Physico-chemical investigation of commercially available honey in Indian market

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***Abstract*- Honey, a natural sweetener with numerous antibacterial and antiseptic properties, stands out as a highly valued and appreciated natural product. Its widespread application in various medicines, addressing conditions from wound healing to cancer treatment, underscores its significance. This study aims to conduct a physiochemical analysis of different commercially available honey brands in the market. The investigation centres on four easily accessible honey brands – Dabur, Patanjali, Zandu, and Apis Himalaya organic tattva. These brands are commonly found in local markets within the region. The analysis encompasses a qualitative examination of honey components such as k^+, ca^{2+}, mg^{2+} iron, and sugars like glucose and fructose. Additionally, the study measures various physical and chemical properties, including acidity, electrical conductivity, *p*H, surface tension, viscosity, and moisture content. To ensure accuracy, the analysis incorporates trusted preliminary examinations, including taste, colour, paper test, genetic memory test, and crystallization test. The overarching goal of this research is to uncover the key differences between the perceived ideal consumption of honey and the reality of what the public actually consumes in their day-to-day lives. Consequently, this study opens up new avenues for further exploration by providing insights into examination practices, consumption patterns, and a comprehensive spectrum of analysis.**

***Key words*- honey, natural sweetener, conductivity, surface tension, viscosity.**

INTRODUCTION

As per Pearson's definition in 1976, honey is described as "the saccharine product gathered by the bees from the nectar of flowers" [1]. Simply put, honey can be identified as a "natural sweet substance produced by honey bees from the nectar of flowers or from secretion of living parts of plants," as stated by Jonathan W. White [2]. This concentrated aqueous solution of invert sugar contains a complex mixture of carbohydrates, amino acids, organic acids, minerals, and aromatic substances. Additionally, honey harbours unstable compounds like enzymes and certain vitamins [3].

Albert Einstein once remarked that removing bees from our ecosystem would lead to the extinction of mankind in four years. This underscores the vital role of bees in cross-pollination among flowers, ensuring variation and giving rise to life through the production of honey, a nectar-based sugar-sweet liquid.

Honey holds a global significance, being widely employed in food, medicine, and flavoring agents across countries. Comprising nearly 80% carbohydrates (35% glucose, 40% fructose, 5% sucrose), honey serves as an excellent energy source [4]. The composition, colour, flavor, and taste of honey are influenced by its origin, including factors such as the type of flower, the bee species, and climatic conditions [5]. Physico-chemical analysis of honey constituents allows for its evaluation. Honey, with a history spanning over 5000 years, has served both as a food and a medicinal substance [6]. Its inherent antibacterial and antiseptic properties position honey as one of the most valued and appreciated natural products. From addressing sore throats to preventing infections, honey has evolved into a significant component of our daily lives. Consequently, understanding the precise quality of the honey we consume becomes imperative [7]. The intriguing aspect of honey lies in its ability to resist spoilage over extended periods, contributing to its timeless appeal and making it a compelling subject for further investigation.

MATERIALS AND METHODS

In ancient times the honey was directly harvested and used in its purest form, but as time changed honey is made commercially available by different brands. The use of preservatives and flavors became a common practice, hampering the purity of this natural sweetener. To ensure the quality of honey from different bands, various physical and chemical experiments were conducted in this work. During harvesting of honey, it is subjected to heat to make it free from any bacteria. But temperature above 35°C may destroy essential enzymes in honey, which attribute to its medicinal properties. The surging global demand for honey and its deteriorating quality prompted the international honey council (IHC) to set some quality standards for commercial honey, which includes moisture content, mineral content, reducing sugar, sucrose content, electrical conductivity, free acidity etc [8].

Looking at the increasing health concerns and increasing levels of adulteration, it is crucial to conduct quality testing of commercially available honey. While honey may not fulfil all human nutritional standard requirements, it exhibits potential as a dietary supplement. This study aims to analyse various physico-chemical properties of few commercially available honey brands available in Indian market.

The study begins with the preliminary examination of 4 selected honey brands, Dabur, Patanjali, Zandu and Organic Tattva. Colour, texture, flavor was tested and inferences were drawn. After preliminary examination presence of potassium, iron and glucose were detected through chemical analysis. Chemical analysis was followed by physical properties determination. Viscosity, Surface tension, *p*H value and electrical conductivity were determined. All the results are compiled and discussed in next section.

RESULTS AND DISCUSSION

PRELIMINARY EXAMINATION

Commercially available honey exhibits a spectrum of colours, ranging from nearly transparent to deep brown, with Flavors that span from pleasantly mild to distinctly bold. These variations attribute to the locations from where honey bees gathered nectar. Generally, honey with a lighter colour tends to have a milder taste compared to its darker counterparts. The subsequent section outlines the observations and findings obtained from these assessments (table 1).

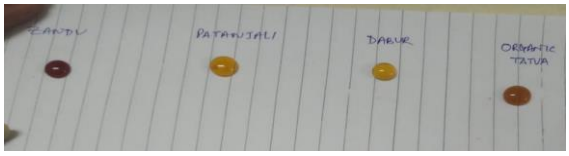

Figure 1 primary colour examination of different honey brands from Indian market

Figure 1 depicts the colour of four honey samples collected from Indian market. Zandu shows dark brown colour, Organic Tattva shows light brown, Dabur and Patanjali shows pale yellow colour of almost same intensity. The colour intensity varies from Zandu > Organic Tattva > Dabur > Patanjali.

FLAVOR EVALUATION

The sweetness of honey serves as a key indicator of its purity. In contrast to common belief, genuinely pure honey tends to lose its sweetness over time. If the sweetness lingers extensively, it may suggest the presence of additive sugars or artificial sweeteners, indicating potential impurity. The sweetness levels observed in the taste test are outlined in table 1.

Table 1Comparative sweetness and rate of flow for honey from different brands

S. No.	Brand	Comparative Sweetness	Miscibility	Time taken to flow from initial to final point (seconds)	Inference
1.	Dabur	Sweet	Moderate	166.8	Low adulteration
2.	Patanjali	Sweetest	High	88.7	High adulteration possible
3.	Zandu	Sweet with flavor of Cinnamon and lemon	High	87	High adulteration possible
4.	Organic tattva	Moderately sweet	Very high	82	May be adulterated

RATE OF FLOW ANALYSIS

Rate of flow (RoF) of honey speaks about its density and water content. Pure honey typically exhibits a thicker consistency, higher density, and consequently a lower RoF as compared to impure honey. Drops of various honey samples were placed on a horizontally positioned white sheet of paper at proper distance. Paper was tilted carefully to allow honey drops roll down, the RoF of the honey drops were observed and recorded. The observations are shown in figure 2 and the time taken to flow from initial to final position is shown in table 1. The high RoF indicates more water content and hence the density of the sample declines from Dabur to Organic Tattva (table 1). For this test glass surface was avoided because of adhesive power of honey with glass and hence a rough surface (paper) was selected for this test.

Figure 2 Rate of Flow (RoF) analysis for various honey samples

CHEMICAL ANALYSIS

Honey is a popular substitute to sugar. It holds viscous nature due to presence of carbohydrates, fructose and glucose (80-85%), along with 15-17% water and other elements such as calcium, potassium, and iron. In present study, we conducted qualitative analyses to identify the presence of iron, potassium, and glucose in the honey samples. The Benedict's test was employed to detect the presence of glucose, wherein the colour of the solution changes when sample containing glucose reacts with Benedict's reagent [9]. Depending upon the quantity of the glucose or sugar present in the sample, colour may vary from green to brick red. About 100 g of honey contains around 52 mg of potassium, a mineral crucial for maintaining body fluid balance and the sodium-potassium pump. However, honey's relatively low potassium content makes it less significant source of the mineral. Each 100 g serving of honey contains approximately 0.4 mg of iron. Iron is a vital component of blood facilitating oxygen transport throughout the body [10].

QUALITATIVE ANALYSIS

Test for Potassium: About 1ml of aqueous honey solution from each sample underwent examination with a few drops of picric acid. The appearance of a yellow precipitate (picrate) confirmed the presence of potassium ions [11]. Results indicated that only Dabur honey tested positive for potassium, while Patanjali, Zandu, and Organic Tattva exhibited an absence of the ion.

Test for Iron: 1ml aqueous sample solution from each honey brand was treated with 1-2ml of dilute sulphuric acid for acidification, followed by adding a pinch of potassium thiocyanate. The development of a red coloration confirmed the presence of iron [12]. All tested honey samples were found to contain iron.

Test for Glucose: Approximately 1ml of aqueous honey sample solution in a clean test tube was subjected to the addition of 5-10ml of freshly prepared Benedict's reagent. The mixture was heated on a water bath for 10-15 minutes, and a colour change was observed in each honey solution. Results indicated that all honey samples exhibited a brick-red colour change, indicating a high content of glucose [13].

PHYSICAL PROPERTIES ANALYSIS

ESTIMATION OF VISCOSITY

Viscosity is the internal property of a fluid. It refers to the internal resistance to it's flow. In 1844, Hagen-Poiseuille proposed an equation for coefficient of viscosity of liquids (η) (Equation 1) [14].

$$V = \frac{\pi p t r^4}{8 \eta l} \qquad (1)$$

Where η= *coefficient of viscosity*
t= time of flow
l= distance travelled by the liquid
V= volume of liquid
r= radius of drop
p= pressure

As the temperature increases, viscosity falls, due to less molecular friction and reduced hydrodynamic forces. The viscosity of honey depends on many factors including composition and temperature. Relative viscosity is the ratio of the absolute viscosity of the fluid to the viscosity of water (or reference fluid) at a certain temperature.

$$\frac{\eta_1}{\eta_2} = \frac{d_1 t_1}{d_2 t_2} \qquad (2)$$

Here
η_1 is coefficient of viscosity for liquid1

η_2 is coefficient of viscosity for liqui2
t_1 time of flow for liquid 1 from point A to B in Ostwald viscometer.
t_2 time of flow for liquid 2 from point A to B
d_1, d_2 are densities of liquid 1 and 2 respectively, as shown in figure 3(a).
Procedure: The Ostwald viscometer was mounted with the help of clamp and stand. Water was filled up to the mark C (figure 3 (a)) and sucked through the capillary till mark A. Water was allowed to flow from mark A to B and time of flow was noted with the help of stop watch. This procedure was repeated for all honey samples too. For each sample two sets were taken. The relative and absolute viscosities of all four honey samples were calculated and reported (Table 2).

ESTIMATION OF SURACE TENSION – DROP COUNT METHOD USING STALAGMOMETER
Surface tension is defined as the tension on the surface of a liquid which tends to minimize it's surface area. It depends on various factors like temperature, solute concentration, intermolecular forces, hydrogen bonding etc.
Surface tension of the samples was determined with the help of stalagmometer (figure 3(b)) using drop count method and water as reference liquid. stalagmometer is mounted with a capillary tube inside it. The size of the drop of liquid depends on radius of capillary and surface tension of the liquid. If two liquids having surface tension γ_1 and γ_2 are allowed to fall through the same capillary then their weights (w_1, w_2) and masses (m_1, m_2) can be written as in equation (3),

$$\frac{\gamma_1}{\gamma_2} = \frac{w_1}{w_2} = \frac{m_1}{m_2} \qquad (3)$$

Procedure (drop count method): the stalagmometer was cleaned and dried with acetone. The lower end of the stalagmometer was immersed in water and sucked till the upper mark. The pinch cork was adjusted so that 10-15 drops fall per minute.
The stalagmometer was clamped and 10 drops of water were collected in pre weighed specific gravity bottle as shown in figure 3. Weight of the gravity bottle with 10 drops was taken. Above steps were repeated using different honey sample solutions. Observations were recorded and the surface tension of honey samples was calculated accordingly and reported in table 2.

***p*H ANALYSIS**
Knowing the *p*H of food items is very important. Our body acts differently for food items with different *p*H values. *p*H is a scale used to specify how acidic or basic a solution is. Acidic solutions have *p*H less than 7, while basic solutions have a *p*H higher than 7. *p*H is a measure of the concentration of hydronium ions.
$pH = - \log [H^+]$ *(4)*

Honey contains a number of different acids, including 18 aliphatic and aromatic amino acids. Because of its acidic properties, honey should always be processed and stored in non-reactive containers preferably glass containers.
Procedure: 10 gram of each honey sample was dissolved in 100ml of distilled water and *p*H of each solution was measured using *p*H meter. *p*H meter first was calibrated using standard buffers of *p*H 7 and *p*H 4. After calibration, the *p*H of various honey solutions was measured. The *p*H of samples was found to lie between 3.4 – 6.1 which is expected *p*H range for honey.

ESTIMATION OF ELECTRICAL CONDUCTIVITY
It is fundamental property of a material that quantifies how strongly it resists or conducts electricity. It depends on the concentration of minerals salts, organic acids and proteins. All the honey samples in the present study showed electrical conductivity values between 0.008 to 1.2 mS/cm.

Procedure: 10% aqueous solution of honey was taken in a beaker. A conductivity cell was placed in it. Observations were recorded. All honey solutions were found to be conducting in nature (table2).

ESTIMATION OF ACIDITY
The acidity of honey can be measured by titration against sodium hydroxide equivalents or direct measurement of *p*H. The commercial high quality of honey should have the free acidity up to 50 milli equivalents/kg of honey. According to the national honey board, the *p*H of honey ranges from 3.4 to about 6.1, the acidity of honey is directly related to the floral sources that created it. Acidity is more rigorously defined as the amount of acids in a solution that can be titrated with strong base. The acidity of test solutions of honey was estimated via titration method. Which includes two

indicators, phenolphthalein and methyl orange indicator. Phenolphthalein indicator indicates weak acidity and methyl orange indicator indicates strong acidity. Hence, the total acidity was calculated.

$$\text{Total acidity} = \frac{\text{phenolphthalein acidity} + \text{methyl orange acidity}}{2} \quad (5)$$

Procedure: 50 ml of aqueous solutions of honey was taken in a conical flask, 2-3 drops of methyl orange indicator was added. 0.05M solution of NaOH was taken in burette. Honey samples were titrated against NaOH, till the change in colour was observed (yellow to orange), which indicated end point. Titration was repeated with phenolphthalein indicator to observe colour change from pink to colourless. Volumes of NaOH used for both titrations were recorded and the respective acidities were calculated. Hence the total acidity was calculated using equation (5).

Table 2 The estimated physical properties of the honey samples

Brand name	Viscosity (centipoise, cp)	Surface tension (dynes/cm)	*p*H	Conductivity (mS/cm)	Acidity (*g/l*)
Dabur	9013	70.73	3.93	0.58900	0.0005
Patanjali	8961	68.82	4.78	1.11800	0.0005
Zandu	7993	52.08	3.48	0.98300	0.0247
Organic tattva	9246	54.19	4.15	0.00892	0.0011

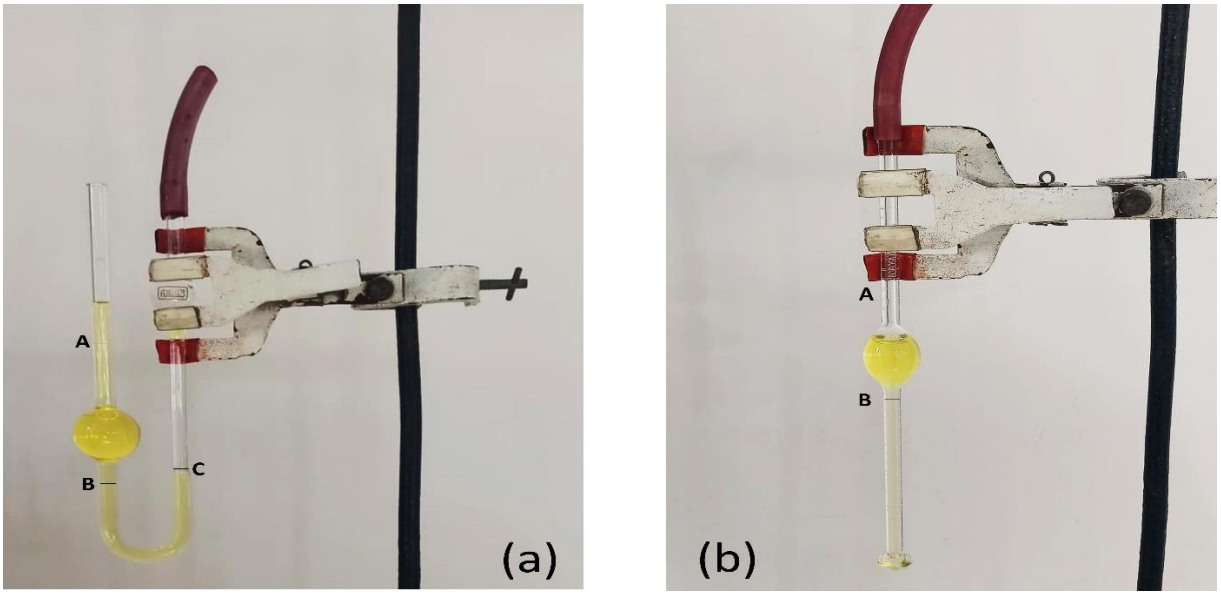

Figure 3 Experimental setup for (a) Viscosity measurement with Ostwald viscometer and (b) Surface tension measurement with stalagmometer

The results of physico-chemical analysis of considered samples speaks about the quality of honey of selected brands. This research does not discourage the purchase or consumption of brands other than Dabur in any way. However, upon comparison with the brands listed above, Dabur emerges as a leader in multiple aspects, as evidenced by the findings. Therefore, it is strongly recommended.

CONCLUSION

The current study examines the quality of honey from various brands, aiming to assist consumers in choosing the right brand based on their health needs. Modern lifestyle-related health conditions such as acid reflux, diabetes, and high/low blood pressure can be exacerbated by consuming the wrong honey regularly. Despite the popular use of honey and lemon solution for weight loss, high sugar content in honey can impact glucose levels. The study includes several preliminary tests to evaluate the quality of honey from four top brands in the Indian market. Additionally, the research highlights the importance of honey and its producers, the bees, for the sustainability of the environment. Among the brands compared, Dabur emerges as a leader in multiple aspects, as demonstrated by the findings.

CONFLICT OF INTEREST

There was no conflict of interest between authors.

CONTRIBUTION
All the authors contributed equally.

ACKNOWLEDGEMENTS
Authors are thankful to Department of Biotechnology (DBT) and ARSD College for supporting this work.

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