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Biochemical Analysis of Different Brands of Unifloral Honey Available at the Northern Region of Bangladesh

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The research work was conducted to investigate the biochemical composition of five different brands of unifloral honey. The honey from Litchi (*Litchi chinensis*), Kadom (*Anthocephalus cadamba*), Til (*Sesamum indicum*), Plum (*Zizyphus mauritiana*) and Mustard (*Brassica campestris*) were used. This paper includes, determination of pH, specific gravity, solubility, moisture content as well as the total and reducing sugar, total protein content, water soluble protein, lipid, ash, dry matter, vitamin A and C and mineral content. It was found that the total content of sugar 77.7-80.30%, reducing sugar 62.30-65.02%, protein 0.30-0.33%, lipid 134-146mg 100 gm⁻¹, vitamin-C 4.8-5.3mg 100 gm⁻¹, pH 3.30 to 4.20, ash 0.65 to 0.80 and specific gravity between 1.33 to 1.36.

Key words: unifloral honey, biochemical analysis

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Introduction

Honey is the most familiar and widely distributed delicious food of Bangladesh. It has an important role as a source of nutrients in human diet, which are necessary for maintaining proper health. Honey is also an excellent adjuvant for acceleration of wound healing (Beraman *et al.*, 1983) and is widely accepted in folk medicine. Honey can be used as a medicine or a sick-diet for the treatment of ulcer and inflammation of stomach (Ioyrish, 1956). The unifloral honey from Litchi, Mustard, Kadom, Plum and Til showed significant antibacterial activity against the test pathogenic bacteria (Khalil *et al.*, 2001). A number of chemical parameters including water activity, free amino acid composition, reducing sugars, total sugars and pH were varied with the origin of honey. In honey, the sugar component was 886.2mg gm⁻¹ honey out of which 32.58% was fructose, 23.40% was glucose and 44.02% was maltose. Whereas, in the honey from the honey stomach sugar component was found to be 20.21mg gm⁻¹ honey, 36.33% being fructose, 29.09% was glucose and 24.58% was sucrose where Bangladeshi honey contain 75-80% sugar in which 65% reducing sugar (Muda, 1985).

Therefore, this paper describes the biochemical analysis of unifloral honey from Litchi (*Litchi chinensis*), Kadom (*Anthocephalus cadmba*), Til (*Sesamum indicum*), Plum (*Zizyphus mauritiana*) and Mustard (*Brassica campestris*) flowers available at the Northern region of Bangladesh.

Materials and Methods

Materials: Five unifloral honey such as honey from Litchi (*Litchi shinensis*), Kadom (*Anthocephalus cadmba*), Til (*Sesamum indicum*), Plum (*Zizyphus mauritiana*), and Mustard (*Brassica campestris*) flowers were collected from BCSIR Laboratory, Rajshahi, Bangladesh.

Production of Unifloral Honey: The unifloral honey is produced by a special procedure from particular kinds of flowers (Khalil *et al.*, 2001)

Determination of pH: pH of the different unifloral honey were determined using electrode assembly pH meter. Standard buffer solution of pH 7.0 or 4.0 were used for this purpose.

Determination of Biochemical parameters:

Determination of specific gravity: The specific gravity of honey was determined by the method of Kalimuddin (1976).

$$\text{Specific gravity of honey} = \frac{\text{Wt. of honeys (gm)}}{\text{Wt. of distilled water (gm)}} \times 100$$

Determination of moisture contents of honey: Moisture content was determined by weighing honey in a porcelain crucible. The crucible with the sample was heated in an electrical oven for about 6 hours at 100°C. It was then cooled in a desiccator and weighed again.

$$\% \text{age of moisture content} = \frac{\text{Weight of moisture obtained (gm)}}{\text{Weight of honey (gm)}} \times 100$$

Determination of ash content of honey: Ash content was determined following the method of A. O. A. C. (1980).

$$\% \text{age of ash content} = \frac{\text{Weight of ash obtained (gm)}}{\text{Weight of honeys (gm)}} \times 100$$

Determination of total protein content of honey: Protein content of the different varieties of unifloral honey was determined by the method of Micro-Kjeldahl. The nitrogen content was calculated using the formula given below:

$$1 \text{ ml of } 0.01\text{N H}_2\text{SO}_4 = 140\mu\text{g of nitrogen in NH}_3$$

Determination of water soluble protein content of honey: Water soluble protein content of honey was determined following the method of Lowry *et al.* (1951).

$$\% \text{ age of protein content} = \frac{\text{Amount of the protein obtained (gm)}}{\text{Weight of honeys (gm)}} \times 100$$

Determination of lipid content of honey: Lipid content of the different varieties of honey was determined by the method of (Bligh and Dyer, 1959).

$$\% \text{ age of lipid content} = \frac{\text{Amount of lipid obtained (gm)}}{\text{Weight of honeys (gm)}} \times 100$$

Determination of total sugar content of honey: Total sugar content of honey was determined calorimetrically by the anthrone method as described in Laboratory Manual in Biochemistry (Jayaraman, 1981).

The amount of total sugar was calculated from the standard curve of glucose.

$$\% \text{ age of total sugar} = \frac{\text{Amount of sugar obtained (gm)}}{\text{Weight of honey (gm)}} \times 100$$

Determination of reducing sugar content of honey: Reducing sugar content of the honey was determined by dinitrosalicylic acid method (Miller, 1972).

$$\% \text{ age of reducing sugar} = \frac{\text{Amount of reducing sugar obtained (gm)}}{\text{Weight of honeys (gm)}} \times 100$$

Estimation of Vitamin-A: Vitamin -A can be determined by Carr-price reaction. In this reaction Vitamin -A form blue color with Trichloro Acetic Acid (TCA) solution in chloroform (CHCl₃). Then a colorimetric or a spectrophotometric method determines the density of the color.

For standard sample: Take 0 to 1.0ml of the standard Vitamin solution in different test tubes and make volume to 1.0ml with CHCl₃. Add 2.0ml of TCA solution in Chloroform (CHCl₃) to each tube and read the absorbency at 620nm against blank, which contains only solvent.

For honey sample: Add 2ml of 95% ethanol (C₂H₅OH) and 4.0ml of petroleum ether (C₂H₅-O-C₂H₅) to the supplied sample. Shake for 3 minutes and take off all the petroleum ether layers. Evaporate to dryness under N₂ gas or by fan. Dissolve in 1ml CHCl₃, then add 2ml of TCA solution and take readings immediately.

Determination of Vitamin-C content of honey: Vitamin-C

content of the honey was determined by the titrimetric method (Bessey and King, 1933).

Determination of mineral content of honey:

(A) Calcium: Calcium content of the honey was determined by the method as described in Practical Physiological Chemistry (Vogel, 1978).

(B) Iron: Iron content of the honey was determined spectrophotometrically by Thiocyanate method as described in Practical Physiological Chemistry (Vogel, 1978).

Results and Discussion

The following was the first reported study on the biochemical analysis of some Bangladeshi unifloral honey. In this work an attempt was made to assess some physical and chemical properties of honey, which are important to the quality of the product.

pH of honey: The pH of all brands of honey was studied within acidic range. The range of the pH of five brands of unifloral honey was from 3.30-4.20 (Table 1). The Litchi honey was minimum acidic i.e. pH 3.30. Whereas the Plum honey was in the lower acidic range of pH 4.2. The pH of others honey i.e. Mustard, Til and Kadom honey were 3.35, 3.50 and 3.85 respectively. From the results it may be concluded that the acidity depends on the sources.

Specific gravity: Table 1 showed that the range of the specific gravity of the unifloral honey was 1.33-1.36. The specific gravity was highest for the Litchi honey i.e., 1.36 and lowest for Plum honey (1.33). The specific gravity of others honey i.e., Mustard, Til and Kadom honey were 1.35, 1.35 and 1.34, respectively.

Moisture content of honey: The moisture content was found to be varied between 13.3-14.8% it was highest for the Plum honey i.e. 14.8% and lowest for Litchi honey i.e. 13.3%. The moisture content of Mustard, Til and Kadom honey were 13.7, 14.3 and 14.5%, respectively (Table 1). The results also revealed that the moisture content of honey varied with the sources of honey.

Dry matter content of honey: The results (Table 1) showed that the dry matter content of five brands of unifloral honey was varied depending on their source. Litchi honey contains large amount of dry matter (86.7%) and the dry matter of Mustard, Plum, Til and Kadom honey were 86.3, 85.2, 85.7 and 85.5%, respectively.

Ash content of honey: The ash content of honey was very little comparatively. The range of ash content of five brands of unifloral honey was 0.655-0.80%. The ash content was highest for Til honey i.e. 0.80% and this was lowest for Litchi honey i.e. 0.655%. The ash content of others honey i.e. Mustard, Plum and Kadom honey were 0.678, 0.79 and 0.70%, respectively (Table 1).

Total protein and water soluble protein content of honey: The total proteins content of honey are shown in Table 2. The Plum honey contains highest amount of protein than that of others and Kadom honey contains less amount of protein. From the results it was found that honey might be used as a good source of protein. The protein content determined by Micro-Kjeldahl method showed a considerably higher value than that given by the Lowry method. This is because Lowry

Table 1: The value of pH, specific gravity, moisture, dry matter and ash of different brands of unifloral honey

Test honey	pH	Specific gravity	Moisture (%)	Dry matter (%)	Ash (%)
Litchi	3.3	1.36	13.3	86.7	0.655
Mustard	3.35	1.35	13.7	85.5	0.678
Plum	4.2	1.33	14.8	86.3	0.79
Til	3.5	1.35	14.3	85.2	0.80
Kadom	3.85	1.34	14.5	85.7	0.70

Table 2: Protein content of the different brands of unifloral honey

Test honey	Total protein content (%)	Water soluble protein (%)
Litchi	0.655	0.43
Mustard	0.74	0.46
Plum	0.744	0.54
Til	0.699	0.47
Kadom	0.656	0.48

method of protein estimation was applied to water extract of honey and took into account, in this case, the water soluble proteins only. Further, Kjeldahl method takes into account of both the protein and non-protein nitrogen.

Total lipid content of honey: Lipid content of the honey is presented in Table 3. The data indicated that honey contained very little amount of lipid, so it cannot be considered as a source of lipid also.

Table 3: Total lipid content of different brands of unifloral honey.

Test honey	Lipid content (mg 100gm ⁻¹)
Litchi	142
Mustard	146
Plum	136
Til	144
Kadom	134

The range of the total lipid content of five different brands of honey was 134-146mg 100 gm⁻¹. The total lipid content was highest for the Mustard honey i.e. 146mg 100 gm⁻¹ and the total lipid content was lowest for Kadom honey i.e., 134mg 100gm⁻¹.

Total sugar content of honey: Total sugar content of the tested unifloral honey is presented in Table 4. The total sugar content of five brands of honey was 77.7-80.3% it was highest for the Litchi honey i.e. space 80.3% and the total sugar content was lowest for Plum honey i.e., 77.7%. The data indicated that honey contained a very large amount of sugar so it can be considered as a good source of sugar.

Table 4: Total sugar content of five different brands of unifloral honey

Test honey	Total Sugar content (%)	Reducing sugar content (%)
Litchi	80.3	65.02
Mustard	79.8	63.20
Plum	77.7	62.30
Til	78.6	64.20
Kadom	78.4	64.40

Reducing sugar content of honey: It was found that all brands of unifloral honey contained high amount of sugar and the content of sugar changes with the changes of sources. The range of reducing sugar content of five brands of unifloral

honey was 62.30-65.02%. The reducing sugar content was highest for the Litchi honey i.e., 65.02% and the reducing sugar content was lowest for Plum honey i.e., 62.30%. The reducing sugar content of others honey i.e. Mustard, Til and Kadom honey were 63.20, 64.20 and 64.40%, respectively (Table 4).

Vitamin-A content of honey: All the test unifloral honey contains very low amount of vitamin-A. So, it is not used as a source of vitamin-A.

Vitamin-C content of honey: The amount of vitamin-C found in the different brands of unifloral honey is given in Table 5. From the results it was known that Litchi honey content comparatively higher concentration of vitamin-C, This might be due to the presence of vitamin-C in Litchi.

Table 5: Vitamin-C content of different brands of unifloral honey

Test honey	Vitamin C (mg 100 gm ⁻¹)
Litchi	6.25
Mustard	4.2
Plum	4.6
Til	4.6
Kadom	4.8

The range of vitamin-C content of five different brands of honey was 4.2-6.25mg 100 gm⁻¹. The vitamin-C content was highest for the Litchi honey i.e. 6.25mg 100 gm⁻¹ and the vitamin-C content was lowest for Mustard honey i.e. 4.2mg 100 gm⁻¹.

Minerals content of honey: Very little amount of calcium (Ca) and iron (Fe) were present in the different brands of honey. So, it is not sufficient to take honey as a source of Ca and Fe (Table 6). The range of Ca content was 6.95-8.11 mg 100 gm⁻¹ and Fe was 1.56-1.95mg 100gm⁻¹ of five

Table 6: Ca and Fe content of honey at five different brands of unifloral honey

Test sample	Ca (mg 100 gm ⁻¹)	Fe (mg 100 gm ⁻¹)
Litchi honey	7.20	1.95
Mustard honey	6.80	1.56
Plum honey	8.11	1.78
Til honey	7.23	1.85
Kadom honey	6.95	1.63

brands of honey. The Ca content was highest for the Plum honey i.e., 8.11mg 100 gm⁻¹ and the Fe content was highest for Litchi honey i.e., 1.95mg 100 gm⁻¹. The Ca content was lowest for the Kadom honey i.e. 6.95mg 100 gm⁻¹ and content was lowest for the Mustard honey i.e., 1.56mg 100 gm⁻¹.

From the research work, it was conclude that the chemical composition of the unifloral honey changes with the source and this changes the nutritive and medicinal value, which specifies the therapeutic value.

References

- Beraman, A, J. Yenai, D. Bell and M. P. David, 1983. Acceleration of wound healing by topical application of honey. *Am. J. Surg.*, 145: 374-76.
- Bessey, O. A. and C. G. King, 1933. The distribution of vitamin-C in plant and animal tissues and its determination., *J. Biol. Chem.*, 103: 687.
- Bligh, E. G. G. and W. Dyer, 1959. Total lipid extraction and purification., *Can. J. Biochem. physiol.*, 37: 911.
- Jayaraman, J., 1981. *Laboratory Manual in Biochemistry* (1st ed.) Wiley Eastern Ltd. New Delhi.
- Kalimuddin, M., A., 1976. *Text Book of Practical Physics* (4th Ed.) Bangladesh, 84.
- A. O. A. C., 1980. *Official methods of Analysis*, 13th Ed., Association of official Analytical chemists, Washington, D.C.
- Khalil, M. I., M. A. Mottalib, A. S. M. Anisuzzaman, Z. S. Sathi, M. Shahjahan, 2001. *Antibacterial Activities of Different Brands of Unifloral Honey Available at the Northern Region of Bangladesh*. The Sciences, Pakistan. (In Press).
- Lowry, O. H., N. J. Rosebrough, A. L. Farr and R. J. Randall, 1951. Protein measurement with the Folin-Ciocalten's reagent., *J. Biol. Chem.*, 193: 265-275.
- Muda, M. Z. B., 1985. Determination of sugar component the nectar honey from the honey stomach of the bee by HPLC. Serdang, Selngor, (Malaysia).
- Miller, G. L., 1972. Use of Dinitrosalicylic acid reagent for determination of reducing sugar. *Anal. Chem.*, 31: 426-428.
- Ioyrish, N. P., 1956. *Lechebnye svoistva meda ipchelinogo yada* (curative Properties of honey and Bee Venom), 3rd edition, Moscow.
- Vogel, A. I., 1978. *Vogel's Text book of Quantitative Inorganic Analysis* (4th Ed.) Longman Group Limited, England, pp: 741, 787, 808,

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