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A preliminary palynological characterization of Spanish thyme honeys

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Pollen was analysed in 25 thyme honey samples from Spain. The honey samples were provided by professional beekeepers, and pollen was obtained by centrifugation. A minimum of 8% of *Thymus* sp. pollen is considered sufficient to typify a honey as unifloral when considering pollen grains from anemophilous and nectarless plants. The quantitative analysis showed that nectar is the main honey source in the samples studied. The qualitative analysis of the samples revealed the presence of 53 taxa belonging to 27 families. The Spanish thyme honeys of the studied region are characterized by their medium to high numbers of grains of pollen (NGP; mean = 155 000) and their medium to high content of honeydew indicator elements (HDE; mean = 8300). *Genista* type (present in 100% of the samples), *Echium plantagineum* and *Reseda luteola* (90%), *Eucalyptus* type (80%), *Calendula* type and *Olea europaea* (75%), and *Cistus monspeliensis* (70%) may be mentioned among the characteristic accompanying species of this honey type. © 2004 The Linnean Society of London, *Botanical Journal of the Linnean Society*, 2004, **146**, 323–330.

ADDITIONAL KEYWORDS: melissopalynology – pollen – *Thymus*.

INTRODUCTION

Honey is the nectar and sugar exudation of plants gathered, modified and stored in honey combs by honeybees. The composition and properties of honeys vary, mainly because of the floral sources utilized by the bees. The characterization of honeys was initiated by the analysis of their pollen content, the objective being to differentiate between exotic honeys and European types produced in Germany and Switzerland (Barbier, 1958; Louveaux & Vergeron, 1964). Pollen spectra revealed appreciable differences among the honeys. Usually, a honey is considered to be produced mainly from one plant (unifloral) if the pollen frequency of that plant is >45%. Some pollen grains such as *Citrus*, *Tilia*, *Robinia* and members of the Lamiaceae, e.g. *Lavandula*, *Rosmarinus officinalis*, *Salvia* and *Thymus*, are under-represented in the honey spectra, and 10–30% of these pollen types are considered sufficient to indicate a honey as unifloral. For over-represented pollen types (*Eucalyptus*, *Castanea sativa*, *Cynoglossum officinale* and *Myosotis*), a honey is regarded as unifloral only when the predominant pollen type represents more than 90% of the total (Maurizio, 1979).

In Spain and Mediterranean countries in general, certain unifloral honeys have been the subject of numerous studies on their pollen and physicochemical properties. They include eucalyptus, orange, sunflower, lavender, heather, rosemary and honeydew honeys among others (Kirkwood, Mitchell & Smith, 1960; Louveaux & Abed, 1984; Accorti *et al*., 1986; Serra-Bonvehí, Gomez-Pajuelo & Gonell-Galindo, 1987; Serra-Bonvehí, 1988; Pérez-Arquillué *et al*., 1994, 1995; Serra-Bonvehí & Ventura-Coll, 1995; Roselló-Caselles *et al*., 1996; Mateo & Bosch-Reig, 1998; Andrade *et al*., 1999; Terrab, Díez & Heredia, 2003a, 2003b).

Although the Lamiaceae is, along with the Leguminosae and Rosaceae, one of the botanical groups of highest interest from an apicultural point of view (Ricciardelli-D'Albore & Persano-Oddo, 1981), certain unifloral honeys such as the thyme ones have been poorly studied in Spain. This is surprising given the great amounts of thyme honey produced and con-

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sumed across the country, notably in Castellón, Granada and Málaga. Most studies characterizing this unifloral honey have been from other countries in the Mediterranean basin (Italy, Greece and Morocco) (Accorti *et al*., 1986; Damblon, 1987, 1988; Persano *et al*., 1995; Thrasyvoulou & Manikis, 1995; Tsigouri & Passaloglou-Katrali, 2000).

Thyme honey is prized by consumers for its golden/dark amber colour, strongly aromatic scent and its slow rate of granulation (Sáenz-Laín & Gómez-Ferreras, 2000), and also attracts scientific interest. Its geographical typification would be of considerable international importance. Thus, the goal of this work is to characterize Spanish thyme honeys and to establish geographical markers that may help to distinguish honeys of this region from those of different geographical origin.

MATERIAL AND METHODS

We analysed pollen in 25 samples of thyme honey selected from 60 Spanish honeys collected in 2002. These honeys show the following characteristics: golden/dark amber colour; the soft odour of thyme flowers; persistent flavour; fine and coarse crystallization. A minimum of 8% pollen from *Thymus* sp. was required to designate these honeys as unifloral from a melissopalynological point of view.

The samples were collected from professional beekeepers; the honey was extracted by centrifugation. The collection sites are listed in the Appendix and shown in Figure 1.

Quantitative analysis

Four slides of each honey sample covering the whole surface of each slide were prepared following Maurizio (1979), without any chemical treatment, and analysed using light microscopy (LM). Counts were made for the numbers of pollen grains (NPG) and honeydew elements (HDE: fungal spores and mycelium) on each slide.

Qualitative analysis

Acetolyzed slides were prepared following Erdtman (1960), slightly modified, and always using a subsample of 10 g of honey. Following the results obtained by Behm, VonderOhe & Henrich (1996), at least 500 pollen grains were counted on four different slides for each honey sample. The different pollen types were identified mainly using a key (Díez, 1987), plus reference slides from the collection at the Department of Botany, University of Seville. When possible, identification was made to a specific level (e.g. *Ridolfia segetum*), but otherwise to a generic level (e.g. *Eucalyptus* type) or to the pollen type (e.g. *Echium plantagineum* type). The pollen types are named according to Persano-Oddo & Ricciardelli-D'Albore (1989). Field observations were made while collecting the samples and, in these cases, the most likely species are quoted

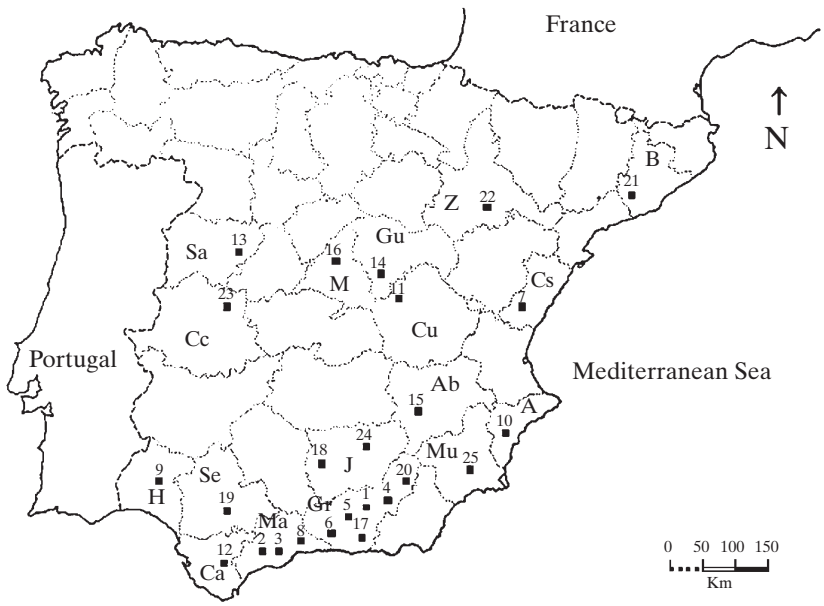

Figure 1. Distribution of the honey samples studied. A, Almería province; Ab, Albacete province; B, Barcelona province; Ca, Cádiz province; Cc, Cáceres province; Cs, Castellón province; Cu, Cuenca province; Gr, Granada province; Gu, Guadalajara province; H, Huelva province; J, Jaén province; M, Madrid province; Ma, Málaga province; Mu, Murcia province; Sa, Salamanca province; Z, Zaragoza province.

in brackets after the pollen type [e.g. *Ononis* type (*Ononis natrix*)].The information on pollen grains from anemophilous and nectarless plants in this study was authenticated by the authors in the field or recorded from literature (Crane, 1979; Herrera, 1985; Talavera *et al*., 1988; Ricciardelli-D'Albore, 1998; López *et al*., 1999; Rodríguez, Ortega & Devesa, 1999).

RESULTS AND DISCUSSION

The results from the quantitative analysis are summarized in Table 1. One sample was rich in pollen grains (Class IV) (Maurizio, 1979), 23 showed a medium content (Classes II and III) and the remaining sample was quite poor in pollen sediments.

The NPG in 10 g of honey ranges between 9500 (sample 25) and 640 000 (sample 21). Although all the samples show the presence of HDE, generally speaking they are sparse, ranging from 1300 (sample 25) to 24 800 (sample 21).

Table 1. Summarized results of the quantitative analysis

Sample	NPG	HDE	BEN	Class*
1	64 600	6 300	70 900	II
2	376 000	14 400	390 000	III
3	127 800	6 400	134 200	III
4	66 100	10 200	76 300	II
5	26 260	4 400	30 660	II
6	488 000	3 200	491 200	III
7	71 800	6 000	77 800	II
8	222 133	14 800	236 933	III
9	149 334	9 666	159 000	III
10	111 360	3 120	114 480	III
11	28 233	2 867	31 100	II
12	84 000	2 800	86 800	II
13	273 200	11 200	284 400	II
14	27 400	3 866	31 266	II
15	158 061	4 561	162 622	III
16	138 586	24 198	162 784	III
17	99 733	9 400	109 133	III
18	68 826	5 534	74 420	II
19	108 200	10 200	118 400	III
20	279 400	5 600	285 000	III
21	640 000	24 800	664 800	IV
22	73 300	14 100	87 400	II
23	44 000	4 400	48 400	II
24	155 733	5 400	161 133	III
25	9 500	1 300	10 800	I

NPG, number of pollen grains; HDE, number of honeydew indicator elements; BEN, number of botanical elements (NGP + HDE). *I, honeys < 20 000 pollen counted grains per 10 g; II, honeys ranging between 20 000 and 100 000 pollen counted grains per 10 g; III, honeys ranging between 100 000 and 500 000 pollen counted grains per 10 g; IV, honeys ranging between 500 000 and 1000 000 pollen counted grains per 10 g.

The results from the qualitative analysis are shown in Table 2, where the pollen types detected and their corresponding percentages on the different samples are shown. We identified 53 pollen types from 27 families.

The best-represented families are Asteraceae (eight types), Cistaceae and Fabaceae (five types) and Brassicaceae (four types). The most frequent families found are: Cistaceae, Fabaceae and Lamiaceae (present in 100% of the samples), Asteraceae (95%), Boraginaceae and Resedaceae (90%), Myrtaceae (80%), Oleaceae and Scrophulariaceae (75%), Brassicaceae (70%), Fagaceae (55%), and Apiaceae and Salicaceae (50%).

The only pollen type present in all the samples along with *Thymus* sp. is *Genista* type (from 1% to 43%). The other pollen types were represented as follows; figures are number of samples in which they are present followed by range of percentages out of total pollen in sample: *Echium plantagineum* type and *Reseda luteola* type, 22 samples, >1–41%; *Eucalyptus*, 20 samples, <1–78%; *Calendula* type and *Olea europaea*, 19 samples, <1–43%; *Cistus monspeliensis*, 18 samples, <1–17%; *Kickxia* type and *Senecio* type, 17 samples, <1–34% and <1–6%, respectively; *Ononis* type, 15 samples, very low percentages (≤ 7%); *Quercus* type, 14 samples, <1–24%; *Centaurea calcitrapa* type and *Pimpinella anisum*, 13 samples, <1–8%; *Halimium* type, 12 samples, 14% (in sample 7); *Trifolium arvensis* type, 11 samples, <1–2%; *Raphanus* type and *Salix triandra* type, 10 samples, <1–28% and <3%, respectively; *Capsella* type, *Lythrum salicaria* type and *Papaver* type, 9 samples; *Helianthemum* type, 8 samples, <1–6%; *Ceratonia siliqua*, *Crepis* type, *Hyosciamus albus* gr., *Muscari* type, Poaceae (> 37 μm) and *Teucrium scorodonia* type, 7 samples, <1–5%; *Cistus salvifolius*, *Anthemis* type, *Campanula erinus* type and *Erica* type, 6 samples, <1–27%; *Onobrychis* type, *Plantago* type, *Solanum nigrum* and *Verbascum* type, 5 samples, <1–5%; *Lactuca* type, 4 samples, very low percentages (< 2%); *Anthyllis cytisoides* type, *Reseda media* type, *Ridolfia segetum*, *Salix fragilis* type and *Silene vulgaris* type, 3 samples, <1–23%; *Carthamus* type, *Polygala vulgaris*, *Rhamnus alaternus* type and *Xanthium* type, 2 samples; *Asparagus acutifolius* type, *Cucumis* type, *Cynoglossum cheirifolium*, *Ecbalium elaterium*, *Sinapis* type and *Thymelaea* type, 1 sample, <1–4%.

The thyme honeys of Spain mainly come from *Thymus mastichina* (L.) L. and *T. capitatus* (L.) Hoffmanns. & Link [= *Thymbra capitata* (L.) Cav.]. They are species with a wide nectar production but with quite low pollen grain content (2300 pollen grains/flower) (Ortiz, 1989), and show a moderately long flowering period of April–September (Valdés, Talavera & Fernandez-Galiano, 1987). As it can be seen in Table 1, the per-

Table 2. Results of the qualitative analysis of pollen types in Spanish thyme honeys, represented as percentages

Pollen type	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22	23	24	25
Apiaceae																									
Pimpinella villosa	1	4	3	–	3	–	–	–	+	+	–	5	–	2	–	–	–	–	+	+	1	2	–	5	–
Ridolfia segetum	–	–	1	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	+	1	–	–	–	–
Asteraceae																									
Anthemis type	–	–	+	+	–	–	–	1	–	–	–	+	–	–	–	–	–	+	–	–	–	+	–	–	–
Calendula type (*Calendula arvensis*)	6	12	9	3	1	–	2	1	2	–	–	8	2	–	1	+	2	–	24	5	2	2	1	8	–
Carthamus type	–	–	–	+	–	–	–	–	–	–	–	–	–	–	+	–	–	–	–	–	–	–	–	–	–
Centaurea calcitrapa type (*Centaurea* spp.)	–	–	–	–	2	–	+	–	–	+	1	+	1	–	8	–	2	–	–	+	+	+	+	–	+
Crepis type (*Andryala* spp., *Leontodon* spp.)	1	–	–	–	–	–	–	–	5	–	–	–	–	–	1	+	–	–	–	–	–	+	+	1	–
Lactuca type (*Cychorium inthybus*, *Scolymus hispanicus*, *S. maculatus*)	–	–	+	–	–	–	–	–	–	+	–	+	–	–	–	–	–	–	–	–	–	–	–	–	2
Senecio type (*Onopordum dissectum*, *Silybum marianum*)	1	3	+	–	6	1	+	–	–	–	1	2	–	–	1	1	3	4	1	2	+	2	–	–	1
Xanthium type	–	–	–	–	–	–	–	–	–	–	–	–	–	–	+	–	+	–	–	–	–	–	–	–	–
Boraginaceae																									
Cynoglossum cheirifolium	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	+	–	–	–
Echium plantagineum type (*E. plantagineum*)	1	23	2	–	–	4	5	1	7	37	–	7	27	5	1	30	+	+	1	1	9	2	41	17	1
Brassicaceae																									
Capsella type	1	9	–	–	–	–	5	3	–	–	–	–	–	–	–	1	–	–	–	–	3	2	+	1	–
Raphanus type	–	–	–	2	–	–	–	–	+	–	–	1	+	–	15	–	–	2	–	+	–	28	7	–	6
Sinapis type	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	1	–	–	–	–	–	–
Caesalpiniaceae																									
Ceratonia siliqua	–	–	–	–	–	–	1	–	–	–	–	–	–	–	–	–	+	1	1	1	–	–	+	–	1
Campanulaceae																									
Campanula erinus	1	–	–	–	–	–	–	–	27	–	1	–	2	–	–	+	–	–	–	–	–	–	1	–	–
Caryophyllaceae																									
Silene vulgaris type	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	+	–	+	+	–	–	–	–
Cistaceae																									
Cistus ladanifer	+	4	–	–	5	–	–	1	26	4	4	+	6	–	–	2	–	2	–	2	+	2	2	3	–
C. monspeliensis	17	3	–	–	–	7	–	1	1	–	3	–	–	+	1	2	16	5	1	6	2	4	2	10	5

C. salvifolius	+	–	2	–	12	–	–	–	–	–	12	–	–	–	–	–	–	–	–	–	1	–	+	–	–
Halimium type (*H. halimifolium*)	–	–	–	8	–	–	14	1	5	10	9	1	–	–	–	–	3	10	–	–	–	1	–	3	1
Helianthemum type	+	–	2	6	–	–	–	–	–	–	–	–	1	–	–	–	–	–	–	–	3	3	+	3	–
Cucurbitaceae																										
Ecbalium elaterium	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	+	–	–	–	–
Cucumis	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	+	–
Ericaceae																										
Erica type (*E. arborea*, *E. australis*)	–	–	–	–	–	–	–	–	–	–	–	–	7	–	+	–	–	+	–	–	+	1	–	1	–
Fabaceae																										
Anthyllis cytisoides type	–	–	–	–	–	–	15	–	–	–	–	1	1	–	–	–	–	–	–	–	–	–	–	–	–
Genista type (*Calicotome villosa*, *Cytisus baeticus*, *Genista hirsuta*, *Ulex borgiae*)	7	5	8	43	3	34	10	1	2	3	10	3	16	7	9	40	24	20	2	1	6	7	4	6	10
Onobrychis type (*O. peduncularis*)	–	+	–	–	–	–	4	–	–	4	–	–	–	–	–	–	–	–	–	–	–	5	–	–	+
Ononis type (*O. natrix. O. pubescens, O. repens*)	–	3	–	–	–	–	–	2	6	2	–	1	7	3	1	1	–	–	+	–	1	3	+	1	2
Trifolium arvensis type (*T. campestre*, *T. repens*)	+	–	–	–	–	–	–	2	–	2	–	–	1	–	+	–	–	–	+	+	1	1	2	1	–
Fagaceae																										
Quercus type (*Q. rotundifolia*, *Q. suber*)	7	–	–	10	–	–	1	1	–	–	1	+	–	–	1	3	6	24	–	–	–	+	8	5	3
Lamiaceae																										
Thymus type (*T. capitatus, T. mastichina*)	10	16	10	10	30	11	31	13	10	11	9	13	10	10	9	10	12	10	10	20	12	14	11	9	8
Teucrium scorodonia type (*T. capitatum, T. scordium*)	+	–	–	1	–	–	1	–	–	–	–	–	–	–	–	–	–	–	1	–	1	1	–	1	–
Liliaceae																										
Asparagus acutifolius type	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	4	–	–	–	–	–	–
Muscari type	–	–	–	–	2	–	–	–	–	–	–	2	1	–	–	–	1	–	–	2	1	–	–	+	–
Lythraceae																										
Lythrum salicaria type (*L. junceum*)	–	3	1	–	–	–	–	–	–	–	–	+	–	+	–	+	–	–	–	+	4	+	–	+	–

Table 2. *Continued*

Pollen type	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22	23	24	25
Myrtaceae																									
Eucalyptus type (*E. camaldulensis*, *E. gomphocephala*)	4	15	55	–	1	–	–	68	1	18	44	51	6	78	–	2	2	–	49	52	40	2	+	7	+
Oleaceae																									
Olea europaea	6	–	1	10	6	4	6	–	–	–	–	+	3	1	2	3	13	10	1	1	4	3	–	5	43
Papaveraceae																									
Papaver type	2	–	1	–	–	–	2	–	–	–	–	–	1	–	–	–	–	–	–	–	1	+	1	1	3
Plantaginaceae																									
Plantago type (*P. coronopus*)	+		–	–	–	–	1	–	–	–	–	–	–	–	–	–	–	–	–	+	–	2	+	–	–
Poaceae																									
Poacea > 37 μm	–	–	–	–	2	–	–	–	–	–	–	–	–	–	–	–	–	–	+	2	1	+	+	+	–
Polygalaceae																									
Polygala vulgaris	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	+	–	–	+
Resedaceae																									
Reseda luteola type (*R. lutea, R. luteola*)	7	–	2	6	4	3	5	2	5	6	1	1	2	–	22	–	14	7	1	1	4	3	8	5	4
Reseda media type	+	–	–	–	–	–	–	–	–	–	–	–	–	–	23	–	–	–	–	–	–	–	–	–	2
Rhamnaceae																									
Rhamnus alaternus	–	–	–	–	–	–	–	–	–	–	–	–	–	+	–	–	–	–	–	+	–	–	–	–	–
Salicaceae																									
Salix fragilis	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	1	1	–	–	–	–	–	–	1
S. triandra	2	–	–	–	–	2	–	1	–	+	–	–	2	–	+	1	–	–	–	+	–	3	1	–	–
Scrophulariaceae																									
Kickxia type (*K. lanigera*)	17	–	1	–	15	34	–	1	3	–	3	–	2	–	1	3	–	1	1	1	1	4	1	–	3
Verbascum type (*V. sinuatum*)	3	–	–	–	–	–	–	–	–	–	–	+	–	–	–	–	–	–	–	–	–	+	2	3	–
Solanaceae																									
Hyosciamus albus	2	–	–	–	–	–	–	–	–	–	–	1	–	–	1	–	–	–	–	–	–	2	2	1	3
Solanum nigrum	1	–	–	–	–	–	–	–	–	–	–	–	–	–	1	–	–	–	–	–	–	+	2	1	–
Thymelaeaceae																									
Thymelaea type (*Daphne gnidium*)	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	1	–	–

+, values below 1%; –, absence of the pollen type.

centage of *Thymus* type pollen ranges between 8% (sample 25) and 31% (sample 7); these are quite low percentages when compared with results from other authors such as Tsigouri & Passaloglou-Katrali (2000). In any case, these low percentages of the *Thymus* type pollen are greatly increased (ranging between 11% in sample 14 and 73% in sample 5) when the pollen grains from anemophilous and nectarless plants (*Calendula* type, *Centaurea calcitrapa* type, *Campanula erinus*, Cistaceae, *Genista* type, *Ononis* type, *Quercus* type, *Olea europaea*, *Papaver* type, *Plantago* type, *Pimpinella villosa*, Poaceae > 37 μm, *Reseda* spp., *Kickxia* type, *Senecio* type, *Verbascum* type) are excluded in the calculation of the percentages (Table 3).

This honey type is characterized by its high pollen content (mean NGP = 155 000), when compared with other honeys with the same floral origin (Accorti *et al*., 1986: NGP = 20 000); 48% of the honeys belong to Class III of Maurizio and 44% to Class II. They are also characterized by their high HDE content ($\overline{X}$ = 8300). The most characteristic accompanying species are *Genista* type, present in 100% of the samples, followed by *Echium plantagineum* type and *Reseda luteola* (90%), *Eucalyptus* type (80%), *Calendula* type and *Olea europaea* (75%), and *Cistus monspeliensis* (70%).

Table 3. Percentage of *Thymus* pollen in each sample after eliminating the pollen grains from anemophilous and nectarless plants

Sample	% *Thymus* pollen
1	42
2	24
3	14
4	71
5	73
6	65
7	53
8	15
9	42
10	15
11	17
12	22
13	18
14	11
15	31
16	22
17	71
18	59
19	14
20	25
21	16
22	23
23	16
24	22
25	35

The thyme honeys from Spain can be clearly differentiated from those of other producing areas such as Italy, which show *Rhus*, *Pistacia*, *Carthamus* and *Myrtus* as accompanying species. They are also clearly differentiated from thyme honeys of Turkey, where *Thymus* is accompanied by *Astrantia*, *Euphorbia* and *Punica*, and Cyprus, characterized by the presence of *Teucrium*, *Sophora*, *Hypericum* and *Myrtus* (Ricciardelli-D'Albore & Vorwohl, 1979). They can be differentiated from the Greek thyme honeys owing to the presence of *Rubus*, *Pistacia lentiscus*, *Salvia*, *Origanum* and *Satureja* in the latter (Tsigouri & Passaloglou-Katrali, 2000). Also, thyme honeys (*Thymus satureioides*) from the Great Atlas (South Morocco) can be differentiated from the Spanish ones by the presence of *Ononis natrix* and *Eryngium ilicifolium* (Damblon, 1988).

The only study previously published on this honey type in Spain was carried out by Roselló-Caselles *et al*. (1996). These authors studied 22 Lamiaceae honeys from the Valencia region (east Spain); seven of these samples were unifloral of thyme, with *Quercus*, *Genista*, Cistaceae and *Papaver*, among others, as accompanying taxa. This is in agreement with the data obtained for sample 7 in this study, which comes from the same geographical area (Valencia region).

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APPENDIX

COLLECTION SITES

1, Lanjarón (Granada);
2, Antequera (Málaga);
3, Antequera (Málaga);
4, Sierra Granada (Granada);
5, Temple (Granada);
6, Collaja de la Jitana (Granada);
7, Almazora (Castellón);
8, Competa (Málaga);
9, Aroche (Huelva);
10, Alcarria (Cuenca);
11, Alcarria (Cuenca);
12, Ronda (Cádiz);
13, Sierra de Frades (Salamanca);
14, Alcarria (Guadalajara);
15, Ossa de Montiel (Albacete);
16, La Peñota (Madrid);
17, Otívar (Granada);
18, Sierra Segura (Jaén);
19, Los Corrales (Sevilla);
20, Sierra Nevada (Granada);
21, Serra de Queralt (Barcelona);
22, Calatayud (Zaragoza);
23, La Mestas (Cáceres);
24, Sierra de Cazorla (Jaén);
25, Campillo (Murcia)