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PHYSICO-CHEMICAL PROPERTIES OF HONEY PRODUCED IN THE CENTRAL REGION OF PUNJAB, PAKISTAN

HAMMAD SHAFIQ[1]*, AZKA SHAHID[1] AND MUHAMMAD ZAKRIA FARID[2]

[1.] DEPARTMENT OF FOOD TECHNOLOGY, UNIVERSITY OF ARID AGRICULTURE RAWALPINDI, PAKISTAN.

[2.] DEPARTMENT OF HORTICULTURE, UNIVERSITY OF ARID AGRICULTURE RAWALPINDI, PAKISTAN.

Corresponding author's e-mail: hammadshafiq_93@yahoo.com

ABSTRACT:

The aim of current study is to determine the physicochemical properties of honey samples collected from the central region of Punjab province of Pakistan. Ten samples were analyzed for several physicochemical parameters including water activity, moisture, acidity, hydroxymethylfurfural (HMF), soluble solids, electrical conductivity, ash contents, sugar content, mineral content and color (L*, a*, b*). The average value for water activity is 0.588, for moisture 17.36 %, for acidity 29.2 meq kg^{-1}, for HMF 13.87 mg kg^{-1}, for soluble solids 80.6 °Brix, for conductivity 0.463 µS cm^{-1}, for ash content 2.12 g $kg^{-1}$and for fructose and glucose is 384.0 g kg^{-1}, 321.0 g kg^{-1} receptively .The honey samples present a good level of quality because the physicochemical values were in the range of approved limits (conforming to codex standards) for all honey samples.

KEY WORDS: *Honey, physicochemical characteristics, Sugars, Minerals.*

INTRODUCTION:

Honey can be said as the miracle food, because of its nutritional and health benefits bestowed to human beings since centuries. It is enjoyed by almost every generation throughout the history of mankind because of its excellent nutritional value and therapeutic potential (Hammock, 2011; Jeffrey and Echazarreta, 1996). Honey is a natural thick, sticky and viscous supersaturated sugar solution, whose composition mainly consists of unique mixture of carbohydrates. Besides sugars honey also contain many other constituent's, though in minor quantity, viz., organic acids and amino acids (like gluconic acid, acetic acid etc.), proteins, enzymes (catalase, glucose oxidase, invertases, phosphatases etc.), lipids, vitamins (like ascorbic acid, niacin etc.), phenolic acids, volatile compounds and minerals (Bogdanov *et al.*, 2004a). Colour, aroma, flavour and chemical properties of honey mostly dependent upon the plant species from which honeybees collects the nectar and also upon the weather, handling and storage conditions (Guler *et al.*, 2007)

Honey from different regions of world has been studied for their physical properties and chemical composition by many researchers (Ahmed *et al.*, 2007; Al *et al.*, 2009; Baroni *et al.*, 2009; Bertoncelj *et al.*, 2007; Castro-Vázquez *et al.*, 2007; De la Fuente *et al.*, 2007; Finola *et al.*, 2007). Honeys from different countries vary greatly in composition, colour and physicochemical properties; very little data is present on the properties of different honey types produced in Pakistan. Therefore, the present study was conducted to ascertain variation in physicochemical properties and mineral composition of the honeys produce in central region of Punjab province of Pakistan.

MATERIALS AND METHODS:

Honey samples

Ten honey samples (H1–H10) were collected from different locations of the central region of Punjab for caring out current study. All samples were collected from the local bee keepers. Honey samples were stored in the holders after collection and then transported to the laboratory, where they were kept at room temperature (~25°C) until analyzed.

Physicochemical analysis

Soluble solids and moisture were measured according to the official method of the Association of Analytical Chemists (AOAC, 1990) by using an Abbe refractometer (Abbe® model 10450). Soluble solids and moisture were expressed as °Brix and %, respectively.

Water activity (a_w) was determined by means of an AquaLab CX2 water activity meter (Decagon Device, USA) according to the procedure reported by Zamora, Chirife and Roldan (2006).

Hydroxymethylfurfural (HMF) was measured according to the spectrophotometric method recommended by Zappala, Fallico, Arena, and Verzera (2005) using a Lambda 45 UV–Vis spectrophotometer (PerkinElmer, USA). Results were expressed in HMF mg kg^{-1} of honey.

Determination of ash content was made by calcination, at 560 °C in a muffle furnace, until constant mass was achieved according to the method of the AOAC (1990) and the results were stated as g kg^{-1}.

Electrical conductivity of a honey was determined using Orion A122 Conductivity Meter (Thermo-Orion, Boston, USA) according to the method reported by Bogdanov (1997) and the results were stated as µS cm^{-1}. Acidity was measured by titration method. First 0.05 N NaOH is added drop by drop to honey solution, titration is stopped when pH 8.5 is achieved (free acidity), instantly 10 ml 0.05N NaOH was added, and without interval back-titration is done with 0.05N HCL until pH 8.3 is achieved (lactone acidity). Total acidity is calculated by adding free plus lactone acidities (AOAC, 1990). Results were expressed in meq kg^{-1}.

Determination of sugar contents

Glucose content of the honey samples was determined by enzymatic oxidation with glucose oxidase reagent (Randox Laboratories Ltd., UK) according to method reported by Buba, Gidado and Shugaba (2013). 2 mL of the reagent was allowed to react with twenty microlitres (20 µL) of the sample or standard, mixed well and incubated at 37°C for 10 min. The absorbance of the sample (A_{sample}) and standard ($A_{standard}$) was read against a reagent blank within 60 min. Glucose concentration was calculated as follows:

$$\textbf{Glucose content (mg/dL)} = (A_{sample}/A_{standard}) \times \textbf{Conc. of standard}$$

Resorcinol reagent method is used for measuring fructose content (Chemists and Horwitz, 1980). 1 mL resorcinol reagent was added in honey sample and mixed thoroughly, and then to this mixture 1 mL of dil. HCl was added. Standard fructose solutions containing 0.2, 0.4, 0.6, 0.8 and 1.0 mg fructose/ mL was also treated with 1 mL of the resorcinol reagent and 1 mL of dil. HCl as above. A blank solution was also prepared along with the standard. The test solution, the standard and blank were then heated in a water bath at 80°C for about 10min, the solution was then removed from the water bath, cooled by immersing in tap water for 5 min and then the absorbance of both the test and standard solution were read against the blank solution at 520 nm. The fructose contents of the honey samples were then extrapolated from a standard curve prepared using the absorbance of the standard.

Determination of mineral elements

Nine elements: Potassium (K), sulfur (S), sodium (Na), magnesium (Mg), iron (Fe), phosphorus (P) silicon (Si), calcium (Ca) and zinc (Zn) were measured by burning honey sample of 10g at 560°C overnight. The resulting ash was then dissolved in 10 mL of a mixture of HNO_3 (1 M) and HCl (1 M) (1:1), making the volume of the resulting solution up to 60 mL with deionized water (González Paramás *et al.*, 2000). Matrix modifiers were used for minerals that showed spectral interference like KCl and La_2O_3. KCl was added for measuring magnesium and calcium; and CsCl for sodium and potasium. Elemental analysis was done on AA-6200 absorption atomic spectrophotometer (Shimadzu, Japan). Analysis of each sample was done in triplicate.

Colour analysis

Colour characteristics was determined by using the CIE L*a*b* method, where lightness L* and two color coordinates, a* (redness–greenness) and b* (yellowness–blueness), were measured by means of a Konica Minolta CL-200A Chroma Meter (Konica Minolta, Tokyo, Japan).

Statistical analysis

All analyses were done in triplicate and the data were expressed as means ± standard deviations, which were calculated using Excel (Microsoft Office, Version 2007).

RESULTS AND DISCUSSION:

Physicochemical properties

The data obtained are presented in Table 1. Moisture content of honey samples varies between 16.22 to 18.89 % and they are in the water range limits (< 20 %) approved by the Codex Alimentarius (Alimentarius, 2001). Moisture content affects the storage life of honey. Higher moisture contents can result in unwanted honey fermentation during storage period, due to the action of osmotolerant yeasts, which result in development of carbon dioxide and ethyl alcohol. Ethyl alcohol is further oxidized to acetic acid thus resulting in a sour taste (Zamora *et al.*, 2006a). Water content in honey depends upon several factors like level of relative humidity during honey production, maturity of honey, temperature in hive and extraction technique used (Zamora *et al.*, 2006b).

The water activity (a_w) of the honey samples varies from 0.568 - 0.616 (Table 1). The values of a_w of honey can varies between 0.49 to 0.65, due to its relatively low water content and high content of sugars (glucose and fructose in particular) (Gleiter *et al.*, 2006; Zamora *et al.*, 2006a). The a_w is an important factor, as it effects the honey stability by limiting or preventing microbial growth. The minimal a_w of 0.6 is required for growth of osmotolerant yeasts (Abramovič *et al.*, 2008); therefore, the honey samples in this study identified as H5, H7, and H8, with a_w values of 0.610, 0.616 and 0.603 respectively could be candidates for fermentation.

Using the values of moisture and a_w, we found a linear correlation, which was as follows:

$$\mathbf{a_w = 0.26207 + 0.018795A}$$

where A is the moisture content (R^2=0.831).

This equation is parallel (intercept and slope) to that calculated by several other authors using honey from various countries (Cavia *et al.*, 2004; Zamora *et al.*, 2006a) . This correlation can be beneficial to calculate the a_w value using only the water content measurement, as measurement of moisture content is relatively easy and fast (by refractometry) as compared with the measurement of the a_w of honey.

Values of soluble solids content (expressed as °Brix) ranged from 79.0 to 81.7 (Table 1). These values are in agreement with those reported for other types of honey. The soluble solid content of honey samples from Portugal ranged from 79.0 to 82.2 °Brix, whereas, for honey samples from India ranged from 76.0 to 88.0 °Brix (Silva *et al.*, 2009).

Hydroxymethylfurfural (HMF) content is used as an indicator of the purity and quality of honey because fresh honeys don’t contain HMF. High level of HMF in honey shows overheating, improper storage conditions and aged honey. The analyzed honey samples in the study showed HMF content values ranged between 9.01 and 21.96 mg kg^{-1}. No honey sample in this experiment (Table 1) exceeded the international regulation which sets a level of maximum HMF to 40 mg kg^{-1} (Alimentarius, 2001). Other studies on honey from various countries have reported the HMF content value: Argentinean honey 14.8 mg kg^{-1} (Finola *et al.*, 2007), Turkish honey 7.26 mg kg^{-1} (Furkan Yardibi and Gumus, 2010), Portugal honey 6.5 mg kg^{-1} (Feás *et al.*, 2010).

Ash content is an indicator of the mineral content in honey. The analyzed honey samples in this study showed ash content values ranged between 1.18 to 4.11 g kg^{-1} (Table 1). Floral or blossom honey is honey type which is gained from the nectar of plants, whereas honeydew honey is honey type which is produced mainly from the excretions of plant sucking insects (Hemiperta) on the living part of plants or secretions of living parts of plants (Alimentarius, 2001). Generally, the ash content of blossom honey is ≤ 6.0 g kg-^{1} as compared to honeydew honey where this value is ≥ 12.0 g kg^{-1} (Ouchemoukh *et al.*, 2007). Thus based on this standard, the honeys analyzed in this work are classified as blossom honey. Ash content of honeys from other regions of the world ranging from 0.79 to 5.49 g kg^{-1} has been reported (Acquarone *et al.*, 2007; Ahmed *et al.*, 2007; Al *et al.*, 2009; Kahraman *et al.*, 2010). The variation in the ash content of different honeys could be because of the beekeeping techniques, process of harvesting, and the pattern of bee feeding (Finola *et al.*, 2007).

Electrical conductivity depend on botanical origin of honey along with other factors like mineral content, organic acids, some complex sugars, proteins and polyols (Terrab *et al.*, 2003). Electrical conductivity values for floral honey should have value < 0.8 mS cm-^{1}, whereas honeydew should have values > 0.8 mS cm^{-1} (Alimentarius, 2001). All honey samples had the conductivity values below 0.8 mS cm^{-1} which advocates that honeys tested in this work were of a floral origin. Conductivity values ranged from 0.325 to 0.737 mS cm^{-1} and the mean conductivity value for the eight honey samples used in this study was 0.436 mS cm^{-1} (Table 1). A linear relationship exist between the electrical conductivity and the ash content, which is expressed as $C = 0.14+1.74\ A$, where C is the electrical conductivity and A is the ash content (Alimentarius, 2001).In this study, the linear relationship between the electrical conductivity and the ash content was expressed as $C = 0.182+1.32\ A$ ($R^2 = 0.973$), where A is the ash content and C is the electrical conductivity. Other equations that describe the relationship between the electrical conductivity and the ash content have been reported (Downey *et al.*, 2005; Nasiruddin Khan *et al.*, 2006).

Acidity in honey is due to the occurrence of organic acids in honey, predominantly gluconic acid (Terrab *et al.*, 2002). Codex Alimentarius specify that free acidity of honey should not be more than 50 meq kg^{-1} (Alimentarius, 2001). The analyzed honey samples in the study showed values of total acidity from 17.3 to 36.6 meq kg^{-1} (Table 1). The average for total acidity of the honey samples of this study (29.2 meq kg^{-1}) was similar to the average (31.70 meq kg-1) reported for Turkish honey (Furkan Yardibi and Gumus, 2010), but lower (48.27 meq kg^{-1}) than the reported for Venezuelan honey (Vit *et al.*, 1998). Also, the total acidity value is lower (18.4 meq kg^{-1}) than that reported for Argentinan honey (Acquarone *et al.*, 2007). The acidity of the honey improves its antioxidant activity, contributes to flavor, and effects against the action of microorganisms (Cavia *et al.*, 2007). Variations in total acidity depends upon the floral source and harvest season (Ojeda de Rodríguez *et al.*, 2004).

Glucose and fructose are the major carbohydrates found in honey and accounts for about 65% to 85% of total soluble solids (De La Fuente *et al.*, 2011). According to Anklam (1998) the ratio between fructose and glucose contents in any honey type depends mainly on the source of the nectar, but White Jr (1978) gave average ratio between fructose and glucose i.e. 1.2:1 which was observed in this study (Table 2). All honey samples used in the study contained more fructose (371.9–411.1 g kg^{-1}) than glucose (304.1–334.4 g kg^{-1}). Honeys with high fructose/glucose ratio show less crystallization due to the change in the level of saturation of glucose in the presence of the higher quantity of fructose (White *et al.*, 1964). In addition, the fructose to glucose ratio also influence honey flavor, since fructose is much sweeter than glucose (Ojeda de Rodríguez *et al.*, 2004). All honey samples also met Codex Alimentarius standard (650 g kg^{-1}) of reducing sugars in honey (Alimentarius, 2001).

Mineral content

Beside the nutritional importance of minerals, mineral content also provide significant information about the geographical origin of honey and indicator of possible environmental pollution (Bogdanov *et al.*, 2004b; Conti, 2000; Pisani *et al.*, 2008). In current study, nine elements were measured: Potassium (K), Sodium (Na), Calcium (Ca), Magnesium (Mg), Phosphorous (P), Sulfur (S), Silicon (Si), Iron (Fe) and Zinc (Zn) (Table 3). K was the most abundant mineral found in all honey samples; with an average content of 716.7 mg kg^{-1} and content values ranging from 276.7 to 1760 mg kg^{-1}. Studies on mineral composition of honey from other geographical localities also exhibit potassium to be the most abundant element; K was also the richest element in honeys from Spain (1935 mg kg^{-1}), form Slavonia (2910 mg kg-1), from Portugal (2590 mg kg^{-1}), from brazil (3353 mg kg-1) and from Israel (3768 mg kg^{-1}) (Dag *et al.*, 2006; Kump *et al.*, 1996; Mendes *et al.*, 2006; Silva *et al.*, 2009)

With an average content of 118.5 mg kg^{-1}, Phosphorous was the second richest mineral found in the honey samples of this study, whose values ranged from 48.8- 394.6 mg kg^{-1}. This mineral also has been found as the second most abundant in honeys of many other botanical origin (Dag *et al.*, 2006; Fernández-Torres *et al.*, 2005). Ca, Na and Si contents in this study was found at average values of 57.4, 96.4, and 40.7 mg kg^{-1}, respectively (Table 3). Two honey samples (H5 and H6) presented the higher Na contents with values of 241.6 and 302.0 mg kg^{-1}. Calcium content in honey from other regions of the World has been found in values ranging from 32 to 270 mg kg^{-1} (Nanda *et al.*, 2003; Rodríguez García *et al.*, 2006). In current study, Ca content of honey samples varies from 37.6 to 127.3 mg kg^{-1} and an average content of 57.4 mg kg^{-1}. Si is another mineral element reported in honey samples by others (Dag *et al.*, 2006; González-Miret *et al.*, 2005) with an average content from 8.8 to 141 mg kg^{-1}, while the content values in this study ranged from 24.7 to 90.9 mg kg^{-1}.

Average value of Mg and S for honey in this work was parallel to the values reported for honey samples from other countries (Baroni *et al.*, 2009; Fernández-Torres *et al.*, 2005; Rodriguez-Otero *et al.*, 1994; Vanhanen *et al.*, 2011) however, very high relative values of Mg content have been reported (137–303 mg kg-1) in other studies (Rodríguez García *et al.*, 2006; Terrab *et al.*, 2004b) in comparison with the results obtained in this work (13.1–61.3 mg kg-1). On the other hand, average values for Zn and Fe ranged from 1.51 to 6.80 mg kg-1 and from 0.66 to 4.72 mg kg-1, respectively. Some studies have reported similar values of Zn for honey samples from other countries (Bogdanov *et al.*, 2007; Terrab *et al.*, 2004b), although a studies on honey from india reported 16.77 mg kg-1 Zn (Nanda *et al.*, 2003). Fe contents in honey samples of this study were lower than those found for honeys from Ireland (Downey *et al.*, 2005) and Morocco (Terrab *et al.*, 2003a), however, a similar average content was reported by (Bogdanov *et al.*, 2007) for honey from Switzerland.

Color characteristics

Color plays a vital role in acceptability of product as it is the major sensory property observed by the consumer. However, little is known about consumer color acceptability of honey. Bogdanov, Ruoff, and Oddo (2004a) reported that in Germany, Austria and Switzerland dark color honeys have more consumer acceptability. According to Murphy, Henchion and O'Reilly (2000) dark golden color is more preferred by Irish peoples. The color of honey is one of the most variable attributes and it depends upon botanical origin, along with other factors like ash content, temperature and time of storage as well as the presence of antioxidant pigments like flavonoids and carotenoids (Baltrušaitytė *et al.*, 2007; Terrab *et al.*, 2004). Honey samples having an L* value 450 are classified light colored honeys, while honeys having an L* value 550 are considered as darker honeys (González-Miret *et al.*, 2005). Therefore, according to this criterion, the Pakistan honeys from the Punjab province studied can be considered as dark honeys since the L* value of the honey samples varies from 21.18 to 35.26 (Table 4). The a* values ranged from 1.04 to 8.00 and b* values varies from 5.57 to 29.91, thus it is obvious that all the samples can be considered as darker and they have red and yellow components.

CONCLUSIONS:

In general, the Pakistani honey from the province of Punjab had a good level of quality according to the results obtained from their physicochemical analysis, which are in agreement with the international regulations. With respect to the color parameter L*, all honey samples can be classified as dark honeys. Mineral contents were found to be in the range of honeys from other countries. Potassium is the most abundant of the element.

REFERENCES:

Abramovič, H., M. Jamnik, L. Burkan and M. Kač. 2008. Water activity and water content in Slovenian honeys. *Food control*, 19 (11): 1086-1090.

Acquarone, C., P. Buera and B. Elizalde. 2007. Pattern of pH and electrical conductivity upon honey dilution as a complementary tool for discriminating geographical origin of honeys. *Food Chemistry*, 101 (2): 695-703.

Ahmed, J., S. Prabhu, G. Raghavan and M. Ngadi. 2007. Physico-chemical, rheological, calorimetric and dielectric behavior of selected Indian honey. *Journal of Food Engineering*, 79 (4): 1207-1213.

Al, M. L. et al. 2009. Physico-chemical and bioactive properties of different floral origin honeys from Romania. *Food Chemistry*, 112 (4): 863-867.

Alimentarius, C. 2001. Revised codex standard for honey. *Codex stan*: 12-1981.

AOAC. 1990. Official methods of Analysis of the AOAC. Association of Official Analytical Chemists Inc.

Baltrušaitytė, V., P. R. Venskutonis and V. Čeksterytė. 2007. Radical scavenging activity of different floral origin honey and beebread phenolic extracts. *Food Chemistry*, 101 (2): 502-514.

Baroni, M. V. et al. 2009. Composition of honey from Córdoba (Argentina): Assessment of North/South provenance by chemometrics. *Food Chemistry*, 114 (2): 727-733.

Bertoncelj, J., U. Doberšek, M. Jamnik and T. Golob. 2007. Evaluation of the phenolic content, antioxidant activity and colour of Slovenian honey. *Food Chemistry*, 105 (2): 822-828.

Bogdanov, S. 1997. Charakterisierung von Schweizer Sortenhonigen. Agrarforschung, 4: 427-430.

Bogdanov, S., K. Ruoff and L. P. Oddo. 2004a. Physico-chemical methods for the characterisation of unifloral honeys: a review. *Apidologie*, 35 (Suppl. 1): S4-S17.

Bogdanov, S., K. Ruoff and L. Persano Oddo. 2004b. Physico-chemical methods for the characterisation of unifloral honeys: a review. *Apidologie*, 35 (Suppl. 1): S4-S17.

Castro-Vázquez, L., M. Díaz-Maroto and M. Pérez-Coello. 2007. Aroma composition and new chemical markers of Spanish citrus honeys. *Food Chemistry*, 103 (2): 601-606.

Cavia, M., M. Fernáez-Muiño, J. Huidobro and M. Sancho. 2004. Correlation between moisture and water activity of honeys harvested in different years. *Journal of food science*, 69 (5): C368-C370.

Cavia, M. M., M. A. Fernández-Muiño, S. R. Alonso-Torre, J. F. Huidobro and M. T. Sancho. 2007. Evolution of acidity of honeys from continental climates: Influence of induced granulation. *Food chemistry*, 100 (4): 1728-1733.

Chemists, A. o. O. A. and W. Horwitz. 1980. Official methods of analysis. AOAC Washington, DC.

Conti, M. E. 2000. Lazio region (central Italy) honeys: a survey of mineral content and typical quality parameters. *Food Control*, 11 (6): 459-463.

Dag, A., O. Afik, Y. Yeselson, A. Schaffer and S. Shafir. 2006. Physical, chemical and palynological characterization of avocado (Persea americana Mill.) honey in Israel. *International journal of food science & technology*, 41 (4): 387-394.

De La Fuente, E., A. Ruiz-Matute, R. Valencia-Barrera, J. Sanz and I. Martinez Castro. 2011. Carbohydrate composition of Spanish unifloral honeys. *Food Chemistry*, 129 (4): 1483-1489.

De la Fuente, E., M. Sanz, I. Martínez-Castro, J. Sanz and A. Ruiz-Matute. 2007. Volatile and carbohydrate composition of rare unifloral honeys from Spain. *Food Chemistry*, 105 (1): 84-93.

Downey, G., K. Hussey, J. Daniel Kelly, T. F. Walshe and P. Martin. 2005. Preliminary contribution to the characterisation of artisanal honey produced on the island of Ireland by palynological and physico-chemical data. *Food Chemistry*, 91 (2): 347-354.

Feás, X., J. Pires, M. L. Estevinho, A. Iglesias and J. P. P. De Araujo. 2010. Palynological and physicochemical data characterisation of honeys produced in the Entre-Douro e Minho region of Portugal. *International journal of food science & technology*, 45 (6): 1255-1262.

Fernández-Torres, R. et al. 2005. Mineral content and botanical origin of Spanish honeys. *Talanta*, 65 (3): 686-691.

Finola, M. S., M. C. Lasagno and J. M. Marioli. 2007. Microbiological and chemical characterization of honeys from central Argentina. *Food Chemistry*, 100 (4): 1649-1653.

Furkan Yardibi, M. and T. Gumus. 2010. Some physico-chemical characteristics of honeys produced from sunflower plant (Helianthus annuus L.). *International journal of food science & technology*, 45 (4): 707-712.

Gleiter, R., H. Horn and H.-D. Isengard. 2006. Influence of type and state of crystallisation on the water activity of honey. *Food chemistry*, 96 (3): 441-445.

González-Miret, M. L., A. Terrab, D. Hernanz, M. A. Fernandez-Recamales and F. J. Heredia. 2005. Multivariate correlation between color and mineral composition of honeys and by their botanical origin. *Journal of agricultural and food chemistry*, 53 (7): 2574-2580.

González Paramás, A. M. et al. 2000. Geographical discrimination of honeys by using mineral composition and common chemical quality parameters. *Journal of the Science of Food and Agriculture*, 80 (1): 157-165.

Guler, A., A. Bakan, C. Nisbet and O. Yavuz. 2007. Determination of important biochemical properties of honey to discriminate pure and adulterated honey with sucrose (< i> Saccharum officinarum</i> L.) syrup. *Food chemistry*, 105 (3): 1119-1125.

Hammock, D. A. 2011. Medicinal Uses of Honey. *Nutrition*, 2011.

Jeffrey, A. E. and C. M. Echazarreta. 1996. Medical uses of honey. *Rev Biomed*, 7 (1): 43-49.

Kahraman, T., S. K. Buyukunal, A. Vural and S. S. Altunatmaz. 2010. Physico-chemical properties in honey from different regions of Turkey. *Food chemistry*, 123 (1): 41-44.

Kump, P., M. Nečemer and J. Šnajder. 1996. Determination of trace elements in bee honey, pollen and tissue by total reflection and radioisotope X-ray fluorescence spectrometry. *Spectrochimica Acta Part B: Atomic Spectroscopy*, 51 (5): 499-507.

Mendes, T. M., S. N. Baccan and S. Cadore. 2006. Sample treatment procedures for the determination of mineral constituents in honey by inductively coupled plasma optical emission spectrometry. *Journal of the Brazilian Chemical Society*, 17 (1): 168-176.

Nanda, V., B. Sarkar, H. Sharma and A. Bawa. 2003. Physico-chemical properties and estimation of mineral content in honey produced from different plants in Northern India. *Journal of Food Composition and Analysis*, 16 (5): 613-619.

Nasiruddin Khan, M., M. Qaiser, S. M. Raza and M. Rehman. 2006. Physicochemical properties and pollen spectrum of imported and local samples of blossom honey from the Pakistani market. *International journal of food science & technology*, 41 (7): 775-781.

Ojeda de Rodríguez, G., B. Sulbarán de Ferrer, A. Ferrer and B. Rodríguez. 2004. Characterization of honey produced in Venezuela. *Food Chemistry*, 84 (4): 499-502.

Ouchemoukh, S., H. Louaileche and P. Schweitzer. 2007. Physicochemical characteristics and pollen spectrum of some Algerian honeys. *Food Control*, 18 (1): 52-58.

Pisani, A., G. Protano and F. Riccobono. 2008. Minor and trace elements in different honey types produced in Siena County (Italy). *Food Chemistry*, 107 (4): 1553-1560.

Rodríguez García, J. C. et al. 2006. Preliminary chemometric study on the use of honey as an environmental marker in Galicia (northwestern Spain). *Journal of agricultural and food chemistry*, 54 (19): 7206-7212.

Silva, L. R., R. Videira, A. P. Monteiro, P. Valentão and P. B. Andrade. 2009. Honey from Luso region (Portugal): Physicochemical characteristics and mineral contents. *Microchemical Journal*, 93 (1): 73-77.

Terrab, A., M. J. Díez and F. J. Heredia. 2002. Characterisation of Moroccan unifloral honeys by their physicochemical characteristics. *Food Chemistry*, 79 (3): 373-379.

Terrab, A., L. González-Miret and F. J. Heredia. 2004. Colour characterisation of thyme and avocado honeys by diffuse reflectance spectrophotometry and spectroradiometry. *European Food Research and Technology*, 218 (5): 488-492.

Terrab, A., A. G. González, M. J. Díez and F. J. Heredia. 2003. Mineral content and electrical conductivity of the honeys produced in Northwest Morocco and their contribution to the characterisation of unifloral honeys. *Journal of the Science of Food and Agriculture*, 83 (7): 637-643.

Vit, P., L. P. Oddo, M. L. Marano and E. S. de Mejias. 1998. Venezuelan stingless bee honeys characterized by multivariate analysis of physicochemical properties. *Apidologie*, 29 (5): 377-389.

White, J., I. Kushnir and M. H. Subers. 1964. Effect of storage and processing temperatures on honey quality. *Food Technol*, 18 (4): 153-156.

Zamora, M. C., J. Chirife and D. Roldán. 2006a. On the nature of the relationship between water activity and % moisture in honey. *Food Control*, 17 (8): 642-647.

Zamora, M. C., J. Chirife and D. Roldán. 2006b. On the nature of the relationship between water activity and% moisture in honey. *Food control*, 17 (8): 642-647.

Table 1. Physicochemical parameters of honey samples collected from central region of Punjab, Pakistan

Honey Samples	Moisture %	Water Activity	Soluble solids (°Brix)	HMF (mg kg^{-1})	Conductivity (mS cm^{-1})	Ash (g kg^{-1})	Acidity (meq kg^{-1})
H1	17.14	0.577±0.001	81.2±0.10	9.01±1.79	0.345±0.013	1.27±0.011	27.8±0.76
H2	16.22	0.573±0.001	81.6±0.05	12.91±2.94	0.325±0.016	1.18±0.012	26.3±0.76
H3	17.52	0.591±0.001	80.3±0.06	21.96±5.04	0.615±0.014	3.34±0.010	36.6±2.02
H4	16.25	0.568±0.001	81.7±0.11	18.54±3.05	0.359±0.011	1.31±0.016	27.2±0.77
H5	18.03	0.610±0.001	79.0±0.05	14.17±0.93	0.512±0.020	1.53±0.019	17.3±0.83
H6	17.36	0.570±0.003	81.0±0.11	12.10±1.74	0.380±0.015	2.45±0.021	26.8±1.59
H7	18.89	0.616±0.002	79.1±0.15	10.12±2.32	0.430±0.010	1.96±0.014	35.5±1.60
H8	17.53	0.603±0.002	81.1±0.10	12.26±1.58	0.737±0.016	4.11±0.012	36.4±1.27
H9	17.46	0.580±0.001	82.4±0.2	10.87±0.80	0.583±0.012	2.02±0.02	30.0±1.27
H10	17.26	0.596±0.002	78.8±0.2	16.87±0.50	0.589±0.020	2.22±0.2	28.4±1.27
Mean	17.36	0.588±0.018	80.6±0.86	13.87±4.48	0.463±0.138	2.12±0.103	29.2±6.47

Note: Results are expressed as mean values ± standard deviation.

Table 2. Sugars mean composition in honey samples collected from central region of Punjab, Pakistan

Honey Samples	Fructose (g Kg^{-1})	Glucose (g Kg-1)	Fructose + Glucose (g Kg-1)	Fructose/ Glucose
H1	377.5±7.3	318.7±5.8	696.2	1.18
H2	411.1±5.2	311.3±4.2	722.4	1.32
H3	381.5±5.8	304.1±4.6	685.6	1.25
H4	384.3±2.5	334.4±12.7	718.7	1.15
H5	371.9±10.9	307.1±1.8	679.0	1.21
H6	381.1±9.8	325.6±7.5	706.7	1.17
H7	391.8±8.4	305.7±5.7	697.5	1.28
H8	372.8±5.8	321.0±2.8	693.8	1.16
H9	352.0±3.8	302.0±3.8	654.0	1.16
H10	416.0±2.8	340.0±2.3	756.0	1.22
Mean	384.0±13.2	321.0±2.8		

Note: Results are expressed as mean values ± standard deviation.

Table 3. Mineral element content of honey samples collected from central region of Punjab

Samples	K	Ca	P	Na	S	Mg	Fe	Si	Zn
H1	370.7± 10.8	37.6 ±1.4	57.9 ±2.0	24.9 ±3.0	14.1±4.3	17.3±1.0	0.91±0.13	24.7±1.1	1.52±0.11
H2	408.4 ± 8.7	50.3±4.4	83.7±13.0	35.5±3.2	22.4±4.4	21.9±2.2	0.83±0.09	33.9±3.7	1.59±0.16
H3	1380.3± 55.0	127.3±5.6	104.8±14.0	41.8±4.8	41.1±7.5	21.6±1.9	1.60±0.10	90.9±5.6	2.69±0.29
H4	561.3± 44.1	44.2±4.1	77.3±2.5	20.5±3.1	25.2±3.6	20.8±2.7	0.66±0.14	27.7±4.5	2.72±0.23
H5	276.7±44.1	58.6±4.9	48.8 ±47.8	241.6±48.2	24.7±4.5	17.8±2.4	1.12±0.10	50.7±3.5	4.94±0.24
H6	646.0±49.8	50.5±5.5	96.9±3.7	302.0±17.2	38.3±3.9	23.1±1.8	1.41±0.16	30.3±1.1	2.84±0.13
H7	430.6±26.3	39.0±3.8	83.9±4.1	43.3±4.6	21.6±3.2	13.1±1.8	1.33±0.31	25.9±1.9	1.75±0.21
H8	1760.0±36.1	51.4±1.4	394.6±58.5	61.3±4.1	62.9±6.0	61.3±2.0	4.72±0.28	26.9±1.8	6.80±0.45
H9	563.3±36.1	67.4±5.1	128.5±6.1	76.4±6.1	36.3±6.1	19.6±1.1	1.07±0.10	52.7±1.2	2.61±0.41
H10	870.0±26.1	47.4±3.1	108.5±5.1	116.4±16.1	26.1±3.1	29.6±2.1	2.07±0.15	28.7±1.2	3.61±0.32
Mean	716.7±530.7	57.4±28.0	118.5±109.6	96.4±106.5	31.3±15.5	24.6±14.6	1.57±1.26	40.7±21.3	3.11±1.77

Note: Results are expressed as mean values ± standard deviation.

Table 4. Colour characteristics of honey samples collected from central region of Punjab, Pakistan

Honey Samples	Colour Parameter		
	L*	a*	b*
H1	31.98±0.21	4.11±0.31	25.59±1.01
H2	27.72±0.17	5.45±0.43	20.46±1.92
H3	21.40±0.77	8.00±0.45	14.05±1.85
H4	35.26±2.60	2.07±0.07	29.91±0.21
H5	33.74±0.47	1.90±0.03	20.06±0.30
H6	25.89±1.04	1.04±0.09	5.57±0.87
H7	27.15±0.48	6.14±0.24	18.69±0.74
H8	21.18±0.93	6.33±0.20	9.66±0.10
H9	25.10±0.21	3.33±0.22	12.32±0.25
H10	31.22±0.22	5.45±0.31	7.0±0.31
Mean	28.16±4.74	4.39±2.87	9.66±0.10

Note: Results are expressed as mean values ± standard deviation.

Fig 1. Locations of honey samples collecting points