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# Estimation of chemical composition in different honey samples

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Abstract: Natural honey is one of the most widely used products due to its unique properties, hence is used for nutritional and medicinal purposes. The present investigation was aimed at comparing the chemical composition of honey samples of four different companies, namely, Dabur, Patanjali, Al-Shafi (UAE) and Balaji (Local) with that of natural honey in order to find such honey that is closer to the natural honey. In this study, Patanjali honey sample was found to be best among all the honey samples, as its contents (Reducing sugar- 67.2%, flavonoid- 0.4mg, phenolic content- 169.67mg, protein-1.8mg/ml and carbohydrate- 2.8mg/ml) were closer to the

natural honey sample (Reducing sugar- 49.5%, flavonoid-0.2mg, phenolic content- 56mg, protein- 1.9mg/ml and carbohydrate- 2.9mg/ml) as compared to other honey samples.

**Key words:** Reducing sugar, flavonoid, phenolic content, protein, carbohydrate.

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#### Introduction:

Honey has been used as a food and as a traditional medicine since ancient times. It is a natural substance produced by honey bees, *Apis mellifera*, from the nectar of blossoms or honeydews. Honey is a sweet natural food made by bees using water, pollen and nectar from flowers (Cantarelli et. al, 2008). Honey has been used as a remedy for burns, cataracts, ulcers and wound healing, because it has a soothing effect when initially applied to open wounds.

Honey is supersaturated solution of sugars (mainly 38 % fructose and 31 % glucose), 80-85%

 carbohydrates, 0.3% proteins, enzymes, amino acids, minerals, trace elements, vitamins and other phytochemicals. It also contains 15-17% water and 0.2% ashes. These chemical components are of great importance as they influence the quality, granulation, texture as well as the nutritional and medicinal efficacy of honey (David, 2007).

The major constituents of natural honey are nearly the same. However, the biochemical composition and physical properties of natural honey varies greatly according to the plant species on which the bees forage (Cantarelli et. al, 2008; Ebenezer and Olugbenga, 2010).

The properties of honey also vary depending on the differences in climatic condition and vegetation of the areas and the natural honey is one of the best widely sought products due to its unique nutritional and medicinal properties, which are attributed to the influence of the different groups of substances it contains (Buba et. al, 2013).

Honey is recognized as having different biological properties, including antioxidants. Phenolic acid and flavonoids are the main antioxidants in this apiary product. An important parameter of honey is its colour which reflects the floral source.

A wide range of minor constituents such as certain enzymes and amino acids that are present in the honey are known to have antioxidant properties. Antioxidants activity is the ability and potential of honey to reduce oxidative reaction within the food systems.

Honey also has disadvantages like it increases risk of cholesterol, diabetes and also lowers homocysteine, another blood marker associated with disease.

A number of companies sell honey under different brand names. The present investigation was aimed at the comparison of the chemical composition of the different honey samples with the natural honey and to select the best one for consumption.

#### Materials and Methods:

Five samples of honey viz., Patanjali (Indian brand), Dabur (Indian brand), Balaji (Local), Al-Shafi (Foreign brand) was collected and Natural honey was collected from rural area.

Reducing sugar content of honey samples was carried out by Layne Enyon method as described in AOAC (Association of Official Analytical Chemistry, 2000).

 $10 \mathrm{gm}$  of different samples of honey were taken in suitable titration flask and then dissolved into  $75 \mathrm{ml}$  of  $\mathrm{CO}_2$  free water. It was then titrated against standard sodium hydroxide solution using 4-6 drops of phenolphalein as an indicator. Readings were taken at the scale where pink color persisted for  $10 \mathrm{\,seconds}$ .

Acidity was calculated using the following formula:

Acidity = 
$$\frac{0.23 \text{ x V}}{\text{M}}$$

The total phenolic content was estimated using the Folin-Ciocalteau methods described by Singleton et al. (1999).

The total flavonoid content was estimated as described by Chang et al. (2002).

Protein content of the honey was estimated by Azeredo et al. (2003).

25 gm of different samples of honey were dissolved separately in 50 ml of 80% of ethanol. It was then filtered. The filtrate of hone.y extract was then centrifuged for 10 min at 2500 rpm. To 1ml

honey sample, 1ml of 15% of phenol and 5 ml conc. $H_2SO_4$  were added and left for 20 min. Absorbance was taken at 490 nm by UV spectrophotometer.

Sample Preparation: 1 gm of each of the honey samples was weighed separately. 20 ml of conc. H<sub>2</sub>SO<sub>4</sub> was added to it. 10 ml of conc. HNO<sub>3</sub> was added to it. The sample mixtures were kept in 250 ml glass beakers. The samples were digested at 80°C for 3 h. The filtrates were collected and transferred to 100 ml volumetric flask. The volume was made up to the mark with the help of distilled water. The samples were analyzed for presence of heavy metals using AAS (Atomic Absorption Spectroscopy).

Sample Preparation: Small quantity of each of the honey samples, was taken in mortal and pestle. Potassium bromide of IR grade was added to the samples. Samples were mixed well until homogenous mixtures were obtained. The pellet of samples was prepared with the help of dye-punch and analyzed for presence of functional groups using FT-IR (Fourier Transform Infrared Spectroscopy).

#### Results and Discussion:

Maximum amount of reducing sugar was found in Patanjali (67.2 %) followed by Dabur (53.3 %); and minimum amount of reducing sugar was found in Balaji (25.5%). The results are shown in Figure 1.

It was found that Balaji honey sample had maximum acidity (0.62%) and natural honey had minimum acidity content (0.41%); and rest of the honey samples had values in between. The results are shown in Figure 2.

The total phenolic content varied greatly among the different honey samples. The lowest

value of phenolic content was found in Balaji honey sample (48.34 mg) whereas the highest value (169.67 mg) was obtained for Patanjali. The results of total phenolic content for all the specified honey samples are shown in Figure 3.

The value of total flavonoid content in honey samples varied from 0.25-3.34 mg with the lowest and highest value observed in Natural and Balaji, respectively. The data are shown in Figure 4.

It was found that the Natural honey contained the highest amount of protein content (1.9 mg/ml); followed by Patanjali (1.8 mg/ml); and lowest in Balaji (1.3 mg/ml). The data are presented in Figure 5.

It was found that the Natural honey contained the highest amount of carbohydrate content (2.9 mg/ml); followed by Patanjali (2.8 mg/ml); and was lowest in Balaji (2.3 mg/ml). The data are presented in Figure 6.

It was found that maximum content of calcium was present in Patanjali (1.337 $\mu$ g/ml); followed by Natural honey (1.263  $\mu$ g/ml) and minimum calcium content was found in Al-Shafi (0.963  $\mu$ g/ml). Calcium content of different honey samples are shown in Figure 7.

It was found that maximum content of zinc was present in Al-Shafi (0.419μg/ml); minimum amount of zinc was found in Natural honey (0.350μg/ml); followed by Patanjali (0.357μg/ml). Figure 8 shows the amount of zinc content in different honey samples.

It was found that amount of iron in Ai-Shafi was 0.075  $\mu$ g/ml; in Patanjali 0.040  $\mu$ g/ml; in Dabur 0.084  $\mu$ g/ml; in Balaji 0.095  $\mu$ g/ml; and in Natural honey it was 0.072  $\mu$ g/ml as shown in Figure 9.

Infrared spectroscopy analysis showed the presence of alkyl halide, alkene, anhydrous carbonyl, acid, amine, alkane, amide and aromatic compounds in Al-shafi sample. The related graph is shown in Figure 10.

In Patanjali sample, alkyl halide, alkene, amine, ester, amide and aromatic compounds were present. The related graph is shown in Figure 11.

In Dabur sample, alkenes, ester, alkylehylide, alcohol, amine and aromatic compounds were present. The related graph is shown in Figure 12.

In Balaji sample, alkyl halide, amine, acid, amide, ester and aromatic compounds were present. The related graph is shown in Figure 13.

In the Natural honey sample, alkene, ester, alkyl halide, alcohol, and aromatic compounds were present. The related graph is shown in Figure 14.

#### **Conclusion:**

The present investigation shows that the five honey samples namely Al-Safi, Natural, Dabur, Balaji, Patanjali contained phenolic compounds, flavonoids, acidic compounds and reducing sugar. The different chemical components of four different honeys were estimated and compared with the Natural honey. Patanjali honey sample was found to be best among all honey samples, as its contents were closer to that of the Natural honey sample.

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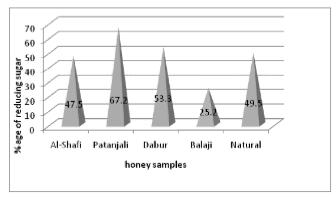


Fig. 1. Percentage of reducing sugar of different honey samples

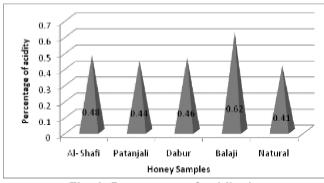


Fig. 2. Percentage of acidity in different honey samples

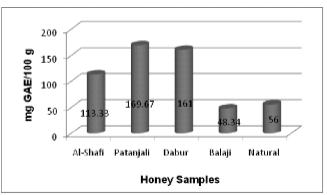


Fig. 3. Phenolic content of different honey samples

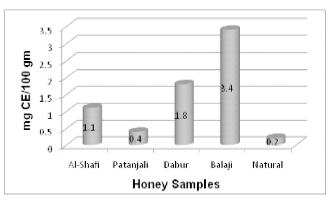


Fig. 4. Total flavonoid content of different honey samples

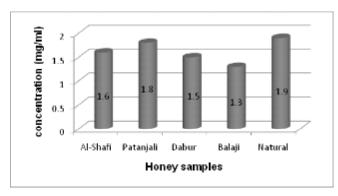


Fig. 5. Protein content of different samples of honey

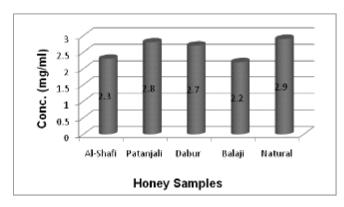


Fig. 6. Carbohydrate content of different samples of honey

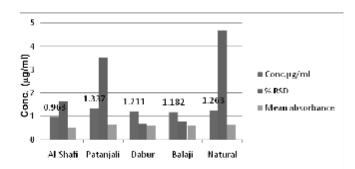


Fig. 7. Calcium content in different honey samples

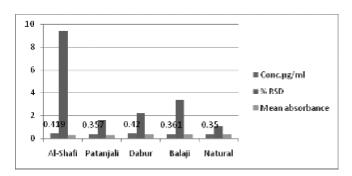


Fig. 8. Zinc content in different honey samples

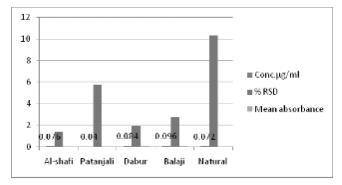


Fig. 9. Iron content in different honey samples

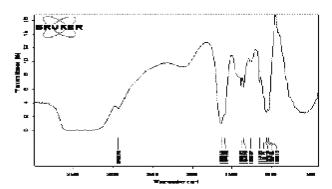


Fig. 10. Detection of functional groups in Al-shafi honey

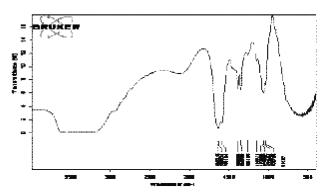


Fig. 11. Detection of functional groups in Patanjali honey

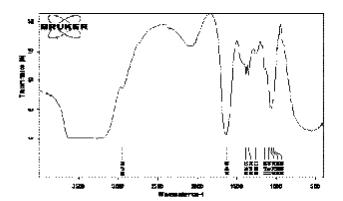


Fig. 12. Detection of functional group in Dabur honey

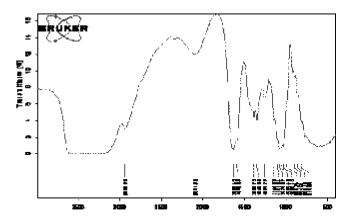


Fig. 13. Detection of functional group in Balaji

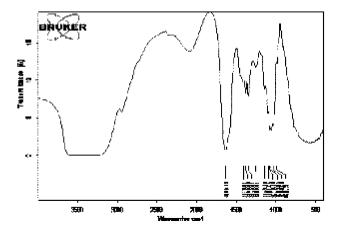


Fig. 14. Detection of functional group in Natural honey

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