

## Rainbow Trout Composition and Fatty Acid Content in Estonia

L. Timberg<sup>1,2</sup>, R. Kuldjärv<sup>1,2</sup>, K. Koppel<sup>1,2</sup> and T. Paalme<sup>1,2</sup>

<sup>1</sup>Competence Centre of Food and Fermentation Technologies, Akadeemia tee 15B, 12618 Tallinn, Estonia; e-mail: loreida@tftak.eu; rain@tftak.eu; kadri@tftak.eu

<sup>2</sup>Tallinn University of Technology, Department of Food Processing, Ehitajate tee 5, 19086 Tallinn, Estonia; e-mail: tpaalme@staff.ttu.ee

**Abstract.** Rainbow trout (*Oncorhynchus mykiss*) is the most popular aquaculture species in Estonia. The aim of the present study was to examine and compare moisture, protein, lipid and fatty acid (FA) compositions in Rainbow trout from different fish farms in Estonia and that farmed in Finland and Norway. The total lipid content in different Rainbow trout varied more than 5.5 fold, but FA proportions were very similar in all Rainbow trout. However, it is important to note that Estonian farmed Rainbow trout had generally lower lipid content and therefore also a lower amount of essential FAs.

**Key words:** Fatty acid, lipid, Rainbow trout (*Oncorhynchus mykiss*)

### INTRODUCTION

Natural resources of fish can no longer fulfil demands of fish consumers; the shortage is forcing the aquaculture sector to expand. There are about 15 fish farms in Estonia where Rainbow trout is cultured. The annual volume of Estonian farmed Rainbow trout is about 700 tons, but many fish farms are expanding; production is expected to double in the next few years. Therefore, the Estonian aquaculture sector is interested in producing Rainbow trout which has high nutritional value, stable quality and is also compatible with Rainbow trout farmed in other countries.

Fat is one of the most important components of fish meat. It attracts consumers' attraction due to the fatty acid (FA) profile, especially n-3 and n-6 FAs (Ruxton et al., 2004; Breslow, 2006). Therefore, the main aim of the study was to characterize and compare the moisture, protein, lipid and FA profiles of Rainbow trout from different fish farms in Estonia and imported Rainbow trout available in Estonian supermarkets.

### Experiment design

Rainbow trout samples from ten different aquaculture facilities in Estonia were acquired (samples E1–E10). Fish were gutted, packed in ice and transported to the laboratory on the day of slaughter; all analyses were performed the next day. Three samples of Rainbow trout (imported) cultured in other countries were purchased from Estonian supermarkets (sample T1–Finland, T2–Norway, and T3–Finland). The imported trout had been slaughtered 4–6 days before purchase and had already been gutted, packed in plastic bag, and transported to the laboratory within an hour after purchase, where the fish was immediately packed in ice. All analyses were performed

the following day. All measurements were carried out in three repetitions.

The moisture content of the fish samples was measured using a halogen moisture analyzer (HR 83, Mettler Toledo, Switzerland). The protein content of the fish samples was measured by Kjeldahl method (Velp Scientifica UDK 142, Italy). The lipid content of fish was measured by Soxhlet method (Velp Scientifica SER 148 Solvent Extractor, Italy).

The fatty acid profile of trout samples was determined as fatty acid methyl esters (FAMES). The Bligh & Dyer (1959) method was used for lipid extraction. The FAMES were prepared according to the standard EVS–EN ISO 5509:2000. The prepared methyl esters samples were injected into the gas chromatograph (Agilent 7890A GC System) equipped with a flame ionization detector (FID) at a split ratio of 1:10. Helium served as the carrier gas (flow 1 ml per min). Agilent J&W GC Column HP–88 (60 m x 0.25 mm x 0.2 µm) was used for the separation of FAMES. The analytical conditions were: injector port temperature –250°C and detector temperature –280°C. The oven was programmed from 125–230°C. Retention times of FAMES of the standard mixture were used to identify chromatographic peaks of the sample. Supelco 37 Component FAME mix was used as standard FAME mixture. Fatty acid content in the samples was calculated, based on the peak area ratio and expressed as g fatty acid per 100 g lipid.

All necessary reagents were purchased from Sigma-Aldrich, Germany.

### Statistical Analysis

XLSTAT (2010, AddInsoft, France) was used for lipid, moisture, protein, and FA ( $P = 0.05$ ) Analysis of Variance (ANOVA) between samples. Principal Component Analysis (PCA) was used to visualize relations between samples. Statistically significant correlations (Pearson,  $P = 0.05$ ) are given in this paper. Samples were clustered using K-means clustering according to the lipid content.

## RESULTS AND DISCUSSION

Moisture, lipid and protein composition of Estonian farmed and imported Rainbow trout are shown in Table 1. The moisture content was in the range of 63.8–73.4%, the lipid content was in the range of 2.1–11.6%, and the protein ranged from 19.7–23.1%. There was a strong negative correlation between water and lipid content of the Rainbow trout ( $R = -0.92$ ,  $P = 0.05$ ). According to the lipid content clustering analysis (k-means) was performed, which indicated that there were three Rainbow trout groups: group 1 (G1)–lipid content 2.1–3.9%; group 2 (G2)–lipid content 4.6–7.1%; and group 3 (G3)–lipid content 9.3–11.6%.

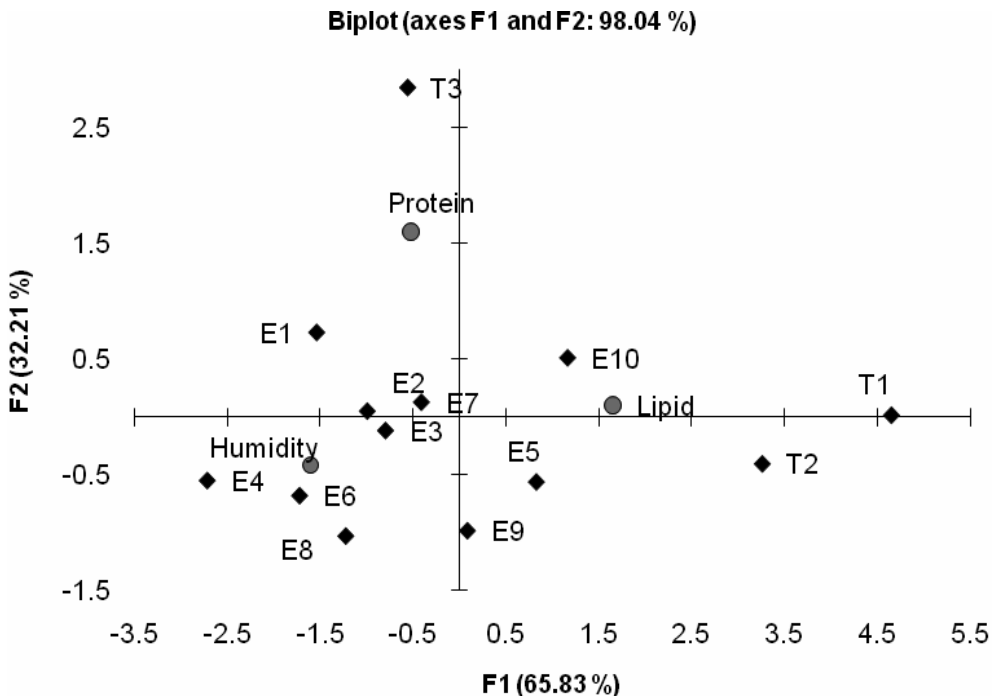
The PCA plot (Figure 1) shows the location of the Rainbow trout in multivariate space according to the first (PC1) and second (PC2) principal component. The first and second principal components explained 98% of the total variance between the samples. The PCA plot confirms Rainbow trout grouping into three groups as the first principal component divides the samples according to their lipid contents. Sample T3 was higher in protein content than the rest of the samples.

The FA contents of Estonian farmed and imported Rainbow trout are shown in Table 2. In all samples, C16:0, C18:1, and docosahexaenoic acid (DHA, C22:6n–3) were dominant, which has also been observed by other researchers (Blanchet et al., 2005; Suzuki et al., 1986). The linoleic acid (18:2n–6) content in all analyzed Rainbow

**Table 1.** Moisture, lipid and protein composition (g per100 g wet meat) in Rainbow trout.

Sample	Moisture	Protein	Lipid
E1	70.3 ± 0.4	21.3 ± 0.0	3.0 ± 0.2
E2	71.1 ± 0.3	20.7 ± 0.3	4.6 ± 0.1
E3	71.1 ± 0.3	20.5 ± 0.2	4.9 ± 0.0
E4	73.4 ± 0.7	20.4 ± 0.3	2.1 ± 0.2
E5	67.6 ± 1.2	19.8 ± 0.0	5.3 ± 0.4
E6	72.4 ± 0.2	20.2 ± 0.5	3.5 ± 0.1
E7	70.3 ± 0.4	20.7 ± 0.2	5.3 ± 0.2
E8	71.8 ± 1.5	19.8 ± 0.2	3.9 ± 0.1
E9	70.0 ± 1.4	19.6 ± 0.9	5.4 ± 0.4
E10	67.6 ± 0.9	20.8 ± 0.9	7.1 ± 0.6
T1	63.8 ± 0.6	19.9 ± 0.2	11.6 ± 0.3
T2	65.2 ± 1.0	19.7 ± 0.1	9.3 ± 0.5
T3	68.2 ± 0.2	23.1 ± 0.3	5.2 ± 0.1

\*Values are mean ± SE



**Figure 1.** PCA of the moisture, lipid and protein content of Rainbow trout samples.

**Table 2.** Fatty acid composition (g per 100 g lipid) in Rainbow trout.

Fatty acids	E1	E2	E3	E4	E5	E6	E7	E8	E9	E10	T1	T2	T3
C14:0	4.4 ± 0.1	4.8 ± 0.1	4.2 ± 0.1	4.9 ± 0.2	4.9 ± 0.1	4.0 ± 0.1	4.5 ± 0.3	3.8 ± 0.1	5.2 ± 0.2	4.9 ± 0.1	4.2 ± 0.1	4.2 ± 0.0	5.0 ± 0.2
C16:0	17.4 ± 0.1	17.7 ± 0.3	16.0 ± 0.1	17.7 ± 0.2	14.7 ± 0.2	15.6 ± 0.2	15.1 ± 0.5	16.0 ± 0.3	16.9 ± 0.3	16.6 ± 0.1	13.8 ± 0.4	13.1 ± 0.0	15.0 ± 0.3
C18:0	3.4 ± 0.0	3.0 ± 0.1	3.1 ± 0.0	2.8 ± 0.0	2.2 ± 0.0	2.8 ± 0.1	2.7 ± 0.2	3.2 ± 0.1	3.1 ± 0.1	3.2 ± 0.1	3.0 ± 0.1	2.8 ± 0.1	3.9 ± 1.1
C16:1	5.6 ± 0.1	5.3 ± 0.1	5.3 ± 0.1	5.5 ± 0.1	5.1 ± 0.0	4.8 ± 0.0	5.3 ± 0.1	4.8 ± 0.0	5.6 ± 0.2	5.2 ± 0.1	5.4 ± 0.1	4.9 ± 0.0	5.9 ± 0.2
C18:1	23.9 ± 0.3	25.9 ± 0.4	30.8 ± 0.2	19.5 ± 0.2	27.4 ± 0.4	27.0 ± 0.2	28.9 ± 0.3	28.2 ± 0.1	26.7 ± 0.3	28.7 ± 0.1	32.6 ± 0.5	32.7 ± 0.0	23.8 ± 0.5
C20:1	2.5 ± 0.0	2.7 ± 0.0	3.1 ± 0.0	3.6 ± 0.0	3.4 ± 0.1	2.5 ± 0.0	3.0 ± 0.0	2.4 ± 0.0	3.6 ± 0.0	3.6 ± 0.0	3.3 ± 0.1	3.3 ± 0.0	6.2 ± 0.0
C18:2	7.7 ± 0.1	8.7 ± 0.1	9.7 ± 0.1	7.0 ± 0.1	9.5 ± 0.3	8.9 ± 0.1	9.6 ± 0.0	11.1 ± 0.0	8.5 ± 0.1	9.3 ± 0.1	11.6 ± 0.2	11.9 ± 0.1	9.7 ± 0.2
C18:3	2.6 ± 0.0	3.1 ± 0.0	3.5 ± 0.0	2.6 ± 0.0	3.8 ± 0.1	3.7 ± 0.0	3.9 ± 0.0	3.4 ± 0.0	3.7 ± 0.0	3.9 ± 0.0	4.4 ± 0.1	4.4 ± 0.0	2.2 ± 0.1
EPA	7.3 ± 0.1	6.8 ± 0.0	5.0 ± 0.1	5.7 ± 0.1	5.4 ± 0.2	6.1 ± 0.1	6.1 ± 0.2	6.4 ± 0.1	4.6 ± 0.1	4.2 ± 0.0	5.0 ± 0.0	4.8 ± 0.0	5.1 ± 0.1
DHA	22.4 ± 0.6	19.2 ± 0.3	16.6 ± 0.4	27.8 ± 0.7	20.7 ± 0.9	21.9 ± 0.5	18.0 ± 1.0	17.9 ± 0.3	19.1 ± 0.8	17.4 ± 0.2	13.8 ± 0.2	14.8 ± 0.1	20.6 ± 0.9
SFA	25.4 ± 0.1	25.8 ± 0.4	23.5 ± 0.1	25.6 ± 0.4	22.0 ± 0.3	22.6 ± 0.3	22.7 ± 0.9	23.3 ± 0.5	25.5 ± 0.4	25.1 ± 0.3	21.3 ± 0.6	20.5 ± 0.1	24.0 ± 0.8
MUFA	32.9 ± 0.4	34.8 ± 0.5	39.9 ± 0.3	29.6 ± 0.2	36.7 ± 0.6	35.1 ± 0.2	37.9 ± 0.3	36.2 ± 0.1	36.7 ± 0.5	38.2 ± 0.0	42.0 ± 0.6	41.6 ± 0.0	36.5 ± 0.8
PUFA	41.7 ± 0.5	39.4 ± 0.1	36.6 ± 0.3	44.8 ± 0.6	41.2 ± 0.3	42.3 ± 0.6	39.4 ± 1.3	40.5 ± 0.4	37.8 ± 0.8	36.7 ± 0.2	36.7 ± 0.2	37.9 ± 0.1	39.5 ± 0.8
n3	32.5 ± 0.6	29.3 ± 0.2	25.3 ± 0.4	36.3 ± 0.7	30.1 ± 0.6	31.9 ± 0.6	28.2 ± 1.2	27.8 ± 0.4	27.7 ± 0.9	25.7 ± 0.2	23.5 ± 0.2	24.3 ± 0.1	28.1 ± 0.9
n6	9.2 ± 0.1	10.2 ± 0.1	11.3 ± 0.1	8.5 ± 0.1	11.1 ± 0.3	10.4 ± 0.1	11.2 ± 0.1	12.8 ± 0.0	10.1 ± 0.0	11.0 ± 0.1	13.2 ± 0.2	13.6 ± 0.1	11.3 ± 0.2
n3:n6	3.6 ± 0.1	2.9 ± 0.1	2.2 ± 0.1	4.3 ± 0.1	2.7 ± 0.1	3.1 ± 0.1	2.5 ± 0.1	2.2 ± 0.0	2.7 ± 0.1	2.3 ± 0.0	1.8 ± 0.0	1.8 ± 0.0	2.5 ± 0.1

\*Values are mean ± SE. Only the major FAs ( $\geq 2\%$  and above) are listed. The other FAs are C12:0, C14:1, C18:3n-6, C20:0, C20:1, C20:2n-6, C20:3n-6, C22:0, C20:3n-3, C22:1, C20:4n-6, C22:2n-6, C24:0. The full table is available on request from the corresponding author.

trout ranged from 7.0–11.9g per 100g lipid. Linoleic acid was the major n–6 polyunsaturated fatty acid (PUFA) in Rainbow trout and is responsible for the n–3 per n–6 PUFA ratios of 1.8–3.6, which correlates well with earlier research results by Blanchett et al. (2005). According to n–3 per n–6 ratio Rainbow trout quality can be optimized, because linoleic acid comes mostly from plant–derived oils that are used in fish feed. According to Bell et al. (2001) n–3 per n–6 ratio can be improved by using feed which consists of at least two–thirds fish meal. DHA in Rainbow trout varied from 13.8 to 27.8g per 100g lipid, which is higher compared to salmon DHA values (13.115.2g per 100g lipid; Blanchett et al., 2005). Most fish species can desaturate and elongate 18:2(n–6) and 18:3(n–3) to their C20 and C22 homologues (Henderson, 1996) and this also explains strong negative correlations found in this study between DHA and 18:2n–6 (–0.85) and 18:3n–3 (–0.72), since lower PUFA diet stimulates the conversion of 18:3n–3 to DHA (Bell et al., 2001). EPA in Rainbow trout varied from 4.2 to 7.3g per 100g lipid, which is also in agreement with previous studies by Blanchett et al (2005).

The total lipid content in different Rainbow trout varied mostly more than 5.5 fold, but FA proportions were very similar in all trout. In order to evaluate possible significant differences among three Rainbow trout groups with different lipid composition, ANOVA analysis was performed on FA composition of groups, and results are shown in Table 3. Saturated fatty acid (SFA) content was similar in groups G1 and G2, but different in group G3. Monounsaturated fatty acid (MUFA) and n–3 content was significantly different in all fish groups. PUFA content was similar in groups G2 and G3, but different in group G1. Significant difference among all fish groups was noted in two dominant FAs: 18:1 and DHA.

**Table 3.** Fatty acid composition (g per 100 g lipid) in Rainbow trout groups G1, G2 and G3 with different lipid content

Fatty acids	G1	G2	G3
C14:0	4.3 ± 0.5 <sup>b</sup>	4.8 ± 0.4 <sup>a</sup>	4.2 ± 0.1 <sup>b</sup>
C16:0	16.7 ± 1.0 <sup>a</sup>	16.0 ± 1.1 <sup>a</sup>	13.4 ± 0.5 <sup>b</sup>
C18:0	3.1 ± 0.3 <sup>a</sup>	3.0 ± 0.6 <sup>a</sup>	2.9 ± 0.1 <sup>a</sup>
C16:1	5.2 ± 0.4 <sup>a</sup>	5.4 ± 0.3 <sup>a</sup>	5.1 ± 0.3 <sup>a</sup>
C18:1	24.7 ± 3.5 <sup>c</sup>	27.6 ± 2.2 <sup>b</sup>	32.7 ± 0.3 <sup>a</sup>
C20:1	2.7 ± 0.5 <sup>b</sup>	3.7 ± 1.1 <sup>a</sup>	3.3 ± 0.0 <sup>a, b</sup>
C18:2n–6	8.7 ± 1.6 <sup>b</sup>	9.3 ± 0.5 <sup>b</sup>	11.8 ± 0.2 <sup>a</sup>
C18:3n–3	3.1 ± 0.5 <sup>b</sup>	3.4 ± 0.6 <sup>b</sup>	4.4 ± 0.0 <sup>a</sup>
C20:5n–3 (EPA)	6.4 ± 0.6 <sup>a</sup>	5.3 ± 0.8 <sup>b</sup>	4.9 ± 0.1 <sup>b</sup>
C22:6n–3 (DHA)	22.5 ± 3.7 <sup>a</sup>	18.8 ± 1.6 <sup>b</sup>	14.3 ± 0.6 <sup>c</sup>
SFA	24.2 ± 1.4 <sup>a</sup>	24.1 ± 1.4 <sup>a</sup>	20.9 ± 0.6 <sup>b</sup>
MUFA	33.5 ± 2.6 <sup>c</sup>	37.2 ± 1.6 <sup>b</sup>	41.8 ± 0.4 <sup>a</sup>
PUFA	42.3 ± 1.7 <sup>a</sup>	38.7 ± 1.7 <sup>b</sup>	37.3 ± 0.6 <sup>b</sup>
n–3	32.1 ± 3.2 <sup>a</sup>	27.8 ± 1.8 <sup>b</sup>	23.9 ± 0.5 <sup>c</sup>
n–6	10.2 ± 1.7 <sup>b</sup>	10.9 ± 0.5 <sup>b</sup>	13.4 ± 0.2 <sup>a</sup>
n–3:n–6	3.3 ± 0.8 <sup>a</sup>	2.6 ± 0.2 <sup>b</sup>	1.8 ± 0.0 <sup>c</sup>

\*Values are mean ± SE. Mean values denoted with a, b, c are significantly different in Rainbow trout groups with different lipid composition ( $P = 0.05$ ).

## CONCLUSIONS

High lipid content and optimum FA composition is considered to be positive criteria for the nutritional value of Rainbow trout. The total lipid content in different Rainbow trout varied more than 5.5 fold, but FA proportions were very similar in all samples. DHA and EPA, as most important essential FAs, content in all analyzed Rainbow trout was sufficient and generally higher in Estonian farmed trout than in imported trout, and correlated well with results from previous research. However, it is important to note that Estonian farmed Rainbow trout had generally lower lipid content. Because of that the amounts of essential FAs in the same size portion of fish in weight were on average 1.6 fold smaller in Estonian farmed Rainbow trout.

Estonian Rainbow trout smolts come from the same hatcheries and are fed the same commercial feed as Rainbow trout farmed in other countries. The difference in Estonian Rainbow trout lipid content is mainly influenced by environment, particularly temperature: optimum is from 8 to 15°C, but in Estonia there are long periods where fish do not feed (cold winters and hot summers). Environmental influence needs to be compensated by proper feeding regimes and a longer growth period. In order to maintain and raise the quality and stability of Estonian farmed Rainbow trout it is vital for Estonian fish farmers to unify the level of lipid content of their fish.

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