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Productive feeding of rainbow trout: properties, effects on physiological state and interior indicators

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Abstract. It is advisable to use secondary resources of the fish industry with high protein content as a raw material for the development of biologically complete feeds in aquaculture. The work uses standard and original research methods. A complex studies of the chemical composition, physico-chemical and microbiological quality indicators of the experimental feed proved its balance in amino acid composition. The degree of essential amino acids was 91.5 to 107.3%. The biological value of feed protein is 95.18%. It was proved that the amount of free ammonia released through the fish gills is directly proportional to the amount of feed eaten, inversely proportional to the feed coefficient and equal to 0.05. The pH value changed on the 42nd day from 7.6 to 7.7 but remained within the range of pH 6-9, which is optimal for the life support of fish. The absolute increase in the amount of individuals in the control group was 1.2% higher compared with the experimental one. At the same time, feed costs for the entire feeding period were 0.99. Fish survival was 98%. In both cases, bifidobacteria were isolated from the resistant microflora. Notably, the bifidobacteria titer was 3 lg units higher in the trout of the experimental group in comparison with the control group, which does not exceed permissible norms. Thus, the obtained feeding is not inferior in quality to the imported feeding. Therefore, it can be recommended for feeding rainbow trout grown in recirculating aquaculture systems.

1. Introduction

Currently, the development of domestic aquaculture is hindered by dependence on the high-quality of the imported feedings, which have significantly risen in price due to ruble devaluation. In addition, logistics costs are included in the price. According to officials, delivery of feeds to fish farms increases their cost by 30-40% and in some cities adds to the price as much as 100%. The costs of Russian fish feed reach about 65-70% of the production cost while in Europe the feed component accounts for 25-35%.

Today, many domestic companies offer a wide range of feed compounds for fish. However, Russian feed compounds production is characterized by small volumes of output, low quality and a narrow range of raw materials, thus, fish farms often opt for imported products. It is stipulated by the fact that world feed compounds producers have a large base, including the thorough studies of more than 60 species of fish, for which various feed recipes have been developed. The pivotal suppliers are enterprises located in EU countries: Norway, Italy and France. We can state that imported feed production destabilizes the development of Russian aquaculture.

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However, high prices, complete dependence on the exchange rate, inconvenience that periodically arises due to supply disruptions (veterinary or customs problems) are not the only issues. Domestic fish farmers are not always satisfied with imported feed since the conditions for growing fish on domestic farms differ from the conditions and standards adopted in foreign countries. In addition, imported feed is mainly intended for certain fish species (e.g. trout and salmon) widely grown in foreign aquaculture, the formulations of foreign feeds are insufficiently developed for typical fish species in Russia and cannot provide high efficiency.

Therefore, the problem of finding new and alternative ways to obtain feed compounds, improve the quality and reduce the cost of their production is relevant and is one of the main tasks of the agricultural sector of the economy.

2. Problem setting

At the present stage of the fishing industry development, the President of the Russian Federation approved a list of instructions on the development of the fishery complex, which affects a number of aspects of the fishery complex related to ensuring the manufacturing of fish products with a deep degree of processing, developing processing facilities and increasing the rate of renewal of the fishing fleet and import substitution.

A significant proportion of waste is generated during the manufacturing fish products of wide consumer demand. Mainly, these are protein-containing ones, which are currently in low demand.

Considering a large amount of heads and entrails (23-33%) during the initial cutting of fish, the main objective is to involve them in the main production of animal feed as they currently represent direct losses.

Since the secondary protein-containing fish resources are the most expensive recipe components in the development of productive feeding for rainbow trout. It is advisable to use dry fish mixtures obtained from by-products and waste from the fishing industry as the main raw materials. Analysis of the chemical and fractional composition of proteins as well as microbiological indicators helps assess the prospect of using protein-containing raw materials of the fishing industry in feed production rather positively [5].

3. Materials and methods

Efficiency of using the obtained productive feeding Fish Food for growing rainbow trout was studied on the basis of the Department of Quality Management and Technology of Aquatic Bioresources in the conditions of the Aquabioresource Innovation and Technology Center, Voronezh Federal State Budgetary Educational Establishment of Higher Education, Voronezh.

Rainbow trout was fed in a fish breeding recirculating aquaculture system for intensive breeding of various fish species with an initial level of oxygen dissolved in water with the proportion of 10 ± 0.5 mg/l and an initial water temperature of 15 ± 0.2 °C (according to the technological recommendations "Food for trout and whitefish for the RAS installation»: 2018-2019, Alltech Coppens, the Netherlands).

Rainbow trout with an initial average weight of 320 g were taken for experimental feeding. They were put into production pools with the capacity of 5.5 m³ at standard stocking densities. The initial weight of the fish from experimental group was 50.170 kg and that of the control group was 50.200 kg. An experimental group of fish was fed with Fish Food and a control group was fed with productive feeding Alltech Coppens at the ratio of 1.4% of the fish weight in the group (according to technological recommendations "Food for trout and whitefish for RAS installation": 2018-2019, Alltech Coppens, Netherlands). The experimental feeding took 56 days.

The experimental and control lots of fish (10 individuals) were weighed with a frequency of 1 time over 14 days. The studies of the water hydrochemical parameters were carried out throughout the entire feeding period, specifically, the value of dissolved oxygen and temperature were measured daily, and the content of NH₃, NO₂, NO₃, and pH in the water was monitored bi-weekly.

The length and weight of the fish was determined according to State standard 1368-2003.

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Studies of the organoleptic and physical parameters of rainbow trout in the experimental and control groups were carried out according to State standard 7631-2008.

The chemical composition of rainbow trout (moisture, fat, protein, alkali, carbohydrates) was determined according to State standard 7636-85.

Analysis of the qualitative and quantitative composition of the fish intestines microflora during feeding was determined according to the methodological recommendations "Methods of bacteriological research of opportunistic microorganisms in clinical microbiology" (approved by the Ministry of Health of the RSFSR on December 19, 1991).

Hydrochemical parameters of water were performed by standard methods in accordance with State standard 15372-87.

The average daily fish gain was determined by the G.G. Winger formula:

$$Cw (\%) = \frac{2(Me-Mb)}{(Me+Mb)\tau} 100,$$
(1)

where Mb is fish mass at the beginning of feeding, g;

Me s fish mass at the end of feeding, g;

 τ is length of feeding in days.

Water temperature and the content of dissolved oxygen in the water were determined using a HANNA instruments HI 9142 thermometer.

Feed costs were determined according to the recommendations of Shcherbinina, Gamygina (2006) according to the formula:

$$3 = \frac{E_B}{R},\tag{2}$$

where EB is the amount of feeding, kg; R is end product, kg.

4. Results and discussion

Studies of the Fish Food chemical composition showed that the protein content in the developed feed is 39.8%. As is known from the literature [5, 6], the high activity of digestive enzymes help rainbow trout digest a greater amount of animal proteins (30–50%) introduced into the granular feed than when eating natural food (10–18%).

The increased content of native protein in the feeding contributes to the growth and more efficient feeding utilization. It should also be noted that the feed coefficient decreases with a high proportion of protein and, accordingly, it affects the expenses of an increased number of 1 kg of fish [1, 2].

The carbohydrate content of Fish Food being the major source of energy for rainbow trout is 20.5%. It has been established that not more than 9-12% of digestible carbohydrates are necessary for the rainbow trout and their total content in granular feed should not exceed 25-30%.

The fiber content of Fish Food is 2.0%. It is not digested by trout but has a positive effect on the secretion of digestive enzymes, which leads to the increased protein digestibility.

The obtained feeding contains 19.7% of fat being the most important component in the nutrition of rainbow trout. Trout can digest a significant amount of fat (up to 25% of the total feed composition). It is known that more protein is consumed when there is a lack of fat in feed in order to cover energy costs, and, as a result, fish growth decreases and feed consumption increases. According to the FAO Technical Guidelines for Responsible Fisheries, "Inland Fisheries" (2010), the optimal ratio between protein and fat recommended when compiling granular feed is from 2:1 to 3:1. In the experimental feed, this ratio was equal to 2.1:1, which is the norm.

The fish demand for protein depends on the amino acid composition of the protein, which contains essential amino acids. The protein in fish feed is considered complete if it contains all the essential amino acids and is in a balanced state [2].

As is well known, every irreplaceable amino acid has its own application in the body. Their lack or excess leads to changes in a fish organism.

Productive feeding protein contains all the essential amino acids that are in a balanced state. According to comparative analysis of certain fish species need for essential amino acids (Shcherbina, Salkova, 1987; Saenko, 1996; Kaushik, Cuzon, 1999, etc.), it was found that Fish Food satisfies the requirement of rainbow trout for such essential amino acids as valine, lysine, and phenylalanine for more than 99%. It is known that an insufficient content of valine in the fish diet leads to impaired coordination and hyperesthesia while lack of lysine in feeding leads to impaired blood circulation, a drop in the number of red blood cells and a decreased concentration of hemoglobin in them, and, most significantly, violation of nitrogen balance.

Thus, the Fish Food is balanced with regards to amino acid composition and it satisfies the need for rainbow trout in essential amino acids from 91.5 to 107.3%. The feeding amino acid scores difference coefficient is 4.82% and the biological value of the Fish Food protein is 95.18%.

Mineral nutrition of fish is an integral part of the overall nutrition. Fish, like all animals, need both macro- and microelements.

It has been experimentally established that Fish Food contains all vital biotic elements that are of biological significance for the fish organism. It should also be noted that satisfaction of the rainbow trout needs for mineral elements ranges from 78.3 to 99.0%. As is well known, fish gets minerals not only with food but also from the water. They penetrate through the integument of the body, gills and esophagus. With the balanced feed, the need for fish in minerals is minimal.

Vitamins unlike proteins, carbohydrates and fats are not the sources of energy and they are required in a very small amount. They perform catalytic functions and the lack of vitamins in the feed causes a deep disturbance in fish metabolism, which leads to diseases and often death [3, 4].

The obtained feed is balanced in vitamin composition and satisfies rainbow trout's needs. Vitamin composition of feed was achieved by introducing vitamin premix PF-1B into the feed formulation.

Studies of the qualitative and quantitative chemical composition of the obtained Fish Food for rainbow trout showed its basic nutrients balance.

An obligatory indicator of the fish feed quality is harmlessness, which is proved by the own experimental results: the value of TBC (total bacterial count) was 1.5*105 CFU/g with an acceptable value of not more than 5*105 (according to Appendix No.1 to the technical regulation "On feeding safety and feed additives", paragraph 2.2).

The normal course of all physiological processes and the increase in fish mass prove that nutritional requirements are fully satisfied. This means that all the substances and compounds necessary for the body are present in the feeding, which means that these feeds have the necessary nutritional value [6, 7].

Therefore, the developed Fish Food for rainbow trout meets the requirements of the technical regulations and can be recommended to all types of cryophilous fish.

The following physicochemical quality indicators were determined with the objective to assess the functionality of the feed: granules humidity, water resistance and friability. The results of the studies showed that the Fish Food meets the requirements of State standard 10385-2014 with regards to physico-chemical parameters.

Thus, the comprehensive studies of the chemical composition, physico-chemical quality indicators and microbiological indicators of the developed Fish Food are able to satisfy the needs for basic nutrients. According to physicochemical and microbiological indicators, Fish Food complies with the regulatory documentation for these types of feeding. Therefore, Fish Food can be recommended for intensive cultivation of rainbow trout in recirculating aquaculture systems.

The analysis of hydrochemical indicators for the entire feeding period of the experimental and control groups of trout are presented in Table 1, which shows that such indicators as the amount of free ammonia released through the fish gills are directly proportional to the amount of feed eaten and inversely proportional to the feed coefficient, and is equal to 0.05.

Table 1. Hydrochemical indicators (according to the technological recommendations Food for trout
and whitefish for RAS installation": 2018-2019, Alltech Coppens, Netherlands)

Indicator	Fish group	Duration of feeding, days				
Indicator	Fish group	0	14	28	42	56
Dissolved oxygen, mg/l	Experimental	10 ± 0.5 taking into account the supply of additional oxygen to maintain the necessary level in water (with daily measurements)			l oxygen to	
	Control				easurements)	
Water temperature, °C	Experimental Control	15±0.2 (with daily measurements)				
NH ₃ (ammonium	Experimental	0.05	0.05	0.05	0.05	0.05
nitrogen), mg/l	Control	0.05	0.05	0.05	0.05	0.05
NO ₂ (nitrites), mg/l	Experimental	0.1	0.2	0.2	0.2	0.2
_	Control	0.1	0.2	0.2	0.2	0.2
NO ₃ (nitrates), mg/l	Experimental	10	15	15	15	15
	Control	10	15	15	15	15
	Experimental	7.6	7.6	7.6	7.7	7.7
рп	Control	7.6	7.6	7.6	7.7	7.7

The pH value changed on the 42nd day from 7.6 to 7.7 but remained within the range of pH 6-9, which is optimal for the life support of fish. The concentration of nitrates and nitrites during the entire time of feeding increased slightly and was within the normal range. This is primarily due to the increase in the mass of experimental fish lots and, accordingly, to the increase in the mass of the introduced feed. Thus, the introduction of Fish Food into the diet of rainbow trout does not change the hydrochemical parameters of water.

It is important to evaluate the effectiveness of the obtained product in comparison with foreign feed manufacturers (Netherlands), which occupy a leading position in the Russian feed market, due to the fact that fish feeding in recirculating aquaculture systems is the main factor ensuring fish growth.

The test results of the experimental productive feeding for rainbow trout showed its high productive properties (Table 2).

Crowth indicators	Fish group		
Glowin indicators	Experimental	Control	
Initial mass, kg	50,170	50,200	
End mass, kg	106,809	107,526	
Absolute gain, kg	56,639	57,326	
The average daily gain, kg/day	1.011	1.024	
The average daily gain,%:			
on the 14 th day	1.38	1.38	
on the 28 th day	1.36	1.37	
on the 42^{nd} day	1.31	1.32	
on the 56^{th} day	1.29	1.30	
Feed costs, units	0.99	1.00	
Survival rate,%	98	98	
Growing period, days	56	56	

 Table 2. Rainbow trout growth indicators

Table 2 shows that the indices of fish growth in the experimental and control groups practically did not differ from each other. A slight increase in growth was noted in individuals of the control group in comparison with the experimental ones. The absolute increase in rainbow trout in the control group was higher by 1.2% (0.687 kg) compared with the control one but the feed costs for the entire feeding

period were less when feeding the experimental lot and was equal to 0.99. The survival of fish in the experimental and control groups was 98%.

Thus, the analysis of rainbow trout cultivation indices in the laboratory experiment revealed the Fish Food effectiveness.

It has been established that the introduction of Fish Food into the diet of rainbow trout produces a positive effect on the chemical composition of fish flesh (Table 3).

Indicator		Rainbow trout			
		Experimental group	Control group		
Moisture, %		74.25	72.98		
Protein, %		17.88	17.08		
Fat, %		5.21	5.41		
Alkali, %		1.29	1.13		
Nitrogen-free substances, %	extractive	1.17	3.60		

Table 3. Chemical composition of fish body

Analysis of the chemical composition of rainbow trout body (Table 3) showed that the introduction of Fish Food increases the protein content in the fish body by 4.5% in comparison with the situation when fish is fed with imported Coppens. As well, a 3.7% decrease in the mass fraction of fat in the body of the experimental fish group and a 12.4% increase in the alkali content compared with the control one are noted, which proves the appropriateness of using domestic production for fish grown in aquaculture.

The impact of the experimental productive feeding Fish Food and control production feeding Coppens on the interior performance of rainbow trout was also evaluated (Table 4).

	Mas	s, kg	Size, cm	
Duration of feeding, days	Experime	Control	Experime	Control
	ntal	Control	ntal	Control
0	0.320	0.320	30.5	30.5
56	0.720	0.725	37.5	38.0

Table 4. Interior indicators of rainbow trout in the control and experimental groups

Table 4 shows that the mass and size characteristics of rainbow trout in the experimental and control fish groups are almost identical.

Studies of organoleptic indicators of rainbow trout in the experimental and control groups showed compliance with State standard 24896-2013 (Figure 1, (a, b)).

Fish has the following appearance indicators: the surface is clean, color is natural and inherent in this species of fish, with a thin layer of mucus; signs of disease are absent, the gills are red. Eyes are bright, transparent, without damage. Odor is characteristic of live fish of this species without extraneous odors.

The condition of the internal organs of rainbow trout in the experimental and control groups have showed that the position, shape and color of the internal organs are normal and any changes are absent. Heart obesity has not been revealed, the structure of the liver and spleen tissue is dense, and the kidneys are without tissue growths (Figure 1 (a, b)).

Basing on the undertaken examination of the appearance and condition of the rainbow trout's internal organs, it is possible to conclude that feeding with Fish Food does not entail pathological changes in fish.

According to scientists (Intizarov, 1994; Vorobiev, 1999), the normal microflora of fish performs a number of vital functions. It has a certain qualitative and quantitative composition for each species and is a powerful barrier to pathogenic microorganisms (Table 5).

It is proved that in the intestines of fish there is a non-pathogenic anaerobic resident microflora that does not exceed permissible norms.



Table 5. Qualitative and quantitative composition of microflora (micro view) of the contents of the	
rainbow trout intestines	

Indicator	Rainbow trout			
Indicator	Experimental group	Control group		
Total seeding, CFU/ml	3.4.103	2.1.103		
Bifidobacterium titer	10-6	10-3		
Lactobacillus titer	Not stated	Not stated		
Entrobacteria	Enterobacter spp.			
	1.1.103	-		
Staphylococci	-	2.9.101		
Enterococci	-	-		
Bacilli	Bacillus spp.			
	6.9.102	-		

5. Conclusion

Thus, on the basis of comprehensive studies of the obtained productive feeding Fish Food, we can conclude that the obtained food is not inferior to imported food (Netherlands), therefore, it can be recommended for feeding rainbow trout in industrial aquaculture.

In addition, the application of Fish Food together with dry fish mixtures obtained from the secondary products of cutting silver carp salmon is the most effective option for rainbow trout cultivation against the economic background. It is stipulated by the fact that a high cost of non-domestic feeding negatively affects the profitability of rainbow trout cultivation under industrial conditions; specifically, approximately 70% of the cost of farmed fish falls on the cost of feed. Consequently, the use of Fish Food will solve the problem of import substitution of feed in aquaculture and will provide enterprises with complete and high-quality feeding.

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