Enzymatic processing as a tool for increasing the biological value of pumpkin puree

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Abstract. The purpose of the article is to obtain a puree with a high content of carotenoids from the pulp of the "Rossiyanka" pumpkin by destroying the polymers of the raw material using the enzyme preparations Amylorizin and Protozyme (water:preparation ratio – 10:1). Enzymeolysis of the native puree was carried out in a laboratory reactor with a volume of 2 dm3 at a temperature of 70±2 °C. It was being continuously stirred for 60 minutes, then dried to a moisture content of 5%. The effectiveness of enzymatic treatment was assessed by carotenoid content. It has been established that complete hydrolysis of pumpkin pulp starch is ensured by a 60-minute treatment with a multienzyme composition containing 25 U/g of Amylorizin, 100 U/g and 150 U/g of Protozym, which makes it possible to increase the content of β-carotene by 8.4-10.3% and improve the organoleptic characteristics of the resulting puree: the texture becomes uniform without any graininess typical for the native puree. It makes the appearance and palatability of the puree more attractive, there is no flavor of raw vegetables, pleasant and harmonious aroma and taste of steamed pumpkin prevail.

1 Introduction

Effective processing of agricultural products using biotechnology methods is one of the global trends in the development of agriculture and the food industry. Pumpkin *Cucurbita* spp., as a source of raw materials, has a number of undeniable advantages and high technological potential. In the Russian Federation, about 70 food cultivars of large-fruited pumpkin *Cucurbita maxima* Duch and hard-bark *Cucurbita pepo* L. are grown almost everywhere in a wide range of agro-climatic conditions; in regions with a warmer climate - about 20 varieties of nutmeg *Cucurbita moschata* Duch. They cultivars differ in composition and content of main biologically active substances, in particular carotenoids; the fruits have good keeping quality and the ability for long-term storage and year-round processing [1].

The main part of pumpkin dry matter is represented by polysaccharides – soluble and insoluble (dietary fiber) [2]. There are studies that have in vivo reliably established a connection between the consumption of polysaccharides from pumpkin pulp and a decrease in the level of glucose in the blood serum of rats with diabetes induced by alloxan or

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streptozotocin [3–5]. Studies [6–8] show the positive dynamics of consuming pumpkin powder and other pumpkin pulp products for reducing the concentration of glucose in the blood of patients with insulin-dependent diabetes (type 1 diabetes) and non-insulin-dependent diabetes (type 2 diabetes).

Three fractions of pectin substances were found in pumpkin fruits: water-soluble, soluble in EDTA and alkalis. It has been experimentally established that pumpkin pectin substances have the effect of retaining glucose and bile acids [9, 10], and also have a stimulating effect on the development of beneficial microflora of the gastrointestinal tract. In vivo experiments on male Sprague-Dawley rats showed a significant decrease in the levels of cholesterol and fatty acids in the blood when fed according to a diet with pumpkin pulp [8].

Pumpkin pulp contains a high amount of carotenoids (on average 7.5 mg/100 g) – the main source of vitamin A, necessary for the normal functioning of the visual apparatus and embryonic development, maintenance of epithelial tissue, as well as the immune system. [11]. The main carotenoid in the peel and pulp of pumpkin is β-carotene (from 0.05 mg/100 g to 29.4 mg/100 g); it also contains lutein (0.03–12.9 mg/100 g) [12], lycopene, cryptoxanthin and cis-carotene. There is information about the positive use of pumpkin carotenoids to effectively compensate for the deficiency of carotenoids in the blood plasma caused by smoking and alcohol consumption [13], as well as in the development of diets for gerontological purposes [14].

Expanding the range of juice drinks using vegetables, in particular pumpkin, as the main raw material is an urgent task for the food industry. Despite the wide range of pumpkin products available on the market - purees, juices, nectars, candied fruits, chips, marmalade, etc. and the implemented scientific developments [15–18], the volume of products produced is not large enough.

The traditional technology for the production of pumpkin puree consists of preparing the raw materials (washing, chopping into segments, removing seeds, crushing), processing the crushed pumpkin mass in scalders until softened, grinding to a homogeneous mass with a particle size of no more than 0.4 mm. The negative aspects of this technology include the relatively harsh conditions for heat treatment of pumpkin mass – exposure at 100–105 ° C for 10–25 minutes, which inevitably leads to degradation of carotenoids and a decrease in the biological value of the resulting pumpkin puree. In fact, the duration of heat treatment of pumpkin pulp is directly dependent on the starch content in it, since, being a polymer, it fully participates in the formation of the intercellular framework. In addition, the presence of a fairly high amount of starch (up to 4.5%) [19] gives fresh pumpkin an unpleasant taste of “raw starch”, as well as a specific chewing sensation. Biotechnological methods for preprocessing raw materials can significantly increase the consumer properties of pumpkin puree by removing the unpleasant taste of starch.

Thus, the main goal of the study was the selection of enzyme preparations and conditions for their use to increase the biological value of semi-finished pumpkin products.

2 Materials and Methods

Eating-ripe fresh pumpkin fruits of the “Rossiyanka” cultivar (without damage or signs of deterioration), harvested in 2023, were used as research objects. Puree samples were obtained using a Viteg HG-15D homogenizer at a rotor speed of 2400 rpm.

To determine the gelatinization temperature of pumpkin starch and the subsequent development of enzymatic puree technology, an EAK-2M express consistency analyzer (OOO “Izobretatel'” (“Inventor”), Russia) was used.

To ferment biopolymers of native pumpkin puree (primarily starch and proteins), enzyme preparations Amylorizin and Protozyme (OOO “Biopreparat”, Russia) were used.

To carry out fermentolysis, native pumpkin puree was heated to reach temperature gelatinization of the starch in the raw material. Then, aqueous solutions of the enzyme preparations Amylorizin and Protozyme (water:preparation ratio – 10:1) were added to the puree with stirring continuously. Fermentolysis of native pumpkin puree was being continued for 60 minutes with constant stirring and thermostatting.

The determination of β-carotene content was carried out according to GOST ISO 6558-2–2019 Fruits, vegetables and their processed products. Determination of carotene content by spectrophotometric method (method A).

3 Results and Discussion

One of the factors influencing the efficiency of enzymatic hydrolysis of starch is its gelatinization temperature, and it is known that for different starch-containing crops the temperature of starch gelatinization is different. When analyzing the available scientific and technical literature, no information was found on the gelatinization temperature of pumpkin starch, so in the experiment it is necessary to establish not only the dosage of the enzyme for starch hydrolysis, but also to select the minimum temperature and duration of the process. Figure 1 shows the change in the consistency of pumpkin puree depending on the heating temperature of the mass.

Native pumpkin puree is a homogeneous mass with the color characteristic of the raw material. The smell is vegetal, characteristic; the taste is of raw vegetables, slightly sweet, not very harmonious. When obtaining native pumpkin puree, the phenomenon of syneresis was observed – a slight separation of free moisture from the solid part of the pumpkin homogenate, and the pumpkin puree itself, based on the totality of its rheological properties, can be classified as a pseudoplastic liquid. The most significant factors influencing the rheological behavior of pumpkin puree include: particle size, total dry matter content and the total amount of undissolved substances, including biopolymers. As a result of heating pumpkin puree in the temperature range between 65-75 °C, the puree acquired a “silky” surface, the phenomenon of syneresis was not observed.

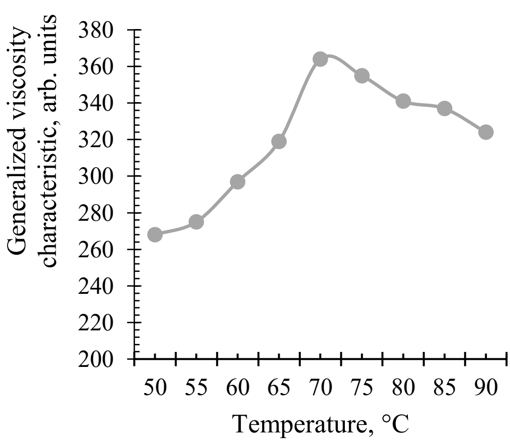

Fig 1. Curves showing the gelatinization process of pumpkin puree starch

Fermentolysis of native pumpkin puree was carried out in a laboratory reactor with a volume of 2 dm3 with continuous stirring and a temperature of 70±2 °C for 60 minutes. During the fermentolysis process, the content of carotenoids was controlled, since it is known that carotenoids are in a bound state with polymers of raw materials, primarily with proteins. Thus, the targeted exposure with enzyme preparations on pumpkin pulp polymers should ensure the release of carotenoids from the matrix, which will increase their bioavailability in food systems. For fermentolysis the following dosages of enzyme preparations were used together: Amylorizin – 25 U/g of raw material (an experimentally found dosage that ensures complete hydrolysis of pumpkin starch within 60 minutes); Protozyme – 5, 10 and 15 U/g of raw material. The efficiency of fermentolysis was assessed by the amount of carotenoids. This indicator was chosen as a control, since it is known that in plant raw materials carotenoids are mainly in a bound state with proteins and other polymer molecules, thus the destruction of the protein framework should lead to the accumulation of carotenoids in pumpkin puree during fermentolysis. The results obtained during the experiment are generally described by the regression equation (r2 = 0.934):

$$Car=4.069+0.0068x+0.0011y,$$

where Car is the mass concentration of carotenoids, mg/100 g; x – duration of fermentolysis, min; y – dosage of the enzyme preparation “Protozyme”, U/g of raw material.

Thus, by purposefully influencing the biopolymers of pumpkin pulp, it is possible to achieve homogeneity of pumpkin puree and increase its plastic properties, which is important by the substantiation and selection of main and auxiliary equipment while designing technologies and configuring production lines.

It has been experimentally established that as a result of enzymatic multi-enzyme composition containing Amylorizin at a dosage 25 U/g of raw material, the starchy part of the puree is completely hydrolyzed, which is accompanied by an increase in sugar content (5.37±0.07% per a.d.s.) and an objective increase in the sweetness of the puree with a simultaneous improvement in the texture and deliciousness of the puree.

Fermentolized pumpkin puree was cooled to room temperature and its organoleptic characteristics were compared with native pumpkin pulp puree. During testing, it was found out that as a result of fermentolysis, the organoleptic characteristics of native pumpkin pulp puree were essentially improved. Thus, a significant increase in the sensation of sweetness in taste was noted, the puree texture became homogeneous, while the graininess of native purees was eliminated. All this makes the semi-finished product more attractive in appearance and more appetizing. The tones of raw vegetables disappear in the flavor, the fermented puree acquires a pleasant and harmonious aroma and taste of steamed pumpkin. The use of a multi-enzyme composition, including enzyme preparations Amylorizin and Protozyme, makes it possible to increase the carotenoid content in the puree by 8.4-10.3%. It is explained by the destruction of carotenoid complexes and the protein matrix of pumpkin pulp, which ultimately leads to better digestibility of carotenoids in the human body. The studies revealed a tendency towards a decrease in the content of carotenoids in pumpkin puree during fermentolysis for more than 60 minutes, which is likely due to the onset of the terminal stage of oxidative destruction of carotenoids.

4 Conclusion

In the course of the research, the following conclusions were obtained:

1) a multi-enzyme composition containing the enzyme preparations Amylorizin (25 U/g of raw material) and Protozyme (10 and 15 U/g of raw material) was selected, which allows to increase the β-carotene content by 8.4-10.3% due to the destruction of stable carotenoid complexes and polymers raw materials;

2) a mathematical relationship was established between the content of carotenoids in fermented pumpkin puree and the conditions of fermentolysis (its duration and dosage of the proteolytic enzyme preparation Protozyme);

3) it has been shown that the use of fermentolysis can improve the organoleptic characteristics of pumpkin puree – flavor and texture, as well as palatability.

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