

# Protein substances of oat bran and their influence on conformational transformations in dough and bread from wheat flour

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## Abstract

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**Introduction.** The aim of the work was to determine the degree of completeness of protein substances of oat bran and its influence on conformational transformations in the structure of dough semi-finished products and bread made from wheat flour.

**Materials and methods.** Oat bran, its amino acid composition and influence on the content of protein substances in bread were investigated. Conformational transformations of structural elements in dough and bread were investigated by infrared spectroscopy in the near-infrared region.

**Results and discussion.** The content of essential amino acids in oat bran is significantly higher than in premium wheat flour. The limiting amino acid in wheat flour is lysine, the amino acid score of which is 0.44. In oat bran the limiting amino acid is methionine, the amino acid score of which is 1.14, and the amino acid score of lysine is much higher than in wheat flour – 1.62. The limiting amino acid in bread is lysine, the amino acid score of which is 0.46. With an increase in the percentage of replacement of wheat flour with oat bran, the amino acid score of lysine increased by 19.5–52.2%. This indicates that the protein of this raw material helps to increase the level of essential amino acids in bread, which enriches its protein profile. The obtained spectra of the dough samples after kneading showed that the introduction of oat bran does not cause conformational changes in the dough system, since not enough time passed for the interaction of the biopolymers of the raw materials. The infrared spectra of the dough at a wavelength of 2100 nm showed that the dietary fibers of oat bran delay the development of the gluten network, the structure of the protein matrix of the dough with bran will be less stable and more weakened than that of the control sample.

**Conclusion.** The introduction of oat bran with a higher content of protein, dietary fiber and a complete amino acid profile into the recipe of wheat bread helps to improve the biological value of bread with this raw material. However, the deterioration of the development of the gluten framework of the dough, and therefore of the specific volume, porosity and dimensional stability of bread, requires the use of technological methods to minimize the negative impact of bran on the quality of finished bakery products.

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## Introduction

The problem of ensuring food security in the world is acute. Also, one of the vectors is orientation towards the sustainable development of production. In this concept, the disposal of food industry waste occupies an important place.

Waste-free production and the use of processed products of various crops are becoming widespread. From this point of view, bran is a valuable raw material (Ivanov et. al., 2021). Their use is recommended, in particular, in the technology of bakery production as a source of dietary fiber, which is especially necessary for people suffering from diseases of the gastrointestinal tract, such as irritable bowel syndrome (IBS).

Bran of various plants are used. Wheat bran is known to have a negative effect on the dough structure, resulting in a decrease in the specific volume of bread, which was confirmed using a 2D image analysis method, where wheat bran was observed to change the pore size distribution in the bread crumb. The properties of the dough, such as stickiness and extensibility, decreased with the increase in the amount of bran. Wheat bran increased dough aeration during kneading (Packkia-Doss et. al., 2019).

The results of using corn bran showed that their mixture with wheat flour has more water absorption and water retention capacity by 4–7% compared to wheat flour. However, the gas-holding capacity of the dough from such a mixture decreased. This led to a decrease in the volume of bread. In addition, moisture content and hardness increased with the addition of corn bran. The water activity of bread increased slightly with the addition of corn bran after 4 days of storage. The consumer properties of bread improved (Hussain et. al., 2021).

The use of barley bran in the amount of 25% instead of wheat flour increased the water absorption capacity of the dough to 71.5%, and also reduced the retrogradation of starch by 26.44%. The composite flour contained up to three times more  $\beta$ -glucan and significantly more total phenolics, including flavonoids. The content of slowly digestible and resistant starch increased. Inclusion of barley bran reduced starch retrogradation due to more amount of soluble starch and soluble amylose (Gujral et. al., 2018).

Oat bran is a more valuable source of protein and dietary fiber than hulled oat products (Sterna et. al., 2016; Zhong et. al., 2019). When adding oat bran to bread, the specific volume decreased with an increase in the amount of bran. In addition, a change in the fatty acid profile was observed, namely an increase in the content of unsaturated fatty acids, as well as an increase in the amount of dietary fiber.

Oat bran has good biological properties. The results showed that bran contains a large amount of  $\beta$ -glucan, which can be easily extracted. In addition, a decrease in the level of glucose in the blood of rats and a better digestibility of products with oat bran were found (Khan et. al., 2020).

It was established that the addition of bran to wheat dough reduces the density of the dough at the end of kneading mainly due to the increased water absorption capacity. Size of the bran particles has a small effect on this indicator. The addition of oat bran reduced the gas-holding capacity of the dough and the volume of the bread, indicating that oat bran exerted its effect mainly during proofing (Campbell et. al., 2008).

The effect of bran addition on gluten secondary structure and water behavior in wheat dough can be studied using Fourier transform infrared spectroscopy. Studies in the frequency range of 700-4000  $\text{cm}^{-1}$  revealed that the addition of wheat bran to wheat dough caused a redistribution of bound water in the gluten-bran system. This redistribution of water affected the secondary structure of gluten in the dough, as evidenced by changes in the spectrum of the second derivative in the amide I region. In the hydrated state, the  $\beta$ -turn (in the form of a  $\beta$ -helix) was the main secondary structure (60%) in the wheat dough. The addition of bran

caused the transformation of  $\beta$ -helices into  $\beta$ -sheets and random structures. However, the degree of this transformation in the presence of bran was proportional to the moisture content in the dough. This study showed that when wheat bran is added to gluten dough, the redistribution of water contributes to the partial dehydration of gluten and the transformation of  $\beta$ -helices into intermolecular  $\beta$ -sheet structures. This trans-conformation can be the reason for the low quality of bread with bran (Bock et. al., 2013). However, no data were found on the study of the influence of oat bran on the conformational transformations in the gluten network of the dough.

Therefore, the aim of the work was to determine the degree of completeness of the protein substances of oat bran and their influence on the conformational transformations in the structure of dough semi-finished products and bread made from wheat flour.

## **Materials and methods**

### **Materials**

The materials for research were premium wheat flour and oat bran.

Dough samples were prepared from wheat flour, pressed baker's yeast in the amount of 3% by weight of flour and salt in the amount of 1.5% by weight of flour. Samples were also prepared with replacement of wheat flour in the recipe with oat bran in the amount of 5%, 7%, 10% and 15%. The control was a sample without additional raw materials.

### **Essential amino acid composition**

Method of ion exchange chromatography was used for determination of amino acid composition in oat bran, wheat flour and bread (Litvynchuk et. al., 2022). The first stage is hydrolysis of proteins and the second is determination of their quantitative estimation using automatic analyzer of amino acids (T-339, Mikrotechna, Czech Republic). polystyrene sulfonate ion ex-change resins were used in Li- citrate buffer one column mode. Li-citrate buffers have pH 2.75 $\pm$ 0.01; pH 2.95 $\pm$ 0.01; pH 3.2 $\pm$ 0.02; pH 3.8 $\pm$ 0.02; pH 5.0 $\pm$ 0.2. The elutions of amino acids conduct in turn by these buffers. Amino acids were detected using photometer (Unicam SP 800, Great Britain) at a wave-length of 560 nm by a rectification with a ninhydrin solution. The results of detection were registered by a variplotter in form the peaks of absorption of light of ninhydrin-positive substances in an eluent. These peaks are in direct ratio concentrations of this substance in solution. Temperature of thermostatic was T1=38.5 °C; T2= 65 °C. Correlation of solution of ninhydrin reagent and eluents was 1 to 2. The prototype was diluted in Li-citrate buffer by pH 2.2 $\pm$ 0.02 and inflicted on an ion exchange column. The mass of every amino acid expressed as g per 100 g protein. Amino acid SCORE is expected according to the certificate scale of THEO/WHO (Choi et. al., 2012).

### **Near-infrared reflection spectroscopy**

Infraprid spectrometer (Labor-Mim, Hungary) was used to research the reflection spectra from shredded samples and a smooth surface in near infrared range from 1330 to 2370 nm. Firstly, the spectrometer recorded the reflectance spectrum from reference I0, secondly a reflection spectrum from the researched sample. The spectra are represented as the reflectivity of R in relative units (the ratio of the intensities I/I0 = R), depending on the

wavelength in nm (Shevchenko et. al, 2022). The intensity of reflection was measured in wheat flour, oat bran, lecithin, in dough after kneading and after 3.5 hours of fermentation and in bread. The reflection intensity was expressed through the recalculation of relative reflection coefficient to spectral index (Yip et. al, 2012).

### Statistical analysis

The data represents the mean of a minimum three replicates  $\pm$  standard deviation (S.D.). Graphical presentation of experimental data was performed using standard statistical processing programs – Microsoft Excel 2010.

## Results and discussion

### Essential amino acid composition of wheat flour and oat bran

The main nutrients contained in the raw materials play a decisive role in forming the properties of the dough system, in particular the gluten frame, and the quality of bread. The chemical composition of wheat flour and oat bran differs significantly. Oat bran has an increased content of protein – 17.1% and dietary fiber – 15.4% compared to premium wheat flour – 11.3% and 3.5%, respectively. The protein content and composition of raw materials affects not only the formation of the structural and mechanical properties of the dough, but also determines the protein composition of the finished bread (Amjid et. al, 2013).

It was established that the content of essential amino acids (EAA) in oat bran is significantly higher than in wheat flour (Table 1). This will enrich the protein profile of bread with this raw material. The limiting amino acid in wheat flour is lysine, the amino acid score of which is 0.44. In oat bran, the limiting amino acid is methionine, the amino acid score of which is 1.14, and the amino acid score of lysine is much higher than in flour – 1.62.

The fact that the amino acid score of the limiting amino acid in oat bran is higher than 1 indicates that the protein of this raw material is complete, which makes it possible to predict an increase in the digestibility of protein in bread when bran is added to the recipe.

**Table 1**  
**Amino acid composition of oat bran and premium wheat flour (g/100 g of the raw material)**

EAA	Premium wheat flour	Oat bran
Valin	0.42 $\pm$ 0.01	0.96 $\pm$ 0.01
Isoleucine	0.36 $\pm$ 0.01	0.67 $\pm$ 0.01
Leucine	0.71 $\pm$ 0.02	1.37 $\pm$ 0.02
Lysine	0.23 $\pm$ 0.01	0.76 $\pm$ 0.01
Methionine	0.40 $\pm$ 0.01	0.34 $\pm$ 0.01
Threonine	0.28 $\pm$ 0.01	0.50 $\pm$ 0.01
Tryptophan	0.13 $\pm$ 0.01	0.33 $\pm$ 0.01
Phenylalanine	0.52 $\pm$ 0.01	0.91 $\pm$ 0.01

The amino acid composition of raw materials determines the biological value of finished bakery products. The amino acid profile of bread baked with the replacement of part of wheat flour with rice flour was determined. The results are shown in Table 2. The amino acid score of each amino acid in the bread samples was also calculated (Table 3).

**Table 2**  
**Amino acid composition of bread baked with replacement of part of wheat flour with oat bran**  
**(g/100 g of bread)**

EAA	Control sample	Oat bran to replace wheat flour, %			
		5	7	10	15
Leucine	0.71	0.77	0.79	0.83	0.88
Isoleucine	0.39	0.42	0.43	0.44	0.47
Methionine	0.31	0.32	0.32	0.32	0.33
Lysine	0.23	0.28	0.30	0.32	0.36
Phenylalanine	0.68	0.70	0.71	0.73	0.75
Threonine	0.28	0.30	0.31	0.32	0.34
Valin	0.43	0.41	0.41	0.40	0.39
Tryptophan	0.09	0.11	0.12	0.13	0.15

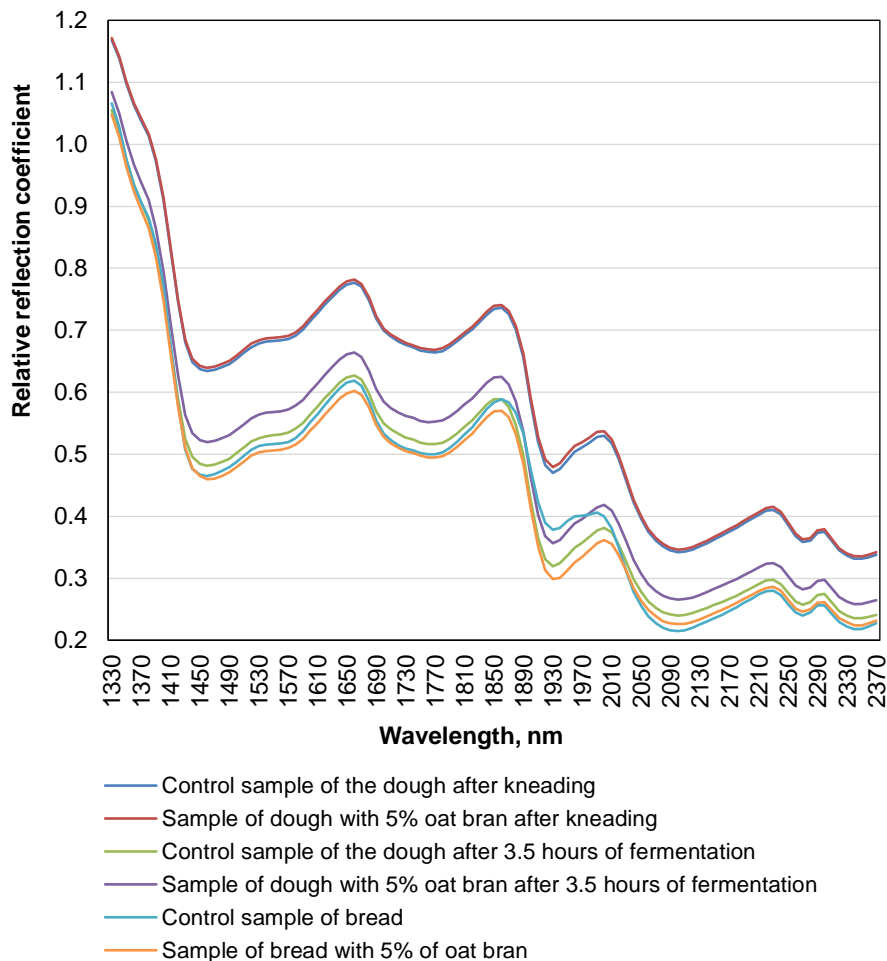
**Table 3**  
**Amino acid score of amino acids in bread baked with replacement of part of wheat flour with**  
**oat bran**

EAA	Control sample	Oat bran to replace wheat flour, %			
		5	7	10	15
Leucine	1.11	1.19	1.23	1.27	1.35
Isoleucine	1.06	1.13	1.15	1.19	1.25
Methionine	0.97	0.98	0.98	0.99	1.00
Lysine	0.46	0.55	0.58	0.63	0.70
Phenylalanine	1.23	1.27	1.28	1.30	1.34
Threonine	0.77	0.82	0.84	0.86	0.91
Valin	0.93	0.90	0.89	0.87	0.84
Tryptophan	0.98	1.19	1.27	1.38	1.57

The limiting amino acid in bread is lysine, the amino acid score of which is 0.46. With an increase in the percentage of replacement wheat flour with oat bran, the amino acid score of lysine increased by 19.5–52.2%. This indicates that the protein of this raw material helps to increase the level of essential amino acids in bread, which enriches its protein profile.

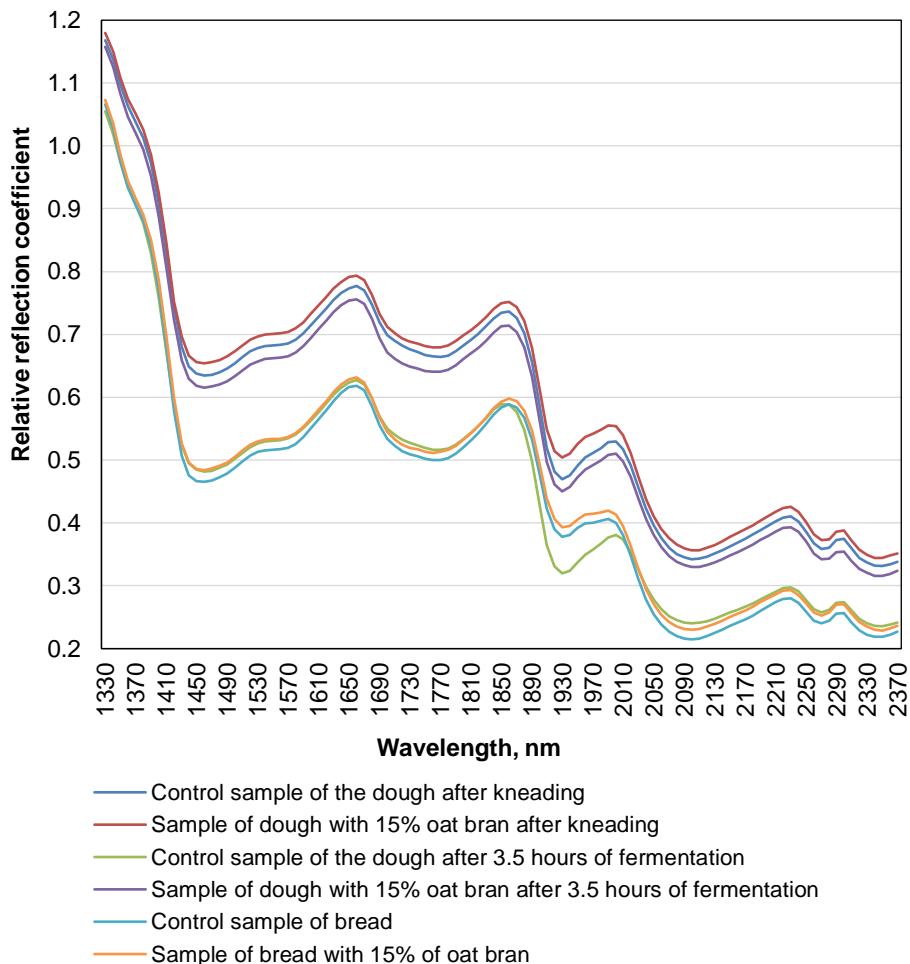
The difference in the chemical composition of wheat flour and oat bran should affect the change in the basic structural units of dough and bread with these components in the recipe. To identify and analyze these components, it is advisable to use the reflection spectrum in the near infrared region (Baslar et. al., 2011). Dough and bread samples were prepared with the minimum and maximum researched replacement of wheat flour with oat bran – 5% (Figure 1) and 15% (Figure 2).

All six spectra (control dough sample and dough samples with 5% replacement of wheat flour with oat bran (after kneading and after 3.5 hours of fermentation), as well as finished bread samples) have a similar character and the same extremes. However, there are differences in the intensity of reflection that identifies them.



**Figure 1. Reflection spectra of dough samples (after kneading and after 3.5 hours of fermentation) and bread samples with 5% replacement of wheat flour with oat bran**

The obtained spectra showed that the control sample of the dough after kneading and the sample of the dough with 5% replacement of wheat flour with oat bran are completely identical throughout the investigated wavelength interval, that is, the spectra are completely superimposed on each other. Even their initial reflection intensity value was the same. This indicates that such a small percentage of substitution does not cause conformational transformations in the dough system. In addition, it takes time for the interaction of the biopolymers of the raw materials, and since the analysis of the samples was done immediately after mixing, not enough time had passed for such an interaction. The lowest extremum is observed at a wavelength of 2100 nm for all samples. This length characterizes the state of the protein substances of the dough (Kröncke et. al., 2022). The relative reflection coefficient is 0.37.



**Figure 2. Reflection spectra of dough samples (after kneading and after 3.5 hours of fermentation) and bread samples with 15% replacement of wheat flour with oat bran**

Spectra of samples of the dough after 3.5 hours of fermentation are situated below on the graph. This indicates the course of conformational processes in the dough during fermentation. Moreover, the dough sample after fermentation with the replacement of flour with 5% oat bran is characterized by a greater intensity of reflection than the fermented control sample. That is, replacing part of the flour with oat bran affects the conformational transformations in the dough system. At a wavelength of 2100 nm, the value of the relative reflectance of the sample with replacement and the control sample was 0.28 and 0.25, respectively. This is explained by the fact that oat bran, which has a globular structure of proteins, due to which they do not participate in the formation of gluten, and also contain a large amount of dietary fibers, are embedded in the gluten framework formed by the gluten proteins of flour and delay its development (Alfaris et. al., 2022). Technologically it can be predicted that the structure of the protein matrix of bran dough will be less stable and more weakened.

The infrared spectra of the baked bread samples conditionally match, not taking into account differences due to different humidity at a wavelength of 1930 nm. This is explained by the fact that high temperatures lead to the destruction of protein macromolecules with the splitting of peptide bonds when baking bread (Zhou et. al., 2021). It is also observed that the indicated spectra of bread are located close to the fermented control dough sample (however, in terms of the intensity of reflection, they are lower). 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. At the same time, although oat bran in the studied amount affects the structural and mechanical properties of the products, it is not capable of significantly changing them.

In general, the spectra have a similar character as when replacing 5%, but the spectra obtained with the replacement of 15% flour with bran can be conditionally divided into two groups. The first (with a higher reflection intensity) includes three spectra: for two samples of dough after kneading (both control sample and with replacement of 15% oat bran) and a sample of dough with 15% oat bran after 3.5 h of fermentation. The relative reflectance of this sample at the wavelength of 2100 nm is 0.35, which is significantly higher than that of the control sample and the sample with 5% replacement. This is explained by the fact that a greater amount of dietary fiber delays the development of the gluten network to a greater extent and more  $\alpha$  – protein structures move to  $\beta$  – structures (Jing et. al., 2016).

The second group has a lower initial reflection intensity (by 0.12 units). The three reflection spectra of this group, which include bread samples and a control sample of fermented dough, also conditionally coincide with each other. Their difference is manifested only at the wavelength of 1930 nm, which is due to the different moisture content in the samples. From a technological point of view, the addition of 15% oat bran will contribute to less dilution of the dough ball during the fermentation process and better shape retention.

Thus, it can be concluded that the introduction of oat bran with a higher content of protein, dietary fiber and a complete amino acid profile into the recipe of wheat bread helps to improve the biological value of bread with this raw material. Also, the addition of bran affects the structural elements of the dough and the structural and mechanical properties of bread. However, the deterioration of the development of the gluten framework of the dough, and therefore of the specific volume, porosity and dimensional stability of bread, requires the use of technological methods to minimize the negative impact of bran on the quality of finished bakery products.

## Conclusion

1. It was established that the content of essential amino acids in oat bran is significantly higher than in wheat flour. The limiting amino acid in wheat flour is lysine, the amino acid score of which is 0.44. In oat bran, the limiting amino acid is methionine, the amino acid score of which is 1.14, and the amino acid score of lysine is much higher than in flour – 1.62.
2. The limiting amino acid in bread is lysine, the amino acid score of which is 0.46. With an increase in the percentage of replacement of wheat flour with oat bran, the amino acid score of lysine increased by 19.5-52.2%. This indicates that the protein of this raw material helps to increase the level of essential amino acids in bread, which enriches its protein profile.

3. The obtained spectra of the dough samples after kneading showed that the introduction of oat bran does not cause conformational transformations in the dough system, since not enough time passed for the interaction of the biopolymers of the raw materials.
4. The infrared spectra of the dough at a wavelength of 2100 nm showed that the dietary fibers of oat bran delay the development of the gluten network, the structure of the protein matrix of the dough with bran will be less stable and more weakened than that of the control sample.
5. The introduction of oat bran with a higher content of protein, dietary fibers and a complete amino acid profile into the recipe of wheat bread helps to improve the biological value of bread with this raw material. However, the deterioration of the development of the gluten framework of the dough, and therefore of the specific volume, porosity and dimensional stability of bread, requires the use of technological methods to minimize the negative impact of bran on the quality of finished bakery products.

## References

- Alfaris N. A., Gupta A. K., Khan D., Khan M., Wabaidur S. M., Altamimi J. Z., Alothman Z. A., Aldayel T. S. (2022), Impacts of wheat bran on the structure of the gluten network as studied through the production of dough and factors affecting gluten network, *Food Science and Technology (Campinas)*, 42(3), DOI: 10.1590/fst.37021
- Amjid M.R., Shehzad A., Hussain S., Shabbir M.A., Khan M.R., Shoaib M. A. (2013), Comprehensive review on wheat flour dough rheology, *Pakistan Journal of Food Sciences*, 23, pp. 105–123.
- Baslar M., Ertugay M.F. (2011), Determination of protein and gluten quality-related parameters of wheat flour using near-infrared reflectance spectroscopy (NIRS), *Turkish Journal of Agriculture and Forestry*, 35(2), pp. 139–144, DOI: 10.3906/tar-0912-507.
- Bock J. E., Damodaran S. (2013), Bran-induced changes in water structure and gluten conformation in model gluten dough studied by Fourier transform infrared spectroscopy, *Food Hydrocolloids*, 31(2), pp. 146–155, DOI: 10.1016/j.foodhyd.2012.10.014
- Campbell G. M., Ross M., Motoi, L. (2008), Bran in bread: effects of particle size and level of wheat and oat bran on mixing, proving and baking. In: Campbell G. M., Scanlon M. G., Pyle D. L. (Eds.), *Bubbles in food 2: novelty, health and luxury*, St. Paul: Eagan Press, pp. 337–354. <http://dx.doi.org/10.1016/B978-1-891127-59-5.50037-7>
- Choi Y.S., Kim H.W., Hwang K.E., Song D.H., Park J.H., Lee S.Y., Choi M.S., Choi J.H., Kim C.J. (2012), Effects of pumpkin (*Cucurbita maxima Duch.*) fiber on physicochemical properties and sensory characteristics of chicken frankfurters, *Food Science of Animal Resources*, 32, pp. 174–183, DOI: 10.5851/kosfa.2012.32.2.174
- Gujral H. S., Sharma B., Khatri M. (2018), Influence of replacing wheat bran with barley bran on dough rheology, digestibility and retrogradation behavior of chapatti, *Food Chemistry*, 240, pp. 1154–1160, DOI: 10.1016/j.foodchem.2017.08.042
- Hussain, M., Saeed, F., Niaz, B., Afzaal, M., Ikram, A., Hussain, S., Mohamed, A. A., Alamri, M. S., Anjum, F. M. (2021), Biochemical and nutritional profile of maize bran-enriched flour in relation to its end-use quality, *Food science & nutrition*, 9(6), pp. 3336–3345, DOI: 10.1002/fsn3.2323
- Ivanov V., Shevchenko O., Marynin A., Stabnikov V., Gubenia O., Stabnikova O., Shevchenko A., Gavva O., Saliuk A. (2021), Trends and expected benefits of the

- breaking edge food technologies in 2021–2030, *Ukrainian Food Journal*, 10(1), pp. 7–36, DOI: 10.24263/2304-974X-2021-10-1-3.
- Jing X., Yang C., Zhang L. (2016), Characterization and Analysis of Protein Structures in Oat Bran, *Journal of Food Science*, 81(10), DOI: 10.1111/1750-3841.13445
- Khan M. A., Amir R. M., Ameer K., Rakha A., Faiz F., Hayat I., Nadeem M., Ahmed Z., Riaz A., Ashraf I. (2020), Characterization of oat bran  $\beta$ -glucan with special reference to efficacy study to elucidate its health claims for diabetic patients, *Food Science and Technology*, 41(1), pp. 105–112. <http://dx.doi.org/10.1590/fst.39019>
- Kröncke N., Benning R. (2022), Determination of Moisture and Protein Content in Living Mealworm Larvae (*Tenebrio molitor L.*) Using Near-Infrared Reflectance Spectroscopy (NIRS), *Insects*, 13, p. 560, DOI: 10.3390/insects13060560
- Litvynchuk S., Galenko, O., Cavicchi, A., Ceccanti, C., Mignani, C., Guidi, L., Shevchenko, A. (2022), Conformational Changes in the Structure of Dough and Bread Enriched with Pumpkin Seed Flour, *Plants*, 11, 2762, DOI: 10.3390/plants11202762
- Packkia-Doss P. P., Chevallier S., Pare A., Le-Bail A. (2019), Effect of supplementation of wheat bran on dough aeration and final bread volume, *Journal of Food Engineering*, 252, pp. 28–35, DOI: 10.1016/j.jfoodeng.2019.01.014
- Shevchenko A., Litvynchuk S. (2022), Influence of rice flour on conformational changes in the dough during production of wheat bread, *Ukrainian Journal of Food Science*, 10(1), pp. 5–15, DOI: 10.24263/2310-1008-2022-10-1-3
- Sterna V., Zute S., Brunava L. (2016), Oat Grain Composition and its Nutrition Benefice, *Agriculture and Agricultural Science Procedia*, 8, pp. 252–256, DOI: 10.1016/j.aaspro.2016.02.100.
- Yip W.L., Gausemel I., Sande S.A., Dyrstad K. (2012), Strategies for multivariate modeling of moisture content in freeze-dried mannitol-containing products by near-infrared spectroscopy, *Journal of Pharmaceutical and Biomedical Analysis*, 70, pp. 202–211, DOI: 10.1016/j.jpba.2012.06.043.
- Zhong L., Ma N., Wu Y., Zhao L., Ma G., Pei F., Hu Q. (2019), Characterization and functional evaluation of oat protein isolate-Pleurotus ostreatus  $\beta$ -glucan conjugates formed via Maillard reaction, *Food Hydrocolloids*, 87, pp. 459–469, DOI: 10.1016/j.foodhyd.2018.08.034.
- Zhou Y., Dhital S., Zhao C., Ye F., Chen J., Zhao G. (2021), Dietary fiber-gluten protein interaction in wheat flour dough: Analysis, consequences and proposed mechanisms, *Food Hydrocolloids*, 111, 106203, DOI: 10.1016/j.foodhyd.2020.10620