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Results of Preclinical Studies to Assess the Effect of Food Composition Components on Reproductive Health

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ABSTRACT

The paper presents the results of studies on the effect of food composition (FC) components on the clinical and biochemical status of laboratory animals in the in vivo experiment. The FC consisted of, g/100 g: albumin light - 30; MZWG - 45; white beans - 25. The experimental results have shown the absence of a biocidal effect to P. caudatum, which indicates the absence of potential toxic effects on the human body and confirms that the FC is biologically safe for human health. Assessment of the biotic potential indicates effective assimilation of the FC nutrient components. Counting the number of P. caudatum organisms developing on the substrate, including FC, revealed a smaller (by 22.3%) negative effect in all control points relative to chicken egg protein. The FC standardized relative bioavailability against the control had a significant value (77.7 %), which allows it to be classified as a raw protein source to develop targeted food formulations. In-vivo experiments in white inbred mice of the BALB/c line have shown that the development of laboratory animals in the early stages proceeded without significant deviations in the indicators by group. At the end of the experiment (day 36), a significant increase in weight of experimental group 2 laboratory mice, whose feed contained the studied enricher, was established, indicating a high digestibility of feed with FC. The dynamics of clinical and biochemical parameters indicates not only the normal course of metabolic processes but also a higher level of digestibility of carbohydrates of the experimental group feed. The calcium and phosphorus level in the experimental group was higher against the control one by 12.5 % and 10.5 %, respectively, which indicates the high availability and digestibility of trace elements, both available in the feed and introduced with FC. A similar level of alkaline phosphatase against a stable content of trace elements confirms the collagen synthesis by osteoblasts at a normal level. This fact indicates the predominance of osteosynthesis processes in the bone tissue of laboratory mice. The data obtained testify to the possibility of the FC under study use in food technologies in the development of specific commercial product lines to improve the nutritional status of the population of reproductive age.

Key words: Food composition, Dietary factors, Preclinical studies, Clinical and biochemical status, Reproductive health

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INTRODUCTION

The problems of improving the nutritional status and reproductive health of the population attract considerable attention in the world community. One of the priority tasks of the World Health Organization (WHO) policy is to improve the health of people of reproductive age, whose level determines the reproduction of the population, its social and physical health [1]. Members of the society of reproductive age are an important socio-demographic group in any state. Its age, quantitative and qualitative composition is considered as the essential reproduction factor, and maintenance and provision of comfortable life support of this population category is a global task of any state. The WHO strategy in this direction involves the organization and implementation of measures to reduce the mortality rate and improve the reproductive health of the planet. To date, there is a significant imbalance in the

demographic development system of individual states. While there are numerous projects in the Central European countries to improve the health of citizens of reproductive age, there is a low standard of living in a significant number of states, accompanied by the collapse of the social protection system, unemployment, and hunger. As a result, they have a high rate of maternal and infant mortality, a large number of abortions and sexually transmitted diseases, a low level of contraception, which leads to the presence of a wide range of diseases for men and women of reproductive age [2-10].

The health status of future parents, the availability of all necessary nutrients before conception, during pregnancy, childbirth, breastfeeding, body weight at the onset of pregnancy, and during its development have a huge impact on the birth of full-fledged members of society, with performance, social and physical health. In order to adjust their physiological nutrient-dependent states, sustainable consumption of necessary nutrients with food is essential. This implies, primarily, the availability and economic opportunity of the population to consume high-quality food of special orientation, the presence of a wide range of available food lines, providing alimentary correction of the nutritional status of men and women of reproductive age [2,7,11,12].

In this connection, there is a growing need to develop research and solutions under the specific needs of social groups in the development of new product lines of food aimed at eliminating the deficiency of nutrients in the human body [2,7,12-15].

Continued availability and replenishment of the body with alimentary sources of energy, native protein, vitamins, and minerals is necessary to improve the nutritional status of men and women of reproductive age. Then, parents can ensure a timely and continuous supply of necessary nutrients to the fetus, which will ensure the birth of healthy offspring in the future.

A valuable source of native protein of animal origin is light albumin [16-19]. This secondary product of slaughter animal blood processing obtained at meat processing plants contains up to 88 % of protein. Light albumin refers to inexpensive and available types of valuable bioresources. Compared with other protein enrichers, light food albumin proteins are the most complete and balanced source of amino acids (especially high content of leucine and lysine), closest in composition to dried egg protein, the price of which is significantly higher. On organoleptic indicators, albumin belongs to high-tech enrichers. It is a fine-grain loose powder without dusty inclusions having a slightly salty flavor, neutral color (white to light cream), and odorless. It is essential that albumin protein is characterized by high digestibility (up to 95%). This quality is explained by the fact that albumin, being a split peptide form, is not subjected to enzymatic processing in the stomach. Importantly, compared to other blood protein constituents, albumin has a lower molecular weight of about 68,000 daltons, which ensures its high solubility and ability to easily penetrate intercellular spaces. It dissolves well in water and saline solutions which are very common in food processing, allowing its incorporation into almost any food composition. Light albumin pH is in the range of 7.0-8.5, and with increasing salt concentration it practically does not change, positively affecting the stability of food systems. It is also characterized by high functional and technological characteristics. It has high water-binding and emulsifying properties, which makes it possible to significantly increase the shelf life of products, in particular in vacuum packaging, and refuse the introduction of additives into the composition of brine preparations to retain moisture. Albumin holds fat very well (FHC=120-125%), gives the product a denser structure and juiciness. Albumin already begins to exhibit its properties at 65°C and higher, with its structure becoming an irreversible gel, very similar to the protein of a chicken egg. As the temperature increases, the gel density increases, and, very importantly, this property is not lost when it cools down later, but the density continues to increase. This factor is very convenient for subsequent storage of finished products in refrigerators [17-21].

As a source of a wide range of vitamins and minerals, cake flour of wheat germ (CFWG), BAA "Vitazar" was chosen, obtained by cold pressing from wheat germ, a by-product of flour mills. CFWG is a cheap, affordable, and safe by-product of food industries containing essential substances necessary for the reproductive health of men and women who plan to have healthy offspring. CFWG contains a considerable amount of protein

fraction (33-35%), such valuable components as polyunsaturated fatty acid (PUFAs), macro- and trace elements, fat-soluble vitamins (A, D, E) and water-soluble ones (B vitamins), polycosanol, pentosans, and dietary fibers. A considerable amount of CFWG proteins contain essential amino acids, PUFAs, including valuable fatty acids such as linoleic and linolenic (omega-6 and omega-3). The beneficial effects of PUFAs on the cardiovascular, digestive, endocrine systems, active participation in metabolic processes of the body, especially in hormonal and fat metabolism, cleansing the body of wastes are known. Their consumption is essential to improve the health of people of reproductive age, especially for women during pregnancy and breastfeeding. The presence of polycosanol in CFWG (1.5-8.0 mg/100), which has an exceptional effect on the metabolism of low-density lipoprotein (LDL) cholesterol, is essential in the diet of future parents. Policosanol accelerates the transition of high-density lipoproteins (HDL), reduces their levels, and increases the HDL fraction. Active components of policosanol significantly reduce the formation of cholesterol and cleave existing cholesterol in the body, have an antioxidant effect. A significant share of the reproductive population is obese and overweight, so the presence of this component is promising in their diets, especially for pregnant and lactating women. Besides, polycosanol has antiplatelet action, increases the susceptibility of people to insulin, and reduces stress symptoms, which is also essential. The presence of ergosterol (0.6-0.9 mg/100g), retinol (0.6-0.8 mg/100g), abnormally high content of α-tocopherol (28.0-31.0 mg/100g), pentosans (up to 10%) and carotenoids, which have a positive effect on enhancing reproductive function, bone formation, vision, normalizing blood pressure, strengthening the walls of blood vessels, regulating blood glucose, improving the calcium and phosphorus uptake allow prediction of extremely beneficial CFWG effect on future parents, especially women during pregnancy and the postpartum period. A wide range of B vitamins, mg/100 g: B1 (2.5-3.5), B2 (0.5-0.6), B3 (14.0-15.0), B5 (8.0-9.0), B6 (1.0-1.5), B9 (0.5-2.5) suggests the positive effect on the body when eating foods with this enricher. The presence of thiamine is essential in carbohydrate metabolism, accompanied by the formation of energy resources. Riboflavin is essential in the regulation of biochemical processes of protein cleavage and enzyme formation. Niacin has a positive effect on fat metabolism and is an integral part of many enzymes, regulates the gastrointestinal tract (GIT), accelerates the repair and formation of skin, hair, and nails. Pantothenic acid is involved in the formation of sex hormones and regulates the genitourinary system. Pyridoxine protects against anemia, prevents diseases of the nervous system, atherosclerosis, prevents toxicoses. It is essential and necessary to support and restore the vital functions of the body of a person in reproductive age, planning to give birth to healthy children, especially for pregnant and lactating women. Folic acid is undoubtedly indicated in diets for pregnant women. It takes an active part in the processes of hemopoiesis, the formation of hematopoietic systems, structures, and organs. It significantly affects the intrauterine growth of the fetus and the subsequent period of the newborn rapid growth. More than 20 mineral elements are found in CFWG (Zn, Mn, Mg, Ca, K, Fe, Na, Se, P) which are essential for the fetus intrauterine growth and health maintenance of the future mother. Selenium deficiency reduces the body's resistance to pathogenic effects and the organism's antioxidant function; with its deficiency, pregnant women may have thyroid hyperplasia. Insufficient intake of zinc food sources significantly slows the formation of T-lymphocytes. This trace element is responsible for controlling gene expression during cell replication and differentiation, its presence in the first stages of embryonic growth is essential. With zinc deficiency, there is a direct correlation of increased likelihood of preterm birth, labor bleeding, decreased labor activity in childbirth, placental abruption. A high percentage of pregnant women's anemia (about 90%) refers to iron deficiency anemia (IDA). Iron deficiency leads to serious health problems for the mother and the newborn: the threat of miscarriage, the birth of premature babies, weakened immunity. Iron induces reactive oxygen intermediates, contributing to the pathogenic mechanism. In the final weeks of pregnancy, one in five women is deficient in calcium, which is associated with the construction and growth of the skeleton, teeth, and bone system of the growing baby in the womb. It is also responsible for the formation of the muscle tissue structure, many

organs, and systems. Pregnant women with low calcium content in the body are more likely to have eclampsia; systemic vascular changes and bone demineralization occur; there are disorders in the placenta, entailing hypertensive and hemodynamic disorders of the fetoplacental complex [22-24].

White beans, a very promising global source of raw materials, are considered to be an undemanding crop and their yield is about 17-20 cwt/ha. The area under beans in the world ranks second among legumes after soybeans and exceeds 250 million ha [11, 22, and 25]. In terms of chemical composition, white beans are not inferior to such legumes as lentils, peas, and soybeans. Presence of native substances in the bean seeds, mg/100 g: vitamins E (3.8), PP (2.1), B2 (0.2), B3 (1.2), B6 (0.9), B9 (9.1), minerals Fe (14.1), K (1100), Ca (150), Mg(103), S (159), P (541) and fiber make it an available raw material source for producing functional foods to improve human reproductive function, the importance of these native substances for the reproductive age population was mentioned above. Beans are a valuable source of native vegetable proteins (about 25%), a significant amount of which are essential amino acids. The composition of bean proteins is similar to that of animal proteins, and they are 70-80% digestible. Bean seeds have high functional and technological properties; have a positive effect on organoleptic indicators and biopotential of finished products. Bean proteins have a high emulsifying, water-binding, fat-absorbing ability, which is beneficial for technological food systems in which they are introduced. Also, an essential advantage of beans relative to alternative sources of animal materials is their low cost which makes it possible to replace the required part of the animal origin protein with a more economically advantageous type of raw materials in the industry of protein food products [14,25,26].

Providing men and women of reproductive age with essential nutrients by including targeted foods in their diets at all stages of the life cycle is essential, especially during pregnancy planning. Mothers-to-be need to monitor the intake and availability of particular nutrients early in pregnancy, throughout pregnancy, and breastfeeding. Compliance with these conditions is an effective method of preventing alimentary-dependent states of children of early, more mature and adolescent age, preventing the development of an intrauterine fetal disease, pregnancy failure, and hypogalactia. The development of a range of special-purpose food products given the needs of each person in society depending on the physiological characteristics of the body, geography, the presence of pathological factors, and the level of physical activity is relevant. For women, the gestational age and the presence and quality of lactation processes in the body are essential [2,7,27].

The work aimed to develop the technology and FC component composition to improve the nutritional status and reproductive health in preclinical studies.

MATERIALS AND METHODS

The study objects were vegetable beans of "White flat" grade (GOST "34299-2017 Fresh Vegetable Beans. Technical Conditions"); cake flour of wheat germ (TU 9295-014-18062042-06 "Wheat germ flour food destination "VITAZAR"); albumin food light (Plasma powder) 70B, produced under Regulation (EC) No 853/2004 and TR CU 021/2011, TR CU 022/2011, TR CU 029/2012, TR CU 038/2013.

Protein fraction digestibility was performed by an in-vivo method using the following indices: the biotic potential of the *P. caudatum* population and standardized relative bioavailability (an index characterizing the digestibility of product proteins). For the experiment, samples were prepared so that the protein volume concentration in the *P. caudatum* culture medium was 4 mg/cm3. Serial dilutions were performed after that. Ready samples were examined at 0.17; 0.34 and 0.68 mg/cm3, which corresponded to the calculated peptide concentrations of 1; 2 and 4 mg/cm3, respectively.

In the study of digestibility and bioavailability of products, egg protein (albumin) data were used as control values. In the control, egg protein was in conventional concentrations (1, 2, 4 mg/cm3). The solvent was distilled water. Growing *P. caudatum* infusoria under laboratory conditions took place without additions to the substrate, so the effect of components of non-protein nature on protein digestion was excluded in the standard. Protein digestibility values were

calculated at the moment when the infusoria population biotic potential reached its maximum values.

In preclinical evaluation studies of a diet with FC, two experimental and one control group were organized. Each group consisted of 15 white inbred BALB/c mice aged 15 days. Throughout the experiment, mice were kept under standard vivarium conditions, with natural light, without restricted access to food and water, in standard polycarbonate cages of 5 animals each. All animals passed a two-week adaptation period before starting the study. The animals were randomized and divided into 2 groups of 15 animals each for the experiment. The intact animals of group 1 (control) received standard FC-120-1 pelleted feed (LLC "Laboratorosnab", Russia); the animals of group 2 (experiment) were on the diet with the replacement of 25% of feed by FC.

The food weight per laboratory animal was within (4.5±0.5) g/day with their continuous access to water. Experimental time was 21 days, with a daily recording of animal weight data throughout the experiment. Blood sampling was performed from the mice's tail veins on the 0-th, 21-th day, and at the end of the experiment. Control was carried out on several clinical and biochemical parameters using Vital Development Corporation (Russia, St. Petersburg) test-systems. Total protein was determined by the biuret method, cholesterol – by the enzymatic colorimetric method, glucose – by the glucose oxidase test without deproteinization, low-density lipoproteins (LDL)–by the enzymatic colorimetric method with selective protection (without precipitation), high-density lipoproteins (HDL)–by the enzymatic method with immune inhibition (without precipitation), phosphorus–by the UV method without deproteinization, calcium–by the colorimetric method with Arsenazo III, alkaline phosphatase activity in serum, and plasma – by the optimized kinetic method according to the instructions of diagnostic kit for quantitative analysis. Ready reagents and test samples were introduced according to the recommended scheme [5].

A scheme to determine the glycemic index was as follows: for 20 days all animals were fed by the standard, the laboratory mice were left without food for 16 hours with access to drinking on the 20th day. After that, to calculate the glycemic index, carbohydrate loading of glucose at 2 g/kg concentration was once injected directly into the stomach using a noninvasive probe for the control group. At this, a single feeding according to the procedure was carried out with regular and FC-supplemented feed. Glucose levels were monitored in whole blood sampled from the tail veins of laboratory mice before the test, as well as 15, 30, 45, 60, 75, 90, 105, and 120 minutes after carbohydrate loading using a OneTouch Select Simple® glucometer.

Germination and preparation of beans were carried out as follows. Bean seeds were cleaned from impurities, washed with tap water at (19 ± 1)ºC, and put on trays for swelling. Water was changed one time every 8 hours. Further, the vessel with germinated seeds was placed in the steam convection apparatus "Rational" SCC61WE-3NAC400V50/60 with the "Dry heat" regime (with parameters of 0.09 kW air convection power and temperature 30°C) was pre-set. Beans were dried within 15-20 minutes up to the humidity of 11-14% and then ground in a Glasser-2M blender to a particle size of 0.3-0.5 mm.

Statistical processing of the experimental results was performed through the Statistica 6.0 software package for Windows by methods of descriptive statistics (mean, error of mean, and standard deviation). The results are presented as M ± SE. The Student's test was used to determine the reliability of the obtained values. Differences were considered reliable at $p \leq 0.05$.

RESULTS AND DISCUSSION

The development of production technology and FC composition was carried out given predicted consumer properties of the object and end products. White bean seeds, albumin, and CFWG, having high biopotential and prospects in food enrichment to improve the reproductive function of the population, were chosen as FC components.

Bean seeds contain anti-nutritional substances–proteinase inhibitors that significantly inhibit the activity of GIT proteolytic enzymes, so they were pretreated [27]. The parameters of swelling and germination (temperature – 18-20ºC, duration of swelling–36 h) of beans were determined to reduce trypsin inhibitory

activity and obtain the end products of the required quality. It was determined during the study that the trypsin inhibitor content in native bean seeds was at a level of 4.74 g/kg (in g inactivated trypsin per kg of product). It was found experimentally that during bioactivation, the trypsin inhibitor content decreased to 1.21 g/kg. Then, the germinated seeds were dried under gentle conditions and grounded according to the parameters specified above.

Given the human body to retain all vital functions (including reproductive) needs not only native protein of animal and vegetable origin in the right amount but also in the right ratio. It was chosen for FC in the range of 50-60:50-40. Optimum content selection of FC components was carried out using a software product in the MathCad environment. The FC components (g/100 g: albumin light – 30, CFWG – 45, white beans – 25) and preparation technological stages were determined. Protein content in FC was 49.3 g/100 g, while the ratio of animal and vegetable protein was in the ratio of 57:43, respectively.

Experimental data confirm the high bioavailability of the developed FC–75 %. The main indices of the content of essential amino acids and their balance had relatively high values (usability index–0.64, comparable redundancy index–3.0). The value of the amino-acid score dissimilarity index was at the level of 25% and confirmed a small value of amino-acid score excess in FC.

The FC biologic effectiveness under in-vivo conditions was assessed on *P. caudatum* population. Substrates with a protein content of 1, 2, and 4 mg/cm3 were used in experimental studies of the *P. caudatum* population. Keeping track of organisms and their counting was performed every 24 h (lag phase), 48 h (logarithmic phase), 72 h (retarded growth phase), and 96 h (stationary phase). The experimental results showed the absence of biocidal effect to P.caudatum indicating the absence of potential toxic effects on the human body. Counting the number of organisms developing on the substrate, including FC, relative to chicken egg protein revealed a lower (by 22.3%) generative effect in all control points (Table 1).

The population biotic potential (BP), generations/hour, was determined according to the known relationship:

$$BP=Nt/2000/t \quad Eq(1)$$

where Nt–number of *P. caudatum* organisms that developed in the egg-protein-based medium and the experimental sample at a certain incubation time; t – incubation time.

BP of *P. caudatum* population cultured on a substrate with FC compared to infusoria grown on a substrate with chicken egg protein was also significantly lower (in all investigated concentrations) during the whole life cycle (Table 2). It indicates that FC composition can meet body needs due to its high biological value.

The FC standard relative product bioavailability (SRPB) was calculated according to the known relationship:

$$SRPB=NFC/NC, \quad (2)$$

where NFC –number of *P. caudatum* organisms grown on the object under study (FC) at a certain incubation time, NC –number of *P. caudatum* organisms grown on the control medium (egg protein) at the same incubation time.

The FC SRPB value was calculated after 48 h of incubation because the FC BP reached its maximum at this stage (Table 2). When calculating the SRPB values, the absence of the biocidal effect of the studied object on *P. caudatum* was

Table 1: *P. caudatum* population cultured in medium containing chicken egg protein (control) and FC ($p<0.05$).

Protein content, mg/cm3	Population at time of exposure, h			
	24	48	72	96
in egg-protein-based medium				
1	14140 ± 1227	31800 ± 1280	35500 ± 1399	32200 ± 1050
2	19760 ± 1360	39990 ± 1250	38100 ± 1140	34400 ± 1120
4	30250 ± 1200	62350 ± 1850	52300 ± 1290	49400 ± 1350
in FC-based medium				
1	10900 ± 1110	23166 ± 1050	19052 ± 1050	16068 ± 1070
2	11977 ± 1050	31445 ± 1110	22150 ± 1110	18896 ± 1140
4	15896 ± 1060	48423 ± 1110	30100 ± 1060	27240 ± 1110

taken into account. The calculations showed that the FC SRPB value was 77.7% which is slightly (by 22.3%) lower than the bioavailability of chicken egg protein (100%) confirming the high FC bioavailability.

The study of the FC effect on the clinical and biochemical status of the laboratory animals proceeded for 36 days. The in-vivo experiment on white mice has shown that the development of animals at early stages proceeded without significant abnormalities in both groups. Animal survival (experiment and control) for the entire observation period was 100%. It indicates the absence of feed toxic effects on the organs and tissues of the laboratory animals. The body weight gain plotted against time was the same in both groups during the first 21 days of the study. Comparison of animal weight gain in the experimental group (group 2) against the intact one (group 1 – control) showed that at the end of the experiment the weight of animals in group 2 was 17.6% more than the control one indicating high digestibility of feed with FC.

In parallel with the mice' weight control, their clinical and biochemical status was studied (Table 3). The results show that the glucose content in the blood plasma of the laboratory animals in groups 1 and 2 varied slightly within the physiological norm (4.6±0.1 mmol/L).

The highest glucose content (4.5±0.82 mmol/L) was noted in the second group of laboratory mice on the 36th day of the experiment, which is 2.3% higher than in the intact group mice (group 1 –

Table 2: BP of *P. caudatum* cultured in FC-based medium (p<0.05).

Protein content, mg/cm3	Exposure time, h			
	24	48	72	96
in egg-protein-based medium				
1	0.29 ± 0.007	0.33 ± 0.010	0.25 ± 0.012	0.17 ± 0.007
2	0.40 ± 0.012	0.41 ± 0.011	0.26 ± 0.020	0.18 ± 0.014
4	0.63 ± 0.008	0.65 ± 0.009	0.36 ± 0.021	0.26 ± 0.005
in FC-based medium				
1	0.22 ± 0.005	0.24 ± 0.007	0.13 ± 0.009	0.08 ± 0.009
2	0.24 ± 0.004	0.33 ± 0.006	0.15 ± 0.011	0.09 ± 0.005
4	0.33 ± 0.005	0.50 ± 0.004	0.21 ± 0.005	0.14 ± 0.005

Table 3: Clinical and biochemical status of animals during the study of the FC effect on white inbred BALB/c mice in the in-vivo experiment.

Factor	Group 1	Group 2
Glucose. mmol/L		
0 days	4.5 ± 0.21	
21 days	4.4 ± 0.29	
36 days	4.4 ± 0.26	4.5 ± 0.82
Norm	4.6 ± 0.1	
Total protein. g/l		
0 days	54 ± 1.8	
21 days	55 ± 2.0	
36 days	54 ± 1.3	55 ± 1.1
Norm	55 ± 1.0	
Cholesterol level. mmol/L		
0 days	2.2 ± 0.10	
21 days	2.1 ± 0.09	
36 days	2.3 ± 0.27	2.2 ± 0.28
Norm	2.2 ± 0.10	
LDL. mmol/L		
0 days	0.44 ± 0.05	
21 days	0.51 ± 0.06	
36 days	2.46 ± 0.12	1.27 ± 0.11
Norm	4.6 ± 0.1	
HDL. mmol/L		
0 days	0.71 ± 0.14	
21 days	0.84 ± 0.12	
36 days	3.61 ± 0.16	2.96 ± 0.15
Norm	3.2 ± 0.1	

control), indicating a more active carbohydrate digestion process of dietary intake in group 2.

The content of total blood protein in laboratory animals (Table 3) in both groups throughout the experiment was at a level of 54-55 g/l, which corresponds to the physiological norm. This fact confirms the standard course of metabolic processes in the control and experimental groups. At the end of the experiment, the blood plasma cholesterol level in group 1 (control) was 2.3±0.27 mmol/L, which slightly (4.5%) exceededs the physiological norm. At the same time, the cholesterol content in the experimental group was within the physiological standard, indicating the absence of FC negative effect on fat metabolism of laboratory animals. LDL values in the experimental and control groups were at the level of the physiological standard while HDL increased in all mice by about 4.6 times (Table 3). These data do not confirm the presence of negative facts but testify to the presence of active parietal digestion processes and intake of lipids of experimental animals.

The metabolism of some trace elements was monitored during the studies. Table 4 results show the absence of significant deviations from the normal content of phosphorus and calcium in the blood of laboratory mice. However, on the 36th day of the experiment, the elevated level of phosphorus and calcium in the blood plasma in mice of the experimental group (group 2) against the control one by 12.5% and 10.5%, respectively. This fact indirectly confirms a higher digestibility and availability of trace elements in the feed with FC. Against the background of a stable level of trace elements, the same values of alkaline phosphatase confirm the presence of a normal process of collagen synthesis by osteoblasts with the moderate activity of osteoclasts. It indirectly confirms the predominance of osteosynthesis processes in the bone tissue of living organisms.

The results obtained confirm the necessity and potential possibility of expanding the commercial product lines of functional orientation to improve the human body homeostasis. Developments in this direction allow the use of new combinations of resource-saving bioactive raw materials having pharmaceutical properties, improve the nutritional status of the population of reproductive age, and contribute to the partial solution of demographic world problems.

CONCLUSION

As a result of the experimental studies, it has been found out that FC shows no signs of toxicity and is biologically safe for human health. Besides, the BP assessment indicates the effective nutrient uptake of FC compared to chicken egg protein. The FC SRPB against the control (chicken egg protein) has a significant value (77.7 %), which allows it to be classified as a raw protein source to develop targeted food formulations.

In-vivo experiments in white inbred mice of the BALB/c line have shown that the development of laboratory animals in the early stages proceeded without significant deviations in the indicators by group. At the end of the experiment (day 36), a significant increase in weight of experimental group 2 laboratory mice, whose feed contained the studied enricher, was established, indicating a high digestibility of feed with FC. Trends in clinical and biochemical parameters indicate not only the normal course of metabolic processes. They show a higher level of digestibility of carbohydrates of the experimental group feed (group 2). The calcium and phosphorus level in

Table 4: Information on the metabolism of macro- and trace elements in the blood plasma of laboratory animals in the in-vivo experiment on the effect of FC on clinical and biochemical status of white inbred BALB/c mice.

Factor	Group 1	Group 2
	Phosphorus, mmol/L	
0 days	0.8 ± 0.05	
21 days	0.7 ± 0.04	
36 days	0.8 ± 0.04	0.9 ± 0.04
	Calcium, mmol/L	
0 days	1.9 ± 0.13	
21 days	1.7 ± 0.15	
36 days	1.9 ± 0.14	2.1 ± 0.16
	Alkaline phosphatase, µmol/L	
0 days	113 ± 5	
21 days	116 ± 7	
36 days	115 ± 3.5	115 ± 3.5

the experimental group was higher against the control one by 12.5 % and 10.5 %, respectively, which indicates the high availability and digestibility of trace elements, both available in the feed and introduced with FC. A similar level of alkaline phosphatase against stable content of trace elements confirms the collagen synthesis by osteoblasts at a normal level. This fact indicates the predominance of osteosynthesis processes in the bone tissue of laboratory mice. The results obtained make it possible to recommend the FC under study for inclusion in the product formulations of assortment product lines of directed action to improve the reproductive health of the population.

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