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Seasonal Variation of Total Lipid and Total Fatty Acid in Muscle and Liver of Rainbow Trout (*Oncorhynchus mykiss* W., 1792) Reared in Derbent Dam Lake

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Abstract

Seasonal and monthly variations in the amount of total lipid and fatty acid in muscle and liver of rainbow trout (*Oncorhynchus mykiss*) reared in Derbent Dam Lake were investigated. It was determined that the amount of total lipids and fatty acids both in muscle and liver, varied by months and seasons ($P<0.05$). The amount of total lipid in muscle and liver reached its maximum in autumn. The amounts of total fatty acid both in muscle and liver were determined to be higher in summer, autumn and winter seasons than in spring. Furthermore, the amount of total lipid and fatty acid was higher in liver than in muscle.

Key Words: lipid, fatty acid, muscle, liver, *Oncorhynchus mykiss*

Introduction

Sea and freshwater fish, which constitute the majority of water products, make up an important part of animal food sources for human. Fish are quite different from the other animal food sources, because they provide low energy and have high-level proteins, which contain all essential amino acids. So they are beneficial nutrition sources (Weatherley and Gill, 1998).

Fish are not only beneficial protein source but also contain considerable amount of unsaturated fatty acids, and thus the studies on lipid biochemistry have been considered so important recently (Atchison, 1975; Farkas and Csengeri, 1976; Farkas *et al.*, 1978; Dave *et al.*, 1976; Akpınar, 1986a).

The lipids are the most important biochemical compounds of fish (Akpınar, 1986b). Fish store the lipids in various organs; particularly in muscles and liver. On the contrary, the mammals store in adipose tissue. A great amount of these lipids are transferred to the different parts of the body to be used for various physiological actions (Yılmaz, 1995).

The freshwater fish constitute a great food potential for human. A large part of these fish species are cultivated forms. Since some other new food sources which have the same content are found there is a growing need for information about the chemical composition of the various species and their food contents. For the cultivation of these fish, some important characteristics, such as nutrition properties, biochemical structure and growth conditions need to be known. It is of great importance to know the seasonal variations of the lipids and the lipid amount of the fish, which is economically important and willingly consumed.

The fish lipids are known to contain n-3 series unsaturated fatty acids which reduce the level of serum triglyceride and cholesterol. As a result of this sudden heart attacks ratio and the risk of thrombosis, which is mainly the reason for heart attacks are reduced. Some researchers reported that the n-3 fatty acids facilitate some cancer treatments such as breast tumours (Konar *et al.*, 1999; El-Sayed *et al.*, 1984). In addition to the clear benefits of fish lipids in treatments, it is observed that lack of these essential fatty acids causes some symptoms to appear, such as slow growth, deformation of tail fin, faded and fatty liver, skin depigmentation and being shocked in case of stress (Ackman and Eaton, 1976). The aim of this study is to investigate both the seasonal and monthly variations in the amount of total lipid and fatty acids both in muscle and liver of *Oncorhynchus mykiss* reared economically, which is very important in recent years. Little is known about the variations in the lipid and fatty acids according to the physiological situation of the fish. Therefore, new information will contribute to further projects.

Materials and Methods

The samples of *O. mykiss* used in this study were collected monthly in Derbent Dam Lake from Rainbow trout production Establishments of Derbent between February 1997, and January 1998. Fish were killed by hitting on the heads and brought to the laboratory. Samples were stored at -30°C in a freezer until they were used (Gill and Weatherley, 1984). After measuring the total length and weight of the fish, their gonads were taken out and their sex were determined immediately. Finally, the weight of portion, which can be eaten was determined after removing the pieces such as head, skin, bone and viscera, which are not edible. The body muscles were

removed from the bones and cut into small pieces. Then they were put in a clean blander which had been sterilized beforehand and minced for 30 seconds. For better homogenization of the tissues, 0.25% acetic acid 30-40 ml was added to the minced pieces and they were minced again for 30 seconds until we get a homogenous mixture; like dough (Lathi, 1987). We took 5gr of mixture out of it and used a method in order to purify and extract total lipids and total fatty acids considering the advices of Folch *et al.* (1957), which take place in Christie's (Christiansen *et al.*, 1989).

After extraction, the total lipids in crude extract were separated and weighed. The total lipids were saponified, and the unsaponified portion was discarded. The saponified part was acidified by the addition of 6 M HCI until it reached pH 1; so, total fatty acids were obtained and then total amounts were fixed by weighing. All weights were done in mg using an AND, HM-200 series, 0.0001 g sensitive balance.

Fish samples were taken between the 15th and 25th day of every month. It was taken into consideration that the intervals were equal; so, we had the change to make a comparison between these samples. The livers of the samples were weighed separately and an average value was obtained. Total lipid and fatty acid amounts obtained from the liver extraction were evaluated using percentage based on the liver wet weight. Total fatty acid percentage was also found by means of the total lipid. Monthly average values were obtained from these data. Therefore, a comparison was made between the months. Same procedures were applied to five g of (5 g) muscle of individual fish.

Data obtained were compared using variant analysis and the results were shown as X±SE in tables (Snedecor and Cochran, 1967.).

Results

Monthly variation of total lipid and fatty acid in Liver

When we looked at the samples collected monthly during a year, we saw that the weight of the livers of the samples was the lowest in May and the highest in April. There was some increase in the amount of total lipid depending on wet weight of livers from May to January, which continued during in February and March ($p<0.05$). Generally, the variations in the amount of total fatty acids, except for November and December, showed a variation parallel to the variation in total lipid amount (Table 1).

Monthly variation of total lipid and total fatty acid in Muscle

The amount of total lipid obtained from muscle of *O. mykiss* during generation period (December-May) had a tendency to decrease. It reached the minimum level in May and increased from June to September ($p<0.05$). During the generation season, total fatty acid amount showed regular and important decrease ($p<0.05$); but it increased significantly from June to October ($p<0.05$). It has been observed that the variations of total lipid and total fatty acid in muscle were parallel to each other (Table 2).

Discussion

The amount of total lipid obtained from various species and subspecies of fresh water fish was investigated, and it was observed that the values were between 0.6-30% (Atchison, 1975; Farkas and Csengeri, 1976; Farkas *et al.*, 1978; Dave *et al.*, 1976). In various organs of the fish, the fatty acids composition and lipids contents were affected by the species, sex, age, water temperature, degree of pollution, nutritional condition seasonal variation, fish origin (whether the species was wild or hatchery) (Gill and Weatherley, 1984; Lathi, 1987; Folch *et al.*, 1957; Christiansen *et al.*, 1989). Recent studies have shown that lipid and fatty acid composition was influenced by seasonal variations (Akpınar, 1986a; Akpınar, 1986b; Konar *et al.*, 1999; Agren *et al.*, 1987; Dutta *et al.*, 1985; Yılmaz *et al.*, 1995; Yılmaz *et al.*, 1996). The same research determined that the variations of the levels of lipids both in the liver and other organs are the results of irregular seasonal variations and the water temperature, which affect the fish diet (Agren *et al.*, 1987; Dutta *et al.*, 1985; Yılmaz *et al.*, 1995; Yılmaz *et al.*, 1996; Carroll, 1986; Kinsella, 1987).

The amount of total lipids and fatty acids both in muscle and liver of *O. mykiss* has shown different variations by months and the parts of the body. At the same time, it was observed that the liver stored lipids and fatty acids more than muscles (Table 1, 2, 3, 4). As some researchers explained, these variations have resulted from the biological features of species. In the life span of fish, the most important biological properties are reproduction and nutrition physiology (Atchison, 1975; Farkas and Csengeri, 1976; Farkas *et al.*, 1978; Dave *et al.*, 1976). The fish generally store lipids in their own liver and muscle tissues, but during the process of storage which tissues are important varies according to the fish species. It was reported that active fish stored their lipids in muscle tissues; but the fish inactive living at the bottom of water store their lipids in liver (Castell *et al.*, 1972). Although *O. mykiss* is an active one, it stores more total lipids and fatty acids in liver, since it is a cultivated form (Table 1, 2). Storage lipids vary during reproduction and nutrition periods. It was observed that, especially in the reproduction period, the lipids mobilised from the livers and muscles to the gonads for development of gonads (Castell *et al.*, 1972). While, both in muscle tissues and liver during the reproduction period, the decrease in the amount of total lipid and fatty acid justifies our result. It has

Table 1. The monthly variation of total lipid and fatty acid in liver of *O. mykiss*

Month	Wet weight (g)	Total lipid (mg/g)	Total lipid (%)	Total fatty acid (mg/g)	Total fatty acid percentage by means of total lipid (%)
January	2.53±0.03^{d}	0.349±0.02^{a}	14.5±1.01^{a}	0.191±0.01^{a}	60.8±1.31^{a}
February	2.71±0.06^{c}	0.400±0.07^{b}	15.5±1.12^{b}	0.262±0.09^{c}	64.2±1.08^{a}
March	2.72±0.07^{c}	0.339±0.06^{a}	12.8±1.40^{b}	0.250±0.04^{c}	73.7±1.16^{c}
April	2.99±0.04^{c}	0.333±0.03^{a}	11.2±1.27^{b}	0.193±0.01^{a}	58.6±1.15^{a}
May	1.96±0.04^{a}	0.265±0.09^{c}	13.5±1.30^{a}	0.184±0.01^{a}	61.8±1.03^{a}
June	2.34±0.09^{b}	0.341±0.04^{a}	14.6±1.21^{a}	0.217±0.01^{a}	64.6±1.18^{a}
July	2.45±0.04^{d}	0.347±0.04^{a}	14.8±1.19^{a}	0.241±0.02^{a}	75.1±1.05^{c}
August	2.34±0.02^{b}	0.461±0.09^{b}	18.5±1.15^{c}	0.299±0.08^{b}	64.1±1.21^{a}
September	2.36±0.07^{b}	0.478±0.08^{b}	20.3±1.20^{c}	0.290±0.02^{b}	60.7±1.24^{a}
October	2.00±0.04^{a}	0.433±0.07^{b}	22.3±1.13^{c}	0.299±0.01^{b}	54.1±1.19^{b}
November	2.68±0.03^{c}	0.461±0.04^{b}	17.2±1.10^{b}	0.214±0.02^{a}	46.6±1.03^{d}
December	2.33±0.01^{b}	0.507±0.03^{d}	21.8±1.07^{c}	0.235±0.01^{a}	46.4±1.02^{d}

The data are shown as mean±SE (n=3)
The data written by same character in every column are not different statiscally (p>0.05).

Table 2. The monthly variation of total lipid and fatty acid in muscle of *O. mykiss*

Month	Total lipid (mg/5g)	Total lipid (%)	Total fatty acid (mg/5g)	Total fatty acid percentage by means of total lipid (%)
January	0.252±0.02^{a}	5.06±0.03^{b}	0.174±0.02^{a}	68.8±1.13^{c}
February	0.167±0.01^{dc}	3.33±0.01^{bc}	0.083±0.02^{b}	50.3±1.04^{a}
March	0.137±0.05^{c}	2.69±0.05^{d}	0.077±0.02^{b}	60.9±1.12^{a}
April	0.113±0.01^{b}	2.27±0.04^{d}	0.061±0.02^{b}	54.7±1.09^{a}
May	0.110±0.03^{b}	2.20±0.01^{d}	0.084±0.03^{b}	76.0±1.17^{d}
June	0.130±0.04^{c}	2.60±0.03^{d}	0.103±0.02^{c}	78.1±1.03^{d}
July	0.270±0.03^{a}	5.40±0.01^{b}	0.205±0.04^{d}	76.1±1.20^{d}
August	0.293±0.05^{a}	5.87±0.07^{c}	0.219±0.01^{d}	74.7±1.30^{d}
September	0.315±0.05^{d}	6.31±0.02^{c}	0.241±0.04^{bc}	76.8±1.24^{d}
October	0.302±0.04^{d}	6.04±0.03^{c}	0.249±0.03^{bc}	82.4±1.03^{d}
November	0.252±0.04^{d}	5.04±0.01^{b}	0.193±0.01^{c}	81.3±1.27^{d}
December	0.277±0.03^{d}	5.55±0.04^{b}	0.198±0.01^{c}	71.5±1.18^{c}

Table 3. The seasonal variation of total lipid and fatty acid in liver of *O. Mykiss*

Month	Total lipid (mg/g)	Total lipid percentage by means of wet weight (%)	Total fatty acid by means of wet weight (mg/g)	Total fatty acid percentage by means of total lipid (%)
Winter	0.418±0.05^{b}	17.3±0.02^{c}	0.225±0.05^{a}	56.4±1.31^{b}
Spring	0.326±0.04^{b}	12.5±0.04^{d}	0.209±0.04^{a}	64.7±1.13^{c}
Summer	0.383±0.08^{c}	16.1±0.05^{c}	0.252±0.06^{b}	67.9±1.70^{c}
Autumn	0.457±0.06^{c}	19.9±0.04^{c}	0.267±0.02^{b}	58.4±1.05^{c}

Table 4. The seasonal variation of total lipid and fatty acid in muscle of *O. mykiss*

Month	Total lipid (mg/5g)	Total lipid percentage by means of wet weight (%)	Total fatty acid by means of wet weight (mg/5g)	Total fatty acid prcentage by means of total lipid (%)
Winter	0.232±0.05^{c}	4.65±0.03^{c}	0.146±0.05^{b}	61.6±1.40^{a}
Spring	0.127±0.04^{a}	2.52±0.02^{d}	0.080±0.03^{d}	64.6±1.14^{a}
Summer	0.236±0.09^{c}	4.64±0.02^{c}	0.177±0.08^{a}	76.3±1.30^{b}
Autumn	0.282±0.05^{b}	5.65±0.05^{b}	0.204±0.04^{c}	74.6±1.51^{b}

been observed that the lipids both in muscle and liver vary by months and seasons throughout the year.

When fish find enough food they can control their reproduction and the period of storing lipid. The cycles of storing lipid are directly connected with food abundance. If there is scarcity of food in their environment the variation is low, but if it is abundant, the variation is higher during the year (Ackman and Eaton, 1976; Kluytmans and Zandee, 1973; Kinsella *et al.*, 1977; Mute *et al.*, 1989). Much more energy is needed during the development of gonads; so plenty of food must be available in that period (Wang *et al.*, 1990). The decrease in the amount of total lipid and fatty acid in liver and muscle of fish during the periods of gonad development and reproduction shows that fish supply the required energy from the stored lipids during this period (Gill and Weatherley, 1984; Aggelousis and Lazos, 1991; Stansby *et al.*, 1990; Ackman, 1967; Akpınar, 1987a).

The studies showed that the amount of total lipid in fish species reached maximum level at the end of spawning and during nutrition season; but that amount diminished during reproduction season. Of the species, some spawn in winter, some in spring and summer, and some in winter and spring seasons. Depending on the spawning period, the beginning of nutrition season changes from one species to another. While some species prefer the summer season for this period, some of them prefer the autumn and spring seasons. It was determined that generally reproduction season of the rainbow trout reared in our country since 1969 was between December and May (Çelikkale, 1988). According to the results, the amount of total lipid and fatty acid in muscle and liver has reached its maximum level in autumn; it has reached its minimum in spring. The decrease of lipid amount in this period with the increase of reproduction function, confirmed the opinion that the storing lipids have been consumed (Akpınar, 1987b; Vlaming *et al.*, 1978) and nutrition period has been in summer and autumn. Thus it was understood that since the nutrition period is in spring and autumn, the maximum amount of lipid and fatty acid is available in those seasons (Table 3, 4).

It was expressed that in a study carried out on *Perca fuliviatilis*, *Coregonus albula* and *O. mykiss*, the storage lipids in liver were consumed during gonadial developments. In addition to liver lipids, the stored lipids in muscle tissues were also used during spawning period (Agren *et al.*, 1987; Ackman, 1967). As can be seen in our results, in autumn, winter and summer seasons, the amount of total lipids and fatty acids has been at maximum level, but it has been at minimum level in spring season. Thus we can draw the conclusion that during spring season, especially until the process of reproduction is completed in May, the decrease of lipid amount both in muscle tissues and liver is for the reproduction; and storing lipid in summer season is necessary for winter (Table 1, 2, 3, 4).

With the increase of population, we should benefit from fish and fish products both for a balanced nutrition and for the food needed. Besides this, we must pay attention to the proportion of carbohydrate and lipid of food rations of the fish during reproduction period, which will be used as a source of food or reared and which have carnivorous characteristic.

Thus both lipid requirements of the stock will be covered and the healthy spawns will be reproduced and the right amount of lipid and fatty acids will be supplied for the growth and the development of the following life cycles.

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