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The partial contribution of specific airborne pollen to pollen induced allergy

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Abstract

The air that we inhale contains simultaneously a multiple array of allergenic pollen. It is well known that such allergens cause allergic reactions in some 15% of the population of the Western World. However little is known about the quantitative aspect of this phenomenon. What is the lowest concentration of pollen that might trigger allergic responses? As people are exposed to heterogeneous and variable environments, clarification of the partial contribution of each of the major airborne pollen allergens and determination of its role in invoking allergy are of prime importance. *Objectives*: (1) Assessment of a possible correlation between the concentration of airborne pollen and incidence of allergy. (2) Estimation of the lowest average concentrations for various species of airborne pollen that elicit allergic symptoms when exceeded. (3) Determination of the extent of the variations in manifestation of allergy symptoms that can be explained by fluctuations in the concentration of individual species of airborne pollen. *Methods*: The study was conducted during 14 months with a rural population in Israel. The participants completed a detailed questionnaire and were skin prick tested with the common airborne allergens. The appearance of clinical symptoms, i.e. nasal, bronchial, ocular or dermal, were reported daily by the patients. Concentrations of the airborne pollen and spores were monitored in the center of activity of the residents during one day every week, using three 'Rotorod' pollen traps. The pollen grains were identified by light microscopy. *Results*: The pollen spectrum was divided into time-blocks presenting the main pollination periods of the investigated species. The correlation between the concentration of airborne pollen of the relevant species and the clinical symptoms of the patients was determined for each time block. The correlation differed for different clinical symptoms and for different pollen allergens. Highest correlation with airborne pollen counts was found for patients with nasal and bronchial symptoms. The onset of the clinical symptoms by sensitive patients started, in each of the relevant groups, once the weekly average concentration of the airborne pollen crossed a threshold level. Under the limitations of the present study, this level was estimated to be 2–4 pollen m^{-3} air for olive, 3–5 pollen m^{-3} air for grasses, 4–5 pollen m^{-3} air for *Artemisia,* 10–20 pollen m^{-3} air for pecan and 50–60 pollen m^{-3} air for cypress. *Conclusions*: Fluctuations in specific airborne pollen grains explained up to 2/3 of the variation in clinical allergy responses. Those were: 69% of the variation for cypress (March–April), 66% for the grasses (March–April), 49% for the pecan (May–June) and 62% for *Artemisia* (Autumn).

1. Introduction

Pollen grains of several plant species evoke allergy responses. However, in order to clarify the detailed effects of each species, and to quantify such relationships, two parameters have to be established:

(1) The threshold concentrations of airborne pollen that provoke allergy symptoms when exceeded.
(2) The contribution of each of the specific pollen allergens to the eventual manifestation of clinical symptoms by the patients.

A positive correlation between clinical symptoms, by allergic patients, and concentrations of airborne pollen and spores was reported by several investigators (Keynan et al., 1991a; Negrini et al., 1992; Charpin et al., 1993; Geller-Bernstein et al., 1996; D'Amato et al., 1996; Minero et al., 1998; Florido et al., 1999; Charpin, 2000; Kontou-Fili et al., 2000; Frenz, 2001; Subiza, 2001; Carinanos et al., 2002). In most cases the annual or seasonal incidence of the clinical symptoms were correlated with the variations in the total pollen counts. However, the summation of all monitored airborne allergens seem to weaken any correlation that might have been compiled for individual allergens. A correlation between pollen rain and allergy symptoms should be more accurate if the counts of specific pollen were continuous, limited to the specific pollination season and calculated for levels above the threshold values.

In view of such a challenge we attempted to tackle the following questions:

- What is the time course of airborne pollen concentrations in the close vicinity of a defined population of patients?
- What is the time course of the manifestation of allergy symptoms by such patients?
- How much of the variation in expression of allergy symptoms can be explained by fluctuations in the concentration of airborne pollen of individual species?

2. Patients and methods

2.1. *The site*

Netzer Sireni is a Kibbutz (a communal rural village) in the Coastal Plain of Israel. It is located some 11 km east of the Mediterranean coast. The altitude is some 70 m. The mean daily summer temperature is 25–26 °C and the mean daily winter temperature 13–15 °C. The mean day-time annual relative humidity is 60–75%. The mean annual rainfall (30 years average) amounts to 515 mm, and is concentrated over the period between November and April. In this kind of village the population travels relatively little, since many of the inhabitants work on the site, and many of the cultural and social activities also take place there.

The village is surrounded by citrus, avocado, pecan, mango, and olive orchards, as well as by grain fields. The inhabitants live in one story houses that are surrounded by gardens, rich in various ornamental and fruit plants. Large areas of the spaces between the houses, and around the village centers of activity, are covered with lawns, planted with various species of turf grasses.

2.2. *Seasonal time blocks*

The investigation was conducted during 14 months. In order to avoid interference of irrelevant data, in the calculations of the correlation between airborne pollen and allergy, we divided the recorded data into the pollination seasons of the major allergens. The following seven time blocks were thus created and investigated: March–April, March–May, March–June, April–May, April–June, May–June, and September–October.

2.3. *Pollen and spores monitoring*

Pollen and spores were monitored using 'Rotorod' samplers (Model 1987, Sampling Technologies, Inc., Los Altos Hills, CA, USA). In order to obtain a representative count at the main sites of the inhabitants activity, three samplers were used simultaneously. The sampling height was 80 cm above the ground. Measurements were made during one day (30 min per hour) every week (except for the16th and 53rd weeks) . The rods were stained with basic Safranine and the pollen and spores were identified and counted by light microscopy. Average pollen concentrations (number of pollen m^{-3} air) were calculated for each of the measurements, according to the manufacturer's directives.

2.4. *Patients*

Out of 324 inhabitants of Netzer Sireni, whose atopic profile was determined before (see Rachmiel et al., 1996), 69 atopic patients have agreed to participate again in the present study, 33 men and 36 women. Twelve of the patients were below the age of 17, 46 were between 17 and 50, and 11 were above the age of 50. The participants completed a detailed questionnaire and were skin prick tested (SPT) with the common airborne pollen and four mould spore extracts (Center Laboratories, New York). The clinical symptoms, i.e. nasal, bronchial, ocular or dermal, were reported daily by the patients and validated by the local nurse.

The patients were divided into groups based on strong SPT responses (wheal diameter exceeding 6 mm). The grass group (34 patients), pecan group (26 patients), the cypress group (22 patients), the olive group (21 patients), the *Artemisia* group (22 patients) and a group of strong responders to cypress, olive and pecan (COP) (18 patients).

2.5. *Statistical analysis*

Data analysis was performed using the Statistix 3.5 software package. Pearson's correlation was used to establish the dependence (r) of the clinical symptoms on each of the pollen allergens. This analysis was used to explain the percentage of the variance in the fluctuations in the clinical symptoms that can be explained by the concurrent changes in pollen counts.

For additional methodological details see Kosman et al. (1994, 1997) and Garty et al. (1998).

3. Results

3.1. *Airborne pollen*

Combined count of all airborne pollen showed a major peak (1627 pollen m^{-3} air) between March and May (Figure 1). High counts were contributed at that time by pecan (1165 pollen m^{-3} air), by cypress (560 pollen m^{-3} air) and by various species of the Urticaceae (785 pollen m^{-3} air). A minor 'hump' in the annual curve, mostly contributed by pollen of *Artemisia* and *Casuarina*, was identified during the autumn (September–October).

During the summer months (July–August) and during the early winter (November–December) the pollen rain in Netzer Sireni was relatively low, with the mean total pollen concentrations in the range of 9–28 pollen m^{-3} air.

3.2. *Specific pollen counts*

Pollen of some plant species (Cupressaceae, *Carya*, *Olea*, *Artemisia* and *Casuarina*) was conspicuous during relatively short time periods (Figures 2 and 3). Pollen of grasses, pollen of various species of the Chenopodiaceae and Amaranthaceae, and pollen of *Parietaria* and *Eucalyptus* (Figures 3 and 4) was present throughout the year.

The spring peak actually started during the last week of February 1997 and was dominated by pollen of various species of the Cupressaceae (*Cupressus, Thuja* and *Callitris*), that constituted 52% of the total count. The highest pollen count was recorded during the first week of April.

Towards the end of May the total pollen count was still high (up to 1265 pollen m^{-3} air), but the species composition has changed with pecan pollen replacing the cypress as the dominant airborne species. During the peak flowering season of pecan trees, i.e. towards mid-May, pecan pollen accounted for almost 95% of the total pollen count in

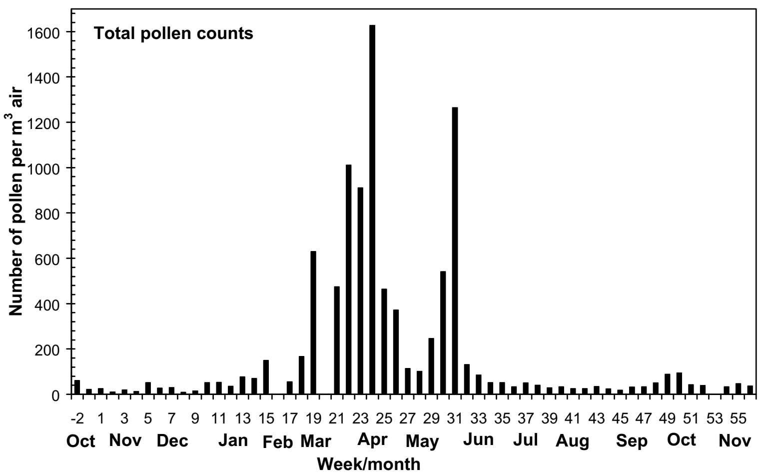

Figure 1. Time course of total airborne pollen counts.

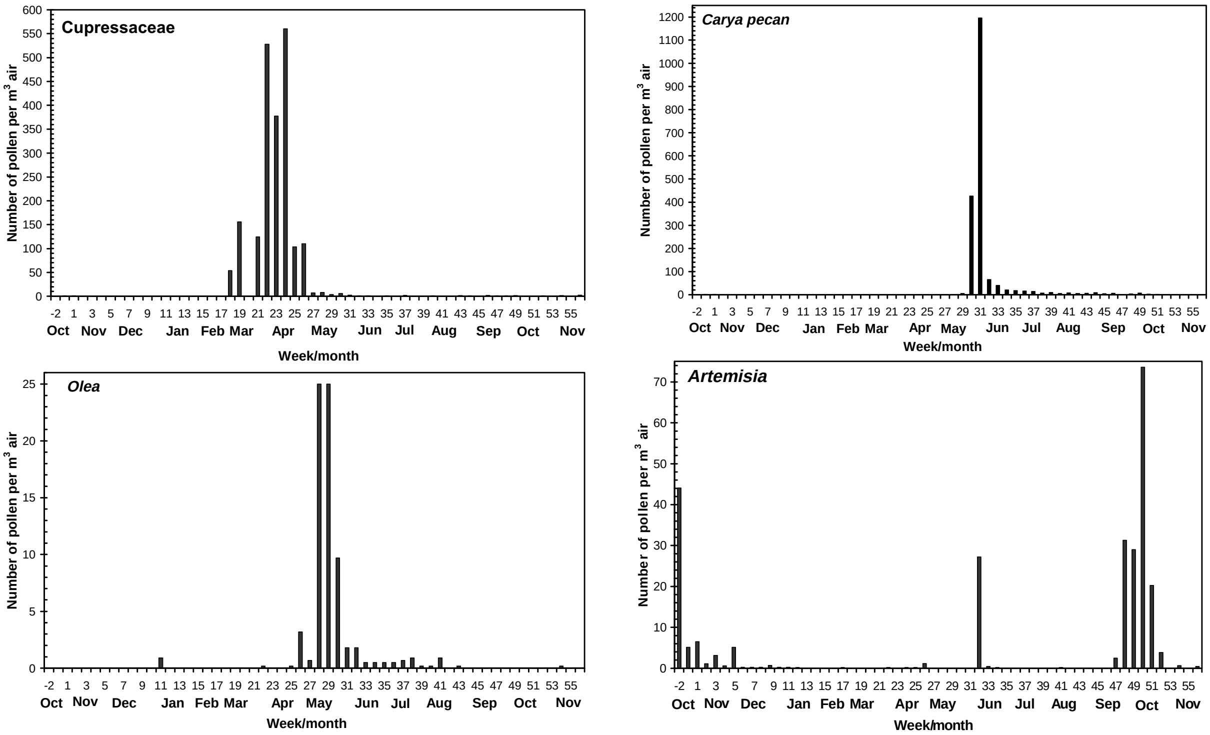

Figure 2. Time course of airborne pollen counts of Cupressaceae, *Carya*, *Olea* and *Artemisia*.

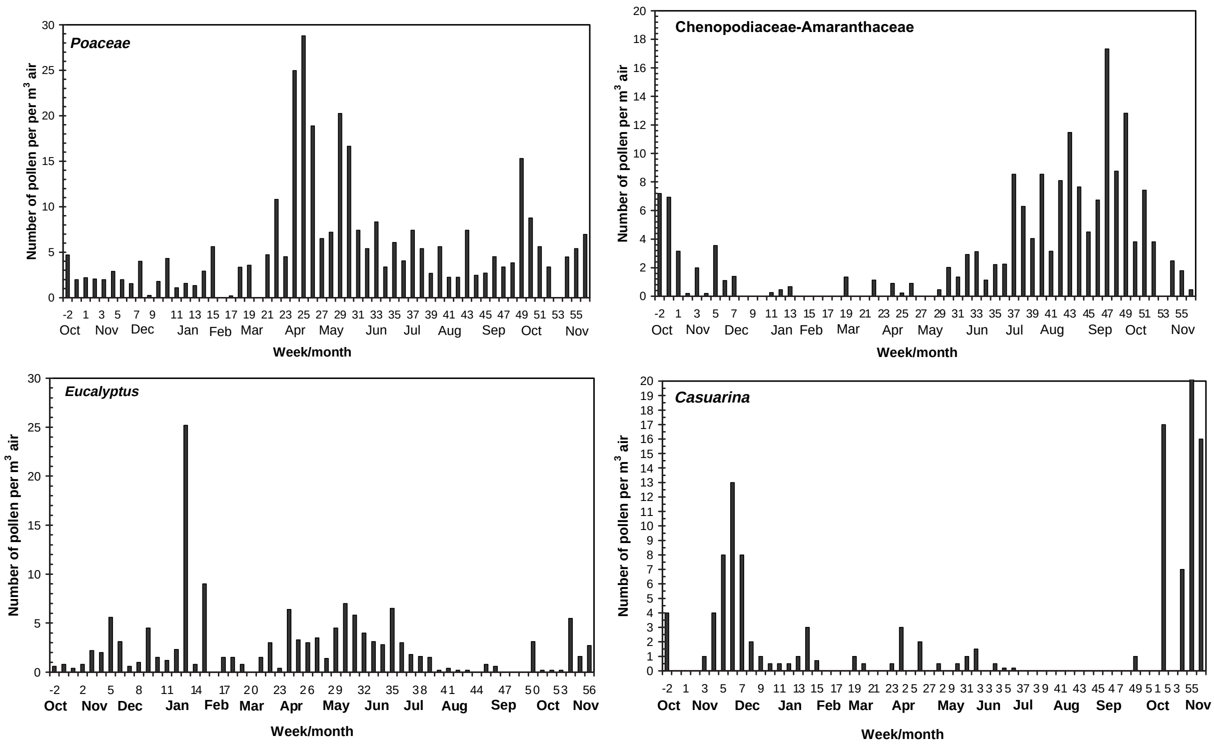

Figure 3. Time course of airborne pollen counts of Poaceae, Chenopodiaceae–Amaranthaceae, *Eucalyptus* and *Casuarina*.

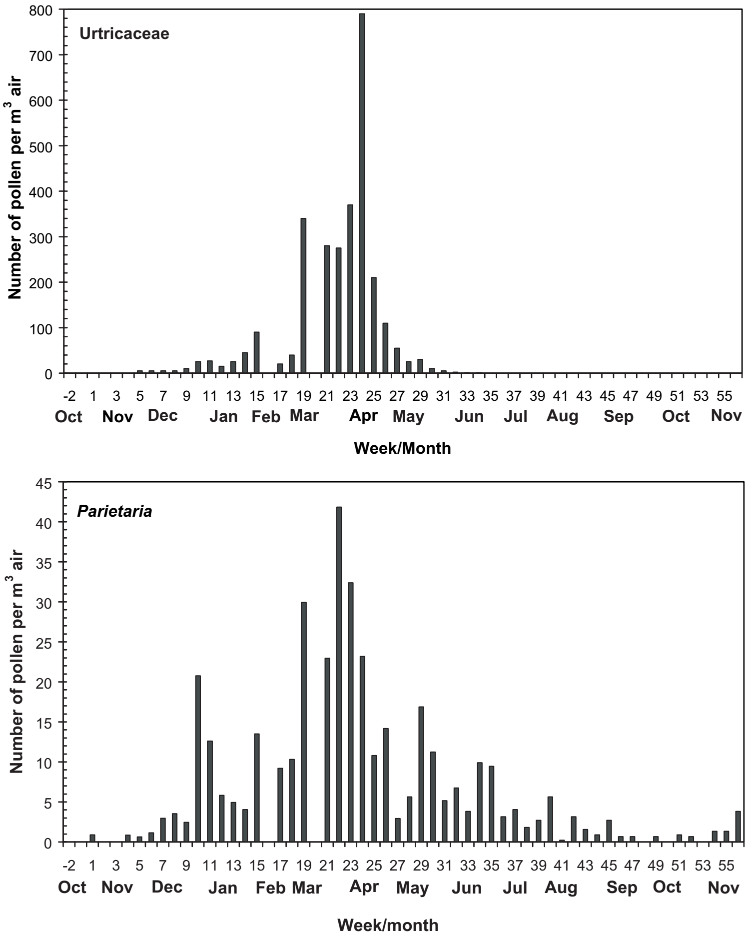

Figure 4. Time course of airborne pollen counts of non-specified Urticaceae and of *Parietaria*.

Netzer Sireni (up to 1165 pollen m^3 air). Airborne pecan pollen counts have dropped remarkably towards the end of May, but a small number of pecan pollen grains was still monitored till the beginning of October.

Grass pollen grains were recorded throughout the year (Figure 3). However, the relative contribution of grasses to the total pollen count was rather small, as compared with the contribution of the cypress and pecan trees. During the peak flowering season of the grasses (April–May) their weekly contribution to the total pollen count in the Kibbutz was only some 5%. A conspicuous rise in grass pollen counts was observed during the autumn.

During the entire year of the present investigation, the contribution of olive pollen to the total airborne pollen count was minor (up to $\sim$25 pollen m^{-3} air), in spite of the fact that many olive trees grow in that area.

The autumn rise in pollen count was much smaller as compared with that of the spring peak. During the first week of October the total pollen count reached a mean value of only 94 pollen m^{-3} air. Nevertheless, it should be stressed that almost 70% of the airborne pollen rain during that period was comprised of *Artemisia* pollen, apparently of *Artemisia monosperma* (Keynan et al., 1991b). It is interesting to note that a short peak of *Artemisia* pollen was found also during the last week of May and the first week of June. This period is out of the flowering season of the local wild species of *Artemisia*, and the appearance of such pollen can be attributed only to *Artemisia arborescens*, a herb that was apparently introduced into the Holy Land by the crusaders.

A peak of *Casuarina* pollen counts (Figure 3) was observed during autumn and early winter and followed the pattern of *Artemisia*.

Pollen of the Urticaceae was recorded almost throughout the year. Peak flowering was observed during the first week of April when *Urtica* reached concentrations of 785 pollen m^{-3} air (Figure 4) and constituted almost half of the total pollen count. Airborne pollen grains of *Parietaria* were monitored throughout the year but peaked during March and April. At that time *Parietaria* pollen comprised only some 4% of the total pollen count. Because of the morphological similarity of pollen of *Parietaria* and pollen of various species of the *Urtica,* some misidentifications might have occurred especially during the period of overlapping flowering.

Pollen of various members of the Chenopodiaceae and Amaranthaceae exhibited a distinct peak during the summer and autumn months (July–October) though relatively high concentrations of such pollen were found throughout the year.

The concentration of airborne spores (Figure 5) showed smaller seasonal variations as compared with those of the pollen. Nevertheless, a peak can be detected for *Alternaria* and *Stemphylium* during the summer months (May–July) and for *Pithomyces* during the autumn (October–December).

3.3. *Manifestation of clinical symptoms and correlation with specific pollen counts*

The main allergic responses were nasal symptoms, with almost half of the participants (46.3%) complaining during April (Figure 6). Respiratory symptoms were less common and reached some 18.9% of the population during the third week of March. Most of the ocular complaints (23.3%) were recorded during mid-April the third week of May.

Complaints of patients with all three types of allergy symptoms were lowest during the summer. During the second week of August only 7.6% of the patients complained of nasal symptoms, 3.6%

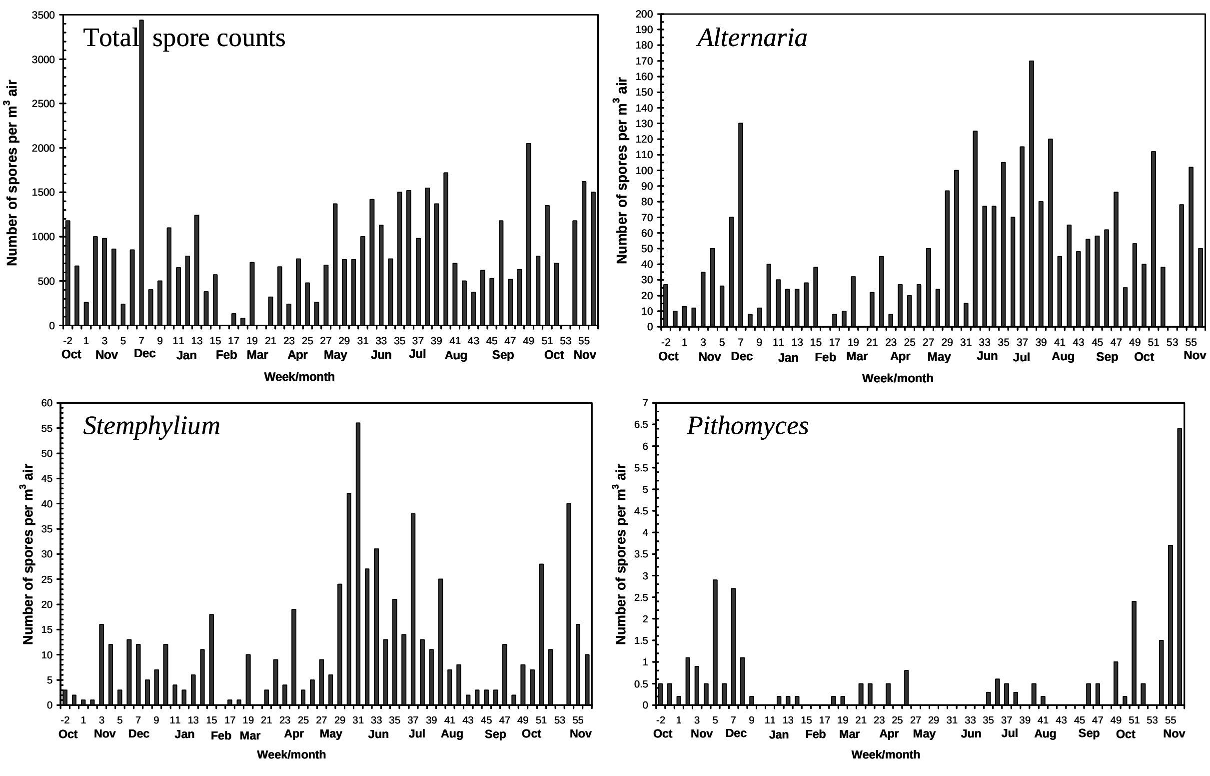

Figure 5. Time course of the total airborne spore counts and that of the spore counts of *Alternaria, Stemphylium* and *Pithomyces.*

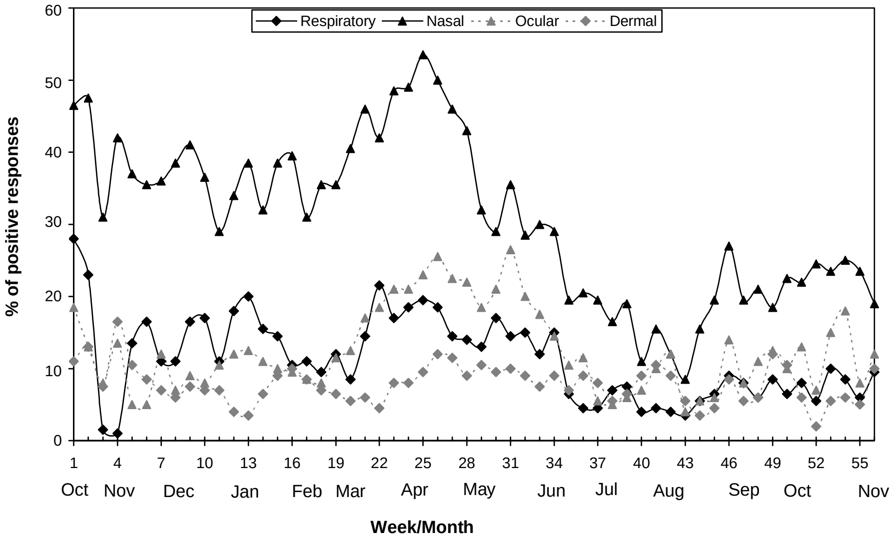

Figure 6. Time course of different clinical responses of allergic patients.

of ocular symptoms and 2.8% of respiratory symptoms. This corresponded well with the low concentrations of all airborne pollen that were monitored during that period.

As stated before, the patients were divided into groups according to their clinical symptoms and to the relevant allergens: The correlation was computed for various time blocks. An example of the variation in allergy symptoms that is explained by the simultaneous changes in airborne pollen counts [r^2(%)] during the relevant time-blocks is presented in Table 1.

Initiation of the clinical symptoms started, in each of the relevant groups, once the concentration of the airborne pollen crossed the species specific threshold level and ended when the concentration dropped below that level (see Figure 7). This was estimated to be 2–4 pollen m^{-3} air for olive, 3–5 pollen m^{-3} air for grasses, 4–5 pollen m^{-3} air for *Artemisia*, 10–20 pollen m^{-3} air for pecan and 50–60 pollen m^{-3} air for cypress (Waisel et al., 2003).

The relevant results are as follows:

The cypress group included 22 symptomatic patients, with only 3 of them monosensitized. Patients' complaints started during February, when the cypress pollen counts reached some 50 pollen m^{-3} air. The clinical responses changed during the season. A high correlation between the concentration of the airborne cypress pollen and respiratory complaints of the patients was established for the March–April period ($r = 0.83$; $r^2(\%) = 69\%$). However, when the time-block was expanded (March–June) more of the nasal than bronchial complaints corresponded to the variations in airborne cypress pollen load.

The pecan group included 26 patients with 15 of them being symptomatic. These included the 6 patients that were allergic to pecan as well as to olive and 7 of the patients that were allergic to cypress, olive and pecan (COP). The complaints of the pecan group patients started early in May when the concentrations of airborne pecan pollen reached 10–20 pollen m^{-3} air. Most of the clinical complaints of the pecan group were for respiratory symptoms [$r^2(\%) \sim 50\%$] and only few of the patients complained about ocular or nasal symptoms.

The correlation between the concentration of airborne pecan pollen and allergy symptoms of the pecan group was moderate and explained only 59% of the variance even during the peak of the pecan flowering season. Expansion of the period beyond May reduced the explained variance considerably.

The clinical symptoms of the 34 patients sensitive to grass pollen were relatively moderate. However, many of the patients developed clinical responses even when the average concentration of the grass pollen was as low as 3–5 pollen m^{-3} air. An example is presented in Figure 7. The correlation between the allergy responses of the grass

Table 1. Percent of the variance of three clinical symptoms that are explained by the variations in pollen counts (total or specific) in each of the investigated groups

Group	Time-block	Symptoms		
		Nasal	Ocular	Bronchial
Cypress vs. total pollen	March–April	41%	52%	**76%**
	March–June	25%	ns	ns
Cypress vs. cypress pollen	March–April	23%	30%	**69%**
	March–June	**48%**	12%	16%
Olive vs. total pollen	March–April	**56%**	45%	55%
	March–June	ns	ns	**35%**
Olive vs. olive pollen	March–April	ns	5%	ns
	March–June	ns	ns	ns
Pecan vs. total pollen	March–April	ns	50%	**59%**
	March–June	ns	27%	**41%**
Pecan vs. pecan pollen	March–April	ns	ns	ns
	March–June	2%	**5%**	2%
Grasses vs. total pollen	March–April	27%	48%	**58%**
	March–June	ns	ns	**18%**
Grasses vs. grass pollen	March–April	**66%**	45%	35%
	March–June	**31%**	25%	24%
Artemisia vs. total pollen	September–October	ns	ns	ns
Artemisia vs. *Artemisia pollen*	September–October	ns	ns	**62%**

Best correlation in bold; ns – not significant.

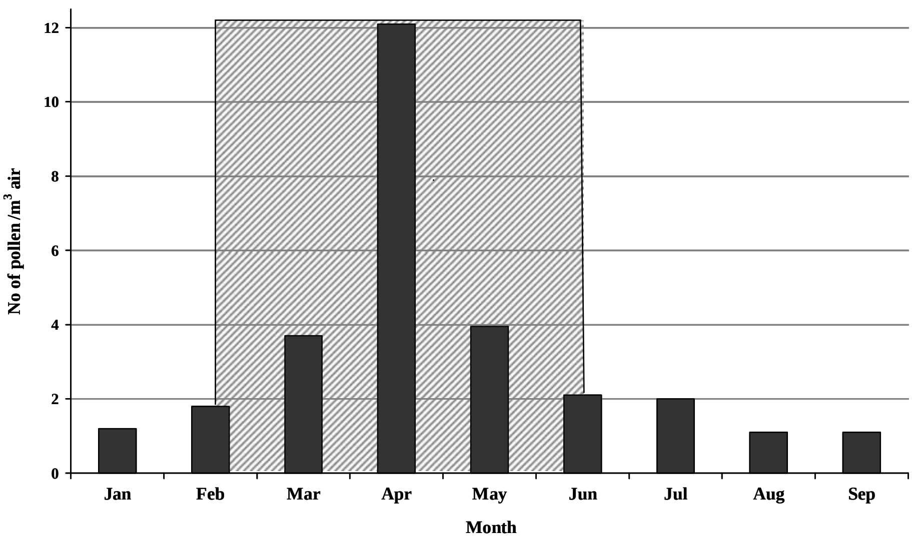

Figure 7. Determination of threshold pollen concentrations. The time course of airborne grass pollen load and the 'window' when clinical responses were presented. Pollen concentrations outside the 'window' were below the threshold value and did not affect the patients.

group and the recorded concentrations of airborne grass pollen varied for the different clinical symptoms. The fluctuations in airborne grass pollen explained 66% of the variance in nasal complaints, during the peak flowering season of the grasses (March–April), but less than half of the variations

Table 2. Percent of the variance of three clinical symptoms of the COP group that are explained by the variations in pollen counts of the respective allergen

Group	Time-block	Symptoms		
		Nasal	Ocular	Bronchial
COP vs. total pollen	March–April	44%	72%	**83%**
	March–June	36%	31%	**53%**
COP vs. cypress pollen	March–April	25%	42%	**76%**
	March–June	36%	28%	**66%**
COP vs. olive pollen	March–April	ns	ns	ns
	March–June	ns	ns	ns
COP vs. pecan pollen	March–April	ns	ns	ns
	March–June	ns	ns	ns
COP vs. grass pollen	March–April	34%	**35%**	30%
	March–June	22%	**34%**	29%

Best correlation in bold.

in ocular or bronchial symptoms. The correlation decreased significantly when the time-block was expanded beyond the peak flowering season (cf. Tables 1 and 2). Still, this denotes a considerable level of incidence and might be the result of the perennial nature of grass pollen distribution in that kibbutz.

The olive group included 21 patients, with nine of them monosensitized to olive. In general patients developed clinical symptoms even when olive pollen concentration was lower than 4 pollen m^{-3} air. It is interesting to note that variations in airborne olive pollen concentrations showed a weak correlation with the clinical responses.

Twenty two patients comprised the *Artemisia* group. In general patients have developed clinical symptoms even when *Artemisia* pollen concentration was as low as 4–5 pollen m^{-3} air. Variations in *Artemisia* pollen counts explained some 62% of the variance of the bronchial complaints of the *Artemisia* group during the autumn (September–October). This is the main flowering season of the native *Artemisia monosperma*. No correlation was found between the *Artemisia* pollen counts and the ocular or nasal symptoms but this was one of the few allergens that showed some correlation with dermal symptoms.

Higher values of explanation, were obtained for the effects of cypress pollen during the pollination period on the patients of the COP group, meaning that polysensitization, and probably other factors as well, exacerbated the clinical symptoms of those patients (cf. Charpin and Vervolet, 1996, 1997; Vieira et al., 1998; D'Amato, 2002).

Up to 83% of the variance in bronchial complaints of the COP group,were explained by the variations in the total pollen counts during March-April. Only some 76% of the variance was explained by the cypress pollen counts and 30% by the grass pollen counts (Table 2). During that period the total pollen counts explained 72% of the ocular symptoms of the COP group but only 42% of the variance due to exposure to cypress pollen and 35% by exposure to grass pollen.

The concentrations of airborne spores in Netzer Sireni varied throughout the year. However, fluctuations in airborne spore counts were relatively small as compared with those of the pollen.

3.4. *The intensity of the symptoms*

Allergy should not be assessed only by the mere appearance of clinical symptoms but also by their intensity. The intensity of the individual symptoms also varied during the year. Severe nasal symptoms were reported from November till May and the mildest during July–August. Strong ocular symptoms, especially by patients of the COP group, were reported from March till May. When the time-block was expanded, e.g. when the correlation was calculated for the period between March and June, the variations in total pollen counts explained lower percentage of the clinical symptoms.

No seasonal pattern was found for conspicuous dermal symptoms. However, the incidence of such symptoms was generally low (~10% of the participants).

4. Discussion

Two complex questions were addressed in the present investigation: (a) what is the threshold concentration of airborne pollen and spores that elicit allergy symptoms? (b) is it possible to distinguish the effects of specific allergenic pollen from that of the total pollen rain and how do we scale the partial weighted effect of each of them during different seasons? Both parameters that are involved herein, i.e. the aerobiological effectors and the clinical manifestation, are highly variable. As the pollen rain is comprised of a multitude of pollen, in concert, the question is if we can get a reliable correlation with their clinical effects, and determine the 'allergenic risk' (cf. Thibaudon, 2003) for each species?

The airborne flora of Netzer Sireni was grouped into three types of plants:

Plant species with a short pollination period: Airborne pollen of such plants appear during a limited time either in the spring (e.g. *Urtica*, *Olea*, *Carya*, etc.) or in the autumn (e.g. *Artemisia* and *Casuarina*).

Plant species with an extended season of pollen release: These include plant species that belong to the Chenopodiaceae and Amaranthaceae as well as various members of the Cupressaceae. As different species of those families differ in their flowering seasons, such cross reacting allergens are airborne during prolonged periods. A good example for the contribution of various species to the seasonal pattern of the airborne flora is the case of *Artemisia*. The main source of such an allergen in the Coastal Plain of Israel is the native *Artemisia monosperma* that flowers during the early autumn (cf. Keynan et al., 1991b). However, a second species, i.e. *A. arborescens* was introduced to Israel, probably by the Crusaders, and naturalized around their fortresses. Nowadays this species, called 'Shiba' by the immigrants from North Africa, is used as an ornamental and herbal plant. *Artemisia arborescens* flowers during the spring and summer and its presence in residential areas expands the exposure period of allergic patients to *Artemisia* pollen by several months. The same phenomenon has developed by the introduction of various species of the Cupressaceae and by various species of the Anacardiaceae (Waisel et al., 1991).

Plant species with perennial pollen dispersal: Such plants include members of the Poaceae, of some Urticaceae (e.g. *Parietaria*),of *Eucalyptus* as well as of some additional but less conspicuous species. Though the concentrations of airborne pollen of such plant groups may fluctuate considerably throughout the year their allergenic effects seem to be perennial (Keynan et al., 1991a).

4.1. *Estimation of threshold levels*

Determination of the threshold level, i.e. pollen concentrations that elicit allergic symptoms, is a complex task that depends on the combined effects of several factors: the seasonal pollen release and dispersal, the load of allergens, the environmental quality, the exact time and duration of exposure and the genetic and physiological status of the patients. Still, an estimation of threshold values must be obtained before any meaningful correlation can be calculated (cf. Chapman, 2000; Waisel, 2001).

Though the presented data are based on averages, we certainly noticed that the patients of Netzer Sireni started to suffer already when the average values were rather low, e.g. 2–4 pollen m^{-3} air for plants with small pollen yields (olive, grasses and *Artemisia*) but higher values for the massive pollen sources (pecan and cypress).

4.2. *Prevalence*

A general correlation was reported before between the density of olive trees and the incidence of allergy symptoms during the flowering season (Geller-Bernstein et al., 1996; Filon et al., 1998). A high correlation between the level of airborne olive pollen and the incidence of rhinitis was reported for Jaen, Spain (Florido et al., 1999). A similar correlation was found between the allergy symptoms among flower growers and their exposure to the flowers that they have cultivated (Goldberg et al., 1998). In those cases the pollen concentrations were enormously high, and therefore the deduction of the threshold levels would have been insignificant. The threshold level (~400 pollen grains m^{-3} air), as reported by Florido et al., (1999) for the patients of the olive rich region of Jaen, is by two orders of magnitude higher than what the patients in Netzer Sireni were confronting. Olive has contributed only little to the total airborne flora of Netzer Sireni during the late spring. However, in view of the low threshold level for olive pollen, exposure to such pollen, even at the recorded concentrations, still could be crucial for sensitive patients.

4.3. *Clinical manifestation and airborne pollen concentrations*

Variations in clinical symptoms are well known (cf. D'Amato et al., 1996; Charpin and Vervolet, 1997; Siracusa et al., 1997; Geller-Bernstein et al., 2000; Dolors Riera et al., 2002) and were reaffirmed for Netzer Sireni (cf. Rachmiel et al., 1996). Apparently, the incidence of allergy symptoms increased during peaks blooming seasons and were less conspicuous when the pollen rain was near or at the suggested average threshold concentrations. Still, because of the heterogeneity of the involved airborne allergens, the quantification of the dose–response relationships between each of the allergens and each of the allergies remains an enigma (cf. Subiza 2001; Tobias et al., 2003; Atkinson and Strachan, 2004). Nevertheless, it should be noted that the correlation between the clinical manifestation and the total pollen load was higher than that for the individual plant species (even for those allergens to which the patients showed the highest sensitivity).

Threshold values are only one parameter which is needed to solve the cause and effect equation. The genuine magnitude of exposure of each of the individual patients is another obstacle in the search for a solution. For example, a good part of the leisure time of the people in the Kibbutz is spent outdoors on the lawns and their exposure there to grass pollen is probably high. However the actual exposure of such patients may differ by an order of magnitude according to their position on the lawn. People that recline on the lawns are exposed to the highest concentrations of grass pollen. Sitting people on the same site inhale only 10% of the grass pollen whereas standing people are exposed only to 1% of the pollen concentration that affects the reclining patients (Waisel et al., 2003). Exposure depends also on the time that the patients spend on the lawn, with the mowing situation and with the time of irrigation. Thus, the actual exposure of the patients to allergenic pollen is a critical issue for the calculation of a correlation between allergens and allergy. However, it is practically impossible to obtain such data for human patients.

Another interesting question is the difference between the magnitude of exposure as well as of the timing and the various clinical responses. For example, rhinitis develops before asthma in 2/3 of the studied cases in Perugia and appears 1 month earlier in the season (Siracusa, 2001; Siracusa et al., 2003). Spring symptoms peaked in May for rhinitis but were as late as June for asthma. So, when we discuss the effects of pollen on the occurrence of allergy, and the pollen thresholds, the specific symptoms must be defined.

Special attention should be given to the differences in explanation of the variance in allergy symptoms when dealing with total pollen counts, as compared with that of the pollen rain of specific species. High responses of the patients to a high total pollen rain may express the result of additive or combined effects rather than a direct response to an individual allergen.

Artemisia was an exception. A high explanation value was obtained for airborne *Artemisia* pollen, but a negligible value for the total pollen count. This might be connected somehow with the relatively lower occurrence of other diseases during that time of the year.

The complex relationships between the cause-pollen and result-allergy are not fully explained yet (cf. Atkinson and Strachan, 2004). However, the presented information contributes to the understanding of such relationships by providing a prelude to the knowledge of what might be the partial contribution by each of the allergens and of what is its estimated threshold level. Such information is crucial for attempts to reduce the incidence of pollen-induced-allergy by elimination of the most noxious allergenic plants from the surrounding area of the suffering people.

No doubt that results with smaller variations would have had a higher significance. However, such information can be reached only under two circumstances: (a) allergy responses should be based on monosensitized patients only and (b) that pollen counts will be obtained by personal pollen traps and continuously monitored. In spite of the fact that such goals are still far away they should be sought in the future.

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