Article

A Study of the Effects of a Diet with Functional Foods on the Adaptogenicity of First-Year Students to the Student Lifestyle

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Abstract: Background: The period of rapid transition from school to university is associated with a complex of negative stress factors caused by social and professional adaptation, irregular daily routine, sleep, and nutrition. During this period, the inclusion of functional foods in students' diets has an increasingly beneficial effect on their health. This study aimed to evaluate the impact of diets with functional foods (vegetable and protein–vegetable) on health indicators while minimizing the negative impact of adaptation and acclimatization on the body of first-year university students at the start of their studies. Materials and Methods: A total of 150 first-year students were randomly selected. Biochemical tests, enzyme immunoassays, and analyses of serum immunoglobulin levels and mineral and vitamin content in first-year students' blood were performed. Results and discussion: Protein–vegetable products are more potent compared to plant-based ones in increasing the body's natural resistance to all types of stress due to their high protein content. The other functional product had a higher content of carbohydrates. In addition, they differed in the content of minerals and vitamins. Conclusions: The importance of a rational diet increases exponentially during the period of adaptation and acclimatization when the emotional, mental, and physical workload increases.

Keywords: functional food; students; prevention; immunity; blood test; vitamins; antioxidants

1. Introduction

As a result of modern factors such as unfavorable environmental situation, decreased physical activity, bad habits, and poor nutrition, the body's resistance to harmful factors is reduced, not only among the middle-aged and elderly, but also among young people, particularly adolescents and students [1–3].

Students are a social group of young people who live a unique lifestyle and have a unique mentality [2]. Existing research indicates that a number of factors [1–4], including anxiety [5,6], depression [7–9], low physical activity [10–12], stress (especially during exams) [13–15], and improper and irregular nutrition [16–18], can have a cumulative negative impact on the mental and physical health of young people when they enter university. Many first-year students must adjust to both the new social and educational environment [19,20] as well as new external conditions such as climate, time zone changes, and so on [21–23] after relocating to the place of study. Numerous studies have shown that

first-year students frequently struggle to adjust to new situations, have an unbalanced sleep and rest schedule [24–26], and are more likely to develop a variety of bad habits [27–30]. It is now understood that transitioning from high school to university necessitates biological, social, and professional adaptation [31,32]. Influencing the state of the body through a balanced diet, contributing to disease prevention and increasing endurance and life expectancy, is of particular interest in this context [33–37].

Students' adjustment to university life is a pressing issue today because it will have a significant impact on their future professional success and personal growth [2]. In a changing reality, many modern students struggle with self-development [6–9]. First-year students are often the most exposed to this problem. First of all, university study conditions are very different from those in schools. Every student must adjust to new circumstances, demands, and a different structure for the educational process than at school [5]. A first-year student should, on the one hand, demonstrate more initiative and independence in learning, but on the other, this necessitates greater responsibility for their education (a university student, more so than a high school student, must independently organize his educational space) [1]. A first-year student faces additional psychological challenges as a result of having to adjust not only to new learning conditions but also to big city life [21–23]. The first year of university study is a constant source of stress for students (financial issues, housing issues, self-catering, difficulty processing a large amount of information) [19,30,31]. Furthermore, because university education is the first step in acquiring profession, the first-year student goes through primary professional socialization and begins to engage in professional activities, which entails additional adaptation processes, but only in a specific professional field. The study of first-year students' adaptation to the learning environment is particularly relevant in this regard [13,16,30].

Good nutrition is largely responsible for the success of adaptation [35]. Food products that have been enhanced with specific ingredients to meet the body's needs are therefore a promising object for research [2–4]. Therefore, any research that contributes to a scientific understanding of the purposeful management of aspects of physical and psychological health through the use of food products made from natural biologically active substances and functional ingredients is relevant [36–38].

Promotion of a healthy lifestyle and nutrition has become a state policy in developed countries. Japan, England, the United States, Germany, France, China, and many other developed and developing countries are implementing national programs to preserve and improve national health. A similar program exists in Russia. An integral part of these programs is the production of food products and food ingredients, which can significantly improve the biochemical composition of consumer products. The inclusion of functional foods (FFs) in the daily diet has already proven its effectiveness in optimizing nutrition, preventing infectious and non-communicable diseases, and improving the quality of life [39,40]. Vegetables and fruits are the best examples of such foods, as they contain many biologically active elements [41].

Plant antioxidants, such as polyphenols, ascorbic acid, betacyanins, carotenoids, tocopherols, etc., play an especially important role in the human body. These compounds, which enter the human body as part of vegetable FPs, protect protein molecules, DNA, and cell membrane components from damage and inactivation caused by oxidative stress and other chronic pathologies [40,41].

Vegetable-derived FPs include physiologically active, soluble, and insoluble dietary fibers (pectins, etc.), vitamins (vitamin E, tocotrienols, folic acid, etc.), minerals (calcium, magnesium, iron, selenium, etc.), fats and substances associated with fats (polyunsaturated fatty acids, plant sterols, conjugated isomers of linoleic acid, structured lipids, sphingolipids, etc.), polysaccharides, secondary plant compounds (flavonoids/polyphenols, carotenoids, lycopene, etc.), probiotics, prebiotics, and synbiotics [39].

Riboflavin, or vitamin B2, is involved in redox reactions in the human body, while retinol, or vitamin A, is involved in cell membrane activity and is essential for human growth and development and mucous membrane function and participates in photorecep-

tion (light perception). Tocopherol, or vitamin E, is a powerful antioxidant that slows the aging process in the body and is involved in the sexual gonads in both men and women. Furthermore, vitamin E is required for normal immune system function; it improves cell nutrition, promotes peripheral circulation, prevents blood clots and strengthens vessel walls, is required for tissue regeneration, reduces the possibility of scarring, ensures normal blood clotting, lowers blood pressure, supports nerve health, ensures muscle function, prevents anemia, and alleviates Alzheimer's disease and diabetes [30–32].

Iron is a vital element for the body. Iron is a heme-containing protein that participates in oxygen transport (hemoglobin and myoglobin). Iron is also found in cytochromes, which are complex proteins that belong to the chromoprotein class and are involved in tissue respiration processes. Mg is important for reproductive functions and normal functioning of the central nervous system. Magnesium is involved in the synthesis of neurotransmitters, improves muscle reflexes, ensures the development of connective and bone tissue, increases fat utilization, and enhances the effects of insulin. Copper is involved in maintaining the elasticity of ligaments, tendons, skin, walls of lung alveoli, capillary walls, and bone strength. Copper is found in the protective membranes of nerve fibers and is involved in the pigmentation process because it is a component of melanin. Copper affects carbohydrate metabolism by enhancing glucose oxidation and inhibition of muscle and liver glycogen breakdown. Copper has anti-inflammatory properties and aids in the fight against bacterial agents. Copper is a cofactor of antioxidant defense enzymes and helps neutralize the effects of free radicals [31,35].

Zinc is an essential trace element that everyone requires to stay healthy. In terms of concentration in the body, this element is second only to iron among micronutrients [42]. Zinc is found in cells throughout the body. It is necessary for the proper functioning of the protective (immune) system of the body. It plays a role in cell division, cell growth, wound healing, and carbohydrate breakdown. Zinc is also necessary for the sense of smell and taste. During pregnancy, infancy, and childhood, the body needs zinc for proper growth and development. Zinc also enhances the effects of insulin. The epidermal, gastrointestinal, central nervous, immune, skeletal, and reproductive systems are the systems most clinically affected by zinc deficiency [42].

In student youth, nutritional status violations, as well as a lack of functional foods with therapeutic and prophylactic properties in the diet, lead to the development of chronic fatigue syndrome [29–31]. Theoretical studies have demonstrated the importance of incorporating essential nutritional factors into scientifically based diets. In practice, however, there are almost no data on the development of such diets for young students [34–36].

This study aimed to evaluate the impact of diets with functional foods (vegetable and protein–vegetable) on health indicators while minimizing the negative impact of adaptation and acclimatization on the body of first-year university students at the start of their studies. The hypothesis in this study was that there might be a way to lessen the detrimental effects of lifestyle changes on first-year students.

2. Materials and Methods

2.1. Research Objects

Functional food products—protein–vegetable product (PVP) "SportAktiv SportLine" (Santeville, Nizhny Novgorod, Russia) and vegetable product (VP) "Sila Prirody" (Santeville, Nizhny Novgorod, Russia)—were used as study objects. These functional foods are produced using cryogenic technologies [43]. The functional food products were assigned to the following treatment groups: TG 1—protein–vegetable product, TG 2—vegetable product. Protein–vegetable functional product contained seeds of rose hips (20%), kelp (16%), parsley (15%), watermelon (13%), celery (10%), egg protein (10%), oats (8%), and spinach (8%).

Vegetable functional product contained Jerusalem artichoke (50%) and beans (50%). Both functional products were produced in the form of tablets and were consumed orally. Three tablets (0.3 g each) of the functional product were taken twice daily, at breakfast and

dinner, for 15 days. The study used registered functional products that are approved for sale in the EEC [44].

2.2. Conditions for Selecting and Forming Groups of Students

The study included a randomized controlled clinical trial of two functional products in order to gain insights into their efficiency in preventing diseases. For this study, 150 first-year male students (mean age 18 years) were randomly selected from students of the I. Kanat Baltic Federal University (Kaliningrad, Russia) and divided into three equal groups (50 each): two groups that were treated with functional foods (TG 1 and TG 2) and a control group that was not treated with functional foods (CG). Informed voluntary consent to participate in the study (approved by the Volga Medical Research University Ethics Committee, no. 8/8.05.2019) was acquired from all participants. Prior to the clinical trials, all students underwent a medical examination and were found to be healthy. Disadaptation changes in the bodies of students were caused by increased absorption of vitamins and minerals in the process of simultaneous acclimatization and adaptation to the new social environment [43].

The study was conducted in accordance with the recommendations of the Consolidated Standards for Test Reporting (CONSORT) (Figure 1) [45]. Inclusion criteria included: signed written informed consent to participate in the study, a medical report on health status (no history of type 1, 2 diabetes, hepatitis A and B, ENT diseases, GI diseases in the acute phase, benign or malignant formations), an average age of 18 years, being a first-year student from another climatic region. Exclusion criteria: presence of acute respiratory illness, allergic reactions, fever, protocol violation, any other condition that, in the opinion of the investigator, may increase the risk to the participant, or reduce the likelihood of obtaining reliable results necessary to achieve the study objectives. The products were manufactured by Santeville (Nizhny Novgorod, Russia) in tablet form and packaged in identical plastic containers numbered underneath according to a random computer-generated list.

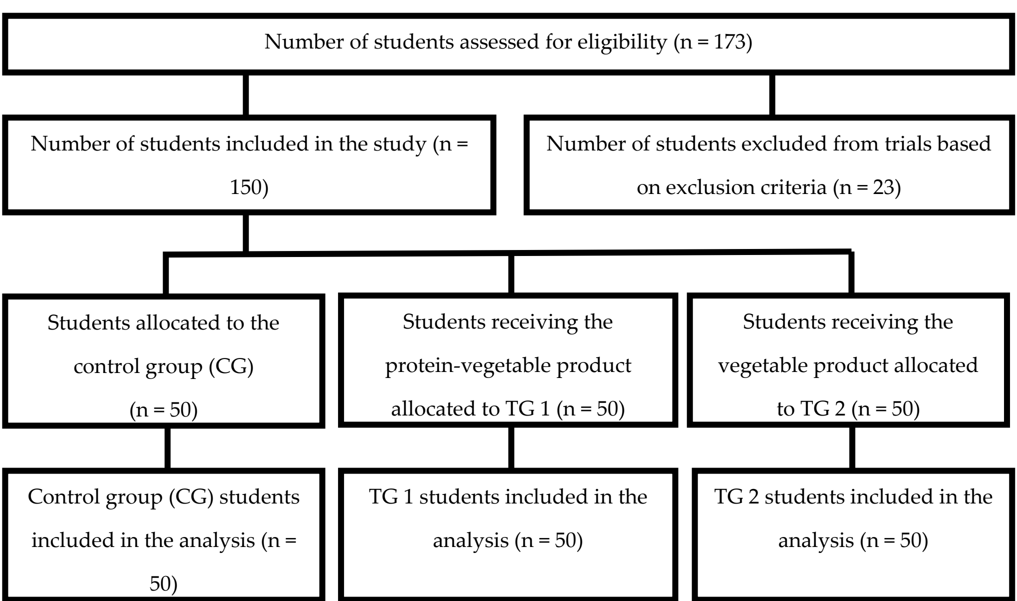

Figure 1. CONSORT flow diagram of student selection.

All students had the same living conditions and daily diet; they lived in a student hostel and had their food delivered (organized catering). The daily calorie intake was 4400 Kcal. The curriculum of the first-year students included intensive sports training, which was an important factor in this research. Physical activity for students in the study groups was three times a week, of moderate intensity, for an average of 80 min each time [43]. The control group (CG) had regular meals with no PVP and VP introduced. The

daily caloric intake was determined taking into account gender, age, and physical activity coefficient, according to [46].

2.3. Methods of Blood Sample Analysis

The studies on the PVP and VP as well as serum and blood samples were performed at the Research Institute of Hygiene and Occupational Pathology of Rospotrebnadzor (Novgorod, Russia), license number L041-00110-52_00575051 issued on 04/06/2014.

The content of vitamins A (retinol and carotenoids), E (alpha-tocopherol), and K (K1) was analyzed in FPs. The content of vitamin A (retinol), vitamin E (alpha-tocopherol), and mineral elements was measured in blood serum; the content of vitamin B2 (riboflavin) was measured in blood samples. The number of pyruvic acid (PVA) breakdown products demonstrates saturation of the body with vitamin B1 (high levels of the vitamin determine a decrease in the vitamin supply of the body). Blood sampling was performed after 15, 30, and 45 days of observation. Blood was drawn in a university outpatient clinic that is licensed to perform this type of analysis. After overnight fasting, blood was drawn from the ulnar vein. Blood samples were collected using a vacuum tube with heparin. For a complete blood count, the samples were delivered to the clinical laboratory within one hour.

Serum/blood samples were refrigerated before being sent to the testing laboratory and transported according to frozen biomaterial transport protocols. The indicators studied were total protein, protein fractions (albumin, α1-, α2-, β-, and γ-globulin), hormones (cortisol and testosterone), and serum immunoglobulins (IgA, IgM, and IgG).

A Sysmex XS-800 hematology analyzer (Sysmex Corporation, Tokyo, Japan) was used to perform a complete blood count. Protein levels were determined using a ROKI-6T photometer (Olvex Diagnosticum, St. Petersburg, Russia) and Olvex Diagnostic reagents (Olvex Diagnosticum, St. Petersburg, Russia) [47]. Protein fractions were determined on a UEF-01-Astra electrophoresis unit (NPC Astra, Ufa, Russia) using CliniTest-EF reagents manufactured by Rosmedbio (Moscow, Russia). A set of sera and cellulose acetate membranes were used. A densitometer was used to scan the electrophoregram [48,49].

The concentration of calcium, chloride, inorganic phosphorus, iron, magnesium, potassium, and sodium in blood serum samples was determined using Olvex diagnostic reagents (Olvex Diagnosticum, St. Petersburg, Russia). The tests were performed on a CLIMA MC-15 biochemical analyzer (DALLAS SEMICONDUCTOR, Dallas, TX, USA). Concentrations of iron and magnesium were determined by the colorimetric method without deproteinization in combination with nitro-PAPS and xylidyl blue chromogen nitro-PAPS and xylidyl blue chromogens [50,51].

Potassium concentration was determined by the nephelometric method without deproteinization using tetraphenylborate [52]. The spectrophotometric method without deproteinization [53] was used to determine the content of inorganic phosphorus in blood serum samples. This method uses the ability of phosphates to form a phosphorus–molybdate complex with ammonium molybdate; the optical density of this complex and the concentration of inorganic phosphorus in the sample are proportional [54]. The sodium concentration [55] was determined by the colorimetric method in combination with a precipitating reagent, and the chloride level was determined by the same method in combination with mercury rhodanide [56].

A Quantum-II/2 atomic absorption spectrometer determined zinc and copper concentrations in blood serum. The content of vitamins A, E (in blood serum), and B2 (in blood) was determined using a "Fluorat-02-ABLF-T" biofluid analyzer. Retinol fluorescence was measured in a hexane extract of serum previously exposed to water and ethanol to determine vitamin A content (range of measurable values 0.1–1.0 $\mu g/cm^3$) [57]. Alpha-tocopherol fluorescence was measured in a hexane extract of serum pretreated with water and ethanol to determine vitamin E content (measurable value range 2–15 $\mu g/cm^3$) [57]. Vitamin B1 levels were measured by the content of pyruvic acid breakdown product. A high content of pyruvic acid breakdown products indicates a low concentration of the

vitamin in the sample. The reaction of pyruvic acid with 2,4-diphenylhydrazine produces hydrazine. It turns brownish red in an alkaline environment. Pyruvic acid content and staining intensity are directly proportional (the range of pyruvic acid concentration was 2.5–20.0 $\mu g/cm^3$ without sample dilution) [58]. The Berch, Bessey, and Lowry method of measuring vitamin B2 mass concentration was used to determine vitamin B2 concentration in whole blood. The concentration range of 0.1–1.0 $\mu g/cm^3$ without sample dilution was determined [59]. Using trichloroacetic acid followed by acid hydrolysis of flavinadenindinucleotide and fluorometric determination of riboflavin content, different forms of riboflavin were isolated from blood.

The levels of vitamins A, E, B1, and B2 in functional foods were measured in the same way that they were in blood. The content and concentration of minerals was determined by atomic absorption spectrometry using Kvant-2AT and Kvant.Z-ETA.

Immunoassay was used to measure hormone content in blood serum (concentration of cortisol and testosterone). Reagent kits manufactured by Vector Best and the AccuBind ELISA Microwells kit were used in this study. Clinical and laboratory parameters characterizing metabolic status, hematopoiesis, humoral immunity, and hormones (cortisol and testosterone) were used to assess the effectiveness of FPs in maintaining students' health.

2.4. Statistical Analysis

All data were processed using the software Statistica 6.1 (StatSoft Inc., 2007, Tulsa, OK, USA). All experimental values were presented as mean ± standard deviation (SD). Descriptive statistics analysis was performed. To determine the reliability of differences between samples in parametric distributions, the Student's *t*-test was used; in nonparametric distributions, the Mann–Whitney and Wilcoxon tests were used. Differences between means were considered significant when the confidence interval was smaller than 5% ($p < 0.05$).

3. Results

3.1. Comparative Analysis of Macro- and Micronutrients in Native Raw Materials and Functional Products

Table 1 presents sociodemographic characteristics (gender, age, employment and occupation, and place of residence before participation in the study). Three groups of volunteers were formed; all underwent an initial examination, were randomized, and were subjected to analysis. During the study, none of the volunteers stopped taking the product or refused observation.

Table 1. Sociodemographic and clinical characteristics of the study groups.

Characteristics	CG	TG 1	TG 2
Number	50	50	50
Male	50	50	50
Female	0	0	0
Age (completed years)	18	18	18
Student	50	50	50
Not a student	0	0	0
From another region	50	50	50
Not from another region	0	0	0
Passed the initial medical examination	50	50	50
Disqualified	0	0	0
Randomized	50	50	50
Scheduled for intervention	0	50	50
Not scheduled for intervention	50	0	0
The intervention was performed	0	50	50
Stopped taking the product	0	0	0
Refused intervention	0	0	0
Analyzed	50	50	50

Tables 2 and 3 demonstrate the biological and energy value, as well as the micronutrient content, of vegetable and protein–vegetable functional food samples. The protein–vegetable functional product contains three times the protein of the vegetable product. The protein–vegetable functional product contains 1.4 times the carbohydrates of the vegetable product. The protein–vegetable functional product has ten times the calories of the vegetable product.

Table 2. Biological and energy value of vegetable and protein–vegetable functional products.

Macronutrients	Content, g Per 100 g of Product	
	VP	PVP
Protein	10.0	30.0
Fats	1.0	1.0
Carbohydrates, g	45.0	65.0
Energy value, Kcal	30.0	309.0

VP—vegetable functional product; PVP—protein–vegetable functional product.

Table 3. The content of micronutrients in vegetable and protein–vegetable functional products.

Micronutrients	Content, g Per 100 g of the Product	
	VP	PVP
	Vitamins	
E	0.484	0.05
A	8.29	0
B_2	0.58	0.24
	Minerals	
Cu	0.68	0.27
Fe	55.58	21.0
Zn	3.0	2.12
Mg	2.15	0.93

VP—vegetable functional product; PVP—protein–vegetable functional product.

The vegetable functional product contains 9.7 and 2.4 times more vitamins E and B than the protein–vegetable product. Vitamin A is absent in the protein–vegetable functional product. The vegetable functional product is superior to the protein–vegetable product in terms of mineral content as well: Cu is 2.5 times higher, Fe is 2.7 times higher, Zn is 1.4 times higher, and Mg is 2.3 times higher.

3.2. Analysis of Blood Samples

The total protein in the blood of all groups was within the reference limits (64–83 g/L) and did not change significantly during all stages of observation (Table 4), although there were certain variations in the fractional structure of proteins. To be more specific, the albumin fraction in the CG at each stage of the study was significantly higher compared with the initial values (8.2% and 15.5% ($p < 0.05$), respectively). In TG 2, albumin increased 4.6% at the end of the final stage of the study ($p < 0.05$), while in TG 1 it did not change.

The concentrations of α1- and α2-globulins reached the upper reference limit. The concentration of α1-globulins in the CG and TG 2 did not change throughout the entire observation period. In TG 1 this indicator significantly decreased (by 12.4% and by 14.6% ($p < 0.05$)) after the end of therapy with functional foods and at the end of the final phase of the study. At the same time, at the initial stage, the concentration of α1-globulin in TG 1 went beyond the upper reference limit.

Table 4. Dynamics of protein metabolism values in biomaterial samples (average).

Indicators (Reference Values) [47]	Observation Period						
	Values			Dynamics, %		p	
	1	2	3	2	3	2	3
Total protein, g/L (64–83):							
treatment 1	74.15 ± 0.68	75.9 ± 1.00	74.60 ± 0.80	+2.3	+0.6	0.211	0.272
treatment 2	73.05 ± 0.48	72.1 ± 0.56	68.70 ± 3.76	−1.4	−6.0	0.089	0.225
control	73.20 ± 0.60	71.8 ± 0.70	74.80 ± 0.90	−1.9	+2.1	0.325	0.398
Albumin, % (46.9–61.4):							
treatment 1	55.71 ± 0.70	54.29 ± 0.64	55.44 ± 0.88	−2.6	−0.5	0.371	0.629
treatment 2	53.86 ± 0.65	55.96 ± 0.63	56.36 ± 0.69	+3.9	+4.6	0.252	0.005
control	51.95 ± 1.00	56.23 ± 0.80	57.91 ± 0.74	+8.2	+11.5	0.025	0.029
Alpha-1 globulins, % (2.2–4.2):							
treatment 1	4.37 ± 0.17	3.83 ± 0.10	3.73 ± 0.14	−12.4	−14.6	0.012	0.008
treatment 2	4.19 ± 0.11	4.08 ± 0.15	4.02 ± 0.12	−2.6	−4.1	0.262	0.212
control	4.21 ± 0.13	4.05 ± 0.20	4.24 ± 0.71	−3.8	+0.7	0.952	0.593
Alpha-2 globulins, % (7.9–10.9):							
treatment 1	10.20 ± 0.23	9.66 ± 0.25	10.02 ± 0.49	−5.3	−1.8	0.132	0.747
treatment 2	10.86 ± 0.46	9.50 ± 0.32	10.22 ± 0.42	−12.7	−5.9	0.027	0.375
control	11.68 ± 0.44	10.06 ± 0.30	9.91 ± 0.54	−13.9	−15.5	0.015	0.036
Beta globulins, % (10.2–18.3):							
treatment 1	11.40 ± 0.26	12.31 ± 0.23	11.61 ± 0.42	+8.0	+1.8	0.025	0.658
treatment 2	11.89 ± 0.27	11.63 ± 0.30	11.84 ± 0.42	−2.2	−0.4	0.708	0.809
control	11.79 ± 0.29	12.13 ± 0.18	11.30 ± 0.52	+2.8	−4.2	0.678	0.646
Gamma globulins, % (17.6–25.4):							
treatment 1	18.28 ± 0.53	19.85 ± 0.58	19.19 ± 0.85	+8.6	+4.9	0.023	0.029
treatment 2	19.18 ± 0.53	18.76 ± 0.53	17.50 ± 0.55	−2.2	−8.8	0.411	0.032
control	20.54 ± 0.80	16.69 ± 0.73	16.63 ± 0.59	−18.7	−19.0	0.038	0.028

1—Beginning (21 days); 2—End (37 days); 3—45 days into observation (67th day). Data presented as a mean ± SD (n = 3). Values did not have significant differences ($p > 0.05$) as assessed by post hoc test.

The concentration of α2-globulins in the CG was initially above the norm. It decreased 13.9% ($p < 0.05$) by the time of the interim blood test. This decrease was 15.5% ($p < 0.05$) at the end of the final stage of the study. In TG 2, the fraction of this type of globulin also dropped significantly by 12.7% ($p < 0.05$) by the interim tests, but later on, it did not differ from the initial value. In TG 1, the concentration of α2-globulins did not change and remained stable.

What is interesting in Table 3 is that in the CG and TG 2, there were no changes in the concentration of β-globulins during the observation period. In students of TG 1, by the end of the protein–plant FP therapy, the concentration of β-globulins increased 8% ($p < 0.05$), but 30 days after the end of the intake, it went down to its initial value.

The concentration of γ-globulins in the blood serum of the CG at the second and third stages of observation were significantly lower than at the initial stage (18.7% and 19.0% ($p < 0.05$) respectively), and were below the norm. In TG 2, an 8.8% decrease ($p < 0.05$) below the norm in this protein was observed only after 45 days. The CG showed a 19.0% drop in γ-globulins whereas in TG 2 the decrease was not so large—8.8%—which is 2.1 times less than in the CG. In TG 1, after completing therapy with functional foods, the level of γ-globulins rose by 8.6% ($p < 0.05$) and remained high 45 days into observation (4.9% ($p < 0.05$)).

The dynamics of blood serum parameters showed that 100% of the CG subjects had an increased level of albumin at the time of the interim test, and one fifth of subjects exceeded the reference limit. By the end of the observation period, an increased level of

albumin relative to the initial value was observed in 90% of the subjects. The initial α1- and α2-globulin levels were above the normal level in 50–60% of students. At the end of observation, high levels of these globulins were registered in 40% (α1) and 30% (α2) of the subjects. In addition, at the second and third stages of observation, the level of α1-globulins increased or remained the same relative to the initial value in 40–60% of students. An increased α2-globulin level at the second stage of the clinical trial was noted in 60% of the CG students and, by the end of observation, in 10% of students. A reduced level of β-globulins at the initial stage was registered in 20% of the students and, after 45 days, in 40%. A slight drop in β-globulin (though within the norm) relative to the baseline values occurred in 60–70% of the subjects. At the beginning of the observation, 10% of students had γ-globulin higher than the norm; 30% of subjects showed below-the-norm γ-globulin values. After 15 days, no subjects had high levels of γ-globulins, and 80% of subjects had much lower γ-globulin values compared with the initial ones. By the end of the observation, the number of students having low γ-globulin reached 50%, and its decrease from the initial value was noted in nine subjects out of ten.

In TG 1, the level of albumin did not exceed the reference limits, and the number of students having an increased level of albumin relative to the initial value did not exceed one third at each stage of the observation period. At the beginning of the observation, the level of α1-globulins was higher than normal in 45% of the subjects. At the interim stage, it was above the norm in 30% of the subjects, and by the end of observation, it was higher only in one out of ten students. At the 2nd and 3rd stages of observation, 60% and 80% of the students demonstrated a decrease in the level of α1-globulin relative to the initial value. The level of α2-globulins at the beginning of observation was higher than the norm in 60% of students and, at the end of observation, only in 20%. A drop relative to the initial individual data was registered in 65% and 60%, respectively, at stages 2 and 3. An increase in β-globulins was noted in 65% of the students by the end of the protein–plant FP intake, and one third of the subjects retained this level of content until the end of the final stage of the study. At the initial stage, the level of γ-globulin was reduced in 35% of the participants. By the end of the course of treatment with functional foods, the percentage of subjects with reduced γ-globulin levels was 25%, and after another 30 days (46th day) it reached 30%.

There were practically no differences in the albumin fraction between TG 2 and the CG. However, after 15 and 45 days, an increase in albumin was registered in 80% and 75% of the subjects, respectively. Increased α1-globulin levels were noted in 40% and 30% of the subjects. However, the level of α1-globulin decreased in 60% of the individuals in this group. No differences in α2-globulin levels were found in the TG 2 compared to the CG. At the same time, a 15–20% decrease in the level of β-globulins in relation to the initial values was registered in a smaller number of subjects. The levels of γ-globulins gradually rose in a larger number of subjects during the stages of observation, while a decrease was noted in a smaller number of subjects than in the CG.

Red blood cell counts at all stages of observation were within the normal range (Table 5). From these data it can be seen that the number of erythrocytes, hemoglobin concentration, and hematocrit did not change significantly during the observation period. However, the mean erythrocyte volume considerably increased in each group. The mean erythrocyte hemoglobin level did not change in TG 1, while in TG 2 the mean erythrocyte hemoglobin decreased by 1.1% ($p < 0.05$) at the end of the final stage of the study. The same response was noted in the CG.

The average hemoglobin level decreased markedly at the second and third stages of observation in all students. The production of erythrocytes increased in the TG 1 group by 2.5% ($p < 0.05$) after a course of treatment with functional foods; at the end of the observation period, it was 2.9% higher than the initial value ($p < 0.05$). TG 2 subjects also showed a 2.3% increase in erythrocyte production ($p < 0.05$), but at the end of the clinical trial, there was no significant deviation from the initial value. No such patterns were observed in the CG.

Table 5. Dynamics of erythrocyte count and other indicators in biomaterial samples during treatment with functional products (average).

Indicators (Reference Values) [47]	Observation Period						
	Content			Dynamics, %		*p*	
	1	2	3	2	3	2	3
Erythrocytes, 4.3–5.7 × 10^{12} cells/L:							
treatment 1	5.13 ± 0.08	5.02 ± 0.05	5.01 ± 0.05	−2.2	−2.4	0.139	0.959
treatment 2	4.90 ± 0.06	5.00 ± 0.06	4.94 ± 0.06	+2.0	+0.8	0.125	0.278
control	5.07 ± 0.07	5.08 ± 0.08	5.00 ± 0.09	+0.2	−1.4	0.866	0.439
Hemoglobin, 3.2–17.3 g/dL:							
treatment 1	14.48 ± 0.15	14.74 ± 0.17	14.60 ± 0.13	+1.8	+0.8	0.156	0.275
treatment 2	14.48 ± 0.16	14.74 ± 0.17	14.60 ± 0.14	+1.8	+0.8	0.156	0.181
control	15.39 ± 0.18	15.04 ± 0.17	15.07 ± 0.19	−2.3	−2.1	0.059	0.132
Hematocrit, 0.39–0.49 L/L:							
treatment 1	0.44 ± 0.01	0.43 ± 0.01	0.43 ± 0.01	−2.3	−2.3	0.123	0.963
treatment 2	0.42 ± 0.01	0.43 ± 0.01	0.42 ± 0.01	+9.5	±0.0	0.356	0.963
control	0.43 ± 0.01	0.44 ± 0.01	0.43 ± 0.01	+2.3	±0.0	0.069	0.131
Average erythrocyte volume, 80–95 fl:							
treatment 1	85.11 ± 0.96	85.88 ± 0.65	85.86 ± 0.60	+0.9	+0.8	0.009	0.003
treatment 2	85.33 ± 0.64	85.88 ± 0.67	86.33 ± 0.63	+0.7	+1.2	0.001	0.004
control	85.11 ± 0.90	85.95 ± 0.99	86.15 ± 0.93	+0.9	+1.2	0.021	0.009
The average amount of hemoglobin in an erythrocyte, 27–31 pg:							
treatment 1	29.60 ± 0.25	29.53 ± 0.26	29.59 ± 0.26	−0.3	−0.1	0.466	0.998
treatment 2	30.35 ± 0.25	30.00 ± 0.23	30.10 ± 0.29	−1.2	−1.1	0.003	0.002
control	30.35 ± 0.39	30.00 ± 0.37	30.10 ± 0.23	−1.2	−1.1	0.028	0.037
Relative width of distribution of erythrocytes by volume, standard deviation 42 ± 5 fl:							
treatment 1	42.93 ± 0.60	44.04 ± 0.55	44.19 ± 0.59	+2.5	+2.9	0.001	0.003
treatment 2	42.50 ± 0.27	43.49 ± 0.37	42.96 ± 0.36	+2.3	+1.1	0.001	0.116
control	42.07 ± 0.50	42.39 ± 0.68	42.46 ± 0.69	+0.7	+1.1	0.722	0.024
Relative erythrocyte distribution width, coefficient of variation 11.5–14.5%:							
treatment 1	13.55 ± 0.16	13.55 ± 0.16	13.54 ± 0.16	−0.4	0.111	0.111	0.081
treatment 2	13.14 ± 0.08	13.18 ± 0.09	13.55 ± 0.17	+0.3	0.712	0.710	0.525
control	13.15 ± 0.09	13.18 ± 0.10	13.05 ± 0.13	+0.2	0.713	0.710	0.125

1—Beginning (21 days); 2—End (37 days); 3—45 days into observation (67th day). Data presented as a mean ± SD ($n = 3$). Values did not have significant differences ($p > 0.05$) as assessed by post hoc test.

In TG 1, white blood cell count increased by 12.0% ($p < 0.05$) by the end of the course and the level of content remained unchanged until the end of the final stage of the study (Table 6). In TG 2 and CG, no relevant changes in the white blood cell count were registered.

These tests also showed that, during the period of observation, the number of lymphocytes increased. In TG 1 there was a considerable rise in the number of lymphocytes by 9.4% ($p < 0.05$), which continued up until the end of the final stage of the study (by 8.6% ($p < 0.05$). In TG 2 and CG, a significant increase of this parameter in comparison with the initial value was registered only at the end of observation—22.7% (TG 2, $p < 0.05$) and 24.9% (CG, $p < 0.05$). The relative level of lymphocytes (fraction of total leukocytes, %) in TG 1 at the second and third stages of observation was more significant than that at the initial stage, exceeding it by 14.3% ($p < 0.05$) and 36.7% ($p < 0.05$), respectively. In TG 2, a strong upward tendency was identified, reaching a 15.1% ($p < 0.05$) increase in the level of lymphocytes at the end of observation. There were no statistically significant differences in the CG data.

Table 6. Dynamics of leukocyte content in biomaterial samples during treatment with functional products (average).

Indicators (Reference Values) [47]	Observation Period						
	Content			Dynamics, %		*p*	
	1	2	3	2	3	2	3
Leukocytes, 4.2–9 × 10 ^{9} cells/L:							
treatment 1	6.96 ± 0.22	7.86 ± 0.30	7.69 ± 0.21	+12.0	+10.4	0.046	0.023
treatment 2	6.86 ± 0.22	6.98 ± 0.28	7.62 ± 0.37	+1.7	+11.0	0.541	0.495
control	6.57 ± 0.48	6.31 ± 0.34	7.95 ± 0.35	−3.9	+24.9	0.641	0.139
Lymphocytes, 1.5–4.0 × 10^9 L:							
treatment 1	6.96 ± 0.22	7.86 ± 0.30	7.69 ± 0.21	+12.0	+10.4	0.046	0.023
treatment 2	6.86 ± 0.22	6.98 ± 0.28	7.62 ± 0.37	+1.7	+11.0	0.541	0.495
control	6.57 ± 0.48	6.31 ± 0.34	7.95 ± 0.35	−3.9	+24.9	0.641	0.139
Relative lymphocyte count, 25–40%:							
treatment 1	33.65 ± 1.30	38.46 ± 1.50	40.60 ± 1.60	+14.3	+36.7	0.001	0.001
treatment 2	30.16 ± 2.30	33.60 ± 1.30	34.72 ± 1.40	+11.4	+15.1	0.243	0.004
control	30.77 ± 2.10	33.85 ± 2.30	35.10 ± 1.80	+10.0	+14.0	0.312	0.065
Neutrophils, 2.0–7.7 × 10^9 L:							
treatment 1	3.58 ± 0.19	4.20 ± 0.26	4.00 ± 0.19	+17.3	+11.7	0.001	0.059
treatment 2	3.51 ± 0.27	3.63 ± 0.25	3.74 ± 0.22	+3.4	+6.5	0.988	0.984
control	4.24 ± 0.34	3.80 ± 0.22	4.11 ± 0.34	−10.4	−3.1	0.295	0.778
Relative neutrophil count, 42–72%:							
treatment 1	50.70 ± 1.20	51.47 ± 1.40	51.63 ± 1.54	+1.5	+1.8	0.988	0.966
treatment 2	57.29 ± 2.66	54.10 ± 1.33	52,85 ± 1.86	−5.6	−9.4	0.217	0.026
control	54.50 ± 1.86	53.20 ± 2.30	52.67 ± 1.90	−4.3	−3.4	0.678	0.575
Combination of monocytes, basophils, eosinophils 0.2–0.8 × 10 ^{9} L:							
treatment 1	0.81 ± 0.07	0.82 ± 0.07	0.9 5 ± 0.09	+1.2	+17.2	0.871	0.001
treatment 2	0.89 ± 0.05	0.86 ± 0.07	0.92 ± 0.06	−3.4	+3.3	0.512	0.347
control	0.99 ± 0.11	0.77 ± 0.11	0.98 ± 0.14	−22.3	−1.2	0.125	0.952
Relative count of a combination of monocytes, basophils, eosinophils, 5–10%:							
treatment 1	11.98 ± 0.70	11.56 ± 0.80	12.00 ± 0.80	−3.5	+0.1	0.655	0.785
treatment 2	12.53 ± 0.70	12.28 ± 0.70	12.42 ± 0.90	−2.0	−0.9	0.544	0.845
control	14.69 ± 1.48	12.92 ± 2.00	12.31 ± 1.30	−12.1	−16.2	0.386	0.514

1—Beginning (21 days); 2—End (37 days); 3—45 days into observation (67th day). Data presented as a mean ± SD ($n = 3$). Values did not have significant differences ($p > 0.05$) as assessed by post hoc test.

Neutrophils in TG 1 rose by 17.3% ($p < 0.05$) at the end of the final stage of the study within the normal limits of nutritional intervention. At the same time, at the end of the final stage of the study, this value changed slightly compared to the initial one ($p < 0.05$). The number of neutrophils during the stages of observation in the TG 2 and CG groups did not change significantly, remaining within normal limits. Their relative number in TG 1 and CG did not change during the stages of observation, and in TG 2 this parameter decreased by 9.4% ($p < 0.05$) by the end of observation.

In TG 1, there was a considerable 17.2% ($p < 0.05$) increase in the level of monocytes, basophils, and eosinophils at the end of the final stage of the study. The relative values of these blood-forming cells did not change during the stages of observation.

By the end of the course the leukocyte count increased 12.0% ($p < 0.05$) in TG 1 and remained unchanged ($p < 0.05$) until the end of the final stage of the study (Table 6). In TG 2 and CG, no statistically relevant changes were registered. During the period of observation, there was an increase in the number of lymphocytes. In TG 1, a marked increase in the number of lymphocytes by 9.4% ($p < 0.05$) was registered, which continued up to the end

of the final stage of the study (8.6 % ($p < 0.05$)). In TG 2 and CG, this parameter grew in comparison with the initial value, however, this growth was registered only at the end of the observation—22.7% (TG 2, $p < 0.05$) and 24.9% (CG, $p < 0.05$), respectively.

The relative count of lymphocytes (fraction of total leukocytes, %) in TG 1 at the second and third stages of observation was 14.3% ($p < 0.05$) and 36.7% ($p < 0.05$) higher than at the initial stage, respectively. TG 2 showed a 15.1% ($p < 0.05$) increase by the end of observation. No statistically relevant changes were registered in the CG.

Neutrophils in TG 1 rose by 17.3% ($p < 0.05$) regarding the reference values by the end of the period of the nutritional intervention. However, until the end of the final stage of the study, this value did not differ significantly from the baseline ($p < 0.05$). The number of neutrophils during the stages of observation in the TG 2 and CG groups did not change, remaining within the norm. Their relative number in the TG 1 and CG was stable during all stages of observation, whereas this rate in the TG 2 group decreased slightly by 9.4% ($p < 0.05$) by the end of the final phase of the study.

TG 1 showed an upward trend in the combination count of monocytes, basophils, and eosinophils, which at the end of the final phase of the study was 17.2% ($p < 0.05$). The relative value of these blood-forming cells did not change during the stages of observation.

The highest platelet counts ($(243.9 \pm 8.50) \times 10^9$ cells/L) were observed for group 3 after the protein–vegetable product TG 1 diet (Table 7). The highest mean platelet volume was observed in control group 1 (10.82 ± 0.38 fl).

Table 7. Dynamics of thrombocyte content in biomaterial samples during treatment with functional products (average).

Indicators (Reference Values) [47]	Observation Period						
	Content			Dynamics, %		*p*	
	1	2	3	2	3	2	3
Platelets, 180–400 × 10^9 cells/L:							
treatment 1	231.80 ± 6.50	242.97 ± 6.90	243.9 ± 8.50	+4.8	+5.6	0.042	0.371
treatment 2	229.45 ± 10.40	245.40 ± 16.80	235.95 ± 13.20	+6.9	+2.7	0.044	0.201
control	221.10 ± 11.20	229.80 ± 9.30	240.2 0 ± 11.40	+3.9	+8.6	0.092	0.059
Relative PDW [a], 15–17%:							
treatment 1	13.55± 0.20	13.05 ± 0.20	12.84 ± 0.20	−3.7	−5.3	0.022	0.212
treatment 2	14.00 ± 0.40	13.45 ± 0.37	13. 30 ±0.40	−4.0	−5.0	0.032	0.017
control	13.97± 0.70	13.78 ± 0.80	14.35 ± 0.90	−1.4	+2.7	0.957	0.474
Average platelet volume 7.5–11 fl:							
treatment 1	10. 54 ± 0.19	10.33 ± 0.23	10.26 ± 0.21	−2.0_	−2.7	0.064	0.015
treatment 2	10. 68 ± 0.16	10.41 ± 0.16	10.23 ± 0.17	−2.5	−4.3	0.006	0.002
control	10. 82 ± 0.38	10.54 ± 0.39	10.53 ± 0.38	−2.6	−2.5	0.073	0.012
p-LCR, 13–43%:							
treatment 1	34.29 ± 1.02	32.49 ± 1.05	32.12 ± 1.00	−5.3	−6.3	0.042	0.031
treatment 2	30. 78 ± 1.30	28. 77 ± 1.37	27.5 0 ±1. 40	−6.6	−10.7	0.002	0.003
control	32.15 ± 3.00	29.99 ± 3.10	29.94 ± 2.90	−6.8	−6.9	0.059	0.036

[a] PDW—platelet volume distribution width. 1—Beginning (21 days); 2—End (37 days); 3—45 days into observation (67th day). Data presented as a mean ± SD ($n = 3$). Values did not have significant differences ($p > 0.05$) as assessed by post hoc test.

In the CG, serum immunoglobulin A was initially at the lower limit of the norm, decreasing 37.5% ($p < 0.05$) at the end of the final stage of the study. At each stage of observation, it was lower than the norm in 90.0% of CG subjects (Table 8). The mean IgM index in the CG was initially within the normal range. After 15 days, the mean value was dramatically higher (80.4% ($p < 0.05$)) than the initial value. In the CG it rose in 100.0% of subjects, and in 30.0% of students its value reached the upper limit of the norm. After

45 days, the mean IgM value was 54.0% higher than that of the baseline ($p < 0.05$). IgG at the baseline and at the observation stages was within its reference values.

Table 8. Dynamics of serum immunoglobulin content in biomaterial samples during treatment with functional products (average), g/L.

Indicators (Reference Values) [47]	Observation Period						
	Content			Dynamics, %		p	
	1	2	3	2	3	2	3
IgA, 0.9–4.5:							
treatment 1	0.938 ± 0.100	1.034 ± 0.120	0.902 ± 0.110	+10.2	−3.9	0.266	0.656
treatment 2	0.881 ± 0.110	0.862 ± 0.100	0.871 ± 0.110	−2.2	−1.2	0.492	0.836
control	0.685 ± 0.090	0.740 ± 0.090	0.428 ± 0.030	+8.0	−37.5	0.166	0.026
IgM, 0.6–3.7:							
treatment 1	1.258 ± 0.140	1.176 ± 0.140	1.803 ± 0.110	−6.6	+43.3	0.176	0.009
treatment 2	0.930 ± 0.110	1.230 ± 0.120	1.565 ± 0.170	+32.2	+68.2	0.008	0.029
control	0.870 ± 0.070	1.570 ± 0.360	1.340 ± 0.160	+80.4	+54.0	0.007	0.006
IgG, 8.0–17.0:							
treatment 1	10.820 ± 1.190	11.980 ± 1.280	15.540 ± 1.390	+10.7	+43.3	0.556	0.001
treatment 2	9.090 ± 0.790	12.260 ± 1.670	12.590 ± 1.390	+34.8	+38.5	0.001	0.001
control	15.920 ± 1.020	15.460 ± 1.390	11.490 ± 1.800	−3.1	−27.8	0.858	0.139

1—Beginning (21 days); 2—End (37 days); 3—45 days into observation (67th day). Data presented as a mean ± SD ($n = 3$). Values did not have significant differences ($p > 0.05$) as assessed by post hoc test.

In TG 1 and TG 2, mean IgA values were within a normal range and did not change much during the stages of observation. IgM was within the norm in all TG 1 subjects. However, it increased 43.3% ($p < 0.05$) after 45 days of observation, whereas TG 2 showed a 32.2% ($p < 0.05$) increase of this parameter after 15 days of observation and a 68.2% ($p < 0.05$) increase at the end of the observation period.

In TG 1 the content of IgG increased compared to reference limits by 43.3% ($p < 0.05$) at the end of the final stage of the study, while TG 2 demonstrated a 34.8% ($p < 0.05$) increase in IgG on the 15th day of observation and a 38.5% ($p < 0.05$) increase at the end of observation.

The testosterone level in the CG during all the stages of observation was within the normal range (Table 9). However, after only 15 days of the study there was a substantial drop in its level by 29.6% ($p < 0.05$). It continued to fall and, after 45 days, the decrease was 32.8% ($p < 0.05$). It is noteworthy that at each stage of observation 100% of the subjects showed a downward tendency in the level of testosterone. In TG 1, the level of the hormone decreased only towards the end of the observation period and the decrease was not significant. In TG 1, 40.0% of subjects had an increase in testosterone by the end of the FP course, and in 25.0% of subjects, this increase persisted for one month after the end of the FP course. A minor drop in testosterone was also observed in TG 2 students. It decreased in all the subjects but was still within the normal range.

Blood cortisol in CG subjects was above the reference limits in 100.0% of subjects during the first and second examinations. Individually, it was initially high in 50.0% of students and, after 15 days, in 70.0%. After another month, its mean value was within the upper limit of the norm, but 60.0% of the individuals examined had a cortisol level higher than the upper reference value. In TG 1 and TG 2, blood cortisol was within normal limits during the whole period of observation. However, at the initial stage, cortisol was above the norm in 40.0% of TG 1 subjects and, after 15 days, in 50.0%; in TG 2 it was above the norm in 40.0% and 60.0% of students, respectively. After 45 days, only 25.0% of TG 1 subjects and 40.0% of TG 2 subjects had an increased cortisol level.

Table 9. Dynamics of hormone content in biomaterial samples (average), nmol /L.

Indicators (Reference Values) [47]	Observation Period						
	Content			Dynamics, %		p	
	1	2	3	2	3	2	3
Testosterone, 8.72–38.17:							
treatment 1	24.73 ± 1.98	25.53 ± 1.65	21.19 ± 1.77	+3.2	−14.3	0.343	0.191
treatment 2	27.40 ± 1.20	18.10 ± 1.30	17.4 ± 1.10	−34.0	−36.5	0.311	0.291
control	26.18 ± 1.17	18.43 ± 1.80	17.59 ± 0.90	−29.6	−32.8	0.022	0.019
Cortisol, 200.0–700.0:							
treatment 1	636.8 ± 31.8	673.3 ± 29.6	672.0 ± 35.0	+5.8	+5.5	0.454	0.361
treatment 2	680.4 ± 21.2	681.5 ± 22.7	654.6 ± 31.2	+0.1	−3.8	0.791	0.276
control	750.6 ± 47.9	761.0 ± 38.4	685.6 ± 28.5	+1.4	−8.7	0.871	0.241

1—Beginning (21 days); 2—End (37 days); 3—45 days into observation (67th day). Data presented as a mean ± SD ($n = 3$). Values did not have significant differences ($p > 0.05$) as assessed by post hoc test.

Vitamin A was above the reference limits in subjects of all groups (Table 10)—no individuals with low levels of the vitamin were found. By the end of the FP course, a considerable increase in the mean values relative to the baseline data was observed; in TG 1 it was 21.3% ($p < 0.05$) and in TG 2—15.5% ($p < 0.05$). After another 30 days, each group retained these high levels. However, in TG 1 the concentration of vitamin A was 23.6% ($p < 0.05$) higher than the baseline, and in TG 2 a slight drop was observed, but the value was still 10.9% ($p < 0.05$) higher than the baseline. CG subjects showed no changes.

Table 10. Dynamics of vitamin content in biomaterial samples (average).

Indicators (Reference Values) [47]	Observation Period						
	Content			Dynamics, %		p	
	1	2	3	2	3	2	3
A, µg/mL (0.3–0.6):							
treatment 1	0.89 ± 0.04	1.08 ± 0.04	1.10 ± 0.03	+21.3	+23.6	0.042	0.002
treatment 2	0.91 ± 0.04	1.05 ± 0.04	1.01 ± 0.04	+15.5	+10.9	0.040	0.004
control	0.83 ± 0.05	0.91 ± 0.05	0.91 ± 0.04	+9.6	+9.6	0.262	0.270
E, µg/mL (8–18):							
treatment 1	7.86 ± 0.34	8.70 ± 0.54	9.19 ± 0.41	+10.7	+16.9	0.02	0.004
treatment 2	8.43 ± 0.39	8.63 ± 0.54	10.08 ± 0.4	+2.4	+19.5	0.736	0.002
control	7.75 ± 0.43	6.78 ± 0.39	6.78 ± 0.53	−12.5	−12.5	0.019	0.039
B_1, µg/mL (7–14):							
treatment 1	21.40 ± 0.90	20.60 ± 1.00	20.60 ± 1.00	−3.8	−3.6	0.008	0.061
treatment 2	15.70 ± 0.06	14.50 ± 0.67	18.60 ± 1.08	−8.1	+18.4	0.069	0.001
control	19.70 ± 1.30	16.36 ± 0.85	17.60 ± 1.00	−17.0	−10.7	0.001	0.003
B_2, µg/% (10–50):							
treatment 1	6.10 ± 0.03	6.90 ± 0.41	7.83 ± 0.20	+13.5	+28.3	0.033	0.001
treatment 2	6.81 ± 0.23	7.04 ± 0.33	7.29 ± 0.32	+3.3	+7.3	0.123	0.017
control	5.91 ± 0.40	5.85 ± 0.30	6.08 ± 0.30	−0.9	+2.8	0.621	0.195

1—Beginning (21 days); 2—End (37 days); 3—45 days into observation (67th day). Data presented as a mean ± SD ($n = 3$). Values did not have significant differences ($p > 0.05$) as assessed by post hoc test.

Fifteen days into observation, in the CG vitamin E was 12.5% ($p < 0.05$) lower than the baseline value and remained stable until the end of observation. Initially, a decreased vitamin E concentration was found in 40.0% of the subjects and, after 15 days of observation, in 80.0%, remaining the same in 80.0% of subjects up to the end of observation. Initially, in TG 1, vitamin E concentration was also below the norm, but after a course of taking

functional foods and at the end of the final stage of the study it was within the normal range, having grown 10.7% ($p < 0.05$) and 16.9% ($p < 0.05$), respectively. At the beginning of the clinical trial, only 40.0% of subjects had normal levels of this vitamin. During the FP course, 65.0% of subjects reached the norm, and at the end of the observation 70.0% of individuals had normal levels of this vitamin. Other subjects also showed positive dynamics; however, the concentration of vitamin E did not reach the normal values. In TG 2, the initial mean value of this vitamin was well within the normal range (in 55.0% of subjects). During the FP course, the mean value did not change significantly, although the percentage of individuals having the normal level of vitamin E rose to 65.0%, and an increase in serum vitamin E was noted in 45.0% of subjects. By the end of the observation, normal levels of this vitamin were registered in 80.0% of the TG 2 students, with an increase of 19.5% ($p < 0.05$).

The concentration of vitamin B1 in all groups was above the reference limits. Vitamin B1 levels in the CG dwindled during all periods of observation, with a 17.0% ($p < 0.05$) decrease after the course of FP intake and a 10.7% ($p < 0.05$) decrease below the baseline values at the end of the observation. In TG 1 no such dynamic was noted. However, in TG 2, the concentration of vitamin B1 fell by 18.4% ($p < 0.05$) by the end of the final stage of the study.

The tests showed that, initially, most subjects of all groups had vitamin B2 deficiency. After the FP intake, TG 1 subjects showed a rise of 13.5% ($p < 0.05$) and, by the end of the final stage of the study, 28.3% ($p < 0.05$), though still not reaching the normal value. A rise in the concentration of vitamin B2 by stages of observation was noted in 25.0% and 75.0% of subjects, respectively. A slight rise of 7.0% ($p < 0.05$) in the mean value of the vitamin was also noted in TG 2 by the end of observation. The increase in the concentration of this vitamin was registered in 35.0% (21 days) and in 60.0% (37 days) of students.

When assessing blood mineral levels, we found that the concentration of iron in the CG and TG 2 did not change much at any stage of the observation and remained within the normal range (Table 11). In TG 1 a significant 15.2% rise in iron ($p < 0.05$) was observed at the end of the FP treatment, which continued up to at the end of the final stage of the study (a 20.8% increase ($p < 0.05$)). At the initial stage, 35.0% of the subjects had iron levels below the norm, but at the end of the study and at the end of the observation period, just 5.0% of students still retained their low initial value. In TG 2, at the beginning of the FP course 15.0% of the subjects had their iron level below the norm, but at the end of the FP intake only 5.0% had iron deficiency. After 45 days, 100.0% of subjects had normal iron values.

In the CG, magnesium was within the norm during all periods of observation, but after 15 days of adaptation, its concentration fell to much lower than the initial value, with a massive drop of 22.6% ($p < 0.05$), and after 45 days of adaptation it was 12.8% lower ($p < 0.05$). A gradual decrease by stage of observation relative to the initial values was noted in 100.0% and 80.0% of the observation groups. In TG 1, at the beginning of the study, magnesium was generally below the norm, and 75.0% of subjects showed considerable deviations. At the end of the FP course, there was a 6.3% increase ($p <0.05$) in magnesium, and the percentage of individuals with low or decreased magnesium levels went down to 40.0%. By the end of observation, 100.0% of subjects had adequate magnesium concentrations and its average value increased by 34.9% ($p < 0.05$). In TG 2, magnesium was within the norm, but after 15 days its concentration decreased 14.3% ($p < 0.05$), and, by the end of the final stage of the study, it was the same as the baseline value. At the beginning of the study, 25.0% of the individuals had magnesium levels lower than the norm. Later on, during all stages of observation, it was within normal limits in all those tested.

Table 11. Dynamics of mineral content in biomaterial samples (average).

Indicators (Reference Values) [47]	Observation Period						
	Content			Dynamics, %		*p*	
	1	2	3	2	3	2	3
Iron, mM/L (11.6–31.3):							
treatment 1	13.60 ± 0.73	15.67 ± 1.00	16.43 ± 1.00	+15.2	+20.8	0.029	0.021
treatment 2	16.10 ± 0.85	17.48 ± 0.87	17.02 ± 0.85	+8.5	+5.7	0.331	0.464
control	14.20 ± 1.15	16.84 ± 0.86	16.65 ± 0.93	+18.5	+17.2	0.066	0.139
Magnesium, mM/L (0.66–1.07):							
treatment 1	0.63 ± 0.01	0.67 ± 0.01	0.85 ± 0.01	+6.3	+34.9	0.002	0.001
treatment 2	0.91 ± 0.05	0.78 ± 0.01	0.86 ± 0.01	−14.3	−5.5	0.007	0.233
control	0.99 ± 0.02	0.77 ± 0.01	0.86 ± 0.01	−22.6	−12.8	0.007	0.016
Phosphorus, mM/L (0.87–1.45):							
treatment 1	1.12 ± 0.03	1.29 ± 0.03	1.35 ± 0.03	+16.1	+20.5	0.001	0.001
treatment 2	1.17 ± 0.03	1.12 ± 0.03	1.24 ± 0.05	−4.3	+5.9	0.231	0.178
control	1.26 ± 0.03	1.13 ± 0.03	1.30 ± 0.03	−10.3	+3.4	0.041	0.153
Calcium, mM/L (2.15–2.57):							
treatment 1	2.61 ± 0.02	2.59 ± 0.01	2.58 ± 0.02	−0.8	−1.1	0.123	0.135
treatment 2	2.57 ± 0.01	2.53 ± 0.02	2.50 ± 0.02	−1.6	−2.7	0.066	0.085
control	2.56 ± 0.02	2.51 ± 0.02	2.54 ± 0.20	−1.9	−0.8	0.344	0.507
Potassium, mM/L (3.6–5.5):							
treatment 1	5.19 ± 0.12	5.04 ± 0.10	5.25 ± 0.11	−2.9	+1.1	0.288	0.735
treatment 2	5.45 ± 0.12	5.11 ± 0.11	5.13 ± 0.13	−6.3	−5.9	0.064	0.356
control	5.31 ± 0.15	5.10 ± 0.08	5.17 ± 0.09	−4.0	−2.7	0.678	0.313
Sodium, mM/L (135–150):							
treatment 1	145.60 ± 2.31	149.60 ± 0.35	150.50 ± 0.48	+2.7	+3.3	0.135	0.552
treatment 2	148.00 ± 0.45	148.70 ± 0.19	149.00 ± 0.18	+0.5	+2.0	0.065	0.356
control	149.80 ± 0.57	147.60 ± 0.36	150.00 ± 0.38	−1.5	+0.1	0.378	0.065
Chlorine, mM/L (97–108):							
treatment 1	100.20 ± 1.11	100.30 ± 0.40	104.30 ± 0.40	+0.1	+4.1	0.222	0.078
treatment 2	100.8 ± 0.43	100.10 ± 0.37	101.40 ± 0.77	−0.7	+0.6	0.147	0.177
control	102.10 ± 0.47	103.40 ± 0.48	102.80 ± 0.41	+1.3	−0.9	0.074	0.099

1—Beginning (21 days); 2—End (37 days); 3—45 days into observation (67th day). Data presented as a mean ± SD ($n = 3$). Values did not have significant differences ($p > 0.05$) as assessed by post hoc test.

Initially, the concentration of phosphorus was within the reference values. In the CG, after 15 days of the study, there was a 10.3% ($p < 0.05$) decrease in the concentration of the mineral. By the end of the observation, it did not differ from the initial value. In TG 1, a remarkable 16.1% ($p < 0.05$) increase in phosphorus levels was observed after the FP intake, which continued up to the end of the final stage of the study (increase by 20.5% ($p < 0.05$)). In TG 2, the concentration of phosphorus did not change during the stages of observation.

The content of potassium, calcium, sodium, and chlorine in both groups of students did not differ significantly from the control values. No statistically significant changes in the concentrations of these minerals were observed ($p > 0.05$) in both groups. However, sodium, which in the CG initially and after 15 days of observation was within the norm, exceeded it by the end of the observation period. There was an upward tendency in the values of this mineral at all stages of observation. At the beginning, it was higher than the norm in 30.0% of subjects, and by the end of observation it was up in 70.0% of subjects. In TG 1, sodium was within the normal range. Initially, it was higher than the norm in 30.0% of subjects, by the end of the course only in 5.0%, and at the end of the observation period just in 10.0%. The sodium level in TG 2 did not differ much at different stages of

observation and was within the normal range. Initially, it was higher than the norm in 15.0% of students, after 15 days in 5.0%, and after 45 days in 30.0% of those observed.

In the CG, at the beginning and after 15 days of observation, chlorine was within the norm, and after 45 days, the mean value was above the norm. By the second stage of observation, there was an increase in the chlorine concentration relative to the initial value in 60.0% of the subjects and, by the end of the final stage of the study, in 90.0%. In TG 1 there was a 1.3% increase in the level of chlorine by the end of the FP intake. However, by the end of the final stage of the study, it was significantly lower than the initial value (by 1.7% but still within the norm). In 100.0% of subjects, it remained normal during all stages. In TG 2, by the end of observation the concentration of chlorine was 6.5% higher than the baseline value. No individuals exceeded the norm during the stages.

4. Discussion

Adaptation is one of the main concepts in biology. It is the result (and/or process) of interaction between living organisms and the environment, which leads to their optimal adaptation to life and activity [60]. Adaptation of a first-year student is a complex, dynamic, multilevel, and multifaceted process of restructuring the needs and motivational sphere and the set of existing skills, abilities, and habits in accordance with new tasks, goals, perspectives, and conditions for their implementation. A significant adaptive situation arises as a result of changes in the conditions of learning when entering university. This aspect of adaptation is called didactic adaptation, which includes adaptation to new forms and methods of work and adaptation to new forms of control [60]. The adaptation of first-year students is closely tied to their academic success.

The student's psychophysiological and psychological characteristics are restructured during the adaptation process. The specifics of the adaptation of university students are determined by the conditions of training and their individual characteristics. The most important component of adaptation is nutrition [60].

This study was carried out during the most stressful period for students—the beginning of their university education. This is a period of abrupt transition from secondary to tertiary education, from living with their parents to living in a student hall of residence, from home-cooked food to organized catering. This period was also accompanied by increased physical activity. In addition, nine out of ten students who participated in the clinical trial had to adapt to new climatic conditions. As is known, the climate of the Baltic Sea may have a negative impact on health [61]. The cumulative negative impact of environmental factors weakens the adaptive capacity of the body, particularly so during the initial period of adaptation and acclimatization. Naturally, there is a need for mitigation and correction of the disadaptation reactions of the body.

Numerous studies have proven the effectiveness of functional products manufactured by cryogenic technology for the prevention of diseases. More importantly, functional food products are essential for compensating vitamin and mineral deficiencies, increasing the body's resistance to disease, and boosting the metabolism of nutrients [62–66]. In the present study, we opted for plant and protein–plant raw materials.

Cortisol is a primary stress hormone [67,68]. There is growing evidence of its association with increased anxiety, depression, and low physical activity [69,70]. In this clinical investigation, signs of cortisol stress were registered in each group of subjects. However, in TG 1 they were much less pronounced compared with TG 2, 25.0% versus 40.0%, respectively. A decrease in testosterone within the normal range was observed in the students of both groups, but its mean values did not change significantly. A similar effect is reported for athletes recovering from overtraining syndrome (a disorder of reduced performance and fatigue caused by excessive training, lack of recovery, and poor quality of sleep) [71]. Interestingly, in TG 1 testosterone levels increased by the end of the FP intake in 40.0% of subjects and remained the same in 25.0% for one month of observation; however, they decreased in all subjects of TG 2. Therefore, we can assume that the students of TG 1 had a more efficient metabolism of nutrients compared with TG 2.

This assumption was also confirmed by the increase in the concentration of vitamins. For example, the level of vitamin A in TG 2 by the end of the FP intake increased more significantly than in TG 1 (21.3% versus 15.5%). After 30 days, in TG 1, it was 23.6% higher than the initial value. In TG 2, there was a decrease relative to the same value after the FP intake, i.e., in the first case, the effect was much longer. Vitamin A deficiency is more common during infection, and vitamin A supplements are known to reduce severe morbidity and mortality from infectious diseases [72]. Vitamin E deficiency can cause many serious conditions such as hemolytic anemia, cerebellar ataxia, and cognitive difficulties [73]. During different stages of the clinical trial, vitamin E in TG 1 was 10.7% and 16.9% higher than the baseline value. It increased to the normal level in 70% of subjects and a rise was registered in all subjects. In TG 2, an increase in this vitamin was noted in 45% of subjects, and an increase to the normal level was found in 10%.

Thiamine and riboflavin are water-soluble B vitamins that function as cofactors in biochemical reactions and are vitally important for normal growth and development, healthy skin, proper functioning of the nervous system and heart, as well as for red blood cell formation. Their insufficiency can be a result of alcohol abuse, malabsorption syndromes, strict veganism, and malnutrition [74]. In TG 1, 100% of students had a low level of vitamin B_1. The FP intake resulted in the normalization of the concentration of vitamin B_1 in 25.0% of subjects; 40.0% of students were within the normal range at the end of observation. In TG 2, an increase in vitamin levels after the FP treatment was observed in 40% of subjects, but after one month, this percentage decreased to just 10.0%. After the FP therapy, there was an increase in the concentration of vitamin B_2 in TG 1: an increase in 25.0% and 75.0% of subjects, respectively, at different stages of observation. In TG 2, an upward positive dynamic was noted in 35.0% and 60.0% of the students at different stages of observation.

Minerals are the most important micronutrients in the human body. Changes in their levels may be a consequence of various health conditions. For instance, intensive physical exercise can cause a decrease in the concentration of Fe, Mg, and *p* in erythrocytes [75]. Depression, especially in women, is usually associated with deficiencies of these minerals [76]. A significant increase in the concentration of iron (a 15.2% rise) was noted in TG 1 at the end of the FP therapy and continued up to the end of the observation (a 20.8% rise). In TG 2, these dynamics were not observed. However, in 15.0% of subjects initially having low iron levels, the concentration of iron was restored to normal. In TG 1, the FP intake resulted in a considerable increase in the level of magnesium though, initially, students of the group had lower magnesium levels. In TG 2, there were only minor changes within the norm. The concentration of phosphorus grew in TG 1, while in TG 2 the phosphorus concentration did not change during the stages of observation. Sodium and chlorine in the observation groups were normal in 100% of the subjects.

In the treatment groups and the control group, an increase in the average volume of the erythrocytes was identified. However, in the treatment groups the increase stood at +0.9% and +0.8% after 15 and 45 days of observation (TG 1) and +0.7% and +1.17% (TG 2). The average hemoglobin content in the erythrocytes in TG 1 did not change, whereas in TG 2 it fell considerably by the end of the final stage of the study. The decrease in the mean concentration of hemoglobin in TG 1 was 1.2% and 1.0% at different stages of observation, and in TG 2 it stood at 1.0% and 2.0%, respectively. In TG 1 subjects there was a 2.6% increase in the production of erythrocytes by the end of the FP therapy and a 2.9% increase 30 days after it. In TG 2 subjects, there was also a 2.3% increase in erythrocyte production, but after one month of observation, there were no significant changes compared to the baseline value. This indicated more pronounced compensatory reactions to stress in TG 1 students compared to TG 2.

Immunoglobulins play an important role in the body's protective reaction (γ-globulins) and participate in the formation of humoral and cellular immunity (β-globulins). In TG 1, the fraction of α1-globulins was initially higher than the norm. After the FP intake, it significantly decreased to the normal values; a decrease was also noted after 45 days

of observation. There were no such changes in TG 2. The concentration of β-globulins increased in TG 1 by the end of the FP course, while that of TG 2 did not change during the observation period. An increase in the level of γ-globulins was registered in TG 1 by the end of FP intake; it remained unchanged for 45 days of observation. A decrease in γ-globulin values was observed in TG 2, but only after 45 days of observation. These data indicated less pronounced inflammatory reactions and early production of immunoglobulins in TG 1.

Natural immunoglobulin M (IgM) antibodies are pentameric or hexameric macro-immunoglobulins, which were highly conservative during evolution. IgM is initially expressed during B-cell ontogenesis and is the first antibody secreted after contact with foreign antigens [77]. IgM was within the normal range in TG 1 and significantly increased only after 45 days of observation; in TG 2 it increased after 15 days of observation. Immunoglobulin G is the dominant immunoglobulin that can initiate various effector functions, such as complement activation [78]. In TG 1, IgG increased within the reference limits by the end of the observation period. In TG 2, an increase in IgG was noted by the 15th day of observation, and the growth continued even further. This evidenced a higher level of humoral protection in the students of TG 1.

TG 1 showed an increase in the number of lymphocytes by the end of the FP intake, and this effect continued to the end of the observation period. In TG 2, an increase in lymphocytes was noted only by the end of observation. The relative content of lymphocytes (fraction of the total number of leukocytes) in TG 1 was more significant at the second and third stages of observation than at the start. In TG 2, significant differences were identified by the end of the follow-up. The relative number of neutrophils in TG 1 decreased substantially during observation, and in TG 2 only at the end of the observation period. The obtained data proved an earlier reaction of TG 1 subjects (stimulation of both humoral and cellular immunity, reduction of inflammation signs) in response to increased physical activity or the introduction of a pathogen.

The obtained results correlate well with the results of other studies [79,80]. It has been demonstrated that introducing vegetable and protein–vegetable functional products into the diet of new university students is necessary in order for them to fully psychologically and physically adapt. A complete adaptation includes three stages: familiarization with a new place of study, understanding of a student's rights and obligations, and the start of learning; in-depth understanding of the rules and norms of the educational institution and quality education; and conscious and innovative learning improvement. According to a study [80], students frequently experience the problem of increased fatigue during adaptation to the start of training. The so-called "squirrel-in-a-cage" syndrome can be used to describe the lifestyle of first-year students [80]. This syndrome develops with a change in lifestyle due to sleep–wake routine failure, putting the body under a lot of stress. The consumption of vegetable and protein–vegetable functional foods that are balanced in terms of nutritional, energy, and biological values and are enriched with vitamins and micronutrients is one aspect that can mitigate this situation [79–81].

Thus, the comparative analysis of the efficiency of the two FPs produced from plant–protein or plant raw materials showed a considerable advantage of the former. This may have been due to the fact that it had a higher content of proteins. In addition, they differed in the content of vitamins and minerals.

Young people face adaptation challenges dictated by the need for greater maturity. If a person does not have time to adapt to new situations, there is a risk of maladaptation. Adolescence is characterized by the deepening and differentiation of cognitive processes, the development of cognitive independence, the need for peer communication, the emergence of a hierarchically structured system of value orientations, professional self-determination, the emergence of life plans, and the formation of the ability to self-actualize. During these adaptation processes, nutrition is restructured, and it is necessary to form proper food habits for the consumption of functional food products [82].

Limitations on nutritional accommodations arise for students with disabilities. In order to ensure academic program mastery and compensation or restoration of the physi-

ological costs of the body for the adaptation process, people with disabilities require an individualized program of nutrition and adaptation. Universal design for people with disabilities and special needs refers to the creation of objects, furnishings, and special food programs that are as usable for as many people as possible without the need for adaptation or special design [83].

5. Conclusions

The evidence from the study suggests that there are negative changes in the health parameters of students during their adaptation to a new physical and social environment. Higher physical activity and the harsh Baltic Sea climate contribute to the negative cumulative effect. This research showed that higher stress levels resulted in changes in the metabolism of proteins, vitamins and minerals, which led to water–electrolyte imbalance. During this period, the humoral immunity decreased, and the introduction of any pathogen triggered inflammatory reactions.

Another major finding was that a rational diet enriched with functional food products is increasingly beneficial during periods of increased mental and physical activity. To replenish energy, the diet should have a balanced composition of macro- and micronutrients obtained from protein and plant raw materials to prevent changes in the metabolism of proteins, vitamins, and minerals, increase water–electrolyte balance and humoral immunity, and reduce the development of inflammatory reactions to pathogens. The structure of the diet should be appropriate to the age and needs of students.

Finally, the analysis of protein metabolism, vitamin and mineral balance, reaction of red and white blood cells, serum immunoglobulin, and reactions to inflammation showed the advantage of plant-and-protein products over functional foods produced from plant raw materials only. This is explained by the higher content of proteins of animal origin in their composition and a higher concentration of minerals and vitamins. The results of the study contribute to a better understanding of the mechanisms of action of functional foods in the prevention and treatment of various diseases.

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