THE INFLUENCE OF RICE PROTEIN CONCENTRATE ON THE TECHNOLOGICAL PROCESS OF WHEAT BREAD PRODUCTION

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Abstract

Bakery products from wheat flour are low in protein and it is inferior and also absorbed at a low level. To solve this problem sources of complete proteins may be introduced into the formulation of bakery products. But animal proteins can be allergens. An alternative can be concentrates, hydrolysates and isolates of proteins of vegetable origin, particularly, obtained from rice. Microbiological, biochemical and conformational changes in dough and bread from wheat flour were influenced by adding rice protein concentrate. Gas-forming capacity of the dough with rice protein concentrate decreased by 8.3–20.8 % compared to the control sample where there was increase of the dosage. Gas formation occurred less intensively in the dough with rice protein concentrate, because fermentation was delayed due to a decrease in the availability of nutrients. The first peak on the gas formation graph in case of the dough with the addition of 4–8 % rice protein concentrate was seen after 65 minutes, in the control sample it was after 60 minutes. When adding 16 % of the additive, the first peak of gas formation was not clearly defined. The second peak of gas formation for control sample was observed after 150 min and for samples with rice protein concentrate a bit later and it was not clearly defined. Infrared spectra showed that relative reflection coefficient of samples with rice protein concentrate was lower both for dough after kneading and after fermentation. The addition of 4 and 8 % rice protein concentrate did not affect the amount of formed sugars, and the addition of 16 % reduced this indicator by 1.6 %. The amount of fermented sugars decreased by 3.0–7.8 %. So, changes during the technological process of bread-making can be followed due to the obtained results for obtaining bread with high protein content.

Keywords: bread, flour, rice protein concentrate, gas-forming capacity, spectroscopy, dough, reflectance.

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1. Introduction

Bakery products from wheat flour are low in protein [1]. It is inferior and also absorbed at a low level [2]. To solve this problem sources of complete proteins of animal and vegeTable origin may be introduced into the formulation of bakery products. But animal proteins can be allergens, so it will affect the health of consumers. An alternative can be concentrates, hydrolysates and isolates of proteins of vegetable origin, particularly, obtained from rice.

Rice protein is extracted mainly from rice bran or milled rice by alkaline or enzymatic methods, it is considered to be hypoallergenic [3].

Research on obtaining protein concentrates from rice bran in non-stabilized and heat-stabilized forms by alkaline extraction and then isoelectric precipitation was conducted. Protein extraction capacity of stabilized rice bran at particle size $75-150 \,\mu\text{m}$ was lower. Maximum nitrogen solubility of both unstabilized and stabilized rice bran protein concentrate was at pH 8.0–71.5 % for unstabilized protein and 50.9 % for stabilized protein. There were also differences in amino acid content. Heat stabilization caused protein denaturation due to which the quality of proteins was affected [4]. Although the protein content in rice is small (about 3 %) it has a high nutritional value, is considered to be particularly useful for human consumption. The nutraceutical and antioxidant properties of rice are also associated with reduced risk of hypercholesterolemia [5].

Rice and its processing products are widely used in the manufacture of gluten-free products, since the protein substances of this raw material do not contain gluten. It was established the possibility of using pure protein and protein-calcium concentrates from different varieties of rice – white and brown, in the technology of gluten-free butter cake. The method of isolation of rice protein concentrates included treatment of flour with amylase and xylanase followed by extraction of proteins with dissolved hydrochloric acid in the presence of calcium citrate. Foaming ability and foam stability of the obtained rice concentrates were high, which allowed to use them in the amount of 50 % to the mass of egg melange in the technology of butter cake to increase its biological value and structural and mechanical properties [6].

Rice protein was also added to the recipe of gluten-free bread. Its effect on the properties of the protein matrix was investigated by evaluating dough rheology, bread physicochemical properties, crumb texture, and microstructure. The results of the rheological properties showed that appropriate concentrations of rice protein increased viscoelastic properties and stored energy of the dough. Excessive concentrations weakened the structural strength, there was observed decrease in the maximum peak of dough development when stress was applied. So, the addition of rice protein improved the properties of the matrix with increased elasticity and hardness. The protein reduced moisture loss and provided a nice colored crust. According to the properties of re-fermentation, this protein significantly reduced specific volume of bread when the concentration exceeded 3.0 g/100 g. The addition of protein formed a compacted structure as it was shown by the microstructure [7].

In wheat bread technologies, rice processing products are not so widely used, as it is believed that they negatively affect the technological process of manufacturing bakery products [8].

The aim of the work was to find the effect of rice protein concentrate on microbiological, biochemical and conformational changes in dough and bread from wheat flour.

2. Materials and Methods

2.1. Object of research

Research was conducted with dough as semi-finished products and baked bread. For dough preparation wheat flour of premium grade, pressed baker's yeast and salt were used. Rice protein concentrate was added in the amount of 4, 8 and 16 % to the mass of flour in order to provide 20, 30 and 40 % of the daily protein requirement. A sample without rice protein concentrate was the control sample.

2. 2. Gas-forming capacity of the dough

Gas-forming capacity was determined as the amount of carbon dioxide in cm³ emitted during the time period of fermentation and keeping of dough from 100 g of flour at a temperature of 30 °C. Volumetric method was used to determine this indicator, namely the volume of CO₂ emitted at constant temperature and pressure [9, 10].

2. 3. Kinetics of accumulation of sugars in the dough

Kinetics of accumulation of sugars was characterized by the amount of formed and fermented sugars. The amount of formed sugars was determined during the fermentation of the dough by the difference between their content in the yeast-free dough after kneading and after 180 minutes of fermentation. The amount of fermented sugars was determined by the formula:

$$U = \frac{C_{\min} \cdot \sum_{j=1}^{8} A_{ej}}{\sum_{i=1}^{8} A_j} S_f = (A+B) - C, \tag{1}$$

where A – amount of sugars at the beginning of fermentation of yeast dough, % to dry matter of dough;

B – amount of sugars formed in yeast-free dough, % to dry matter of dough;

C – amount of sugars contained in yeast dough after 180 minutes of fermentation, % to dry matter of dough.

The accelerated iodometric method was used to determine the kinetics of sugar accumulation in the dough [11].

2. 4. Near-infrared reflection spectroscopy

Reflectance spectroscopy in the near-infrared region is based on the principle of evaluation of the diffuse reflection that occurs during the penetration of optical radiation through the surface layer of the sample. After that the excitation of the vibrational modes of the analyzed molecules, and than the scattering of optical radiation in all directions took place. For measurement of the reflectance spectra of ground samples with a smooth surface Infrapid spectrometer (Labor-Mim, Hungary) was used. The wavelength range was from 1330 to 2370 nm. The reflection spectra from the standard and from the studied sample were recorded. The results were presented as reflectance R, depending on the wavelength [12]. The intensity of reflection was measured in raw materials, dough after kneading and after 3.5 hours of fermentation, and in bread. The reflection intensity was expressed by recalculating the relative reflection coefficient to the spectral index [13].

2. 5. Statistical analysis

The obtained results were presented as average data after repeating the experiments 3 times±standard deviation. Graphical presentation of experimental data was made in Microsoft Excel 2010 (USA).

3. Results and Discussion

During the technological process of manufacturing bakery products, microbiological and biochemical processes take place [14]. Microbiological processes in dough are important in bread technology. In the process of dough fermentation biotransformation of polymers of raw materials takes place. Their intensity depends on the chemical composition of the recipe components. Microbiological processes in the dough with rice protein concentrate were evaluated by the amount of released carbon dioxide during the fermentation and keeping of the dough and the dynamics of its release (**Fig. 1**).





Gas-forming capacity of the dough with rice protein concentrate decreased by 8.3-20.8 % compared to the control sample with an increase of the dosage. It indicated deterioration in the enzymatic capacity of yeast. The gas-forming capacity was determined primarily by the carbo-

hydrate-amylase complex of the recipe raw materials, as well as the sugar-forming ability of flour and the attackability of starch by amylase enzymes [15, 16]. The decrease of the studied indicator when applying rice protein concentrate is explained by the decrease in the availability of nutrients in its presence. This delayed the fermentation process and the amount of released carbon dioxide decreased. Protein concentrate complexes were formed with wheat flour starch, which prevented the release of nutrients for yeast. Such a tendency can be reflected in the indicators of bread quality, in particular, cause a decrease in its specific volume [17]. A decrease in yeast fermentation activity in dough in the presence of rice protein concentrate will influence the dynamics of carbon dioxide release during the process of dough fermentation and keeping (**Fig. 2**).



Fig. 2. Dynamics of gas formation in the dough samples during fermentation and keeping: 1 – control sample; 2 – with 4 % rice protein concentrate; 3 – with 8 % rice protein concentrate; 4 – with 16 % rice protein concentrate

It was found that gas formation occurred less intensively in the dough with rice protein concentrate, because fermentation was delayed due to a decrease in the availability of nutrients. The first peak of gas formation on the graph of the dynamics of the release of carbon dioxide in the dough with the addition of 4-8 % rice protein concentrate was observed after 65 minutes, and in the control sample – after 60 minutes. This is explained by the fact that the additive decreased the amylolytic enzymes activity of wheat flour. When adding 16 % of the additive, the first peak of gas formation was not clearly defined. With an increase in the dosage of rice protein concentrate the amount of released carbon dioxide in the dough decreased. The reason for this is a decrease in the availability of starch for amylolysis. The second peak of gas formation for control sample was observed after 150 min and for samples with rice protein concentrate a bit later and it was not clearly defined.

The main difference between the protein composition of wheat flour and rice protein concentrate, which determines the completely different behavior of these raw materials when kneading the dough, is that the rice protein concentrate does not contain gluten proteins, and therefore cannot participate in the formation of the gluten frame, which determines structural and mechanical properties of the dough, affects the conformation of the transformation in the process of its fermentation.

To identify basic structural units in dough and bread with rice protein concentrate, the reflection spectrum in the near infrared region was used [18]. Dough and bread samples were prepared with the minimum and maximum researched addition of replacement of rice protein concentrate -4% (Fig. 3) and 16 % (Fig. 4).

All spectra had a similar character and the same extremes. But there are differences in the intensity of reflection that identifies them.

The obtained results showed that spectra of the samples of dough after kneading were situated higher than those after fermentation. In addition, the spectrum of the sample with rice protein concentrate was lower than control sample. This was due to significant differences in the composition of wheat flour, which mainly determines the behavior of the structural elements of the dough and additive. In particular, rice protein concentrate had a high protein content, which was very different in composition from flour protein. This can be seen immediately after mixing at a wavelength of 2100 nm. The relative reflection coefficient of the control sample of the dough after kneading was 0.34, while the sample with protein concentrate was 0.32. This indicated that even such a small percentage caused conformational transformations.









Spectra of samples of the dough after 3.5 hours fermentation were situated below on the graph. But the trend was opposite, because the spectrum of the control sample had lower values of relative reflectance coefficient over the entire wavelength range compared to the protein-supplemented sample. This is mainly due to the change in the structure of gluten in the presence of rice protein concentrate, because proteins of vegetable origin weaken its structure and prevent the formation of a branched framework [19]. This was confirmed by the values of relative reflectance coefficient at a wavelength of 2100 nm - 0.28 and 0.25 for the sample with added protein and for control sample.

The infrared spectra of the baked bread samples conditionally matched. A characteristic extremum was observed at the wavelength 2294 nm that characterizes the amino acid composition. The graph shows that there were significant changes in the protein structure after baking. After all, the spectra overlapped. This is explained by the effect of high temperatures on the protein structure, in particular, the denaturation process, as well as the interaction of amino acids of rice protein concentrate with flour carbohydrates [20].

The spectra of bread are located close to the fermented control dough sample. This is explained by the fact that the determining role in the formation of the final structure of the bread frame is played by the components of the flour, content of which is major in the recipe.

In general, the spectra of samples with 16 % rice protein concentrate had a similar character as when replacing 4 %, but the sample of dough with 16 % after 3.5 h of fermentation was close to the sample after mixing.

Such conformations should also affect biochemical processes during dough fermentation.

The biochemical processes in the dough mean the breakdown of proteins and starch of flour under the action of enzymes of yeast and other microorganisms as well as the flour's own enzymes [21]. In such case sugars and nitrogenous substances are accumulated in the dough. The content of sugars depends on the process of their accumulation and fermentation intensity ratios during dough fermentation [22].

It was established that the addition of 4 and 8 % rice protein concentrate did not affect the amount of formed sugars, and the addition of 16 % reduced this indicator by 1.6 % (**Table 1**).

The amount of fermented sugars decreased by 3.0-7.8 %. This is due to the fact that proteins of rice protein concentrate form complexes with wheat flour starch. This caused deterioration of access of enzymes to starch grains.

Indicators	Control sample	Rice protein concentrate, %		
		4	8	16
	Yeast-free dough			
After kneading	2.10 ± 0.10	$2.10{\pm}0.10$	$2.10{\pm}0.10$	2.11±0.10
After 3 hours of fermentation	3.39±0.17	$3.39{\pm}0.17$	$3.39{\pm}0.17$	3.40±0.17
Accumulated sugars	1.29 ± 0.03	$1.29{\pm}0.03$	$1.29{\pm}0.03$	1.27 ± 0.03
	Yeast dough			
After kneading	2.15±0.12	2.15±0.12	2.16±0.12	2.18±0.12
After 3 hours of fermentation	$1.78{\pm}0.08$	1.83 ± 0.08	$1.88{\pm}0.08$	$1.92{\pm}0.08$
Fermented sugars	$1.66{\pm}0.06$	1.61 ± 0.06	1.57 ± 0.06	$1.53{\pm}0.06$

Table 1

Accumulation and fermentation of sugars during dough fermentation (in terms of maltose), % on dry matter

Note: results given as: M±SD (mean±standard deviation) of triplicate trials

Changes during the technological process of bread-making can be followed due to the obtained results for obtaining bread with high protein content. The obtained results can be used for the development of recipes of products for special purpose with healthy properties. The direction of providing future studies is to carry out research on structural and mechanical characteristics in dough systems with rice protein concentrate and bread quality indicators.

Restrictions when conducting research concerned martial law conditions in Ukraine due to the impossibility of conducting research during an air raid, which delayed the timing of their conduct and processing of results.

4. Conclusions

Microbiological, biochemical and conformational changes in dough and bread from wheat flour were influenced by adding rice protein concentrate.

Gas-forming capacity of the dough with rice protein concentrate decreased by 8,3-20.8 % compared to the control sample with an increase of the dosage. Gas formation occurred less intensively in the dough with rice protein concentrate, because fermentation was delayed due to a decrease in the availability of nutrients. The first peak of gas formation in the dough with the addition of 4-8 % rice protein concentrate was observed after 65 minutes, while in the control sample – after 60 minutes. When adding 16 % of the additive, the first peak of gas formation was not clearly defined. The second peak of gas formation for control sample was observed after 150 min and for samples with rice protein concentrate a bit later and it was not clearly defined.

Infrared spectra showed that relative reflection coefficient of samples with rice protein concentrate was lower both for dough after kneading and after fermentation.

The addition of 4 and 8 % rice protein concentrate did not affect the amount of formed sugars, and the addition of 16 % reduced this indicator by 1.6 %. The amount of fermented sugars decreased by 3.0-7.8 %.

The obtained results allow to follow changes during the bread-making technological process for obtaining bread with high protein content.

Conflict of Interest

The authors declare that there is no conflict of interest in relation to this paper, as well as the published research results, including the financial aspects of conducting the research, obtaining and using its results, as well as any non-financial personal relationships.

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The study was performed without financial support.

Data availability

Manuscript has no associated data.

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