

Allergy to pine nut

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SUMMARY

Background: food allergy is highly prevalent in our environment, especially among atopic patients. *Pinus pinea* is common in our region and its fruit, the pine nut, is allergologically important. Several cases have been reported in the literature that demonstrate the existence of common antigenic bands between pine nut and almond. In this study we try to assess this finding and the possible existence of common allergens by *in vitro* techniques.

Methods and results: we present a 10-year-old boy, previously diagnosed of seasonal rhinoconjunctivitis with sensitisation to grass and olive pollen, who had an anaphylactic reaction after eating pine nut. We performed *in vivo* (prick test, prick-by-prick) and *in vitro* tests (total and specific IgE determinations [CAP-FEIA]), histamine release test, and immunoblotting (SDS-PAGE). We also reviewed the literature through the MEDLINE database in PubMed.

Conclusions: because pine nut is commonly consumed in our environment, the prevalence of allergic reactions is probably considerable and these reactions take place at an early age. We demonstrate the existence of common antigenic proteins between pine nut and peanuts.

Key words: Anaphylaxis. Pine nut. Immunoblotting. Cross reactivity to nuts. Food allergy.

Allergol et Immunopathol 2002;30(2):104-108.

INTRODUCTION

It is estimated that the prevalence of food allergy ranges from 1.4 % (1) to 2.4 % (2) among adults, and

from 0.3 % to 7.5 % among children, and it is around 10 % among atopic individuals (3).

About 3.6 % of the patients who came to an Allergology service have food sensitisation.

Pine nut is the fruit of the *Pinus pinea* L. belonging to the *Pinaceae* family, of the gymnosperm class. It is also called *Pinus sativa* Quer. and *Pinus domestica* Mathiol. It blooms from March to May, and in Europe it can be found along the Mediterranean basin where it was spread by the Romans, occupying nowadays 2.84 % of the woodland. In Spain the areas of greatest incidence are Huelva, Sevilla and Cadiz, followed by the Northern plateau, appearing in Valladolid (where it is exported as fruit tree), in Zamora, Ávila and Segovia, apart from other areas in Spain (4).

Because of the case report we review the literature and assess the possible existence of common allergens by immunoblotting.

CASE REPORT

Ten year old boy diagnosed previously of seasonal rhinoconjunctivitis for grass and olive pollen sensitisation.

The patient is under treatment with sublingual immunotherapy to grass pollen since September 2000, presenting a good evolution and staying with no nasal or conjunctival symptoms.

The patient comes to our clinic presenting abdominal pain; pharyngeal, retroauricular, axillar and toe pruritus; and labial and palpebral angioedema, after eating caramel pine nut with good response to the parenteral treatment with methylprednisolone and dexchlorpheniramine.

RESULTS

Blood tests and erythrocyte sedimentation were within normality. Skin prick tests were positive to al-

mond (5 mm) and peanut (5 mm), and negative to hazelnut, sunflower seed, walnut, pistachio and pine (*pinus* spp. Prick by prick test was positive to pine nut (12 mm) (fig. 1). Specific IgE by CAP FEIA (Pharmacia, Uppsala, Sweden) to *pinus edules*: 8.32 kU/l, *Pinus strobus*: negative, lolium: 21.6 kU/l and olive: 5.71 kU/l. Total IgE value was 64.5 kU/l. Basophil activation test was positive to pine nut (80.7 %) and peanut (87 %), and negative to pine and almond. Histamine release to pine nut was 38.3 %, to peanut 40.43 %, to almond 2.13 % and to *pinus* sp. 0 %, basal histamine 2.88 ng/ml, total histamine 78.21 ng/ml; anti IgE 38.38 %.

Immunoblotting by SDS-PAGE was performed using pine nut, almond and peanut extract in polyacrylamide gel (fig. 2), the gel revealed about 12 proteic bands with molecular weights from 20 to 140 kDa (fig. 3). In the nitrocellulose membrane, we observed bands that react with the patient's specific IgE, especially the 30 and 44 kDa ones. Subsequently, we performed immunoblotting inhibition, by incubating the patient serum with peanut extract for 24 hours. Afterwards another immunoblotting was performed using pine nut, almond and lolium extracts. The result was that most bands disappeared especially the 30 and 44 kDa (fig. 4), remaining for pine nut a 50 kDa band, for almond a 25 kDa band and for Lolium a 35 kDa band.

DISCUSSION

We find in the literature several publications on food allergy to this fruit, and a total of 21 case reports (table I).

Koepke JW et al (5) in 1990 proved by electrophoresis 30 proteic bands, three of which were in the range of 66 to 68 kDa. In 1996, the existence of cross reactivity between *pinus pinea* and *pinus cembra* was described for the first time in a patient who presented allergic reaction to pine nuts after eating parrot's food (6). Roux et al (7) in 1998 described the case of a patient with systemic reaction to pine nut who previously suffered from sensitisation to almond.

De las Marinas et al (8) presented in 1998 one patient with previous demonstrated sensitisation to almond, who had suffered severe anaphylaxis after eating pine nuts. They proved the existence of 12 antigenic bands of molecular weights between 17 and 133 kDa and finding a common 50 kDa band between almond and pine nut with inhibition blotting and confirmed by inhibition CAP. Nubira et al (9) in 1998 described four cases of allergy to pine nuts among children aged 12 months to 6 years, with the particularity that none of them was aware of having eaten pine nuts be-



Figure 1.—Prick-Prick to pine nut.

fore. They concluded that due to the great disguised consumption of this nut in our country, the prevalence of allergic reactions was probably greater here and happened at an earlier age. Moneret-Vautrin et al (10) in 1998 published a case of allergy to walnut and pine nut stimulated by the increase of vascular permeability and the vasodilatation produced by an angiotensin converting enzyme inhibitor. In 2000 anaphylaxis by pine nut and cross reactivity to pine pollen proteins is proven

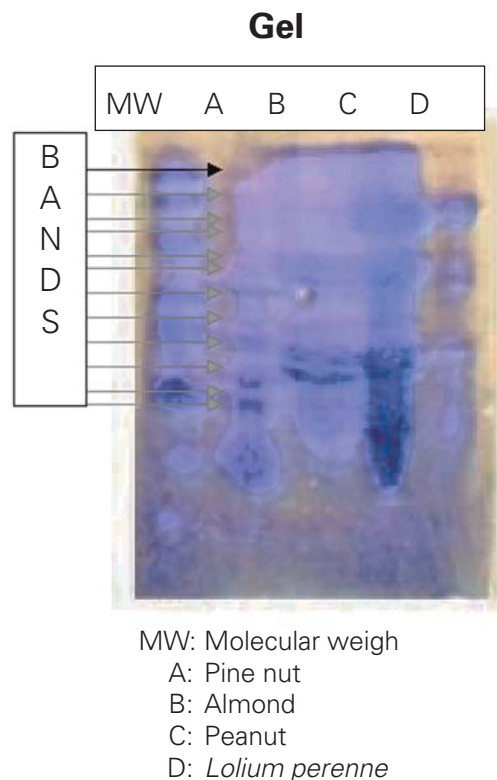


Figure 2.—Immunoblotting gel to pine nut, almond and peanut.

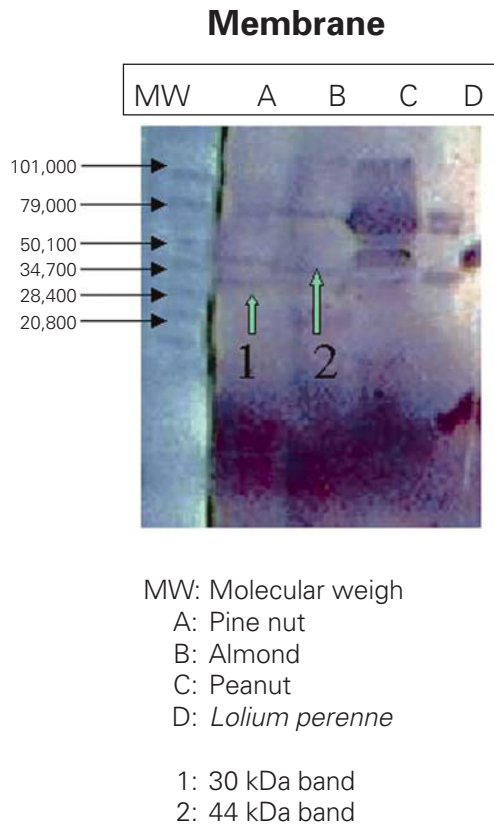


Figure 3.—Immunoblotting membrane to pine nut, almond and peanut.

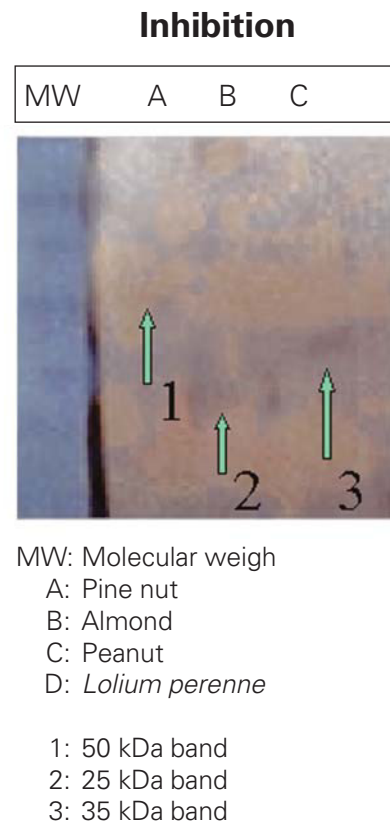


Figure 4.—Inhibition immunoblotting to pine nut, almond and peanut.

(11). In an extract of pine pollen the existence of several 8 to 90 kDa bands is demonstrated, as well as an 8 to 11 kDa band. Subsequently, García-Menaya et al. (12) detected through SDS-PAGE a new 17 kDa monoclonal band not described previously.

Out of the 22 cases of allergy to pine nuts found in the literature, eleven were atopic. Regarding the symptoms, only one patient had anaphylactic shock, 12 had anaphylaxis, including the present case, and the other ten had urticaria with or without angioedema. In most cases the nuts were disguised with other foods, or the patient ignored having eaten it before. Only in four cases the reaction is due to a direct ingestion, like in our case.

Regarding the diagnostic tests, prick test was positive in all the patients, seven of them underwent also prick by prick tests, and two rub test. We can therefore state that skin test is reliable in this pathology. Specific IgE levels went from class 2 to class 6. Oral provocation test had to be done in four patients, two of them children, and the result was positive in all of them. In our case, the evident symptoms clearly advised against it.

Up to now, we have found no cross reactivities described between peanut and pine nut. In our re-

sults, the fact that the 30 and 44 kDa bands disappear in the inhibition immunoblotting demonstrates the existence of these common antigenic proteins between pine nut and peanut.

The finding described makes it necessary to avoid both foods in these patients' diet, even though the symptoms appear only with one of them.

It is interesting to highlight the apparent lack of cross reactivity with almond unlike the case described in 1998 (8). Nevertheless our patient was advised to avoid it as well as peanuts.

Cross reactivity with *Lolium* could not be demonstrated in this patient, since in the inhibition immunoblotting the 30-44 kDa bands remained.

RESUMEN

Introducción: la prevalencia de alergia alimentaria es elevada en nuestra población y mayor todavía cuando nos referimos a pacientes atópicos. El pino piñonero está ampliamente distribuido por nuestra geografía por lo que su fruto, el piñón, ha adquirido bastante importancia alérgica.

Table I
PUBLISHED CASES OF PINE NUT ALLERGY

Case	Age	Sex	Atopy	Symptoms	Ingestion	Skin Test (mm)	Total IgE KU/L	Specif. IgE KU/L	Oral Provoc	Place and year	First Author (et al.)
1	32	F	Yes	Anaphylaxis	Unknown	Rub test	ND	ND	NP	USA 1958	M. Santos
2	13	F	No	Anaphylaxis	Unknown	Prick 10 15	ND	ND	NP	USA 1967	A.J. Fine
3	21	M	No	Urticaria	Unknown	Prick 4 4	ND	ND	NP	USA 1987	A.J. Fine
4	43	F	No	Anaphylaxis	Unknown	Prick 18 24	ND	ND	NP	USA 1969	C.J. Falliers
5	28	F	Yes	Urticaria	Unknown	Rub test	ND	ND	NP	USA 1989	C.J. Falliers
6	17	M	Yes	Digestive	Unknown	Prick 10 12	ND	ND	NP	USA 1989	C.J. Falliers
7	8	F	Yes	Urticaria	Unknown	Prick 8 9	ND	ND	+	Spain 1990	A. Armentia
8	21	M	No	Anaphylaxis	Cookies with pine nuts	Positive Prick test	ND	+	NP	Denver 1990	Koepke JW
9	28	F	No	Anaphylaxis	Unknown	Prick test	ND	2.5 PRU	+	Denmark. 1990	N. H. Nielsen
10	21	M	No	Anaphylaxis	Unknown	Prick 20 20	ND	ELISA	NP	USA 1990	U. K. Koepke
11	54	F	No	Anaphylaxis	Bird food	Prick 7 Id + Pinnea and cembra	184 Pinnea 44.6	RAST +	NP	Rotterdam 1996	A. Jansen
12	28	M	Yes	Urticaria	Salad with pine nuts	Prick (-) Prick by Prick (+)	2472	ELISA 11 (C-3)	NP	Switzerland 1998	N. Roux
13	35	M	Yes	Anaphylaxis	Salad	Prick by Prick (+)	1011	ELISA 6.25 (C-3)	NP	Switzerland 1998	N. Roux
14	28	M	Yes	Anaphylaxis	Almonds and meat balls	Id: 10 12 Prick 6 6	485	CAP FEIA 5.27	NP	Spain 1997	D. de las Marinas
15	5	M	Yes	Urticaria	Walnuts and pine nuts	Prick 15 10 Prick by Prick 10 7	ND	CAP 6	Positive	Spain 1998	N. Nubira
16	7	M	Yes	Urticaria and angioedema	Cake with pine nuts	Prick 15 15 Prick by Prick 9 6	ND	CAP 44	NP	Spain 1998	N. Nubira
17	4	M	Yes	Urticaria and angioedema	Pine nuts	Prick 15 15 Prick by Prick 27 9	ND	CAP 63	NP	Spain 1998	N. Nubira
18	5	F	Yes	Angioedema	Cookies with pine nuts	Prick 10 15 Prick by Prick 10 7	ND	CAP 2	NP	Spain 1998	N. Nubira
19	53	M	Yes	Anaphylaxis	Salad with pine nuts	Prick +	ND	CAP C-4	Positive	Germany 1998	A.V. Beyer
20	87	F	No	Anaphylactic shock	Cake	Prick 10	ND	RAST-	NP	France 1998	D.A. Moneret
21	29	F	No	Anaphylaxis	Cake with pine nuts	Prick by Prick 74	ND	RAST C-2	NP	Italy 2000	G. Senna
22	22	M	No	Anaphylaxis	Pine nuts	Prick by prick 12	Alastat 698 U/ml	Alastat 0.79	NP	Spain 2000	J.M. García-Menaya

Sex: M = Male, F = Female.

ND: Not described.

NP: Not performed.

La bibliografía refleja varios casos descritos que ponen en evidencia la existencia de bandas antigénicas comunes entre piñón y almendra. En este estudio, se valorará este hallazgo así como la posible existencia de alérgenos comunes mediante técnicas *in vitro*.

Pacientes, métodos y resultados: presentamos el caso de un niño de 10 años de edad con el diagnóstico previo de rinoconjuntivitis estacional por sensibilización a pólenes de gramíneas y oleáceas, que acude por reacción anafiláctica tras ingesta de piñón. Para su estudio se realizaron test diagnósticos tanto

in vivo (prick test, prick by prick), como *in vitro* (IgE total y específica [CAP-FEIA], test de liberación de histamina, inmunoblotting [SDS-PAGE]).

Se realiza revisión de la bibliografía existente mediante la base de datos del Medline en PubMed.

Conclusiones: debido al gran consumo de piñón en nuestro medio, probablemente la prevalencia de reacciones alérgicas es mayor y tengan lugar más tempranamente.

Demostramos la existencia de proteínas antigénicas comunes entre el piñón y el cacahuete.

Los hallazgos encontrados obligan en estos pacientes a la evitación de ambos alimentos de la dieta, aunque la sintomatología se exprese únicamente con uno de ellos.

Palabras clave: Anafilaxia. Piñón. Inmunoblotting. Reactividad cruzada a frutos secos. Alergia alimentaria.

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