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Effect of Sugary Feeding Periods on Physicochemical Characteristics of Bees' Honey

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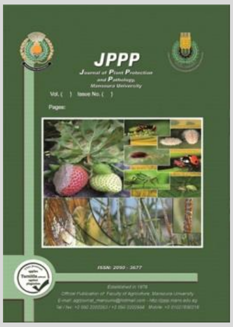

ABSTRACT

The objective of the present study is to know effect of sugary feeding periods for honeybee colonies [beginning (FSB), mid (FSM) and end (FSE) of the flowering seasons] on some physicochemical properties of of bees' honey. All data were statistically tested using analysis of variance and discriminating analysis to distinguish between the three types of honey for parameters that can be determined easily in routine honey control. The characterization of three types of honey was carried out based on their quality parameters [Moisture, total soluble solids (TSS), electrical conductivity, Specific gravity, viscosity, pH, total acidity, free acids, lactones, glucose, fructose, sucrose, maltose content, HMF and Diastase and Invertase enzymes]. The results showed that the three honey types could not be assigned by 100% into their actual groups even when all parameters were used simultaneously in the analysis. Two samples of the honey (FSM and FSE) were also assigned. Among different parameters used moisture, TSS, viscosity, sucrose content. The results were discussed in the light of some beekeeping managements before and after harvesting of honey, and the effectiveness of the chosen parameters.

Keywords: floral honey; sugar-feeding honey; physical content; chemical contents; classification; discriminating analysis.

INTRODUCTION

Honey is a natural sweet material all over the world and viscous liquid produced by honeybee (*Apis mellifera* L.) that collect the nectar from blossoms, secretions of plants and from secretions of some plant sucking insect (Soares *et al*., 2008). Honeybees collect this primary material and convert it into honey by combining with certain specific substances called enzymes deposit, dehydrate and store in the comb to ripen (Conti *et al*., 2007). Moisture content of bees' honey represents a major importance to its stability against granulation and fermentation. The low moisture content conserves honey from microbiological activity and thus it can be preserved for longer periods (Buba *et al*., 2013; Akhtar *et al*., 2014; El-Metwally, 2015). Honey also contains water (13–20%) (Alvarez-Suarez *et al*., 2013). Honey contains at least 181 components (White 1975). Although the major fundamental of honey are nearly the same in all honey samples, physical properties and the precise chemical composition of natural honeys differ according to the plant species on which the bees forage (Cantarelli *et al*., 2008; James *et al*., 2009). Honey consists primarily of sugars, at most fructose (40–50%) and glucose (32–37%), little amounts of sucrose (<2%) and mineral constituents (ash less than 0.1%). Surveys of floral honey compositions have established that the three major components are fructose, glucose, and water (Doner, 1977). In addition, di- and tri-saccharides, and some higher sugars have also been identified (Crane, 1990; Horn and Huellmann, 2002). Invertase activity ranged from 46.25 to 184.68 unit/kg, with an average of 88.61 unit/kg (Boussaid *et al*., 2014). Diastase number of examined honey samples ranged between 3.0 and 100 unit/kg, with an average of 18.32. With regard to glucose oxidase content, it ranged from 0.0 to 10.0 unit/g, with a mean value of 0.72 unit/g (El-Metwally, 2015). In Egypt, many beekeepers use sugar syrup to feed bee colonies in different periods to speed up brood rearing, and this feeding affects honey production and quality. Thus, this paper was carried out to evaluate the effect of sugary feeding at different periods on bees' honey quality. In this way, we verifying the quality of honey through comparing with the international standards (Codex, 2001).

MATERIALS AND METHODS

Honey samples

Nine honey samples were collected; (feeding until the beginning of flowering season (FSB), feeding until mid of flowering season (FSM) and feeding until the end of flowering (FSE)). The three honey samples/ groups were taken at May of 2018 and 2019 seasons and were analyzed a week after their arrived in the laboratory. In this study, the magnitude of 13 parameters was determined, to describe the different types of honey. These parameters were moisture content (%), total of soluble solids (TSS), electrical conductivity (EC), pH, free acidity, total acidity, lactone, fructose, glucose, sucrose, HMF, Diastase and Invertase. The determination of all parameters was carried out according to Bogdanov *et al.* (2004). Sugar concentration was expressed in g/100 g, and the electrical conductivity was expressed in milli Siemens per cm. Total soluble solids (TSS) was determined by using Abbe Refractometer and

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expressed in percentage. All values of free acidity and lactone were calculated to mill equivalents per kilogram.

Statistical analysis:

For each variable, F test was used to test the effectiveness of each parameter to discriminate the three types of natural honey. Discriminating analysis was used to distinguish between the three types of honey by separating the variables in 4 sets: physical, acidity sugar variables, and all other variables.

For each set of variables, the best one was determined by comparing the percentage of correctly classified cases. Wilks' Lambda test was used to test which discriminate function contributes significantly to the discrimination between studied groups. The significance of Wilk's Lambda was tested by the chi-square statistic. Discriminating analysis was used also to classify the three types of honey to each other and to 18 samples during two years. For this purpose, each group of honey type was divided into two subsets. One subset was used to estimate the discriminant function, and the other was classified based on the function rule derived for the first subset. The same procedure was repeated by classifying the first subset according to the function of the second one (Jobson, 1992). The percentage of correct classification was then used to determine the reliability of the classification rules.

RESULTS AND DISCUSSION

1. Physical properties:

The physical properties of the main Khfer El-shiehk honey presented in this study for clover (*Trifolium alexandrenum*) and Eucalyptus (*Eucalyptus globulus*) honeys.

The results presented in Table 1, showed that there are significant differences between the studied honey kinds in moisture content. The moisture content of different honey kinds was 22±1, 21±1 and 18±1% with FSB, FSM and FSE respectively, in the second year non-significant within but the lowest recorded with FSE were 19±1.277%. These results are agreeing with those obtained by Nour (1998) and Androde *et al*. (1999), who indicated that the moisture content honey was14.6-22.1%.

And indicated that there are significant differences in studying of two years between the Egyptian honey kinds in the total soluble solids percentage (TSS %); the TSS % of the studied in honey kinds were varied from (78±1, 78±0.5%), (79±2, 80±0.625%) and (82±0.0, 81±0.436%) with FSB, FSM and FSE honeys, respectively. These results are in harmony with that obtained by Mesallam and El-Shaarawy (1986), demonstrated that the TSS % of imported honey was 79.0-82.0%. A higher TSS % values were found by Abu-Tarboush *et al*. (1992). They pointed out that the TSS % values of different Saudi honey kinds were 81.8-86.6%. In the same line, Al-Khalifa and Al-Arify (1999), reported that the TSS% of unifloral Saudi honeys was 82.7-84.33%.

Non-significant differences in the electrical conductivity (EC) among the studied honey types they were ranged 0.005±0.001% to 0.007±0.002% in two studying year. The comparison of the obtained results with the values limited showed that all samples of honeys within the limit for EC ≤ 800µS/cm.

The electrical conductivity is a good criterion of the botanical origin of honey and today it is determined in routine control instead of the ash content. The differences in the electrical conductivity value in honey kinds may be attributed to the concentrations of minerals; some other constituents such as organic acids, also proteins and possibly some complexes like sugars.

Table 1. Effect of sugary feeding periods of flowering of flowering season (beginning FSB, mid FSM and end FSE) on some physical properties of bees' honey during May of 2018 and 2019 seasons.

	Treatments Parameters	Feeding until the beginning of flowering	Feeding until mid of flowering	Feeding until the end of flowering	F. value	L.S.D$_{(0.05)}$
First year	Moisture (%)	22±0.458a	20±1.609a	19±1.277a	5.74^{NS}	---
	TSS (%)	78±0.500b	80±0.625a	81±0.436a	18.42**	1.40
	Electrical conductivity (%)	0.005±0.002a	0.007±0.001a	0.007±0.002a	1.20^{NS}	---
	Specific gravity (g/ml)	1.397±0.4038a	1.402±0.265a	1.410±0.292a	0.01^{NS}	---
	Viscosity(Poise)	34.9±0.964b	34.9±1.007b	48.00±1.169a	124.23**	2.66
Second year	Moisture (%)	22±1.000a	21±1.000a	18±1.000b	9.75*	2.62
	TSS (%)	78±1.000b	79±2.000b	82±0.000a	9.75*	2.62
	Electrical conductivity (%)	0.005±0.001a	0.005±0.002a	0.005±0.001a	0.001^{NS}	---
	Specific gravity (g/ml)	1.397±0.100a	1.345±0.346a	1.417±0.464a	0.07^{NS}	---
	Viscosity(Poise)	34.9±2.100b	31.66±0.822c	69.00±0.889a	683.34**	3.11

Means in the same row followed by different letters are significantly different (L.S.D Test at 0.05).
***: Significant; **: Highly significant; NS: Non-significant.**

Specific gravity of tested honey samples obtained ranged between 1.345±0.346 and 1.417±0.464 g/ml. Maximum density average value, 1.417 g/ml was found in FSE honey samples, while, minimum average value, 1.345±0.346 g/ml, was showed in FSM honey samples. Valdes -Silverio *et al. (*2018) recorded that, the specific gravity of eucalyptus honey samples from Ecuador were ranged from 1.4 to 1.46. Zidan (2019) no significant the specific gravity was 1.415 ±0.018 to 1.417 ±0.073 of all samples were found of Sidr honeys produced in Arab countries (Egypt, Algeria, Libya and Yemen) this value meet honeys quality European Legislation, European Commission (2001).

High significant differences between the studied honey kinds in the viscosity value. The viscosity value of studied honeys was 34.9±2.1, 31.66±0.822 and 69.00±0.889 poise with FSB, FSM and FSE respectively. In the second year of studying FSE honey had the highest value of viscosity (48.00±1.169) poise followed by FSB and FSM honeys recorded 34.9±0.964, 34.9±1.007 poise respectively. Viscosity values ranged from 13.6 to 69.0 poise in Egyptian honey, while it ranged between 48.1 and 87.5 poise in Iraqian honey samples. Therefore, Viscosity of Iraqian honey was more than those of Egyptian ones (Fathy *et al.*, 2015).

2. Chemical composition for honeys

The average values of free acidity, lacton, total acidity and pH of the FSB, FSM and FSE honey samples in this studying is indicated in Table2, Free acidity content for all tested samples in the first year has no-significant among all of them, recorded 13.5±0.50, 13.5±1.00 and 12.5±1.00 meq /Kg with FSB, FSM and FSE respectively. The highest value of free acidity (13.5±1 meq/kg.) was recorded in FSB honey sample, in contrast the lowest value was found in FSE sample which was 12.5±1.00 meq/kg. lactone values of the all tested honey samples were non-significant among, recorded ranged from 16±0.50 to 16±1.32 meq. /kg in the first year. The high recorded value 18±1.323 meq. /kg in FSB honey samples, while the lowest amount 17±0.500, 17.5±1.0 meq. /kg was noticed in FSM and FSE samples. For total acidity of analyzed honey samples in the first year it was ranged from 28.5±0.50 to 29.5±1.32 meq/kg. The second year in all honey samples ranged 29±1.0 to 29±1.732 meq/kg (Table 2).

Table 2. Effect of sugary feeding periods (beginning FSB, mid FSM and end FSB) of flowering season on bees' honey acidity during May of 2018 and 2019 seasons.

	Treatments Parameters	Feeding until the beginning of flowering	Feeding until mid of flowering	Feeding until the end of flowering	F. value	L.S.D$_{(0.05)}$
First year	Free acidity (meq)/Kg	13.5±0.50a	13.5±1.00a	12.5±1.00a	1.71^{NS}	---
	Lactone (meq)/Kg	16±1.32a	16±1.00a	16±0.50a	0.001	---
	Total acidity (meq)/Kg	29.5±1.32a	29.5±1.32a	28.5±0.50a	1.71^{NS}	---
	PH	4.5±0.50a	4.2±0.30b	4.5±0.40a	9.00*	0.23
Second year	Free acidity (meq)/Kg	11±0.500b	12±0.866ab	13.5±1.000a	9.50*	1.60
	Lactone (meq)/Kg	18±1.323a	17±0.500a	17.5±1.000a	0.50^{NS}	---
	Total acidity (meq)/Kg	29±1.323a	29±1.732a	29±1.000a	0.001^{NS}	---
	PH	4.5±0.458a	4.06±0.306a	4±0.265a	1.26^{NS}	---

Means in the same row followed by different letters are significantly different (L.S.D Test at 0.05). *: Significant; **: Highly significant; NS: Non-significant.

Codex Alimentarius Standard, (1998) recommend value of total acidity of honey not more than 50.0 meq/kg. These results are in good accordance with those recorded by Crecente and Latorre (1993); Lopez *et al.* (1995) and Costa *et al.*, (1998) who reported that the total acidity content of honey were 20.0 - 61.30 meq/kg. On the same line, Paramas *et al.* (1999) noted that total acidity of sixty Spanish honey samples were 21.8 - 69.6 meq/kg.

There are significant differences in pH value between honey kinds tested in first year and non-significant among them in second year. FSE honey had the lowest value of pH (4±0.265, 4.5±0.4), followed by FSM honey (4.06±0.306, 4.2±0.30), and FSB honey which had the highest pH value (4.5±0.50, 4.5±0.458) (table 2). These results are in accordance with those reported by Mesallam and El-Shaarawy (1986); Abu-Tarboush *et al.* (1992) and Latorre *et al.* (1998), who recorded that the pH value of honey was 3.60-5.40. In the same line, Hassan and Abd El-Aal *et al.* (1997) and Paramas *et al.* (1999), noted that the pH value of honey was 3.20-5.55. In general pH value affected somewhat by the amounts of the various acids present, but mostly by the mineral content likely calcium, sodium, potassium and other ash constituents, as example honey rich in ash generally show high pH value (White, 1976).

The data given in Table 3, showed that there are significant differences among the studied honey kinds in fructose content, the maximum value was 42.9±0.20 and 43.4±0.529% with FSE honey followed by FSM honey which was 39.8±0.62 and 40±0.854%, while the lowest was 37.7±0.46 and 39.1±0.889% with FSB honey in the two years respectively. There are significant difference among FSE honey and others studied of honey kinds in glucose content, the recorded maximum were (35.2±0.66, 35±0.458%) with FSE honey followed (32.7±0.66, 32.3±0.781%) with FSM honey and the lowest with FSB honey which was (30.7±0.66, 31.4±0.721%) respectively. From the previous investigations in Egypt, El-Sherbiny *et al.* (1980) found that fructose content was ranged from 38.9 to 41.96% in Egyptian honeys, (citrus, clover, and cotton). He also found that glucose content was ranged from 33.66% to 36.50%. The range of fructose and glucose contents in several studies of Egyptian honeys were determined by Hassan and Abd Elaa (1997) (33.18-38.82% and 28.14-39.72% respectively), Nour (1988) (32.76-41.94% & 30.72-39.25% respectively) and Rateb (2005) (36.8-43.0% & 27.1-34.0% respectively). There are high significant differences among the honey kinds of studied in sucrose content during two years.

FSE honey the high maximum value was (3.2±0.28, 7.4±0.745%) followed FSM which was (2.2±0.34, 1.6±0.187%) and the lowest value was (0.83± 0.07, 0.9±0.090%) with FSB respectively. From the previous results, the percentage of fructose, glucose and sucrose contents was a distinguishing mark between the honey that collected during FSB, FSM and FSE flowering season. Sucrose content in Egyptian honey was found by many researchers such as Hassan and Abd-Elaa (1997) to be 0.20-2.82%; by Nour (1988) to be 1.76-12.7% with a mean value of 6.69% and Rateb (2005) to be 0.05-8.0% with an average of 1.979%. It was noticed that the sucrose percentage of all the honey samples was less than the maximum conventional limit of 5% recommended by the European Community (European Economic Commity, 2002). Non-significant maltose contents between the tested of honey samples during two years. The first year a range of 1.7±0.23 to 1.99%, and range of 2.3±0.321 to 2.5±0.445 in the second year (Table 3). Metwaly (2010) found that no significant differences were found between maltose content in citrus (mean value 2.01%), clover, (3.33%) and cotton (3.32%) honeys.

High significant differences were found between of the reducing sugars (F+G) among of all examined FSB, FSM and FSE honey samples ranged which between

68.4±0.72 to 78.1±0.75% and 70.5±1.552 to78.4±0.265% during two years respectively (Table 3). El-Metwally (2015) noticed that the mean values of fructose, glucose and total reducing sugars were 33.33, 28.24 and 61.56%, respectively of some Egyptian honey samples.

Table 3. Effect of sugary feeding periods (beginning FSB, mid FSM and end FSB) of flowering season on sugar percentages in bees' honey during May of 2018 and 2019.

	Treatments Parameters	Feeding until the beginning of flowering	Feeding until mid of flowering	Feeding until the end of flowering	F. value	L.S.D(0.05)
First Year	Fructose	37.7±0.46c	39.8±0.62b	42.9±0.20a	103.51**	1.01
	Glucose	30.7±0.66b	32.7±0.66b	35.2±0.66a	17.25*	---
	Sucrose	0.83± 0.07c	2.2±0.34b	3.2±0.28a	88.32**	0.497
	Maltose	1.99±0.20a	1.8±0.17a	1.7±0.23a	1.99 NS	---
	F+G	68.4±0.72c	72.5±1.28b	78.1±0.75a	238.42**	1.238
	G/W	1.6±0.09a	1.57±0.07a	1.71±0.08a	1.75 NS	---
	(G -W)/ F	0.31±0.03a	0.3±0.02a	0.34±0.03a	1.73 NS	---
Second Year	Fructose	39.1±0.889c	40±0.854b	43.4±0.529a	319.24**	0.498
	Glucose	31.4±0.721b	32.3±0.781b	35±0.458a	16.85*	1.792
	Sucrose	0.9±0.090b	1.6±0.187b	7.4±0.745a	198.36**	0.995
	Maltose	2.5±0.445a	2.3±0.321a	2.5±0.494a	4.88 NS	---
	F+G	70.5±1.552b	72.3±1.249b	78.4±0.265a	68.73**	1.961
	G/W	1.43±0.040b	1.62±0.160ab	1.84±0.105a	13.33*	0.224
	(G -W)/ F	0.24±0.020b	0.31±0.050ab	0.37±0.021a	10.23*	0.0778

(Fructose + Glucose): Reducing sugars: 65% or more (Normal Range).
Sucrose: 5% or less for clover, cotton and medical plants honey (Normal Range).
Means in the same row followed by different letters are significantly different (L.S.D Test at 0.05).*: Significant; **: Highly significant; NS: Non-significant.

In the first year the glucose to water ratio (G/W) between all examined FSB, FSM and FSE honey samples were non-significant ranged 1.57±0.07 to 1.71±0.08%, and significant among honey samples reached 1.43±0.040 to 1.84±0.105% in the second year (Table 3). El Sohaimy el al. (2015) estimated that glucose to water ratios were ranged from 0.72 to 1.56. In the same table, general average values of (glucose – water)/ fructose ((G-W)/F) ratios for all analyzed FSB, FSM and FSE honey samples of 0.3±0.02 to 0.34±0.03% was non-significant in the first year. But it was significant among all honey samples ranged 0.24±0.020 to 0.37±0.021% during the second year. Glucose and fructose constituted the primary sugars in all honey. The percentage of fructose should exceed that of glucose in honey of good quality (Kaakeh and Gadelhak, 2005). Honey samples with a glucose- water to fructose ((G-W)/F) ratio higher than 0.5 predicted rapid granulation and a ratio less than 0.2 predicted slow granulation (Manikis and Thrasivoulou, 2001).We concluded from this point that FSB honey samples are slow granulation when compared to FSM and FSM honey samples.

The data presented in Table 4, indicated that there are Non-significant differences between the studied honey kinds concerning their HMF content. The HMF content of the studied honey kinds, was ranged from 6.79 to 8.18 mg/kg. FSB honey had the lowest value (7.46± 0.22 mg/kg) followed by FSM honey (7.62± 0.21 mg/kg), while the FSE honey had the highest value of HMF (7.70± 0.27 mg/kg). The HMF content of the studied honey kinds were considered lower than the values recommended by Codex Alimentaruis Standard (1998) which recommended HMF contest not more than 60mg/kg. Fatehe (2013) found that the HMF concentrations in certain Egyptian honey samples ranged from zero to 13.44 mg/ kg. El-Metwally (2015) recorded that the HMF content in investigated Egyptian honey samples was 15.05 mg/kg.

Table 4. Effect of sugary feeding periods (beginning FSB, mid FSM and end FSB) of flowering season on Hydroxy methyl furfural (HMF) in bees' honey during May of 2018 and 2019 seasons.

Treatments Test years	Feeding until the beginning of flowering	Feeding until mid of flowering	Feeding until the end of flowering	F. value	L.S.D(0.05)
First year	7.68± 0.38a	7.68± 0.28a	7.68± 0.17a	0.001 NS	---
Second year	7.23± 0.23a	7.55± 0.41a	7.72± 0.39a	1.67 NS	---
Range	6.79 - 8.13	7.06 - 8.18	7.26 - 8.14	----	----
Average	7.46± 0.22a	7.62± 0.21a	7.70± 0.27a	2.73 N.S.	---

Means in the same row followed by different letters are significantly different (L.S.D Test at .05).
***: Significant; **: Highly significant; NS: Non-significant.**

The data present in table 5, show the Diastase and Invertase activity. The diastase number ranged 5.45 to 6.82 units in all tested of honey types. Diastase the honey was not significant in the first year. The high significant value (6.82±0.15units) was noticed in FSB honey samples followed FSM honey samples 6.10±0.48 units, while, FSE honey samples had the lowest value (5.56±0.42 units). These results with agreement the diastase number of some Egyptian honey samples ranged between 3.0 and 100.0 u/kg, with general mean value of 18.32 u/kg. The diastase activity of certain Argentinean honey types ranged between 3.9 and 39.3 Goth units (Aloisi, 2010).

There are non-significant differences between the studied of all honey samples concerning their invertase enzyme activity content. In the first year FSB honey samples recorded high values 41.5± 1.32 unit/kg, followed FSM honey samples 41.32±0.95unit/kg. and 41.32± 0.46unit/kg. for FSE. These results nearly similar with second year results. Dinko (2014) recorded that invertase activity of some Bulgarian multifloral honey was 6.06 ± 5.92. Invertase is the enzyme responsible for converting sucrose to fructose and glucose which are the main sugars in honey (White, 1975). Many beekeepers care for activating their colonies early in the season, so that they would produce high quantity of forager bees at the beginning of flowering season. The foragers are thought to be responsible for producing enzymes which are important in converting nectar to honey (Costa and Cruz-Landim, 2002).

Table 5. Effect of sugary feeding periods (beginning FSB, mid FSM and end FSB) of flowering season on Diastase and Invertase (U/Kg) activity in bees' honey during 2018 and 2019 seasons.

	Treatments Test years	Feeding until the beginning of flowering	Feeding until mid of flowering	Feeding until the end of flowering	F. value	L.S.D$_{(0.05)}$
Diastase enzyme	First year	6.82±0.15a	6.00±0.78a	5.66±0.33a	5.02^{NS}	---
	Second year	6.60±0.30a	6.20±0.53a	5.45±0.48b	9.35*	0.75
	Average	6.71±0.19a	6.10±0.48a	5.56±0.42a	2.19^{NS}	---
	Range	6.60 – 6.82	6.00 – 6.20	5.45 – 5.66	---	---
Invertase enzyme	First year	41.5± 1.32a	41.32±0.95a	41.32± 0.46a	0.15^{NS}	---
	Second year	41.2± 1.07a	41.1±0.86a	41.22± 0.46a	0.10^{NS}	---
	Average	41.2± 1.19a	41.1± 0.91a	41.22± 0.46a	0.09^{NS}	---
	Range	40.9- 42.31	40.24- 41.96	40.71 - 41.73	---	---

Means in the same row followed by different letters are significantly different (L.S.D Test at 0.05).
***: Significant; **: Highly significant; NS: Non-significant.**

Discrimination analysis of three main types of Egyptian honey

The objective of the present study is to use discriminating analysis to distinguish, three bee honey types by testing the equality of the averages investigated of the physicochemical parameters according to the sugary feeding periods by parameters that can be determined easily in routine honey control.

The characterization of three honey types (feeding until the beginning of flowering, feeding until mid of flowering and feeding until the end of flowering) and sugar-feeding honey was carried out based on their quality .The parameters under this study were, the magnitude of 15 parameters chosen for describing the different types of honey. Total of soluble solids (TSS), electrical conductivity (EC), viscosity, pH, free acidity, total acidity, lactone, moisture, fructose, glucose, sucrose, maltose, HMF, diastase and invertase. The determination of all parameters was carried out according to Bogdanov *et al.* (2004). Discriminating analysis was used to discriminate between three types of natural honey by separating the variables in 4 sets: physical, acidity sugar variables, enzymes activity and all 15 variables.

The first function discriminated the three groups significantly from each other Table 6 (Chi-square = 66.200, P<0.000) as shown in Fig. (1) the 3 groups could be separately obviously from each other.

By using the standardized coefficients of two functions it could be known each character participle effectively in the separation. For each set of variables, the best one was determined by comparing the percentage of correctly classified cases. Wilks' Lambda test was used to test which discriminating function contributes significantly to the discrimination between groups studied. The significance of Wilk's Lambda was tested by the chi-square statistic.

Table 6. Multivariate analysis of variance for testing the equality of the averages estimated of the physicochemical parameters according to the groups' sugary feeding periods.

Parameters	Wilks' Lambda	F	df1	df2	P ≤ 0.05
Moisture %	0.541	6.369	2	15	0.010**
T.S.S %	0.432	9.853	2	15	0.002**
Electrical conductivity %	1.000	0.000	2	15	1.000^{NS}
Specific gravity (g/ml)	0.995	0.035	2	15	0.965^{NS}
Viscosity(Poise)	0.347	14.145	2	15	0.000***
Free acidity (meq)/Kg	0.985	0.111	2	15	0.896^{NS}
Lactone (meq)/Kg	0.978	0.167	2	15	0.848^{NS}
Total acidity (meq)/Kg	0.896	0.870	2	15	0.439^{NS}
PH	0.970	0.232	2	15	0.796^{NS}
Fructose	0.984	0.125	2	15	0.884^{NS}
Glucose	0.992	0.061	2	15	0.941^{NS}
Sucrose	0.549	6.161	2	15	0.011**
Maltose	0.997	0.020	2	15	0.980^{NS}
Diastase (DN)	0.889	0.937	2	15	0.414^{NS}
Invertase (U/Kg)	0.997	0.021	2	15	0.979^{NS}
HMF	0.982	0.134	2	15	0.876^{NS}
Discriminant Scores from Function 1 for Analysis 1	0.000	47531.196	2	5	0.000***
Discriminant Scores from Function 2 for Analysis 1	0.219	8.927	2	5	0.022**

Discriminating analysis was used also to classify the three types of honey to each other and to 9 samples of honey. For this purpose, each group of honey types was divided into two subsets. One subset was used to estimate the discriminate function, and the other was classified based on the function rule derived for the first subset. The same procedure was repeated by classifying the first subset according to the function of the second one (Jobson, 1992). The percentage of correct classification was then used to determine the reliability of the classification rules. Results discriminate analysis showed that two discriminate functions were formed significant among these honey samples. Parameters analysis showed significant differences on moisture, TSS**,** viscosity and sucrose according to honey types Wilks' Lambda between 0.347 to 0.549 (P = < 0.011 to 0.000) (Table 6). The discriminate two functions was used for the classification of honey types according to the sugary feeding periods , since it explained 100% of total variance and a good canonical correlation equal to0.997. In addition, the standardized canonical discriminate function coefficients correlation for each of the significant physicochemical parameters that contributed to the sugary feeding periods' discrimination of honey types (Table 7). In the end summary regarding the identification of the variables with the highest discriminatory power, higher the absolute value of a standardized canonical coefficient, the more significant the variable is for the determination of honey types. Remarkable, discrimination ability of conventional physicochemical parameters, thus multivariate data evaluation of traditional physical and chemical measures and may also be helpful to establish new criterion for a more reliable description of the honey types and for the determination of their honey types.

Table7. Standardized Canonical Discriminate Function Coefficients

Parameters	Function 1	Function 2
Fructose	7.063	-18.235
Sucrose	-2.863	22.368
Maltose	-.417	9.407
HMF	-5.603	5.198
Discriminate Scores from Function 1 for Analysis 1	2.748	1.508

By using the standardized coefficients of two functions it could be known each character participle effectively in the separation. Evaluation of the different characters in the two functions in table 7, the best character which participle in the separation in different groups in function 1 are fructose (7.063), sucrose (-2.863), maltose (-.417) and HMF(-5.603) are less effective in the separation in 3 groups. On the other hand, the best character participle effectively in the separation by the function 2 is sucrose (22.368).

Classification and discriminate of the three groups to each other:

The three groups were classified to each other to know the misclassification rate of each group in relation to other groups. As shown in table 8, the three groups (FSB, FSM and FSE honeys) could be classified correctly (100, 66.7 and 50 % for FSB, FSM and FSE honeys, respectively) to their groups. The results showed that FSB, FSM, and FSE honey could not be assigned by 100% into their actual groups even when all parameters were used simultaneously in the analysis. FSM and FSE honey samples were assigned as sugar-feeding honey. Among different parameters used, moisture, TSS, viscosity, sucrose content, accounted for the most distinguishing parameters between the different honey types. The results were discussed in the light of some beekeeping managements before and after harvesting of honey, and the effectiveness of the chosen parameters.

Table 8. Discriminatory power of the developed statistical model for the classification of sugary feeding periods for the honey bee colonies.

		Treatments	Predicted Group Membership: Feeding till beginning	Feeding till mid	Feeding till end	Total
Original	Count	Feeding till beginning	6	0	0	6
		Feeding till mid	0	4	2	6
		Feeding till end	0	3	3	6
	%	Feeding till beginning	100.0	0	0	100.0
		Feeding till mid	0	66.7	33.3	100.0
		Feeding till end	0	50.0	50.0	100.0
Cross-validated[a]	Count	Feeding till beginning	3	2	1	6
		Feeding till mid	0	0	6	6
		Feeding till end	0	6	0	6
	%	Feeding till beginning	50.0	33.3	16.7	100.0
		Feeding till mid	0	0	100.0	100.0
		Feeding till end	0	100.0	0	100.0

a. Cross validation is done only for those cases in the analysis. In cross validation, each case is classified by the functions derived from all cases other than that case.
b. 72.2% of original grouped cases correctly classified.
c. 16.7% of cross-validated grouped cases correctly classified.

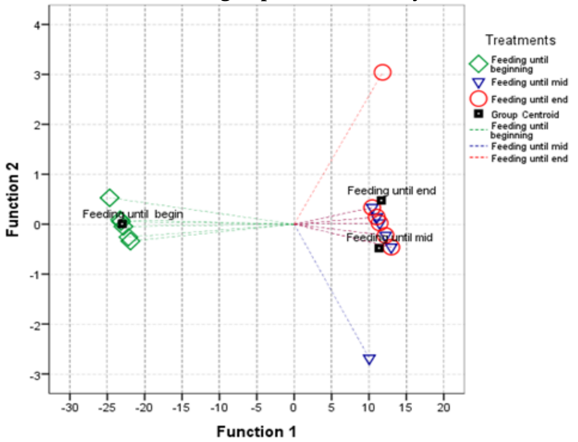

Fig. 1. Canonical discrimination functions

The pervious results, ease of application and reproducibility, have been previously reported in the literature in studies involving Spanish (Serrano *et al.*, 2004 and Karabagias *et al.*, 2017), Moroccan (Chakir *et al.*, 2016) and Greek (Karabagias *et al.*, 2014) unifloral honeys, in agreement with the present results. Ruoff *et al.* (2007) stated that several exceptions are listed in the above-mentioned standards, thus indicating the limited value of this measure and for the discrimination of honey types.

REFERENCES

Abd El-Aal, E. M.; Ziena, H. M. and Youssef, M. M. (1993). Adulteration of honey with high fructose corn syrup: Detection by different methods. Food Chemistry,48, 1209-1212.

Abu-Tarboush, M. H.; Hassan, A. K. and El-Sarvage M. S. (1992). Floral kind identification and Quality evaluation of some honey kinds. Food Chemistry 46, 3-17.

Akhtar, S., Ali, J., B. Javed, Hassan S., Abbas, S. and Siddique, M. (2014). Comparative physiochemical analysis of imported and locally produced Khyber Pokhtunkhwa honey. Global J. Biotechnol. Biochem 9(3): 55- 59 .

Al-Khalifa, A. S. and AL-Arify, I. A. (1999). Physicochemical characteristics and pollen spectrum of some Saudi honey. Food Chemistry 67, 21-25.

Aloisi, P. V. (2010). Determination of quality chemical parameters of honey from Chubut (Argentinean Patagonia). Chil. J. Agr. Res. 70(4): 640-645.

Alvarez-Suarez, J. M., Giampieri, F. and Battino, M. (2013). Honey as a source of dietary antioxidants: Structures. Bioavailability and evidence of protective effects against human chronic diseases. Current Medicinal Chemistry 20 (5): 621 – 638.

Androde, B.; Amaral M.; Teresa, P.; Joa, C.; Seabra, M. and Antonis, C. (1999). Physicochemical attributes and pollen spectrum of Porluguese heather honeys. Food Chemistry 66, 503-510.

Bogdanov, S., Ruoff, K., P., and Oddo, L. (2004). Physico-chemical methods for the characterisation of unifloral honeys: A review. Apidologie, Special Issue, 35(Suppl. 1), S4–S17.

Boussaid, A., Chouaibi, M., Rezig, L., Hellal, R., Donsi, F., Ferrari, G. and Hamdi, S. (2014). Physicochemical and bioactive properties of six honey samples from various origins from Tunisia. Arabian Journal of chemistry 11(2): 265-274.

Buba, F., Gidado A., Shugaba A. (2013). Analysis of biochemical composition of honey samples from North- East Nigeria. Biochem. Anal. Biochem 2(3):(139-150).

Cantarelli, M. A., Pellerano, R. G., Marchevsky, E. J. and Camina, J. M. (2008). Quality of honey from Argentina: Study of chemical composition and trace elements. J. Argent. Chem. Soc., 96(1-2): 33-41.

Chakir, A.; Romane, A.; Marcazzan, G.L. and Ferrazi, P. (2016). Physicochemical properties of some honeys produced from different plants in Morocco. Arab. J. Chem., 9: S946–S954.

Codex Alimentarius (1998). Draft revised for honey at step 6 of the codex procedure. Cx P 5/10.2, Cl 1998/12-S 1998. F.A.O., Roma, Italy (c.f.Rateb, 2006).

Codex Alimentarius Commission (2001). Draft revised for honey of the Codex Procedure. FAO; Rome, Italy.

Conti, M. E., Stripeikis, J., Campanella, L. D. C. & Tudino, M. B. (2007). Characterization of Italian honeys (Marche Region) on the basis of their mineral content and some typical quality parameters, Chemistry Central Journal, 1: 14, 1 -10.

Costa RAC and Cruz-Landim C (2002). Enzymatic activity of hypopharyngeal gland extracts from workers of *Apis mellifera* (Hymenoptera, Apidae, Apinae). Sociobiology 40: 403-411.

Crane, E. (1990). Bees and beekeeping. Science, Practice and World Resources. Cornstock Publ., Ithaca, NY., USA. 593 p.

Crecente, P. and Latorre, H. (1993). Paltern recognition analysis applied to classification of honey from tow geographic origins. Agric.Food.Chem,41, 560-564.

Dinkov, D. H. (2014). Quality parameters of Bulgarian kinds of bee honey. Mac. Vet. Rev. 37:35-41.

Doner, L. W., While, J.W. and Phillips, J.W. (1977).Gas-liquid chromatographic test for honey adulteration by high fructose corn syrup. J. AOAC 62(1): 186-189.

El Sohaimy, S. A., Masry, S. H. D. and Shehata, M. G. (2015). Physicochemical characteristics of honey from different origins Annals of Agricultural Science 60(2) 279-287.

El-Sherbiny, G. A.; Rizk S. S.; El-Ashwah, F. A.; Heikal, H. A. (1980). Chemical composition of citrus honey produced in A.R.E . Agri. Res. Rev, 58: 289-297.

European Commission (2001). Council Directive 2001/110 relating to honey. Official Journ al of the European Communities. The antibacterial activity of honey1. eucalyptus honeys by discriminate analysis. Food Chem., 87: 619–625.

European Economic Comminity (2002). Council Directive of 20 December 2001 relating to honey. Official Journal of the European Communities, 110: 47 – 50.

Fathy, H. M., Awadalla, S. S., El-Batran, L. A. and Al-Mashhadani, M. H. A. (2015).Studies on some physical properties of Egyptian and Iraqian honey. J. Plant Prot. And Path., Mansoura Univ., 6(11):1567 – 1577.

Hassan, A. R. and Abd-Elaa, H. A.(1997). Physicochemical analysis of some honey type in Egypt. Sugar and S. Subst. in Fd. Proc. & Nutr. Ismailia, 14-16 Oct.

Jobson, J. D. (1992). Applied multivariate data analysis. Vol. II:Categorial and multivariate methods. Springer-Verlag.

Kaakeh, W. and Gadrlhak G.G. (2005). Sensory evaluation and chemical analysis of Apis mellifera honey from the Arab Gulf Region. Journal of Food and Drug Analysis, 13: 331-337.

Karabagias, I.K.; Badeka, A.V.; Kontakos, S.; Karabournioti, S.; Kontominas, M.G. (2014). Botanical discrimination of Greek unifloral honeys with physico-chemical and chemometric analyses. Food Chem., 165: 181–190.

Karabagias, I.K.; Louppis, P.A.; Karabournioti, S.; Kontakos, S.; Papastephanou, C. and Kontominas, M.G. (2017). Characterization and geographical discrimination of commercial Citrus spp. honeys produced in different Mediterranean countries based on minerals, volatile compounds and physicochemical parameters,using chemometrics. Food Chem. 217: 445–455.

Latorre, M.; Pena, R.; Pita, C.; Botana, A.; Garcia., S. and Herrero C. (1998). Chemometric classification of honey succoring to their kinds. I-metal content data .Food Chemistry 66, 263-268.

Lopez, A.; Latorre, M.; Fernandez, M.; Garcia, M.; Garcia, S. and Herreo, C. (1995). Chemometric classification of honeys according to their kind based on quality control data. Food chemistry, 55, (3) 281-287.

Manikis, I. and Thrasivoulou, A. (2001). Relation of physicochemical characteristics of honey and the crystallization sensitive parameters. Apiacta 36: 106-112.

Mesallam, S. and El-Shaarawy, I. (1986).Quality attributes of honey in Saudi Arabia. Food chemistry , 2S, 1-11.

Metwaly, A. A. A. (2010) . The chemical composition of different types of honeybee produced from different botanical origin. Thesis Msc, Fac. of Agric. Al-Azhar Uni. 125p.

Nour, M. E. (1988). Some factors affecting quality of Egyptian honey. Ph. D. Thesis, faculty of Agric. Cairo Univ.

Paramas, A.M.; Gonzalez, J.A.; Gomez, B.R.; Garcia V.J.; Tereso, R.P.; Ramon, A.A. and Jose, S.M . (1999).Geographical discrimination of honey by using mineral composition and common chemical quality parameters. J. Sci. Food Agric. 80,157-165.

Ruoff, K.; Werner, L.; Verena, K.; Jacques, O. B.; Katharina, V.; Werner, V. and Renato, A. (2007). Authentication of the botanical origin of honey using profiles of classical measure and discriminate analysis. Apidologie 38: 438–452.

Serrano, S.; Villarejo, M.; Espejo, R. and Jodral, M. (2004). Chemical and physical parameters of Andalusian honey: classification of citrus and eucalyptus honeys by discr iminate analysis. Food Chem. 87: 619-625.

Soares, J., Soares, N., Pires, M. L., Novaes, S. and Lacerda, J. J. (2008). Honey Calssification from Semi-Arid, Atlantic and Transitional Forest Zones in Bahia, Brazil, J. Braz, Chem. Sec., 19, 502 – 508.

White, J. W. (1975). Physical characteristics of honey. In: Honey, a comprehensive survey, Crane (ed.), Heinemann, London, U. K: 207- 239.

White, J. W. (1976). Honey. Unpublished observations. U.K.

Zidan, E. W. (2019). Classical classification and discrimination analysis of physicochemical characters of Sidr honey produced in some Arab countries. Egypt. J. Plant Prot. Res. Inst. 2 (2): 387 – 397.

تأثير فترات التغذيه السكريه لطوائف نحل العسل على الخصائص الفيزيائيه والكيميائيه لعسل النحل

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الهدف من هذه الدراسه هو معرفة تأثير فترات التغذيه السكريه لطوائف نحل العسل في بداية ومنتصف ونهاية موسم التزهير على بعض الخواص الفزيائيه على جودة عسل النحل المنتج، تم اختبار جميع البيانات إحصائياً بإستخدام تحليل التباين والتحليل التمييزي للتمييز بين ثلاثة أنواع من العسل، المعلمات التى يمكن تحديدها بسهوله في مراقبة العسل الروتينيه، تم إجراء توصيف ثلاثة أنواع من العسل بناءً على معايير الجوده الخاصه بها (الرطوبه – المواد الصلبه الذائبه الكليه – التوصيل الكهربائي – الثقل النوعى – اللزوجه – الأس الهيدروجينى – الحموضه الكليه – الأحماض الحره – اللاكتونات – الجلوكوز – الفركتوز – السكروز – المالتوز – محتوى HMF – إنزيمي Diastase, Invertase) ،تم إختبار جميع البيانات إحصائياً بإستخدام تحليل التباين والتحليل التفاضلي الذى تم إستخدامه فى هذه الدراسه كالتعرف على الأنماط كأداة تصنيف للتعرف على أهم المتغيرات فى التصنيف، وأظهرت النتائج أنه لا يمكن تخصيص أنواع العسل الثلاثه بنسبة 100% فى مجموعاتها الفعليه حتى عند إستخدام جميع المعلمات فى وقت واحد فى التحليل وعينات من العسل تم تخصيصها لعسل منتصف ونهاية موسم التزهير ومن بين المعلمات المختلفه المستخدمه (الرطوبه – المواد الصلبه الذائبه – اللزوجه – محتوى السكروز) وتمت مناقشة النتائج فى ضوء بعض إدارات تربية النحل قبل وبعد حصاد العسل وفاعلية المعايير المختاره.