Original Scientific paper
10.7251/AGRENG2002144B
UDC 638.132(65)

STUDY OF MELLIFEROUS PLANTS VISITED BY THE WORKER HONEYBEE, *APIS MELLIFERA* INTERMISSA IN BOUMERDES, ALGERIA

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ABSTRACT

This work allows to establish a list of melliferous plants visited by the worker bee, *Apis mellifera intermissa* in Boumerdes through melissopalynological analysis. Eight samples of honey were collected in summer from different regions of Boumerdes province in Algeria: Mizrana (H1), Tizéruine (H2), Tagdempt (H3), Sherraba (H4), Baghlia (H5), Boudouaou (H6), Sidi Daoud (H7) and Si Mustapha (H8). For the analysis, we adopted the classical method of Louveaux *et al*., (1970). The results obtained showed that the melissopalynological analysis indicated the pollen grains of *Eucalyptus* were numerous as more than 45% of the pollen in six (6) of the eight samples of Boumerdes (H1, H2, H3, H4, H7 and H8). The pollen of *Hedysarum coronarium* was dominant in the sample H5 (67.60%). In seven samples honey recognized multifloral by beekeepers, *Eucalyptus* and *Citrus* taxa were respectively super represented and underrepresented in the honey H7 (87.46%) and H6 (19.80%). Thus, the number of taxa listed in the honeys studied varies from 14 to 19. The total number of pollen grains counted for the 8 samples varied from 646 to 3580.

Keywords: *Honey, melliferous plants, melissopalynology, Boumerdes province.*

INTRODUCTION

Honey contains pollens grains provided from foraging activity of honeybees that reflet the environmental location of beehives (Von Der Ohe *et al.*, 2004). The pollen is a natural source of protein for bees and it is necessary for the honeybee's development (Radev *et al*., 2018). The composition of pollen harvested by honeybees can vary greatly on the time of year, the botanical origin and the geographic region from which the pollen is obtained (Von Der Ohe *et al.,* 2004). The only surefire way to issue the "identity card" of a honey is to do its pollen

analysis, because honey contains countless grains of pollen (several million in a single kilogram of honey) and honeydew elements. Each honey carries within it the mark of its origin defined by the pollen of the flowers pollinated by the bees which harvested the nectar (Louveaux, 1985). Pollen analysis of honey or melissopalynology is a valuable diagnostic tool for the possibility to determine the botanical and geographical origin of honey (Von Der Ohe *et al*., 2004).

The importance of identified taxa in honeys provides an image of the plant environment in the bees' forage area (Louveaux, 1985). In the world, several studies are devoted to the study of botanical ressources through palynological analysis (Feller, 1979; Valencia-Barrera *et al*., 1994, Kaya *et al.* 2005; Sajwani *et al*., 2007; Ayansola & Davies 2012; Dukku, 2013; Iritie *et al*., 2014). In Algeria, this field is far from being explorable (Ouchemoukh *et al*., 2007; Chefrour *et al*., 2009; Benaziza-Bouchema & Schweitzer, 2010; Makhloufi *et al*., 2010).

The purpose of this work is to establish a list of melliferous plants visited by the worker bee, *Apis mellifera intermissa* in Boumerdes through melissopalynological analysis.

MATERIAL AND METHODS

Samples collection

Honeys samples from *Apis mellifera intermissa* are collected in summer from different regions of Boumerdes province (Algeria): Mizrana (H1), Tizéruine (H2), Tagdempt (H3), Sherraba (H4), Baghlia (H5), Isser (H 6), Sidi Daoud (H7) and Si Mustapha (H8). They were obtained by centrifugation and stored at 10 ° C until analysis.

Palynological analysis

For the palynological analysis of honey, we adopted the classical method described by the International Commission for Bee Botany ICBB (Louveaux *et al*., 1970). This method consists in dissolving 10 g of each honey sample in 100 ml of warm distilled water at 40 ° C. After centrifugation at 3000 rpm for 5 to 10 minutes and after decantation, the pellet is put on a slide covered with coverslip and analysed by using light microscope at the magnification 400 to 1000x .

Pollen determination level

According to Louveaux *et al.,* (1970), when detailed knowledge is lacking or when, for reasons of time, a finer determination has to be abandoned, the pollen can be attached to a larger group (form or type). These two terms are used to indicate all genera or species represented by the same morphological type.

Expression of results

According to Gadbin (1980), the counts were carried out until the percentages and the number of taxa stabilized. Identified taxa being grouped into "Pollen dominant" (more than 45% of the total pollen grains counted), "secondary pollen« (16-45%), «important minor pollen" (from 3 to 15%), "rare pollen» (less than 3%). The

identification of different pollen taxa is based on pollen reference collections and publications (Louveaux, 1977; Gadbin, 1980; Sawyer, 2010).

A honey is classified as unifloral (monofloral) if it contains pollen in quantities exceeding 45% on the remaining pollen identified. Honey containing a variety of pollen types, none of which reaches 45%, are classified as multifloral (Darrigol 1979; Sawyer, 2010). According to Sawyer (2010) and Corvucci *et al.*, (2015), these are general some exceptions depending on the type of pollen. Many pollen types are underrepresented (*Citrus* spp, *Tilia* spp, *Helianthus*, *Rosmarinus…*) or overrrepresented (*Casanea sativa*, *Eucalyptus* spp…). For instance, to characterize acacia honey as unifloral, *R.pseudoacacia* pollen must be over 15%, *Citrus* must have at least 10% of *Citrus* spp, 20 to 30% for *Tilia.* Von Der Ohe *et al*., (2004) recommended content over 83% for *Eucalyptus* spot be unifloral honey. Several pollen types that could not be identified were designed as unknowns.

RESULTS AND DISCUSSION

The pollen analysis improves our knowledge on the honey flora of the different sites studied and on the food resources available to bee *Apis mellifera intermissa* under the conditions of our country. The results obtained are shown in Table 1.

Table 1. Different types of pollen from Boumerdes honey samples (in percent)

Familly	Species	H1	H2	H3	H4	H5	H6	H7	H8
Apiaceae[1]	*Daucus carotta*	0.93	0.68	0.33	1.46	1.87	13.15	0.86	0.22
Asteraceae	*Chrysanthemum myconis*	1.4	-	-	0.69	2.06	-	-	-
	Taraxacum officinais	-	11.17	-	-	1.20	-	-	-
	Galactites tomentosa	3.75	7.86	0.13	0.12	1.68	02.32	0.36	0.02
	Helianthus annus	4.96	-	-	-	1.93	-	-	-
	Inula viscosa	0.6	-	0.08	0.54	0.46	-	0.5	0.05
Borraginaceae	*Borrago officinalis*	3.69	0.05	-	-	-	02.63	0.21	-
	Echium plantagineum	2.48	2.27	6.59	16.94	3.34	4.02	1.8	5.81
Caryophyllaceae	*Lycnis sp*	-	-	-	-	0.06	-	-	-
Convolvulaceae	*Convolvulus arvensis*	-	1	0.08	-	-	-	-	0.3
Cruciferae[2]	*Raphanus raphanistrum*	-	2.22	-	0.3	-	-	-	0.08
	Brassica napus	1.34	2.22	0.39	0.82		-	1.73	0.96
	Sinapis arvensis	1.54	1.16	0.08	-	-	-	-	-
Ericaceae	*Erica arborea*	0.2	-	0.02	0.69	-	2.32	-	-
Lamiaceae[3]	*Lavandula stoechas*	0.13	-	039	0.30	1.60	2.78	0.5	0.08
	Rosmarinus officinalis	0.2	0.2	-	0.17	-	-		-

Myrtaceae	*Eucalyptus sp*	49.36	58.04	73.07	64.21	11.69	04.02	87.46	81.81
Oxalidaceae	*Oxalis prescaprae*	3.02	0.47	0.61	0.56	-	1.85	0.57	0.2
Papillionaceae [4]	*Trifolium sp*	2.95	0.21	0.33	0.77	4.94	-	0.36	0.85
	Hedysarum coronarium	10.33	3.28	3.49	6.45	67.60	29.55	2.31	4.61
	Robinia pseudoacacia	1.81	1.48	0.15	0.34	0.53	04.95	0.07	0.58
Rosaceae	*Pyrus- Prunus* type	7.57	7.93	4.83	4.45	0.40	3.71	1.8	2.57
	Rubus type	-	-	-	-	0.26	2.01	-	-
Rutaceae	*Citrus sp*	2.88	0.1	0.75	0.47	-	19.80	0.66	1.58
Tiliaceae	*Tilia sp*	-	-	-	-	-	4.64	-	-

- : absent, H: Honey, 1. Apiaceae or Umbelliferae, 2. Cruciferae or Brassicaceae, 3. Lamiaceae or Labiateae, 4. Papillionaceae or Fabaceae

The exploitation of Table 1 shows that Mizrana honey (H1) is characterized by the dominance of *Eucalyptus* sp pollen (49.36%). Compared to the other samples of honeys studied, this pollen taxon represents the lowest percentage. H1 is characterized by the abundance of *Hedysarum coronaruim* (Sulla) and *Pyrus-Prunus* type. The microscopic analysis of Tizeruine honey (H2) reveals the dominance of Eucalyptus sp (58.04%). This honey is very characteristic of the abundance of *Taraxacum officinalis*, *Galactites tomentosa*, and *Pyrus- Prunus* type. The Sherraba region (H4) is characterized by the dominance of *Eucalyptus* sp (64,21%). This pollen taxon is associated with *Hedysarum coronarium* and *Pyrus-Prunus* type which reaching the level of important isolated pollen. *Echuim plantagineum* reaches the level of the secondary pollen. In samples H7, H8 and H3, the predominance of *Eucalyptus* reflects the important harvest on this species, which reaches, as pollen analysis shows, the highest percentage respectively 87.46%, 81.81% and 73.07%. In Tagdemt region (H3), *Echuim plantagineum*, *Pyrus Prunus* type and *Hedysarum coronarium* respectively reached 6.59%; 4.83% and 3.49%. Comparatively to others honey, *Hedysarum coronarium* reached the lower percentage (2.31%) in the sample of Sidi Daoud (H7). In later region (H8), *Eucalyptus* pollen was accompanied respectively with *Echuim plantagineum*, *Hedysarum coronarium* and *Pyrus -Prunus* type.

Boumerdes regions were characterized by another's pollen types. *Hedysarum coronarium* pollen (67.60 %) and *Citrus* (19.80%) were dominant respectively in samples of Baghlia (H5) and Boudouaou (H6). *Eucalyptus* (11.69%), *Trifolium* sp (4.94%), *Echium plantagineum* (3.34%) and *Chrysanthemum myconis* pollen grains (2.06%) were concentrated in the first sample. The *Citrus* honey (H6) contained *Robinia pseudoacacia* (4.95%), Eucalyptus (4.02%), *Echium plantagineum* (4.02%), *Pyrus -Prunus* type (3.71%) and *Rubus* type 2.01%.

Microscopic analysis reveals that the honeys generally contain similar pollen, but in different combinations and percentages so the pollen spectrum shows some pollen characteristic and exclusive of a region. For example, pollen of *Helianthus*

annus is present only in honey H1 and H5. *Convolvulus arvensis* is only found in honeys H2, H3 and H8 (Table 2). The pollen grains of *Lycnis* and *Tilia* were present respectively only in samples H5 and H6. The pollens of *Daucus carotta*, *Galactites tomentosa, Echuim plantagineum, Hedysarum coronarium, Robinia pseudoacacia* are ubiquitous in all the samples studied.

In the family Asteraceae, we noted 5 types of pollen namely *Chrysanthemum myconis*, *Taraxacum officinalis, Galactites tomentosa*, *Helianthus annus* and *Inula viscosae.* In the Borraginaceae family, only 2 types are identified (*Borrago officinalis* and *Echium plantagineum*). The Cruciferae has 3 taxa *Raphanus raphanistrum*, *Brassica napus* and *Sinapis arvensis*. Lamiaceae contain 2 pollen species *Lavandula stoechas* and *Rosmarinus officinalis*. As for Papillionaceae, the *Trifolium* sp, *Hedysarum coronarium* and *Robinia pseudoacacia* are noted. For Rosacea, two taxa were identified (*Pyrus -Prunus* type and *Rubus* type).

Thus, the number of taxa listed in the honeys studied varies from 14 to 19 (Table 1 and Figure 1). This number is far from real, especially for samples H6, H5 and H2 (Figure 2). Note that the total number of pollen grains counted for the 8 samples from the different regions of Boumerdes varied from 646 to 3580 (Figure 3).

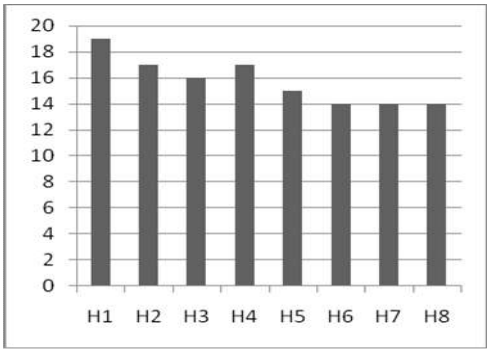

Figure 1 : Number of identified taxa pollen in the honey samples (H)

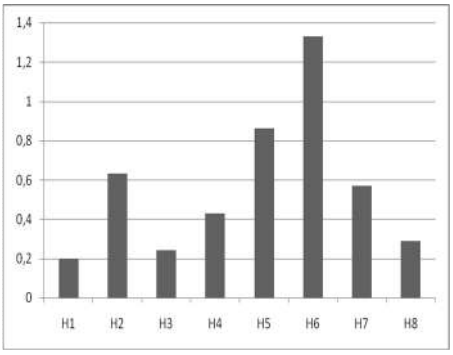

Figure 2 : Frequency of indeterminate honey samples(H)

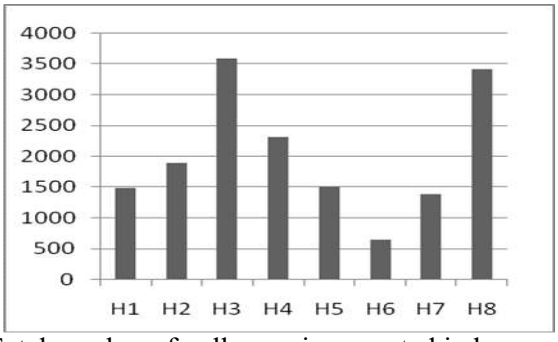

Figure 3: Total number of pollen grains counted in honey samples (H)

If we consider that families, Boumerdes regions are generally characterized by the richness of Myrtaceae, Papillionaceae, Borraginaceaes, Asteraceae and Rosaceae. Cruciferae, Rutaceas and Oxalidaceas are infrequent. The families of Apiaceae, Convulvulaceas, Ericaceaes and Lamiaceaes were found in trace amounts (Table 2 and Figure 4). There are 8 to 12 families in the different samples studied (Figure 5).

Table 2. Different Families of botanical resources visited by honeybees in Boumerdes honey samples (%)

Familly	H1	H2	H3	H4	H5	H6	H7	H8
Apiaceae	0.93	0.68	0.33	1.46	1.87	13.15	0.86	0.22
Asteraceae	10.46	19.03	0.11	1.35	7.33	2.32	0.86	0.07
Borraginaceae	6.17	2.32	6.59	16.94	3.34	6.65	2.01	3.81
Caryophyllaceae	-	-	-	-	0.06	-	-	-
Convolvulaceae	-	1	0,08	-	-	-	-	0.3
Cruciferae	2.88	5.60	0.47	112	-	-	1.73	1.04
Ericaceae	0.2	-	0.02	0.69	-	2.32	-	-
Lamiaceae	0.33	0.2	0.39	0.47	1.60	2.78	0.5	0.08
Myrtaceae	49.36	58.04	73.07	64.21	11.69	04.02	87.46	81.81
Oxalidaceae	3.02	0.47	0.61	0.56	-	1.85	0.57	0.2
Papillionaceae	9.11	5.44	4.58	8.12	73.07	36.35	3.31	6.34
Rosaceae	7.57	7.93	4.83	4.45	0.66	5.79	1.8	2.57
Rutaceae	2.88	0.1	0.75	0.47	-	19.80	0.66	1.58
Tiliaceae	-	-	-	-	-	4.64	-	-

- : absent

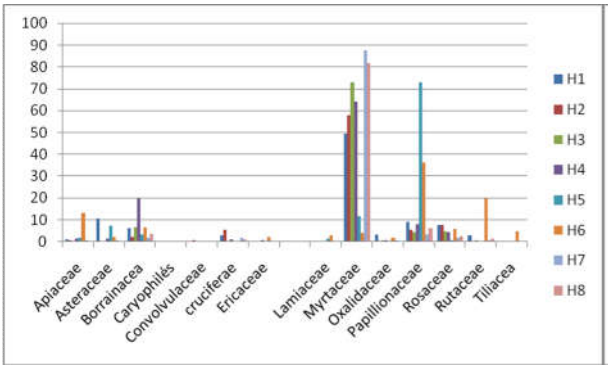

Figure 4: Occurrence of different families in Boumerdes honeys (H)

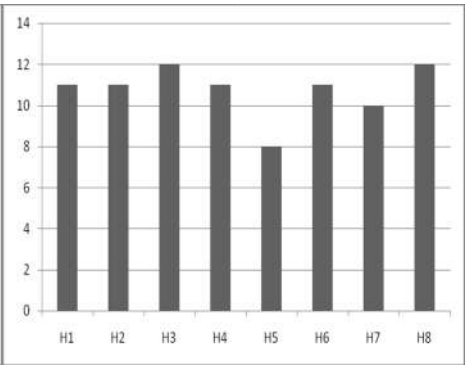

Figure 5: Number of families in each honey of Boumerdes

This work studies a number of plants that are sources of nectar and pollen for the honeybees (*Apis mellifera intermissa*) of Boumerdes regions. In seven samples honey recognized multifloral by beekeepers, Eucalyptus and *Citrus* taxa were respectively super represented and underrepresented in the honey H7 and H6. According to many researchers, the unifloral honey must contain a minimum of

45% of a single pollen type. Honey containing a variety of pollen types, none of which reaches 45%, are classified as multifloral (Darrigol 1979; Sawyer, 2010). According to Von Der Ohe *et al.,* (2004); Sawyer, (2010) and Corvucci *et al.*, (2015), these are general guidelines but many pollen types are underrepresented (*Citrus* sp) or overrrepresented (*Eucalyptus* spp). For instance, to characterize *Citrus* honey as unifloral, pollen must have at least 10% of *Citrus* sp, while for Eucalyptus honey, a content content over 83% is required to classify honey as unifloral. In the study, only the honey H7 and H6 respected this rule. Pollens taxa of *Eucalyptus* and *Citrus* reached respectively 87.46% and 19.80%. According to Sajwani *et al.,* (2007), *Eucalyptus* honey is a common type in Argentina and South Florida and California. Eucalyptus pollen is rare in honey of Oman. Louveaux and Abed (1984) have shown the importance of *Eucalyptus* in honey from the North Africa. This observation is confirmed by Ouchmoukh *et al.,* (2007); Chefrour *et al.* (2009); Benaziza-Bouchema and Schweitzer (2010) in Algerian honey. In a general way the secondary pollen present in these honeys is generally homogeneous. The pollens of *Citrus, Olea*, *Echium*, *Chamaerops*, Umbelliferae, are most often present in the Maghreb spectra. Botanical families such as Apiaceae and Ericaceae (Ouchemoukh *et al*., 2007), Papillionaceaes and Rosaceae (Chefrour *et a*l., 2009) are best represented respectively in honeys of northern and northeastern Algeria. According to Makhloufi *et al*., (2010), the accompanying pollen of *Eucalyptus* honey is *Echium plantagineum*, *Olea europaea*, *Trifolium* spp., *Papaver rhoeas*, *Centaurea* sp. and Apiaceae. Terrab *et al*., (2001), show that T*rifoluim*, *Citrus*, *Lythrum*, *Mentha* frequently appear in honeys from Northwest Morocco. Valencia-Barrera *et al*., (1994) noted in the province of Leon (Espagne), the most frequent families were Rosacea and Fabacea. Also, they reported in 27 honeys recognized multifloral honeys some pollen taxa were overrrepresented: *Lotus cornilatus*, *Trifoluim repens*, *Castanea sativa and Genista florida* type. Myrtaceae, Fabaceae, Asteracee, Proteaceae, Euphorbiacea, Fabaceae and Cistaceae were the best families represented respectively in Australian and Portuguese honey. Fabaceae, Asteraceae, Boraginacea, Cistacea and Scrophulriacea are the families with the highest quantity of pollen types in the samples from Spain (Seijo *et al*., 2003). The Asteracea, Fabaceae and Mimosacea are the most important families in Oman (Sajwani *et al*., 2007). In this work, *Eucalyptus* pollen is accompanied as previously mentioned by Papillionaceae, Myrtaceae and Rosaceaes. Valencia-Barrera *et al*., (1994) found in the province of Leon (Espagne), 22 types of pollen belonging to 5 families. A melissopalynological study of Turkys and Omani honeys, 122 pollen types representing 50 plants families and 86 taxa respectively were noted by Kaya *et al*., (2005) and by Sanjwni *et al*., (2007).

In our work, Boumerdes which characterises by the wide diversity of melliferous plants, these samples are also characterized by a low number of pollen types (14 to 19). Sajiwani *et al*., (2007) reported the low number of pollen types suggesting that the concentrated pollen of the dominant taxa provided enough nectar and pollen for honey production. Louveaux (1968) reports that various factors may be involved in determining the choice of forager. The plants that provide pollen and nectar at once

are preferred to others. Common plants existing in dense stands are also preferred. Localization and various ecological factors may also play an important role. This author also reports that the bee selected the rich pollens which bring him the maximum of the proteins. According to Rabiet (1984), bees do not choose the pollen they collect only according to its quality; the harvest of this material is generally subordinate to nectar sugar content and its abundance. According to Bertrand (1967), the bees harvest preferably those which contain 40 to 50% of sugar, and from one day to another they remember what time a flowers reaches its most favorable concentration.

CONCLUSIONS

Pollen forms varied in frequency and abundance probably due to an abundance of geographical distribution of the plant and the effect of environmental conditions on nectar production. For the identification of pollen types and the interpretation of pollen taxa, extensive experience, the establishment of the phytogeographic map and the pollen Atlas from our country are essential.

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