

English plantain and psyllium: lack of cross-allergenicity by crossed immunoelectrophoresis

Marjorie S Morgan, PhD*; Larry G Arlian, PhD*; DiAnn L Vyszanski-Moher*; James Deyo, DVM, PhD†; Thomas Kawabata, PhD‡; and Enrique Fernandez-Caldas, PhD‡

31312

Background: English plantain (*Plantago lanceolata*) weed pollen and psyllium (*Plantago ovata*) husk dust are inhalant allergens. Because of the phylogenetic relationship between these plant species, cross-allergenicity has been a concern.

Objective: The purpose of this study was to investigate the possible cross-allergenicity of plantain and psyllium.

Methods: Homologous and heterologous crossed immunoelectrophoresis (CIE) were performed using a commercial English plantain pollen extract and an extract of psyllium seed embryo. Crossed radioimmuno-electrophoresis (CRIE) was performed using sera from subjects who were RAST positive only to plantain (group A), RAST positive only to psyllium (group B), RAST positive to both plantain and psyllium (group C), or RAST negative to both (group D).

Results: All of the group A plantain subjects showed IgE binding to at least one of the six plantain allergens in homologous plantain CRIEs while only one of the sera from the group B subjects reacted very weakly to these plantain allergens. In homologous psyllium CRIE, all group B subjects showed pronounced IgE binding to 2 to 7 of the seven psyllium allergens. Several of the plantain subjects demonstrated only very weak binding to psyllium allergens. Heterologous CRIEs demonstrated little relevant IgE binding.

Conclusions: These results indicate that there is little cross-allergenicity between psyllium husk and English plantain pollen.

INTRODUCTION

English plantain (*Plantago lanceolata*) is a weed found in many temperate regions of the world. In the United States, this plant is commonly found in the northwest coastal regions and in most of the eastern half of the country. It is found in urban as well as rural areas and has a pollen season lasting from May to October.^{1,2} Pollen from this plant has been widely reported to cause hay fever and asthma. In 1925, Bernton reported that 4.3% (5/116) of subjects with hay fever in Washington,

DC, showed cutaneous sensitivity to plantain.¹ More recent studies have shown that 20% to 34% of subjects with respiratory allergies surveyed in England were RAST positive to plantain,^{3,4} while 4% of 49,660 allergic individuals tested in Italy showed sensitivity.⁵

Plantain is a member of the Plantaginaceae family of plants which also includes psyllium (*Plantago ovata*). Psyllium, also known as ispaghula, is grown mainly in Mediterranean countries, Asia and the Indian sub-continent.⁶ The psyllium seed coat (husk) yields a hydroscopic, mucilaginous material that is a good source of soluble dietary fiber and preparations made from psyllium husk have been used in this country as bulk-forming laxatives for more than 60 years.⁶

Several million Americans use psyllium preparations on a daily basis and relatively few allergic reactions have been published. The first documented

case of psyllium allergy was reported in 1941 and involved a worker in a psyllium manufacturing plant.⁷ Since then, most reported cases of psyllium sensitivity have involved either individuals working in psyllium processing plants⁸⁻¹⁵ or nurses, pharmacists, or other health care workers who dispense psyllium laxative formulations in health care facilities.¹⁶⁻³¹ Both of these groups of workers are exposed to psyllium dust which is produced either during the manufacturing process or when medication is improperly dispensed. This suggests that inhalation is the primary route of sensitization to this allergen.

The phylogenetic relationship between English plantain and psyllium has led to much speculation concerning possible cross-allergenicity between psyllium husk preparations and plantain pollen.^{7-10,12,14,20,23,27,31} Although it was recently demonstrated that the allergens present in psyllium husk preparations are the result of contamination by other psyllium seed components,^{32,33} the possibility that plantain-sensitive individuals react to these residual psyllium allergens was not addressed. The objective of this study was to investigate the possible cross-allergenicity of psyllium and plantain. Using crossed radioimmuno-electrophoresis (CRIE), we have found that no significant cross-allergenicity can be demonstrated.

MATERIALS AND METHODS

Human Sera

Sera were obtained from 38 atopic individuals, none of whom was receiving immunotherapy for English plantain or psyllium (Table 1). RASTs for English plantain, psyllium (ispaghula), and total IgE were performed on all sera.

From *the Department of Biological Sciences, Wright State University, Dayton, Ohio; †the Procter & Gamble Co., Cincinnati, Ohio; and ‡the Division of Allergy and Clinical Immunology, University of South Florida, Tampa, Florida.

Funding for this study was provided by The Procter & Gamble, Co., Cincinnati, Ohio.

Received for publication November 14, 1994.

Accepted for publication in revised form February 1, 1995.

Table 1. RAST Scores and Total IgE Levels for Sera Used

Subject Number	RAST Score		Total IgE, units/mL
	English Plantain	Psyllium	
Group A - Plantain-sensitive only			
1	III	0/I	270
2	III	I	562
3	II	I	>1000
4	II	0/I	279
5	II	0/I	417
6	III	I	436
7	III	I	610
8	III	0/I	573
9	III	0/I	280
10	IV	0/I	ND
11	IV	0/I	796
12	IV	0/I	294
13	IV	0	304
14	III	0	259
15	IV	I	>1000
Group B - Psyllium-sensitive only			
16	0	III	33
17	0	III	43
18	I	V	ND
19	0	I/II	19
20	0	III	47
21	I	III/IV	349
Group C - Plantain and Psyllium sensitive			
22	II	II	>1000
23	II/III	IV	321
24	II	III/IV	ND
25	IV	IV	405
26	II	III	124
27	III	III	213
28	II	V	123
29	III	III/IV	785
Group D - Controls			
30	0	0	6
31	0	0	50
32	I	0/I	782
33	0/I	0	125
34	0	0	25
35	0	0	34
36	0/I	0	78
37	0	0	31
38	0/I	0	58

ND = not determined.

Subjects 1 to 15 (group A) were RAST positive ($\geq$ class II) to English plantain and RAST negative ($\leq$ class I) to psyllium. These were subjects who presented at an allergy clinic in an area where English plantain is prevalent. Subjects 16 to 21 (group B) were RAST positive to ispaghula (all except 19 were $\geq$ class III) and negative ($\leq$ class I) to plantain. The group B

subjects were all workers in a bulk-laxative manufacturing plant who were initially identified as psyllium-sensitive through routine occupational RAST monitoring. Group C was composed of eight individuals (subjects 22 to 29) who were RAST positive to both English plantain and psyllium (both $\geq$ class II). Nine individuals (30 to 38, group D) were included as negative

controls. Several of these subjects had elevated circulating IgE levels and none was sensitive to either plantain or psyllium (both $\leq$ class I).

Extracts

Although psyllium laxatives and cereals are made from the husk of the psyllium seed, the allergens present in these preparations are actually contributed by contaminating embryo and endosperm seed components.^{32,33} Additionally, the viscous nature of psyllium husk extracts makes them unsuitable for CIE analysis.³³ We have therefore used a psyllium seed embryo extract as the source of psyllium antigens/allergens in this study.

An extract of psyllium seed embryo (*Plantago ovata*) was prepared as described in the literature.³³ English plantain pollen extract (*Plantago lanceolata*), 1:10 (wt/vol) was obtained from Hollister-Stier (Spokane, WA).

Rabbit Antisera

Rabbit antisera to psyllium seed embryo, psyllium seed husk, and to English plantain pollen extract were produced in New Zealand White rabbits as described in the literature.³³

CIE and CRIE

The numbers of antigens and IgE-binding allergens in the English plantain pollen and psyllium embryo extracts were studied using crossed immunoelectrophoresis (CIE) and crossed radioimmunoelectrophoresis (CRIE).³³⁻³⁶ Homologous reactions were performed by electrophoresing 45 μ L of English plantain pollen extract or 20 μ L of psyllium embryo extract in the first dimension at 200 V for 30 minutes. Second dimension anodal gels contained 300 μ L of rabbit antiserum to English plantain or 180 μ L of antiserum to psyllium husk in 3 mL of 1% agarose, while the cathodal gels contained 10 μ L of English plantain antiserum or 60 μ L of psyllium embryo antiserum in 1 mL of agarose. Second dimension gels were electrophoresed at 50 V overnight. Heterologous reactions were performed similarly except that plantain extract was electrophoresed into antiserum to

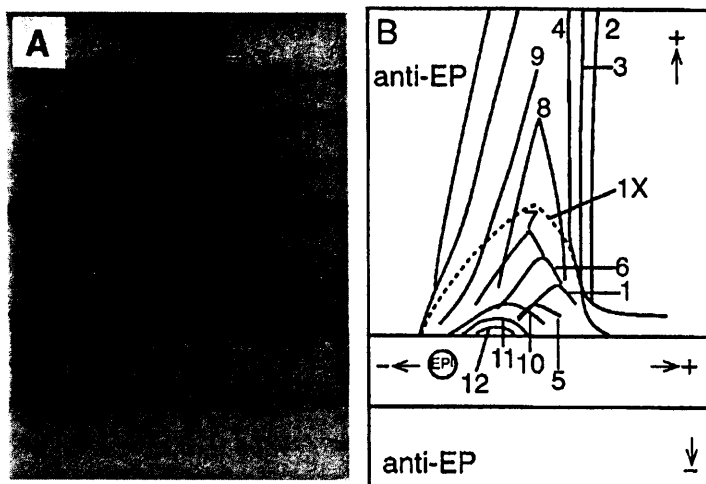


Figure 1. English plantain pollen extract (EP) reacted with rabbit anti-EP serum. (A) Coomassie blue stained homologous CIE and (B) schematic profile of homologous CIE pattern.

psyllium husk or embryo while psyllium extract was reacted with English plantain antiserum. Crossed immunoelectrophoresis plates were stained with Coomassie Brilliant Blue R-250 to visualize antigen/antibody precipitates.

IgE-binding allergens were visualized using CRIE.³³⁻³⁶ Briefly, CIE gels were incubated overnight in human serum diluted 1:20 (vol/vol) in 0.1 M sodium phosphate, 0.2% BSA, 1% Tween 20, 1% NaN₃, pH 7.5. Gels were washed and incubated with 1 million cpm ¹²⁵I-labeled antihuman IgE (Kallestad™ Allercoat™ RAST isotope reagent, Sanofi Diagnostics Pasteur, Inc., Chaska, MN) diluted in 20 mL 0.1 M sodium phosphate, 4% calf serum, 1% Tween 20, 1% NaN₃, pH 7.5. Gels were washed, dried, and placed in cassettes with tungsten-enhancement screens and Kodak X-OMAT RP film at -80 °C. Film was exposed for three, five or nine days. To standardize and to make comparisons both within and among sera, all gels were processed identically in one batch with one lot of reagents. The relative intensity of IgE binding was assessed using a - (no radiostaining on 9-day autoradiograms) to + + + + (pronounced radiostaining) scale.

RESULTS

Reaction of Plantain Extract with Plantain Antiserum (Homologous)

The antigenic profile of English plantain pollen extract was examined using crossed immunoelectrophoresis (CIE). When commercial English plantain pollen extract was reacted with antiserum from rabbits immunized with this plantain extract, 12 Coomassie blue staining antigens were detected in the extract (Fig 1). All antigens were electronegative and migrated toward the anode. Five of these 12 antigens (antigen/antibody precipitates) bound

IgE when screened using the 38 patients' sera assayed by crossed radioimmunoelectrophoresis (CRIE) (Fig 2A, Table 2 and 3). Crossed radioimmunoelectrophoresis analysis also revealed one additional allergen (designated 1X) not visualized on the Coomassie blue stained gel (Fig 2A). Sera from all 15 of the plantain-sensitive (by RAST) individuals in group A showed IgE binding to 1 to 3 of the plantain allergens (Table 2). Thirteen of these subjects (87%) showed binding to antigen #8. When subjects recognized antigen #3 as an allergen, radiostaining was distinctly visible on 3-day autoradiograms suggesting that this was the most potent allergen (Fig 2B). No subject had circulating IgE to all of the six allergens (Table 2).

Only one of the sera of the six psyllium-sensitive (by RAST) subjects in group B exhibited any IgE binding to plantain allergens and this binding was barely detectable and limited to only one of the six plantain allergens (Fig 2C, Table 3). Sera from three of the eight group C subjects also demonstrated IgE binding to 1 to 3 plantain allergens (Table 3). None of the negative control sera showed radiostaining indicative of IgE binding to any plantain allergen, even though several had high levels of circulating IgE directed at other allergens (eg, house dust mites).

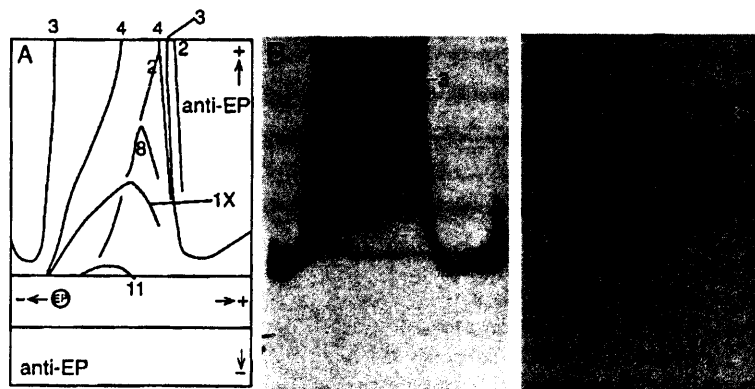


Figure 2. Autoradiograms of IgE binding to CIE gels of English plantain pollen extract (EP) reacted with rabbit anti-EP serum. (A) Schematic composite CRIE profile, (B) representative 9-day autoradiogram using serum from plantain-sensitive subject 13, and (C) representative 9-day autoradiogram using serum from psyllium-sensitive subject 21.

Table 2. IgE Binding to English Plantain Pollen Extract Antigens Precipitated with Rabbit Anti-English Plantain Pollen Serum

Subject No.	English Plantain Antigen Number (EP × EP)					
	#2	#3	#4	#8	#11	#1X
Group A - Plantain-sensitive only (by RAST)						
1	—	—	—	X	X	—
2	—	—	—	X	X	—
3	—	—	—	X	—	—
4	—	—	—	X	X	—
5	—	—	—	X	—	X
6	—	—	—	X	X	—
7	—	—	—	X	—	—
8	—	—	—	X	—	—
9	—	—	—	X	X	X
10	X	—	X	X	—	—
11	—	X	X	X	—	—
12	—	X	X	X	—	—
13	—	X	X	X	—	—
14	—	—	X	—	—	—
15	X	—	X	—	—	—

EP × EP = English plantain pollen extract reacted with English plantain pollen antiserum; X = IgE binding observed; and — = no IgE binding seen by nine days.

Reaction of Psyllium Extract with Psyllium Antisera (Homologous)

When psyllium extract was reacted with psyllium husk antiserum, four Coomassie blue staining antigen/antibody precipitates and seven radiostaining allergens could be detected (Fig 3, Table 4). Psyllium allergens #2 and 3 could be seen as well-defined peaks on CIE and as darkly-radiostaining peaks on CRIE (Fig 3A, C). All of the psyllium-sensitive persons in groups B and C showed binding to both peaks #2 and 3 with 6 of the 14 having clearly visible peaks on three days of autoradiography which indicated high levels of circulating IgE directed at these allergens (Table 3 and 4). Most of these subjects also showed IgE binding to both allergens #1 and 3X. Two subjects (group B subject 16 and group C subject 23) had serum IgE directed at all seven psyllium embryo antigens.

In contrast to the strong IgE binding exhibited by sera from psyllium-sensitive subjects in both groups B and C, plantain-sensitive subjects in group A showed only very weak IgE binding to allergens #2 and 3 and this was only visible on 9-day autoradiograms (Table 3, Fig 3D). No sera from group A (plantain-sensitive only) subjects had IgE directed at epitopes on

allergens #1X, 2X, or 4. As expected, none of the sera from the control subjects showed IgE binding to any psyllium antigen.

Reaction of Plantain Extract with Psyllium Antisera (Heterologous)

When plantain pollen extract was reacted with rabbit antiserum to either psyllium seed husk (Fig 4A, B) or psyllium seed embryo (Fig 4C), only one diffuse Coomassie blue staining antigen/antibody precipitate was faintly visible on the CIE plate. Five plantain-sensitive subjects (Table 3; three from group A and two from group C) had faintly detectable (equivocal) serum IgE that bound to the cross-reacting antigen in English plantain extract that was precipitated by psyllium husk antiserum. Significantly, none of the group B subjects (RAST positive to psyllium and negative to plantain) showed any serum IgE directed at this antigen. Additionally, no subject had circulating IgE that bound to the plantain antigen precipitated by the psyllium embryo antiserum (Table 3).

Reaction of Psyllium Extract with Plantain Antiserum (Heterologous)

To study further the cross-reactivity between psyllium and English plantain, we

reacted psyllium embryo extract with antiserum to English plantain pollen (Fig 5). Three Coomassie blue stained antigen/antibody precipitates were visible on the CIE gels, all of which bound IgE from the psyllium-sensitive subjects' sera. All but one of the psyllium-sensitive subjects in both groups B and C exhibited serum IgE that bound to at least one of the psyllium antigens precipitated with antiserum to English plantain (Table 3). Five of these subjects had IgE antibodies directed at all three antigens; however, in this heterologous CRIE format, sera from only three of the 15 plantain-sensitive subjects in group A (20%) showed any IgE binding to psyllium antigen #1 and this very weak radiostaining was barely detectable even after nine days of autoradiography (Fig 5D). None of these persons had circulating IgE that bound to either psyllium antigen #2 or 3. As before, none of the negative control subjects exhibited any allergen-specific IgE.

DISCUSSION

There has been much speculation about the possibility of allergenic cross-reactivity between English plantain pollen and psyllium husk,^{4,7-10,12,14,20,23,27,31,33,37,38} however, no studies have directly investigated this. In this study we first

Table 3. Relative IgE Binding in Homologous and Heterologous CRIE by Serum from Study Subjects

Subject No.	Homologous CRIE		Heterologous CRIE		
	EP × EP	PE × PH	EP × PH	EP × PE	PE × EP
Group A - Plantain-sensitive only (by RAST)					
1	+++	+	+	-	-
2	++	+/-	-	-	-
3	+/-	+/-	-	-	-
4	++	+	-	-	-
5	++	+/-	-	-	-
6	++	+	+/-	-	-
7	+	+/-	-	-	-
8	+	+/-	-	-	-
9	++++	+	-	-	+/-
10	++	+/-	+	-	+/-
11	++++	+	-	-	-
12	++++	+	-	-	-
13	++++	-	-	-	-
14	+	+/-	-	-	+/-
15	+++	-	-	-	-
Group B - Psyllium-sensitive only (by RAST)					
16	-	++++	-	-	++
17	-	+	-	-	+
18	-	+	-	-	+
19	-	+/-	-	-	+/-
20	-	+++	-	-	++++
21	+/-	+	-	-	+
Group C - Plantain and Psyllium sensitive (by RAST)					
22	++	+/-	+	-	-
23	-	++++	-	-	++++
24	-	+++	-	-	++
25	+/-	+	-	-	+
26	-	+++	-	-	++++
27	++	+	+	-	++
28	-	+++	-	-	++++
29	-	+/-	-	-	+
Group D - Negative Controls					
30	-	-	-	-	-
31	-	-	-	-	-
32	-	-	-	-	-
33	-	-	-	-	-
34	-	-	-	-	-
35	-	-	-	-	-
36	-	-	-	-	-
37	-	-	-	-	-
38	-	-	-	-	-

EP × EP = English plantain pollen extract reacted with English plantain pollen antiserum, PE × PH = psyllium embryo extract reacted with psyllium husk antiserum, EP × PH = English plantain pollen extract reacted with psyllium husk antiserum, EP × PE = English plantain pollen extract reacted with psyllium embryo antiserum, PE × EP = psyllium embryo extract reacted with English plantain pollen antiserum, - = no IgE binding observed on 9-day autoradiograms, +/- = very weak (equivocal) IgE binding observed, and + to ++++ = moderate to strong IgE binding (moderate to high IgE levels in serum).

characterized the allergenicity of extracts of each plant species by directly examining serum IgE sensitivity to each plant species using four groups of subjects with different sensitivity profiles. We then examined the possibility of cross-reactivity

between extracts made from each plant species by comparing the four reference groups' sera IgE binding directly to (1) antigens in each extract (homologous CIE) and (2) cross-reacting antigens precipitated by heterologous CIE.

Our CIE analysis of a commercial English plantain pollen extract showed that it contained 12 Coomassie blue staining antigens precipitated by serum antibodies from rabbits immunized with this same extract. Crossed radio-immunoelectrophoresis analysis using

the sera of 38 atopic individuals determined that of these 12 antigens, five were IgE-binding allergens. Crossed radioimmuno-electrophoresis analysis also revealed the presence of an additional IgE binding allergen that was not stained by Coomassie blue. These results are in general agreement with those previously reported.^{37,38} Baldo et al³⁷ used the CIE and CRIE techniques and reported that extracts of English plantain pollen contained 16 antigens with at least six of these binding IgE. Another study also used CRIE to demonstrate five allergens in plantain extract.³⁸ Differences in the quality of the extracts, antisera, and the techniques used likely account for the slight discrepancy in the total number of allergens reported by different laboratories. Regardless, our results coupled with those of other researchers suggest that commercial extracts of English plantain pollen contain a minimum of six different allergens.

Previous examinations of the allergenicity of various psyllium seed components led to conclusions that psyllium seed embryo and endosperm, but not husk itself, were the sources of psyllium allergens in 95% pure commercial-grade husk material.^{32,33} Crossed radioimmuno-electrophoresis analysis using sera from 11 psyllium-sensitive individuals demonstrated the presence of four and five allergens in psyllium embryo and endosperm, respectively. Indirectly, it was demonstrated that these same allergens were also present in husk extracts but they originated from the endosperm and embryo that contaminated the husk material. Because the antigens in psyllium husk extract are more concentrated in an embryo extract and embryo extract can be electrophoresed in the CIE system while husk extract cannot,³³ we used embryo extract to detect serum IgE in the groups of reference sera used in this study. In the present study using psyllium embryo extract and antiserum to psyllium husk we detected seven immunoreactive allergens. By using a larger set of reference sera with varied sensitivities to psyllium, we have identified three addi-

tional weak allergens that were not detected in previous studies.^{32,33}

Other studies on plantain/psyllium cross-antigenicity have presented conflicting results. Some studies reported observing cross-reactivity between English plantain and psyllium^{7,8,10,14} while others did not.^{9,12,23,31} Most of these reports, however, have been case studies and used either RAST or cutaneous testing exclusively. Cross-reactivity cannot be directly investigated with these diagnostic techniques. In addition, agreement between RAST and skin test results for English plantain has been reported to be poor.³⁹

In the present study, we used RAST to select the reference sera and CRIE (homologous and heterologous) to examine directly plantain/psyllium cross-antigenicity and allergenicity. Crossed radioimmuno-electrophoresis was used because individual allergens can be visualized and are not altered as is the case with SDS-PAGE and Western blotting (eg, proteins denatured by heat and/or detergent treatment). The reference sera we used represented groups of patients with differing allergic sensitivities to English plantain (pollen) and psyllium (ispaghula). One group (A) was highly plantain sensitive (by

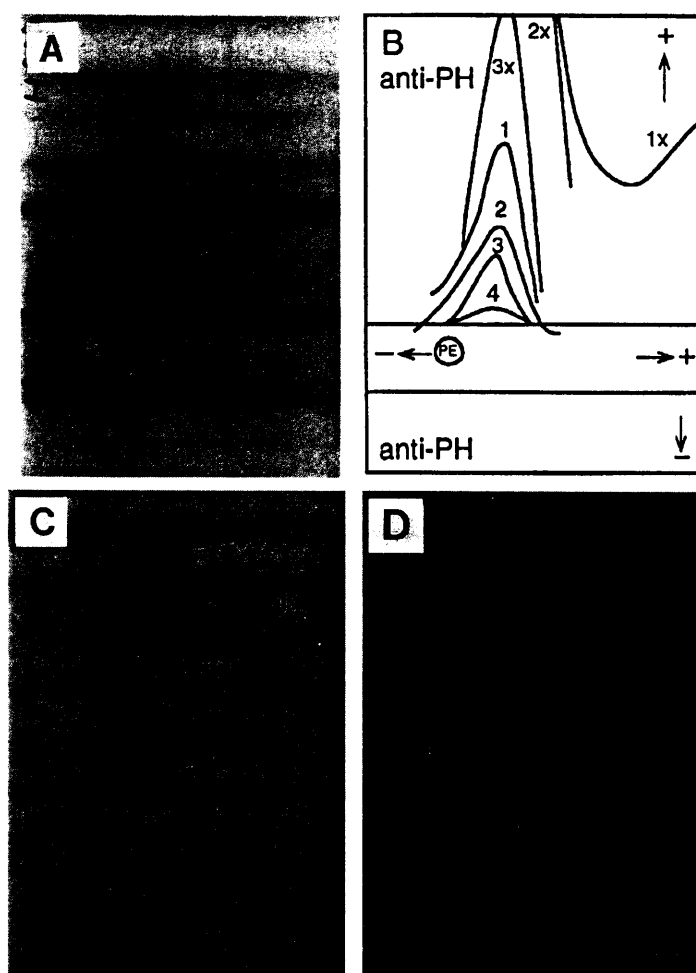


Figure 3. Psyllium embryo extract (PE) reacted with rabbit anti-psyllium husk (PH) serum. (A) Coomassie blue stained CIE, and (B) schematic composite CRIE profile. Three allergens (#1X, 2X, and 3X) did not stain with Coomassie blue and were not visible on the CIE gel. (C) Representative 9-day autoradiogram using serum from psyllium-sensitive subject 16. (D) Representative 9-day autoradiogram using serum from plantain-sensitive subject 1.

Table 4. IgE Binding to Psyllium Embryo Extract Antigens Precipitated with Rabbit Anti-Psyllium Husk Serum

Subject No.	Psyllium Embryo Antigen Number (PE × PH)						
	#1	#2	#3	#4	#1X	#2X	#3X
Group B - Psyllium-sensitive only (by RAST)							
16	X	X	X	X	X	X	X
17	X	X	X	—	—	—	—
18	X	X	X	—	—	—	X
19	—	X	X	—	—	—	—
20	X	X	X	—	—	—	X
21	X	X	X	—	—	—	X
Group C - Plantain and psyllium sensitive (by RAST)							
22	—	X	X	—	—	—	—
23	X	X	X	X	X	X	X
24	X	X	X	—	—	—	X
25	X	X	X	—	—	—	—
26	X	X	X	—	—	—	X
27	X	X	X	—	—	—	X
28	X	X	X	—	—	—	X
29	—	X	X	—	—	—	—

PE × PH = Psyllium embryo extract reacted with psyllium husk antiserum, X = IgE binding seen at nine days, and — = no IgE binding seen by nine days.

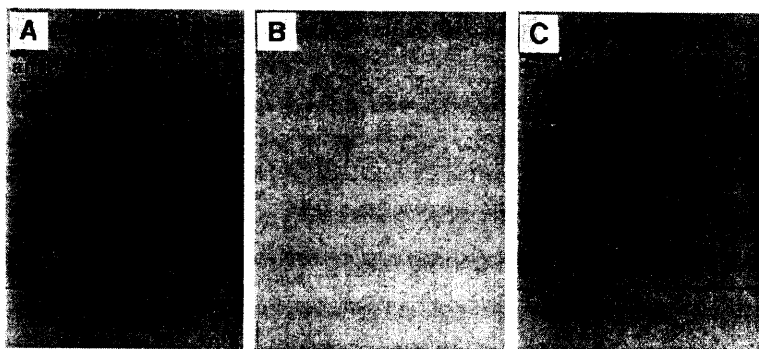


Figure 4. Heterologous CIE of English plantain pollen extract (EP) reacted with rabbit anti-psyllium serum. (A) Coomassie blue stained CIE of EP reacted with anti-psyllium husk (PH) serum, (B) representative 9-day autoradiogram using serum from plantain-sensitive subject 1, and (C) Coomassie blue stained CIE of EP extract reacted with rabbit anti-psyllium embryo (PE) serum.

RAST) with low psyllium sensitivity while the opposite group (B) was highly psyllium sensitive with low plantain sensitivity. A third group (C) was RAST positive to both plant species. When homologous English plantain CIE gels were incubated in sera from psyllium-sensitive individuals (group B), only one of six (17%) showed equivocal IgE binding to plantain allergens. Similarly, when homologous psyllium CIE gels were incubated in sera from plantain-sensitive subjects, only very weak binding to psyllium allergens was observed.

The heterologous CRIEs however, provided an even more dramatic demonstration of the low level of relevant cross-allergenicity between these two plant species. When English plantain extract was precipitated by psyllium antisera, none of the psyllium-sensitive (group B) and only two of the plantain/psyllium-sensitive (group C) individuals showed only very weak binding to the single diffuse precipitate. In the reciprocal heterologous reaction of psyllium embryo extract precipitated with antiserum to plantain pollen, sera from only three of the 15 plantain-

sensitive subjects in group A showed only extremely faint radiostaining indicating very low levels of IgE directed at the one cross-reactive psyllium allergen.

These results provide direct evidence that there is very little cross-allergenicity between antigens in extracts of psyllium and English plantain. The little IgE binding that was exhibited in heterologous reactions was displayed by sera that showed strong binding to the same allergen in the corresponding homologous reactions suggesting that the cross-allergenicity that was observed is probably of no clinical significance. This was the case when serum IgE from psyllium-sensitive subjects bound to psyllium allergenic epitopes on psyllium antigens precipitated by plantain antiserum. Conversely, only plantain-sensitive subjects had IgE that reacted with English plantain allergenic epitopes on plantain antigens precipitated by psyllium antiserum. No psyllium subject had IgE that bound to English plantain epitopes precipitated by psyllium antiserum and only three plantain-sensitive sera had slight amounts of IgE that bound to the psyllium antigen precipitated by plantain antiserum. These results indicate that

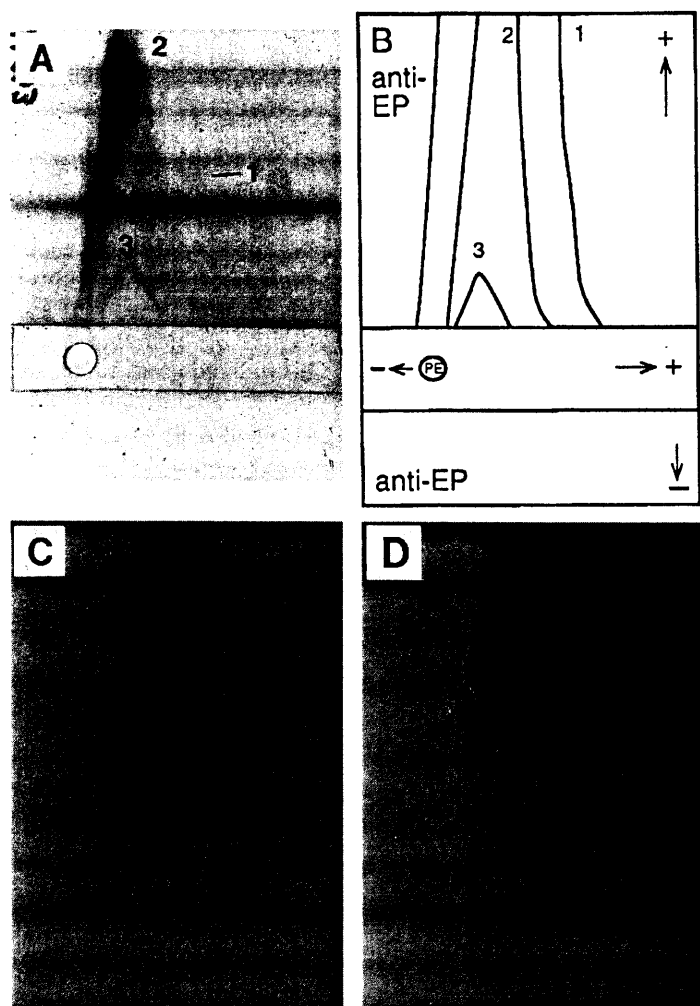


Figure 5. Psyllium embryo extract (PE) reacted with rabbit anti-English plantain (EP) serum. (A) Coomassie blue stained CIE, (B) schematic composite CRIE profile, (C) representative 9-day autoradiogram using serum from psyllium-sensitive subject 16, and (D) representative 9-day autoradiogram using serum from plantain-sensitive subject 10.

there is little reason for concern for cross-allergenic reactions or cross-sensitivity between English plantain and psyllium.

ACKNOWLEDGMENTS

The authors wish to thank Dr. David Sutton for helpful discussions in the planning of this study and Kelley Love for technical assistance.

REFERENCES

1. Bernton HS. Plantain hay-fever and asthma. *J Am Med Assn* 1925;84: 944-6.
2. Solomon WR. Aerobiology and inhalant

- allergens: Pollens and fungi. In: Middleton E Jr, Reed CE, Ellis EF, Adkinson NF Jr, Yunginger JW, Busse WW, eds. *Allergy principles and practice*. 4th ed. St. Louis: Mosby, 1993:480-98.
3. Merrett TG, Pantin CFA, Dimond AH, Merrett J. Screening for IgE-mediated allergy. *Allergy* 1980;35:491-501.
4. Mehta V, Wheeler AW. IgE-mediated sensitization to English plantain pollen in seasonal respiratory allergy: identification and partial characterisation of its allergenic components. *Int Arch Allergy Appl Immunol* 1991;96: 211-7.
5. Negrini AC, Arobba D. Allergic pollen and pollinosis in Italy: recent

- advances. *Allergy* 1992;47:371-9.
6. Youngken HW. Studies on commercial psyllium seeds. *J Am Pharm Assn* 1932;21:1265-73.
7. Ascher MS. Psyllium seed sensitivity. *J Allergy* 1941;12:607-9.
8. Busse WW, Schoenwetter WF. Asthma from psyllium in laxative manufacture. *Ann Intern Med* 1975; 83:361-2.
9. Göransson K, Michaelson NG. Ispaghula powder: an allergen in the work environment. *Scand J Work Environ Health* 1979;5:257-61.
10. Rosenberg S, Landay R, Klotz SD, Fireman P. Serum IgE antibodies to psyllium in individuals allergic to psyllium and English plantain. *Ann Allergy* 1982;48:294-8.
11. Machado L, Olsson G, Stalenheim G, Ketterström O. Dust exposure challenge tests as a measure of potential allergenicity and occupational disease risk in handling of ispaghula products. *Allergy* 1983;38:141-4.
12. Gauss WF, Alarie JP, Karol MH. Workplace allergenicity of a psyllium-containing bulk laxative. *Allergy* 1985;40:73-6.
13. Kirby J, Bardy JD, Malo JL, et al. Prevalence of IgE sensitization to psyllium in two pharmaceutical companies [Abstract]. *J Allergy Clin Immunol* 1986;77:134.
14. Bardy JD, Malo JL, Sequin P, et al. Occupational asthma and IgE sensitization in a pharmaceutical company processing psyllium. *Am Rev Respir Dis* 1987;135:1033-8.
15. McConnochie K, Edwards JH, Fifield R. Ispaghula sensitization in workers manufacturing a bulk laxative. *Clin Exp Allergy* 1990;20:199-202.
16. Gross R. Acute bronchospasm associated with inhalation of psyllium hydrophilic mucilloid. *JAMA* 1979;241: 1573-4.
17. Machado L, Zetterström O, Fagerberg E. Occupational allergy in nurses to a bulk laxative. *Allergy* 1979;34:51-5.
18. Suhonen R, Kantola I, Björkstén F. Anaphylactic shock due to ingestion of psyllium laxative. *Allergy* 1983;38: 363-5.
19. Machado L, Stalenheim G. Respiratory symptoms in ispaghula-allergic nurses after oral challenge with ispaghula suspension. *Allergy* 1984;39: 65-8.
20. Seggev JS, Ohta K, Tipton WR. IgE mediated anaphylaxis due to a psylli-

- um-containing drug. *Ann Allergy* 1984;53:325-6.
21. Zaloga GP, Hielwimmer UR, Engler RJ. Anaphylaxis following psyllium ingestion. *J Allergy Clin Immunol* 1984;74:79-80.
22. Schoenwetter WF, Steinberg P. Psyllium hypersensitivity, nurses, and geriatric units. *Ann Intern Med* 1985;103:642.
23. Pozner LH, Mandarano C, Zitt MJ, et al. Recurrent bronchospasm in a nurse. *Ann Allergy* 1986;56:14-5,44-7.
24. Cartier A, Malo JL, Dolovich J. Occupational asthma in nurses handling psyllium. *Clin Allergy* 1987;17:1-6.
25. Nelson WL. Allergic events among health care workers exposed to psyllium laxatives in the workplace. *J Occupat Med* 1987;29:497-9.
26. Jancelewicz Z, Dolovich J, Sussman GL. Psyllium allergy in hospital personnel: a report on twelve patients. *Can Fam Physician* 1988;34:573-6.
27. Kaplan MJ. Anaphylactic reaction to "Heartwise." *N Engl J Med* 1990;323:1072-3.
28. Lantner RR, Espiritu BR, Zumerchik P, Tobin MC. Anaphylaxis following ingestion of a psyllium-containing cereal. *JAMA* 1990;264:2534-6.
29. Malo JL, Cartier A, L'Archeveque J, et al. Prevalence of occupational asthma and immunologic sensitization to psyllium among health personnel in chronic care hospitals. *Am Rev Respir Dis* 1990;142:1359-66.
30. Sussman GL, Dorian W. Psyllium anaphylaxis. *Allergy Proc* 1990;11:241-2.
31. James JM, Cooke SK, Barnett A, Sampson HA. Anaphylactic reactions to a psyllium-containing cereal. *J Allergy Clin Immunol* 1991;88:402-8.
32. Vyszynski-Moher D, Arlian L, Lawrence A, et al. Psyllium antigenic analysis and lack of cross-reactivity between psyllium and English plantain [Abstract]. *J Allergy Clin Immunol* 1990;85:152.
33. Arlian LG, Vyszynski-Moher DL, Lawrence AT, et al. Antigenic and allergenic analysis of psyllium seed components. *J Allergy Clin Immunol* 1992;89:866-76.
34. Svendsen PJ, Weeke B, Johansson B-G. Chemicals, solutions, equipment and general procedures. *Scand J Immunol* 1983;17(Suppl 10):3-20.
35. Hoiby N, Axelsen NH. Crossed immunoelectrophoresis as modified for quantitative purposes. *Scand J Immunol* 1983;17(Suppl 10):125-34.
36. Weeke B, Sondergaard I, Lind P, et al. Crossed radioimmuno-electrophoresis (CRIE) for identification of allergens and determination of the antigenic specificities of patients' IgE. *Scand J Immunol* 1983;17(Suppl 10):265-72.
37. Baldo BA, Chensee QJ, Howden MEH, Sharp PJ. Allergens from plantain (*Plantago lanceolata*): studies with pollen and plant extracts. *Int Arch Allergy Appl Immunol* 1982;68:295-304.
38. Dreborg S, Sjogren I, Eriksson NE, Einarsson R. Selection of patients for biological standardization as exemplified by standardization of mugwort, goosefoot and English plantain pollen allergen extracts/preparations. *Allergy* 1987;42:485-95.
39. Pascual HC, Reddy PM, Nagay H, et al. Agreement between radioallergosorbent test and skin test. *Ann Allergy* 1977;39:325-7.

Request for reprints should be addressed to:
Larry G Arlian, PhD
Department of Biological Sciences
Wright State University
Dayton, Ohio 45435