (2008) reported raw protein, oil, raw cinder and

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moisture content of rainbow trout as 20.33%, 4.1%, 1.22% and 74.18%, respectively. Kuş (2012) identified the protein, lipid, cinder and moisture values for rainbow trout as 19.94%, 6.45%, 1.21% and 72.26%, respectively. Dikel (1999) reported raw protein, lipid, raw cinder and dry matter content for freshwater rainbow trout as 19.11%±0.26, 0.96%±0.01, 1.60%±0.02, and 21.67%±0.10 respectively, and the raw protein, lipid, raw cinder and dry matter content for saltwater rainbow trout as 18.46%±0.41, 1.45%±0.017, 1.58%±0.005, 21.50%±0.21, respectively. As a result of our study, rainbow trout fed with black cumin oil supplements have higher raw protein, raw cinder, lipid and proportion of dry matter when compared to the results reported in the studies mentioned.

Analyses performed at the end of the 144 day feeding schedule have identified 23 different fatty acids in all groups. At the end of the feeding period, Group 2 had the highest proportion of saturated fatty acids (SFA) with 21.54%, while Group 4 had the lowest SFA value (18.17%). The analysis comparing the total SFA values at the end of the feeding period shows that there is a statistically significant differences between groups 1, 2 and 4 (p<0.05). A comparison of the MUFA values at the end of the feeding period showed no significant difference between the groups (p<0.05). Group 6 had the highest MUFA value while Group 2 had the lowest. A comparison of the PUFA values at the end of the feeding period showed that the interval was between 32.09% and 38.56%. The control group had the lowest PUFA values while Group 2 had the highest PUFA values.

After the 144 day feeding period in the scope of the study, the total SFA interval was between 18.17% and 21.54%. Haliloğlu *et al*. (2001), found that the ∑SFA value for rainbow trout was 31.92%; Beyter (2008) came up with a range between 23.07% and 24.52%. Yavuzer (2011) found that the ∑SFA value for rainbow trout in the wild was 19.00% while the ∑SFA value for cultivated rainbow trout was 18.25%. The results achieved in this study are lower than those reported by Haliloğlu *et al*. (2001) and Beyter (2008), but higher than those reported by Yavuzer (2011).

Haliloğlu *et al*. (2001), reported that in rainbow trout ∑MUFA was 30.81%, the EPA (C20:5n3) value was 3.07% and the DHA (C22:6n3) value was 19.17%. In his study, Beyter (2008) fed rainbow trout with three different feeds and found that the eicospetaenoic acid (C20:5n3-EPA) values were 3.13%, 2.60%, and 2.20% respectively, while the docosahexaenoic acid values were (C22:6n3-DHA) 20.32%, 8.69% and 10.83% and the ∑MUFA values ranged between 33.00% and 36.90%. We found a higher range of ∑MUFA values (37.86% - 39.98%) than these two studies, however the DHA (10.51% - 14.76%) and EPA (1.83% - 2.10%) values were lower.

Similarly, Beyter (2008) fed rainbow trout with 3 different commercial feeds; the pre-feeding PUFA value was found to be 44.33% and the post-feeding

PUFA values were found to be 42.14%, 34.13% and 40.03% for the 3 groups. Öz and Dikel (2015) found that the total SFA, MUFA and PUFA values for wild rainbow trout were 28.04%±0.54, 24.69%±0,73 and 35.07%±0.95, respectively while the total SFA, MUFA and PUFA values for cultivated rainbow trout were 20.74%±0.67, 26.57%±0.65, and 51.12%±0.97, respectively.

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