

**SPECTRUM OF POLLEN COLLECTED BY HONEYBEES IN BURSA
LOWLAND AREA IN HIGH SEASON**

Bursa Ovasında Bal Arılarının Yoğun Sezonda Topladıkları Polenlerin Yayılmımı

(Extended Abstract in Turkish can be found at the end of this article)

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Abstract: Pollen spectrum collected by honeybee foragers (*Apis mellifera anatoliaca*) was recorded during the main pollen flow in Bursa (N-W Anatolia) lowland region in order to prepare a calendar of honeybee used pollen loads, which will be useful for regional and itinerant beekeepers. Fifty one plant taxa were identified and eighteen of them had percentages higher than 1%. Dominant taxa were; Brassicaceae (13.00%), *Papaver* spp. (11.99%), *Paliurus spina-christi* (10.23%), Cistaceae (8.03%), *Ranunculus* spp. (7.29%) and Rosaceae (6.32%), while *Echium* spp. (4.34%), *Trifolium repens* (4.33%), *Quercus* spp. (4.33%), *Salix* spp. (4.29%), *Plantago* spp. (3.87%), *Vicia* spp. (3.48%), *Trifolium pratense* (3.15%), Asteraceae (2.35%), *Helianthus annuus* (2.15%) and Fabaceae (2.11%) were appeared with medium percentages. There is a strong reliance on some indigenous plant species for pollen foraging activity but a number of cultivars are also seen within the samples. The most diverse period for collecting various pollen types was June.

Keywords: Pollen, Pollen spectrum, Pollen calendar, *Apis mellifera anatoliaca*, Bursa

INTRODUCTION

The mutual relationship between honeybees and plants are a good example for ecological studies. Many flowering plants require the transfer of their pollen to the same or another flower of the same species for pollination and reproduction of new generations (Free 1993). In this respect honeybees are considered one of the most effective pollen vectors. Alternatively, plants need pollen and nectar from honeybees for their nutritional need (Winston 1987, Free 1993).

The decision to collect pollen by honeybee foragers depends on the number of larvae (brood), amount of stored pollen in the colony, as well as forager genotype and available resources in the environment (Pankiw et al. 1998). Besides pollen grains, the pollen pellets contain lipidic dyes from flower anthers. Several colours of pollen pellets, changing from white and cream to dark brown, presenting yellow, orange, red, greenish and grey

degrees, occur depending on the botanical taxa and the chemical composition of these substances (Stanley and Linskens 1974, Almeida-Muradian et al. 2005).

The beekeeping industry is expanding in Turkey but most of the traditional beekeepers are unaware of the vegetation that honey bee survival depends on. Melissopalynological studies have significant application in beekeeping industries. Analyses of pollen grains collected from honey bee colonies provide relevant information on the type of plants visited and also increase our knowledge of honeybee behaviour. Studies identifying plants used by honey bees as pollen or nectar sources have been used to develop floral calendars for beekeeping (e.g. Sharma 1970, Sorkun and Inceoglu 1984a-b-c, Thrasivoulou and Manikis 1995, Tsigouri et al. 2004, Webby 2004, Andrada

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and Tellería 2005, Bhusari et al. 2005, Terrab et al. 2005, Silici and Gokceoglu 2007).

The aim of this work was to provide data that can be used to create a floral calendar and to identify important pollen sources in the area of Mustafakemalpasa, Bursa. Such data are important to beekeepers and farmers so they can make informed decisions about the blooming periods of plants important to honeybees.

MATERIAL AND METHODS

Sampling was performed in Mustafakemalpasa-Bursa located in the northwest part of Turkey. The study area was situated $39^{\circ} 57' 32''$ N, $28^{\circ} 30' 28''$ E at an altitude of Ca. 50 m above sea level. Bursa is the fourth largest city in Turkey. It is located near the southwest part of the Marmara Sea at the northwest foot of Mount Uludag (2543 m). The region is drained by the Nilufer River and nearby is lake Uluabat. The climate is generally warm during most of the year consistent with the Mediterranean region. We selected this region because it is visited frequently by beekeepers in Anatolia. The region is a transitional zone as most of the plants which grow naturally belong to Mediterranean elements but there are also European-Siberian and Iran-Turanian elements. Consequently Mediterranean maquis elements are present in the region. However, whether naturally growing or planted, pine species such as *Pinus pinea* L., *Pinus brutia* Ten. and oak species such as *Quercus infectoria* Olivier, *Qercus robur* L. and *Quercus pubescens* Willd. populations also occur. Surrounding the study area are extensive agricultural areas with *Helianthus annuus* L., *Brassica napus* L., *Zea mays* L., *Punica granatum* L., *Morus nigra* L. *Morus alba* L., *Malus sylvestris* Miller, *Prunus domestica* L., *Persica vulgaris* Miller, *Amygdalus communis* L. and *Pyrus communis* L. under cultivation.

To obtain pollen loads, we used five colonies of *Apis mellifera anatoliaca* Maa placed in Langstroth-type hives. We collected pollen loads a short time after the first heavy pollen loads were brought to the hive by foragers and continued sampling until the main pollen flow decreased. We removed the accumulated pollen loads from the bottom pollen drawers of each hive every three days from April 10th until July 1st 2005. This period was selected based on previous research with *A. mellifera anatoliaca* (Bilisik et al. 2008). We collected 130 samples at +4°C until analysis. To identify the pollen and its botanical source, 500 pollen loads

were separated randomly and pollen loads were classified according to their colour (Kirk 1994). A piece of each pollen load of each colour was mixed with glycerine-jelly, and stained using basic fuchsin (Wodehouse 1935) and pollen grains were examined using light microscopy and compared with the reference slide collection of the Uludag University Botany department. From this data, the percentages of the each taxon of pollen grains were calculated.

RESULTS

A total of 51 different types of pollen grains were identified of which 18 had percentages higher than 1%. Only 0.22% of the total pollen loads failed to be identified (Table 1). Overall, 24 pollen types were identified to genus, 22 to family and 5 to species level. Dominant pollen types are; Brassicaceae (13.00%), *Papaver* spp. (11.99%), *Paliurus spinachristi* (10.23%), Cistaceae (8.03%), *Ranunculus* spp. (7.29%), Rosaceae (6.32%), *Echium* spp. (4.34%), *Trifolium repens* (4.33%), *Quercus* spp. (4.33%), *Salix* spp. (4.29%), *Plantago* spp. (3.87%), *Vicia* spp. (3.48%), *Trifolium pratense* (3.15%), Asteraceae (2.35%), *Helianthus annuus* (2.15%), Fabaceae (2.11%), Liliaceae (1.53%), *Thalictrum* spp. (1.25%). These represented 94.01% of the total.

The most common pollen types frequented by honeybees (>5%) were the Brassicaceae species. They are widespread in the region with the most common being *Brassica napus* L., *Brassica nigra* (L) Koch., *Sinapis arvensis* L., *Raphanus raphanistrum* L. Brassicaceae pollen percentages were variable during the sampling period which we believe was the result of the availability of pollen from the different species of this family. The maximum values of this type of pollen were obtained in June, especially during the 18th sampling period.

Papaver spp. was the second most important pollen loads collected. Honeybees collected *Papaver* spp. pollen for seven weeks. They gathered this type of pollen for nearly all the sampling period and maximum percentages can be seen in the first half of May (Figure 1). Maximum values of *Papaver* spp. pollen loads were collected during the 7th, 8th and 11th sampling periods (Table 1). Honeybee foragers collected *Paliurus spinachristi* pollen grains for one month; maximum values were between 14th-18th sampling periods (Figure 1). They represented 10.23% of all (Table 1).

Table 1: Pollen types which collected by honeybee foragers and their three-day variation with the percentages.

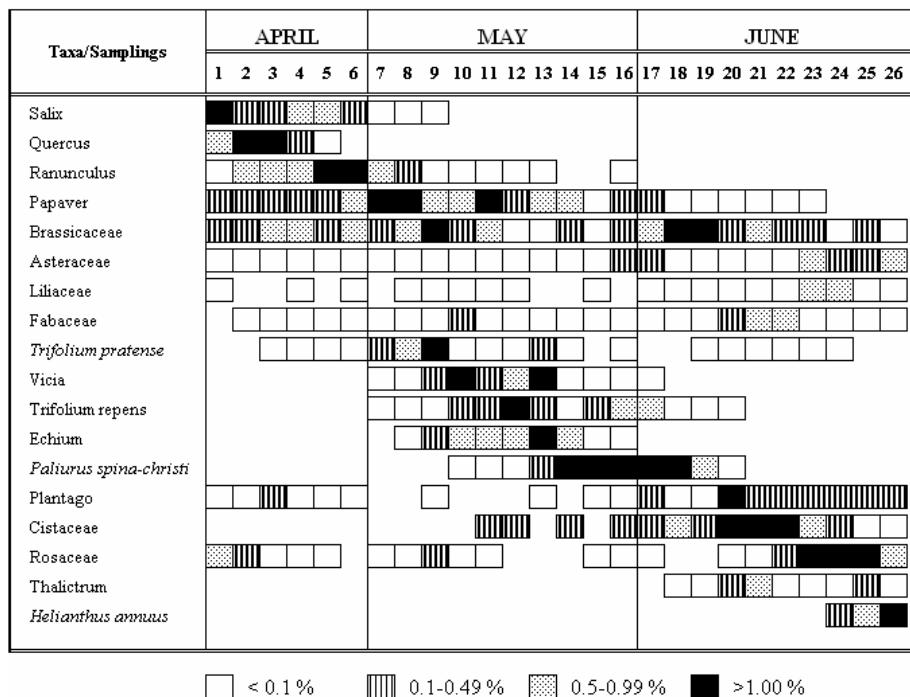
TAXA/SAMPLINGS	APRIL						MAY										JUNE							TOTAL									
	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22	23	24	25	26							
Acer	0,01	0,05	0,06	0,38	0,02																					0,53							
Asteraceae	0,01	0,03	0,01	0,02	0,05	0,01	0,01	0,01	0,01	0,01	0,01	0,02	0,01	0,01	0,15	0,20	0,04	0,03	0,01	0,01	0,08	0,50	0,24	0,20	0,65	2,32							
Boraginaceae		0,01	0,01			0,01		0,01									0,17		0,01	0,01	0,01	0,01	0,01	0,06	0,13	0,01	0,42						
Brassicaceae	0,20	0,27	0,66	0,65	0,37	0,63	0,46	0,58	1,04	0,37	0,64	0,09	0,01	0,12	0,02	0,34	0,57	1,80	2,53	0,42	0,63	0,25	0,16	0,04	0,13	0,06	13,00						
Campanulaceae			0,01	0,01		0,01	0,01																				0,02						
Calyptegia																											0,05	0,01	0,06				
<i>Castanea sativa</i>																		0,02	0,06	0,35	0,05	0,03						0,51					
Carduus																											0,02	0,03	0,15	0,53	0,04	0,77	
Caryophyllaceae																	0,01	0,01	0,01	0,01	0,01									0,03			
Centaurea																											0,02	0,06	0,06	0,02	0,02	0,10	0,28
Cichorioideae		0,01					0,01	0,01	0,01								0,01	0,01	0,01	0,01	0,00	0,01	0,04	0,09	0,22	0,20	0,60						
Cistaceae											0,01	0,03	0,18	0,20	0,08	0,49	0,10	0,38	0,47	0,54	0,36	1,31	1,11	1,79	0,62	0,26	0,05	0,08	8,03				
Convolvulus	0,01	0,01			0,01												0,01	0,01									0,03	0,01	0,07				
Cucurbitaceae																														0,04	0,04		
Cupressus	0,01	0,03	0,01		0,01		0,01	0,01																				0,07					
Cyperaceae	0,01			0,01														0,01	0,01									0,13	0,16				
Echinops																												0,01	0,02	0,03			
Echium							0,01	0,12	0,77	0,95	0,64	1,03	0,73	0,01	0,09															4,34			
Epilobium																												0,01	0,01	0,01			
Euphorbia	0,03	0,02	0,01	0,01																									0,07				
Fabaceae		0,02	0,01	0,08	0,01	0,01	0,01	0,01	0,05	0,11	0,01	0,02	0,01	0,01	0,01	0,01	0,01	0,01	0,07	0,41	0,63	0,57	0,06	0,01	0,01	0,01	2,11						
Geraniaceae	0,01	0,01					0,01	0,01																				0,02					
<i>Helianthus annuus</i>																												0,13	0,73	1,30	2,15		
Juglans	0,01	0,01	0,01		0,04	0,01																						0,07					
Labiatae			0,01	0,01					0,01	0,01	0,01	0,01															0,01	0,04	0,08				
Liliaceae	0,01			0,01		0,01		0,01	0,01	0,01	0,01						0,01		0,01	0,01	0,01	0,02	0,01	0,09	0,68	0,61	0,07	0,01	1,53				
Malvaceae																	0,01	0,01									0,01	0,01	0,01	0,04			
Oleaceae		0,01																											0,01	0,04			
Onobrychis																			0,01		0,01								0,02				
<i>Palmaria spinosa-christi</i>																	0,05	0,05	0,07	0,12	1,65	3,22	1,82	1,35	1,30	0,57	0,03				10,23		
Papaver	0,15	0,28	0,41	0,42	0,43	0,77	2,16	1,98	0,88	0,89	1,36	0,28	0,66	0,70	0,08	0,24	0,23	0,01	0,02	0,02	0,01	0,01	0,03						11,99				
Pinus	0,01	0,01	0,01	0,01		0,01		0,01				0,00	0,01				0,01											0,05					
Plantago	0,01	0,01	0,12	0,08	0,02	0,01		0,01				0,01					0,01	0,04	0,11	0,06	0,09	1,05	0,40	0,15	0,44	0,39	0,43	0,45	3,87				
Poaceae	0,02	0,07	0,04	0,05	0,01	0,03	0,02	0,01	0,01	0,02	0,07	0,05	0,02	0,01	0,01	0,02	0,02	0,01	0,01	0,07	0,20	0,05	0,01	0,01	0,01	0,79							
Populus	0,01	0,01	0,01	0,01																									0,02				
Quercus	0,89	1,78	1,34	0,31	0,02																							4,33					
Ranunculus	0,02	0,55	0,97	0,88	2,15	1,99	0,58	0,10	0,01	0,02	0,01	0,01	0,01				0,01										7,29						
Rosaceae	0,53	0,23	0,01	0,10	0,01		0,05	0,07	0,36	0,02	0,02						0,02	0,01	0,01									6,32					
Rubiaceae												0,01	0,01															0,01					
Salix	1,93	0,45	0,15	0,72	0,66	0,32	0,03	0,03	0,01								0,01	0,07	0,08	0,01	0,01	0,07	0,04	0,05	0,07	0,01	0,01	4,29					
Sanguisorba																	0,01												0,43				
Scabiosa																												0,01	0,01	0,02			
Scrophulariaceae	0,01	0,01	0,01	0,02			0,01				0,03	0,01																0,08					
Sympathium					0,01	0,01																						0,01					
Thalictrum																				0,01	0,04	0,13	0,73	0,03	0,07	0,07	0,12	0,07	1,25				
Tilia																				0,01	0,01	0,02	0,06	0,03			0,01	0,01	0,12				
<i>Trifolium pratense</i>			0,01	0,03	0,03	0,05	0,42	0,96	1,17	0,03	0,01	0,05	0,35	0,01		0,01					0,01	0,01	0,01	0,01	0,01	0,01		3,15					
<i>Trifolium repens</i>							0,02	0,01	0,03	0,18	0,32	1,67	0,39	0,07	0,31	0,61	0,63	0,05	0,02	0,04								4,33					
Umbelliferae					0,01			0,01	0,00	0,01	0,13	0,01	0,01	0,01			0,01	0,01				0,01					0,01	0,01	0,20				
Urticaceae	0,00	0,01	0,01	0,05	0,03	0,01		0,00		0,06																		0,17					
Vicia							0,07	0,04	0,11	1,10	0,20	0,73	1,13	0,03	0,04	0,04	0,01										3,48						
Unidentified	0,02	0,02	0,01	0,01	0,01	0,01	0,01		0,01	0,01	0,01	0,01	0,01	0,01			0,01	0,01	0,02	0,01		0,01	0,01	0,01	0,01	0,01	0,17						

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Cistaceae pollen loads were collected primarily between 9th and 26th sampling periods. Their values reached a maximum during the 20-22nd sampling period (Figure 1) and they constituted 8.03% of the total (Table 1). Ranunculus spp. pollen loads were collected in the spring between the 1- 16th sampling periods with the highest amounts collected during the 5-6th sampling period (Figure 1). Ranunculus spp. pollen loads represented 7.29% of the total (Table 1).

Rosaceae pollen loads were collected intermittently by honeybees during the study period and probably reflect different flowering times for the species lumped within this group such as *Rubus sanctus*, *R. discolor* and cultivated *Fragaria* and *Rosa* species. Pollen loads of this family represented 6.32% of all (Table 1) and maximum values were found during the 23-25th sampling period (Figure 1).

Figure 1: Bee collected predominant pollen types and their seasonal variations in the study area.



DISCUSSION

DISCUSSION
 In April, honeybee foragers collected 30 types of pollen grains mainly from *Salix* spp., *Quercus* spp., *Ranunculus* spp., *Papaver* spp. and *Brassicaceae* (Table 1). *Quercus* spp. and *Salix* spp. known as wind pollinated plants but pollen usage of honeybees on willow trees is an ordinary case. High percentages of *Quercus* spp. pollen loads were surprising to us based on previous research in northwest Anatolia (Bilisik et al. 2007, Bilisik et al. 2008). We believe this reflects the availability of this particular pollen compared to others. For example, the 5th and 6th sampling period reflects the availability of pollen from *Ranunculus* spp. flowers (Figure 1). This is important for beekeepers because these pollen providing plants in April help for quick Spring build up of honeybee colonies in the region.

Honeybee foragers collected 34 different types of pollen and 8 of them passed the 1% threshold in May; *Papaver* spp., *Palirurus spina-christii*, *Echium* spp., *Brassicaceae*, *Trifolium repens*, *Vicia* spp., *Trifolium pratense* and *Cistaceae* were most frequented plants used as a pollen source (Table 1). In the first samples of May, there is a high constancy to poppy pollens by honeybee foragers. Our observations suggest that honeybee foragers do not prefer *Papaver* spp. pollen grains after a heavy rain. That

was probably the reason of the shedding of the poppy petals; for honeybees could not find a running track to collect any more pollen grains from poppy flowers thus they turned to other sources such as Brassicaceae species and *Trifolium repens*. Foragers mostly collect *Trifolium repens* pollen grains in 12th sampling period and with the beginning of the flowering period of *Paliurus spina-christii* they mostly focused on this plant (Figure 1). In June, honeybee foragers mostly collected Brassicaceae, Cistaceae, Rosaceae, *Plantago* spp., *Paliurus spina-christii*, *Helianthus annuus*, Asteraceae, Fabaceae, Liliaceae and *Thalictrum* spp. pollen grains (Table 1). After leaving *Paliurus spina-christii* pollen, honeybees turned to another species of the Brassicaceae family and they visited the nectar-less plant *Plantago* spp. the Cistaceae species during the 20th sampling period. After this, they shifted to Rosaceae and from Rosaceae to *Helianthus annuus* (Figure 1). After June, with the beginning of the main nectar flow, pollen foraging activity was decreased in northwest Anatolia (Biliski et al. 2008).

Honeybees collect more pollen from some plant species than from others, and a pollen calendar prepared from these data can be used by beekeepers of the region (Figure 1). In order to know more reliably what species of

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plant the honeybees are utilizing, a more detailed study is required that can identify pollen to the species level. Although being dominated within the calendar, Asteraceae, Liliaceae, Fabaceae and Thalictrum spp. pollen grains have never been seen up to the percentage of 1 % in any samples. This might be a result of nutrition levels of pollen grains offered by some species over others because amino acid composition or protein content of pollen grains has been implicated as a reason of attraction (e.g. Kim and Smith 2000, Pernal and Currie 2001, Cook et al. 2003). The palynological composition of pollen loads collected by honeybee colonies reflected the local flora in our investigation.

Finally, the results demonstrate that honeybee foragers concentrate on a few plant species at a time despite the rich and diverse flora in the study area. The pollen types which were recorded in high levels are abundant in the vicinity of the hives. Based on our results Brassicaceae, Papaver spp., Paliurus spina-cristii, Cistaceae, Ranunculus spp. and Rosaceae are the main pollen taxa collected by Anatolian honeybees in the area.

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Özet: Anadolu arısının polen tercihlerini, bölgenin polen kaynaklarını ortaya koymak ve yöresel veya gezgin arıcılar için kullanışlı olan bal arısı polen takviminin çıkartılması amacı ile Bursa ovasında, ana polen akımı döneminde bal arılarının (*Apis mellifera anatoliaca*) topladıkları polenlerin yayılımı araştırılmıştır. Bu amaç doğrultusunda Bursa'nın Mustafakemalpaşa ilçesinde Langstroth tip beş adet kovan seçilmiştir. Kovanlardan polenler, yoğun polen akımı döneminin başlangıcından bitimine kadar olan 10 Nisan–1 Temmuz 2005 tarihleri arasında, her kovanın çekmecesinden rastgele alınarak örnekleştirilmiştir. Bu yolla 130 örnek elde edilmiş ve örnekler analiz edilene kadar +4°C'de saklanmıştır. Bal arılarının bölgede kullandıkları bitki kaynaklarını teşhis edebilmek için her örnek şişesinden rastgele 500 polen pelleti seçilerek renklerine göre ayırt edilmiştir (Kirk 1994). Her bir renge ait pollen pelletinden alınan bir parça Wodehouse metoduna (1935) göre preparat haline getirilmiş ve polenler ışık mikroskopunda incelenmiştir. Elde edilen bilgiler doğrultusunda her taksona ait polen miktarı yüzde cinsinden hesaplanmıştır. Renklerine göre ayrılan polen pelletlerinin analiz edilmesi ile elli bir takson teşhis edilmiş ve bunların on sekiz tanesi toplamın %1'inden fazla bulunmuştur. %0,17'si ise tanımlanamayan olarak kaydedilmiştir. Dominant taksonlar; Brassicaceae (%13,00), *Papaver* spp. (%11,99), *Paliurus spina-christi* (%10,23), Cistaceae (%8,03), *Ranunculus* spp. (%7,29), Rosaceae (%6,32), *Echium* spp. (%4,34), *Trifolium repens* (%4,33), *Quercus* spp. (%4,33), *Salix* spp. (%4,29), *Plantago* spp. (%3,87), *Vicia* spp. (%3,48), *Trifolium pratense* (%3,15), Asteraceae (%2,35), *Helianthus annuus* (%2,15), Fabaceae (%2,11), Liliaceae (%1,53), *Thalictrum* spp. (%1,25) olup bunlar toplamın %94,01'ini oluşturmaktadır. Sonuç olarak bal arılarının bölgenin zengin florasına rağmen birkaç bitki türü üzerinde yoğunlaştıkları görülmüştür. Bal arılarının bazı bitkilerin polenleri için net bir tercih göstergeleri bölge arıcıları ile gezginci arıcılar için işlevsel olabilecek ve polen tiplerinin varyasyonunu gösteren bir takvim hazırlanmasına zemin hazırlamıştır. Polen toplama aktivitesinde bazı yerli bitkilere kuvvetli bir bağlılık görülmüş olmasına rağmen örneklerde birçok kültür formlarının polenlerine de rastlanmıştır. Haziran ayı farklı polen tiplerinin toplanması açısından çeşitliliğin en yüksek olduğu dönem olarak bulunmuştur. Özellikle Nisan ayında polen sağlayan bitkiler arı kolonilerinin ilkbahar gelişiminde oldukça rol oynamaktadır.

Anahtar Kelimeler: Polen, Polen yayılımı, Polen takvimi, *Apis mellifera anatoliaca*, Bursa.