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## THE PECULIARITIES OF CRYSTAL FORMATION DURING FREEZING OF BROCCOLI

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**Abstract.** The peculiarities of crystallization during the freezing of the inflorescences broccoli of Parthenon sort, zoned cabbage in Ukraine, have been researched and analyzed. The mass fraction of moisture and the form of its connection with dry substances in freshly picked cabbage and depending on the methods of pretreatment before freezing were determined: blanching for 3 minutes at a temperature of 85°C and exposition in the 3% salt solution for 20 minutes.

It is established that in the general part of moisture prevails osmotically-absorbed. When blanching cabbage, the amount of the colloid-bound moisture is significantly reduced and the amount of osmotic-absorbed increased. It is predefined by partial denaturation of the protein and by the decline of him to moisture keeping ability. Exposure in solution of NaCl substantially does not influence on the change of forms of the moisture binding.

Study of the process of the crystals formation was undertaken by the method of differentially thermal analysis. Analysis of the exothermic processes shows that the optimum temperature for storage of non treating broccoli is  $-14.5^{\circ}$ C, at which crystallization occurs all forms of physical and chemically bonded water, which helps to maximize the preservation of consumer properties of cabbage. The inflorescences exposure in the salt solution, compared with the most commonly used blanching operation, also contributes to reducing the energy intensity of the production, as it provides the crystallization of all forms of water at a temperature of  $-10^{\circ}$ C, which is substantially lower than the temperature regulated by the regulatory documents  $-18^{\circ}$ C.

A forecast model of the dependence of the crystallization initiation temperature on the mass fraction of moisture has been developed. The use of the presented model will help to manage the process of crystallization to improve the quality of the frozen broccoli cabbage.

**Keywords:** cabbage broccoli, formation of crystals, quick-frozen cabbage broccoli.

# ОСОБЛИВОСТІ КРИСТАЛОУТВОРЕННЯ ПІД ЧАС ЗАМОРОЖУВАННЯ КАПУСТИ БРОКОЛІ

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Анотація. Досліджено та проаналізовано особливості кристалоутворення під час заморожування суцвіть районованого в Україні сорту капусти броколі Партенон. Визначено масову частку вологи та форми її зв'язку із сухими речовинами у свіжозібраній капусті та залежно від способів попередньої обробки перед заморожуванням: бланшування

протягом 3 хв при температурі 85°C та витримування у 3 % розчині кухонної солі протягом 20 хв.

Встановлено, що в загальній частці вологи переважає осмотично-поглинена. При бланшуванні капусти, частка колоїдно-зв'язаної вологи суттєво знижується та збільшується частка осмотичної-поглиненої. Це зумовлено частковою денатурацію білка та зниженням його вологоуримувальної здатності. Витримування у розчині кухонної солі суттєво не впливає на зміну форм зв'язку вологи.

Дослідження процесу кристалоутворення проведено методом диференційно-термічного аналізу. Аналіз екзотермічних процесів засвідчує, що оптимальною для зберігання замороженої капусти броколі без обробки є температура — 14,5°С, за якої відбувається кристалізація всіх форм фізико-хімічно зв'язаної вологи, що сприяє максимальному збереженню споживних властивостей капусти. Витримка суцвіть у розчині кухонної солі, порівняно із найбільш використовуваною на практиці технологічною операцією бланшуванням, також сприяє зниженню енергоємності виробництва, оскільки забезпечує кристалізацію всіх форм води при температурі –10°С, що є суттєво нижчою від регламентованої нормативними документами температури зберігання —18°С. Розроблено прогнозну модель залежності температури

## Хімія харчових продуктів і матеріалів / Chemistry of food products and materials

ініціації кристалоутворення від масової частки вологи. Використання представленої моделі сприятиме можливості керувати процесом кристалоутворення задля підвищення якості швидкозамороженої капусти броколі.

Ключові слова: капуста броколі, кристалоутворення, швидкозаморожена капуста броколі

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## Introduction. Formulation of the problem

Quick-frozen fruits and vegetables are long-term storage products whose quality can be guaranteed to be stable for 12-24 months of storage if the appropriate low-temperature storage conditions are observed: temperatures below -18°C, relative humidity 95% [1]. The ability of frozen food to be stored for so long a time is due to the phase change of moisture and its transformation from the partially ordered state into the ordered one with the formation of a crystal lattice [2,3]. Water, the proportion of which in fruits is 82–90%, in vegetables 74–93%, is a solution of substances of different composition and concentration. The presence of substances in the cellular juice that form a true solution causes a change in the characteristic properties of water: an increase in the temperature of boiling, a decrease in water activity and cryoscopic temperature. The cryoscopic temperature is subject to Raoult's law that states that a decrease in the freezing point of the solution, as compared with the freezing point of the pure solvent, is proportional to the total molar concentration of the dissolved substances. Thus, the cryoscopic temperature depends on the concentration of the dissolved substances, the degree of their dissociation, the properties of the solvent, and is not constant [4].

However, in structured systems, which include plant material, cryoscopic temperature and, consequently, the formation of ice crystals depend not only on the properties of the solution, but also on the anatomical and morphological features of fruits and vegetables and are limited to the structural elements of tissues [5-7]. The state of cell membranes of plant material, their permeability, concentration and degree of hydration of chemical components dissolved in the cellular and intercellular fluid determine the features of ice formation in fruit and vegetable products. That is why in different types of fruits and vegetables, the cryoscopic temperature is not constant and varies in a wide range. Therefore, in spite of some common features and patterns that take place during freezing and low temperature storage of plant material (reduction of the kinetic energy of molecules, increase in the viscosity of the intracellular solution, increase in the concentration of electrolytes, delayed diffusion processes), the dynamics of physical, chemical, biochemical changes is not always consistent with the general laws of chemical kinetics and requires in-depth study.

Due to a sufficiently high moisture content in fruits and vegetables, different strength of its connection with soluble and insoluble substances, under conditions of low temperature refrigeration, recrystallization of ice crystals formed is possible, partial diffusion of uncrystallized water from the interior to the external parts of the product with condensation on its surface in the form of rime, which significantly reduces the quality of frozen foods. International Academy of Cold recommends storing frozen fruit and vegetable products at a temperature not higher than -18°C, as fluctuations and increase of the recommended temperature lead to activation of biochemical processes, recrystallization of ice crystals in fruit and vegetable tissues, formation of frost on the surface, clumping and, consequently, reduction of consumption product properties [2].

In recent years, scientific literature [8–14] have provided data on the feasibility of storing quick frozen fruit and vegetable products at temperatures below – 23°C, during which crystallization of all forms of moisture occurs, including the one that is most closely held with protoplasm biocolloids.

Based on the foregoing, it is important to study the peculiarities of crystallization during the freezing of various species and varieties of fruits and vegetables to establish the temperature at which the maximum amount of bound moisture crystallizes. This will allow determining the temperature at which crystallization of all forms of physico-chemically bound moisture occurs and recommend it as the optimum for long-term refrigerated storage of frozen fruit and vegetables. Storing frozen fruit and vegetables at certain temperatures will prevent the appearance of one of the most common defects of frozen fruit and vegetable products – 'snow cap.'

## **Analysis of recent research and publications**

The generalization of the results of research by foreign and domestic scientists, in particular A. Bartolome [13], I.G. Legaretta [8], J. Thalheimer [9], C. Kennedy [10], J. Evans [1], O.V. Vasilishina [6], S.A. Bolshakow [15], D.M. Odarchenko [7], T.D. Pylypenko [16], G.B. Chizhov [2,3] has found that the process of freezing is a set of successive stages: cooling the product to the cryoscopic temperature; actually freezing; futher freezing.

The completion of the cooling stage of the product till it achieves the cryoscopic temperature is the beginning of the crystal formation process. In the scientific literature of recent years, there are controversial data on the cryoscopic temperature of individual berries (temperature ranging  $-0.8^{\circ}$ C to  $-5^{\circ}$ C) and vegetables (ranging from  $-0.4^{\circ}$ C to  $-2.4^{\circ}$ C) [1,17-20].

These discrepancies can be explained by the fact that the plant material is a structured system with different concentration of the aqueous solution in different parts of fruit and vegetables and in structural elements of their tissues. Since the solution concentration inside the cells is higher than in the intercellular space, the cryoscopic temperature of the cell solution is 0.2–0.4°C lower than the intercellular one.

Crystals of ice, which are formed at the cryoscopic temperature, almost do not contain dissolved substances. With a further decrease in temperature, the amount of soluble matter turns out to be dissolved in a smaller amount of water. Accordingly, the concentration of the solution increases, and the cryoscopic temperature decreases, which is accompanied by subsequent crystallization of moisture [18]. Thus, free moisture, which is contained in the intercellular space and is a solvent of organic and inorganic substances, turns into ice primarily, and the size, shape and localization of ice crystals depend on the state of cell membranes, the concentration of solutes, the degree of protein hydration, and the rate of freezing.

At low freezing rates, crystallization starts from one nucleation center whose size increases with further lowering of the temperature. An increase in the freezing rate is accompanied by an increase in the number of crystallization centers and a decrease in the size of the crystals. Thus, the cryoscopic temperature is an important parameter of the freezing regime, since the rapid passage of the crystallization zone during freezing contributes to the formation of fine ice crystals, which positively affects the quality of frozen fruits and vegetables and preserves the structure of plant material after its defrosting [2,3].

Actually, freezing is the advancement of the ice front from the surface to the center of the product. The narrower the range of the freezing process, the higher the quality of the product is. The completion of the freezing process is considered to be the complete phase transformation of the moisture of fruits and vegetables from the liquid state into solid [16]. It is also known that in the plant material, a small proportion of the moisture does not crystallize even at temperatures below  $-70^{\circ}$ C [8,14,21-23].

According to the classification based on the nature and energy of the chemical bond of water in food products, it can be free (physico-mechanical) and linked (physico-chemical and chemical). Free water participates in chemical and biochemical reactions, and at 0°C it turns into ice. The bound water is adsorbed by intracellular systems and is firmly retained by colloids of proteins and carbohydrates and has altered physical properties due to its interaction with non-aqueous components, in particular, the lower crystallization temperature. Physico-chemical water is divided into colloid-bound and osmotically-absorbed, which with varying strength are kept physico-chemical bonds. A stronger bond is the colloid-bound water, which is held by the surface molecules of colloidal substances (pro-

teins and carbohydrates) at the boundary of the solidwater separation. Osmotically absorbed or structural water, which is held by the forces of osmosis and diffusion, has less retention power. At the same time, it should be noted that the division into free and bound moisture is relatively conditional, since almost all the water contained in food products is in a bound form but is retained by tissues of varying strength.

The results of the study of water mobility during freezing of plant material has confirmed that in vegetative cells, free moisture freezes at a temperature 0 to  $-4^{\circ}$ C, weakly bound at -4 to  $-14^{\circ}$ C, firmly bound at -14 to  $-30^{\circ}$ C [11].

The last stage of the freezing process is the freezing of the product to a temperature that isregulated by regulatory documents and within frozen fruit and vegetables should not exceed –18°C. Accordingly, the temperature of their storage and distribution should be the same as fluctuations and violations of the recommended temperatures lead to the activation of biochemical processes, the recrystallization of ice crystals in the fruit tissues, and the reduction of the product's properties.

The analysis of scientific literature reveals lack of efforts aimed at identifying the features of the crystallization of moisture when freezing broccoli and confirms the relevance of scientific research.

The aim of this work is to study the process of crystallization during the freezing of inflorescences of broccoli. This will enable to determine the optimum temperature for its low temperature storage.

Achievement of the goal provided for solving the following **tasks**:

- 1. To determine the moisture content in the variety Parthenon and the form of its connection with dry substances:
- 2. To determine the parameters of exothermic processes during the freezing of broccoli;
- 3. To develop a prediction model for the dependence of the crystallization initiation temperature on the mass fraction of moisture in broccoli.

## Research Materials and Methods

The research was carried out at the Laboratory of Plant Physiology and Microbiology of the Institute of Horticulture of the National Academy of Agrarian Sciences of Ukraine.

The object of the study was broccoli's inflorescences of sort Parthenon, which was included in the State Register of Plant Varieties (Experiment 1); blanched inflorescences for 3 minutes at a temperature of 80°C (experiment 2); inflorescences that was subjected by exposition in 3% salt solution for 20 minutes (experiment 3).

The study of the crystallization process was carried out by the method of differential thermal analysis, using the device for differential thermal analysis of plant objects designed in the Laboratory of plant physiology and microbiology of the Institute of Horticulture

of the National Academy of Sciences of Ukraine. Chromel-alumel thermocouples were used as temperature sensors, one of which was introduced into the central part of the curd, the other, for comparison, into a sample which did not contain water (standard). The dimensions of the standard corresponded to the size of the sample. The samples were cooled in a two-stage semiconductor micro-refrigerator type TLM-2 with a uniform velocity with a decrease in temperature of 2°C/min in the range +10 to -40°C. Since the transformation of water into ice is an exothermic process that results in hidden heat, then the thermograms recorded exothermic peaks, the analysis of which allowed determining the features of crystallization. Graphic data was converted into digital, statistical processing of which was carried out using the Excel program.

The mass fraction of moisture was determined by the thermogravimetric method till the establishment of a constant mass; forms of communication of moisture with a dry substance, including the share of colloid-bound and osmotically-absorbed by H. M. Pochinok's method of [24].

## Results of the research and their discussion

It is known that the freezing and transformation of water contained in fruits and vegetables from liquid into solid state is an exothermic process. As a result of the heat release during the phase transition of water on thermograms, high-temperature and low-temperature exotherms that appear in the form of peaks were recorded. Their analysis made it possible to determine the characteristics of crystallization. The range of observation of the exothermic process, starting with the temperature of the initiation of ice formation, is divided into sections in 1°C, each of which determines the amplitude of the exothermic process (Fig. 1).

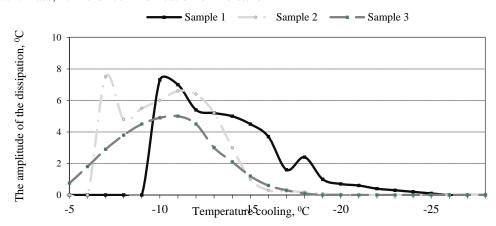


Fig. 1. Exotherms of ice formation in the broccoli curd

The presence of several peaks on the isotherms, their amplitude and position on the temperature scale are determined not only by different freezing temperatures of the overcooled aqueous solutions, but also by the total moisture content, its forms of connection with the dry matter, and, possibly, the features of the tissue structure.

The graphs of the exothermic process are translated into numerical values, the mean of which are given in Table 1.

Table 1 - Parameters of exothermic processes of ice formation in the curds of broccoli, °C

	The temperature of the exothermic processes			Temperature range of ice formation			The amplitude of the exothermic processes of ice formation		
Variants	Initiation	maximum of HTE	maximum of LTE	НТЕ	LTE	curd as a whole	front ini- tiation	maximum of HTE	maximum of LTE
Experiment 1	-11.3	-12.0	-14.5	1.0	11.0	12.0	7.7	9.8	6.3
Experiment 2	-6.3	-6.3	-10.5	0.3	13.0	13.3	7.6	7.6	6.7
Experiment 3	-4.0	-5.0	-10.0	1.5	13.5	15.0	0.7	1.6	5.0

Notes: HTE is high temperature exotherm; LTE is low temperature exotherm.

The above data suggest that the process of crystallization of moisture in the samples of experimental variants 2 and 3 begins much earlier (in the temperature range -4 to -6.3°C) than in the samples of experiment 1 (-11.3°C), and correlates with the total content moisture (r=0.72). The results of determination of the content and forms of moisture communication are confirmed by its higher total content in the samples of experimental variants 2 and 3 (Table 2). Obviously, these differences are due to a partial residue of water on the

curd surface after the technological operations (keeping in the table salt solution and blanching) and the loosening of the moisture bond with the dry matter during the heat treatment of the cabbage (experiment 2) due to the loss of the native properties of the protein.

Table 2 – Mass fraction of moisture and forms of communication of moisture from the dry substance, %

Variants	Mass frac- tion of mois- ture	Colloid- associated moisture	Osmotically absorbed moisture
Experiment 1	$87.42 \pm 4.37$	$35.75 \pm 1.79$	$51.37 \pm 2.56$
Experiment 2	$93.94 \pm 4.69$	$5.44 \pm 0.28$	$88.49 \pm 4.42$
Experiment 3	89.12± 4.45	$31.06 \pm 1.55$	$58.06 \pm 2.91$

A higher crystallization initiation temperature in samples of experimental variant 3, in comparison with those of variant 1, can also be explained by the fact that a salt solution in which the curd has been kept can retain moisture on its surface.

Establishing the temperature of initiation of crystallization is of great practical importance in the technology of low temperature preservation, since starting from this temperature, the maximum cooling rate should be provided for the formation of crystals of the minimum size, which will positively effect on the quality of frozen products.

In the analysis of thermograms, peaks were observed on the exotherms high-temperature (HTE) and low-temperature (LTE) ones. This confirms the differences in the crystallization of osmotically absorbed and colloid-bound moisture, which are due to different strengths of bonding with dry substances.

The presence of the HTE peaks at a temperature of -5°C (Experiment 3), -6.3 C (Experiment 2) and -12.0°C (Experiment 1) confirms the process of crystallization of osmotically absorbed moisture, which has a less effective connection with the dry substance, compared to that of colloid-linked moisture.

The authors' previous research agrees with the data of scientific literature on the presence of 2 to 3% of protein in broccoli, which, together with carbohydrates (3–4%), provides the moisture content of molecules of colloidal substances. The data of Table 2 indicate a sufficiently high content of colloidal-related moisture – 35.75% in absolute terms (40.9 % of the total water

$$y = 1907.73 + 1224.36x - 11.16x^2 - 0.095x^3 - 0.0019x^4 - 0.000015x^5 + 1.15 \cdot 10^{-6}x^6 - 7.33 \cdot 10^{-9}x^7$$

Fisher's statistical criterion was used to estimate the adequacy of the model obtained (2) to predict the initiation temperature. The estimated value of the criterion is

$$F_T = \frac{R^2}{1 - R^2} \cdot \frac{f_2}{f_1} \tag{3}$$

where  $R^2$  is the coefficient of determination,  $f_1$  and  $f_2$  are the number of degrees of freedom of explained and unexplained variance. If we designate the number of explanatory variables (factors) through k, and the

content). Blanching of the cabbage (Experiment 2) results in a partial protein denaturation and reducing of its moisture capacity. When blanching the cabbage, the proportion of colloidal-bound moisture decreases with an increase in the osmotic-absorbed particle. Keeping in the table salt solution is accompanied by slight changes in the forms of moisture bonding.

Significant differences in the temperature ranges of ice formation in curds of cabbage of the experimental samples have not been established.

Since the initiation temperature of crystal formation is one of the essential factors in the formation of the quality of frozen products and is determined by the moisture content, we have developed a forecast model of its dependence on moisture.

To determine the most influential factor at the initiation temperature, the Forward Selection method was used, according to which a factor best correlating with the parameter was chosen out of a number of factors.

The correlation coefficient was calculated according to the formula:

$$r_{xy} = \frac{\sum_{i=1}^{9} (x_i - \overline{x})(y_i - \overline{y})}{\sqrt{\sum_{i=1}^{9} (x_i - \overline{x})^2 \cdot \sum_{i=1}^{9} (y_i - \overline{y})^2}}$$
(1)

where,  $x_i$ ,  $y_i$  are the value of the i-th factor and the indicator obtained in the experiment;  $\overline{x}$ ,  $\overline{y}$  – average values of the factor and the indicator.

The result: 
$$r_{x_1y} = -0.871$$
,  $r_{x_2y} = -0.759$ ,  $r_{x_3y} = -0.335$ .

All correlation dependencies are inverse in character, that is, an increase in the value of the factor causes the corresponding decrease in the indicator. The most significant was the effect of the total mass fraction of moisture on the initiation temperature, which is why it was this very factor ( $x_1$ ) we chose to construct a forecast model.

The Curve Expert system was used to select the best forecast model. The best for prediction is a polynomial fit with a determination coefficient  $R^2 = 0.882$ .

The regression equation used for prediction has the form:

number of experimental values through n, for our model  $f_1 = k = 1$ ,  $f_2 = n - k - 1 = 7$ .

$$F_T = \frac{0.882}{1 - 0.882} \cdot \frac{7}{1} = 52.322$$

The critical value of the Fisher criterion  $F_k=F(0.05; 1; 7)=5.591$ .

Since  $F_T > F_k$ , the resulting polynomial model (2) is adequate and can be effectively used to predict the initiation temperature, depending on the value of the mass fraction of moisture.

(2)

## Хімія харчових продуктів і матеріалів / Chemistry of food products and materials

The presented research results are only part of the complex work aimed at stabilizing the consumer properties of frozen broccoli during long-term low-temperature storage. It is promising to study the chemical composition of broccoli during its storage for 24 months at temperatures of -10°C and -18°C, respectively, recommended by the authors and regulated by regulatory documents.

## **Conclusions**

- 1. The total moisture content of Parthenon broccoli is 87,42%, of which 51,37% is osmotically absorbed, 35,75% is colloid-bound. When blanching the cabbage, the proportion of colloidal-bound moisture decreases with an increase in the osmotic-absorbed particle. The maintenance in the solution of the kitchen salt is accompanied by slight changes in the forms of moisture bonding.
- 2. Based on the analysis of exothermic processes, it has been established that the optimum for storage of
- raw frozen broccoli is the temperature below  $-14.5^{\circ}$ C, at which the maximum crystallization of all forms of physico-chemically bound moisture takes place, while biochemical processes are impossible. This temperature contributes to the maximum preservation of the consumer properties of cabbage. Keeping in the salt solution, compared with blanching (because the latter is a technological operation most used in practice), also contributes to reducing the energy intensity of production, since it provides crystallization of all forms of water at a temperature of  $-10^{\circ}$ C, which is significantly lower than the recommended storage temperature  $-18^{\circ}$ C.
- 3. The forecast model of dependence of the crystallization initiation temperature on the mass fraction of moisture and its practical use will enable producers to control the quality of frozen fruit and vegetable products by providing the maximum cooling of the product in the crystallization zone to form small ice crystals inside the product.

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