

Immediate skin test reactivity to common aeroallergens in patients with respiratory allergies: A comparative analysis of allergen-induced skin reactions and their histamine controls

Luis R. Lopez, MD, Yovina Noriega, and Ricardo Losno, MD Lima, Peru

The results of the immediate skin test response to a panel of 16 common aeroallergens performed in a group of 659 consecutive patients with symptoms suggestive of a respiratory allergy were analyzed. A group of 108 healthy individuals served as control subjects. Ninety-four percent of the patients and 87% of the control subjects had at least one allergen-induced reaction (wheal ≥ 2 by 2 mm). The prevalence of positive skin reactions to each aeroallergen was equally high in both groups. However, if a skin reaction is considered as positive only when an allergen-induced wheal is equal or larger compared to the 50% of the wheal obtained with the histamine control in that individual, 70% of the patients had positive skin reactions and only 38% of the control subjects were positive ($p < 0.05$). Similarly, the prevalence rates to five aeroallergens (pollen, Fusarium, Mucor, Pullularia, and Curvularia) in the patient group were reduced to those levels observed with the control group, suggesting they are clinically less important. The age and not the sex influenced both the prevalence rates ($p < 0.001$) and the mean size ($p < 0.01$) of allergen and histamine-induced skin reactions. Lower prevalence rates and mean size values were observed in the youngest group (0 to 9 years). Moreover, there was an inverse relationship between lower skin reactivity with more younger subjects in our patient population. These results indicate that patients and healthy individuals have similar mechanisms for skin reactivity. The size of the allergen-induced wheal reaction compared to the histamine-induced control wheal reaction appears to be an important consideration for the correct clinical interpretation of the results. The observation that there were more younger patients with lower skin reactivity suggests that other mechanisms not detected by skin testing may play a role in the pathogenesis of respiratory allergies. (J ALLERGY CLIN IMMUNOL 1988;81:1143-8.)

The immediate skin test response to allergens represents the most common diagnostic procedure used in routine clinical evaluations of allergic diseases and has been demonstrated to correlate well with specific IgE levels¹ and with clinical symptoms and provocative challenges.² Therefore, the knowledge of the prevalent local aeroallergens and the characteristics of their reactions elicited by skin tests represent important considerations for the correct clinical interpreta-

Abbreviation used

PBS: Phosphate-buffered saline

From the Department of Microbiology (Immunology Section) Universidad Peruana Cayetano Heredia, Lima, Peru.

Supported in part by a grant from Consejo de Ciencia y Tecnologia (CONCYTEC) of Peru.

Received for publication Nov. 20, 1986.

Accepted for publication Dec. 26, 1987.

Reprint requests: Ricardo Losno, MD, Universidad Peruana Cayetano Heredia, Department of Statistics, PO Box 5045, Lima, Peru.

tion of the results. In Lima, Peru, the occurrence of respiratory symptoms believed to be IgE related is impressively common. However, a single retrospective study published in 1977 estimated the incidence of bronchial asthma in Peru to be only 0.9% in the general population without properly considering the characteristics of etiologic factors and diagnostic procedures.³ More recent and systematic studies regarding the prevalence of aeroallergens and their skin test reactivity in our population are lacking. Lima is located at sea level on the western desert coast of Peru, with minimal indigenous vegetation that eliminates pollens as relevant local aeroallergens. In contrast,

TABLE I. Skin test wheal reactivity

Allergen	Patients (N = 659)		Control subjects (N = 108)	
	All reactions		All reactions	
	$\geq 2 \times 2$ mm (%)	$\geq 50\%$ Histamine reaction* (%)	$\geq 2 \times 2$ mm (%)	$\geq 50\%$ Histamine reaction* (%)
<i>Cladosporium</i>	70	29	62	15
<i>Candida</i>	69	30	60	13
<i>Alternaria</i>	67	29	48	7
<i>Penicillium</i>	65	28	48	9
<i>Aspergillus</i>	64	22	52	12
<i>Helminthosporium</i>	63	23	56	11
Dog	62	21	59	15
House dust	61	23	52	10
<i>Hormodendrum</i>	60	22	48	12
<i>Rhizopus</i>	59	21	52	10
Cat	56	22	51	8
<i>Curvularia</i>	54	13	44	5
<i>Pullularia</i>	48	8	40	1
Pollen	45	8	41	8
<i>Mucor</i>	32	5	14	2
<i>Fusarium</i>	20	2	11	0

* Allergen-induced wheals equal or larger compared to the 50% of the wheal obtained with histamine in that individual were recorded as positive.

mild temperatures throughout the year associated with high humidity and drizzle during the winter months allow molds and house dust mites to proliferate, assigning them an important role as aeroallergens.

This study was undertaken to determine the prevalence of skin test reactivity to common local and clinically relevant aeroallergens in patients with respiratory allergic disorders, the value of the size of the allergen-induced wheal reaction compared to the histamine-induced control wheal reaction in establishing the diagnosis of atopy, as well as the role of age and sex on the skin test reactivity.

MATERIAL AND METHODS

Subjects

The results of the immediate skin test response to a panel of 16 common aeroallergens performed during 1983 on a group of 659 consecutive patients were analyzed. These patients were referred to us for skin testing from the allergy and pulmonary medicine clinics. The skin tests were part of the initial clinical evaluation of those patients presenting with history of symptoms and physical examination findings suggestive of a respiratory allergy, i.e., rhinitis and/or bronchial asthma. The following symptoms were taken into consideration to include the patients in the study: paroxysms of sneezing, nasal stuffiness and itching, nasal airflow obstruction in association or not with intermittent reversible cough, wheezing, or dyspnea. Other pertinent clinical in-

formation as personal and familial history of atopy (IgE-related diseases), as well as the nature and seasonal variations of symptoms, were also recorded. In this group, there were 355 male and 304 female subjects with a mean age of 15.5 years and an age range of 2 to 72 years.

A group of 108 selected healthy individuals served as control subjects. They all had no personal or family history of atopy. Special attention was directed toward selecting healthy volunteers in order to resemble demographically our patient group, i.e., similar racial and cultural background, socioeconomic status, and place of origin. There were 46 male and 62 female subjects in this group, with a mean age of 24.8 years, and the age range was between 2 and 70 years.

Allergenic extracts

Sixteen common aeroallergenic extracts were selected for scratch skin testing according to the climatologic conditions of Lima, which were purchased from Center Laboratories, Inc., Port Washington, N.Y. All extracts used came from the same lot and were kept at 5° C to assure stability during the study.

The following glycerinated allergenic extracts (50% glycerine in PBS) were used: pollen mix (grasses), 1:20; house dust concentrated; cat epithelium, 1:20; dog epithelium, 1:20; *Aspergillus* mix, 1:10; *Penicillium* mix, 1:10; *Alternaria*, 1:10; *Fusarium*, 1:10; *Mucor*, 1:10; *Pullularia*, 1:10; *Curvularia*, 1:10; *Rhizopus*, 1:10; *Hormodendrum*, 1:10; *Helminthosporium*, 1:10; *Cladosporium*,

1:10; and *Candida albicans*, 1:10. Histamine phosphate, 2.75 mg/ml (1 mg/ml histamine base, 1:1,000), and 50% glycerine in PBS were routinely used as positive and negative controls, respectively.

Immediate skin testing

All skin tests were performed, and the results were read by the same experienced person throughout the study. All subjects were instructed to stop their medications for at least 24 hours before the skin testing. The scratch technique was used on the alcohol-cleansed volar surface of both forearms. In brief, the skin was manually scratched with skin scarifiers (Center Laboratories, Inc.) and plastic templates in order to ensure the reproducibility of the length of the scratch to 3 mm. A drop of the allergen extract was immediately applied to the scratched skin surface. The results were routinely read after 20 minutes, and two perpendicular diameters of the wheal reaction were measured and recorded for prevalence studies. Twofold serial dilutions of histamine in 50% glycerine-PBS produced a decrease in the wheal size, and with a dilution of 1/16, the wheal reaction measured 2 by 2 mm. This size represented the smallest and easiest to measure wheal reaction by this technique. Repeated histamine skin tests on several occasions during the study were performed in one healthy individual. The wheal reactions obtained demonstrated no significant interassay or intra-assay (<10%) size variation. All wheal reactions >2 by 2 mm were considered as positive, and for analysis of the size of the reaction, the diameters measured were multiplied. In some analyses, the allergen-induced skin reactions were recorded as positive only when the area of this reaction was equal or larger compared to the 50% of the area obtained with histamine in that individual.

Statistical analysis

Standard computer programs were used to process the data, and the statistical methods used to analyze the results include Student's *t* test, Chi-square test, and one- and two-way analysis of variance. A *p* value of <0.05 was considered significant.

RESULTS

Prevalence of skin test reactivity to common aeroallergens

Ninety-four percent of the patients had at least one allergen skin reaction (wheal ≥ 2 by 2 mm) of the 16 aeroallergens tested, compared to 87% in the control group. In this group, 4% of the patients had positive reactions with the 50% glycerine-PBS negative control, and the largest wheal size recorded was 3 by 3 mm (9mm²). Ninety percent of the histamine-induced wheal reactions were >5 by 5 mm (25 mm²). If a skin reaction is considered as positive only when an allergen-induced wheal is equal or larger compared to the 50% of the wheal obtained with the histamine-induced control wheal in that individual, the prevalence rate decreased to 70% in the patient group and

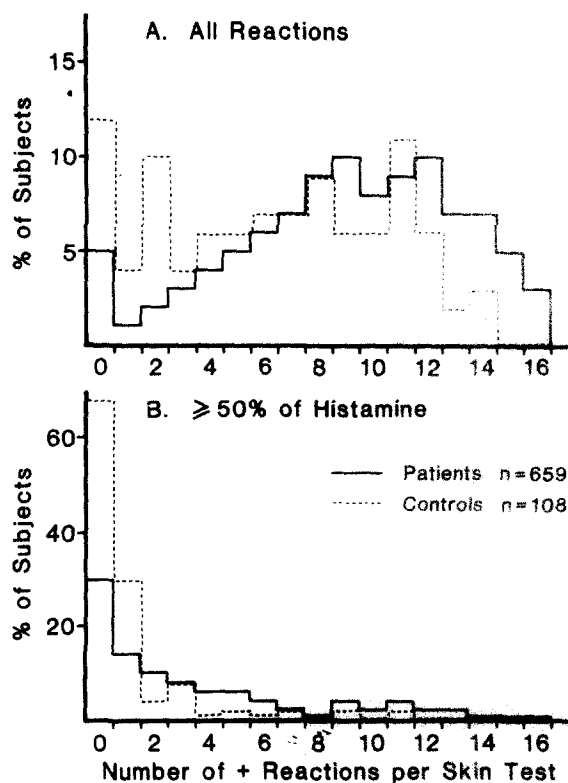


FIG. 1. Percent of subjects who exhibited one or more positive reactions to all allergens in the population studied. **A.** All wheal reactions >2 by 2 mm were considered positive. **B.** Only those reactions equal or larger compared to the 50% of the reaction obtained with histamine control in that individual were considered positive. The number of control subjects without positive reactions in their skin tests was statistically significant ($p < 0.001$) compared to the number of patients without positive reactions.

to 38% in the control group ($p < 0.05$). The prevalence of skin reactions (wheal ≥ 2 by 2 mm) for both groups, patients and control subjects, to each aeroallergen is presented in Table I. The prevalence rate varied with the allergen, but most subjects had rates between 45% to 70% in the patient group. The prevalence rates in the control group were slightly less compared to those obtained in the patient group, and in both cases, *Fusarium* and *Mucor* had the lowest rates of reactivity. These prevalence rates alone could not clearly differentiate the patients from the healthy subjects. However, when the size of the allergen-induced reactions were compared to the 50% of the area obtained with histamine in each individual, the prevalence rate in the control group is further decreased. Furthermore, in the patient group, the greatest reductions in prevalence rates were observed with pollen, *Fusarium*, *Mucor*, *Pullularia*, and *Curvularia* to the level observed in the control group.

The percentage of subjects who exhibited one or

TABLE II. Influence of age on the prevalence rates of aeroallergen skin test reactivity (wheal ≥ 2 by 2 mm) in the patient group*

Allergen	N = 347	N = 115	N = 75	N = 69	N = 30	N = 23
	0-9 yr (%)	10-19 yr (%)	20-29 yr (%)	30-39 yr (%)	40-49 yr (%)	>50 yr (%)
<i>Cladosporium</i>	68	74	60	79	83	82
<i>Candidia</i>	66	75	56	74	90	73
<i>Alternaria</i>	65	64	72	71	70	74
<i>Penicillium</i>	61	70	65	75	76	65
<i>Aspergillus</i>	59	65	66	78	66	74
<i>Helminthosporium</i>	58	69	65	72	80	74
Dog	56	66	69	68	70	69
House dust	53	69	72	72	76	56
<i>Hormodendrum</i>	55	62	70	66	70	61
<i>Rhizopus</i>	56	61	62	62	73	69
Cat	51	61	56	71	66	61
<i>Curvularia</i>	49	58	57	63	76	60
<i>Pullularia</i>	42	57	52	50	70	43
Pollen	38	54	56	58	43	43
<i>Mucor</i>	26	39	41	42	40	30
<i>Fusarium</i>	15	27	25	26	20	30
Mean	51	61	59	64	67	60
Mean (control group)	43	58	62	47	66	57

*Two-way analysis of variance: factor age (column) $p < 0.001$ and factor allergen (row) $p < 0.001$.

more positive reactions to all allergens is illustrated in Fig. 1. A bimodal curve was obtained when a wheal reaction ≥ 2 by 2 mm was recorded as positive (Fig. 1, A). The patient group had 5% of subjects with no reactions on their skin tests, and most had nine to 12 reactions to aeroallergens. The control group demonstrated more subjects with no positive reaction in their skin test (12%). Fig. 1, B illustrates the distribution of the number of positive skin test reactions considering only those allergen-induced wheal reactions equal or larger in size compared to the 50% of wheal size obtained with their histamine-induced reactions. Thirty percent of the patients had no positive reactions in their skin tests, compared to 62% of the subjects in the control group ($p < 0.001$).

Prevalence of skin test reactions according to the sex and age of the population studied

The prevalence and the size of the skin test reactions (wheal ≥ 2 by 2 mm) were not influenced by sex in either study group (not presented). To determine the influence of age on the prevalence and size of the skin test reactions, the subjects were divided into six groups according to their ages. The prevalence of positive skin test reactions (wheal ≥ 2 by 2 mm) to each aeroallergen in the patient group is presented in Table

II. Results obtained with the control group revealed similar distribution of prevalence rates. The lowest rates were observed in the group of subjects 9 years of age and younger. The peak rate was observed in subjects whose age ranged from 30 to 49 years, followed by a decrease in patients older than 50 years. The influence of the age on the prevalence rates of allergen-induced wheal reactions was statistically significant ($p < 0.001$).

The size distribution of the skin test reactions (wheal ≥ 2 by 2 mm) to aeroallergens according to their age is presented in Table III. Again, the lowest mean wheal size was observed in the group of patients younger than 9 years of age, followed by a gradual increase, reaching a maximum mean size in the group of patients whose age ranged from 30 to 39 years, decreasing thereafter in older patients. The influence of age on the size distribution of allergen-induced wheal reactions was statistically significant ($p < 0.01$). The mean size of the wheal reactions observed in the control group was considerably lower to that observed in the patient group. Furthermore, the same size distribution according to the age was also observed.

The histamine-induced wheal reactions in both the patient and control groups had the same size distri-

TABLE III. Influence of age on the size mean values of aeroallergen skin test reactions (wheal $\geq 2 \times 2$ mm) in the patient group*

Allergen	N = 347	N = 115	N = 75	N = 69	N = 30	N = 23
	0-9 yr (mm ²)	10-19 yr (mm ²)	20-29 yr (mm ²)	30-39 yr (mm ²)	40-49 yr (mm ²)	>50 yr (mm ²)
<i>Cladosporium</i>	12.9	13.8	14.7	16.1	16.0	14.4
<i>Candida</i>	13.1	14.3	16.3	13.8	15.2	14.7
<i>Alternaria</i>	13.1	14.1	14.5	13.8	12.5	14.0
<i>Penicillium</i>	12.8	13.5	15.2	14.8	13.6	13.4
<i>Asperigillus</i>	11.6	13.4	12.2	13.5	11.6	14.2
<i>Helminthosporium</i>	10.5	14.3	14.8	17.3	15.3	12.4
Dog	12.0	12.1	11.1	15.1	13.7	13.7
House dust	10.9	12.1	12.8	14.8	10.4	12.4
<i>Hormodendrum</i>	11.2	12.4	12.2	12.1	12.9	14.8
<i>Rhizopus</i>	11.0	11.8	11.4	14.4	13.2	12.3
Cat	10.4	12.6	11.7	13.4	14.7	11.6
<i>Curvularia</i>	9.9	10.0	10.8	13.4	11.0	9.8
<i>Pullularia</i>	9.2	8.9	8.6	11.0	10.9	11.3
Pollen	8.5	10.3	11.0	10.8	9.1	7.9
<i>Mucor</i>	8.6	9.1	8.6	9.0	9.9	9.0
<i>Fusarium</i>	7.9	9.0	7.8	8.1	8.2	10.5
Mean	10.6	11.9	12.1	13.2	12.4	12.2
Mean (control group)	4.0	8.1	9.5	8.0	7.1	7.7

*Two-way analysis of variance: factor age (column) $p < 0.001$ and factor allergen (row) $p < 0.001$.

bution according to the age of the subject as that presented in Table III. An inverse relationship between the mean wheal size of the histamine-induced reactions and the distribution of our patient population according to their age is illustrated in Fig. 2.

DISCUSSION

Our study population represents a group of 659 consecutive patients attending two specialized outpatient clinics. The nature of their symptoms strongly suggested an allergic respiratory disorder, and the immediate skin test was performed to measure atopy. This could explain the high incidence (94%) of patients with at least one aeroallergen-induced skin test reaction. Furthermore, the incidence of positive reactions to each aeroallergen is also high (Table I). In contrast, comparable results were obtained when a population of healthy subjects (87%) was skin tested under similar conditions and with the same aeroallergens. These results can be explained on the basis that most of the aeroallergens used are locally prevalent and that the sensitivity of the scratch skin test applied was high enough to detect specific IgE responses in most subjects studied.²

Previous studies from several regions of the world⁴⁻⁸ have demonstrated various degrees of allergen

skin test reactivity (up to 50%) in the general population. The interpretation of these results is that the allergen skin reactivity is common and that the actual prevalence may depend on several factors, such as the skin test procedure, the number of allergens, the criteria used to define a positive reaction, the potency of the allergen extracts, the age of the subjects, and geographic differences.⁹ These factors may explain the high incidence of positive reactions in our control populations.

To be able to distinguish the healthy subjects from the patient population, we considered, as positive reactions, those with an allergen-induced wheal size $\geq 50\%$ of the wheal size obtained with their respective histamine-induced reactions. Since there is a wide individual variation in the skin test reactivity, we chose to compare the allergen-induced wheal reaction to that obtained with histamine during the test. The mean wheal diameter and the allergen-histamine wheal ratio have been used in the evaluation of allergic sensitivity to common allergens for epidemiologic purposes.¹⁰ In this way, the prevalence rate of positive skin tests is significantly reduced in the control group from 87% to 38%, compared to the patient population (from 94% to 70%). Regarding the incidence of positive reactions to each aeroallergen (Table I), the pa-

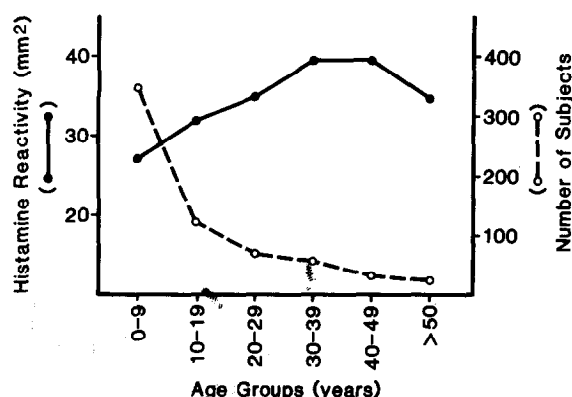


FIG. 2. Inverse relationship between the mean histamine-induced wheal size and the distribution of patients according to their age.

tients had rates from 20% to 30% in most of the aeroallergens, except in grass pollen (8%), *Fusarium* (2%), *Mucor* (5%), *Pullularia* (8%), and *Curvularia* (13%). These incidence rates are similar to those obtained in the control group for most of the aeroallergens used, and thus, we may consider them as less clinically relevant aeroallergens in Lima, Peru. The wheal size of the allergen-induced skin reaction compared to the wheal size induced by histamine appears to be important in the clinical interpretation of skin test results.

The distribution of the number of allergen-induced skin reactions demonstrated that most subjects in the patient group had nine to 12 reactions and eight to 11 reactions in the control group. There were more subjects with no reactions in their skin tests in the control group (12%) compared to 5% in the patient population. However, to be able to observe a greater difference between the healthy and the patient groups, the distribution of the number of allergen-induced reaction was analyzed considering as positive those reactions >50% of the histamine-induced wheal size. In this manner, 62% of healthy subjects had no reactions in their skin test compared to 30% in the patient group, thus representing the truly nonatopic group in our patient population studied.

The age of the subjects and not their sex appears to affect both the prevalence rates and the size of the skin reactions to allergens and histamine.⁷ The lowest prevalences and smaller wheal sizes were observed in the younger (less than 9 years) subjects from both the patient and control groups. However, the control population demonstrated smaller wheal size reactions in all age groups compared to that observed in our patients. The mechanism by which the age of the subject influences the size of the skin reaction is not clear.

An inverse correlation between higher serum IgE levels and smaller histamine-induced wheal size reactions has been reported in young individuals from a community population¹¹ and infants of atopic parents.¹² Our data suggest that IgE may not be the only mechanism involved in this observation. Decreased numbers or less active histamine receptors in our younger subjects, rather than small amounts of histamine released by their skin mast cells, appear more likely to occur because histamine also elicited smaller skin responses. The inverse relationship between more younger subjects with allergic symptoms with smaller skin reactivity suggests that other mediators and/or mechanisms not present in the skin may play an important role in the pathogenesis of the allergic reaction in target organs. However, from these data it appears clear that healthy individuals have similar mechanisms for skin sensitivity but, to a lesser degree, compared to symptomatic individuals.

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