STUDY OF PHYSICOCHEMICAL PROPERTIES OF WILD HONEY SAMPLE FROM SANGAMNER TALUKA OF MAHARASHTRA

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ABSTRACT
Honey is the thick (opaque) viscous liquid produced by honey bees from the nectars of flowers. It is used in breakfast to retain energy in daily life. Honey’s nutritional value is determined by the amount of glucose & minerals present in honey. It is dependant on the season of honey. The physico-chemical features of honey may be used to determine overall quality of the product.

The physico-chemical characteristics of wild honey sample Collected from Sangamner Taluka of Maharashtra State are investigated in the current research to determine overall quality of honey.

Honey samples from to villages in the Sangamner Taluka were used in investigation. The moisture content, ash content, PH, colour intensity, electrical conductivity, viscosity, specific gravity, glucose, sucrose, fructose, and total protein content of the collected samples were all determined by using various methods. Those honeys with low moisture content, high glucose content & high mineral content have a high calorific value have longer shelf life than others. The findings of the research indicates that the wild honey samples collected are of high quality & which is gives high nutritional content.

The outcome of this research the nutritious benefits of honey in the daily diet & suggest that it might be utilized as a source of extra income for farmers.

KEY WORDS : Honey, physical properties, chemical propertie Quality, Glucose.

INTRODUCTION
Honey, one of the major products, is a sweet Viscous natural Fluid made from the nectar of plants Properties and compositions of bee honey depends on its geographical Floral origin, season & enviromental Factors [1]. Honey bees (Apis mellifera) collect a liquid secretion from flowers, called nectar [2-5]. Honey & beekeeping have a long history in India. Honey was the first Sweet food tasted by the ancient Indian inhabiting rock shelters & forests [6-7].

The genus name Apis is Latin for 'bee' & 'mellifera' is the latin for honey bearing referring to the species production of honey for the winter [8-9]. All India Beekeepers Association was formed during 1938-39 and the First Beekeeping research station was established in Punjab in 1945 by the Indian council of agricultural research. The production of honey in India increased significantly towards the late 1990s. 70% of honey production comes from informal segments [10-11]. Honey is the major bee product which has important nutritional value & provide significant economic contributions. Honey has become one of the most commercial agriculture products in many countries in the World [12-13].

The physicochemical properties of honey are helpful for the comparison of natural honey samples from different locations. The physico chemical properties provide the parameters for characterization & classification of honey [14].

The present study deals with the different Physico chemical parameters of honey samples found in three different locations of three villages (chincholi gurav, Talegaon Dighe, Paregaon BK) of a Sangamner Taluka.

The sweet substance produced by honey bees from the nectars or from secretion on living plants which bees collect, transform & Store in honey combs[15]. It is concentrated aqueous Solution of Invert sugar that comprises a mixture of other compounds like carbohydrate amino & organic acids aromatic substances minerals pigment Waxes & Pollen grains to make it complex[16,17,18].
The quality of honey sample depends on various Physiological factors such as climate, soil etc. Honey contains sugar, protein, moisture, vitamins,minerals, Enzymes polyphenus. [19]. Honey is sweet & viscous substance made by several bee honey bees. C the best known of which are honey bees [20-21]. Honey is sweet because of it's high concentration of the monosaccharides Fructose & glucose. It has about the same relative Sweetness as sucrose (table sugar) [22-23].

METHODS

Collection of Sample- In collection of samples Honey sample were collected from three different location of villages in Sangamner taluka, during September 2021 – October 22. Total 12 different honey samples were collected as follows . 6 from agricultural area, 4 from forest area, 2 from road sides. Area wise honey sample collected in equal quantities i.e.100 gm each& this collected honey sample put in air tight sterilized Plastic containers. They were labelled, brought to the laboratory & stored at 0.4°c until analysis completed [24].

1) Agricultural Area- In agricultural area there are 6 honey sample were collected. Farmers engaged in the cultivation of traditional crops likes onion, Wheat, gram, Sorghum, maize, Soybeans etc. in this area provided & independent system in which the bees assist to carryout cross pollination. This agricultural area includes location viz, Chincholi, Paregav, Talegaon Dighe. The distance between the two sampling is approximately 1-10 km [25].

2) Forest area- The Paregaon Khurda is situated 12 km away from sangamner Taluka. The forest is famous for woody plants, frost Irrigation, Gravel crusher project etc. There are total 4 samples of honey collected from the paregaon Khurda forest area. The following wild plants species were found in forest area Acacia (bavul), neem tree (Azadirachta indica), chinch (Tamarindus indica) .Aarla (phyllanthus emblica)[26].

3) Road side area- There are 2 honey sample from road side area. The distance between these two collected sample near about 25 meter from the road side. This road side area location viz, Talegaon Dighe, Paregaon Khurda, chincholi gurav. The distance between the two sampling is near by 1-4 km [27].

<table>
<thead>
<tr>
<th>Std for DNSA method (sr.no.)</th>
<th>Conc.of glucose (μmol)</th>
<th>Amount of working solution (μl)</th>
<th>Volume of DW (μl)</th>
<th>Amount of DNSA (ml)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>1</td>
<td>100</td>
<td>1400</td>
<td>3</td>
</tr>
<tr>
<td>2</td>
<td>2</td>
<td>200</td>
<td>1300</td>
<td>3</td>
</tr>
<tr>
<td>3</td>
<td>3</td>
<td>300</td>
<td>1200</td>
<td>3</td>
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<tr>
<td>4</td>
<td>4</td>
<td>400</td>
<td>1100</td>
<td>3</td>
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<tr>
<td>5</td>
<td>5</td>
<td>500</td>
<td>1000</td>
<td>3</td>
</tr>
<tr>
<td>6</td>
<td>6</td>
<td>600</td>
<td>1500</td>
<td>3</td>
</tr>
</tbody>
</table>

Physicochemical Analysis
Honey samples were analyzed for pH,electrolyte conductivity, moisture content, protein content, Glucose, fructose & Fructose Glucose ratio. This physicochemical analyses done following AOAC method. (Association of Official Agricultural chemists) [28].

Physicochemical Parameters-

1) Determination of pH

The pH of honey sample was measured by pH-meter. The pH of 10% WIV solution of honey sample prepared boiled warm water & measured pH of this given sample by using pH-meter instrument. The pH meter was calibrated by using externally standard buffer of pH 4.0, 7.0 & pH 9.0 to measuring the pH of sample.

2) Determination of electrical conductivity

The electrical conductivity was determined by a conductivity meter (model No:660B). Electrical conductivity meter was first calibration with water and then conductivity meter was dipped into honey solution (10%) & reading was noted after stable of instrument. Honey contains electrolytes, in the form of acids and minerals, it exhibits varying degrees of electrical conductivity. Measurements of the electrical conductivity are used to determine the quality of honey in terms of ash content [29].

3) Determination of moisture content

The moisture content of honey samples were determined by using refractometer reading at 20°C & Percentage moisture obtained from AOAC Standard moisture was determined by using refractometer . The refractometer was calibrated by adjusting zero. After that 2-3 drops of honey noted in the record for all honey samples.

4) Determination of Total Reducing sugar Assay:

3, 5 Dinitrosalicylic acid (DNSA, IUPAC Name) 2-hydroxy-3, 5- dinitrobenzoic acid) is an aromatic compound that reacts with reducing sugars & Other reducing molecules to form 3-amino-5-nitosalicylic acid, which absorbs light strongly at 540 nm (In case of glucose).

Procedure : Take three test tube of 20 ml & took an amount of glucose Stock Solution in each test tube as per table given below prepared a blank, in which case added 500 μl of DW instead of sample for incubation. Put 15 min in water bath. Add 3 ml ONSA kept tube.(Glucose solution + DW+ DNSA) in boiling H2O bath for 15 min. Cool at room temperature & Absorbance was measure at 540 nm.
5) Determination of Glucose, Fructose & Fructose-Glucose ratio

Glucose Percentage is determined iodometrically in a weak alkaline medium & the value is Subtracted from reducing sugar percentage to arrive of fructose percentage & Fructose-glucose ratio.

\[
\text{Glucose} \% = \text{Normality of sodium thiosulphate solution} \times (B-S) \times 0.009005/(0.1N \times \text{Wt of sample}) \times 100.
\]

\[
\text{Fructose} \% = \text{Reducing sugar} \% - \text{Glucose} \%
\]

\[
\text{Fructose Glucose ratio} = \frac{\text{Fructose} \%}{\text{Glucose} \%}
\]

6) Determination of sucrose content

In this type of determination sucrose content of the honey samples determined according to the procedure of Lane & Eynon method (1923)[30]. 2.6 gm of honey were weighed & then transferred to a 500ml volumete flask. standardized Fehling A & B solution were transferred to a 250ml conical flask, with 7ml H2O & 15 ml of honey solution. The conical flask was heated & 1ml methylene blue was added. titration was carried out adding the diluted honey solution until the indicator was decoloured. Determining sucrose content was carried out by inversion, adding to mi dil HCl, 50 ml dilute honey solution & H2O to 100ml Volumetric flask. Heating in H2O baththen cooling & dilute upto mark. Finally, the Lane-Enyon method way applied and sucrose content was obtained by difference.

Apparent sucrose= (Invert sugar 100 gm honey after inversion) – (sugar content before inversion) x 0.95

The result were expressed as gm apparent sucrose/100g honey.

7) Determination of lipid content

Lipid content was determined by Bligh & Dyer (1959) method[31]. Homogenize the sample 20 g with 16ml D.W. 40 ml of chloroform and 80 ml of methanol at the speed of 9500rpm for 1 min at 4 degree c. Add 40 ml chloroform and homogenize for 30 seconds. After centrifugation of the homogenate at 2000 rpm at 4 degree c for 20 min transfer the supernatant in to a seaparatory funnel & allow it to separate. Determine lipid content gravimetrically by measuring triplicate aliquots of the chloroform layer into tared containers, evaporate the solvent & weight Calculate the lipid content.

RESULT

The results of honey sample having different Parameters collected in three different locations of Sangamner Taluka were compared with the Codex Alimentarius & European standards[32]. Its observed that studied physicochemical Parameters are within the normal range (Table 1 & 2).

1) pH : The mean pH values Of honey samples obtained from three different locations of sangamner Taluka were within range 3.8-5.2. The pH is an important parameter during extraction of honey. It increases shelf life & quality of honey[33].

2) Electrical conductivity : The mean values of electrical conductivity in honey samples obtained from three different locations of Sangamner taluka were within the range of 0.07 to 0.116 mS/cm. EC is one of the most considerable factors for determination of the physical characteristics of honey. It is also useful for the Identification of honey quality & purity [34].

3) moisture content : The mean value of moisture content in honey sample obtained from three different locations of Sangamner taluka were within the range 15.98 - 18.28%.

4) Reducing sugar: The mean values of reducing sugar in honey sample from the different locations of Sangamner taluka were within the range of 58.92-64.72%. Honey is a mixture of two reducing sugars namely glucose & Fructose.

5) Glucose: The mean concentration of glucose in honey sample obtained from three different locations of Sangamner taluka were within the range of 20.22-24.12%.

6) Fructose: The mean concentration of fructose in honey sample obtained from three different locations of Sangamner taluka were within the range of 32.12-42.60%.

7) Fructose-Glucose ratio: The mean concentration of fructose-glucose ratio in honey sample obtained from three different locations of Sangamner taluka were within the range of 1.10-1.92.

8) Sucrose: The mean concentration of sucrose in honey sample obtained from three different locations of Sangamner taluka were within the range of 1.014-1.432g/100g.

9) Lipid: The mean concentration of sucrose in honey sample obtained from three different locations of Sangamner taluka were within the range of 1.014-1.432g/100g.

CONCLUSION

Total results of physicochemical parameters indicates that the nutritional quality of honey was different from species to species and from location to location. The honey obtain from agricultural and forest areas has highest nutritional quality than honey obtain from road side area. The average value of the physicochemical parameters found in the honey samples showed that honey harvested from the studied area is safe for human decay referring to Codex Alimentarius standards [35,36,37].
**Table- 1 :**

Physical properties of honey sample from three different location of sangamner taluka of Ahmednagar district.

<table>
<thead>
<tr>
<th>Collection Site</th>
<th>PH</th>
<th>Electrical conductivity (mS/cm)</th>
<th>Moisture Content (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Agricultural Area</td>
<td>4.4 ± 0.16</td>
<td>0.088 ± 0.002</td>
<td>17.20 ± 0.90</td>
</tr>
<tr>
<td>Forest Area</td>
<td>3.8 ± 0.10</td>
<td>0.06 ± 0.004</td>
<td>15.98 ± 0.92</td>
</tr>
<tr>
<td>Road side Area</td>
<td>5.2 ± 0.20</td>
<td>0.116 ± 0.005</td>
<td>18.28 ± 0.80</td>
</tr>
<tr>
<td>Standards of codex,1998 &amp; 2019</td>
<td>3.4- 6.1</td>
<td>0.8 mS/ cm</td>
<td>22%</td>
</tr>
</tbody>
</table>

**Table- 2 :**

Chemical properties of raw honey sample from the different location of sangamner taluka of Ahmednagar district.

<table>
<thead>
<tr>
<th>Parameters</th>
<th>Agar</th>
<th>Glucose %</th>
<th>Fructose %</th>
<th>G/F Ratio</th>
<th>Sucrose g/100g</th>
<th>Lipid g/100g</th>
</tr>
</thead>
<tbody>
<tr>
<td>Agricultural Area</td>
<td>62.10 ± 2.12</td>
<td>23.82 ± 0.42</td>
<td>38.02</td>
<td>1.62</td>
<td>1.32 ± 0.20</td>
<td>0.32 ± 0.06</td>
</tr>
<tr>
<td>Forest Area</td>
<td>64.72 ± 1.2</td>
<td>22.28 ± 0.88</td>
<td>42.01</td>
<td>1.68</td>
<td>1.62 ± 0.12</td>
<td>0.28 ± 0.02</td>
</tr>
<tr>
<td>Road side Area</td>
<td>58.92 ± 1.32</td>
<td>24.78 ± 0.40</td>
<td>36.16</td>
<td>1.32</td>
<td>1.06 ± 0.32</td>
<td>0.34 ± 0.06</td>
</tr>
<tr>
<td>Standards of codex,1998 &amp; 2019</td>
<td>&gt;60 %</td>
<td>23-32%</td>
<td>31.2-42.4%</td>
<td>&gt;0.95</td>
<td>&lt;5 g/100g</td>
<td>0.10-0.50 g/100g</td>
</tr>
</tbody>
</table>

± Indicates standard deviation

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