Scientific Bulletin. Series F. Biotechnologies, Vol. XXVIII, No. 1, 2024
ISSN 2285-1364, CD-ROM ISSN 2285-5521, ISSN Online 2285-1372, ISSN-L 2285-1364

QUALITY EVALUATION OF SOME COMMERCIAL HONEY SAMPLES

Carmen Mihaela TOPALA, Sorin Ionut PRITA, Loredana Elena VIJAN

Pitesti University Centre, National University of Science and Technology Politehnica Bucharest, 1 Targu din Vale Street, Pitesti, Romania

Corresponding author emails: loredana.vijan@upb.ro, loredana.vijan@upit.ro

Abstract

Honey is one of the most valuable natural product used for thousands of years in human nutrition. The quality of honey must be controlled analytically with the aim of preserving the consumer from commercial speculation. The values of the physicochemical parameters for ten commercial honey samples attest that all samples appeared to conform to the European Legislation. However, food products such as honey are easily adulterated both in domestic and foreign trade. An alternate analytical technique used to detect adulterations and authenticity of honey implies both FTIR spectrum screening and multivariate analysis of samples. The spectral data of ten honey samples were acquired in the range of 4000–400 cm^{-1}. Out of them, there were two adulterant materials. A spectral range of 1800 to 650 cm^{-1} was selected in order to achieve a satisfactory cluster discrimination. Based on spectral variations, cluster analysis may also be used to categorize and differentiate between pure and contaminated honey.

Key words*: 5-hydroxymethylfurfural, ATR-FTIR spectroscopy, cluster analysis, multivariate analysis, polyphenols.*

INTRODUCTION

Honey is the most important primary product of apiculture, from a qualitative point of view, as well as from an economic perspective. It was also the first apiarian product used by mankind from ancient times. Throughout time, in almost all the cultures there is evidence of honey being used as a food source with therapeutic benefits and as a symbol in magical and religious ceremonies (Eteraf-Oskouei & Najafi, 2013; Al-Ghamdi & Ansari, 2021).

According to the Explanatory Dictionary of the Romanian Language, honey is “a yellowish semi-liquid substance with a sweet taste and a pleasant aroma, very rich in sugars, vitamins, and enzymes, gathered and produced by bees from the nectar of the flowers” (Coteanu et al., 2016). The nectar that the bees suck from the flowers turns into honey through a kind of "kiss" between the bees that participate in the honey maturation process, through which small drops of nectar are transferred from one bee to another, moment when the nectar is enriched with the enzymes that ennoble it, then the excess water is removed. The production of honey by bees begins with the collection of nectar or manna and ends with the filling of the cells of the honeycomb, in which the honey is stored. The requirement to convert complex carbohydrates (sucrose, maltose, etc.) into simple, assimilable sugars (glucose and fructose) so that the bees may completely digest them throughout the winter months gives rise to enzymatic processing. Afterwards, the nectar or manna is diluted with saliva and by reducing the water content, the enzymatic processes are blocked and the storage space is decreased (Giosanu et al., 2022; Vîjan et al., 2023; Albu et al., 2024).

Honey has water, carbohydrates, pollen, minerals, enzymes, vitamins, pigments, aromatic compounds and acids in its composition. Carbohydrates include fructose (38%), glucose (31%), sucrose (1%), maltose and other disaccharides (7%), melezitose (characteristics of manna honey). Minerals in honey, such as: potassium, sodium, calcium, magnesium, chlorine, sulphates, phosphates, silicon, have a low content in flower honey, but they have a high content in manna honey. These minerals are responsible for the extraordinary qualities of honey. Thiamine (vitamin B_1), riboflavin (vitamin B_2), nicotinic acid (vitamin B_3), vitamin K, folic acid (vitamin M), biotin (vitamin H), and pyridoxine are among the vitamins found in honey. Honey contains trace levels of vitamins, amino acids, and proteins due to pollen (5%) in the honey. The enzymes in honey are: glucose oxidase,

catalase, phosphatase, diastase (which transforms starch into dextrins) and invertase (which turns sucrose into fructose and glucose). In flower honey, the enzymes have a double origin: vegetable (the enzymes in the nectar) and animal (the enzymes in the bee's saliva). The composition and therapeutic effects of different types of honey are determined by the various plants visited by bees, respectively by their active principles (Hills et al., 2019; Giosanu et al., 2022; Vîjan et al., 2023; Albu et al., 2024).

The source of the nectar determines the honey's scent, which can range from subtle (acacia) or bitter (chestnut honey) to the distinct perfume of linden, raspberry, etc. The colour of honey can vary from almost colourless (acacia honey) to dark brown (manna honey).

A good honey (16–18% moisture) is viscous at normal temperature (20°C). If the humidity is higher than 21%, the honey flows like water, which means that it was obtained and/or stored inadequately, and it will degrade quickly due to its poor quality.

Honey acidity is maximum 4 mEq/kg for flower honey and 5 mEq/kg for manna honey. Normal pH values for honey are between 3.4 and 6.1.

One of the factors used to confirm the authenticity of honey is electrical conductivity. In flower honey, the electrical conductivity is very different according to the type of honey. The pollen spectrum is the basic criterion in order to be able to correctly assess the type of honey, due to the fact that the morphology of the pollen grains is characteristic for each individual plant species (Pérez et al., 2007; Castro-Vázquez et al., 2008; Bogdanov, 2009; Alvarez-Suarez et al., 2010; Bodó et al., 2021; Giosanu et al., 2022; Vîjan et al., 2023; Albu et al., 2024).

Since the physicochemical parameters of honey types are generally similar, it is difficult to distinguish between different types of honey based on the physicochemical analysis of this product (Giosanu et al., 2022; Vîjan et al., 2023).

The food sector finds FTIR (Fourier transform infrared) spectroscopy to be an alluring technique since it offers quick, easy, and non-destructive evaluations of chemical and physical components. The development of multivariate data analysis techniques and advancements in FTIR instruments make this technology perfect for large-scale, quick screening and for the description of minor food components down to parts per billion (ppb) levels.

In order to verify the quality of the tested honey samples, one analysed ten honey samples as regards several physicochemical parameters, such as: moisture, ash, pH, free acidity (FA), electrical conductivity (EC), total sugar content (TSC), 5-hydroxymethylfurfural (5-HMF), and several biochemical parameters, such as: total polyphenol content (TPC), total flavonoid content (TFC), total tannin content (TTC), and the antioxidant activity. Additionally, multivariate analysis and ATR-FTIR spectroscopy were combined to identify and assess honey adulteration.

MATERIALS AND METHODS

Chemicals and Reagents

All the chemicals and reagents were acquired from Merck in Darmstadt, Germany.

Honey samples

Ten honey samples, eight from primary honey producers and two commercial honey samples (Greek and Manna honey) were analyzed. The eight samples of honey from primary producers from Romania are: two sunflower (S) from Argeș - Costesti (AG-C) and Argeș - Gliganu (AG-G), three acacia samples (A) from Argeș - Costesti (AG-C), Argeș - Mosoaia (AG-MO) and Argeș - Vedea (AG-V) and three multifloral samples (M) from Tulcea - Casimcea (TL-C), Arges - Mozaceni (AG-MZ) and Giurgiu-Bolintin (GR-B). Greek and Manna honey were purchased from the supermarket.

Physicochemical determinations

By means of a C-561 Consort multimeter, one determined the electrical conductivity (EC) and the pH according to the methodology suggested by Vîjan et al. (2023). The EC results were expressed as microsiemens per centimeters (μS/cm).

Free acidity (FA) was determined by titrating aqueous solutions of honey using 0.1 M sodium hydroxide solution to pH 8.30 according to the

methodology suggested by Vîjan et al. (2023). The FA results were expressed in milliequivalents of acids per kilogram of honey (mEq/kg).
Moisture and ash (mineral content) were determined gravimetrically by oven drying at 105–110°C, and by the calcination of dry residue at 550–600°C, respectively, until the samples were brought to a constant mass. The moisture and ash results were expressed as percent (%).
By colorimetric analysis, according to the methodology suggested by Dubois et al. (1956) one determined the total sugar content (TSC) expressed as g glucose/100 g. Similarly, by colorimetric analysis, according to the methodology suggested by Vîjan et al. (2023), one determined 5-hydroxymethylfurfural (5-HMF) content expressed in milligrams 5-HMF per kg (mg 5-HMF/kg).

Bioactive compounds determinations
By colorimetric analysis and in agreement with the methodology suggested by Vîjan et al. (2023), one determined the total polyphenol content (TPC), the total flavonoid content (TFC), and the total tannin content (TTC). The results were expressed as mg gallic acid equivalent (GAE)/100 g for TPC and TTC, and mg catechin equivalent (CE)/100 g for TFC, respectively.
The antioxidant activity, expressed as a percent of inhibition of 2,2-diphenyl-1-picrylhydrazyl (DPPH I%), was calculated according to the approach proposed by Vîjan et al. (2023).

UV-Vis and ATR-FTIR analysis
The spectral measurements were made with a UV-Vis Perkin-Elmer Lambda25 spectrometer and an FTIR Jasco 6300 spectrometer. Pike Technologies' ATR attachment with a diamond crystal enables direct sample FTIR spectra collection without any special further processing. The FTIR spectra were recorded in the region of 4000-400 cm^{-1}, detector TGS, and apodization Cosine. JASCO Spectra Manager II was used to process the spectrum data. Between measurements, the ATR crystal was washed with pure acetone and then dried with soft tissue (Topală & Tătaru, 2019; Topală et al., 2020). All measurements were made at room temperature (T= 23°C). For every sample, three replicate spectra were obtained, and the average spectrum was calculated.
Infrared Spectra were exported from Spectra Manager, in ASCII (dx) format, into the Unscrambler Software (Edition X 10.4, Camo Oslo Norway) for chemometric analysis. Spectra were pre-processed using the second-derivative transformation, the Savitzky-Golay derivation (Topală et al., 2020; Vîjan et al., 2023). The principal component analysis (PCA) model was developed using cross-validation. PCA was performed on the entire spectral range (4000 to 400 cm^{-1}), Validation: Cross Validation, Algorithm: Singular Value Decomposition (SDV). The spectral range 1800–650 cm^{-1} was chosen for Hierarchical Cluster Analysis (HCA).

Statistical Analysis
At least three replications' worth of data were presented as mean ± standard deviation.

RESULTS AND DISCUSSIONS

The parameters of honey samples, presented in Tables 1 and 2 (i.e., electrical conductivity (EC), pH, free acidity (FA), moisture, ash, total sugar content (TSC), 5-hydroxymethylfurfural (5-HMF), total phenolic content (TPC), total flavonoid content (TFC), total tannin content (TTC), and antioxidant activity (DPPH I%) showed strong oscillations depending on the honey harvest zone.
EC values ranged from 206.83 to 519.17 µS/cm, which suggested that the analyzed samples came from nectar honey (certified with EC values lower than 800 µS/cm, and ash values below 0.6%), according to Directive 2014/63/EU.
The pH values recorded for the analyzed honey samples range within the standard limits (pH 3.40–6.10) that ensure the freshness of the honey.
The free acidity values that ranged from 6.83 to 24.13 mEq/kg (below the allowed maximum of 50 mEq/kg, according to Bogdanov, 2009) attest the freshness of all the samples.
All honey samples presented a safe moisture content for storage and consumption (12.66–18.74%), below the maximum value (20%) established by the Codex Alimentarius (2001) standard.

The 5-HMF content of all the tested samples was below the maximum level of 40 mg/kg required by the International Honey Commission Standard, which attests the fact that no heat treatment had been applied to the honey (Bogdanov, 2009). The honey from Manna and Greece had the greatest 5-HMF levels (33.94 and 36.07 mg/kg, respectively).

Table 1. Honey electrical conductivity (EC), pH, free acidity (FA), moisture, ash, total sugar content (TSC) and 5-hydroxymethylfurfural (5-HMF) based on the geographical origin (GO) of the honey

Honey	GO	EC (µS/cm)	pH	FA (mEq/kg)	Moisture (%)	Ash (%)	TSC (g glucose/100 g)	5-HMF (mg/kg)
S	AG-C	448.67±1.15 a	3.93±0.01 a	15.50±0.19 a	14.92±0.13 a	0.18±0.01 a	59.99±0.03 a	19.08±0.09 a
	AG-G	519.17±1.15 a	3.97±0.01 a	17.83±0.19 a	16.14±0.13 a	0.23±0.01 a	61.65±0.03 a	19.65±0.09 a
	P	*<0.001*	*<0.001*	*0.002*	*0.001*	*0.177*	*<0.001*	*<0.001*
A	AG-C	283.83±1.15 a	4.45±0.01 a	8.83±0.19 a	12.90±0.13 c	0.11±0.01 b	62.55±0.03 b	16.22±0.09 a
	AG-MO	239.00±1.15 a	4.33±0.01 a	6.83±0.19 a	14.32±0.13 b	0.17±0.01 a	68.46±0.03 a	16.17±0.09 a
	AG-V	206.83±1.15 a	4.45±0.01 a	6.83±0.19 a	15.85±0.13 a	0.13±0.01 ab	60.35±0.03 b	15.82±0.09 a
	P	*<0.001*	*<0.001*	*<0.001*	*<0.001*	*<0.001*	*<0.001*	*<0.001*
M	AG-MZ	385.33±1.15 a	4.12±0.01 b	11.17±0.19 a	13.65±0.13 a	0.16±0.01 a	78.36± 0.03 a	24.97±0.09 a
	GR-B	377.38 ±1.15 a	4.42±0.01 a	9.43±0.19 a	13.22±0.13 a	0.15±0.01 a	62.88± 0.03 b	25.02±0.09 a
	TL-C	363.17±1.15 a	4.55±0.01 a	6.92±0.19 b	12.66±0.13 b	0.12±0.01 a	60.92± 0.03 b	25.92±0.09 a
	P	*<0.001*	*<0.001*	*<0.001*	*<0.001*	*0.034*	*<0.001*	*<0.001*
Greek honey		437.00±2.28	4.75±0.05	24.13±0.25	14.62±0,28	0.16±0.02	58.05±0.18	33.94±0.49
Manna honey		440.67±1.51	4.56±0.06	23.62±0.12	14.10±0.17	0.17±0.08	59.26±0.07	36.07±0.33

Means with the same letter in each column are not significantly different at a 5% level, according to Duncan`s Multiple Range Test.
P values for GO influence significance were presented according to the One-Way Analysis of Variance (at a significance level of α=.05).
S = sunflower honey, A = acacia honey, M = multifloral honey. AG-C = Arges-Costesti, AG-G = Arges-Gliganu, AG-MO = Arges-Mosoaia, AG-MZ= Arges-Mozaceni, AG-V=Arges-Vedea, GR-B = Giurgiu-Bolintin, TL-C=Tulcea-Casimcea

Table 2. Total phenolic (TPC), flavonoid (TFC) and tannin content (TTC), and antioxidant activity (DPPH I%) depending on the geographical origin (GO) of the honey

Honey	GO	TPC (mg GAE/100 g)	TTC (mg GAE/100 g)	TFC (mg EC/100 g)	DPPH (I%)
S	AG-C	186.71±0.01 a	73.99±0.01 a	17.70±0.05 a	27.71±0.02 a
	AG-G	198.47±0.01 a	84.11±0.01 a	20.30±0.07 a	28.62±0.02 a
	P	*<0.001*	*<0.001*	*<0.001*	*<0.001*
A	AG-C	118.69±0.01 a	77.62±0.01 a	12.16±0.05 a	13.36±0.02 c
	AG-MO	81.41±0.01 b	62.13±0.01 a	12.16±0.07 a	19.21±0.02 b
	AG-V	70.45±0.01 b	48.45±0.01 a	11.18±0.07 a	23.13±0.02 a
	P	*<0.001*	*<0.001*	*<0.001*	*<0.001*
M	AG-MZ	140.51±0.01 a	50.05±0.01 a	20.05±0.06 a	26.49±0.02 a
	GR-B	134.05±0.01 a	43.65±0.01 a	19.55±0.05 a	25.48±0.02 a
	TL-C	130.15±0.01 b	40.25±0.01 a	14.73±0.04 b	23.41±0.02 a
	P	*<0.001*	*<0.001*	*<0.001*	*<0.001*
Greek honey		102.39±1.05	28.72±1.03	12.93±0.24	18.14±0.71
Manna honey		116.49±0.02	37.41±0.02	12.34±0.32	21.62±0.04

The antioxidant properties of honey are mostly caused by the phenolic compounds produced under abiotic and biotic stress conditions. The results from Table 2 showed that sunflower honey had the highest phenolic, tannin, and flavonoid content, whereas acacia, Greek and Manna honey presented the lowest phenolic, and flavonoid content. Our results showed that all analysed honey samples had corresponding values of the quality indicators, in accordance to the EU legislation and the Codex Alimentarius standard (Codex Alimentarius, 2001; Council Directive 2001/110/EC; Directive 2014/63/EU).

The FTIR Spectroscopy is used in combination with multivariate analysis to detect adulterated honey and assess honey quality. The honey samples spectra were analyzed in the present

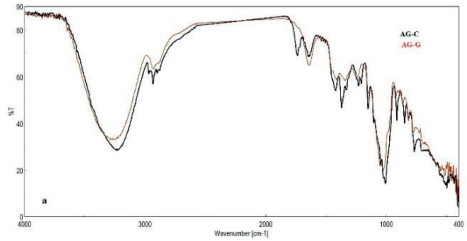
study in two regions: 1800–650 cm^{-1} and 3000–2800 cm^{-1}. According to Sivakesava and Irudayaraj (2001), the ideal spectral range for distinguishing between pure honey and honey contaminated with simple sugars (glucose, fructose, and sucrose) is between 1800 and 750 cm^{-1}. The fingerprint spectral is region of 1500–800 cm^{-1} and is mainly due to the absorptions of monosaccharides and disaccharides present in the honey (Limm et al., 2003; Damto et al., 2023; Vîjan et al., 2023).

The bands at 1474–1199 cm^{-1} are responsible for bending of O–C–H, C–C–H and C–O–H (Tewari & Irudayaraj, 2004). According to Gok et al., 2015, the anomeric region was a peak between 950 and 750 cm^{-1} and was often preferred for the spectral analysis of carbohydrates. The area that stands out in the assessment and description of honey is the band from 890 to 810 cm^{-1} typical for the C-H deformation or vibration anomeric region of carbohydrates (Tul'chinsky et al., 1976; Damto et al., 2023).

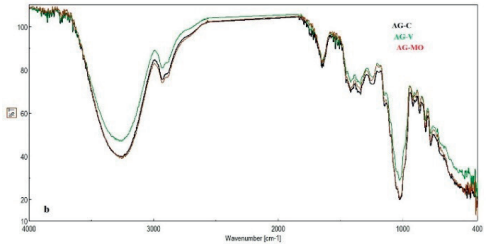
Figure 1 and Table 3 show the ATR-FTIR spectra of the tested honey with major high bands. Generally, one noted some differences in the FTIR spectrum analysis of the honey samples.

Table 3. ATR-FTIR Assignments for honey samples

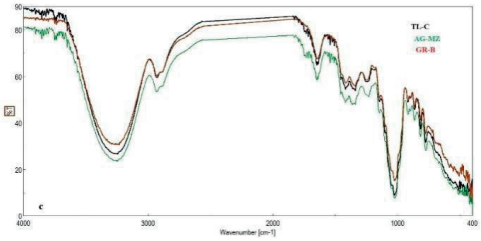
Honey	Sunflower (S)		Acacia (A)			Multifloral (M)			Greek	Manna
Harvest zone	AG-C	AG-G	AG-C	AG-MO	AG-V	TL-C	AG-MZ	GR-B	from trade	
ν(C-H) tretching of carboxylic acids + ν(NH_3) of free aminoacids	2935	2933	2930	2930	2935	2930	2933	2930	2931	2931
CH_3 sym stretch	2881	2887	2888	2892	2892	2883	2879	2883	2884	2883
C=O stretch in unconjugated ketones, carbonyls in ester groups (frequently of carbohydrate origin)	1736	1744	1743	1732	1747	1733	1743	1741		1743
δ(O-H) from H_2O	1646	1644	1646	1646	1646	1646	1646	1645	1645	1645
δ(O-H) in C-OH group + δ(C-H) in the alkenes	1420	1415	1418	1418	1418	1418	1416	1417	1417	1417
Stretching C-O, deformation C-H, deformation N-H	1373	1373	1357	1339	1360	1373	1362	1340	1339	1341
ν(C–H) + ν(C–O) in carbohydrates	1231 1152	1244 1148	1243 1145	1252 1145	1242 1145	1243 1146	1232 1146	1244 1146	1266 1105	1254 1100
ν(C-O) in C-OH group + ν(C-C) in carbohydrates	1046 1008	1049 1028	1051 1024	1048 1024	1049 1025	1046 1024	1048 1024	1044 1024	1046 988	1048 1023
δ(C–H)	915	917	917	917	916	917	916	917	923	918
Ring vibrations (mainly from carbohydrates)	850	862	863	864	863	863	862	863	859	863
-C–H bending (mainly from carbohydrates)	814	816	816	816	815	816	816	816	831	817
Anomeric region of carbohydrates	768	776	776	775	776	775	774	776	767	776



(a)



(b)



(c)

Figure 1. ATR-FTIR spectra of honey samples with different botanical origins: sunflower (a), acacia (b), and multifloral (c)

The spectra of natural honey and commercial honey have similar characteristics and spectral overlaps but differ in the wavelength of the characteristic peaks (Figure 2 and Table 3). However, the Greek honey sample's spectra showed a shift to a longer wavelength and a

broadening of the absorption bands at the range 950–810 cm^{-1}, typical for the C-H deformation or vibration anomeric region of carbohydrates (Figure 2).

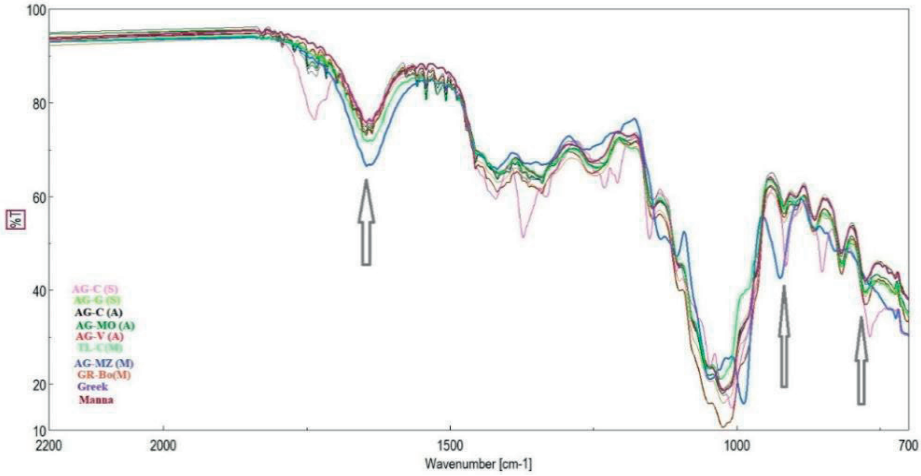
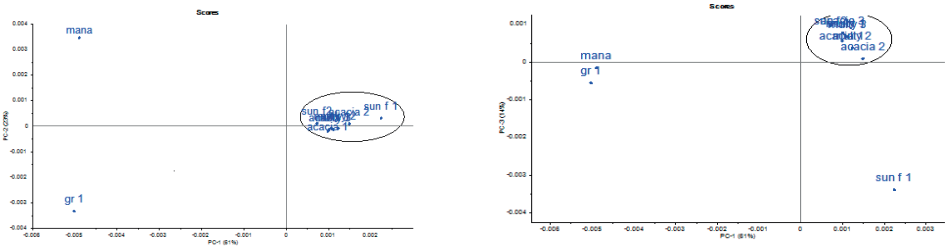


Figure 2. FT-IR spectra of honey samples with the indication of spectral changes for commercial honey (Greek and Manna honey)

From these results it is possible to differentiate two groups of honey, commercial (probably adulterated) and nonadulterated honey. To make these results clearer, PCA was applied to FTIR spectra of all groups, obtaining evident discrimination.

A clear splitting of the data can be observed as depicted in Figure 3 by the first two principal components in the 4000–650 cm^{-1} region. The first three principal components (PCs) for the honey samples under consideration account for 98% of the total variance (PC1=61%, PC2=23%, and PC3=14%). This suggests that the three components were sufficient to provide a good separation between the groups. Commercial honey (Greek and Manna honey) is separated from sunflower, acacia, and multifloral honey obtained from primary honey producers.



(a) (b)

Figure 3. 2-D scores obtained from PCA of FTIR spectra of honey for the first two PCs (a), and PC3 versus PC1 (b)

Hierarchical cluster analysis (HCA) involves a measurement of the similarity between objects about to be clustered and samples with the maximum similarities were clustered preferentially (Yi et al., 2013). Thus, it was possible to separate two groups, adulterated and nonadulterated honey. The spectral area from 1800–650 cm^{-1} was selected for successful discrimination of clusters (Damto et al., 2023). The results obtained are represented in Figure 4 in the form of dendrogram. The clusters determination for the analysed honey samples indicates a clear separation in two distinct categories, a category of commercial and other non-adulterated honey.

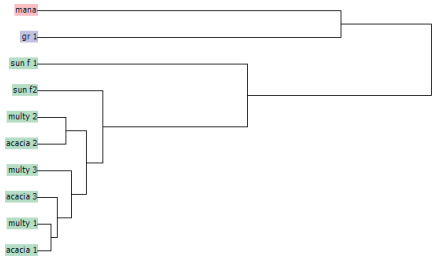


Figure 4. Hierarchical clustering of all samples in the 1800–650 cm^{-1} (fingerprint) spectral region

CONCLUSIONS

The physicochemical parameters values of the ten analyzed honey samples indicate that all samples are conform to the European Legislation. However, the levels of physico-chemical and biochemical parameters of honey samples fluctuated significantly depending on the geographical origin of honey. Thus, sunflower honey was noted for its high content of phenolic compounds, flavonoids and tannins, whereas acacia, Greek and Manna honey presented the lowest phenolic, and flavonoid content.

The adulterated honey was identified by the use of multivariate analysis from FTIR spectrum screening. The Greek and Manna honey samples spectra showed a shift to a longer wavelength and a broadening of the absorption bands as the concentration of sample increases. The bands in the spectral region 1800–650 cm^{-1} were selected for successful discrimination of clusters. Our results suggest that the two commercial honey (Greek and Manna honey) were probably adulterated.

ACKNOWLEDGEMENTS

This research work was carried out with the support of the Romanian Ministry of Research, Innovation and Digitalization and also was financed from "Henri Coandă" program, contract number 4/16.01.2024.

REFERENCES

Albu, A., Simeanu, C., Pop, I.M., Pui, A., Tarcău, D., & Cucu-Man, S.M. (2024). Selected characteristics of multifloral honeys from North-Eastern Romania. *Agriculture*, *14*, 26.

Al-Ghamdi, A.A., & Ansari, M.J. (2021). Biological and therapeutic roles of Saudi Arabian honey: A comparative review. *Journal of King Saud University - Science*, *33(2)*, 101329.

Alvarez-Suarez, J.M., Tulipani, S., Romandini, S., Bertoli, E., & Battino, M. (2010). Contribution of honey in nutrition and human health: A review. *Mediterrean Journal of Nutrition and Metabolism*, *3*, 15–23.

Bodó, A., Radványi, L., K″oszegi, T., Csepregi, R., Nagy, D.U., Farkas, Á., & Kocsis, M. (2021). Quality evaluation of light- and dark-colored Hungarian honeys, focusing on botanical origin, antioxidant capacity and mineral content. *Molecules*, *26*, 2825.

Bogdanov, S. (2009). Harmonized methods of the International Honey Commission. Available online: http://www.ihc-platform.net/ihcmethods2009.pdf (accessed on 10 March 2023).

Castro-Vázquez, L., Díaz-Maroto, M.C., González-Viñas, M.A., de la Fuente, E., & Pérez-Coello, M.S. (2008). Influence of storage conditions on chemical composition and sensory properties of citrus honey. *Journal of Agricultural and Food Chemistry*, *56*, 1999–2006.

Codex Alimentarius (2001). Draft Revised Standard for Standard for Honey (at Step 10 of the Codex Procedure). *Alinorm*, *1*, 19–26.

Coteanu, I., Seche, L., & Seche, M. (2016). DEX. Dicționarul explicativ al limbii române, Editura Univers Enciclopedic, 1376.

Council Directive 2001/110/EC of 20 December 2001 relating to honey. (2002). *Official Journal of the European Communities*, L10/47-L10/52.

Damto, T., Zewdu, A., & Birhanu, T. (2023). Application of Fourier transform infrared (FT-IR) spectroscopy and multivariate analysis for detection of adulteration in honey markets in Ethiopia, *Current Research in Food Science*, *7*, 100565.

Directive 2014/63/EU of the European Parliament and of the Council amending Council Directive 2001/110/EC relating to honey. (2014). *Official Journal of the European Communities*, *57*, L164.

Dubois, M., Giles, K.A., Hamilton, J.K., Rebers, P.A., & Smith, F. (1956). Colorimetric method for determination of sugars and related substances. *Analytical Chemistry*, *28(3)*, 350–356.

Eteraf-Oskouei, T., & Najafi, M. (2013). Traditional and modern uses of natural honey in human diseases: a review. *Iranian Journal of Basic Medical Sciences*, *6(6)*, 731–742.

Giosanu, D., Topală, C., Vîjan, L., Rădulescu, A., & Mazilu, I. (2022). Application of spectroscopy in analysis of some floral honey samples collected from four south Romanian counties in 2021. *Current Trends in Natural Sciences*, *11(22)*, 162–171.

Gok, S., Severcan, M., Goormaghtigh, E., Kandemir, I., & Severcan, F. (2015). Differentiation of Anatolian honey samples from different botanical origins by ATR-FTIR spectroscopy using multivariate analysis. *Food Chemistry*, *170*, 234–240.

Hills, S.P., Mitchell, P., Wells, C., & Russell, M. (2019). Honey supplementation and exercise: A systematic review. *Nutrients*, *11*, 1586.

Limm, W., Karunathilaka, S.R., & Mossoba, M.M. (2023). Fourier transform infrared spectroscopy and chemometrics for the rapid screening of economically motivated adulteration of honey spiked with corn or rice syrup. *Journal of Food Protection, 86(4)*, 100054.

Pérez, R.A., Iglesias, M.T., Pueyo, E., Gonzalez, M., & de Lorenzo, C. (2007). Amino acid composition and antioxidant capacity of Spanish honeys. *Journal of Agricultural and Food Chemistry*, *55*, 360–365.

Sivakesava, S., & Irudayaraj, J. (2001). Prediction of inverted cane sugar adulteration of honey by Fourier Transform Infrared Spectroscopy. *Journal of Food Science, 66(7)*, 972–978.

Tewari, J., & Irudayaraj, J. (2004). Quantification of saccharides in multiple floral honeys using transform infrared micro attenuated total reflectance spectroscopy. *Journal of Agricultural and Food Chemistry*, *52*, 3237–3243.

Topală, C.M., & Tătaru, L.D. (2019). ATR-FTIR spectroscopy coupled with chemical and chemometric analysis to distinguish between some sweet wines. *Revista de Chimie*, *70(7)*, 2355–2361.

Topală, C.M., Vîjan, L.E., Giura, S., & Botu, M. (2020). Attenuated total reflection Fourier transform infrared (ATR-FTIR): a method for the biochemical study of walnut leaves. *Current Trends in Natural Sciences*, *9(17)*, 266–272.

Tul'chinsky, V.M., Zurabyan, S.E., Asankozhoev, K.A., Kogan, G.A., & Khorlin, A.Y. (1976). Study of the infrared spectra of oligosaccharides in the region 1,000-40 cm^{-1}. *Carbohydrate Research*, *51(1)*, 1–8.

Vîjan, L.E., Mazilu, I.C., Enache, C., Enache, S., & Topală, C.M. (2023). Botanical origin influence on some honey physicochemical characteristics and antioxidant properties. *Foods*, *12*, 2134.

Yi, T., Chen, Q., He, X., So, S., Lo, Y., Fan, L., Xu, J., Tang, Y., Zhang, J., Zhao, Z., & Chen, H. (2013). Chemical quantification and antioxidant assay of four active components in *Ficus hirta* root using UPLC-PAD-MS fingerprinting combined with cluster analysis. *Chemistry Central Journal*, *7(1)*, 115.