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# Mineral content and botanical origin of Spanish honeys

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#### **Abstract**

Eleven elements (Zn, P, B, Mn, Mg, Cu, Ca, Ba, Sr, Na and K) were determined by inductively plasma coupled spectrometry in 40 honey samples from different places of Spain and four different botanical origins: Eucalyptus (*Eucalyptus sp.*), Heather (*Erica sp.*), Orange-blossom (*Citrus sinensis*) and Rosemary (Rosmarinus officinalis). K, Ca and P show the higher levels with average concentrations ranged between 434.1–1935 mg kg<sup>-1</sup> for K; 42.59–341.0 mg kg<sup>-1</sup> for Ca and 51.17–154.3 mg kg<sup>-1</sup> for P. Levels of Cu (0.531–2.117 mg kg<sup>-1</sup>), Ba (0.106–1.264 mg kg<sup>-1</sup>) and Sr (0.257–1.462 mg kg<sup>-1</sup>) are the lowest in all honey samples. Zn (1.332–7.825 mg kg<sup>-1</sup>), Mn (0.133–9.471 mg kg<sup>-1</sup>), Mg (13.26–74.38 mg kg<sup>-1</sup>) and Na (11.69–218.5 mg kg<sup>-1</sup>) concentrations were found strongly dependent on the kind of botanical origin.

Results were submitted to pattern recognition procedures, unsupervised methods such as cluster and principal components analysis and supervised learning methods like linear discriminant analysis in order to evaluate the existence of data patterns and the possibility of differentiation of Spanish honeys from different botanical origins according to their mineral content. Cluster analysis shows four clusters corresponding to the four botanical origins of honey and PCA explained 71% of the variance with the first two PC variables. The best-grouped honeys were those from heather; eucalyptus honeys formed a more dispersed group and finally orange-blossom and rosemary honeys formed a less distinguishable group.

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Keywords: Honey; Metals; Chemometrics; Pattern recognition; Eucalyptus (Eucalyptus sp.); Orange-blossom (Citrus sinensis); Rosemary (Rosmarinus officinalis); Heather (Erica sp.)

## 1. Introduction

Honey is produced by bees from nectar of plants, as well as from honeydew. Bees and plants are the sources of some honey components as: carbohydrates, water, traces of organic acids, enzymes, aminoacids, pigments; and others like pollen and wax arise during honey maturation.

Honey composition depends on great extent on the nectar sources. The monosacharides, glucose and fructose are

the main components of honey, mineral content ranges from about 0.04% in pale honeys to 0.2% in some dark honeys and protein content of honey is usually lower than 0.5% [1].

The Commission of European Union has adopted a new Council Directive, 2001/110/ECC, which repeals the Directive 74/409/ECC [2]. The new Directive establishes the types of honeys that can be marketed in the European Union and gives general definitions related to honey, including general and specific honey compositional characteristics such as, hydroximethyl furfural content, humidity, enzymatic activities and pesticide levels in honey, but those parameters have not been found to have a real relationship to geographical or botanical origin of honey. Pollen recognition has been the traditional method to determine the botanical origin of honey, but this technique is tedious and has limitations:

Abbreviations: ICP-AES, inductively coupled plasma-atomic emission spectroscopy; NW, North-western; PR, Pattern recognition; CA, Cluster analysis; PCA, Principal components analysis; LDA, Linear discriminant analysis; DF, discriminant function; PC, principal component

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counting procedure, identification and interpretation are difficult and requires trained analysts. Other studies [1] have sought analytical markers, such as aroma compounds, sugar profile, flavonoid pattern, non-flavonoid phenolics, isotopic relations, protein or amino acid content to honey classification.

Pattern recognition techniques have been widely applied to food chemistry in recent years [3–6]. The subject of food authenticity covers a wide range of products, and it is of a great economic and social importance, for sectors involved in food production and consumers. Authenticity helps to guarantee the characteristics and quality of food products and helps to prevent overpayment due to adulteration or consumers mislead as a result of ambiguous, improper product labelling. Honey dilution with water and addition of sugars and syrups are the main honey adulteration methods. Besides, in recent years, there has been a rising presence of monofloral honeys in markets, more expensive than multi-floral ones, so possible adulteration by honey mixtures must be controlled.

Different procedures for the determination of honey mineral content have been proposed in some previous papers [7–27]; some of them with classification purposes [24–26], but always according to geographical origin and none of them established a classification of honey according to its botanical origin. Stein [9] determined Cd, Pb and Mn in honey samples, in different seasons of the year, using atomic absorption furnace and three different methods, they obtained identical results by the three methods; Serra [11] determined the levels of Ca, Mg, K, Na, Fe, Cu, Cr and Pb in Spanish eucalyptus honey; Sevimli et al. [12] determined As, Cr, Sb, K, Br, Zn, Fe and Co in Turkish honey by neutron activation analysis; Fodor and Molnar [13] analysed seven honey samples produced in industrial areas to investigate environmental contamination, Al, B, Ca, Cd, Cu, Fe, K, Mg, Mn, Na, P, Pb and Zn were determined; Rodriguez Otero et al. [14] determined Na, K, Ca, Mg, Cu, Fe, Mn and P in Galician honeys; Viñas et al. [16] determined Pb, Zn, Cu and Cd in honey samples from different type and origin by electrothermal atomic absorption spectrometry; Latorre et al. [25,26] determined 11 metals in honeys from Galicia with classification purposes, they differentiate between honeys from Galicia and non-Galician ones, and processed and industrially commercialised Galician honeys using pattern recognition methods. Despite the fact that some authors correlate the mineral content of honey only with its geographical origin [1], the aim of this paper is, first to contribute to the scarce data about mineral content of Spanish honeys and secondly establish whether mineral content can explain botanical origin.

## 2. Materials and methods

## 2.1. Apparatus

Elemental analysis was carried out on a Fisons-ARL 3410 inductively coupled plasma-atomic emission spectrometer

Table 1
Operating parameters for ICP-OES

RF frequency	27.12 MHz			
Operating power	650 W			
Coolant argon flow rate	$7.51\mathrm{min}^{-1}$			
Plasma/nebuliser argon flow rate	$0.81  \mathrm{min}^{-1}$			
Burner type	Minitorch			
Nebuliser type	Meinhard			
Viewing height	8 mm			
Sample flow rate	$2.3\mathrm{mlmin^{-1}}$			
Metals	Emision wavelength (nm)			
Zn	213.868			
P	214.914			
В	249.796			
Mn	257.610			
Mg	279.553			
Cu	324.772			
Ca	393.369			
Sr	407.771			
Ba	455.424			
Na	589.003			
K	766.473			

(Fisons Instruments, Valencia, CA). Table 1 shows the analytical lines for each element, as well as the instrumental conditions.

## 2.2. Reagents and solutions

All chemicals were of analytical-reagent grade or better. Standard solutions were prepared by adequate dilution of a multi-element standard (1000 mg l<sup>-1</sup>) obtained from Merck (Darmstadt, Germany). Nitric acid 65% and perchloric acid 85% were from Merck. All solutions and dilutions were prepared with ultrapure water (Milli-Q, Milipore, Bedford, MA).

## 2.3. Honey samples

Forty samples of honey were analysed. The samples were obtained from stores and cooperatives, representing the most common types of honey readily available to consumers in Spain, i.e. eucalyptus (Eucalyptus sp.), orangeblossom (Citrus sinensis), rosemary (Rosmarinus officinalis) and heather (Erica sp.). A palinological study of the honey samples was performed in order to guarantee the labelled botanical origin [28]. An identification code was assigned to each group of samples, E for eucalyptus, OB for orangeblossom, R for rosemary and H for heather ones. All the samples from the same botanical origin are from different geographic origin to avoid formation of natural grouping between samples due to the same geographic origin. In addition, samples from the same geographic origin but different botanical class have been analysed to guarantee classification is due to botanical origin. Table 2 shows the origin of the analysed samples.

Table 2 Studied honey samples

Sample	Geographical origin	Botanical origin		
E1	Huelva	Eucalyptus		
E2	Madrid	Eucalyptus		
E3	Badajoz	Eucalyptus		
E4	Sevilla	Eucalyptus		
E5	Salamanca	Eucalyptus		
E6	Cadiz	Eucalyptus		
E7	Caceres	Eucalyptus		
E8	Ciudad Real	Eucalyptus		
E9	Vizcaya	Eucalyptus		
E10	La Coruña	Eucalyptus		
H1	Huelva	Heather		
H2	Caceres	Heather		
Н3	Avila	Heather		
H4	Galicia	Heather		
H5	Asturias	Heather		
Н6	Cadiz	Heather		
H7	Guadalajara	Heather		
Н8	Palencia	Heather		
H9	Badajoz	Heather		
H10	Castellon	Heather		
OB1	Huelva	Orange-blossom		
OB2	Murcia	Orange-blossom		
OB3	Castellon	Orange-blossom		
OB4	Valencia	Orange-blossom		
OB5	Alicante.	Orange-blossom		
OB6	Tarragona	Orange-blossom		
OB7	Sevilla	Orange-blossom		
OB8	Malaga	Orange-blossom		
OB9	Cordoba	Orange-blossom		
OB10	Almeria	Orange-blossom		
R1	Huelva	Rosemary		
R2	Avila	Rosemary		
R3	Caceres	Rosemary		
R4	Cuenca	Rosemary		
R5	Guadalajara	Rosemary		
R6	Badajoz	Rosemary		
R7	Cadiz	Rosemary		
R8	Granada	Rosemary		
R9	Salamanca	Rosemary		
R10	Segovia	Rosemary		

### 2.4. Analytical determinations

Eleven metals (Zn, P, B, Mn, Mg, Cu, Sr, Ca, Ba, Na and K) were determined in each honey sample. 1 g of honey was treated with 10 ml of concentrated nitric acid, in open PTFE vessels, and heating until nearly dry. This procedure was repeated with 15 ml of a 2:1 (HNO<sub>3</sub>/HClO<sub>4</sub>) mixture until complete mineralization [29]. The residue was dissolved, at room temperature, in 1 ml of concentrated nitric acid, transferred to a 25 ml volumetric flask and diluted with milli-Q water. Samples were measured by ICP-AES according to the parameters shown in Table 1. Blank solutions were prepared under identical conditions and the average signal was subtracted from analytical signals of honey samples.

Recovery assays and standard addition procedures with known spiked samples were carried out in order to assure analytical data with adequate precision and accuracy; good results were obtained.

The statistical package STATISTICA version 6.0 (2001) for windows, from Statsoft was used for all the chemometric calculations.

### 3. Results and discussion

## 3.1. Mineral content

Metal content of the studied honeys is shown in Table 3. Heather and eucalyptus, darker honeys, have a higher mineral content than the rest of the analysed honeys. This result is consistent with Ankalm [1] that concludes that dark honeys have a higher mineral content than pale ones. In general, K, Ca and P show the highest levels with average concentrations ranged between  $639-1845 \text{ mg kg}^{-1}$ ;  $111-257 \text{ mg kg}^{-1}$  and  $63.8-143 \,\mathrm{mg \, kg^{-1}}$ , respectively. Latorre et al. [25] obtained similar values for K in Spanish honeys (botanical origin not specified). Eucalyptus honeys from Spain were analysed by Serra [11] showing similar levels for Ca, but slightly lower levels of K and Cu than in the present study. K and Ca levels also agree with those reported by Packer and Giné [21] in eucalyptus honey. Cu, Ba and Sr are the elements with the lowest concentrations in all samples. Cu has been widely analysed in previous papers [8,11,13,14-22,24,25], with levels ranging from 0.057 to  $2,200\,\mathrm{mg\,kg^{-1}}$  in honeys from different botanical and geographical origin; the values obtained in the present study are comprise in that range. Zn, Mn, Mg and Na concentrations are strongly dependent on the honey botanical origin. Results obtained for Zn  $(1.332-7.825 \text{ mg kg}^{-1})$ , Mn  $(0.133-9.471 \text{ mg kg}^{-1})$ , Mg  $(13.26-74.38 \text{ mg kg}^{-1})$  and Na (11.69–218.5 mg kg<sup>-1</sup>) agree with values found previously [19].

## 3.2. Data analysis

Using the mineral content found in the analysed honey samples as chemical descriptors, pattern recognition (PR) methods [30,31] (cluster analysis, principal components analysis and linear discriminant analysis) were applied to establish whether there are defined groups between eucalyptus, heather, orange-blossom and rosemary honey samples. A data matrix was constructed using the analysed elements as columns and the honey samples as rows. Cluster analysis was applied to the auto-scaled data, the Euclidean distance was used to calculate the sample similarities and a hierarchical agglomerative procedure was employed to establish clusters. The results obtained are shown as a dendogram in Fig. 1. Four clusters corresponding to each botanical origin were found at a linkage distance level of 4.6. From the left, the first cluster (A) is composed of rosemary honey samples; cluster (B) is composed of orange-blossom samples; cluster (C) is formed by eucalyptus samples and cluster (D) by heather honey samples.

Table 3 Honey samples mineral content  $(mg kg^{-1})^a$ 

	Zn	P	В	Mn	Mg	Cu	Ca	Sr	Ba	Na	K
H1	3.211±1.053	154.3±1.191	6.988±0.169	4.539±0.721	61.34±6.815	0.809±0.048	263.0±20.95	0.271±0.068	0.225±0.051	97.66±8.35	1771±21.0
Н3	3.101±0.943	144.2±1.081	$6.878\pm0.059$	4.429±0.611	$55.84\pm6.705$	$0.699\pm0.062$	$252.0\pm20.84$	$0.260\pm0.042$	$0.214\pm0.042$	86.66±7.65	1760±38.2
H10	$2.632 \pm 0.568$	142.6±5.821	5.150±0.665	$3.485\pm0.936$	66.26±2.152	2.117±0.126	201.6±27.97	$0.354 \pm 0.036$	$0.215 \pm 0.065$	47.09±7.860	1845±42.2
H7	$2.422 \pm 0.358$	121.6±5.611	$4.940\pm0.455$	$3.275\pm0.726$	61.16±1.942	$1.907 \pm 0.084$	195.6±27.76	$0.333 \pm 0.174$	$0.194\pm0.004$	48.90±5.761	1824±41.9
Н6	2.275±0.430	130.8±3.185	2.962±0.471	2.438±0.295	71.18±4.673	$0.554\pm0.097$	326.0±31.72	0.461±0.016	$0.217 \pm 0.032$	63.60±5.957	1920±21.9
H4	$2.425 \pm 0.580$	145.8±3.335	$3.112\pm0.621$	$2.588\pm0.445$	$74.38 \pm 4.823$	$0.704\pm0.247$	341.0±31.87	$0.476\pm0.166$	$0.232\pm0.010$	$78.60\pm6.107$	1935±22.0
H8	2.921±0.290	$148.5\pm5.884$	6.069±0.919	4.012±0.527	63.80±2.459	1.463±0.654	232.3±30.71	0.313±0.042	$0.220\pm0.005$	72.38±9.87	1808±36.8
H9	2.871±0.240	143.5±5.834	6.019±0.869	3.962±0.477	61.80±2.409	1.413±0.200	227.3±30.66	$0.308\pm0.008$	0.215±0.045	67.38±6.21	1803±36.8
H5	$2.454\pm0.178$	136.7±5.884	4.056±1.094	2.961±0.524	$68.72\pm2.460$	$1.336\pm0.781$	263.8±26.95	$0.408 \pm 0.053$	$0.216\pm0.001$	55.34±8.255	1882±37.3
H2	2.554±0.278	146.9±5.984	4.156±1.194	3.061±0.624	73.22±2.560	1.436±0.090	273.8±30.95	$0.418\pm0.153$	$0.226\pm0.011$	65.34±7.900	1892±27.8
E1	5.438±1.037	113.0±0.048	7.065±0.935	7.427±0.300	36.61±5.447	$0.578\pm0.057$	184.4±22.28	$0.408\pm0.013$	$0.444 \pm 0.001$	168.6±1.689	1094±19.4
E4	5.327±0.927	$101.9\pm0.062$	4.967±0.825	7.316±0.190	35.51±5.337	$0.542\pm0.046$	173.3±12.08	0.597±0.008	0.555±0.010	157.5±1.579	1083±18.3
E2	5.821±0.979	140.3±0.942	3.163±1.272	6.600±0.408	47.64±2.970	$0.645\pm0.091$	177.7±10.75	$0.823 \pm 0.058$	0.493±0.174	116.2±1.820	1425±20.7
E5	5.611±0.769	119.3±0.732	2.953±1.062	6.390±0.198	45.54±2.760	$0.533\pm0.070$	156.7±4.897	$0.613\pm0.052$	0.393±0.036	137.2±1.610	1404±18.6
E3	$7.672\pm1.017$	96.61±2.098	2.314±0.862	9.318±0.195	36.88±1.505	0.517±0.003	269.3±11.93	$0.635\pm0.048$	$0.522\pm0.099$	218.5±9.639	1217±13.7
E8	7.825±1.167	111.9±2.248	2.467±1.012	9.471±0.345	38.38±1.655	$0.670\pm0.018$	254.0±12.08	$0.788 \pm 0.025$	0.675±0.114	203.2±4.095	1232±17.8
E6	5.426±0.816	116.0±1.662	4.842±0.612	7.950±0.547	41.50±1.627	$0.636\pm0.088$	203.0±11.22	1.175±0.011	$0.380\pm0.018$	150.0±1.724	1581±30.7
E9	5.375±0.766	111.0±1.612	4.792±0.562	7.900±0.497	41.00±1.577	$0.586\pm0.083$	198.0±11.17	1.125±0.039	$0.330\pm0.032$	145.0±1.674	1576±25.9
E7	$6.089\pm0.927$	116.5±0.777	4.346±1.814	6.324±0.201	40.65±4.477	$0.594\pm0.052$	208.6±6.35	$0.760\pm0.034$	$0.360\pm0.072$	197.0±3.995	1329±21.7
E10	6.194±1.027	127.0±0.877	4.451±1.914	$6.429\pm0.145$	41.70±4.577	$0.699\pm0.062$	219.1±5.575	$0.865\pm0.040$	$0.465 \pm 0.082$	186.5±8.987	1340±22.8
O1	5.297±0.617	$76.72 \pm 0.477$	8.120±1.042	0.456±0.127	19.22±2.779	< 0.531	42.70±4.512	0.691±0.132	< 0.106	43.44±7.124	618.9±8.31
O6	3.943±0.200	75.61±0.367	8.01±0.93	$0.35\pm0.02$	18.11±2.67	< 0.531	42.59±4.40	$0.58\pm0.02$	< 0.106	42.34±2.09	608.8±8.98
О3	4.054±0.286	53.27±0.458	6.926±0.725	0.133±0.006	15.35±1.947	< 0.531	51.52±4.842	$0.665\pm0.038$	< 0.106	11.69±0.960	455.0±7.67
O7	3.845±0.076	51.17±0.248	6.716±0.515	0.200±0.059	13.26±1.737	< 0.531	49.43±4.632	0.456±0.012	< 0.106	23.59±1.501	434.1±7.46
O2	2.950±0.021	57.64±2.191	5.419±1.133	4.268±0.056	42.34±1.482	< 0.531	80.70±2.021	$0.573\pm0.075$	1.113±0.124	69.37±8.379	839.5±7.37
O8	3.101±0.171	59.15±2.341	5.154±1.283	$0.276\pm0.071$	23.85±1.632	< 0.531	82.21±2.171	$0.724\pm0.225$	1.264±0.089	70.88±8.529	854.6±7.52
O4	3.268±0.739	63.19±4.466	6.506±1.345	$0.286\pm0.058$	20.86±1.059	< 0.531	59.2±4.02	1.462±0.028	< 0.106	39.39±8.762	634.9±6.19
O5	3.217±0.689	58.09±4.416	6.454±1.295	$0.234\pm0.008$	20.36±1.009	< 0.531	54.0±3.09	1.410±0.022	< 0.106	38.87±8.710	629.7±5.67
09	4.272±0.345	68.32±4.362	5.723±0.412	0.491±0.076	19.41±1.268	0.548	89.6±3.102	0.916±0.046	< 0.106	27.41±5.485	647.3±21.3
O10	4.375±0.445	69.35±4.462	5.826±0.512	$0.388 \pm 0.060$	20.44±1.368	< 0.531	90.9±3.202	$1.019\pm0.146$	< 0.106	28.44±5.588	657.6±5.98
R1	$1.441 \pm 0.007$	77.04±2.027	6.657±0.374	$0.586\pm0.042$	19.70±3.013	$0.636\pm0.057$	152.9±17.91	0.257±0.023	0.122±0.023	33.19±10.84	494.0±19.5
R9	$1.332\pm0.006$	75.95±1.918	6.548±0.265	$0.695\pm0.031$	20.79±2.904	< 0.531	142.0±16.90	0.366±0.012	$0.111\pm0.012$	32.10±5.897	483.1±15.7
R2	$3.254\pm0.624$	78.39±1.768	5.340±2.132	0.525±0.119	20.97±0.929	< 0.531	206.7±28.93	$0.480\pm0.076$	$0.175\pm0.076$	22.05±4.931	630.9±18.2
R3	3.057±0.427	76.42±1.571	5.143±1.935	$0.722\pm0.099$	22.94±0.732	$0.693\pm0.025$	187.0±28.73	$0.283 \pm 0.056$	0.155±0.056	20.08±4.734	611.2±16.0
R7	$3.864\pm0.151$	87.91±2.016	2.646±0.896	$1.374\pm0.164$	33.46±2.281	$0.574\pm0.043$	128.3±20.12	$0.386\pm0.049$	$0.155\pm0.047$	16.17±0.157	872.1±13.0
R8	4.008±0.295	89.35±2.160	5.790±1.040	1.230±0.179	34.90±2.425	< 0.531	142.7±20.98	0.530±0.193	$0.170\pm0.061$	16.31±0.301	886.5±10.0
R4	4.496±0.584	105.8±1.376	8.456±1.984	$0.819\pm0.143$	37.82±1.221	< 0.531	140.4±28.59	$0.531 \pm 0.032$	$0.044\pm0.049$	36.80±0.962	754.2±8.46
R6	4.436±0.524	105.2±1.316	8.396±1.924	$0.759\pm0.083$	37.22±1.161	< 0.531	134.4±28.53	$0.471 \pm 0.026$	$0.138\pm0.043$	36.20±0.902	748.2±8.97
R5	3.263±0.140	87.29±11.49	5.774±2.118	0.826±0.335	26.99±1.578	$0.550\pm0.060$	121.0±20.03	0.413±0.104	$0.124\pm0.050$	27.05±8.308	687.8±13.2
R10	$3.362 \pm 0.239$	$88.28 \pm 1.267$	$5.873 \pm 2.217$	$0.925 \pm 0.434$	$27.98 \pm 1.964$	$0.649 \pm 0.069$	130.9±18.90	$0.512 \pm 0.203$	$0.134 \pm 0.060$	$28.04 \pm 7.045$	697.7±8.45
LD	0.032	0.957	0.160	0.032	0.003	0.160	0.006	0.003	0.032	1.596	15.960
LC	0.106	3.192	0.532	0.106	0.011	0.532	0.021	0.011	0.106	5.320	53.19

<sup>&</sup>lt;sup>a</sup> Average of three determinations  $\pm$  S.D.

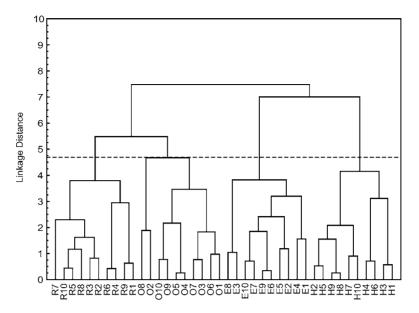


Fig. 1. Dendogram of cluster analysis. (Category codes: E, eucalyptus; H, heather; OB, orange-blossom; R, rosemary).

Principal components analysis was performed on the autoscaled data. From the loadings of original variables in the two first considered principal components (Table 4), P, Mg, Ca and K are the dominant variables in the first principal component (PC1) that represents 43.48% of the total variance, while Ba, Sr, Cu and Zn show the lower values. The dominant variables appear at negative values of PC1, all being very close. From this fact, it could be inferred that they provide the same kind of information. Principal component two (PC2) explains up to 28.11% of the total variance. Examining the loadings, Zn results the most dominant variable in this PC. The first two principal components account for 71.59% of the total variance and were considered to be sufficient for such data.

Examining the score plot of the objects in the space defined by the two principal components considered, Fig. 2, a natural separation of the four honey groups of different botanical origin was found. On the bottom left side of the biplot a compact group composed of heather honey samples can be observed. Most of the eucalyptus honeys appear at the top left side as a more dispersed group, rosemary honeys forms a

Table 4 Loadings of the variables for the two first PC's (principal components)

Variable	PC1	PC2
Zn	-0.220	0.843
P	-0.902	-0.289
В	0.617	-0.307
Mn	-0.768	0.551
Mg	-0.873	-0.401
Cu	-0.202	-0.651
Ca	-0.896	-0.180
Sr	0.223	0.625
Ba	-0.022	0.601
Na	-0.657	0.688
K	-0.929	-0.236

compact group on the bottom right side, and a more dispersed group of orange-blossom honeys appear on the top right side of the biplot. Despite rosemary and orange-blossom samples appear very close in the biplot, it could be considered they formed two different groups, rosmary honeys are on the negative side of the biplot and orange-blossom on the positive side.

Principal components analysis shows that metal content could provide enough information to develop a botanical classification for the studied honey. In addition, the agreement between the two unsupervised techniques used, PCA and cluster analysis, is consistent with this idea.

Unsupervised techniques only have visualising capabilities, so a supervised pattern recognition method such as linear discriminant analysis (LDA) was performed with the purpose of obtaining classification rules for assigning categories to samples. After applying LDA, three discriminant functions

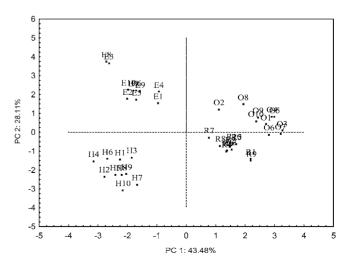


Fig. 2. Principal components analysis (PC1 vs. PC2).

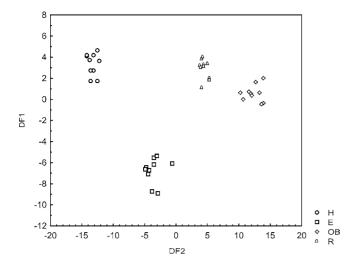


Fig. 3. Linear discriminant analysis (DF<sub>1</sub> vs. DF<sub>2</sub>).

were calculated. Fig. 3 shows the scatter plot of honey samples in the plane defined by the discriminant functions  $DF_1$  and  $DF_2$ . The four types of honeys appear completely separated in the plot. The samples corresponding to heather honey are situated as a compact group at the top left side of the plot. At the right side appears the rosemary and orange-blossom honeys forming two clearly defined groups and at the lower side of the plot forming a more dispersed group, it appears the eucalyptus honey samples.

Validation was performed using a cross-validation method with  $\approx\!25\%$  of samples as the evaluation set. The same process was repeated several times with different compositions of both training and evaluation sets, ensuring that all samples were included in the evaluation set at least once. Recognition ability, calculated as the percentage of members of the training set that were correctly classified, and prediction ability, calculated as the percentage of members of the evaluation set correctly classified were 100 and 97%, respectively.

Using LDA analysis a highly successful classification between the four honey groups studied was achieve, so, the determination of the mineral content of monofloral honeys and the later chemometric study can be an useful alternative to the usual pollen studies to obtain botanical classification.

#### 4. Conclusions

In this work, an attempt to establish a classification of some Spanish honeys from different botanical and geographical origin was made using pattern recognition techniques. Until now, few attemps have been made on honey from different botanical origin classification attending its mineral content. PCA and cluster analysis show that metal contents could provide enough information to develop a classification between the four botanical origins studied. LDA was applied to ob-

tained classification rules and it was validated by means of cross-validation, obtaining good recognition and prediction abilities, 100 and 97%, respectively. The discriminant model obtained could save time and money in the determination of the botanical origin of honey.

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