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COMPARATIVE INDICATORS OF FATTY ACID COMPOSITION OF RAINBOW TROUT (*ONCORHYNCHUS MYKISS* WALBAUM, 1792) FROM VARIOUS AQUACULTURE FACILITIES IN MACEDONIA

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ABSTRACT

The objective of this study was to make a comparative analysis of the fatty acid composition of the rainbow trout (*Oncorhynchus mykiss* Walbaum, 1792) with consumption size of 250-300 g, from two different aquaculture facilities - coldwater fishponds (fishpond A and fishpond B) in Macedonia, at different ambient conditions in the aquatic environment, and with the use of pelleted food from different manufacturers.

Considering the results of the fatty acid composition of the rainbow trout from the fishpond A and the fishpond B, it can be concluded that the content of the saturated fatty acids (SFA) is higher in the fishpond B (20.303%) compared to the fishpond A (16.155%), while the content of the monounsaturated fatty acids (MUFA) and the polyunsaturated fatty acids (PUFA) is greater in the fishpond A (MUFA - 53.364%; PUFA - 30.435%) compared to the fishpond B (MUFA - 52.359%; PUFA - 27.268%).

Overall, by a comparative analysis of the qualitative properties of the rainbow trout from the both aquaculture facilities, the ambient conditions prevailing in the aquatic environment, and the used forage mixtures for feeding fish, it can be concluded that the both aquaculture facilities are producing fish with high nutritional quality in terms of the fatty acid composition.

Keywords: rainbow trout (*Oncorhynchus mykiss* Walbaum, 1792), fatty acid composition

INTRODUCTION

Freshwater and marine fish species are a good source of essential fatty acids (**Özoğul *et al.*, 2007**). Variations in lipid and fatty acid compositions between and within fish species depend on many factors such as food availability, season, environmental temperature, geographic location, sex, diet and age, physical and chemical properties of water, rearing conditions, physical activity and nutritional habits of the fish (**Gorgun and Akpinar 2007**). Therefore, the fatty acid composition of fish absolutely differs from feeding conditions. According **Jankowska *et al.* (2003)** fatty acid composition is the major factor that determines the quality and flavor aspects of fish meat.

Oncorhynchus mykiss (Walbaum, 1792), commonly known as rainbow trout, inhabits cold headwaters, creeks, rivers and lakes. It is widely used in aquaculture in many countries because of its rapid growth and high market value due to its flesh quality (**Sarma *et al.*, 2013**). The low fat content, the protein content of high biological value, and the content of a significant amount of essential fatty acids make the rainbow trout one of the most highly rated fish in the human diet.

Fish oils contain n-3 polyunsaturated fatty acid (PUFA), particularly, eicosapentaenoic acid (EPA; C20:5 n3) and docosahexaenoic acid (DHA; C22:6 n3) (**Aslan *et al.*, 2007**). The importance of the long-chain PUFA has gained attention due to the prevention of human coronary artery disease (**Mozaffarian *et al.*, 2005**), improvement of retina and brain development (**Crawford, 1993**), reduce the risk of heart attacks and also decrease incidence of breast cancer, rheumatoid arthritis, multiple sclerosis, psoriasis and inflammation (**Daviglus *et al.*, 1997**).

The objective of this study was to make a comparative analysis of the fatty acid composition of the rainbow trout (*Oncorhynchus mykiss* Walbaum, 1792) with consumption size of 250-300 g, from two different aquaculture facilities - coldwater fishponds (fishpond A and fishpond B) in Macedonia, at different ambient conditions in the aquatic environment, and with the use of pelleted food from different manufacturers.

MATERIAL AND METHODS

Examinations were performed on rainbow trout (*Oncorhynchus mykiss* Walbaum, 1792) with consumption size of 250-300 g from two aquaculture facilities - coldwater fishponds (fishpond A and fishpond B) in Macedonia.

The fishpond A is located in Demir Hisar region (Macedonia) and it is supplied with water from the river Crna, spring Zheleznec. In this fishpond pelleted food Aller silver (6 mm) from manufacturer Aller aqua (Poland) is used, with following content (per specification): fish meal, wheat, fish oil, soy, hemoglobin powder, hydrolyzed protein, protein concentrates from sunflower and oil rape. Besides the main components, the food contains the following components (per specification): phosphate (1%), calcium (0.8%), sodium (0.2%), vitamin A (10,000 ie/kg), vitamin D3 (1000 IE/kg), antioxidant E 324 ethoxyquin (100 mg/kg), trace elements E2 iodine (3 mg/kg), E4 copper (5 mg/kg), E5 manganese (12 mg/kg), E6 zinc (70 mg/kg), and pigments Astaxanthin E161j (50 mg/kg) and Canthaxanthin E161g (25 mg/kg).

The average mass of rainbow trout samples from fishpond A was 285 g, while the average length, 29 cm.

The fishpond B is located near the city of Bitola (Macedonia) and it is supplied with water from the reservoir Strezhevo. In this fishpond pelleted food Troco prime 18 (4.5 mm) from manufacturer Coppens International (Netherlands) is used, with following content (per specification): fish meal, wheat, fish oil, soy, wheat gluten, hemoglobin powder and oil rape. Besides the main components, the food contains the following components (per specification): phosphorus (0.96%), calcium (1.6%), sodium (0.3%), vitamin A (10.000 ie/kg), vitamin C (150 mg/kg), vitamin E (200 mg/kg), vitamin D3 (799 ie/kg), antioxidants E 324 ethoxyquin (100 mg/kg) and E321 butilied hidroksitulen (40 mg/kg), as well as trace elements E1 iron (75 mg/kg), E2 iodine (5 mg/kg), E4 copper (5 mg/kg), E5 manganese (20 mg/kg) and E6 zinc (80 mg/kg).

The average mass of rainbow trout samples from fishpond B was 267 g, while the average length, 27.2 cm.

During examinations, the following methods for determination of fatty acid composition in fish meat were used:

-Determination of fatty acid composition by gas chromatography - AOAC method 996.06.

Methods for physical – chemical properties of water in fish ponds that are used:
-pH determination - ISO 1052:1994;
-Chloride determination - ISO 9297:1989;
-Spectrophotometric determination of nitrates - HACH DR 400 procedure Method 8039;
-Spectrophotometric determination of iron - HACH DR 400 procedure Method 8365;
-Spectrophotometric determination of nitrites - HACH DR 400 procedure Method 8507;
-Turbidity determination of translucency - ISO 7027:1999;
-Spectrophotometric determination of ammonia - HACH DR 400 procedure Method 8038;
-Determination of chemical oxygen demand - Merck Method Spectroquant 1.18752.0001;
-Total nitrogen determination - Merck Method Spectroquant 1.14537.0001.

RESULTS AND DISCUSSION

Considering the results of the physical – chemical analysis of water in fishpond A and fishpond B, we`ve obtained the following results (Table 1):

Table 1 Physical – chemical properties of water in fishpond A and fishpond B

Parameters	**Fishpond A**	**Fishpond B**
Represent of oxygen - saturation	76 %	78 %
5-day biochemical consumption of O_2 at 20 °C	1,25 mg/l	1,30 mg/l
Chemical oxygen demand	4,95 mg/l	3,00 mg/l
Dry residue of filtered water	36,0 mg/l	39,0 mg/l
pH	8,3	7,15
Visible waste	No	No
Visible color	No	No
Noticeable odor	No	No
Fe	0,00 mg/l	0,030 mg/l
Nitrites	0,0015 mg/l	0,0960 mg/l
Nitrates	0,00 mg/l	0,00 mg/l
Ammonia	0,00 mg/l	0,160 mg/l
Turbidity	1,0 NTU	1,0 NTU
Chlorides	6,40 mg/l	6,80 mg/l
Total phosphorous	0,0048 mg/l	0,0070 mg/l
Total nitrogen	0,320 mg/l	0,300 mg/l

Based on the physical - chemical properties of water from these two aquaculture facilities, water is classified into class II (according to the Regulation on water classification Official Journal of RM 18/99), which has allowed for fish production.

Considering the results of the fatty acid composition of the rainbow trout (Oncorhynchus mykiss Walbaum, 1792) from fishpond A and fishpond B, we`ve obtained the following results (Table 2, Figire 1):

Table 1 Fatty acid composition of rainbow trout (*Oncorhynchus mykiss* Walbaum, 1792) from fishpond A and fishpond B

Lipid numbers	**Name**	**Type**	**Fishpond A (%)**	**Fishpond B (%)**
C12:0	Lauric acid	SFA	/	0.098
C14:0	Myristic acid	SFA	1.330	2.007
C15:0	Pentadecanoic acid	SFA	0.068	0.102
C16:0	Palmitic acid	SFA	10.897	14.335
C17:0	Heptadecanoic acid	SFA	0.097	0.118
C18:0	Stearic acid	SFA	2.993	3.389
C20:0	Arachidic acid	SFA	0.602	0.131
C21:0	Heneicosanoic acid	SFA	0.128	0.123
TOTAL SFA			**16.155**	**20.303**
C14:1	Myristoleic acid	MUFA	/	0.027
C16:1	Palmitoleic acid	MUFA	2.946	5.040
C17:1	Cis-10- Heptadecanoic acid	MUFA	0.189	0.275
C18:1 n9 c	Oleic acid	MUFA	47.005	43.317
C20:1	Cis-11- Eicosenoic acid	MUFA	2.081	2.657
C22:1 n9 c	Cis - Erucid acid	MUFA	0.937	0.694
C24:1	Nervonic acis	MUFA	0.206	0.349
TOTAL MUFA			**53.364**	**52.359**
C18:2 n6 t	Linoleic acid	PUFA	0.051	0.100
C18:2 n6 c	Linoleic acid	PUFA	19.075	15.962
C18:3 n6	γ- linolenic acid	PUFA	5.061	3.520
C20:2 n6	Eicosadienoic acid	PUFA	0.261	0.312
C20:3 n6	Eicosatrienoic acid	PUFA	0.231	0.286
C18:3 n3	α- linolenic acid	PUFA	0.919	0.596
C20:3 n3	Eicosatrienoic acid	PUFA	0.305	0.301
C20:5 n3	Eicosapentaenoic acid	PUFA	1.083	1.655
C22:6 n3	Docosahexaenoic acid	PUFA	3.449	4.536
TOTAL PUFA			**30.435**	**27.268**
Total UFA			83.799	79.627
Total PUFA n-6			24.679	20.180
Total PUFA n-3			5.756	7.088
n-3/n-6			0.233	0.351
UFA/SFA			5.187	3.921
PUFA/SFA			1.883	1.343
PUFA/MUFA			0.570	0.520

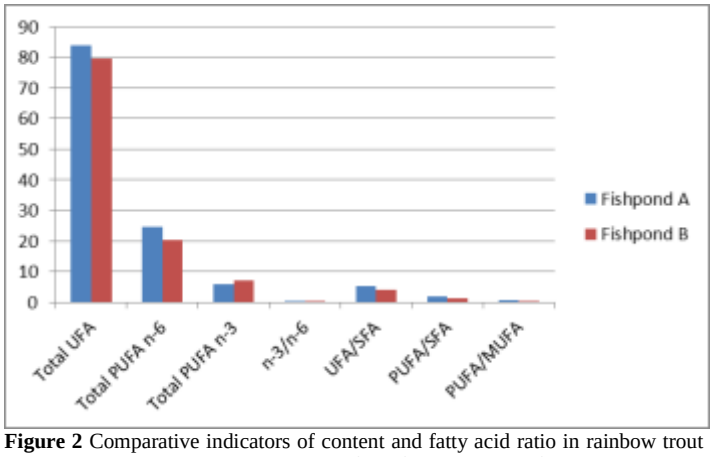

Figure 2 Comparative indicators of content and fatty acid ratio in rainbow trout (*Oncorhynchus mykiss* Walbaum, 1792) from fishpond A and fishpond B
Legend: UFA – unsaturated fatty acid, PUFA – polyunsaturated fatty acid, SFA – saturated fatty acid, n 3 – omega 3 fatty acid, n 6 – omega 6 fatty acid

Considering the results of the fatty acid composition of the rainbow trout from the fishpond A, it can be concluded that in total fatty acid content, saturated fatty acid (SFA) participates with 16.155 %. Of those, the most dominant are palmitic (10.897 %), stearic (2.993 %) and myristic (1.330 %) fatty acid. Monounsaturated fatty acids (MUFA) have the greatest participation in this fishpond, with 53.364 %. From those, the most dominant is oleic (47.005 %), followed by palmitoleic acid (2.946 %). Polyunsaturated fatty acids (PUFA) participate with 30.435 % in total fatty acid content, from which, the most dominant are linoleic (19.075 %), γ- linolenic (5.061 %), docosahexaenoic DHA (3.443 %) and eicosapentaenoic EPA (1.083 %) fatty acid. N-6 (omega 6) fatty acids participate with 24.679 %, while n-3 (omega 3) with 5.756 %, so the n-3/n-6 ratio is 0.233. UFA/SFA ratio is 5.187, PUFA/SFA is 1.883, while PUFA/MUFA is 0.570.
Considering the results of the fatty acid composition of the rainbow trout from the fishpond B, it can be concluded that in total fatty acid content, saturated fatty acid (SFA) participates with 20.303 %. Of those, the most dominant are palmitic (14.335 %), stearic (3.389 %) and myristic (2.007 %) fatty acid. Monounsaturated fatty acids (MUFA) have the greatest participation in this fishpond, with 52.359 %. From those, the most dominant is oleic (43.317 %) and palmitoleic acid (5.040 %). Polyunsaturated fatty acids (PUFA) participate with 27.268 % in total fatty acid content. From this type of acid, linoleic acid participates with the greatest percent (15.962 %), followed by γ- linolenic (3.520 %), docosahexaenoic DHA (4.536 %) and eicosapentaenoic EPA (1.655 %) fatty acid. N-6 fatty acids participate with 20.180 %, while n-3 with 7.088 %, so the n-3/n-6 ratio is 0.351. UFA/SFA ratio is 3.921, PUFA/SFA is 1.343, while PUFA/MUFA is 0.520.
Regarding the amount of SFA, the results of our research are in correlation with the findings of **Mustafa and Dikel (2015)** (20.74%), but slightly lower compared to those of ***Blanchet et al.*** *(2005)* (26.9%), ***Celik et al.*** (2008) (27.65%), **Danabas (2011)** (26.81 - 27.93%), **Vranić *et al.* (2012)** (29.14%), **Çagiltay *et al.* (2015)** (22.37 - 24.6%) and **Tkaczewska *et al*. (2015)** (19.82 - 27.90%).
The content of MUFA in our research closely correlated with the findings of **Çagiltay *et al.* (2015)** (42.55 - 43.68%), but higher than the results indicated by **Blanchet *et al.* (2005)** (32.5%), **Halilоglu *et al.* (2002)** (30.81%), **Mustafa and Dikel (2015)** (26.57%), **Danabas (2011)** (25.3 - 31.4%) and **Vranić *et al.* (2012)** (33.05%).
The results of PUFA`s amount in our research are correlated with the data presented by **Çagiltay *et al.* (2015)** (30.86 - 32.17%), higher than the results of **Celik *et al.* (2008)** (9.23%), and lower than those quoted by **Blanchet *et al.* (2005)** (40.6%) and **Mustafa and Dikel (2015)** (51.12%).
From this statement it can be concluded that the fatty acid profile is affected by extrinsic and intrinsic factors such as genetics, development phase, environmental condition and dietary lipids.
Nutritional value of rainbow trout fillets was measured by n-3/n-6 ratio. It has been suggested that this ratio is a good standard to compare the nutritional value of oils presented in meat fish. According **Osman *et al.* (2001)**, n-3/n-6 ratio of 1:1 or 1:1.5 can contribute to a healthy diet in humans. In our study, the value of the n-3/n-6 ratio for the fishpond A was 0.233, while for the fishpond B was 0.351.
According **H.M.S.O. (1994)** ideal n-6/n-3 ratio of fatty acids is up to 4. Values greater than the maximum value are harmful to health and may promote cardiovascular diseases (**Moreira *et al.*, 2001**). In our research n-6/n-3 ratio was ranged from 4.28 for fishpond A to 2.84 for fishpond B, from which it can be concluded that favorable is those from fishpond B.
According **H.M.S.O. (1994)** a minimum value of PUFA/SFA ratio is recommended as 0.45, which is in correlation with our findings of 1.88 for fishpond A and 1.34 for fishpond B, indicating the meat of good nutritional quality because the obtained values are higher than the minimum (0.45) which is recommended for a healthy diet in humans.
According **AFSSA (2003)**, the UFA/SFA ratio of fatty acids in fish fat is very important and its value should be over 3. This statement is in correlation with our UFA/SFA values which for fishpond A was 5.187 and for fishpond B was 3.921.

CONCLUSION

Considering the results of the fatty acid composition of the rainbow trout from the fishpond A and the fishpond B, it can be concluded that the content of the saturated fatty acids (SFA) is higher in the fishpond B (20.303%) compared to the fishpond A (16.155%), while the content of the monounsaturated fatty acids (MUFA) and the polyunsaturated fatty acids (PUFA) is greater in the fishpond A (MUFA - 53.364%; PUFA - 30.435%) compared to the fishpond B (MUFA - 52.359%; PUFA - 27.268%).
In terms of the amount of n-6 fatty acids, it is higher for the fishpond A (24.679%) compared to the fishpond B (20.180%), while the amount of n-3 is greater for the fishpond B (7.088%) compared to the fishpond A (5.756%).
The resulting value of the relations UFA/SFA, PUFA/SFA and PUFA/MUFA is increased with the fishpond A (5.187, 1.833 and 0.570, respectively) compared to the fishpond B (3.921, 1.343 and 0.520, respectively), while the value of the ratio n-3/n-6 is greater for the fishpond B (0.351) in comparison with the fishpond A (0.233).
In the rainbow trout from the fishpond A, the content of the eicosapentaenoic (EPA) fatty acid is 1.083%, while the one of the docosahexaenoic (DHA) fatty acid is 3.449%. In the rainbow trout from the fishpond B, the content of the EPA is 1.655%, while the one of DHA is 4.536%, which is higher in comparison with the fishpond A.
The variations in the fatty acid composition of the fish meat depend on many factors, but primarily on the season, the ambient temperature, the geographical location, the gender, the diet, the age of the fish, the physical and the chemical characteristics of the water, the rearing conditions, the physical activity, etc.
Overall, by a comparative analysis of the qualitative properties of the rainbow trout from the both aquaculture facilities, the ambient conditions prevailing in the aquatic environment, and the used forage mixtures for feeding fish, it can be concluded that the both aquaculture facilities are producing fish with high nutritional quality, in terms of the fatty acid composition.

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