academicJournals

Vol. 7(7) pp. 168-173, July 2013
DOI: 10.5897/AJFS2013.0986
ISSN 1996-0794 ©2013 Academic Journals
http://www.academicjournals.org/AJFS

African Journal of Food Science

Full Length Research Paper

Physicochemical characterization of some multifloral honeys from honeybees *Apis mellifera* collected in the Algerian northeast

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Accepted 4 June, 2013

The characterization of some Algerian northeast honey was carried out on the basis of their physico-chemical properties: moisture, hydroxymethylfurfural, diastase activity, pH, free, total and lactonic acidity, electrical conductivity, mineral and proline content. Studied samples are found to be low in moisture and therefore safe from fermentation, low in HMF level and high in diastase activity. Additionally, the diastase activity and the HMF content are widely recognized parameters indicating the freshness of honey. The quality of honey samples differs on account of various factors such as season, packaging and processing conditions, floral source, geographical origin and storage period. It is important that precautions should be taken to ensure standardization and rationalization of beekeeping techniques, manufacturing procedures and storing processes to improve honey quality.

Key words: Honey, physico-chemical characterisation, HMF, diastase activity.

INTRODUCTION

Honey is a very important energy food and is used as an ingredient in hundreds of manufactured foods, particularly in cereal-based products, for its sweetness, colour, flavour, caramelisation and viscosity (Rodrigez and Ferrer, 2004). The composition of honey depends on the plant species visited by the honeybees and the environmental, processing and storage conditions (Bertoncelj et al., 2007; Guler et al., 2007). Honey produced by the honeybee is a natural super saturated sugar solution, which is mainly composed of a complex mixture of carbohydrates. In addition, to carbohydrates, it also contains certain minor constituents, including proteins, enzymes (invertase, glucose oxidase, catalase, phosphatases), amino and organic acids (gluconic and acetic acids), lipids, vitamins (ascorbic acid, niacin, pyridoxine), volatile chemicals, phenolic acids, flavonoids and carotenoid-like substances and minerals (Blasa et al., 2006). Fresh honey is usually heated in order to facilitate processing and to maintain good quality. However, excessive heat treatment leads to the formation of hydroxymethylfurfural (HMF) and reduced honey quality. HMF value is virtually absent or very low in fresh honey and is high in honey that has been heated, stored in inadequate conditions, or adulterated with invert syrup (Nozal et al., 2001). Chemical properties of honey such as pH, mineral content and total acidity also affect HMF content. The presence of organic acids and low water activity also favours the production of HMF (Kalabova et al., 2003). The Codex Alimentarius (2001) and International Honey Commission (2002) set the maximum concentration of HMF to 40 mg/kg for honey from non-tropical regions and 80 mg/kg for honey from tropical regions.

Extremely high (>500 mg/kg) HMF values demonstrate adulteration with invert syrup (Coco et al., 1996). There is a vast number of floral species characteristic of Algeria

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Figure1: Maps of the geographical origin of the honey samples.

visited by the honeybees. However, reports of the chemical composition of Algerian honeys are scarce (Makhloufi et al., 2007; Ouchemoukh et al., 2007). The potential honey production is very high in the Algerian northeast; this is related to the botanical wealth and the biodiversity of this region. Therefore, this study was carried out to evaluate the physicochemical quality of twenty Algerian Northeast honey samples from *Apis mellifera*.

MATERIALS AND METHODS

Honey samples

Twenty blossom honeys from various regions of Algerian Northeast were used. The samples of honey collected from bee-keepers were classified according to their botanical origin using the method established by the International Commission of Bee Botany described by Louveaux et al. (1978). The result shows that all samples are multifloral honey (no dominant pollen) with the presence of Secondary pollen (16 to 45%): *Trifolium* sp, *Eucalyptus*, *Hedysarium coronarium*, *Echium* and *Rosaceae*. Minor pollen (3 to 15%): Prunus / *Pyrus*, *Fabaceae*, *Carduus* and *Erica arborea*. Pollen very minor or isolated (<3%): *Allium* spp, *Apiaceae*, *Lavandula asphodelus*, *Lavandula stoechas*, *Lamiaceae*, *Brassicaceae*, *Taraxacum*, *Euphorbiaceae*, *Rhamnaceae*, *Asteraceae*, *Prunus / Pyrus*, *Mimosa pudica*, *Helianthus*, *mimosa bimucromata*, *Arctium* and X (indeterminate species). All the samples were collected in six different region of Northeast Algeria during the years 2004 to 2007 (Figure 1 and Table 1).

Physico-chemical analysis

Water content was determined by refractometry, pH was assessed in a 10% (w/v), total acidity, free acid, lactone and proline contents were measured in fresh honey samples using AOAC methods (Helrich, 1990). Proline content was determined by the measurement of the absorbance at 510 nm of the resulting product between proline and ninhydrin. Mineral content and electrical conductivity were determined by the methods of Bogdanov et al. (1997). Ash content was determined by heating 5 g of honey at 625°C in a muffle furnace. Electrical conductivity was measured in a 20% (w/v) solution of honey. The protein content was determined by the method of Bradford (1976), the quantity of proteins was estimated at 595 nm in relation to bovine serum albumin standard. HMF and diastase activity were measured as recommended by the

Table 1. Botanical and geographical origins of honey samples

Samples	Botanical origin	Geographic origin	Year collected
S1	multifloral	El taref	2006
S2	multifloral	Tebessa	2007
S3	multifloral	Tebessa	2007
S4	multifloral	El taref	2007
S5	multifloral	Annaba	2007
S6	multifloral	El taref	2004
S7	multifloral	Annaba	2004
S8	multifloral	El taref	2004
S9	multifloral	Skikda	2004
S10	multifloral	Skikda	2004
S11	multifloral	El taref	2004
S12	multifloral	SoukAhras	2004
S13	multifloral	Tebessa	2004
S14	multifloral	Guelma	2004
S15	multifloral	SoukAhras	2004
S16	multifloral	Annaba	2005
S17	multifloral	Guelma	2005
S18	multifloral	SoukAhras	2005
S19	multifloral	Tebessa	2006
S20	multifloral	Annaba	2006

Codex Alimentarius Commission (CAC, 1989).

RESULTS

Table 2 shows the mean values, standard deviations and ranges of the data of the different physico-chemical parameter analyses. The parameters indicating product maturity had values that fell into the limits set by current European Community requirements.

DISCUSSION

Moisture content

The moisture content in the investigated samples ranged from 13.7 to 18.09% (Table 2). All tested Algerian honeys had moisture content below 20%, which is the maximum prescribed limit for the moisture content as per the Codex standard for honey (Codex Alimentarius, 2001). The different moisture content of honey depends on harvest season, the degree of maturity reached in the hive and moisture content of the plant nectar source (Finola et al., 2007).

pH

The pH values of honey are of great importance during extraction and storage, since acidity can influence the texture, stability and shelf life of honey (Terrab et al., 2003). All honeys are acidic, having a pH in the range of 3.33 to 4.6 (Table 2). The acidity of honey is due to the presence of organic acids, particularly gluconic acid; and inorganic ions such as phosphate and chloride (Nanda et al., 2003). These results agreed with data reported by Azeredo et al. (2003). In general, honey is acidic in nature irrespective of its variable geographical origins. The pH values of Algerian, Brazilian, Spanish, Turkish and Indian honeys have been found to vary between 3.49 to 4.53, 3.10 to 4.05, 3.63 to 5.01, 3.67 to 4.57 and 3.49 to 4.43, respectively (Azeredo et al., 2003; Ouchemoukh et al., 2007; Kayacier and Karaman, 2008; Sudhanshu et al., 2010).

Free acidity of honeys

Free acidity of all twenty samples fell within the permitted range proposed by Codex Alimentarius (2001) of not more than 50 milliequiv acid/kg. The free acidity of honey samples in this study ranged from 25.763 to 44.884 milliequiv acid/kg (Table 2).

Lactonic acidity of honeys

Lactonic acidity of the honey ranged from 7.063 to 10.9 milliequiv acid/kg (Table 2).

Total acidity of honeys

Total acidity of the honey ranged from 34.363 to 54.317

Table 2. Physico-chemical characteristics of some Algerian honey samples.

Samples	Moisture %	pH	Electric Conductivity (µs/cm)	Mineral content %	Free acidity (meq/Kg)	Lactonic acidity (meq/Kg)	Total acidity (meq/Kg)	HMF (mg/Kg)	DN Schade unite	Proline content mg/Kg
S1	15.4 ±0.23	4.4 ±0.02	309 ±1	O.128 ±0.001	25.763±0.37	8.6±0.01	34.363±0.38	4.1 ±0.45	198.0 ±0.45	454.3±0.01
S2	13.8 ±0.453	4.6 ±0.03	686 ±2	0.441 ±0.084	28.098±0.05	9.073±0.4	37.171±0.45	9.7 ±1.15	117.23 ±O.12	110.7 ±0.52
S3	14.1 ±0.125	4.1 ±0.03	314 ±2	0.35 ±0.067	41.231±0.50	10.9±0.75	52.131±1.25	10.3 ±0.16	100.33 ±2.67	100 ±0.388
S4	17.7 ±0.651	4.1 ±0.035	1050.5±1.5	0.518 ±0.013	32.7±0.35	7.983±0.00	40.683±0.35	7.4±0.91	154.1 ±2.11	364 ±0.632
S5	13.7 ±0.587	4.4 ±0.023	326.5 ± 0.5	0.111 ±0.054	35.7±0.18	10.873±0.50	46.573±0.68	4.0 ±0.33	202.123 ±3.56	502.5 ±0.752
S6	15.21 ± 0.685	3.33 ± 0.351	349 ± 5.13	0,37 ± 0.036	29.9±1.00	7.063±1.00	36.963± 2	10.217 ± 0.529	44.45 ± 4.0	403.36 ± 0.557
S7	16.63 ±0.34	4.027 ±0.006	499.5 ± 1.5	0,44 ± 0.095	44.884±2.517	9.10±0.90	54.317 ±1.303	11.26 ± 0.276	51.82 ± 4.06	305.67 ±0.58
S8	16.18 ±0.16	3.47 ± 0.334	269.33 ± 1.15	0,233 ± 0.030	28.023±0.70	8.0±1.05	36.02± 1.75	1.12 ± 0.11	94.23 ± 3.6	407.593 ± 0.713
S9	17.177 ±0.26	3.8 ±0.025	312.67 ± 1.15	0,283 ± 0.015	27.02±2.00	10.02±0.02	37.04 ± 2.02	2.115 ± 0.094	105.63 ± 4.41	203.99 ± 0.165
S10	17.14 ± 0.140	3.5 ±0.055	286 ±2.00	0,263 ± 0.032	33.3±1.00	10.067±1.017	43.367±2.017	7.276 ± 0.216	76.85 ±1.61	201.133 ±1.0
S11	16.243±0.084	4.34 ±0.270	147.97 ±0.55	0,11 ± 0.01	40.0±0.481	10.38±1.00	50.38± 1.481	4.483 ± 0.379	83.81 ± 4.81	206.98 ± 0.076
S12	15.043 ± 1.092	4.2 ±0.396	266.67 ±9.02	0,213 ± 0.020	31.68±1.87	11.033±1	42.713± 2.87	6.217 ± 0.758	86.08± 4.01	506.86 ± 1.091
S13	15.633 ±1.041	4.0 ±0.215	307 ±1.07	0,363 ± 0.028	42.033±0.475	9±1.50	51.033 ± 1.975	3.747 ± 0.357	76.96 ± 4.95	309.5 ±0.608

Table 2. Contd.

S14	16.07 ± 0.113	3.84 ± 0.064	328.83±3.69	0,234 ± 0.173	27.5±0.5	9.5±0.5	37 ± 1.00	14.153 ± 0.055	29.28 ± 1.95	156.167 ± 1.258
S15	18.09 ± 0.131	4.05 ± 0.055	67.83 ±2.57	0,019 ± 0.001	35.3±0.20	8.033±1.32	43.333 ±1.52	8.599 ± 1.678	43.67 ± 1.10	104.56 ± 0.833
S16	15.81 ± 0.685	3.45±0.151	449 ± 1.13	0,27 ± 0.006	28.9±1.0.11	8.063±0.00	36.963±0.11	5.217 ± 0.629	54.45 ± 3.0	303.3 ± 0.56
S17	16.633 ± 0.349	4.02 ± 0.001	599.5 ± 1.0	0,45 ±0.015	44.884±0.193	9.10±0.40	54.317 ±0.59	11.0 ± 0.176	52.82 ± 4.06	405.67 ±0.5
S18	17.18 ± 0.101	4.47 ± 0.134	369.33 ± 0.15	0,238 ± 0.003	27.023±0.485	9.0±1.5	36.02± 1.985	1.12 ± 0.117	95.23 ± 2.6	307.003 ± 0.813
S19	16.17 ± 0.164	3.91 ± 0.125	412.67 ± 1.25	0,383 ± 0.015	28.02±.00	9.02±2.02	37.04 ± 2.02	1.11 ± 0.004	100.3 ± 0.41	103.99 ± 0.275
S20	16.14 ± 0.150	4.50 ± 0.055	487 ± 2.00	0.283 ± 0.012	38.3±2.420	9.067±0.03	43.367±2.25	6.36 ± 0.116	76.00±1.61	301.18±1.003

milliequiv acid/kg (Table 2). The acidity of honey is due to the presence of organic acids, particularly gluconic acid, in equilibrium with their lactones or esters, and inorganic ions such as phosphate and chloride (El-Sherbiny and Rizk, 1979; Al-Khalifa and Al-Arify, 1999).

Electrical conductivity of honeys

The electrical conductivity as measured in most honeys is less than 700 µS/cm (Table 2). The sample S4 has the highest conductivity (1050.5 µS/cm). This parameter depends on the mineral, organic acid, protein, complex sugars and polyol content and varies with botanical origin (Terrab et al., 2003).

Mineral content

The mineral content of the honey ranged from 0.019 to 0.518% (Table 2). These differences in mineral content are dependent on the type of soil in which the original nectar bearing plant was located (Anklam, 1998).

The percentage mineral content is considered as a quality criterion indicating the possible botanical origin of honey. The variability in the mineral content of honeys could be due to harvesting processes, beekeeping techniques and the material collected by the bees while foraging on the flora (Finola et al., 2007).

Proline content

Proline comes mainly from the salivary secretions of *A. mellifera* during the conversion of nectar into honey (Bergner and Hahn, 1972). The concentrations of proline ranged between 103.66 and 506.86 mg/kg (Table 2).

Amylase activity

The amylase activity is usually expressed as diastase number, symbol DN, also known as Gothe units. The amylase activity ranged from 29.28 to 202.123 Gothe units (Table 2) and the remaining DN was still above the minimum limit (8 DN). Consequently, it has been recommended that other quality indicators, such as invertase activity, which is more heat-sensitive than amylase, should be used (Oddo et al., 1999).

HMF content

Since HMF can be formed either by Maillard reaction (heating of reducing sugars in the presence of proteins), or by dehydration under acidic conditions. The HMF values were low (<15 mg/ kg) (Table 2). The diastase activity and the

HMF content are widely recognized as parameters indicating the freshness of honey (Mendes et al., 1998; Terrab et al., 2002).

Conclusion

The result of this study indicated that honey samples purchased from Northeast Algeria, were mostly of good quality. The research describes the variability of some physicochemical characteristics of twenty Algerian honeys. The results obtained agreed with requirements of European Community Directive. Honey samples differ in quality on account of various factors such as season, the origin of honey, the activity of the bee, the food of the bee, the period and technique of extraction of honey, conditions of storage and the freshness of honey. The comparison of these honey varietals with published international standards allowed us to have an idea on the quality of our honeys produced in the Algerian Northeast.

ACKNOWLEDGEMENT

We are grateful to beekeepers that collaborated with us in providing honey samples.

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