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PROSPECTS OF USING THE CRYOSTABILISING PROTEIN-POLYSACCHARIDE COMPOSITION TO MANUFACTURE SEMI-FINISHED CHOPPED MEAT PRODUCTS

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Abstract. One of the technological ways of improving the quality of frozen semi-finished chopped meat products is the use of protein-polysaccharide mixtures in their composition. The mechanism of the cryoprotective action of these mixtures involves the formation of the amorphous structure of the product, decrease in the number of crystallisation centres, and reduction of water activity, which is especially important for long-stored meat products at sub-zero temperatures. For this purpose, a protein-polysaccharide composition has been developed. It consists of blood plasma proteins, sodium caseinate, and dietary fibres of flax and plantain (*Plantago*) in the ratio 1:1:2:2. The regularities of the effect of the selected composition on the physicochemical properties of model forcemeat systems have been established. It has been proved that this composition allows eliminating the disadvantages of low-functional meat raw materials in the semi-finished meat products and counteracting the negative effects of their long-term storage in the frozen state. It has been found that 3% of the composite cryoprotectant mixture decreases the cryoscopic temperature of forcemeat systems by 2.09–2.81°C, reduces the mass fraction of frozen moisture by 1.7% and the water activity index by 0.031–0.067. The mathematical modelling and processing of the experimental data in the system of the mathematical package Mathcad 15 have allowed establishing that semi-finished chopped meat products with a mass fraction of the protein-polysaccharide composition contain up to 4% of frozen water when stored for up to 30 days at -10°C. Besides, the chemical makeup and functional and technological properties of model forcemeat systems containing different amounts of the protein-polysaccharide composition have been studied to develop recommendations how to use it as a cryoprotectant. It has been proved that using 3% of the protein-polysaccharide composition is technologically practical, since it is an effective cryoprotectant of forcemeat systems during their low-temperature processing. The research results obtained allow purposefully forming and stabilising the required functional, technological, structural, mechanical, and sensory characteristics of the final product.

Key words: semi-finished chopped products, blood plasma protein, sodium caseinate, dietary fibre, cryoprotectant.

Introduction. Formulation of the problem

One of the main tasks of the meat processing industry has always been to provide the Ukrainian people with quality food. The quality of finished products largely depends on the specific functional and technological properties of meat raw materials and on the physicochemical and microbiological processes involved in their technological treatment. So, it is necessary to use a comprehensive approach to study and evaluate the ingredients that determine the quantitative and qualitative characteristics of meat products.

That is why scientists should focus primarily on how meat products made from low-grade raw materials can be enriched with animal proteins and dietary fibres, which have high functional and technological properties. Usually, freezing storage of food is accompanied with protein denaturation and/or aggregation with partial loss of functional and technological properties. Water plays a major role in the process. Changing the concentration of salt solutions while water is freezing results in changes in the pH and strength of ionic interactions in the layer close to a protein molecule. These transformations lead to the dehydration and aggregation of protein macromolecules [1-3]. Semi-finished chopped products retain their quality after they are unfrozen only if the recommended refrigeration conditions have been created. Violation of the storage temperature requirements reduces the consumption properties of frozen semi-finished products: low temperatures can ruin the muscle fibre of meat raw materials if crystallisation of free water is excessive (it depends on the functional and technological properties of meat raw materials and semi-finished products) [4-5]. However, the molecules of a cryoprotectant can approach or bind to the protein molecules by any functional group and thus form hydrogen or ionic bonds as if covering their surface [4-6]. For this reason, the use of animal proteins and dietary fibre can make up for the poor quality of low-grade meat raw materials, prevent the negative effects of low temperatures during long-term storage, and improve the composition and quality of semi-finished frozen meat products. Given the above, semi-finished minced meat products should be frozen in the presence of effective cryoprotectants that can directly effect on the change in the phase state of moisture, as well as on the microflora activity, rheological characteristics, and quality of final products.

Analysis of recent research and publications

Low-temperature processing of meat and semi-finished meat products should be carried out under conditions ensuring that the consumer properties of meat products are as close to the native ones as possible and change neither during their long-term refrigerated storage nor during unfreezing. The action of substances with cryoprotective properties is aimed at creating hydrogen bonds with water molecules, which prevents their transformation into ice, contributes to the weakening of the effect of crystallisation, changes its character, prevents the aggregation and denaturation of macromolecules, and helps to preserve the integrity of cell membranes [7-9].

The properties of cryoprotectants in various biological objects and food systems have been researched in the works by P. Kapel, J. Lafon, J. Lebel, L. Sarafanov, A. Sutton, D. L. Huffman, B.E. Harrison, F. Kholodova, M. Yancheva, and others. These authors characterise cryoprotectants as substances that prevent or retard the growth of ice crystals, counteract the destructive effect of such damaging factors as formation of intracellular ice and dehydration of tissues, and protect cells against osmotic changes. Cryoprotectants can create hydrogen bonds with water molecules, thus preventing them from arranging into ice, weaken the effect of crystallisation by changing its character, prevent the aggregation and denaturation of protein macromolecules, and contribute to preserving the integrity of cell membranes [10-13].

Cryoprotectants penetrate into cells and interfere with the formation of ice crystals by creating hydrogen bonds with water molecules contained in cellular structures. The most studied ingredients with cryoprotective properties are glycerol, propylene glycol, ethyl glycol, dimethyl sulphoxide [14-16]. The principle of non-penetrating cryoprotectants is reducing the growth rate of crystals and protecting cells from osmotic changes. Non-penetrating cryoprotectors include two groups of substances: oligosaccharides (the best known of them are sucrose, trehalose, maltose, and lactose) and macromolecular compounds (albumin, blood plasma, polyvinylpyrrolidone) [17-18].

Cryoprotective properties are known to be characteristic of some food ingredients of polysaccharide nature (hydrocolloids, dietary fibres), which are high molecular weight substances, soluble and insoluble in water. They are widespread in nature, different in origin, chemical composition, properties, and area of application [17-18]. A promising direction aimed at minimising unwanted changes in meat raw materials is using cryoprotective food ingredients to prevent or slow down the growth of ice crystals in biotechnological objects and counteract the adverse effects of intracellular ice formation or tissue dehydration.

Scientists have proved that in technologies of meat products, it is practical to use different ratios and concentrations of different hydrocolloids, such as carrageenans, various gums – those of xanthan, guar, carob, tara tree, karaya, gum arabic, carboxymethyl cellulose, methyl cellulose, sodium alginate. These ingredients improve the functional, technological, and organoleptic characteristics, increase the output of meat products.

The analysis of literary sources on how freezing and low-temperature storage of semi-finished minced meat products effect on their quality suggests that these issues have not been sufficiently studied. This confirms the necessity of further research in this sphere.

The purpose of the work is to study the cryostabilising ability of the protein-polysaccharide composition added to semi-finished chopped meat products for purposeful formation and stabilisation of their functional, technological, structural, mechanical, and organoleptic characteristics during their long-term storage in the frozen state.

Objectives of the study:

1. On analysing scientific and technical information, to justify the way of solving the problem of the negative effects that freezing and long-term

storage at sub-zero temperatures have on the quality of frozen semi-finished meat products;

2. To investigate the cryostabilising ability of the protein-polysaccharide composition in the model samples of forcemeat systems and semi-finished chopped meat products by studying their physical, chemical, structural, and mechanical properties.

3. To study the effect of the protein-polysaccharide composition developed on the quality parameters of semi-finished chopped meat products under conditions of their long-term storage in the frozen state.

Research materials and methods

The composition of the pre-developed protein-polysaccharide composition: blood plasma protein Vepro 75 PSC (company *Veos*), sodium caseinate *DairyCo*, flax and plantain fibre (TU U 15.8-24239651-007:2007), in the ratio 1:1:2:2. The degree of hydration of the protein-polysaccharide composition is 1:6. Samples of model forcemeat systems were prepared as follows: for the control sample, the forcemeat was made according to the recipe of cutlets *Po-Domashnemu* (by the technological instruction in DSTU 4437:2005). When making the forcemeat, the protein-polysaccharide composition developed was added to the test cutlets in the amounts 2.0, 2.5, 3.0, and 3.5% to replace, respectively, 2.0, 2.5, 3.0, and 3.5% of second-grade beef and 12.0, 15.0, 18.0, 21% of bread [22].

Model forcemeat systems of the cutlets were made on the basis of second-grade beef and medium-fat pork at the ratio 1:1. The forcemeat samples obtained were stirred at 12°C for 15 min, moulded, and frozen at minus 18°C. The samples were stored at minus 10°C for 30 days. The test pieces of the cutlets were heat-treated before freezing, after freezing, and after unfreezing. The unfreezing was carried out at 20±2°C for 1.5–2 hours. The cutlets were fried until the temperature in their centre was 78–80°C.

The penetration tension of the semi-finished chopped products was determined with a penetrometer Ulab3-31M, by the depth of the indenter's immersion in the test sample at 20°C. Three measurements were made on the open surface of the sample at a distance of at least 10 mm from the edge of the product and at the maximum distance from the points of other measurements, so that the deformed part of the surface did not enter the measurement area. Then the penetration value was converted into the value of the penetration tension.

The water activity (a_w) of the model forcemeat systems and semi-finished chopped meat products was determined with an analyser Rotronic HygroPalm-23 [22,23]. The cryoscopic temperature of the model forcemeat systems and semi-finished chopped meat products was measured by the thermal analysis method based on the construction of curves of temperature change over time [23].

The moisture content was determined according to DSTU (State Standard of Ukraine) ISO 1442:2005 *Meat and meat products. Moisture content test method (control method) (ISO 1442:1997, IDT).*

The mass fraction of fat was determined according to DSTU ISO 1443:2005 *Meat and meat products. Method of determining the total fat content.*

The mass fraction of protein was determined by the Kjeldahl method: *Agricultural food products. General guidelines for the determination of nitrogen content by the Kjeldahl method (ISO 1871:1975, IDT).*

The mass fraction of mineral substances was determined by the gravimetric method after burning the organic substances in a muffle furnace at 500–700°C for 5–6 hours to constant mass [23].

The organoleptic characteristics of the cutlets were determined according to DSTU 4823.2:2007 *Meat products. Organoleptic evaluation of quality parameters. General requirements.*

The moisture-binding capacity (MBC) and moisture-holding capacity (MHC) were determined by centrifugation [18].

The moisture-binding capacity of the model meat systems (MBC, %) was evaluated using the method of R. Grau and R. Hamm (in V. Volovinska and B. Kelman's modification) by the ratio of the mass fractions of free and bound moisture [18].

The stability of the emulsion was determined by centrifugation of the test tube with the emulsion for 5 min at 1500 min^{-1}, followed by immersion of the tube in boiling water, repeated centrifugation, and calculation of the percentage of undestroyed emulsion.

The mathematical modelling was performed using the MathCAD software package.

Results of the research and their discussion

In order to counteract the negative effects of freezing on the functional and technological properties of second-grade meat raw materials and semi-finished chopped products, and to prevent significant crystallisation and slowdown in the freezing process, the cryoprotective properties of the protein-polysaccharide composition (PPC) based on animal proteins and vegetable fibre have been studied [7]. Blood plasma proteins Vepro 75 PSC and sodium caseinate were selected as cryostabilising components. Protein concentrates, as high molecular weight substances, can reduce the growth rate of crystals and protect muscle cells from osmotic and temperature fluctuations. They are also capable of stabilising meat systems during storage.

Flax and plantain fibre was used as a polysaccharide component, because it improves the consistency of food, adsorbs water, reduces weight loss, and enriches products with ballast substances [5,6]. Its principle of action is based on the formation of an amorphous structure in the food system, on reducing the number of crystallisation centres in it, and on a decrease in the water activity.

The chemical composition and functional and technological properties (FTP) of model forcemeat systems containing different amounts of the PPC have been studied to develop recommendations how to use it as a cryoprotectant. The results of the studies are presented in Table 1.

Table 1 – Chemical composition and functional and technological properties of model forcemeat systems containing PPC (n=3; P≥95)

Parameters	Control	Test samples containing the protein-polysaccharide composition, %			
		2.0	2.5	3.0	3.5
Moisture content, %	69.95±3.18	71.47±3.35	71.50±3.40	72.67±3.39	72.56±3.41
Mass fraction of protein, %	9.89±0.34	10.61±0.41	10.63±0.28	10.60±0.31	10.65±0.29
Mass fraction of fat, %	19.46±0.81	17.19±0.77	17.14±0.71	16.00±0.79	16.06±0.78
Ash content, %	0.70±0.02	0.73±0.02	0.73±0.01	0.73±0.02	0.73±0.02
pH	6.09±0.20	6.05±0.21	6.06±0.17	6.07±0.19	6.07±0.07
Moisture-binding capacity, %	78.20±3.6	79.09±3.5	79.85±3.6	80.10±3.7	81.14±3.5
Moisture-holding capacity, %	67.90±3.0	74.50±3.1	74.70±3.1	75.1±3.0	75.79±3.0
Fat-holding capacity, %	68.00±3.2	74.40±3.2	74.52±3.1	74.63±3.4	74.57±3.4
Emulsifying capacity, %	69.80±3.2	79.90±3.3	79.86±3.1	79.91±3.3	79.88±3.4
Emulsion stability, %	77.30±3.1	89.40±3.3	89.95±3.1	90.40±3.2	90.02±3.5
Penetration tension, Pa	17.25±0.74	24.87±0.95	27.37±1.06	27.54±1.08	27.63±1.07
Output	74.58±3.2	80.76±3.4	81.30±3.4	81.78±3.3	81.83±3.3

The research results confirm that the introduction of PPC in forcemeat systems as cryoprotectants increases the moisture-holding (by 9.7–17.3%) and fat-holding (by 9.4–9.7%) abilities of model meat systems, which improves the structure of model semi-finished chopped products. When the gelation temperature of the selected polysaccharides is close to the denaturation temperature of the meat proteins, water is separated from the meat proteins and absorbed by the protein-polysaccharide complexes.

An important parameter that determines the quality of semi-finished products is the stability of the forcemeat systems of semi-finished chopped products. This parameter characterises the content of bound water and fat in a meat system [8]. The results of the studies indicate that the stability of the meat systems of the test samples increased by 15.7–16.5%, compared to the control sample.

The results of the study characterise the ability of meat systems to absorb and hold moisture during heat treatment [9]. Determining the losses from the heat treatment of the model forcemeat systems has shown that the output is higher for the PPC-containing samples. This effect can be explained by the fact that during heating, the immobilised moisture is held better due to the capillary effect characteristic of food fibres, and due to the high technological properties of the thermostable proteins of blood plasma Vepro 75 PSC and sodium caseinate [10-12]. The most significant changes in the organoleptic quality parameters (lack of juiciness, brittleness of structure, lower output, losses from heat treatment that are bigger by 6.18–7.25%) were observed in the control samples after freezing, storage for 30 days, and defrosting.

The results of the organoleptic study of the model cutlets has shown that the use of the PPC developed does not change significantly the sensory properties of meat systems. The samples with 3.0% of PPC, juicier and with a stronger structure, were rated the best in comparison with the control sample (Fig. 1).

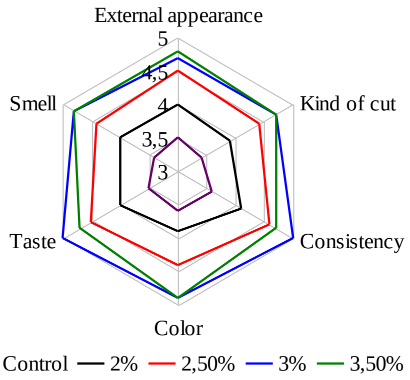

Fig. 1. Quality profiles of the model samples of semi-finished chopped meat products by the organoleptic characteristics and recipe composition

When determining the weight loss in the heat-treated model samples of semi-finished products, it was found that the use of 2.0–3.5% of PPC in their composition allowed reducing the weight loss, regardless of how they are treated. Thus, the weight loss in the unfrozen and heat-treated test samples of cutlets decreased, compared to the control sample, by 10.24–14.67%, respectively (Table 2).

Table 2 – Weight loss in heat-treated model forcemeat systems with protein-polysaccharide compositions

Type of treatment	Control	Test samples containing the protein-polysaccharide composition, %			
		2.0	2.5	3.0	3.5
Heat treatment before freezing	29.33±1.12	20.25±0.93	17.50±0.78	16.23±0.78	16.42±0.79
Freezing	2.64±0.12	1.96 ±0.09	1.76±0.07	1.65±0.080	1.66±0.08
Heat treatment after defrosting	33.68±1.39	23.44±1.14	20.49 ±0.98	18.93±0.92	19.02±0.90

All processes in the freezing and defrosting cycle occur in an extremely short period and in small volumes of the medium, with no possibility of measuring the temperature of the sample and the environment with thermometers and controlling the change in the solution's state of matter. That is why the kinetics of freezing the moisture in forcemeat systems was described mathematically.

Using the MathCad 15 mathematical package, we have obtained the multivariate regression equation:

$$Z=0.344-0.164x+0.041x^2+0.166y-0.003y^2-0.009xy$$

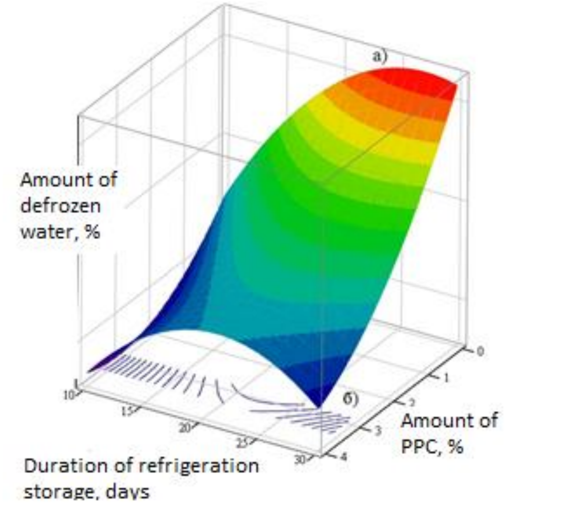

Fig. 2. Response plane (a) and the optimal lines showing the level (b) of dependence of the amount of frozen water on the amount of the PPC and on the duration of refrigeration (3D graph)

To verify the adequacy of the model obtained, the root-mean-square deviation (σ=0.012%) has been calculated, which indicates quite a high degree of reproducibility of the storage process at 10°C using the response plane.

On the basis of the search for extrema of the mathematical relation, the rational PPC content and the recommended duration of refrigeration treatment have been determined, with the minimum amount of frozen water equal to:

$$Y(x, y) \rightarrow \min \text{ with } y=3\%;\ y=10\text{-}30$$

So, it can be concluded that the use of 3% of the PPC can minimise the amount of frozen moisture over the period of storage required by the technology, at minus 10°C.

The preserving effect of the freezing process is aimed at reducing the activity of water a_{WB}, which contributes to the extension of the storage of meat systems [8]. The PPC as a cryoprotectant in model forcemeat systems also lowers the water activity by 0.031–0.067 compared to the control sample. The results of the study of water activity of samples 1–4 with 2.0, 2.5, 3.0, and 3.5% of the PPC are presented in Fig. 4.

The polynomial obtained (1) demonstrates the combined influence of the technological factors – the amount of the protein-polysaccharide composition (x) and the duration of refrigeration processing (y) – on the amount of frozen moisture (z) in the model samples of the semi-finished products under the experimental conditions. Fig. 2 and 3 show the response plane and the lines of constant values indicating how the amount of frozen water depends on the amount of the PPC and on the duration of refrigeration.

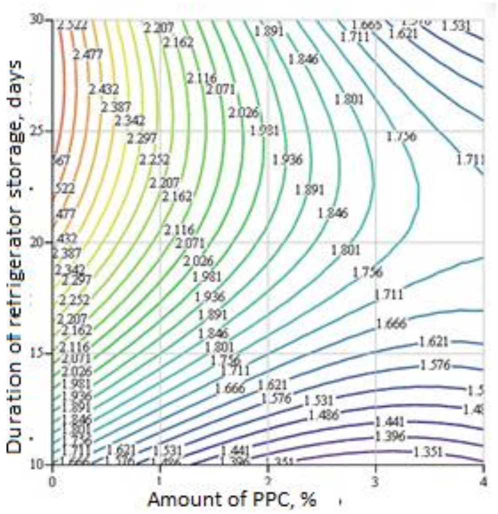

Fig. 3. Lines of constant values of the dependence of the amount of frozen water on the amount of protein-polysaccharide composition and the duration of refrigeration

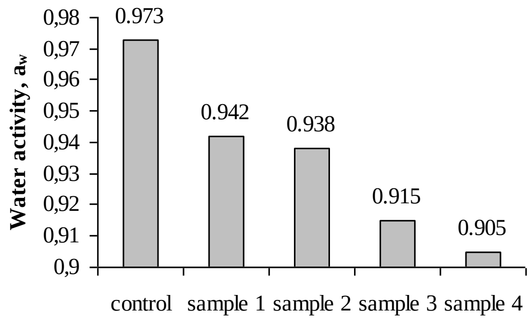

Fig. 4. Dynamics of changes in the water activity index of the heat-treated samples of semi-finished chopped products after 30 days of storage

The decrease in the water activity a_w when the PPC content in the test samples increases also leads to a decrease in the temperature at which moisture crystallisation begins in forcemeat systems and, accordingly, determines a change in the character of the water crystallisation process in the cellular structure of muscle tissue [8,9,10].

The values of the cryoscopic temperature of the test samples are shown in Fig. 5.

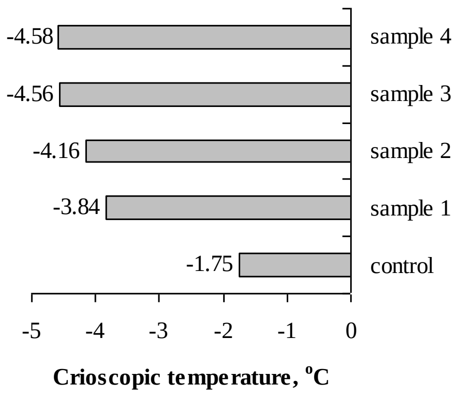

Fig. 5. Cryoscopic temperature of the samples of semi-finished chopped products

The results confirm the advantages of using the PPC in an amount of 3% as a composition of ingredients whose actions are aimed at cryoprotecting forcemeat systems from the effects of low temperatures and a decrease in the water activity index a_w.

Water freezing changes the concentration of soluble compounds in aqueous solutions of meat systems. This, in turn, affects the change in the pH and the strength of ionic interactions in the layer closest to the protein molecule. This phenomenon is due not only to dehydration and aggregation, but also to the breakdown of glycogen remaining in the meat before freezing, and to the formation of lactic acid [11-13]. In the chopped semi-finished products, during their freezing, storage, defrosting, and heat treatment, there was a slight decrease in the value of active acidity by 0.02–0.04 pH units compared to the initial pH.

The PPC has hydrophilic properties, so its use as a component of forcemeat systems prior to their freezing can reduce the negative effects of physicochemical factors and protect the meat systems from cell destruction during cryopreservation. Besides, cryopreservation can slow down oxidation processes, prevent denaturation of proteins, molecular complexes, and pigments, and also dearomatisation of raw materials.

This research proves the functional, technological, and cryostabilising properties of the PPC. The selected ratio between proteins and polysaccharides allows increasing the moisture-holding, fat-holding, and emulsifying capacities, and forms stable properties of model forcemeat systems.

So, there are two promising directions of the use of protein-polysaccharide compositions in the production of semi-finished chopped meat products:

- stabilisation of the functional and technological properties of meat raw materials;
- cryostabilisation of meat systems in the manufacture of frozen semi-finished products.

The prospects of further research consist in studying the structural and mechanical characteristics of PPC-containing forcemeat systems after defrosting.

Conclusion

1. The protein-polysaccharide complex stabilises the organoleptic, structural, and mechanical properties of model samples of semi-finished chopped products, which helps to decrease losses during heat treatment and increase the output of finished products after long-term storage at sub-zero temperatures.
2. The protein-polysaccharide complex decreases the cryoscopic temperature of forcemeat systems by 2.09–2.81°C, decreases the mass fraction of frozen moisture by 1.7%, and increases the moisture-holding capacity by 9.7–15.3%, compared to the control sample, which has a positive effect on the quality of finished products.
3. Due to the cryoprotective action of the ingredients of the protein-polysaccharide complex contained in forcemeat systems in an amount of 3%, the water activity indicator decreases by 0.031–0.067, which helps to prolong storage of meat systems.

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ПЕРСПЕКТИВИ ВИКОРИСТАННЯ КРІОСТАБІЛІЗУЮЧОЇ БІЛКОВО-ПОЛІСАХАРИДНОЇ КОМПОЗИЦІЇ У ВИРОБНИЦТВІ М'ЯСНИХ ПОСІЧЕНИХ НАПІВФАБРИКАТІВ

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Анотація. Одним з технологічних способів підвищення якості заморожених м'ясних посічених напівфабрикатів є використання у їхньому складі кріопротекторних композиційних сумішей. Механізм дії кріопротекторів пов'язаний з утворенням аморфної структури продукту, зменшенням кількості центрів кристалізації та зниженням активності води, що особливо важливо для м'ясопродуктів тривалого зберігання при мінусових температурах. З цією метою розроблено склад білково-полісахаридної композиції, що складається з білків плазми крові, натрію казеїнату, харчових волокон льону та подорожнику за співвідношення 1:1:2:2. Встановлено закономірності впливу обраної композиції на фізико-хімічні властивості модельних м'ясних фаршевих систем. Доведено можливість використання даної композиції з метою усунення недоліків низькофункціональної м'ясної сировини у складі напівфабрикатів м'ясних посічених заморожених та нівелювання негативних наслідків їх тривалого зберігання в замороженому стані. Встановлено, що композиційна кріопротекторна суміш у кількості 3% знижує кріоскопічну температуру м'ясних фаршевих систем на 2.09–2.81ºС, зменшує масову частку виморожeної вологи на 1.7% та на 0.031–0.067 знижує показник активності води. За результатами проведеного математичного моделювання та обробки експериментальних даних у системі математичного пакету MathCad 15 визначено вміст виморожeної води у складі м'ясних посічених напівфабрикатах з масовою часткою білково-полісахаридної композиції у кількості до 4% за тривалості їх зберігання до 30 діб при температурі мінус 10ºС. З метою розроблення рекомендацій щодо використання білково-полісахаридних композиційних як кріопротектору додатково досліджено хімічний склад та функціонально-технологічні властивості модельних м'ясних фаршевих систем з різним її вмістом. Доведено технологічну доцільність використання білково-полісахаридної композиції у кількості 3%, яка є ефективним кріопротектором у складі м'ясних фаршевих систем під час низькотемпературного оброблення. Одержані результати дослідження дозволяють цілеспрямовано формувати та стабілізувати задані функціонально-технологічні, структурно-механічні та органолептичні показники готового продукту.

Ключові слова: посічені напівфабрикати, білок плазми крові, казеїнат натрію, харчові волокна, кріопротектор.

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