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Elemental analysis of different floral honeys of East Godvari Dist., Andhra Pradesh, India.

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Abstract: *The Honey is a natural product which is synthesized by honey bees by collecting nectar from different kinds of flowers having both food and medicinal values which are useful to the mankind and other organisms. Minerals are one of the most important elements of the biological (vital) activities of the living organisms. In our investigation we studied elemental composition of some floral honeys of particular area and their ash contents and also mentioned the importance of essential (micro to macro) and non essential elements (whose functions are unknown).*

Keywords: *Meleto Palionology, Frequency classes, Pollen morpho types, Chromoproteins, Metal activators, Unifloral, Multifloral.*

I. Introduction

The Honey is an excellent unique sweetening agent which contains both medicinal and food values and can be preserved for a long time without adding any preservative. The properties gained by honey are its ingredients which are about 300 different chemicals. These chemicals give colloidal suspension nature to honey. In the composition of honey, nearly 30 micro to macro elements have been observed[1]. And these elements play a very important role in the biological reactions of living organisms. Even though their quantities are less their importance is very essential or high. We have investigated the elements (mineral contents) of the honey by recognizing the origin of honey. Origin of honey again investigated by pollen.

We have selectively chosen the various floral honeys of East Godavari Dist., investigated and identified their maximum elemental composition. We tried to explain the roles, the importance and deficiency causing diseases of each element.

II. Importance Of Metal Elements In Biological Reactions

Out of 13 metals required by the human body, we detected 9 micro elements in all East Godavari honey (EGH) samples in which Fe, Cu, Mn, Zn and Mg play very important role in biochemical reactions. These metals directly links with proteins and known as metalproteins or chromoproteins. Generally they are in the enzymes and acts as metal activators and helps in group transfer, redox and hydrolysis reactions of the organisms[1]. And the rest of the metals have their own significance in biochemical reactions of the organism. The human body requires Na, K, Mg and Ca are in macro levels and Zn, Cu, are in micro levels. Fe is in between micro and macro levels.

III. Mineral Content Of The Honey:

Minerals in honey are grouped and expressed as ash content in most studies, but analysis of individual elements has been largely ignored. The present study determined ash content, electrical conductivity and 3 individual elements like K, Na, Mg, Mn, Cu, Fe, Zn and Ni of the 3 different honey types under study. The related data are presented, where the K / Na ratio is also included. The values of ash content are expressed as percent of honey weight, and those of individual elements are in ppm.

IV. Samples

Pollen Analysis: Honey samples were procured from different areas of East Godavari district, Andhra Pradesh, India in different seasons[2]. The samples were subjected to qualitative and quantitative pollen analysis following the methodology recommended by the International Commission for Bee Botany (ICBB) (Louveaux et al 1978). The pollen morphotypes were identified with the help of reference slides mentioned in the Central Bee Research Institute (CBRI, Pune) palynarium.

V. Methods

The pollen types recovered and identified were placed under four frequency classes as mentioned bellow. The three E.G samples were investigated for their origin by using pollen analysis of honey is known as Meleto palionalysis[3].

1. Predominant pollen type: More than 45% of the total pollen grains counted.
2. Secondary pollen type: Between 16 and 45% of the total pollen grains counted.
3. Important minor pollen type: Between 3 and 15% of the total pollen grains counted.
4. Minor pollen type: Less than 3% of the pollen grains counted.

The honey sample was treated as Unifloral if the prepared slide contains a predominant pollen morphotype. If several morphotypes are represented, the honey sample was termed as Multifloral[4]. Basing on the above information honey samples were identified out of three, two are Unifloral and one is as Multifloral.

List of Unifloral honeys of East Godavari district, Andhra Pradesh – India investigated

1. Eucalyptus globulus
2. Borassus flabellifer
3. Syzygium cumini

Fig 1a. Eucalyptus globulus

Fig 1b. Borassus flabellifer

Fig 1c. Syzygium cumini

TABLE I. Frequency classes and frequencies of Pollen morphotypes in the Unifloral honey samples of the present study

Honey type	Frequency class	Pollen morphotype	Frequency (%)
EGH1	P	Eucalyptus globulus	60
	S	Schleichera oleosa	25
	I	Ageratum thumb conyzoides	14
	M	Phoenix Sylvestris	1
EGH2	P	-Nil-	0
	S	Borassus flabellifer	62
		Pongamia pinnata	22
		Croton bonplandianum	16
EGH3	P	Syzygium cumini	58
	S	Flacourtia indica	29
	I	Borassus flabellifer	13
	M	-Nil-	0

P= Predominant S= Secondary I= Important minor M= Minor

VI. Determination Of Total Ash (AOAC 1975):

About 25g of the well homogenated honey sample was placed in an ashing vessel of known weight. The materials in the vessel was carbonized under the hood initially on a low heat to prevent spattering.Then, the silica crucible was transferred to a muffle furnace maintained at 550^0C and incarcerated for about 6hrs. The dish was taken out, cooled to room temperature and the ash was wetted with minimum amount of water, followed by the addition of a few drops of concentrated HNO_3.The contents were dried on low heat and again turned into ash at 550^0C, till carbon free ash was obtained. The crucible was cooled down to 100^0C and transferred to a desiccator for further cooling to room temperature and later weighed. (The ash was reserved for the estimation of mineral matter).

$$\%\ of\ Ash = \frac{Weight\ of\ Ash}{Weight\ of\ sample} \times 100 \qquad (1)$$

Fig 2a. Schleichera oleosa

Fig 2b. Ageratum thumb conyzoides

Fig 2c. Phoenix Sylvestris

Fig 3a. Pongamia pinnata

Fig 3b. Croton bonplandianum

Fig 3c. Flacourtia Indica

a) Preparation of sample solution suitable for mineral component (determination of ash):

a) The ash obtained was dissolved in 10ml concentrated HCl, boiled and evaporated to near dryness on a hot plate.

b) The residue was redissolved in 10ml HCl of 2N normality by boiling gently and filtered through the fast ash less filter (Whatman 41) into 50ml volumetric flask. The residue and paper were washed thoroughly with water collecting all the filtrates in to the same volumetric flask. The solution was made with 50ml with water mixed well.

c) The concentration of metals in ash solutions under consideration was measured directly or diluted with 0.5N HCl to obtain solutions within the range of standards.

b) Detailed procedure for each metal as follows :

a) Determination of Calcium : The determination was done by comlexometric titration .

Reagents :

i. Ethylene diamine tetra acetic acid salt (0.1ml).
ii. Murexid indicator (ammonium purpurate) .
iii. Sodium hydroxide solution (2N) .

b) Determination of sodium and potassium: Flame photometer was used for this purpose.

Reagents:

i. Stock solution - 1000 ppm (1 ml = 1 mg).
ii. Stock potassium solution -1000 ppm (1 ml = 1 mg).

*Procedure***:** The sample was aspirated after waiting for a few minutes the readings of sodium and potassium were noted .

$$\text{Sodium}(\text{mg/l}) = \frac{\text{Reading} \times \text{ppm of Na std.}}{100} \quad (2)$$

$$\text{Potassium}(\text{mg/l}) = \frac{\text{Reading} \times \text{ppm of K std.}}{100} \quad (3)$$

c) **Determination of Mg, Cu, Zn, Ni, Fe and Mn :** The ash samples of Mg, Cu, Zn, Ni, Fe and Mn were aspirated in the calibrated Atomic absorption spectrophotometer (A.A.S) and the concentrations of the elements in ppm were recorded. The parameters used for the elements are as follows.

TABLE II. Parameters used for the elements

S.No	Element	Lamp Current (mA)	Fuel	Support	Wave length (nm)	Slit width (nm)	Reagent (1:1 and 1 ml stock solution)
1	Mg	4	Acetylene	Air	285.5	0.1	HNO_3 in Dist. Water
2	Cu	4	Acetylene	Air	324.7	0.5	HNO_3 in Dist. Water
3	Zn	5	Acetylene	Air	213.9	1.0	HNO_3 in Dist. Water
4	Ni	4	Acetylene	Air	232.0	0.2	HNO_3 in Dist. Water
5	Fe	5	Acetylene	Air	248.3	0.2	Hcl in Dist. Water
6	Mn	5	Acetylene	Air	279.9	0.2	HNO_3 in Dist.Water

Procedure : The ash sample was aspirated in the calibrated A.A.S and the concentration of each element was noted in ppm .

$$\text{Content of element (in ppm)} = \frac{C \times D \times 50}{W} \quad (4)$$

Where C = Concentration of metal (in µg/ml) read from instrument ,
D = Dilution factor , if the original ash solution (of 50 ml) was distilled further .
W = Weight (in g) of the sample taken for ashing .

VII. Results

Among the three unifloral honey types of East Godavari district, Eucalyptus honey (EGH 1) showed the highest percent of ash content and the lowest value was that of Syzygium (EGH 3). Regarding individual elements, Borassus honey (EGH 2) had the highest value of Fe, Ni and Na. Similarly Eucalyptus honey was rich in Cu and Mn.The lowest values of the different elements were Fe, Ni, Zn, Mn , Mg, Na and K - all in Syzygium honey, and Cu was present in lowest quantity in Syzygium honey. Among the three multifloral honeys from East Godavari District, EGH 1 showed the highest ash content. EGH 3 had the lowest percentage. The individual elements Fe, Ni, Zn, Mn were comparatively.

Of the unifloral honeys, Syzygium honey (EGH 5) from East Godavari District was rather poor in both ash content (%) and total minerals. The difference could be attributed to the difference in the mineral content in the soil of the different areas. Since plants obtain minerals in soil, aeration between the mineral content of honeys and of soil may be expected. Such relationship was shown by Varju (1970) who investigated the mineral composition of Acacia honeys and the soils. The honeys of Acacia trees grown in soils rich in Ca and Fe were richer in these elements. Such findings suggest the potential use of honey as an indicator in environmental contamination (Jones 1987). Morse and Lisk (1980) determined 16 elements in honey and the high concentration of certain elements were related to the sours of these metals additional reports of the elemental composition of honeys in areas with and without pollution are required to see the possibility of utilizing honeys as indicator of environmental pollution, and also the relationship between the mineral composition of honey samples and their geographical origin. Uren et al.(1998) showed that only honeys with low total mineral content may be affected easily by the contamination sources , and may have elevated levels of possible contaminants.

The values of ash content obtained in recent study are comparable with similar data reported earlier for Indian honeys. The values of the East Godavari District honey are either close to or exceed the values of ash content reported for honeys in other countries like USA (White and Doner 1980) and (Whits et al. 1962) , Calcia (Rodriquez-Otero et al .1994) : Molise region Esti et al.1997). And several other countries mentioned in White (1975). Rodriquez-Otero et al (1994) mentioned that the Calcian honeys have in general a higher mineral content than the honey from various European countries. The Calcian honeys also appear to exceed the ash content values of Indian honeys.

Majority of honey types content of K greatly exceeded the other elements. Thus the study found K as the most abundant of the element determined. Potassium accounted for a higher % of ash weight in the Calcium honeys (Rodriquez-Otero et al 1994) and also in US honey for dominance of K over other elements has also been reported for Calibri an honey and Molise region honeys (Esti et al .1997). The Molise workers found a

positive correlation between pH and ash content of honeys and attributed the same to the high cationic content, particularly K that influences the Stalinized fraction of the acids. A similar positive correlation exists between ash content and pH of the honey types of the present study as seen above.

VIII. Conclusion

Elemental analysis of some unifloral honeys of East Godavari Dist. Of A.P, India. We investigated three unifloral honeys for the elemental composition. All unifloral honeys contain all elements i.e, Na, K, Ni, Fe, Mg, Zn, Cu, Mn and Ca. However, some unifloral honeys are rich with some elements (Cu, Zn, Mn are trace elements). Out of three unifloral honeys of East Godavari Dist. Of A.P, India.

Eucalyptus honey with highest ash content and content predominant elements are Cu, Mn along with other elements. Borassus honey another unifloral is rich in Ni, Na, and Fe along with other elements. If we go through the importance in human biological system Ni functions are not completely established, however it comes to know that Ni has important key role in the human biology. Na other elements present in this honey are very good electrolyte and who maintains body fluid osmotic pressure. Fe is the most important macro element of the life. K activity is most important in biological systems like Na, Zn meta improves mental ability. Syzygium also famous unifloral honey containing all elements, but in very low quantities. Ash content is also very low.

Syzygium contains all minerals in small quantities. Each element is having its own importance in human biological system. Hence we can recommend each floral honey as an additional food supplement accordingly instead of taking other food supplements. Because honey is a very fine mixture of different foods (components) which are essential for the human body and having no adverse results if we consume.

Pure honey does not adhere to the paper and it processes tart taste. Most of the honeys contain their flower color and smell. Different branded honeys are available in the market. They are pure, but we cannot say its origin. For unifloral honeys one can contact beekeeper and he can guide us in the proper way.

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