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THE EVALUATION OF BEE BREAD QUALITY

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Annotation. The aim of this study was to evaluate bee bread quality. Bee bread is product with long history; it was used by many civilizations for centuries. But information and experimental results about bee bread is still missing nowadays. From this reason, in this work is presented model about evaluation of basic quality indicators of bee bread.

For important indicators of bee bread quality is considered – locality of bee bread harvesting, harvesting period, technology of harvesting, storing of bee bread, plant origin, morphology characteristic, chemical composition, content of heavy metal, residues of pesticide content, presence of microorganisms and others. These indicators are very important for determining of complex bee bread quality and also give information about economic value and practical using of bee bread. But nowadays many scientific works are oriented in one direction for evaluating only one indicator. In this work we presented our results and also results from scientific literature about bee bread.

Key words: quality indicators, characteristic, pollen, bee bread.

Introduction. The term "bee bread" is reserved for the original bee pollen stored in the combs. The bee bread has already been processed by the bees for storage with the addition of various enzymes and honey, which subsequently ferments. This type of lactic acid fermentation is similar to that in yoghurts (and other fermented milk products) and renders the end product more digestible and enriched with new nutrients. One advantage is almost unlimited storability of beebread in comparison with dried or frozen pollen in which nutritional values are rapidly lost. The natural process carried out by the bees can more or less be repeated artificially with dry or fresh bee-collected pollen. It is important however, to provide the correct conditions during the fermentation process (Krell, 1996).

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According to Zuluaga, et al. (2015) "bee bread" is a product of the hive obtained from pollen collected by bees, to which they added honey and digestive enzymes and subsequently stored in the combs, starting a lactic fermentation which gives it greater power conservation. A proper hive management promotes bee-bread collection, aimed at marketing it for human consumption since it can be considered as food supplement due to its content of a wide range of nutrients. One of the contributions to their high nutritional value is the presence of significant amounts of proteins, vitamins and phenolic compounds as natural antioxidants. The potential application of "bee bread" as a food and as a nutraceutical supplement depends in large part on its chemical composition which varies directly with the flora of the region and the time of collection by the bees.

The process of bee bread formation starts with gathering of pollen, then a bee mixes it with flower nectar or honey and saliva, and carries to the beehive, where non flying bees fill the mixture into honeycomb cells for ³/₄ of the cell volume. Residual cell volume is filled with honey, thus protecting the pollen mass from oxygen. An anaerobic lactic fermentation process takes place and bee bread is forming. Bee bread differs from pollen by lower pH (3.8–4.3), it contains less proteins and fats, but more carbohydrates and lactic acid. Bee bread has a better bioavailability because the walls of pollen, which cannot be destructed by gastrointestinal liquids, have been partly destructed by fermentation and the functionally and energetically rich content of pollen can be assimilated and used easier (Mizrahi and Lensky, 1997).

The chemical composition of pollen is multiform. It contains about 24 % of water and different organic and inorganic substances. It contains proteins, amino acids, carbohydrates, fats, vitamins, carotenoids, flavonoids, phenolic acids, enzymes, phytohormones, growing stimulators, micro and macro elements.

The content and variability of components can be different not only in pollen from different plants, but also from one and the same plant, which grows in different places and from the gathering time. In bee bread, proteins are partly cleaved to amino acids, fats are destructed, the content of carbohydrates and lactic acid increases, changes of other components are not significant (Shapiro and Shemetkov, 2012).

The activity of pollen (content of vitamins and enzymes) decreases after 2 or 3 months of storage. Bee bread keeps its activity longer. Pollen has antimicrobial, antioxidant hepatoprotective, immuno-modulating and antiradiation activity, adaptogenic properties. It stimulates protective forces of a human body, normalizes metabolism, has a positive influence on the liver, nervous and endocrine system functions, and enhances regeneration of tissues, physical and mental persistence of a human body. The effects of bee bread are similar, moreover the use of bee bread is more effective and valuable, because the components in bee bread are partly fermented and can be easier assimilated in the organism (Bogdanov, 2011). The main objective of this study was to prepare model for evaluate of bee bread quality.

Model for evaluation of bee bread quality. For recognition of the economic value of bee bread is necessary to evaluate the following indicators:

1. Locality of bee bread harvesting. A characteristic of the area in which was created bee bread by bee colonies is very important – emissions of volatile organic components, heavy metal contamination, contamination of soils by hazardous substances and other major indicators.

2. *Harvesting period of bee bread.* For quality assurance of bee bread is also important harvesting period from honeycomb cells. The highest quality is achieved if bee bread is removed after completion of the fermentation process. Later harvesting period can causes a certain degree of bee bread degradation by aging. Many beekeepers are taken bee bread from honeycomb cells in the winter season with freezing.

3. *Quality of fermentation process.* Bee bread is formed by lactic acid fermentation of pushing bee pollen pellet with honey in honeycomb cells. Lactic acid formed in the fermentation process preserves the pollen pellet that bees use for their nutrition. For the quality of bee bread is considered when pollen pellet by fermentation process coalesced into a compact form (Figure 1). If the pollen pellet separate – fermentation process was not yet completed (Figure 2).



Fig. 1. Bee bread after lactic acid fermentation process (Photo: A. Oravec, 2015)



Fig. 2. Bee bread with not-completed lactic acid fermentation (Photo: A. Oravec, 2015)

4. *Technology of bee bread harvesting.* Technology of bee bread harvesting from honeycomb cells significantly affects the quality of the bee bread peaces and their integrity, which is documented by Figure 3.



Fig. 3. The shape of bee bread after good quality harvesting from honeycomb cells technology by Brovarský, (2012) (Photo: A. Oravec, 2015) One of the oldest methods of bee bread harvesting from honeycomb cells is freezing and gradual harvesting with needles (Figure 4).



Fig. 4.The shape of bee bread after bad quality harvesting from honeycomb cells (Photo: A. Oravec, 2015)

5. *Storing of bee bread.* The bee bread can be storing in fresh, dried and frozen form. Bad storage condition significantly reduces the nutritional value of bee bread, increase microorganism content and can significant change colors. Long-term storage of bee bread causes unsuitable dark color (Figure 5) Unsuitable storage conditions of bee bread significantly changes the dry matter content, which is documented in Figure 6. The significant differences in dry matter content between samples are conditioned by the chemical composition (Figure 6).



Fig. 5. Long-term storage of bee bread in honeycomb cells causes unsuitable dark color (Photo: A. Oravec, 2015)



Fig. 6. The changes of dry matter content (%) in 4 samples of bee bread storing in laboratory conditions during 8 days

1. *Plant origin of bee bread. Determination* plant origin of bee bread is very difficult but very important. Obtaining process of monofloral bee bread is very difficult, but it is not impossible (Figure 7); if we have monofloral bee bread is easy

to determine plant origin. In the case of polyfloral bee bread (Figure 8) is identification of representative species possibly with the help of a microscope and pollen atlases, but it is not easy because in some cases, the similarity between the types of pollen grains in the form is very high. The second problem is that, within each category, there is a wide intra-shaped variation (Figure 9) and also variation in the surface of pollen grains (Figure 10).



Fig. 7. Monofloral bee bread (Photo: A. Oravec, 2015)



Fig. 8. Polyfloral bee bread (Photo: A. Oravec, 2015)



Fig. 9. The compare of pollen shape from selected group of wild cherry (Photo: R. Ostrovsky, 2013)

2. Morphology characteristic of bee bread. The important quality indicators of bee bread are shape, uniformity in shape and size of bee bread (Figure 3 and 4). Between the samples of bee bread may be smaller or larger differences in the basic indicators such as weight, height and width of bee bread (Table 1).



Fig. 10. The compare of pollen surface from selected group varieties of apple tree (Photo: R. Ostrovsky, 2013)

3. *Chemical composition of bee bread.* Chemical composition of bee bread is very important, because give us information about nutritional value. For this purpose, it is important to know the basic content of protein, carbohydrate, fat, and the essential vitamins and minerals in bee bread. For

practical use of bee bread it is necessary to know the variability of the content of these components. Nowadays is bee bread also using as dietary supplement, but in this case it must be declared on the label content of basic nutritional components. The results in Table 2 demonstrates that the number of components, we have found a very important variation – we determined the value of coefficients of variation from 6.10% (dry matter) to 65.26% (carbohydrates). The significant differences in the chemical composition are related to the plant species. It is well known that the chemical composition of pollen which forms bee bread is specific for plant species.

Genotypes	n	mın	max	X	S	V%
Weight of bee bread (mg)						
FE-1	30	275	405	341.66	39.64	11.60
FE-2	30	276	390	357.10	22.79	6.38
FE-3	30	252	363	305.10	28.19	9.24
FE-4	30	260	392	323.80	23.79	7.35
FE-5	30	181	300	239.90	31.56	13.16
Height of bee bread (mm)						
FE-1	30	8.50	13.12	11.18	1.00	8.99
FE-2	30	10.38	12.88	11.79	0.61	5.19
FE-3	30	7.96	12.54	10.25	1.00	9.84
FE-4	30	8.25	11.12	10.18	0.61	6.07
FE-5	30	7.80	12.37	10.03	1.33	13.35
Width of bee bread (mm)0						
FE-1	30	3.38	6.03	5.33	0.60	11.33
FE-2	30	4.72	6.04	5.46	0.30	5.54
FE-3	30	3.22	6.26	5.36	0.57	10.54
FE-4	30	5.20	6.15	5.74	0.24	4.21
FE-5	30	3.30	5.89	5.19	0.47	9.08

1. The variability of basic morphological indicators of bee bread*

*Source: Results of authors.

2. Variability in basic chemical content of bee bread*

Component	Ν	Unit	Minimum	Maximum	Mean	V %
Dry matter	7	g/100g	71.28	83.65	73.61	6.10
Proteins	7	g/100g	13.99	20.03	16.90	14.24
Fats	7	g/100g	6.48	8.32	7.03	8.47
Carbohydrates	7	g/100g	2.09	17.03	8.54	65.26
Ash	7	g/100g	2.16	4.31	2.87	24.91
Dietary fiber	7	g/100g	26.43	43.75	38.26	14.70
Phosphor	7	mg/kg	4346.00	7440.00	5210.71	20.68
Potassium	7	mg/kg	4536.00	9892.00	6152.43	31.25
Magnesium	7	mg/kg	768.00	3332.00	1735.00	54.51
Calcium	7	mg/kg	1116.00	2064.00	1648.00	17.83
Natrium	7	mg/kg	50.00	76.00	58.29	15.14

*Source: Results of authors.

4. Secondary metabolites content. It is well known that practical using of bee bread in pharmacy, cosmetics and other areas is related to the content

of bioactive components. It is well known that the pollen grains from each species are rich for biological active substances. This finding is significant on the one hand for the specific use of bee bread, but on the other hand, it is also disadvantageous because sometimes of the processing industry is not suitable bee bread from each plant species or polyfloral bee bread.

5. *Heavy metal content.* The heavy metal content can be determined in a bee organs, plant parts, and therefore also in bee products (Table 3). The quality of bee bread significantly reduces excessive content levels of heavy metals. It is very important for each bee bread sample determine the essential content of heavy metals.

Paramotors	Trace elements and metalloids; mg/kg						
Farameters	Cu	Zn	Pb	Cd	Ni	Со	
Whole bees	15.23±0.88	84.08±8.41	2.42±0.32	0.11±0.01	1.21±0.11	1.29±0.17	
Fecal mass	28.70±1.44	98.01±15.93	9.59±2.04	0.17±0.02	5.19±0.32	2.05±0.19	
Bee honey	0.36±0.01	1.96±0.04	0.05±0.001	0.03±0.001	0.17±0.01	0.09±0.004	
Pollen	11.54±0.76	10.00±0.21	0.92±0.04	0.16±0.03	0.69±0.05	0.80±0.04	
Wax	2.03±0.14	12.24±0.73	0.28±0.01	0.27±0.01	0.57±0.10	0.25±0.02	

3. Content of trace elements and metalloids in bee body and bee products (Zhelyazkova et al. 2010)

6. *Residues of pesticide contentio.* The quality of bee bread significantly decreases the levels of residues of agropesticide. Only a small number of laboratories in the world are able to determine all that may contain residues after agropesticide. It shows the transparent work by the team of authors Johnson et al. (2010), which describe in pollen, honey, bees wax and the contents of more than 120 agropesticide. In Table 4 we present only selected part of their results.

7. *Radionuclides content.* The radioactivity in other bee products has been less investigated. Measurements of ¹³⁷Cs radioactivity in the Ukraine showed very high averages for products harvested between 1986 and 1989: pollen: 11070 Bq/kg, propolis: 34310 Bq/kg. In France considerably lower values were measured at the same time: honey, 29 Bq/kg; pollen, 283 Bq/kg (Canteneur, 1987). In Italy, measurements of honey and pollen harvested in 1986 showed ¹³⁷Cs values in honey between 30 and 360 Bq/kg, while those in pollen varied between 1000 and 2500 Bq/kg (Porrini et al., 2002). Pollen and propolis are considered to be better indicators for radioactive contamination than honey (Alexenitser and Bodnarchuk, 1999).

8. *Microbiological characteristic of bee bread.* There is no pollen from plant, bee pollen pellet or bee bread, which did not contain any microorganisms. The question is which this is, whether pathogenic or not pathogenic if they are used by human. Unfortunately many microorganisms significantly reduce the quality of bee bread. It is therefore necessary microbiological characteristics of each sample of bee bread and knows the presence of the species. Identification is sometimes very complicated. In Table 5 we present the number of bacteria and fungi provided on bee pollen pellet derived from different plant species. In table 6 we presented some types of

microorganisms in the evaluation samples of bee pollen pellet. The results clearly confirm the significant differences between the samples.

Total postigida	CL 4 5 5	Maximum detection in ppb			
Total pesticide	ULA33	Wax	Pollen	Bee	Honey
Allethrin	PYR	139	11	24	n.d.
Amicarbazone	HERB	n.d.	98	n.d.	n.d.
Amitraz	FORM	46060	1117	13780	555
Atrazine	S HERB	31	49	15	81
Azinphos methyl	OP	817	643	91	n.d.
Azoxystrobin	S FUNG	278	107	n.d.	4
Bifenthrin	PYR	56	13	12	3
Boscalid	S FUNG	388	962	33	n.d.
Bromopropylate	MITI	135000	11	2245	245
Captan	PS FUNG	400	10000	1740	1
Carbaryl	PS CARB	820	94000	5800	42
Carbendazim	S FUNG	133	149	14	27
Carbofuran	S CARB	22	1400	669	645
Chlorothalonil	FUNG	53700	98900	878	10
Chlorpyrifos	OP	890	830	57	15
Coumaphos	OP	94131	5828	2777	2020
Cyfluthrin	PYR	45	34	14	9
Cyhalothrin	PYR	17	1672	2	0.8
Cypermethrin	PYR	131	1900	26	92
Cyprodinil	S FUNG	106	344	19	n.d.
DDT-p,p''	OC	>40	45	7	658

4. Determine of selected residues of agropesticide in bee products according to Johnson et al. (2010) results

5. Number of bacteria and microscopic fungi detected on pollen samples (Brindza et al. 2010)

Pollen sample No.	Bacteria: CFU/g of pollen	Microscopic fungi: Ranges of CFU/g of pollen
1	300 630 - 1089	
2	1100	566 - 1100
3	13125	2761 - 4688
4	1125	112 - 664
5	1250	606 – 911
6	1000	3733 - 3978
7	100	165 - 690
8	1200	107 - 343

9. *Color of bee bread.* The bee bread quality increases natural color from pollen pellet which form bee bread. Dark colors of bee bread in many cases already indicate bad storage or late harvesting from honeycomb cells.

10. *Smell of bee bread.* The significant quality indicator of bee bread is smell. Bee bread with excellent quality characterizes typical fruit and natural odors.

Samples of	Bacteria	Microscopic fungi			
Pollen no. 1	Serratia marcescens Agrobacterium radiobacter Fermenting and non- fermenting Gram-negative rods	Alternaria chartarum Aspergillus flavipes Aureobasidium pullulans (De Bary) Arnaud Cladosporium sphaerospermum Penz. Humicola grisea Traaen Monodictys castanae (Wallr) Hughes Mucor racemosus Fres. Penicillium sp			
Pollen no. 2	Serratia marcescens Agrobacterium radiobacter Fermenting and non-fermenting Gram- negative rods	Aspergillus repens (Corda) De Bary Cladosporium sphaerospermum Penz. Mucor spinosus Tiegh. Paecilomyces varioti Bainier. Penicillium sp. Rhizopus arrhizus A. Fisher Rhizopus nigricans Ehrenb.			

6. Bacterial and fungal species and groups detected on pollen samples and on bumble bee individuals

Conclusion

Bee bread is considered as very important and major bee product. Most of bee bread is used in folk medicine in almost all countries and as a dietary supplement. Its production has not been sufficient in the past, mainly because of the complicated process of its harvested from the honeycomb cells. At present, the production of bee bread increased mainly due to the use of new technologies in its collection. Bee bread can therefore become a major product for pharmaceutical, cosmetic and food industry applications. For potential customers and fixing prices of bee bred is necessary to have elaborated a baseline standard for the assessment of its economic value based on its basic indicators of quality. In this work we described some important indicators of the quality of bee bread as a proposed model for the processing of a basic bee bread quality standard.

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References

1. Alexenitser M. L. Beekeeping products as bioindicators of radioactive contamination in a certain zone / M. L. Alexenitser, L. I. Bodnarchuk // In Apiacta, 1999. – Vol. 34. – P. 93–96.

2. Brindza J. Pollen microbial colonization and food safety / J. Brindza, J. Gróf, K. Bacigálová, P. Ferianc, D. Tóth // In *Acta Chimica Slovaca*, 2010. – Vol. 3. – P. 95–102.

3. Canteneur R. Le contrôle de la radioactivité à travers l'abeille et les produits du rucher / R. Canteneur, 1987 : L'Abeille de France. – P. 87–90.

4. Johnson M. R. Pesticides and honey bee toxicity / M. R. Johnson, M. D. Ellis, Ch. M. Mullin, M. Frazier // Usa: In *Apidologie*, 2010. – Vol. 41. – P. 312–331.

5. Krell R. Value-added products from beekeeping / R. Krell // In FAO Food and Agriculture Organization of the United Nations Roma, 1996. – 409 p.

6. Mizrahi A. Bee products: properties, applications, and apitherapy / A. Mizrahi, Y. Lensky // USA : Springer, 2012. – P. 93–101.

7. Porrini C. Use of honey bees as bioindicators of environmental pollution in Italy / [C. Porrini, S. Ghini, S. Girotti, A. G. Sabatini, ats.] // In: Devillers, J., Pham-Delègue M. H. (Eds.) /// Honey bees: Estimating the environmental impact of chemicals. Taylor & Francis : London and New York, 2002. – P. 186–247.

8. Shapiro D. Investigation of biologically active compounds of pollen of honey plants. [cited 2012 Jul 10;4 screens] / D. Shapiro, M. Shemetkov // Available from : URL: http://airbees.com/articles/pollen/199-issledovanie-biologicheski-aktivnyh-soedineniypylcy-medonosnyh-rasteniy.html

9. Zhelyazkova I. Content of heavy metals and metalloids in bees and bee products from areas with different degree of anthropogenic impact / I. Zhelyazkova, S. Atanasova, V. Barakova, G. Mihaylova // In *Agricultural Science And Technology*, 2010. – Vol. 3. – P. 136–142.

10. Zuluaga C. M. Chemical, Nutritional and Bioactive Characterization of Colombian Bee-Bread / C. M. Zuluaga, J. M. Serrato, M. C. Quicazan // In *Chemical Engineering Transactions*, 2015. – Vol. 43. – P. 175–180.

ОЦІНКА ЯКОСТІ ПЕРГИ БДЖОЛИНОЇ

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Анотація. Визначено необхідність оцінки якості перги бджолиної. Цей продукт відомий і використовується багатьма цивілізаціями вже протягом багатьох століть. Але, незважаючи на це, в науковій літературі недостатньо експериментальних результатів і знань про цей продукт бджільництва. Тому в даній роботі представляємо модель оцінки основних показників якості перги бджолиної. Найважливішими показниками якості вважаємо місцевість збору перги, період збору, технологію збору, спосіб зберігання, рослинне походження, морфологічну характеристику, основний хімічний склад, вміст важких металів, залишків пестицидів, присутність мікроорганізмів та інші.

Необхідно, щоб для кожного зразка було оцінено всі показники. Комплексна оцінка дозволяє визначити економічну цінність зразка та його практичне застосування. Багато наукових робіт орієнтовані лише в одному напрямі, щоб оцінити тільки один показник. Під час презентації моделі було використано власні результати і висновки з літературних джерел.

Ключові слова: перга, модель оцінки, якість, показники якості.

ОЦЕНКА КАЧЕСТВА ПЕРГИ ПЧЕЛИНОЙ

Я. Бриндза, З. Шубертова, В. Броварской, С. Величко, И. Крефт, Э. Иванишова

Аннотация. Определена необходимость оценки качества перги пчелиной. Этот продукт известен и используется многими цивилизациями уже на протяжении многих веков. Но, несмотря на это, в научной литературе недостаточно экспериментальных результатов и знаний об этом продукте пчеловодства. Поэтому в данной работе представляем модель оценки основных показателей качества перги пчелиной.

Важнейшими показателями качества считаем местность сбора перги, период сбора, технологию сбора, способ хранения, растительное происхождение, морфологические характеристики, основной химический состав, содержание тяжелых металлов, остатков пестицидов, присутствие микроорганизмов и другие. Необходимо, чтобы для каждого образца были оценены все показатели. Комплексная оценка позволяет определить экономическую ценность образца и его практическое применение. Многие научные работы ориентированы только в одном направлении, чтобы оценить только один показатель. Во время презентации модели были использованы собственные результаты и выводы из литературных источников.

Ключевые слова: перга, модель оценки, качество, показатели качества.

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POLLEN AND BEE POLLEN FEATURES OF SWEET CHESTNUT (CASTANEA SATIVA MILL.)

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Annotation. The aim of this work was to study general morphological characteristics of pollen grains and bee pollen of sweet chestnut (Castanea sativa Mill.). The studies were carried on pollen extracted from genotypes,

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