DIAGNOSTIC VALUE OF RADIOALLERGOSORBENT (RAST) TEST IN ALLERGIC DISEASES

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SUMMARY

The diagnosis of allergic disease relies on complete clinical history and physical examination. In vitro tests for allergen-specific antibodies can be performed as primary confirmatory tests to strengthen the diagnosis. RAST is a noncompetitive, heteregenous, solid-phase immunoradiometric assay. It is indicated for patients who have severe cutaneous disease, dermatographism, cannot discontinue medications that interfere with skin testing, or have experienced severe anaphylaxis. Disadvantages of in vitro testing are inereased cost, delays in providing results, and laboratory reliability. Selection of a reliable laboratory is fundamental. Although conventional RAST is less sensitive than skin testing, specific IgE measurements correlate well with the results of skin testing and the clinical picture. Second generation of RAST- type assays are more quantitative, reproducible and automated than the older ones and thus more diagnostically competitive with their in vivo prneture skin test counterparts. Specific IgE leveis that are measured with different commercial assays cannot be considered interehangeable or equivalent. For food and respiratory allergy, both serum IgE antibody performed by second generation RAST assays and prick skin test results are considered equivalently acceptable. In latex allergy, the diagnostic sensitivity of latex-specific IgE antibody assays has been shown to be inadequate and thus can be used as complementary to the prick skin test results. Maximal clinical sensitivity is needed for evaluating patients with suspected venom and drug allergies because of the potential life-threatening systemic reactions.

In conelusion, IgE antibody seroIogy test should be reserved as a confirmatory test when intradermal skin test cannot be performed or contradictory with the history.

Key words: Allergy, RAST, specific IgE

INTRODUCTION

Allergy is a worldwide growing problem. Diagnosis and management of allergic disease relies on compiete clinical history and physical examination. Detection of allergen specific antibodies by skin test and/or serologically can be performed as primary confirmatory tests to strengthen the diagnosis. In this way potential allergens that might be contributing to the allergic disease process can be identified, the patient and his family can avoid exposures, and the elinician can manage the disease appropriately. In vivo provocation tests are usually considered secondary level confirmatory tests that are preferred when clinical history and allergen-specific IgE antibody tests do not correlate [2,8].

Among in vitro tests, total serum IgE was initially used as diagnostic marker for allergic disease. But as there is a wide overlap in total IgE leveis between atopic and nonatopic populations, allergen specific IgE is preferred and the most important laboratory analyte in the diagnostic work-up for allergic disease [6].

Although conventional RAST assay is less sensitive than skin testing, specific IgE measurements correlate well with the results of skin testing and the clinical picture. Skin testing places patients at some small risk for anaphylaxis, and also causes mild itehing during and some time after the test, especiaily in atopic people, whereas there is no such patient risk and itehing associated in vitro

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testing. RAST is indicated for patients who have severe cutaneous disease, dermatographism, cannot discontinue medications that interfere with skin testing, or have experienced severe anaphylaxis. Disadvantages of in vitro testing are increased cost, delays in providing results, cross-reactions and laboratory reliability. Selection of a reliable laboratory is fundamental. It is also important to obtain simultaneous total IgE levels because of false positive RAST results may occur in sera with extremely high levels of IgE due to non-specific binding [15].

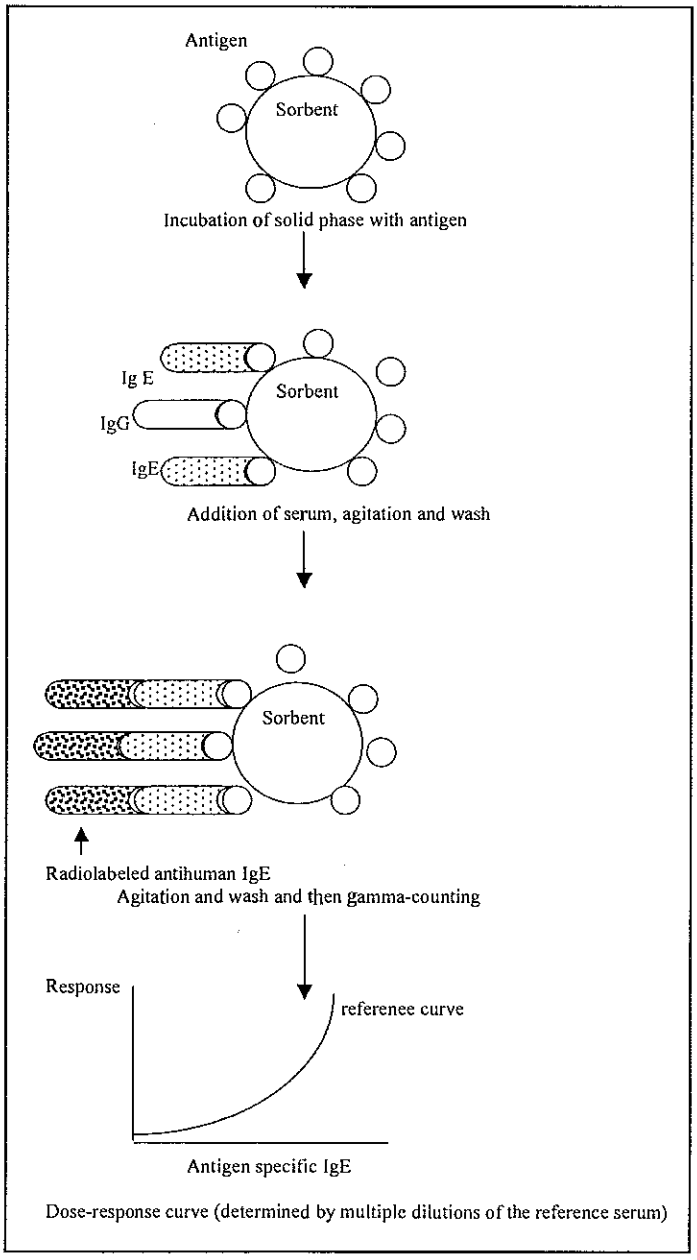
METHODOLOGY OF RAST

Phadebast RAST (Pharmacia, Uppsala, Sweden) was the first assay reported for the detection of allergen specific IgE antibodies. This assay was a noncompetitive, hcteregenous, solid-phase immunoradiometric (radiolabeled antibody) assay. The allergosorbent was prepared by covalently coupling allergen onto cellulose (paper) disc. Addition of human serum to the allergosorbent permitted allergen specific antibodies of ali isotypes that is IgE and if present, IgE to bind. Following a first buffer wash to remove unbound serum proteins, radioiodinated antihuman IgE was added to detect bound IgE. After a second buffer wash to remove unbound radiolabeled antihuman IgE, bound reactivity was measured in a gamma counter as a reflection of the amount of specific IgE in the initial serum specimen. IgE antibody results were reported in arbitrary Phadebas RAST units per mL of IgE antibirch based on a calibration curve produced by the binding of dilutions of an IgE antibirch reference serum, each to their own birch pollen allergosorbent (Figure 1) [2,5].

Second Generation of Rast-Type Assays

The basic RAST chemistry has remained essentially unchanged over more than 35 years. Development of a large number and quality of allergen extracts and new matrix materials such as cellulose sponged for aller-

Figure 1. Schcmatic diagram of the radioallergosorbent test



gosorbents, various polyclonal and monoclonal anti-IgE detection antibody combinations, nonisotopic labels, improved automation and calibration systems have resulted in second generation of RAST- type assays that display superior analytical sensitivity and specificity. Some of these assays have not obtained Food and Drug Administration (FDA) clearance and are thus markcted outside the USA (Table 1) [4]. They are more quantitative, reproducible and automated than the earlier assays and more diagnostically competitive with their in vivo puncture skin test counterparts. They have the ability to generate quantitative IgE antibody measurements in mass per volume (microgram per liter) units. But specific IgE leveis that are measured with different commercial assays can still not be considered interchangeable or equivalent since allergen extracts vary in their composition and allergic potency between manufacturers due to a number of factors. These include the season In which the raw material collected, the degree of difficulty in identifying a pure source of material, the presence of morphologically similar raw materials that may cross-contaminate and the differences in extraction process during allergen-reagent production by assay manufacturers. There are also issues of stability during storage, heterogeneity of human IgE antibody containing sera used for quality control, and different criteria for acceptance of the finished allergen-containing reagent by different manufacturers. Thus allergosorbents from different manufacturers detect different populations of IgE antibodies for any given allergen specificity [2,4,5].

Table 1. Some commercially available allergen-specific IgE antibody immunoassays

Manufacturer	Assay name	Interpolated units
Abbott*	MATRIX	SIgE/ml
Chiron Diagnostic Corporation*	Magic Lite	IU/ml
MAST Immunosystems	MAST (CLA)	mV
Pharmacia UpJohn	Phadebas RAST	PRU/ml
Pharmacia UpJohn	CAP system	kIUA/l
Sanofi Diagnostics/ Pasteur	RAST/EAST	AU/ml
Diagnostic Products Corporation	Ala STAT	IU/ml

**These immunoassays are not actively used in North America.*

INTERPRETATION OF RAST IN CLINICAL USAGE

Rast In Food Allergy

For food allergy, serum IgE antibody performed by second generation RAST assays and skin prick test (SPT) results arc considered equivalently acceptable. But caution should be used when interpreting both food skin tests and RAST, because only a small fraction of patients who have a positive skin test or specific IgE result will react when a positive doublc-blind, placebo-controlled food challenge (DBPCFC) is performed. The avoldance of foods in diet never should be based on skin or in vitro test results unless accompanying relevant clinical history and symptoms. It is also important to remember that food allergy can oecur through non-IgE mediated reactions, too [6,8].

Measurement of specific IgE quantitatively enhances the diagnostic value of RAST assay, especially in food allergy. In 1997, Sampson et al [12] retrospectively investigated sera from 196 children and adolescents with a mean age of 5.2 years with atopic dermatitis. They determined some cut-off levels for specific IgE antibodies to cow's milk (32 kUa/L), chicken egg (6 kUa/L), peanut (15 kUa/L) and fish (20 kUa/L) with a predieted clinical reactivity more than 95% certainty defined by DBPCFC or a convincing history of

food-induced anaphylaxis. Sampson later, extended his observations with a prospective study of 100 children and adolescents with a mean age of 3.8 years that been referred for evaluation of food allergy and again confirmed precision of the cut-off values for specific IgE levels defined in his previous study [13]. The important conclusion of these studies is that quantitative measurements of specific IgE can eliminate the need for DBPCFC in a significant number of children suspected of having IgE mediated food allergy [5]. Boyano et al [1] prospectively investigated specific IgE leveis for egg-white, egg-yolk, egg-ovalbumin and egg-ovomucoid in 81 children aged less than 2 years old with a history of immediate hypersensitivity reaction after egg ingestion. The diagnosis of egg allergy was defined by an open controlled oral challenge test in the absence of a history of severe food-induced anaphylaxis. They determined that the presence of specific IgE antibodies to egg white of ≥0.35 kUa/L was to be sufficient to establish the diagnosis in 94% of the children. Monti et al [9] prospectively compared the outcome of a first oral egg challenge and the results of albumin and yolk SPTs and RASTs in 107 children with atopic dermatitis, with a median age of 5 months who had never ingested egg. They reported that specific IgE leveis >99 kUa/L for albumin and >17.5 kUa/L for yolk verified a positive challenge with 100% specificity. But they also observed positive challenge tests with specific IgE leveis <0.35kUa/L. The significance of this report rests again in the potential of eliminating challenge tests in food allergy in certain group of children who had specific IgE levels equal or above the specific established cutt-offs. But it must always kept in mind that these cut-offs can be used to confirm a symptomatic allergy and not to rule out an allergy and clinical reactions to the challenge can occur below the established cut-offs, so that challenges always should be preferably performed in an appropriately equipped environment. The three investigators defined different cut-offs for the same allergen, egg in their reports. This discordance could be due to the differences in age and selection criteria of the populations analyzed.

Rast in Inhalant Allergies

There are several studies evaluating RAST for respiratory allergens. Shafer et al [14] compared the diagnostic values of SPT and RAST to aeroallergens with respect to incidence of hay fever cases in school children at different cut-off points. The incidence of hay fever cases predicted rather poorly though somewhat bettcr by SPT than by RAST with a cut-off <0.35 kUa/L. When cut-off point for RAST was considered 1.5 kUa/L, almost identical predictive values for both tests were obtained and both tests performed better results in negative than positive prediction. This study shows that the predictive capability of RAST depends on the chosen cut-off point. Pasterollo et al [11] reported the optimal cut-off values for specific IgE that could discriminate between patients with symptomatic and those with asymptomatic allergy. They showed that patients with symptomatic allergy had a higher mean specific IgE value than those with asymptomatic allergy and healthy control subjects. Optimal cut-off value in differentiating patients with symptomatic from those with asymptomatic allergy was 11.7kUa/L. The optimal cut-off values for seasonal allergens and perennial allergens were 10.7 kUa/L and 8.4 kUa/L, respectively. This study shows that the predicted cut-off values for specific serum IgE antibody levels may be useful in clinical practice to distinguish symptomatic patients from sensitized asymptomatic patients. In an other study, Wood et al [17] evaluated the predictive value of SPT, intradermal test and RAST in the

diagnosis of cat allergy and found that RAST was highly specific but somewhat less sensitive than SPT. They supported the clinical usage of SPT and RAST together, in the diagnosis of cat allergy.

Multiallergen IgE Screening Assays

The multiallergen IgE antibody screen is a qualitative RAST-type test that evaluates a patient's serum for the presence of IgE antibodies specific for a mixture of approximately 15 indoor and outdoor aeroallergens that are account for a largc majority of allergic respiratory disease. A pediatric form of a similar qualitative multiallergen screening test evaluates common food specific IgE (egg, milk, peanut, wheat, soybean) in addition to IgE for weed, grass and tree pollen, molds, pet epidermal and dust mite aeroallergens. A negative multiallergen screen reduces the probability that IgE antibodies are involved in the patient's clinical problems to less than 5%. These screening assays are most useful in confirming the absence of significant atopic disease in individuals who are suspected of having an intrinsic or non-IgE mediated respiratory, cutaneous, or gastrointestinal disease process. Such a test can minimize the need for multiple in vivo and in vitro ailergen-specific IgE measurements in patients with a low clinical probability of atopic disease. But the use of this screening test in unselected populations is likely to generate many false-positive results since IgE antibody responses are much frequent than symptomatic disease [6]. There are several studies supporting the predictive value of multiallergen test. Williams et al [16] evaluated 145 children and adolescent patients for allergic disease. They determined allergic ones regarding to history, physical examination skin prick tests and sp IgE measurements to common seven allergens (mite, oak, ragweed, grass, dog, cat, alternaria). They compared skin prick test and spIgE with UniCAP Phadiatop test results. Ali patients with resolved diagnosis 143 of 145, (103 with allergy and 40 not) were identified correctly by the UniCAP Phadiatop test. The UniCAP Phadiatop test was shown to be highly sensitive and specific in differentiating individuals who were sensitized to common inhalants from those who were not. Paganelli [10] reported that Phadiatop in detecting atopic sensitization to common inhalant antigens agreed with clinical diagnosis in 764 of 836 (91.4%) cases. The clinical sensitivity and specificity for Phadiatop was 93% and 89%, respectively.

Rast in Latex Allergy

In latex allergy, the diagnostic sensitivity of FDA-cleared latex-specific IgE antibody assays has been shown to be inadequate. Hamilton et al [3] compared the diagnostic performance of 3 established FDA-cleared in vitro assays (DPC Microplate Alastat, Pharmacia CAP system FEIA, and Hycor HYTECH) for the detection of natural latex-specific IgE. When the SPT was used as the reference method, The PharmaciaCAP system and DPC Alastat microplate displayed 76% and 73% diagnostic sensitivity, respectively, whereas their diagnostic specificity in comparison with the SPT was 97%. These data indicated that both assays misclassified approximately 25% of latex-sensitized cases as falsely IgE antibody-negative. The HyTECH, in contrast, displayed a specificity of 73%, which indicates that it produces 27% false-positive results when compared to skin test. There are similar studies done with similar results. As a result, latex-sp IgE test can be a diagnostic confirmatory test. It must be performed in a reference laboratory, and the assay methods used to measure antibody should either be FDA-cleared or well documented.

Rast in Venom and Drug Allergies

Maximal clinical sensitivity is needed for evaluating patients with suspected venom and drug allergies because of the potential life-threatening systemic reactions. In these cases, the IgE antibody serology results are viewed as complementary to the intradermal skin test results [2]. Generally, in vitro testing is less sensitive, being positive in only 80% of individuals who have positive venom skin test responses [7].

CONCLUSION

In conclusion, RAST enables objective measurement of ailergen-specific IgE antibody leveis and shows the extent of allergic sensitization, but not the allergic disease. It should be reserved as a confirmatory test when intradermal skin test cannot be performed or contradictory with the history.

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