



## Chemical composition of the Black Sea trout (*Salmo labrax* Pallas, 1814): A comparative study

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### ABSTRACT

In this study, the chemical composition of the Black Sea trout (*Salmo labrax*), which were obtained from different environmental and feeding conditions, were evaluated. Wild trouts were captured from Altındere and Çağlayan rivers, while culture form obtained from aquaculture facilities. According to the results, wild forms had the highest crude protein and fat compared to culture forms, while moisture and crude ash was higher in culture forms. Glycine, alanine, glutamic acid, and aspartic acid were higher in the individuals caught from wild, whereas culture forms had the highest isoleucine, threonine, and valine. All essential amino acids were detected in all groups, and total essential amino acids exhibited the highest values in the culture forms. While the total monounsaturated and polyunsaturated fatty acids showed the highest values in the wild, they were in lower amounts in the culture forms. Linoleic acid and linolenic acid, which are essential for humans, were detected in all groups. Docosahexaenoic acid (DHA), linoleic acid, and eicosapentaenoic acid (EPA) were found to be the highest polyunsaturated ones, respectively. In filial generations, there are no statistical differences found neither in total essential amino acids nor in fatty acid contents between different generations of Black Sea trout.

**Keywords:** *Salmo trutta labrax*, Chemical quality, Seafood, Aquaculture, Salmon

## Introduction

Fish and other aquatic food sources are known to be biologically beneficial and indispensable throughout life in the human diet. In scientific studies, it has been shown that aquatic foods contain a high proportion of polyunsaturated fatty acids and essential amino acids, which are necessary for the human diet (Sahena et al., 2009). Essential amino acids and essential fatty acids are known that micronutrients are requisite for the maintenance of metabolic activities, the protection and development of organs and tissues (Ballantyne, 2011). Despite the continuous increase in consumer expectations, many species are at risk of extinction in the reason of uncontrolled fishing, environmental conditions, etc. This case is also an essential problem with the view to provide qualified food resources. In the food sector, one of the ways of providing qualified raw materials regularly and continuously is to use aquaculture products.

Today, Salmonids have become an integral part of the aquaculture sector because of their high economic value (Yeakley & Hughes, 2013). In Europe, Atlantic salmon (*Salmo salar*) and rainbow trout (*Oncorhynchus mykiss*) cover 60% of the total aquaculture and have great importance to the sector, economically (Liu et al., 2016). Therefore in Turkey, Black Sea trout (*Salmo labrax* PALLAS, 1814), which is an endemic subspecies of brown trout, is promising species for the aquaculture sector. Black Sea trout can be found as three forms in Turkey; sea, stream, and lake (Tabak et al., 2001). The natural distribution of the Black Sea trout is briefly can be called the Black Sea and the rivers flowing into the Black Sea (IUCN, 2020). It predominantly distributed in the northeast coast of the Black Sea, the Azov and Caspian Sea basins (Okumuş et al., 2004). In Turkey, primarily due to excessive hunting pressure, Black Sea trout stocks become endangered in nature (Çakmak et al., 2019).

The decrease in the natural stocks of the Black Sea trout has led researchers to the culturing of this fish. The first studies started in 1998 with the sampling of broodstock fish from the rivers Firtına, Çaglayan, and Kapistre, which poured into the Black Sea. As a result of the studies carried out in recent years, Black Sea trout have been cultured, and finally, the fifth filial generation was achieved when the study conducted (Çakmak et al., 2018). During the domestication process, the culture characteristics and meat yield of the Black Sea trout have been enhanced with selectivity programs and have become an alternative aquaculture species in the Eastern Black Sea region (Çakmak et al., 2019). Parallel with this development, broodstock belongs to the third filial generation is donated to local facilities in the Eastern Black Sea Region to promote the local aquaculture industry. Nowadays, 19 fish

farms are culturing Black Sea trout extensively with the amount of 2000 tons per year (Çankırılıgil et al., 2017; Turkish Statistical Institute, 2018). The aquaculture sector should focus on the cultivation of locally endemic species similar to Black Sea trout (Teletchea & Fontaine, 2014). With all these developments, for the food sector and the consumer, the meat quality is one of the most important subjects to be investigated in aquaculture species. Therefore, in this study, the meat quality of the Black Sea trout individuals, which they obtained from wild, fish farms, and different filial generations such as third (F3), fourth (F4), and fifth (F5) generations were compared.

## Material and Methods

### *Chemicals, Reagents and Other Consumables*

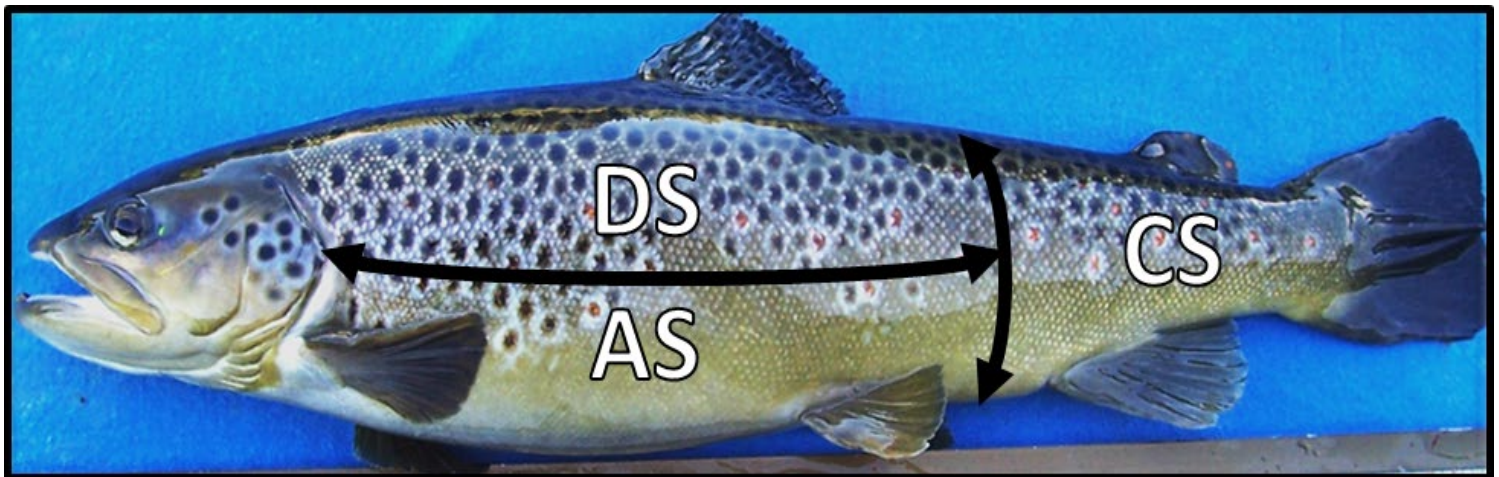
The chemicals, reagents and other consumables that used in all analyses are; hydrochloric acid huming 37% (Merck, 1.13386.2500), sulphuric acid (Merck, 1.00731.2511), boric acid (Merck, 100731.2511), sodium hydroxide (Merck, 1.06462.1000), methanol (Merck, 1.06009.2500), chloroform (Merck, 1.02445.2500), N-heptan (Merck, 1.04365.2500), hekzan (Merck, 1.04368.2500), boron trifluorid methanol complex (BF<sub>3</sub>) (Merck, Germany, 801663.0100), sodium chloride (Merck, 1.06404.1000), sodium sulphate (Merck, 1.06648.1000), Kjeldahl catalyst tablet containing 3.5 g K<sub>2</sub>SO<sub>4</sub>, 0.0035 g Se, borate buffer (Agilent, U.S.A., Agt-5061-3339), o-phthalaldehyde reagent (OPA) (Agilent, Agt-5061-3335), 9-fluorenylmethyl chloroformate reagent (FMOC) (Agilent, Agt-5061-3337), acetonitrile GC grade (Merck, 1.00030.2500), methanol GC grade (Merck, Germany, 1.06018.2500), sodium phosphate dibasic solution (Na<sub>2</sub>HPO<sub>4</sub>) (Merck, 1.06342.1000), amino acid standard solutions which is mixture of L-alanine, L-arginine, L-aspartic acid, L-cystine, L-glutamic acid, glycine, L-histidine hydrochloride monohydrate, L-isoleucine, L-leucine, L-lysine hydrochloride, L-methionine, L-phenylalanine, L-proline, L-serine, L-threonine, L-tyrosine, L-valine stored in 0.1N HCl (Agilent, Agt-5061), amino acid standards of amino acids sensitive to acidic pH such as L-glutamine, L-asparagine, L-tryptophan and L-4-hydroxyprolin in the powder form (Agilent, Agt-5062-2478), Zorbax extend C18 column for amino acids (Agilent, 3.5µm, 4.6x150 mm) (Agt-764953-902), GC column for fatty acids (Shimadzu, 50 m), fatty acid methyl ester standard (FAME's) (Supelco<sup>TM</sup> Component FAME mix, 47885-U), autoclave bottle (100ml) (Isolab, 061.01.100), Whatmann filter (1.2 µm, 0.45 µm) (Aldrich, WHA1001045), 1.5ml amber vials with politetrafloroetilen caps (Agt-5182-0716), vial insert (0.2 mL, konic) (Isolab,

097.05.110) and syringe filters (Isolab, 0.45 $\mu$ m, politetraflo-roetilen) (Isolab, 094.01.002), syringe (10 mL) (Isolab, 094.91.010), pipette tip (1000  $\mu$ L) (Isolab, 005.01.003).

### Fish Material and Sampling

The study material was selected as wild and culture forms of Black Sea trout (*Salmo labrax* PALLAS, 1814). The Black Sea trout individuals belong to river form were caught from rivers of Altindere and Çağlayan in May 2017, and June 2018, respectively, and they were compared to cultured ones. Culture forms of the Black Sea trout were obtained from aquaculture facilities operated in the same rivers. In addition to this, a culture form was obtained from aquaculture facilities operated in Borçka Dam Lake, which is an important production area for the *Salmo labrax*. There is no wild form of Black Sea trout that was captured from Borçka Dam Lake with the reason of this lake is not the natural habitat of the *Salmo labrax*. All fish samples were selected approximately equal to each other in terms of weight ranged from 240.22 to 260.31 g. Finally, different filial generations of the Black Sea trout such as third (F3), fourth (F4) and fifth (F5) generations

which were cultured in 2009-2010, 2012-2013 and 2016-2017 breeding seasons, respectively were obtained from Central Fisheries Research Institute in order to determine possible differences between achieved culture lines. Individuals belong to first (F1), and second (F2) filial generations did not exist anymore; that is why they were not analyzed in this study. In the analysis, while three individuals were used for each river in the analysis of wild forms, ten individuals were used for culture forms and culture lines (approximately 200 g). Ultimately, obtained fish were filleted and homogenized with for the chemical analysis. Besides that, fillets of individuals belong to culture lines were divided into three parts called the dorsal, abdomen and caudal muscle tissues to determinate possible differences be formed during the domestication period throughout the years. Besides, liver tissues were analyzed to determine possible excessive fat accumulation. All samplings and other treatments were carried out following ethical rules of ARRIVE guidelines (Kilkenny et al., 2010). Obtained fillets stored at +4 °C for analyses. The Black Sea trout and the partition of the fillets were shown in Figure 1, whereas sampling locations were shown in Figure 2.



**Figure 1.** The Black Sea trout (*Salmo labrax* PALLAS, 1814): Fillets of the Black Sea trout were divided into three parts which are dorsal section (DS), abdomen section (AS) and caudal section (CS) as shown in the figure for the analysis of different culture lines such as third (F3), fourth (F4) and fifth (F5) lines.



**Figure 2.** Sampling stations and aquaculture facilities that fish obtained from. a: Sampling station on the Altındere River ( $40^{\circ}40'08.77''\text{N}$ ,  $39^{\circ}39'58.86''\text{E}$ ), b: An aquaculture facility operated in Altındere River ( $40^{\circ}42'10.23''\text{N}$ ,  $39^{\circ}39'06.04''\text{E}$ ), c: Central Fisheries Research Institute's research units ( $40^{\circ}57'35.56''\text{N}$ ,  $39^{\circ}51'17.62''\text{E}$ ), d: Sampling station on the Çağlayan River ( $41^{\circ}14'16.01''\text{N}$ ,  $41^{\circ}15'55.51''\text{E}$ ), e: An aquaculture facility operated in Çağlayan River ( $41^{\circ}15'19.70''\text{N}$ ,  $41^{\circ}13'49.52''\text{E}$ ), f: An aquaculture facility operated in Borçka Dam Lake ( $41^{\circ}19'15.22''\text{N}$ ,  $41^{\circ}43'44.24''\text{E}$ ).

### *Determination of Proximate Composition*

Water (moisture) analysis was carried out, according to Horwitz (2000). Homogenized samples weighted as 1 g to petri plates and dehydrated with drying oven at  $100^{\circ}\text{C}$  for 24 hours and calculated according to method. Crude protein analysis was carried out with the Kjeldahl method (AOAC, 2000). Fish meats digested with 15 mL  $\text{H}_2\text{SO}_4$  and Kjeldahl catalyst at  $120^{\circ}\text{C}$  and distilled with NaOH. Obtained samples were titrated with 0.1 N HCl and calculated as percentage. The crude fat analysis was conducted according to the method of Folch et al. (1957). Crude fat extracted with methanol-chloroform complex (2:1) and were filtrated with  $1.2\ \mu\text{m}$  Whatman filters. Finally obtained mixtures evaporated at  $65^{\circ}\text{C}$  with a rotary vacuum evaporator (Eyela, N-N 1521) and calculated as percentage according to the method. Crude ash analysis was carried out according to Horwitz (2000). Homogenized samples weighted and burned with muffle furnace (Protherm) at  $600^{\circ}\text{C}$  for 6 hours. Obtained ash was weighted and calculated as percentage.

### *Amino Acid Analysis*

Firstly, fish meat was digested with the HCl at  $110^{\circ}\text{C}$  in 24 hours as a preliminary treatment for the amino acid analysis (Çankırılıgil et al., 2020). Obtained hydrolysates were filtered by  $0.45\ \mu\text{m}$  PTFE syringe filters and diluted as  $10^{-1}$  with pure water. In the following, samples transferred to 1.5 mL amber vials having PTFE caps and stored until the analysis. The amino acid analysis was done under the method of Henderson et al. (Henderson et al., 2000) in HPLC (Agilent Infinity II) system equipped with a diode-array detector and Agilent standards were used (Agilent, Agt-5061). In the analysis,  $0.5\ \mu\text{L}$  of the samples were derivatized with borate, OPA, and FMOC by auto-sampler. Derivatized samples were injected into the system having an amino acid column as a solid phase and mixture of  $\text{MeOH}:\text{ACN}:\text{H}_2\text{O}$  (%45:%45:%10) and 40 mM  $\text{Na}_2\text{HPO}_4$  solution which has 7.8 pH adjusted with 10 N NaOH as a mobile phase. The gradient conditions of the mobile phase were shown in table 1. Detection was carried out in two wavelengths as 262 nm for FMOC amino acids and 338 nm for OPA amino acids. All samples were analyzed for the five times, and detected pikes were auto-integrated with

system's software. Finally, obtained data were compared with calibration curves which constituted via the Agilent amino acid standards and expressed as g/100g.

**Table 1.** HPLC mobile phase gradient conditions

Time (min)	A (MeOH:ACN:H <sub>2</sub> O)	B (Na <sub>2</sub> HPO <sub>4</sub> )	Flow (mL/min)
1.90	0%	100%	2
18.1	57%	43%	2
18.6	100%	0%	2
22.3	100%	0%	2
23.2	0%	100%	2

### Fatty Acid Analysis

Firstly, 0.15 g fat weighted from crude fat samples, which were obtained before and 5 mL 0.5 N methanolic NaOH, were put in volumetric flasks for evaporation, which is executed with Soxhlet evaporator at 65°C. During evaporation, 5 mL BF<sub>3</sub> and 2 mL heptane were added to mixtures in the 15th and 17th minutes, respectively. Obtained mixtures were blended with saturated NaCl, and emerged phase in the samples were filtered with 0.45 µm syringe filters for the analysis. The fatty acid analysis was performed with gas chromatography (Shimadzu, GC-17A) having 50 m fatty acid column and flame ionization detector and fatty acid methyl ester standard (FAME's) (Supelco™ Component FAME mix, Germany, 47885-U) was used. Heptane was injected into all samples with auto-injectors in the amount of 1 µL as a dissolver. The column oven temperature was adjusted as 140 °C in starting and stabilized in 240 °C with increasing by 20 °C every minute, whereas the detector and injector block temperature was 260 °C. Helium (He) with 30 mL/dk flow, hydrogen (H) with 40 mL/dk flow, and air with 400 mL/dk flow were used as carrier gasses with 22.8 mL/dk total flow. All samples were analyzed five times and obtained data expressed as a percentile (IUPAC International Union of Pure and Applied Chemistry, 1979).

### Data Analysis

IBM SPSS 23 software was used in statistical analysis. Results of all chemical analyses were analyzed with one-way ANOVA method after the normality and homogeneity were checked by Anderson–Darling and Levene tests, respectively.

## Results and Discussion

The proximate composition of the Black Sea trout obtained from different environmental conditions was shown in Table 2. According to food legislation, if the moisture content of the food is higher than the 50 %, it called water content instead of moisture. So, the term of water used in this article due to fish meat has 60-80 % water content, parallel with our results. The highest amount of water was found in cultured fish, crude protein and crude fat ratios were found in fishes sampled from Altındere and Çağlayan rivers, and the highest amount of crude ash was determined in third, fourth and fifth filial generations ( $P \leq 0.05$ ).

The proximate composition of the Black Sea trout filial generations according to different body parts, as shown in Table 3. When the muscle tissues from different body parts of the Black Sea trout were examined, no statistical differences could be detected in individuals of all generations ( $P \geq 0.05$ ). The highest crude protein content was found in the dorsal and caudal parts, and the highest crude fat content was in the abdominal ( $P \leq 0.05$ ). In the research, the liver fat ratio was found to be higher than muscle tissues ( $P \leq 0.05$ ), and no statistical difference was found between generations ( $P \geq 0.05$ ). Meat quality of fish depends on some specific environmental features such as species, sex, length, age, reproduction stage, temperature (Nurnadia et al., 2011). Altındere and Çağlayan rivers, which are the sampling area of fish which caught from nature, are high flow rated and cold in spring due to melting snow waters (Fidan et al., 2017). In addition to the crude fat content of fish, which grow in cold waters, the amount of long-chain fatty acids is also high (Farkas et al., 1980). Besides, trout, which is usually caught from nature and reach high swimming speeds, shows more muscle development than the culture forms (Sanger & Stobier, 2001; Totland et al., 1987). Therefore, crude protein and fat ratios were found to be high in-stream forms. In salmonids, the myotomal muscle bundles (white muscle tissue) in the dorsal and caudal parts are responsible for providing the pushing force required to swim and contain more muscle bundles than the abdomen. The abdominal part is the muscle part where fat accumulation is frequently seen in trouts (Totland et al., 1987; Videler, 1993). Therefore, while the amount of crude protein ratio was higher in the dorsal and caudal parts, the amount of crude fat was higher in the abdomen ( $P \leq 0.05$ ). Similarly, lipids accumulate in muscle tissue and adipose fin in fish and are stored in the liver (Özel et al., 2017).

**Table 2.** Proximate composition of the Black Sea trout obtained from different conditions (%)

	Water	Crude protein	Crude fat	Crude ash
<b>Wild</b>				
Altındere River	71.61±0.18 <sup>d</sup>	17.94±0.10 <sup>a</sup>	8.11±0.21 <sup>a</sup>	1.35±0.03 <sup>b</sup>
Çağlayan River	72.07±0.20 <sup>c</sup>	17.90±0.09 <sup>a</sup>	7.89±0.18 <sup>a</sup>	1.26±0.04 <sup>c</sup>
<b>Culture</b>				
Altındere River	73.34±0.16 <sup>a</sup>	17.77±0.12 <sup>ab</sup>	6.52±0.08 <sup>c</sup>	1.26±0.04 <sup>c</sup>
Çağlayan River	73.38±0.21 <sup>a</sup>	17.68±0.11 <sup>b</sup>	6.56±0.09 <sup>c</sup>	1.25±0.05 <sup>c</sup>
Borçka Dam lake	72.79±0.19 <sup>b</sup>	17.52±0.18 <sup>b</sup>	7.10±0.11 <sup>b</sup>	1.40±0.03 <sup>a</sup>
<b>Filial Generations</b>				
F3 Generation	73.22±0.19 <sup>a</sup>	17.81±0.09 <sup>ab</sup>	6.22±0.12 <sup>d</sup>	1.45±0.06 <sup>a</sup>
F4 Generation	73.33±0.21 <sup>a</sup>	17.75±0.08 <sup>ab</sup>	6.24±0.17 <sup>d</sup>	1.40±0.02 <sup>a</sup>
F5 Generation	73.34±0.29 <sup>a</sup>	17.69±0.09 <sup>b</sup>	6.13±0.18 <sup>d</sup>	1.38±0.02 <sup>ab</sup>

Values are expressed as mean ±SD, mean values in a column with different superscripts were significantly different ( $P \leq 0.05$ ).

**Table 3.** Proximate composition of the body parts of Black Sea trout's filial generations (%)

	Water	Crude protein	Crude fat	Crude ash
<b>F3 Generation</b>				
Dorsal section	74.73±0.25 <sup>a</sup>	17.97±0.18 <sup>a</sup>	4.98±0.04 <sup>d</sup>	1.32±0.03 <sup>b</sup>
Abdomen section	70.82±0.23 <sup>c</sup>	16.39±0.20 <sup>b</sup>	10.63±0.15 <sup>b</sup>	1.16±0.04 <sup>c</sup>
Caudal section	73.51±0.21 <sup>b</sup>	17.93±0.14 <sup>a</sup>	6.22±0.17 <sup>c</sup>	1.29±0.06 <sup>b</sup>
Liver tissue	64.89±0.18 <sup>d</sup>	14.96±0.19 <sup>c</sup>	14.02±0.16 <sup>a</sup>	1.41±0.03 <sup>a</sup>
<b>F4 Generation</b>				
Dorsal section	74.75±0.21 <sup>a</sup>	18.10±0.21 <sup>a</sup>	5.16±0.15 <sup>d</sup>	1.35±0.04 <sup>b</sup>
Abdomen section	70.32±0.19 <sup>c</sup>	16.41±0.20 <sup>b</sup>	10.79±0.16 <sup>b</sup>	1.11±0.03 <sup>c</sup>
Caudal section	73.49±0.20 <sup>b</sup>	17.95±0.11 <sup>a</sup>	6.13±0.07 <sup>c</sup>	1.28±0.05 <sup>b</sup>
Liver tissue	64.22±0.13 <sup>d</sup>	15.33±0.08 <sup>c</sup>	14.32±0.10 <sup>a</sup>	1.38±0.06 <sup>a</sup>
<b>F5 Generation</b>				
Dorsal section	74.41±0.17 <sup>a</sup>	18.02±0.13 <sup>a</sup>	5.06±0.08 <sup>d</sup>	1.31±0.04 <sup>b</sup>
Abdomen section	70.44±0.16 <sup>c</sup>	16.26±0.09 <sup>b</sup>	10.59±0.19 <sup>b</sup>	1.15±0.04 <sup>c</sup>
Caudal section	73.23±0.21 <sup>b</sup>	18.23±0.14 <sup>a</sup>	6.09±0.08 <sup>c</sup>	1.30±0.06 <sup>b</sup>
Liver tissue	65.34±0.11 <sup>d</sup>	15.02±0.16 <sup>c</sup>	14.24±0.11 <sup>a</sup>	1.42±0.05 <sup>a</sup>

Values are expressed as mean ± SE, mean values in a column with different superscripts were significantly different ( $P \leq 0.05$ ).

**Table 4.** Amino acid composition of the Black Sea trout obtained from different conditions (g/100g)

Amino acids	Wild forms		Culture forms			Filial Generations		
	Altındere River	Çağlayan River	Altındere River	Çağlayan River	Borçka Dam Lake	F3 Generation	F4 Generation	F5 Generation
ASP	0.84±0.04 <sup>a</sup>	0.89±0.04 <sup>a</sup>	0.68±0.03 <sup>b</sup>	0.65±0.04 <sup>b</sup>	0.70±0.03 <sup>b</sup>	0.71±0.04 <sup>b</sup>	0.68±0.03 <sup>b</sup>	0.73±0.03 <sup>b</sup>
GLU	1.76±0.03 <sup>a</sup>	1.73±0.02 <sup>a</sup>	1.60±0.03 <sup>b</sup>	1.62±0.04 <sup>b</sup>	1.70±0.02 <sup>ab</sup>	1.59±0.05 <sup>b</sup>	1.55±0.05 <sup>b</sup>	1.58±0.04 <sup>b</sup>
ASN	0.58±0.04 <sup>a</sup>	0.59±0.04 <sup>a</sup>	0.56±0.02 <sup>a</sup>	0.61±0.06 <sup>a</sup>	0.60±0.03 <sup>a</sup>	0.60±0.05 <sup>a</sup>	0.58±0.03 <sup>a</sup>	0.59±0.06 <sup>a</sup>
SER	1.43±0.12 <sup>a</sup>	1.43±0.09 <sup>a</sup>	1.48±0.08 <sup>a</sup>	1.50±0.12 <sup>a</sup>	1.49±0.11 <sup>a</sup>	1.46±0.13 <sup>a</sup>	1.46±0.07 <sup>a</sup>	1.43±0.08 <sup>a</sup>
GLN	0.85±0.05 <sup>a</sup>	0.85±0.04 <sup>a</sup>	0.81±0.03 <sup>a</sup>	0.81±0.05 <sup>a</sup>	0.79±0.10 <sup>a</sup>	0.83±0.04 <sup>a</sup>	0.78±0.07 <sup>a</sup>	0.79±0.06 <sup>a</sup>
HIS	0.49±0.03 <sup>a</sup>	0.49±0.05 <sup>a</sup>	0.44±0.03 <sup>a</sup>	0.46±0.03 <sup>a</sup>	0.50±0.04 <sup>a</sup>	0.45±0.04 <sup>a</sup>	0.48±0.02 <sup>a</sup>	0.48±0.06 <sup>a</sup>
GLY	0.83±0.03 <sup>a</sup>	0.85±0.04 <sup>a</sup>	0.68±0.04 <sup>b</sup>	0.69±0.03 <sup>b</sup>	0.75±0.06 <sup>ab</sup>	0.67±0.04 <sup>b</sup>	0.65±0.05 <sup>b</sup>	0.63±0.04 <sup>b</sup>
THR	1.18±0.04 <sup>b</sup>	1.20±0.02 <sup>b</sup>	1.30±0.03 <sup>a</sup>	1.26±0.04 <sup>ab</sup>	1.24±0.03 <sup>ab</sup>	1.29±0.03 <sup>a</sup>	1.27±0.03 <sup>a</sup>	1.26±0.03 <sup>a</sup>
ALA	0.50±0.03 <sup>a</sup>	0.48±0.03 <sup>a</sup>	0.26±0.01 <sup>b</sup>	0.27±0.02 <sup>b</sup>	0.28±0.02 <sup>a</sup>	0.24±0.01 <sup>b</sup>	0.23±0.02 <sup>b</sup>	0.25±0.01 <sup>b</sup>
TYR	0.16±0.02 <sup>a</sup>	0.15±0.02 <sup>a</sup>	0.17±0.02 <sup>a</sup>	0.17±0.01 <sup>a</sup>	0.16±0.02 <sup>a</sup>	0.16±0.02 <sup>a</sup>	0.16±0.01 <sup>a</sup>	0.16±0.01 <sup>a</sup>
CYS	0.14±0.01 <sup>a</sup>	0.15±0.01 <sup>a</sup>	0.12±0.02 <sup>a</sup>	0.13±0.01 <sup>a</sup>	0.13±0.02 <sup>a</sup>	0.11±0.02 <sup>a</sup>	0.12±0.02 <sup>a</sup>	0.12±0.01 <sup>a</sup>
VAL	0.69±0.04 <sup>b</sup>	0.68±0.06 <sup>b</sup>	0.86±0.05 <sup>a</sup>	0.85±0.04 <sup>a</sup>	0.82±0.05 <sup>a</sup>	0.87±0.03 <sup>a</sup>	0.88±0.03 <sup>a</sup>	0.86±0.03 <sup>a</sup>
MET	0.84±0.06 <sup>a</sup>	0.85±0.04 <sup>a</sup>	0.83±0.11 <sup>a</sup>	0.84±0.09 <sup>a</sup>	0.82±0.05 <sup>a</sup>	0.84±0.05 <sup>a</sup>	0.86±0.06 <sup>a</sup>	0.86±0.07 <sup>a</sup>
TRP	0.08±0.01 <sup>a</sup>	0.08±0.01 <sup>a</sup>	0.06±0.01 <sup>a</sup>	0.07±0.01 <sup>a</sup>	0.06±0.01 <sup>a</sup>	0.08±0.01 <sup>a</sup>	0.08±0.01 <sup>a</sup>	0.07±0.01 <sup>a</sup>
PHE	0.75±0.04 <sup>a</sup>	0.74±0.05 <sup>a</sup>	0.78±0.09 <sup>a</sup>	0.78±0.06 <sup>a</sup>	0.76±0.05 <sup>a</sup>	0.75±0.05 <sup>a</sup>	0.74±0.03 <sup>a</sup>	0.76±0.05 <sup>a</sup>
ISO	0.84±0.04 <sup>b</sup>	0.87±0.03 <sup>b</sup>	1.02±0.05 <sup>a</sup>	0.98±0.05 <sup>a</sup>	0.99±0.04 <sup>a</sup>	0.99±0.06 <sup>a</sup>	0.96±0.06 <sup>a</sup>	0.97±0.05 <sup>a</sup>
LEU	1.48±0.10 <sup>a</sup>	1.51±0.12 <sup>a</sup>	1.53±0.13 <sup>a</sup>	1.49±0.09 <sup>a</sup>	1.55±0.11 <sup>a</sup>	1.50±0.08 <sup>a</sup>	1.52±0.10 <sup>a</sup>	1.53±0.07 <sup>a</sup>
LYS	1.41±0.11 <sup>a</sup>	1.44±0.09 <sup>a</sup>	1.37±0.12 <sup>a</sup>	1.38±0.09 <sup>a</sup>	1.39±0.09 <sup>a</sup>	1.39±0.11 <sup>a</sup>	1.41±0.13 <sup>a</sup>	1.43±0.06 <sup>a</sup>
<b>TEAA</b>	6.19±0.10 <sup>b</sup>	6.27±0.09 <sup>b</sup>	6.45±0.12 <sup>a</sup>	6.39±0.08 <sup>a</sup>	6.39±0.15 <sup>a</sup>	6.42±0.11 <sup>a</sup>	6.45±0.09 <sup>a</sup>	6.48±0.11 <sup>a</sup>
<b>TAA</b>	14.95±0.13 <sup>a</sup>	15.08±0.21 <sup>a</sup>	14.55±0.16 <sup>b</sup>	14.56±0.21 <sup>a</sup>	14.73±0.20 <sup>ab</sup>	14.53±0.22 <sup>a</sup>	14.41±0.23 <sup>a</sup>	14.50±0.18 <sup>a</sup>

Values are expressed as mean ±SE. Mean values in a row with different superscripts were statistically different ( $P \leq 0.05$ ). ASP; aspartic acid, GLU; glutamic acid, ASN; asparagine, SER; serine, GLN; glutamine, HIS; histidine, GLY; glycine, THR; threonine, ALA; alanine, TYR; tyrosine, CYS; cysteine, VAL; valine, MET; methionine; TRP; tryptophan, PHE; phenylalanine, ISO; isoleucine, LEU; leucine, LYS; lysine, TEAA; total essential amino acids, TAA; total amino acids.

**Table 5.** Fatty acid composition of the Black Sea trout obtained from different conditions (%)

Fatty acids	Wild forms		Culture forms			Filial Generations		
	Altındere River	Çağlayan River	Altındere River	Çağlayan River	Borçka Dam Lake	F3 Generation	F4 Generation	F5 Generation
C10:0	0.08±0.02 <sup>c</sup>	0.12±0.02 <sup>b</sup>	0.09±0.01 <sup>c</sup>	0.14±0.02 <sup>ab</sup>	0.20±0.02 <sup>a</sup>	0.18±0.01 <sup>a</sup>	0.16±0.002 <sup>ab</sup>	0.14±0.02 <sup>ab</sup>
C12:0	0.11±0.02 <sup>b</sup>	0.15±0.02 <sup>b</sup>	0.12±0.02 <sup>b</sup>	0.23±0.03 <sup>a</sup>	0.20±0.01 <sup>a</sup>	0.21±0.02 <sup>a</sup>	0.17±0.01 <sup>ab</sup>	0.17±0.01 <sup>ab</sup>
C14:0	1.83±0.03 <sup>d</sup>	1.90±0.03 <sup>d</sup>	2.79±0.07 <sup>bc</sup>	2.67±0.05 <sup>c</sup>	2.88±0.03 <sup>b</sup>	2.91±0.06 <sup>ab</sup>	3.00±0.04 <sup>a</sup>	3.10±0.06 <sup>a</sup>
C15:0	5.48±0.29 <sup>a</sup>	5.39±0.27 <sup>a</sup>	5.67±0.18 <sup>a</sup>	5.62±0.23 <sup>a</sup>	5.71±0.28 <sup>a</sup>	5.60±0.26 <sup>a</sup>	5.61±0.24 <sup>a</sup>	5.55±0.22 <sup>a</sup>
C16:0	11.20±0.31 <sup>b</sup>	11.25±0.35 <sup>b</sup>	12.81±0.32 <sup>a</sup>	12.89±0.28 <sup>a</sup>	12.88±0.40 <sup>a</sup>	12.61±0.41 <sup>a</sup>	12.56±0.44 <sup>a</sup>	12.54±0.47 <sup>a</sup>
C17:0	1.96±0.11 <sup>b</sup>	2.01±0.12 <sup>ab</sup>	2.27±0.14 <sup>a</sup>	2.16±0.17 <sup>a</sup>	2.28±0.11 <sup>a</sup>	2.17±0.13 <sup>a</sup>	2.15±0.15 <sup>a</sup>	1.91±0.09 <sup>b</sup>
C18:0	3.40±0.22 <sup>a</sup>	3.54±0.24 <sup>a</sup>	3.61±0.29 <sup>a</sup>	3.58±0.22 <sup>a</sup>	3.44±0.25 <sup>a</sup>	3.47±0.27 <sup>a</sup>	3.44±0.22 <sup>a</sup>	3.55±0.18 <sup>a</sup>
C20:0	1.52±0.10 <sup>b</sup>	1.54±0.09 <sup>b</sup>	1.50±0.12 <sup>b</sup>	1.67±0.08 <sup>ab</sup>	1.74±0.11 <sup>a</sup>	1.73±0.08 <sup>a</sup>	1.78±0.13 <sup>a</sup>	1.75±0.13 <sup>a</sup>
C21:0	0.81±0.02 <sup>b</sup>	0.88±0.03 <sup>a</sup>	0.84±0.04 <sup>ab</sup>	0.79±0.02 <sup>b</sup>	0.91±0.03 <sup>a</sup>	0.85±0.02 <sup>ab</sup>	0.88±0.03 <sup>a</sup>	0.84±0.03 <sup>ab</sup>
C22:0	0.96±0.11 <sup>a</sup>	1.03±0.13 <sup>a</sup>	0.67±0.08 <sup>b</sup>	0.68±0.09 <sup>b</sup>	0.60±0.06 <sup>bc</sup>	0.52±0.05 <sup>c</sup>	0.51±0.05 <sup>c</sup>	0.58±0.04 <sup>bc</sup>
C24:0	0.46±0.03 <sup>a</sup>	0.49±0.03 <sup>a</sup>	0.33±0.04 <sup>b</sup>	0.28±0.03 <sup>bc</sup>	0.31±0.03 <sup>b</sup>	0.22±0.03 <sup>c</sup>	0.24±0.03 <sup>c</sup>	0.26±0.03 <sup>bc</sup>
<b>TSFA</b>	27.81±0.33 <sup>c</sup>	28.30±0.30 <sup>c</sup>	30.70±0.32 <sup>b</sup>	30.71±0.26 <sup>ab</sup>	31.15±0.30 <sup>a</sup>	30.47±0.35 <sup>b</sup>	30.52±0.41 <sup>b</sup>	30.40±0.30 <sup>b</sup>
C15:1	0.09±0.01 <sup>a</sup>	0.10±0.01 <sup>a</sup>	0.08±0.01 <sup>a</sup>	0.08±0.01 <sup>a</sup>	0.05±0.01 <sup>b</sup>	0.08±0.01 <sup>a</sup>	0.09±0.01 <sup>a</sup>	0.08±0.01 <sup>a</sup>
C16:1	9.84±0.12 <sup>a</sup>	9.66±0.14 <sup>a</sup>	9.42±0.16 <sup>b</sup>	9.36±0.12 <sup>b</sup>	9.56±0.09 <sup>b</sup>	9.48±0.15 <sup>b</sup>	9.45±0.16 <sup>b</sup>	9.36±0.18 <sup>b</sup>
C17:1	1.12±0.03 <sup>a</sup>	1.13±0.03 <sup>a</sup>	1.02±0.02 <sup>b</sup>	1.13±0.03 <sup>a</sup>	1.10±0.02 <sup>a</sup>	1.12±0.02 <sup>a</sup>	1.08±0.03 <sup>ab</sup>	1.07±0.04 <sup>ab</sup>
C18:1	12.33±0.21 <sup>a</sup>	12.45±0.23 <sup>a</sup>	12.24±0.19 <sup>a</sup>	12.29±0.12 <sup>a</sup>	12.14±0.20 <sup>a</sup>	12.36±0.18 <sup>a</sup>	12.23±0.22 <sup>a</sup>	12.18±0.29 <sup>a</sup>
C20:1	1.30±0.04 <sup>a</sup>	1.33±0.04 <sup>a</sup>	1.20±0.04 <sup>b</sup>	1.15±0.03 <sup>c</sup>	1.24±0.05 <sup>ab</sup>	1.19±0.04 <sup>bc</sup>	1.22±0.03 <sup>b</sup>	1.26±0.03 <sup>ab</sup>
<b>TMUFA</b>	24.68±0.36 <sup>ab</sup>	24.67±0.42 <sup>ab</sup>	24.68±0.51 <sup>ab</sup>	24.56±0.33 <sup>ab</sup>	24.09±0.26 <sup>b</sup>	24.95±0.51 <sup>a</sup>	24.88±0.51 <sup>a</sup>	24.67±0.42 <sup>ab</sup>
C18:2	11.20±0.30 <sup>a</sup>	11.09±0.27 <sup>a</sup>	10.03±0.20 <sup>b</sup>	10.26±0.24 <sup>b</sup>	10.06±0.12 <sup>b</sup>	10.40±0.26 <sup>b</sup>	10.32±0.25 <sup>b</sup>	10.26±0.16 <sup>b</sup>
C18:3	1.36±0.05 <sup>a</sup>	1.35±0.04 <sup>a</sup>	1.20±0.05 <sup>b</sup>	1.20±0.04 <sup>b</sup>	1.23±0.04 <sup>b</sup>	1.23±0.03 <sup>b</sup>	1.18±0.05 <sup>b</sup>	1.27±0.04 <sup>ab</sup>
C20:2	0.62±0.05 <sup>a</sup>	0.63±0.04 <sup>a</sup>	0.70±0.05 <sup>a</sup>	0.68±0.06 <sup>a</sup>	0.65±0.02 <sup>a</sup>	0.64±0.06 <sup>a</sup>	0.64±0.04 <sup>a</sup>	0.66±0.06 <sup>a</sup>
C20:3	0.94±0.05 <sup>a</sup>	0.99±0.06 <sup>a</sup>	0.78±0.06 <sup>b</sup>	0.77±0.04 <sup>b</sup>	0.84±0.04 <sup>ab</sup>	0.82±0.05 <sup>b</sup>	0.86±0.05 <sup>ab</sup>	0.85±0.05 <sup>ab</sup>
C20:4	1.20±0.04 <sup>a</sup>	1.23±0.05 <sup>a</sup>	1.10±0.06 <sup>ab</sup>	1.09±0.07 <sup>b</sup>	1.11±0.04 <sup>ab</sup>	1.04±0.06 <sup>b</sup>	1.06±0.05 <sup>b</sup>	1.06±0.06 <sup>b</sup>
C20:5	5.46±0.18 <sup>a</sup>	5.29±0.11 <sup>a</sup>	4.80±0.25 <sup>b</sup>	4.86±0.19 <sup>b</sup>	5.16±0.09 <sup>ab</sup>	4.99±0.12 <sup>b</sup>	5.02±0.15 <sup>b</sup>	5.10±0.15 <sup>ab</sup>
C22:5	1.42±0.32 <sup>a</sup>	1.49±0.24 <sup>a</sup>	1.36±0.26 <sup>a</sup>	1.38±0.27 <sup>a</sup>	1.50±0.31 <sup>a</sup>	1.42±0.20 <sup>a</sup>	1.40±0.28 <sup>a</sup>	1.39±0.29 <sup>a</sup>
C22:6	25.22±0.31 <sup>a</sup>	24.86±0.25 <sup>a</sup>	24.34±0.24 <sup>b</sup>	24.32±0.21 <sup>b</sup>	24.21±0.23 <sup>b</sup>	24.03±0.41 <sup>b</sup>	24.11±0.34 <sup>b</sup>	24.30±0.29 <sup>b</sup>
<b>TPUFA</b>	47.22±0.36 <sup>a</sup>	46.93±0.34 <sup>a</sup>	44.71±0.29 <sup>b</sup>	44.56±0.044 <sup>b</sup>	44.76±0.41 <sup>b</sup>	44.57±0.50 <sup>b</sup>	44.59±0.32 <sup>b</sup>	44.89±0.41 <sup>b</sup>

Values are expressed as mean ± SE, mean values in a row with different superscripts were statistically different ( $P < 0.05$ ). TSFA; total saturated fatty acids, TMUFA; total monounsaturated fatty acids, TPUFA; total polyunsaturated fatty acids, C10:0; capric acid, C12:0; lauric acid, C14:0; myristic acid, C15:0; pentadecylic acid, C16:0; palmitic acid, C17:0; margaric acid, C18:0; stearic acid, C20:0; arachidic acid, C21:0; heneicosylic acid, C22:0; behenic acid, C24:0; lignoceric acid, C14:1; myristoleic acid C15:1; pentadecenoic acid, C16:1; palmitoleic acid, C17:1; heptadecenoic acid, C18:1; oleic acid, C20:1; eicosenoic acid, C18:2; linoleic acid; C18:3;  $\alpha$ -linolenic acid, C20:2; eicosadienoic acid, C20:3; dihomogamma linolenic acid, C20:4; arachidonic acid, C20:5; eicosapentaenoic acid, C22:5; docosapentaenoic acid, C22:6; docosahexaenoic acid



Amino acid compositions of the Black Sea trout individuals obtained from the different environments were shown in Table 4. According to results, while the total amino acid amount was detected as highest in the wild fish, the total amount of essential amino acids was detected highest in the culture forms ( $P \leq 0.05$ ). The most abundant amino acids were found as leucine, glutamic acid, serine, and lysine in all groups, respectively ( $P \leq 0.05$ ). Glycine, alanine, glutamic acid, and aspartic acid were found higher in the wild forms, whereas isoleucine, threonine, and valine were found higher in the culture forms ( $P \leq 0.05$ ). There are no statistical differences determined between all culture forms, including filial generations ( $P \geq 0.05$ ). The eight essential amino acids were detected in all fish groups, including tryptophan, even though it was found as a minimum quantity. While threonine, valine, and isoleucine were found highest in the culture forms ( $P \leq 0.05$ ), other essential amino acids were found statistically the same between groups ( $P \geq 0.05$ ). Amino acids are essential compounds for nutrition. Essential amino acids are necessary for many vital tasks in metabolism, such as protein synthesis, gene expression, cell division, and hormone secretion (Wu et al., 2010). Amino acids and proteins need to be taken daily in all diets for healthy eating and long life (Fontana & Partridge, 2015; Mirzaei et al., 2014). For these reasons, it is crucial to determine the amino acid composition of the species, such as the Black Sea trout, whose growth potential is increasing day by day. As well as the amount of amino acid in fish species may differ from species to species (Kaushik & Seiliez, 2010), the species may also vary according to environmental factors and feeding conditions (Ballantyne, 2011). Moreover, amino acid composition and muscle structure of Salmonidae species can be show differences due to the development of myotomal bundles caused by swimming activity between actively swimming species and non-swimming ones, even though in some species (Totland et al., 1987; Videler, 1993). As an anadromous species, Black Sea trout can be migrated between sea and throughout the whole stream in wild forms (Aydın & Yandı, 2002). Thus, crude protein and total amino acid contents of the wild forms were found higher compared to others due to muscle development. All essential amino acids, just as isoleucine, leucine, lysine, methionine, phenylalanine, threonine, valine, and tryptophan were detected in all groups. The lowest amino acid determined as tryptophan among all amino acids due to analyzing procedures. In the amino acid analysis, fish meat was treated with extreme conditions such as high temperature and low pH. Tryptophan is instable such extreme conditions (Çankırılıgil et al., 2020), and it can be lost entirely (Cuq & Firedman, 1989). Thus, tryptophan was found lowest.

The fatty acid composition of the Black Sea trout individuals was shown in Table 5. According to results, while the highest

total saturated fatty acid (SFA) was detected in culture forms obtained from Borçka Dam Lake, values of wild forms were detected lower compared to other groups ( $P \leq 0.05$ ). Filial generations were found more abundant in terms of total monounsaturated fatty acids among groups ( $P \leq 0.05$ ). Oleic acid was found highest monounsaturated fatty acid in the Black Sea trout is all individuals and followed by palmitoleic acid. Besides that, these two fatty acids found highest in the wild forms among groups ( $P \leq 0.05$ ). Polyunsaturated fatty acids were specified as the highest fatty acid group with the ratios ranges from  $44.56 \pm 0.04$  to  $47.22 \pm 0.36$ , and they were found higher in wild forms than culture forms ( $P \leq 0.05$ ). Linoleic acid and  $\alpha$ -linolenic acid, which are essential for humans, were found in all forms of Black Sea trout. The most abundant polyunsaturated fatty acids were detected as DHA, linoleic acid, and EPA, respectively ( $P \leq 0.05$ ). Whereas linoleic acid,  $\alpha$ -linolenic acid, dihomo gamma-linolenic acid, arachidonic acid, DHA, and EPA were found highest in wild forms ( $P \leq 0.05$ ), there are no statistical differences detected in the eicosadienoic acid and docosapentaenoic acid between groups ( $P \geq 0.05$ ). One of the essential quality parameters that makes fish meat nutritious is the content of long-chain fatty acids (Lund, 2013; Sahena et al., 2009). Regular intake of polyunsaturated fatty acids, especially EPA and DHA are recommended for healthy nutrition in humans. It has been reported that fish oil is reducing deaths from such heart diseases and certain types of cancer with its effects on lowering insulin resistance, preventing infections, reducing embolisms, and blood viscosity (Simopoulos, 1991, 2002). According to our results, Black Sea trout rich in terms of beneficial fatty acids, DHA, and EPA, along with essential ones such as linoleic acid and alpha-linolenic acid. Marine fish living in cold waters are more affluent in long-chain fatty acids that are important for human nutrition (Farkas et al., 1980; Innis, 1991). Besides, pelagic fishes of cold marine waters have the highest DHA and EPA contents compared to others (Hossain, 2011). The trout individuals were used in this study were caught in rivers having approximately 11 °C water temperature. The environmental conditions are similar to why the culture forms were obtained from the aquaculture facilities on the same river pending the same period. However, as aforementioned before, wild Black Sea trout migrates between sea and freshwater (Kaushik & Seiliez, 2010) and are exposed to very different salinity and temperature conditions. Conversely, cultured trouts are stocked in a fixed area with a high stock density compared to the trouts living in nature (Mazur & Iwama, 1993). The fishes used in our study are at the same age and close length, yet environmental conditions vary. Besides that, wild trouts feed on crustase, diptera, molluscs, and fish species, which are rich in terms of polyunsaturated fatty acids (Kolanowski et al., 2007; Teixeira & Cortes, 2006). Thus, it

is possible to conclude that the differences in fatty acid composition are caused by environmental conditions as well as the feeding regime. Ultimately, it is crucial to know the fatty acid profile of the Black Sea Trout caught from different environmental conditions.

## Conclusion

The Black Sea trout, an endemic species to the Eastern Black Sea, has been widely cultured in recent years and has a high economic return. Although the process of aquaculture of the species has been limited to the last 20 years, scientific studies on the species are extensive. In parallel with the scientific studies and the spread of the aquaculture of the species, consumer demand is increasing day by day. This species, which is preferred by consumers, is rich in terms of meat quality. Although individuals sampled from nature are more abundant in some essential amino acids and unsaturated fatty acids, it has been determined that culture forms values close to individuals from nature. The culture forms obtained from different aquaculture facilities show similar results to different culture lines (F3, F4, F5). With the breeding studies to be carried out on the Black Sea trout, the aquaculture of this nutritious species can be increased.

## Compliance with Ethical Standard

**Conflict of interests:** The authors declare that for this article they have no actual, potential or perceived conflict of interests.

**Ethics committee approval:** All experiments were carried out with approval (ETIK-2017/1) of the Ethical Committee of Animal Experiments of Central Fisheries Research Institute considering the ethical rules of ARRIVE (Animal Research: Reporting of in Vivo Experiments) and European Union directive named as 2010/63/EU.

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