

J. F. Florido-Lopez^a
P. Gonzalez-Delgado^a
B. Saenz de San Pedro^a
C. Perez-Miranda^b
J. M. Arias de Saavedra^b
J. F. Marin-Pozo^b

^a Allergy Unit,

^b Laboratory Service, Hospital General
de Especialidades 'Ciudad de Jaen',
Jaen, Spain

Allergy to Natural Honeys and Camomile Tea

Abstract

Precipitation of food allergy reactions is well known in some patients with pollinosis when they consume natural food, such as honey or camomile tea. We present 9 patients with hay fever, with or without asthma, who experienced systemic allergic reactions after ingestion of natural honeys from two local areas (Andujar and Granada) and/or camomile tea. Pollen analysis showed a high level in sunflower pollen (23.6% of pollen grains) in the honey from Andujar but not in that from Granada. The diagnosis of food and respiratory allergy was based on history, skin prick tests and specific IgE activity against pollen from *Compositae*. Conjunctival challenge with camomile extract also gave positive results. The above allergological tests and the inhibition studies carried out, suggest that pollen of *Compositae* may be responsible for allergic reactions to certain natural foods and that the reactions are mediated by an IgE-related mechanism.

Key Words

Food allergy
Honey allergy
Camomile allergy
Compositae pollen
Pollinosis

Introduction

The *Compositae* family consists of more than 30,000 plant species. Thus, it is one of the largest families known, being larger even than the grasses [1].

Because of their wide distribution, members of *Compositae* are a frequent cause of hay fever and asthma in many parts of the world [2]. Pollen from these plants also contaminates some natural foods such as honey, bee pollen, etc. Infusions from some of these plants are also used as drinks (e.g. camomile tea), which can elicit allergic reactions in sensitive patients [3–5] or even allergic conjunctivitis when are used as an antiinflammatory ocular lavage [6]. In our region, some of these foods and drinks (honeys and camomile tea) are habitually consumed and may be acquired directly from bee keepers or from small local shops.

This report described a group of patients with sensitivity to pollen of *Compositae* who presented rhinoconjunctivitis

and/or bronchial asthma and who had experienced allergic reactions after ingesting camomile tea and/or honey from our region.

Materials and Methods

Nine patients (7 females and 2 males) with mean age of 36 (range 19–72) years were included in this study. These patients all had a history of systemic allergic reactions subsequent to ingestion of honey from two local areas (Andujar and Granada) and/or after drinking camomile tea.

As shown in table I, all patients had a history of pollinosis during the spring and summer months, and none of them had received immunotherapy with *Compositae* pollen extracts.

Allergens

Honeys and Camomile. Qualitative and quantitative analysis of honeys from Granada and Andujar was performed by the Botanic Unit of the Autonomous University of Barcelona [7] (table 2).

Received:
May 11, 1994
Accepted after revision:
April 4, 1995

Correspondence to: Dr. J. Fernando Florido Lopez
Unidad de Alergia
Hospital General de Especialidades 'Ciudad de Jaen'
Av. Ejercito Español 10
E-23007 Jaen (Spain)

© 1995 S. Karger AG, Basel
1018–2438/95/1082–0170
\$8.00/0

Extracts of honeys from Granada and Andujar were prepared by weighing 2 g of each honey and dissolving it in 20 ml of a buffered of sodium phosphate solution (PBS) (pH 7.2; 0.01 M). After stirring for 1 h at room temperature, the sample was centrifuged for 10 min at 5,000 g. The supernatant was recovered and sterilized by passage through a filter with pores of 0.22 µm in diameter (Milipore, Bedford, Mass., USA).

Two grams of dried *Matricaria chamomilla* were defatted with acetone and macerated in 20 ml of PBS. The mixture was stirred for 24 h at 4°C, centrifuged (5,000 g, 20 min) and filtered as described above.

The final concentration of these three extracts was 1/10 w/v and their protein concentrations were determined by the method of Lowry et al. [8].

Other Pollen Extracts

The three above-mentioned extracts were used in conjunction with commercial pollen extracts of mugwort (*Artemisia vulgaris*), dandelion (*Taraxacum vulgare*) and olive tree (*Olea europaea*) (Alergia e Inmunología Abelló S.A., Madrid, Spain), and of marguerite (*Chrysanthemum leucanthemum*) and sunflower (*Helianthus annuus*) (Leti CBF, Madrid, Spain).

Skin Tests

Skin tests were performed by the prick-test method on the volar surface of forearm of each patient. We used extracts of the above-listed pollen, Granada honey (2.9 mg/ml), Andujar honey (3.02 mg/ml) and camomile (3.5 mg/ml). Histamine dihydrochloride (10 mg/ml) and PBS glycerol were used as positive and negative controls, respectively. All skin test sites were examined after 15 min and a wheal that had a diameter more than 3 mm greater than that of the negative control was considered positive.

Conjunctival Challenge

Since camomile tea is commonly applied to the conjunctival mucosa as an antiinflammatory agent, we included this test in our protocol using camomile extract.

One drop of PBS was deposited in the conjunctival sac as a negative control; if no reaction was observed, one drop of a serie of 10-fold dilution of the allergen in PBS was placed in the sac every 15 min. A dilution of 1:10⁵ (w/v) was used for the initial application and the test was considered positive if congestion of the conjunctival mucosa and itching of the eye were evident.

Twenty control subjects (10 atopic and 10 nonatopic) were prick-tested with honey extracts and subjected to prick tests and conjunctival challenge with the camomile extract.

Titration of Total and Specific IgE

Total Serum IgE. IgE levels were measured with the fluoroimmunoassay Pharmacia CAP System (Kabi Pharmacia Diagnostics, Uppsala, Sweden). Measurements were made in accordance with the manufacturer's instructions.

Specific IgE. The specific IgE was also determined by the Pharmacia CAP System or alternatively by the Phadezym Rast method. The CAP System is a system for titration of total and specific IgE. The assay can be performed as a radioimmunoassay or as a fluorometric assay. In this study, the fluorometric method was used. The solid phase consists of a flexible hydrophilic carrier polymer encased in a capsule (ImmunoCAP). The carrier consists of a CNBr-activated derivative of cellulose.

Table 1. Patient data

Patient No.	Age years	Sex	Clinical symptoms		Food implicated
			respi-ratory	food induced	
1	24	F	RC	rhinitis, vomiting	C
2	37	F	RC	pruritus, vomiting angioedema	C, H
3	48	M	RC, A	rhinitis, pruritus angioedema	C, H
4	35	F	RC	pruritus, cough	C, H
5	20	F	RC	urticaria, asthma angioedema	C, H
6	52	F	RC	abdominal pain urticaria, rhinitis	C, H
7	19	M	RC, A	urticaria, angioedema	H
8	72	F	RC	anaphylaxis, urticaria	C
9	19	F	RC	pruritus, rhinitis	H

RC = Rhinoconjunctivitis; RC, A = rhinoconjunctivitis, asthma; H = honey; C = camomile tea.

Table 2. Pollens in local honeys

Honey	Pollen types	%
Andujar	Echium	32.1
	Helianthus annuus	23.6
	Eucalyptus	22.4
	Lythrum	4.7
	Cistaceae	2.0
	Rosaceae	1.8
	Olea	1.8
	Plantago	1.4
	Hypocoum	1.2
	Cyperaceae	1.2
Granada	Liliaceae	1.0
	Xanthium type	1.0
	Citrus	21.5
	Eucalyptus	20.5
	Olea	15.2
	Salix	11
	Fabacea	9.9
	Rosmarinus	6.7

The system included two sets of calibrators: 2–2,000 ku/l for total IgE and 0.35 to 100 ku/l for specific IgE.

ImmunoCAP capsules with extracts of honey from Granada and Andujar were specially prepared for this study by Kabi Pharmacia Diagnostics. We provided 100 g of each primary material and serum from one of our patients for quality control.

The Phadezym IgE RAST method was used to measure IgE specific for sunflower pollen and camomile and the results are expressed in Phadebas RAST units/ml (PRU/ml).

Table 3. Results of prick* and conjunctival tests

Patient No.	Andujar honey	Granada honey	Camomile	Mug-weed	Sun-flower	Mar-guerite	Dandelium	Olive	Histamine	CPT ¹
1	12	0	12	14	9	8	10	5	8	1/100
2	6	0	7	14	8	6	11	4	6	1/10
3	15	6	11	18	23	20	18	7	8	1/10
4	6	12	7	10	11	6	12	5	7	1/10
5	10	3	5	25	10	10	5	4	7	1/10
6	9	0	9	20	8	6	8	0	6	1/100
7	14	6	10	18	11	10	16	13	7	1/10
8	10	0	7	14	10	12	23	4	8	1/10
9	16	0	9	n.d.	n.d.	n.d.	n.d.	2	7	1/100

* Diameter of largest wheal on skin-prick test (mm).

¹ Positive camomile conjunctival provocation test (w/v).

Inhibition Assay

We attempted to inhibit binding of specific IgE (CAP inhibition) to Andujar honey by incubating pooled serum, which had been obtained by mixing equal parts of serum from 5 of our patients (numbers 4, 5, 6, 7 and 8) with soluble extracts of camomile and mugwort pollen (358 and 280 µg protein/ml, respectively). The protein concentration of these extracts was also determined by Lowry's method. In brief, duplicate 100-µl aliquots of serial twofold dilutions in PBS of each of the competing fluid-phase antigens, were incubated for 2 h at room temperature with an equal volume of serum from the serum pool. Positive control (serum pool unabsorbed) as well as negative control (serum of a subject who was insensitive to pollens of *Compositae* and who had a low level of IgE) were included by mixing 100 µl of each control sample with 100 µl of PBS. After incubation, fluorometric assay was performed in accordance with the manufacturer's instructions.

Percent inhibition for each dilution was calculated as follows:

$$\text{Inhibition} = \frac{[(\text{ku/l of + control}) - (\text{ku/l of sample})]}{[(\text{ku/l of + control}) - (\text{ku/l of - control})]} \times 100.$$

Percent inhibition was plotted against the logarithm of the protein concentration and the best-fit lines were determined from the linear regression. The concentration for 50% inhibition (C_{50}) and Pearson's correlation coefficient were also determined [9].

Results

Skin Tests and Conjunctival Challenge

Results of skin tests and conjunctival challenges are shown in table 3. Skin tests and conjunctival challenges were negative in all 20 control subjects. All patients showed a clear positive reaction when extracts of *Compositae* pollens, camomile and Andujar honey were tested. However, only 4 of them reacted positively with Granada honey.

The conjunctival challenge with camomile extract was also positive in all patients at low-level dilutions (1/10 or 1/100).

Titration of Total and Specific IgE

As seen in table 4, the majority of patients had low levels of total IgE, below 100 kU/l, which is considered in our laboratory as the value that allows us to distinguish between atopic and nonatopic populations.

Determinations of specific IgE for *Compositae* pollens (table 4) gave clearly positive results, as did the seven determinations for specific IgE to camomile extract.

The two honeys, however, gave different results. All patients had high levels of specific IgE for Andujar honey but only one had a high level of specific IgE for Granada honey (No. 7). The other patients had either very low or undetectable levels of the latter IgE.

Inhibition Assay

Results of CAP inhibition assays with Andujar honey as the solid phase are shown in figure 1. Maximum inhibition achieved with mugwort pollen and camomile extracts was 89 and 76%, respectively. The inhibitory capacity of mugwort extract ($C_{50} = 1.96$ µg/ml) was about 20 times greater than that of camomile ($C_{50} = 45.72$ µg/ml).

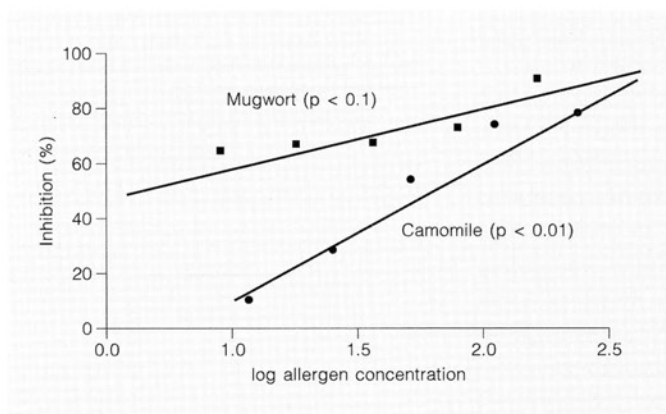
Discussion

The cases presented herein are consistent with the association of inhalant and food allergy reactions to *Compositae* pollen, with such reactions being mediated by an IgE-related mechanism.

In our group of patients, as in groups in other studies, food allergies appeared to be caused by pollen that contaminated both the honeys and camomile tea [10–12]. However, if certain bee components are included in honey, such as bee

Table 4. Specific and total IgE

Patient No.	CAP, kU/l						RAST, PRU/ml		Total IgE
	Andujar honey	Granada honey	mug-weed	dandelion	mar-guerite	olive	camomile	sunflower	
1	1.85	0.6	11.3	8.2	13.5	<0.35	8.5	8.3	180
2	1.48	<0.35	11	7.13	7.36	<0.35	5.18	5	40.6
3	3.51	<0.35	7.54	8.43	7.99	2.74	n.d.	3.74	71.8
4	9.54	0.70	17.7	17.2	21.4	6.11	10.48	19.7	82
5	11.9	<0.35	38.1	92.9	27.7	<0.35	52.4	35	71
6	2.16	<0.35	42.8	11.2	46	<0.35	n.d.	21.30	70.9
7	11.9	6.54	27.5	20.7	28.1	55.6	24.30	20.50	746
8	8.33	0.83	37.8	16.8	35.9	<0.35	16.20	18.20	291
9	12.7	<0.35	39.3	37.5	34.6	<0.35	14.34	16.35	59.8

**Fig. 1.** Inhibition assay (CAP); the solid phase was Andujar honey that contained 23.6% sunflower pollen.

venom enzymes or secretions from pharyngeal glands, they could also be responsible for this sensitization [13].

Skin tests and measurements IgE specific for Granada honey were negative in the majority of our patients, and this class of honey lacked pollen of *Compositae*.

The majority of species of *Compositae* are entomophilous, but in some of them, such as mugwort and short ragweed, the pollination is anemophilous [14]. For this reason, it is difficult to ascertain whether the route of sensitization was via digestive tract or inhalation in our group of patients. The subject of possible absorption of pollen via the digestion is very controversial but, in the absence of high levels of *Compositae* pollens in the air, as in the present cases, and in view of the mildness of respiratory symptoms (only 2 of the 9 patients suffered from asthma), the possibility of digestive sensitization cannot be ignored.

The inhibition achieved with mugwort extract and, to a lesser extent, with camomile extract, in the binding of spe-

cific IgE to Andujar honey (rich in sunflower pollen) suggests that the symptoms presented by our patients upon ingesting the honey were the result of the contaminating pollen rather than of other possible food components. Nevertheless, a complex extract was used, and the dilutions included a range that was too small to allow one to draw any conclusions about cross-reactivity between these pollens, which has already been demonstrated by other authors [15, 16].

Olive tree pollen is the most frequent cause of pollinosis in our area [17]. For this reason and also because of its presence in Granada honey, we included it in our study protocol.

The fact that our patients also exhibited symptoms in response to Granada honey has two possible explanations: first, some of them were sensitized to pollen of *Olea europaea*, and, second, and more probably, differences might exist both in the composition of identically labelled products and in their marketing due to the absence of rigorous sanitary controls.

In conclusion, we believe that the old recommendation that patients allergic to *Compositae* pollen avoid consuming natural products such as honeys and camomile tea should be followed. This recommendation should, moreover, be emphasized since the consumption of these products has become popular as a consequence of the curative and energizing properties that have been attributed to them. Only preparations with clear identification of contaminating pollens that do not contain *Compositae* pollens are suitable for consumption by such patients.

References

- 1 Yman L: Allergenic plants; in: Botanical Relations and Immunological Cross-reactions in Pollen Allergy. Uppsala, Pharmacia Diagnostics, 1982.
- 2 Wodehose RP: Hay Fever Plants: Their Appearance, Distribution, Time of Flowering and Their Role in Hay Fever. New York, Hafner, 1971.
- 3 Lin FL, Vaughan TR, Vandewalker ML, Weber RW: Hypereosinophilia, neurologic and gastrointestinal symptoms after bee-pollen ingestion. *J Allergy Clin Immunol* 1989;83:793-796.
- 4 Pauli G, Hutt N, De Blay F, Hoyet C, Bessot J, Leuschner R: Food allergy following the ingestion of pollen sacs. *Allergologie* 1989;12:87-88.
- 5 Bousquet J, Campos J, Michel FB: Food intolerance to honey. *Allergy* 1984;39:73-75.
- 6 Subiza J, Subiza JL, Alonso M, Hinojosa M, Garcia R, Jerez M, Subiza E: Allergic conjunctivitis to chamomile tea. *Ann Allergy* 1990;65:127-132.
- 7 Torreguitart A, Perez-Obiol R: Caracterización polínica de mieles de frutal; in Civis J, Valle F (eds): *Actas de Palinología*. Salamanca, Universidad de Salamanca, 1988.
- 8 Lowry OH, Rosebrough NJ, Farr AL, Randall RJ: Protein measurement with the Folin phenol reagent. *J Biol Chem* 1951;193:265-275.
- 9 Perrick D, Stafford CT, Armstrong E, DuRant RH: Modification of the fluorescent allergosorbent test as an inhibition assay for determination of cross-reactivity among aeroallergens. *J Allergy Clin Immunol* 1991;87:98-103.
- 10 Fernandez Rodriguez C, Pascual Marcos C, Martin Esteban M, Lopez Squillary C, Ojeda Casas JA: La miel como alergeno. *Rev Esp Alergol Immunol Clin* 1987;2:189.
- 11 Bousquet J, Dhivert H, Clauzel A, Hewitt B, Michel F: Occupational allergy to sunflower pollen. *J Allergy Clin Immunol* 1985;75:70-74.
- 12 Subiza J, Subiza JL, Hinojosa M, Garcia R, Jerez M, Subiza E: Anaphylactic reaction after the ingestion of chamomile tea: A study of cross-reactivity with other composite pollens. *J Allergy Clin Immunol* 1989;84:353-358.
- 13 Helbling A, Peter Ch, Berchtold E, Bogdanov S, Müller U: Allergy to honey: Relation to pollen and honey bee allergy. *Allergy* 1992;47:41-49.
- 14 Lewis W, Vinay P: North American pollinosis due to insect-pollinated plants. *Ann Allergy* 1979;42:309-318.
- 15 Fernandez C, Martin-Esteban M, Fiandor A, Pascual C, Lopez Serrano C, Martinez Alzamora F, Diaz Pena Jose M, Ojeda Casas JA: Analysis of cross-reactivity between sunflower pollen and other pollens of the *Compositae* family. *J Allergy Clin Immunol* 1993;92:660-667.
- 16 Alonso M, Subiza JL, Garcia R, Jerez M, Hinojosa M, Subiza E: Reactividad cruzada entre *Centaurea*, *Helianthus* y *Artemisia* (Abstract). *Rev Esp Alergol Immunol Clin* 1988;3:37.
- 17 Subiza E, Subiza JL, Jerez M: Arboles, Hierbas y plantas de interes Alergológico en España. *Sociedad Española de Alergia e Inmunología Clínica, Tratado de Alergología de Inmunología Clínica IV*, Madrid, 1986;pp 257-366.