Original Article

QUALITY ATTRIBUTES OF LOCAL AND IMPORTED HONEYS COMMERCIALIZED IN ALGERIA

Kheira Dahmani[1]
Jinane B. Houdeib[2]
Amina Zouambi[1]
Badis Bendeddouche[1]
Miguel Fernández-Muiño[3]
Sandra M. Osés[3]
M.Teresa Sancho[3]*

[1]National Veterinary School ENSV, Algiers, Algeria
[2]Technical Institute of Farming, Central Laboratory, Algiers, Algeria
[3]Universidad de Burgos, University of Burgos, Spain

*corresponding author: mtsancho@ubu.es

Abstract

This study was aimed to assess quality, authentication parameters and trolox equivalent antioxidant capacity (TEAC) of Algerian and imported honeys sold in Algerian markets. Results indicated that 80% Algerian samples fulfilled international standards, whereas only 21.4% imported honeys were in agreement with the current regulations. 13.3% Algerian samples and 7.1% imported honeys showed values of proline lower than 180 mg/kg, which is the recommended limit for authentic honeys. Comparing Algerian and imported honeys, electrical conductivity, degrees Brix, diastase activities and proline contents were higher in Algerian honeys, in contrast to moisture percentages, hydroxymethylfurfural contents and acid phospatase activities that were higher in imported honeys. Methanolic extracts of Algerian samples were richer both in total phenolics and flavonoids determined in alkaline medium. There were not significant differences between Algerian and imported samples concerning pH, free acid, invertase, total carotenoid, total phenolics of raw honeys and TEAC, as well as regarding total flavonoids determined in neutral medium and o-diphenols of honeys' methanolic extracts. Principal components analysis showed a good separation between Algerian and imported samples, only one multifloral Algerian honey being misclassified. Our research showed that a legal frame for Algerian honeys is of utmost importance. Spurge-labeled honeys were grouped, showing interesting common features that should be taken into account in a future regulation, in which a protected designation of origin for spurge honeys could be considered.

Keywords: Algerian honeys, phenolics, quality parameters, TEAC, total carotenoids

INTRODUCTION

Honey is a natural complex sweet food produced by honeybees from nectar or honeydew, whose composition mainly depends on botanical and geographic origins and climatic conditions (Bogdanov, 2017; Machado De-Melo et al., 2018). Honey quality assessment is usually based on legislated specifications. Besides the parameters included in international regulations (Thrasyvoulou et al., 2018), the enzymes invertase and acid phosphatase, as well as the amino acid proline, are interesting authentication parameters (Alonso-Torre et al., 2006; Bogdanov, 2017). Some of them, together with phenolic compounds, carotenoids and other constituents contribute to honey antioxidant capacity (Álvarez-Suárez et al., 2010; Ciappini & Stoppani, 2014; Rodríguez-Flores, Escuredo, & Seijo, 2016; Piszcz & Głód, 2019).

In Algeria, honey is consumed on a large scale, so that apart from Algerian honeys, other

imported honeys from different countries are widely commercialized. Nevertheless, Algeria does not have a legal regulation for honey, so that the consumers are more exposed to honey adulterations and frauds. Published research on Algerian honeys shows a great variability of results regarding compositional parameters (Diafat et al., 2017). Some papers stress the compliance/lack of compliance of some Algerian honeys with international standards (Ouchemoukh, Louaileche, & Schweitzer, 2007; Otmani et al., 2019), whereas other studies highlight the importance of Algerian honeys characterization and/or the possibility of obtaining specific quality labels (Makhloufi et al., 2010; Haderbache, Mouna, & Arezki, 2013; Mekious et al., 2015; Ouchemoukh et al., 2017; Zerrouk et al., 2018). Antioxidant activity and parameters related to this capacity have been less researched in Algerian honeys (Khalil et al., 2012; Mouhoubi-Tafinine, Ouchemoukh, & Tamendjari, 2016; Habati et al., 2017; Ouchemoukh et al., 2017; Otmani et al., 2019). Strategic improvements in beekeeping are well-known to have a positive impact on economic development (Ramadani et al., 2019; Sari et al., 2020), which is of paramount importance in North African countries. Enhancing Algerian honeys' quality through the application of good beekeeping practices and regulatory tools will contribute to increased consumers' safety and to boosted Algerian apiculture sector. Therefore, the purpose of this work was to research quality, authentication, antioxidant-related parameters and trolox equivalent antioxidant capacity (TEAC) in Algerian and imported honeys in order to help establish legislation criteria for honeys commercialized in Algeria.

MATERIAL AND METHODS

Samples

This study was carried out on fifteen representative Algerian honeys ("A") and on fourteen representative imported honeys ("I"). All samples were kept in 250 mL glass flasks and stored in the dark at fresh room temperature until analysis (during 2017-2018). All assays were performed in duplicate.

"A" samples were acquired from beekeepers in different regions of Algeria during 2016-2017. Their best-before dates ranged from 2018 and 2021. Their floral origins were declared by the beekeepers on the basis of beehive locations and nectar sources; 5 spurge- (*Euphorbia* sp.) labeled honeys (A-S), 5 jujube- (*Ziziphus* sp.) labeled honeys (A-J), and 5 multifloral- labeled honeys (A-M). Samples A-S1, A-S2 and A-S5 came from El bayadh, A-S3 and A-S4 came from Ain sefra, A-J1 and A-J2 came from Ain oussera, A-J3 came from Laghouat, A-J4 and A-J5 came from Djelfa, A-M1 came from Chrea, A-M2 came from Chiffa, A-M3 and A-M4 came from Bouira and A-M5 came from Lakhdaria.

"I" samples were collected in the Algerian retail market. They came from honey export companies of different European, American, Asian and African countries. The actual geographical origins of "I" honeys were uncertain, because the countries in which the registered offices of the export companies were placed, did not necessarily coincide with the countries in which the honeys had been harvested. Furthermore, the honeys could be blends of honeys from several countries, because it is not compulsory in Algeria to indicate the real geographical origin of honeys on the label. According to the labels of "I" honeys, their harvesting year was 2016, their best-before dates ranged from 2019 and 2021, and their botanical origins were one buckwheat (*Fagopyrum esculentum* Moench), one clover (*Trifolium* spp.), one citrus (*Citrus* spp.), one eucalyptus (*Eucalyptus* spp.), one wild lavender (*Lavandula* spp.), one heather (*Erica* spp., *Calluna vulgaris* (L.) Hull), two wildflowers (indicating the presence of goldenrod (*Solidago* spp.) and *Aster* spp.) as well as six multifloral honeys. In Algeria there is neither legislation for honeys and nor control about the truthfulness of the labels, so the origins, best-before dates and other labeled data of honeys may not be true. This study was aimed to compare Algerian honeys and imported honeys sold in Algeria, regardless of their botanical origin, in order to assess the quality of honeys sold in Algeria.

Procedures

Electrical conductivity (mS/cm), moisture (%), degrees Brix (ºBx), pH, free acid (meq/kg), hydroxymethylfurfural (HMF) content (mg/kg), diastase activity (Schade scale), and proline (mg/kg) were determined using the methods approved by the International Honey Commission (2009) and AOAC (2016). Invertase activity (U/kg) was analyzed quantifying 4-nitrophenol formed by the reaction of honey invertase with 4-nitrophenyl-α-D- glucopyranoside (Siegenthaler, 1977; Huidobro et al., 1995). Acid phosphatase activity (mg P/100 g honey/24 h) was assessed measuring the quantity of 4-nitrophenol released from 4-nitrophenylphosphate by honey acid phosphatase (Günther & Burckhart, 1967; Alonso-Torre et al., 2006). Total carotenoid content (TCC) (mg β-carotene/kg honey) was determined measuring the absorbance at 450 nm of the filtrated supernatant after honey extraction with acetone-hexane (4:6) (Álvarez-Suárez et al., 2010). Phenolic compounds were extracted using an Amberlite XAD-2 resin column (Baltrušaitytė, Venskutonis, & Čeksteryte, 2007), being eluted with methanol. Total phenolic content (TPC) (mg gallic acid/100 g honey) was determined in both raw honeys and extracts following the Folin-Ciocalteu method (Singleton & Rossi, 1965; Sancho et al., 2016).

Total flavonoids' contents (TFC) were assessed on honey methanolic extracts. Quercetin-type TFC (mg quercetin/100 g honey) was determined by the reaction of flavonols and the flavone luteolin (Pękal & Pyrzynska, 2014) with $AlCl_3$ in neutral medium. Catechin-type TFC (mg catechin/100 g honey) was assessed by the reaction of flavan-3-ols (catechin, epicatechin), phenolic acids (such as chlorogenic acids), rutin and luteolin (Pękal & Pyrzynska, 2014) with $AlCl_3$ in an alkaline medium. o-Diphenol content (µg cathechol/100 g honey) was analysed for first time on honey. The procedure was done on methanolic extracts in triplicate. 1 mL honey extract was mixed with 1 mL 0.5N HCl, 1 mL Arnow's reagent (10.00 g $NaNO_2$ and 10.00 g Na_2MoO_4 in 100 mL H_2O), 10 mL H_2O and 2 mL 1N NaOH. After 30 s, the absorbance was read

at 515 nm. A calibration curve with cathechol was drawn (Afoakwa et al., 2012). TEAC (µmol trolox/100 g honey) was determined on honeys with the procedure based on the decolorization of the $ABTS^{•+}$ radical, which is a function of antioxidants' concentration (Re et al., 1999; Sancho et al., 2016).

Statistical analyses were carried out with Statgraphics Centurion XVI.II. As the purpose of our research was to compare "A" and "I" samples regardless of their origins, one-way ANOVA was used to analyze the values of "A" and "I" honeys for the parameters in which assumptions of normality, independence of cases and homoscedasticity were fulfilled. For the rest of parameters, the Kruskal-Wallis test was employed. Statistically significant differences were considered at $p < 0.05$. No statistical analysis was applied to compare Algerian samples (A-S, A-J and A-M), because on the one hand, their botanical origins had not been confirmed, and on the other hand the number of honeys for each group (five) was not representative enough. To check possible groupings of the samples, data were also analyzed through the Principal Component Analysis (PCA).

RESULTS

Tab. 1 shows the averages, standard deviation and minimum and maximum values for the studied "A" and "I" honeys. One-way ANOVA was applied to the results of moisture percentages and proline contents. Kruskal-Wallis was applied to the results of electrical conductivity, degrees Brix, pH, free acid, HMF content, diastase, invertase and acid phosphatase activities, TCC, TPC, TFC, o-diphenols content and TEAC. Tab. 2 shows the percentages of samples that did not fulfill international regulations.

According to the current quality standards (Codex 2001, OJEC 2002), electrical conductivity values fulfilled the legal limit for blossom honeys (not more than 0.8 mS/cm), with the average of "I" honeys about the half of "A" samples. All honeys were in an acceptable moisture range below the limit of 20%, with the moisture percentage significantly higher in the

Table 1.

Average, standard deviation, minimum and maximum values of physicochemical parameters of the analysed Algerian (n=15) and imported (n=14) honeys. A-S: Algerian spurge-labeled honeys. A-J: Algerian jujube-labeled honeys. A-M: Algerian multifloral-labeled honeys.

	A-S (n= 5)	A-J (n= 5)	A-M (n= 5)	Algerian honeys (n= 15)	Imported honeys (n= 14)
Properties/Parameters	Mean±SD (range)	Mean±SD (range)	Mean±SD (range)	Mean±SD (range)	Mean±SD (range)
Electrical conductivity (mS/cm)	0.26±0.09 (0.15-0.39)	0.35±0.16 (0.19-0.56)	0.28±0.18 (0.09-0.56)	0.30±0.14[b] (0.09-0.56)	0.17±0.12[a] (0.04-0.48)
Moisture (%)	15.20±0.97 (14.00-16.50)	16.70±1.30 (15.00-18.00)	16.70±1.82 (15.00-19.50)	16.20±1.50[a] (14.00-19.50)	17.71±1.31[b] (14.50-19.50)
Degrees BRIX (°Bx)	83.30±0.84 (82.00-84.00)	81.80±1.25 (80.50-83.00)	82.20±1.89 (79.00-83.50)	82.43±1.45[b] (79.00-84.00)	81.18±1.05[a] (80.00-84.00)
pH	3.93±0.20 (3.70-4.21)	3.88±0.92 (2.53-4.99)	3.84±0.30 (3.62-4.34)	3.88±0.53[a] (2.53-4.99)	3.60±0.38[a] (3.18-4.24)
Free acid (meq/kg)	22.20±5.72 (15.00-30.00)	31.20±18.21 (6.00-50.00)	39.40±22.94 (27.00-80.00)	30.93±17.53[a] (6.00-80.00)	22.21±7.63[a] (13.00-43.00)
HMF (mg/kg)	9.05±1.75 (6.80-11.52)	15.38±20.06 (0.00-46.70)	46.14±73.70 (3.59-177.0)	23.52±44.15[a] (0.00-177.0)	103.60±147.08[b] (19.31-580.83)
Diastase number (Schade units)	15.18±3.90 (10.60-20.50)	17.16±13.58 (5.30-40.00)	16.74±14.44 (0.20-32.60)	16.36±10.83[b] (0.20-40.00)	7.11±2.26[a] (3.40-11.70)
Invertase (U/kg)	155.78±42.44 (118.81-228.56)	114.64±105.58 (19.39-207.58)	124.64±92.60 (7.87-233.88)	132.90±78.07[a] (7.87-233.88)	109.73±112.24[a] (10.52-377.48)
Acid phosphatase (mg P/100 g/24h)	38.62±3.86 (34.48-44.86)	48.51±13.09 (33.87-59.09)	41.96±7.99 (33.56-55.06)	43.03±9.46[a] (33.56-59.09)	55.62±8.34[b] (26.86-59.41)
Proline (mg/kg)	839.42±215.38 (552.79-1126.13)	832.98±644.22 (112.01-530.27)	1006.01±659.70 (20.56-1877.69)	892.80±512.88[b] (20.56-1877.69)	487.66±275.32[a] (28.64-976.89)
Total carotenoid (mg β-carotene/kg)	8.84±2.92 (5.90-12.47)	9.75±3.64 (4.68-14.93)	20.46±22.06 (4.38-59.07)	13.02±13.23[a] (4.38-59.07)	7.20±3.74[a] (1.62-16.56)
Total phenol raw honey (mg gallic acid/100 g)	38.81±20.00 (24.52-74.08)	53.50±25.71 (33.56-96.26)	47.09±17.28 (24.10-62.86)	46.46±20.67[a] (24.10-96.26)	57.87±25.31[a] (33.86-114.88)
Total phenol honey extract (mg gallic acid/100 g)	15.05±1.74 (12.64-17.51)	25.15±14.43 (12.30-47.02)	20.56±5.41 (14.33-27.97)	20.25±9.33[b] (12.30-47.02)	15.07±7.95[a] (8.14-34.88)
Total flavonoids-*neutral medium*-(mg quercetin/100 g)	11.17±7.51 (2.94-20.49)	7.01±2.99 (3.90-10.64)	8.63±4.26 (2.51-14.08)	8.94±5.20[a] (2.51-20.49)	10.73±15.16[a] (2.48-59.93)
Total flavonoids-*alkaline medium*-(mg catechin/100 g)	30.28±3.09 (27.79-35.48)	30.87±4.71 (26.89-38.70)	33.20±8.49 (26.58-47.77)	31.45±5.60[b] (26.58-47.77)	26.95±6.71[a] (13.37-41.31)
o-Diphenols (μg cathechol/100 g)	11.79±13.97 (2.78-32.59)	11.50±9.11 (2.09-23.97)	18.84±20.22 (5.19-53.27)	14.20±14.46[a] (2.09-53.27)	14.67±10.40[a] (0.19-44.55)
TEAC (μmol trolox/100 g)	221.80±80.60 (143.18-336.79)	273.86±139.73 (121.12-448.78)	187.25±119.83 (21.76-334.82)	227.64±113.56[a] (21.76-488.78)	243.01±205.80[a] (38.82-733.55)

Different superscript letters (a–b) indicate significant differences ($p < 0.05$) for the same parameter.

"I" samples in comparison with the "A" honeys. Most samples showed pH values lower than 4.5, with no significant differences between pH of "A" and "I" honeys. Only A-M1 exceeded the 50 meq/kg free acid legal limit. The free acid of the rest of the samples was below 50 meq/kg.

Among "A" honeys, A-S samples exhibited the lowest HMF average. A-J5, A-M5 and 6 "I" honeys exceeded the limit of 40 mg/kg. A-J5 and A-M5 also exhibited the lowest values for diastase number and did not meet the criteria for this parameter (3 or 8 on the Schade scale). 71.4% "I" samples did not fulfill the minimum diastase activity. "A" samples exhibited significantly lower values of HMF and higher values of diastase number. Proline contents were also significantly higher in "A" honeys. A-M5 and one "I"

honey showed proline contents below the limit of 180 mg/kg that was proposed for authentic honeys (Bogdanov, 2017).

A-S honeys showed higher mean values for invertase activity than A-J, A-M and "I" samples, although there were no significant differences between the invertase activities of "A" and "I" honeys. Both the lowest and the highest values were exhibited by two A-M honeys; the sample with the lowest value was A-M5, the same sample that did not have the minimum quality required for honeys (Codex, 2001; OJEC, 2002). A-S honeys showed the lowest average of acid phosphatase activity, in contrast to the highest average of A-J samples. "I" honeys showed significantly higher acid phosphatase activities than "A" samples. However, the lowest acid

Table 2.
Unacceptable percentage of Algerian (n= 15) and imported (n= 14) honey samples according to international regulations. A-S: Algerian spurge-labeled honeys. A-J: Algerian jujube-labeled honeys. A-M: Algerian multifloral-labeled honeys.

PARAMETER	REGULATION Codex (2001), OJEC (2002)	Unacceptable percentage of samples				
		A-S (n= 5)	A-J (n= 5)	A-M (n= 5)	Algerian honeys (n= 15)	Imported honeys (n= 14)
Electrical conductivity (mS/cm)	< 0.8 mS/cm	0	0	0	0	0
Moisture (%)	< 20%	0	0	0	0	0
Free acid (meq/kg)	< 50 meq/kg	0	0	20	6.7	0
HMF (mg/kg)	< 40 mg/kg	0	20	20	13.3	42.8
Diastase number (Schade units)	> 8 (Schade scale)	0	20	20	13.3	71.4
	Specifications of different countries (Thrasyvoulou et al., 2018)					
Invertase (U/kg)	Germany: > 60 U/kg (Auslese Honig) > 80 U/kg (Feine Auslese Honig)	0 0	40 40	20 20	20 20	35.7 42.9
	Poland: > 250 mg/kg	0	40	20	20	21.4
Proline (mg/kg)	Turkey: > 120 mg/kg (acacia, rosemary honeys) > 180 mg/kg (bakery, canola, citrus, lavender, lime and eucalyptus honeys) > 300 mg/kg	0	40	20	20	7.1 7.1 14.3

phosphatase activity was exhibited by an "I" buckwheat-labeled honey.

All "A" samples exhibited higher averages than "I" honeys for TCC, but no significant differences were found between the TCC results of "A" and "I" samples. Among "A" honeys, A-J samples and their methanolic extracts showed the highest TPC averages, whereas A-S samples exhibited the lowest. Comparing methanolic extracts of "A" and "I" honeys, TPCs of "A" samples were significantly higher than those of "I" honeys. No significant differences were found regarding quercetin-type TFC and *o*-diphenols content between "A" and "I" samples, in contrast to the higher values of catechin-type TFC exhibited by "A" honeys in comparison with "I" samples. Both the highest and the lowest individual values of catechin-type TFC were observed in A-M honeys. A-M5 sample showed the highest value for this parameter. Regarding extracts of "I" honeys, the lowest catechin-type TFC was exhibited by the extract of one sample of acacia-labeled honey and the highest result was shown by the

extract of a buckwheat-labeled honey. Among "A" samples, A-M honeys showed the highest averages of *o*-diphenols, and A-M5 honey had the highest content. The values of "I" honeys varied from 0.19 µg cathechol/100 g in one citrus-labeled honey to 44.55 µg cathechol/100 g in a multifloral-labeled honey.

In our study, A-J honeys exhibited higher TEAC averages than A-S and A-M samples, and the TEAC values were statistically similar for both "A" and "I" honeys. PCA (Fig. 1) showed a good separation of "A" and "I" samples, using the parameters that showed the strongest loadings (both positive and negative), on component 1 and component 2: electrical conductivity, moisture, degrees Brix, free acid, diastase, proline, acid phosphatase, TCC and TPC (honey extract). Component 1 and component 2, with eigenvalues greater than or equal to 1.0, explained 59.30% of the variance. Component 2 exhibited the differences regarding free acidity and TPC (honey extract), whereas component 1 comprised the information about the rest
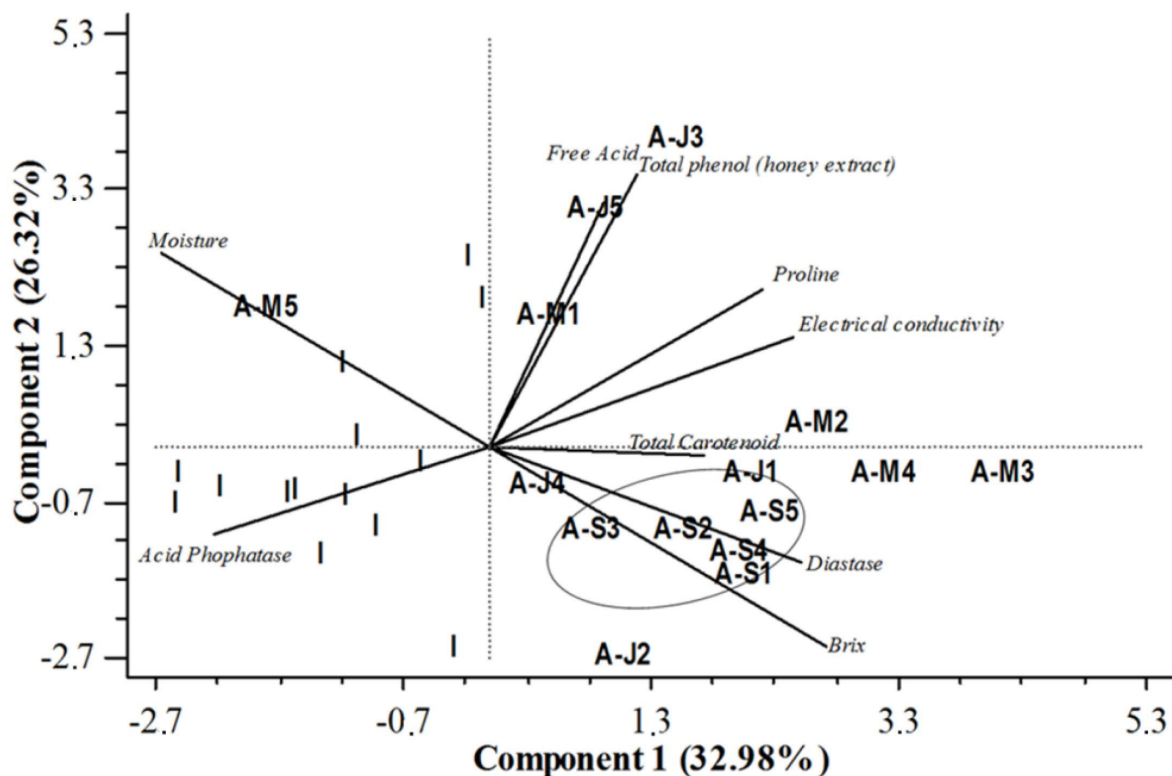

Fig. 1. Principal component analysis of electrical conductivity, moisture, degrees Brix, free acid, proline, diastase, acid phosphatase, total carotenoid and TPC (honey extract) in Algerian honeys labeled as spurge (A-S), jujube (A-J) and multifloral (A-M), as well as in imported (I) honeys.

of parameters. Component 1 separated the honeys into two groups, "I" samples on the left and "A" samples on the right. "A" honeys showed higher values of electrical conductivity, degrees Brix, proline, TCC and diastase activity, while "I" honeys exhibited higher moisture percentages and acid phosphatase activities. PCA clearly included A-M5 sample within the group of "I" honeys because the characteristics and poor quality of this sample were similar to those of most "I" honeys.

DISCUSSION

Electrical conductivity is related to ash content and also illustrative of the source of nectar or honeydew (Machado De-Melo et al., 2018). Our electrical conductivity values agreed with those described in the literature for honeys from different countries (Silva, Sousa, & Taveira, 2017), and for other Algerian samples of the same botanical origins (Haderbache, Mouna, & Arezki, 2013), although Zerrouk et al. (2018) reported a mean value (0.65 mS/cm) for electrical conductivity of Algerian *Ziziphus lotus* honeys higher than our average (0.35 mS/cm) for A-J honeys.

Both moisture and degrees Brix are indicators of honey maturity. Our results for moisture were similar to those described in other Algerian samples (Ouchemoukh, Louaileche, & Schweitzer, 2007; Diafat et al., 2017; Zerrouk et al., 2018), in that they were higher than the average (13.2 g/100 g) reported by Khalil et al. (2012), and the average (13.9 g/100g) by Mekious et al. (2015) in Algerian jujube honeys. In contrast, Habati et al. (2017) described higher moisture values in Algerian samples (16.1-20.4 g/100 g). The different values and ranges of moisture might be due to the harvesting season, moisture content of plants and degree of ripeness reached in the hive (Bogdanov, 2017). Degrees Brix values of all samples were within the usual values described in the literature for other honeys (Silva, Sousa, & Taveira, 2017).

Both pH and free acid are related to organic acids' content. pH is influenced by inorganic ions, extraction and storage conditions, affecting texture, stability and shelf life of honey (Terrab et al., 2004). Two A-J honeys exhibited pH higher than 4.4, agreeing with previous research (Haderbache, Mouna, & Arezki, 2013; Mekious et al., 2015) that reported relatively high pH of jujube honeys. In general, our results of free acid agreed with those of literature references (Benaziza-Bouchema & Schweitzer, 2010). However, our average for A-S honeys was higher than the mean value (15.13 meq/kg) described by Haderbache, Mouna, & Arezki (2013) in other Algerian spurge honeys. Likewise, our average for A-J honeys was also higher than the mean value (14.75 meq/kg) obtained by Haderbache, Mouna, & Arezki (2013) and the average (12.5 meq/kg), obtained by Zerrouk et al. (2018) in other Algerian jujube samples. These contrasting results of free acid might be due to distinct geographic harvesting season (Silva, Sousa, & Taveira, 2017), hive locations and storage conditions. The high value of free acid exhibited by sample A-M1 (80 meq/kg) showed that this honey was likely spoiled.

HMF is a freshness, heating and ageing parameter (Bogdanov, 2017). Most HMF results for our "A" samples agreed with those of other Northwestern Algerian honeys (Achouri et al., 2019). As for the two samples which exceeded the HMF limit, A-J5 was from Djelfa, an Algerian location with a very warm and dry-summer climate, and A-M5 was the sample that did not fulfil other quality criteria.

Diastase activity is also a freshness parameter of honey (Codex, 2001; OJEC, 2002) with minimum limits in several legislations, some of them related to HMF contents. A-J2 sample exhibited the highest result for diastase number (40 Schade units). It came from the region of Ain Ouessara, a semi-arid area, in which jujube honey is particularly prestigious. This sample also showed a high value of invertase and non-detectable HMF content, which confirmed its quality related to ageing and heating. Our diastase activities for "A" honeys were similar to those described by Haderbache, Mouna, & Arezki (2013) for Algerian spurge honeys but about half of the average described by the latter authors for jujube Algerian honeys. Our

values of diastase activity for A-S samples (all of them from North Algeria), were similar to those found by Haouam et al. (2019) in Northern Algerian honeys (ranging from 9.62 to 29.47 Shade units). The results of "I" honeys' HMF and diastase activity were far from desirable, highlighting the poor quality of honeys from abroad that are sold in Algerian markets.

Proline, the most abundant amino acid of honey. It has been suggested as a parameter of honey ripeness. The vast majority of our proline results were concordant with literature values (Elamine et al., 2018). In Poland and Turkey there are regulations regarding minimum proline concentrations in honey (Thrasyvoulou et al., 2018). Two A-J honeys and one A-M sample fulfilled neither the Polish specification (not less than 250 mg/kg), nor the Turkish specification (not less than 300 mg/kg) with respect to proline. As for "I" honeys, three samples did not fulfill the Polish regulation, and two of whom did not either fulfill the Turkish legislation. The "I" honey that fulfilled the Turkish specification for proline but not the Polish one was labeled as citrus honey and contained 189.1 mg proline/kg honey, which is above the minimum limit of proline (180 mg/kg) required in Turkey for citrus honey.

Invertase, which hydrolyzes the sucrose of nectar and honeydew into fructose and glucose, has been proposed as a quality parameter of honey (Machado De-Melo et al., 2018). Three "A" samples (2 A-J honeys and 1 A-M honey) and five "I" honeys exhibited invertase results lower than 60 U/kg which is the minimum invertase value required in Germany (Thrasyvoulou et al., 2018). One "I" sample, whose invertase activity was 72.5 U/kg, did not fulfill the minimum value of 80 U/kg for invertase activity required in Germany for higher quality honeys (Thrasyvoulou et al., 2018).

Two honeys, one "A" (A-M5) and one "I", showed the values of several parameters that did not correspond to those of authentic/acceptable honeys. A-M5 exhibited a low electrical conductivity (0.09 mS/cm), a high HMF content (177 mg/kg) and the lowest diastase (0.20 Schade scale) and invertase (7.87 U/kg) activities, and the lowest proline concentration (20.56 mg/

kg) which lacked the minimum quality required for commercialization. The "I" sample showed the lowest electrical conductivity (0.04 mS/cm), the highest HMF content (580.83 mg/kg), low diastase (7.9 Schade scale) and invertase (13.44 U/kg) activities and a low proline concentration (28.64 mg/kg) with values that were far from desirable for a genuine honey. High values of honey acid phosphatase were related to honey fermentation. Our values for "A" samples were lower than the results described in the literature for other honeys (Alonso-Torre et al., 2006).

Carotenoids are pigments that contribute to antioxidant activity. The TCC of honey depends on the geographical origin, season, and environmental factors (Boussaid et al., 2014). Among "A" samples, the TCC average of A-M honeys was the highest. Mouhoubi-Tafinine, Ouchemoukh, & Tamendjari (2016) described values from 3.0 to 10.1 mg β-carotene/Kg on honeys from different regions of North Algeria. A study on Cuban honeys described values between 1.17 and 5.57 mg β-carotene/Kg (Álvarez-Suárez et al., 2010). In Tunisian samples Boussaid et al. (2014) reported a TCC ranging from 1.16 to 4.72 mg β-carotene/Kg.

Literature describes positive correlations between TPC and the antioxidant activity of honey (Álvarez-Suárez et al., 2010; Ciappini & Stoppani, 2014; Meinen, Camilleri, & Attard, 2014). TPC measured on raw honeys usually provides higher results, because the determination includes other reducing substances, and the extraction helps to remove some water-soluble phenols. Our TPC averages agreed with the results reported by Mouhoubi-Tafinine, Ouchemoukh, & Tamendjari (2016), who described a range of TPC from 15.84 mg/100 g (methanolic extract) to 61.63 mg/100 g (aqueous solution) in Algerian honeys. In contrast, our TPC values for "A" samples were lower than those obtained by Habati et al. (2017) in five honeys from South Algeria (53.93-123.05 mg gallic acid/100 g raw honey) and lower to the results of Elamine et al. (2018) in twelve honeys from Morocco (59.32-123.65 mg gallic acid/100 g raw honey). Differences in honeys' TPC could be due to habitat differences and harvesting year,

but to get more accurate results, TPC should be measured on honey extracts. Our values for quercetin-type and catechin-type TFC were higher than those obtained for other honey extracts in the literature (Khalil et al., 2012; Sancho et al., 2016). Interestingly, "A" honey samples showed lower quercetin-type TFC values than "I" samples conversely to the higher results for catechin-type TFC of "A" honeys in comparison with "I" samples. Algerian honeys likely contain higher amounts of flavan-3-ols, rutin, luteolin and phenolic acids than honeys from other countries. Therefore, a study of the composition of flavonoids of Algerian honeys would be interesting because it could contribute to a future characterization of Algerian honeys. o-Diphenols were related to antioxidant activity (Afoakwa et al., 2012). No significant differences were found between the o-diphenol contents of "A" and "I" honeys. As stated above, sample A-M5 was unacceptable for commercialization according to the parameters established in international regulations, however its content of both catechin-type TFC and o-diphenols was the highest of all analyzed samples. Thus, the sample A-M5 was likely a honey adulterated with vegetable syrups boiled up with honey or sugars.

Currently, antioxidant activity is considered an interesting parameter to characterize honeys. Comparing our TEAC results with those of the literature, Álvarez-Suárez et al. (2010) obtained similar results for Cuban morning glory (201.4 μmol trolox/100 g) and singing bean (195.8 μmol trolox/100 g) honeys, higher for linen vine honeys (294.5 μmol trolox/100 g), and lower for black mangrove (122.0 μmol trolox/100 g) and Christmas vine (103.5 μmol trolox/100 g) honeys. Our study confirms the interesting antioxidant potential of Algerian honeys. Khalil et al. (2012) came to the same conclusion after carrying out FRAP and DPPH assays on other Algerian samples.

With regard to PCA, all A-S honeys were perfectly grouped because of the common physicochemical features exhibited by these samples that can be summarized in lower values for moisture, free acid and acid phosphatase activities as well as higher values for degrees Brix and a narrower range of TPC (honey extract), in comparison with A-J and A-M honeys. Therefore, future legislation of Algerian honeys could establish particular features for Algerian spurge honeys, after an analysis of a significant number of spurge honeys from different Algerian locations of different harvesting years that must include melissopalinology and sensory characterization. It could help contribute to a possible protected designation of origin for Algerian spurge honeys. 80% "A" samples fulfilled international standards, whereas only 21.4% "I" honeys did so. 13.3% "A" samples and 7.1% "I" honeys showed proline values lower than 180 mg/kg, which is the recommended limit for authentic honeys. Algeria has no legal regulation for honey, and countries, where there are such regulations, likely take advantage of this fact and export their unacceptable honeys to Algeria. Therefore, Algeria should establish a honey directive as soon as possible to avoid adulterations and frauds. To improve the beekeeping sector, good beekeeping practices including proper honey handling should be adopted in Algeria, except for honeys from very warm areas, in a similar way that European legislation did for honeys from tropical climate regions.

In comparison with imported honeys, Algerian honeys exhibited higher values for electrical conductivity, degrees Brix, diastase activities, proline contents, as well as TPC and catechin-type TFC in their methanolic extracts. Conversely, they showed lower results for moisture percentages, HMF contents and acid phosphatase activities.

PCA showed a clear distinction between "A" and "I" honeys. Only one "A" sample, whose quality lacked international standards, was classified within the "I" group. Spurge-labeled honeys were grouped because of their common features of moisture, degrees Brix, free acid, acid phosphatase and TPC (honey extract). Thus, a detailed characterization of spurge honeys in the near future would be promising for the Algerian beekeeping sector.

REFERENCES

Achouri, M.Y., Selka, M.A., Chenafa, A., Brahim, S., Messafeur, M.A., Toumi, H. (2019). Teneur en 5-hydroxyméthylfurfural (HMF) dans les miels du Nord-Ouest de l'Algérie. *Toxicologie Analytique et Clinique*, *31*(2), 100-105. https://doi.org/10.1016/j.toxac.2019.03.001

Afoakwa, E.O., Takrama, F.S., Quao, J., Budu, A.S., Saalia, F.K. (2012). Changes in total polyphenols, *o*-diphenols and anthocyanin concentrations during fermentation of pulp pre-conditioned cocoa (*Theobroma cacao*) beans. *International Food Research Journal*, *19*(3), 1071-1077.

Alonso-Torre, S.R., Cavia, M.M., Fernández-Muiño, M.A., Moreno, G., Huidobro, J.F., Sancho M.T. (2006). Evolution of acid phosphatase activity of honeys from different climates. *Food Chemistry*, *97*(4), 750-755. https://doi: 10.1016/j.foodchem.2005.06.010

Álvarez-Suárez, J.M., Tulipani, S., Díaz, D., Esteves, Y., Roman-Dini, S., Giampieri, F., Battino, M. (2010). Antioxidant and antimicrobial capacity of several monofloral Cuban honeys and their correlation with color, polyphenol content and other chemical compounds. *Food and Chemical Toxicology*, *48*(8-9), 2490-2499. https://doi:10.1016/j.fct.2010.06.021

Association of Official Analytical Chemists-International [AOAC]. (2016). Official Methods of Analysis. 20th edition. Arlington. Virginia. USA.

Baltrušaityte, V., Venskutonis, P.R., & Čeksteryte, V. (2007). Radical scavenging activity of different floral origin honey and beebread phenolic extracts. *Food Chemistry*, *101*(2), 502-514. https://doi:10.1016/j.foodchem.2006.02.007

Benaziza-Bouchema, D., & Schweitzer, P. (2010). Caractérisation des principaux miels des régions du Nord de l'Algérie. *Cahiers Agricultures, 19*(6), 432-438. https://doi: 10.1684/agr.2010.0432

Bogdanov, S. (2017). The Honey Book, Bee Product Science. Retrieved July 27, 2018, from http://www.bee-hexagon.net.

Boussaid, A., Chouaibi, M., Rezig, L., Hellal, R., Donsia, F., Ferrari, G., Hamdi, S. (2014). Physicochemical and bioactive properties of six honey samples from various floral origins from Tunisia. *Arabian Journal of Chemistry, 11*(2), 265-274. https://doi: 10.1016/j.arabjc.2014.08.011

Ciappini, M.C., & Stoppani, F.S. (2014). Determination of antioxidant capacity, flavonoids, and total phenolic content in eucalyptus and clover honeys. *Journal of Apicultural Science*, *58*(1), 103-111. https://doi.org/10.2478/jas-2014-0010

Codex (2001) Codex Alimentarius standard for honey 12-1981. Revised Codex Standard for Honey, Standards and Standard Methods (Vol. 11).

Diafat, A.E.O., Benouadah, A., Bahloul, A., Meribai, A., Mekhalfi, H., Bouaziz, F., Arrar, L. (2017). Physicochemical properties and pollen analyzes of some Algerian honeys. *International Food Research Journal, 24*(4), 1453-1459.

Elamine, Y., Aazza, S., Lyoussi, B., Antunes, M.D., Estevinho, L.M., Anjos, O.,... Miguel M.G. (2018). Preliminary characterization of a Moroccan honey with a predominance of *Bupleurum spinosum* pollen. *Journal of Apicultural Research, 57*(1),153-165. https://doi: 10.1080/00218839.2016.1265759

Günther, F., & Burckhart, O. (1967). Bestimmung der sauren Gesamtphosphatase in Honig. *Deutsche Lebensmittel-Rund-schau, 63*, 41-44.

Habati, M., Gherib, A., Bakchiche, B., & Benmebarek, A.A. (2017). Study on the physicochemical, antioxidant properties and mineral content of five honeys produced in the central region of Algeria. *Scientific Study and Research: Chemistry and Chemical En-*

gineering, Biotechnology, Food Industry, 18(2), 121-134.

Haderbache, L., Mouna, B., & Arezki, M. (2013). Ziziphus lotus and Euphorbia bupleuroides Algerian honeys. World Applied Sciences Journal, 24(11), 1536-1543. https://doi: 10.5829/idosi.wasj.2013.24.11.7525

Haouam, L., Dailli, H., Bruneau, E., & Tahar, A. (2019). The quality of honeys influenced by the traditional heating method. Journal of Microbiology, Biotechnology and Food Sciences, 8(6), 1276-1280. https://doi.org/10.15414/jmbfs.2019.8.6.1276-1280

Huidobro, J.F., Santana, F.J., Sanchez, M.P., Sancho, M.T., Muniategui, S., Simal-Lozano, J. (1995). Diastase, invertase and β-glucosidase activities in fresh honey from north-west Spain. Journal of Apicultural Research, 34(1), 39-44. https://doi:10.1080/00218839.1995.11100884

International Honey Commission (2009) Harmonised methods of the International Honey Commission. Retrieved July 29, 2019, from http://www.ihc-platform.net

Khalil, I., Moniruzzaman, M., Boukraâ, L., Benhanifia, M., Islam, A., Islam, N., Sulaiman, S.A., Gan, S.H. (2012). Physicochemical and antioxidant properties of Algerian honey. Molecules, 17(9), 11199-11215. http://doi:10.3390/molecules170911199

Machado De-Melo, A.A.M., De Almeida-Muradian, L.B., Sancho, M.T., & Pascual-Maté, A. (2018). Composition and properties of Apis mellifera honey: A review. Journal of Apicultural Research, 57(1), 5-37. https://doi: 10.1080/00218839.2017.1338444

Makhloufi, C., Kerkvliet, J.D., D'Albore, G.R., Choukri, A., & Samar, R. (2010). Characterization of Algerian honeys by palynological and physico-chemical methods. Apidologie, 41(5), 509-521. http://doi:10.1051/apido/2010002

Meinen, N., Camilleri, L., & Attard, E. (2014). The antioxidant activity of Maltese honey. Journal of Apicultural Science, 58(1), 51-60. https://doi.org/10.2478/jas-2014-0004

Mekious, S., Houmani, Z., Bruneau, É., Masseaux, C., Guillet, A., Hance, T. (2015). Caractérisation des miels produits dans la région steppique de Djelfa en Algérie. Biotechnology, Agronomy and Society and Environment, 19(3), 221-231.

Mouhoubi-Tafinine, Z., Ouchemoukh, S., & Tamendjari, A. (2016). Antioxidant activity of some Algerian honey and propolis. Industrial Crops and Products, 88, 85-90. http://doi:10.1016/j.indcrop.2016.02.033

Official Journal of the European Communities [OJEC]. EU. (2002). Council Directive 2001/110/EC of 20 December 2001 relating to honey. Official Journal, L 10, 47-52.

Otmani, I., Abdennour, C., Dridi, A., Kahalerras, L., & Halima-Salem, A. (2019). Characteristics of the bitter and sweet honey from Algeria Mediterranean coast. Veterinary World, 12(4), 551-557. https://doi.org/10.14202/vetworld.2019.551-557

Ouchemoukh, S., Louaileche, H., & Schweitzer, P. (2007). Physicochemical characteristics and pollen spectrum of some Algerian honeys. Food Control, 18(1), 52-58. https://doi:10.1016/j.foodcont.2005.08.007

Ouchemoukh, S., Amessis-Ouchemoukh, N., Gómez-Romero, M., Aboud, F., Giuseppe, A., Fernández-Gutiérrez, A., Segura-Carretero, A. (2017). Characterisation of phenolic compounds in Algerian honeys by RP-HPLC coupled to electrospray time-of-flight mass spectrometry. LWT-Food Science and Technology, 85, 460–469. https://doi:10.1016/j.lwt.2016.11.084

Pękal, A., & Pyrzynska, K. (2014). Evaluation of aluminium complexation reaction for flavonoid content assay. Food Analytical Methods, 7(9), 1776-1782. https://doi:10.1007/s12161-014-9814-x

Piszcz, P., & Głód, B.K. (2019). Antioxidative properties of selected Polish honeys. Journal of Apicultural Science, 63(1), 81-91. https://doi.org/10.2478/jas-2019-0003

Ramadani, V., Hisrich, R.D., Dana, L.-P., Palalic, R., &

Panthi, L. (2019). Beekeeping as a family artisan entrepreneurship business. *International Journal of Entrepreneurial Behaviour and Research, 25*(4), 717–730. https://doi.org:10.1108/IJEBR-07-2017-0245

Re, R., Pellegrini, N., Proteggente, A., Pannala, A., Yang, M., Rice-Evans, C. (1999). Antioxidant activity applying an improved ABTS radical cation decolorization assay. *Free Radical Biology and Medicine, 26*(9-10), 1231–1237. https://doi:10.1016/s0891-5849(98)00315-3

Rodríguez-Flores, S., Escuredo, O., & Seijo, M.C. (2016). Characterisation and antioxidant capacity of sweet chestnut honey produced in North-West Spain. *Journal of Apicultural Science, 60*(1), 19-30. https://doi.org/10.1515/jas-2016-0002

Sancho, M.T., Pascual-Mate, A., Rodrıguez-Morales, E.G., Oses, S.M., Escriche, I., Periche, A., Fernandez-Muino, M.A. (2016). Critical assessment of antioxidant-related parameters of honey. *International Journal of Food Science and Technology, 51*(1), 30–36. https://doi: 10.1111/ijfs.12988

Sari, F., Kandemir, I., Ceylan, D.A., & Gül, A. (2020). Using AHP and PROMETHEE multi-criteria decision making methods to define suitable apiary locations. *Journal of Apicultural Research, 2020*, 1-12. https://doi.org/10.1080/00218839.2020.1718341

Siegenthaler, U. (1977). Eine einfache und rasche Methode zur Bestimmung der alpha Glucosidase (Saccharase) im Honig. *Mitteilungen aus der Gebiete der Lebensmittelunters und Hygiene, 68*, 251-258.

Silva, L.R., Sousa, A., & Taveira, M. (2017). Characterization of Portuguese honey from Castelo Branco region according to their pollen spectrum, physico-chemical characteristics and mineral contents. *Journal of Food Science and Technology, 54*(8), 2551-2561. https://doi: 10.1007/s13197-017-2700-y

Singleton, V.L., & Rossi, J.A. (1965). Colorimetry of total phenolics with acid reagents. *American Journal of Enology and Viticulture, 16*, 144-158.

Statgraphics Centurion XVI.II. Statgraphics Technologies, Inc. The Plains, Virginia, USA.

Terrab, A., Recamales, A.F., Hernanz, D., & Heredia, F.J. (2004). Characterisation of Spanish thyme honeys by their physicochemical characteristics and mineral contents. *Food Chemistry, 88*(4), 537-542. https://doi:10.1016/j.foodchem.2004.01.068

Thrasyvoulou, A., Tananaki, C., Goras, G., Karazafiris, E., Dimou, M., Liolios, V., Kanelis, D., Gounari, S. (2018). Legislation of honey criteria and standards. *Journal of Apicultural Research, 57*(1), 88-96. https://doi:10.1080/00218839.2017.1411181

Zerrouk, S., Seijo, M.C., Escuredo, O., & Rodríguez-Flores, M.S. (2018). Characterization of *Ziziphus lotus* (jujube) honey produced in Algeria. *Journal of Apicultural Research, 57*(1), 166–174. https://doi: 10.1080/00218839.2017.1399663