The Effects of Some Phytobiotics on Biochemical Composition of *Oreochromis Niloticus* Meat Reared in a Recirculating Aquaculture System

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Abstract

The purpose of this experiment is the evaluation of biochemical composition of Nile tilapia meat by administrating several phytobiotics in feed. *Oreochromis niloticus,* with an initial average weight of 125.41±34.33 g/exemplar, were reared, during six weeks in a recirculating aquaculture system. The phytobiotics from this experiment were administered in feed in a concentration of 1%/kg feed and consist in: thyme (Thymus vulgaris), fenugreek (Trigonela foenum graecum), neem (Azadirachta indica). Thus, the experimental variants were: V1-control, V2-thyme, V3-fenugreek and V4-neem. Fish were fed with SOPROFISH pelleted feed with 38% crude protein and 7% crude fat. During the experiment, the physico-chemical parameters of technological water were situated in normal range for optimal growth. At the end of the experiment, significant differences ($p<0.05$) were recorded between the experimental variants, regarding to moisture, protein content, fat content and dry matter; unsignificant differences were recorded regarding the percentage of ash ($p>0.05$; $p=0.68$). The highest value of moisture (87.25%) was registered in V2; the lowest value of fat content (0.05%) was recorded in V4 and the lowest protein content (10.79%) in V2. In conclusion, thyme, fenugreek and neem administration, in concentration of 1%/kg feed, influenced significantly the biochemical composition of *Oreochromis niloticus*.

Keywords: biochemical composition of meat, Oreochromis niloticus, phytobiotics, recirculating aquaculture system

1. Introduction

Globally, like many other animal products, fish represents a main source of food, with high nutritional value, used in human diet.

Sahu et al. (2000) reported that among the commercial characteristics of fish flesh quality is becoming more important to the aquaculture industry [1].

The nutritional value of fish meat consists in the content of moisture, dry matter, protein, lipids, vitamins and minerals, plus the caloric value of fish [2, 3].

Fish body composition appears to be largely influenced by feed composition. An increase in other parameters such as feeding rate and fish size also results in enhanced adipose deposition and decreases water content in the fish body [4]. Besides the food composition, fish body composition is influenced by age and growing conditions.

Nile tilapia is an important species in fresh water aquaculture. The success of Nile tilapia farming is mainly attributed to its ease of culture and desirable qualities as a food fish [5]. *Oreochromis niloticus* are principally herbivorous, although occasionally they can be omnivorous. Tilapia (including all species) is the second most important group of farmed fish after carps, and the most widely grown of any farmed fish [6].

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Phytobiotics represent a wide range of bioactive compounds that can be extracted from various plant sources. In recent years, some interesting and novel applications of phytobiotics in the animal production appeared [7].

The phytobiotics used, in this research, were: thyme, fenugreek and neem.

Thyme, a plant species native from Western Mediterranean area, is now widely cultivated as spice throughout temperate climate. The thyme plant contains a number of important compounds, such as the phenols: thymol (44–60%) and carvacrol (2.2–4.2%) [8]. The therapeutic properties of thyme, on fish, are: antiseptic, antioxidant, digestion stimulant etc. [9].

Fenugreek is a leguminous plant having maple-like flavor, traditionally used as spice in India. It is also added as aromatic condiment to different kinds of manufactured foods [10]. Fenugreek is used, among other things, for stimulating fish appetite [9].

In India, the neem is known as "Sacred Tree"/"Village Pharmacy" and has been used for over two millennia for their medicinal properties: neem products are believed to be antihelmintic, antifungal, antidiabetic, antibacterial, antiviral [11].

In recent years, there are many research about effects of several phytobiotics on immune response, disease resistance and growth performance.

The aim of this research is to investigate the influence of thyme, fenugreek and neem's on biochemical composition of *Oreochromis niloticus* meat, reared in an intensive recirculating aquaculture system conditions.

2. Materials and methods

The research was conducted in the pilot recirculating system and at the research laboratory of Aquaculture, Environmental Science and Cadastre from "Dunarea de Jos" University, Galati. The experiment was conducted during a six weeks.

The recirculating system design includes four rearing units, with a volume of $1m^3$ each, and a series of water quality conditioning units [12]. The conditioning units for water quality are designed to maintain in an optimum range the main physical-chemical parameters of the water: oxygen content, temperature, the pH and the concentration of ammonia nitrite.

A total number of 180 Nile tilapia, with an initial average weight of 125.41±34.33g, were randomly distributed in four rearing units. Fish were fed with SOPROFISH pelleted feed, with 38% crude protein. The feed biochemical composition is shown in Table 1.

Table 1. The biochemical composition of SOPROFISH 38/7 pelleted feed

Composition	Quantity
Protein %	38
Water %	10
Fat %	7
Ash %	10
Cellulose	4
Total Ca	1.6
Total P	1.2
Total Na	0.2
Vitamin A (IU/kg)	15000
Vitamin D (IU/kg)	2500
Vitamin E (mg/kg)	90
Vitamin C (mg/kg)	200
Lysine %	2.3
Methionine+Cysteine %	1.2
Ingredients: fish meal, soybean protein content, corn, wheat.	

During the experiment, the feed was supplemented with three phytobiotics (*Thymus vulgaris*, *Trigonela foenum graecum*, *Azadirachta indica*), in concentration to 1% phytobiotic/ kg feed. Thus, the experimental variants were organized as follows: V1 – control, V2 - 1% thyme/kg feed, V3 - 1% fenugreek/kg feed and V4 - 1% neem/kg feed.

Fish were fed four times per day with a daily ration of 3.4% from fish body weight.

The determination of biochemical composition from Nile tilapia meat was made at the end of the experiment, from fresh meat. When collecting the samples, the uniformity of exemplars, was taken into account, to eliminate the errors which consist in weight differences between exemplars.

The biochemical determination was performed on muscle tissue samples. The samples were then weighed and minced in a tissue grinder, to ensure homogeneous samples for the analysis.

The proximate composition of diets was carried out using the Association of Analytical Chemists methods [13].

For the analysis of *Oreochromis niloticus* meat biochemical composition was performed to

determine the body percentage of moisture, protein content, fat content, ash, dry matter.
Proteins were determined with Gerhardt type equipment by using Kjeldahl method, fats were determined by Soxhlet solvent extraction method (petroleum ether) with Raypa extraction equipment, dry matter was determined by heating at temperature of 105±2ºC using Sterilizer Esac and ash was evaluated by calcification at temperatures of 550±20°C, in a Nabertherm furnace.
The data were statistically analyzed, in Microsoft Excel, using descriptive statistics and ANOVA test.

3. Results and discussion

The meat biochemical composition of meat varies considerably, depending on several factors like: fish age, size, sex, environmental conditions (temperature, dissolved oxygen, pH, salinity, etc), feeding mode and the type of feed used [14].
During the experiment, the physico-chemical parameters of technological water were situated into normal range for optimal growth (O_2 – 8.54±0.91 mg/l; T – 26.22°C; nitrite – 0.16±0.01 mg/l; nitrate – 65.48±1.16 mg/l; ammonia – 0.16±0.03 mg/l).
The average individual weight, at the end of the experiment registered the following values: V1 – 251.00 g/fish, V2 – 268.33 g/fish, V3 – 253.89 g/fish and in V4 – 251.56 g/fish.
Fish body composition appears to be largely influenced by feed composition. An increase in other parameters, such as feeding rate and fish size, results also in a enhanced adipose deposition and a moisture content decrease of fish body [4].
The main biochemical parameters (moisture, protein content, fat content and ash) of Nile tilapia meat, reared in a recirculating industrial aquaculture system Figure 1.

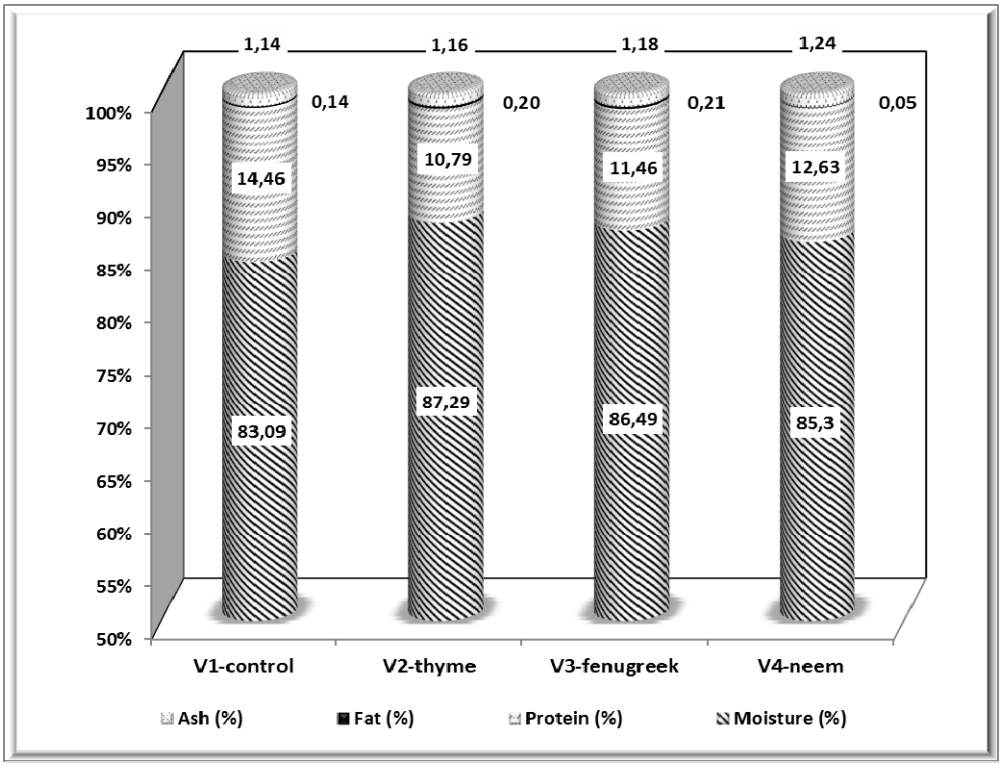

Figure 1. The biochemical composition of *Oreochromis niloticus* meat, fed with several phytobiotics

In the current experiment, the results of meat biochemical composition analysis, showed significant differences ($p<0.05$) in statistical terms, between V1 and others variants where phytobiotics were administrated, in terms of moisture level (p=0,000011), protein content (p=0,0000099) and fat content (p=0.0024).

In terms of ash content insignificant differences ($p>0.05$; $p=0.68$) were recorded between the experimental variants. The highest value of ash content was recorded in V4 variant – 1.24%. Olagunju et al., in 2012, affirm that an ash content between 1.17 and 1.79 % of fish, revealed that they are a good source of minerals such as calcium, potassium, zinc, iron and magnesium [15].

The highest value of moisture was registered in V2 variant (87.29%) and the lowest in V1 (83.09%). Abd-Alla, in 1994, found a percentage range between 80.50 and 84.00% for fish muscles moisture content, from various fish cultures [16].

The protein content from the tissue samples ranged from 10.79% to 14.46%. The lowest value of protein content was recorded in V2, followed by V3, V4 and V1 variant. This is, also, confirmed by one of the technological growth indicators, respectively retained protein (RP). The value of retained protein was the lowest in V2 compared with other experimental variants [17].

Hossain et. al., in 2002, reported that fish fed diet containing higher levels of Sesbbania seeds had lower body crude protein and lipid content, compared with the control diet [18].

Concerning fat content, the lowest value was registered in V4 (0.05%) and the highest was reported in V3 (0.21%). Ackman et al., in 1989, affirm that fish can be grouped into four categories, according to their fat content: lean fish (<2%), low fat (2 to 4%), medium fat (4 to 8%), and high fat (>8%) [19]. In case of Nile tilapia, some researchers have reported fat content values from 0.7 to 8.5% [20, 21], while De Castro in 2007 reported a value of 16.7% of the lipid content [22]. Ionescu et al., in 2006, said that the percentage of fat, in fish meat, varies between 0.2–25% [23].

Table 2. The biochemical composition of Nile tilapia meat

	Moisture %	**Protein %**	**Fat %**	**Ash %**
Gaber, 2000 [24]	78.9±0.5	16.6±0.5	2,75±0,16	2,6±0,2
Agbo, 2008 [25]	73.69 ± 0.41	15.18 ± 0.21	6.43 ± 0.14	4.16 ± 0.05
Kumar et al., 2011 [26]	73.6±2.0	17.3±0.5	5.6±1.2	3±1.1
Suleiman and James, 2011 [27]	74.08±3.08	21.05±0.64	7.32±0.17	2.98±0.36
Bag et al., 2012 [28]	75.50±1.21	13.36±0.20	4.60±0.05	5.10±0.06
Bozaoglu and Bilguven, 2012 [29]	80.06±0.11	13.62±0.21	2.47±0.27	2.06±0.12
Hernández-Sánchez and Aguilera-Morales, 2012 [14]	72-80	13-25	0.79-8.5	0.5-1.5
Mabroke et al., 2013 [30]	74.49±0.3	16.37±1.0	1.16±0.0	7.92±1.1

In Table 2 are presented the values of the main biochemical parameters, of Nile tilapia meat, reported by different authors.

El-Zaeem et al., in 2012, found a difference between the biochemical meat compositions of Nile tilapia, grown in the natural environment, against that of the Nile tilapia derived from aquaculture. They found an increase in moisture content and protein content in case of fishes derived from aquaculture [4].

In 2003, Richter et al. studied the effect of dried moringa leaf meal, as alternative protein source for Nile tilapia (*Oreochromis Niloticus*), on body chemical composition. They reported that increasing the levels of moringa leaves in diets increases the moisture body percentage, while lipid and gross energy values decreased significantly [31].

Sultana et al., in 2012 studied the influence of spirulina flakes, rice bran and mustard oil cake on juvenile Nile tilapia. They showed that protein content ranged between 13.32-10.19%, the highest value being registered at tilapia fed with spirulina in a concentration 2% [32].

In case of dry matter, significant differences ($p<0.05$, $p=0.025$) were obtained between the experimental variants. The highest value of dry matter was registered in V2 variant (1% thyme/kg feed) – 23.60±0.63%, followed in descending order by V3 – 22.25±0.13 %,V4 – 21.23±0.24% and V1 - 20.36±0.21%. Giving the fact that dry matter contains all the elements with nutritious value, the quality of meat is defined primarily by water/dry matter ratio [33].

4. Conclusions

The supplementation of the diet with phytobiotics significantly influenced the biochemical composition of *Oreochromis niloticus* meat:

- The highest value of moisture was registered in V2 variant – 87.25%, that also registered the highest individually average weight at the end of the experiment.
- The use of phytobiotics led to a decrease in crude protein content compared to the control variant.
- A significant decrease in fat content was recorded in V4. In V2 and V3, the percentage of fat content was higher compared with V1 variant. The percentage obtained reveals that tilapia is a species with a lean meat.
- Regarding ash content, it was observed an increase in variants in which phytobiotics were administered.
- The highest dry matter value was registered in V2 variant (1% thyme/kg feed) – 23.60±0.63% and the lowest in V1 - 20.36±0.21%.

In conclusion, thyme, fenugreek and neem administration, in concentration of 1%/kg feed, influenced significantly the biochemical composition of *Oreochromis niloticus*.

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