

MELISSOPALNOLOGICAL ANALYSIS OF HONEY SAMPLES COLLECTED FROM ŞIRNAK CITY

Şırnak İli'nden Toplanan Balların Melissopalinojik Analizleri

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ÖZ

Bu çalışma ile Türkiye'nin Güneydoğu Anadolu Bölgesi'nden 23 adet bal örneği toplanılmış ve bitkisel kökenleri tanımlanmak üzere araştırılmıştır. Mikroskobik analiz sonuçlarına göre, 27 bitki familyasına ait 24 taksonun polenleri teşhis edilmiştir.

Melissopalinojik analiz sonucunda; 21 bal örneği multifloral (karışık çiçek balı) ve iki tanesi ise unifloral (*Myosotis* balı) olarak tanımlanmıştır.

Mikroskobik analizin ikinci basamağı olarak, örneklerin 10 gram baldaki toplam polen sayıları (TPS₁₀) hesaplanmış ve bu değerlerin 1 117 ile 82 005 arasında olduğu tespit edilmiştir.

Anahtar Kelimeler: Güneydoğu Anadolu, Bal, Melissopalinojisi, TPS₁₀

ABSTRACT

In this study, 23 honey samples collected from South-Eastern Anatolia Region of Turkey were investigated to identify their botanical sources. According to the microscopic analysis results, pollen grains identified belong to the 24 taxa of 27 plant families.

As a result of melissopalynological analysis, 21 of the honey samples are characterized as multifloral and two of them are as unifloral (*Myosotis* honey).

As the second step of the microscopic analysis, the total pollen number of in 10 grams honey (TPN₁₀) of the samples were calculated and it was observed that the values were vary between 1 177 and 82 005.

Key words: South-Eastern Anatolia, Honey, Melissopalynology, TPN₁₀

GENİŞLETİLMİŞ ÖZET

Giriş: Türkiye'nin gerek iklim özellikleri gerekse bitki çeşitliliği bakımından içerdiği yedi coğrafik bölgesi de arıcılık için oldukça yüksek potansiyele sahiptir. Gençlerin ve kadınlarımızın da bu konuda istihdamına yönelik çalışmalar ve teşvikler Türkiye'nin çeşitli bölgelerinde yapılmaktadır. Türkiye için önemli bir pazar olan arıcılık sektörü özellikle Doğu Anadolu Bölgesi'nde kırsal kalkınma için önemli bir iş koludur.

Türkiye'de üretilen ballar bitkisel içerik bakımından çok çeşitlilik göstermekte olup, bunlar arasında üçgül balı, narenciye balı, kestane balı, ormangülü balı, yayla balı ve salgı balları sayılabilmektedir. Çoğu yöremizde kendine has ve ün yapmış ya da markalaşmış bal üretimleri mevcuttur. Ancak ülkemizde üretilen ballara ait yapılan çalışmalar genellikle yöre bazında olmayıp, Türkiye'nin farklı bölgeleri'nden toplanılan ballar üzerinden gerçekleştirilmiştir. Bunun yanında, ülkemiz balları ile ilgili yapılan çalışmaların çoğunluğu, balların fizikokimyasal karakterize edilmesine yönelik olmuştur. Yani çalışılan balların bitkisel kökeni melissopalinojik analizler ile tanımlanmadan fizikokimyasal analizlerine dayalı çalışmalar yapılmaktadır. Dolayısıyla tam bir karakterize etme gerçekleştirilememektedir.

Gereç ve yöntem: Bu çalışma ile daha önce detaylı olarak çalışılmamış olan Türkiye'nin Doğu Anadolu Bölgesi'nde bulunan Şırnak ili'ne ait 23 bal örneği incelenmiştir. Analizler melissopalinojik açıdan nitel ve nicel olarak gerçekleştirilmiştir.

Bulgular: Örnekler nitel açıdan incelendiğinde, analiz edilen 23 bal örneğinin bitkisel kökeni olarak çoğunlukla; Asteraceae, Apiaceae, Berberidaceae, Betulaceae, Boraginaceae, Brassicaceae, Campanulaceae, Caryophyllaceae, Caprifoliaceae, Chenopodiaceae, Cistaceae, Convolvulaceae, Dipsecaceae, Fabaceae, Fagaceae, Geraniaceae, Lamiaceae, Liliaceae, Malvaceae, Plantaginaceae, Poaceae, Polygonaceae, Ranunculaceae, Rosaceae, Rubiaceae, Rutaceae ve Salicaceae familyalarına ait bitki taksonları tespit edilmiştir. Dominant oranda sadece Boraginaceae familyasına ait *Myosotis* sp. taksonuna ait polenlere rastlanılmıştır. Apiaceae, Berberidaceae, Caryophyllaceae, Fabaceae, *Lotus* sp., *Onobrychis* sp., *Plantago* sp., Rosaceae, ve *Trifolium* sp. taksonlarına ait polenlere ise bazı örneklerde sekonder oranlarda rastlanılmıştır.

Nicel analiz kısmında ise; incelenen bal örneklerinin 10 gramında bulunan toplam polen miktarları hesaplanmış ve polence zenginlik düzeyleri gözlemlenmiştir. Bu değerlerin 1 177 ila 82 005 aralığında değişiklik gösterdiği tespit edilmiştir. En düşük değer Beytüşşebap ilçesinin Yeşilöz köyünden toplanan A16 örneğinde gözlemlenirken en yüksek değer ise yine Beytüşşebap ilçesinin Toptepe Köyü'nden toplanan A22 örneğinde gözlemlenmiştir.

Her iki örneğinde nitel melissopalinojik analiz sonuçlarına bakacak olursak benzerlik göstermekte olup; *Onobrychis* sp. taksonuna ait polenlere sekonder oranda rastlanılmıştır.

Sonuç: Çalışma sonucunda yöre ballarının çoğunluğunun karışık çiçek balı olduğu, yani baskın olarak tek bir nektar kaynağından değil çok sayıda farklı bitkilerden kaynaklandığı gözlemlenmiştir. Örneklerden iki tanesinin ise *Myosotis* balı olduğu tespit edilmiştir. Bu iki bal ise unifloral olarak nitelendirilmiştir. *Myosotis* balı olarak nitelendirilen A7 balı; Beytüşşebap ilçesinin Akarsu köyünden, A8 örneği ise yine Beytüşşebap ilçesinin Beşagaç köyü'nden toplanmıştır.

Bu sonuçlar, Şırnak balları ile ilgili yapılacak olan ileriki çalışmalara ışık tutabilecek niteliktedir ve yöreden elde edilebilecek *Myosotis* balları ile ülke pazarına yeni bir ürün girdisi sağlanabileceği düşünülmektedir.

INTRODUCTION

Honey is described as “the sweet substance produced by honeybees from the nectar of blossoms or from secretions on living plants, which the bees collect, transform and store in honey combs” (Codex Alimentarius Commission, 2001).

The content and quality of honey shows varieties according to the climatic conditions, the environmental temperature, the botanical source that bees prefer to collect nectar, honey bees species, harvesting and storage conditions (Alvarez-Suarez et al. 2010). Also its colour has a wide range of spectrum, including white, amber, red, brown and almost black (Ndife et al.,2014). The taste, smell and

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color of honey changes according to the nectar of flowers collected by bees. Its flavour and texture also vary owing to the botanical source (Alvarez-Suarez et al. 2010).

The most known biological activity of honey is its antimicrobial effect. Besides antimicrobial activity, honey has been found to contain significant biological activities, especially antioxidant activity (Bogdanov et al. 2008).

Melissopalynological analysis is a type of method that investigate the pollen grains and spores in honeys to determine the source of honey. It can give information about the botanical and geographical sources of the sample (Kaya et al. 2005). Microscopical analysis of honey is based on the fact that the raw materials of honey (nectar and honeydew) have certain constituents which remain identifiable in the ripe honey. For the nectar, these constituents are generally pollen grains from the plants producing the nectar. For the honeydew, these constituents are usually fungus spores and hyphae that come from the forest trees (Lieux 1972).

From the honey types, monofloral honeys, that are originated from dominantly one plant species, are most attractive compare to the multifloral honeys. It is possible to determine their origin from the dominant pollen grains by microscopic investigation. Besides this, multifloral honeys are originating from the nectar of several plant species (Barth 2004).

Studies in Turkey identified flowering plants containing nectar through pollen analysis in honey samples were started with Quistani (1976), the first research for the pollen analysis of Turkish honey. Sorkun and Yuluğ (1984), investigated the honey samples from Erzurum, Gür (1993), investigated Elazığ honey, Kaplan (1993), Konya honey, Türker; (1993) Gümüşhane honey, Silici (1995), Antalya honey, Yılmaz (1969), İzmit honey, Kemancı (1999), Marmaris honey, Mercan et al. (2007), honey samples from İzmir, Sivas Afyon and Muğla. Also Sorkun and Doğan (1995), Can et al. (2015) were investigated the honey samples from various regions of Turkey by melissopalynological analysis.

By this study we aimed to determine botanical sources of some honey samples collected from Şırnak city located in South-Eastern Region of Turkey, by melissopalynological analysis.

MATERIALS AND METHODS

23 honey samples were collected from Şırnak city which is located in South-Eastern Region of Turkey in 2017.

Seven of them were collected from the village Toptepe, seven from Yeşilöz, four from Akarsu village, one sample each from Beşağaç, Boğazören and Söğütçe villages and two samples from Uludere (Figure 1, Table 1).

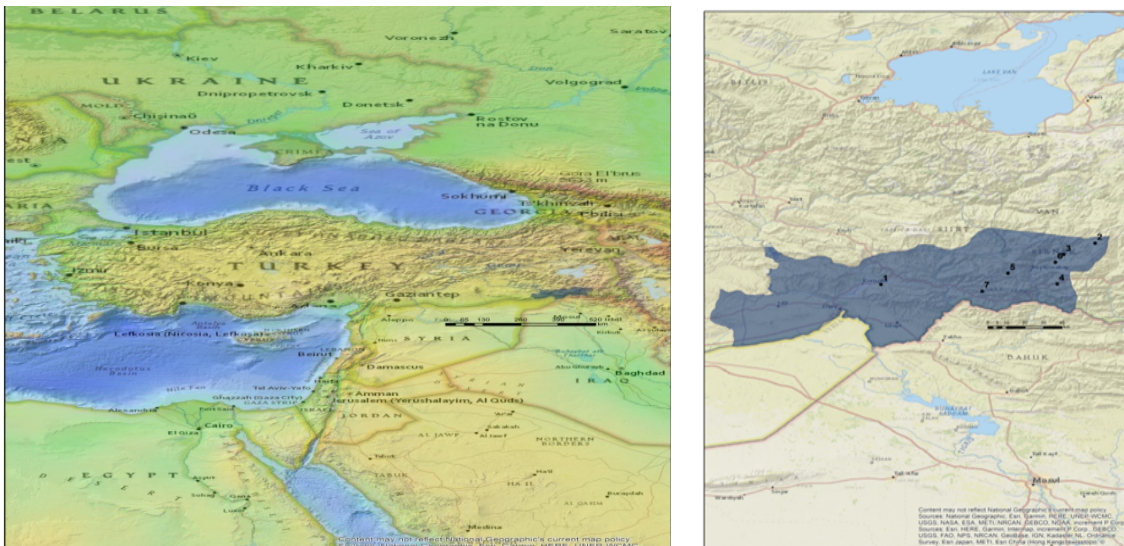


Figure 1. The location of Şırnak city (This map prepared with ArcGIS pro 2.2)

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Table 1. The collection areas of the honey samples

Lattitude	Longitude	City	Location-	Sample Numbers
1 37,4652000	42,39	Şırnak	Beytüşşebap-Toptepe Village	A1, A11, A17, A19, A20, A22, A23,
2 37,6980194	43,431	Şırnak	Beytüşşebap-Yeşilöz Village	A2, A4, A9, A14, A16, A18, A21,
3 37,6338500	43,27	Şırnak	Beytüşşebap -Akarsu Village	A3, A5, A7, A15,
4 37,4679556	43,24	Şırnak	Beytüşşebap -Beşağaç Village	A8
5 37,5280833	43,00	Şırnak	Beytüşşebap Village	Boğazören A6
6 37,5904278	43,23	Şırnak	Beytüşşebap Söğütçe	A12
7 37,4269333	42,88	Şırnak	Uludere Uzungeçit Plateau	A10, A13

Melissopalynological Analysis

Preparation of honey samples for qualitative and quantitative melissopalynological analysis was performed by Louveaux et al. (1978). Stock honey sample was well stirred and 10 grams of it was weighed in a centrifuge tube. Then 20 ml distilled water was added and to melt down the honey, tube was left for 10-15 minutes in a water bath of 45°C. The mixture was centrifuged in 3500 rpm for 45 minutes. After centrifugation, the supernatant was poured and from the residual sediment slides were prepared by using glycerine gelatine with basic fuchsin. Then the slides were investigated under the microscope. The classification of Zander (1935) was used in this study to indicate pollen and associated nectar source: dominant pollen (over 45%), secondary pollen (16-45%), minor pollen (1-15%); trace pollen (less than 1%).

The total pollen number in 10 grams honey (TPN₁₀) of all samples was calculated according to the method described by Moar (1985). The honey samples (10g) were classified according to total pollen number (TPN₁₀) as Group I: TPN<20000; Group II: 20000<TPN<100000; Group III: 100000<TPN<500000; Group IV: 500000<TPN<1000000 and Group V: TPN>1000000 (Maurizio 1975).

RESULTS

According to the microscopic analysis results, pollen grains were identified belong to the taxa of Asteraceae, Apiaceae, Berberidaceae, Betulaceae, Boraginaceae, Brassicaceae, Campanulaceae, Caryophyllaceae, Caprifoliaceae, Chenopodiaceae, Cistaceae, Convolvulaceae, Dipsecaceae, Fabaceae, Fagaceae, Geraniaceae, Lamiaceae, Liliaceae, Malvaceae, Plantaginaceae, Polygonaceae, Ranunculaceae, Rosaceae, Rubiaceae, Rutaceae, Salicaceae. The microscopic analysis results of the honey samples are given in Table 2. With regard to Zander's classification; pollen grains belong to the *Berberis* sp., *Onobrychis* sp., *Trifolium*, *Plantago* taxa and Apiaceae, Caryophyllaceae, Fabaceae, Rosaceae families were found in secondary ratios. Besides this only pollen of *Myosotis* sp. was found in dominant ratio in investigated two samples with high ratios (72.77%-80.63%) owing to higher pollen producing potential of *Myosotis* (A7, A8) (Fig. 2,3,4).

The TPN₁₀ values are vary between 1 177 and 82 005. According to the Moar (1985); 9 of 23 samples are included in group II, 14 of total are in group I (Table 2).

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Table 2. Microscopic analysis results of honey samples (for samples A1-11) (D> 45%, S:16-45%, M:1-15%, T<1%).

Plant Family	Plant taxa	A1	A2	A3	A4	A5	A6	A7	A8	A9	A10	A11
Asteraceae		M	M	M					M			M
	<i>Centaurea sp.</i>	M							T			
	<i>Taraxacum sp.</i>	M										T
Apiaceae		M	S			M	S	M	M	M	M	T
	<i>Daucus sp.</i>		T						T			
Berberidaceae		M	M		S	S	M	T	T	S	M	M
Boraginaceae		M					M				M	T
	<i>Cerintho sp.</i>		M									
	<i>Heliotropium sp.</i>		T									
	<i>Myosotis sp.</i>	M	M				M	D	D			
Brassicaceae		T	M						T			
Campanulaceae		M	M				M	M	M			M
Caryophyllaceae		T	M	M	M		M					
Caprifoliaceae	<i>Scabiosa sp.</i>	T	T									
Cistaceae		T				M			T	M	M	M
Fabaceae		S	M	M	M	M	S	M	M	S	M	S
	<i>Astragalus sp.</i>			M		M	M	T	M			
	<i>Vicia sp.</i>		T				M					
	<i>Lathyrus sp.</i>							M				
	<i>Lotus sp.</i>							T				T
	<i>Onobrychis sp.</i>	M	M				M			M	S	
	<i>Trifolium sp.</i>	M	M	M	S	S	S	M	M		M	S
	<i>T. pratense</i>		M									
	<i>Castanea sativa</i>							T				
Geraniaceae			T									T
Lamiaceae		M	M				M	T		M	M	
	<i>Teucrium sp.</i>	M										T
Liliaceae		M										
Malvaceae					M							
Plantaginaceae												
	<i>Plantago sp.</i>	T	M	M	M	M				M	S	
Poaceae		M		M				T	T			T
Polygonaceae	<i>Rumex sp.</i>		T									
Ranunculaceae					M	M				M		
Rosaceae		M	M	M		M	M	M		S		M
	<i>Sanguisorba sp.</i>			M								

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Rubiaceae			T										
	<i>Galium sp.</i>	T	T		M				T				
Salicaceae	<i>Salix sp.</i>	M	M		M	M					M	T	
TPN₁₀ values		27 009	21 985	15 713	9666	12 410	21 244	14 890	12 539	6322	2133	38 664	

Plant Family	Plant taxa	A12	A13	A14	A15	A16	A17	A18	A19	A20	A21	A22	A23
Asteraceae		M	M	M	M	M		M		M	M		M
	<i>Centaurea sp.</i>			T							M		
	<i>Taraxacum sp.</i>			T			M				T		
Apiaceae		M	S	M	M	M	M				T		M
	<i>Daucus sp.</i>		M	T									
Berberidaceae			S	M	M						M		M
Betulaceae									M				
Boraginaceae				M	M								
	<i>Alkanna sp.</i>				T						T		
	<i>Echium sp.</i>			T	T		M				M		M
	<i>Cerinthe sp.</i>			T									
	<i>Heliotropium sp.</i>		M										
	<i>Myosotis sp.</i>			T									
Brassicaceae				T			M	M			M		T
Campanulaceae			M	T							T		T
Caryophyllaceae		S		T				M			T		T
Caprifoliaceae	<i>Scabiosa sp.</i>												
Chenopodiaceae		M											
Cistaceae				T								M	T
Convolvulaceae		M											
Geraniaceae			M							M	T		
Dipsacaceae													T
Fabaceae		M	M	S	S	S	S			S	S	S	S
	<i>Astragalus sp.</i>		M										
	<i>Vicia sp.</i>										T		
	<i>Lathyrus sp.</i>												
	<i>Lotus sp.</i>	M		T	M				M	S	M		M
	<i>Medicago sp.</i>										T		
	<i>Onobrychis sp.</i>	M	M	S	M	S	M	M		S	M	S	S
	<i>Trifolium sp.</i>	M	M	M					M		M	M	M
	<i>T. pratense</i>			T									

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Lamiaceae		M	M	M	M		M			M	M	M	
	<i>Teucrium sp.</i>	M	T	T			M		M	M			
Liliaceae			T	T			M						
Plantaginaceae			T										
	<i>Plantago sp.</i>	M	T	M	M		M	M	M	M		M	
Poaceae			T	M			M					M	
Polygonaceae	<i>Rumex sp.</i>										T		
Ranunculaceae							M		M				
Rosaceae		M	M	T	M	M	M	M		T		M	
Rubiaceae													
	<i>Galium sp.</i>										T		
Rutaceae			M	T							T		
Salicaceae	<i>Salix sp.</i>	M		M	M	M	M	M		M		M	
TPN₁₀ values		2071	3314	55 833	17 972	1177	5029	3945	7000	48 330	61411	82 005	39 168

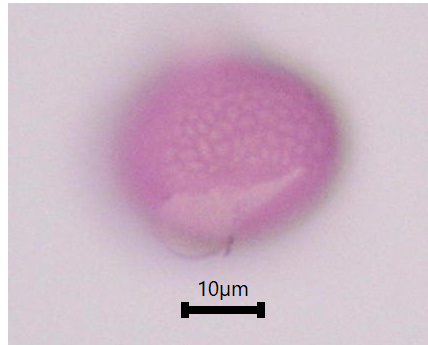
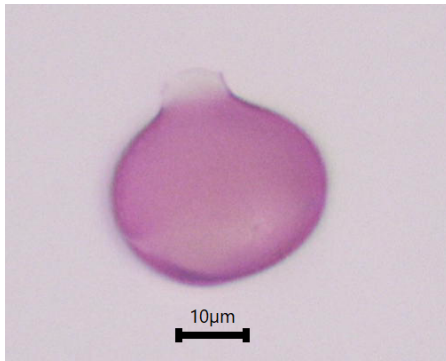


Figure 2. Microphotograph of *Trifolium* sp. pollen grain(X400)

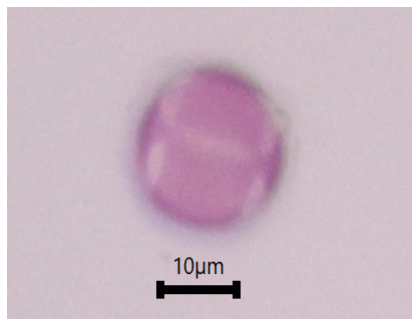


Figure 3. Microphotograph of *Berberis* sp. pollen grain (X400)

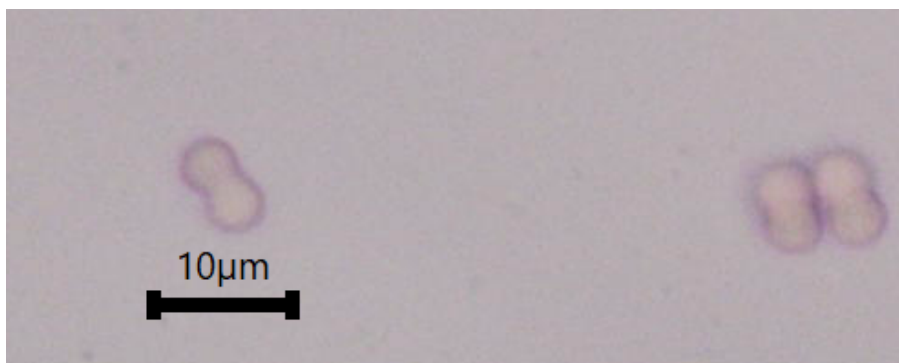


Figure 4. Microphotograph of *Myosotis* sp. pollen grain(X400)

DISCUSSION

Turkey has a rich flora and suitable climatic conditions for beekeeping and producing high quality bee products. Especially the Eastern parts of Turkey exhibits a high diversity of plant species. Owing to plant species richness of the country, honeys produced in Turkey have really high quality that represent so many nectar source in it.

As a result of melissopalynological analysis, only two of the investigated of 23 honey samples were found as unifloral that is sourced dominantly from *Myosotis* nectar. The other 21 samples were evaluated as multifloral honey that are mostly sourced from nectar of *Berberis* sp., *Onobrychis* sp., *Trifolium* sp., *Plantago* sp., Apiaceae, Caryophyllaceae, Fabaceae, Rosaceae plant taxa.

According to the previous studies, it is the first investigation about botanical origin of honey which are produced in Şırnak. We observed that there is no any detailed information about botanical source of honey samples of Şırnak city. Generally the researches are about the different locations of Turkey.

There are so many researches about Turkish honey (Yılmaz and Yavuz, 1999; Küçük et al., 2007; Cengiz et al., 2018) but they are mostly based on physicochemical analysis and not comprise a specific location as we done.

Can et al. (2015) analyzed melissopalynological and physico-chemical properties of sixty-two honey samples from the regions of Aegean, Marmarean and Black Sea of Turkey. They found 11 unifloral honeys (chestnut, heather, chaste tree, rhododendron, common eryngo, lavender,

Jerusalem tea, astragalus, clover, acacia), two different honeydew honeys (lime and oak), and 7 different multifloral honeys. The samples of their study showed that physico-chemical and biological characteristics of honeys are closely related to floral sources, and dark colored honeys evaluated as oak, chestnut and heather, having high therapeutic potential.

Kaya et al., 2005 investigated the 13 floral honeys from various regions of Turkey (Kırklareli, Marmaris, Manisa, Yozgat, Çankırı, Bolu, Balıkesir, Aydın, Bartın, Elazığ, Tekirdağ, Rize). They found one unifloral honey and 12 multifloral honeys. Pollen have been identified belonging to the 86 taxa. The dominant group of pollen grains consisted of: *Hedera helix*, *Gossypium*, *Trifolium*, *Sophora*, *Rhododendron*, *Castanea sativa*, *Peganum harmala* and *Helianthus*.

Mercan et al. (2007) investigated the honey samples from İzmir, Sivas, Afyon, Muğla and found Chenopodiaceae pollen grains as dominant in İzmir, *Anthemis*, *Papaver*, *Rumex*, *Trigonella*, *Onopordum*, Umbelliferae in Sivas, *Erica*, *Centaurea*, Chenopodiaceae, Amaranthaceae, *Helianthus annuus* in Afyon, *Erica*, Umbelliferae in Muğla.

Yılmaz and Küfrevioğlu (2001) investigated honey samples from South-Eastern Anatolia but they studied chemical properties of the samples. They don't give any information about the botanical sources of honey from this region.

Çam et al. (2010) investigated, 30 honey samples from Ankara city markets. They identified pollen grains belong to the 46 taxa. The pollen grains of Fabaceae, Aceraceae, Boraginaceae, Poaceae, Asteraceae, Apiaceae, Caryophyllaceae, Rosaceae,

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Hedysarum, Brassicaceae, Fagaceae as dominant and secondary while the pollen grains of *Plantago*, Cistaceae, Geraniaceae, Cucurbitaceae, Liliaceae, Caryophyllaceae, *Rumex*, Plantaginaceae, *Echium*, Campanulaceae, Salicaceae, Chenopodiaceae, Lamiaceae, Ericaceae identified as the minor group.

Compare to the previous researches, we observed that, there is no any data about *Myosotis* honey for Turkey. So this can be the first record for the *Myosotis* honey for Turkey. Besides this, the investigated multifloral honey samples show similar pollen contents with the other plateau honeys of Turkey that are sourced from any other region of Turkey.

This research can be helpful for future researches about any investigations of honeys (physicochemical etc.) from South-Eastern of Turkey.

CONCLUSION

Melissopalynological analysis is important to qualify and characterize the honey samples according to their botanical sources and also reflects the plant spectrum of the location that honeys collected. It also gives information about the hygiene and adulteration of the honeys.

Şırnak is a favorable location for beekeeping. Its flora comprises various plant species that have nectar potential for honeybees. Owing to the pollen analysis of honey samples from Şırnak, the plant spectrum of the region was reflected. The results are overlapping with the flora of Şırnak (TÜBİVES, 2019).

As a result we can say that this research is the first detailed melissopalynological investigation about Şırnak honeys. According to the results, the samples show mostly multifloral honey characteristics. That is derived from so many botanical sources and there is no any dominant nectar source. Only two of them evaluated as unifloral honeys. So, they derived from mostly from one species.

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