

In vitro screening for inhalant allergy with multi SX 1 RAST® (Phadiatop®)

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A new *in vitro* screening test for inhalant allergy (Phadiatop®, Pharmacia Uppsala, Sweden) was evaluated for its effectiveness in identifying allergic patients. The test is based on the radio-allergo-sorbent-test (RAST®). Specific IgE antibodies to different inhalant allergens are detected simultaneously. Serum samples from 300 patients and controls were run with Phadiatop. 96% of 150 patients with proved allergic disease were correctly identified by Phadiatop. 92% of 150 individuals without clinically relevant allergic hypersensitivity were found correctly to be negative. Allergic disease was proved or excluded by case history, skin prick test, RAST and, in some cases, additionally by provocation challenge.

Key words: immunoglobulin E (IgE); inhalant allergens; Phadiatop®.

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Diagnosis of allergy is usually established by specialists to whom patients are referred with symptoms of suspected allergic origin by general practitioners. Selection for referral is difficult because such diseases as intrinsic asthma and vasomotor rhinitis show a similar course (16, 17). From the routine laboratory methods available, only elevated serum IgE levels relate to the allergic cause of the disorder (6, 7, 8, 9). However, total IgE normal values are highly age dependent (10, 14, 15). Serum analysis for several allergen specific antibodies, e.g. RAST, is costly even if newer methods reduce the technical requirements (1, 2).

For about 3 years, a new *in vitro* screening test for inhalant allergy (Phadiatop®, SX 1 RAST, Pharmacia Uppsala, Sweden) has been available as radio-immuno-assay (RIA, RAST), and as enzyme-immuno-assay (EIA, ELISA) respectively (11). The aim of our study was to evaluate the reliability of this method in identifying allergic patients.

In the following "atopy" or "atopic" is used for IgE-mediated hypersensitivity with or without disease, whereas "allergy" or "allergic" is used for IgE-mediated disease.

MATERIAL AND METHODS

Patients

A total of 150 patients (55 female, 95 male) in the age range 1 to 56 years (mean 13 10/12 y) with IgE-mediated inhalant allergy of the airways, and 150 control persons without suspicion of inhalant allergy (69 female, 81 male) in the age range 1 to 79 years (mean 32 7/12 y) participated in the study. They were selected at random from our out-patient department and clinic; sex and age distribution in both groups corresponds to the overall distribution of our patients.

Table 1

Results and efficiency of Phadiatop compared with total IgE determination (IgE PRIST)

		Phadiatop		Total-IgE > 100 kU/l	
		+	-	+	-
Clinical allergy diagnosis	+ 150	144	6	128	22
	- 150	12	138	30	120
Total	300	156	144	158	142
% Efficiency		94.0		82.7	
% Sensitivity		96.0		85.3	
% Specificity		92.0		80.0	

Diagnosis of allergy

After taking the case history (including specific case history for allergy) and physical examination, patients and controls underwent skin prick tests with a battery of 22 important ubiquitous allergens (house dust mite, animal epithelia, pollen, moulds and food allergens). Skin prick tests were carried out by a standardized method (12). A wheal and flare reaction greater than that produced by 1 mg/ml histamine was interpreted as positive. The allergens used were biologically and immunologically standardized extracts (Beecham-Wülfig, Bencard, Neuss, Germany). In cases where history and skin test results were discrepant, allergen-specific IgE antibodies were determined by RAST, and if allergological findings were unclear we additionally performed nasal ($n = 10$) or bronchial ($n = 4$) provocation tests as well. For the latter we used the extracts of the same manufacturer (Beecham-Wülfig, Bencard, Neuss, Germany). A nasal provocation test was regarded as positive if nasal resistance was at least double that after a control solution, and if excessive sneezing occurred. A bronchial provocation test was regarded as positive if FEV_1 fell below 80% of the control value for at least 5 min.

Allergic disease could be excluded by the provocation tests in four subjects with negative case history and positive skin test and RAST. The provocation tests confirmed allergic disease in one patient with positive case history, doubtful skin test and negative RAST and in one patient with questionable case history, positive skin test and RAST. In the other eight cases who underwent provocation tests, the diagnosis of allergic disease had already been confirmed by the presence of other allergies. By this procedure we could confirm a clinically relevant inhalant allergy in the 150 patients and exclude it in the 150 controls.

IgE tests

Serum samples of all subjects were tested for total IgE by RIA (Phadebas IgE PRIST) (3) and for specific IgE antibodies to common inhalant allergens by Phadiatop RIA. On the Phadiatop discs 10 representative allergenic components of pollen (grasses, trees, weeds) house dust mites, animal epithelia (cat, dog, horse), and mould spores are coupled (according to the manufacturer's specification). As in the RAST procedure (18), paper discs are incubated with patient serum; after washing ^{125} I-labelled anti-IgE binds to specific IgE bound to the allergens. Radioactivity is measured after washing and compared with that of a reference serum. Radioactivity above the reference level is interpreted as atopic, below that level as non-atopic. The qualitative result is given as a yes/no answer (according to the manufacturer's recommendations).

Statistical analysis of results

The ability of the test to distinguish allergic from non-allergic subjects is defined by sensitivity, specificity, and efficiency. Sensitivity is a test's ability to give a positive finding when the person tested has an allergy. Sensitivity (%) is the ratio of positive test subjects to all allergic subjects tested $\times 100$. Specificity is the ability of a test to give a negative finding

when the person tested is allergy-free. Specificity (%) is the ratio of negative test subjects to all non-allergic subjects tested $\times 100$. Efficiency (%) is the ratio of all correctly classified subjects to all subjects tested $\times 100$. Phadiatop was performed as a single test, the reproducibility of the test was not evaluated in our study.

RESULTS

In 282 of 300 subjects Phadiatop results agreed with the clinical diagnosis of allergy; efficiency was 94% compared with 82.7% for total IgE (248/300) when applying 100 kU/l as cut-off value (19) (Table 1). In the allergic group Phadiatop detected 144 of 150 patients (sensitivity 96%), whereas 138/150 controls were correctly found negative (specificity 92%). In comparison, total IgE determination detected 128 of 150 allergic patients (sensitivity 85.3%), and 120 of 150 controls (specificity 80%) (Table 1). Thus with Phadiatop, six (4%) "false negative" and 12 (8%) "false positive" results were found in relation to our diagnosis. For total IgE, the respective figures

are 22 (14.7%) "false negative" and 30 (20%) "false positive" results (Table 1).

When applying age-dependent cut-off values for total IgE (mean + 1 SD or 95th percentile), according to Kjellman et al. (10) or Ringel et al. (14) the efficiency of total IgE determinations is decreased to 81% resp. 80.7% (sensitivity 88.7% resp. 86.7%, specificity 73.3 resp. 74.7%, Fig. 1).

If there are both a positive Phadiatop and a total IgE level > 100 kU/l, then specificity increases to 96.7% and sensitivity decreases to 82.7%. If either or both are classified as positive, sensitivity rises to 98.7%, specificity falls to 75.3% (Table 2).

Phadiatop showed six "false" negative results in patients with a clear history and positive challenge test. In two of these patients we found weak skin reactions and no elevated specific IgE antibody titers (RAST class 0), one of them had elevated total IgE (274 kU/l). The other four patients had clearly positive results in SPT and RAST (RAST class 2-4) to the allergens on the Phadiatop disc, three of them had elevated total IgE titers (159, 324, and 1065 kU/l).

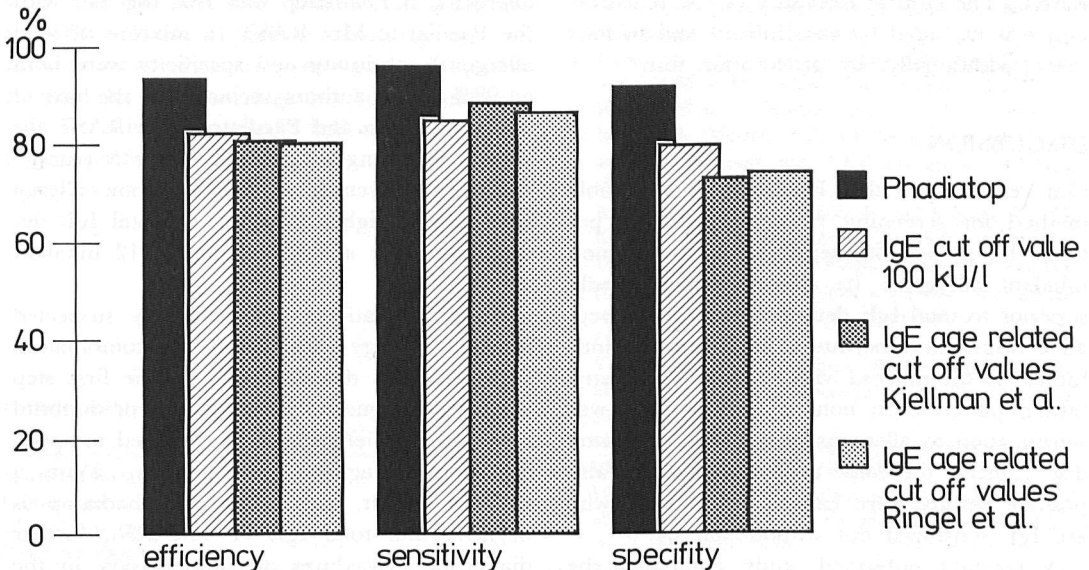


Fig. 1. Efficiency of Phadiatop test compared with total IgE using different cut-off values.

Table 2

Results and efficiency of Phadiatop in combination with total IgE determination

		Phadiatop/total IgE > 100 kU/l			
		+/+	+/-	+/+	-/-
			-/-	+/+	+/+
		+	-	+	-
Clinical allergy diagnosis	+ 150	124	26	148	2
	- 150	5	145	37	113
Total	300	129	171	185	115
% Efficiency		89.7		87.0	
% Sensitivity		82.7		98.7	
% Specificity		96.7		75.3	

Twelve patients who were "false" positive with Phadiatop had positive test reactions and specific IgE antibodies without clinical symptoms. Thus, Phadiatop correctly identifies sensitization to inhalant allergens in these 12 patients, as also demonstrated by skin test and RAST. The clinical relevance of the sensitization was excluded by case history and in four cases, additionally, by provocation test.

DISCUSSION

Our results show that Phadiatop is a suitable method for screening respiratory disease patients for atopy with sensitization to common inhalant allergens. Its efficiency is markedly superior to total IgE determination. False negative results may be due to local sensitizations limited to the mucosa with irregular IgE distribution patterns. In none of our patients was sensitization to allergens not on the Phadiatop disc causing the false negative results. False positive results were caused by patients who are IgE sensitized but without allergy.

A recently published study compared the efficiency of Phadiatop and total IgE determinations. Selected patients with positive skin

test and positive specific IgE antibodies (RAST) revealed a sensitivity of 95 % for Phadiatop and 79 % for total IgE, specificity was found to be 94 % for Phadiatop but only 56 % for total IgE (5). One insect venom- and one wheat flour-allergic patient were included in this study, showing negative results as they could not be detected by Phadiatop. The results are not directly comparable with ours as the investigations applied 120 kU/l as a cut-off for total IgE.

In another study (11) Phadiatop results were compared with allergen-specific IgE analysis by RAST. The authors found agreement of the results in 95.7 % of the investigated cases.

Merrett & Merrett (13) found Phadiatop to be an improvement over a grass/mite/cat RAST atopy screen they had used previously. Phadiatop was not only able to identify a similar number of atopics as their RAST screen, it also succeeded in identifying patients with mould allergy.

Zimmerman & Forsyth investigated 109 children, 55 of whom had allergic diseases (20). They found Phadiatop had 87.3 % sensitivity for identifying atopics and 100 % specificity compared with case history, clinical investigation and a RAST screen of 13 allergens. If Phadiatop was used together with the Paediatric Mix RAST (a mixture of food allergens) sensitivity and specificity were both at 98 %. The authors recommend the use of both Phadiatop and Paediatric Mix RAST for allergy screening in children under 5 years.

The effectiveness of Phadiatop in allergy screening is higher than that of total IgE determination or a RAST panel of 12 inhalant allergens (4).

Thus, in patients with clinically suspected inhalant allergy, Phadiatop in combination with total IgE determination, is the first step in allergy diagnosis. With positive or doubtful results the patient should be referred to specific allergy diagnosis (allergologist). With a negative result, particularly if Phadiatop is negative and total IgE < 100 kU/l, further diagnostic procedures are unnecessary in the majority of cases and symptomatic therapy may be indicated.

Conclusion

Phadiatop is a reliable *in vitro* test for inhalant allergens for the screening of patients with suspected atopic disease. Its efficiency is markedly superior to that of total IgE determination.

Phadiatop is mainly designed for the non-specialist in allergology (general practitioners, physicians, pediatricians, otolaryngologists) to supply information about IgE-mediated inhalant allergy in patients with suspected atopy. The test is even more effective in combination with total IgE determination.

A suitable screening procedure can be used by any physician (Table 3):

1. Case history and clinical inspection/examination.
2. Phadiatop and total IgE determinations.
3. RAST food allergen multidisc "Pediatric Mix" (eggwhite, cow milk, wheat flour, peanut, soy (20).

In general, these laboratory tests alone are not suitable to prove or exclude an allergic disease, and they can not replace detailed allergy diagnosis. With a positive result in one of these immunological tests the patient should be referred for further diagnosis to an allergologist for skin tests, specific IgE antibody determinations for selected allergens, and, if necessary, provocation tests.

Table 3

Recommended diagnostic procedure in suspected inhalant allergy (details see text)

Screening for atopy:
Case history
Clinical investigation
Phadiatop/total-IgE
Multiple allergen allergosorbent test infant food

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