

## THE INFLUENCE OF HONEY ENRICHMENT WITH BEE POLLEN OR BEE BREAD ON THE CONTENT OF SELECTED MINERAL COMPONENTS IN MULTIFLORAL HONEY

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### ABSTRACT

Bee products, such as honey, pollen, and bee bread, are an excellent source of bioactive ingredients, including minerals, having a health-supporting effect. However, due to the specific sensory properties of bee pollen and bee bread, the best way to include them in a diet is to add them to honey. Therefore, the aim of this paper was to evaluate the influence of the added bee pollen or bee bread on selected minerals content in multifloral honey. The mineral content was analyzed using absorption atomic spectrometry (FAAS) with prior dry mineralization. On the basis of obtained results, it was found that the addition of bee pollen or bee bread to honey significantly influences the content of selected macro- and microelements, excluding sodium. The greatest increase in mineral content was observed for magnesium, iron, and zinc. Enrichment of honey with the highest dose of bee pollen or bee bread resulted in an over 20-fold increase in the Mg and Fe content, and an over 14-fold increase in the Zn content. Honey enriched with the maximum addition of bee pollen was characterized by a higher content of K, Ca, Mg, Fe, and Cu compared to honey with bee bread. Due to a fact that both bee pollen and bee bread are good sources of minerals, their addition to honey significantly increases its ability to cover daily demand for macro- and microelements.

**Keywords:** honey; bee pollen; bee bread; mineral composition

### INTRODUCTION

Honey is a naturally sweet substance produced by honey bees *Apis mellifera* from plant nectar, secretions of live parts of plants or insect excretions. Honey has been valued for centuries, in different countries and cultures, due to a diversified range of its preventive and health-supporting properties (Bogdanov et al., 2008; Socha, Habryka and Juszcak, 2016; Socha, Habryka and Juszcak, 2018). Consumption of natural honey contributes to improved immunity and enriches diet in many valuable nutrients and bioactive substances. It is an excellent form of easily digestible sugars, vitamins, organic acids, and many other biologically active substances (Bogdanov et al., 2008; Wesolowska and Dżugan, 2017). Due to its highly diversified chemical composition, it is used as a treatment supporting agent (Bogdanov et al., 2008). Apart from direct consumption, it is used to sweeten food and drinks, baking, preserving food, and preparing of mead (Dżugan, Ruszel and Tomczyk, 2018).

Honey contains mineral ingredients deriving mainly from plant juice creating nectar, or from honeydew. The mineral ingredients content in honey depends on its variety and environmental conditions (Kačániová et al., 2009; Grembecka and Szefer, 2013). Differences in contents of bioelements between varieties, or even within the same

variety of honey result from their varying content in obtained plant material (Pohl, Sergiel and Stecka, 2009; Kędzierska-Matysek et al., 2013; Dżugan et al., 2017). Levels of these compounds are the lowest in light nectar honey, while they are the highest in honeydew honey (Gonzalez-Miret et al., 2005). Potassium is the main mineral ingredient found in honey, and its content corresponds to half of the total mineral content. Magnesium is found at a relatively stable level in honey, while the calcium content can vary significantly, depending on the honey origin (Dżugan et al., 2017). Phosphorus, iron, manganese, silicon, nickel, and sulfur are found in smaller quantities, while trace elements include copper, barium, cobalt, zinc, tin, palladium, aluminum, tungsten, chromium, titanium, molybdenum, vanadium, cadmium, and others (Solayman et al., 2016). Differences in macro- and microelements contents within the same honey variety results from the biodiversity of material, which contributes to a significant diversification of its mineral composition (Gonzalez-Miret et al., 2005; Kędzierska-Matysek et al., 2013). The number of mineral ingredients found in honey is correlated with species of plants from which the material is harvested. The botanical origin, in turn, is closely correlated with the location of an apiary, because soil composition and climatic conditions also determine the presence of metals in

melliferous plants. Environmental pollution and other anthropogenic processes should also be considered as an additional source of metals in honey, including Cu, Fe, and Zn (Kačaniová et al., 2009; Pohl, Sergiel and Stecka, 2009; Roman and Popiela, 2011; Solayman et al., 2016).

Apart from honey, bee products also include pollen, bee bread, propolis, royal jelly, bee venom, and wax. Pollen, depending on plants visited by bees, forms pollen loads of a color characteristic for a given plant species. Its composition is varied and depends on the origin and on weather conditions prevailing during anther forming and maturing (Gabriele et al., 2015; Kędzia and Holderna-Kędzia, 2016). Bee pollen is a diversified plant product rich in biologically active ingredients. Over 200 active substances were found in it (Gabriele et al., 2015; Kieliszek et al., 2018). Bee pollen is also rich in valuable bioelements essential for human health. They include macroelements such as phosphorus, potassium, magnesium, and calcium, microelements such as iron, manganese, zinc, and trace elements like copper, cobalt, and nickel (Kędzia and Holderna-Kędzia, 2016).

The bee bread is formed from plant pollen preserved by bees. Bees place flower pollen loads in honeycomb cells, wetting it with the secretion of their salivary glands and honey. Then bees pack it tightly in the cells to secure it against the access of air (Kieliszek et al., 2018; Bakour et al., 2019). In anaerobic conditions, this mixture of pollen, honey, and bee saliva is fermented by *Lactobacillus* bacteria, which contribute to the formation of antibacterial peptides, hydrogen peroxide, and organic acids. Enzymes found in bee saliva and bacteria themselves contribute to differences in the chemical composition of fresh pollen loads and bee bread (Kieliszek et al., 2018; Bakour et al., 2019). When compared to pollen, bee bread is characterized by higher nutritional value, better digestibility, and richer chemical composition (Socha, Habryka and Juszcak, 2016).

The literature review indicates that bee products such as bee pollen and bee bread are an excellent source of bioactive ingredients, and their addition to the diet may significantly enrich it with bioactive substances, including minerals, having health-supporting effects. Due to the specific sensory properties of bee pollen and bee bread, apparently, the best way to include them in a diet is to add them to honey (Kňazovická et al., 2011; Juszcak et al., 2015).

Therefore, the aim of this paper was to evaluate the influence of the added bee pollen or bee bread on selected minerals content in multifloral honey.

### Scientific hypothesis

Enrichment of honey with bee pollen or propolis at a sensoric acceptable level causes a significant increase in the content of mineral components.

## MATERIAL AND METHODOLOGY

### Materials

The multiflower honey (District Beekeeping Cooperative "Pszczelarz", Krakow, Poland) and the bee pollen and bee bread (Biopharmaceutical Laboratory „Aria”, Krakow, Poland) were used as experimental materials. Based on the preliminary sensory assessment, honey was enriched with bee pollen or bee bread in a quantity ranging from 5% to 25%. The use of the maximum pollen addition to honey, at a level of 25%, was supported by conducted preliminary sensory evaluation. The increasing levels of pollen or bee bread in honey changed the perception of its color, smell, texture, and palatability.

### Methods

Determination of the chosen mineral components content was made by atomic absorption spectrometry (ASA) in accordance with the requirements of the Polish Standard (PN-EN 14082, 2004). Mineralization of the tested sample was carried out at 600 °C in a muffle furnace (SNOL 8.2/1100, Lithuania) for 12 h. After mineralization, the ash content was calculated. The mineralized samples were dissolved in hot 6.0M hydrochloric acid and then in 0.5 M nitric acid. The chosen mineral components in honey were determined with an Avanta Sigma atomic absorption spectrometer (GBC, Australia), flame technique (FAAS), using acetylene/air or, for calcium, acetylene/nitrogen monoxide). The levels of macroelements including calcium, magnesium, sodium, and potassium as well as microelements: iron, zinc, copper, and manganese were determined. The quantitative analysis was carried out on a base of calibration curves made for standard solutions (Merck, Germany). The work parameters of the spectrometer are given in Table 1.

**Table 1** Spectrometer parameters used in the mineral composition analysis.

Element	Wavelength (nm)	Working range (µg.ml <sup>-1</sup> )	Equation of the calibration graph	R <sup>2</sup>
K	769.9	2.0 – 12.0	y = 8.947x + 0.017	0.9998
Na	589.6	0.2 – 1.5	y = 6.320x - 0.023	0.9983
Ca	422.7	1.0 – 4.0	y = 42.421x - 0.343	0.9905
Mg	202.6	0.5 – 1.5	y = 0.799x + 0.025	0.9872
Fe	248.3	0.2 – 0.4	y = 23.894x – 0.422	0.9948
Zn	213.9	1.0 – 0.8	y = 3.925x + 0.071	0.9981
Cu	324.7	0.1 – 1.5	y = 12.368x + 0.066	0.9995
Mn	279.8	0.2 – 0.4	y = 8.988x + 0.028	0.9973

**Statistical analysis**

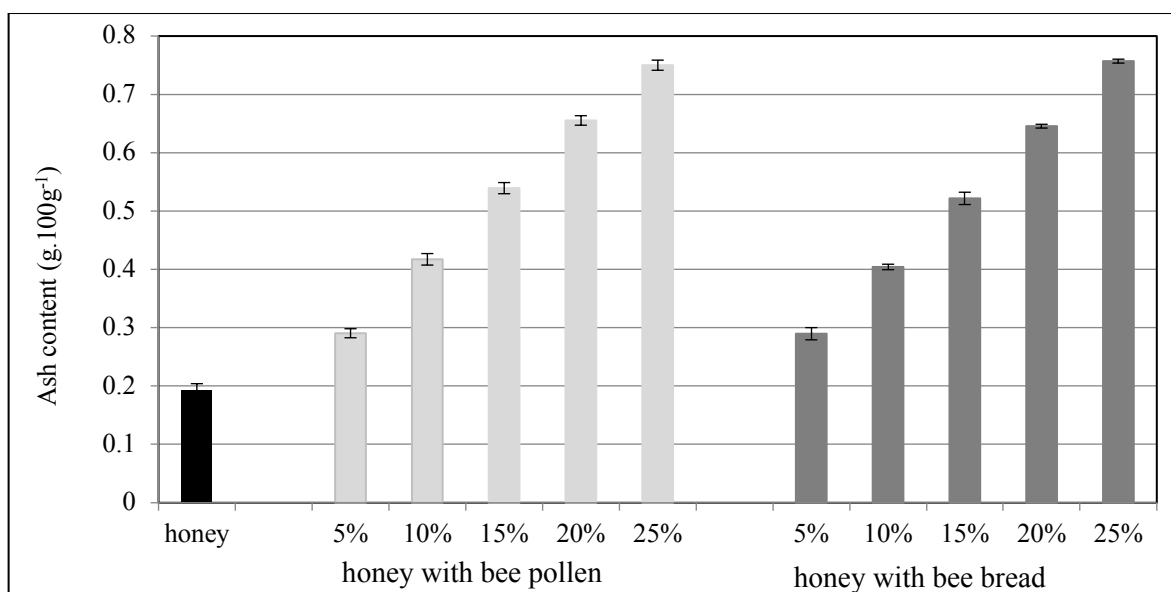
The analyses were made in triplicate, and the results were expressed as mean values ± standard deviations. In order to determine the significant differences between the means, the data were treated by one-way analysis of variance, and the Fisher test at a significance level  $\alpha = 0.05$  was calculated.

In order to assess the impact of both the type and amount of additive level (i.e., bee pollen or bee bread) to honey a two-way analysis of variance was performed. The values of Pearson's linear correlation coefficients between ash content and the content of individual mineral components were calculated, and their significance was verified by the Student's t-test at the significance level of 0.05. Calculations were performed with statistical software package Statistica 11.0 (StatSoft Inc., USA).

**RESULTS AND DISCUSSION**

The physical and chemical properties and nutritional value of honey are significantly influenced by its ash content, which also reflects the content of total and individual minerals. The ash content is a parameter depending on botanical origin, and it is also correlated with honey color (Gonzalez-Miret et al., 2005; Pohl, Sergiel and Stecka, 2009). The ash content in the studied multifloral honey and in honey enriched with bee pollen or bee bread is shown in Figure 1. The determined ash content was 0.19 g per 100 g of pure multifloral honey.

The obtained results are within the extensive range specified for Polish multifloral nectar honeys (Kędzierska-Matysek et al., 2013). The increasing addition of pollen or bee bread contributed to a significant ( $p < 0.05$ ) proportional increase in the honey ash content.



**Figure 1** Ash content of multifloral honey and samples enriched with pollen or bee bread.

**Table 1** Macroelements content in multifloral honey and samples enriched with pollen or beebread.

Type of ingredient	Amount of ingredient (%)	K (mg.100g <sup>-1</sup> ±SD)	Na (mg.100g <sup>-1</sup> ±SD)	Ca (mg.100g <sup>-1</sup> ±SD)	Mg (mg.100g <sup>-1</sup> ±SD)
honey bee pollen	0	271.4 ±3.8 <sup>j</sup>	8.1 ±0.1 <sup>a</sup>	13.9 ±0.4 <sup>j</sup>	2.3 ±0.2 <sup>j</sup>
	5	299.4 ±6.2 <sup>i</sup>	8.3 ±0.4 <sup>a</sup>	23.9 ±2.5 <sup>h</sup>	11.5 ±0.5 <sup>i</sup>
	10	372.7 ±4.8 <sup>g</sup>	7.8 ±0.9 <sup>a</sup>	31.4 ±1.6 <sup>g</sup>	21.4 ±0.4 <sup>g</sup>
	15	486.4 ±5.1 <sup>e</sup>	8.1 ±0.7 <sup>a</sup>	45.4 ±0.9 <sup>d</sup>	33.4 ±0.3 <sup>e</sup>
	20	548.1 ±10.8 <sup>c</sup>	8.1 ±0.5 <sup>a</sup>	53.0 ±1.2 <sup>b</sup>	41.2 ±0.7 <sup>c</sup>
	25	639.3 ±9.5 <sup>a</sup>	8.1 ±0.9 <sup>a</sup>	67.8 ±1.0 <sup>a</sup>	50.7 ±0.3 <sup>a</sup>
bee bread	5	350.8 ±3.0 <sup>h</sup>	7.6 ±0.7 <sup>a</sup>	21.5 ±0.4 <sup>i</sup>	13.1 ±0.4 <sup>h</sup>
	10	364.6 ±2.8 <sup>g</sup>	8.5 ±0.7 <sup>a</sup>	31.4 ±1.7 <sup>g</sup>	22.0 ±0.1 <sup>g</sup>
	15	419.5 ±7.2 <sup>f</sup>	8.0 ±0.4 <sup>a</sup>	34.2 ±0.2 <sup>f</sup>	29.4 ±0.2 <sup>f</sup>
	20	524.6 ±1.8 <sup>d</sup>	7.5 ±0.5 <sup>a</sup>	40.1 ±1.4 <sup>c</sup>	35.8 ±0.2 <sup>d</sup>
	25	617.8 ±3.3 <sup>b</sup>	8.0 ±0.2 <sup>a</sup>	50.1 ±2.6 <sup>c</sup>	46.2 ±0.5 <sup>b</sup>
<b>Two-way ANOVA - p</b>					
<b>Factor I (Type)</b>		$p < 0.05$	$p = 0.487$	$p < 0.05$	$p < 0.05$
<b>Factor II (Amount)</b>		$p < 0.05$	$p = 0.936$	$p < 0.05$	$p < 0.05$
<b>Factor I x Factor II</b>		$p < 0.05$	$p = 0.398$	$p < 0.05$	$p < 0.05$

Note: Mean values assigned with the same letters in particular columns are non-significant at the 0.05 level of confidence.

Table 1 list average contents of macroelements in multifloral honey and in honeys enriched with bee pollen or bee bread. In the plain multifloral honey, potassium was an element found at the highest level, and its content corresponded to over 90% of the total minerals determined. Its average content amounted to 271.4 mg per 100 g (Table 1), and this confirms earlier results for multifloral honeys (Grembecka and Szefer, 2013; Kędzierska-Matysek et al., 2013; Juszcak et al., 2018). The addition of pollen to honey significantly ( $p < 0.05$ ) increased its potassium levels. The greatest increase in potassium content, to 639.3 mg.100g<sup>-1</sup> was noted for 25% concentration of bee pollen in honey.

This observation confirms results obtained by Juszcak et al. (2018) for the commercial samples of honey with pollen added, who determined its potassium content at a level of 494 mg to 651 mg per 100 g. A significant ( $p < 0.05$ ) increase in potassium levels in honey enriched with pollen results from the high content of that ingredient in pollen. According to Kędzia and Holderna-Kędzia (2016), potassium content in pollen ranges from 284.3 mg to 2000 mg per 100 g. The potassium content in honey enriched with bee bread increases in a similar way, where a 5% addition already increased the content of that element to 350.8 mg.100g<sup>-1</sup> and a 25% addition increases that content to the level of 617.8 mg.100g<sup>-1</sup> (Table 1). Much higher average potassium content in commercial samples of honey enriched with bee bread was observed by Juszcak et al. (2018), and this resulted from a significantly higher addition of bee bread. The conducted two-way analysis of variance has shown that the potassium content in enriched honey significantly ( $p < 0.05$ ) depends both on the type and the level of the added ingredient. Furthermore, a significant positive linear correlation ( $r = 0.9823$ ) was found between ash and potassium content. A significant increase in potassium content following the addition of bee products to honey significantly influences its nutritional value. While 100 g of multifloral honey covers ca. 13% of the daily potassium demand, products enriched with pollen or bee bread cover over 39% of that demand.

The sodium content in the analyzed multifloral honey amounted to 8.1 mg.100g<sup>-1</sup>, representing ca. 2.7% of the total determined mineral ingredients (Table 1), and this confirms previous observations for Polish multifloral honey (Kędzierska-Matysek et al., 2013). The addition of bee products did not result in a significant ( $p > 0.05$ ) change in contents of this element, and this observation was confirmed by the two-way analysis of variance (Table 1). Studies conducted by Grembecka and Szefer (2013) show that the bee pollen sodium content amounts to 5.86 mg.100g<sup>-1</sup>, while Bakour et al. (2019) determined the sodium content in bee bread at a level of 14.2 mg.100g<sup>-1</sup>.

Thus, the literature data indicates that the sodium content of bee pollen and bee bread is similar to its content in honey, therefore, their addition does not significantly ( $p > 0.05$ ) influence the content of that macroelement.

The calcium content in the analyzed multifloral honey amounted to 13.9 mg.100g<sup>-1</sup>, representing ca. 4.7% of the total determined mineral ingredients (Table 1), and is within an extensive range reported for Polish multifloral honey (Grembecka and Szefer, 2013, Kędzierska-Matysek et al., 2013, Juszcak et al., 2018).

Increasing addition of bee pollen to honey increased the calcium content from 23.9 mg to 67.8 mg per 100 g of the product, while enrichment with bee bread increased the content of this element from 21.5 mg to 50.1 mg.100g<sup>-1</sup> (Table 1). The literature data indicate that the calcium content in pollen is much higher than in multifloral honey (Grembecka and Szefer, 2013), hence its significant ( $p < 0.05$ ) increase after the enrichment. The conducted two-way analysis of variance has shown that the calcium content in enriched honey significantly ( $p < 0.05$ ) depends both on the type and the level of the added substance. Furthermore, a significant positive linear correlation ( $r = 0.9625$ ) was found between ash and calcium content. Although honey is not a good source of calcium because at the determined content per 100 g (Table 1) it covers less than 2% of daily demand for this element, yet when honey is enriched with pollen or bee bread, the coverage of the daily calcium demand rises to ca. 6% to 8% per 100 g of the product.

Table 2 Microelements content in multifloral honey and samples enriched with pollen or bee bread.

Type of ingredient	Amount of ingredient (%)	Fe (mg.100g <sup>-1</sup> ±SD)	Zn (mg.100g <sup>-1</sup> ±SD)	Cu (mg.100g <sup>-1</sup> ±SD)	Mn (mg.100g <sup>-1</sup> ±SD)	
honey	0	0.34 ±0.03 <sup>k</sup>	0.19 ±0.00 <sup>i</sup>	0.10 ±0.00 <sup>i</sup>	1.20 ±0.01 <sup>k</sup>	
	bee pollen	5	1.29 ±0.05 <sup>j</sup>	0.53 ±0.01 <sup>h</sup>	0.12 ±0.01 <sup>h</sup>	1.29 ±0.02 <sup>j</sup>
		10	3.90 ±0.09 <sup>f</sup>	0.95 ±0.00 <sup>f</sup>	0.16 ±0.01 <sup>g</sup>	1.48 ±0.01 <sup>i</sup>
		15	4.55 ±0.17 <sup>e</sup>	1.67 ±0.01 <sup>d</sup>	0.28 ±0.02 <sup>de</sup>	1.91 ±0.00 <sup>h</sup>
		20	6.67 ±0.04 <sup>c</sup>	2.25 ±0.05 <sup>c</sup>	0.41 ±0.02 <sup>c</sup>	2.07 ±0.01 <sup>g</sup>
		25	9.67 ±0.17 <sup>a</sup>	2.73 ±0.02 <sup>b</sup>	0.57 ±0.04 <sup>a</sup>	2.35 ±0.01 <sup>e</sup>
bee bread	5	2.59 ±0.07 <sup>i</sup>	0.87 ±0.00 <sup>g</sup>	0.15 ±0.00 <sup>g</sup>	2.15 ±0.01 <sup>f</sup>	
	10	3.73 ±0.03 <sup>g</sup>	1.50 ±0.00 <sup>e</sup>	0.18 ±0.01 <sup>f</sup>	2.89 ±0.02 <sup>d</sup>	
	15	3.38 ±0.03 <sup>h</sup>	2.27 ±0.05 <sup>c</sup>	0.26 ±0.02 <sup>c</sup>	3.61 ±0.02 <sup>c</sup>	
	20	5.01 ±0.24 <sup>d</sup>	2.74 ±0.01 <sup>b</sup>	0.30 ±0.02 <sup>d</sup>	4.21 ±0.01 <sup>b</sup>	
	25	7.26 ±0.19 <sup>b</sup>	3.75 ±0.02 <sup>a</sup>	0.46 ±0.01 <sup>b</sup>	4.92 ±0.04 <sup>a</sup>	
Two-way ANOVA – p						
Factor I (Type)		$p < 0.05$	$p < 0.05$	$p < 0.05$	$p < 0.05$	
Factor II (Amount)		$p < 0.05$	$p < 0.05$	$p < 0.05$	$p < 0.05$	
Factor I x Factor II		$p < 0.05$	$p < 0.05$	$p < 0.05$	$p < 0.05$	

Note: Mean values assigned with the same letters in particular columns are non-significant at the 0.05 level of confidence.

In the analyzed multifloral honey, magnesium was at the level of 2.3 mg.100g<sup>-1</sup> (Table 1), and this corresponds to over 0.74% of the total mineral ingredients determined. The determined magnesium content was within the extensive range reported for Polish multifloral honeys (**Grembecka and Szefer, 2013; Kędzierska-Matysek et al., 2013; Juszcak et al., 2018**). The addition of bee pollen or bee bread significantly ( $p < 0.05$ ) influences an increase in the magnesium content, and this observation was confirmed by the results of the two-way analysis of variance (Table 1).

As reported by **Grembecka and Szefer (2013)**, floral pollen is richer in magnesium than honey, therefore, its addition contributed to a significant ( $p < 0.05$ ) increase in contents of that element to a level of 50.7 mg.100g<sup>-1</sup> in the enriched honey. In honey enriched with bee bread content of that element was not much lower (Table 1). Although honey is not a good source of magnesium because at the determined content per 100 g (Table 1) it covers only about 0.6% of daily demand for this element, yet when honey is enriched with pollen or bee bread, the coverage of the daily magnesium demand rises to ca. 12% to 13% per 100 g of the product.

Determinations also included contents of iron, zinc, copper, and manganese in multifloral and enriched honey. The average contents of these microelements are presented in Table 2. Iron is an element that in the studied honey represented 0.11% of all determined mineral compounds, and its content amounts to 0.34 mg.100g<sup>-1</sup> (Table 2). The obtained result was within the extensive range reported for Polish multifloral honey (**Grembecka and Szefer, 2013, Kędzierska-Matysek et al. 2013**). The addition of bee pollen to honey at a level of 25% contributed to the significantly ( $p < 0.05$ ) increase in the iron content to the level of 9.67 mg.100g<sup>-1</sup>, while the addition of the bee bread increased the iron content to the value of 7.26 mg.100g<sup>-1</sup>. Slightly lower values for the iron content in samples of commercial honey enriched with pollen or bee bread were reported by **Juszcak et al. (2018)**. According to **Grembecka and Szefer (2013)**, bee pollen is a rich source of iron, and its content ranges from 3.26 to 3.96 mg.100g<sup>-1</sup>. For this reason, honey enrichment with these bee products significantly ( $p < 0.05$ ) contributes to the increase in the level of that microelement. The conducted two-way analysis of variance has shown that the iron content in enriched honey significantly depends both on the type and the level of the added substance. Furthermore, a significant positive linear correlation ( $r = 0.9471$ ) was found between ash and iron content. A significant increase in iron content following the addition of bee products to honey significantly influences its nutritional value. While 100 g of multifloral honey covers ca. 2.5% of the daily iron demand, enriched honey cover as much as 69% of that demand for bee pollen and ca. 52% for bee bread.

In the analyzed honey, the zinc content was at the level of 0.19 mg.100g<sup>-1</sup> (Table 2), and this corresponds to over 0.06% of the total mineral ingredients determined. The obtained values are consistent with data reported by **Grembecka and Szefer (2013)** for Polish honey, and by **Kačaniová et al. (2009)** for Slovak honey. Other report indicate a slightly higher Zn content in Polish multifloral honey (**Kędzierska-Matysek et al., 2013; Juszcak et al., 2018**). With the rising addition of bee pollen, the Zn content in tested samples also significantly ( $p < 0.05$ ) increased, and

for the addition of 25% it reached 2.73 mg.100g<sup>-1</sup> (Table 2). The zinc content at the maximum bee bread concentration was 3.75 mg.100g<sup>-1</sup> (Table 2). The similar zinc content in commercial samples of honey enriched with pollen was reported by **Grembecka and Szefer (2013)** and **Juszcak et al. (2018)**. According to **Grembecka and Szefer (2013)**, bee pollen is a rich source of zinc, and its content in this substance amounts to 2.9 mg.100g<sup>-1</sup>. For the bee bread used as a honey ingredient, the increase of the zinc content was higher than in the case of pollen, and with the maximum supplementation, the content of that microelement reached 3.75 mg.100g<sup>-1</sup>. As the two-way analysis of variance demonstrated, both the type and the level of the additive have a significant ( $p < 0.05$ ) influence on zinc content in enriched honey. Furthermore, a strong positive linear correlation ( $r = 0.9690$ ) was found between ash and zinc content. An increase in zinc content following enrichment of honey with bee products significantly influences its nutritional value. While 100 g of multifloral honey covers ca. 2% of the daily zinc demand, enriched honey cover that demand in ca. 27% for pollen and ca. 37% for bee bread.

The copper content in both multifloral honey and honey enriched with bee products was the lowest of all mineral ingredients. In the multifloral honey, the copper content amounted to 0.10 mg.100g<sup>-1</sup> (Table 2), and this corresponds to just 0.03% of all elements determined. The obtained results are similar to values reported by **Kędzierska-Matysek et al. (2013)** for Polish honey, and by **Kačaniová et al. (2009)** for Slovak honey. As the conducted two-way analysis of variance has shown, both the type and the level of the additive have a significant ( $p < 0.05$ ) influence on the copper content in enriched honey (Table 2) and the addition of pollen or bee bread increases the content of the discussed element about five times. Furthermore, a strong positive linear correlation ( $r = 0.9603$ ) was found, indicating a relationship between ash and copper content. The average reference human demand for copper is low and amounts to 1 mg. While 100 g of multifloral honey covers ca. 10% of the daily copper demand, enriched honey cover that demand in ca. 57% for pollen and ca. 46% for bee bread.

In the analyzed honey, the manganese content was at the level of 1.2 mg.100g<sup>-1</sup> (Table 2), and this corresponds to 0.40% of the total mineral ingredients determined (**Grembecka and Szefer, 2013; Kędzierska-Matysek et al., 2013; Juszcak et al., 2018**). The determined value is within the extensive range reported for Polish multifloral honey. An addition of bee pollen or bee bread to honey caused a significant ( $p < 0.05$ ) increase in the manganese content. For bee pollen, the manganese content increased about two times, while for bee bread it increased four times. The conducted two-way analysis of variance has shown that the manganese content in enriched honey significantly ( $p < 0.05$ ) depends both on the type and the level of the bee product added. Honey can be considered a very good source of manganese, as at the determined level of this ingredient, 100 g of honey covers 60% of the daily demand. Honey enrichment with bee products, which increased the manganese content, also caused honey's ability to cover the daily demand for that element to a level of 117% for pollen and as much as 246% for bee bread.

## CONCLUSION

Based on obtained results it was found that the addition of pollen or bee bread to honey significantly influences the content of ash and selected macro- and microelements, excluding sodium. Significant positive linear correlations between the ash content and the content of some elements were observed. The greatest increase in mineral content was observed for magnesium, iron, and zinc. Enrichment of honey with the highest dose of bee pollen or bee bread resulted in an over 20-fold increase in the Mg and Fe content, and an over 14-fold increase in the Zn content. Honey enriched with the maximum addition of bee pollen was characterized by a higher content of K, Ca, Mg, Fe, and Cu compared to honey with bee bread. In turn, honey enriched with bee bread was characterized by a higher content of Zn and Mn. Due to a fact that both bee pollen and bee bread are good sources of minerals, their addition to honey significantly increases its ability to cover daily demand for macro- and microelements.

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## Acknowledgments:

The research was financed by the Ministry of Science and Higher Education of the Republic of Poland.

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