Original Research Paper

Volume - 7 | Issue - 5 | May - 2017 | ISSN - 2249-555X | IF : 4.894 | IC Value : 79.96

Chemistry

A COMPARATIVE STUDY OF ACIDS IN MULTIFLORAL HONEYS OF EAST GODAVARI DISTRICT, A.P., INDIA

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ABSTRACT Honey is a natural product known to man since ancient time. It can be preserved for a long time with out adding any preservatives. Its food and medicinal value made it unique among all natural products. It is a colloidal suspension of nearly 300 different compounds. This wonderful tasty product synthesized by honey bees by collecting nectar of various flowers. Honey is acidic in nature. No major change reported in pH even after storage for prolonged periods. Honey is acidic in nature. Acidity of honey results from the presence of Organic Acids. Many acids have been reported to be present in honey. Acidity of honey may be expressed as free acids, lactone acids or tabal acids which account for less than 0.5 g of solids. This small quantity of acids contributes to the flavour and tart taste to the honey and also responsible in part for the stability of honey against Micro Organisms. Total acidity of 3 different multiifloral honeys are studied. These honeys were collected or procured in different seasons from different areas of East Godavari District, A.P., India.

KEYWORDS :Meleto Paleonology, Frequency Classes, Pollen Morphotypes, Unifloral, Multifloral, Milli Equivalent, Glucose Oxidase.

INTRODUCTION

Several acids like glutamic, formic, acetic, tartaric, malic, succinic acids have been reported in honey. Of these acids, gluconic acid predominant. The most important and major being gluconic acid. It arises from the action of glucose oxidase in honey on dextrose (Glucose) gluconic acid in solution in equilibrium with gluconolactone (or) internal ester, often the acids include acetic, butyric, lactic, succinic, tartaric, pyroglutamic and citric acids the free acidity of which is provided by all free acids as a whole while the lactone acidity is due to glucomo lactone.

Since inorganic ions such as Phosphate, Chloride and Sulphate are also present in honey, their corresponding acids may also exist in honey (white 1975; Echigo and Takanaka 1974.)

POLLEN ANALYSIS:

Honey samples were procured from different areas of East Godavari District, Andhra Pradesh, India in different seasons.[5]. 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.

The pollen types recovered and identified were placed under four frequency classes as mentioned below. The three E.G. samples were investigated for their origin by using pollen analysis of honey is known as Meleto Palionalysis [6].

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 Multiflora [4]. Basing on the above information honey samples were identified. Three are Multifloral.

TABLE - 1
FREQUENCY CLASSES AND FREQUENCIES OF POLLEN MORPHOTYPES IN THE MULTIFLORAL HONEY SAMPLES OF THE PRESENT STUDY

Honey Type	Frequency Class	Pollen Morphotype	Frequency (%)
EGH (1)	P	Brassica Juncea	54
	S	Litchi chinensis	41
	I	Poaceae	5
	M	-Nil-	0
EGH (2)	P	Limonia acidissima	67
	S	Delonix regia	21
	I	Syzygium cumini	12
	M	-Nil-	0
EGH (3)	P	Trifolium alexandrinum	58
	S	Helianthus annuus	37
	I	Syzygium cumini	14
	M	Carthamus tinctorius	1

1. **Fig 1a. Brassica Juncea**

Fig 1b. Litchi chinensis

Fig 1c. Poaceae

P = Predominant, S = Secondary, I = Important, M = Minor
On the basis of above information, all 3 honey samples were identified as Multifloral.

EGH (1)Brassica Juncea
EGH (2)Limonia acidissima
EGH (3)Trifolium alexandrinum

METHODS

Determination of free acidity, lactone and total acidity (White et al, 1958):

The sample (log) was weighed accurately in a 250 ml beaker and 75 ml of Carbon Dioxide free distilled water added to it. The honey was dissolved and stirred with a magnetic stirrer. The electrodes of a pH meter were placed in the solution and the initial pH recorded. The solution was then titrated with 0.05 N NaOH. The NaOH was added at such a rate that individual drops must tend to merge in to a steady stream (5 ml / min). Addition of NaOH was stopped when the pH was reached 8.5. Immediately 10 ml of 0.05 N NaOH was added using a 10 ml Pipette and the pH was brought back to 8.3 by rapidly adding 0.05 N HCl from a 10 ml burette.

Fig 2a. Limonia acidissima

Fig 2b. Delonix regia

The amount of NaOH added from the burette minus the "blank" correction give the measure of free acid present and the amount of HCl subtracted from 10 ml was the measure of free acid present. The sum of the free acid and lactone was the total acidity. All the values were calculated to ml 0.1 N alkali per 100 grams sample or milli-equivalents per Kg. The titration rate given was as rapid as found consistent with acceptable reproducibility. It was found that the titration to pH 8.5 was equivalent to maintenance of Phenol Phthalein Pinic for 10 seconds, since the pH has fallen to 8.3 in that time.

Determinations were made in the following way.
Free acidity = (ml 0.05 N NaOH from burette ml blank) x 50g sample.
Lactone = (10.00 ml 0.05 N HCl from burette) x 50 / g sample.
Total acidity = Free acidity + Lactone

Fig 3a. Trifolium alexandrinum

Fig 3b. Helianthus annuus

Fig 3c. Syzygium cumini

Fig 3d. Carthamus tinctorius

Determination of pH:

Digital pH meter was used for the purpose. The instrument was calibrated with standard buffer solutions of pH 4.0 and 7.0 and the readings were taken at 300C. Honey sample of 20 g was dissolved in 100 ml CO2 free water and the pH readings were taken.The acidity of honey was expressed in different ways by different workers. Total acidity or free acids as percent (%) of total solids (See white, 1975) [8]. But the better expression is milli equivalent per Kilogram of honey. (Fogler, 1975) [7]. The most studies, acidity is expressed as free acidity, whether gluconic acid or formic acid.

But more appropriate to measure free acidity and Lactone acidity. Addition of these two gives total acidity to facilitate calculations of percentage of acids in milliequivalent (mequ per Kg of honey) (Fasler, 1975). [7,8].

No major change reported in pH.

1% formic acid=208 mequ/Kg
1% gluconic acid=50 mequ/Kg
1% glucolactone=56 mequ/Kg

DISCUSSION:

The present study examined free acidity as well as Lactone acidity and thus total acidity in milli-equivalents / Kg of honey. One cannot recognize origin of any honey without meleto paleonological studies. A consumer knowledge insufficient recognize pure honey. Majority of people don't know that there are two types of honeys by their origin i.e unifloral honeys and multi floral honeys. Honey is known/familiar to the people as "pattu thena or putta thena" in telugu language. From our extensive studies about honey we could able to reveal the facts about honeys. Quality of the honeys are decided by pollen morpho types and its percentage. In multi floral honeys also importance of percentage of each pollen morpho types decides its quality. Mostly predominant (in sense maximum percentage of pollen morpho type present in the honey but not in the sense of frequency classes) pollen morpho types. All the parameters of honey are inter related, for example Ash(Type of element & % of each element) pH & acidity. We have observed the same thing in case of multi floral honeys of east Godavari district AP, India. We have studied and compared the total acidity & pH of 3 different multi floral honeys. Acidity of multi floral honeys of east Godavari district is having close relation with US honeys than other multi floral honeys of other countries. Our studies are multi floral honeys and its results are as follows:

TABLE - 2
DECREASING ORDER OF ACIDITY IN MULTIFLORAL HONEYS OF E.G. DT., A.P., INDIA

Honey Type	Name of the Sample	Acidity
EGH (1)	Brassica Juncea	Decreasing Order
EGH (2)	Limonia acidissima	↓
EGH (3)	Trifolium alexandrinum	

TABLE - 3
INCREASING ORDER OF FREE ACIDS IN MULTIFLORAL HONEYS OF E.G. DT., A.P., INDIA

Honey Type	Name of the Sample	Acidity
EGH (1)	Brassica Juncea	Increasing Order
EGH (2)	Limonia acidissima	↓
EGH (3)	Trifolium alexandrinum	

TABLE - 4
DECREASING ORDER OF LACTONE CONTENT IN MULTIFLORAL HONEYS OF E.G. DT., A.P., INDIA

Honey Type	Name of the Sample	Acidity
EGH (1)	Brassica Juncea	Decreasing Order
EGH (3)	Trifolium alexandrinum	↓
EGH (2)	Limonia acidissima	

TABLE - 5
DECREASING ORDER OF Ph IN MULTIFLORAL HONEYS OF E.G. DT., A.P., INDIA

Honey Type	Name of the Sample	Acidity
EGH (1)	Brassica Juncea	Decreasing Order
EGH (2)	Limonia acidissima	↓
EGH (3)	Trifolium alexandrinum	

order, but they are differing in their lactone content order. In above 3 cases EGH1 is dominating other 2 multi floral honeys. Much difference is not found in the value of EGH2 & EGH3 (pH & total acids), but the values of total acidity of EGH1 showing quite high values than other 2 honeys (EGH2 & EGH3). We can give the following explanation for the above mentioned values or facts by means of their individual pollen morpho types and their percentages.

CONCLUSIONS:

From the above information, we concluded as a sample EGH1 morphotypes, Brassica and Litchi are contributing more towards its acidity and pH value. The third morpho type contribution is very less i.e. 5% only towards the total acidity & pH (almost 0).

In case of EGH2 main contributing morphotypes are Limonia 67% and Delonix regia 21%, Syzygium contribution is very less. Hence it can occupy 2nd place in pH and 3rd place in Free acid.

As in EGH3, Trifolium contribution 50%. Second major pollen morpho type is 37% i.e. Helianthus and other one Syzygium is 12 % only and hence it got second place in lactone values.

Anyhow the major pollen morpho types are deciding factors for the quality and other parameters of the honey. Basing on the above facts EGH1 occupies 1st place and 2nd by EGH2 and 3rd by EGH3 respectively. The preservative properties also in the same order as we expected.

The honey collected from bee keepers are good to consumers because he can give some information about the seasonal flowers of the particular honey sample. Honeys are available in market by different trade names but majority don't give any information regarding to the season or floral type. Hence we cannot consider them as either multi floral or unifloral.

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