THE EFFECT ON BIOCHEMICAL COMPOSITION OF DIFFERENT CONCENTRATIONS PROBIOTIC FED TO JUVENILE CARP *(Cyprinus carpio, L. 1758)* IN A RECIRCULATING AQUACULTURE SYSTEM

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Abstract

Knowledge about biochemical composition of flash fish allow estimation about: general maintenance conditions, methods of availability to natural and supplementary trophy basis, nutritive value of flash and establishing the impact of environmental conditions to metabolic processes. In this study, some biochemical determinations of carp (Cyprinus carpio, L. 1758) tissues were investigated. The biochemical analysis of the meat has been effectuated in the experiment that took place during 60 days, in four breeding units of 500 liters in volume each. Four kind of variants were compared: V1-pellets with 30% crude protein, without probiotics; V2-pellets with 30% crude protein, with probiotics of 2.24×109 CFU/kg food; V3-pellets with 30% crude protein, with probiotics of 3.84×109 CFU/kg food; and V4-pellets with 30% crude protein, with probiotics of 7.04×109 CFU/kg food. We used BioPlus® 2B probiotics (a mixture of Bacillus licheniformis (DSM 5749) and Bacillus subtilis (DSM 5750). After analyses the biochemical composition in stage one was: V1-16.89% proteins, 1.57% lipids; V2-16.47% proteins, 2.5% lipids; V3-16.72% proteins, 1.87% lipids; V4-15.52% proteins, 1.69% lipids and in the second stage was: V1-14.47% proteins, 1.63% lipids; V2-15.70% proteins, 3.12% lipids; V3-14.45% proteins, 1.85% lipids; V4-15.24% proteins, 2.35% lipids. The obtained results showed that are insignificant differences (p>0.05) in the levels of proteins, fats and ash between the control variant (V1) and experimental variants (V2, V3, V4) where probiotic supplements were used.

Key words: carp, protein, fats, dry substance, RAS

INTRODUCTION

Some studies have attributed the enhancement of animal growth to the nutritional benefits of probiotic bacteria, such as vitamin production, availability of minerals and trace elements and production of important digestive enzymes [3].

Though, probiotics have been shown to be effective in a wide range of species for the promotion of growth, enhanced nutrition, immunity and survival rate. Some studies were conducted on the use of probiotics in the diets of the fresh water species [11], [12].

Probiotics in aquaculture have been shown to have several modes of action: competitive exclusion of pathenogenic bacteria through the production of inhibitory compounds; improvement of water quality; enhancement of immune response of host species and enhancement of nutrition of host species through the production of supplemental digestive enzymes [12].

Because *Bacillus* bacteria secrete many exoenzymes [7], these bacteria have been used widely as putative probiotics. Growth performance of common carp and nutritional quality has extensively been studied in recirculating system and survival and growth of common carp larvae are influenced by both stocking density and food type [5], [9], [10].

Bacillus species are capable of secreting enzymes such as protease and can result infaster digestion of the protein components in the diet and convert them to simpler

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peptides and amino acids by peptilytic and proteolysis enzymes [2].

The purpose of this experiment was to determine the biochemical composition of juvenile carp meat in conditions of feeding them with feed embedded with Probiotics *BioPlus®2B*, for 8 weeks.

MATERIAL AND METHOD

The research was conducted in the pilot recirculating system of Aquaculture, Environmental Science and Cadastre from "Dunarea de Jos" University, Galati. The recirculating system is represented by four rearing units with a volume of 0.500 L each and the and the conditioning unit of the water quality.

The biological material used for this experiment was represented by 320 juvenile carp with a initial average weight (±SEM) of 182 g/fish.

The experiment was conducted over two experimental stages, separated in time by periodic weighing, which were aimed to adjust the amount of feed distributed to fish due to accumulation of biomas and changes in water temperature. Weighing control was carried out after 30 days from the beginning of the experiment.

In this experiment, feeding intensity was 2% from body weight (BW), in the first stage, while in the second stage due to the lower water temperature, feeding intensity was 1.2% from BW. The daily amount of feed was distributed in three meals/ day, manually.

The fish were fed with *ALLER CLASSIC* pellets with 4 mm diameter and with an content of 41% crude protein and 7% lipids, in which has been incorporated a probiotic product *BioPlus*®2B (a mixture composed by *Bacillus licheniformis* (DSM 5749) and *Bacillus subtilis* (DSM 5750) in proportion of 1:1) in different concentrations:

- V1 (B1) –pellets without probiotic;
- V2 (B2) – with probiotic concentration of 2.24×10^9 CFU/kg feed;
- V3 (B3) – with probiotic concentration of 3.84×10^9 CFU/kg feed;
- V4 (B4) – with probiotic concentration of 7.04×10^9 CFU/kg feed.

Table 1 The biochemical composition of *ALLER CLASSIC*

Parametres	Measure unit	Quantity
Crude protein	%	30
Lipids	%	7
NFE	%	43
Ash	%	7
Fibre	%	5
Vitamin A (IE)	UI/kg	2500
Vitamin D3(IE)	UI/kg	500
Vitamin E	mg/kg	150
Gross energy	kcal/MJ	4325/18.1
Convertible energy	kcal/MJ	3353/14.0

To determine the biochemical composition of carp meat, biological material samples were taken at the end of each experimental stage. When the samples were collected we ensure about the uniformity of fish in order to eliminate errors due to differences in mass of the specimen.

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.

RESULTS AND DISCUSSIONS

The biochemical composition of fish meat is a good indicator to assess the general maintenance state of biological material, the food utilization and the impact of environmental conditions on fish body [4].

Growth performance of common carp and nutritional quality has extensively been studied in recirculating system and survival and growth of common carp larvae are influenced by both the stocking density and the type of food [5], [9], [10].

The biochemical composition of muscles tissue, in the first stage of the experiment, is presented in the figure below:

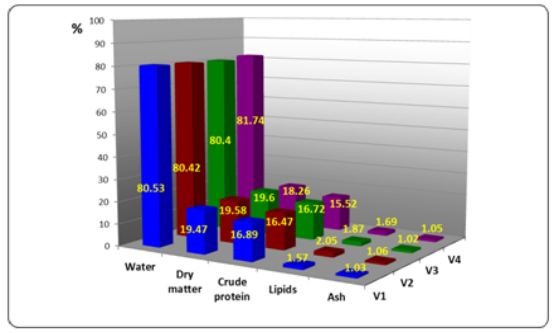

Figure 1 The biochemical composition of the muscular tissue in the first experimental stage

From the obtained data regarding protein content of muscle tissue, no significant differences ($p<0.05$) were found between control variant and the others experimental variants, where different concentrations of probiotic was used, fact that reveals no influence over carp meat biochemistry.

Regarding the fat content of carp muscle tissue, there were registered significant differences ($p<0.05$) between the experimental variants. Fat retention was significant for V2 version, in which case it reached to a percentage of 2.05%. However, the fat retention had a limited variation range, due to feed composition and environmental conditions. Optimal lipid content of feed should be between 5-12%.

It is also known that the fat content of fish muscle tissues is inversely proportion with the increase of dietary protein level, this being due to the low ratio of digestible energy to feed protein content.

The biochemical composition of the muscle tissue from the second experimental stage is shown in the figure below:

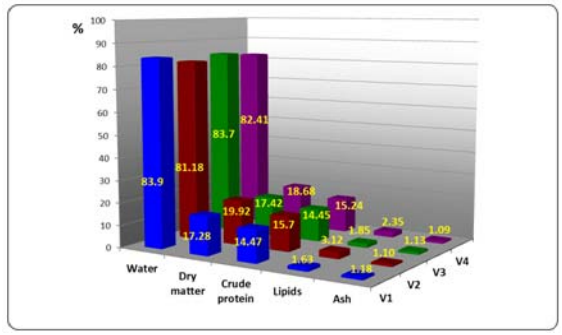

Figure 2 The biochemical composition of the muscle tissue in the second experimental stage

From the data obtained regarding the fat content of carp muscle tissue, it can be stated that significant differences ($p<0.05$) between the experimental variants were recorded: V2 fat content (3.12%), compared to the control variant (1.63%).

Comparing carp protein retention, it can be stated that differences between V1(14.47%) and V3 (14.45%) respectively, V2 (15.70%) and V4 (15.24 %) were not significant ($p<0.05$), fact which suggest that the accumulation of protein in fish meat is directly proportional with its weight gain, in all of the four experimental variants.

Regarding the biochemical composition of carp in the second experimental stage, it can be stated that protein, fat and ash content are slightly lower comparing with previous stage, differences being recorded also between control version and the others experimental variants. Also, the small variation seen between the control variant and the others experimental variants is explained by the fact that the probiotic which

was used, does not have a significant influence on biochemical composition of carp meat. Mocanu M. et al. (2012) support that adding probiotics in fish feed, in very high doses, does not improve biochemical composition of meat, but an appropriate dose of probiotic can induce an improvement in the nutritional qualities of meat and also those reported by other authors which showed that introduction of probiotic *BioPlus®2B* in different concentrations in rainbow trout feed, leads to an increase of protein content and a decreas of fats and water percentage from meat [6].

CONCLUSIONS

Fish protein contains all essential amino acids which are easy to digest. The protein digested and assimilated is mostly incorporated in the muscles of the fish [1] reported that protein content, which is a vital constituent of living cells, tends to vary relatively little in healthy fish unless drawn upon during particular demands of reproduction or during food deprivation periods. Similar to fat, the protein content in the body of fishes change depending on time of year, environmental condition, stage of maturity of the gonads, state of nutrition and age.

In conclusion, the biochemical composition of carp meat, reared in a recirculating system, under the influence of technological factors presented above, has lower values at the end of the experimental period, due to the decrease of environmental temperature and the non-corresponding assimilation of key nutrients, presented in administrated feed.

Regarding the experimental variants, not significant differences ($p>0.05$) were registered between control variant and other variants, where different concentration of probiotic was used, fact that reveals their lack of influence over the biochemical composition of carp meat, that was supposed as it appears due to the used of probiotic in feed, in tested concentrations.

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