Effect of hemp seed by-products on wheat dough fermentation

Mariia Blazhenko, Nataliia Falendysh, Vladyslav Shpak

National University of Food Technologies, Kyiv, Ukraine

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Corresponding author:

Mariia Blazhenko
E-mail: blagmary@ukr.net

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Abstract

Introduction. The aim of the research was to determine the chemical composition of hemp seed by-products, namely hemp seed flour, hemp seed protein, and hemp seed kernels and the effect of their addition on the fermentation activity of yeast in wheat dough.

Materials and methods. Hemp seed flour, hemp seed kernels, and hemp seed protein were obtained from the non-narcotic variety of hemp "Liryna" (Ukraine). Starch content was determined by the Evers polarimetric method, fractional composition of proteins by electrophoresis, amino acid composition by the method of ion exchange chromatography.

Results and discussion. The by-products of hemp processing, namely hemp seed protein, hemp seed flour, hemp seed kernels, contained protein 4.8, 3.4, and 2.5 times higher, and fat 10.4, 8.8, and 30 times higher than first-grade wheat flour. While, the content of carbohydrates in hemp by-products was lower than in wheat flour. The percentage content of starch, mono- and disaccharides, and dietary fiber in hemp seed processing products was as follows: 24.5, 23.2, and 52.3% in hemp seeds protein; 18.0, 20.3, and 61.7% in hemp seed flour; 23.5, 19.8, and 56.7% in hemp seed kernels, respectively. The content of the globular protein fraction was predominant in hemp seed processing products from 49.6% to 61.7%, while in first grade wheat flour their content was only 10.7%. The content of the albumin fraction in hemp seed processing products varied from 28.1% to 32.1% compared to 7.2% in first grade wheat flour. It was determined that the content of essential amino acids in hemp by-products was higher than in first grade wheat flour. The amount of released carbon dioxide decreased from 10 to 70 ml with the addition of hemp seed by-products: 10% hemp seed flour instead of wheat flour, 15% hemp seed protein instead of wheat flour and the combined application of 15% hemp seed protein to replace wheat flour and 7% of hemp seed kernels to the mass of flour, however, the fermentation process intensified. The first peak of gas formation was observed simultaneously in all samples after 60 min of fermentation. The second peak of gas formation in the dough with the replacement of part of the wheat flour with hemp seed-processing products was after 120 min of fermentation, which was 30 min earlier than in the control sample. At the same time, the largest amount of carbon dioxide released in the dough with the replacement of 10% of wheat flour with hemp seed flour.

Conclusions. Hemp by-products had a relatively higher content of proteins, fats, micro- and macro-elements, a balanced composition of carbohydrates, amino- and fatty acids. Different from wheat flour chemical composition affected the course of the dough fermentation process and caused changes in the carbohydrate-amylase complex of the dough.

Introduction

Enrichment of bread and bakery products with non-traditional plant raw materials is a new direction in the manufacturing of functional foods (Ivanov et al., 2021; Stabnikova et al., 2021; Shevchenko, 2022; Shevchenko et al., 2023). To increase the nutritional and biological value of bread and intensify the technological process of its production, hemp seed by-products could be used (Hayit and Gül, 2020). Hemp seeds contain 20–25% easily digestible proteins with a high content of essential amino acids, up to 34% of dietary fiber, as well as a large amount of macro- and microelements (Farinon et al., 2020). In addition to high nutritional value, hemp seeds were also characterized by the presence of various biologically active substances, including phenolic compounds, as well as bioactive peptides with antioxidant, anti-inflammatory, neuroprotective, and antihypertensive properties (Farinon et al., 2020).

The possibility of using hemp seeds in the production of bread was demonstrated (Rusu et al., 2021). It was found that the content of proteins and essential amino acids, lipids and unsaturated fatty acids, fiber and minerals increased in bread when adding 5% hemp flour, which did not significantly affect the rheological properties of bread.

Incorporation of 5 – 40% (w/w) hemp seed flour in wheat flour increased the total phenolic content in the bread, thus significantly increasing its antioxidant activity (Capcanari et al., 2023). It was found that the protein content in the bread increased from 11.11% (control sample) to 18.18% (for a sample with 40% of hemp seed flour). However, the addition of hemp seed flour led to a decrease in the porosity of the bread, an increase in the hardness, and a decrease in the elasticity of the dough.

Replacement of 5 to 20% wheat flour with hemp flour in the production of cookies led to an increase in the content of ash, protein, fat, total phenolic content and antioxidant activity of the final product (Ertas and Aslan, 2020). The highest content of ash, 2.11%, was found in cookies prepared with 20% of roasted hemp flour (in control it was just 1.33%), and the highest content of protein, 11.81%, was detected in cookies with 20% of raw hemp flour (in control it was 8.55%). The antioxidant activity of cookies with 20% of roasted hemp flour was 40.88% and cookies made with 20% of raw hemp flour had an antioxidant activity 38.21%. At the same time, cookies with raw hemp flour up to 20% and roasted hemp flour up to 15 % demonstrated good overall acceptability.

Addition of 10% hemp flour or 5% hemp protein decreased elasticity of bread crumbs Addition of hemp by-products increased the dough yield, which was 165.38% for dough with 10% of hemp flour and 163.91% for dough with 5% of hemp protein (Mullerova et al., 2016). Sensory analysis of gluten-free bread with the addition of 5% hemp flour did not show significant difference from the control bread and it was acceptable to consumers (Hayward and McSweeney, 2020).

Thus, application of hemp seed by-products in manufacturing of bread needs to be deepened and detailed. The aim of the present study was to determine the effect of hemp seed by-products, namely hemp seed flour, hemp seed protein, and hemp seed kernels on the fermentation activity of yeast in wheat flour dough.

Materials and methods

Materials

Hemp seed flour, hemp seed protein and hemp seed kernels were used for research (Desnaland LLC, Ukraine).

Hemp seed flour was obtained from the seeds after oil extraction by cold pressing, without the use of heat or chemicals. Hemp seed protein was obtained from hemp seeds that had been cold-pressed and oil extracted, and further sifted. Ready-to-eat hemp seeds were shelled steamed kernels (Irakli et al., 2019).

The dough was prepared from first grade wheat flour, pressed baker's yeast – 2% to the mass of flour, table salt – 1.5% to the mass of flour, white sugar – 3 – 5% to the mass of flour, sunflower oil – 2% to the mass of flour, barley-malt extract – 10% to the mass of flour. 10% hemp seed flour was added to replace equal amount of wheat flour, 15% hemp seed protein to replace equal amount of wheat flour, and composition of 15% hemp seed protein to replace equal amount of wheat flour containing 7% of hemp seed kernels to the mass of flour. The control was dough without hemp by-products. Therefore, the aim of the research was to determine the effect of hemp seed byproducts, namely hemp seed flour, hemp seed protein and hemp seed kernels on the fermentation activity of yeast in wheat flour dough.

Methods

Determination of protein content

Determination of the protein content in first grade wheat flour, hemp seed flour, hemp seed protein, and hemp seed kernels was carried out by the biuret method.

1.5 g of the product was weighed with an accuracy of 0.0001 g and placed in a dry conical flask with a capacity of 250 – 300 ml with a stopper. A portion of the product was completely moistened with 2 ml of carbon tetrachloride to remove fat. Then 100 ml of the biuret reagent was added, the flask was closed with a cork and shaken with a non-mechanical shaker for 60 min. After that, obtained extract was centrifuged for 10 min at 4500 rpm. The transparent centrifuge was placed in the cuvette of the photoelectrocolorimeter with a solution layer thickness of 5 mm. The optical density of the hood was measured at a wavelength of 550 nm. Based on the obtained value of the optical density of the protein extract, the protein content was determined using a calibration graph for the biuret method. The result was expressed as a percentage of the dry matter of the product (López-Bascón and de Castro, 2020).

Determination of fat content

Determination of the fat content in first grade wheat flour, hemp seed flour, hemp seed protein, and hemp seed kernels was carried out by the method of multiple extraction of fat from the appropriate weight of the product.

From the sample of the above-mentioned raw materials, two measurements were separated, each with a mass of (10±0.1) g. One was transferred to a filtering separatory funnel, (25±0.5) ml of diethyl ether was added, closed with a ground stopper and extraction was carried out by intensive shaking for 1 min. The obtained fat extract was sucked by a water-jet pump into a receiver attached to the filtering funnel, into which 2 – 3 ml of diethyl ether was poured before the extraction to prevent possible losses of the concentrated part of the first extract. The extract from the receiver was filtered through a pleated paper filter into a pre-dried and weighed flask. Similarly, the extraction was repeated in the same flask. Ether was driven off the flask using a Soxhlet apparatus. Then the flask with fat was placed in a drying cabinet and dried for 2 hours at a temperature of (105 ± 5) ºC, cooled in a desiccator and weighed with an error of ± 0.0002 g (López-Bascón and de Castro, 2020).

Determination of the starch content

The determination was made by the Evers method (Koshova et al., 2020). 5±0.01 g of the product was put into a volumetric flask, where 25 ml of 0.31 mol/l HCl solution was added. An additional 25 ml of HCl was put to rinse the residue into the flask. The flask was shaken, water was brought up to the neck and after 15 minutes 25–30 ml of distilled water was poured. For the precipitation of proteins and clarification, 5 ml of ammonium molybdate solution or 5 ml of phosphorous-tungstic acid solution or 2 ml of Carrez reagent I and II was added to the flask. After 5 minutes, the solution was filtered. On the saccharimeter, the angle of rotation of the plane of polarization was measured, the length of the polarizing tube was 200 mm. In parallel, a control experiment was conducted, which differred in that the flask must be kept in a boiling water bath for 15 minutes.

The starch content was calculated according to the formula (1):

$$C = \frac{\left(a_g - a_k\right) \cdot F \cdot 100}{100 - W}, \tag{1}$$

where a_g is the value of the angle of rotation of the plane of polarization by optically active substances in the main experiment, degree of saccharimeter;

a_k is the value of the angle of rotation of the plane of polarization by water-soluble optically active substances in the control experiment;

F is Evers coefficient, $1000/\left[a\right]_D^{20}$

W is the moisture content in the product, %.

Determination of mono- and disaccharides content

The determination was made by the Schorl method (Pokrzywnicka and Koncki, 2017). 30 ml of extract or hydrolyzate was put to a 250 ml conical flask, 10 ml of Feling I and Feling II reagents was added with a pipette, boiled for 2 minutes and cooled. To determine the amount of unreacted copper, 10 ml of a 30% potassium iodide solution and 10 ml of 25% sulfuric acid were added. The released iodine was immediately titrated with a 0.1 mol/l solution of sodium thiosulfate to a light yellow color, 2 ml of a 1% solution of soluble starch was added as an indicator, and the titration continued until the blue color disappeared.

In parallel, a control experiment was conducted in which everything was done similarly, replacing the sugar solution with the appropriate amount of distilled water.

Sugar content x, % to dry matter, in the product was calculated according to the formula (2):

$$x = \frac{\left(V_k - V_p\right) \cdot K \cdot 100 \cdot 100}{H\left(100 - W\right)}, \tag{2}$$

where V_k is the amount of 0.1 mol/l sodium thiosulfate solution used for titration in the control experiment, ml;

V_p is the amount of 0.1 mol/l sodium thiosulfate solution used for titration in the working solution, ml;

K is conversion factor for the type of sugar;

H is the amount of the product corresponding to the volume of extract or hydrolyzate taken for reaction with Fehling reagents, mg;

W is the moisture content in the product, %.

Determination of dietary fiber content

A general study was conducted to determine the total content of dietary fiber in products by the enzymatic-gravimetric method. Dietary fiber was calculated as the residual weight minus the weight of protein and ash (Shevchenko and Litvynchuk, 2022).

Determination of ash content

Ash content in wheat flour, hemp seed flour, hemp seed protein, and hemp seed kernels was determined by burning the sample with the subsequent determination of the unburned residue. Ashing was carried out using a nitric acid accelerator (Sahayam et al., 2009). The sample was weighed: for flours and protein – 1.5–2.0 g, for hemp seed kernels – 1.0–1.5 g into two crucibles pre-heated to a constant mass and cooled in a desiccator.

Weighed crucibles with a measuring scale were placed near the door of a muffle furnace heated to a temperature of 400–500 °C, the measuring scale was charred, preventing ignition of dry distillation products. After the separation of dry distillation products stopped, the crucibles were placed in a muffle furnace and the door was closed. The contents of the crucibles were ashed until they turned into a loose mass of gray color, cooled in air to room temperature, 2–3 drops of nitric acid were added and placed near the door of the muffle furnace, and the acid was carefully evaporated to dryness without allowing boiling. Then the crucibles were placed inside the muffle furnace, heated to a temperature of 600 – 900 °C, and ashed for 20–30 minutes. When there were no dark spots at the bottom of the crucible, ashing was considered complete. After the crucible was cooled to room temperature in a desiccator and weighed.

The ash content of product X, % to dry matter, was calculated according to the formula (3):

$$X = \frac{m_3 \cdot 100 \cdot 100}{m_н \cdot (100 - W)}, \quad (3)$$

where m_3 – mass of ash, g, $m_н$ – mass of the product, g, W – mass fraction of moisture in the product, %.

Determination of the moisture content

5±0.001 g of the product was put to the dried bags, placed in an open oven at a temperature of 100–105 °C for 4 hours. Further, they were cooled in a desiccator, weighed and again placed for drying, the end of drying was considered when the difference between the weight was no more than 0.004 g or the weight of the sample began to increase.

The moisture content (W) was calculated according to the formula (4):

$$W = \frac{m_1 - m_2}{m} \cdot 100, \quad (4)$$

where m is mass of the product, g;

m_1 is mass of the product with the bushing before drying, g;

m_2 is mass of the product бюксою the bushing after drying, g (Pasichnyi et al., 2023).

Determination of fractional composition of proteins

Fractional protein composition of raw materials was carried out by electrophoresis.

Electrophoresis was performed at a current of 40 mA and a voltage of 100–150 V based on two gel plates with 12 samples each. The speed of movement of proteins in the gel was controlled by bromophenol blue (0.1 mol/l). The protein separation time was 12 hours.

Determination of amino acid composition

The content of amino acids was determined using the method of ion exchange chromatography. The quantity of amino acids was determined by hydrolysis of proteins using automatic amino acid analyzer T–339 with polystyrene sulfonate ions. Amino acids were detected at a wavelength of 560 nm by rectification with a ninhydrin solution on a Unicam SP 800 photometer (Litvynchuk et al., 2022).

Determination of fatty acid composition

Fatty acid content was determined by gas-liquid chromatography (Chowdhury, 2007). The gas-liquid chromatograph was equipped with a flame ionization detector and a stainless steel column. The column was heated at 180 ºC for about 2 hours to achieve thermal stability before use. The operating mode was programmed in the oven at a temperature of 150 °C (holding time 5 min), the rate of increase from 80 ºC/min to 190 °C (holding time 0 min), from 20 °C/min to 200 °C (holding time 10 min), injection temperature 250 0 C and 312 Studies on the Fatty Acid 42(3) 2007 detector temperature 250 ºC. Nitrogen was used as a carrier gas with a flow rate of 20 ml/min.

Determination of micro- and macroelements content

The content of micro- and macroelements was determined using atomic absorption spectrometry and optical emission spectrometry with inductively coupled plasma (Başgel and Erdemoğlu, 2006; Malik et al., 2008).

The method of atomic absorption spectrophotometry was based on the phenomenon of absorption of resonant radiation with the corresponding wavelength by free atoms of the element, which were determined to be formed as a result of spraying the analyzed solution in the flame of a combustible mixture: air-acetylene, air-propane-butane.

Determination of gas-forming capacity of the dough

The kneaded dough was divided in such a way that each piece of dough contained 25 g of flour. Each piece was placed in a vessel for fermentation and kneaded with a rolling pin. The vessel was placed in the water thermostat of the AG–1 device, closed with a rubber stopper with a tube. The cylinder was filled with oil; the access of carbon dioxide to the tube was opened. Results were foxed every 30 minutes during 5 hours. The average arithmetic value of two parallel determinations was multiplied by 4 to remove from 25 g of flour to 100 g.

Results and discussions

Chemical composition of hemp seed by-products

Food products, including bakery products, are the main source of energy and nutrients for the human body (Drobot and Shevchenko, 2017).

Hemp by-products were not sufficiently studied in terms of their chemical composition and technological properties. It was appropriate to determine the general chemical composition of hemp seed by-products: hemp seed flour (Figure 1a), hemp seed protein (Figure 1b), and hemp seed kernels (Figure 1c) in comparison with first grade wheat flour (Table 1).

Figure 1. Hemp seed by-products:
hemp seed flour (a), hemp seed protein (b), hemp seed kernels (c)

The protein content in hemp seeds was 4.8 times higher, in hemp seed flour 3.4 times higher, in hemp seed kernels 2.5 times higher than in first-grade wheat flour. The total fat content of hemp seed by-products was also higher than that of first grade wheat flour. Hemp seed protein, hemp seed flour, and hemp seed kernels contained 10.4, 8.8, 30 times more fat, respectively, than wheat flour.

It was found that the content of carbohydrates in hemp by-products was lower than in first grade wheat flour. However, the content of total carbohydrates in wheat flour was largely provided by starch (92% of the total amount of carbohydrates), while in hemp by-products the content of total carbohydrates had more balanced composition among starch, mono- and disaccharides and dietary fibers: in hemp seed protein – 24.5, 23.2, and 52.3%, in hemp seed flour – 18.0, 20.3, and 61.7%, in hemp seed kernels – 23.5, 19.8, and 56.7%, respectively.

Table 1

Chemical compositions of first grade wheat flour and hemp seed by-products

Components	**Content, %**			
	First grade wheat flour	**Hemp seed protein**	**Hemp seed flour**	**Hemp seed kernels**
Protein,	11.6±0.23	55.4±0.26	38.1±0.42	29.3±0.24
Fat	1.4±0.45	14.5±0.13	12.3±0.23	42.1±0.26
Carbohydrates				
Starch	68.0±0.31	4.0±0.20	6.0±0.52	4.4±0.16
Mono- and disaccharides	1.8±0.12	3.8±0.33	6.8±0.11	3.7±0.14
Fiber	3.5±0.43	8.5±0.32	20.7±0.24	10.6±0.17
Ash	0.75±0.11	4.8±0.22	4.9±0.15	4.2±0.19
Moisture	13.0±0.28	9.0±0.30	11.2±0.18	5.7±0.26

Results are presented as mean ± standard deviation of three replicates.

In the study of Capcanari et al. (2023), the protein, fat and dietary fiber contents in hemp seed flour were 31.6, 8.2%, and 23%, which are consistent with the values obtained in the present study.

Fractional compositions of proteins of first grade wheat flour and hemp seed by-products are shown in Table 2.

Table 2

Fractional compositions of proteins of first grade wheat flour and hemp seed by-products

Products	Content, %				
	albumins	**globulins**	**prolamins**	**glutelins**	**non-protein nitrogen**
First-grade wheat flour	7.2±0.52	10.7±0.65	23.5±0.35	58.6±0.58	5.4±0.32
Hemp seed flour	32.1±0.94	58.4±0.96	6.6±0.26	2.9±0.21	9.7±0.41
Hemp seed protein	30.2±0.87	61.7±0.98	5.5±0.24	2.6±0.19	12.8±0.49
Hemp seed kernels	28.1±0.73	49.6±0.84	20.9±0.45	1.4±0.15	11.9±0.47

Results are presented as mean ± standard deviation of three replicates

Fraction of glutelins dominated in first grade wheat flour where it consisted 58.6%, while content of glutelins in the proteins of hemp seed by-products was very low (Table 2). This is consistent with the results reported in (Apetroaei et al., 2024).

The globular protein fraction was predominant in hemp seed processing products, and its content varied from 49.6% to 61.7%, while in first grade wheat flour their content was only 10.7%. The content of the albumin fraction in hemp seed processing products varied from 28.1% to 32.1% compared to 7.2% in first grade wheat flour.This confirmed that hemp by-products will not contribute to the formation of the gluten framework, as they do not contain gluten proteins (Marinopoulou et al., 2024).

It was shown that the amino acid composition of seed proteins differs depending on the varieties of hemp (Russo et al., 2015). The amino acid compositions of first grade wheat flour and hemp seed by-products used in the present study are shown in Table 3.

The content of essential amino acids in hemp by-products was higher than in first grade wheat flour. In particular, the content of leucine was 2.4, 2.3, and 1.1 times, content of lysine 5.8, 5.2, and 3.2 times, content of methionine 3.6, 3.3, and 1.6 times, threonine 3.4, 3.3, and 1.8 times, phenylalanine 2.7, 2.5, and 1.3 times higher in hemp seed protein, hemp seed flour, and hemp seed kernels compared to their contents in first grade wheat flour, respectively. This indicated the feasibility of increasing the biological value of bread when such raw materials are added to the recipe.

The chemical composition of first grade wheat flour and hemp by-products differed significantly in terms of total fat content. The research results showed (Table 4) a diverse fatty acid composition; however, it was necessary to distinguish the content and ratio of ω – 3 and ω – 6 fatty acids. They prevent the development of atherosclerosis, coronary heart disease, stroke, myocardial infarction, have antioxidant properties, According to World Health Organization recommendations, the ideal ratio of ω–6/ω–3 fatty acids in food products is considered to be 4:1 (Simopoulos, 2002; Stabnikova and Paredes-Lopez, 2024; WHO, 2003).

Analysis of the fatty acid composition showed that the ratio of ω–6/ω–3 fatty acids was the closest to the ideal: for hemp seed kernels it was 3.91, while for hemp seed flour it was 3.5:1, and for hemp seed protein it was 3.4:1 (Table 4).

Table 3

Amino acid compositions of first grade wheat flour and hemp seed by-products

Amino acids	Content, mg/100 g of protein			
	First grade wheat flour	Hemp seed protein	Hemp seed flour	Hemp seed kernels
Alanine	330±15	1556±31	1462±30	735±27
Arginine	400±20	3589±52	3411±45	1647±32
Aspartic acid	340±16	2263±62	2224±26	1359±62
Valin	470±22	885±31	910±56	445±28
Histidine	200±12	870±32	806±47	41±3
Glycine	350±15	1272±47	1319±66	740±56
Glutamic acid	3080±57	4445±88	4625±91	2870±87
Isoleucine	430±22	782±24	813±38	374±23
Leucine	806±35	1951±42	1877±63	913±62
Lysine	250±17	1458±39	1300±52	788±48
Methionine	190±12	686±25	630±24	302±22
Proline	970±64	1358±38	1305±40	673±32
Serin	500±32	1597±37	1514±48	824±35
Tyrosine	250±25	1078±36	955±36	469±28
Threonine	311±33	1056±34	1029±37	555±56
Phenylalanine	500±45	1350±35	1271±42	653±65
Cysteine	200±19	594±24	545±22	197±20

Results are presented as mean ± standard deviation of three replicates.

Table 4

Fatty acid compositions of hemp by-products

Fatty acids	Content, % to the total mass		
	Hemp seed flour	Hemp seed protein	Hemp seed kernels
Saturated fatty acid			
Myristic acid (C14:0)	0.32±0.06	0.35±0.01	0.20±0.01
Palmitic acid (C16:0)	6.44±0.12	8.01±0.16	7.22±0.42
Stearic acid (C18:0)	3.04±0.08	4.66±0.05	3.87±0.01
Arachinic acid (C20:0)	4.37±0.03	4.1±0.05	4.53±0.06
Geneicosanoic acid (C21:0)	1.13±0.08	1.09±0.06	1.11±0.07
Monounsaturated fatty acid			
Palmitoleic acid (C 16:1, ω-7)	0.13±0.01	0.14±0.01	0.11±0.01
Oleic acid (cis-18:1, ω -9)	12.87±0.13	12.59±0.012	12.68±0.08
Elaidic acid (9-trans-C18:1, ω-9)	0.1±0.01	0.64±0.01	0.11±0.01
Gondoic acid (C20:1, ω-9)	0.003±0.00012	0.41±0.02	0.33±0.07
Polyunsaturated fatty acids			
Linoleic acid (C18:2, ω-6)	54.55±0.20	51.44±0.2	54.97±0.2
γ-linolenic acid (C18:3, ω-6)	0.14±0.01	0.15±0.01	0.05±0.01
α-linolenic acid (C18:3, ω-3)	15.8±0.19	15.18±0.18	14.17±0.18
Arachidonic acid (C20:4, ω-6)	0.04±0.01	0.08±0.01	0.01±0.001
Ratio of ω-6 : ω3	3.5 : 1	3.4 : 1	3.9 : 1

Results are presented as mean ± standard deviation of three replicates.

Hemp by-products were less refined than first-grade wheat flour, in particular, hemp seed flour was whole-ground, so, it could be assumed that they contain high levels of macro- and microelements (Table 5).

By-products of hemp processing contain large amounts of magnesium, which is essential for the proper functioning of the body participating in the regulation of blood pressure, blood sugar levels and preventing the development of osteoporosis (Glasdam et al., 2016). Content of magnesium in hemp seed protein, hemp seed flour and hemp seed kernels was by 29.1, 26.8, and 22.1 times higher than in first grade wheat flour. Content of potassium, which reduces the risk of cardiovascular diseases, regulates heart rhythm, and maintains normal muscle tone (McLean and Wang, 2021) was by 9.5, 8.9, and 8.6 times higher in hemp seed protein, hemp seed flour and hemp seed kernels, than its content in wheat flour. The amount of calcium that regulates blood clotting, ensures the strength of bones and teeth, and is important for the functioning of human immune system (Beto, 2015) was by 12.8, 12.0, and 3.3 times higher in hemp seed protein, hemp seed flour and hemp seed kernels than its content in wheat flour. Content of zinc, which is essential for normal function of brain and immune system (Kiouri et al., 2023) was by 11.6, 10.5, and 9.9 times higher in hemp seed protein, hemp seed flour and hemp seed kernels than its content in wheat flour. Content of selenium, which play an important role in thyroid hormone metabolism, immune responses, and protection from oxidative damage (Stabnikova et al., 2022) was by 2.4, 3.6, and 8.2 times higher in hemp seed protein, hemp seed flour and hemp seed kernels than its content in wheat flour.

Table 5

Content of macro- and microelements in first grade wheat flour and hemp seed by-products

Macroelements	**First grade wheat flour**	**Hemp seed protein**	**Hemp seed flour**	**Hemp seed kernels**
	Content, mg/g			
Magnesium	0.22±0.01	6.40±0.08	5.9±0.05	4.86±0.05
Calcium	0.15±0.01	1.92±0.02	1.8±0.02	0.5±0.01
Phosphorus	1.08±0.03	11.17±0.02	9.2±0.08	8.2±0.09
Potassium	1.07±0.02	10.20±0.15	9.6±0.07	9.2±0.08
Microelements	**Content, µg/g**			
Manganese	57.0±0.08	129.0±0.26	152.9±0.27	68.2±0.16
Zinc	7.0±0.06	80.9±0.19	73.8±0.20	69.6±0.17
Iron	12.6±0.06	147.9±0.23	127.5±0.23	72.1±0.19
Cobalt	0.1±0.01	0.9±0.01	0.6±0.01	0.2±0.01
Copper	7.4±0.12	19.1±0.09	15.9±0.15	10.7±0.05
Selenium	0.34±0.01	0.83±0.01	1.22±0.02	2.77±0.02

Results are presented as mean ± standard deviation of three replicates.

Hemp seed flour used in the study of Capcanari et al. (2023) had similar content of minerals, mg/g: magnesium, 8.83; phosphorus, 18.51; potassium, 15.94; µg/g: zinc, 120; iron, 110, and copper, 20.

Effect of hemp seed by-products on wheat dough fermentation

The effect of hemp by-products on the carbohydrate-amylase complex of the dough was determined. The main process during the ripening of the dough is fermentation, during which carbon dioxide is released, which ensures the loosening of the dough.

It was found that the amount of released carbon dioxide in the samples with the replacement part of wheat flour with hemp by-products was lower compared to the control sample (Figure 2). In the dough with replacement of 10% of wheat flour with hemp seed flour 10 ml carbon dioxide released less compared to the control. When replacing 15% of wheat flour with hemp protein, 20 ml carbon dioxide released less and when replacing 15% of wheat flour with hemp seed protein and adding 7% of hemp seeds, 70 ml carbon dioxide released less.

The decrease in gas formation in dough with hemp seed by-products compared to the control is explained by the features of the protein composition of additives, namely the differences in the fractional composition of proteins compared to proteins of wheat flour (Table 2).

Glutelins and gliadins are the main types of wheat flour proteins responsible for the formation of the gluten framework of the dough (Urade et al., 2018). The protein composition of hemp seed by-products consists mostly of globular (albumin and globulin) proteins (Table 2), which have a compact spatial structure, greater strength, thereby not contributing to the stretchability of the dough and the creation of an elastic gluten frame compared to glutelins in the composition of wheat flour proteins (El- Sohaimy et al., 2022; Marinopoulou et al., 2024).

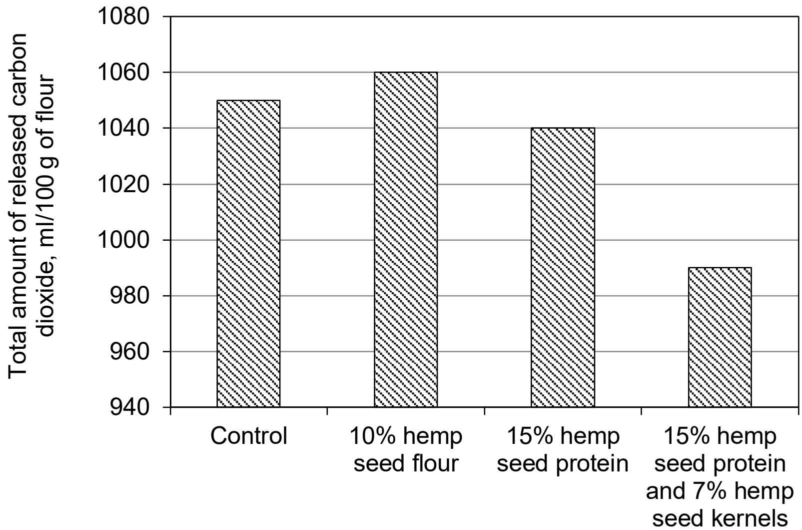

Figure 2. Influence of hemp by-products on amount of released carbon dioxide in the dough during fermentation

The dynamics of carbon dioxide release during 5 hours of fermentation is shown in Figure 3.

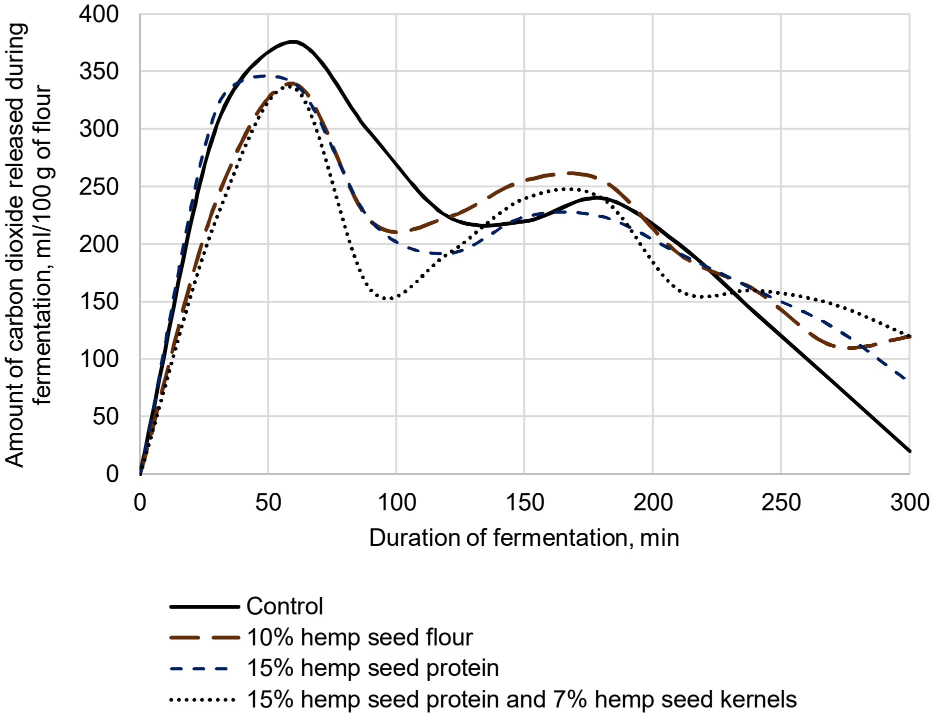

Figure 3. Influence of hemp by-products on dynamics of gas formation in dough during fermentation

It was found that the sugars of the control (dough without hemp by-products) were fermented most intensively (Figure 3). However, the first peak of gas formation was observed simultaneously in all samples after 60 min of fermentation. The second peak of gas formation, which indicates the beginning of maltose fermentation (Hackenberg et al., 2018), in the dough with the replacement of part of the wheat flour with hemp by-products began after 120 min of fermentation, which was 30 min earlier than in the control sample. At the same time, the highest amount of carbon dioxide was released in dough with the replacement of 10% of wheat flour with hemp by products.

The dynamics of gas formation in the dough with the replacement of 10% of wheat flour with organic hemp flour of the "Richoil" brand was studied (Tomashpolska et al., 2021). The first peak was observed after 150 min of fermentation, which was 90 min more compared to our results, which indicated a longer dough fermentation process. The addition of hemp seed flour to wheat bread recipe reduced gas formation. This is explained by the absence of gluten proteins and the increased fiber content in hemp flour (Istrate et al., 2021).

Replacing 30–50% of wheat flour with hemp seed flour in wheat bread recipes worsened the elasticity of the dough, reduced the gas formation (Mikulec et al., 2019), and, therefore, reduced the specific volume of bread. So, it was recommended to use no more than 30% hemp seed flour in wheat bread recipes. It was shown that the replacement of 10, 15 and 20% of wheat flour with hemp seed by-products led to a decrease in the amount of carbon dioxide formed during the fermentation of test samples of the dough for 180 minutes by 23-

30% compared to the control sample wheat dough without the addition of hemp seed by-products (Sokolova et al., 2020).

Based on the results of the research, it was found that hemp seed by-products have a rich chemical composition, a high content of protein and polyunsaturated fatty acids, they were balanced in terms of amino and fatty acid composition, and have an increased content of dietary fiber and micro- and macroelements compared to wheat flour. Therefore, hemp seed by-products can be considered promising raw materials for the use in bread production. However, the chemical composition different from wheat flour affected the course of the dough fermentation process and changes in the carbohydrate-amylase complex of the dough.

Conclusions

1. Determination of the chemical composition showed that the hemp by-products had 2.5–4.8 times more total protein and 8.8–30 times more fats, as well as a balanced composition of carbohydrates than first grade wheat flour.

2. As a result of determining the fractional composition of proteins, it was found that wheat flour of the first grade contained mostly the glutenin fraction, unlike hemp by-products, which were dominated by the albumin and globulin fractions. Hemp by-products had higher content of essential amino acids than in first grade wheat flour.

3. It was found that hemp by-products contained $\omega - 3$ and $\omega - 6$ fatty acids. In particular, in hemp seed kernels the ratio was 1:3.9, while in hemp seed flour – 1:3.5, in hemp hemp seed protein – 1:3.4.

4. The content of micro- and macroelements in hemp seed by-products was several times higher than in wheat flour of the first grade. Magnesium content was more than 20 times higher compared to wheat flour, potassium content – 8–9 times higher, zinc – 9–11 times higher, calcium content in hemp seed flour and hemp seed protein was 12 times higher compared to wheat flour, but hemp seed kernels - lower 3.3 times.

5. The amount of released carbon dioxide in the dough with the replacement of part of wheat flour with hemp by-products was lower compared to the control dough by 10–70 ml during 5 hours of dough fermentation.

6. The total amount of released carbon dioxide in dough with the addition of hemp by-products was less compared to the control sample, however, the second peak of gas formation, which indicates the fermentation of maltose, occured faster by 30 min, which indicates the intensification of the dough fermentation process. This result makes it possible to speed up the dough ripening process and accordingly reduce the time spent on the technological process of bread production with the appropriate recipe composition.

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