

Comparative Evaluation of the Multiple RAST and Discrete RAST for Inhalant Allergy

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With the conventional, discrete RAST various tests are required to detect IgE of different specificities in the same serum. To overcome this problem and to reduce the costs, a multiple RAST with seven different mixtures was compared with the individual mixture constituents and with the 12 individual allergens currently in use in our department. One grass pollen mixture (gx3), two weed pollen mixtures (wx3, wx4), two tree pollen mixtures (tx3, tx6), one mould mixture (mx1) and one epithelial mixture (ex1) were used. A mixture of mites was not evaluated as there is only one important pathogenic organism in our regions (*Dermatophagoides pteronyssinus* or House dust mite). For grasses the gx3 mixture offered no advantage over the discrete RAST. The weed mixture wx3 was more sensitive than the most common discrete RAST's but at the cost of specificity. The wx4 mixture should not be used because the specificity is too low. The tree mixtures were not significantly more sensitive than the most common individual tree allergens, and were less specific. Mould mixtures should not be used because there is little cross-reactivity between the individual allergens, thus using a mixture would necessitate the subsequent determination of individual allergens, and the number of tests and the cost would be even higher. Neither should a mixture be used for epithelia because one wants to detect allergies to individual allergens. Moreover, the sensitivity of the epithelial mixture was too low. In general, we suggest the use of a limited panel of discrete RAST's instead of mixtures.

Key words: discrete; multiple; RAST.

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Several clinical studies have shown good correlation between the radio-allergosorbent test (RAST), skin tests and allergen provocation in patients suspected of having inhalant allergies (1-6, 10-11). However, a large number of discrete RAST (d-RAST) allergens exist, and it would be expensive to test all the relevant allergens to avoid false negative results. A multiple antigen RAST (m-RAST) screening procedure, in which multiple common allergens are determined in the same test, could therefore be more appropriate. In this study we evaluated mixtures of allergens of the same class in comparison with the individual constituents.

MATERIAL AND METHODS

Patients and controls

Patients. 94 patients with allergic asthma and/or rhinitis were studied, and a detailed history,

guided by a standard questionnaire was obtained from each patient.

Skin prick tests were preformed with the following commercially available allergens (HAL, Haarlem Allergen Laboratories, Haarlem, The Netherlands): grass pollen mixture, weed pollen mixture, tree pollen mixture, *Alternaria tenuis*, *Aspergillus fumigatus*, *Cladosporium herbarum*, dog dander, cat dander and *Dermatophagoides pteronyssinus*. Allergen provocation was performed with the same allergens as for skin testing, as described earlier (12). Allergen provocation was only performed when there was any discrepancy between the history and/or the results of skin test and/or the specific IgE determination (RAST).

The result of the allergen provocation was used for final classification of the patients into allergic or nonallergic diseases.

Controls. 16 individuals without symptoms of asthma, rhinitis, cough or atopic eczema (past or present), were studied.

Discrete RAST

The Phadebas RAST method (Pharmacia, Brussels, Belgium) was used. The following allergens were used in the evaluation of each patient: *Anthoxanthum odoratum* or Sweet vernal grass (g1), *Lolium perenne* or Rye grass (g5), *Phleum pratense* or Timothy grass (g6), *Poa pratensis* or Meadow grass (g8), *Secale cereale* or Cultivated rye (g12), *Holcus lanatus* or Velvet grass (g13), *Ambrosia elatior* or Common ragweed (w1), *Artemisia vulgaris* or Mugwort (w6), *Chrysanthemum leucanthemum* or Marguerite (w7), *Taraxacum vulgare* or Dandelion (w8), *Plantago lanceolata* or Plantain (English) (w9), *Che-nopodium album* or Goosefoot (w10), *Salsola kali*

or Saltwort (prickly) (w11), *Solidago virgaurea* or Golden rod (w12), *Rumex acetosella* or Sheep sorrel (w18), *Acer negundo* or Box elder (t1), *Alnus incana* or Grey alder (t2), *Betula verrucosa* or Common silver birch (t3), *Corylus avellana* or Hazel (t4), *Fagus grandifolia* or American Beech (t5), *Quercus alba* or Oak (t7), *Ulmus americana* or Elm (t8), *Juglans californica* or Walnut (t10), *Plantanus acerifolia* or Maple leaf sycamore, London plane (t11), *Salix caprea* or Willow (t12), *Copulus deltoides* or Cottonwood (t14), *Penicillium notatum* (m1), *Cladosporium herbarum* (m2), *Aspergillus fumigatus* (m3), *Alternaria tenuis* (m6), cat dander (e1), horse dander (e3), cow dander (e4) and dog dander (e5).

Multiple RAST

The same Phadebas RAST-method was used with the following mixtures: gx3: a mixture of

Table 1
Results for the grass mixture (gx3) and its most common constituent (g6)

	False + ve	False-ve	Spec (%)	Sens (%)	PV + ve (%)	PV-ve (%)
gx3	2/62 (3.2 %)	4/48 (8.3 %)	95.7	93.8	98.4	91.7
g6	0/60 (0.0 %)	4/50 (8.0 %)	100.0	93.8	100.0	92.0

False + ve: false positive, false-ve: false negative, spec: specificity, sens: sensitivity, PV + ve: predictive value of a positive result, PV-ve: predictive value of a negative result.

Table 2
Results for the weed mixtures (wx3 and wx4) and their most common individual constituents (w6 and w9).

	False + ve	False-ve	Spec (%)	Sens (%)	PV + ve (%)	PV-ve (%)
wx3	5/34 (14.7 %)	4/76 (5.3 %)	91.1	87.9	85.3	94.7
wx4	14/48 (29.2 %)	0/62 (0.0 %)	81.6	100.0	70.8	100.0
w6	0/25 (0.0 %)	8/85 (9.4 %)	100.0	75.8	100.0	90.6
w9	0/24 (0.0 %)	9/86 (10.5 %)	100.0	72.7	100.0	89.5

For abbreviations see Table 1.

Table 3
Results for the tree mixtures (tx5 and tx6) and their most common constituents (t2, t3 and t4).

	False + ve	False-ve	Spec (%)	Sens (%)	PV + ve (%)	PV-ve (%)
tx5	4/32 (12.5 %)	2/78 (2.6 %)	95.0	93.3	87.5	97.4
tx6	3/38 (7.9 %)	4/72 (5.5 %)	95.8	89.7	92.1	94.4
t2	0/23 (0.0 %)	7/87 (8.0 %)	100.0	76.7	100.0	92.0
t3	0/34 (0.0 %)	5/76 (6.6 %)	100.0	87.2	100.0	93.4
t4	0/24 (0.0 %)	6/86 (7.0 %)	100.0	80.0	100.0	87.0

For abbreviations see Table 1.

g1, g5, g6, g12, g13; wx3: a mixture of w6, w9, w10, w12; wx4: a mixture of w1, w7, w8, w11, w18; tx5: a mixture of t2, t4, t8, t12, t14; tx6: a mixture of t1, t3, t5, t7, t10; mx1: a mixture of m1, m2, m3, m6; ex1: a mixture of c1, c3, c4, c5.

For both d-RAST and m-RAST, results were considered negative for RAST classes 0 and 1 and positive for classes 2, 3, and 4.

RESULTS

For *grasses* (Table 1) the sensitivity and specificity of the mixture were equivalent to the most frequently positive individual constituent (g6). Another very common allergen (g8), not present in the mixture, gave comparable results, with a sensitivity of 89.2% with respect to the mixture constituents of gx3.

The *weed* mixture wx3 (Table 2) had a higher sensitivity than w6 and w9, the constituents most commonly responsible for allergic symptoms in our regions (87.9% versus 75.8% and 72.7% respectively), but 5/34 positive patients (14.7%) were false positive to this mixture. With the combination w6 and w9, only two patients would be missed and the sensitivity would increase to 93.9%.

The *tree* mixture tx6 (Table 3) was of the same value as t3, the constituent most commonly responsible for symptoms, whereas the sensitivity of the tx5 mixture was slightly higher. The sensitivity of three other common individual allergens, t11 (not present in the mixture), t2 and t4, was lower (60.0%, 76.7% and 80% respectively) than the tx5 mixture.

For *moulds* the sensitivity of the mixture was fairly high (86.4%), and only 1/20 patients (5.0%) was false positive.

For *epithelia* the mixture was less sensitive (78.1%), but there were no false positive results.

DISCUSSION

Establishing a diagnosis of allergic asthma and/or rhinitis is important because not only is allergy frequently present in the population, but also because of the important therapeutic consequences.

The clinical history can be very useful in seasonal allergies, but can be atypical, especially in perennial allergies.

Several studies have shown good correlation between the results of RAST and the results of skin tests and allergen provocation in patients suspected clinically to have inhalant allergies. However, to exclude false negative results entails a large number of discrete RAST's, thus making the test expensive. To overcome this problem, a multiple RAST screening procedure has been used (8, 9). In contrast to our study, these authors examined only crude mixtures containing very different allergens (*Derma-tophagoides*, grass pollen and, respectively, cat dander (8) or tree pollen (9)). They found high specificity and sensitivity with the m-RAST compared to the d-RAST and skin tests. However, as a positive result did not define the specificity of the IgE antibody, subsequent determination of the individual constituents would be needed. We therefore considered it useful to examine the results of a mixture of different allergens of the same type (e.g. different grass pollen species in one mixture) versus the individual components.

It has to be stressed that the manufacturers compose mixtures with antigens of the same botanic subclass and do not choose mixtures composed of the allergens in that class most commonly responsible for symptoms. However, our aim was to evaluate the mixtures, by comparing their sensitivity and specificity with those of their most common individual constituents. Consequently, this study was not designed to evaluate the value of RAST procedures *per se*, nor to determine the prevalence of positive RAST tests in an allergic population.

Our results with m-RAST showed a lower overall sensitivity and specificity than was found by Merret & Merret (8) (90 and 90% versus 97 and 100% respectively). This could be because we evaluated a large panel, sometimes with even more discrete allergens than were present in the mixture (e.g. the t11 individual allergen was not present in the tx5 or tx6 and these mixtures had low sensitivity). For grasses the gx3 mixture offered no advantage over the d-RAST g6. The weed mixture, wx3,

was slightly more sensitive than the d-RAST's w6 and w9, but this was at the cost of specificity. The wx4 mixture should not be used because the specificity is too low.

The trec mixtures were not significantly more sensitive than the t3 allergen, and were less specific.

In contrast to grasses, weeds and trees, there is very little cross-reactivity between the different mould genera (7). Thus one should not speak of false negative results for individual mould allergens if another mould genus in the mixture is positive. Only for the mixture can one speak of false negative and false positive results. Although the mould mixture had higher sensitivity than the individual mould allergens, and high specificity, it should not be used because there is little cross-reactivity between individual mould allergens. Using the mixture would necessitate subsequently determining the individual allergens, thus further increasing the number of tests and the expense. Moreover, as the number of positive mould RAST tests is low, the predicted sensitivity and specificity will not be very accurate.

Because with epithelia one wants to detect allergies to individual allergens, a mixture should not be used on a theoretical basis. Moreover, the epithelial mixture was not more frequently positive than the c1 allergen. On the other hand, we could confirm the findings of Wüthrich et al. (13) who demonstrated that there exists considerable cross-reactivity between cat and dog dander, which makes even the interpretation of a discrete RAST very difficult.

In general, we would suggest the use of a limited panel of discrete RAST's instead of mixtures. If mixtures are to be used, they should be based on combinations of allergens which most frequently give rise to symptoms, rather than mixing plant species: e.g. by using a combination w6 and w9, only two patients would be missed instead of four with the mixture wx3.

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