The result of the research is the development of a technology for the production of germinated flaxseed using plasma-chemically activated aqueous solutions. The object of research was flaxseed. An urgent technological problem is the intensification of the bioactivation process of flaxseed and its effective disinfection. The expediency of using plasma-chemically activated aqueous solutions as an intensifier of the process of flaxseed germination and an effective disinfectant of food raw materials was experimentally proven. It is shown that the use of plasma-chemical activation of process solutions not only accelerates flaxseed germination, but also contributes to a more active accumulation of biologically valuable components in flax raw materials. The composition of flaxseed as a raw material derivative was analyzed. Germinated flaxseed, which is considered a high-value component of food products, was studied separately. An increase in the moisture content of flaxseed during the soaking process by 0.7–1.7 % was recorded. Seedling development increases by 2–9 mm. The germination energy and capacity increase by 5–12 %. The biomass of germinated seeds increases by 39–56 %. In the process of germination, the content of proteins in flaxseed increases from 21.88 to 23.71 %, reducing sugars from 2.37 to 4.02 %. The total content of amino acids increases from 3.64 to 10.38 % compared to the control, and 10 times compared to the raw material. A significant accumulation of vitamins was noted: B_1, B_2, B_3, B_5, B_6, B_7, B_9, C, E. In addition, plasma-chemically activated solutions effectively disinfect germinated flaxseed.

The technology can be applied in the production of enrichment components of food products. The developed technology will receive special attention in the production of functional food products

Keywords: flaxseed, plasma-chemical activation, germination, germinated seeds, biologically active substances

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DEVELOPMENT OF A TECHNOLOGY FOR THE PRODUCTION OF GERMINATED FLAXSEED USING PLASMA-CHEMICALLY ACTIVATED AQUEOUS SOLUTIONS

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1. Introduction

High-quality, nutritious and chemically safe food is one of the main factors that ensure human health, quality and duration of active life. To preserve and strengthen public health, as well as to prevent diseases caused by unbalanced and poor nutrition, an active search for ingredients that can improve the quality of everyday food is underway.

The use of germinated (bioactivated) plant raw materials to create healthy foods in recent years is one of the trends in modern food science. Food products began to be considered a highly effective means of maintaining physical and mental health and preventing many diseases. The selection of raw materials has a significant impact on the expansion of the range of healthy food products. To optimally realize the potential of various plant raw materials, various methods of process activation and regulation of biochemical transformations are widely used.

Flaxseed is a source of the main functional food ingredients and biologically active substances that can have a positive effect on the human body. Flaxseed is rich in essential polyunsaturated fatty acids, dietary fiber, complete proteins and other highly valuable components. Therefore, it is widely used as a functional ingredient to adjust the chemical composition and nutritional value of products.

A promising direction of flaxseed processing is its germination for further use in food production and directly in nutrition. In order to maximize the biochemical potential of flaxseed and increase its nutritional value, its germination or bioactivation is promising. This process allows the maximum accumulation of biologically active components in raw materials. Bioactivation is currently widely used in the pretreatment of plant raw materials, especially various seeds and grains. Technologies of bioactivation for each type of raw material differ in their peculiarities, process modes in which the accumulation of useful substances in plant material (grains or seeds) is intensified. Therefore, the study of the flaxseed bioactivation process and influencing factors correcting the process is a promising scientific direction.

Intensive germination is an innovative technology of preparing flaxseed for consumption and production of healthy foods. In the modern food industry, the priority is to increase the biological and nutritional value of raw materials by obtaining bioactivated food products and components: from germinated seeds of various plants, even exotic ones, to microgreens.

The relevance of research lies in the development of technology for obtaining germinated (bioactivated) flaxseed in order to provide modern food production with a functional ingredient. The scientific value of the technology lies in the creation of innovative food products based on germinated flaxseed, which can become a valuable component of healthy food. This will make it possible to expand raw material sources for creating functional food products, improve the nutritional properties of products and, accordingly, the digestibility of target products.

2. Literature review and problem statement

Flaxseed and products of its processing have a number of useful properties, which are confirmed by scientific research [1–3]. The choice of grain raw materials is of great importance for expanding the range of healthy food products. As natural ingredients, oil crops and products of their processing have become popular. These include flaxseed, which in its biochemical composition meets the requirements for functional products. In the world, there is an increase in the demand for flaxseed and an increase in the volume of its processing. The seeds of this crop belong to the category of natural functional food products [1].

Flax (Linum usitatissimum) has been used as a source of oil for centuries. Flax raw material attracts considerable interest of the general public of scientists. The studies explain the potential benefits of using flax and its processing products to improve public health, including the prevention of chronic non-infectious diseases [2]. Due to the high content of protein, α-linolenic oil, lignans and fiber, the demand for flaxseed and flaxseed oil as a source of food raw materials is increasing [3]. Flaxseed is one of the richest sources of bioactive compounds [4].

Flax continues to occupy a leading position among healthy food components. Flaxseed is used in the prevention of cardiovascular diseases. This is due to the fact that the seeds are a complex source of alpha-linolenic acid, phytoestrogen, lignans, soluble dietary fiber, etc. [5]. Lignans have a diverse spectrum of biological activity. Regular consumption of flaxseed can affect the concentration of total cholesterol. It was also proven that flaxseed has pronounced immunomodulatory, antioxidant and anti-inflammatory effects [5]. The protein complex of flaxseed has a balanced amino acid composition [6, 7]. The shell of flax seeds contains a large number of polysaccharides, which are soluble dietary fibers. Omega-3 and lignan phytoestrogens of flax seeds have a positive effect on the health of a wide range of consumers [8]. Flaxseed is being studied as an alternative or supplement to medications for reducing risk factors associated with the progression of cardiovascular disease. Thus, alpha-linolenic acid (ALA) serves as an antihypertensive agent. Derivatives of enterolignans obtained from secoisolariciresinol (SDH) serve as antioxidants. Accordingly, dietary fibers effectively reduce cholesterol levels [9]. And the list of potential benefits of flaxseed consumption is expanding every year. An unsolved problem is to provide the food industry with high-value flax raw materials and products of its processing.

The main consumer product of oil flax, which has a constant demand on the world market, is its seed [6]. Flaxseed and flax flour are actively used in food industries, such as bakery, meat, and dairy industries [6]. Flaxseed is currently an independent functional food product. The bioactive properties of defatted protein hydrolyzates of flaxseed make them suitable for use as preservatives in natural food products [10]. Nut and flaxseed spreads have recently been used as a substitute for butter [10]. As a functional food ingredient, flax or flaxseed oil is added to baked goods, juices, milk and dairy products, cakes, dry pasta, macaroni and meat products [11]. However, the issue of introducing flaxseed into a wider range of food products is not resolved.

In addition, the priority scientific direction is to provide modern food production with germinated (bioactivated) flaxseed, which can be widely used to enrich food products with useful and highly nutritious substances.

Analysis of scientific and technical sources [9, 10] proves that in modern conditions, the production of fortified food products is one of the priority areas of the food processing industry. Making products with improved consumer properties attracts the attention of both producers and consumers [12]. An unsolved issue is the introduction of functional ingredients of plant origin into the industrial circulation, which will provide the body with nutrients and expand the range of healthy food products [12].

The task is to maximize the biochemical potential of flaxseed and increase its nutritional value. Thus, seed germi-

nation or bioactivation is promising. The intensive method of seed germination is widely used in the production of bioactivated food products and components. In fact, germination is an enzymatic biomodification of seeds by their own proteinases, which increases the availability and digestibility of nutrients due to their breakdown into lower molecular forms [6]. Despite the research and their significant results in the field of food biochemistry of seed germination, scientific and practical interest in the course of these processes remains high. Thus, from a practical point of view, it is important to determine the duration of the process in which a specific raw material acquires increased biological activity due to the accumulation of products of hydrolytic cleavage of biopolymers [13]. However, an unresolved issue is the forced adjustment of the process of transformation of substances in seeds during bioactivation.

During germination, the content of total protein in flaxseed increases. Germinated flaxseed has a significantly higher ratio of unsaturated fatty acids compared to saturated fatty acids and higher calculated values of oxidizability, in addition, an increased content of vitamin C is noted [14]. The high antioxidant capacity of germinated flaxseed is observed [15, 16]. Also, during the bioactivation of flaxseed, an increase in the content of amino acids is noted, so germination can be used as an effective method of increasing the nutritional value of flaxseed [17]. Germinated flaxseed is rich in nutrients, has a wide range of functional properties, is promising in processing, and has scientifically proven health benefits due to the presence of bioactive molecules, namely essential fatty acids, lignans and dietary fiber [18–20]. However, an important factor in bioactivation processes is the selection of optimal germination modes [21], which will ensure a directed flow of biochemical transformations. An unresolved problem is the selection of substances that can intensify the germination process.

There is also the problem of providing industrial production with such important components as germinated flaxseed. All this allows us to state that it is appropriate to study the development of a new intensive technology for obtaining germinated flaxseed, which could ensure the increased demand of food industry enterprises for biologically active components of natural plant origin [11].

From a scientific point of view, the production of germinated flaxseed using an intensive and environmentally friendly technology is interesting. Scientific and technical sources [23–27] give various methods of intensifying seed germination. The main methods used in the intensification of germination are as follows: biotechnological, chemical, physical, physicochemical, complex and others [23, 25–28]. Thus, the most common methods to intensify germination include stratification of flaxseed with an H_2SO_4 solution. Solutions of metal-containing nanoparticles (calcined and uncalcined zinc oxide, zinc, magnesium oxide, silver, copper and iron) are used. The technology using metal salts (zinc acetate, magnesium sulfate, silver nitrate, copper sulfate, and iron (III) chloride) [22], gibberellic acid activation, and ultrasound treatment of soaked seeds are widely used. However, the issue of reducing the use of chemical compounds in the production of germinated seeds, which will allow the production of environmentally safe food products, remains unresolved. The reason for this may be factors related to the high price of effective and non-toxic intensifiers. In addition, an urgent issue is the disinfection of germinated flaxseed. For this purpose, aqueous solutions treated with carbon dioxide are used in classical technologies [28, 29], but an active search for environmentally safe disinfectants continues.

An option to overcome the relevant difficulties can be to use plasma-chemical technologies in the food industry. This approach is used in the work [30], it is stated that plasma-chemical technologies have recently been widely used in the food industry [30–32]. Special attention of scientists is drawn to correcting the course of processes in plant raw materials. However, studies of their influence on the process of flaxseed germination have not been carried out before.

Plasma-chemical activation of water and aqueous solutions allows using the properties of water without its chemicalization by foreign chemicals [26]. During water activation, processes are implemented that contribute to changing the reactogenic properties of aqueous solutions. Thus, the properties of activated water, which arise after plasma-chemical treatment, can be rationally used in various directions of innovative technologies. Water activated by non-equilibrium contact plasma has antiseptic and antibacterial properties [27]. Plasma-chemically activated water is a cluster structure and can exhibit new properties that were little studied before, but are of considerable interest from a practical point of view [26]. Activated water has a specific composition, hydrogen peroxides and superoxide compounds, excited particles and radicals are the most easily detectable. These components play an important role in oxidation-reduction processes, as well as accelerate the transport of moisture into grain material, and correct biochemical transformations in malt grain [28]. Plasma-chemical activation changes the structure of water, namely, water clusters are actively crushed under the action of contact plasma. This leads to the effect of more active penetration of such crushed particles through the membranes and shells of seeds during their soaking and subsequent germination [26]. Clusters contain microparticles of hydrogen peroxide, which upon contact with raw materials are able to form active oxygen and water, which positively affects biochemical transformations and activates the germination process [27].

If we compare the technology of plasma-chemical activation of water with other technologies of seed bioactivation, we can note its chemical safety. This is because its components are decomposed during seed processing and the finished product has no foreign chemical impurities [26]. The shortcomings of the technology include the lack of large-scale industrial use of plasma-chemically activated aqueous solutions.

All this allows us to state that it is appropriate to conduct a study on the selection of a safe intensifier and, accordingly, modes of flaxseed germination. This will make it possible to improve existing technologies and obtain high-quality and chemically pure highly nutritious plant raw materials.

3. The aim and objectives of the study

The aim of the study is to develop a technology for the production of germinated flaxseed using plasma-chemically activated aqueous solutions. This will make it possible to obtain a biologically active component of food products, namely germinated (bioactivated) flaxseed.

To achieve the aim, the following objectives were accomplished:

– to examine changes in the moisture content of the material during moisture absorption by seed (swelling);

– to investigate visual changes in flaxseed during germination, germination energy and capacity of flaxseed;

– to study changes in the biomass of germinated flaxseed;

– to examine the composition of flaxseed of different degrees of germination;

– to investigate the microbiological state of germinated flaxseed;

– to develop a flowchart for the production of germinated flaxseed using plasma-chemically activated aqueous solutions.

4. Research materials and methods

4. 1. Research object and hypothesis

The object of research is flaxseed.

Plasma-chemically activated aqueous solutions were chosen for the research. These solutions are already used in the technology of malting [27], but the scope of this technology for processing solutions needs to be expanded.

The main research hypothesis is obtaining a concentrate of biologically active substances of flaxseed by germination in plasma-chemically activated aqueous solutions. As an intensifier of the flaxseed germination process, plasma-chemically activated aqueous solutions were used.

Plasma-chemically activated aqueous solutions were obtained in the Specialized Laboratory of Plasma Processing of Process Solutions of Food Industries and KNP-TECHNOLOGY Scientific and Production Enterprise LLC (Dnipro, Ukraine). The research was carried out on the basis of the research and production laboratory for determining the quality of grain and grain products, Department of Food Technologies, Dnipro State Agrarian and Economic University (Ukraine).

4. 2. Research materials and equipment used in the experiment

4. 2. 1. Preparation of plasma-chemically activated aqueous solutions

Activation of water for flaxseed moistening was carried out using the technology of processing solutions with cold non-equilibrium plasma [12, 33]. For this purpose, a laboratory setup for plasma-chemical activation of aqueous solutions was used [34]. Thus, the laboratory three-arc plasma-chemical setup consists of a reactor, anodes, cathode, reflux condenser, power source, and vacuum pump. For flaxseed soaking, plasma-chemically activated aqueous solutions were obtained, the characteristics of which are shown in Fig. 1. The concentration of hydrogen peroxide in activated water was determined by iodometry.

Thus, during the activation process from 10 to 60 minutes, it was possible to obtain a concentration of the active substance (hydrogen peroxide) in process water from 300 to 700 mg/l. According to these peroxide concentrations, the following experimental groups were formed: 1 – control (ordinary tap water was used to process the samples); 2 – plasma-chemically activated water (H_2O_2 concentration of 300 mg/l); 3 – plasma-chemically activated water (H_2O_2 400 mg/l); 4 – plasma-chemically activated water (H_2O_2 600 mg/l); 5 – plasma-chemically activated water (H_2O_2 650 mg/l); 6 – plasma-chemically activated water (H_2O_2 700 mg/l).

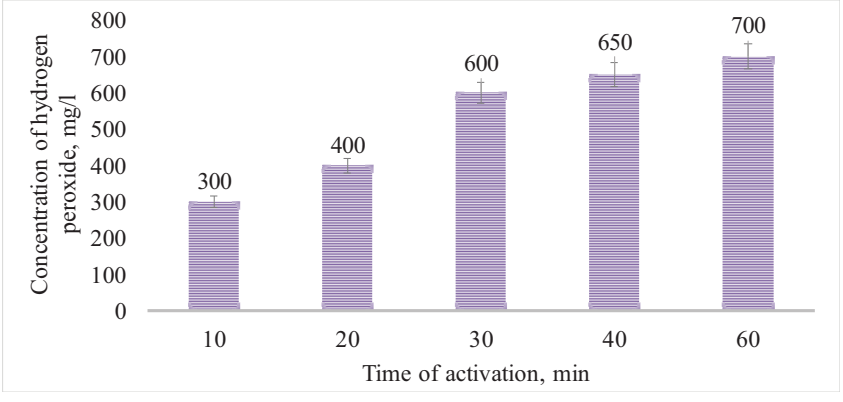

Fig. 1. Characteristics of plasma-chemically activated aqueous solutions for soaking and germination of flaxseed

4. 2. 2. Selection of raw materials for germination and features of obtaining germinated flaxseed

Flaxseed (Linum usitatissimum L.) was chosen as a raw material for germination. The studied flaxseed had the following characteristics, %: proteins – 21.88±1.09; fats – 42.89±2.14; moisture – 3.71±0.18; acid value 3.15±0.15. The seeds are flat, glossy, brown in color.

Germination of test flaxseed samples was carried out in laboratory malthouses [12]. The test samples were treated with activated aqueous solutions according to the standard scheme [12, 34, 35]. The duration of the soaking process was 48 hours, the water duty – 2:1 (aqueous solution: seed), during this time, the flaxseed was completely saturated with the activated process water solution.

The germination process lasts 2–7 days at a temperature of 17–21 °C, with periodically adding an aqueous solution and mixing the raw materials. Further processing of germinated flaxseed depends on process needs (cooling, grinding, drying).

After germination, the moisture content of the material is 46–58 %. In order to preserve the biologically active substances that arose during the germination of raw materials, it is recommended to gradually increase the temperature regime of the drying stage. Seed drying was carried out in the following modes:

– up to 30 % moisture content at a temperature of up to 40 °C;

– up to 10 % – at temperatures from 40 to 70 °C;

– up to 6 % – at a temperature of 70–85 °C [12].

The total drying time of the material was 30–60 hours. Grinding of germinated seeds was carried out on a Miller-350 laboratory mill [36].

4. 3. Methods of determining the parameters of sample properties

4. 3. 1. Method of studying changes in the moisture content of the material during moisture absorption by flaxseed (swelling)

In the course of research, the moisture content was recorded starting from the 1st day of the germination process. The determination was carried out by drying the seeds by a standard method, namely, using a drying cabi-

net. Control was carried out using a SuperPro automatic moisture meter (Manufacturer: SUPERTECH AGRO-LINE AGROELECTRONICS, Denmark).

For the mathematical processing of the experimental materials and justification of the conclusions, analysis of variance without repetitions was used in the study, which determines the influence of two factors on the resulting characteristic [37, 38]. It was assumed that the considered factors have, respectively, M and N gradations, that is, sample observations are denoted as X_{mn}, m=1,...,M, n=1,...,N. Let X denote the total mean over the entire sample, i. e.:

$$X=\Sigma_m \Sigma_n X_{mn}/MN.$$

X_m^{rows} denote the mean for the gradation m of the factor corresponding to the data rows, i. e.:

$$X_m^{rows}=\Sigma_n X_{mn}/N. \quad (1)$$

$X_n^{columns}$ – the mean for the gradation n of the factor corresponding to the data columns, i. e.:

$$X_n^{columns}=\Sigma_m X_{mn}/M. \quad (2)$$

The variance of the resulting characteristic by the factor corresponding to the rows of the data table is calculated as:

$$MS_{rows}=N\cdot\Sigma_m(X_m^{rows}-X)^2/(M-1),$$

the variance of the resulting characteristic by the factor compared to the columns of the data table is found by the formula:

$$MS_{columns}=M\cdot\Sigma_n(X_n^{columns}-X)^2/(N-1),$$

the variance of errors not explained by the considered factors is equal to:

$$MS_{error}=$$
$$=\Sigma_m\Sigma_n\left(X_{mn}-{}^{rows}_{m}-{}^{columns}_{n}X+X\right)^2/\left((M-1)(N-1)\right).$$

The analysis of variance consists in comparing the calculated Fisher criterion:

$$F_{rows}=MS_{rows}/MS_{error} \text{ or } F_{columns}=MS_{columns}/MS_{error},$$

with a critical value F_{crit} with a significance level α and degrees of freedom:

$$df_0=(M-1)(N-1),\ df_1=M-1,\ df_2=N-1.$$

If:

$$F_{rows}>F_{crit}(\alpha;\ df_1;\ df_0) \text{ or } F_{columns}>F_{crit}(\alpha;\ df_2;\ df_0), \quad (3)$$

then the resulting characteristic significantly depends on the dynamics of the corresponding factor. If the inequality holds:

$$F_{rows}\le F_{crit}(\alpha;\ df_1;\ df_0) \text{ or } F_{columns}\le F_{crit}(\alpha;\ df_2;\ df_0),$$

then the corresponding factor does not have a significant effect on the resulting characteristic.

4. 3. 2. Method of studying visual changes in flaxseed during germination

In the course of the research, the day of seedling appearance and changes in the flax seedling length were recorded. The parameters were recorded in a sample of 50 seeds.

4. 3. 3. Methods of determining the germination energy and capacity of flaxseed

For research, a standard method of malt production was used, 6 analytical groups of 500 pieces were selected from flaxseed.

To forecast the quality of flaxseed germination, the germination capacity and energy were determined [12]. Control – flax seeds that were not subjected to any physical or chemical treatment. All experiments were repeated five times.

4. 3. 4. Methods of determining the biomass of germinated flaxseed

Biomass was examined by weighing the respective samples. The goal was to record the intensity of seedling development and the increase in the mass of food raw materials during the germination process.

In the course of the study, hypotheses were put forward about the similarity of the relative response of flaxseed to the use of plasma-chemically activated aqueous solutions in a pair of seedling length and biomass indicators and in a pair of germination energy and capacity indicators. Testing of these hypotheses was carried out by the T-test for the sample mean with unknown variance [37, 38]. The sample elements were defined as differences in the values of the analyzed indicators $\Delta_k=Y1_k-Y2_k$, k=1,..,K. The test statistic with the assumption of a zero sample mean value was calculated by the formula:

$$T=\Sigma_k \Delta_k/(K\cdot\Sigma_k(\Delta_k-\Delta)^2/(K-1))^{0.5}. \quad (4)$$

The hypothesis of a non-zero sample mean was rejected on the basis of inequality:

$$|T|\le T(\alpha;\ df), \quad (5)$$

where $T(\alpha;\ df)$ is the critical value of two-tailed Student's T-distribution with the level of significance α and degrees of freedom $df=K-1$.

4. 3. 5. Methods of determining the chemical composition, content of amino acids and vitamins in germinated flaxseed

The protein content in the seeds was determined by the Kjeldahl method, the fat content by the Rushkovsky method, the carbohydrate content by the liquid chromatography method, the fiber amount by the standard method, and reducing sugars by the ferricyanide method [12].

The analysis of the amino acid content in flaxseed was carried out by ion-exchange liquid column chromatography on a T339 automatic amino acid analyzer, manufactured in the Czech Republic, Prague. The vitamin composition of germinated flaxseed was also determined using ion-exchange liquid column chromatography and other standard methods [12].

To test the hypothesis about the preservation of relative proportions of the biochemical and vitamin composition of

germinated flaxseed when using plasma-chemically activated aqueous solutions, the T-test for sample means with different variances was applied [37, 38]. Test statistics were calculated by the formula:

$$T=(\Delta 1-\Delta 2)/(S1/N1+S2/N2)^{0.5},$$

where $N1$ and $N2$ are the size of the first and second samples, consisting, respectively, of the elements $\Delta 1_i$, $i=1,..,N1$, $\Delta 2_j$, $j=1, .., N2$, $\Delta 1$ and $\Delta 2$ denote the sample means, namely:

$$\Delta 1=\Sigma_i \Delta 1_i/N1,\ \Delta 2=\Sigma_j \Delta 2_j/N2.$$

$S1$ and $S2$ denote the sample unbiased (corrected) variances, namely:

$$S1=\Sigma_i (\Delta 1_i-\Delta 1)^2/(N1-1),\ S2=\Sigma_j (\Delta 2_j-\Delta 2)^2/(N2-1).$$

The hypothesis about the difference in sample means was rejected on the basis of inequality:

$$|T|\le T(\alpha;\ df), \quad (6)$$

where $T(\alpha;\ df)$ is the critical value of two-tailed Student's T-distribution with the level of significance α and degrees of freedom df found by the formula:

$$df=\left[\left(\begin{matrix}S1/N1+\\+S2/N2\end{matrix}\right)^2/\left(\begin{matrix}(S1/N1)^2/(N2-1)+\\+(S2/N2)^2/(N1-1)\end{matrix}\right)\right].$$

To test the hypothesis about a statistically significant increase in the amino acid content in dynamics during the germination of bioactivated flaxseed, the T-test for the sample mean with unknown variance was applied [37, 38]. In this case, the test statistic (4) was compared with the critical value $T(\alpha;\ df)$ of the right-tailed Student's T-distribution with the level of significance α and degrees of freedom $df=K-1$. If:

$$T>T(\alpha;\ df), \quad (7)$$

then the sample mean is significantly different from 0. If the inequality holds:

$$T\le T(\alpha;\ df), \quad (8)$$

then the alternative hypothesis of a statistically significant non-zero sample mean is rejected.

4. 3. 4. Methods of studying the microbiological state of germinated flaxseed

The total microbial count (QMAFAnM) was determined by the classical method of inoculation on agar nutrient media followed by incubation of the cultures and counting of cultivated colonies of microorganisms [39].

5. Results of studies of process indicators of obtaining bioactivated flaxseed

5. 1. Study of changes in the moisture content of flaxseed during moistening and germination

To determine the optimal concentration of hydrogen peroxides in plasma-chemically activated aqueous solutions, monitoring of changes in the moisture content of flaxseed during its moistening was carried out (Table 1).

Table 1

Changes in the moisture content of flaxseed during moistening and germination, %

Experiment	Hydrogen peroxide concentration, mg/l	Day					
		0	1	2	3	4	5
1	0	3.7	49.8	52.7	54.5	57.5	57.0
2	300	3.7	50.1	52.9	54.8	57.6	57.2
3	400	3.7	50.8	53.1	55.0	57.9	57.5
4	**600**	**3.7**	**51.5**	**53.7**	**55.9**	**58.2**	**57.8**
5	650	3.7	51.2	53.4	55.5	58.0	57.6
6	700	3.7	50.9	53.1	54.6	57.8	57.1

To substantiate changes in the moisture content of flaxseed during moistening and germination, a two-factor analysis of variance without repetitions performed using spreadsheet tools was applied to Table 1. The initial data are selected from $M=6$ rows about the experiments performed and $N=5$ columns, corresponding to 1–5 days of moistening and germination. Based on ratios (3):

$$19.25=F_{rows}>F_{crit}(0.05;\ 5;\ 20)=2.71,$$

$$1213.48=F_{columns}>F_{crit}(0.05;\ 4;\ 20)=2.87,$$

it was found that the hydrogen peroxide concentration (from 0 to 700 mg/l) and soaking duration (from 1 to 5 days) cause the appearance of significantly different moisture indicators of flaxseed at a standard significance level of $\alpha=0.05$. According to formulas (1), (2), it was found that the maximum average indicators $X_4^{rows}=55.42\ \%$ and $X_4^{columns}=57.83\ \%$ correspond to experiment 4 and the fourth day of seed treatment, and these values had the smallest variations of 7.99 and 0.07, which confirms their greatest stability.

5. 2. Study of visual changes in flaxseed during germination, germination energy and capacity of flaxseed

The swelling rate of flaxseed can be monitored by visual observation and recording changes in the geometric parameters of the seeds. The results of the studies are given in Table 2.

Important indicators characterizing the quality of germinated flaxseed and the level of bioactivation of raw materials are germination energy and capacity (Table 3). Evaluating these indicators, it can be argued that the samples with their maximum values will have the highest content of biologically active food components.

The hypothesis about the similarity of the relative response of the germination energy and capacity indicators of bioactivated flaxseed was tested according to Fig. 2, illustrating changes in these indicators compared to the control experiment (Table 4).

Inequality (5), calculated by spreadsheet tools at $K=5$ and with a typical significance level of 0.05, $2.14=T\le T(0.05;\ 4)=2.78$, shows a similar reaction of flaxseed in terms of germination energy and capacity when using plasma-chemically activated aqueous solutions, which is consistent with a positive close to 1 Pearson correlation coefficient of 0.98.

Table 2

Visual changes in flaxseed during germination

Experiment	Hydrogen peroxide concentration, mg/l	Germination day, seedling length in mm				
		1	2	3	4	5
1	0	mucilagination and swelling of the polysaccharide shell, increase in seed size	seed swelling	a sprout appeared	3	8
2	300		seed swelling	a sprout appeared	5	10
3	400		a sprout appeared	5	10	12
4	**600**		a sprout appeared	**6**	**12**	**14**
5	650		a sprout appeared	5	11	13
6	700		a sprout appeared	4	9	11

Table 3

Germination energy and capacity of flaxseed when using plasma-chemically activated aqueous solutions

Experiment	Water	Hydrogen peroxide concentration, mg/l	Germination rates, %	
			energy	capacity
1	tap	–	82±1.7	88±1.3
2	activated	300	87±1.2	93±1.1
3	activated	400	92±1.9	97±1.5
4	**activated**	**600**	**96±1.4**	**100±1.2**
5	activated	650	93±1.1	99±1.4
6	activated	700	91±1.5	96±1.2

Table 4

Changes in the germination energy and capacity of flaxseed when using plasma-chemically activated aqueous solutions, r.p.

Experiment	Germination energy, $Y1_k$	Germination capacity, $Y2_k$
2	5	5
3	10	9
4	14	12
5	11	11
6	9	8

5. 3. Study of changes in the biomass of germinated flaxseed

In parallel, the weight of flaxseed biomass at the end of the germination process was studied, the relevant data are shown in Table 5. This indicator allows you to predict the yield of the finished food component (germinated flaxseed).

Table 5

Biomass of germinated flaxseed

Experiment	Water	Hydrogen peroxide concentration, mg/l	Biomass	
			5th day of germination, g	relative change in biomass, %
1 (control)	Tap	–	1.26	100
2	Activated	300	1.76	139
3	Activated	400	1.85	147
4	**Activated**	**600**	**1.97**	**156**
5	Activated	650	1.84	146
6	activated	700	1.79	142

The hypothesis about the similarity of the response of the seedling length (Table 2) and biomass indicators (Table 5) of germinated flaxseed was tested according to the data in Table 6, illustrating the relative changes of these indicators compared to the corresponding control experiment.

Table 6

Changes in the seedling length and biomass of flaxseed on the 5th day when using plasma-chemically activated aqueous solutions

Experiment	Seedling length, $Y1_k$	Biomass, $Y2_k$
2	0.25	0.40
3	0.50	0.47
4	0.75	0.56
5	0.63	0.46
6	0.38	0.42

Inequality (5), calculated by spreadsheet tools at K=5 and with a typical significance level of 0.05:

$$0.61=T\leq T(0.05;\ 4)=2.78,$$

evidences a similar response of flax seeds in terms of seedling length and biomass when using plasma-chemically activated aqueous solutions, which is consistent with a positive close to 1 Pearson correlation coefficient of 0.92.

5. 4. Study of the composition of germinated flaxseed

A significant stage in the research of flaxseed germination is to determine its composition. For a comprehensive analysis of bioactivated seeds, samples treated with plasma-chemically activated solutions with a peroxide concentration of 600 mg/l were selected. The main components included in their composition are listed in Table 7. It is important to compare the biochemical composition of ordinary and germinated flaxseed, which will allow us to more thoroughly characterize the features of the germination process from a chemical point of view.

The hypothesis about the preservation of relative proportions of the biochemical composition of flaxseed when using plasma-chemically activated aqueous solutions was verified. This was done by the T-test for sample means with excellent variances according to the data in Table 8, containing the modules of the difference between the parameters of the control and the initial raw material ($\Delta 1_i$) and the experiment and the initial raw material ($\Delta 2_j$) at $N1=N2=7$.

Table 7

Biochemical composition of flaxseed during germination, %

Indicator	Initial raw materials (non-germinated flaxseed)	Control (flaxseed germinated without adding activator)	Experiment (flaxseed germinated using plasma-chemically activated aqueous solutions)
Proteins	21.88	22.65	23.71
Fats	42.89	40.72	39.91
Fiber	6.78	5.29	5.01
Hemicellulose	12.64	12.04	11.95
Reducing sugars, including	2.37	3.64	4.02
Monosaccharides	1.31	1.62	1.84
Disaccharides	1.06	2.02	2.18

Table 8

Changes in the biochemical composition of flaxseed when using plasma-chemically activated aqueous solutions, r.p.

Indicator	$\Delta 1_i$	$\Delta 2_j$
Proteins	0.77	1.83
Fats	2.17	2.98
Fiber	1.49	1.77
Hemicellulose	0.6	0.69
Reducing sugars	1.27	1.65
Monosaccharides	0.31	0.53
Disaccharides	0.96	1.12

Based on inequality (6), calculated by spreadsheet tools with a typical significance level of 0.05:

$$1.09=T\leq T(0.05;\ 11)=2.20,$$

the relative proportions of the biochemical composition of the control and experimental flaxseed samples were preserved. This testifies to the inviolability of the natural qualities observed in the initial raw materials.

A step-by-step analysis of germinated flaxseed was performed, namely, monitoring changes in the amino acid composition throughout the entire germination process of the studied raw material. The results of the experiments are given in Table 9.

Important for the human body, both essential and non-essential, amino acids were found in the samples. When using plasma-chemically activated aqueous solutions, the content of amino acids tended to increase steadily.

Table 9

Amino acid content in germinated flaxseed, mg/g of protein

Amino acid	Germination day				
	0	2	4	5	6
Control (flaxseed germinated without adding activator)					
Alanine	0.25	2.48	2.50	3.08	4.05
Arginine	1.38	3.77	3.31	3.78	4.42
Asparagine	0.36	0.85	1.18	1.65	2.06
Aspartic acid	0.48	0.84	0.72	0.84	0.98
Cystine	0.05	0.12	0.26	0.28	0.34
Glycine	0.21	0.92	1.48	2.16	2.61
Glutamine	0.07	5.06	8.65	9.31	10.42
Glutamic acid	0.41	3.15	3.58	3.97	4.61
Histidine	0.14	1.22	1.41	1.87	2.11
Lysine	0.18	1.12	1.21	1.46	1.65
Leucine	0.01	1.78	1.72	2.14	2.42
Isoleucine	0.05	1.10	1.12	1.21	1.34
Methionine	0.01	0.54	0.53	0.54	0.57
Phenylalanine	0.14	1.40	1.08	1.42	1.55
Proline	0.29	1.15	2.46	3.82	4.36
Serine	0.07	1.88	1.99	3.19	3.38
Threonine	0.12	0.96	1.02	1.15	1.26
Tryptophan	0.39	0.91	1.29	1.63	1.72
Tyrosine	0.11	1.12	1.18	1.21	1.25
Valine	0.05	1.40	1.54	1.68	1.77
Total content	4.77	31.77	38.13	46.39	52.87
Experiment (seeds germinated using plasma-chemically activated aqueous solutions)					
Alanine	0.25	2.54	2.60	3.35	4.31
Arginine	1.38	4.05	3.42	3.86	4.58
Asparagine	0.36	1.21	1.24	1.81	2.64
Aspartic acid	0.48	1.18	0.77	0.89	1.02
Cystine	0.05	0.20	0.27	0.30	0.41
Glycine	0.21	1.14	1.52	2.24	3.02
Glutamine	0.07	6.01	8.74	9.45	10.89
Glutamic acid	0.41	3.41	3.65	4.07	4.98
Histidine	0.14	1.32	1.58	1.93	2.35
Lysin	0.18	1.16	1.29	1.59	1.77
Leucine	0.01	1.87	1.84	2.21	2.49
Isoleucine	0.05	1.18	1.16	1.29	1.39
Methionine	0.01	0.55	0.53	0.56	0.59
Phenylalanine	0.14	1.41	1.17	1.49	1.56
Proline	0.29	1.26	2.59	3.96	4.42
Serine	0.07	1.94	2.14	3.25	3.85
Threonine	0.12	1.01	1.18	1.19	1.42
Tryptophan	0.39	1.07	1.38	1.68	1.81
Tyrosine	0.11	1.14	1.19	1.23	1.29
Valine	0.05	1.42	1.67	1.74	2.08
Total content	4.77	35.07	39.84	48.08	56.86

The hypothesis about a statistically significant increase in the content of amino acids in flaxseed germinated using plasma-chemically activated aqueous solutions was tested

according to the data in Table 8 about 2–6 days of germination (K=4) in relation to 20 individual amino acids and their total content. The analyzed indicators $Y1_k$ and $Y2_k$, k=1, .., K corresponded to the content of amino acids in the experiment and control. The critical value of the right-tailed Student's T-distribution with a typical level of significance α=0.05 and degrees of freedom df=3 was $T(\alpha; df)$=2.35.

Hence, on the basis of inequalities (7) with the test statistics T of 4.62; 3.19; 3.58; 2.52; 2.56; 2.84; 3.59; 4.50; 7.41; 6.06; 2.61; 6.18; 3.08; 4.26; 3.58 calculated by formula (4), a statistically significant increase in the total content of amino acids was found. In particular, alanine, arginine, asparagine, cystine, glutamic acid, histidine, lysine, leucine, isoleucine, methionine, proline, threonine, tryptophan and tyrosine for the considered germination period of bioactivated flaxseed.

On the other hand, although the flaxseed of the experiment showed increased indicators for aspartic acid, glycine, glutamine, phenylalanine, serine and valine compared to the control, but on the basis of inequalities (8) with the test statistics T of 1.64; 2.24; 2.08; 2.18; 1.90; 2.03 calculated by formula (4), these advantages in dynamics turned out to be statistically insignificant with a significance of α=0.05.

The composition of vitamins of germinated flaxseed is given in Table 10. Flaxseed contains a wide range of B vitamins, especially vitamin B1. It is an important component of carbohydrate metabolism in the human body. Because with a lack of vitamin B_1, the human body is not able to fully absorb sugars.

The hypothesis about the preservation of the relative proportions of the vitamin composition of germinated flaxseed when using plasma-chemically activated aqueous solutions was tested. This was done by the T-test for sample means with excellent variances according to the data in Table 11, containing the modules of the difference between the parameters of the control and raw material ($\Delta 1_i$) and the experiment and raw material ($\Delta 2_j$) at $N1$=$N2$=9.

Inequality (6), calculated by spreadsheet tools with a typical significance level of 0.05:

$$0.99=T\leq T(0.05; 15)=2.13,$$

confirms that the germinated flaxseed retains the vitamin composition in the experimental sample compared to the control.

Table 11

Changes in the vitamin composition of germinated flaxseed, mg r.p.

Vitamin	$\Delta 1_i$	$\Delta 2_j$
Vitamin B_1	0.48	0.6
Vitamin B_2	0.12	0.37
Vitamin B_3	0.57	0.78
Vitamin B_5	0.39	0.57
Vitamin B_6	0.12	0.29
Vitamin B_7	0.003	0.005
Vitamin B_9	0.011	0.022
Vitamin C	0.76	0.97
Vitamin E	0.35	0.44

5. 5. Study of MAFAnM contamination of germinated flaxseed

In order to investigate the contamination with MAFAnM, the germination of flaxseed samples was performed in working process solutions with a hydrogen peroxide concentration of 300–700 mg/l, and the microbiological state of germinated seed QMAFAnM was determined. The results obtained are shown in Table 12.

Table 12

Study of the microbiological state of germinated flaxseed

Experiment	Water	Hydrogen peroxide concentration, mg/l	QMAFAnM, CFU/g
1 (control)	Tap	–	$8.2\times10^6\pm1.5$
2	Activated	300	$5.7\times10^4\pm1.7$
3	Activated	400	$1.9\times10^2\pm1.6$
4	**activated**	**600**	–
5	Activated	650	–
6	Activated	700	–

5. 6. Development of a flowchart for the production of germinated flaxseed using plasma-chemically activated aqueous solutions

For the future industrial implementation of the proposed technology, a flowchart for the production of germinated flaxseed using plasma-chemically activated aqueous solutions was developed (Fig. 2).

Table 10

Vitamin composition of germinated flaxseed

Vitamin	Initial raw materials (non-germinated flaxseed)	Control sample (seeds germinated without adding activator)	Experimental sample (seeds germinated using plasma-chemically activated aqueous solutions)
Vitamin B_1, mg %	0.64	1.12	1.24
Vitamin B_2, mg %	0.24	0.36	0.61
Vitamin B_3, mg %	3.21	3.78	3.99
Vitamin B_5, mg %	0.58	0.97	1.15
Vitamin B_6, mg %	0.68	0.80	0.97
Vitamin B_7, mg %	6	9	11
Vitamin B_9, mg %	112	123	134
Vitamin C, mg %	0.58	1.34	1.55
Vitamin E, mg %	0.54	0.89	0.98

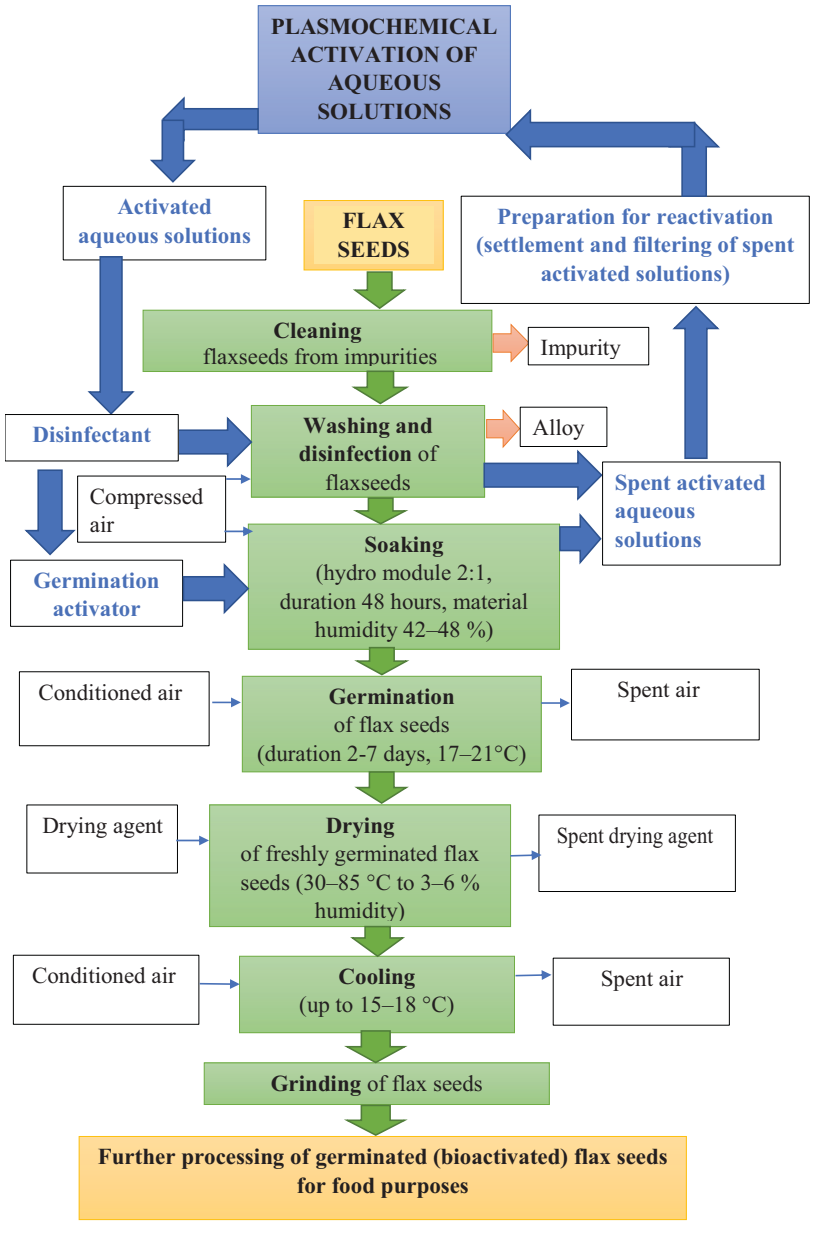

Fig. 2. Flowchart for the production of germinated flaxseed using plasma-chemically activated aqueous solutions

6. Discussion of the results of research on the technology of obtaining a biologically active component of food products (germinated flaxseed)

Analysis of the results of Table 1 allows us to note that at the initial stage of germination, the seeds swell as a result of intensive moisture absorption by flaxseed. On the first day of germination, the moisture content increased from 3.7 to 51.5 %. Over the next four days, the moisture content stabilized and changed little. Moreover, in the samples treated with plasma-chemically activated solutions, the moisture content was higher on average by 0.7–1.7 %. The highest figure was recorded when using activated solutions with a peroxide concentration of 600 mg/l. That is, at the beginning of the process, there was an intensification of moisture transport to the seeds, which is a prerequisite for a more intensive process of flaxseed germination. And also a guarantee of intensive accumulation of biologically active substances in the studied food raw materials.

After examining changes in the moisture content of the flaxseed material, a more intense moisture transport to the seeds can be noted. Thus, the moisture content increases from 0.7 to 1.7 %, depending on the peroxide concentration in the solution. Moreover, the concentration of peroxides in the activated solution within 600 mg/l has an optimal effect on the seeds (Table 1). It is important to accelerate the diffusion of water into flaxseed at the initial stage. The intensity of water absorption in the first hours of soaking affects the entire subsequent process of germination and, accordingly, the course of biochemical reactions in the raw material. The seed shell is partially permeable and therefore allows only water and aqueous solutions to diffuse into the seed. Ions penetrate inside through the micro-cracks in the shell and can affect the embryo. The chaotic movement of

ions in plasma-chemically activated water allows accelerating water diffusion into flaxseed due to a more active influx of charged particles to the surface of the raw material [26]. This aspect confirms that when using activated water as a moistening agent, due to its specific composition, there is a more active transport of active moisture to the seed. That is, plasma-chemically activated water is rapidly diffused into the grain (absorbed). Therefore, the increase in the moisture content of the material (Table 2) indicates the active transport of activated aqueous solutions to flaxseed, which contributes to the bioactivation processes in it [26]. It should be noted that in the developed technology, compared to the known ones [6, 40], a more intense increase in the moisture content of the material is observed, which confirms the active absorption of solutions by flaxseed. All this causes a redistribution between the flaxseed substances, increases the proportion of soluble substances and decreases the proportion of insoluble substances in the seeds.

Analyzing the results of visual observation of flaxseed germination, at the initial stage of germination (swelling), the seeds intensively absorb moisture, as a result, the seeds begin to germinate, as shown in Table 2. Starting from the second day of observation, seed germination is recorded in samples treated with plasma-chemically activated aqueous solutions. If you analyze the intensity of seedling development, it should be noted that when using plasma-chemically activated solutions, their length increased by 2–9 mm on the 4th day compared to the control. Accordingly, on the fifth day, it increased by 2–6 mm. Therefore, the effect of intensive germination is observed in all samples, the maximum result was obtained at a concentration of hydrogen peroxide in the solution of 600 mg/l.

The study of visual changes in flaxseed during germination (Table 2) confirmed the hypothesis of intensive germination. This became especially noticeable when monitoring the length of the flax seedling. Thus, when using plasma-chemically activated aqueous solutions, the seedlings were larger by 2–9 mm. In addition, the biomass of flaxseed increased by 39–56 % during germination. That is, biochemical processes occurred efficiently enough to obtain high-quality food raw materials.

Analyzing the data in Table 3, it should be noted that there is an increased germination activity compared to the control. The process efficiency increases from 5 to 12 %. Thus, the total process duration in industrial conditions will be 3–5 days, as evidenced by 96 % germination on the third day and 100 % germination of seeds on the 5th day of the process. At the same time, in the control experiment (using tap water), the process lasts 10 days. The process of flaxseed germination accelerated by 2 times.

Studies of germination indicators (germination energy and capacity) of flaxseed with the proposed activator showed an increase in the process efficiency, the indicators increased from 5 to 12 %. For 100 % seed germination, the process duration is 5 days, which is half the control process without using an activator (with tap water). Among the concentrations of plasma-chemically activated aqueous solutions studied in the work, it is advisable to use a solution with a concentration of 600 mg/l. It was this concentration that made it possible to achieve 100 % germination capacity of flaxseed. Comparing the action of plasma-chemically activated solutions with the action of other activators [23–27], a significant increase in germination rates should be noted, on average by 2–3 %.

Analyzing the data obtained, it should be noted that the biomass of germinated flaxseed exceeded the control sample by 39–56 %, which is evidence of the intensive course of germination processes in flaxseed. In the samples treated with plasma-chemically activated aqueous solutions, increased growth of biomass was noted. Thus, samples with a concentration of 600 mg/l showed the maximum increase in the indicator, namely, the biomass of germinated flaxseed increased by 56 %. Therefore, as a result of flaxseed germination, 56 % more product will be obtained, which is a positive technological result.

The composition of germinated flaxseed was investigated in the work. It was found that germinated flaxseed obtained using plasma-chemically activated aqueous solutions has all the signs of an effective course of the germination (bioactivation) process. Namely, an increase in protein content from 21.88 % to 23.71 % is recorded, the content of reducing sugars increases from 2.37 % to 4.02 %, the concentration of fiber in flaxseed decreases from 6.78 % to 5.01 %, and hemicellulose – from 12.64 % to 11.95 %. A decrease in the amount of complex carbohydrates with a simultaneous increase in the amount of mono- and disaccharides is observed. Thus, the resulting food product will have a greater amount of biologically active substances in its composition and thereby a high nutritional value. Comparing the obtained results with the results of other researchers [13], a more intense accumulation of proteins should be noted, on average by 2–3 %.

The data in Table 7 demonstrate that germinated flaxseed obtained using plasma-chemically activated aqueous solutions has all the signs of an effective course of the germination (bioactivation) process. Germination is a rather complex process, which changes the entire biochemical complex of food raw materials. At the same time, secondary formation of amino acids occurs, when the proteins contained in the endosperm are hydrolyzed to form free amino acids. In the future, they are used to form the embryo. In the experiments, an increase in protein content is recorded due to the leaching of non-protein substances from the seeds during soaking and synthesis of new proteins. The carbohydrate complex of flaxseed is also prone to changes during germination. Thus, the content of reducing sugars in it increases, and this is because the formation of sugars during flaxseed germination occurs mainly due to the hydrolytic splitting of fats under the action of lipase. Fatty acids and glycerol are formed from them, which later turn into sugars. At the stage of intensive germination, complex biopolymers, such as fiber and hemicellulose, are decomposed under the action of enzymes into simpler ones that can dissolve well. As a result, the fiber content in flaxseed decreases.

In the study of the proposed technology, an increased content of amino acids was found, compared to the control sample by 3.64–10.38 % (Table 9). An increase in the amount of amino acids was observed: alanine, arginine, asparagine, aspartic acid, cystine, glycine, glutamine, glutamic acid, histidine, lysine, leucine, isoleucine, methionine, phenylalanine, proline, serine, threonine, tryptophan, tyrosine, valine. These results prove the veracity of the research hypothesis about the possibility of obtaining a concentrate of biologically active substances. This is mainly due to the use of an activator represented by plasma-chemically activated water (process solution). It contributes to the accumulation and accelerates the action of proteolytic enzymes in raw materials [26].

Analyzing the data in Table 9, it can be stated that as a result of more intense flaxseed germination, amino acids are synthesized very actively. Thus, if we compare the results of the second day of germination, the total content of amino acids in the samples treated with plasma-chemically activated aqueous solutions increased by 10.38 % compared to the control. If we compare the content of amino acids with the original raw material, that is, flaxseed, the total content of amino acids increased 8 times. If we analyze the amount of amino acids at the final stage of germination, then, accordingly, the total amount of amino acids when using the proposed technology increased by 7.54 %, compared to the control, and was 10 times higher than in the original raw material. This confirms the research hypothesis about the possibility of obtaining a food concentrate of biologically active substances in the product of the germination process using an activator.

The composition of vitamins in germinated flaxseed was investigated. An increase in the content of the following vitamins was recorded: B_1, B_2, B_3, B_5, B_6, B_7, B_9, C, E, compared to both the initial raw material (flaxseed) and the control sample (germinated flaxseed without activator). Analyzing the data in Table 10, it was found that the experimental samples have an increased content of vitamins. This also shows the expediency and importance of using germinated flaxseed in food in order to enrich the diet and provide it with a vitamin complex contained in bioactivated flaxseed.

All plant materials contain microorganisms on their surface, in particular, pathogenic ones. Since the process of flaxseed germination involves soaking and moistening seeds, such microflora in favorable conditions (high humidity and heat) is reproduced throughout the entire process. The seed microbiota may have undesirable effects on the finished product if it is not heat-treated. In this regard, it is necessary to determine the ways of aseptic treatment of germinated flaxseed in order to improve the quality. Plasma-chemically activated aqueous solutions are known to have a disinfecting effect [34]. So, it is advisable to use them in the fight against microbiological contamination of food raw materials.

The microbiological state of germinated flaxseed was studied, which confirmed the positive effect of plasma-chemically activated solutions on the microbiological state of food raw materials. It is known that plasma-chemically activated aqueous solutions can inhibit pathogenic microorganisms and disinfect raw materials [34, 36]. It was found that among solutions with different activator concentrations, solutions with a concentration of 600 mg/l have a disinfecting effect on flaxseed. At this concentration, the germinated material contained no microorganisms at all.

The total microbial number for germinated flaxseed in plasma-chemically activated solutions of different concentrations was determined. The quantity of microorganisms was determined by counting colonies found on standard agar media. The standard QMAFAnM indicator for non-traditional raw materials (including flaxseed) is less than $5*10^4$. It was found that the proposed disinfectant is effective in the developed technological process. Solutions with an activator concentration of 600 mg/l were found to have a disinfecting effect on flaxseed. At this concentration, the germinated material contained no microorganisms at all. Due to the specific composition, plasma-chemically activated aqueous solutions have a long-lasting disinfecting effect. Therefore, when the microflora re-enters during the germination process, it dies.

For the industrial implementation of the presented technology, a flowchart for the production of germinated flaxseed using plasma-chemically activated aqueous solutions was developed (Fig. 2). In addition to the classic technological operations, such as seed cleaning, washing and disinfection, soaking, germination, drying, grinding and further processing, it includes a new technological solution, namely, the introduction of plasma-chemical activation of aqueous solutions into the process. Activated solutions are used at the stage of seed washing and disinfection as a disinfecting agent. In addition, it is advisable to use them at the stage of soaking, in order to accelerate the process of seed moistening, with the subsequent intensification of the germination process. Next, the spent activated aqueous solutions are settled, filtered and sent for repeated plasma-chemical activation. Such a closed cycle of water use when applying the technology of plasma-chemical activation of aqueous solutions makes it possible to significantly save water resources [35].

The results of studies of the process parameters and changes in the composition of germinated flaxseed allow creating an effective processing of flaxseed raw materials to obtain a biologically active component of food products of universal purpose. The obtained product can be used without heat treatment, dried or ground into flour, and used as a component of high-tech food products for confectionery, dairy, meat or fish products.

The shortcomings of the studies include the lack of data on changes in the lipid part of flaxseed in the process of germination. These data are planned to be obtained during the continuation of research on the technology of production of germinated flaxseed using plasma-chemically activated aqueous solutions.

The limitations of this study may be related to the provision of sufficient amounts of plasma-chemically activated aqueous solutions to industrial enterprises. However, this issue is currently being resolved, as KNP-TECHNOLOGY Scientific and Production Enterprise LLC is actively working on the project of serial production of plasma-chemical industrial installations. This, in turn, will expand the prospects and opportunities for providing the food processing industry with activated process solutions.

The prospect of the research consists in the development of recipes for functional food products using the obtained biologically active ingredient (germinated flaxseed).

7. Conclusions

1. Changes in the moisture content of flaxseed in the process of moistening with plasma-chemically activated aqueous solutions were investigated. It was found that the moisture content increased by 0.7–1.7 %. The highest figure was recorded when using activated solutions with a peroxide concentration of 600 mg/l.

2. Visual changes in the process of flaxseed germination were studied, so the seedling length was recorded to increase by 2–9 mm. The maximum result was obtained at a concentration of hydrogen peroxide in the solution of 600 mg/l. The indicators of flaxseed germination (germination energy and capacity) were analyzed, they increased by 5–12 %.

3. The study of changes in the biomass of germinated flaxseed with plasma-chemically activated solutions showed an increase by 39–56 %.

4. The study of the chemical composition of germinated flaxseed using plasma-chemically activated aqueous solutions revealed an increase in protein content by 1.83 % and reducing sugars by 1.65 %. The total content of amino acids increased by 3.64–10.38 %, and compared to the original raw material (flaxseed), by 10 times. An increased amount of the following vitamins was found in the germinated seeds: B_1, B_2, B_3, B_5, B_6, B_7, B_9, C, E.

5. The study of the microbiological state of the material confirmed the disinfecting effect of plasma-chemically activated solutions in the treatment of flaxseed with an initial QMAFAnM index of $8.2*10^6$. The use of a hydrogen peroxide concentration of 600 mg/l in the process liquid ensured the absence of microorganisms in the final germinated product.

6. A flowchart for the production of germinated flaxseed was developed, a feature of which is the introduction of plasma-chemical activation of aqueous solutions into the technological process.

Conflict of interest

The authors declare that they have no conflict of interest in relation to this study, whether financial, personal, authorship, or otherwise, that could affect the study and its results presented in this paper.

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Data availability

The manuscript has no associated data.

References

1. Kraevska, S., Yeshchenko, O., Stetsenko, N. (2019). Optimization of the technological process of flax seed germination. Food Science and Technology, 13 (3). doi: https://doi.org/10.15673/fst.v13i3.1453
2. Gutiérrez, C., Rubilar, M., Jara, C., Verdugo, M., Sineiro, J., Shene, C. (2010). Flaxseed and flaxseed cake as a source of compounds for food industry. Journal of Soil Science and Plant Nutrition, 10 (4), 454–463. doi: https://doi.org/10.4067/s0718-95162010000200006
3. Sebei, K., Debez, A., Herchi, W., Boukhchina, S., Kallel, H. (2007). Germination kinetics and seed reserve mobilization in two flax (Linum usitatissimum L.) cultivars under moderate salt stress. Journal of Plant Biology, 50 (4), 447–454. doi: https://doi.org/10.1007/bf03030681
4. Wang, H., Wang, J., Guo, X., Brennan, C. S., Li, T., Fu, X. et al. (2016). Effect of germination on lignan biosynthesis, and antioxidant and antiproliferative activities in flaxseed (Linum usitatissimum L.). Food Chemistry, 205, 170–177. doi: https://doi.org/10.1016/j.foodchem.2016.03.001
5. Katare, C., Saxena, S., Agrawal, S., Prasad, G. (2012). Flax Seed: A Potential Medicinal Food. Journal of Nutrition & Food Sciences, 02 (01). doi: https://doi.org/10.4172/2155-9600.1000120
6. Gutte, K. B., Sahoo, A. K., Ranveer, R. C. (2015). Bioactive Components of Flaxseed and its Health Benefits. International Journal of Pharmaceutical Sciences Review and Research, 31 (1), 42–51. Available at: https://www.researchgate.net/publication/273441906_Bioactive_Components_of_Flaxseed_and_its_Health_Benefits
7. Kraevska, S., Stetsenko, N., Korol, O. (2017). Comparing Between The Amino Acid Composition Of Flax Seeds Before And After Germination. Agrobiodiversity for Improving Nutrition, Health and Life Quality, 253–257. doi: https://doi.org/10.15414/agrobiodiversity.2017.2585-8246.253-257
8. Singh, K. K., Mridula, D., Rehal, J., Barnwal, P. (2011). Flaxseed: A Potential Source of Food, Feed and Fiber. Critical Reviews in Food Science and Nutrition, 51 (3), 210–222. doi: https://doi.org/10.1080/10408390903537241
9. Edel, A. L., Aliani, M., Pierce, G. N. (2015). Stability of bioactives in flaxseed and flaxseed-fortified foods. Food Research International, 77, 140–155. doi: https://doi.org/10.1016/j.foodres.2015.07.035
10. Ghosal, S., Bhattacharyya, D. K., Bhowal, J. (2022). Production, characterization, and storage stability of nutritionally enriched flaxseed-based spread. Journal of Food Processing and Preservation, 46 (5). doi: https://doi.org/10.1111/jfpp.16574
11. Goyal, A., Sharma, V., Upadhyay, N., Gill, S., Sihag, M. (2014). Flax and flaxseed oil: an ancient medicine & modern functional food. Journal of Food Science and Technology, 51 (9), 1633–1653. doi: https://doi.org/10.1007/s13197-013-1247-9
12. Kovalova, O., Vasylieva, N., Stankevych, S., Zabrodina, I., Haliasnyi, I., Gontar, T. et al. (2023). Determining the effect of plasmochemically activated aqueous solutions on the bioactivation process of sea buckthorn seeds. Eastern-European Journal of Enterprise Technologies, 2 (11 (122)), 99–111. doi: https://doi.org/10.15587/1729-4061.2023.275548
13. Minevich, I. E., Nechiporenko, A. P., Goncharova, A. A., Sitnikova, V. E. (2021). Dynamics of macronutrients during short-term germination of flax seeds. Proceedings of Universities. Applied Chemistry and Biotechnology, 11 (3), 449–459. doi: https://doi.org/10.21285/2227-2925-2021-11-3-449-459
14. Herchi, W., Bahashwan, S., Sebei, K., Ben Saleh, H., Kallel, H., Boukhchina, S. (2015). Effects of germination on chemical composition and antioxidant activity of flaxseed (Linum usitatissimum) oil. Grasas y Aceites, 66 (1), e057. doi: https://doi.org/10.3989/gya.0463141
15. Villeneuve, S., Power, K. A., Guévremont, E., Mondor, M., Tsao, R., Wanasundara, J. P. D. et al. (2014). Effect of a Short-Time Germination Process on the Nutrient Composition, Microbial Counts and Bread-Making Potential of Whole Flaxseed. Journal of Food Processing and Preservation, 39 (6), 1574–1586. doi: https://doi.org/10.1111/jfpp.12385

16. Wu, Y., Wang, H., Wang, Y., Brennan, C. S., Anne Brennan, M., Qiu, C., Guo, X. (2020). Comparison of lignans and phenolic acids in different varieties of germinated flaxseed (Linum usitatissimum L.). International Journal of Food Science & Technology, 56 (1), 196–204. doi: https://doi.org/10.1111/ijfs.14619
17. Li, X., Li, J., Dong, S., Li, Y., Wei, L., Zhao, C. et al. (2019). Effects of germination on tocopherol, secoisolarlciresinol diglucoside, cyanogenic glycosides and antioxidant activities in flaxseed (Linum usitatissimumL.). International Journal of Food Science & Technology, 54 (7), 2346–2354. doi: https://doi.org/10.1111/ijfs.14098
18. Kajla, P., Sharma, A., Sood, D. R. (2014). Flaxseed – a potential functional food source. Journal of Food Science and Technology, 52 (4), 1857–1871. doi: https://doi.org/10.1007/s13197-014-1293-y
19. Liu, J., Shim, Y. Y., Tse, T. J., Wang, Y., Reaney, M. J. T. (2018). Flaxseed gum a versatile natural hydrocolloid for food and non-food applications. Trends in Food Science & Technology, 75, 146–157. doi: https://doi.org/10.1016/j.tifs.2018.01.011
20. Bekhit, A. E.-D. A., Shavandi, A., Jodjaja, T., Birch, J., Teh, S., Mohamed Ahmed, I. A. et al. (2018). Flaxseed: Composition, detoxification, utilization, and opportunities. Biocatalysis and Agricultural Biotechnology, 13, 129–152. doi: https://doi.org/10.1016/j.bcab.2017.11.017
21. Kurt, O. (2012). A predictive model for the effects of temperature on the germination period of flax seeds (Linum usitatissimum L.). Turkish Journal of Agriculture and Forestry, 36 (6), 654–658. doi: https://doi.org/10.3906/tar-1202-13
22. Bayat, M., Zargar, M., Murtazova, K. M.-S., Nakhaev, M. R., Shkurkin, S. I. (2022). Ameliorating Seed Germination and Seedling Growth of Nano-Primed Wheat and Flax Seeds Using Seven Biogenic Metal-Based Nanoparticles. Agronomy, 12 (4), 811. doi: https://doi.org/10.3390/agronomy12040811
23. Kovaliova, O., Chursinov, Y., Kalyna, V., Khromenko, T., Kunitsia, E. (2020). Investigation of the intensive technology of food sprouts using organic acids. EUREKA: Life Sciences, 2, 45–53. doi: https://doi.org/10.21303/2504-5695.2020.001204
24. Pivovarov, O., Kovaliova, O. (2019). Features of grain germination with the use of aqueous solutions of fruit acids. Food Science and Technology, 13 (1). doi: https://doi.org/10.15673/fst.v13i1.1334
25. Kovaliova, O., Tchoursinov, Y., Kalyna, V., Koshulko, V., Kunitsia, E., Chernukha, A. et al. (2020). Identification of patterns in the production of a biologically-active component for food products. Eastern-European Journal of Enterprise Technologies, 2 (11 (104)), 61–68. doi: https://doi.org/10.15587/1729-4061.2020.200026
26. Pivovarov, O., Kovaliova, O., Koshulko, V. (2020). Effect of plasmochemically activated aqueous solution on process of food sprouts production. Ukrainian Food Journal, 9 (3), 576–587. doi: https://doi.org/10.24263/2304-974x-2020-9-3-7
27. Kovaliova, O., Pivovarov, O., Koshulko, V. (2020). Study of hydrothermal treatment of dried malt with plasmochemically activated aqueous solutions. Food Science and Technology, 14 (3). doi: https://doi.org/10.15673/fst.v14i3.1799
28. Pivovarov, O., Kovaliova, O., Khromenko, T., Shuliakevych, Z. (2017). Features of obtaining malt with use of aqueous solutions of organic acids. Food Science and Technology, 11 (4). doi: https://doi.org/10.15673/fst.v11i4.728
29. Fang, Y., Franke, C., Manthei, A., McMullen, L., Temelli, F., Gänzle, M. G. (2021). Effects of high-pressure carbon dioxide on microbial quality and germination of cereal grains and beans. The Journal of Supercritical Fluids, 175, 105272. doi: https://doi.org/10.1016/j.supflu.2021.105272
30. Zhao, Y., Patange, A., Sun, D., Tiwari, B. (2020). Plasma-activated water: Physicochemical properties, microbial inactivation mechanisms, factors influencing antimicrobial effectiveness, and applications in the food industry. Comprehensive Reviews in Food Science and Food Safety, 19 (6), 3951–3979. doi: https://doi.org/10.1111/1541-4337.12644
31. Bourke, P., Ziuzina, D., Boehm, D., Cullen, P. J., Keener, K. (2018). The Potential of Cold Plasma for Safe and Sustainable Food Production. Trends in Biotechnology, 36 (6), 615–626. doi: https://doi.org/10.1016/j.tibtech.2017.11.001
32. Lin, C.-M., Chu, Y.-C., Hsiao, C.-P., Wu, J.-S., Hsieh, C.-W., Hou, C.-Y. (2019). The Optimization of Plasma-Activated Water Treatments to Inactivate Salmonella Enteritidis (ATCC 13076) on Shell Eggs. Foods, 8 (10), 520. doi: https://doi.org/10.3390/foods8100520
33. Pivovarov, O. A., Kovaleva, O. S., Chursinov, J. O. (2020). Prevention of biofouling of industrial reverse water supply systems by plasma water treatment. 3 nd International Scientific and Technical Internet Conference "Innovative development of resource-saving technologies and sustainable use of natural resources". Petroșani: UNIVERSITAS Publishing, 50–52. Available at: https://dspace.dsau.dp.ua/handle/123456789/5295
34. Pivovarov, O., Kovalova, O., Koshulko, V., Aleksandrova, A. (2022). Study of use of antiseptic ice of plasma-chemically activated aqueous solutions for the storage of food raw materials. Food Science and Technology, 15 (4). doi: https://doi.org/10.15673/fst.v15i4.2260
35. Kovaliova, O., Pivovarov, O., Kalyna, V., Tchoursinov, Y., Kunitsia, E., Chernukha, A. et al. (2020). Implementation of the plasmochemical activation of technological solutions in the process of ecologization of malt production. Eastern-European Journal of Enterprise Technologies, 5 (10 (107)), 26–35. doi: https://doi.org/10.15587/1729-4061.2020.215160
36. Pivovarov, O., Kovaliova, O., Koshulko, V. (2022). Effect of plasma-chemically activated aqueous solutions on the process of disinfection of food production equipment. Food Science and Technology, 16 (3). doi: https://doi.org/10.15673/fst.v16i3.2392
37. Greene, W. (2017). Econometric Analysis. London: Pearson Learning Solutions, 1176.
38. Wooldridge, J. M. (2012). Introductory Econometrics: A Modern Approach. Boston, MA: Cengage Learning, 910.
39. Erkmen, O. (2022). Microbiological analysis of foods and food processing environments. Academic Press. doi: https://doi.org/10.1016/c2021-0-01219-0
40. Vasylieva, N. (2013). Forecasting of prices in the field of crops-growing in Ukraine and regions. Economic Annals-XXI, 11-12 (2), 26–29. Available at: http://ea21journal.world/index.php/ea-v136-07/