THE ANALYSIS OF FOUR HONEY TYPES FROM TWO INDEPENDENT PRODUCERS FROM SALAJ COUNTY

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Abstract

Honey represents an apiarian product obtained from flower nectar which is transformed by bees in honey, being deposited in comb cells, representing a food reserve for bees population. Also, honey has a great importance for people, being an important foodstuff, having therapeutic action and a great importance for cosmetics. Honey contains simple sugars such as glucose and fructose, being easily assimilable.

In present study were analyzed four honey samples from two producers from Salaj County. All samples were analyzed inorderto see if the existing rules in the field were respected. Such rules are established taking in account some quality requirements which must meet the honey when is picked up from the producer. Withal was established if the analyzed samples were falsified by conscious addition of the substances that increases the quantity of honey. All analysis were effectuated considering the legal regulation in the field.

Keywords: honey, human health, sugar, health properties

INTRODUCTION

Honey is the most recognized and famous natural food and its composition is extremely variable, depending on its botanical and geographical origins (Ciulu et al., 2016). Honey has been demonstrated to possess anti-inflammatory property and most commercially available therapeutic honey is derived from flowering Leptospermum scoparium (manuka) plants from New Zealand (Cokcetin et al., 2016). In Romania are founded two major categories of honey: floral honey (nectar honey), which can be mono or poly floral one, and extraflora one (manna or forest honey), the well know types of monofloral honey are acacia, lime, sunflower, mint, rapeseed, corianderhoney. Polyphenols founded in honey are mainly from the flavonoids class, such as: quercetin, luteolin, kaempferol, apigenin,

crizin, galangin, which are compounds with well known antioxidant properties.

A large number of studies published into the scientific literature shown the large number of honey application as aliment or as drug having antiproliferative and antimetastaticeffects in treatment of brain tumors (Moskwa J. et al., 2014), considerable therapeutic effect in: chronic wounds, ulcers and burns healing (Al-Waili et al., 2011). Also, honey proved good antimicrobial effects against microbes from oral cavity (Safii et al., 2017), and a mixture obtained from honey and mustard seeds proved effective endodontic effects (Sobhi, Manzoor, 2004). In order to produce all expected therapeutic effects, honey must meet certain qualitative requirements.

An important requirement is represented by the phenolic profile of honey which is due to the different floral sources, being a consequence of the natural presence of such compound into the plants used as sources by bees (Chis, Purcarea, 2011; Chis et al., 2012;Ciulu et al., 2016; Purcãrea, Chis, 2011;Szabo et al., 2013).

Organoleptic examination of honey consists of analyzing colour, consistency, smell, taste and purity of the honey. In general the examination of honey is done for several purposes: appreciation of its quality and purity, determination of whether it was falsified or not, determination of the deterioration or degradations state. The purpose of present study was to analyze four types of honey by organoleptic examination and physical/chemical analysis in order to identify whether they were falsified, taking in account that honey is one of the most falsified foods in the world. When consuming for long time fake honey instead of protecting our health by the benefit normally offered by genuine honey, we compromise our health.

MATERIALS AND METHODS

Organoleptic examination: in present paper were analyzed four different honeys provided by two producers from Sălajcounty. From first producer were analyzed one acacia honey and one polyfloral honey, and from second one were analyzed one sunflower honey and onepolyfloral honeys. (Mărghitaş L.A., 2005; SR 2213-12:2009; SR 784-1:2009; SR 784-3:2009; SR 784-3:2009; SR 784-3:2012)

Color: honey color analysis was performed by visual inspection in daylight. Honey consistency was evaluated by analyzing honey flowing on a wand. The smell and the taste of honey are two analyses which are made directly by smelling and by tasting the honey samples. Natural honey can contain some impurities represented by remains of combs, propolis, pollen,

bee remnants. Determination of physical impurities represents mandatory analysis for honey. Physical and chemical analyzes were carried out after the honey was homogenized using a glass rod to uniformly disperse the impurities throughout the mass, filtered through double gauze and leave it to rest until all embedded air has been removed and honey have cleared. For crystallized honey, it was subjected to fluidification by heating it at 40-45°C so all the crystals melt. Then, the honey was homogenized using a glass rod, and filtered in order to remove all impurities.

Humidity determination can be done using two methods: refractometric and oven drying. Refractometric method is based on the fact that we have a direct correlation between the refractive index of honey and its dry matter content, which allow us to determine the water percentage. Refractive index determination was made within the temperature range of 15 to 25°C, and then corrected for the temperature of 20 °C. Oven drying method consists in drying the sample to a constant mass. Mass loss is calculated as percentage and represents the humidity of the honey. In order to obtain real results were performed several measurements and were taken in account only two determination between which there was no more than 0.1 % difference.

Ash determination can be carried out by calcining the honey sample in a furnace until a constant mass was obtained. The residue obtained is calculated as percentage and is representing the ash content.

Determination of acidity was performed by dissolving the honey sample in waterand titrating resulted solution with sodium hydroxide 0.1 Nin the presence of phenolphthalein.

Determination of reducing sugar by Elser method: reduction sugar contained in honey can convert copper sulphate in cuprous oxide (red precipitate) in alkaline medium, by heating the system. Inverting sugar for artificial feeding of bees represent a process in which sucrose decomposes in glucose and fructose.

Determination of diastase index – in natural honey exist several enzymes which are acting on starch contained in honey. Amylase is the most resistant enzyme at thermal treatment. The diastase index has low values (even 0) when the honey was subject of brutal heating treatment, or when the honey was subject of falsification.

Determination of sucrose by Elser method is made by calculation the difference between the amount of direct reducing sugar before and after an acidic hydrolysis (inverting) and corrected with a conversion factor of reducing sugar directly in sucrose equivalent.

Usual methods for determination of falsified honey:

Determination of artificial inverted sugar (Fiehe reaction) – which consist in identification of hidroxymethylfurfurol, which is an intermediate product obtained during the degradation of hexoses treated by heating with an acid (acidic hydrolysis). When the sugar is artificial inverted are produced glucose and fructose which suffer some degradation producing in this way hydroxymethylfurfurol.

Identification of industrial glucose (Fiehe reaction) – by adding industrial glucose during honey falsification process lead at some increase of the glucose content, in comparison with the fructose content. In natural honey the ration between glucose and fructose content is less than 1. By adding industrial glucose, the glucose concentration increase, fructose concentration decrease, leading at modification of glucose – fructose ration, and the honey is not respecting the legal regulations.

Identification of flour, starch or starch thickening substances – such compounds are added in honey in order to correct the consistency of the honey. Presence of such compounds can be identified by organoleptic analysis, when must be observed the opalescent, milky appearance of the honey. Honey flavour does not change, but honey is prone to early deterioration.

Identification of glue or gelatine added if it is desired to correct the honey consistency can be made oraganoleptically. Honey jammed with gelatine has a thin gel appearance. When the honey was falsified by adding gelatine or glue a flocking like abundant precipitate appears immediately. In case of natural honey a white cloudiness can be observed.

RESULTS AND DISSCUSIONS

Results obtained for the organoleptic examination of the four honey samples are depicted in table 1.

Organalantia analysis of honor complex

Table 1.

| | | Oraganoicpue and | irysis or noney samples | |
|-----------------------|------------------|-------------------------------------------------------------|---------------------------------------------------------------------------------------------|-------------------------|
| Honey | Colour | Smell and taste | Consistency | Impurity content [%] |
| Sun flower honey | Yellow- gold | Sweet taste, undefined aroma | Total crystallized, fine sandy appearance, does not flow from wand | 0,05 |
| Acacia honey | Light yellow | Pleasant, sweet taste, pronounced acacia flower aroma | Transparent, perfectly fluid or fluid thin, uniform, without signs of crystallization | 0,03 |
| Polyfloral honey 1 | Orange yellow | Sweet taste, pleasant flavour | Fluid with a fine crystallization | 0,04 |
| Polyfloral honey 2 | Brown orange | Sweet taste, pleasant aroma | Fluid with a crystallization start | 0,04 |

Humidity determination of analysed samples was performed by using two methods described earlier and the results are presented in table 2.

Table 2.

| framally accommution | | | | | |
|----------------------|------------|-----------------------------------|--------------------------|--|--|
| Honey | Refractive | Humidity (%), determined by using | Humidiy(%) determined by | | |
| Honey | index | refractometric method | drying the samples | | |
| Sun flower honey | 1,4886 | 19,18 | 19,20 | | |
| Acacia honey | 1,4875 | 19,60 | 19,69 | | |
| Polyfloral honey 1 | 1,4882 | 19,35 | 19,40 | | |
| Polyfloral honey 2 | 1,4881 | 19,43 | 19,40 | | |
| | | | | | |

Humidity determination

Analyzing data presented in scientific literature was observed that generally acacia honey have low water content (Lazarevic et al., 2012). From obtained experimental data can observe that the analyzed samples are from the class of honey produced in temperate climate.

For determination of ash content were performed several analysis in order to obtain a difference of maximum 0.02 % against average value. Obtained experimental data are depicted in table 3. Muhammad Shahnawayet all reported that the maximum ash content (0.082 %) was founded in berry honey, followed by herbal honey (0.074 %) and acacia one (0.054 %) (Shahnawaz et al., 2013).

| | Ash content of analyzed honey | | | |
|----------|-------------------------------|-----------------------|------------------|--|
| | Honey Ash content [%] | | | |
| | Sun flower honey | 0,14 | | |
| | Acacia honey | 0,12 | | |
| | Polyfloral honey 1 | 0,11 | | |
| | Polyfloral honey 2 | 0,13 | | |
| Honey ac | idity represents a qu | ality parameter which | is used in order | |

Honey acidity represents a quality parameter which is used in order to appreciate the freshness of honey (Mădaş M.N., 2013). Commercial honeys with a good quality have a maximum acidity of 4 mvales per 100 g of product. The average pH of good quality honeys is typical situated between 3.4 and 6.1 (National Honey Board Food Technology). The average pH of analyzed honey samples was 3.9. Obtained data are presented in table 4 and are in accordance with the data presented in scientific literature, all analyzed samples falling within the accepted pH range.

Table 4

Table 3

| Acidity values for | analyzed honey |
|--------------------|----------------|
| Honey | Acidity |
| Sun flower honey | 3,85 |
| Acacia honey | 4,00 |
| Polyfloral honey 1 | 4,15 |
| Polyfloral honey 2 | 3,60 |

Reducing sugar from analyzed samples was determined by using Elser method, and the obtained experimental data are depicted in table 5.

Paduaing sugar contant of analyzed honory

Table 5

| Reducing sugar content of analyzed honey. | | | | |
|-------------------------------------------|----------------------------|--|--|--|
| Honey | Reducing sugar content [%] | | | |
| Sun flower honey | 74.07 | | | |
| Acacia honey | 73.28 | | | |
| Polyfloral honey 1 | 74.40 | | | |
| Polyfloral honey 2 | 58.47 | | | |
| | | | | |

The diastatic index of honey may have values under the limits set by food safety regulation if the honey has been brutally heated, when the honey is very old, and was not kept at right temperature, it comes from bees fed only with sugar syrup, was falsified with artificially inverted sugar syrup. If the diastase index approaches or exceeds 50 we can affirm that analyzed honey was falsified with malt extract or with other sweet syrups prepared by enzymatic hydrolysis of starch. Experimental values for diastatic index of analyzed honey samples are presented in table 6.

Table 6.

| J | Diastatic | index | obtained | for | ana | lyzed | hone |
|---|-----------|-------|----------|-----|-----|-------|------|
| | | | | | | ~ | |

| T | 1 | |
|---|----------------------|----------------------|
| | Diastatic index obta | ined for analyzed ho |
| | Honey | Diastatic index |
| | Sun flower honey | 10,87 |
| | Acacia honey | 7,14 |
| | Polyfloral honey 1 | 13,16 |
| | Polyfloral honey 2 | 13,16 |

Determination of sucrose was carried out by the Elser method, and the results obtained for the four types of honey were weighed in table 7.

Table 7.

| Sucrose content in analyzed honey | | | | |
|-----------------------------------|---------------------|--|--|--|
| Honey | Sucrose content [%] | | | |
| Sun flower honey | 3,61 | | | |
| Acacia honey | 4,80 | | | |
| Polyfloral honey 1 | 4,27 | | | |
| Polyfloral honey 2 | 8.07 | | | |

Usually the falsified honey can be determined by using some usual methods which are standardised one (Mărghitas L.A., 2005; SR 2213-12:2009; SR 784-1:2009; SR 784-1:2009; SR 784-3: 2009; SR 784-3/C91: 2012)

In real word a lot of honey producer are interested to obtain huge benefits by trading huge quantities of honey, but they are not able to produce such amounts and they falsify produced honey. Honey falsification can be made by producers by mixing it with different substances such as: flour, glucose, sugar, starch, gelatine, etc.

In real practice were founded two different used to falsify honey: direct and indirect falsification. Direct falsification is made by the producers in order to correct some physical characteristics of produced honey. Indirect falsification is a consequence of incomplete and incorrect knowledge of the processing procedures applied to honey. In order to determine the falsified honey can apply several methods.

1.- Determination of artificially inverted sugar is possible by using Fiehe reaction (obtained data are presented in figure 1).





Fig. 1. Determination of artificially inverted sugar in analysed samples (sun flower, acacia, first polyfloral, and second polyfloral honey samples).

Fig. 2. Analysis of honey falsification using industrial glucose

From performed analysis was observed that the second polyfloral honey presented a red coloration, which persisted also after three hours. The other three honey samples presented a pink coloration, which was not persistent one (data depicted in figure 1).

2.- Identification of industrial glucose was made using Fiehe reaction (obtained data are depicted in figure 2).By analyzing the four test tubes can observe that in last one a milky coloration appears (in this tube was analyzedpolyfloral honey obtained from second producer).

3.- Identification of flour, starch or starch compounds addition – is possible by using iodine reaction (obtained data are depicted in figure 3).By analyzing data presented in figure 3 can observe that the analyzed samples were not falsified by using flour, starch or starch compounds.



Fig. 3. Identification of presence of flour, starch or starch compounds in honey



Fig. 4. Identification of gelatine or glue honey falsification

4.- Identification of the usage of glue or gelatine. Presence of bones glue or of gelatine is possible because these compounds form in presence of tannin some abundant precipitates (obtained data are depicted in figure 4).By analyzing data presented in figure 4 can conclude that the analyzed samples were not falsified by adding glue or gelatine.

CONCLUSIONS

From the data obtained from organoleptic analysis of analyzed honey samples can conclude that all samples respect the regulation limits regarding the quality. By comparing the experimental data obtained for humidity, ash content, acidity, reducing sugar, diastatic index, and sucrose content with the standard values can conclude that all analyzed samples respect the legislation and can be commercialized. Also all samples were analysed in order to see if they were falsified. Experimental data confirm that all samples were not falsified and can be commercialized.

REFERENCES

- Al-Waili N., K. Salom, A.A. Al-Ghamdi, 2011, Honey for Wound Healing, Ulcers and Burns Data Supporting Its Use in Clinical Practice, Review The Scientific World Journal, 11, 766–787.
- Chis A., C. Purcarea, 2011, The effect of thermal tratament on fresh monofloral honey types from Bihor county, AnaleleUniversității din Oradea, Fascicula: Ecotoxicologie, ZootehnieşiTehnologii de IndustrieAlimentară, 71-76
- Chis A., E. Szabó, M. Borbély, N. Czipa, C. Purcărea, 2012, Comparative study of acacia and rape honey, AnaleleUniversitatii din Oradea. Fascicula: Ecotoxicologie, ZootehniesiTehnologii de IndustrieAlimentara, 325-332
- Ciulu M., N. Spano, M.I. Pilo, G. Sanna, 2016, Recent Advances in the Analysis of Phenolic Compounds in Unifloral Honeys, Molecules, 21(4):451
- Cokcetin N.N., M. Pappalardo, L. T. Campbell, P. Brooks, D.A. Carter, S.E. Blair, E.J. Harry, 2016, The Antibacterial Activity of Australian *Leptospermum* Honey Correlates with Methylglyoxal Levels, PLoS ONE, 11(12), e0167780.
- Lazarević K.B., F. Andrić, J. Trifković, D. Milojković-Opsenica, 2012, Characterization of Serbian unifloral honeys according to their physicochemical parameters, Food Chemistry, 132, 2060-2064.
- Mădaş M.N., 2013, Cercetăriprivindindicii de calitateşiautenticitateaimierii de salcâm (robiniapseudoacacia), Teza de Doctorat, Cluj-Napoca
- 8. Mărghitaș L.A., 2005, Albineleșiproduselelor, Editura Ceres, București
- Moskwa J., M.H. Borawska, R. Markiewicz-Zukowska, A. Puscion-Jakubik, S.K. Naliwajko, K.Socha, J. Soroczynska, 2014, Polish Natural Bee Honeys Are Anti-Proliferative and Anti-Metastatic Agents in Human Glioblastomamultiforme U87MG Cell Line, PLOS ONE, 9(3): e90533
- National Honey Board Food Technology/ Product Research Program. Available on internet from: http://www.honey.com/images/downloads/ph-acidsinhoney.pdf.
- Purcãrea C., A. Chis, 2011, Chemical and Biochemical Characterization of Three different types of honey from Bihor County, AnaleleUniversitatii din Oradea, Fasicula: Ecotoxicologie, ZootehniesiTechnologii de IndustrieAlimentara, 313-318
- Safii S.H., G.R. Tompkins, W.J. Duncan, 2017, Periodontal Application of Manuka Honey: Antimicrobial and Demineralising Effects In Vitro, Int J Dent., ID 9874535
- Shahnawaz M., S.A. Sheikh, M. Hussain, A. Razaq, S.S. Khan, 2013, A study on the determination of physicochemical properties of honey from different valleys of Gilgit-Baltistan, International Journal of Agricultural Science Research, 2(2), 049-053
- Sobhi M.B., M.A. Manzoor, 2004, Efficacy of camphorated paramonochlorophenol to a mixture of honey and mustard oil as a root canal medicament, J Coll Physicians Surg Pak., 14(10), 585-588
- 15. SR 2213-12:2009 ProdusedulciPartea 12: Determinareaconținutului de zahăr
- 16. SR 784-1:2009 Miere de albinePartea 1: Cerințe de calitate la preluarea de la producători
- 17. SR 784-1:2009 Miere de albinePartea 2: Cerințe de calitate la comercializare
- 18. SR 784-3: 2009 Miere de albinePartea 3: Metode de analiză
- 19. SR 784-3/C91: 2012 Miere de albinePartea 3: Metode de analiză
- 20. Szabó E., M. Borbély, P. Sipos, C. Purcărea, A.Chiş, 2013, Analysis of total polyphenol contents and colour of brewed beer samples, AnaleleUniversității din Oradea, Fascicula: Ecotoxicologie, ZootehnieşiTehnologii de IndustrieAlimentară, XII/B, 373-377