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Hematological and biochemical blood indices of rainbow trout depending on the level of protein in feeds

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Abstract. Protein nutrition of fish is one of the main factors of ensuring their active growth. Finding the optimal concentration of protein is an actual direction of research. The purpose of the experiment was to establish the effect of different protein nutrition levels in commercial rainbow trout feeding on hematological and biochemical parameters of their blood. For this purpose, five experimental groups were formed by the method of analogs. The experiment lasted 210 days and was divided into two periods: equalization (10 days) and main (200 days). During the equalization period, the feeding ration was the same for fish in the control and experimental groups. In the main period, the level of protein in experimental feeds for different experimental trout groups ranged from 44 to 52% per 1 kg. It was found that different levels of rainbow trout's protein nutrition did not cause significant changes in blood morphological composition. It is proved that rearing trout using compound feeds with a crude protein content of 50% and 52% promotes the increase of red blood cells, hemoglobin, white blood cells and phagocytic activity in blood. The use of 44% and 46% low crude protein diets in fish feeding reduces the morphological components in blood. As a result of studies on the phagocytic activity of neutrophils in fish blood, a slight increase was revealed under the influence of the studied factor. The analysis of the obtained results showed that with increasing levels of crude protein in the diets of rainbow trout, the concentration of total protein and its fractions in blood serum increased. Accordingly, the amount of total protein in blood of fish in the control group was 60.0 g/L, in the experimental groups – it was within the range of 59.6-65.3 g/L. The increase in the level of rainbow trout's protein nutrition in the experimental groups 4 and 5 caused an increase in the number of albumins, β-globulins and γ-globulins compared to control. As a result of experimental studies, an increase in the mineral status of rainbow trout blood during use of feeds with the crude protein content of 50-52 % was established. No differences were found during the assessment of trout white blood cell counts. The use of the obtained results during the production of compound feed will allow having a positive effect on the hematological parameters of commercial rainbow trout

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Introduction

Many researchers confirm the need for hematological studies to clarify the physiological status of fish, because using physiological methods it is possible to establish the presence of pathological disorders in a particular system and to identify the degree and nature of deviations from the normal fish body's status. Also, a significant number of researchers emphasize the great practical importance of studying blood parameters in different species of fish depending on the growing conditions. Therefore, the study of changes in the blood system of rainbow trout fish depending on protein nutrition is one of the criteria to assess the course of adaptive and compensatory reactions. Thus, the study of different rainbow trout protein nutrition levels' impact on hematological and biochemical parameters is necessary to determine the protein need for fish in order to ensure the successful operation of cold-water fish farms in Ukraine.

Analysis of Recent Researches and Publications

Biologically complete protein nutrition is one of the main conditions that determine the efficacy of feed nutrients, productivity, health and reproductive functions of fish. The generalization of modern research in the field of fish nutrition taking into account their species, breed and age characteristics, as well as the issue of efficient use of protein feeds and supplements is a necessary condition for solving the problem of protein in industrial fish breeding.

The possibility of protein balancing of compound feed for rainbow trout, both using plant components and including fish meal (Brezas, Hardy, 2020) or meal from poultry offal (Rimoldi *et al*. 2018), has been proven. For optimal growth and effective use of feed, it is recommended (Ahmed, Ahmad, 2020) that young rainbow trout receive feed with protein in the range of 450-471 g/kg. Protein supplements can affect immunity and hematological indicators of rainbow trout. In particular, a positive effect of adding hydrolyzed powder of *Saccharomyces cerevisiae* extract to the feed was established (Tukmechi, Bandboni, 2014), in turn, the addition of silkworm pupae leads to a decrease in the number of erythrocytes and hemoglobin (Shakoori *et al*., 2015).

Analysis of fish blood based on appropriate biochemical research methods is one of the main stages of monitoring the feeding full value condition. However, the variety of factors, including technological, that can affect the blood biochemical composition, laboratory diagnostic methods, the lack of perfect differential diagnostic programs, especially in the conditions of changing nutritional parameters within the physiological norm, significantly complicate the interpretation of data and assessment of fish protein nutrition full value (Sherman *et al*., 2002; Karabulut & Yandi, 2010).

Biochemical studies of rainbow trout blood permit expansion of the understanding of the relationships between blood parameters and mechanisms of homeostasis regulation and to accumulate data for summarizing them at the level of reference values.

It is known that the main component of cells and tissues in the animal body are proteins, which are associated with all vital functions.

Their content in feed affects the level of fish productivity and economic efficiency of production. Accordingly, biochemical blood tests can detail the signs of possible abnormalities. Protein deficiency in the diet is known to be accompanied by certain changes in nitrogen compound metabolism, in particular, a decrease in the serum total protein content mainly due to albumin and sometimes a- and b-globulins and residual nitrogen.

An unbalanced protein diet disrupts the synthesis of proteins, enzymes, hormones and fixation of biologically active substances, which requires the blood test study of general clinical and biochemical parameters along with specific criteria in assessing full value condition of feeding (Kim & Kaushik, 1992; Ullah-Khan *et al*., 2019).

Equally important are the hematological parameters in trout, as blood is one of the most reactive tissues of the fish body. With the help of morpho-physiological analysis of blood and its physicochemical properties, it is possible to accurately and objectively assess the condition of the fish body. Species features of blood are hereditary. These include the composition and morphology of cell forms, quantitative parameters and their seasonal and age dynamics (Mahmud *et al*., 1996).

Materials and Methods

Experimental studies on *Oncorhynchus mykiss* (Walbaum, 1792) two-year-old rainbow trout were carried out in the "Shipot" farm, Perechyn district of Transcarpathian region.

The purpose of the research and economic experiment was to establish the effect of different commercial rainbow trout's protein nutrition levels on hematological and biochemical parameters in their blood.

For this purpose, five experimental groups were formed by the method of analogs (Table 1). During the equalization period of the experiment, which lasted 10 days, the feeding ration was the same for fish in the control and experimental groups. In the main period of the experiment (200 days), the protein level in the feed of trout from the experimental groups was regulated by changing the individual components in the feed (using combined mathematical methods to optimize the calculation by the Agro Soft Win Opti software).

Table 1. *Design of the research and economic experiment*

Fish group	Stocking density at the beginning of the experiment, fish/m²	Mean body weight at the beginning of the experiment, g	Experimental periods	
			equalization (10 days)	main (200 days)
			protein content per 1 kg of feed, %	
1 – control	50	55.3±2.48	48	48
2 – experimental	50	56.1±2.13		44
3 – experimental	50	54.8±2.37		46
4 – experimental	50	55.1±3.13		50
5 – experimental	50	54.5±2.99		52

Rainbow trout were fed 4-6 times a day during the experimental period, at regular intervals during the day. The required amount of feed was calculated according to the indices of individual fish weight and ambient temperature at the time of feeding.

Weighing of experimental trout was performed once every 10 days using electronic

scales in a calibrated water-logged container with an accuracy of 0.1 g. The rearing of commercial two-year-old rainbow trout was carried out in ponds with an area of 100 m^2 at the stocking density of 50 fish/m^2, and the water level in them of 1 m. The total number of trout in experimental studies was 25 thousand fish. The conditions of experimental fish keeping met the regulatory requirements in salmon farming (Kanydev, 1985; SOU-05.01-37-385:2006).

Blood samples were collected from the heart using an injection needle inserted from the abdomen along the sagittal line between the pectoral fins. Heparin was used as an anticoagulant.

The concentration of red blood cells was determined by *in vitro* method using a storage solution, a microscope and a Goryaev's chamber, white blood cells – by an indirect method on a blood smear, hemoglobin – by a hemoglobin cyanide method with a spectrometer. In addition, the total serum protein was determined refractometrically (Kondrakhin *et al.*, 2004; Vlizlo *et al.*, 2012).

Indices of constructive metabolism, namely: glucose content in the fish blood was determined by the method of Kondrakhin *et al.* (2004). The principle of the method is based on the ability of glucose oxidase, while oxidizing, to form a complex compound that turns the solution pink. The color intensity of the solution is proportional to the glucose content in the sample. The blood glucose concentration was calculated according to a standard solution in mmol/L. The content of total protein in blood plasma was determined using sets of reagents from "Lachema" (Czech Republic) and standard solutions of substrates.

The research results were processed by the method of variation statistics (Plokhynskyi, 1969) using the STATISTICA 7.0. and MS Excel software using built-in statistical functions.

Results of the Research and Discussion

Characterizing various changes in the body's vital activities, the blood composition permits to reflect the characteristic properties of metabolism at different stages of the development and under different conditions of the body's existence.

Blood cells are the first in the body to respond to abiotic and biotic environmental factors, they are easily redistributed, thus ensuring the development of adaptive and immune responses.

The results of the studied feeding factor's influence on the hematopoiesis status in the rainbow trout fish body are shown in Table 2.

Table 2. *Hematological parameters in rainbow trout, n=5*

Index	Fish group				
	1st	2nd	3rd	4th	5th
Red blood cells, T/L	1.22±0.07	1.16±0.05	1.18±0.05	1.28±0.09	1.31±0.08
Hemoglobin, g/L	122±1.33	111±2.09**	116±1.97*	123±2.01	125±2.10
White blood cells, g/L	22.9±0.09	21.6±0.11	22.0±0.12	23.5±0.08	24.01±0.21**
Phagocytic activity, %	45.8±2.31	42.3±2.12	44.5±2.54	46.8±2.63	47.3±2.41

Note: **p<0.05; **p<0.01 – compared to group 1*

The results of our studies indicate that different levels of rainbow trout's protein nutrition do not cause significant changes in blood morphological composition. It was found that rearing trout on feed with a crude protein content of 50% (experimental group 4) and 52%

(experimental group 5) leads to an increase in blood red blood cells, hemoglobin, white blood cells and phagocytic activity, while the use of fish diets with low crude protein content (experimental groups 2 and 3) leads to reduced morphological components of the blood. Thus, the number of red blood cells in fish of these groups was within the range of 1.16-1.18 T/L, which is by 5.1% and 3.3% less compared to the control index. The hemoglobin content was also lower by 9.1% and 5.2% than in the control. The above difference is statistically significant ($p<0.01$ and $p<0.05$).

Similar nature of changes was observed in rainbow trout according to the number of white blood cells in the blood, which in fish of the control group was 22.9 g/L, and the experimental groups ranged from 21.6 to 24.1 g/L. There is no significant difference in this index between the control and experimental groups.

Considering the changes in the phagocytic activity of neutrophils in the blood of experimental fish, there is a slight increase under the influence of the studied factor.

Phagocytic activity of neutrophils in the blood of fish in experimental group 5 was higher compared to the control and was 47.3% vs. 45.8%. And when fish consumed feed with a crude protein content of 46% (group 3) and 50% (group 4), the phagocytic activity index was at the control level.

Thus, rearing the rainbow trout on compound feeds with different levels of crude protein did not significantly change their morphological components in blood. However, there is a slight positive effect of crude protein on the number of red blood cells, hemoglobin and phagocytic activity of neutrophils. The above difference between the analogues of the experimental groups was within physiological fluctuations.

Depending on the level of crude protein in feeds consumed by rainbow trout, certain fluctuations were found in blood biochemical parameters (Table 3) but for most of them, no significant differences were found.

The above data show that with increasing levels of crude protein in the diets of rainbow trout, the concentration of total protein and its fractions in blood serum increased. Thus, the amount of total protein in blood of fish from the control group was 60.0 g/L, in the analogs from the experimental groups, this figure was within the range of 59.6-65.3 g/L.

The increase in the feed of crude protein to 52% provided the highest level of the total protein in blood: in fish of experimental group 5, it significantly exceeded the value in the control group by 5.3% ($p<0.05$). In this case, there is also a probable increase in the total protein concentration and compared to trout, which received in the feed 50% of crude protein: the total protein amount in blood serum of fish in the experimental group 4 compared to the analogs of the experimental groups 2 and 3, was higher by 5.03% and 4.2%, respectively, and the control group index was probably lower by 4.3%.

The level of total protein in blood serum of trout in the experimental groups 2 and 3 approached the control index, and was 59.6 g/L and 60.1 g/L against 60.0 g/L, respectively.

The study of the blood protein fractional composition in rainbow trout groups, which were fed compound feeds with different levels of crude protein, minor changes were also found. Thus, the increase in the level of protein nutrition in rainbow trout of experimental groups 4 and 5 caused an increase in the number of albumins, β-globulins and γ-globulins, compared to similar indices of fish in the control group.

It was also found that rearing fish on feed with a reduced crude protein content of up to 44% (experimental group 2) and 46% (experimental group 3) did not significantly affect the

changes in the fractional composition of blood proteins. Similar patterns were observed for the activity of enzymes, which varied depending on the studied factor. Thus, feeding rainbow trout compound feeds with a crude protein content of 50-52% (experimental groups 4 and 5) showed higher alanine aminotransferase activity by 10.0% ($p<0.05$) and 26.1% ($p<0.05$), and aspartate aminotransferase – by 6.7% and 15.6% ($p<0.05$), respectively.

Table 3. *Blood biochemical parameters in rainbow trout of the experimental groups, n=5*

Index	Fish group				
	1st	2nd	3rd	4th	5th
Total protein, g/L	60.0±1.68	59.6±1.65	60.1±1.71	62.6±1.82	65.3±1.45*
Albumins, g/L	24.5±1.06	23.8±1.14	24.0±2.04	24.9±2.13	26.3±1.88
Globulins, g/L	35.2±1.17	34.9±1.24	35.1±1.36	37.8±1.47	38.1±1.35
α-globulins	11.8±0.95	11.3±0.96	12.0±1.03	12.6±1.08	13.2±1.09
β-globulins	12.5±1.12	12.2±1.32	12.6±1.08	12.9±1.14	13.9±1.09
γ-globulins	10.9±1.11	11.4±1.31	10.5±0.99	12.3±1.47	11.0±1.39
Alanine aminotransferase, U/L	23.00±0.58	20.00±1.79	22.00±1.65	25.30±0.69*	29.00±1.76*
Aspartate aminotransferase, U/L	45.00±2.03	41.00±2.14	43.00±1.74	48.00±1.62	52.00±1.59*
Glucose, mmol/l	2.94±0.06	2.76±0.09	2.82±0.11	3.01±0.21	3.15±0.19
Carotene, mg%	0.52±0.02	0.50±0.03	0.51±0.02	0.54±0.07	0.56±0.06
Total calcium, mmol/L	2.84±0.16	2.82±0.21	2.84±0.32	2.91±0.28	2.93±0.28
Inorganic phosphorus, mmol/L	1.67±0.16	1.61±0.23	1.64±0.38	1.69±0.21	1.71±0.41

Note: **p<0.05 compared to group 1*

The enzyme activity, such as aspartate aminotransferase, in fish of the experimental groups 2 and 3, which were fed compound feeds with a crude protein content of 44-46% approached the level of the control, while, compared to the analogs of experimental groups 4 and 5, there was some decrease in its activity.

The glucose content in blood of rainbow trout in the control group was 2.94 mmol/L, in blood of the experimental groups' fish – 2.76-3.15 mmol/L, however, different levels of crude protein in the feed had different effects on the value in fish of the experimental groups. The highest level of glucose was in blood of trout in the experimental group 5 and was 3.15 mmol/L, which is by 7.1% more compared to the control and by 4.7-14.1% compared to the analogs in experimental groups 2-4. Similar nature of the changes is observed in the serum carotene content of two-year-old rainbow trout. Its concentration in fish of the control group was 0.52 mg %, while in fish of the experimental groups ranged from 0.50 to 0.56 mg %. There is no significant difference in this index between the control and experimental groups.

The rate of enzymatic reactions also depends on the concentration of minerals in the serum, in particular calcium and phosphorus. The results of experimental studies revealed an increase in the mineral status of rainbow trout blood when they consumed feed with the crude protein content of 50-52%.

When assessing the indices of the trout's white blood (Table 4), no differences in the number of white blood cells depending on the action of paratypic factor were found. Differential white blood cell count in certain types of

white blood is important for their detection. Changes in the conditions of fish keeping and feeding can increase the functions of some white blood cells and weaken them in others.

Table 4. *Dynamics of changes in leukocyte formula and the total lipids content in blood of rainbow trout, n=5*

Index	Fish group				
	1st	2nd	3rd	4th	5th
White blood cells, g/L	22.9±1.09	21.6±1.11	22.0±1.12	23.5±1.08	24.0±1.21
Neutrophils, %	19.9±1.12	19.25±1.33	19.42±1.47	20.12±1.28	21.20±1.39
Polymorphonuclear, %	3.60±0.12	3.71±0.41	3.83±0.32	4.20±0.65	4.50±0.34*
Lymphocytes, %	60.45±2.06	59.65±2.12	60.14±2.47	61.35±2.36	67.12±2.03*
Monocytes, %	16.80±1.98	16.05±2.03	16.65±1.54	17.40±1.71	17.90±1.65
Total lipids, mmol/L	3.66±0.13	3.52±0.31	3.58±0.29	4.09±0.09*	4.01±0.07*

Note: **p<0.05 compared to group 1*

In particular, during experimental studies, it was found that changes in the feeding conditions of rainbow trout, contributed to some changes in the number of neutrophils in fish blood. Under the influence of the studied feed factor, their growth was found in trout of the experimental groups 4 and 5. Thus, the number of neutrophils was the highest (21.2%) in blood of fish in the experimental group 5, which were grown on compound feeds with increased to 52% levels of crude protein. At the same time, the reduction of crude protein levels to 44-46% (groups 2 and 3) contributed to a decrease in the number of neutrophils compared to the control.

The ratios of polymorphonuclear rods, lymphocytes and monocytes in blood of rainbow trout of the experimental groups, although slightly higher than those in the control, were within physiological fluctuations. When feeds with the crude protein content of 50% (experimental group 4) and 52% (experimental group 5) were used for feeding fish, the content of polymorphonuclear rods significantly increased and the content of lymphocytes and monocytes decreased compared to the control analogs.

Rearing of commercial trout on feed with a crude protein content of 52% (group 5) led to a reliable increase in the number of lymphocytes in blood, while the feeds with the crude protein level of 44% (group 2) and 46% (group 3), on the contrary, led to a slight decrease in their content.

For a more detailed assessment of the studied feed factor's influence on the functional state of the rainbow trout's body, a study of the total blood lipids was performed. In fish of the control group, its level was 3.66 mmol/L; in analogs of the experimental groups, it was 3.52-4.09 mmol/L. The highest level of total lipids (4.09 mmol/L) in blood of two-year-old rainbow trout was in experimental group 4, which were reared on compound feeds with a high (up to 50%) levels of crude protein. It was also found that with the decrease in the level of crude protein in the feed of two-year-old rainbow trout to 44-46% (experimental groups 2 and 3), there was a slight decrease in this index in blood plasma.

Thus, the results of physiological and biochemical studies showed that increasing the levels of protein nutrition in rainbow trout promoted better hematopoiesis, which led to their high commercial qualities.

Conclusions

1. It was found that the number of white blood cells in blood of rainbow trout, which have received compound feed with the protein content of 48% was 22.9 g/L, in blood of fish consuming feed with a protein content of 52%, this figure was higher by 4.8% ($p<0.05$).

2. Increase of crude protein in the production feed from 48 to 52% provides an increase in the total blood protein by 8.8% ($p<0.05$).

3. It is proved that when using rainbow trout feed with the crude protein content of 50-52%, the higher alanine aminotransferase activity by 10.0% ($p<0.05$) and by 26.1% ($p<0.05$), respectively, and by 6.7% and 15.6% ($p<0.05$) that of aspartate aminotransferase, was observed in blood of fish compared to fish that consumed feed with the protein level of 48%.

4. The number of neutrophils was the largest and amounted to 21.2% in blood of fish reared on compound feeds with an increase to 52% level of crude protein. Reducing crude protein levels from 48% to 44-46% contributed to some reduction in neutrophils.

5. It was found that the total amount of blood lipids in two-year-old rainbow trout, which consumed feed with the protein content of 50-52%, increased by 9.6-11.7% ($p<0.05$) compared to those who were reared on compound feed with the protein level of 48%.

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Гематологічні та біохімічні показники крові райдужної форелі залежно від рівня протеїну у комбікормах

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Анотація. Протеїнове живлення риб є одним з основних факторів забезпечення активного їх росту, а виявлення оптимальної концентрації протеїну є актуальним напрямком досліджень. Метою досліду передбачалося встановити вплив різних рівнів протеїнового живлення товарної райдужної форелі на гематологічні та біохімічні показники їхньої крові. Для цього за методом аналогів було сформовано п'ять піддослідних груп. Дослід тривав 210 діб та поділявся на два періоди: зрівняльний (10 діб) та основний (200 діб). У зрівняльний період раціон годівлі був однаковим для риб контрольної та експериментальних груп. В основний період рівень протеїну в експериментальних комбікормах для різних піддослідних груп форелі коливався від 44 до 52 % у 1 кг. Встановлено, що різні рівні протеїнового живлення райдужної форелі не спричиняють суттєвих змін у морфологічному складі крові. Доведено, що вирощування форелі на комбікормах із вмістом сирого протеїну 50 % та 52 % сприяє збільшенню в крові кількості еритроцитів, гемоглобіну, лейкоцитів та фагоцитарної активності. Використання в годівлі риб раціонів зі зниженим вмістом сирого протеїну (44 % та 46 %) призводить до зменшення морфологічних компонентів крові. У результаті досліджень фагоцитарної активності нейтрофілів у крові риб виявлено незначне їх зростання під дією досліджуваного фактора. Аналізом отриманих результатів встановлено, що зі збільшенням рівнів сирого протеїну в раціонах райдужної форелі, концентрація загального білку та його фракцій в сироватці крові підвищувалась. Відповідно, кількість загального білку в крові риб контрольної групи становила 60,0 г/л, у дослідних груп – була в межах 59,6-65,3 г/л. Підвищення рівня протеїнового живлення райдужної форелі 4 та 5 дослідних груп викликало зростання кількості альбумінів, β-глобулінів та γ-глобулінів, порівняно з аналогічними показниками риб контрольної групи. У результаті проведених експериментальних досліджень встановлено підвищення мінерального статусу крові райдужної форелі за її годівлі комбікормами з вмістом сирого протеїну на рівні 50-52 %. Під час оцінки показників білої крові форелі відмінностей в кількості лейкоцитів не виявлено. Врахування отриманих результатів під час виробництва комбікормів дозволить позитивно вплинути на гематологічні показники товарної райдужної форелі

Ключові слова: товарна райдужна форель, годівля риб, комбікорми, протеїн, гематологічні показники, лейкоцитарна формула, біохімічні показники крові

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Impact of bior and butofanon some parameters of lipid metabolism in adult quail placed in reconditioning

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Abstract. Taking into account the current trends in the development of the most developed branch of animal husbandry – poultry farming, it should be noted that the use of ecologically clean biological preparations based on plant raw materials is a relevant topic for research in the cultivation of various types of poultry, in particular quail. The purpose of the study is to analyze the effect of the BioR preparation, made according to the original technology from the biomass of cyanobacteria Spirulina platensis, on the physiological and metabolic status of groups of quail that are subject to recovery. The biological material was 150 quail at the end of the laying cycle divided into 3 batches of 50 birds. The tested preparation was administered intramuscularly to the quails two times consecutively at the onset of the study and secondly 14 days after the first administration at a dose of 0.5 ml/head. In another experimental batch, the commercial product Butofan was administered to compare the obtained results. Birds of the control group received 0.5 ml of the solution of NaCl of 0.9% in both terms. The birds included in the study were analogous in terms of race, age, body weight, and physiological status. During all time of the study, quails were monitored: clinical parameters, body weight, and the number of eggs. In addition, for laboratory investigations, blood was collected from five quails at the start of the study before the tested preparations were administered and then 2 times during the study. It has been established that the tested remedies do not cause adverse reactions or deviations in quails' health. Moreover, BioR showed adaptive properties, reflected in body temperature lower by 0.32°C than the control group and 0.18°C than the group treated with Butofan. Similar manifestations were also found in birds' breathing. Biochemical research performed on quails that have benefited from the BioR remedy reveals a true decrease in total lipids in one investigation and an increase in triglycerides at this stage and their decrease at the end of the study. Additionally, both BioR and Butofan remedies induced an increase in the blood serum of ß-lipoproteins at both stages of research, while cholesterol values did not show any essential changes. In conclusion, we can state that the BioR product has a beneficial effect on quail, including lipid metabolism. It has been proven that due to the use of the drug, a pronounced normalizing effect on the lipid metabolism of the experimental group of quails was obtained, due to a decrease in total lipids, which caused a slight increase in cholesterol in the blood of birds

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Keywords: Quails, BioR remedy, lipid metabolism, physiological status, reconditioning

Introduction

Aviculture is a relatively new branch in zootechnics, and in the national economy with many directions of activity focused on obtaining hybrids with high genetic-productive potential, offering birds optimal terms of growth and exploitation, feeding properly to the species, age, and category of birds. Despite the decline of livestock in the Republic of Moldova, aviculture in the past twenty years reanimated and even extended particularly great [1].

To be specified the fact that extension of this branch occurred not only as a result of scientifically-technical progress but also due to the gustative and dietetic qualities of poultry products (meat, eggs), to their accessibility to socially vulnerable classes [2]. At the same time, another part of this problem can be focused on the short reproduction time and bird breeding, accessibility, and their morph-physiological possibility to be elevated in industrial conditions of poultry factories [3], [4]. More than that, contrary to other branches of zootechnics, aviculture deals with the growth and exploitation of many species of birds. However, the approach to the growth and exploitation of birds is different from one species to another, depending on the knowledge level, their adaptability, and economic development on one hand and microclimate conditions for different species of birds on the other hand [5].

Currently, priority is given to medicinal products of natural origin, especially vegetables. Thus, these requirements are current and necessary for zootechnics specialists, and animal owners [6], [7], [8]. At the same time, it is worth mentioning that a medicinal remedy from Spirulina platensis – BioR was developed in Moldova [9]. This remedy is of plant origin and corresponds to all requirements submitted to drugs, both for human and animal use, being successfully tested on humans and animals [10], [11]. It is therefore of interest to study this BioR preparation in parallel with another known drug remedy – Butofan.

Material and Methods

In the study, the biological material was represented by 150 quail at the end of the laying cycle divided into 3 lots of 50 birds. The test remedy was administered intramuscularly to the quails two times consecutively at the onset of the study and secondly 14 days after the first administration at a dose of 0.5 ml /head. Another experimental lot was administered with the alternative product Butofan to compare the results obtained. Birds of the control group received 0.5 ml of sol. NaCl of 0.9%. The pattern of administration of the test preparations is shown in Table 1.

Table 1. *Scheme of administration of BioR and Butofan preparations to adult quail placed in reconditioning*

Group of Animals	Nr. Of Birds	Dose and time of administration, ml/head		Mode of administration
		1st administration (at the onset of the study)	2nd administration (on the 14th day after the 1st administration)	
CG	50	0.5 ml 0.9% sol. NaC1	0.5ml 0.9% sol. NaC1	intramuscularly
EG-I	50	0.5 ml BioR	0.5ml Bi0R	
EG-2	50	0.2 ml Butofan	0.2 ml Butofan	

The quails included in the study were similar in terms of body weight, age, and physiological status and were in identical conditions on feeding and maintenance, microclimate, and veterinary services. Birds during the study were continuously monitored and clinically investigated (individual weighing was performed periodically with an interval of 10-14 days, and temperature and respiratory movements were measured per minute).

Research Results

Following investigations over 50 days, the drugs tested, both BioR and Butofan, were well tolerated by adult quails, with no adverse reactions at the

injection site of the studied drugs in the pectoral muscles and also in the whole body. Similar series of studies was described in scientific works [12], [13], but they were conducted for only 30 days with the use of other types of drugs, the action of which, in Shasha's opinion, could cause side effects and side reactions at the place of administration of the investigated drug. The authors of scientific papers [14], [15] established that the tested means used in conducting similar experiments caused adverse reactions and abnormalities in the health of quails and guinea fowls.

It has been established that the tested remedies do not cause adverse reactions or deviations in quails' health. Moreover, BioR showed adaptive properties, reflected in body temperature lower by 0.32°C than the control group and 0.18°C than the group treated with Butofan. Similar manifestations were also found in birds' breathing.

Of particular importance are the lipids and the nature of their metabolism in the study of the functional status of the body, individual cells, and subcellular elements, as well as in the evaluation of the quality of products obtained from agricultural birds. The results of the influence of these remedies on the main indicators of lipid metabolism are presented in Table 2.

The data from table 2 show that the total lipid content in the blood serum for quail in the control group in the first study period was 12.5% higher than at the beginning of the experiment. Using the test formulations, the analysis showed that the best results of the total lipid versus control were significantly reduced in EG-I treated with BioR by 37.9% ($p<0.01$) and EG-2 with Butofan, with 31.1% ($p<0.05$) lower than in control. At the end of reconditioning, this indicator in the control group was significantly lower by 1,6 than in the previous study ($p<0.05$). These results repeat the late dynamics set in the first study in the experimental groups, confirming the validity of these findings and the analogy of the quail groups used. Indicators at the end of studies in experimental groups confirm this hypothesis when the total lipid content is close to the same level, which allows us to talk about the adaptogenic properties of both remedies studied.

Table 2. *Influence of tested remedies on indicators of lipid metabolism at adult quails*

Indicators	Fon	Group of animals		
		CG	EG-I	EG-2
Total lipids, g/l 1st administration 2nd administration	791.84±75.05	890.51±99.30 566.08±5.06*	553.27±12.53** 593.22±10.20	595.48±50.79* 596.98±3.67
Cholesterol, mmol/l 1stadministration 2nd administration	5.72±0.16	5.68±0.05 5.46±0.06	5.67±0.04 5.59±0.16	5.50±0.04 5.85±0.19
Triglycerides, mmol/l 1 st administration 2nd administration	3.49±0.31	2.92±0..28 3.10±0.47	3.75±0.43 2.89±0.15	3.12±0.16 2.86±0.34
ß-lipoproteins, g/l 1 st administration 2nd administration	116.14±4.60	88.78±8.72* 91.74±7.06	99.60±8.07 107.28±3.31	124.66±10.61* 100.30±5.09

*NB: * $p<0.05$;** $p<0.01$*

According to table 2 in quail serum in the first stage, the cholesterol index was almost at the same level. The difference of this indicator was also insignificant at the end of the experiment, with a slight increase of 2.4-7.1% compared to the control.

The data in table 2 indicates a low triglyceride content in the serum in the control group, which was 16.3% lower compared to the font. At the same time, quails in the experimental groups recorded an insignificant increase of this indicator by 6.8-28.4%, compared to the control group. In addition, at the end of the study, on the contrary, there was an increase in the triglyceride content in the control group, which was 6.2% compared to the first stage of the experiment. At the same time, it is attested that only in the experimental groups the difference of this indicator decreased compared to the control group by 6.8-7.7%. From table 2, it can

be seen that, before starting the study, the medium ß-lipoprotein content was 116.14±4.60 g/l, and the level at the control group at the first stage of the studies was 23.6% lower compared to the baseline data (p<0.05). During the study, there was an increase in ß-lipoprotein content in the blood serum: in the first stage – by 12.2-40.4% (p<0.05, for EG-2) and in the final stage of research with 9.3-16.9%, respectively in both experimental groups compared to the control group.

The study shows that the BioR remedy obtained by advanced Spirulina platensis technology, administered twice a year, to adult quail at the end of the laying cycle, during the study, has a good local and general tolerance. In addition, the tested remedies are moderately involved in improving lipid metabolism in adult quail placed on reconditioning, which is also confirmed by better productive parameters in birds treated with biologically active remedies, especially BioR.

Conclusions

Based on experimental data on the use of BioR, it has been established that this remedy has a pronounced normalization effect on lipid metabolism. This reflected a decrease in total lipids and contributed to an insignificant increase in blood cholesterol in quail during the research, and also contributed to increased serum triglycerides at the first stage and an insignificant increase in beta-lipoproteins, suggesting that the tested remedy improves lipid metabolism in quail.

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Вплив препаратів BioR та Butofanon на ліпідний обмін дорослих перепелів на відгодівлі

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Анотація. Враховуючи сучасні тенденції розвитку найбільш розвиненої галузі тваринництва – птахівництва, слід зазначити, що використання екологічно чистих біопрепаратів на основі рослинної сировини є актуально тематикою для дослідження при вирощуванні різних видів птиці, зокрема і перепелів. Метою дослідження є аналіз впливу препарату БіоР виготовленого за оригінальною технології з біомаси ціанобактерій Spirulina platensis на фізіолого-метаболічний статус у груп перепелів, що підлягають відновленню. Біологічний матеріал складався зі 150 перепелів, взятих в кінці циклу яйцекладки, розділених на 3 групи по 50 голів у кожній. Досліджуваний препарат вводили перепелам внутрішньом'язово два рази поспіль на початку дослідження і вдруге через 14 днів після першого введення в дозі 0,5 мл/голову. В іншій експериментальній групі був введений комерційний продукт Бутофан для порівняння отриманих результатів. Птахи контрольної групи в обидва терміни отримували 0,5 мл розчину NaCl 0,9 %. Птахи, включені в дослідження, були аналогічними за породою, віком, масою тіла та фізіологічним статусом. Протягом всього періоду дослідження за перепелами спостерігали, відзначаючи такі показники: клінічні, масу тіла, кількість яєць. Крім того, для лабораторних досліджень кров брали у п›яти перепелів на початку дослідження перед введенням тестованих препаратів, а потім 2 рази під час дослідження. Встановлено, що випробувані засоби не викликають побічних реакцій і будь-яких відхилень у стані здоров'я перепелів. Більш того, Біор показав адаптивні властивості, що виявилось в температурі тіла нижче на 0,32 °С, ніж у контрольній групі, і на 0,18 °С, ніж в групі, що одержувала Бутофан. Подібні прояви були виявлені і в диханні птахів. Біохімічні дослідження перепелів, яким вводився препарат Біор, виявили справжнє зниження загальної кількості ліпідів в даному дослідженні і підвищення тригліцеридів на цьому етапі і їх зниження в кінці дослідження. Крім того, препарати БіоР і Бутофан викликали підвищення рівня ß-ліпопротеїнів в сироватці крові на обох етапах дослідження, в той час, як значення холестерину не показали будь-яких істотних змін. Продукт БіоР сприятливо впливає на перепелів, в тому числі на їх ліпідний обмін. Доведено, що за рахунок застосування препарату отримано виражену нормалізуючу дію на ліпідний обмін дослідної групи перепілок, за рахунок зменшення загальних ліпідів, що спричинило незначне підвищення холестерину в крові птахів

Ключові слова: перепели, препарат Біор, ліпідний обмін, фізіологічний сатус, рекондиціонування

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Improvement of the technology of functional pre-made meat products in a dough shell

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Abstract. In modern conditions of increasing globalization and intensifying competition in the food industry in general and in the meat processing industry in particular, there is a need to find new ways to increase the competitiveness of enterprises, guarantee the quality and safety of products, ensure proper working conditions for personnel, and reduce the negative impact on the environment. The research aims to improve the technology of pre-made products in dough shell production and to determine the rational parameters of the production process by introducing new ingredients into the recipe. The content of toxic elements was determined based on the Ukrainian Laboratory of Quality and Safety of Agricultural Products of the National University of Life and Environmental Sciences of Ukraine. The main stages of production of pre-made products in dough shells were analyzed, the technology was improved considering a set of safety and quality studies, risks and critical control points, sources of their occurrence were identified, and preventive actions were developed. The amino acid composition of the protein component and functional and technological quality indicators of the minced meat samples (moisture retention capacity, emulsifying capacity, and stability of meat emulsions) were evaluated. To determine the changes that occurred in the experimental prepared products enriched with vegetable components, a study of changes in their physicochemical properties after freezing and thawing was conducted. The research results showed that in the experimental sample containing the food additive Elamin, the amount of bound moisture in meat systems gradually decreases during storage, but the mass fraction of the concentrate in the amount of 0.3 kg per 100 kg of raw material allows for retaining free moisture. Based on the research results, the technology of functional pre-made meat products in a dough shell was improved, and technical specifications and technological instructions state standard 10.1-00493706-075:2019 “Frozen semi-finished products in a dough shell “Healthy dumplings” were developed for implementation in production

Keywords: minced meat, elamine, wheat fiber, soy protein, recipe, technology, functional product

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Relevance

The progressive deterioration of the environment results in the contamination of traditional food products with unusual pollutants, which traditionally include chemical plant protection products, mineral fertilizers, pesticides, etc. On the other hand, the modern diet lacks essential micronutrients, vitamins, and other biologically active substances, which has led to an increase in the incidence of chronic diseases not only of the gastrointestinal tract but also of cancer and cardiac diseases (Bal-Prylypko, Palamarchuk, and Nikolaenko, 2019; Balogun *et al*., 2018). Given the development of negative trends, the direction of modifying the composition of products with components that help reduce the deficiency of biologically active substances in the human body has become widespread (Bu *et al*., 2020; Cherednichenko *et al*., 2021). Currently, further research on the development of new formulations and improvement of technologies for products known as functional foods, which are intended for systematic consumption by all groups of the healthy population, remains relevant.

Analysis of Recent Research and Publications

The technologies for creating functional foods are based on the modification of traditional foods, which increases the content of useful ingredients in them to a level comparable to the physiological norms of their consumption, which, according to various estimates, is 10-50% of the average daily requirement (Cherednichenko and Bal-Prylypko, 2019).

Ukraine has several deficient substances that need to be included in the daily diet of the population. In particular, most regions of Ukraine, especially Polissia and the Carpathian mountain zone, are characterized by iodine deficiency (Nikolaienko and Bal-Prylypko, 2020; Ren *et al*., 2022). The daily requirement for this micronutrient is highlighted in the World Health Organization (WHO) recommendations (Table 1).

Table 1. *Iodine intake (micrograms per day)*

#	Population group	Norm
1	Children to 5 years	90
2	Children from 6 to 12 years	120
3	Adolescents and adults	150
4	Pregnant women	250

One of the ways to eliminate iodine deficiency is to include seafood in the daily diet, particularly cod liver (iodine content 3700 mcg/kg), salmon (2000 mcg/kg), hake (1600 mcg/kg), tuna (500 mcg/kg) (Dang *et al*., 2020). The current level of consumption of these products remains low, so for the majority of the population, the most affordable way to supplement traditional foods with iodine compounds, namely potassium iodate KJO_3, is the most affordable way recommended by WHO (Cherednichenko & Bal-Prylypko, 2020; Rahmasari & Yemiş, 2022).

It is possible to solve the problem of iodine deficiency by adding sea salt and seaweed to food, which contains a significant amount of iodine in an organically bound form favorable for absorption. The most affordable and economically feasible is the introduction into the formulation of traditional products of the food additive “Elamin”, obtained by processing the brown seaweed *Laminaria*, which is used as a dietary supplement for adults and children (Wang *et al*., 2019; Felici *et al*., 2020). The iodine it contains, due to its organic form, is

absorbed by 90-95%, and to compensate for its lack, it is enough to introduce about 0.1 grams of the drug into the daily diet. In addition to iodine, the supplement contains 0.01-0.02% of minerals (K, Na, Ca, Co, Mg, Fe, Zn, S, N, P, Cl, Cu, Ag, Al, Cr, Mn, B, Br) and vitamins (A, B, D, E). Alginic acid salts, which are part of the "Elamin" dietary supplement, are unique natural sorbents that selectively bind radionuclides, heavy metal salts, and toxic substances and remove them from the body (Stadnyk *et al*., 2020).

The research aims to improve the technology of functional semi-finished meat products in a dough shell by introducing iodine compounds, which are characterized by a deficiency in certain regions of Ukraine, into the recipe, as well as to bring the amino acid composition of the protein component of the product to the WHO-approved standards of ideal protein.

To achieve the aim, the following scientific objectives were defined: based on the analysis of WHO recommendations and current research, to identify promising areas for eliminating iodine deficiency in Ukraine; to develop recipes for minced meat for semi-finished products in a dough shell enriched with iodine compounds; to determine changes in the functional and technological properties of the experimental samples of minced meat compared to the control; to investigate the organoleptic characteristics, chemical composition and biological value of semi-finished products in a dough shell made with minced meat according to new recipes.

Materials and Methods

The research object was liver-based minced meat and pre-made products made from them in a dough shell. Minced meat according to Ukraine state standard 6028:2008 was chosen as a control. The formulation of the test samples included the food additive "Elamin" under technical standard 00382119-02-99, as well as wheat fiber according to technical standard 21586560.001.99 (sample 1) and soy protein according to Ukrainian state standard 4595:2006 (sample 2).

Experimental research was carried out using the methods of physicochemical, organoleptic, chemical, and biochemical research. Functional and technological parameters of minced meat (moisture retention, emulsifying ability, emulsion stability) were determined by centrifugation (Kumar *et al*., 2021; Kim *et al*., 2020). The organoleptic evaluation of semi-finished products was carried out on a five-point scale under Ukraine state standard 4823.2:2007, based on expert evaluation at the Department of Meat, Fish and Seafood Technology of the National University of Life and Environmental Sciences of Ukraine. The mass fraction of ash was determined by the weight method, after mineralization of the product in a muffle furnace at a temperature of 500-600°C based on Ukraine state standard ISO 936:2008. The mass fraction of protein was determined under state standard 25011-81 by the Kjeldahl method. The mass fraction of total fat content was determined by the Soxhlet method, based on the change in sample weight after fat extraction with a solvent under the Ukraine state standard 8380:2015. The amino acid composition of proteins was determined by ion-exchange chromatography, samples were prepared by acid hydrolysis, and free amino acids were extracted with dilute hydrochloric acid, precipitated with sulfosalicylic acid, and separated by filtration.

Results and Discussion

To enrich the pre-made products in the dough casing with iodine, the food additive "Elamin" was introduced into the recipe of the experimental samples. The formulations of minced meat pre-made products in the dough are given in Table 2.

Table 2. *Recipes for minced meat pre-made products in dough, kg*

Raw material	Receipt		
	Control	Sample 1	Sample 2
For minced mead, 100 kg of unsalted material			
Fried liver	65.0	59.7	59.7
Butter	5.0	5.0	5.0
Hydrated wheat fiber	-	5.0	-
Hydrated soy protein	-	-	5.0
Boiled buckwheat	16.0	16.0	16.0
Chicken eggs or egg melange	4.0	4.0	4.0
Fresh chopped onion	10.0	10.0	10.0
Elamine hydrated	-	0.3	0.3
Total	100	100	100
Spices and materials per 100 kg of minced meat			
Rock salt	1.2	-	-
Sea salt	-	1.1	-
Rock salt withlow natrium	-	-	1.1
Ground red pepper	0.05	0.05	0.05
Ground black or white pepper	0.2	0.2	0.2
Ground allspice pepper	0.1	0.1	0.1
Ground coriander	0.2	0.2	0.2

In the experimental samples per 100 kg of minced meat, the liver content was reduced by 5.3 kg compared to the control by adding 0.3 kg of the hydrated food additive "Elamin" to their formulations, and 5.0 kg of hydrated soy protein was added to sample 1, and hydrated wheat fiber to sample 2 (1:2 hydration ratio, without aging). In the formulation of the prototypes, table salt was replaced with sea salt (sample 1) and with table salt with a reduced sodium content (sample 2).

The next step in the research was to study changes in the functional and technological properties of minced meat. The ability of minced meat to retain moisture, its emulsifying ability, and the stability of the resulting emulsions were determined. Replacing meat raw materials with fiber or soy protein leads to a significant increase in the amount of moisture retained by minced meat. This is also facilitated by the replacement of table salt (sodium chloride) used for salting with food-grade salts characterized by a reduced sodium content and enriched with hydrophilic magnesium and potassium chlorides (Fig. 1). Minced meat modified with vegetable additives showed an increased ability to form emulsions (Fig. 2). Emulsified minced meat modified with additives is also characterized by increased stability (Fig. 3).

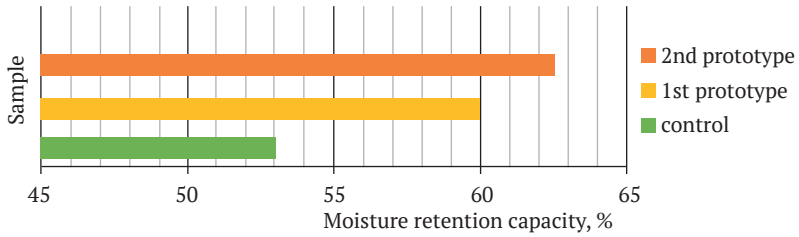

Figure 1. Moisture retention capacity of minced meat

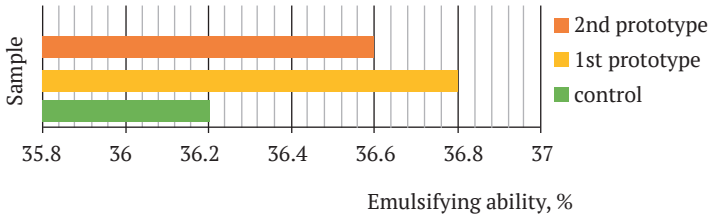

Figure 2. The emulsifying ability of minced meat

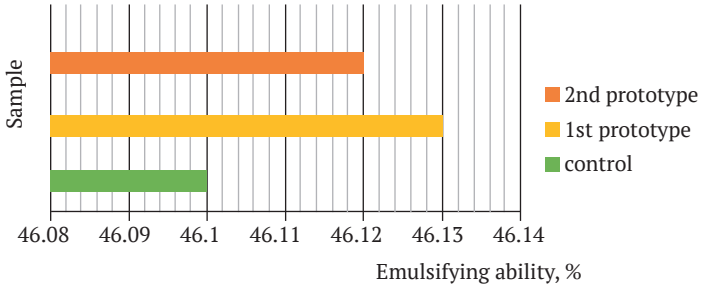

Figure 3. Stability of minced meat emulsions

Based on the studied samples of minced meat, semi-finished products in a dough shell "Healthy dumplings" were made. The study of the organoleptic characteristics of semi-finished products revealed that partial replacement of the liver with additives of plant origin in combination with the use of the food additive "Elamin" and a decrease in the amount of sodium cation leads to an improvement in the consistency of the product and has virtually no effect on the appearance and aroma of the product (Table 3).

Table 3. *Organoleptic evaluation of quality indicators of meat semi-finished products in the dough shell "Healthy dumplings"*

Samples	Appearance	Appearance and color, cut	Aroma	Taste	Consistency	Moisture	Overall marks	Overall mark
Control	4.9	4.9	5	4.8	4	4.7	28.3	Very good
1-prototype	4.9	4.9	4.9	4.5	4.9	4.9	29	Very good
2-prototype	5	5	5	4.7	4.9	4.9	29.5	Excellent

The highest overall score of organoleptic quality indicators on a 5-point scale was obtained for minced meat with the replacement of 5% liver with hydrated soy protein – the 2nd experimental sample (Fig. 4).

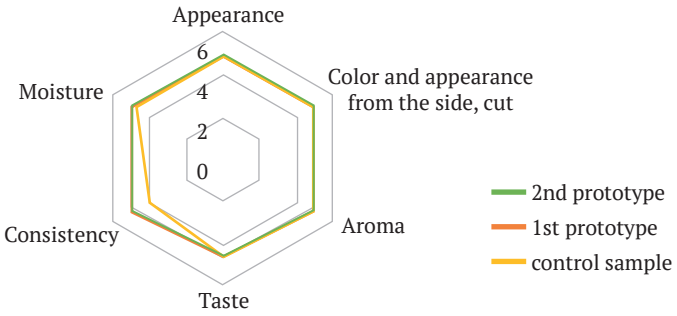

Figure 4. Diagram of quality indicators of pre-made products in dough shell "Healthy dumplings"

In the development of new types of functional meat products, the chemical composition of the experimental samples and their comparison with the control samples is of key importance. The results of studying the general chemical composition of minced meat for pre-made products in a dough shell are shown in Table 4.

Table 4. *Chemical composition of minced meat, %*

Component	Samples		
	Control	1-prototype	2-prototype
Moisture	55.00±2.44	58.21±3.12	59.15±3.44
Protein	10.90±1.02	11.34±2.12	11.63±2.17
Fat	26.51±2.78	24.82±1.98	24.5±2.04
Ash	2.20±0.24	3.16±0.44	3.08±0.41

According to the results of chemical composition studies, an increase in the content of minerals in the experimental samples was noted, and their mass fraction increased by 1.3-1.5 times compared to the control. This fact is explained by the higher content of micro- and macroelements in the used additives compared to meat raw materials.

The results of the study of the amino acid composition of proteins of the control and experimental samples of pre-made products in the dough shell are shown in Table 5.

Table 5. *Amino acid composition of proteins, mg/100 g of product*

Attributes	Samples		
	control	1-prototype	2-prototype
Essential amino acids:			
Isoleucine	518.63	492.18	522.71
Leucine	892.48	891.56	902.08
Sum of methionine and cysteine	412.83	409.51	446.37
Lysine	780.12	741.05	805.7
Sum of phenylalanine and tyrosine	874.86	878.63	940.78
Threonine	458.12	431.45	466.71
Tryptophan	143.33	128.58	139.64
Valine	670.54	647.96	698.66
Nonessential amino acids:			
Alanine	556.28	544.44	579.76
Arginine	677.86	678.57	717.64

Table 5. Continued

Attributes	Samples		
	control	1-prototype	2-prototype
Aspartic acid	776.51	763.77	806.93
Histidine	427.15	422.75	456.42
Glycine	486.58	770.63	514.36
Glutamic acid	1198.87	1171.19	1169.53
Proline	551.73	514.8	549.86
Serine	409.1	397.53	417.71
The total amount of amino acids	9834.9	9584.6	10134.86

The data obtained demonstrate that the qualitative composition of protein substances changes significantly in the experimental dumplings samples compared to the control sample. This indicates that the replacement of the main raw material (liver) with soy protein (2nd experimental sample) contributes to an increase in the number of amino acids in semi-finished products. An important indicator that characterizes the biological value of a protein is its compliance with the ideal protein. The correspondence of the amino acid content to the ideal protein is shown in Table 6.

Table 6. *Compliance with the ideal protein amino acid composition of the protein component of functional meat pre-made products*

Names of essential amino acids	The amino acid content in the protein component of minced meat, g/100 g of protein			
	ideal protein	control	1-prototype	2-prototype
Valine	5.0	5.91	5.57	6.41
Isoleucine	4.0	4.57	4.23	4.80
Leucine	7.0	7.87	7.67	8.28
Lysine	5.5	6.88	6.37	7.39
Methionine + cystine	3.5	3.64	3.52	4.10
Threonine	4.0	4.04	3.71	4.28
Tryptophan	1.0	1.26	1.11	1.28
Phenylalanine + tyrosine	6.0	7.72	7.55	8.63
Amount of amino acids	36	41.89	39.73	45.17

The table shows that the content of each essential acid and their total amount in the control, 1st experimental, and 2nd experimental samples exceeds their amount in the ideal protein and is 41.89 g/100 g of protein, 39.73 g/100 g of protein and 45.17 g/100 g of protein, respectively. The results obtained indicate the absence of limiting amino acids and the high biological value of the product.

Conclusions and Perspectives

Based on the results of complex research, the technology of functional pre-made meat products in a dough shell has been improved. It has been shown that the prototype of minced meat No. 2 enriched with the food additive Elamin and soy protein is characterized by improved functional, technological, and organoleptic properties, an increase in the mass fraction of protein, and compliance with its amino acid composition with the ideal protein. Technical specifications and technological instructions 10.1-00493706-075:2019 "Frozen semi-finished products in a dough shell "Healthy dumplings" were developed for implementation in production. Further research will be aimed at determining the mineral composition of the dumplings to recommend them as a functional food for the prevention of iodine deficiency.

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Удосконалення технології м'ясних напівфабрикатів функціонального призначення у тістовій оболонці

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Анотація. В сучасних умовах посилення процесів глобалізації та загострення конкуренції в харчовій промисловості загалом та у м'ясопереробній зокрема виникає необхідність пошуку нових шляхів підвищення конкурентоспроможності підприємств, гарантування якості та безпечності продукції, забезпечення належних умов праці персоналу, зменшення негативного впливу на навколишнє середовище. Метою досліджень є удосконалення технології виробництва напівфабрикатів у тістовій оболонці та визначення раціональних параметрів технологічного процесу виробництва, за рахунок введення у рецептуру нових складових. Визначення вмісту токсичних елементів проводили на базі Української лабораторії якості і безпеки продукції агропромислового комплексу Національного університету біоресурсів і природокористування України. Проаналізовано основні етапи виробництва напівфабрикатів у тістовій оболонці, удосконалено технологію з урахуванням комплексу досліджень безпечності та якості, визначено ризики та критичні контрольні точки, джерела їх виникнення та розроблено попереджувальні дії. Проведена оцінка амінокислотного складу білкової компоненти та функціонально-технологічних показників якості дослідних зразків м'ясного фаршу (здатність до утримання вологи, емульгувальна здатність, стабільність м'ясних емульсій). Для визначення змін, що відбувалися з дослідними напівфабрикатами збагаченими рослинними компонентами, було проведено дослідження зміни їх фізико-хімічних властивостей після їх заморожування-розморожування. Результати досліджень показали, що у дослідному зразку з вмістом харчової добавки «Еламін», кількість зв'язаної вологи у м'ясних системах протягом зберігання поступово зменшується, але масова частка концентрату у кількості 0,3 кг на 100 кг сировини дозволяє утримати вільну вологу. На основі результатів досліджень удосконалена технологія м'ясних напівфабрикатів функціонального призначення у тістовій оболонці, а для впровадження у виробництво розроблені технічні умови і технологічна інструкція ТУ У і ТІ У 10.1-00493706-075:2019 «Напівфабрикати у тістовій оболонці заморожені «Вареники оздоровчі»

Ключові слова: м'ясний фарш, еламін, пшенична клітковина, соєвий білок, рецептура, технологія, функціональний продукт

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Genetic determination of reproduction rate traits in dairy cattle breeding

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Abstract. The relevance of the study is due to the need to control economically important attributes of the reproduction of dairy cattle, which have a weak genetic determination and substantially depend on the management and feeding of cows. The purpose of the study is to determine the relationship between the duration of the voluntary waiting period, days open, and period of insemination, assess the impact of the father factor on them. An examination of the influence of genetic and environmental factors on the voluntary waiting period, days open, and milk productivity was conducted. A new feature is highlighted – the period of insemination, which was determined as the difference between the lengthening of the open day period and the voluntary waiting period. Studies were conducted on Holstein cows (a total of 605 heads), the average yield of which was 8777 kg. As a source of primary information, data from the Uniform Agri programme was used, which was processed using variance and correlation-regression analysis. The probable influence of the father factor on milk during 305 days of lactation (11.5%), days open (5.4%), and the period of insemination (5.2%) was revealed. A positive relationship has been established between milk yield and days open (0.115, $p \leqslant 0.01$), milk yield and period of insemination (0.165, $p \leqslant 0.01$), days open and voluntary waiting period (0.257, $p \leqslant 0.01$), days open and period of insemination (0.955, $p \leqslant 0.01$). The dependence of the duration of days open and the voluntary waiting period on such paratypical factors as the age of cows and the month of their calving was examined. There is a tendency to reduce the duration of days open and the voluntary waiting period in cows with each subsequent lactation. It was identified that the duration of the voluntary waiting period was the greatest in cows that calved in November, March, and May, and the smallest – in August. Cows that calved in March had a longer duration of days open, and with calving in February – the shortest. Conclusions are drawn about the possibility of using the value of the period of insemination in selection programmes. The possibility of using reproduction level attributes in selection programmes is proved

Keywords: reproduction rate, period of insemination, milk production, voluntary waiting period, days open, Holstein breed

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Relevance

In the practice of breeding improvement of farm animals, there is a tendency to expand controlled traits for their further use in selection programmes. On the one hand, this is caused by the need to control new economically important attributes, and on the other – by the possibilities of genomics (Ruban *et al.*, 2019; Fedota *et al.*, 2018; Cole *et al.*, 2012), when selection based on genetic markers requires additional monitoring of changes in the phenotypic manifestation (Ruban *et al.*, 2016; Fedota *et al.*, 2016). Recently, this has led to the emergence of a new research area called phenomenon (Bilder *et al.*, 2009; Houle *et al.*, 2010). The term phenomenon reveals the need for more detailed (more regular in time) control of breeding traits (Greenwood *et al.*, 2016; Koltes *et al.*, 2019; Ruban *et al.*, 2019), and analysis of changes in these attributes reveals the features of the genotype-environment interaction (Ruban, 1999; Ruban, 1987).

Reproduction rates are known to have a substantial impact on the economy of production (Cole *et al.*, 2020), but in breeding, these traits have a weak genetic determination and are more dependent on livestock management and feeding (Ali *et al.*, 2019; Brzáková *et al.*, 2019). So the vast majority of successful fertilisation of a cow depends on the professionalism of the artificial insemination technician and the veterinarian, who monitor the state of recovery processes in the period after calving. The system for monitoring the state of reproduction of cows additionally includes such a feature as the waiting period (Mitioglo *et al.*, 2017), which allowed testing the working hypothesis regarding the possibilities of using this indicator in the breeding process.

Analysis of recent studies and papers

A voluntary waiting period or VWP is defined as the interval from the date of calving to the date of the first insemination of a cow, made by a specialist based on an assessment of the state of the cow›s reproductive organs, or other management decisions (Fleming *et al.*, 2019). The duration of this period is substantially influenced by the artificial insemination technician or veterinarian, based on an assessment of the general condition and a thorough examination of the cow›s reproductive organs (Miller *et al.*, 2007). VWP is influenced by the specific features of calving planning, the use of sexual heat synchronisation schemes, and most importantly, the qualification of veterinarians and artificial insemination technicians, so the value of this period varies greatly between farms and animals of the same herd (Fleming *et al.*, 2019).

The most common reasons for changing the duration of this period were the condition of cows after calving – 50%, the calving season – 18%, the productivity level – 18%, the calving number – 14%, and other reasons – 14% (Dejarnette *et al.*, 2007)

In the conditions of Dutch farms, the optimal VWP was 6 weeks. The average economic losses from extending the VWP for more than 6 weeks were 9, 26, and 52 euros for the duration of the waiting period of 9, 12, and 15 weeks, respectively (Inchaisri *et al.*, 2011). In the practice of North American dairy farms, VWP within 60 days (8.5 weeks) is considered normal (Miller *et al.*, 2007; Stangaferro *et al.*, 2018).

In tropical climates, the calving season has a substantial impact on VWP values (Silva *et al.*, 2017).

Relationships were found between VWP of less than 50 days, the period of days open – DO with the corresponding probability value ($p \leqslant 0.05$), and fertilisation in the first 200 days of lactation ($p \leqslant 0.01$), compared with the period of voluntary waiting that was more than 50 days (Fodor *et al*., 2018)

VWP lasting 45 to 70 days was associated with a shorter rumination time during estrus and a lower pregnancy index after the first insemination (Yazlık *et al*., 2018).

According to the data (Ali *et al*., 2019), the heritability ratio (h^2) of the DO interval in different livestock breeds was 0.09±0.121, and a substantial genetic correlation was also found between DO and milk yield during lactation (r=0.24; $p \leqslant 0,01$). This means that selection to increase milk yield affects the increase in DO duration.

In herds of the Swiss breed in the United States, the genetic correlation between DO and milk forms, DO and the height of posterior udder attachment was positive – 0.52 and 0.55, respectively, and between the duration of the service period and productive life was negative – -0.60 (Gibson & Dechow, 2018).

Purpose of the study. The purpose of the study was to determine the influence of the father factor on the duration of the voluntary waiting period (VWP), days open (DO), and period of insemination (PoI), the task was to determine the relationship of these periods with each other, and with milk productivity, assess the impact of paratypical (organised) factors on them.

Materials and Methods

The study was conducted in the conditions of an agricultural Ltd "Agroko", Chornobai district, Cherkasy region. A sample was formed for analysis.

The animals were kept using a tethered system and milked into a milk pipeline. Cows were fed a mixed diet, which was balanced in terms of energy, protein, and vitamin-mineral components.

Insemination of cows was conducted by a technique of artificial insemination by rectocervical method. Ultrasound examinations recorded the presence or absence of problems of the sexual apparatus (ovarian cyst or hypofunction, subclinical and clinical endometritis), after which a decision was made regarding the possibilities of insemination. A portable ultrasound scanner was used to evaluate the results of fertilisation, and the control was performed by a veterinarian. The results were entered into a computer herd management programme – Uniform Agri, the database of which stores information about the reproduction of herd animals (dates of calving, insemination, launch, number of the producer whose sperm was used for insemination), and signs of productivity (hopes, etc.).

Mathematical data processing was performed using IBM SPSS Statistics and using programme modules such as general linear model, graphical, correlation, and regression analysis.

The main intervals (attributes) that comprehensively characterise the complex process of cow reproduction and were used for analysis are shown in Fig. 1. For this purpose, the international terminology regarding these intervals is used, which is regulated by the International Committee for Animal Recording (ICAR). Additionally, such a feature as the period of insemination is highlighted – PoI. The PoI value was calculated for each cow as the difference between the DO and VWP values.

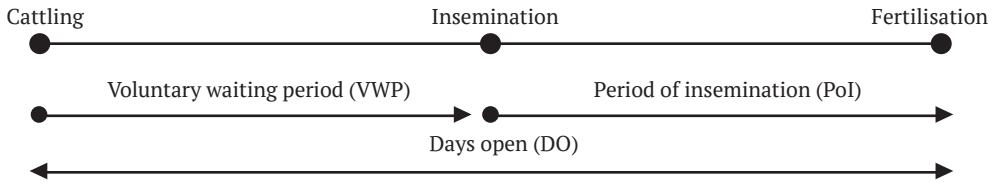

Figure 1. Attributes that characterise the level of reproduction of a cow

Paternal origin (the father factor) was used as a fixed genetic factor to calculate the genetic influence on breeding traits. The sample of cows was divided into two maximum equal groups, depending on the size of milk yields and considering the nature of the sample distribution to determine changes in the duration of the estimated periods (VWP, DO, PoI) from the level of productivity (Fig. 2). Based on the nature of the distribution, two almost equal groups were formed. Thus, cows that had a lactation yield of less than 8777 kg were included in the low-productive group (n=287), and in accordance with the milk yield of the higher number, a group of highly productive cows was formed (n=316).

Results and Discussion

The productivity of controlled cows was in the range of 4500-14000 kg of milk without the existing values of asymmetry and excess. The maximum productivity is recorded at 14 tons per cow. The sample was presented by originating from 18 breeding bulls of the Holstein breed (import of sperm from the USA, Canada).

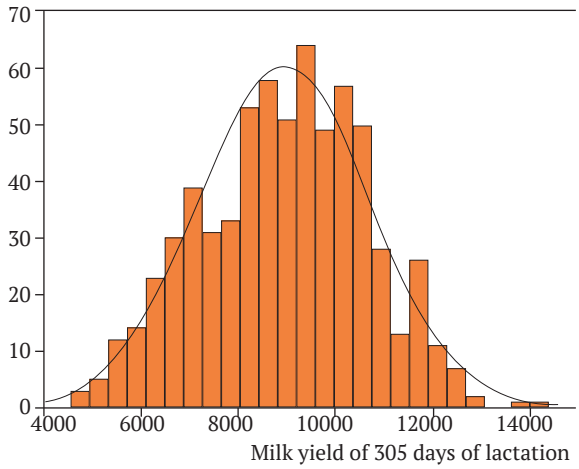

Figure 2. Value of the normal distribution of herd cows by milk yield (total 605 heads)

The highest average milk yield over 305 days of lactation was detected in Armstrong's daughters, whose productivity exceeded Audacity's offspring by 906 kg of milk and, respectively, by 755 kg compared to the herd average (Table 1). Armstrong's daughters were also characterised by the shortest DO period (98 days) and the longest voluntary waiting period (59 days). Cows offsprings of Audacity, in addition to the lower milk yield, were characterised by the longest DO value.

Table 1. *Values of productivity indicators and reproduction characteristics in daughters of different producers*

Producer	Number of cows	Milk yield for 305 days of lactation, kg	Periods, days		
			VWP	PoI	DO
		M±m	*M±m*	*M±m*	*M±m*
Matrix	24	9326±260.9*	66±3.0	52±14.5	118±14.3
He ET	76	9476±152.7***	65±3.5	78±8.9	147±16.3
Billtest	39	9254±225.9*	60±3.0	87±14.0	147±17.7
Audacity	26	8626±319.9	74±5.4	53±9.9*	152±22.4
Armstrong	26	9532±238.9***	59±2.1	38±8.9***	98±9.5**
Benjamin	68	8928±203.0	75±9.8	71±7.9	150±17.1
Other	316	8169±102.8***	78±11.0	77±4.5	154±19.2
Total	605	8777±67.8	72±8.5	72±3.1	146±17.8

Note: ** p≤0.05; ** p≤0.01; *** p≤0.001*

There was also a substantial variability between producers over the period of insemination – PoI. Thus, the average value of this trait ranged from 38 days (Armstrong's daughter) to 87 days (Billtest's daughter). This trend coincided for the daughters of these producers with the average duration of the voluntary waiting period. A longer voluntary waiting period was characterised by cows from producer Benjamin (75 days), which is 5% more than the average value of this indicator for the herd. In this regard, the degree of influence of the father factor on the main breeding characteristics was evaluated (Table 2).

Table 2. *Assessment of the influence of the "parent" factor on the evaluated attributes (n=605 heads)*

Attribute	Degree of influence	F	Probability
Milk yield of 305 days of lactation	0.115	4.212	0.000***
VWP	0.027	0.890	0.592
DO	0.054	1.852	0.017***
PoI	0.052	1.758	0.027***

Note: ****– p≤0.001*

It was identified that the influence of the father factor on the variability of the evaluated traits ranged from 2.7 to 11.5%. The probable influence of the genetic factor analysed on VWP was not confirmed, along with this, a substantial effect was determined on such signs as milk during 305 days of lactation (0.115, $p \leq 0.001$), PoI interval (0.052, $p \leq 0.001$) and DO (0.054, $p \leq 0.001$).

Analysis of correlation links between milk yield for 305 days of lactation and the period of days open was positive +0.257 ($p \leq 0.01$), which indicates a deterioration in the reproduction function with increased milk yield. Signs such as DO and PoI had a positive substantial correlation with milk yield at 305 days of lactation ($p \leq 0.01$) (Table 3).

Table 3. *Correlation coefficients between the main breeding characteristics of cows*

Paired attributes	Correlation coefficient
VWP –DO	0.257**
DO – milk yield of 305 days	0.115**
VWP – Milk yield of 305 days	-0.141**
DO – PoI	0.955**
PoI – Milk yield of 305 days	0.165**

Note: ***– p≤0.01*

With a decrease in VWP values, there is an increase in milk yield in cows (*r*=-0.141), which indicates the best recovery processes in cows in the post-calving period.

Regression analysis between the duration of the service period and milk yield during lactation showed that with an increase in milk yield by +3.38 kg, the duration of the service period increases by 1 day (Fig. 3).

Frequency distribution over the period of insemination (Fig. 4) proves that in the analysed sample, cows are most often found with a duration in the range of up to 50 days (terms of manifestation of two sexual cycles), which indicates a relatively high level of fertilisation and especially after passing the period of voluntary waiting, which was characterised by the restoration of sexual functions in cows.

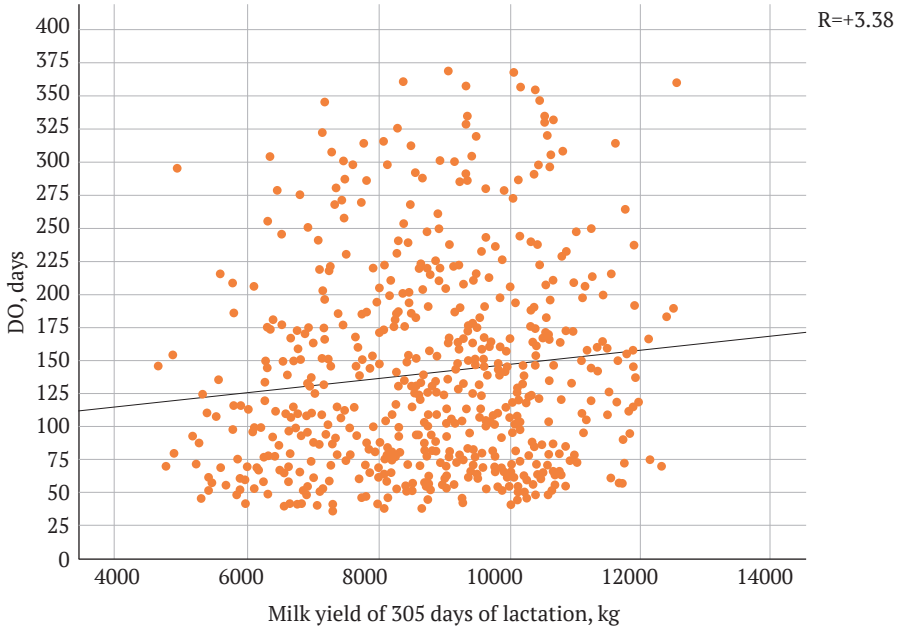

Figure 3. Distribution and regression relationship between milk yield at 305 days of lactation and the DO value in cows (*r*=+3.38)

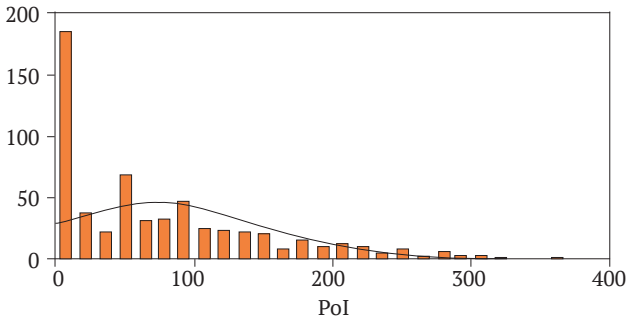

Figure 4. Distribution of herd cows by value (PoI)

Some cows were successfully fertilised in the following time periods, which is primarily due to the level of productivity and such consequences as a negative balance in live weight, the degree of fatness. So the data in Table 4 demonstrates that for the group of highly productive cows, there is a substantial difference for all periods included in the analysis (VWP, DO, PoI) compared to the low-performance group.

Table 4. *Elongation dependence of VWP, DO, and PoI depending on the performance level*

Group	Heads	Milk yield, kg	Duration, days		
			VWP	DO	PoI
Low-performance-	287	7391±59.0	70±1.30	137±4.78	66±4.46
High-performance-	316	10230±53.2	65±0.94	148±4.58	82±4.56
Difference between groups	-	-2839***	**-5.2*** **	**+11.00****	**+16.00****

Note: ***p≤0.01, ***– p≤0.001*

The possible changes in the duration of VWP, DO, and PoI periods from the cow's lactation age (Fig. 5) and the calendar month of calving, or the period of the year were analysed.

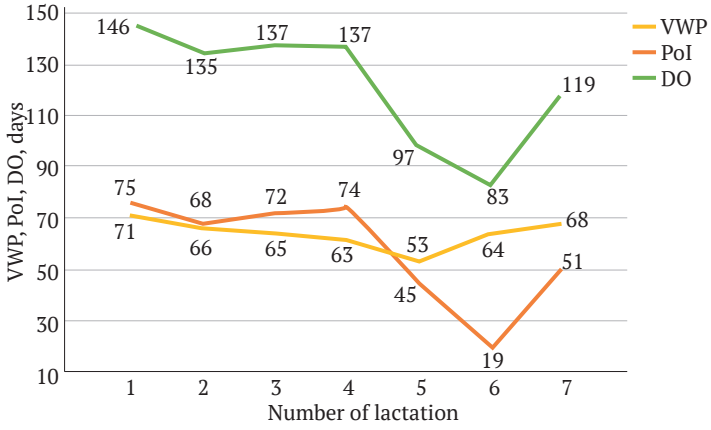

Figure 5. Dependence of DO, VWP, and PoI periods on the cow's lactation number

There is a general tendency to reduce the lengthening of periods that characterise the level of reproduction with the age of lactation (up to the sixth lactation), which indicates a better level of reproduction in full-aged cows than in first-calved and second-calved cows. The duration of the voluntary waiting period and the period of days open varied substantially depending on the month of calving of cows in the herd (Fig. 6). Thus, the longest duration of DO was in cows that calved in March, which indicates a modifying effect of environmental factors on this trait. A study by American specialists (Oseni *et al.*, 2003), which was conducted on Holstein cows in various farms, noted that cows that calved in March had the longest period of days open.

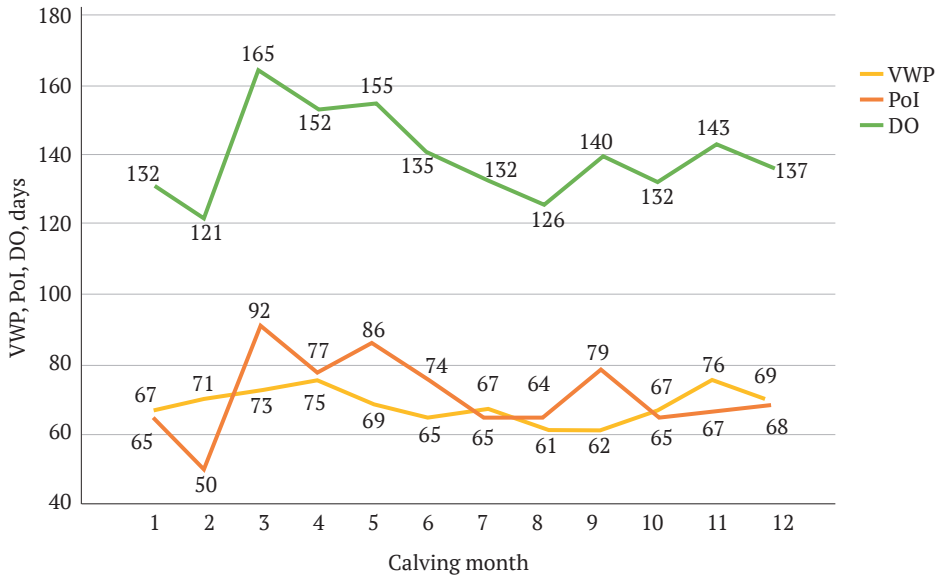

Figure 6. Dependence of DO, VWP, and PoI periods on the cow's calving month

Thus, a group of important traits was identified and a general scheme was developed for the correct assessment of the influence of genetic and non-genetic factors on the level of reproduction.

Detailed research in this area will continue if more representative samples are formed.

Conclusions and Prospects

1. A substantial influence of the father factor on such signs as milk yield of 305 days of lactation (0.115, $p \leq 0.001$), period of insemination (0.052, $p \leq 0.001$), and the period of days open (0.054, $p \leq 0.001$) was identified, which allows improving these traits by breeding.

2. Possible changes towards increasing the duration of DO and PoI with an increase in the level of productivity of cows were established.

3. The dynamics of changes in the characteristics of the level of reproduction depending on the age of cows and the month of calving is noted, which indicates the need to assess the influence of these factors when constructing a general model for assessing genetic value.

4. The substantial variability of the period of insemination among the daughters of different

producers indicates the possibility of using this indicator to correctly assess the influence of genetic and non-genetic factors, provided that more representative samples are formed.

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Генетична детермінація ознак рівня відтворення в молочному скотарстві

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Анотація. Актуальність дослідження обумовлена необхідністю контролю економічно важливих ознак відтворення молочної худоби, які мають слабку генетичну детермінацію та суттєво залежать від менеджменту і годівлі корів. Мета досліджень – визначити зв'язок між тривалістю добровільного періоду очікування, відкритих днів та періоду осіменінь, а також оцінити вплив на них фактору «батько». Проведено дослідження впливу генетичних та факторів зовнішнього середовища на добровільний період очікування, відкриті дні та молочну продуктивність. Виділено нову ознаку – період осіменінь, яка визначалась як різниця між подовженістю періоду відкритих днів та добровільного періоду очікування. Дослідження проводились на коровах голштинської породи (всього 605 гол), середній надій яких склав 8777 кг. Як джерело первинної інформації були використанні дані програми Uniform Agri, обробка яких здійснювалась з використанням дисперсійного та кореляційно-регресійного аналізу. Виявлено вірогідний вплив фактору «батько» на надій за 305 днів лактації (11,5 %), відкриті дні (5,4 %), період осіменінь (5,2 %). Встановлено позитивний зв'язок між надоєм і відкритими днями (0,115, $p \leq 0,01$), надоєм та періодом осіменінь (0,165, $p \leq 0,01$), між відкритими днями і добровільним періодом очікування (0,257, $p \leq 0,01$) та між відкритими днями і періодом осіменінь (0,955, $p \leq 0,01$). Досліджено залежність тривалості відкритих днів і добровільного періоду очікування від таких паратипових факторів, як вік корів та місяць їх отелення. Відмічена тенденція зменшення тривалості відкритих днів та добровільного періоду очікування у корів з кожною наступною лактацією. Встановлено, що тривалість добровільного періоду очікування була найбільшою у корів, які отелилися у листопаді, березні, травні, а найменшою – у серпні. Корови, які отелились в березні мали більшу тривалість відкритих днів, а з отеленням в лютому – найкоротшу. Зроблені висновки про можливості використання значення періоду осіменінь в програмах відбору. Доведена можливість використання ознак рівня відтворення в програмах відбору

Ключові слова: рівень відтворення, період осіменінь, молочна продуктивність, добровільний період очікування, відкриті дні, голштинська порода

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Research and development of a technology for the production of healthy sausages

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Abstract. Due to the general deterioration of the environmental situation and the associated increase in the negative impact on consumer health, studies have been conducted to develop a healthy sausage recipe. The research aims to develop the composition of minced meat characterized by minimal sensitivity to oxidants and balanced mineral content and implement therapeutic and preventive properties to the product made from it. The research was conducted on minced meat. The Ukrainian state standard 4436:2005 minced meat recipe was used as a sample. The values of the parameters were determined by standardized methods, and the concentration of sodium chloride was determined by the Mohr method. Organoleptic quality indicators of sausages of standardized and experimental composition were determined by the method of expert evaluation. A method of inhibiting the processes of oxidative deterioration of the meat mixture under the influence of oxygen by introducing rosemary extract into the mixture was determined. The possibility of reducing the dosage of sodium cation by replacing rock salt with sea salt enriched with kelp was determined, which also made it possible to reduce the dosage of toxic sodium nitrite. The content of beef in the meat raw materials of the prototype was reduced to 30%, and that of semi-fat pork to 26% due to the inclusion of blood plasma protein (1.0%), orange dietary fiber (0.5%), and water for their hydration (7%). In the composition of spices and auxiliary materials of the prototype, table salt (2.2%) was replaced by sea salt (2.1%), and the content of sodium nitrite was reduced from 0.0075% to 0.0050% due to the introduction of rosemary extract (0.15%) and the bacterial preparation "Iprovit LRR" (0.05%). The possibility of extending the guaranteed shelf life of sausages from 3 to 10 days was shown and the possibility of replacing rock salt with sea salt in minced meat was determined, which, while maintaining a sense of sufficient salinity of the product, allowed the reduction of the dosage of sodium cation by 30% and enrich the minced meat with trace elements – calcium, magnesium and acutely deficient iodine. The developed recipe of "Healthy" sausages can be used for industrial production at meat processing enterprises

Keywords: quality, sausages, antioxidant, rosemary, sea salt, food additives

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Introduction

The priority goal of all food industries is to ensure the high quality and safety of their products. The decrease in product safety is primarily due to the accumulation of harmful compounds in the product, which are formed during the oxidation of organic substances under the influence of external and internal ionizing radiation, as well as oxygen and peroxides that arise from its interaction with the components of the meat mixture and free radicals (highly reactive oxygen and nitrogen compounds). Given the significant share of meat products in people's diets and the short shelf life of their guaranteed shelf life, technology developers and manufacturers of innovative products are tasked with slowing down the process of product spoilage and thus extending the guaranteed shelf life without compromising consumer properties.

Literature Overview

Oxidation processes characteristic of meat systems take occur in water-rich environments with the presence of oxygen and peroxides formed with its participation, mainly hydrogen peroxide H_2O_2. In both cases, due to their instability in slightly acidic environments, atomic oxygen is formed in the product mass. With its participation, oxygen and nitrogen compounds characterized by the presence of an unpaired electron, which is highly active in chemical terms, are also formed in the product mass. Their influence on the components of meat leads to the oxidation of lipids, the main result of which is the deterioration of organoleptic quality indicators, primarily the rancidity of products and, in certain cases, the formation of hazardous substances [1].

To minimize the rate of these processes, the food industry uses several methods, including controlling the storage conditions of finished products, and careful selection of packaging methods and packaging materials. But most often, this is done by adding ingredients with antioxidant properties to the composition, which primarily interact with free radicals, characterized by the presence of unpaired electrons, and protect the components of the meat raw material from decomposition [2, 3].

To extend the guaranteed shelf life of meat products, plants and plant extracts containing so-called flavonoids (bioflavonoids) are widely used, such as parsley, garlic, buckwheat, blueberries, and other berries, bananas, citrus fruits, as well as red wine, dark chocolate (with a cocoa bean content of more than 70%), black and green tea. In general, a flavonoid molecule (a terpene compound) contains a "skeleton" of 15 carbon atoms, including two phenolic and one heterocyclic ring characterized by the presence of unsaturated chemical bonds sensitive to free radicals [4-6].

Currently, more than 5,000 compounds belonging to this class are known to be characterized by a low level of toxicity, which allows them to be introduced into everyday dietary products, including meat compositions [7-9].

The effectiveness of the combined effect of natural antioxidants and antimicrobial compounds during the cold storage of nitrite-free sausage was determined [10].

There are technologies for meat and fish products using spicy and aromatic plants or their extracts, which not only improve the flavor and aroma characteristics but also improve the quality and safety of the finished product [11].

Scientists have shown the positive effect of using spices (nutmeg, black pepper, fresh garlic, mustard powder) and their oil extracts to increase the resistance of fats to oxidation. Natural antioxidants not only increase the

nutritional and biological value but also serve as a free radical trap for fatty acids [12].

Among the flavonoid-rich antioxidant supplements of plant origin, a special place belongs to rosemary, the leaves, and stems of which contain a sufficiently large amount of acids: rosmarine, caffeic, carnosic, as well as ascorbic acid, and carotenoids that can inhibit the oxidation of fats [13, 14]. The flavonoids present in the plant are also characterized by medicinal properties, in particular, antimutagenic and antitumor effects: the introduction of 1% rosemary extract into the food of mice reduced the level of breast cancer by 47% [15]. At least 29 substances characterized by antioxidant activity (99.87% of the total extract weight) were found in rosemary processing products, including 95.1% of monoterpenes and 4.77% of sesquiterpenes: 1,8-cineole, camphor, α-pinene, β-pinene, but among these, the extract contains the highest amount of rosmarinic acid, characterized by the presence of a chemically active double bond [4, 15] (Fig. 1).

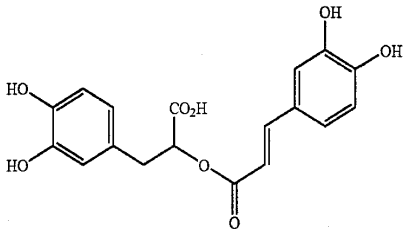

Figure 1. Rosmarinic acid

The result of adding rosemary or rosemary extract to minced meat compositions is a powerful antioxidant effect, which allows products containing lipids to be stored for a long time without spoilage.

The *research goal* is to develop a minced meat recipe with minimal sensitivity to oxidants and a balanced mineral composition and impart therapeutic and prophylactic properties to the product made from it.

To achieve this goal, the following objectives were defined: based on the results of studying the dynamics of changes in the acid and peroxide numbers of fat during storage of control and experimental samples of sausages, to determine the optimal amount of rosemary extract added to minced meat to extend the shelf life; based on the mineral composition of sea and table salt and the results of the organoleptic evaluation of the salinity level of minced meat, to determine the feasibility of replacing ordinary table salt with sea salt in experimental samples of sausages; to develop a recipe and technological conditions for the production of healthy sausages.

Material and Methods

The research was conducted under comparative conditions using sausage minced meat, the composition of which is normalized by the Ukrainian state standard 4436:2005 "Boiled sausages, sausages, bratwursts, meat bread". The prototype was additionally supplemented with porcine blood plasma protein, orange dietary fiber, rosemary extract, and a bacterial preparation containing lactic acid bacteria and microorganisms of the Staphylococcus *carnosus strain*. The difference between the experimental sample was also the replacement of ordinary rock salt with sea salt enriched with kelp. The composition of the tested samples is shown in Table 1.

Table 1. *Composition of the tested samples of minced meat, %*

The main minced meat ingredients	Control	Research
Beef	33	30
Pork half-fat	33	26
Fat pork	34	34
Plasma protein	-	1.0
Water for plasma protein hydration	-	2.0
Orange dietary fiber Citri-Fi 100	-	0.5
Water for the hydration of dietary fiber	-	6.5
Overall	100	100
Spices and materials		
Rock salt	2.2	-
Sea salt with kelp	-	2.1
Sugar	0.16	0.16
Sodium nitrite	0.0075	0.0050
The bacterial drug “Iprovit LRR”	-	0.050
Rosemary extract	-	0.15
Water for the hydration of raw meat	35.0	30.0

The meat raw materials used to make the control and experimental samples of minced meat were cut into pieces and ground in a meat grinder, salt and the calculated amounts of sodium nitrite and sugar were added. The mixtures were stirred in a mixer with a frame stirrer at a speed of 30 rpm for 10 minutes until a homogeneous consistency was achieved. The salted meat was kept for 48 hours in the refrigerator (t°=3-4°C) for maturation. The calculated amounts of water were added to the dry orange fibers and plasma protein used in the experiment (hydration of the substances was performed separately) and mixed in a mixer with a frame stirrer at a speed of 30 rpm for 10 minutes until a homogeneous consistency was achieved. To determine the optimal dosage in terms of extending the predicted guaranteed shelf life of sausages, rosemary extract was added to the experimental composition in the amount of 0.05, 0.1, 0.15, and 0.20% – by weight of meat raw materials. The rest of the ingredients in the amounts given in Table 1 were added in random order and mixed in a mixer in the same mode, the minced mixture was ground in a meat grinder repeatedly, and the calculated amount of water was added. The resulting minced meat was filled into the intestinal membranes and kept at room temperature for 3 hours before heat treatment. The casing filled with minced meat was sterilized in a thermal chamber until the temperature in the center of the minced meat reached 40-45°C, after which the frying temperature was increased until the temperature in the center of the sausage reached about 70°C. The finished product was transferred to a cooling chamber and kept at 2-5°C for 5 hours.

After aging, samples were taken from the finished products and analyzed under standard methods. The dynamics of changes in the peroxide and acid numbers of fat of the experimental sausage samples during 10-day storage were monitored in comparison with similar indicators of sausages of the composition normalized by Ukrainian state standard 4436:2005 “Boiled sausages, sausages, bratwursts, meat bread”. The

acid number of lipids was determined under the Ukrainian state standard method 4350:2004 (ISO 660:1996), the peroxide number of lipids – under Ukrainian state standard EN ISO 3960:2019, and the mass fraction of table salt - by the Mohr method according to State Standard 9957-2015. The degree of salinity and organoleptic properties of the control and experimental product samples were evaluated on a five-point scale at the Department of Meat, Fish, and Seafood Technology of the National University of Food Science and Technology of Ukraine.

Results and Discussion

The sausages produced under the methodology described above were aged for 10 days. Changes in the peroxide and acid number of fat of the products of the control and rosemary extract-modified formulations were monitored. In the first stage, the optimal amount of the additive was determined (Fig. 2), according to the results of which it was found that the optimal content of the extract additive was 0.15%. The conclusion was based on the fact that with an increase in dosage from 0% to 0.15%, the peroxide number, which characterizes the number of oxidized lipids, progressively decreased compared to the control. However, with the further addition of the extract, the peroxide number of samples with additions of 0.15% and 0.20% was practically the same. This, in our opinion, is evidence of the sufficiency of the number of flavonoids present in the sample with a concentration of 0.15%, and the inexpediency of further increasing their concentration in additives.

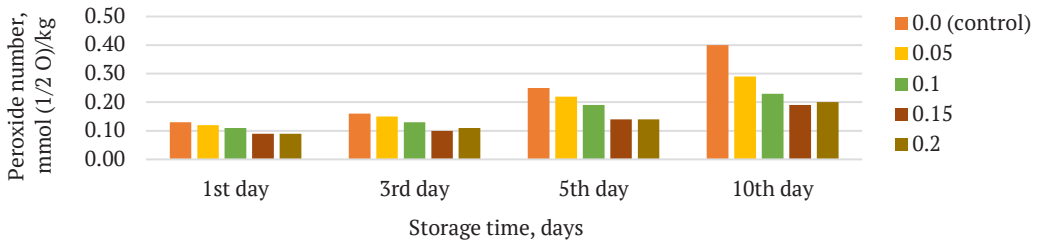

Figure 2. Dynamics of changes in the peroxide number of control and experimental samples of sausages

Considering the sufficient amount of rosemary extract added at 0.15%, the dynamics of the growth of the acid number of minced meat was studied only in the control and optimal composition according to the preliminary results (Fig. 3).

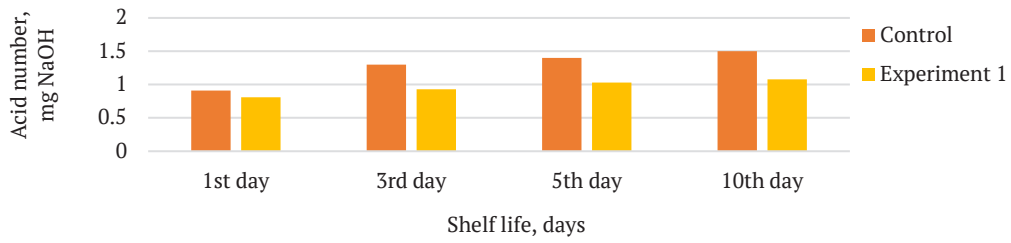

Figure 3. Dynamics of changes in the acid number of fat of control and experimental samples of sausages during storage

Here, as in the previous series of experiments, the positive role of rosemary extract was revealed: the acidity of the samples in the rosemary-modified samples increased more slowly compared to the control. It was also determined that the acidity level of the sample modified by the extract at the end of the study was even lower than that achieved in the control on the 3rd day of aging. Considering that the guaranteed shelf life of sausages with a standardized composition (control) is set at three days, we believe that the proposed modification of the sausage mince recipe allows us to count on extending the warranty period for the product, which is the subject of further research. The next step in the development of a functional product was to determine the possibility of reducing the intake of excess sodium cation, the content of which in sausage products reaches 2.5% in some cases [16], for which purpose ordinary rock salt was replaced with sea salt in the experiments. A comparison of the chemical composition of the salts used for salting (Table 2) shows that such a replacement not only reduces the amount of sodium cation added to the minced meat by about 30%, but also enriches the product with useful trace elements such as potassium, calcium, magnesium, and iodine.

Table 2. *Mineral composition of dry matter of sea and rock salt (%)*

Attribute	Sea salt	Rock salt, Ukrainian state standard 3583:2015	
		first grade	"Extra"
Sodium	30.6	38.4	38.7
Calcium	1.2	0.35	0.02
Potassium	1.1	0.1	0.008
Magnesium	3.7	0.05	0.01
Iodine	5×10^{-6}	-	-

An interesting fact was that despite the significantly lower content of sodium chloride in sea salt, the results of the organoleptic evaluation showed that the salinity level of minced meat prepared using sea salt and first-grade table salt practically did not differ (Fig. 4). On this basis, it was concluded that it is advisable to use sea salt for salting.

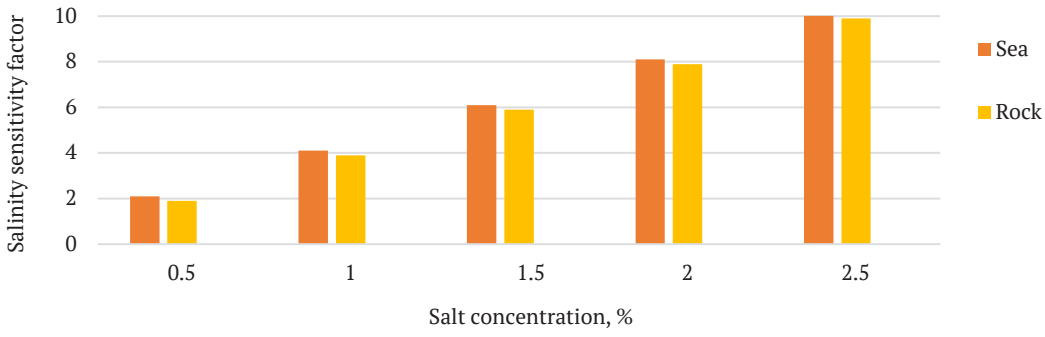

Figure 4. Results of organoleptic evaluation of the degree of salinity of minced meat mixtures

Considering the results of experimental studies, a recipe for healthy sausages and technical specifications for the product 10.1-00493706-064:2019 "Health sausages" of the Ukrainian technical specifications (UTS) were developed (Table 3).

Table 3. *Recipe for sausages "Healthy" UTS 10.1-00493706-064:2019*

Ingredient	Amount
Unsalted raw product, kg (per 100 kg)	
Grade 1 lean beef	30
Pork no-fat	26
Pork lean semi-fat	34
Plasma protein	1.0
Water for the hydration of raw ingredients	8.5
Fiber (citrus fibers Citri-Fi 100)	0.5
Total	100
Water, kg	30.0
Spices and materials, g (per 100 kg of unsalted raw materials)	
Rock salt	-
Sea salt	2100
Sugar	160
Sodium nitrite	5.0
Ground black or white pepper	160
Ground allspice pepper	100
Ground nutmeg or cardamom	50.0

The health benefits of the product are associated with several factors: 1) the product has an extended shelf life, which means a reduced amount of substances that can harm health, primarily lipid oxidation products; 2) the product has a significantly lower sodium content due to the use of sea salt and a reduced dosage of sodium nitrite in the minced meat.

Conclusions

Adding 0.15% rosemary extract to minced meat can reduce the peroxide number of fat in the meat mixture during storage for 10 days and slow down the growth rate of fatty acids in the minced meat.

Replacing rock salt, which is traditionally used for salting, with sea salt allowed us to reduce the amount of sodium cation, enrich the mineral composition, maintain a feeling of sufficient saltiness, and enhance the health benefits of the product.

The direction of further research is on the nutritional value of sausages to recommend their use in diets of therapeutic and preventive orientation.

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Обґрунтування та розроблення технології виробництва сосисок з оздоровчими властивостями

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Анотація. У зв'язку із загальним погіршенням екологічної ситуації та пов'язаним з цим посиленням негативного впливу на стан здоров'я споживачів проведені дослідження з розроблення рецептури сосисок, які характеризуються оздоровчими властивостями. Метою виконання роботи було розроблення складу м'ясної фаршевої композиції, що характеризується мінімальною чутливістю до дії окисників та збалансованим мінеральним складом і надає виробленому з неї продукту лікувально-профілактичних властивостей. Дослідження проводили на м'ясних фаршах. За контроль використано рецептуру м'ясного фаршу за ДСТУ 4436:2005. Значення показників визначали стандартизованими методами, а концентрацію хлориду натрію – методом Мора. Органолептичні показники якості сосисок стандартизованого та дослідного складу визначали методом експертної оцінки. В роботі визначено спосіб гальмування процесів окислювального псування м'ясної суміші під дією оксигену шляхом введення в суміш екстракту розмарину. Визначено можливість зменшення дозування катіону натрію заміною кам'яної солі на збагачену ламінарією морську сіль, що дозволило також зменшити дозування токсичного нітриту натрію. У складі м'ясної сировини дослідного зразка вміст яловичини було зменшено до 30 %, свинини напівжирної – до 26 % внаслідок включення до рецептури білку плазми крові (1,0 %), апельсинових харчових волокон (0,5 %), а також води для їх гідратації (7 %). У складі прянощів та допоміжних матеріалів дослідного зразка кухонну сіль (2,2 %) було замінено на сіль морську (2,1 %), а також зменшено вміст нітриту натрію з 0,0075 % до 0,0050 % внаслідок введення екстракту розмарину (0,15 %) та бактеріального препарату «Іпровіт ЛРР» (0,05 %) Контролю підлягали динаміка зміни їх пероксидного і кислотного чисел жиру. Показана можливість подовження гарантованого терміну зберігання сосисок з 3 до 10 діб і визначена можливість заміни у фарші кам'яної солі на морську, що за збереження відчуття достатності солоності продукту дозволило зменшити дозування катіону натрію на 30 % і збагатити фарш мікроелементами – кальцієм, магнієм та гостродефіцитним йодом. Розроблену рецептуру сосисок «Оздоровчі» доцільно використовувати для промислового виробництва на м'ясопереробних підприємствах

Ключові слова: якість, сосиски, антиоксидант, розмарин, морська сіль, харчові добавки

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Determination of the biological value of minced semi-finished products with a complex food additive by the enzymatic method

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Abstract. Providing the population with protein products of animal origin is becoming an increasingly difficult task at present. Therefore, the development of new meat products using animal protein is quite relevant. One of the criteria for determining the biological value of a product is the degree of its digestibility. The method is to determine the kinetics of acid and alkaline hydrolysis of protein components in minced semi-finished products in the "in vitro" system. For the study, minced semi-finished products (cutlets) were used: a control sample (the main raw material is beef cutlet meat and fat pork) and three prototypes with partial replacement of beef cutlet meat with a complex food additive based on animal and vegetable raw materials – 0.5%, 0.75%, and 1.0%, respectively. The hydration of the supplement is 1:15. The food supplement contains in its composition, g/100g: sodium alginate – 60, whey protein – 16, soy fibre – 24. Determination of the biological value of a protein by the enzymatic method is a fairly objective way to determine the ability of a protein to be broken down by proteolytic enzymes in the gastrointestinal tract. A device for protein hydrolysis was used by the enzymatic method to determine the biological value of minced semi-finished products. The essence of the method is six-hour hydrolysis by enzymes of the finished product. The first stage is fermentation with pepsin (3 hours), and the second stage is fermentation with trypsin (3 hours) with an hourly selection of hydrolysis products to determine the degree of digestibility at a certain stage of the study. It is proved that the addition of a food mixture to the recipe of minced semi-finished products is advisable since it allows providing them with better digestibility indicators. Therewith, the digestibility of the prototypes was slightly higher

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than the control one. The digestibility index for the control was 69%, and for the experimental subjects – 68-74%. The expediency of using a complex food additive based on animal and vegetable raw materials for the production of minced meat semi-finished products with the share of replacement of the main raw materials from 8% to 16% with an increase in quality indicators was experimentally established, namely, the digestibility of minced semi-finished products has been confirmed by the "*in vitro*" system, pepsin-trypsin

Keywords: minced meat cutlets, food mixture, pepsin-trypsin proteinase system, digestion

Relevance

Today in Ukraine and the world there is an increased demand for the consumption of fast-food products (frozen and chilled minced meat semi-finished products). The increase in their popularity is due to the long shelf life of products for consumption and their fast cooking time. The production of quick-frozen products is one of the most promising areas for the development of industrial processing of meat raw materials. Recently, animal proteins have become increasingly popular in the production of the above-mentioned products. Therefore, the development of new food products using animal protein, namely whey protein, is quite relevant.

Analysis of recent studies and papers

The analysis of recent studies indicates a steady interest of specialists in expanding and improving the range of minced meat semi-finished products by introducing various additives into their composition. One of the most effective ways to compensate for alimentary insufficiency in nutrition is the regular inclusion of functional food additives of various preventive purposes in the diet. A special feature of functional food additives obtained based on balanced compositions is their multicomponence and the realisation of the functional properties of raw food ingredients that can create structural systems.

There are many food additives and mixtures on the modern market, and each of them is limited in its component composition, so a new complex food additive was created based on animal and vegetable raw materials, which provides high functional and technological indicators of minced meat semi-finished products.

The food supplement is a mixture of the following components: as a biologically active component, it contains sodium alginate – 60%, as dietary fibre – soy fibre – 24%, and as a protein – whey protein – 16%. Each component of the mixture has its own functionality.

There is a group of polysaccharides, different from starch that is not digested by digestive enzymes. In a physiological sense, they are combined into a group of dietary fibres. Dietary fibre is a diverse composition and structure of polymers of natural origin. They are widely distributed in plant products: cereals, vegetables, and legumes.

Fibre (cellulose) is the most common high-molecular non-starchy polysaccharide. It is the main component and supporting material of plant cell walls. Fibre does not dissolve in water and under normal conditions does not undergo hydrolysis by acids. For the production of meat products, natural dietary fibres were developed, which are fibre, the raw material of which is soy.

The use of sodium alginate is based on the ability to form gels, that is, to work as a thickener, gelling agent and emulsifier. Sodium alginate is able to stabilise the structural and mechanical properties of the product. The widespread use of alginates is associated with such properties as increased viscosity, the ability to swell and gel, stabilisation of aqueous, and water-fat solutions.

One of the promising raw materials for research in this area, which contains a complex of biologically active substances and is rational in terms of economic indicators, is whey, based on which whey protein is produced.

The biological value of whey is determined by the content of protein nitrogenous compounds (primarily essential amino acids), carbohydrates, lipids, mineral salts, vitamins, organic acids, enzymes, immune bodies, and trace elements. Milk protein is considered one of the main components of meat products, which, however, is responsible for the taste, and the nutritional value of the finished product or semi-finished product.

The higher the biological value of food, the more it meets the physiological needs of the body. The rate at which hydrolysis of food proteins occurs is one of the indicators of their biological value (BV) since it allows for predicting the degree of protein utilisation by the tissues of living organisms. The BV of a food product can be determined by several methods: chemical, biological, and enzymatic. A fairly effective method of enzymatic hydrolysis, according to which the rate of protein digestion in the gastrointestinal tract is determined to examine this indicator, a proteinase system containing pepsin and trypsin consistently acts on dietary protein. Therewith, hydrolysis products are constantly removed from the reactive medium by dialysis. This method mimics the conditions under which food proteins are hydrolysed in the body.

The purpose of the study – analysis and determination by the enzymatic method of the biological value (degree of digestibility) of minced semi-finished products with a complex food additive based on animal and vegetable raw materials with its use in experimental samples, respectively, 0.5%, 0.75%, and 1.0%.

Materials and Methods

The objects of study are control and experimental samples of minced semi-finished products. The study was conducted in the laboratory of the Department of Technology of Meat and Meat Products of NUFT. The method of enzyme hydrolysis (pepsin-trypsin system) for determining the biological value of minced meat semi-finished *invitro* products was used as a basis. A device was used to conduct the study that has external and internal vessels separated by a semipermeable membrane (Fig. 1).

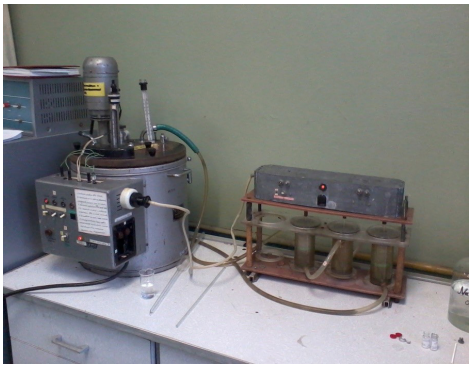

Figure 1. Hydrolysis device

Minced semi-finished products (cutlets) were used for research. Four samples were formed from them. The control sample (C) contained only the main raw materials – beef cutlet meat and fat pork. In three experimental samples, the main raw materials were partially replaced with a complex food additive based on animal and vegetable raw materials. The

food supplement contains: sodium alginate – 60 g/100g, whey protein – 16 g/100g and soy fibre – 24 g/100g. Dietary supplement administration was 0.5% (S1), 0.75% (S2), and 1.0% (S3). The hydration of the supplement is 1:15.

The research technique was as follows: firstly, frozen semi-finished products were subjected to heat treatment (frying) and then cooled. A sample containing approximately 150 mg of protein was selected from the finished product. Placed in the internal container of the device, 15 ml of 0.02 n hydrochloric acid solution with a pH of 1.7 was added. 60 ml of hydrochloric acid of the same concentration was added to the outer container. The inner container was immersed in the outer one until the liquid level in them was equal to adhere to the isotony. The samples were incubated in a water bath at a temperature of 37°C for 15 minutes with constant stirring. 15 mg of crystalline pepsin was then added to the internal container. Fermentation was conducted for 3 hours with constant stirring. During each hour, the hydrolysate was collected for further studies to determine the degree of digestibility. After fermentation, the mixture from the inner container was neutralised with a solution of sodium hydroxide, and then 10 ml of bicarbonate buffer was added. The mixture from the outer container was replaced completely with bicarbonate buffer. The liquid in both containers is at the same level. After temperature control for 15 minutes, 15 mg of crystalline trypsin was added to the internal vessel and further fermentation was performed for 3 hours. The hydrolysate was also collected each hour. The degree of digestion of product proteins is assessed by the difference between the amount of protein taken for the study and the protein that remained after sequential treatment of the product with pepsin and trypsin. The amount of accumulated hydrolysis products in the dialysate was determined by the Lowry method (the method is based on the staining of products during the interaction of Folin's reagent with an alkaline solution of proteins). According to it, the tyrosine content was identified in the samples of minced semi-finished products formed as a result of enzymatic hydrolysis using the enzymes pepsin and trypsin. The amount of protein in the solutions was determined according to the calibration graph. It was made using a standard tyrosine solution.

Results and Discussion

The biological value (BV) of proteins characterises their quality, ability to provide plastic processes and synthesis of metabolically active substances, and is explained by to the presence of essential amino acids in them, their ratio with substitutes and digestibility in the gastrointestinal tract.

It is known that animal proteins of different origins are digested differently. The speed of digestion is affected by a number of factors: the type of raw material, the degree of grinding, and heat treatment.

Figure 2 shows that minced semi-finished products at the initial stage are characterised by high rates of digestion under the action of pepsin. Digestion over the course of three hours occurred gradually without drastic changes. The best indicator of digestibility was prototype No. 3. It contains the largest amount of a food supplement based on whey protein. The control sample had the lowest digestibility index. This is due to the fact that the composition includes beef meat and pork fat, these types of meat raw materials have a relatively low rate of protein digestion compared to experimental samples that contain pure whey protein that is fully digested during the first three hours – pepsin.

Figure 3 shows the results obtained from further study with trypsin. In the 4th hour of the study, trypsin was added to pepsin hydrolysate. Subsequent studies were determined by relatively small indicators of digestion. Prototypes 2 and 3 had the best performance.

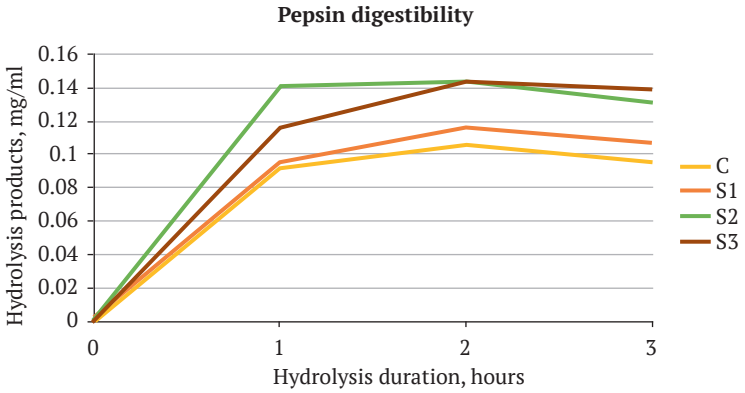

Figure 2. Pepsin protein digestibility (*in vitro*)

Figure 4 shows that the digestibility of all prototypes has a certain pattern. This is due to the gradual and partial replacement of the main raw materials of minced meat semi-finished products with a complex food additive. In the composition of the finished minced semi-finished product, the content of the food mixture (hydrated 1:15) is from 8 to 16%. The best digestibility index was demonstrated by prototypes No. 2 and No. 3, which were 72.0% and 74.0%, respectively, and the control sample – 69.0%. This is due to the fact that whey proteins are rapidly and almost completely digested by pepsin and trypsin, while raw meat is slower and not fully digested by trypsin.

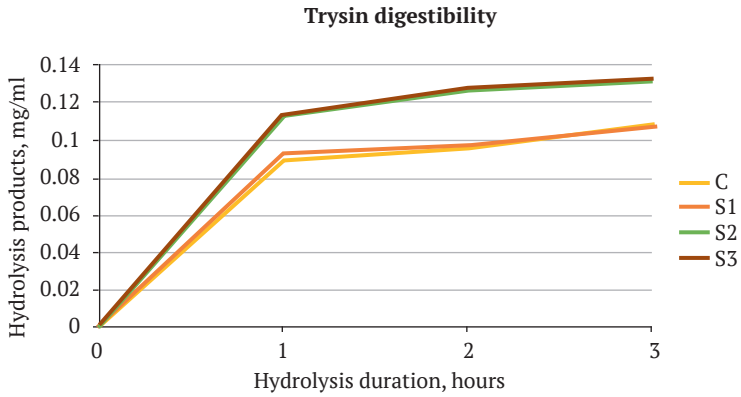

Figure 3. Trypsin protein digestibility (*in vitro*)

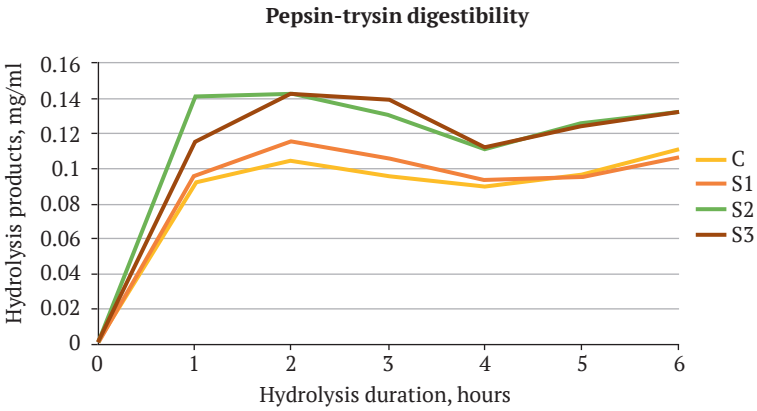

Figure 4. Rate of protein digestion by the pepsin-trypsin system

According to the obtained data, in accordance with the value of the concentration of accumulation of hydrolysis products of proteins of minced semi-finished products percentage to tyrosine, the value of digestion of proteins of semi-finished products changed.

Conclusions and Prospects

The biological value (digestibility under conditions of *in vitro*) of minced meat semi-finished products by the enzymatic method of protein hydrolysis. It was found that minced semi-finished products with the addition of 0.75% and 1.0% additives had better digestion indicators relative to the control by 3% and 5%, respectively. This indicates an improved biological value of the product in general. Therefore, the replacement of raw meat with a food additive, which after hydration is from 12% to 16% of the total weight, is advisable and can be used for the production of minced semi-finished products.

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Визначення біологічної цінності січених напівфабрикатів з комплексною харчовою добавкою ферментативним методом

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Анотація. Забезпечення населення білковими продуктами тваринного походження стає все більш складним завданням нашого часу. Тому розробка нових м'ясних продуктів з застосуванням білка тваринного походження є досить актуальним. Одним із критеріїв визначення біологічної цінності продукту є ступінь його перетравності. Методикою є визначення кінетики кислотного та лужного гідролізу білкових складових речовин у січених напівфабрикатах у системі «*in vitro*». Для досліджень використовували січені напівфабрикати (котлети): контрольний зразок (основна сировина – м'ясо котлетне яловиче та свинина жилована жирна) та три дослідних зразки з частковою заміною м'яса яловичого котлетного на комплексну харчову добавку на основі тваринної та рослинної сировини – 0,5 %, 0,75 % та 1,0 % відповідно. Гідратація добавки становить 1:15. Харчова добавка містить у своєму складі г/100г: альгінат натрію – 60, білок молочної сироватки – 16, соєву клітковину – 24. Визначення біологічної цінності білка ферментативним метод є досить об'єктивним способом визначення здатності білка до розщеплення протеолітичними ферментами у шлунково-кишковому тракті. Для визначення біологічної цінності січених напівфабрикатів ферментативним методом використовували прилад для гідролізу білків. Суть методу полягає у шестигодинному гідролізі ферментами наважки готового продукту. Перший етап – ферментація пепсином (3 години), другий етап – трипсином (3 години). З погодинним відбором продуктів гідролізу для визначення ступеня перетравності на певному етапі дослідження. Доведено, що додавання до рецептури січених напівфабрикатів харчової суміші являється доцільним, оскільки дозволяє отримати січений напівфабрикат з кращими показниками перетравлюваності. При цьому перетравлюваність дослідних зразків була дещо вищою за контрольний. Показник перетравлюваності для контролю становив 69 %, а для дослідних – 68-74 %. Експериментально встановлено доцільність використання комплексної харчової добавки на основі тваринної та рослинної сировини для виробництва м'ясних січених напівфабрикатів з часткою заміни основної сировини від 8 % до 16 % з підвищенням показників якості, а саме підтверджено перетравність посічених напівфабрикатів системою «*in vitro*», пепсин-трипсин

Ключові слова: котлети м'ясні січені, харчова суміш, система протеіназ пепсин-трипсин, перетравлення

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Influence of body structure types and pronouncement of meat forms of bulls on their sexual activity

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Abstract. The relevance of the study is determined by the need to find ways to improve the attributes of the reproduction of bulls that affect the economic efficiency of their use. The purpose of the study was to identify the relationship between the features of the exterior of bulls and their sexual activity, other signs of reproduction. Bulls of the Ukrainian meat breed were divided into two groups using the size Index: large-sized and compact. The pronouncement of meat forms in bulls was determined at the age of 15 months on a 60-point scale. Sexual activity was characterised by the duration of the manifestation of sexual reflexes from bringing the animal to the mount to mounting it. In experimental bulls, the duration of the period of productive use, the number of deliveries to the mount and attempts of mounting it, and the number of ejaculates received and culled were recorded. It was found that the tendency to distribute active moderate and calm manifestations of sexual reflexes in bulls of different types of body structure and the development of meat forms are similar. Most of all, there was a proportion of moderate sexual activity, but specific trends were observed within the groups. The proportion of high sexual activity was higher in compact bulls compared to large-sized peers and animals with better pronounced meat forms. Further, large-sized bulls tend to increase the duration of productive use, predominate in the proportion of mounts made and are characterised by a decrease in the proportion of ejaculate culling. Bulls with more pronounced meat forms tend to increase the duration of productive use, which is due to their higher breeding value based on the characteristics of meat productivity. In terms of the proportion of mounts made and the percentage of ejaculates culled, there was no difference between animals with different meat forms. The results obtained prove that compact bulls are more suitable for intensive use at an early age, and large-sized producers are more effective in adulthood. The better development of meat forms somewhat reduces the sexual activity of young bulls. At a more mature age, meat forms practically do not affect the results of using bulls. The results obtained allow selecting the type of body structure of bulls that have the highest sexual activity

Keywords: libido, meat cattle, artificial vagina, ejaculate

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Relevance

The problem of the reproduction of meat cattle is becoming increasingly urgent in animal husbandry. During the creation of the Ukrainian meat breed, artificial insemination of females was used by a complex reproductive crossing of the Kian, Charolez, Simmental, and grey Ukrainian breeds. The use of 4 breeds with different exteriors led to the production of new genotypes of animals with different types of body structures and the pronouncement of meat forms. The sexual activity of bulls also varied substantially, which sometimes led to inefficient use of males and high cost of sperm and calves. Getting bulls with a high libido can make it easier to manage cattle breeding.

Analysis of Literature Sources

Bulls are evaluated according to the signs of reproduction, which are divided (Savchuk, 1985) into indirect (anatomical structure of the genitals, size of the testes, girth of the scrotum, age of puberty, severity of sexual dimorphism) and direct (sexual activity, spermoproductivity, fertilising ability of sperm). The physiological maturity of bulls lasts from 2 to 5 years of age (Naumenko *et al*., 2009). In addition to the desired origin, exterior and constitution, the producer must show high sexual activity and produce high-quality sperm. During natural mating or receiving sperm for artificial insemination, producers exhibit five basic sexual unconditioned reflexes: attraction, hugging, erection, mating, and ejaculation. Sexual desire and hugging reflex bulls show not only on the female but also on the mount, which ensures the receipt of sperm in the artificial vagina. Not all producers give a quick mounting, which is manifested by conditioned reflexes. It is known that the sexual behaviour of producers is not affected by their sperm mass and physical and morphological aspects (De Oliveire *et al*., 2007). It is influenced by the parameters of the testes, which are determined by measuring and calculating the volume of the scrotum (Gippolito *et al*., 2019).

The sexual behaviour of a bull during natural mating does not always coincide with the manifestation of libido in the arena. Sexual activity can be affected by the dominance of other producers and stress, which worsens the final results. Bulls that have a strong libido manifestation provide a higher level of pregnancy and sperm production output compared to low and medium manifestation (Islam *et al*., 2018), so it is important to identify bulls with low activity of sexual behaviour in the arena when taking sperm into the artificial vagina. The problem of increasing the sexual activity of bulls and the quality of their sperm production is recommended (Kozyr & Barabash, 2018) to be solved by mechanical pressure and electrical stimulation of erogenous zones. The earliest manifestation of puberty and sexual behaviour in bulls is being investigated as one of the new features that allow for accelerated animal use (da Silva Neto *et al*., 2020). In turn, the sexual activity of animals is associated with the breed (Rehman *et al*., 2016) or a genetic group of animals (Schenk, 2018). Sexual behaviour depends not only on the individual characteristics of the bull, but also on the conditions of its use. The addition of variable factors can improve libido (Schenk, 2018), although the activity of sexual reflexes depend on the season of the year (Rinku *et al*., 2019; Piddubna, Zakharchuk, 2020).

Some signs of the performance of bulls are also associated with the manifestation of libido. In the Ongole grade breed (Affandhy *et al*., 2018) it was identified that young bulls have higher sexual activity and sperm quality at the intermediate live weight. In the complex of measures to improve the breeding value of meat bulls, the emphasis is placed on the choice of animals with a large live weight of the desired type of body structure, which most corresponds to the productive area of the livestock. Animals of compact

and large-sized types are distinguished by type. During their selection, special attention is paid to the pronouncement of meat forms. Previously, it was identified that large-sized bulls are characterised by a large volume of ejaculate and the number of spermatozoa with rectilinear translational movement in it, and large-sized bulls with better-expressed meat forms show a decrease in the volume of ejaculate, the concentration of sperm in one ML and their fertilising ability (Koropets, Ugnivenko, 2019).

Since the type of body structure and the development of musculature and subcutaneous adipose tissue are associated with metabolic processes in the animal's body, they may be associated with the activity of sexual reflexes.

The purpose of the study was to establish the manifestation of sexual activity and features of the reproductive ability of bulls of the Ukrainian meat breed of various types of body structure and the severity of meat forms when receiving sperm in an artificial vagina.

Material and Research Methods

The study was conducted at the Volia breeding plant in the Zolotinskyi District of the Cherkasy region. Until the age of 8 months, the bulls were suckled, after which they were tested for their own productivity. During the test, they investigated sexual behaviour by taking sperm on an artificial vagina, the type of body structure and pronouncement of meat forms, and signs of reproductive ability. A mount was used to test the libido of bulls in the arena. Bulls were tested individually and classified into three main degrees of sexual activity (active, calm, and moderate). Active (A) – a high degree of sexual activity was characterised by a short duration (from 10 to 60 seconds) of all sexual reflexes to mount. Moderate (M) – the degree of manifestation of sexual activity was characterised by a slightly longer course of reflexes. It lasted from 60 to 120 seconds. Calm (C) – a relatively low degree of sexual activity, in which the time from the animal to the mount to the attempt of mounting lasted more than 120 seconds.

Receiving sperm from bulls started at the age of 12 months. From the age of 14 months, they were used to get ejaculate once a week. Two ejaculates (double mounts) were taken from young bulls up to 18 months old, and up to four ejaculates per week were taken from adults. After taking sperm on an artificial vagina, the volume of ejaculate and the concentration and motility of sperm were evaluated in bulls.

The conditional compactness or large stature of bulls was determined according to the method proposed by the study (Ugnivenko *et al.*, 2010). It is based on the size index determined based on the oblique length of the body with a stick and the height in the sacrum. Based on the comparison of the index distribution, two conditional types were distinguished: large-sized (large-sized and long-bodied) and compact (short-sized and short-bodied). The pronouncement of meat forms of bulls was determined at 15 months of age on a 60-point scale (Prakhov *et al.*, 1990). Animals on this basis were divided into two groups – the better pronouncement (the score is higher than the average for the group of experimental bulls) and the worse pronouncement (the score is lower than the average).

Results and Discussion

Sexual activity and features of the use of bulls do not depend much on the type of structure of their body and the pronouncement of meat forms. In large-sized and compact bulls, there is a tendency to reduce the active and calm degrees of reflex manifestation, but this substantially increases the number of producers with a moderate degree of sexual activity (Table 1). The proportion of high sexual activity tends to increase in compact bulls compared to large-sized peers.

Table 1. *Sexual activity of bulls with different types of body structure and pronouncement of meat forms*

Feature	Degree of activity	Body structure type		Pronouncement of meat forms	
		large-sized (*n*=9)	compact (*n*=10)	better (*n*=20)	worse (*n*=20)
		M±*m*	*M*±*m*	*M*±*m*	*M*±*m*
Proportion of degrees of sexual activity, %	A	7.6±0.5	12.2±1.1	8.4±0.2	12.3±0.4
	M	71.3±4.9	69.4±4.6	71.4±2.1	69.7±1.9
	C	21.1±2.1	24.4±2.0	20.2±1.0	18.0±0.9

This is due to the fact that compact forms are characteristic of more precocious animals. That is, they reach physiological maturity at an earlier age, which to a certain extent has a positive effect on sexual activity. Large stature slows down puberty.

For more pronounced meat forms, which are evaluated on a 60-point scale, the best rating is also given to bulls with accelerated adipose tissue growth. The development of obesity contributes to a decrease in sexual activity. This affected the fact that the share of active manifestations of sexual reflexes that show libido in bulls with better pronounced meat forms is 4.1% less. Although the overall distribution of the frequency of sexual activity of bulls with different development of meat forms is similar, this trend must be considered.

Large-sized bulls tend to increase the number of leads to the mount and mountings on the artificial vagina. They have a longer duration of productive use (Table 2). Therewith, large-sized producers are characterised by a decrease in the proportion of ejaculate culling.

Table 2. *Features of using bulls for different types of body structure*

Feature	Body structure type		Pronouncement of meat forms	
	large-sized (*n*=9)	compact (*n*=10)	better (*n*=20)	worse (*n*=20)
	M±*m*	*M*±*m*	*M*±*m*	*M*±*m*
Number of leads to the mount, times	214±32.1	154±28.1	164±21.3	125±15.6
Number of completed mounts, times	196±27.0	120±25.8	135±19.2	99±14.7
Share of made mounts, %	80.3±4.2	71.8±7.5	82.3±2.4	79.2±3.4
Ejaculate culled, %	13.7±2.3	19.3±4.7	8.1±2.4	9.3±3.2
Duration of productive use of bulls, days	672±102.8	531±87.1	550±72.6	416±47.0

Bulls with pronounced meat forms tend to increase the number of leads and mountings and the duration of productive use. This is to some extent due to their higher breeding value in terms of meat productivity. In terms of the proportion of mounts made from the total number of leads and the percentage of ejaculates culled, there is practically no difference between animals with different manifestations of meat forms. Thus, there were no grounds for limiting the use of bulls with good muscle and subcutaneous tissue development due to the deterioration of signs of their reproductive ability. In turn, the compact type of bull, which is associated with the tendency to the precocity of animals, may have a slightly worse quality.

Conclusions and Prospects

In large-sized young bulls, a decrease in the share of active manifestation of sexual reflexes is observed in comparison with compact ones, which is due to their relatively late maturation, but during the period of productive use, these animals show a tendency to increase the share of mounts

made and a decrease in the number of ejaculates culled.Bulls with better pronounced meat forms during the test tend to reduce the proportion of leads to the mount with an active manifestation of sexual reflexes. Further, there is no difference in the proportion of mounts made and high-quality ejaculates between bulls with different development of meat forms, but the breeding value of meat productivity affects the duration of lifelong use.

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Вплив типів будови тіла та вираженості м'ясних форм бугаїв на їх статеву активність

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Анотація. Актуальність досліджень визначена необхідністю пошуку шляхів поліпшення ознак відтворення бугаїв, які впливають на економічну ефективність їх використання. Метою дослідження було виявити зв'язок між особливостями екстер'єру бугаїв і їх статевою активністю та іншими ознаками відтворення. Бугаїв української м'ясної породи з використанням індексу великорослості розділяли на дві групи: великорослі та компактні. Вираженість м'ясних форм у бугайців визначали в 15-місячному віці за 60-бальною шкалою. Статеву активність характеризували за тривалістю прояву статевих рефлексів від підведення тварини до опудала до садки на нього. У дослідних бугаїв фіксували тривалість періоду продуктивного використання, кількість підведень до опудала і спроб садок на нього та кількість отриманих і вибракуваних еякулятів. Було встановлено, що тенденція розподілу активного помірного і спокійного прояву статевих рефлексів у бугайців різних типів будови тіла і розвитку м'ясних форм подібна. Найбільше була частка помірного прояву статевої активності, але в межах груп спостерігались специфічні тенденції. Частка прояву високої статевої активності була більша у компактних бугайців проти великорослих ровесників і тварин з краще вираженими м'ясними формами. Надалі великорослі бугаї мають тенденцію до збільшення тривалості продуктивного використання, переважають за часткою зроблених садок та характеризуються зменшенням частки вибракування еякулятів. Бугайці за краще виражених м›ясних форм мають тенденцію до збільшення тривалості продуктивного використання, що обумовлено їх вищою племінною цінністю за ознаками м'ясної продуктивності. За часткою зроблених садок та відсотком вибракуваних еякулятів різниці між тваринами з різним проявом м'ясних форм не виявлено. Отримані результати доводять, що для інтенсивного використання в ранньому віці більш придатні бугайці компактного типу, а в зрілому віці більш ефективні великорослі плідники. Кращий розвиток м'ясних форм дещо знижує статеву активність молодих бугаїв. У більш зрілому віці на результати використання бугаїв м'ясні форми практично не впливають. Отримані результати дозволяють добирати за типом будови тіла бугаїв, які мають найвищу статеву активність

Ключові слова: лібідо, м'ясна худоба, штучна вагіна, еякулят

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