

QUALITATIVE PROPERTIES OF RAINBOW TROUT (*ONCORHYNCHUS MYKISS* WALBAUM, 1792) FROM AQUACULTURE FACILITY IN BITOLA REGION (MACEDONIA)

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Abstract: The main goal of this research was to determine the qualitative properties of the rainbow trout (*Oncorhynchus mykiss* Walbaum, 1792) from aquaculture facility – salmonid fishpond Strezhevo which is situated in Bitola region (Republic of Macedonia). The qualitative properties of the rainbow trout are established by determination of the chemical and the fatty acid composition of the fish meat, the energy value of the meat and the microbiological analysis for the total number of microorganisms on fish skin and presence of *Salmonella* sp. and *Listeria monocytogenes*. The main purpose of the research produced additional analyzes that determine the physical - chemical properties and also a microbiological analysis of the water in which the rainbow trout resides, the chemical composition of feed used for feeding of the rainbow trout, the condition factor (CF) and the feed conversion. The results obtained during the examination of the chemical composition of the rainbow trout meat from the fishpond Strezhevo determined the mean value of 74.533% water, 20.600% protein, 3.366% fat and 1.38% ash. The energy value of the meat was 484.635 kJ/100 g. Considering the results of the fatty acid composition of the rainbow trout from the fishpond Strezhevo, it can be concluded that the content of the saturated fatty acids (SFA) is 20.303%, the monounsaturated fatty acids (MUFA) is 52.359% and the polyunsaturated fatty acids (PUFA) is 27.268%. In terms of the amount of n-6 fatty acids, it is 20.180%, while the amount of n-3 is 7.088%.

Keywords: rainbow trout, chemical composition, fatty acid composition

Introduction

Rainbow trout (*Oncorhynchus mykiss* Walbaum, 1792) is one of the most consumed fish species in Macedonian kitchen. It is grown in many aquaculture facilities – salmonid fishponds in our country, due to its fast growth and

exceptional nutritive quality. According *Simonović (2001)* this fish species is farmed intensively for consumption and it is tolerant to environmental conditions.

The main goal of this research was to determine the qualitative properties of the rainbow trout (*Oncorhynchus mykiss* Walbaum, 1792) from aquaculture facility – salmonid fishpond Strezhevo which is situated near city of Bitola (Republic of Macedonia).

The qualitative properties of the rainbow trout are established by determination of the chemical and the fatty acid composition of the fish meat, the energy value of the meat and the microbiological analysis for the total number of microorganisms on fish skin and presence of *Salmonella* sp. and *Listeria monocytogenes*. The main purpose of the research produced additional analyzes that determine the physical - chemical properties and also a microbiological analysis of the water in which the rainbow trout resides, the chemical composition of feed used for feeding of the rainbow trout, the condition factor (CF) and the feed conversion.

Material and Methods

Examinations were performed on rainbow trout (*Oncorhynchus mykiss* Walbaum, 1792) with consumption size of 250-300 g from aquaculture facility – salmonid fishpond Strezhevo. During these examinations, 18 samples from rainbow trout were analyzed. The average mass of rainbow trout samples was 267 g, while the average length, 27.2 cm.

The fishpond Strezhevo is located near the city of Bitola (Macedonia) and it is supplied with water from the accumulation Strezhevo. During these examinations, three samples of fishpond water were analyzed for each microbiological and physical-chemical property, in the same time when the fish catching was organized, accordingly. The results below (Table 1) represent average value from these three repetitions.

In this fishpond pelleted feed 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 feed contain 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 butilat hidroxitoluen (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).

Based on absolute indicators of mass and body length of the fish, the condition factor (CF) is calculated according to formula $CF = (BW / L^3) \times 100$ (Ricker, 1975), where BW is fish body weight and L is total fish length.

The feed conversion ratio (FCR) is calculated according to the formula $FCR = F/G$, where F is consumed feed (g) and G is fish growth (kg).

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

- Determination of moisture content - ISO 712:2009;
- Spectrophotometric determination of total nitrogen according Kjeldahl - HACH DR 400 procedure method 2410;
- Determination of total fat by gravimetric method (Soxhlet extraction) - AOAC method 2003.6
- Determination of ash in an oven at 700°C - ISO 3593:1981;
- Determination of fatty acid composition by gas chromatography - AOAC method 996.06.

Methods for physical – chemical properties of water in fishpond that were 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.

Methods for microbiological analyses of fish meat that were used:

- Horizontal method for detection and enumeration of *Listeria monocytogenes* - ISO 11290 - 1:2008;
- Horizontal method for detection and enumeration of *Salmonella* sp. - ISO 6579 - 2008;
- Horizontal method for the enumeration of microorganisms - ISO 4833:2003.

Methods for microbiological analyses of water that were used:

- Detection and enumeration of coliform bacteria and *Escherichia coli* - ISO 9308 - 1:2000;

- Detection and enumeration of intestinal enterococci and *Streptococcus faecalis* - ISO 7899 - 2:2000

Energy value (EV) of rainbow trout meat was calculated according to formula: $E.V. (\text{kJ}/100\text{g}) = \text{proteins} (\%) \times 17.16 + \text{fats} (\%) \times 38.96$ (Vitčenko et al., 1981).

For data processing, standard statistical methods (Microsoft Office Excel 2010, Data Analysis ToolPak) were used.

Results and Discussion

Considering the results of the physical – chemical and microbiological analysis of water in fishpond Strezhevo, we've obtained the following results:

Table 1. Physical – chemical analysis of water in fishpond Strezhevo (Bitola, Macedonia)

Parameters	Amount
Represent of oxygen - saturation	78 %
5-day biochemical consumption of O ₂ at 20 °C	1.30 mg/l
Chemical oxygen demand	3.00 mg/l
Dry residue of filtered water	39.0 mg/l
pH	7.15
Visible waste	No
Visible color	No
Noticeable odor	No
Fe	0.030 mg/l
Nitrites	0.0960 mg/l
Nitrates	0.00 mg/l
Ammonia	0.160 mg/l
Turbidity	1.0 NTU
Chlorides	6.80 mg/l
Total phosphorous	0.0070 mg/l
Total nitrogen	0.300 mg/l

Table 2. Microbiological analysis of water in fishpond Strezhevo (Bitola, Macedonia)

Parameters	Amount
The probable number of thermo-tolerant coliform bacteria in 100 ml	30
Streptococcus of faecal origin in 100 ml	0

Based on the physical - chemical and microbiological analysis of water from fishpond Strezhevo (Bitola, Macedonia), water is classified into class II (according to the Regulation on water classification Official Journal of RM 18/99), which is allowed for fish production.

Considering the results of the chemical composition of pelleted feed used in this fishpond, we've obtained the following results:

Table 3. Chemical composition of feed (Coppens International, Netherland) used in fishpond Strezhevo (Bitola, Macedonia)

Components	Amount (%)
Proteins	42
Fats	18
Carbohydrates	16
Ash	6.2

In this fishpond pelleted feed Troco prime 18 (4.5 mm) from the manufacturer Coppens International (Netherland) is used. By analyzing the chemical composition of the feed we received the following results: 42% proteins, 18% fats, 16% carbohydrates and 6.2% ash.

In practice, in conditions of intensive farmed fish production, it is found that the application of high quality feed is one of the most important factors affecting the fish growth, the feed conversion ratio and chemical composition of the fish meat. Today, complete forage mixtures in pelleted form are used for rainbow trout feeding.

In our examinations for the condition factor (CF) of rainbow trout, we established a value of 1.3267, which is in close correlation with the findings of *Çagiltay et al. (2015)* which in their research obtained the values of 1.24, 1.29 and 1.22, respectively. *Kiessling et al. (1991)* emphasize that under the influence of stable growing conditions, the fish growth is directly related to the feed utilization and fish age. Changes in the feed amount have impact on this ratio, so the increased amount of feed leads to greater deposition of fat and vice versa, reduced feed intake can requirements increased fat content in the fish tissues, compared to fish with the same size due to poor feed conversion ratio.

In our examinations we established a value of 1.18 for feed conversion ratio. From an economic point of view, it is very important to determine the most way of diet (continuous, intensive at the beginning or intensive feeding during the late phase of growth) that primarily reflects the chemical and fatty acid composition of fish meat.

Considering the results of the chemical composition of rainbow trout meat from fishpond Strezhevo, we've obtained the following results:

Table 4. Chemical composition of rainbow trout meat from fishpond Strezhevo (Bitola, Macedonia)

Chemical parameters	$\bar{x} \pm SD$	min	max
Water	74.533 \pm 0.573	73.80	75.20
Proteins	20.600 \pm 0.571	20.10	21.40
Fats	3.366 \pm 0.880	2.60	4.60
Ash	1.380 \pm 0.120	1.22	1.51

Legend: \bar{x} - mean value; SD – standard deviation; min – minimum value; max – maximum value

The nutritional value of fish meat is determined by the amount of proteins, fats, minerals and vitamins, and it depends on the fish species, age, cultivation methods, the composition of the consumed feed, as well as the season.

The results obtained during the examination of the chemical composition of rainbow trout meat from the fishpond Strezhevo show the mean value of 74.533% water, 20.600% protein, 3.366% fat and 1.38% ash. The energy value of the meat was 484.635 kJ/100 g.

Variations in the chemical composition of fish meat are closely related to the proportion feed intake, so the percentage of proteins in muscle tissue increases slightly during the feed period, while the percentage of fat increases faster.

The protein content in salmonid fish is related to the fish size and it is controlled endogenous, while the fat content depends on many endogenous and exogenous factors. Protein content is stable during the growth period except in insufficient and unbalanced diet.

When the fat content in feed can cross most that fish can metabolize, fat will be deposited in muscle tissue. By increasing the fish weight and age, the metabolism is focused on increasing the percentage of dry matters, or water content reduction and fat accumulation in the fish muscle. Also, we conclude that the fat content in the rainbow trout meat is indirectly proportional to the water content.

According to the fat content, fish are classified in: lean fish (fat content of less than 5%), moderate fatty fish (5-10%) and fatty fish (more than 10% fat) (*Jabeen & Chaudhry, 2011*). According to the received amount of fat, rainbow trout from fishpond Strezhevo is being classified as lean fish (3.366% fat).

Bud et al. (2008) noticed that the total fat content in rainbow trout can range from 2.7 to 9%, depending on age, physiological condition, time of catch, individual differences, etc. The main factor of which the fat content in fish depends, is the content of fat in feed. According *Shimeno and Shikata (1993)*, the diet, dietary feed supplements, the amount of feed consumed as well as growth increase, generally influenced the increase of the fat content in fish meat. Other factors such as temperature, mobility and the addition of steroids, indirectly stimulate the diet and increase the fat content (*Viola et al., 1992*).

Considering the results of the fatty acid composition of the rainbow trout meat from fishpond Strezhevo, we've obtained the following results:

Table 5. The content of SFA (saturated fatty acid) in rainbow trout meat from fishpond Strezhevo (Bitola, Macedonia)

Lipid numbers	Name	Amount (%)
C12:0	Lauric acid	0.098
C14:0	Myristic acid	2.007
C15:0	Pentadecanoic acid	0.102
C16:0	Palmitic acid	14.335
C17:0	Heptadecanoic acid	0.118
C18:0	Stearic acid	3.389
C20:0	Arachidic acid	0.131
C21:0	Heneicosanoic acid	0.123
TOTAL SFA		20.303

Considering the results of the SFA (saturated fatty acids) composition of the rainbow trout from the fishpond Strezhevo, it can be concluded that from total fatty acid content, SFA participate with 20.303%. From those, the most dominant are palmitic (14.335%), stearic (3.389%) and myristic (2.007%) fatty acid.

Table 6. The content of MUFA (monounsaturated fatty acid) in rainbow trout meat from fishpond Strezhevo (Bitola, Macedonia)

Lipid numbers	Name	Amount (%)
C14:1	Myristoleic acid	0.027
C16:1	Palmitoleic acid	5.040
C17:1	Cis-10- Heptadecanoic acid	0.275
C18:1 n9 c	Oleic acid	43.317
C20:1	Cis-11- Eicosenoic acid	2.657
C22:1 n9 c	Cis - Erucid acid	0.694
C24:1	Nervonic acis	0.349
TOTAL MUFA		52.359

Monounsaturated fatty acids (MUFA) have the greatest participation in the fish meat from this fishpond, with 52.359%. From those, the most dominant is oleic (43.317%) and palmitoleic (5.040%) fatty acid.

Polyunsaturated fatty acids (PUFA) participate with 27.268% in total fatty acid content. From this type of acids, linoleic participate with the greatest percent (15.962%), followed by γ - linolenic (3.520%), docosahexaenoic DHA (4.536%) and eicosapentaenoic EPA (1.655%) fatty acid.

Table 7. The content of PUFA (polyunsaturated fatty acid) in rainbow trout meat from fishpond Strezhevo (Bitola, Macedonia)

Lipid numbers	Name	Amount (%)
C18:2 n6 t	Linoleic acid	0.100
C18:2 n6 c	Linoleic acid	15.962
C18:3 n6	γ - linolenic acid	3.520
C20:2 n6	Eicosadienoic acid	0.312
C20:3 n6	Eicosatrienoic acid	0.286
C18:3 n3	α - linolenic acid	0.596
C20:3 n3	Eicosatrienoic acid	0.301
C20:5 n3	Eicosapentaenoic acid	1.655
C22:6 n3	Docosahexaenoic acid	4.536
TOTAL PUFA		27.268

The fatty acid composition in different, but also within the same fish species shows some variations (*Valente et al., 2007; Robin and Skalli, 2007*), and a number of factors such as temperature, water quality, type and availability of feed, season, age, gender, reproductive status, geographical location and individual differences are considered as significant factors that contribute to the occurrence of these variations.

Table 8. A review of fatty acid content in rainbow trout meat from fishpond Strezhevo (Bitola, Macedonia)

Total SFA	20.303
Total MUFA	52.359
Total PUFA	27.268
Total UFA	79.627
Total PUFA n-6	20.180
Total PUFA n-3	7.088
n-3/n-6	0.351
n-6/n-3	2.84
UFA/SFA	3.921
PUFA/SFA	1.343
PUFA/MUFA	0.520

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.

The fat quality in fish is defined by the relationship between n-6/n-3 and PUFA/SFA ratio (*Ahlgren et al., 1996*).

According *HMSO (1994)* ideal ratio of n-6/n-3 fatty acids is up to 4. Values greater than the maximum value are harmful to health and can lead to cardiovascular diseases (*Moreira et al., 2001*). In our examinations, the ratio n-6/n-3 is 2.84, which is in correlation with provided recommendations.

According *HMSO (1994)*, the recommended minimum amount of PUFA/SFA ratio is 0.45 which is in correlation to our findings with value of 1.34.

The type and amount of fatty acids in fish muscle tissue is directly related to their diet, but also to other factors such as the fish size and age, reproductive status, season, geographical location, etc. which can influence the fatty acid profile of fish meat.

Considering the results of the microbiological analysis for the total number of microorganisms on fish skin and presence of *Salmonella* sp. and *Listeria monocytogenes*, we've obtained the following results:

Table 9. Microbiological analysis of rainbow trout from fishpond Strezhevo (Bitola, Macedonia)

Parameters	Total number of microorganisms (log cfu/cm ²)	<i>Salmonella</i> sp.	<i>Listeria monocytogenes</i>
\bar{x}	6.66	0	0

Legend: \bar{x} - mean value

In terms of determining the total number of microorganisms on the skin of rainbow trout, in our examinations we obtained average value of 6.66 log cfu/cm², which is in correlation with the findings of *Adams and Moss (2008)* which concluded that the total number of microorganisms on the fish skin surface ranged from 2.00 - 7.00 log cfu/cm².

Conclusions

- The results obtained during the examination of the chemical composition of the rainbow trout meat from the fishpond Strezhevo (Bitola, Macedonia) show the mean value of 74.533% water, 20.600% protein, 3.366% fat and 1.38% ash.
- Energy value of rainbow trout meat from fishpond Strezhevo (Bitola, Macedonia) is determined as 484.635 kJ/100 g.
- Considering the results of the fatty acid composition of the rainbow trout meat from the fishpond Strezhevo (Bitola, Macedonia), it can be concluded that saturated fatty acids (SFA) participate with 20.303%, monounsaturated fatty acids (MUFA) with 52.359%, while the polyunsaturated fatty acids (PUFA) with 27.268% from total fatty acid content.
- N-6 fatty acids participate with 20.180%, while n-3 with 7.088%, so the n-3/n-6 ratio is 0.351, while the n-6/n-3 ratio is 2.84.
- UFA/SFA ratio is 3.921, PUFA/SFA is 1.343, while PUFA/MUFA is 0.520.

- The type and amount of fatty acids in fish muscle tissue is directly related to their diet, but also to other factors such as the fish size and age, reproductive status, season, geographical location, etc. which can influence the fatty acid profile of fish meat.
- In conditions of intensive farmed fish production, proper diet with adequate amount of high quality feed is the most important parameter that affects the fish growth, the feed conversion, as well as, chemical and fatty acid composition of fish meat.

Kvalitativne osobine kalifornijske pastrmke (*Oncorhynchus Mykiss* Walbaum, 1792) iz ribnjaka u regionu Bitola (Makedonija)

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Rezime

Glavni cilj ovog istraživanja bio je da se utvrde kvalitativne osobine pastrmke (*Oncorhynchus mykiss* Walbaum, 1792) iz ribnjaka za salmonidne vrste riba Strezhevo koji se nalazi u regionu Bitolja (Republika Makedonija).

Kvalitativna svojstva kalifornijske pastrmke se utvrđuje određivanjem hemijskog sastava i sastava masnih kiselina ribljeg mesa, energetske vrednosti mesa i mikrobiološkom analizom za ukupan broj mikroorganizama na koži ribe i prisustvo *Salmonella* sp. i *Listeria monocytogenes*. Glavni cilj istraživanja je uključivao i dodatne analize koje određuju fizičko - hemijske osobine, kao i mikrobiološku analizu vode u kojoj pastrmka boravi, hemijski sastav hrane koja se koristi za ishranu kalifornijske pastrmke, faktor stanja/kondicije (CF) i konverziju hrane.

Rezultati dobijeni tokom ispitivanja hemijskog sastava mesa kalifornijske pastrmke iz ribnjaka Strezhevo pokazuju srednju vrednost 74,533% sadržaja vode, 20,600% proteina, 3,366% masti i 1,38% pepela. Energetska vrednost mesa je 484,635 kJ/100 gr.

S obzirom na rezultate sastava masnih kiselina pastrmke iz ribnjaka Strezhevo, može se zaključiti da je sadržaj zasićenih masnih kiselina (SFA) bio 20,303%, mononezasićenih masnih kiselina (MUFA) 52,359% i polinezasićenih masnih kiselina (PUFA) 27.268%. U pogledu količine n-6 masnih kiselina, dobijena je vrednost od 20.180%, dok je količina n-3 bila 7.088%.

Ključne reči: kalifornijska pastrmka, hemijski sastav, sastav masnih kiselina

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