ORIGINAL ARTICLE

Allergy to Laxative Compound (*Plantago ovata* seed) Among Health Care Professionals

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Abstract

Background: The seeds of *Plantago ovata* (psyllium, ispaghula) used in the manufacture of bulk laxatives are known to be the cause of occupational allergy (rhinitis, asthma) in health care and pharmaceutical workers.
Objective: We studied the prevalence of *P ovata* seed allergy among health care workers in geriatric care homes and compared it with a group of health care professionals not exposed to *P ovata* seed. Cross reactivity with *Plantago lanceolata* pollen was also studied.
Methods: Two groups of health professionals were recruited: 58 health care workers from geriatric care homes who were exposed daily to laxatives containing *P ovata* and 63 nonexposed health care professionals. The prevalence of allergy and sensitization to *P ovata* seed was determined based on clinical history, skin prick test, and analysis of specific immunoglobulin (Ig) E. IgE immunoblotting was performed to calculate the molecular weights of the *P ovata* seed allergens. Cross reactivity to *P lanceolata* pollen was studied by enzyme allergosorbent test (EAST) and immunoblot inhibition techniques.
Results: The prevalence of sensitization and clinical allergy to *P ovata* seed in the exposed group was 13.8% and 8.6%, respectively. No sensitization was observed in the nonexposed group. IgE-binding proteins of 17, 20, 25, 32-34, 54, 73-77, and >97 kDa were identified. EAST inhibition and immunoblot inhibition demonstrated the existence of cross reactivity between *P ovata* seed and *P lanceolata* pollen extracts.
Conclusions: The rate of sensitization to *P ovata* seed is high among health care workers in geriatric care homes (13.8%). A mild cross reactivity between *P ovata* seed and *P lanceolata* pollen was observed.

Key words: *Plantago ovata. Plantago lanceolata*. Occupational allergy. Prevalence. Health care workers. Cross reactivity.

Resumen

Antecedentes. Las semillas de *Plantago ovata* (psyllium, ispaghula) se usan en la fabricación de laxantes de volumen y han sido descritas como causantes de alergia ocupacional (rinitis y asma) en personal sanitario y trabajadores farmacéuticos.
Objetivos: Se ha estudiado la prevalencia de alergia a semillas de *P ovata* entre sanitarios de residencias geriátricas y se ha comparado dicha prevalencia con la obtenida en un grupo de sanitarios no expuestos a semillas de *P ovata*. En una segunda parte, se ha analizado la posible reactividad cruzada con el polen de *Plantago Lanceolada*.
Métodos: Se reclutaron dos grupos de trabajadores sanitarios: uno compuesto por 58 trabajadores procedentes de residencias geriátricas donde diariamente se exponen a laxantes con *P ovata* y otro de 63 trabajadores sanitarios no expuestos de forma habitual a este tipo de laxantes. Mediante la historia clínica, las pruebas cutáneas y la determinación de inmunoglobulina (Ig) E específica se calculó la prevalencia de alergia y sensibilización a semillas de *P ovata*. Empleando la inmunodetección de proteinas (inmunobloting) se determinó el peso molecular de los alérgenos de las semillas de *P ovata*. La reactividad cruzada entre el polen de *P lanceolata* y las semillas de *P ovata* se evaluó utilizando técnicas de inhibición del blotting y EAST (enzyme allergosorbent test) inhibición.
Resultados: La prevalencia de sensibilización y alergia clínica a semillas de *P ovata* en el grupo de expuestos fue de 13,8% y 8,6% respectivamente. No se obtuvo ninguna sensibilización en grupo de no expuestos. Se encontraron proteinas fijadoras de IgE correspondientes a los siguientes pesos moleculares: 17,20,25, 32-34, 54, 73-77 y >97 kDa. Mediante las técnicas de EAST e inmunobloting inhibición se demostró la existencia de reactividad cruzada entre los extractos de semillas de *P ovata* y de polen de *P lanceolata*.
Conclusiones: La sensibilización a semillas de *P ovata* es alta entre los trabajadores de las residencias geriátricas (13,8%). Se ha observado reactividad cruzada entre las semillas de *P ovata* y el polen de *P lanceolata.*

Palabras clave: *Plantago ovata. Plantago lanceolada*. Alergia ocupacional. Prevalencia. Trabajadores sanitarios. Reactividad cruzada

Introduction

Plantago ovata is a member of the Plantaginaceae family. Its seeds, also known as psyllium and ispaghula, are widely used as bulk laxatives presented as powder or granulate formulations. Metamucil, Plantaben, Cenat are some of the laxatives commercially available in Spain. In the United States of America, Canada, and Australia, the seeds of *P ovata* are also added to breakfast cereals to increase dietary fibre and reduce serum cholesterol levels [1].

The first case of allergic reaction to *P ovata* seed was described by Ascher [2] as early as 1941. Since then, many cases of occupational allergy (rhinitis, asthma), anaphylaxis, and asymptomatic eosinophilia [3] have been reported. In general, *P ovata* seed sensitization occurs after inhalation of *P ovata* seed powder, the particles of which can be as small as 2 μm [4]. It seems that the risk of sensitization is higher with laxatives in powder form (for example Plantaben) than with granulated forms or with laxatives that produce fewer airborne particles [5]. Three risk groups have been described [6]: pharmaceutical workers who handle *P ovata* seeds in the manufacture of bulk laxatives; health care professionals who usually prepare these laxatives for their patients; and finally consumers who take this kind of laxative for themselves. Thus, except for the patients, it could be considered as an occupational disease.

Cases of occupational asthma have been described in both pharmaceutical manufacturing workers and health care professionals [6-9]. The prevalence of occupational asthma in health care professionals was described as 4% and sensitization to *P ovata* seed ranges according to the method used for diagnosis from 5% (skin prick test) to 12% (specific immunoglobulin [Ig] E levels) [10]. In pharmaceutical manufacturing workers, the prevalence of occupational asthma was 3.6%, and sensitization to *P ovata* seed was 27.9% based on prick tests and specific IgE determinations [11]. Cases of anaphylactic reaction have been reported in all risk groups after ingestion of laxative or breakfast cereals containing *P ovata* seed [1,9,12-17]; in most of those subjects, sensitization occurred by inhalation of P ovata seed dust in the workplace. Sensitization by inhalation may occur in the absence of symptoms (rhinitis, asthma) that might alert to the future possibility of anaphylaxis by ingestion [13]. Recently, a case of psyllium-associated anaphylaxis and death was reported [18].

Plantago lanceolata or English plantain pollen is a well-known aeroallergen that can cause seasonal allergy (rhinoconjunctivitis and asthma) through an IgE-mediated mechanism [19]. Because of the phylogenetic relationship between *P ovata* and *P lanceolata*, cross reactivity studies between *P ovata* seed and *P lanceolata* pollen allergens have been done, and while most of them suggest a lack of cross reactivity [20,21], at least 1 showed the existence of immunologic cross reactivity between *P ovata* seed and *P lanceolata* pollen [19].

The aim of the present study was to determine the prevalence of sensitization and clinical allergy to *P ovata* seed among health care professionals in geriatric care homes who usually manipulate laxatives with *P ovata* seed powder and compare it with a similar sample of health care professionals at a general hospital who were not exposed to those laxatives. In a second part of the study we determined the molecular mass of allergenic components from *P ovata* seed and evaluated the existence of cross reactivity between these allergens and proteins from *P ovata* pollen.

Material and Methods

A cross-sectional epidemiologic study was performed in the town of Vitoria-Gasteiz in the Basque Country, Spain. The study protocol was approved by the Institutional Review Board of Santiago Apóstol Hospital and informed consent was obtained from all subjects.

Subjects

Two random samples of health care professionals (nurses and auxiliary nurses) were included in the study. The exposed sample was selected from 269 workers in 3 geriatric care homes belonging to the Regional Social Welfare Institute (Instituto Foral de Bienestar Social). Fifty-eight subjects (7 men and 51 women; 13 nurses and 45 auxiliary nurses) with a mean age of 40 years (range, 31 – 60 years) were included. All of them were exposed to *P ovata* seed in the workplace during preparation and administration of laxatives to patients. As a control group, 63 nonexposed subjects (3 men and 60 women; 44 nurses and 19 auxiliary nurses), with a mean age of 38 years (range, 23-58 years), were randomly enrolled from among the 394 health care professionals of Santiago Apóstol Hospital. They were not usually exposed to *P ovata* seed-containing laxatives. An additional group of 5 subjects was recruited for immunoblotting and cross-reactivity studies. All of them were health personnel from the same geriatric institutions with known allergy to *P ovata* seed (demonstrated by clinical history and laboratory test) and had similar demographic characteristics to the study subjects.

Prevalence Study

Analysis of prevalence included a complete clinical history, skin prick test, and specific IgE measurements. Subjects with positive prick test or positive specific IgE to *P ovata* seed were considered as sensitized. When symptoms appeared, the patient was considered as allergic to *P ovata* seed. Finally, subjects who showed at least 1 positive immediate skin reaction to any of the allergenic sources tested other than *P ovata* seed were considered atopic.

Clinical history. In all subjects, a medical history was obtained (including allergies) and a physical examination performed. Information about years of work, period of exposure to *P ovata* seed powder, and symptoms presented when manipulating laxatives with *P ovata* seed were obtained. Occupational asthma and rhinitis were suspected when, according to the clinical history, patients reported any asthma-related symptom such as wheezing, cough, and shortness of breath or presented rhinoconjunctivitis symptoms like sneezing, itchy eyes, or blocked nose during or after handling laxatives

containing *P ovata* seed. Information about potential food allergy reactions after ingestion of products containing *P ovata* seed and any symptoms related to latex were also recorded.

Skin prick test. Skin prick tests were performed according to European Academy of Allergy and Clinical Immunology recommendations [22] on the volar side of the forearm using prick lancets (ALK-Abelló, Madrid, Spain). The tests were done with commercial laxatives (Plantaben, Metamucil, and Cenat), extract of *P ovata* seed (Bial-Aristegui, Bilbao, Spain), latex, nuts, and common inhalants (dust mites, *Phleum pratense* pollen, *P lanceolata* pollen, cat and dog furs, and *Alternaria alternata*); 1 sterile device was used for each test. Histamine dihydrochloride (10 mg/mL) and sterile 0.9% saline were used as positive and negative controls, respectively. Positive reactions were defined as a wheal of at least 3 mm as compared with the negative control, 15 minutes after puncture.

Total and specific IgE measurements. Total IgE and specific IgE to *P ovata* seed, *P lanceolata* pollen, and latex extracts were measured in all the subjects. In addition, specific IgE was also measured for those allergenic sources to which subjects appeared to be sensitized by prick test. IgE determinations were performed with a Pharmacia CAP system (Pharmacia, Uppsala, Sweden) according to the manufacturer's instructions. Any values greater than 0.35 kU/L were considered positive.

Cross-Reactivity Study

The cross reactivity study was carried out with sera from 3 patients: 2 of them were sensitized to *P ovata* seed and *P lanceolata* pollen (patients 1 and 11) and the other (patient 12) was only sensitized to *P ovata* seed.

P ovata *seed extract (Plantaben).* To obtain the *P ovata* seed extract, the commercially available Plantaben was dissolved in 50 mM phosphate-buffered saline at pH 7.5 (1.6%, weight by volume) and extracted overnight by magnetic stirring at 4ºC. After centrifugation, supernatant was dialyzed against distilled water. The dialyzed extract was filtered through a 0.22 μm pore diameter membrane and freeze dried. Protein determination was performed by the Bradford method [23].

Enzyme allergosorbent test inhibition. Enzyme allergosorbent test (EAST) inhibition was carried out according to the method described by Yman et al [24]. Patient sera were incubated with serial 10-fold dilutions (0.001, 0.01, 0.1, 1, and 10 mg/mL) of *P ovata* seed and *P lanceolata* pollen extracts at 4ºC overnight. Specific IgE measurement was then performed with the corresponding solid phase (*P ovata* seed or *P lanceolata* pollen extracts). Solid phase was obtained by coupling the extract solutions (10 mg/mL) to 6 mm-diameter cyanogen bromide-activated paper discs, as described by Ceska and Lunqvist [25].

Immunoblotting and inhibition. Sodium dodecyl sulfate (SDS) polyacrylamide gel electrophoresis was carried out according to the method of Laemmli [26]; 12.5% and 4% acrylamide was used for separating and stacking gels, respectively. Samples were dissolved in 0.125 M Tris-HCl, pH 6.8, and dissociated with 0.1% SDS and 5% ß-mercaptoethanol at 100ºC for 5 minutes. Twenty micrograms of protein (determined by Bradford assay) was applied per lane. Separated protein bands were electrophoretically transferred to polyvinylidene diflouride membranes, essentially as described by Towbin et al [27], and blocked for 1 hour at room temperature with 0.1% Tween-20 in Tris-buffered saline. Membranes were incubated overnight at 4ºC with patient sera followed by incubation with anti-human IgE-horseradish peroxidase conjugate and detection by chemoluminescence as recommended by the manufacturer (ECL-Plus; Amersham Pharmacia Biotech, Uppsala, Sweden). For immunoblot inhibition, patient sera were preincubated overnight at 4ºC with the inhibitory phase (*P ovata* seed extract or *P lanceolata* pollen extract).

Sera from the same 3 patients were used to carry out both EAST inhibition and the immunoblot inhibition.

Statistical Analysis

Sample size was calculated on the basis of a known *P ovata* sensitization prevalence of 5% to obtain a 95% confidence interval (CI) with an alpha error of 5%. Prevalence was calculated in each group as the percentage of affected patients in the group with the associated 95% CI. Qualitative variables were expressed as percentages and were compared by χ^2 test. Quantitative values were shown as means (SD) and compared by *t* test or Mann-Whitney test, when appropriate. *P* values less than .05 were considered statistically significant. All analyses were performed using SPSS version 10.0.

Results

Prevalence of Sensitization and Clinical Allergy

Sensitization to *P ovata* seed was defined as a positive prick test or positive specific IgE to *P ovata*. We found 8 out of 58 subjects to be sensitized in the exposed group, resulting in a prevalence of sensitization to *P ovata* seed of 13.8% (95% CI, 6% - 25%). However, none of the 63 nonexposed subjects was found to be sensitized.

All the sensitized subjects had a positive prick test (8/58; 13.8%) but only 4 (6.9%) of them were positive for specific IgE (> 0.35 kU/L). Five out of the 8 sensitized health care workers reported allergic symptoms on or after handling laxatives containing *P ovata* seed. All of them presented rhinoconjunctivitis and in 2 cases asthma was suspected, so the prevalence of clinical allergy to *P ovata* seed was estimated as 8.6% (95% CI, 3% - 19%). The other 3 subjects were considered as asymptomatic sensitizations. No subjects reported symptoms after ingestion of products containing *P ovata* seeds. The clinical characteristics and response to tests in sensitized individuals are summarized in the Table.

The group of 8 sensitized individuals included 7 women and 1 man (2 nurses and 6 auxiliary nurses) with a mean age of 37 years (range, 34 to 40 years). There were no significant differences in characteristics when compared with the rest

Table. Clinical Characteristics and Response to Tests in Sensitized Subjects

Patient Number[a]	Sex/Age, y	Symptoms	Prick Test, mm				IgE-*P ovata* seed, kU/L	Other Sensitizations (by prick test)
			P ovata seed	Plantaben	Metamucil	Cenat		
1	M/38	Rhinitis	–	–	–	3×3	0.57 (class 1)	Dust mites, grass pollen *P lanceolata* pollen dog fur, latex
2	F/35	Rhinitis	5×5	12×10	7×5	7×7	3.04 (class 2)	Hazelnut, latex
3	F/40	Rhinitis + Asthma	6×6	12×6	6×5	7×6	6.34 (class 3)	No
4	F/40	Rhinitis	6×5	4×4	–	7×5	<0.35 (class 0)	No
5	F/36	No	5×4	–	–	–	<0.35 (class 0)	No
6	F/40	No	–	4×3	–	–	<0.35 (class 0)	Dust mites, cat and dog furs
7	F/34	Rhinitis + Asthma	6×6	7×6	12×5	10×6	0.51 (class 1)	Dust mites, grass pollen, *P lanceolata* pollen
8	F/40	Rhinitis	7×7	12×10	11×7	12×8	1.62 (class 2)	
9	F/36	Rhinitis + Asthma	12×11	14×9	22×11	15×13	2.51 (class 2)	*P lanceolata* pollen
10	F/40	Rhinitis + Asthma + Anaphylaxis	6×6	6×6	8×5	10×6	50.3 (class 5)	Almond
11	F/37	Rhinitis + Asthma	7×6	10×9	10×9	31×6	4.46 (class 3)	*P lanceolata* pollen, sunflower seed
12	F/35	Rhinitis + Asthma	10×7	14×14	14×11	30×7	23.0 (class 4)	Cat and dog furs
13	F/38	No	–	4×4	–	–	<0.35 (class 0)	Cat and dog furs

Abreviations: F, female; M, male; *P lanceolata, Plantago lanceolata; P ovata, Plantago ovata*.
[a] Subjects 1-7 and 13 belong to the prevalence study. Subjects 8-12 had known allergy to *P ovata* seed and were recruited for immunologic studies.

of the exposed population. The sensitized patients had been manipulating laxatives containing *P ovata* seed for a mean of 10 years (95% CI, 6.7-13.5 years), similar to the rest of the exposed subjects.

There were no statistically significant differences in the prevalence of atopy between exposed and nonexposed subjects: 24% (95% CI, 14% - 37%) and 31.7% (95% CI, 20% - 44%), respectively. However, the percentage of atopy in the sensitized group was significantly higher than in the remaining individuals who were exposed but not sensitized: 62% (95% CI, 25% - 89%) and 18% (95% CI, 9% - 31%), respectively ($P < .05$).

Sensitization to *P lanceolata* pollen and latex were also studied (Figure 1). There were no statistically significant differences in latex sensitization between exposed and nonexposed groups: 6.9% (95% CI, 2% - 17%) and 4.8% (95% CI, 1% - 14%), respectively. A higher percentage of *P lanceolata* pollen sensitization was found in the nonexposed (12.6%; 95% CI, 6% - 24%) than in exposed individuals (6.9%; 95% CI, 2% - 17%), although the difference was not statistically significant.

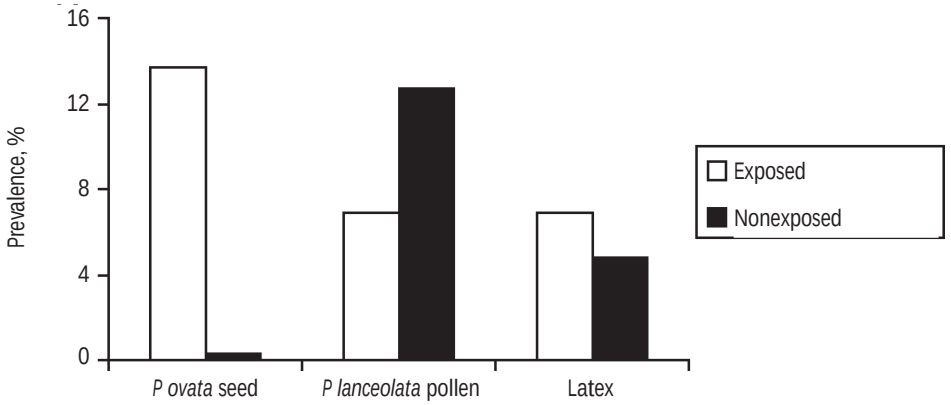

Figure 1. Prevalence of sensitization to *Plantago ovata* seed, *Plantago lanceolata* pollen, and latex in exposed and nonexposed subjects.

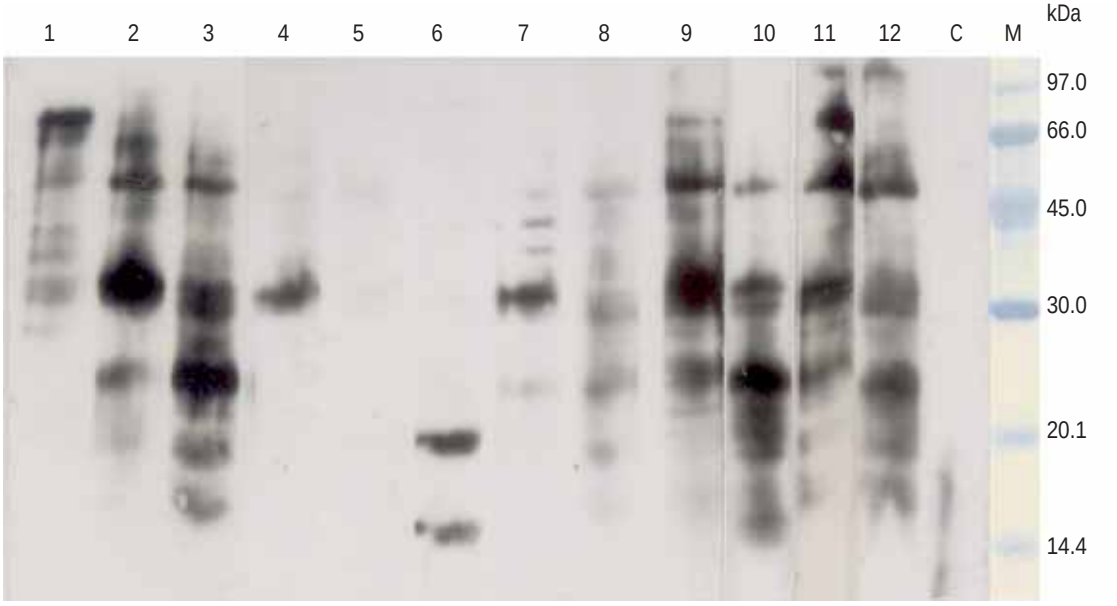

Figure 2. Immunoblotting for specific immunoglobulin E with an extract from a laxative containing *Plantago ovata* seed (Plantaben). Lanes 1-7: sera from subjects included in the prevalence study who were sensitized to *P ovata* seed. Lanes 8-12: sera from subjects with known allergy to *P ovata* seed recruited for immunoblotting and cross-reactivity studies. C indicates negative control (pool of sera from nonatopic subjects); M, molecular mass marker.

Immunoblotting and Cross-Reactivity

IgE immunoblotting revealed several IgE-binding components with an apparent molecular mass of 17, 20, 25, 32-34, 54, and >97 kDa (Figure 2). They appeared with sera from most of the patients, including patient 10, who, apart from respiratory symptoms, had suffered from 2 anaphylactic reactions after ingestion of laxatives containing *P ovata* seed. Immunoblotting with serum from subjects 1, 9, and 11, who were also sensitized to *P lanceolata* pollen, revealed another IgE-binding protein of 73-77 kDa.

Cross reactivity between *P ovata* seed and *P lanceolata* pollen was studied by EAST inhibition and immunoblot inhibition. Results of EAST inhibition using serum from subjects 1 and 11 with P lanceolata pollen as the solid phase showed a strong inhibition when *P ovata* seed was used as inhibitor (Figure 3). However, a mild inhibition was obtained with serum from subject 11 when *P ovata* seed was used as the solid phase and *P lanceolata* pollen as the inhibitor (data not shown).

Immunoblot inhibition results with *P ovata* seed extract as solid phase and *P lanceolata* pollen as inhibitor showed a complete inhibition of IgE binding when serum from subject 1 was used (patient with a much higher sensitization to *P lanceolata* pollen, class 4, than to *P ovata* seed, class

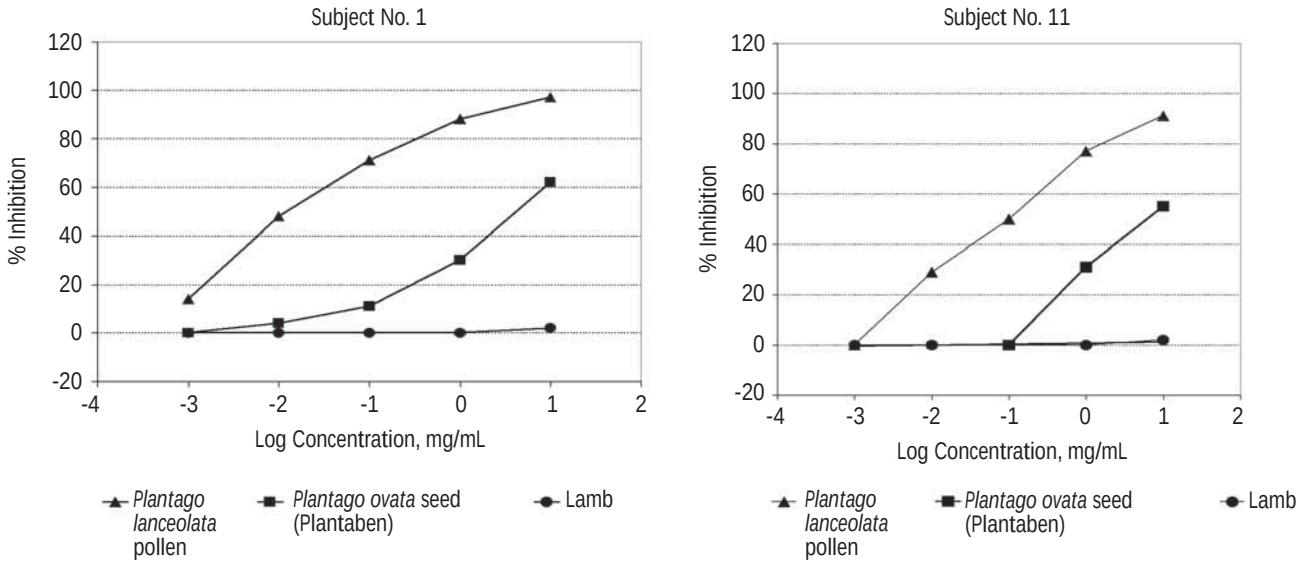

Figure 3. Results of enzyme allergosorbent test inhibition with *Plantago lanceolata* pollen extract as the solid phase.

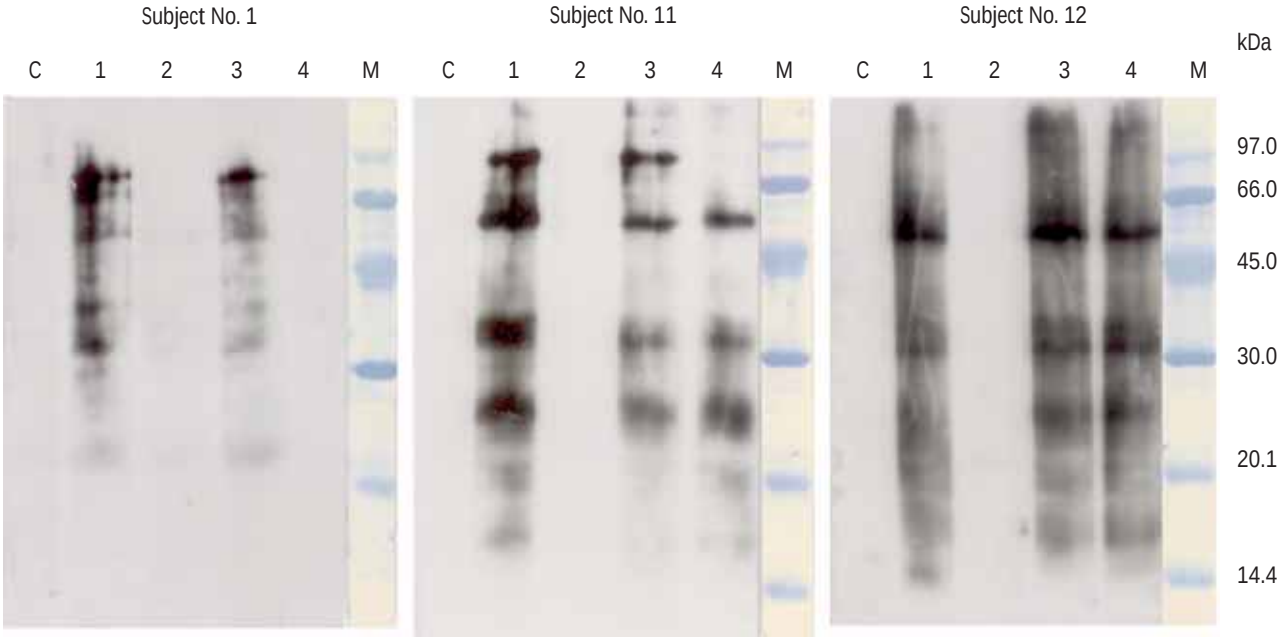

Figure 4. Immunoblot inhibition of *Plantago ovata* seed extract (Plantaben) with *Plantago lanceolata* pollen. Subjects 1 and 11 were sensitized to *P ovata* seed and *P lanceolata* pollen. Subject 12 was only sensitized to *P ovata* seed. Lane C: control serum (pool of sera from nonatopic subjects). Lane 1: patient serum. Lane 2: patient serum preincubated with *P ovata* seed extract, positive inhibition control. Lane 3: patient serum preincubated with lamb meat extract, negative inhibition control. Lane 4: patient serum preincubated with *P lanceolata* pollen extract (5 mg/mL). Lane M: molecular weight marker.

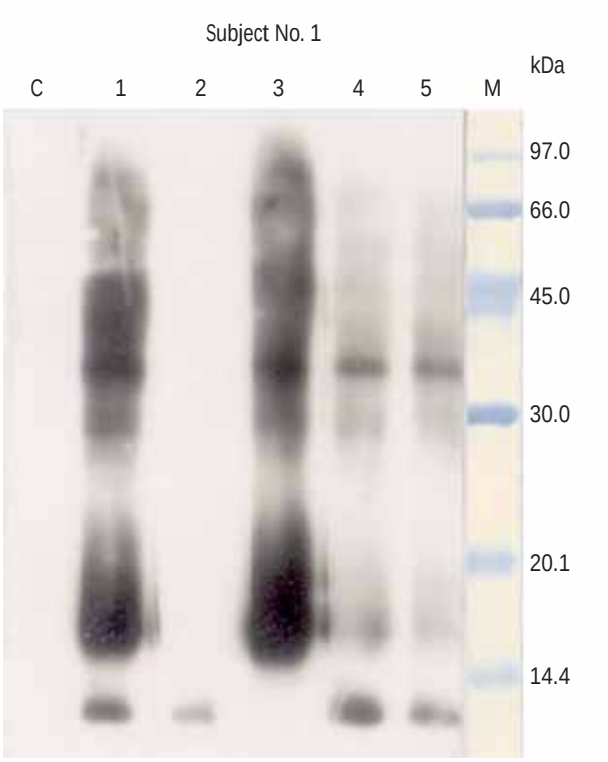

Figure 5. Immunoblot inhibition of *Plantago lanceolata* pollen extract using Plantago ovata seed (Plantaben) as inhibition phase with serum from patient 1. Lane C: control serum (pool of sera from nonatopic subjects). Lane 1: patient serum. Lane 2: patient serum preincubated with *P lanceolata* pollen extract, positive inhibition control. Lane 3: patient serum preincubated with lamb meat extract, negative inhibition control. Lanes 4 and 5: patient serum preincubated with different concentrations of *P ovata* seed extract (5 and 10 mg/mL, respectively). Lane M: molecular weight marker.

1). However when serum from subject 11 was used, only an IgE-binding band of 73-77 kDa disappeared (patient 11 showed a similar level of sensitization to both *P ovata* seed (class 3) and *P lanceolata* pollen (class 2). No inhibition at all was observed with serum from subject 12, who was only sensitized to P ovata seed (Figure 4). When immunoblots were carried out with *P lanceolata* pollen in the solid phase, *P ovata* seed extract as inhibitor, and using serum from subject 1, a high level of inhibition was obtained and only an IgE-binding band of 40 kDa remained (Figure 5). When the test was performed using serum from subject 11, the only apparent inhibition involved a high molecular weight band (data not shown).

Discussion

The main objective of our study was to determine the prevalence of clinical allergy and sensitization to *P ovata* seeds in health care professionals working at geriatric care homes and to compare these results with a nonexposed group of health professionals. The prevalence of sensitization (13.8%) and clinical allergy (8.6%) to *P ovata* seed described in the present study is similar to that obtained by other authors who described a prevalence of IgE sensitization to *P ovata* seed in health care professionals at chronic care hospitals (nurses, auxiliary nurses, and assistants) of between 5% by skin testing and 12% by measurement of specific serum IgE levels (radioallergosorbent test [RAST]) [10]. In our case, sensitization was higher by prick test, as only 4 of the 8 sensitized patients were positive for specific IgE.

Several factors associated with sensitization to *P ovata* seeds were considered in this study, including sex, age, atopy, period of exposure, and pharmaceutical form of *P ovata* laxatives. There were no differences between sensitized individuals and nonsensitized exposed subjects in terms of age, sex, or number of years handling laxatives containing *P ovata* seed. Most workers had been handling *P ovata* seed laxatives for 6 to 15 years, with a mean of 10 years, a similar exposure to that observed in other epidemiologic studies, such as that of Malo et al [10]. As in our study, the group studied by Malo et al was predominantly composed of women between 25 and 45 years of age.

Sensitization to *P ovata* seed usually occurs by inhalation of dust particles, which can be as small as 2 μm [4,13]. Health care professionals working with elderly patients inhale *P ovata* seed particles when they mix the laxative with water, an operation that is repeated many times each day, considering the number of individuals cared for by those professionals. Thus, allergy to *P ovata* seed should be considered an occupational disease and measures should be taken to prevent exposure.

According to the formulation of the laxatives, Plantaben has effervescent properties (due to a mixture of tartaric acid and bicarbonate), leading to significant dispersion of *P ovata* seed particles when it is mixed with water. Among the laxatives containing *P ovata* seeds, Plantaben was the most prescribed in the geriatric care homes studied. The high level of consumption of this laxative and its particular effervescent properties could explain the high prevalence of sensitization to *P ovata* seed observed in health care professionals, demonstrated by positive prick test, not only to Plantaben but also to any laxative containing *P ovata* seed. We are in agreement with proposals to use products with lower amounts of airborne particles or granulated forms to reduce the potential risk of sensitization to P ovata seed [5, 21, 28].

We found an association between *P ovata* sensitization and atopy. The prevalence of atopy in the group of sensitized individuals was higher than in the rest of the sample (62.5% vs 18%). This finding is consistent with previous studies indicating that atopy may be a predisposing factor in *P ovata* seed sensitization [11].

Among the allergens studied, *P lanceolata* pollen and latex have been considered in more detail. Despite the botanical relationship between *P ovata* and *P lanceolata*, we did not find a higher prevalence of allergy and sensitization to *P lanceolata* pollen than to other allergens; in fact, sensitization to *P lanceolata* pollen was even higher in the nonexposed subjects. There were no differences in the prevalence of latex sensitization between exposed (6.9%) and nonexposed subjects (4.8%), a finding that is consistent with the results of previous studies in health professionals [28].

However, the rate of *P ovata* seed sensitization observed (13.8%) was higher than that found for latex sensitization, suggesting that, *P ovata* should be considered as a potential allergen in health workers who handle laxatives containing *P ovata* seed.

Analysis of *P ovata* seed allergenic components by immunoblotting revealed IgE-binding proteins of 17, 20, 25, 32-34, 54, 73-77, and >97 kDa, values which are similar to those reported in previous studies (allergenic proteins ranging from 10 to 66 kDa) [1,21,30]. As described previously [9], we also identified an IgE-binding protein of approximately 77 kDa. In our case, this protein was only observed when *P ovata* seed extract was incubated with serum from subjects sensitized to both *P ovata* seed and *P lanceolata* pollen (subjects 1, 9, and 11). The same IgE-binding proteins were observed when *P ovata* seed was incubated with sera from patients with respiratory symptoms and with serum from a patient who suffered an anaphylactic reaction. Our results suggest that there is no specific immunoblot pattern associated with respiratory or systemic symptoms, as previously suggested [9].

Although most studies have found an absence of cross-reactivity between *P ovata* seed and *P lanceolata* pollen [20,21], some have described a significant level of cross-reactivity (60% inhibition of *P lanceolata* pollen by *P ovata* seed extract in RAST-inhibition assay) [19]. The results obtained in our study with inhibition assays (EAST and immunoblot inhibition) indicate the existence of a low level of cross-reactivity. However, more studies are required to establish the clinical relevance of these findings.

Few cross-reactivity studies have been reported in the literature between pollen and seeds of the same plant. Singh et al [31] described the presence of cross reactivity between pollen and seed from Ricinus communis and Blanco et al [32] between Carica papaya pollen, papaya fruit, and papain. Cross-reactivity studies with other allergens like pollen and seed from sunflower [33] or *Pinus radiata* pollen and pine cones [34] did not reveal a clear association between pollen and seed allergens.

In summary, our results demonstrate that *P ovata* seed could be a potential allergen for those health workers who handle products containing *P ovata* seed powder. The prevalence of sensitization and clinical allergy to *P ovata* seed was even higher than that obtained for latex sensitization in our study group. Furthermore, *P ovata* seed can be responsible not only for respiratory symptoms but could also be a risk factor for systemic reactions in those sensitized subjects who ingest foods containing *P ovata* seed. Avoidance of this allergen is easy and several measures have already been taken in the geriatric care homes studied here, such as changing *P ovata* laxatives for others or using *P ovata* laxatives in formulations that cause less dispersion of airborne particles. Although we have demonstrated immunologic cross-reactivity between *P ovata* seed and *P lanceolata* pollen, further studies will be required to determine its clinical implications.

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