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The Effect of Pancreatic Hydrolysate of Soy Protein on Growth, Development and Amino Acid Composition of Muscle Tissues in Lena Sturgeons

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Abstract

The article contains materials about studying the influence of pancreatic hydrolysate of soy protein on productivity, survival rate, chemical and amino acid composition of the muscle tissues in the Lena sturgeons grown in cages at natural temperature of the 4th fishery zone of the Russian Federation. The research was aimed at increasing productivity of the Lena sturgeon grown in cages. Analysis of the obtained research data shows that the content of crude protein in the muscle tissues of bions in the experimental group was higher by 8.6% (P>0.95) than that in the reference group. The obtained data allow recommending the use of pancreatic hydrolysate of soy protein for cultivating Lena sturgeon as a biostimulant for increasing productivity and usefulness of protein in the muscle tissues.

Keywords: feed, feeding, pancreatic hydrolysate of soy protein, amino acids, Lena sturgeon, system of cages.

INTRODUCTION.

The most important component of supplying protein of animal origin for the world's population is fish. However, in recent years, catching of valuable food fish has decreased, and may only be recovered by means of artificial cultivation - aquaculture.

One of the most mastered object of sturgeon breeding on the territory of the Russian Federation is Siberian sturgeon (of the Lena population) *Acipenser baerii* (Brant, 1869), which, due to its plasticity, has enormous potential for growing in various regions of the country, and may be successfully cultivated in fish farms of various types. It features eurythermy, can withstand water temperature increase to +30 ^{0}C. However, sturgeon grows most intensively at the temperature of 15–25 ^{0}C [1, 2, 3].

Currently one of the most promising and cost-effective forms of industrial aquaculture is box farming. Here, the most important factor for maintaining normal functioning of the fish organisms is complete and balanced nutrition. Proper organization of biologically complete feeding of fish helps maximizing their genetic potential [4, 5, 6]. This increases the value of protein as the main essential factor in the nutrition of animals and fish. The usefulness of protein for the synthetic processes is determined by the fact that it contains over 22 amino acids. Fish synthesize only half of them. In this regard, essential amino acids should come with food, since lack of amino acids in the diets is accompanied by poor appetite, decreased growth rate, overall resistance of the organism, and results in an increased protein consumption per unit of production [2, 7]. To enrich fodder with amino acids, feed additives may be used in the form of a complex of essential and nonessential amino acids.

The source of protein, essential and nonessential amino acids in compound feeds for valuable fish species such as salmon and sturgeon is mainly fish flower, the amount of which in the feed mixture is about 45-50%, or more. However, this component is expensive, and has recently been deficient.

In this context, the use of food additives for enriching the diets with nutrients and improving fish growth is important for fish farming [1, 8]. Most interesting is the complex amino acid feed additive based on dry pancreatic hydrolysate of soy protein of the average decomposition degree containing 20-30% free amino acids and 70-80% lower peptides. It is characterized by the upper limit of molecular mass of around 5 kDa and the ratio of the number of free amino groups to their total number equal to 0.4-0.6, and does not contain genetically modified ingredients. The addition contains a group of essential amino acids, such as lysine, arginine, histidine, threonine, leucine, isoleucine, valine, methionine, tryptophan and phenylalanine.

METHODS

We have studied the effect of pancreatic hydrolysate of soy protein on the productivity of Siberian sturgeon (of the Lena population) *Acipenser baerii* (Brant, 1869) grown in cages at the natural temperature of the 4th fishery zone of the Russian Federation.

The study was performed in a pond with an area of 157 hectares located within the Marksovskiy district of the Saratov region on the basis of the Feeding, Zoo-hygiene and Aquaculture Department of the "Technologies of feeding and growing fish" Research Laboratory of the FSEI HE (Federal State Educational Institution of Higher Education)"The Saratov State Agricultural University n.a. N. I. Vavilov".

Lena sturgeon was cultivated in a system of cages [9], which included four 2.5x2.5x2.5 m cages made of

nodeless latex webbing with 10 mm wall mesh and 3 mm bottom mesh.

200 bions of Siberian sturgeon (Lena population) *Acipenser baerii* (Brant, 1869) were purchased for the experiment from the IE Vertey's fish farm in Saratov district of the Saratov region. Juveniles have been accustomed to eating pelleted feed.

Feeding was performed 4 times in the daytime, at equal intervals, with complete combined feed with the pellet size of 3.0-4.0 mm. The combined feed was obtained by extrusion, and consisted of fish flour (57.5%), soybean meal pellets (20.0%), wheat (1.5%), fish oil (20.0%) and premix (1.0%).

During the experiment, fish in the reference group received dry complete ration pelleted feed. The fish in the experimental group received the same combined feed soaked in a 25% solution of soy protein pancreatic hydrolysate in the ratio of 1:1 according to the developed method [10].

Water temperature was measured three times a day, at 7, 13 and 19 o'clock, the content of dissolved oxygen in water was determined once a week. To adjust daily norms, the growth rate of fish was monitored every 7 days based on test fishings.

Chemical composition of muscle tissues was analyzed according to the methods described by L. V. Antipova, I. A. Glotova and I. A. Rogovy (2004).

The initial moisture was determined according to AFNOR NF V04-401 Meat, meat products and fishery products - Determination of moisture content.

Wet ash was determined according to GOST 31727-2012 (ISO 936:1998). Meat and meat products. Determination of the mass fraction of total ash.

Determination of fat was performed by the skim residue according to GOST 23042-2015. Meat and meat products. Determination of fat.

Determination of protein was performed according to the method of Kjeldahl.

Calcium was determined according to GOST R 55573-2013. Meat and meat products. Determination of calcium by titrimetric methods.

Phosphorus was determined with the use of the colorimetric method GOST 32009-2013 (ISO 13730:1996). Meat and meat products. Spectrophotometric method for determination of mass fraction of total phosphorus.

Nitrogen-free extractive substances (NFES) were determined by the calculation method.

Amino acids were identified with the use of precolumn modification with 6-aminoquinolyl-N-hydroxysuccinimidyl carbamate - AccQ according to the method of Waters AccQ-Tag with the use of the set of WAT 052880 reactants. This method ensures specific quantitative modification of primary amino groups, amino acids and amino sugars, and is characterized by high sensitivity and high separation efficiency.

The efficiency of growing the Lena sturgeon was identified in the end of the experiment by the aquaculture and biological indicators. The obtained experimental data were subjected to biometric processing with regard to the recommendations of the G. F. Lakin (1990) with the use of the MS Excel 2007 software package.

RESULTS

The average temperature in the period of study was higher for this season, and ranged between 16.3 and 38 ^{o}C. Water temperature at the surface was in the range between 17 and 28 ^{o}C, water temperature at the depth of 1 meter was lower than at the surface by 4.3 ^{o}C, the temperature at the bottom of the cage and at the bottom of the pond was 1 ^{o}C lower than the temperature at the surface in the range between 16 and 27.3 ^{o}C. Other hydrochemical parameters, including oxygen content in water (8.0 mg/l), were within the physiological norm. This contributed to high productivity and survival rate of sturgeons, which in the reference group was 93.0%, and in the experimental group - 98.0%. Indicators of growth intensity were determined weekly and analyzed at the end of the studies (Table 1).

At the beginning of the experiment, the sample weight of juveniles in both groups was the same - about 100.0 g. The most intensive growth was observed in the experimental group; by the end of the first month, the weight of the fish in the reference group increased twice, and in experimental - 2.5 times. This allowed in 5 months of the study to obtain fish weight in the reference group of 836.7±5.2 g, and in the experimental group – of 897.4±7.4 g (P>0.999). High growth rates are explained by the fact that the juveniles were adapted to the conditions of feeding and maintenance, and quickly adapted to industrial methods of cultivation. The waste therefore was low, only the weak bions died in the spring and only the largest bions died during the hot summer weather.

Products of sturgeon fish are widely demanded in the consumer market. Therefore, it is important to solve the issues of determining chemical composition and quality characteristics of fish muscle tissues, and their nutritional value (Table 2).

Table 1 – Dynamics of Lena sturgeon weight during the experiment, g

Period of experiment, month	Group	
	reference	experimental
Start of experiment	99.7±1.2	100.2±2.3
1	213.7±3.7	229.7±3.0*
2	396.3±2.9	423.8±4.5**
3	600.3±2.7	641.2±4.2**
4	753.2±4.9	806.5±3.9***
5	836.7±5.2	897.4±7.4***

Note: *P>0.95; ** P>0.99; ***P>0.999 (P – validity criterion)

Table 2 - Chemical composition the air-dry matter in the muscle tissues of Lena sturgeon, %

Substance	Group	
	reference	experimental
Hygroscopic humidity	3.0±0.3	3.3±0.1
Crude protein	45.6±1.2	54.2±3.1*
Crude fat	43.6±2.6	37.8±3.1
Ash	3.1±0.2	3.5±0.2
Nitrogen-free extractive substances (NFES)	7.7±0.2	4.5±0.1

Note: *P>0.95

Table 3 - Amino-acid composition of muscle tissues' protein of Lena sturgeon, g/100 g

Amino acid	Start of experiment	Group				
		reference		experimental		
		end of experiment	+/- to the beginning	end of experiment	+/- to the beginning	+/- to the reference
Asparaginic acid	1.54±0.03	1.59±0.04	0.05	1.84±0.04	0.30***	0.25***
Serine	0.59±0.02	0.61±0.01	0.02	0.73±0.02	0.14***	0.12***
Glutamic acid	2.38±0.05	2.38±0.07	0.00	2.77±0.05	0.39***	0.39***
Glycine	0.62±0.02	0.66±0.02	0.04	0.81±0.02	0.19***	0.15***
Histidine	0.50±0.02	0.49±0.02	-0.01	0.60±0.02	0.10***	0.11***
Threonine	0.62±0.01	0.67±0.02	0.05	0.78±0.02	0.16***	0.11***
Arginine	0.83±0.02	0.84±0.03	0.01	1.03±0.02	0.20***	0.19***
Alanine	0.85±0.02	0.87±0.02	0.02	1.00±0.02	0.15***	0.13***
Proline	0.46±0.01	0.47±0.01	0.01	0.56±0.01	0.10***	0.09***
Cystine	0.10±0.001	0.11±0.01	0.01	0.14±0.001	0.04***	0.03***
Tyrosine	0.49±0.02	0.49±0.02	0.00	0.60±0.01	0.11***	0.11***
Valine	0.71±0.01	0.75±0.02	0.04	0.87±0.01	0.16***	0.12***
Methionine	0.40±0.02	0.39±0.03	-0.01	0.47±0.02	0.07*	0.08*
Lysine	1.48±0.02	1.55±0.05	0.07	1.71±0.07	0.23*	0.16**
Isoleucine	0.67±0.01	0.71±0.02	0.04	0.83±0.01	0.16***	0.12***
Leucine	1.15±0.02	1.21±0.03	0.06	1.42±0.02	0.27***	0.21***
Phenylalanine	0.64±0.02	0.66±0.02	0.02	0.81±0.01	0.17***	0.15***
TOTAL	14.03	14.45	0.42	16.97	2.94***	2.52***

Note: *P>0.95; ** P>0.99; ***P>0.999

DISCUSSION

Analysis of the obtained data showed that crude protein content of the muscle tissue in the bions in the experimental group, compared to the reference group, was higher by 8.6% (P>0.95); it was the evidence of increased plastic exchange, and the effect of feed additives on the chemical composition of muscle tissues. The content of crude fat in the muscle tissues of the reference group was higher than in the experimental group by 13.7%. The contents of inorganic compounds in the muscle tissue in the reference group were by 0.4% lower than in the experimental group. Hence, a conclusion may be drawn that fish in the experimental group better digested and accumulated nutrients in the body.

Based on the importance of amino acid nutrition of fish, accumulation of amino acids in their muscle tissues, and further consumption of fishery products by humans, we determined the quantitative content of amino acids in the muscle tissues of juvenile Lena sturgeon grown in cages during our experiment. The data in Table 3 show that muscle tissues of the Lena sturgeon contain biologically complete protein, and a complete set of essential for fish and nonessential amino acids. At the end of the experiment, in the reference group there was no veracious difference in the protein composition from the similar data in the beginning of the experiment.

However, one can note the increased content of essential amino acids: lysine by 4.7%, leucine by 5.2%, threonine by 8.1%, while the content of tyrosine and glutamic acid did not vary significantly, and the amount of methionine and histidine reduced insignificantly.

In the experimental group, the total content of amino acids was significantly higher than at the beginning of the experiment by 2.94 g (P>0.999), and in the reference group - by 2.52 g (P>0.999). The content of essential amino acids increased by more than 20%, compared to the beginning of the experiment, and to the reference group. Of nonessential amino acids, most attention should be paid to the contents of glutamic acid, as it is actively involved in the biosynthesis of most other amino acids as an amino group donor, its content is higher in the experimental group by 16.4% (P>0.999), than at the beginning of the experiment and in the reference group at the end of the experiment.

CONCLUSION.

Thus, a conclusion may be made about the positive effect of soy protein pancreatic hydrolysate on the usefulness of the muscle tissues' protein in the Lena sturgeon. The use of the "Biopeptide" feed additive for feeding Lena sturgeons in cages increases the intensity of the metabolic processes and the content of crude protein in the muscles, respectively, by 8.6%. The use of soy protein pancreatic hydrolysate increases the content of amino acids in the muscle tissues of sturgeons by 17.4%, compared to the reference group.

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