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Quality of durable cookies enriched with rape bee pollen

Kvalita trvanlivých sušienok obohatených o repkový peľ

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Abstract

The objective of this study was to enrich durable cookies with different additions of rape (*Brassica napus var. napus*) bee pollen to increase nutritional properties of cookie samples and to improve technological and sensorial properties as well. Bee pollen is an important raw material due to its nutritional and functional properties. Cookie samples were prepared by substituting wheat flour with rape bee pollen in the amount of 16 % (1 g of bee pollen per cookie) and 32 % (2 g of bee pollen per cookie) using bee pollen from two localities Lenártovce and Nové Zámky. In baked samples beside sensory properties also chemical parameters and technological parameters of cookies were evaluated. It was found out that with the gradual addition of rape bee pollen the amount of ash content increased and the highest ash content was analysed in variants II and IV (0.71 and 0.77 %) using 32 % addition of rape bee pollen. In terms of reducing sugars, addition of bee pollen caused that the content of reducing sugars in the products increased slightly. The highest reducing sugar content was determined in variant II. (24.59 %). On the other hand amount of crude protein the most considerably raised by addition of 2 g of pollen per cookie. The highest content of crude protein was analysed in variants II and IV (8.72 and 9.00 %). From the results of a linear models in which the dependent variables were the ash, crude protein and moisture it was determined the significant effect ($p < 0.05$) only of the pollen addition. In the case of the model with the dependent variable reducing sugars it was found out significant effect ($p < 0.0001$) of pollen addition and locality and their interactions. With the gradual addition of bee pollen values of technological parameters such as diameter and weight of cookies increased and thickness of products decreased. Based on sensory scores using a 9-point Hedonic scale the best sensorial acceptability (7.4) was found in variant I (1 g of bee pollen per cookie). Cookies were characterized by a pleasant, but little bit cabbage smell. Cookies were easy to chew and were of a sufficient delicate flavour with slight pollen and honey aftertaste. Taking into account all aspects of quality it can be concluded that the most acceptable addition of bee pollen was 16 % (1 g of bee pollen per cookie) from locality Lenártovce (south part of middle Slovakia).

Keywords: bee pollen, nutritional composition, durable cookies, sensory parameters, technological parameters

Abstrakt

Cieľom experimentu bolo obohatiť trvanlivé sušienky o rôzne prídavky repkového (*Brassica napus var . napus*) peľu za účelom zvýšenia nutričných a zlepšenia technologických a senzorických vlastností trvanlivých sušienok. Včelí peľ je dôležitou surovinou z hľadiska svojich nutričných a funkčných vlastností. Pokusné vzorky sušienok boli pripravené tak, že sa nahradila pšeničná múka repkovým peľom v množstve 16 % (t.j. 1 g peľu na sušienku) a v množstve 32 % (t. j. 2 g peľu na sušienku). Aplikovaný repkový peľ pochádzal z lokalít Lenártovce a Nové Zámky. Hotové varianty sušienok boli podrobené senzorickému hodnoteniu a zároveň boli hodnotené aj chemické parametre a technologické parametre sušienok. Výsledky ukázali, že s postupným prídavkom repkového peľu sa obsah minerálnych látok v sušienkach postupne zvyšoval a najvyšší obsah bol nameraný vo variantoch II a IV (0,71 a 0,77 %) s použitím prídavku 32 % repkového peľu. Obsah redukujúcich cukrov sa prídavkom repkového peľu mierne zvyšoval. Najvyšší obsah redukujúcich cukrov bol stanovený vo variante II (24,59 %). Na druhej strane množstvo dusíkatých látok sa značne zvýšilo prídavkom 2 g peľu na sušienku. Najvyšší obsah dusíkatých látok bol nameraný vo variantoch II a IV (8,72 a 9,00 %). Z výsledkov lineárnych modelov, kde závislými premennými boli obsah popola, hrubý proteín a vlhkosť bol zistený preukazný vplyv ($p < 0,05$) iba prídavku peľu. V prípade modelu so závislou premennou obsah redukujúcich cukrov sa potvrdil preukazný vplyv prídavku peľu aj lokality ($p < 0,0001$) a ich interakcie. S postupným prídavkom repkového peľu sa hodnoty technologických parametrov ako je priemer a hmotnosť sušienok zvýšili a hrúbka sušienok sa znížila. Na základe senzorického hodnotenia pomocou 9-bodovej hedonickej stupnice najlepšiu senzorickú prijateľnosť (7,4) preukázal variant I (1 g peľu na sušienku). Sušienky mali príjemnú, mierne kapustovitú vôňu. Boli ľahko žuteľné a vyznačovali sa jemnou chuťou s mierne peľovou a medovou dochuťou. Po zohľadnení všetkých aspektov kvality možno vyvodiť záver, že najprijateľnejší prídavok repkového peľu bol 16 % (1 g peľu na sušienku) pochádzajúceho z lokality Lenártovce.

Kľúčové slová: nutričné zloženie, senzorické a technologické parametre, trvanlivé sušienky, včelí peľ

Podrobný abstrakt

V poslednej dobe sa čoraz viac stretávame s problematikou hospodárskeho využívania obnôžkového peľu v rôznych odvetviach, ktorý sa radí medzi ochucovadlá. Obnôžkový peľ je zaujímavá surovina z nutričného hľadiska, pretože má pozitívne účinky na zdravie. Taktiež môže vylepšiť senzorické vlastnosti hotového výrobku. Cieľom experimentu bolo obohatiť trvanlivé sušienky o rôzne prídavky repkového (*Brassica napus var. napus*) peľu za účelom zvýšenia nutričných a zlepšenia technologických a senzorických vlastností trvanlivých sušienok. Pokusné vzorky sušienok boli pripravené tak, že sa nahradila pšeničná múka repkovým peľom v množstve 16 % (t. j. 1 g peľu na sušienku) a v množstve 32 % (t.j. 2 g peľu na sušienku). Aplikovaný repkový peľ pochádzal z lokalít Lenártovce a Nové Zámky. Hotové varianty sušienok boli podrobené senzorickému hodnoteniu a zároveň boli hodnotené aj chemické parametre a technologické parametre sušienok. Ukázalo sa, že náhrada pšeničnej múky obnôžkovým repkovým peľom má vplyv na kvalitatívne parametre výrobkov. Výsledky preukázali, že s postupným prídavkom repkového peľu sa obsah minerálnych látok v sušienkach postupne zvyšoval a najvyšší obsah bol nameraný vo variantoch II a IV (0,71 a 0,77 %) s použitím prídavku 32 % repkového peľu. Obsah redukujúcich cukrov sa prídavkom repkového peľu mierne zvyšoval. Najvyšší obsah redukujúcich cukrov bol stanovený vo variante II (24,59 %). Na druhej strane množstvo dusíkatých látok sa značne zvýšilo prídavkom 2 g peľu na sušienku. Najvyšší obsah dusíkatých látok bol nameraný vo variantoch II a IV (8,72 a 9,00 %). Prídavok peľu ovplyvnil aj hodnoty antiradikálovej aktivity. Vzorky s vyšším obsahom peľu mali inhibíciu 54,44 a 66,91 %. Rozdiely boli zistené aj pri aplikácii peľu pochádzajúceho z dvoch odlišných pestovateľských lokalít. Vzorky peľu z lokality Lenártovce vykazovali vyššie hodnoty inhibíce (39,34 a 66,91 %) ako vzorky peľu pochádzajúceho z lokality Nové Zámky (32,66 a 54,44 %). S postupným prídavkom repkového peľu sa hodnoty technologických parametrov ako je priemer a hmotnosť sušienok zvýšili a hrúbka sušienok sa znížila, pričom najvyššia hrúbka bola zistená pri kontrolných sušienkach (5,9 mm). Najväčší priemer sušienok bol nameraný vo variante I (53,2 mm) a najvyššia hmotnosť sušienok bola zistená pri variante II (10,82 g). V porovnaní s kontrolou najvýraznejšie sa zvýšila hmotnosť sušienok vo variantoch I a II (10,51-10,82 g) aplikovaním peľu pochádzajúceho z lokality Lenártovce. Hmotnosť sušienok vyrobených z peľu z lokality Nové Zámky bola nižšia (10,27-10,45 g). Na základe senzorického hodnotenia pomocou 9-bodovej hedonickej stupnice najlepšiu senzorickú prijateľnosť (7,4) preukázal variant I (1 g peľu na sušienku). Sušienky mali príjemnú, mierne kapustovitú vôňu. Boli ľahko žuteľné a vyznačovali sa jemnou chuťou s mierne peľovou a medovou dochuťou. Vyšší prídavok repkového peľu (viac ako 2 g na sušienku) pôsobil vo výrobkoch nepriaznivo, pretože zanechával výraznú peľovú až štipľavú chuť a dochuť, ktorá zhoršovala celkový dojem výrobkov. Po zohľadnení všetkých aspektov kvality možno vyvodiť záver, že najprijateľnejší prídavok repkového peľu bol 16 % (1 g peľu na sušienku) pochádzajúceho z lokality Lenártovce.

Introduction

Bee pollen is valuable natural product in terms of chemical composition and content of biologically active substances. The dry matter of pollen contains about 60 % protein, 15 % carbohydrates. Moreover there are also minerals, enzymes, vitamins, nucleic acids, nucleosides, nucleotides, organic acids, colorants (Kačániová, et al., 2010). Pollen is the source of easily digestible protein and essential amino acids (Naumkin, 1984; Čeksterite, 1988, 1991; Campos, et al., 1996). High concentration of essential amino acids is nutritionally very important factor. The content of almost all amino acids in 100 g of mixed bee pollen exceeds the recommended daily minimum (Košlík, 1995). The most important pollen carbohydrates are glucose, dextrin, starch, cellulose, as well as fructose, ribose, pentose and pollenin (Kačániová, et al., 2010). Pollen contains 10 % lipids, sterols, and fatty acids (Veselý, et al., 2003). Fats in bee pollen in 43 % consists of the three most valuable fatty acids - linolenic, linoleic and arachidonic (Košlík, 1995). Pollen contains also minerals which represents app. 3 % of the pollen dry matter. Parts of the pollen are also acids such as malic, tartaric, citric, raspberry, fumaric etc. Further therein a nucleic acid, purine substances, essential oils, flavonoids. Pollen is a rich source of plant metabolites such as flavonoids and other phenolic compounds (Balch and Balch, 1990; Broadhurts, 1999; Silva, et al., 2000; Fatrcová-Šramková, et al., 2010). These

metabolites have anti-inflammatory and antioxidant properties (Kroyer and Hegedus, 2001; Campos, et al., 2003; Gallmann, et al., 2005; Le Blanc, et al. 2009). Pollen also contains triterpenic acids which have anti-inflammatory, stimulating, tonic effect. Pollen extract contains 5-10 flavonoids and many other substances such as carotenoids, growth promoters, agents having an antibiotic effect (Kačániová, et al., 2010). Bee pollen has beneficial health effects. It can be used in the beekeeping in the pharmaceutical industry, in cosmetics, but also in food. Pollen is mainly used as a nutritional supplement for humans in the form of granules, cereal bars, sweets, tablets, etc. (Chlebo and Čermáková, 2001). Bee pollen can also improve the organoleptic properties of the products, giving it a specific flavour and colour.
The bakery industry is one of the largest organized food industries all over the world and in particular biscuits, cookies and cakes are one of the most popular products because of their convenience, ready to eat nature, and long shelf life (Marroquin, et al., 1985).

The objective of the present study was to substitute wheat flour with rape (*Brassica napus var. napus*) bee pollen from two localities in the amount of 16 % and 32 % by preparation of durable cookies and to study the effect of pollen addition on cookies nutritional, technological and sensorial properties as compared to wheat flour cookies.

Material and methods

Material

The ingredients used in cookie production were: wheat flour type 00 extra, vegetable fat, egg yolk, powder sugar and rape bee pollen. Basic ingredients were purchased from local supermarkets.

Rape bee pollen was obtained from two localities: locality Lenártovce (south part of middle Slovakia) and Nové Zámky (south part of west Slovakia) directly from beekeepers in the vegetation year 2012. It was frozen in -18 °C immediately after harvest until analyses were realized. The bee pollen samples were monofloral. The sample from locality Lenártovce contained 9.3 % of pollen pellets from other plant species, and the sample from locality Nové Zámky contained 2.9 % of other pellets. Even our pollen samples were contaminated by pellets from other plant species we can still characterize them as monofloral. The contamination was less than 10 % and according to literature sources (Campos, et al., 2008) it is considered as monofloral bee pollen. The pollen samples were also contaminated by different types of impurities such as – parts of plant, pieces of bee body, soil, sand, etc. All impurities were removed from analysed samples.

For cookies production was used dried and processed bee pollen. Homogenized pollen was dried at the temperature 35 °C until 5 to 10 % moisture. Then it was grinded and sieved to get powdered material.

Methods

Cookie preparation: cookie samples were prepared by substituting wheat flour with rape bee pollen in the amount of 16 % (that represent 1 g of bee pollen per cookie) and 32 % (2 g of bee pollen per cookie) using bee pollen from two localities Nové Zámky and Lenártovce (Table 1). We tried to keep recommended daily consumption dose of bee pollen which represents the amount of app. 10 g of bee pollen (Bogdanov, 2012) and we assumed intake of around 5 -10 cookies per day.

Four variants of cookies were prepared. A control sample was also prepared: 125 g wheat flour type 00 extra, 75 g vegetable fat, 45 g powder sugar, 1 egg yolk. Wheat flour, rape bee pollen, powder sugar, vegetable fat and egg yolk were mixed until the development of dough using a laboratory type mixer for 5 min low speed. The resulting dough was manually shaped into circular form (diameter: 50 mm, thickness: 5 mm). The cookies were baked in automatically controlled electric oven at 180 °C for 12 min. The cookies were cooled down to room temperature app. 20 °C.

Table 1. Variants of cookies with added bee pollen from *B. napus* var. *napus*

Tabuľka 1. Varianty sušienok obohatené o repkový peľ *B. napus* var. *napus*

Variants	Pollen addition (%)	Locality
Control	0	-
Variant I	16	Lenártovce
Variant II	32	Lenártovce
Variant III	16	Nové Zámky
Variant IV	32	Nové Zámky

Antiradical activity (DPPH) of durable cookies: antiradical activity was realized according to methodology of Band-Williams, et al. (1995) and Kutlu, et al. (2011) on spectrophotometer Genesys 20 Model 4001/4. The cookies from each variant were blended and frozen. For extraction of 1 mg blended sample we added 25 ml of methanol p.a. 99.7 %. The prepared solution was shook for 12 hours. Extracts were filtered and mixed with prepared working solution. The basic solution was prepared from – 25 mg DPPH (2.2-difenil-1-pikrilhydrazil), 100 ml methanol. The working solution contained – 10 ml of basic solution and 90 ml of methanol. The absorption was measured from solution prepared from 3.9 ml of working solution and 0.1 ml of filtered sample. We measured absorption at t = 0 minute and t = 10 minutes according to following formula (1).

(1)

$$inhibition\ (\%) = \frac{(A_C - A_{AT})}{A_c} \times 100$$

Notes:
Inhibition of DPPH radicals (%)
A_C – absorption of control at t = 0 min
A_{AT} – absorption of sample at t = 10 min

Chemical analysis: the cookie samples were analysed for their crude protein by the Kjeldahl method AACC-Method 46-13 (1986), for ash content by AACC-Method 08-01 (1983), for reduction sugars according to Schoorl by AACC-Method 80-68.01 (1999), for moisture by ICC-Standard 110/1 (1976).

Technological parameters analysis: the cookie samples were analysed for their diameter (mm), thickness (mm) and weight (g).

Sensory parameters analysis: the cookie samples were sensory analysed for their colour, intensity of colour, odour, intensity of bee pollen odour, shape, crispiness/crumbliness, taste, intensity of bee pollen taste, after taste and the overall impression. Sensory scores were evaluated using a 9-point Hedonic scale.

Experimental data were evaluated by using descriptive statistics, Pearson correlation coefficient and two-way analysis of variance (ANOVA) - linear model and significant differences among means at $P < 0.05$ were determined by Kruskal-Wallis' nonparametric test, using SAS Enterprise Guide 5.1.

Results and discussion

Results regarding chemical parameters showed that with the gradual addition of bee pollen values of crude protein in all variants of cookies increased and the most significantly by 32 % addition (2 g of pollen per cookie) in comparison with the control except variant III (7.18 %), (Table 2). The highest amount of crude protein (9.0 %) was measured in variant IV with 32 % addition of rape bee pollen from locality Nové Zámky and the lowest amount of crude protein (7.18%) was measured in variant III locality Nové Zámky and in control sample (Table 2). According to Dobrovoda (1986) fresh bee pollen contains about 24.06% protein. Pollen is generally low in fat and rich in minerals and vitamins in comparison with many traditional foods for people and is rich in protein (Schmidt, 1996). Ash content ranged from 0.46 to 0.77 % (Table 2). With gradual addition of rape bee pollen amount of ash content increased. The most significantly increased in cookies with higher addition 32 % of bee pollen (2 g of pollen per product). The highest values of ash were recorded in variants II and IV (0.71 and 0.77 %), (Table 2). The lowest ash content was measured in control cookies (0.46 %). Results revealed that even locality influenced values of ash in cookies. Cookies made with addition of rape bee pollen from locality Nové Zámky had higher ash content in comparison with the cookies prepared using rape bee pollen form locality Lenártovce (Table 2). According to Schmidt and Buchmann (1992) the presence of ash in the fresh pollen ranges from 1 to 5 %. It was found out that content of reducing sugars in cookies ranged from 17.11 to 24.59 % (Table 2). The highest content of reducing sugars was measured in variant II (24.59 %) with 32 % addition of rape bee pollen from locality Lenártovce (2 g of pollen per cookie). With gradual addition of rape bee pollen reducing sugar content in cookies samples only slightly increased except variant I (17.11 %), where there was measured the lowest content of reducing sugars (Table 2). There weren't noticeable differences in values of reducing sugars between the localities. According to Dobrovoda (1986) carbohydrates represent in bee pollen 18.5 %. Moreover it was found out that the moisture content with gradual addition of bee pollen increased. The lowest moisture content was measured in control cookies (5.20 %), (Table 2). On the other hand the highest moisture content was measured in variants II and IV with the higher 32 % pollen addition (2 g of pollen per cookie). We used the Kruskal-Wallis nonparametric test to find out significant differences, as we do not confirmed normality of evaluated files. Based on the results of the statistical analysis, we confirmed significant differences (p <0.05) between individual variants (control, VAR 1, VAR 2, VAR 3, VAR 4) in the monitored chemical parameters (ash, reducing sugars, crude protein and moisture). Using two-factor analysis of variance (ANOVA), the significance of effect of two main factors - locality and the addition of pollen (independent variable) and their interactions on evaluated technological and chemical parameters of cookies was investigated. From the results of a linear models in which the dependent variable were the ash, crude protein and moisture we determined the significant effect (p <0.05) only of the pollen addition. In the case of the model with the dependent variable reducing sugars it was found out significant influence of both main effects and their interactions (Table 3).

Table 2. Physical-chemical parameters of cookies with added bee pollen from *B. napus* var. *napus*

Tabuľka 2. Fyzikálno-chemické parametre sušienok s prídavkom repkového peľu *B. napus* var. *napus*

Variants	Ash (%)		Reducing sugars (%)		Crude protein (%)		Moisture (%)	
Control 0% addition	0.46	±0.0688	20.45	±0.2466	7.27	±0.0788	5.20	+0.0435
Variant I 16% addition	0.54	±0.0404	17.11	±0.1329	7.52	±0.1238	7.50	+0.0950
Variant II 32% addition	0.71	±0.0202	24.59	±0.3251	8.72	±0.2591	8.10	+0.0417
Variant III 16% addition	0.65	±0.0338	21.57	±0.1822	7.18	±0.1299	7.60	+0.1484
Variant IV 32% addition	0.77	±0.0497	21.43	±0.0680	9.00	±0.0665	8.30	+0.1877

Legend: variant I – locality Lenártovce, variant II – locality Lenártovce, variant III – locality Nové Zámky, variant IV – locality Nové Zámky. Values are mean±standard error

Table 3. The summary tables for ANOVA – dependent variable reducing sugars

Tabuľka 3. Súhrnné tabuľky ANOVA – závislá premenná redukujúce cukry

Source	DF	Sum of Squares	Mean Square	F Value	Pr > F
Model	3	85.13	28.37	234.64	<.0001
Error	8	0.96	0.12		
Corrected Total	11	86.10			

R-Square	Coeff Var	Root MSE	Reducing Sugars Mean
0.988763	1.64	0.34	21.17

Source	DF	Type III SS	Mean Square	F Value	Pr > F
Locality	1	1.25	1.25	10.37	0.0122
Pollen addition	1	40.40	40.40	334.10	<.0001
Locality*Pollen addition	1	43.47	43.47	359.45	<.0001

Since it was found out a significant interaction effect of independent variables on reducing sugars content of cookies the following tables present the simple effects analysis (Table 4).

Table 4. The pairwise comparisons comprising the simple effects analysis – dependent variable reducing sugars

Tabuľka 4. Párové porovnanie jednofaktorovej analýzy - závislá premenná redukujúce cukry

Locality	Pollen addition	Red. Sug. LSMEAN	LSMEAN Number
Lenártovce	P16	17.11	1
Lenártovce	P32	24.59	2
Nové Zámky	P16	21.57	3
Nové Zámky	P32	21.43	4

Least Squares Means for effect locality*pollen addition
Pr > |t| for H0: LSMean(i)=LSMean(j)

i/j	1	2	3	4
1		<.0001	<.0001	<.0001
2	<.0001		<.0001	<.0001
3	<.0001	<.0001		0.9611
4	<.0001	<.0001	0.9611	

These results (Table 4) show that, compared means of cookies reducing sugars content with the pollen addition 16% from locality Lenártovce (mean of 17.11) and from locality Nové Zámky (mean of 21.57) are significantly different. It was also confirmed the significant difference between means with the pollen addition of 32% between locality Lenártovce (mean of 24.59) and locality Nové Zámky (mean of 21.43). It can be assumed that pollen from different localities can affect the content of reducing sugars in cookies. Regarding locality Lenártovce it was confirmed a significant difference between means with different pollen additions. So we can assume that the higher addition of pollen from locality Lenártovce increased the content of reducing sugars.

Antioxidant capacity

Many authors analysed antioxidant capacity of different bee pollens, from different regions, and they used different methodologies (Carpes, et al., 2009; LeBlanc, et al., 2009; Graikou, et al., 2011; Almaraz-Abarca, et al., 2004). But we did not find any literature sources about the processed products with added bee pollen. We analysed prepared cookies by DPPH method, and results were compared between samples. In the table 5 are presented the results of antiradical activity of analysed samples of cookies with addition of different amounts of rape bee pollen. In comparison with control sample (pollen addition 0 %) is visible that the inhibition increased rapidly in case of variants with added bee pollen. Also the different amount of added pollen (16 or 32 %) influenced the antiradical activity. The samples with higher amount of pollen have inhibition 54.44 and 66.91 % (Table 5). The results from realized analyses show also visible difference between localities. Samples from locality Lenártovce showed higher inhibition (39.34 and 66.91 %) than samples from locality Nové Zámky (32.66 and 54.44 %), (Table 5). It would be interesting to analyse in details statistical significance of these differences.

Table 5. Antiradical activity of cookies with added bee pollen from *B. napus* var. *napus*

Tabuľka 5. Antiradikálová aktivita sušienok s prídavkom repkového peľu *B. napus* var. *napus*

Variants	Pollen addition (%)	Locality	Mean	SD	Confidence intervals
Control	0	-	6.16	0.41	5.15 – 7.17
Variant I	16	Lenártovce	39.34	0.94	35.60 – 43.09
Variant III	16	Nové Zámky	32.66	3.92	30.69 – 34.63
Variant II	32	Lenártovce	66.91	2.85	63.39 – 70.42
Variant IV	32	Nové Zámky	54.44	1.62	49.42 – 59.47

Legend: Mean – average value of three repetitions; SD – standard deviation

From technological parameters it was measured thickness, weight and diameter of cookies. Results regarding technological properties of cookies showed that with higher addition of bee pollen 32 %, cookies diameter decreased, in comparison with lower 16 % addition of bee pollen (Table 6). Variant I and III had larger diameter in comparison to control cookies (Table 6). The highest diameter was measured in variant I (53.2 mm). Regarding localities cookies made from bee pollen form locality Nové Zámky had small diameter in comparison to cookies prepared from bee pollen from Lenártovce. Even thickness of cookies was influenced. The highest thickness was measured in control sample (5.9 mm) and with gradual addition of rape bee pollen thickness of cookies decreased. There weren't noticeable differences among localities (Table 6). On the other hand weight of cookies with gradual addition of rape bee pollen increased. The lowest weight was measured in control cookies (10.19 g), (Table 6). In comparison with control the most considerably increased weight of cookies in variant I and II (10.51 - 10.82 g), bee pollen from locality Lenártovce.

Weight of cookies made from bee pollen from locality Nové Zámky was lower (10.27 - 10.45 g), (Table 6). When assessing the technological parameters (diameter, thickness and weight of cookies), it was confirmed highly significant differences ($p < 0.0001$) between individual variants (control, VAR 1, VAR 2, VAR 3, VAR 4). In the case of analysed technological parameters of cookies (diameter, thickness and weight of cookies) it was confirmed the statistical significance of correlation relations using the Pearson correlation coefficient. We found out that in the case of control (zero addition of pollen) there were not any statistically significant relations ($p > 0.05$) among technological parameters. In datasets with added amount of pollen (16% and 32%) there were confirmed statistically significant relations ($p < 0.05$) among evaluated parameters. It can be assumed that the addition of pollen can positively or negatively affect the technological parameters of cookies (diameter, thickness and weight of cookies).

Table 6. Technological parameters of cookies with added bee pollen from *B. napus* var. *napus*

Tabuľka 6. Technologické parametre sušienok s prídavkom repkového peľu *B. napus* var. *napus*

Variants	Diameter (mm)		Thickness (mm)		Weight (g)	
Control 0% addition	52.1	±0.0216	5.9	±0.0286	10.19	±0.0035
Variant I 16% addition	53.2	±0.0299	4.7	±0.0357	10.51	±0.0031
Variant II 32% addition	52.4	±0.0328	5.0	±0.0234	10.82	±0.0026
Variant III 16% addition	52.8	±0.0244	4.9	±0.0212	10.45	±0.0021
Variant IV 32% addition	51.4	±0.0331	4.9	±0.0378	10.27	±0.0033

Legend: variant I – locality Lenártovce, variant II – locality Lenártovce, variant III – locality Nové Zámky, variant IV – loc. Nové Zámky. Values are mean±standard error

Moreover, regarding technological parameters (dependent variable - diameter, thickness, weight), it was confirmed a statistically significant effect ($p < 0.05$) of locality, of pollen addition and their interactions. Whereas in linear models where the dependent variables were the diameter and weight of cookies, the impact of interaction (locality and pollen addition) was statistically significant, we implemented the simple effects analysis (Table 7, 8).

Table 7. The pairwise comparisons comprising the simple effects analysis – dependent variable diameter

Tabuľka 7. Testovanie kontrastov pomocou Tukeyho testu - závislá premenná priemer

Locality	Pollen addition	Diameter LSMEAN	LSMEAN Number
Lenártovce	P16	53.20	1
Lenártovce	P32	52.35	2
Nové Zámky	P16	52.76	3
Nové Zámky	P32	51.41	4

Least Squares Means for effect locality*pollen addition
Pr > |t| for H0: LSMean(i)=LSMean(j)

i/j	1	2	3	4
1		<.0001	<.0001	<.0001
2	<.0001		<.0001	<.0001
3	<.0001	<.0001		<.0001
4	<.0001	<.0001	<.0001	

Table 8. The pairwise comparisons comprising the simple effects analysis – dependent variable weight

Tabuľka 8. Testovanie kontrastov pomocou Tukeyho testu - závislá premenná hmotnosť

Locality	Pollen addition	Weight LSMEAN	LSMEAN Number
Lenártovce	P16	10.51	1
Lenártovce	P32	10.81	2
Nové Zámky	P16	10.45	3
Nové Zámky	P32	10.26	4

Least Squares Means for effect locality*pollen addition
Pr > |t| for H0: LSMean(i)=LSMean(j)

i/j	1	2	3	4
1		<.0001	<.0001	<.0001
2	<.0001		<.0001	<.0001
3	<.0001	<.0001		<.0001
4	<.0001	<.0001	<.0001	

Tables 7, 8 show that the differences between means regarding each pair were all significant. Based on the gained results it was found out that the average values of the cookies diameter and weight with the pollen addition from locality Lenártovce were slightly higher than the average values of the cookies with the pollen addition from locality Nové Zámky. According to this finding, we can assume that pollen from different localities can affect some technological parameters of cookies. On the other hand, we found that higher addition of pollen slightly decreased average values of cookies diameter and weight.

Bee pollen distinguishes from each other in colour, taste, nutritional composition and other properties (Stoklasa, 1975, Přídal, 2003, Titěra, 2006). Therefore for instances colour of product depends on the chemical composition of bee pollen (Synytsya, et al., 2009). From a sensory point of view, based on sensory scores using a 9-point Hedonic scale the best overall sensorial acceptability showed variant I (7.4), where there was used 16 % addition of rape bee pollen (1g of pollen per cookie), (Figure 1). Cookies were characterized by a pleasant smell, shape of the products was symmetric, they were easy to chew and were enough crispy. The intensity of pollen odour was slightly cabbage but not disturbing. The intensity of pollen taste was slightly after pollen. Taste of cookies was slightly honey with a slight pollen aftertaste (Table 9, Figure 1).

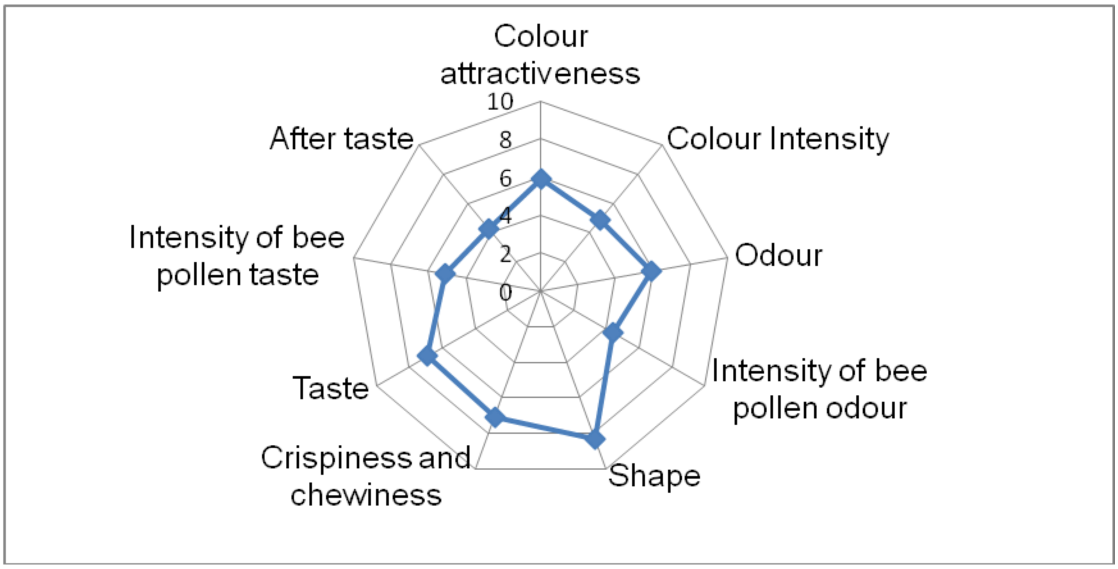

Figure 1. Sensorial profile of cookies made with 16 % addition of bee pollen from *B. napus* var. *napus* – Variant I

Graf 1. Senzorické parametre sušienok s prídavkom repkového peľu *B. napus* var. *napus* v množstve 16 % – Variant I

The worst overall sensory acceptability was found in cookies made with higher 32 % addition of rape bee pollen (variant II and IV). Higher pollen addition caused that the odour and taste of products was deteriorated with prevailing bitter and spicy taste with cabbage odour. The colour of cookies was also negatively influenced with the higher addition of pollen because products had a strong yellow-green colour (Table 9). The shape of cookies with addition of bee pollen was not influenced, cookies kept symmetric shape. Bee pollen addition improved crispiness of cookies. In terms of localities, better sensory properties was found in cookies where there was used bee pollen from locality Lenártovce because pollen from locality Nové Zámky had strongly cabbage taste leaving bitterness and spiciness on the tongue. Cookies containing pollen from locality Lenártovce were sensorial more acceptable.

Table 9. Sensorial attributes of cookies with added bee pollen from *B. napus* var. *napus*

Tabuľka 9. Senzorické parametre sušienok s prídavkom repkového peľu *B. napus* var. *napus*

Attribute	Control 0% addition	Variant I 16% addition	Variant II 32% addition	Variant III 16% addition	Variant IV 32% addition
Colour attractiveness	5.0	5.9	5.9	6.1	6.1
Colour intensity	3.7	4.9	5.7	5.4	5.2
Odour	7.0	5.9	5.3	6.4	6.4
Intensity of bee pollen odour	-	4.4	6.4	5.0	7.1
Shape	8.4	8.3	7.8	7.8	8.1
Crispiness/chewiness	6.5	7.1	7.1	7.4	7.6
Taste	7.6	6.9	4.9	5.7	3.9
Intensity of bee pollen taste	-	5.1	6.8	5.3	7.3
After taste	-	4.3	6.7	4.8	8.4
Overall impression	7.3	7.4	5.4	6.2	4.0

Legend: variant I – locality Lenártovce, variant II – locality Lenártovce, variant III – locality Nové Zámky, variant IV – locality Nové Zámky.

Conclusion

Whereas, relatively little bee pollen is used directly for food today, it has great potential to be used in food products mainly due to its high nutritional value. It can be concluded from the results of this study and suggested that substitute wheat flour with rape bee pollen at maximum 16 % (1 g per cookie) level could be suitable for further food applications with better nutritional, technological and sensorial properties. Important for application is moreover the choice of locality where bee pollen comes from.

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