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THE IMPACT OF DIFFERENT DIET AND ENVIRONMENTAL CONDITIONS ON CHEMICAL COMPOSITION OF RAINBOW TROUT (*ONCORHYNCHUS MYKISS WALBAUM*, 1792) FROM MACEDONIAN AQUACULTURE FACILITIES

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Abstract: *The main purpose of the research was to make an analysis of the influence of ambient conditions and nutrition on the chemical composition of rainbow trout (Oncorhynchus mykiss Walbaum, 1792) from various aquaculture facilities in Macedonia, in different ambient conditions in the aquatic environment, and with the use of pelleted food from different manufacturers. This research produced additional analyzes that determine the physico-chemical properties and also a microbiological analysis of the water in which the rainbow trout grows, the chemical composition of the food used for its feeding, as well as, the energy value of fish meat from these two aquaculture facilities (fishpond A and fishpond B).*
The results obtained during the examination of the chemical composition of the rainbow trout meat from the fishpond A determined the mean value of 71.700% water, 20.366% proteins, 5.633% fats and 1.21% ash, while the ones from the fishpond B determined the mean value of 74.533% water, 20.600% proteins, 3.366% fats and 1.38% ash. The energy value of the rainbow trout meat from the fishpond A was 568.941kJ/100 g, while the one from the fishpond B was 484.635 kJ/100 g.
Overall, by a comparative analysis of the chemical composition of the rainbow trout from both aquaculture facilities, as well as energy value of fish meat, the ambient conditions prevailing in the aquatic environment, and the forage mixtures used for feeding fish, it can be concluded that both aquaculture facilities are producing high nutritional quality fish.

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

1. Introduction

The rainbow trout (*Oncorhynchus mykiss* Walbaum, 1792) is one of the widely distributed species of fresh water in Europe [1]. According to [2] [3] it is one of the most appreciable fish in human nutrition thanks to its low fat content, proteins of high biological value and relatively low cholesterol content as well as valuable quantities of essential fatty acids. Researchers [4][5] considered that the most important parameter which influences fish growth, feed conversion and chemical composition of fish meat is proper nutrition by offering suitable quantities of high-quality feed, especially in intensive fish farming. [6] noticed that in spite of this constatation, salmonid fish species, where the rainbow trout belongs, are the most demanding ones, because nutrition may affect, not only the ratio of proteins, fat and water, but the nutritional value of meat as well, and the fatty acid composition. However, besides that, [7] [8] concluded that the fish nutritional quality might be influenced by genetic factors, pH, oxygen content, water quality and its temperature, type of feed and feeding technology.

The main purpose of the research was to make an analysis of the influence of ambient conditions and nutrition on the chemical composition of the rainbow trout (*Oncorhynchus mykiss* Walbaum, 1792) from various aquaculture facilities in Macedonia at different ambient conditions in the aquatic environment, and with the use of pelleted food from different manufacturers. 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 food used for feeding of the rainbow trout, as well as, the energy value of fish meat from these two aquaculture facilities (fishpond A and fishpond B).

2. Matherials and methods

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

The fishpond A is located in the Demir Hisar region (Macedonia) and it is supplied with water from the river Crna, the 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 concentrate 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 accumulation 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 of 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 the rainbow trout samples from fishpond B was 267 g, while the average length, 27.2 cm.

During examinations, the following methods for determination of the chemical composition of 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 the total fat by gravimetric method (Soxhlet extraction) - AOAC method 2003.6;
- Determination of ash in an oven at 700°C - ISO 3593:1981;

The Methods for physical – chemical properties of the water which are used in the fishponds are:

- 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 water that are 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.

3. Results and discussion

Considering the results of the physical – chemical and microbiological analysis of the water in fishpond A and fishpond B, we`ve obtained the following results (Tab. 1 and Tab. 2):

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

The water quality is a combination of chemical, physical and biological parameters affecting the growth and development of farmed fish. The success of the activities of the commercial aquaculture is dependent on the optimal environmental conditions for the accelerated fish growth and the development at the lowest cost of resources used. The water quality affects the general fish condition and determines the conditions for the growth and health of the farmed fish. As a result, water quality

is a major factor to be taken into account when planning aquaculture production.
Based on the physical - chemical and microbiological properties of the water from these two aquaculture facilities,the 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.

Table 2.

Microbiological properties of water in fishpond A and fishpond B

Parameters	Fishpond A	Fishpond B
The probable number of thermo-tolerant coliform bacteria in 100 ml	10	30
Streptococcus of faecal origin in 100 ml	0	0

Considering the results of the chemical analysis of pelleted food used in fishpond A (Aller aqua, Poland) and fishpond B (Coppens International, Netherlands), we`ve obtained the following results (Tab. 3):

Table 3.

Chemical composition of pelleted food used in fishpond A and fishpond B

Parameters	Percent (%)	
	Fishpond A **Aller Aqua (Poland)** **Aller silver (6 mm)**	**Fishpond B** **Coppens International (Netherlands)** **Troco prime 18 (4.5 mm)**
Proteins	41	42
Fats	24	18
Carbohydrates	18	16
Ash	7	6.2

By comparing the content of the basic chemical components in the food that is used for rainbow trout feeding, it can be established that the food is characterized by similar composition in both fishponds, with deviation only in the amount of fat which in the food from fishpond A (24%) is greater than in the one from fishpond B (18%).
Considering the results of the chemical composition and energy value of the rainbow trout meat from fishpond A and fishpond B, we`ve obtained the following results (Tab. 4):

Table 4.

Comparative indicators of the chemical composition (%) and energy value (κJ/100 g) of the rainbow trout meat from fishpond A and fishpond B

Parameters	Water	Proteins	Fats	Ash	Energy value
Fishpond A	**71.700**^{a} ± 1.003	**20.366** ± 1.268	**5.633**^{b} ± 0.555	**1.210**^{c} ± 0.157	**568.941**^{d}
Fishpond B	**74.533**^{a} ± 0.573	**20.600** ± 0.571	**3.366**^{b} ± 0.880	**1.380**^{c} ± 0.120	**484.635**^{d}

*The differences in the values with the same superscripts are statistically significant on level p<0,001 , the result are $\bar{X}$ **± SD (**mean value **± standard deviation)**

The results obtained during the examination of the chemical composition of the rainbow trout meat from the fishpond A show the mean value of 71.700 % water, 20.366 % proteins, 5.633 % fats and 1.21% ash. On the other hand, the results of the rainbow trout meat from fishpond B are as following: 74.533 % water, 20.600 % proteins, 3.366 % fats and 1.380 % ash. In our tests, the differences in the values of certain chemical parameters and the energy value of rainbow trout meat from fishpond A and fishpond B are statistically significant on level ($p<0.001$) (Tab. 4). The rainbow trout meat from the fishpond A contains significantly less water (71.700%) compared with the one from fishpond B (74.533%). The differences in proteins content in the rainbow trout from fishpond A (20.366%) and fishpond B (20.600%) is quite minimal, and were not statistically significant. Fat content in the rainbow trout meat from the fishpond A (5.633%) is greater compared with the one from fishpond B (3.366%). The average amount of ash is slightly higher in rainbow trout from fishpond B (1.380%) compared with the one in fishpond A (1.210%). Higher energy value is established in the rainbow trout meat from fishpond A (568.941 kJ/100 gr) compared to the meat from fishpond B (484.635 kJ/100 gr).

Our results obtained during the examination of the chemical composition of rainbow trout meat correlated with the findings of [9] (water 71.65%, proteins 19.6%, fats 4.43%, ash 1.36%), [10] (water 74.18%, proteins 20.33%, fats 4.1%, ash 1.22%), [11] (water 75%, proteins 20%, fats 3.8%, ash 1.2%), [12] (water 74.77%, proteins 20.78%, fats 3.18%, ash 1.27%) and [13] (water 73.03%, proteins 20.39%, fats 4.73%, 1.37% ash).

Proteins content determined in our results is higher compared with the results cited in [14] (17.13%), [15] (16.9%) and [16] (17.34 - 17.81%), and lower than the ones in [17] (22.96%). In terms of fat content, our results show higher values than the ones presented by [18] (2.17%) and [16] (1.28 - 2.28%), and lower, compared with the results in [19] (7.02-8.27%), [14] (9.07%)**,** [15] (10.1%) and [20] (11.6%).

The variations in the chemical composition of the fish meat depend on many factors, but primarily on the diet, the ambient temperature, the geographical location, the gender, the fish age, the physical - chemical parameters of the water, the rearing conditions, the physical activity, etc.

It is established that the fat content in fish meat is directly related to the nature and fat content in the food, while the water content of fish meat is inversely proportional to the fat content [21]. This statement correlates with our findings that the higher fat content (24%) in the food consumed by fish from the fishpond A leads to higher fat content in fish meat (5.633%), compared to fat content (18%) in the food used in fishpond B which results in lower fat content in fish meat (3.366%).

The results obtained for the energy value of fish meat indicate that the energy of the rainbow trout meat from fishpond A (568.941 kJ/100 g) is higher compared to the one from fishpond B (484.635 kJ/100 g). These results are directly related with the fat content in fish (5.633% fat in the fish meat from fishpond A and 3.366% fat in the fish meat from fishpond B).

Higher production results obtained in fishpond A compared with the ones from fishpond B arise as a result of the higher fat content and higher energy value of the relevant diet used in fishpond A. Therefore, the higher fat content in fish from the fishpond A compared with the one from the fishpond B stems does not depend only on the difference in the type of nutrients and their participation in the diet, but of the season, gender and water temperature.

Due to the presence of essential fatty acids in fish fat, the higher fat content can be assessed as an important parameter for the fish quality and should not be considered as a negative impact on nutrition.
Overall, by a comparative analysis of the chemical composition of the rainbow trout from the both aquaculture facilities, as well as energy value of fish meat, 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.

4. Conclusion

The results obtained during the examination of the chemical composition of the rainbow trout meat from the fishpond A determined the mean value of 71.700% water, 20.366% protein, 5.633% fat and 1.21% ash, while the ones from the fishpond B determined the mean value of 74.533% water, 20.600% protein, 3.366% fat and 1.38% ash in fish meat.
The energy value of the rainbow trout meat from the fishpond A was of 568.941kJ/100 g, while the one from the fishpond B was of 484.635 kJ/100 g.
The differences in the values of certain chemical parameters and the energy value of rainbow trout meat from fishpond A and fishpond B are statistically significant on level ($p<0.001$).
Overall, by a comparative analysis of the chemical composition of the rainbow trout from the both aquaculture facilities, as well as energy value of fish meat, the ambient conditions prevailing in the aquatic environment, and the forage mixtures used for feeding fish, it can be concluded that both aquaculture facilities are producing high nutritional quality fish.
Fish breeding in quality water with well balanced food and proper implementation of health care, will give better results compared with the ones in conditions of insufficient and poor quality food, and too dense plantation.

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