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PLASMA-CHEMICALLY ACTIVATED WATER INFLUENCE ON STALING AND SAFETY OF SPROUTED BREAD

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Abstract. The article reveals the research results of freshness and safety of sprouted wheat bread made with the use of water additionally treated with nonequilibrium low-temperature contact plasma. Prospects of the use of dispersion of wheat grain for the wholegrain bread production are shown which allow decreasing the grain raw materials loss along the food chain. The ways of prolongation of bread freshness during storage and slowdown of bread staling are analyzed. It is shown that in case of usage of plasma-chemically activated water in the technology wheat grain soaking duration decreases by 30%. The additional water treatment also promotes bread freshness prolongation up to two days. It is determined that water gets the characteristics relevant for the technology after 30–40 minutes of treatment with nonequilibrium low-temperature contact plasma. According to our findings, usage of plasma-chemically activated water provides slowdown of water migration and moisture loss by the crumb. It is determined that the usage of additional water treatment in the technology results in 17–60% increase in hydrophilic properties of the crumb and slows down their reduction during storage of sprouted wheat bread. Results of differential thermal analysis showed changes in various forms of moisture binding in the product during storage and increase in the part of adsorptionally bound moisture by 6–8%, when additional water treatment is used for grain soaking and dough making in sprouted wheat bread making technology. The rate of moisture removal from crumb of sprouted wheat bread made with the use of water subjected to nonequilibrium low-temperature contact plasma was determined through mathematical processing of data and construction of piecewise linear model. It is proved that safety level of usage of plasma-chemically activated water in sprouted wheat bread technology meets the requirements to the content of heavy metals such as mercury, arsenic, copper, lead, cadmium, zinc, and mycotoxins (aflatoxin B1, deoxynivalenol, zearalenone).

Key words: wheat sprouted bread, plasma-chemically activated water, differential-thermal analysis, staling, crumbling, crumb swelling, bonded water, free water, safety characteristics.

ВПЛИВ АКТИВОВАНОЇ ПЛАЗМОЮ ВОДИ НА ПРОЦЕСИ ЧЕРСТВІННЯ І БЕЗПЕЧНІСТЬ ЗЕРНОВОГО ХЛІБА

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Анотація. У статті представлено результати досліджень свіжості і безпечності хліба, виготовленого із диспергованої зернової маси пшениці при використанні у технології додаткової обробки води контактною нерівноважною низькотемпературною плазмою. Показано перспективи застосування диспергування зерна пшениці для виробництва цільнозернового хліба, що дозволяє досягти зниження втрат зернової сировини вздовж продовольчого ланцюга. Проаналізовано шляхи подовження тривалості зберігання хлібом свіжості та затримання процесів черствіння готової продукції. Встановлено, що за умови застосування у технології плазмохімічно активованої води тривалість замочування зерна пшениці скорочується на 30%. При цьому додаткова обробка води сприяє подовженню свіжості виробів на дві доби. Виявлено, що раціональні для технології характеристики вода набуває після 30–40 хв обробки контактною нерівноважною низькотемпературною плазмою. Показано, що використання плазмохімічно активованої води викликає сповільнення процесів міграції вологи та її втрати м'якушкою. Виявлено, що застосування додаткової обробки води у технології призводить до зростання на 17–60% гідрофільності м'якушки та уповільнює її зниження під час зберігання хліба із диспергованої зернової маси пшениці. За результатами диференційно-термічного аналізу виявлено зміни різних форм зв'язку вологи у продукті під час зберігання та показано збільшення на 6–8% частки адсорбційно зв'язаної вологи при використанні у технології цільнозернового хліба додаткової обробки води для замочування зерна і приготування тіста. Шляхом математичної обробки даних та побудови кусково-лінійної моделі, визначено швидкість видалення вологи із м'якушки зернового хліба, виготовленого з використанням води, підданої дії контактної нерівноважної низькотемпературної плазми. Доведено, що рівень безпечності застосування плазмохімічно активованої води у технології хліба із диспергованої зернової маси пшениці задовольняє вимоги щодо вмісту таких важких металів як ртуть, мишяк, мідь, свинець, кадмій, цинк і мікотоксинів (афлотоксину В1, дезоксиніваленолу, зеараленону).

Ключові слова: хліб із диспергованої зернової маси, плазмохімічно активована вода, дериватографічний аналіз, черствіння, кришкуватість, набухання, зв'язана волога, вільна волога, показники безпеки.

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Introduction. Formulation of the problem

According to data provided by FAO, more than one third of all food raw materials in the world is lost, which entails the exhaustion of the labor, ecological, material and energy resources [1]. Furthermore, losses of the grain raw materials such as wheat, rye, and oats are almost 10% higher than the corresponding losses for oil-bearing crops [2]. The greatest losses along the food chain occur at its final stage, i.e. in the process of consumption of food products.

Development of resource-saving technologies is the prerogative of the food industry. At the same time, this effect can be achieved through the integrated approach and reduction of losses at different stages of the food product making [3]. Production of sprouted bread allows excluding the stage of grain processing to obtain flour and preserving all useful biologically active substances in the finished product [4]. Development and improvement of the technological aspects of sprouted bread production is considered in a number of works [5-8]. However, the issue of provision of freshness of such products and, as a consequence, reduction of losses at the stage of consumption of bread is still relevant.

Analysis of recent research and publications

Bread belongs to food products with the relatively high moisture content, which reduces duration of the period of its consumer properties' stability [9]. Besides, loss of quality characteristics of bakery products is an inevitable process. Since the staling, along with microbiological damage, limit the life cycle of bakery products, various attempts are made to extend the period of maintenance of bread freshness. It is known that the indicators of freshness of bread depend on raw materials used for making of bakery products [10-15]. At present time, extension of the periods of product storage is a priority task for the baking industry; therefore, in order to solve this issue, the scientists conduct active studies in this direction.

Decrease in bread quality is associated with the partial loss of moisture, development of negative microflora and staling of products [16-19]. Bread staling results in deterioration of consumer properties of finished products, i.e. increase in crumbling, loss of taste and aroma [20,21]. In its turn, this process combines a number of transformations in the structure of biopolymers [22,23]: reduction of hydrophilic properties of bread, change of the product microstructure, transition of starch from the amorphous to crystalline state. Today there are many technological approaches to extension of the term of bread freshness which include freezing, packaging in conditions of the gas environment of a certain composition, intensification of bioconversion of flour raw materials, usage of various additives of chemical origin, and others [9]. Modern trends in healthy nutrition are aimed at exclusion of artificial additives. Attempts to use various methods of raw materials' treatment might be of interest [24-26]. Water is one of the main ingredients in the baking formulas. Owing to treatment of water using physical and chemical methods, directed regulation of its properties is achieved. In particular, usage of nonequilibrium contact plasma for treatment of water and aqueous solutions showed itself as an efficient method of acceleration of the production processes and increase of quality of bread made traditionally of wheat flour [27] and sprouted bread [6].

Despite a number of advantages, production of sprouted bread demands studying of processes of the product quality loss during storage. Therefore, it is relevant to study the changes in quality of sprouted bread made with the use of plasma-chemically activated water and to evaluate safety of products, since along with the large number of peripheral particles the final product incurs the risk of contamination by heavy metals and mycotoxins.

Aim of the work is the research of processes of changing quality of sprouted bread made with the use of water subjected to nonequilibrium contact plasma during storage and the level of safety of final product.

To achieve the objective, the following **tasks** were specified:

1. Identification of changes in quality indicators which characterize staling processes for the sprouted bread made with the of water treatment by nonequilibrium low-temperature contact plasma in the technology;
2. Establishing of the effect of plasma-chemically activated water on the forms of moisture binding by the differential thermal analysis;
3. Determination of safety indicators of sprouted bread made with the use of plasma-chemically activated water.

Research Materials and Methods

In the course of studies we used winter wheat grain of "Antonovka" grade with the quality indicators as below: grain moisture content – 13.8%, natural weight – 822 g/l, falling number – 381 s, amount of crude gluten – 20.6%, and value of gluten deformation – 62 units. As the recipe components, "Lvivski" pressed bakery yeast (TS U 10.8-00383320-001); dry wheat gluten of CARGILL TM (TS 9189-005-00365517-06); "Agram dark" food additive (TS 9293-024-18256266-03) were used. For soaking the grain and mixing the dough we used untreated drinking water of Dnipro city water main (Table 1) and water subjected to nonequilibrium contact plasma during 10, 20, 30 and 40 minutes, with the characteristics set forth in Table 2. Since the alkaline earth metals do not form any sediment as a result of water plasma-chemical treatment, and amount of other elements dissolved in drinking water is minimal, water after treatment was used without the additional treatment operations. Water was treated in the laboratory of plasma-chemical technologies of "Ukrainian State University of Chemical Technology" SHEI on the experimental unit including power supply unit, vacuum pump and 0.5 dm^3 reactor made of molybdenum glass, with the cooling jacket [28]. Operating parameters of the unit are shown in Table 3. Bottom part of the reactor comprises the fixed cathode of stainless steel. Anode of stainless steel is mounted in the reactor above the surface of water which is poured into reactor at the distance of (7–15)$\cdot 10^{-3}$ m. Pressure in the reactor is 100 KPa. Plasma-chemically activated was stored for 2–3 days at the temperature of 5–10°C.

Wheat grain was soaked at hydro-module 1:1 and temperature of 18–22°C, with the duration of operation of 16 and 24 hours and washing of grain after soaking. Wheat grain was dispersed before formation of the homogenous grain mass. Other components according to the recipe [6] were added to the prepared grain amass, and the dough was mixed.

Changes in the consumer properties of bread were studied in 24, 48 and 72 hours after baking. Tests samples were stored at the relative air humidity of 75%.

The content of forms of bound moisture in bread was determined using derivatograph “Paulik-Paulik-Erdey” Q-1500D. The sample was heated in the temperature range of 20-250°C at the rate of 2.5°/min.

Table 1 – Tap water quality indicators (Dnipro city)

No.	Name of the indicator	Result of measurement
1	Water color, deg.	11.58
2	Odor score	1
3	Turbidity, mg/dm^3	4.75
4	Hydrogen index, pH units	7.30
5	Total alkalinity, mg-eq. HCO_3^-/dm^3	3.29
6	Bicarbonates, HCO_3^-/dm^3, mg/dm^3	200.75
7	Dry residue (insoluble matter), mg/dm^3	290
8	Hardness, mg-eq/dm^3	4.10
9	Calcium, mg/dm^3	70.14
10	Magnesium, mg/dm^3	7.29
11	Sodium and potassium, mg/dm^3	3.00
12	Iron, mg/dm^3	1.52
13	Manganese, mg/dm^3	0.27
14	Chlorides, mg/dm^3	27.65
15	Sulfates, mg/dm^3	8.16
16	Nitrates, mg/dm^3	1.07
17	Phosphates, mg/dm^3	0.045
18	Permanganate oxidation, mg O/dm^3	6.3
19	Nitrites, mg/dm^3	0.02
20	Ammonium (ammonia nitrogen), mg/dm^3	0.2
21	Copper, mg/dm^3	0.160
22	Lead, mg/dm^3	Less than 0.005
23	Zinc, mg/dm^3	0.020

Table 2 – Water characteristics before and after treatment with nonequilibrium low-temperature contact plasma

Sample No.	Duration of plasma action on water, minutes	Water pH	Concentration of peroxide compounds, mg/l	Redox potential, mV
1	–	7.30	–	240
2	10	9.61	100	140
3	20	9.80	200	122
4	30	10.18	400	109
5	40	9.72	500	105

Table 3 – Operating parameters of the unit for water treatment with nonequilibrium low-temperature contact plasma

Parameters	Power supply	Value
Input voltage	Single-phase alternating	~50 Hz; 220 V
Output voltage	Direct, pulsating, regulated within	700–1,500 V
Load current	Maximum value	0.3 A
Ignition voltage	Amplitude Pulse duration	12,000–15,000 V 1.0–1.5 ms

Toxic elements were determined by the method atomic adsorption spectrophotometry using the device Shimadzu AA-6300 (GOST 26927-86, DSTU 7670:2014, GOST 30178-96), mycotoxins – by the method of thin layer chromatography (MRI 2273-80, MU 5177-90). Determination of kinetic characteristics of various forms of moisture binding was carried out by mathematical processing of data of the derivatographic analysis and construction of the piecewise linear model in the MS Excel environment.

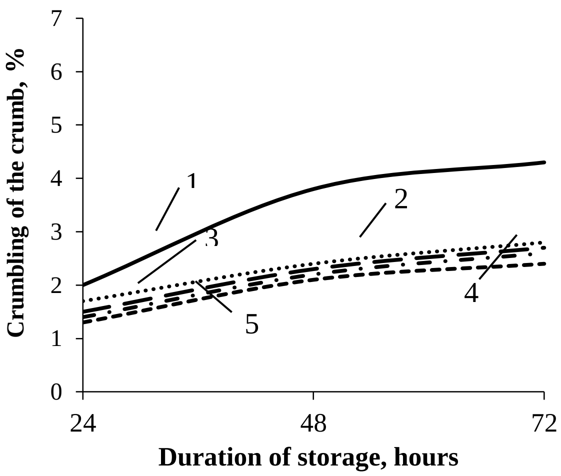
Results of the research and their discussion

Storage of bread is accompanied by gradual loss of its moisture, due to sufficiently high content of water in the product. Sprouted bread, as distinct from wheat varieties of bread, features higher content of shells, which cause the increase in water-absorbing capacity of the dough and, accordingly, growth of moisture in the final product [6]. As it is seen from data of Table 4, losses of moisture during 72 hours of storage for all samples under study vary within 0.6–1.45%. Control samples of sprouted bread are characterized by higher loss of moisture with less time of grain soaking (decreased from 24 to 16 hours). At the same time, compared to control samples, the losses of moisture slow down almost twice when the grain is soaked for 16 hours and dough is mixed with the use of water subjected to nonequilibrium contact plasma. It should be noted that increase in duration of wheat grain soaking on condition of usage of treated water causes certain leveling of the process of loss of moisture slowdown compared to control.

Table 4 – Effect of duration of grain soaking on moisture content of sprouted bread during storage

Duration of bread storage, hours	Duration of water treatment with plasma, minutes				
	0	10	20	30	40
Soaking of grain for 16 hours					
24	48.3	48.9	49.0	48.6	48.6
48	47.9	48.7	48.8	48.2	48.2
72	47.6	48.6	48.7	48.1	48.1
Soaking of grain for 24 hours					
24	47.2	47.3	48.4	48.3	46.9
48	47.1	47.1	48.3	48.1	46.8
72	46.8	46.8	47.9	47.8	46.4

Along with the loss of moisture, decline in consumer characteristics of bread is observed due to changes in the structure of biopolymers of the crumb. These changes provoke the appearance of additional gaps in the starch-protein matrix of bread. Crumbling indicator is important for the consumer and is considered the defining one, since it is directly related to the texture characteristics. It is found that duration of grain soaking has its effect on crumbling of the bread crumb (Fig. 1). For sprouted bread made with the use of tap water in 24 hours after baking this indicator makes 2–3.5%; when the period of storage is increases to 72 hours it grows twofold. At the same time, test samples obtained from grain after 16 hours of soaking are characterized by the change in crumbling within 0.5% (Fig. 1a). Therefore, usage of plasma-chemically activated water in the technology of grain bread promotes decrease in crumbling of the crumb by 15–45%, on condition of the usage of water activated during 30–40 minutes, and soaking of grain for 16 hours.



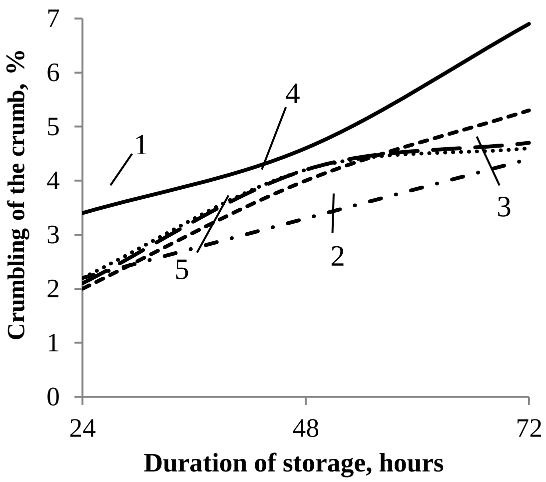
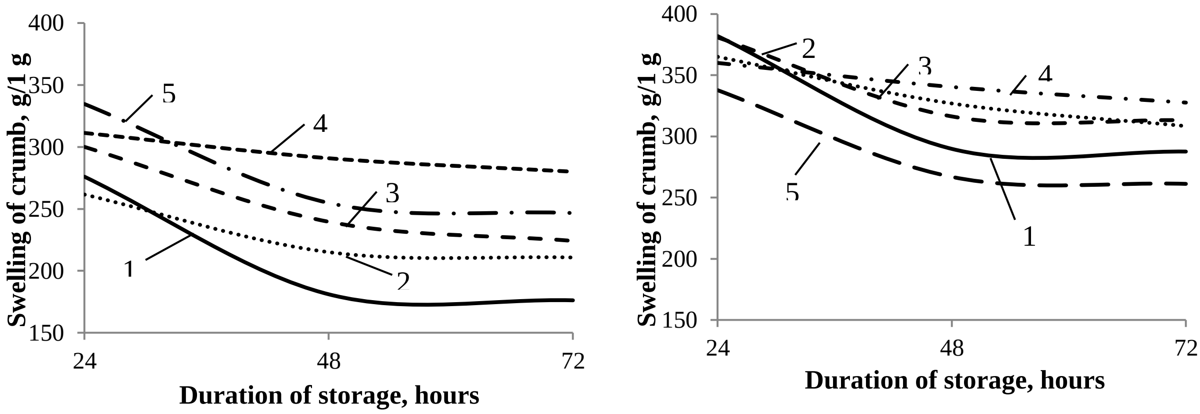


Fig. 1. Change in crumbling of bread crumb depending on grain soaking duration: a – 16 hours; b – 24 hours. Duration of water treatment, minutes: 1 – 0; 2 – 10; 3 – 20; 4 – 30; 5 – 40

One of properties of fresh bread is high moisture susceptibility of the crumb. However, as a result of ageing of biopolymers, this property is gradually lost. Fig. 2 shows the change in the degree of swelling of crumb of sprouted wheat bread during storage. With the increase in duration of grain soaking, the growth of the degree of swelling of the crumb occurs, which is obviously due to activation of the processes of conversion of high-molecular substances into low-molecular ones as a result of activation of biochemical processes at the expense of prolonged action of water on the grain biopolymers. The largest effect from increase in the soaking duration is observed for the control sample of sprouted wheat bread. Hydrophilic properties of the crumb of bread made according to the traditional technology are the most significantly lost during the first 36 hours of storage, so the indicator changes to 24–34% with regard to initial values. Hydrophilic properties of the crumb for the test samples is 17–60% higher compared to control, and they are lost at the lower rate. In some cases, such as the use of plasma-chemically activated water with the duration of treatment of 30 minutes for soaking of grain and mixing of dough, the loss of ability to absorb moisture by the crumb is reduced significantly. Effect of plasma-chemically activated water on wheat starch [29] is evident.



a b

Fig. 2. Change in the degree of swelling of bread crumb depending on the soaking duration: a – 16 hours, b – 24 hours. Duration of water treatment, minutes: 1 – 0; 2 – 10; 3 – 20; 4 – 30; 5 – 40

High moisture content of grain bread requires special attention with regard to the mechanisms of dynamics of changes in the associative bonds of moisture with biopolymers of the crumb. Table 5 shows the derivatographic analysis of the studied bread samples made of whole grain on condition of soaking for 16 hours. It reflects the distribution of moisture by the forms of binding depending on water used in the technology (C – sample made with the use of untreated water; E – sample made with the use of water subjected to nonequilibrium contact plasma for 30 min).

Table 5 – Forms of moisture binding in grain bread crumb during storage

Forms of moisture binding		**Amount of moisture,%**	**Temperature of removal, °C**		**Amount of moisture,%**	**Temperature of removal, °C**	
			Interval	Peak		Interval	Peak
		24 hours of storage			72 hours of storage		
Free moisture	C	83.1	20–108	–	83	20–117	–
	E	74.7	20–78	–	76,9	20–120	–
Mechanically bound	C	47.9	20–82	–	35.9	20–74	–
	E	35.3	20–51	–	29.3	20–79	–
Osmotically bound	C	35.2	82–108	100	47.1	74–117	107
	E	39.4	51–80	80	47.6	79–120	110
Adsorptionally bound	C	16.9	108–245	–	17	117–250	–
	E	25.3	78–248	–	23.1	120–250	–
Of poly-molecular layers	C	11.3	108–214	–	9.8	117–204	–
	E	14	78–208	–	15.5	120–220	–
Of mono-molecular layers	C	5.6	214–245	–	7.2	204–250	–
	E	11.3	208–248	–	7.5	220–250	–

For example, content of free moisture in control samples exceeds similar indicators of test samples by 6–8%. Mechanically bound moisture prevails in the products made with the use of tap water, while osmotically bound moisture – in the products made with the use of plasma-chemically activated water. It indicates the increase in the degree of swelling of proteins which may be conditioned by high penetrating power of treated water, expressed in particular in reducing the surface tension of water after treatment with plasma [30]. The endothermic peak of moisture removal falls on its osmotically bound form for both experimental and control samples, which is generally typical for all bakery products. Concerning the control samples, the ratio of the adsorption moisture of the mono- and poly-molecular layers absorbed by the starch is 2:1, in contrast to the test ones, where the preference is given to the most strongly bound form of moisture. Obviously, this is due to micro-cluster structure of plasma-chemically activated water: large number of -OH groups in the starch molecule causes its tendency to form the hydrogen bonds. Since water changes its structure under action of nonequilibrium contact plasma, it is quite probable that the starch adsorption capacity grows and, consequently, the amount of strongly bound moisture in bakery products with its use increases. During storage for 72 hours, moisture redistribution is the crumb occurs. For the test samples, we found the reduction in the proportion of mechanically bound moisture; the amount of adsorptionally bound moisture decreases to 2%, and the migration of moisture of mono-molecular layers to poly-molecular ones occurs.

Mathematical data processing allowed revealing the rates of moisture removal at certain stages of heat treatment process (Fig. 3). For example, the rate of moisture removal is distributed regardless of the duration of storage of products and water used for soaking of grain and preparation of the dough, as follows: mechanically bound moisture > moisture of swelling > adsorption moisture of mono-molecular layers > adsorption moisture of poly-molecular layers. It should be noted that higher content of moisture of the poly-molecular layers is typical for bread which is made of flour [31]. It is evident that swelling of starch granules occurs during soaking of the grain, and water penetrates deeper into the granule. Along with activation of amylolytic enzymes, it promotes the change in redistribution of adsorptionally bound moisture.

Usage of nontraditional methods of raw material processing usually raises the questions of safety of the final product for the consumer. The use of water subjected to nonequilibrium contact plasma showed no toxic effects with regard to physiological state of hydrobionts in the course of complex biotests [28]. It should be noted that production of bread of whole grain which excludes the removal of shells, along with the existing biological benefits of consumption, may face the environmental problems of the present time. The level of contamination by heavy metals, in particular, in the industrial regions of the country is rather high. It can adversely affect the safety of food products that do not require removal of external shells of plant raw materials capable of accumulating the toxic elements. As a result of studies, it is found that content of toxic elements in the sprouted bread made both using untreated water and plasma-chemically activated water is within quantities permissible by the State standards (Table 3). It should be noted that lead content is the closest to threshold values for both the test and control samples. This is evidently due to ecological conditions of grain growing in Dnipropetrovsk region which is confirmed by the data of the work [32] and the revealed facts of lead metal bearing among the local population.

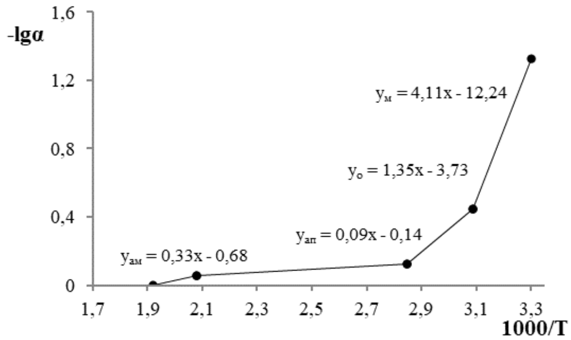


Fig. 3. Piecewise linear function for removal of moisture from sprouted bread with the use of plasma-chemically activated water (treated during 30 minutes)

Table 3 – Sprouted bread safety indicators

Indicator	Requirements of DSTU-P 4583:2006	Bread made of wheat grain with the use of water treated during (minutes)	
		0	30
Toxic elements, weight fraction, mg/kg			
Mercury	0.01	Less than 0.00015	Less than 0.00015
Arsenic	0.1	Less than 0.0025	Less than 0.0025
Copper	5	1.64	0.92
Lead	0,3	0.231	0.206
Cadmium	0.05	Less than 0,01	Less than 0,01
Zinc	25	12.74	13.86
Mycotoxins, mg/kg			
Aflatoxin B1	0.005	Less than 0.001	Less than 0.001
Deoxynivalenol	0.5	Less than 0.2	Less than 0.2
Zearalenone	1	Less than 0.1	Less than 0.1

Today the risks of contamination of food products by mycotoxins attract a lot of attention, especially when the technological conditions of production are favorable for development of the negative microflora. In the conditions of excess moisture created during soaking of grain in the grain bread making technology, microbiological risks are increasing. In terms of the content of mycotoxins, being secondary metabolites of mold fungi, namely aflatoxin B1, vomitoxin and zearalenone, test samples are safe for consumption. It creates the preconditions for expansion of the niche market segment of bakery products with the safe and useful spouted bread.

Conclusions

Loss of moisture by the sprouted bread crump during storage slows down twice when water subjected to nonequilibrium low-temperature contact plasma is used for soaking of grain during the period which is 8 hours shorter compart-ed to that of the traditional technology. During storage of wholegrain bread, usage of plasma-chemically activated water reduces crumbling of the crumb by 15–45%, and 17–60% increase in hydrophilic properties is observed compared to the control, which indicates the product freshness prolongation up to two days.

Usage of water subjected to nonequilibrium low-temperature contact plasma for making of wholegrain wheat bread results in the increase in bound moisture content in the product by 6–8% compared to the control, and binding of free moisture occurs through its absorption by proteins. The rate of moisture removal is distributed regardless of the duration of storage and water used for soaking of grain and preparation of the dough, as follows: mechanically bound moisture > moisture of swelling > adsorption moisture of monomolecular layers > adsorption moisture of polymolecular layers.

The content of heavy metals and mycotoxins in the sprouted bread made both using untreated water and plasma-chemically activated water is within quantities permissible by the state standards.

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