International Food Research Journal 24(4): 1453-1459 (August 2017)
Journal homepage: *http://www.ifrj.upm.edu.my*

Physicochemical properties and pollen analyzes of some Algerian honeys

[1*]Diafat, A.E.O., [1]Benouadah, A., [1]Bahloul, A., [1]Meribai, A., [1]Mekhalfi, H., [1]Bouaziz, F., [1]Techache, D., [1]Laabachi, H. and [2]Arrar, L.

[1] laboratory of characterization and valorization of the natural product Faculty of Nature and Life Science, Elbachir Elibrahimi University, Bordj Bou Arreridj, Algeria.
[2] Laboratory of Applied Biochemistry, Faculty of Nature and Life Science, Ferhat Abbas University, Setif 1, Algeria

Article history

Received: 15 June 2016
Received in revised form: 28 June 2016
Accepted: 20 July 2016

Keywords

Honeys
Physicochemical characterization
Botanical origin
Pollen analysis

Abstract

The samples of 25 honeys, collected from Bordj Bou Arreridj beekeepers, were analyzed for parameters such as pH, free acidity, moisture content, diastase activity, hydroxymethylfurfural (HMF) content, electrical conductivity, polyphenol, flavonoid, proline and proteins, in addition, melissopalynological analyses were carried out for characterization of honeys. The mean values of analyzed honeys were: pH 4.59, moisture 16.81%, free acidity 12.88 mmol/kg, electrical conductivity 0.513 mS/cm, diastase activity 129.49 DN and HMF below11.40 mg/kg, Ash 0.12 mg/kg, polyphenol content 75.28 mg Gallic acid equivalent / 100g, flavonoids content 30.55 mg quercetin equivalent /100 g, insoluble matter 0.46%, proteins 3.05 mg/g, proline 4.07 mg / Kg. The melissopalynological analyzes showed that all honeys were polyfloral. The results of physicochemical parameters, all of the analyzed honeys were found to meet European Legislation.amongst four species of seaweed.

Introduction

Honey is a natural sweet substance produced by honey bees from the nectar of plants (blossom honey), secretions of living parts of plants, or excretions of plant-sucking insects (honeydew honey) (Kirs *et al.*, 2011). This natural complex foodstuff is produced in almost every country and largely used as food source. Honey cannot be considered a complete food by human nutritional standards, but it offers potential as a dietary supplement (Mendes *et al*., 1998). Honey contains about 200 substances such as sugars or monosaccharide (of which fructose and glucose are the main components 65%) and 18% of water, approximately. Proteins, flavor and aroma, phenolic compounds and flavonoids, free amino acids, organics acids, vitamins and minerals constitute minor components of honeys (González-Miret *et al*., 2005). The properties and composition of honey are known to vary widely depending on the region, season, and types of bee, plant source of nectar, period for which it is stored in the honeycomb, mode of harvesting and postharvest storage (González-Miret *et al*., 2005).

Honey is considered as an important part of traditional medicine (Silva *et al*., 2009) It has been used in ethno-medicine since the early humans, and in more recent times its role in the treatment of burns, gastrointestinal disorders, asthma, infected wounds and skin ulcers have also been reported(Al-Mamary *et al*., 2002). The Commission of the European Communities in Council Directive DENLEG 2000/10 (Council Directive, 2000) establishes general and specific compositional characteristics of the main varieties of honeys, which can be marketed in the European Union. According to this directive, the principal labeling requirements are the indication of floral or vegetable origin, source, organoleptical characteristics, physicochemical properties and regional or topographical origins (Council Directive, 2000).

Microscopical analysis, especially the identification and quantification of pollen grains in honey sediment, is the reference method used to determine the botanical origin of sample honeys. Normally, honeys are classified as monofloral, when the pollen frequency of one plant is over 45%. Honey quality can also be affected by heating during the extracting, liquefying or clarifying processes or by ageing during storage with production of 5-hydroxymethyl-2-furfuraldehyde (HMF). Some other physicochemical quality parameters were also investigated in order to determine moisture, ash content, diastase activity, free acidity and water-insoluble solids (Mendes *et al*., 1998; Azeredo *et*

*Corresponding author.
Email: *ilyes132@yhoo.fr*

al., 2003). Adulteration of honey is possible, so its quality must be controlled analytically with the aim of guaranteeing the genuity and preserving the consumer from commercial speculation. The present work was conducted to investigate the quality of 25 sample of honey from Bordj Bou Arreridj region (northeast of Algeria) by physicochemical and melissopalynological analysis.

Materials and Methods

Sample collection

Honey samples were obtained from Bordj Bou Arreridj region. They were collected in an area covered the most important production zones. Samples were then stored at 4°C until analysis.

Pollen analysis

The botanical origin of the samples was determined by microscopical analysis according to the method described by Lutier *et al*. (1993).

Physicochemical characteristics

Honey were analyzed according to methods previously reported for pH, moisture, ash content, electrical conductivity, free acidity, diastase activity, hydroxyl methyl furfural, proteins and proline(AOAC, 1990). The pH was measured by an Inolab pH-meter (7300 N° 09060943), in a solution of 10% (W/V) in distilled water. Free acidity was determined by titrimetric method; the addition of 0.05 M NaOH was stopped at pH 8.5, Moisture was determined by refractometry, using WYA Abbe Refractmeter. All measurements were performed at 20°C, Ash content was measured by calcinations, overnight in furnace at 550°C, until constant mass. The Winkler method (Winkler, 1955) was used to determine the HMF content in honey samples, Electrical conductivity of a honey solution at 20% (dry matter basis) in distilled water was measured at 20°C in a WTW, GmbH conductmeter, and the results were expressed as mS cm^{-1}.

Diastase activity was measured using a buffered soluble starch solution and honey, which was incubated in the thermostatic bath at 40°C. Absorption was followed using a (UV mini-1240_ UV.Vis.Spectrophotometers SHIMADZU); the gravimetric method was used for the determination of the insoluble solids according to the Bogdanov (2002) method.

Proline was measured according to the original method of Ough (1969). Proline and ninhydrin form a colored complex. After adding 2-propanol, the extinction of the sample solution and a reference solution at a wave length 510 nm. The proline content is determined from the ratio. Results are expressed in proline milligrams per kilograms honey. The Bradford method (1976) was used for protein determination. To a 0.1 ml solution of protein extract (honey sample 50% w/v) were added 5 ml of Coomassie Brilliant Blue (CBB) G250. The CBB forms a protein-dye complex. After 2 minutes of incubation, absorbance was measured at 595 nm against bovine serum albumin (BSA) standard solution.

Honey color intensity was determined using a Spectrophotometer (UV mini-1240_ UV. Vis. Spectrophotometers SHIMADZU). The honey samples were diluted to 50% (w/v) with warm water (45-50°C), sonicated for 5 minutes, and filtered to eliminate large particles. The net absorbance was defined as the difference between the spectrophotometric absorbance at 450 and 720 nm (Beretta *et al*., 2005).

Total polyphenols were measured using Prussian blue assay methods described by Price and Butler (1977). Total Polyphenol Content was expressed as mg Gallic acid equivalents (GAE)/100 g. Flavonoids were quantified using AlCl3 reagent (Bahorun *et al*., 1996). 1mL of 10 % (W/V) honey sample are dissolved in distilled water, then 1 ml of AlCl3 (2 % in Methanol) was added, after incubation for10 min, the absorbance was measured at 430 nm. Total flavonoids were expressed as mg Quercetin equivalent (QE)/100 g.

Results and Discussion

The results from the pollen analysis, summarized in Table 1 showed the floral origin of honey samples determined by microscopy pollen analyses. Data indicate that all of honey samples were multifloral. *Daucus carota* was a predominant source used by honey bees in the Bordj Bou Arreridj, once its pollen was detected in 68% of the total analyzed samples. Multifloral honeys contained several pollen types with a considerable percentage of pollen grains from: *Daucus carota, Eucalyptus* sp, *Convolvolus arvensis, Rubus* sp., *Lavandula stoechas, Olea europaea* and *Fraximus* sp.

The results of several physicochemical parameters determined are presented in Table 2. Honey pH is affected by the extraction and storage conditions, which also influences texture, stability and shelf-life. Indeed, pH is useful indexes of possible grow well in alkaline media (Terrab *et al*., 2004). The pH values ranged between 4 .02 and 5 .70 (4.59 ± 0.32). These values are in accordance with acceptable range for honey (Bogdanov, 1999) and similar to those

Table1. Classification of honeys samples

Sample identification	Honey type
H1	Multifloral: (Apiaceae: *Daucus carota)*, (Oxalidaceae: *Oxalis* sp), (Asteraceae: *Anthemis cotula)*, (Convolvulaceae: *Convolvolus arvensis*).
H2	Multifloral: (Convolvulaceae: *Convolvulus arvensis*) (Apiaceae: *Daucus carota)*, (Oxalidaceae: *Oxalis* sp), (Oleaceae: *Fraxinus sp).*
H3	Multifloral: (Apiaceae: *Pimpinella anisium*), (Rosaceae: *Rubus fruticosis)*, (Oxalidaceae: *Oxalis* sp), (Apiaceae: *Daucus carota)*,(Asteraceae: *Cirsium arvense) (* Oleaceae: *Olea europaea).*
H4	Multifloral: *(*Asteraceae: *Cirsium arvense), (*Rosaceae: *Rubus* sp), (Convolvulaceae: *Convolvulus arvensis*), (Myralaceae: *Eucalyptus sp), (*Apiaceae*: Ammi majus),* (Apiaceae: *Daucus carota),* (Oleaceae: *fraxinus sp).*
H5	Multifloral: (Oleaceae: *fraxinus sp), (Brassicaceae: Sinapis arvensis), (*Rhamnaceae*: Zizyphis lotus),* (Apiaceae: *Daucus carota), (*Fagaceae: *Quercus ilex),* (Rosaceae: *Rubus fruticosis).*
H6	Multifloral: *(*Asteraceae: *Cirsium arvense), (poaceae: poaceae type),* (Fagaceae: *Quercus ilex),* (Oleaceae: *fraximus sp), (*Asteraceae: *Chicorium intybu),* (Asteraceae: *Cirsium arvense), (Oleaceae: Olea europaea).*
H7	Multifloral: (Lamiaceae: *Lavendula angustifolia*), (Oleaceae: *Olea europaea),* (Asteraceae: *Achillea sp),* (Pinaceae: *Pinus sp)* (Oleaceae: *fraximus sp),* (Apiaceae: *Pimpinella anisium*), (Asteraceae: *Cirsium arvense) (*Oleaceae: *Olea europaea).*
H8	Multifloral: *(*Oleaceae: *Fraxinus sp), (*Rosaceae: *Rubus* sp), (Myralaceae: *Eucalyptus),* (Apiaceae: *Daucus carota),* (Oleaceae: *Fraxinus sp),* (Lamiaceae: *Lavendula stoechas*), (Myralaceae: *Eucalyptus sp).*
H9	Multifloral: (Rosaceae: *Rubus* sp), (Oleaceae: *fraximus sp), (*Oleaceae: *Olea europaea),* (Convolvulaceae: *convolvolus arvensis*), (Apiaceae: *Pimpinella anisium*), (Myralaceae: *Eucalyptus sp).*
H10	Multifloral: (Rosaceae: *Rubus fruticosis), (*Asteraceae: *Cirsium arvense),* (Lamiaceae: *Lavendula stoechas*), (Apiaceae: *Daucus carota),* (Convolvulaceae: *Convolvulus arvensis*), (Myralaceae: *Eucalyptus sp).*
H11	Multifloral: (Apiaceae: *Daucus carota),* (Oleaceae: *fraxinus sp), (Papaveraceae: papaver rhoeas), (*Oleaceae: *Olea europaea).*
H12	Multifloral: (Apiaceae: *Daucus carota),* (Oleaceae: *fraxinus sp),* (Myralaceae: *Eucalyptus sp), (*Pinaceae*: pinus halpensis), (Papaveraceae: Papaver rhoeas, (*Oleaceae: *Olea europaea).*
H13	Multifloral: (Oleaceae: *Fraximus sp), (*Apiaceae: *Daucus carota),* (Oxalidaceae: *Oxalis* sp), *(Papaveraceae: Papaver rhoeas), (*Rosaceae: *Rubus* sp), (Apiaceae: *Pimpinella anisium*).
H14	Multifloral: *(*Apiaceae: *Daucus carota),* (Oleaceae: *fraximus sp), (Papaveraceae: Papaver rhoeas),* (Convolvulaceae: *Convolvulus arvensis*).
H15	Multifloral: *(*Apiaceae: *Daucus carota),* (Asteraceae: *Chicorium intybus),* (Convolvulaceae: *Convolvulus arvensis*), (Rosaceae: *Rubus fruticosis),* (Oleaceae: *fraximus sp), (*Asteraceae: *Cirsium arvense).*
H16	Multifloral: (Pinaceae*: Pinus halpensis),* (Aquifoliaceae: *Ilex divaricata*), (Cupressaceae: *Cupressus sempervirens), (*Pinaceae*: pinus halpensis),* (Lamiaceae: *Lavendula stoechas*).
H17	Multifloral: (Oleaceae: *fraximus sp), (Lamiaceae: Rosmarinus officinalis)* (Asteraceae: *Chicorium intybus),* (Apiaceae: *Pimpinella anisium*), (Myralaceae: *Eucalyptus sp),* (Convolvulaceae: *convolvolus arvensis*).
H18	Multifloral: (Oleaceae: *fraximus sp), (*Apiaceae: *Daucus carota),* (Oleaceae: *Olea europaea),* (Apiaceae: *Pimpinella anisium*), (Convolvulaceae: *convolvolus arvensis*).
H19	Multifloral: (Oleaceae: *fraximus sp), (*Rhamnaceae*: Zizyphis lotus),* (Asteraceae: *Artemisia campestris*), *(*Apiaceae*: Ammi majus), (*Fagaceae: *Quercus ilex),* (Lamiaceae: *Lavendula stoechas*), (Oleaceae: *fraxinus sp) (*Fabiaceae: *ceratonia sp).*
H20	Multifloral: (Asteraceae: *Chicorium intybus), (*Oleaceae: *Olea europaea),* (Myralaceae: *Eucalyptus sp),(* Poaceae: *poaceae type), (*Apiaceae: *Ammi majus), (*Apiaceae: *Daucus carota).*
H21	Multifloral: *(*Asteraceae: *Cirsium arvense), (*Apiaceae: *Daucus carota),* (Rosaceae: *Rubus fruticosis),* (Oxalidaceae: *Oxalis* sp), *(*Rhamnaceae*: Zizyphis lotus), (*Pinaceae*: pinus halpensis).*
H22	Multifloral: (Oxalidaceae: *Oxalis* sp), (Poaceae: *Poaceae type), (*Apiaceae: *Daucus carota),* (Oleaceae: *fraximus sp), (*Papaveraceae: *Papaver rhoeas),* (Convolvulaceae: *Convolvulus arvensis*).
H23	Multifloral: (Oleaceae: *Fraxinus sp), (*Papaveraceae*: Papaver rhoeas), (*Rosaceae: *Rubus* sp), (Aquifoliaceae: *Ilex divaricata), (*Asteraceae: *Cirsium arvense).*
H24	Multifloral: (Oleaceae: *Fraxinus sp),* (Papaveraceae: *Papaver rhoeas), (*Rosaceae: *Rubus* sp), *(*Apiaceae *: Ammi majus),* (Lamiaceae: *Lavendula stoechas*), (Euphoribiaceae: *Ricinus communis*), (Oleaceae: *Olea europaea).*
H25	Multifloral: *(*Apiaceae : *Daucus carota),* (Oleaceae : *fraxinus sp),* (Oleaceae: *Olea europaea), (*Papaveraceae: *Papaver rhoeas), (*Rosaceae : *Rubus* sp), Fabiaceae: *Ceratonia siliqua*

Table 2. Distribution data for physicochemical parameters in Bordj Bou Arreridj (Algeria) honey samples

Sample	pH	Free Acidity (meq/kg)	Electrical conductivity (µScm-1)	Moisture (%)	Color (mm Pfund)	insoluble matter (mg/100g)	Ash (%)	Diastase activity	HMF (mg/kg)	polyphénol (mg AGE/ 100g)	Flavonoids (mg EQ/ 100 g)	protein (mg / g)	proline (mg / Kg)
H1	5.03	14	255.55	15.0	154.42	23.43	0.0002	130.43	5.84	94.02	34.60	16.48	224.44
H_2	4.40	08	556.41	14.2	235.01	57.13	0.00293	119.52	8.83	108.57	23.17	31.87	434
H_3	5.06	06	351.61	13.8	147.74	67.7	0.01506	130.44	2.24	45.50	10.49	31.31	426
H_4	4.71	04	275.48	16.4	51.17	13.15	0.83093	92.59	12.35	29.07	14.16	18.000	243
H_5	5.15	20	260.98	15.4	334.91	48.91	0.46146	150	8.91	36.78	93.87	16.85	223
H_6	4.02	14	407.79	25.5	74.20	18.02	0.0007	115.38	6.74	124.30	38.56	25.39	343
H_7	5.41	10	687.80	18.4	158.87	17.09	0.00169	84.74	1.95	30.52	18.23	4.90	459
H_8	4.81	18	524.69	17.2	108.74	22.2	0.00079	254.24	7.56	37.80	26.69	10.58	644
H_9	5.70	12	799.27	19.2	359.80	1.13	0.27313	94.34	5.01	83.18	19.25	5.05	714
H_{10}	4.65	20	110.56	13.0	37.43	8.32	0.00911	83.33	10.25	86.07	25.92	7.33	828
H_{11}	4.69	08	344.36	17.6	459.70	32.85	0.0056	202.70	17.14	15.25	26.80	6.06	460
H_{12}	5.30	12	563.66	17.2	249.127	21.7	0.00058	69.77	7.18	60.49	29.38	6.22	444
H_{13}	5.15	20	581.78	15.8	133.99	20.81	0.00249	87.72	36.23	44.35	38.23	9.49	690
H_{14}	4.45	02	311.73	15.4	187.84	8.07	0.2958	122.45	4.72	134.39	25.79	12.55	413
H_{15}	4.25	04	594.47	15.4	131.76	59.07	0.00029	157.89	5.91	34.81	16.79	3.22	258
H_{16}	4.71	08	577.25	18.2	135.85	22.12	0.00326	86.21	20.21	117.79	31.77	6.90	47
H_{17}	4.26	06	327.14	16.8	386.54	43.57	0.05486	198.67	5.16	131.02	11.34	5.52	497
H_{18}	5.39	12	220.21	16.8	122.11	21.3	0.00157	120.97	6.59	94.40	32.25	10.95	775
H_{19}	4.53	14	676.93	17.4	357.57	52.99	0.54366	57.25	27.89	88.16	41.41	16.43	447
H_{20}	4.97	14	687.80	19.2	227.95	50.39	0.0060	72.81	7.86	42.76	22.15	3.66	257
H_{21}	4.90	16	808.33	15.4	227.58	55.6	0.6028	150.00	5.46	20.26	35.01	12.86	1515
H_{22}	4.63	20	540.09	15.8	314.86	20.62	0.00127	254.24	4.16	136.22	30.21	17.74	262
H_{23}	4.20	22	328.04	16.4	301.12	60.15	0.00933	130.43	12.95	84.53	40.36	7.36	1371
H_{24}	4.80	20	530.13	16.6	214.58	19.77	0.00231	211.27	45.25	102.76	39.10	7.63	270
H_{25}	4.50	18	1151.19	16.0	373.542	18.48	0.01327	60.00	8.68	98.98	38.46	7.16	557
Mean	4.59	12.88	512.8593	16.81	219.45	219.45	0.1255	129.50	11.40	75.29	30.56	30.56	528
SEM	0.32	5.0	183.58	1.56	95.54	17.16	01.8	43.67	7.38	34.44	10.20	6.31	229.56

obtained with others Algerian honeys (Ouchemoukh *et al*., 2007).

Percent moisture in the analyzed honeys ranged from 13 to 25.5% (16.81 ± 1.56 %). The water content of honey depends on various factors, like the harvesting season, the degree of maturity reached in the hive and climatic factors. The maximum amount of water contained by honey is regulated for safety against fermentation. All the samples contained less than 20% water, the maximum amount allowed by international and European legislations (The Council of the European Union, 2002).

The ash content is a quality criterion for honey botanical origin; the blossom honeys have lower ash content than honeydew honeys (White *et al*., 1978). The results found (0.0002 to 0.83%), (0.13 ± 0.18 %). These differences in mineral content are dependent on the type of soil in which the original nectar bearing plant was located (Anklam, 1998).

The electrical conductivity of honey is closely related to the concentration of mineral salts, organic acids and proteins. This parameter shows

great variability according to the floral origin and it is important for the differentiation of honeys of different floral origins (Terrab *et al*., 2002). The results obtained for the honey samples under study varied between 110.56 and 1151.19 µS cm^{-1} (512.86 ± 183.58µS cm^{-1}). These values are below the maximum limit indicated by European legislation for nectar honey (800 µS cm^{-1}).The increase in ash content of the honey samples from Bordj Bou Arreridj region was accompanied by the increase of electrical conductivity, as previously reported by Downey *et al*. (2005).

Honey acidity is due to the presence of organic acids, mainly gluconic acid, and to inorganic ions, such as phosphate, sulfate and chloride (Nanda *et al*., 2003; Terrab *et al*., 2004). The values of the acidity of our honey samples vary from 2.00 to 22.00 meq/kg (12.88 ± 5 meq /kg). All honeys are acidic, indicating the absence of undesirable fermentation. Free acidity was within the limits of European legislations (below 50 meq/kg), The results obtained for acidity were in agreement with data reported for other Algerian honeys (Ouchemoukh *et al*., 2007), as well as for samples from other geographical locations (Terrab *et al*., 2002; Terrab *et al*. 2004). The variation of acidity has been attributed to harvest season (DeRodriguez *et al*., 2004).

Hydroxymethylfurfural (HMF) content is widely recognized as parameter of freshness for honey samples. Several factors influence the formation of HMF, such as storage conditions (e.g. temperature) and floral sources (Terrab *et al*., 2002; Fallicogr *et al*., 2004). It is well known that honey heating results in the formation of HMF, which is produced during acid-catalyzed dehydratation of héxoses, such as fructose and glucose (Belitz and Grosch., 1999). The amounts found fell within the European legislation, corresponding to a high degree of freshness (The Council of the European Union, 2002), all samples presented HMF levels below 40 mg/kg of honey, ranging from 2.24 to 27.88 mg/kg (13.06 ± 7.38 mg/kg), and only one samples presented an inappropriate HMF with a value superior to 40 (45.25 mg/kg) but inferior with the values given by the codex 50 mg/kg (Table 2),

Diastase activity is a parameter used to determine if honey has been extensively heated during processing, because the enzyme is susceptible to heating and storage factors. The honey samples exhibited very different values, ranging between 60 and 254.23 Schade units (129.49± 43.67Schade units), (Table 2), generally the diastasic index should not be lower than 8 Schade units (EUD, 2002).

The determination of insoluble matter is very useful for detecting impurities in honey. The insoluble content in water is in the order of 100 mg /100 g (Codex Alimentarius Commission, 1999) In the present study insoluble matter values range between 1.13 mg and 67.7mg/100g (31.38 ± 17.16 mg / 100 g). All samples are consistent with the limit. Honey color intensity depends on the floral origin. It is closely related to its chemical composition, mainly in the presence of colorants such as chlorophylls, carotenoids, flavonoids and derivatives of the tannins and polyphenols. In the present study our samples have different color, the color values are between 37.43 and 459.70 mm Pfund (219.45 ± 95.54 mm Pfund). Monica *et al*, (2007), reported that the nectar of honey color is dark and strong, resulting in the highest mineral content but also the presence of algae and green algae that part of the flora of the forest trees. The color of honeydew honey depends on the raw material composition of the food given to the bees.

The determination of phenolic content of honey is a good parameter for the assessment of its quality and its possible therapeutic potential (Al-Mamary *et al*, 2002), Amiot *et al* (1989) result that honey contains a number of phenolic compounds, in the nature and quantity vary widely depending on the floral origin. Many researchers have found that honey has a dark color with a higher amount of total phenolics. The values of the total polyphenol content of our samples are between 15.25 and 136.22 mg / 100g (75.28 ± 34.44mg / 100 g). Meda *et al* (2005) found that the honeydew honey has a higher content of total polyphenols (114.75 mg / 100 g). Gheldof *et al*, (2002); Meda *et al* (2005) demonstrated a correlation between the antioxidant activity and polyphenol content.

The values of the flavonoid content of our samples are between 10.48 and 93.86 mg EQ /100g (30.55 ± 10.20 mg EQ / 100 g). Analyses of Flavonoids are a very promising technique for studying the floral origin of honey, and evaluation of their quality (Soler *et al*, 1995). Therefore, the flavonoids in honey can make a good source of antioxidants near their effect as an antibacterial, thus increasing its potential therapeutic activity. Frankel *et al* (1998) shows that the correlation between TPC (total polyphenol content) and TFC (total flavonoid content) was found and between TFC and color, and it is suggested that the intensity of the color of honey is related to pigments (carotenoids, flavonoids, etc.), in fact, the increase in color intensity is related to an increase in the concentration of these compounds. Our samples contain relatively low flavonoids content compared to data provided by Ouchemoukh *et al*., (2007) who

found higher concentrations of 64-1304 mg EQ / 100g.

The values of the protein content of our honey samples ranged between 1.37 and 18.56 mg /g (8.42 ± 6.31 mg / g). The results of our study are higher than those obtained by Ouchemoukh *et al*, (2007), indicates that the protein content of the samples analyzed were in honey 3.7 and 9.4 mg/g. Anklam *et al*, (1998) found in their study that the honey protein content is less than 5 mg /g. Proline is the most abundant amino acid in honey is used as a standard for measuring amino acid content. Proline content is an indication of the quality, and maturity of honey is also an indicator of adulteration when it falls below a value of 183 mg /kg (Bogdanov *et al*, 1999). All honey samples have a proline content of over 183 mg / kg, shows the absence of adulteration (Meda *et al*, 2005). The value of the proline contents varies between 223 and 1515 mg / kg, (528 ± 229.56 mg / kg). Some authors report that the high values of proline are typical for honeydew honeys. Ouchemoukh *et al*, (2007), found that the concentrations of proline range of 202 and 680 mg / kg. The results we found indicate that the honeys are good qualities to the absence of any spell of adulteration.

Conclusions

Honeys from Bordj Bou Arreridj region present a good level of quality, once 24 of the 25 studied samples are in agreement with the European honey directive (EUD., 2002), indicating adequate processing, good maturity and freshness.

Acknowledgments

This work was supported by the Algerian Ministry of Higher Education and Scientific Research (MESRS, DGRSDT), Algeria.

References

Al-Mamary, M., Al-Meeri, M. and Al-Habori, M. 2002. Antioxidant activities and total phenolics of different types of honey. Nutrition Research 22: 1041–1047.

Amiot, M.J., Aubert, S., Gonnet, M. and Tacchini, M. 1989. Les composés phénoliques des miels : étude préliminaire sur l'identification et la quantification par familles. Apidologie 20(2): 115-125.

Andrade, P.B., Amaral, M.T., Isabel, P., Carvalho, J., Seabra, R. and Cunha, A. 1999. Physicochemical attributes and pollen spectrum of Portuguese heather honeys. Food Chemistry 66: 503-510.

Anklam, E. 1998. A review of the analytical methods to determine the geographical and botanical origin of honey. Food Chemistry 63: 549–562.

Association of Official Analytical Chemists (AOAC). 1990. Official Methods of Analysis, (15th ed.) Association of Official Analytical. Chemists, Inc. Arlington.

Azeredo, L., Azeredo, M.A.A., de Souza, S.R. and Dutra, V.M.L. 2003. Protein contents and physicochemical properties in honey samples of Apis mellifera of different floral origins. Food Chemistry 80: 249–254.

Bahorun, T., Gressier, B., Trotin, F., Brunet, C., Din, T., Vasseur, J., Gazin, J. C., Pinkas, M.,Luyckx, M. and Gazin, M. 1996. Oxygen species scavenging activity of phenolic extract fromhowthorn fresh plant organs and pharmaceutical preparation. Arzneimittelforschung. Drug Research 46(2): 1086 - 1089.

Belitz, H.D. and Grosch, W. 1999. Food Chemistry Springer, New York.

Beretta, G., Granata, P., Ferrero, M., Orioli, M. and Maffei, F.R. 2005. Standardization of antioxidant properties of honey by a combination of spectrophotometric/ fluorimetric assays and chemometrics. Analytica Chimica Acta 533: 185-191.

Bogdanov, S. 1999. Honey quality and International regulatory standards: review by the International Honey Commission. Bee World 90: 61–69.

Bogdanov, S. 2002. Harmonised methods of the international honey commission. 1-61.

Bradford, M.M. 1976. Rapid and sensitive method for quantification of microgram quantities of protein utilizing the principle dye binding. Analytical Biochemistry 72: 248-254.

DeRodriguez, G.O., Sulbarán de Ferrer, B., Ferrer, A. and Rodriguez, B. 2004. Characterisation of honey produced in Venezuela. Food Chemistry 84: 499–502.

Downey, G., Hussey, K., Kelly, J., Walshe, T. and Martin, P. 2005. Preliminary contribution to the characterisation of artisanal honey produced on the island of Ireland by palynological and physico-chemical data. Food Chemistry 91: 347–354.

European Union Directive, Council Directive 2001/110/ EC relating to honey. 2002.

Fallico, B., Zappala, M., Arena, E. and Verzera, A. 2004. Effects of conditioning on HMF content in Monofloral honeys. Food Chemistry 85: 305–313.

Frankel, S., Robinson, G.E. and Berenbaum, M.R. 1998. Antioxidant capacity and characteristics of 14 unifloral honeys. Journal of Apiculture Research 37: 27–31.

Gheldof, N. and Engeseth, N.J. 2002. Antioxidant capacity of honeys from various floral sources based on the determination of oxygen radical absorbance capacity and inhibition of vitro lipoprotein oxidation in human serum samples. Journal of Agricultural and Food Chemistry 50: 3050-3055.

González-Miret, M.L., Terrab, A., Hernanz, D., Fernández-Recamales, M.A. and Heredia, F.J. 2005 . Multivariate correlation between color and mineral composition of honeys and by their botanical origin. Journal of Agricultural and Food Chemistry 53: 2574–2580.

Kirs, E., Pall, R., Martverk, K. and Laos, K. 2011. Physicochemical and melissopalynological

characterization of Estonian summer honeys. Procedia Food Science 1: 616 – 624.

Silva, L.R., Andreia, P.R.V., Valentão, M.P. and Andrade, P.B. 2009. Honey from Luso region (Portugal): Physicochemical characteristics and mineral contents. Microchemical Journal 93: 73–77

Lutier, P. and Vassière, B. 1993.An improved method for pollen analysis of honey. Review of Palaeobotany and Palynology 78: 129–144.

Martins, R.C., Lopes, V.V., Valentão, P., Carvalho, J.C.M.F., Isabel, P., Amaral, M.T., Batista, M.T. and Andrade, P.B. 2008. Relevant principal component analysis applied to the characterisation of Portuguese heather honey. Natural Product Research 17 : 1560–1582.

Meda, A., Lamien, C. E. and Marco, R. 2005. Determination of the total phenolic, flavonoïde and proline contents in Burkina Fasan honey, as well as their radical scavenging activity. Food Chemistry 91(3): 571-577.

Mendes, E., Brojo-Proença, E., Ferreira, I.M. and Ferreira, M.A. 1998. Quality evaluation of Portuguese honey. Carbohydrate Polymer 37: 219–223.

Monica, S., Finola, Mirta, C., Lasagno, Juan, M. and Marioli. 2007. Microbiological and chemical characterization of honeys from central Argentina. Food Chemistry 100(4): 1649–1653

Nanda, V., Sarkar, B. C., Sharma, H. K. and Bawa, A. S. 2003. Physicochemical properties and estimation of mineral content in honey produced from different plants in Northern India. Journal of Food Composition and Analysis 16: 613–619

Ough, C. 1969. Rapid determination of proline in grapes and wines. Journal of Food Sciences 34: 228–230.

Price, M. P. and Butler, L. G. 1977. Rapid visual estimation and spectrophotometric determination of tannin content of sorghum grain. Journal of Agricultural and Food Chemistry 25:1268–1273.

Soler, C., Gil, M.I., Garcia-Viguera, C. and Tomas-Barberan, F.A. 1995. Flavonoid patterns of French honeys with different floral origin. Apidologie 26: 26–53.

Terrab, A., Díez, M.J. and Heredia, F.J. 2002. Characterization of Moroccan Monofloral honeys by their physicochemical characteristics. Food Chemistry 79: 373–379.

Terrab, A., Díez, M.J. and Heredia, F.J. 2003. Palynological, physico-chemical and colourcharacterisation of Moroccan honeys: II. Orange (*Citrus* sp.) honey. International Journal of Food Science and Technology 38: 387–394.

Terrab, A., González, A.G., Díez, M.J. and Heredia, F.J. 2003. Characterization of Moroccan uniforal honeys using multivariate analysis. European Food Research and Technology 218: 88–95.

Terrab, A., Recalames, A.F., Hernanz, D. and Heredia, F.J. 2004. Characterisation of Spanish thyme honeys by their physicochemical characteristics and mineral contents. Food Chemistry 88 537–542.

White, J.W. 1978 Honey. Advances in Food Research 24: 287–375.

Winkler, O. 1955. Detection and determination of hydroxy-methylfurfural in honey. Lebensm Untersuch Forsch 102: 161–167.