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Original Scientific Article

COMPARISON OF THE CHEMICAL COMPOSITION AND NUTRITIONAL VALUES OF FRESH AND FROZEN RAINBOW TROUT

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Summary: The aim of this work was a comparison of the chemical compositions (including water, carbohydrate, protein, fat, ash content, mineral substances and fatty acids profiles and nutritional value) of fresh refrigerated and frozen rainbow trout (*Oncorhynchus mykiss*). Fillets of rainbow trout (without skin) obtained from aquaculture, and divided into two groups, trout cooled and stored at 0–4 °C and trout frozen and stored at –18 °C, were used. By providing the basic chemical composition of rainbow trout fillets, we determined that the water content was 73 %, protein content 18.5 %, fat content 2.8 %, and ash content 1.15 %; there were no statistically significant differences when comparing fresh and frozen trout. Based on the profile of fatty acids, the most representative n-3 polyunsaturated fatty acids (PUFAs) in rainbow trout were docosahexaenoic acid (DHA) 12.7 % and eicosapentaenoic acid (EPA) 4.5 %. PUFAs n-6 consisted primarily of linoleic acid (14.5 %), mono-unsaturated fatty acids of oleic acid (21.4 %), and saturated fatty acids palmitic acid (13 %). The sum of total PUFAs represents 42.9 %, when n-3 PUFAs represent 22.55 % and n-6 PUFAs 20.36 %. Comparing the nutritional value of rainbow trout fillets calculated from the basic chemical composition, statistically significant differences between fresh (106.34 kcal) and frozen (106.11 kcal) fillets were not found, which means that these different ways of extending shelf life should not affect the nutritional value.

Key words: chemical composition; fatty acids; freezing; nutritional value; trout

Introduction

In 2008, fishing and aquaculture produced worldwide approximately 142 million tons of fish, of which 115 million tons were used for human consumption with an estimated conversion of about 17 kg (body weight) per capita. Aquaculture accounted for 46 % of total fish supply for consumption in 2008, which is an increase from 43 % in 2006. Aquaculture continues to be the fastest growing sector of food-producing animals (1). In 2008, diadromous fish production was dominated by salmon farming (1.5 million tons), milkfish (0.68 million tons), rainbow trout (0.58 million tons) and eel (0.26 million tons) (2). Aquaculture is of increasing importance for the supply of fish such as trout and salmon to industrialised countries. Highly effective production has been attained through the establishment of breeding programmes, the optimisation of feeds, and improved disease treatment (3). The quality of farmed fish and of products made from farmed fish strongly depends on the rearing conditions and, in particular, on the feed composition (4).

The chemical compositions of fish species show differences according to season, migratory

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behaviour, sexual cycle and feeding cycles. These factors are mainly observed in wild fish. Farmed fish in aquaculture may also exhibit differences in chemical composition, but in such cases there factors are monitored so that the chemical composition can be predicted. High-quality dietary fish meat is a result of its higher proportion of simple protein, favourable fat composition, high lipophilic vitamins content, the softness of muscle fibres, the absence of elastin in connective tissue and relatively high mineral content. The composition of fish meat is dependent on the type of fish, season, sexual cycle, aquatic environment, age and sex, when water content varies in the range of 50 to 80 %, protein content 15 to 25 %, fat content 0.1 to 35 % and mineral substances 0.8 to 2 % (5).

Lipids are vital components of fish diets due to their role in providing energy sources, the essential role of some fatty acids, carriers of fat soluble vitamins and resource of polar lipid plus sterols, which are essential structural compounds of cell membranes. Polyunsaturated fatty acids (PUFA) and their derivatives, eicosanoids, are highly biologically active substances of a lipid nature. They are divided according to the position of the first double bond nearest the methyl end of the n-3, n-6, n-9 unsaturated fatty acids. Mammalian bodies are able to synthesize double bonds of n-9 position toward the carboxyl end of fatty acids. However, they are not able to form double bonds to position n-3 and n-6; these fatty acids must be obtained from the diet and, therefore, are considered to be essential (6). Fatty fish, such are tuna, mackerel, herring, anchovy and salmon, and fish oils contain essential n-3 polyunsaturated fatty acids known as eicosapentaenoic acid (EPA) (C20:5 n-3) and docosahexaenoic acid (DHA) (C22:6 n-3) (7).

The aim of this work was the determination of the chemical composition, including water, protein, fat, ash content, mineral substances profile and fatty acids profiles, and the calculation of nutritional value of fresh refrigerated and frozen trout.

Materials and methods

Twenty-four rainbow trout (*Oncorhynchus mykkis*) were raised at the Rybarstvo Požehy farm. Fish with an average body weight of 300 g were eviscerated, washed, cooled, and vacuum packaged to avoid surface dehydration, and subjected to two different treatments. The first group (n = 12) was placed directly into a cold room (0 to 2 °C) and stored for five days. The second group (n = 12) was individually quick frozen (IQF) until the temperature of the fish cores had reached –18 °C. The frozen trout were immediately stored in a freezing room (–18 °C) for six months for further analyses. Subsequently, the samples were thawed in cold conditions (0–2 °C) until the fish cores had reached –1 °C.

The basic chemical parameters, including profiles of mineral substances and fatty acids of twelve fresh rainbow trout, were performed immediately after cooling. The basic chemical parameters (water, carbohydrate, protein and fat contents) were examined in fresh vacuum-packed trout after five days of storage and in frozen samples after six months of storage and subsequent thawing, and nutritional values were calculated.

Determination of chemical composition

The basic chemical composition (water, carbohydrate, protein, fat and ash content) was determined according to AOAC Official methods of analysis (8). Samples for the determination of the profile of mineral substances were prepared using microwave degradation in an MLS-1200 MEGA (fy Milestone) digestion system; analysis of minerals (Ca, Mg, Na, K, Cu, Zn, Mn, Fe) was carried out using Flame Atomic Absorption Spectroscopy (FAAS) Solar 939 (fy Unicam). Phosphorus content was analysed via spectrophotometry.

Determination of profile of fatty acids

Total lipids in samples were isolated using chloroform/methanol according to Čertík et al. (9). Methylesters of fatty acids were measured in rainbow trout oil by gas chromatography using GC-6890 N (Agilent Technologies) according to Čertík et al. (10). On the column DB-23 (50 %-cyanopropyl-methylpolysiloxan, length 60 m, diameter 0.25 mm, width of film 0.25 µm), 1 µl of fatty acids methylesters was automatically injected, and then the samples were analysed under following conditions: carrier gas: hydrogen (44 cm/s at 130 °C), temperature of injection: 220 °C, split: 1:50, FID detector (250 °C, flow of hydrogen: 30 ml/min, flow of oxygen: 500 ml/

min), temperature regime: 130 °C - 1 min, 130–170 °C – 6.5 °C/min, 170–215 °C – 2.7 °C/min, 215 °C - 7 min, 220–240 °C – 20 °C/min, 240 °C - 2 min). Records were evaluated using ChemStation B0103 (Agilent Technologies) and quantified on the basis of retention times of known standards of fatty acids C4–C24 (Sigma, USA).

Calculation of nutrition value

The energy value to be declared in rainbow trout was calculated using the following conversion factors (11): carbohydrate 4 kcal/g and 17 kJ/g; protein 4 kcal/g and 17 kJ/g and fat 9 kcal/g and 37 kJ/g.

Statistical evaluation

The mean values and standard deviations were calculated by using column statistics with processing of six values for each analysed group. Statistically significant differences between groups were calculated using t-test in the program GraphPad Prism 5 (2007). Differences were evaluated as statistically significant when this P value was < 0.05.

Results

In fresh rainbow trout fillets before vacuum packing, the basic chemical composition (water, protein, fat, ash content and profile of mineral substances) listed in Table 1 and the profile of the fatty acids found in fish oil (Table 2, Figure 1) were determined.

By providing the basic chemical composition of rainbow trout fillets, we determined that the average water content was 73 %, protein content 18.5 %, fat content 2.8 %, and ash content 1.15 %. Based on the determination of profile of mineral substances, these are listed by order of concentrations in Table 1.

Based on the profile of fatty acids, the most representative polyunsaturated fatty acids (PUFA) n-3 series found in oil of rainbow trout were docosahexaenoic acid (DHA) at 12.7 % and eicosapentaenoic acid (EPA) at 4.5 %. The PUFA n-6 series consisted primarily of linoleic acid (14.5 %), monounsaturated fatty acids (MUFA) oleic acid (21.4 %), and saturated fatty acids (SFA), palmitic acid (13 %). The amount of total PUFA represents 42.9 % of the n-3 PUFA are 22.554 % and n-6 PUFA 20.361 %.

The basic chemical compositions (water content, protein content, fat content) in the fresh chilled and frozen trout fillets, vacuum packed, were determined (Table 3).

In comparing the chemical composition of both groups, no statistically significant differences (P>0.05) were found.

Based on the chemical composition, the nutritional value of chilled and frozen trout fillets was calculated using conversion factors (11) (Table 4).

Table 1: Chemical composition of fillets from rainbow trout

Parameter	Value
Water g.100 g^{-1}	73.03 ± 0.66
Protein g.100 g^{-1}	18.57 ± 0.52
Fat g.100 g^{-1}	2.81 ± 0.73
Ash g.100 g^{-1}	1.15 ± 0.38
Potassium g.100 g^{-1}	0.425
Phosphorus g.100 g^{-1}	0.240
Calcium g.100 g^{-1}	0.091
Magnesium g.100 g^{-1}	0.051
Sodium g.100 g^{-1}	0.050
Iron mg.100 g^{-1}	2.840
Manganese mg.100 g^{-1}	1.560
Zinc mg.100 g^{-1}	1.020
Copper mg.100 g^{-1}	0.040

Table 2: Profile of fatty acids in rainbow trout oil

Fatty acids	Formulas	Value (%)
Myristic acid	C 14:0	3.2
Pentadecanoic acid	C 15:0	0.3
Palmitic acid	C 16:0	13.0
Palmitoleic acid	C 16:1-9c	5.1
Hexadecadienoic acid	C 16:2-9c,12c	0.8
Heptadecanoic acid	C 17:0	0.3
Stearic acid	C 18:0	3.0
Oleic acid	C 18:1-9c	21.4
Cis-vaccenic acid	C 18:1-11c	3.0
Linoleic acid n-6	C 18:2-9c,12c	14.5
Alpha linolenic acid n-3	C 18:3-6,9,12c	0.4
Gamma linolenic acid n-6	C 18:3-9,12,15c	2.0
Octadecatetraenoic acid n-3	C 18:4-6,9,12,15c	1.2
Eicosenoic acid	C 20:1-11c	4.8
Eicosadienoic acid n-6	C 20:2-11c,14c	0.8
Dihomo-linolenic acid n-6	C 20:3-8,11,14c	0.4
Arachidonic acid n-6	C 20:4-5,8,11,14c	0.8
Eicosatrienoic acid n-3	C 20:3-11,14,17c	0.2
Eicosatetraenoic acid n-3	C 20:4-8,11,14,17c	0.8
Eicosapentaenoic acid n-3	C 20:5-5,8,11,14,17c	4.5
Docosanoic acid	C 22:0	0.1
Erucic acid	C 22:1-13c	3.7
Docosadienoic acid n-6	C 22:2-13c,16c	0.1
Docosapentaenoic acid n-3	C 22:5-7,10,13,16,19c	1.7
Docosahexaenoic acid n-3	C 22:6-4,7,10,13,16,19c	12.7
Tetracosenoic acid	C 24:1	0.0

Table 3: Chemical composition of fresh and frozen fillets from rainbow trout

	Water	Protein	Fat	Carbohydrates
Fresh fillets	76.84 ± 0.58	18.77 ± 0,46	2.74 ± 0.62	1.65 ± 0.28
Frozen fillets	76.90 ± 0.81	18.35 ± 0.66	2.99 ± 1.11	1.45 ± 0.41

Table 4: Nutritional value of fresh and frozen fillets from rainbow trout

Component	Fresh fillets			Frozen fillets		
	$g.100\ g^{-1}$	Kcal	kJ	$g.100\ g^{-1}$	Kcal	kJ
Protein	18.77	75.08	319.09	18.35	73.4	311.95
Fat	2.74	24.66	101.38	2.99	26.91	110.63
Carbohydrates	1.65	6.6	28.05	1.45	5.8	24.65
Energy value in 100 g		106.34	448.52		106.11	447.23

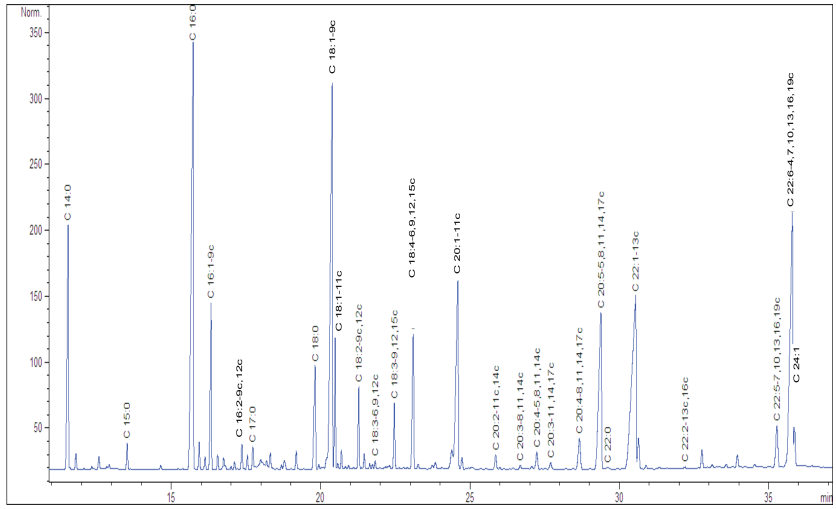

Figure 1: Chromatographic record of fatty acids profile in rainbow trout oil

Comparing the nutritional value of rainbow trout fillets calculated from the average values of basic chemical composition, no statistically significant differences (P>0.05) between fresh (106.34 kcal) and frozen (106.11 kcal) fillets were found.

Discussion

Fish meat consists of water, proteins and other nitrogenous compounds, lipids, carbohydrates, vitamins and minerals. The water content in the whole body of the trout was 70 %, and 73 % in the fillets, protein content 17.25 and 20.03 %, respectively, and the fat content 10.58 % throughout the whole body, compared with 5.18 % in the fillets. The ash content was 2.36 % in the whole fish and 1.48 % in trout fillets (3). These values are comparable to our results achieved in the determination of chemical composition in rainbow trout fillets. However, the biggest differences can be in fat content, when the content of total lipids in different species of freshwater fish can range between 0.6 to 30 %. In the various organs of fish, there were impacts on the content and composition of fatty acids and lipids fish species, sex, age, water temperature, degree of pollution, nutritional status, seasonal variations and origin of fish (12).

Lipids and fatty acids play a significant role in membrane biochemistry and have a direct impact on membrane-mediated processes, such as osmoregulation, nutrient assimilation and transport. However, the nature and quantity of these lipids in fish vary according to species and habitat (6). It is known that n-3 fatty acids are essential for neural development in infants and during the first few years of life, and have beneficial effects on hypertension, inflammation, arrhythmias, psoriasis, aggression, depression, coronary heart disease, inflammatory and auto-immune disorders and cancer (13). The fatty acid composition of fish is highly variable between and within species. It has been indicated that these variations are influenced by a number of factors, such as food availability and nutritional habits of fish, catch area, fish size, age, maturity, season and sampling tissue (14).

Based on the profile of fatty acids, the most representative polyunsaturated fatty acids (PUFA) n-3 series found in oil of rainbow trout were docosahexaenoic acid (DHA) and eicosapentaenoic acid (EPA); the PUFA n-6 series consisted primarily of linoleic acid. The amount of total PUFA is 42.9 %; 22.554 % of which is n-3 PUFA and 20.361 % is n-6 PUFA. The optimal composition of fatty acids in the diet is a decisive factor that plays a role in preventing disease and improving health. Generally, in Europe, consumers are experiencing the effects of an increased intake of fats in their diet; therefore, the factor of composition of the fatty acids and their ratio is even more important. The recommended ratio of polyunsaturated fatty acids is 1 portion of n-3 PUFAs to 2 portions of n-6 PUFAs. In the current diet based on the consumption of red and white meat, PUFAs are imbalanced in the ratio from 1:10 to 1:50. In our study, ratio of polyunsaturated fatty acids in rainbow trout oil 1.1 portion of n-3 PUFAs to 1 portion of n-6 PUFAs was found.

The fatty acid (FA) composition of some tissues of *Salmo trutta labrax* in Turkey was investigated. Fatty acids profiles of muscle, liver, gonad, egg and adipose tissue were compared in terms of total and individual saturated and unsaturated fatty acids. The results of the present study have revealed that the most abundant individual FAs were palmitic, oleic and docosahexaenoic acids (DHA) in most of the tissues. In general, SFA was higher in the muscle tissue while the MUFAs were dominant in the gonads. There was also a significant difference between the PUFA profiles of the tissues. For example, total n-3 PUFAs were 4.8 to 7.7-fold higher than that of the n-6 PUFAs, and the eggs had the highest n-3 PUFA (48.09 ± 9.38) content (15).

The fatty acid composition of liver and muscle tissues of immature and mature *Oncorhynchus mykiss* fed on two different diets were determined. Fatty acid analyses were carried out using gas chromatography. Palmitic acid (C16:0), oleic acid (C18:1 n-9), linoleic acid (C18:2 n-6) and docosahexaenoic acid (C22:6 n-3) were the key components in both liver and muscle tissues of immature and mature rainbow trout of both sexes. The amounts of C22:6 n-3 were higher in the liver (29.04 ± 0.06 – 27.41 ± 0.17 %) and muscle (13.05 ± 0.40 – 11.37 ± 0.21 %) of immature fish than in mature fish, and depended on the composition of the diet. The results of this study have shown that fatty acid composition in fish tissues can vary considerably, depending on the age of fish and their diet (14).

It is well known that lipid oxidation is one of the most significant problems in fish processing and subsequent storage and shelf-life (16). Unsaturated fatty acids are more easily oxidizable than saturated acids, and thus it is assumed that the fatty fish whose higher fat content are subject to oxidative changes faster than lean fish with a standard diet (17). Due to oxidation, the organoleptic characteristics of fish are also ultimately adversely affected (7).

It is known that fish meat has high nutritional and biological value. The muscle tissue contains approximately 15–20 % protein, and fish proteins have, in contrast to mammals and poultry, favourable amino acid composition. Fish is a rich source of mineral substances; small bones softened by processing (e.g. marinated fishery products) can be eaten along with the meat and thus become a valuable source of phosphorus, calcium, iodine and selenium. However, by comparing the nutritional value of rainbow trout fillets calculated from the basic chemical composition, no statistically significant differences between fresh cooled (106.34 kcal) and frozen (106.11 kcal) fillets were found, which means that these different ways of extending shelf life should not affect the nutritional value.

In conclusion, the beneficial effects of a diet rich with n-3 PUFA for humans are a reason for the regular consumption of fish and encourage aquaculturists to find alternatives to increase fish production. Fish can make a significant contribution toward reducing fat in people's diet. Not only is fish low in fat, but it is also a tasty, highly nutritious and wholesome food that can offer an endless variety to menus. Rainbow trout is an excellent meat group choice because it is lower in fat and calories than some foods from the meat group and is also an excellent source of many essential nutrients. The presented fish species with its high n-3 PUFA content verified in this research could be a potential healthy food fish in terms of the positive effects of EPA and DHA in the diet.

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PRIMERJAVA KEMIČNE SESTAVE IN HRANILNE VREDNOSTI SVEŽE IN ZAMRZNJENE ŠARENKE

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Povzetek: Namen našega dela je bil primerjati kemično sestavo (vsebnost vode, ogljikovih hidratov, beljakovin, maščob, pepela, mineralnih snovi in maščobnih kislin) in hranilno vrednost sveže oz. ohlajene in zamrznjene šarenke (*Oncorhynchus mykiss*). Fileti iz šarenk (brez kože), pridobljeni iz ribogojnic, so bili razdeljeni v dve skupini, v ohlajene in shranjene pri 0-4 °C ter v zamrznjene in shranjene pri -18 °C. S kemično analizo smo ugotovili, da vsebujejo 73 % vode, 18,5 % beljakovin, 2,8 % maščob in 1,15 % pepela. Ugotovili smo, da ni statistično značilnih razlik v kemični sestavi filetov iz šarenk med načinoma shranjevanja. Da bi ugotovili vpliv načina shranjevanja na njihovo hranilno vrednost, smo v njih analizirali tudi vsebnost maščobnih kislin. Fileti šarenke vsebujejo največji delež dokozaheksaenojske kisline (DHA) - 12,7 % in eikozapentaenojske kisline (EPA) - 4.5 % izmed n-3 polinenasičenih maščobnih kislin. Med n-6 polinenasičenimi maščobnimi kislinami je najvišji delež linolne kisline (14,5 %), med mononenasičenimi maščobnimi kislinami delež oleinske kisline (21,4 %) in med nasičenimi maščobnimi kislinami delež palmitinske kisline (13 %). Polinenasičene maščobne kisline predstavljajo 42,9 % , med temi n- 3 22,55 % in n-6 20,36 %. Na osnovi kemijčne analize smo primerjali hranilno vrednost filetov šarenke in ugotovili, da ni statistično značilnih razlik med svežimi (106,34 kcal) in zamrznjenimi (106,11 kcal), kar kaže, da različna načina podaljšanja roka uporabnosti ne vplivata na prehransko vrednost.

Ključne besede: kemična sestava; maščobne kisline; zamrzovanje; hranilna vrednost; postrv