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The Physical and Chemical Properties of Bee Honey Produced in North Sinai Governorate, Egypt

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ABSTRACT
Four types of honey were collected from two apiaries in North Sinai Governorate, Egypt, during the 2021 seasons. The honey was obtained from different floral sources, including Citrus (Citrus sp.) in Rafeh region and Algae (*Salvia officinalis*), Myrrh (*Commiphora myrrha*), and Tarot (*Zygophyllum* L.) in El-Arish region. The plant species were determined by examining the pollen grains present in the honey under a microscope. The proximate analysis of the honey samples obtained from various locations in North Sinai, Egypt, showed that there were no significant differences in moisture, fructose, maltose, (fructose + glucose), reducing sugar, F/G, and G/W. However, there were significant differences ($P<0.05$) in glucose, sucrose, pH, free acidity, lactone, and total acidity among the honey samples. Based on these findings, it can be concluded that the quality parameters of all the honey samples studied meet the standards set by international honey regulations. The study also found that the quality and physicochemical properties of the honey samples varied depending on their geographical and botanical origins.

Keywords: honey, floral sources, Citrus, pollen grains

1. Introduction
Honey standardization has therefore, become an importer issue to consider. Every country or a region of countries has suggested special honey standards, on different factors existing in countries. These factors were, environmental conditions, flora type, and individual beekeeping practices. In the past USA standards judged-honey brands by two methods, the first method was the grading, which was based on four factors: Soluble solids, flavor, absence of defect and clarity. While the second method was the color measured by the aid of many instruments as the refractometer. (El-Ansary, l998). Compositions of bee honey depend on its geographical floral origin, season, environmental factors and process of beekeepers (Kaškonienė *et al.,* 2010 and El Metwally, 2015). Honey contains approximately carbohydrates 80% (glucose 35 %, fructose 40%, and sucrose 5 %) and water 20 %, serving as an excellent source of the energy. In addition, it constitutes more than 200 components, including amino acids, vitamins, minerals, enzymes, organic acids, and phenolic compounds (Rodriguez *et al.,* 2004 and Kahraman *et al.,* 2010). The identification of honey plant sources is a subject of a great deal of interest since many years. There were various reasons why the floral origin of honey may be wanted to be known, such as, for quality control in marketing and where there is regulatory concern about the country of origin of honey (Molan, 1998). Honey is mostly made up of sugars, mostly fructose (40–50%) and glucose (32–37%), with a small quantity of sucrose (honey can be variable and dependent on its floral source, geographical origin, environmental factors, and processing (Guler *et al.,* 2007; Alvarez-Suarez *et al.,* 2010 and El Sohaimy *et al.,* 2015). The characterization of three types of floral (citrus, clover,

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cotton) and sugar-feeding honey was carried out based on their quality parameters [pH, glucose, fructose, sucrose, maltose content, total soluble solids (T.S.S.), total acidity, free acids, lactones, electrical conductivity (Nafea & Mazeed 2023). The aim of this study evaluate some physical and chemical properties of North Sinai honeys and their quality according to Egyptian honey standard (2005).

2. Materials and Methods

2.1. Honey samples Collecting:

Thirty samples of honey were harvested from apiaries located in two location of North Sinai regions during seasons of the year depending on floral sources, Citrus (Citrus sp.) in Rafeh region, Algae (*Salvia officinalis*), myrrh (*Commiphora myrrha*) and tarot (*Zygophyllum* L.) in El-Arish region. Honey samples were collected and kept in dark jars until analyses.

Table 1: Illustrated the region, honey types, and samples number for each location

Location	Sample number	English name	Plant source (scientific name)
Rafeh	9	Citrus	*Citrus* sp.
El-Arish	7	Algae	*Salvia officinalis*
	7	Myrrh	*Commiphora myrrha*
	7	Tarot	*Zygophyllum* L.

2.2. Physical Properties of Honeys

2.2.1. Viscosity and The specific gravity

Viscosity of honey was measured according to Munro, (1943), The specific gravity was measured according to Wedmore (1955). The capillary viscometric measurements were performed using an Ostwald viscometer. The viscometer was initially calibrated using doubled distilled water and 20% sucrose solution calculated amount of distilled water was added to the fresh honey sample to raise percentage water content to 25-50 range. The viscosities of the samples were measured for each of the samples at 15,25,35,45 and 60 °C.

2.2.2. Determination of electrical conductivity (EC)

According to the method of Vorwhol (1964). using model EN50081-1 at room temperature, weigh tow gm of honey dissolved in ten ml of distilled water. The results are expressed as ppm.

2.2.3. Determination of total soluble solids (TSS)

The coarse materials within honey sample should be removed before determination the (TSS) therefore; the honey sample was stirred at room temperature and pass through (0.5mm) sieve, the granulated honey samples were liquid in water bath at (40°C), and then the flask which contained honey was cooled and stirred at room temperature, (A.O.A.C.,1990).

2.3. Chemical Properties of Honeys

2.3.1. Determination of moisture content: Determination of moisture content of honey was carried out by measurement its refractive index value (Abbe refractometer at 20ºC) (A.O.A.C, 1995).

2.3.2. Determination of pH, free acids, lactone content and total acidity according the method of White *et al.* (1962).

2.3.2. Determination the quantity of sugars by High Performance Liquid Chromatography (HPLC). Concentration of fructose, glucose, sucrose and maltose in honey samples were determined by HPLC according to the method of Bogdanov and Baumann (1988).

2.4. Statistical Analysis

The results were expressed as the mean ± standard deviation of the mean, and the significant difference between means was evaluated using a one-way analysis of variance followed by a post hoc test for the comparison of significance using the Statistical Package program SPSS version 23.0 (IBM SPSS Statistics, USA). Values of $p<0.05$ were considered statistically significant.

3. Result and Discussion

3.1. Physical Properties of Honeys

Table 2 and Figure 1 provide information about the physical characteristics of different types of honey at different locations. The viscosity of honey is an important parameter that affects various processes such as mixing, filtration, and bottling. The viscosity values ranged from 48.4 ± 0.51 to 69 ± 0.36 and showed a significant difference between the samples tested. The variation in viscosity is primarily influenced by temperature and water content. Previous studies have reported viscosity values of different honey types, ranging from 14.0 to 69.0 poise. Another study found viscosity values ranging from 13.6 to 87.5 poise for Libyan honey types. However, no significant difference was observed in the viscosity values of the examined samples, which ranged from 69 ± 0.08 to 69 ± 0.36 for Matrouh honey samples El-Dereny *et al.,* 2022) and (El-Dereny,2023) viscosity varied between 46.73 and 73.12 Poise.

Table 2: The Physical Properties of Honeys at two locations (Rafeh and El-Arish) in North Sinai regions.

Location	Sample	Honey type	V.* (poise)	S.g *	EC * ppm	TSS (%)*
Rafeh	9	Citrus	69.83±1.04(a)	1.406±0.015(a)	0.0096±0.0005(a)	81.5±1.0(a)
	7	Algae	49.2±1.05(b)	1.41±0.005(a)	0.007±0.004(a)	82.4±0.57(a)
El-Arish	7	Myrrh	69.31±0.57(a)	1.413±0.11(a)	0.008±0.0001(a)	82.0±0.57(a)
	7	Tarot	48.4±0.51(b)	1.402±0.005(a)	0.007±0.001(a)	82.5±1.0(a)
F			69.83	0.596	0.269	1.16
P			0.000	0.635	0.845	0.381

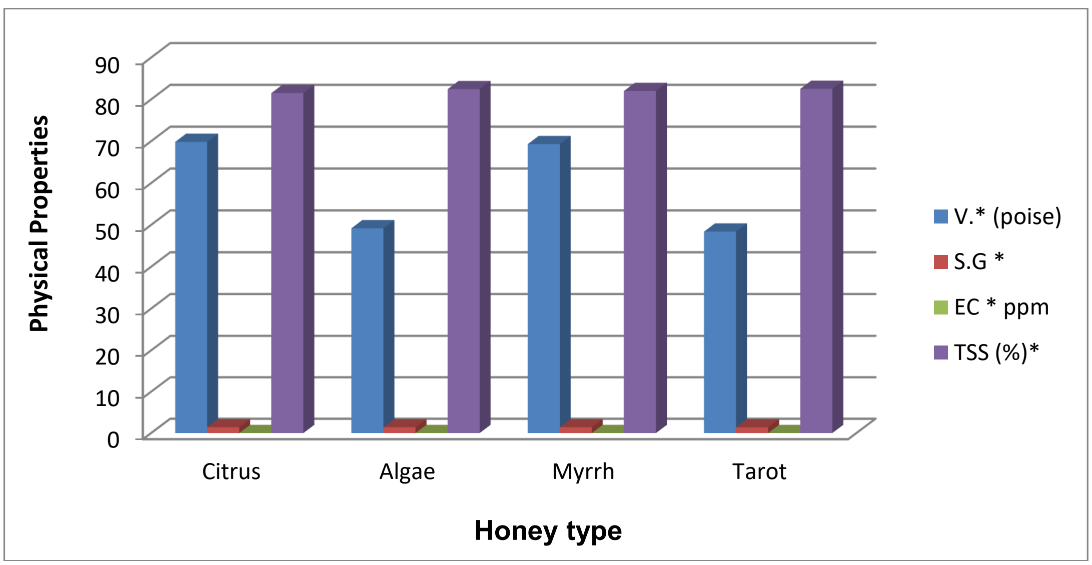

Fig 1: The Physical Properties of Honeys

Density, expressed as Specific gravity, is dependent on water content. As temperature or water content increases, density decreases, while an increase in solids concentration leads to an increase in specific gravity (Oroian, 2013). The specific gravity property of honey is not regulated by European legislation (Council, 2001). The tested honeys' densities (Table 2) and Fig. (1) show that there is no significant difference in specific gravity between the examined samples of honey ($P<0.05$). The specific gravity values ranged from 1.402 to 1.413, and all samples meet the quality standards stated in the European Legislation, European Commission (2001). These values fall within the densities reported by Nafea *et al.* (2009) for different Libyan honey types, specific gravity ranged from 1.39 to 1.43. Zidan

(2019) indicated that the specific gravity ranged from 1.415 ±0.018 to 1.417 ±0.073. The specific gravity ranges for Matrouh honeys were 1.4 ±0.01 to 1.433 ±0.005 (El-Dereny *et al.,* 2022).

The electrical conductivity of honey is an indicator of its authenticity and adulteration. It provides information about the nectar source.

According to Crane (1985), the electrical conductivity (EC) of honey is an excellent indicator of its adulteration and can reflect its original form from nectar. The EC values in table 2 and Fig.1 were 0.0073% for all Algae & Tarot and 0.0083% for Algae alone, with the highest value of 0.0096% found in Citrus. The electrical conductivity, showing no significant difference among the examined honey samples (P<0.05), ranged from 0.007 to 0.0096. All samples meet the quality standards stated in the European Legislation, European Commission (2001). The high values of EC can be attributed to high mineral content (Nour, 1988). Accorti *et al.,* (1986) suggest that EC measurement can replace ash content measurement in official analytical methods. Abde-Hameed (2020) showed that EC values ranged from 110.0 ±10 to 520.0 ±10 ppm. El-Dereny *et al.,* (2022) demonstrated that the EC values ranged from 0.008 to 0.039%. El-Dereny (2023) E.C. 0.21±0.20 and 0.70 ±0.32 (mS/cm).

These results differ from those of Habib *et al.,* (2014) who found EC values ranging from 413.81±178 to 0.48 µS cm-1 for different types of honey obtained from the United Arab Emirates. However, all honey types had EC values within the limits set by the EU directive, Egyptian honey standards, and the Codex Alimentarius Standard.

The data indicates that there is no significant difference in Total Soluble Solids (T.S.S.) values among the tested honey samples (P<0.05), which ranged from 81.5% to 82.5%. Honey with T.S.S. equal to or greater than 81.4% is classified as higher grade (A and B), while a T.S.S. falling between 80% and 81.3% is considered lower grade C (Nyau *et al.,* 2013 and QSAE, 2005). Abdel-Hameed (2020) demonstrated that the T.S.S. of Egyptian honey samples ranged from 79.0% to 87.75%. Nafea & Mazeed (2023) found a correlation coefficient between canonical discriminant functions and the original variables when comparing citrus, clover, and cotton honeys of T.S.S. = -0.66, and for sugar-feeding honeys T.S.S. = -0.55.

3.2. The chemical composition of the floral honeys including its contents of moisture, carbohydrate (glucose, fructose, maltose, sucrose), pH, free acidity, lactone and total acidity.

Table (3) presents the measured values of chemical properties for various types of honey, such as Citrus Algae, Myrrhan, and Tarot. The moisture content, which reflects the ripeness of the honey, ranged from 17.23±0.76% to 18.32±0.57% across the samples. There were no significant differences (p = 0.512, F = 0.83) in the moisture content among the honey samples, indicating that these honeys possess good storage ability. The average moisture content of the samples was found to be below the limit of 20.0 g/100 g set by Codex Alimentarius Commission (2001) and the Egyptian standard (2005), which restricts the nectar honey to a maximum of 20% moisture content. The moisture content is a crucial quality parameter, particularly for the shelf-life of honey (Bogdanov *et al.,* 2008).

Most of the samples demonstrated low moisture contents, with an average value of 17.84%. These values fall within the range reported by Bogdanov (2009) for moisture contents. The significance of moisture in honey arises from its correlation with yeast count; at a moisture content of 17.0%, the fermentation risk due to yeast is minimal.

All of the evaluated honey samples had moisture contents ranging from 17.5% to 19.1% (Nayik *et al.,* 2019). Abdel-Hameed (2020) discovered that the moisture percentages of honey samples spanned from 17.25±0.66% to 21.0±1.11%. El-Dereny (2023) found that moisture ranged from 17.35 to 18.38 %. El-Dereny *et al.,* (2022) moisture ranged of 17.5 - 18.5% as in contrast, Martínez *et al.,* (2018) reported a moisture content of 26% for Yateí honey, which conflicts with the obtained results.

Furthermore, the pH, free acidity, lactonic acids, and total acidity of the honey samples were also evaluated.

The pH level of the honey ranged from 3.3 to 3.93, indicating its acidic nature. The data revealed significant differences (p=0.512, F=0.83) between the honey samples in accordance with the standard pH limit of 3.40-6.10. The Algae honey had the highest pH value, while the Tarot honey had the lowest. The pH values of the honeys were influenced by the presence of different acids, particularly mineral contents. Previous studies have also demonstrated variations in the pH of Saudi honey, ranging from 3.8 to 4.1 (Nafea, 2004). Additionally, Essa *et al.* (2010) found that the pH values ranged from 3.70 to

4.15, with a mean of 3.8. The free acidity of honey depends on the organic acids naturally present, as well as the floral source and bee species (De Sousa *et al.,* 2016).

The total acidity values of the Egyptian honey samples varied from 25 to 43.93 meq/km. The highest free acidity was observed in Algae honey (43.93 meq/km), while the lowest was found in Citrus honey (25 meq/km). The range of free acidity was 9.93-32.07 meq/km, and lactone range was 1.0-27.76 meq/km. The free acidity remained relatively constant during the first 15 months of storage, with a slight tendency to increase afterwards (Cavia *et al.,*2007). After 20 months, most honey samples exhibited a constant increase in free acidity, which can vary due to the presence of free acids in honey. The acidity of honey is influenced by factors such as the source of nectar, the action of enzymes or bacteria and bee species. Free acidity due to chlorides, sulfates and nitrates that produce their corresponding acids (Gonzalez, 2002 and Belay *et al.,* 2013).

Lactones are considered to be verse of acidity, and their value is lower than free acidity (Krauze and Krauze, 1991 and Jimenez *et al.,* 1994). The % of, particularly gluconic acid, organic acids in equilibrium with esters or lactones, and inorganic ions, contribute to the honey acidity. The variation in acidity among different honey types may be attributed to variation in these constituents (Bath and Singh (1999) and Rodriguez *et al.,* (2004). Also, Castro-Vazquez *et al.,* (2008) found that total acidity increased significantly during the six-month storage period for both A. mellifera and A. florea. Abdel-Hameed, (2020) showed that free acidity (11.0 ±1.32 to 68.3 ±0.85), Lactone (7.5 ±0.70 to 17.5 ±0.70), total acidity (18.5 ±1.05 to 86.0 ±0.70). El-Dereny (2023) found that free acids 27.00 ±3.08 and 36.17 ±1.26 (meq./kg),lactone 1.13 ±0.48 and 4.94 ±6.40 (meq./kg) and total acidity 28.13 ±2.95 and 37. 83 ±2.02 (meq./kg).

Table 3: The chemical Properties of Honeys at two locations (Rafeh and El-Arish) of North Sinai regions

Location	**Rafah**	**El-Arish**			**F**	**P**	**LSDs**
Sample	**9**	**7**	**7**	**7**			
Honey Type	**Citrus**	**Algae**	**Myrrh**	**Tarot**			
Moisture (%)	18.0(a) ±1.0	17.23(a)±0.76	18.32(a)±0.57	17.83(a)±1.0	0.833	0.5122	1.489
pH	3.433(ab)±0.25	3.73(ab)±0.4	3.3(b)±0.3	3.93(a)±0.15	2.902	0.1015	0.549
Free acids meq/km	24.0 (b)±1.0	32.07(a)±2.64	13.38©±1.68	9.93(a)±1.2	94.79	0.0001	3.35
lactone meq/km	1.0©±1.0	11.86(b)±1.3	11.83(b)±1.98	27.76(a)±3.4	78.46	0.0007	4.059
Total Acidity meq/km	25.0(b)±1.0	43.93(a)±4.0	25.66(b)±2.6	37.7(a)±4.6	22.91	0.0003	6.33
F /G	1.55±0.21	1.33±0.05	1.38±0.11	1.53±0.17	1.723	0.239	0.275
G/W	1.51±0.04	1.71±0.07	1.58±0.06	1.49±0.191	2.37	0.146	0.219

3.2.1. Sugar contents

Sugars make up the largest portion of the dry matter in honey, which contributes to its characteristic physical properties such as high viscosity, high density, and resistance to spoilage. Monosaccharides make up approximately 75% of the sugars in honey, while disaccharides account for 10-15%, and small amounts of other sugars are also present (da Silva *et al.,* 2016).

The results of the sugar analysis of the honey samples can be seen in Tables 4 and Figures 2, 3.

The fructose content of the samples ranged from 39.21% to 40.33%, with an average of 39.68%. The glucose content varied from 26.53% to 29.6%, with a mean value of 28.04%. The sucrose content ranged from 3.73% to 5.96%, with an average of 4.52%.

The maltose content of the honey samples ranged from 6.66% to 8.36%, with a mean value of 7.62%. The fructose/glucose ratio and glucose/water ratio fell within the range of 1.32 to 1.54 and 1.49 to 1.7, with mean values of 1.44 and 1.57, respectively. The sum of fructose and glucose (fructose + glucose) content ranged from 66.33% to 68.9%, with an average of 67.73%, while the reducing sugar content varied from 70.5% to 73.2%, with an average of 72.25%.

There were no significant differences (P>0.05) in the fructose, maltose, (fructose +glucose), and reducing sugar contents among the different types of honey samples. Similarly, there were no significant differences (P>0.05) in both the fructose/glucose ratio and glucose/water ratio among the different

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honey samples from the two regions. However, the apparent glucose content of honey samples from Algae (29.6±1.35) was significantly (P<0.05) higher than that of citrus (27.12±0.85%), myrrh (28.2±0.87%), and tarot (26.53±2.2%). Additionally, the sucrose content of honey samples from citrus (4.13±1.0) and myrrh (3.3±0.3%) was significantly (P<0.05) higher than that of tarot (3.93±0.15%) and algae (4.27±0.57%).

Honeys with a high fructose/glucose ratio, greater than 1.33, do not crystallize for long periods, while honeys with a ratio less than 1.11 crystallize quickly (Smanalieva and Senge 2009). Generally, slow crystallization of honey should occur when the glucose/water ratio is less than 1.7, while a ratio greater than 2.0 leads to fast and complete crystallization (Dobre *et al.,* 2012). Recent research by Escuredo *et al.,* (2014) has confirmed that the fructose, glucose, and moisture contents, as well as the fructose/glucose and glucose/water ratios, are key indicators for predicting the crystallization phenomenon in honey. Nafea (2004) concluded that different types of honey contain 36.9-41.0% fructose, 28.3-34.2% glucose, 1.1-4.17% sucrose, and 2.8-5.5% maltose. Nafea *et al.,* (2014) reported that the sugar values of some Egyptian honey ranged from 35.1% to 38.9% for fructose, 27.7% to 32.0% for glucose, 0.75% to 2.5% for sucrose, and 2.0% to 5.0% for maltose. El-Dereny (2023) found that medicinal and aromatic honeys sugar were fructose ranged 38.50 ±5.16 and 43.11±3.88 (%), glucose 31.55 ±5.54 to 33.46 ±1.29 (%), sucrose 1.24 ±1.00 and 3.00 ±0.78%, maltose (%) 1.90 ±0.72 to 3.75 ±1.45, G/W 1.71 ±0.08 - 1.90 ±0.09 and F/G 1.40 ±0.17 and 1.25 ±0.22.

Table 4: The sugars contents of honeys at two locations (Rafeh and El-Arish) of North Sinai regions

Location		**Rafah**		**El-Arish**		**F**	**P**	**LSDs**
Sample		**9**	**7**	**7**	**7**			
Honey Type		**Citrus**	**Algae**	**Myrrh**	**Tarot**			
Sugars	**Maltose**	8.36(a) ±0.9	7.13(a)±0.90	8.33(a) ±1.1	6.66(a)±0.71	0.564	0.6515	3.727
	Fructose	39.21(a)±0.88	39.3(a)±0.7	39.86(a)±1.46	40.33(a)±1.5	0.582	0.643	2.243
	Glucose	27.12(ab)±0.85	29.6(a)±1.35	28.93(ab)±0.87	26.53(b)±2.23	2.874	0.1033	2.798
	Sucrose	4.133(a) ±1.0	4.273(b)±0.57	3.733(a)±0.47	5.966(a)±2.3	1.697	0.244	2.4704
Glucose+ fructose		66.33±1.65	68.9±1.99	68.8±1.39	66.9±1.88	1.719	0.239	3.241
Reducing sugar		70.5±0.81	73.2±2.45	72.5±1.87	72.8±3.43	0.813	0.521	4.409

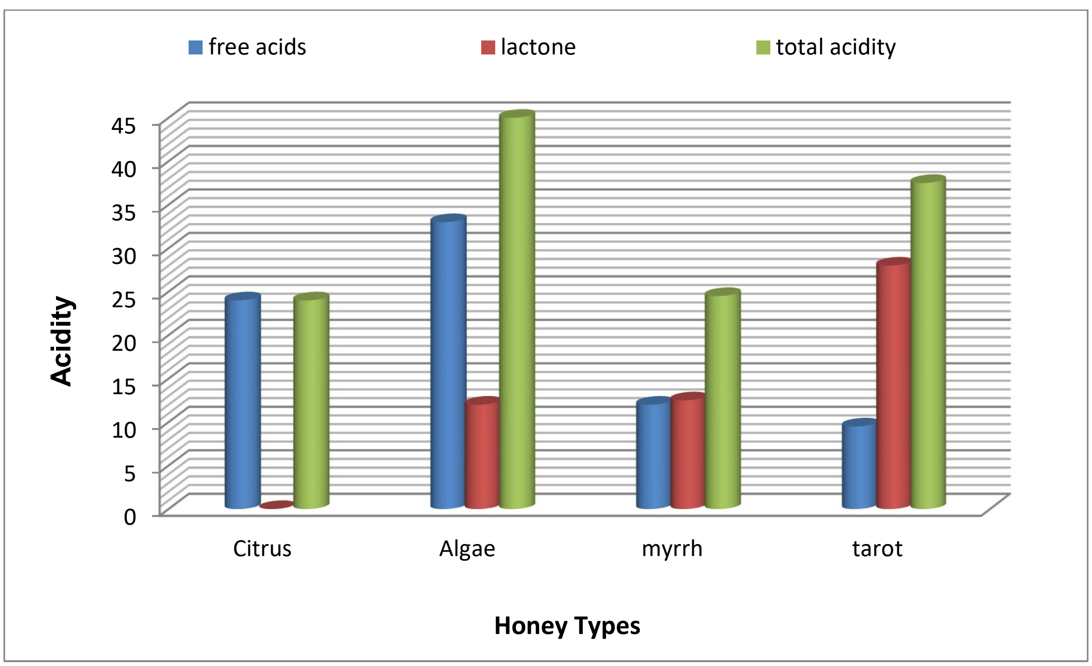

Fig. 2: Acidity of honey at two location

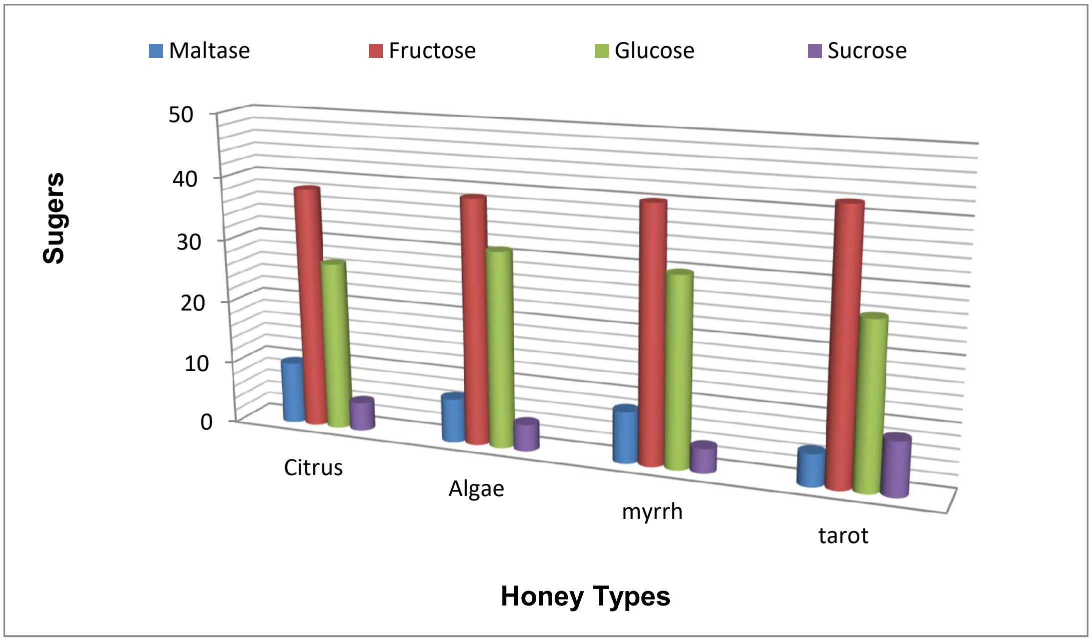

Fig. 3: Sugars of honey at two location

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