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EFFECTS OF KOMBUCHA AND MILK KEFIR DIETARY SUPPLEMENTS ON THE MEAT BODY COMPOSITION OF SIBERIAN STURGEON (*Acipenser baerii*)

Cristian RIMNICEANU[1, 2], Mirela CRETU[1, 2], Iulia Rodica GRECU[1, 2], Marina PIHUROV[1], Gabriela BAHRIM[1], Lorena DEDIU[1, 2]

[1]"Dunărea de Jos" University of Galaţi, 47 Domnească Street, 800008, Galați, Romania
[2]Romanian Center for Modelling Recirculating Aquaculture Systems, "Dunărea de Jos" University of Galați, 800008, Galați, Romania

Corresponding author email: mirela.cretu@ugal.ro

Abstract

This paper aimed to evaluate the effect of a diet supplemented with fermented products on the meat quality of Siberian sturgeon (Acipenser baerii) reared in a recirculation aquaculture system. The fermented product was made by combining artisanal cultures of Kombucha and milk kefir granules grown up in sucrose, black tea, and bovine colostrum. Four experimental groups were established: V1 - a control group that received a commercial diet with 54% crude protein, and V2 to V4 groups, which received the same diet supplemented with 1 g/kg, 2 g/kg, and 3 g/kg of fermented product, respectively. After 35 days of diet administration, the biochemical composition of fresh meat was analysed. The results showed that the addition of fermented products significantly influenced the water, ash, and lipid content of A. baerii meat (p < 0.05), while the protein content was not influenced (p > 0.05) by the administrated diet. In conclusion, it is evident that adding the fermented product in A. baerii meat has an improving effect on body composition

***Key words**: bioactive fermented products, meat composition, sturgeon.*

INTRODUCTION

The overexploitation of sturgeons for caviar and meat, corroborated with the habitat deterioration, led to drastic declines in natural populations. The high demand for these products has driven the development of sturgeon aquaculture, which has become an important industry in many countries.

Siberian sturgeon (*Acipenser baerii*) is a species of fish that has been farmed for thousands of years for its valuable meat, caviar, and other by-products, showing a rapid growth rate (Pyka et al., 2003; Babaei et al., 2017; Williot et al., 2018), higher resistance to pathogens (Kayiş et al., 2016), and can be reared using a wide range of diets and environmental conditions (Bronzi et al., 2011; Xu et al., 2019).

The production of high-quality sturgeon meat requires a balanced and nutritious diet, which is a major challenge for producers. Over the past few years, there has been a huge interest in supplementing the sturgeon's diet with different feed additives, as this has been proven to exhibit a significant impact on the growth performance and biochemical composition of fish meat (Palmegiano et al., 2008; Sayed et al., 2020; Mocanu et al., 2022). Generally, feed additives typically contain a range of beneficial substances, such as probiotics, enzymes, minerals, and vitamins, that can have a positive effect on the growth and well-being of fish. The quality of sturgeon meat is of significant interest to both researchers and producers since it has a significant influence on the overall quality and value of the obtained product.

Kombucha, also known as SCOBY (Symbiotic Culture of Bacteria and Yeasts), is made up of several organic acids, sugars, vitamins, amino acids, biogenic amines, purines, pigments, lipids, proteins, hydrolytic enzymes, ethanol, caffeine, carbon dioxide, polyphenols, anions, minerals, and bacterial metabolites, such as D-saccharic acid-1, 4-lactone (DSL), according to Jayabalan et al. (2014).

Kefir grains consist of a symbiotic culture of bacteria, yeast, and acetic acid bacteria that adhere to a matrix of polysaccharides (Chen et al., 2015). The mixture of kombucha culture and milk kefir grains gives rise to various consortia of naturally occurring microorganisms, such as lactic acid, acetic acid, and yeast. Through their fermentative activity, these microorganisms produce a diverse array of bioactive compounds, including prebiotics, probiotics, postbiotics, and para-probiotics, which have been found to offer *in vitro* and *in vivo* benefits (Pihurov et al., 2021).

Studies conducted in the past have revealed that the inclusion of fermented products in the diet can enhance the sensory characteristics and extend the shelf life of the final product (Ndaw et al., 2008). Fermentation can also lead to the synthesis of metabolites that exhibit antitoxic properties, thereby reducing the bioaccumulation of heavy metals in water and fish tissues, and offering direct protection against oxidative stress induced by metals (Giri et al., 2018). Moreover, these products can serve as a viable alternative to antibiotics, while still promoting the growth and well-being of fish (Choi et al., 2022).

While there is a growing interest among sturgeon farmers to cultivate healthy fish and optimize feed efficiency, there is a lack of available information regarding the nutritional requirements and feeding practices specific to Siberian sturgeon. In this context, the objective of this study was to assess how the dietary inclusion of fermented products affects the body composition of Siberian sturgeon meat.

MATERIALS AND METHODS

Experimental design. The experiment took place at the University Dunărea de Jos, Galați, Romania, at the Romanian Centre for the Modelling of Recirculating Aquaculture Systems (www.moras.ugal.ro, accessed on February 9th, 2021). Four different experimental groups were established, including the control group (V1), which was fed with a commercial diet containing 54% crude proteins and 15% lipids; the V2 group, which received the same diet supplemented with 1 g/kg of the fermented product; the V3 group, which received 2 g/kg of the fermented product; and the V4 group, which received 3 g/kg of fermented product. The fermented product used in our study was obtained in the Microbiology Laboratory of the Faculty of Food Science and Engineering. The product was obtained by combining artisanal cultures of kombucha and milk kefir granules, which were grown in black tea, sucrose, and bovine colostrum.

Fish body composition analysis. After 35 days of diet administration, the proximate composition analyses of the fish body were determined by the standard methods of AOAC, 2003. The fish were eviscerated and filleted, and only the muscular tissue was blended and utilized for subsequent analysis.

Moisture content (%) was assessed by subjecting the samples to a constant temperature of 105 °C in a convection oven (Jeiotech, Jeio Tech Co., Inc, Korea) until a constant weight was achieved (AOAC, 2016). The dry samples were then finely ground and utilized further for the evaluation of protein, fat, and ash levels.

Crude protein content (%). The Dumas method was used to quantify the nitrogen content of the dry samples, which was then converted into crude protein content (%) using the common conversion factor of N×6.25. The combustion of samples was performed at 1100°C using a Primacs SNC 100 (Skalar Analytical B.V., The Netherlands).

The lipid content (%) was determined using the Soxhlet extraction method with petroleum ether as the solvent (C. Gerhardt GmbH & Co. KG, Germany), following the AOAC method from 1997.

The ash content (%) was determined by heating the sample in a muffle furnace (Nabertherm, Applied Scientific Instruments Co., Ltd. Thailand) at 525±25°C for 8 hours. All calculations for protein, fat, and ash content were reported based on the dry weight.

The fish experiments were conducted with the approval of the Ethics Committee of the University Dunărea de Jos from Galați.

Statistical analysis. The statistical analysis of the fish's proximate composition was conducted using SPSS software for Windows, version 21.0 (SPSS Inc., Chicago, United States). Results were considered statistically significant at $p<0.05$. The mean values and standard deviations (S.D) of each species were calculated and presented. Differences between the mean values were determined by One-factor analysis of variance (ANOVA), and Duncan's test was used to identify specific groups with significantly different means.

RESULTS AND DISCUSSIONS

The biochemical composition of fish is crucial for fish preservation. Typically, fish meat comprises 70-84% water, 15-24% protein, 0.1-22% fat, 1-2% minerals, and 0.1-1% carbohydrates. The protein content is particularly important since it contributes to the nutritional value and sensory characteristics of the final product. The fat content also plays a significant role in the taste, texture, and shelf life of fish products (Love, 1970; Desta et al., 2019; Nicolae, 2020; Ahmed et al., 2022). Body composition is a good indicator of the physiological condition of fish (Ali et al., 2005).

The proximate composition of fish can vary greatly between and within species and is influenced by various factors such as food composition, feeding rate and frequency, age, size, sex, genetics, and season/migration (Morris, 2001; Kestin et al.,2001; Mazumder et al., 2008; Begum et al., 2016).

The body composition analysis results of the experimental fish are illustrated in Figures 1-4. There was no significant difference found in the protein content ($p>0.05$) of *A. baerii* meat among the experimental variants. However, significant differences were observed in the water, ash, and lipid content of the meat ($p<0.05$).

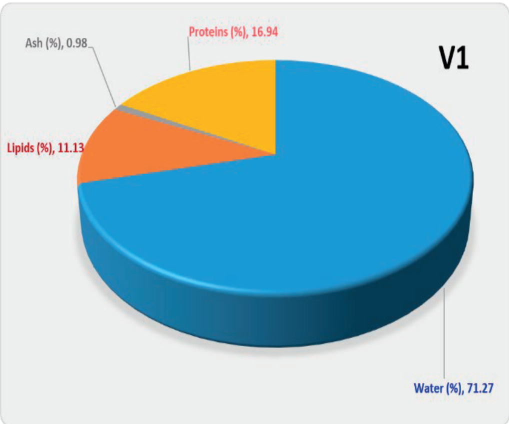

Figure 1. Proximate composition analysis of fish meat in V1 Variant

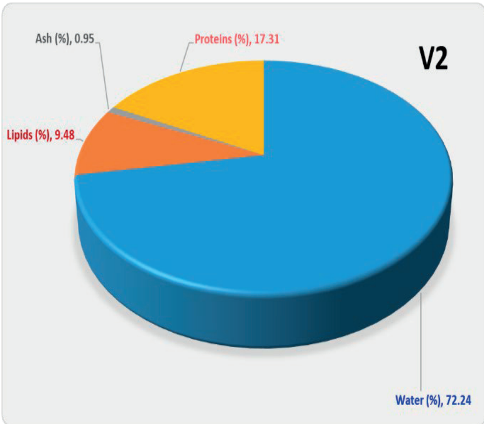

Figure 2. Proximate composition analysis of fish meat in V2 Variant

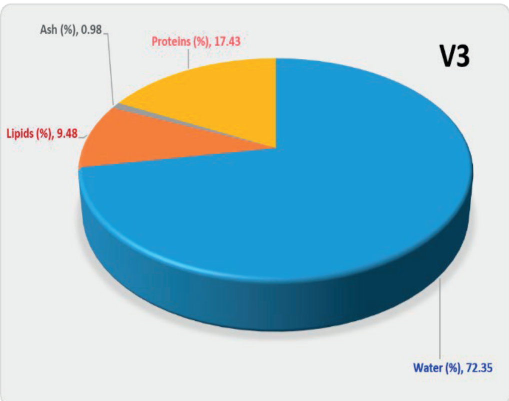

Figure 3. Proximate composition analysis of fish meat in V3 variant

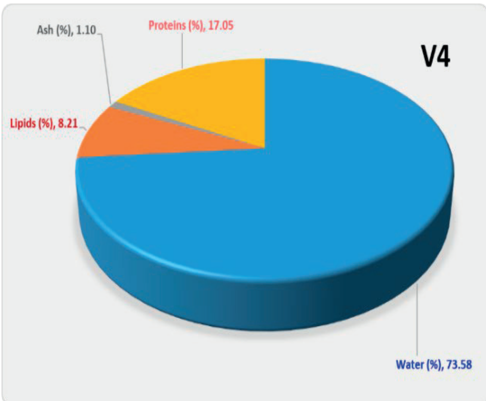

Figure 4. Proximate composition analysis of fish meat in V4 Variant

The inclusion of different levels of the fermented product in *A. baerii* food leads to a significant ($p<0.05$) reduction of the lipid content. The values of the lipids content showed a significantly higher value in the control variant, (11.13±1.29%), with no significant differences between fish fed with feed fermented product at 1 g/kg feed (9.48±0.96%), and 2 g/kg (9.48±0.31%). A significant ($p<0.05$) lower lipid content (8.21±0.87%) was registered in the variant V4 (3 g/kg fermented product). In conclusion, the inclusion of probiotics in the diets of fish resulted in lower levels of crude lipids compared to the control group. This indicates that the fish had a higher protein-to-fat ratio, which is highly desirable in aquaculture practices.

Moisture content and lipid content were found to have an inverse relationship. The water content showed significantly lower ($p<0.05$) moisture contents in the V1 group (71.27±1.03%), followed by fish fed with 1 g/kg fermented product (72.24±1.146%), and 2 g/ kg respectively (72.35±1.44%), with no differences ($p>0.05$), and by the V4 variant, with the highest water content (73.58±1.44%).

The results obtained by us were similar to those reported by Ayoola et al. (2013), for African catfish *Clarias gariepinus,* who registered after 90 days of feeding diets supplemented with the probiotic *Lactobacillus* and *Bifidobacterium,* an increase of water content with the probiotic dose (0; 0.5; 1; 1.5, respectively 2 g probiotic), respectively a linear decrease of the lipids content.

Although there were no statistically significant differences ($p>0.05$) observed in the protein content between the variants, slightly higher values were recorded in variants V2, V3, and V4. Fish meat with higher protein content and lower fat is desirable in aquaculture. The same results were obtained by Opiyo et al., 2019, who supplemented the diet of Nile tilapia with different levels of *Saccharomyces cerevisiae* (1×10^{10} CFU g^{-1}) or *Bacillus subtilis* (1×10^{9} CFU g^{-1}).

The ash content of the samples was found to be significantly higher ($p<0.05$) in the V4 variant (1.10±0.13%) compared to V1, V2, and V3, which showed no significant differences ($p>0.05$) in terms of ash content.

There are reports in the literature indicating that the addition of probiotics does not cause significant changes in the protein, lipid, or ash content of fish meat (Merrifield et al., 2010). A study conducted by Mocanu et al. (2012) also state that the addition of probiotics (BioPlus®2B) to fish feed at excessively high doses does not have a significant impact on the biochemical composition of meat. However, when probiotics are administered at an appropriate dosage, they can potentially improve the nutritional qualities of the meat.

In contrast, some studies have reported a significant increase in the protein content of fish meat, which could be attributed to increased nutrient deposition or higher secretion of proteins by probiotics in the gastrointestinal tract (Lara-Flores & Olvera-Novoa, 2013).

According to a study conducted by Opiyo et al., in 2019, it was found that fish fed with diets supplemented with probiotics, specifically *S. cerevisiae,* exhibited a notable increase in protein content and a decrease in lipid content compared to the control group. The observed increase in protein content may be attributed to enhanced nutrient deposition within the fish.

In conclusion, it is evident that adding the fermented product in *A. baerii* meat has an improving effect on body composition. However, it is important to consider that the effects of probiotics on the composition of fish meat may vary depending on the fish species, type of probiotic, as well as other factors such as diet and environmental conditions.

CONCLUSIONS

The current study examined the impact of dietary fermented products made from a combination of kombucha and milk kefir granules grown in sucrose, bovine colostrum and black tea, on the crude protein and lipid content of fish meat compared to a control group. The findings indicate that the inclusion of fermented products led to an increase in crude protein and a decrease in lipid content, which could be advantageous for fish meat.

However, the effects of the fermented product on fish meat composition may be influenced by various factors such as microorganism type, administration dose and duration, fish species and age, and environmental conditions.

Further research is required to better understand the potential benefits and mechanisms of this product in aquaculture, with the aim of enhancing the proximate composition of *A. baerii* meat.

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