

## Gluten-free muffins based on fermented and unfermented buckwheat flour – content of selected elements

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### Summary

The aim of the study was to determine the content of macro- and microelements in innovative gluten-free muffins. Four types of innovative muffins were obtained from unfermented or fermented, by *Lactobacillus plantarum*, common buckwheat flour suspension mixed with buckwheat flour or commercial gluten-free maize flour. Buckwheat flour was a better source on macro- and microelements compared to maize gluten-free flour. Only the calcium content in gluten-free maize flour was higher than in buckwheat flour. Innovative muffins obtained from unfermented or fermented, by *Lb. plantarum*, common buckwheat flour suspension mixed with buckwheat flour showed the highest content of macroelements: potassium and magnesium, and also higher content of microelements: zinc and manganese. The applied fermentation process significantly decreased copper content in the samples containing fermented buckwheat flour compared to other muffin samples.

### Keywords

microelements; macroelements; *Lactobacillus plantarum*; baking

Approximately 1% of world population suffer for celiac disease, which is intolerance for gluten in genetically pre-disposed individuals. The only available treatment for this disease is a lifelong strict gluten-free diet. The people suffering for celiac disease have a decreased intestinal absorption of several nutrients such as minerals, folic acid and fat-soluble vitamins [1]. Therefore, it is still necessary to search for the components supplementing gluten-free products, which will increase the content of the nutritional and dietary components. Use of different fibre, whole-meal flour, addition of vitamins and minerals lead to an increase in the nutritional level of gluten-free products [2–4].

Buckwheat (*Fagopyrum esculentum* Moench) is known and documented as a component of gluten-free products [5, 6]. Buckwheat flour is a rich source of minerals, flavonoids, phytosterols, phenolic compounds, resistant starch, dietary fibre, lignans, vitamins and antioxidants [7, 8]. Substitution of gluten-free formula with buckwheat flour

increases the levels of total proteins, macro- and microelements, and improvement of technological and overall sensory quality of bread was also noted [9, 10].

Worldwide popular muffins are sweet and high-energy cookies with a good taste and soft texture. Traditionally, they are prepared from wheat flour, eggs, sugar, oil or fat and milk, which means that people suffering from celiac disease cannot consume them [11, 12]. An increase in mineral content in food could be achieved by dietary diversification, mineral supplementation or food fortification. Fruits and vegetables are the main source of minerals in the human diet.

The objective of this work was to determine the level of micro- and macroelements in four types of innovative gluten-free muffins made from unfermented and fermented, by *Lactobacillus plantarum*, common buckwheat flour in comparison with muffins made from commercial gluten-free maize flour.

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## MATERIAL AND METHODS

### Ingredients for muffins formulation

The following ingredients were used: commercial granulated sugar (Korunný cukor, Slovenské cukrovary, Sered', Slovakia), sunflower oil Prommiena (PD Vlára, Nemšová, Slovakia), eggs (medium size 53–63 g, PD Vlára), salt Castello (PD Vlára) and sodium bicarbonate, p. a. (NaHCO<sub>3</sub>; Slavus, Bratislava, Slovakia). Gluten-free maize flour was provided by Dr. Schär (Burgstall, Italy). Flour from common buckwheat was provided by the local industry from North-East Poland.

### Preparation of buckwheat or gluten-free maize flour suspensions

About 25 g of buckwheat flour or gluten-free maize flour was mixed with 100 ml of water, brought to boil and stirred properly in a water bath, until a thick flour suspension was obtained. Glass flasks were closed with twist-off cups and sterilized in autoclave at 121 °C for 15 min, at 200 kPa.

### Preparation of stock culture of *Lb. plantarum*

Lyophilized culture of *Lactobacillus plantarum* S-lak 1 (collection from Stuvital, Bratislava, Slovakia) was recovered in 5 ml of de Man-Rogosa-Sharpe (MRS) medium (Merck, Darmstadt, Germany) for 1 h at 37 °C. The recovered culture was inoculated on MRS agar plates and incubated for 48–72 h at 37 °C under anaerobic conditions. Stock

solution of *Lb. plantarum* was prepared by inoculation of one colony from the plate in 10 ml of MRS medium and maintained at 4 °C for 2 weeks (after that re-inoculated in the same medium). The stock culture (0.2 ml) was suspended in 20 ml of MRS medium and incubated for 24 h at 37 °C without shaking.

### Preparation of fermented suspension of flour

A volume of 1 ml of a 24 h inoculum of *Lb. plantarum* was added into the buckwheat flour suspension, mixed and incubated for 24 h at 25 °C without shaking.

### Muffin-making process

The basic formulas of control and experimental muffins are shown in Tab. 1. The mixture of all ingredients was blended by planetary rotation mixing in a 5-speed mixer KitchenAid Model 5KSM150PS (Artisan, Benton Harbor, Michigan, USA). Portions of 50 g of dough in paper cups were baked at 180 °C for 25 min. The baking tests were carried out in an electric oven (Miwe Condo, Arnstein, Germany). The mass of each fresh muffin, after cooling, was approx. 40 g. Muffins were dried at laboratory temperature for 24 h and milled into powder. The following sample abbreviations were used (Tab. 1): C – control muffins from unfermented gluten-free maize flour suspension with gluten-free maize flour, UB – muffins from unfermented buckwheat flour suspension with buckwheat flour, FB – muffins from fermented buckwheat flour suspension with buckwheat flour, FG – muffins from fermented buckwheat flour suspension with gluten-free maize flour.

Tab. 1. Recipe for experimental muffins.

Ingredients [g]	Control muffins (C)	Gluten-free buckwheat muffins		Gluten-free buckwheat/maize muffins	
		Unfermented (UB)	Fermented (FB)	Unfermented (UG)	Fermented (FG)
Buckwheat flour	–	54	54	–	–
Gluten-free maize flour	54	–	–	54	54
Gluten-free maize flour in suspension	36	–	–	–	–
Unfermented buckwheat flour in suspension	–	36	–	36	–
Fermented buckwheat flour in suspension	–	–	36	–	36
Eggs	116 ± 10	116 ± 10	116 ± 10	116 ± 10	116 ± 10
Sugar	50	50	50	50	50
Sunflower oil	10	10	10	10	10
Salt	0.3	0.3	0.3	0.3	0.3
NaHCO <sub>3</sub>	2	2	2	2	2

C – control muffins from unfermented gluten-free maize flour suspension with gluten-free maize flour, UB – muffins from unfermented buckwheat flour suspension with buckwheat flour, FB – muffins from fermented buckwheat flour suspension with buckwheat flour, UG – muffins from unfermented buckwheat flour suspension with gluten-free maize flour, FG – muffins from fermented buckwheat flour suspension with gluten-free maize flour.

flour, UG – muffins from unfermented buckwheat flour suspension with gluten-free maize flour, FG – muffins from fermented buckwheat flour suspension with gluten-free maize flour.

### Methods

Ash content was determined according to AOAC method No 32.3.08 [13]. The content of selected macro- and microelements was determined by atomic absorption spectrometry (AAS) according to KOREŇOVSKÁ and SUHAJ [14]. The powdered dried muffins (samples of approx. 0.5 g) were digested in a microwave digestion system Milestone MLS 1200 MEGA (Soriso, Italy). For digestion, 4 ml of 65%  $\text{HNO}_3$  and 0.5 ml  $\text{H}_2\text{O}_2$  were added to each sample. After digestion, samples were adjusted to volumes of 10 ml. The content of ash as well as micro- and macroelements in muffins was expressed on dry matter (DM) basis.

### Statistical analysis

The measurements were performed in three repetitions for each type of muffins obtained from two separate baking processes for every formulation. The reported data are the mean results for each formulation with the standard deviation. Obtained results were analysed by one-way ANOVA. Fisher's Least Significant Difference Test at a significance level of  $p < 0.05$  was performed for post-hoc comparison.

## RESULTS AND DISCUSSION

The content of ash as well as micro- and macroelements in flours and muffins is compiled in Tab. 2. Buckwheat flour had a 7-fold higher ash content in comparison to the gluten-free maize flour used for muffin preparation. This finding was in agreement to the general evidence that buckwheat flour is a better source of elements in comparison to the gluten-free maize flour. In our study, buckwheat flour was richer in all studied elements, with an exception of calcium, content of which was higher, but not statistically significantly, in gluten-free maize flour (Tab. 2).

Control muffins (C) prepared from the gluten-free maize suspension and gluten-free maize flour were characterized by significantly lower content of ash in comparison to the four types of innovative muffins. Muffins prepared from gluten-free maize flour with unfermented (UG) or fermented buckwheat flours (FG) showed lower ash content by 11% as compared to muffins prepared from unfermented buckwheat flour suspension with buckwheat flour (UB) ( $17.9 \text{ g}\cdot\text{kg}^{-1} \text{ DM}$ ) and from

fermented buckwheat flour suspension with buckwheat flour (FB) ( $18.3 \text{ g}\cdot\text{kg}^{-1} \text{ DM}$ ).

It should be pointed out that the content of calcium in both flours was almost twice lower as compared to all analysed muffins. In all muffins (C, UB, FB, UG, FG), calcium content was similar (approx.  $600 \text{ mg}\cdot\text{kg}^{-1} \text{ DM}$ ), which was probably associated with the eggs used in the recipes. Muffins of type UB and FB showed the highest content of potassium, however, this finding was directly connected to the high content of this element in the buckwheat flour. Similar observations were made in relation to magnesium contents. In this case, the highest level was found in UB and FB muffins type, followed by UG and FG types, and C muffin. The highest content in UB and FB was closely associated with magnesium content in buckwheat flour, while the lowest one in C type with magnesium content in gluten-free maize flour. The higher magnesium content noted in buckwheat-based gluten-free muffins (UB and FB) is also of interest due to regulatory function of magnesium in distribution of calcium ions in cells. Magnesium ions are natural blockers of calcium channels and, in human cells, a strong competition between calcium and magnesium ions exists [15].

The recommended dietary reference intake of calcium for adults (19–50 years) is 1000 mg Ca per day [16]. Daily ingestion of an adequate amount of calcium is essential for cardiac regulation, blood clotting, muscle contraction, nerve transmission, bone and tooth formation. Approximately 2% of the recommended daily calcium intake could be covered by the inclusion of one muffin to the diet. Regarding potassium, the recommended dietary reference intake of this element for adults (19–50 years) is 4.7 g per day [16]. The experimental muffins UB and FB could provide 2% of the daily requirement for this element, while UG and FG only 1% of the daily requirement.

The significantly highest content of iron was found in control muffins compared to other samples. The higher content of iron in the muffins prepared only from buckwheat flour (UB and FB) compared with gluten-free maize muffins (UG and FG) was noticed. This was connected with high iron content in buckwheat flour. FB muffins had the significantly highest content of zinc compared to other samples. For both muffins, in which fermented buckwheat flour (FB and FG) was used in the recipe, the increase of zinc content was observed. In a review, RAES et al. [17] presented that fermentation had different effects on iron and zinc bioaccessibility, causing an increase of 6% and 40%, respectively, the effect being connected with the cereal/legume used or the type of fermenta-

**Tab. 2.** Ash and elements in experimental flours and muffins.

	Flour		Muffins				
	B	G	C	UB	FB	UG	FG
Ash [g·kg <sup>-1</sup> ]	7.7 ± 0.0 <sup>d</sup>	1.1 ± 0.0 <sup>e</sup>	11.9 ± 0.3 <sup>c</sup>	17.9 ± 0.3 <sup>a</sup>	18.3 ± 0.3 <sup>a</sup>	16.0 ± 0.3 <sup>b</sup>	16.1 ± 0.3 <sup>b</sup>
Macroelements [mg·kg <sup>-1</sup> ]							
Ca	236 ± 5 <sup>b</sup>	320 ± 1 <sup>b</sup>	590 ± 18 <sup>a</sup>	569 ± 40 <sup>a</sup>	582 ± 82 <sup>a</sup>	640 ± 35 <sup>a</sup>	555 ± 6 <sup>a</sup>
K	1796 ± 20 <sup>a</sup>	344 ± 22 <sup>d</sup>	975 ± 6 <sup>c</sup>	1769 ± 43 <sup>a</sup>	1824 ± 30 <sup>a</sup>	1328 ± 55 <sup>b</sup>	1350 ± 1 <sup>b</sup>
Mg	548 ± 1 <sup>a</sup>	78 ± 0 <sup>e</sup>	209 ± 0 <sup>d</sup>	519 ± 6 <sup>b</sup>	533 ± 28 <sup>ab</sup>	341 ± 1 <sup>c</sup>	341 ± 8 <sup>c</sup>
Microelements [mg·kg <sup>-1</sup> ]							
Fe	12.0 ± 0.1 <sup>d</sup>	5.8 ± 0.2 <sup>e</sup>	30.9 ± 1.6 <sup>a</sup>	23 ± 0.1 <sup>b</sup>	24.2 ± 2.4 <sup>b</sup>	19.9 ± 0.3 <sup>c</sup>	19.2 ± 0.3 <sup>c</sup>
Zn	16.8 ± 0.6 <sup>c</sup>	9.3 ± 1.9 <sup>d</sup>	21.5 ± 1.8 <sup>bc</sup>	27.9 ± 2.0 <sup>b</sup>	34.0 ± 3.7 <sup>a</sup>	22.4 ± 0.0 <sup>c</sup>	28.7 ± 1.7 <sup>b</sup>
Mn	5.9 ± 0.0 <sup>a</sup>	1.2 ± 0.2 <sup>d</sup>	0.6 ± 0.0 <sup>e</sup>	3.7 ± 0.0 <sup>b</sup>	3.7 ± 0.2 <sup>b</sup>	1.8 ± 0.2 <sup>c</sup>	1.8 ± 0.1 <sup>c</sup>
Cu	4.1 ± 1.4 <sup>a</sup>	1.4 ± 0.2 <sup>bc</sup>	1.2 ± 0.2 <sup>c</sup>	1.7 ± 0.0 <sup>b</sup>	1.3 ± 0.1 <sup>c</sup>	1.8 ± 0.0 <sup>b</sup>	0.7 ± 0.0 <sup>d</sup>

Data are expressed per kilogram of dry matter as mean ± standard deviation. Values followed by the same letter in the same row are not significantly different at 95% confidence level.

B – buckwheat flour, G – gluten-free maize flour, C – control muffins from unfermented gluten-free maize flour suspension with gluten-free maize flour, UB – muffins from unfermented buckwheat flour suspension with buckwheat flour, FB – muffins from fermented buckwheat flour suspension with buckwheat flour, UG – muffins from unfermented buckwheat flour suspension with gluten-free maize flour, FG – muffins from fermented buckwheat flour suspension with gluten-free maize flour.

tion performed. ABDELSEED et al. [18] observed an increase in iron extractability of sorghum lines after fermentation. One of the highly prevalent problems in the world, especially in the developing countries, is deficiency in iron and zinc and they, which are ranked at 9th and 11th position in the list of major risk factors for global burden of disease, respectively. Health and productivity of adults, and impairment of cognitive development in infants and young children, are known to be related to iron deficiency. Retarded skeletal development and immunodeficiency disorders are closely connected with zinc deficiency [19, 20].

Regarding iron, the recommended dietary reference intake for premenopausal, perimenopausal and postmenopausal women (aged 19–50 years) is 18 mg iron per day. Men of the same age should ingest 8 mg iron per day [16]. Approximately 5% of the recommended iron for women and 11% for men could be covered by one of the experimental muffins. Considering zinc, a component of multiple enzymes and proteins involved in the regulation of gene expression, the experimental muffins could provide 14% of the daily requirement for this element in women.

Manganese content, similar to potassium and magnesium, was the highest in UB and FB samples, which was strongly connected with Mn content in buckwheat flour (higher compared to gluten-free maize flour). On the other hand, similar dependence was not observed for copper. The high copper content in buckwheat flour had no influence on copper content in muffins pre-

pared only from buckwheat flour (UB and FB). The content of copper in all investigated muffins was almost at the same level. However, it should be noted that the applied fermentation process significantly decreased copper content in samples FB and FG compared to other muffins samples. SOKRAB et al. [21] showed significant increase of total and extractable zinc, manganese, copper and cobalt contents after fermentation of maize flour. In the case of manganese and copper, the experimental muffins could provide about 6% of the daily requirement for those elements.

## CONCLUSIONS

The levels of macro- and microelements in four types of innovative gluten-free muffins made from unfermented or fermented, by *Lactobacillus plantarum*, common buckwheat flour, comparing with muffin made from commercial gluten-free maize flour, were analysed in this study. Generally, buckwheat flour was a better source of elements in comparison to the gluten-free maize flour. The four types of innovative gluten-free muffins were characterized by higher contents of ash compared to control muffins. Muffins from unfermented and fermented buckwheat flour mixed with dry buckwheat flour showed the highest contents of macroelements, potassium and magnesium, and also higher contents of microelements, zinc and manganese. The applied fermentation process significantly decreased copper content in the samples

containing fermented buckwheat flour compared to other muffins samples.

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