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Allergenic Pollen in the Atmosphere of Kayseri, Turkey

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SUMMARY **Airborne pollen are important allergens that cause sensitization in allergic rhinoconjunctivitis and asthma. Our aim was to detect the pollen in the atmosphere of Kayseri, to present a pollen calendar, and to detect the allergenic level of these pollen by performing skin tests on patients. Atmospheric pollen were collected by Durham gravimetric samplers in Kayseri between March and November in the years 1996 and 1997. In our study, we observed pollen belonging to 43 different taxa. The total number of pollen per cm^2 was found to be 1,330.8 in 1996 and 1,182.5 in 1997. Most of the pollen were from the taxa *Pinus, Poaceae, Chenopodiaceae/Amaranthaceae, Cupressaceae, Populus* and *Quercus* in decreasing order. In the skin tests, pollen of the taxa *Poaceae* and *Chenopodiaceae* were found to give the most frequent allergic reactions. It was concluded that preparing an airborne pollen calendar could be useful for medical practice. Nevertheless the skin test data did not really correlate with the aerobiologic data, as skin test reactivity is related to the allergenicity of the pollen and not just to ambient exposure.**

Some aeropalynological studies were performed in Turkey previous to our study. Ozkaragoz[1] searched allergic pollen and spores in the atmosphere of Ankara and performed skin test on his patients. Aytug[2] performed aeropalynological studies in the Belgrade Forests of Istanbul by a Hirst-Burkard sampler. Yurdukoru[3] searched the allergic pollen in the atmosphere of Samsun. Ince and Pehlivan[4] searched airborne pollen in Serik (Antalya) in the south of Turkey and devised a pollen calendar. Inceoglu and coworkers[5] searched airborne pollen in Ankara by Burkard samplers, and Ince[6] searched airborne pollen in Kirikkale by a Durham sampler. Both presented pollen calendars. Ince[7] performed some studies about plants that can be allergenic in Turkey. Some investigators performed skin tests on patients.[8-12]

It has been reported that the prevalence of allergic diseases (rhinitis, asthma and atopic dermatitis) has increased progressively over recent years.[13,14] In particular, the prevalence of pollinosis has increased over the last decade.[15]

The aim of our study was to identify the pollen in the atmosphere of Kayseri, to present a pollen calendar, and to detect the allergenic degree of these pollen by performing skin tests on patients. Thus we hope to improve the diagnosis of allergic diseases and to give input into the pollen calendar of Turkey.

MATERIALS AND METHODS

Collection of atmospheric pollen

Kayseri is located at the Inner Anatolian Region (near Cappadocia) of Turkey at an altitude of 1,093 meters (m), at 38°45'N, 35°29'E. Kayseri is part of the Irano-Turanian floristic region.[16] The major flora of the province is steppe vegetation, which is nurtured by the arid and semiarid aspects of the Mediterranean climate, which is a dry sub-humid climate with little or no water surplus. The mean monthly temperatures vary between 24°C in

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July and 1°C in January. Precipitation is unevenly distributed over the year in spring and in the end of summer. Therefore, we collected the samples from the beginning of the spring until the end of the summer. In winter pollination is not enough due to cold weather.

This region has few forests. *Juniperus oxycedrus* L. (Prickly juniper), *J. excelsa* Bieb (Grecian juniper) from *Cupressaceae* (Cypress family), *Quercus pubescens* Wild (Downy oak), *Q. cerris* L. (Turkey oak), are sparsely found in this region, and *Rosa canina* L. (Dog rose) is frequent in the valleys. Inside the city, *Thuja orientalis* L. (Arborwitae oriental) from *Cupressaceae* (Cypress family), *Pinus* L. spp. (Pine), *Acer negundo* L. (Maple), *Elaeagnus angustifolia* L. (Olive Russian), *Fraxinus ornus* L. (Beech), *Betula pendula* Roth (Birch), *Cedrus libani* Rich (Cedar of Lebanon), *Ulmus minor* Miller (Elm), *Syringa vulgaris* L. (Lilac), *Populus tremula* L. (European aspen), *Salix* L. spp. (Willow) and *Robinia pseudoacacia* L. (Black locust) are planted in the parks and along the streets. *Morus* L. spp. (Mulberry), *Juglans regia* L. (Walnut), *Prunus* L. spp. (Cherry), *Armeniaca vulgaris* Lam (Wild apricot), *Amygdalus communis* L. (Almond), *Malus sylvestris* Miller (Apple) are found in the gardens as fruit trees. Species belonging to *Amaranthaceae* (Amaranth family), *Compositae* (Daisy family), *Chenopodiaceae* (Goosefoot family), *Urticaceae* (Neetle family) and *Poaceae* (Grasses) families are found near the streets and in abandoned areas. *Dactylis glomerata* L. (Orchard grass), *Parietaria* L. spp. (Pellitory), *Urtica* L. spp. (Nettle), *Rumex squtatus* L. (Red sorrel), *R. acetosella* L. (Sorrel), *Chenopodium album* L. (Lamb's quarter), *Medicago sativa* L. (Alfalfa), *Trifolium* spp. L. (Clover), *Plantago lanceolata* L. (English plantain), *Plantago major* L. (Greater plantain), *Anthemis cretica* L. (Mayweed), *A. arvensis* L. (Dogfennel), *Artemisia* L. spp. (Wormwood), *Stipa* L. spp. (Needle grass), *Phleum montanum* (L.) Koch (Timothy), *Festuca valesiaca* L. (Fescue), *F. ovina* L. (Sheep fescue), *Poa pratensis* L. (Kentucky bluegrass), *P. nemoralis* L. (Wood bluegrass), *P. bulbosa* L. (Bulbous meadow grass), *Bromus tectorum* L. (Brome), *Agropyron cristatum* (L.) Gaertner (Crested wheatgrass), *Hordeum* spp. L. (Barley) and *Agrostis alba* L. (Redtop) are also found in this region, and species belonging to *Cyperaceae* (Sedge family) and *Juncaceae* (Rush family) are found at the swamp areas. Agriculture of *Triticum acstium* L. (cultivated wheat), *Avena sativa* L. (cultivated qats), *Hordeum vulgare* L. (cultivated barley), *Helianthus annuus* L. (common sunflower), *Beta vulgaris* L. (Beet) *and Medicago sativa* L. (Alfalfa) are grown in the fields.

In this study, a gravimetric method and a Durham sampler were used. The Durham sampler was placed on the roof of a building, at a height of 15 m above ground level. The slide was covered with glycerin jelly mixed with basic fuchsin.[17] The slides were changed daily, and covered with 20 x 20 mm lamella after being heated. Identification and counting of the pollen were performed under a light microscope and the number of pollen per cm^2 was determined. The pollen were collected daily between March 13 and October 31. All procedures were performed by the same botanist.

Skin test of patients

Eighty-nine patients, suffering from seasonal rhino-conjunctivitis and/or seasonal asthma, who presented to the Allergy Polyclinic of Kayseri Erciyes University, Faculty of Medicine, Department of Pulmonology, between 1996 and 1997, were recruited. The mean age of the patients (male: 25, female: 64) was 24.3 ± 4.1 years (range 18-41 years). Informed consent was obtained from all patients. Subjects were excluded from the study if they had received or were undergoing immunotherapy and/or using systemic corticosteroid treatment, or were suffering of one of the following: perennial rhinitis or asthma, pregnancy or lactation, malignancy or other severe systemic diseases. Patients were not allowed to use medicines with antihistaminic effect for at least 1 week before each test. Intracutaneous prick tests with pollen extracts were performed on the patients[17] using 1 mm tip disposable prick lancets. We used only commercially (Stallergenes SA, Antony, France) available aqueous solutions of allergens produced according to international guidelines for allergen preparation.[18] Histamine phosphate (1/200 g/ml)[20] was used as a positive control, and diluent (glycerine, 50%), as a negative control. All extracts were kept at 4°C and handled as recommended by the manufacturer. All vials were discharged when expired. All tests were performed with the same allergen throughout the study. Potency was expressed in micrograms of allergen per milliliter. The skin tests were located on the back of the patients and the injections were spaced 2 cm apart to avoid false positive reactions. The largest weal diameter was assessed 10-15 min-

utes after injecting the antigen. The size of each reaction was measured with a transparent millimeter ruler. Wheal and flare measurements were taken in two perpendicular directions. Both the largest and smallest diameters were recorded, added up and divided by 2. A positive reaction was defined as a weal ≥ 3 mm in the absence of a reaction to the diluent and in the presence of a positive reaction to histamine phosphate. Reactions were ranked as follows: 3-5 mm was 1+, 5-7 mm 2+, 7-10 mm 3+, 10 mm and higher 4+. To prevent interobserver variability, one doctor with special training in allergy, performed all skin tests.

RESULTS

Pollen of 43 taxa were determined in the Kayseri atmosphere between March and October. The period of time and the percentages are given in Table 1. The total amount of pollen per cm^2 was 1,330.8 in 1996 and 1,182.5 in 1997 as shown in Table 1.

Tree and shrub pollen were the most commonly observed pollen, representing 63.2% in 1996 and 50.3% in 1997. The most commonly determined taxa were *Pinus, Poaceae, Chenopodiaceae/Amaranthaceae, Cupressaceae, Populus* and *Quercus* in decreasing order. The pollen with the highest percentage among trees and shrubs were from taxa *Pinus* (Table 1). The second highest percentage of pollen among pollen of trees and shrubs were belonging to taxa *Cupressaceae* (Table 1). Graphics showing pollen variations of amount per cm^2 of these taxa were presented in Fig. 1. As seen in the figure, *Pinus, Poaceae* pollen reached their maximum in June, *Chenopodiaceae/Amarantha-*

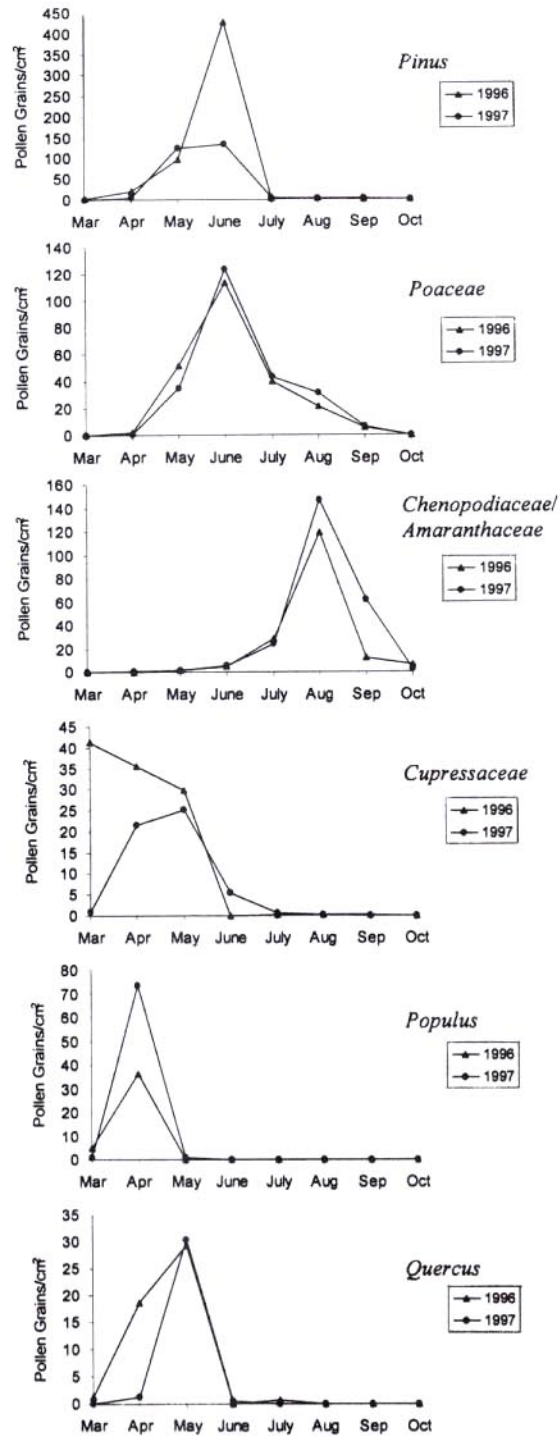

Fig. 1 Monthly trend of the most common pollen per cm^2 in the atmosphere of Kayseri (March- October 1996 and 1997).

Table 1 Percentage and total count of pollen and taxa per cm^2 in the atmosphere of Kayseri (March-October 1996 and 1997)

Group	Taxa	1996		1997	
		Total	%	Total	%
Tree and shrubs	*Abies*	0	0	0.3	0.02
	Acer	14.3	1.07	33.8	2.86
	Alnus	0.3	0.02	0	0
	Betula	11.2	0.84	19.6	1.66
	Carpinus	0	0	1.3	0.11
	Cedrus	7.8	0.59	8.3	0.7
	Cupressaceae	107.5	8.07	54.4	4.61
	Elacagnus angustifolia	0	0	2.8	0.24
	Fabaceae	1.1	0.08	8.6	0.73
	Fagus	1.1	0.08	6.5	0.55
	Fraxinus	8.3	0.62	6.5	0.55
	Juglans regia	8.6	0.65	9.8	0.83
	Morus	17.3	1.3	15.6	1.32
	Oleaceae	2.4	0.18	2.3	0.19
	Pinus	552.6	41.7	270.4	22.9
	Pistacia	3.5	0.26	1.6	0.14
	Platanus	2.5	0.19	4.1	0.35
	Populus	41.3	3.1	75.3	6.38
	Robinia pseudoacacia	0	0	0.5	0.04
	Rosaceae	2.9	0.22	10.3	0.87
	Quercus	49.9	3.74	32.1	2.71
	Salix	5.8	0.44	20.1	1.7
	Sophora	0.8	0.06	10.3	0.87
	Vitis	0.8	0.06	0	0
	Total	**840**	**63.27**	**594.5**	**50.33**
Other herbs	*Apiaceae*	1.6	0.12	10.9	0.92
	Artemisia	12	0.9	6.3	0.53
	Asteraceae	7.1	0.53	13.1	1.11
	Brassicaceae	18.1	1.36	4.6	0.39
	Caryophyllaceae	0.6	0.04	3.3	0.28
	Centaurea	3.1	0.23	4.1	0.27
	Chenopodiaceae/Amaranthaceae	173.7	13	243.4	20.6
	Cyperaceae	3.8	0.29	3.7	0.31
	Galium	0.8	0.06	1.8	0.15
	Papaver	3.6	0.27	5.6	0.47
	Plantago	16.2	1.22	34.1	2.89
	Ranunculus	0	0	0.3	0.02
	Rumex	6.5	0.49	5.8	0.49
	Labiatae	2.5	0.19	0.3	0.02
	Typha	0.6	0.05	1.3	0.11
	Urticaceae	0.6	0.05	1.8	0.15
	Xanthium	2.1	0.16	2.8	0.24
	Total	**252.9**	**18.96**	**343.2**	**28.95**
Grasses	*Poaceae*	234.2	17.5	241	20.4
	Zea mays	0.3	0.02	0.5	0.04
	Total	**234.5**	**17.52**	**241.5**	**20.44**
	Unidentified	**3.4**	**0.25**	**3.3**	**0.28**
	TOTAL	**1,330.8**		**1,182.5**	

ceae pollen in August, *Populus* pollen in April and *Quercus* pollen in May in both 1996 and 1997. *Cupressaceae* pollen reached their maximum level in March 1996 and in May 1997.

A pollen calendar of Kayseri was prepared by taking the mean count per cm^2 of pollen of each taxon during the March-October period as presented in Fig. 2.

Prick test results related to allergens (pollen) and the reaction degree are given in Table 2. *Poaceae* and other herbs seem to have a high allergenicity. *Chenopodiaceae/ Amaranthaceae* is the most common cause of pollen allergy in Kayseri.

DISCUSSION

Ince[6] found in his study performed in Kirikkale that 87% of pollen derived from trees and shrubs, and in the study of Inceoglu,[5] this percentage was 75%. These authors suggested that this high percentage is due to the abundance of trees in the parks and gardens in these two cities, which is also true for Kayseri.

Cupressaceae pollen may be important allergens in March, April and May, the months that when they are observed more intensively (Fig. 1). Pollen of *Poaceae* gave positive reactions in 48-62% of the patients (Table 2). *Poaceae* was also found to be an important allergen in other aeropalynological studies performed in Turkey.[3,4,6,7] Most reactions to this allergen were found in June, but it was also effective in May, July and August (Fig. 1).

In the atmosphere of Kayseri, *Betula* pollen were observed intensively in the last week of April and the first week of May, *Fagus* pollen in the second week of May, *Quercus* pollen in the last week of April and May and *Fraxinus* pollen in the last week of March and the first week of April (Fig. 2). The pollen of these taxa may be effective on allergic patients at the times when they are observed intensively. *Acer* pollen were observed intensively in April, *Salix, Juglans regia* in the first weeks of May and *Populus* in the later weeks of March (Fig. 2). In the skin tests, we obtained 35% positivity to *Acer negundo* pollen, 37% to *Morus alba* pollen, 16% to *Juglans regia* pollen 38% to *Populus alba* pollen and 30% to *Populus tremuloides* pollen (Table 2).

Among herbaceous plants, *Chenopodiaceae/Amaranthaceae* produced most of the pollen in the atmosphere. Its pollen were observed in July, reaching a maximum level in August and lasted through September (Fig. 1). Skin tests performed with pollen of this family (*Kochia scoparia* L., Sahrad *Chenopodium album* L., *Chenopodium botrys* L., *Salsola* L. spp. gave positive reactions at maximum level and intensity (Table 2). Ince[6] stated in his study that pollen of this taxa were important allergens in July and August in the atmosphere of Kirikkale.

Climate and vegetation determine the level of exposure to pollen in a given region, both in quantity and pollen type.[19] This is evident in the medical literature, with different prevalence rates obtained according to geographic and climatic variations. *Parietaria* pollen was considered as one of the most common causes of allergic respiratory symptoms in the Mediterranean and Atlantic area. The presence of very high levels of these pollen in that atmosphere explains this fact.[12] In a study from Kuwait (desert climate) the three most prevalent sensitizing pollen were from *Chenopodium* (70.7%), *Bermuda grass* (62.9%), and *Prosopis* (62.7%).[20] In the skin tests we performed with herbaceous plants, *Artemisia vulgaris* L. pollen gave positive reaction in 66%, *A. annua* L. pollen in 61% and *Plantago lanceolata* L. pollen in 53% of the patients. In Kayseri, *Plantago* pollen can be effective in June and July, and *Artemisia* pollen in August, September, and October (Fig. 2). Some authors also stated that the allergenic effects of pollen of these taxa could be very strong.[7-10]

In a study from Spain, the highest airborne presence was for *Quercus* spp. (17%); followed by *Platanus* spp. (15%), Poaceae (15%), Cupressaceae (11%), *Olea* spp. (9%), *Pinus* spp. (7%), *Populus* spp. (4%), and *Plantago* spp. (4%). The most predominant pollen from January to April were tree pollen (*Cupressaceae, Alnus, Fraxinus, Ulmus, Populus, Platanus,* and *Morus*), which were also abundant in May and June (*Quercus, Olea,* and *Pinus* spp.). The grass pollination period showed a double curve, with the first peak occurring from February to April (8% of yearly grasses), and the second peak from May to July. Among allergenically significant weeds, the most notable was *Plantago*; in contrast, *Rumex, Urticaceae, Chenopodiaceae/Amaranthaceae*, and *Artemisia* spp. had very low atmospheric concentrations.[21] The most significant allergenic pollen were that of grasses, with a prevalence of positive prick

Table 2 Skin prick test results related with allergens (pollen) and their reaction degrees

Groups of plants	Pollen	n	Total + %	1+ %	2+ %	3+ %	4+ %
Poaceaea	*Cynodon dactylon*	89	57	19	14	14	10
	Poa campressa	89	51	21	8	10	12
	Poa pratensis	89	46	16	3	12	15
	Zea mays	89	46	23	6	9	8
	Avena sativa	79	55	15	20	13	7
	Dactylis glomerata	86	51	18	6	12	15
	Lolium multiforum	88	47	19	5	13	10
	Lolium perenne	89	55	24	4	7	20
	Phleum pratense	89	46	17	5	7	17
	Triticum aestivum	78	48	26	6	8	8
	Grass mix	89	62	28	8	12	14
Other herbs	*Xanthium strumarium*	89	53	27	15	11	0
	Rumex acetosella	89	49	29	17	2	1
	Rumex crispus	88	48	26	19	2	1
	Kochia scoparia	88	62	19	10	15	18
	Chenopodium album	89	60	14	9	26	11
	Chenopodium botrys	89	59	22	18	18	1
	Artemisia annua	89	61	38	14	5	4
	Artemisia vulgaris	84	66	37	14	12	3
	Urtica spp.	82	58	46	12	5	1
	Plantago lanceolata	77	63	33	16	3	1
	Salsola spp.	85	65	21	10	8	24
	Medicago sativa	85	65	30	10	16	3
	Trifolium pratense	86	74	44	25	5	0
	Brassica spp	80	60	40	15	5	0
Trees and shrubs	*Morus alba*	83	37	24	12	1	0
	Populus alba	82	38	32	5	1	0
	Populus tremuloides	80	30	23	6	1	0
	Acer negundo	81	39	29	9	2	0
	Juniperus occidentalis	79	5	5	0	0	0
	Juglans regia	74	16	13	2	1	0
	Fraxinus americana	74	46	20	13	9	4
	Corylus americana	87	33	21	7	4	1
	Platanus occidentalis	87	25	17	4	3	1
	Betula mix	76	45	27	15	1	2
	Tree mix	78	52	24	18	9	1

n: patients who were tested with each extract

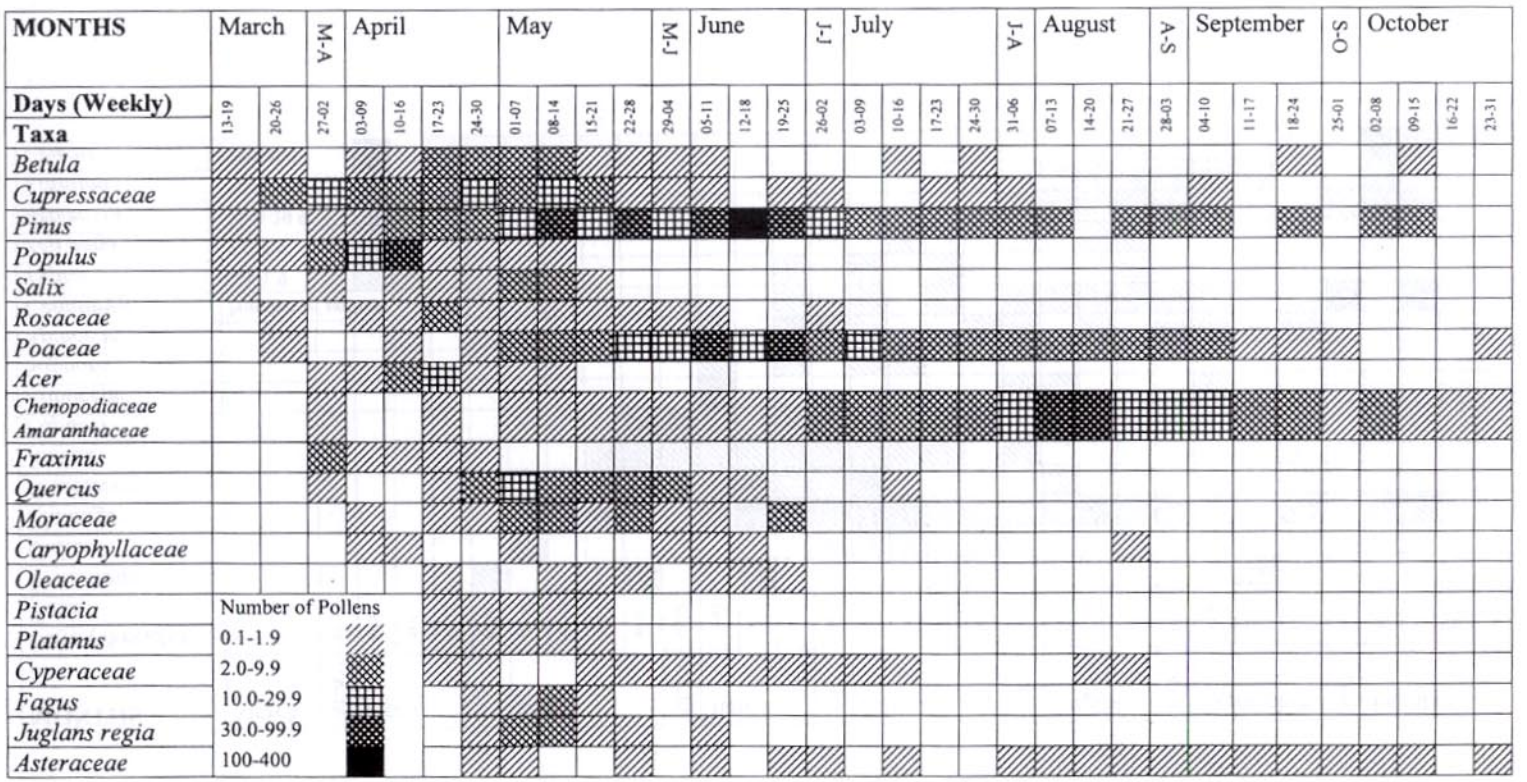

Fig. 2a The pollen calendar of Kayseri in 1996-1997.

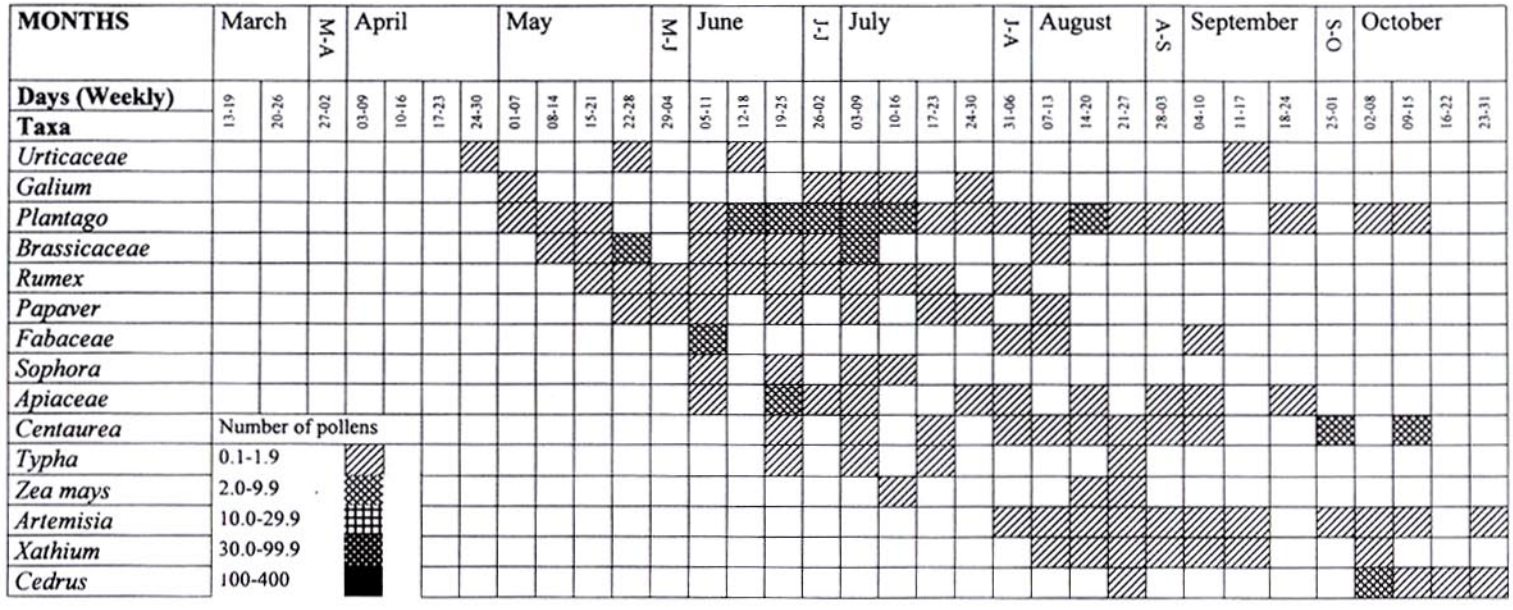

Fig. 2b The pollen calendar of Kayseri in 1996-1997.

test results of 94%, followed by *Olea europaea* (61%), *Plantago lagopus* (53%), *Platanus hybrida* (52%), and *Cupressus arizonica* (20%).

No reaction or only a small reaction was observed in the skin tests performed by *Pinus* pollen, which was the most common pollen in Kayseri.[1,8,16] *Pinus* pollen do not seem to be important allergens for our region. In the skin tests we performed with *Juniperus occidentalis* pollen (belonging to the *Cupressaceae* family, which was the second highest pollen percentage), 5% of the patients gave positive reactions. Other authors showed a higher positive reaction to *Juniperus* spp.[8,9]

Forty-five percent of the patients showed positive reactions to *Betula* pollen, and 46% to *Fraxinus americana* pollen. Chapman and Williams[10] determined that *Betula, Fraxinus, Quercus* and *Fagus* pollen gave positive reactions in 43-58% of patients. Ince[7] determined that pollen of these taxa were allergenic. Levetin and Buck[9] demonstrated that *Acer* and *Morus* pollen showed moderate reactions. *Acer* pollen may be effective in the second and third weeks of April and *Morus* pollen in the first, second and fourth weeks of May (Fig. 2). Some authors revealed that *Salix* and *Populus* pollen also had allergenic effects.[7-10] Pollen of these taxa can be the reason of allergic diseases in the weeks they were intensively found in the atmosphere (Fig. 2). *Platanus occidentalis* pollen gave positive reactions in 23% of the patients (Table 2). But it was detected only minimally in the atmosphere of Kayseri. In a study from another part of Turkey, sensitivity to *Poaceae* was the most com-mon (69%), and among them positivity to *Anthoxanthum odoratum* was 45%. In weeds *Artemisia vulgaris* sensitivity was 24%. Among trees, the *Oleacea* family which is a characteristic species for the Mediterranean region, had the highest sensitivity (22%) in pollen allergic patients.[22]

Pollen of *Rumex* spp., *Urtica* spp., *Xanthium strumarium, Brassica* spp., *Medicago sativa, Trifolium pratense* taxa showed positive reactions in most of the patients (Table 2), but they were not detected in the atmosphere of Kayseri. *Medicago sativa* and *Trifolium pratense,* which were pollinated by the way of insects, had a high allergenic potential. But pollen of these taxa were also not detected in the atmosphere of Kayseri. *Medicago sativa* was cultured in Kayseri and *Trifolium pratense* was a plant of meadow and pasture. Pollen of this plant can cause allergy if direct contact occurs. Other authors also demonstrated allergenic effects of these plants.[8,9,11]

In conclusion, by preparing an airborne pollen calendar of Kayseri, it was found that the date of onset of the pollen season could be specified early enough to be useful for medical practice. Modern pollen spectra from pollen traps have provided useful data for the interpretation of pollen assemblages and detailed information on the nature of the annual pollen distribution of the vegetation of Kayseri. Pollen of the local horticultural plants were the main sensitizing allergens among allergic patients in this environment. Nevertheless the skin test data did not really correlate with the aerobiologic data, probably because skin test reactivity is affected by many different factors, like the allergenicity of the pollen and not just by the ambient exposure.

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