

Occupational allergy to sunflower pollen

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Although the sunflower belongs to the Compositae family, allergy to sunflower pollen is not common. The occurrence of occupational allergy to this pollen species made it possible to characterize cross-reactive patterns of Compositae pollens in a human experimental model. A 24-yr-old man developed rhinitis and conjunctivitis over 5 yr of exposure to sunflower pollens, and asthma developed during the fifth year. All respiratory and ocular symptoms disappeared after he was removed from exposure, but he had a food allergic reaction while he was eating honey containing 30% sunflower pollens. The diagnosis of occupational allergy was based on history, skin prick tests and RAST to the pollen. Bronchial provocation tests performed after removal from exposure confirmed the sensitivity to sunflower pollens but there was no nonspecific hyperreactivity. It was found by RAST inhibition that sunflower pollen does not cross-react with other Compositae pollens tested or with sunflower seed. The honey that elicited food intolerance was demonstrated to inhibit significantly sunflower pollen RAST. (J ALLERGY CLIN IMMUNOL 75:70-4, 1985.)

With more than 30,000 species the Compositae family is the largest of the flowering plants, but it is the most uniform and compact of all orders of Phe-nogamous plants.¹ The large number of species is generally grouped into 14 tribes. Among these, many pollens have a particular allergenic importance (e.g., the short and giant ragweeds). Cross-reactivities among Compositae pollens have been extensively studied, and it was found that pollens from many species shared cross-reacting allergens.² The *Helianthus* or sunflower tribe is the most typical and primitive tribe of the family (Fig. 1).¹ The flowers are nearly always obviously insect pollinated, although often somewhat imperfectly so that some pollen is scattered. Moussitou and Cordero³ reported two patients who worked with sunflower seeds and presented allergic dermatosis directly related to the pollen of the plant. Wodehouse¹ indicated that both wild and cultivated plants of the sunflower tribe may cause hay fever when they are handled by susceptible individu-

Abbreviations used

RAST:	Radioallergosorbent test
FEV ₁ :	Forced expiratory volume in 1 sec
MMEF(R):	Maximum expiratory flow (rate)
V _{max} 50:	Maximum flow at 50% of total lung capacity
SG _{aw} :	Specific conductance
FVC:	Forced vital capacity
PD:	Provocative dose

als or occasionally when their pollens are inhaled. Little is known about the allergenicity of sunflower pollen and seed, and it is not known whether there are some cross-reactive allergens between these pollens and the seed and pollens of other tribes of the Compositae family.

The present article reports a case of occupational rhinitis and asthma induced by both inhalation and ingestion of sunflower pollen. An IgE-mediated reaction was sought and cross-reactivities were investigated in order to determine whether other Compositae pollens and sunflower seeds share allergens with sunflower pollens.

Case report

A 24-yr-old man started working in the Agricultural Department of the University of Montpellier. His job was

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aimed exclusively at the adaptation of sunflowers to dry climates and he was therefore in contact with sunflower plants, seeds, and pollens throughout the year. During the pollination of sunflowers in June, however, the amount of pollen inhaled was far greater than during the rest of the year. During the pollen season of the third year of his employment, he noted isolated episodes of hay fever that were relieved by antihistamines. During that year he had two or three episodes of rhinitis, but symptoms were not severe and the patient attributed them to a cold. During the next pollen season, the patient had severe rhinitis that required systemic corticosteroids, and he remained away from his work for 1 week. When he returned, his rhinitis reappeared, and he had to stop working for 2 wk. After the peak of the pollen season, the patient continued to present episodes of rhinitis while working with sunflower pollen. He avoided close contact with the pollen as much as possible and continued to work without major symptoms. During the pollen season of the fifth year of employment, he had to return to working with pollen and tried to prevent hay fever symptoms by means of pretreatment by oral antihistamine (diphenhydramine, 50 mg) and nasal sprays of beclomethasone and cromoglycate. His nasal symptoms were not severe, but he developed asthmatic attacks while working. Chest symptoms subsided during the weekend and reappeared as soon as he was in contact with pollen. He then ended his work situation definitively.

Six months after this episode, the patient ingested a teaspoon of honey and within minutes developed symptoms suggestive of food allergy. He complained of abdominal pain and experienced vomiting and diarrhea for 2 hr. No nasal or chest symptoms were observed.

There was no previous history of rhinitis or respiratory illness and no personal or family prevalence of allergy. The patient had never smoked. Physical examination results during asymptomatic periods were normal.

MATERIALS AND METHODS

Preparation of allergen extracts

Pollens. Pollens of the following species were extracted: sunflower (*Helianthemum annuus* L.; *Helianthus* tribe), short ragweed (*Ambrosia eliator*; *Ambrosieae* tribe), mugwort (*Artemisia vulgaris*; *Anthemideae* tribe), and dandelion (*Taraxacum officinale*; *Cichorieae* tribe). All these pollens were collected from wild or cultivated population plants over 3 years to avoid possible annual variations. They were microscopically analyzed for purity. Ten percent extracts were prepared in Coca's solution by stirring for 24 hr at +4° C. The supernatant was then passed through a sterilizing grade filter and aliquots were lyophilized for further tests.

Sunflower seeds. Sunflower seeds were prepared by the same procedure. Extraction was performed after crushing and without prewashing the seeds.

Honey. The honey that elicited the food intolerance was microscopically analyzed and found to contain 30% sunflower pollen among the total number of pollens present. A 50% honey solution prepared in Coca's solution was used

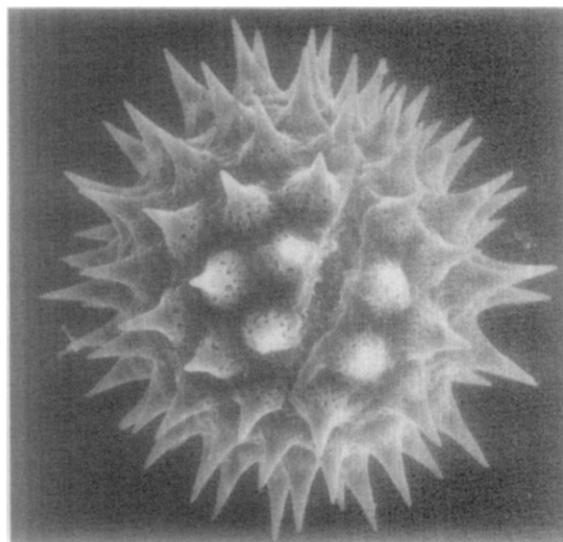


FIG. 1. Sunflower (*Helianthemum annuus* L.) pollen.

for the in vitro tests. Another honey that did not contain any sunflower or Compositae pollens was extracted by the same procedure. The latter extract was used as a negative control to test whether the viscosity of honey might interfere in the RAST.

Direct skin tests. Skin prick tests were performed with extracts specially prepared to investigate this clinical case and with allergens included in the battery screening. Pollen extracts were either standardized and tested in the form of 10,000 BU extracts or they were nonstandardized; the strength of the extract was 5%. Specially prepared extracts had a strength of 5% (w/v). Higher concentrations were not used, because nonspecific reactions may occur. To eliminate the possibility that the specially prepared extracts might contain nonspecific irritants, 10 patients with allergies without previous histories of pollen allergy were also tested. No positive reaction was observed. Prick test results were considered to be positive when the wheal elicited by the extract was at least as large as that induced by a 5% (w/v) codeine phosphate-positive control.

Radioimmunoassays. Total serum IgE was determined by the PRIST (Pharmacia Laboratories; Uppsala, Sweden). Specific IgE to pollen, seed, and honey extracts was measured by the classic RAST (Pharmacia Laboratories; Uppsala, Sweden). RAST discs were prepared according to the method of Ceska et al.⁴

Cross-allergenicity studies with other pollens of the Compositae family, with sunflower seed extracts, and with honey were determined by the RAST-inhibition procedure according to the method of Gleich et al.⁵ Briefly, a volume of 100 μ l of serial threefold dilutions of the test extracts were incubated with 100 μ l of the patient's serum for 1 hr at room temperature. A sunflower allergen disc was then added to each solution and incubated for 3 hr at room temperature. Binding of specific IgE to the sunflower disc was revealed with radiolabeled anti-IgE (Pharmacia AB; Upp-

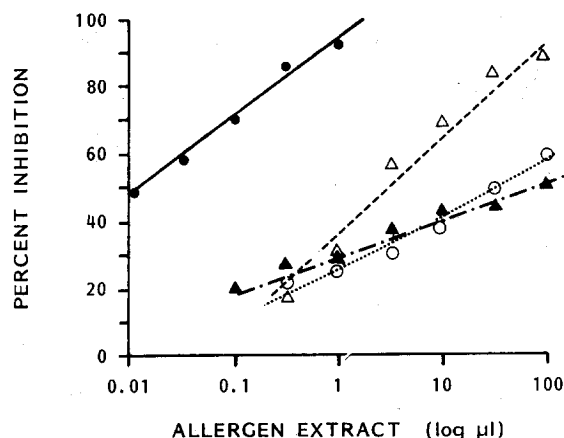


FIG. 2. RAST inhibition studies. ●—●, sunflower pollen inhibitor; △---△, honey containing 30% sunflower pollen inhibitor; ○---○, short ragweed pollen inhibitor; ▲---▲, dandelion pollen inhibitor.

sala, Sweden). The capacity of each test extract to inhibit binding of specific IgE to the sunflower disc was compared to that of a sunflower allergen extract solution.

Bronchial provocation tests. These tests were performed when the patient's clinical status was normal and his baseline pulmonary function test results were within 10% of predicted values. Nonspecific bronchial reactivity was assessed by metacholine⁶ and histamine⁷ challenges. Bronchial challenges to sunflower seeds and pollens were performed with the specially prepared extracts. For these challenges, the delivery system was identical, and pulmonary function was measured by a total body plethysmograph.

Metacholine, histamine, and allergen extracts were delivered with Piglowski's apparatus.⁸ Aerosols were delivered by a modified Tiffeneau nebulizer, which delivers a reproducible output of 0.22 L per minute with a particle size of 2 to 4 μ m (aerodynamic mass median diameter). Reproducibility of aerosol generation is controlled by a constant aerosolization pressure controlled at $\pm 0.5\%$. The nose was clipped, and aerosols were inhaled by tidal breathing through a mouthpiece. Piglowski's apparatus records the amount of air inhaled by the subject. From the concentration of the aerosol, it is easy to calculate the amount of extract inhaled. Pulmonary function was recorded with a total body plethysmograph,⁹ and FEV₁, MMEFR, $\dot{V}_{max50}$ and SG_{aw} were analyzed.

Inhalation tests were started 4 mo after the last natural pollen exposure and were performed as follows: Before any provocative testing, pulmonary function of the patient was assessed. In all investigations, the patient had an FVC and an FEV₁ value within 10% of predicted. Nonallergic bronchial responsiveness to inhaled histamine was determined. An aerosol of saline solution followed by doubling concentrations of histamine acid phosphate (0.03 to 16 mg/ml) were inhaled for 2 min at 5-minute intervals. The PD₂₀FEV₁ and PD₃₅SG_{aw} were recorded before and 30 and 90 sec after each inhalation. Inhalations were continued until there was

a 20% decrease in FEV₁, a 35% decrease in SG_{aw}, or until the maximal concentration had been administered. A similar procedure was used for metacholine inhalations, in which doses ranged from 50 to 3200 μ g. The second investigation occurred 2 wk after the first.

Control and allergen inhalation tests were performed the fourth week. The diluent for the allergen solutions (phosphate-buffered saline solution) was inhaled for 3 min. SG_{aw} was initially measured, since forced expiratory maneuvers may elicit a bronchoconstrictive response, and then FEV₁, MMEFR, and $\dot{V}_{max50}$ were measured. Each of these parameters was assessed before and 10, 15, 20, 30, and 45 min after the inhalation. After the control solutions, allergen solutions were administered by doubling concentrations from 50 to 1600 μ g after the protocol described for control solutions. Inhalations were continued until PD₂₀FEV₁, PD₂₅ MMEF, PD₂₅ $\dot{V}_{max50}$, or a PD₃₅SG_{aw} were reached or until the maximal concentration had been administered. The late-phase reaction induced by allergen was also sought, and after a positive early phase response, the patient was monitored every hour for the following 8 hr.

RESULTS

Skin tests. Skin prick test results were positive only with the sunflower pollen extract (1/20, w/v), and were totally negative with other pollen, mite, dander, or mold extracts. Skin test results with sunflower seed were negative. Honey was not tested by means of skin tests.

IgE measurements. Total serum IgE levels (titrated on two occasions) were within the normal range. After a second symptomatic pollen season, the level was 96 U/ml, and in October of the same year there was little change (83 U/ml). Specific serum IgE levels were determined by a RAST battery screen and with the particular extracts made for this study. RAST was negative for grass, olive, ragweed, mugwort, goldenrod, dandelion, and oxeye daisy pollen extracts. Percentage RAST binding to sunflower pollen ranged from 17.5% to 22.3% on five occasions. In contrast, percentage RAST binding to sunflower pollen in five control subjects was always under 2%. No specific IgE could be detected with sunflower seed extract. Percentage RAST binding to honey containing 30% sunflower pollen was 5.2%. A negative control made with honey that did not contain any sunflower or Compositae pollens had RAST binding under 2%.

RAST-inhibition experiments (Fig. 2). Although the IgE titer of Compositae pollens was not significant and results of skin tests performed with these pollens were negative, their capacity in RAST inhibition was sought in the event that limited cross-reactivity might be found. It is clear from Table I that ragweed, mugwort, and dandelion pollens did not induce significant inhibition in the sunflower pollen

TABLE I. Relative inhibitory capacities of several Compositae pollens and honey containing 30% sunflower pollen

Extract	Correlation coefficient (r)	Parallelism	Ratio of activity
Sunflower pollen	0.97	—	Reference
Short ragweed pollen	0.90	No	5×10^{-4}
Dandelion pollen	0.97	No	1.9×10^{-4}
Mugwort pollen		No inhibitory activity	
Honey	0.96	Yes	7.8×10^{-3}

solid-phase system, and the inhibition curves did not parallel those obtained with sunflower pollen extracts.

By contrast, the honey solution containing some detectable sunflower pollen induced significant inhibition of the sunflower system (Table I) and a parallel response. Specificity of the inhibition with the honey was further tested by investigation of the capacity of the honey solution to inhibit binding of specific IgE to a completely unrelated allergen, *Dermatophagoides pteronyssinus*. The honey solution produced no inhibition in this system, demonstrating that inhibition noted against the sunflower system was not a nonspecific phenomenon related to the viscosity of the honey.

Bronchial challenge testing. Challenges with either metacholine or histamine were negative. Spirometric values did not change after aerosol challenge with doses of metacholine at a concentration of 3200 μ g and histamine at a concentration of 16 mg/ml. In contrast the bronchial challenge performed with sunflower pollen extracts was highly positive at a strength of 200 μ g. The FEV₁ fell 25%, and other parameters were decreased (Table II). The decrease in spirometric values was observed within 15 min and returned to baseline values within 2 hr. No bronchodilator medication was required to reverse bronchoconstriction. No late-phase reaction was observed. Bronchial challenge with sunflower seed extract was entirely negative.

DISCUSSION

Occupational allergens are reported with increasing frequency. Pollens can be responsible for occupational asthma and/or rhinitis, and it has been observed that sugar beet and chrysanthemum pollens¹⁰⁻¹² were involved. Sunflower pollen was found to induce occupational dermatitis,³ but the mechanisms involved were not clear. The case presented herein is consistent with inhalant and food allergic reactions to sunflower pollen mediated by an IgE mechanism. On the other hand, and in contrast to the findings of Moussito and

TABLE II. Sequential changes of spirometric values after bronchial challenge with sunflower pollen extract

	Extract concentration		
	50 μ g	100 μ g	200 μ g
FEV ₁	0%	14%	25%
$\dot{V}_{max50}$	0%	10%	27%
MMEF	2%	20%	35%
SG _{aw}	10%	15%	42%

Cordero,³ there was no association between pollen and seed allergens.

The patient first developed rhinitis, with asthma appearing after a longer exposure to the allergens. Asthma was confirmed by a positive bronchial challenge with allergen, whereas nonspecific bronchial hyperreactivity to metacholine and histamine could not be demonstrated. Although in most instances asthma is associated with airway hyperreactivity, this is not always the case,¹³ and it has been found in occupational asthma to western red cedar that the level of nonspecific hyperreactivity could decrease when the exposed worker is removed from the risk.¹⁴ Present findings confirm that the bronchial hyperreactivity of subjects with asthma may be delayed or may not be present in some patients or under particular circumstances. The patient described herein may not be atopic since he did not develop any other allergic sensitization. This may be a case of the absence of hyperreactivity, but the possibility that hyperreactivity is acquired is now questioned.¹⁵

A food allergy was noted, and it is not rare that an inhalant allergen may cause untoward reactions when it is ingested.¹⁶⁻¹⁹ However, the fact that honey can induce food allergy must be made known to patients allergic to pollens, since some entomophilous pollens may be involved in allergic sensitivities.^{16, 20} A patient who is allergic to Compositae pollens should

take care not to eat honey produced with Compositae pollens.

A search for cross-reactive allergens was made between pollens emitted by plants of the Compositae family. Species from several tribes of this family have been studied, but results of RAST inhibition did not disclose significant cross-reactivity between these pollens. The *Helianthus* tribe appears to be the most primitive, and pollens emitted from its members are allergenically very different from those of other tribes.

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