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Evaluation of Ara h2 IgE thresholds in the diagnosis of peanut allergy in a clinical population

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TO THE EDITOR:

Component resolved specific IgE testing for peanut allergy has recently received much attention as a means of more accurately identifying patients with peanut allergy than the routine use of peanut extract-specific IgE serology or skin testing. Among the peanut component proteins, IgE antibodies to Ara h2, and to a much lesser extent Ara h1, Ara h3, Ara h6, and Ara h9, have been identified as the major driver of clinically relevant allergy.^{1–9} Because sensitization to Ara h2 is found in 40% to 90% of patients with clinical peanut allergy,^{1–8,10} some investigators have proposed using specific IgE to Ara h2 serology as a way to reduce the need for oral food challenges.¹¹ Various IgE anti-Ara h2 cut points have been proposed to predict clinically relevant peanut allergy. Codreanu et al,² for instance, suggested a threshold of 0.23 kU_A/L Ara h2 for identifying peanut allergic subjects and further suggest that “The DBPCFC [double-blind placebo-controlled food challenge] should no longer be considered a mandatory diagnostic procedure, but will remain a useful test for defining the eliciting dose for patients who are candidates for oral immunotherapy.”^{2(p225)} Nicolaou and Custovic¹⁰ similarly proposed 0.35 kU_A/L as a threshold. In their study, this threshold exhibited a 100% diagnostic sensitivity. However, it still remains unclear whether these thresholds should be used to suggest home introduction of peanuts to patients (if they fall below the threshold) or whether they should be used in place of a food challenge (if they rise above the threshold). Nor is it clear how accurate either of these applications will be in a general clinical population. Here, we apply a variety of cutoff values for IgE anti-Ara h2 to a

pediatric allergy clinic to determine whether using component-resolved diagnostics for peanut would result in decreased need for food challenge.

Subjects who had diagnostic peanut oral challenges at the Pediatric Allergy Clinic between 2003 and 2010, had stored serum available within 2 years of their challenge, and were sensitized to peanut were identified retrospectively. Samples were obtained from banked excess serum originally drawn for clinical purposes. Human subjects' approval was given by the Johns Hopkins Institutional Review Board. Because this was a retrospective study, informed consent was waived by the Institutional Review Board.

Peanut food challenges were open. The cumulative maximum food challenge dose was 5 g of peanut protein for children younger than 5 years and 8 g otherwise (equivalent to ~17 and 27 peanuts, respectively). The form of peanuts depended on patient preference and other allergies, but the form was most commonly peanut butter or peanut candies. Food challenges were done for diagnostic purposes, and they were stopped when clear clinical symptoms developed, including symptoms in the following systems: skin (hives, full body flushing), upper respiratory (sneezing, significant rhinorrhea), lower respiratory (wheezing, cough), gastrointestinal (significant persistent abdominal pain, vomiting), and cardiovascular (hypotension, loss of consciousness). Although the threshold varied according to clinical circumstances, most often oral challenges were done when the peanut-specific IgE was <2 kU_A/L in patients with a clear history of reaction and <5 kU_A/L in patients without a clear history.

IgE antibody specific for peanut extract and for the individual peanut components (Ara h1, h2, h3, h8, and h9) were quantified in kU_A/L by the ImmunoCAP-250 (Thermo Fisher Scientific, Waltham, Mass). Each serum sample was additionally analyzed in the ImmunoSorbent Allergen bioChip assay (ISAC103; Thermo Fisher Scientific).³ Semiquantitative IgE antibody results were reported in ISAC Standardized Units (ISUs), with <0.3 ISU being considered negative. The available ISAC peanut allergen specificities examined for this report included Ara h1, Ara h2, Ara h3, and Ara h8.

Diagnostic sensitivity and specificity and positive and negative predictive values were computed with exact binomial confidence intervals for the defined thresholds by comparing individual component-specific IgE levels as measured by ImmunoCAP and ISAC with peanut food challenge outcome. All calculations were performed with STATA/SE 11.0 (Stata Corp, College Station, Tex).

Sixty patients were included in this analysis, including 26 with a history of acute peanut reaction and 34 with a positive test but no previous history of reaction. With peanut food challenge, 26 reacted and 35 did not. One patient initially failed a peanut challenge at age 4.8 years and then later passed at 7.2 years and had sera available from both times (peanut specific IgE of 1.5 kU_A/L at first challenge and 0.8 kU_A/L at second challenge). The mean age at the time of challenge was 7 years (range, 3–19 years) and occurred at a mean of 5 months after the serum was obtained. Generally, the peanut extract-specific IgE for both groups was relatively low (Table I).

Ara h2 was the most commonly recognized peanut component by IgE antibody in sera from patients who failed peanut food challenges, with a diagnostic sensitivity among this population (when sensitization was defined as ≥ 60.1 kU_A/L) of 96% and diagnostic specificity of 54% (Table II). In contrast, IgE anti-Ara h1, -Ara h3, -Ara h8, and -Ara h9 antibodies were insensitive predictors of failed peanut oral food challenge (sensitivity: Ara h1, 23%; Ara h3, 24%; Ara h8, 12%; and Ara h9, 12%) but showed moderate specificity (Ara h1, 80%; Ara h3, 74%; and Ara h9, 82%). The exception was Ara h8, which at a cutoff of 0.1 kU_A/L displayed a poor specificity (56%). In fact, if subjects had detectable anti-Ara h8 IgE, they were more likely to pass a peanut food challenge than if they did not ($P = .01$); however, 3 subjects with positive anti-Ara h8 IgE did fail the challenge (11% of those positive). The sensitivity, specificity, positive predictive value, negative predictive value, and percent misclassified, defined as the total percentage labeled as either allergic or nonallergic in error, for various cutoffs of Ara h2-specific IgE are listed in Table II, including diagnostic sensitivity to Ara h2 as defined by ISAC. A high rate of misclassification was observed for all IgE anti-Ara h2 cutoff levels tested: 26% were misclassified at 0.23 kU_A/L, 21% at 0.35 kU_A/L, 36% at 2 kU_A/L, and 21% at 0.3 ISU with ISAC (Table II). Among those with anti-Ara h2 IgE below 0.35 kU_A/L who reacted to the food challenge, none had lower respiratory or cardiovascular symptoms or required epinephrine. However, 2 patients (10%) with anti-Ara h2 IgE below 2 kU_A/L had lower respiratory symptoms and 5 (25%) required epinephrine.

Previous studies have shown that IgE antibody to peanut components in general, and Ara h2 in particular, correlate well with symptomatic peanut allergy.^{1,2,7,8,10} Less well explored is how peanut component diagnostics perform in the patient population that an allergist will commonly encounter. Here, we used IgE anti-Ara h2 cutoffs that others had previously suggested were accurate in predicting food allergy and found a high rate of misclassification for all cutoffs evaluated. Although the predictive ability is clearly improved over a crude peanut specific IgE, the use of IgE anti-Ara h2 values above these cutoffs as a replacement for food challenge will still result in many patients being classified as peanut allergic who are not truly allergic. Conversely, if patients with levels below these cutoffs are considered nonallergic and advised to consume peanut at home, some patients will react. A limitation to these data is that in some patients a fairly significant interval passed between serum acquisition and food challenge, but this is likely representative of clinical practice. Ultimately, our data underscore the notion that even with more tailored tests of antigen-specific IgE, we are still left with the problem that sensitization does not equal clinical disease. Whether any type of specific IgE testing will match the highly predictive qualities that we have come to expect of diagnostic testing in modern medicine remains an open question. For the present time, we are not yet at a stage where laboratory-based tests can substitute for clinical history and oral food challenge.

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Clinical Implication

- Although the use of component resolved IgE testing for peanut allergy may have improved diagnostic characteristics than the use of peanut extract alone, it cannot yet replace clinical history and oral food challenge.

TABLE I

Subject characteristics

Group	Passed challenge (n = 35)*	Failed challenge (n = 26)*
Age at sample (y), mean (range)	6.0 (2.2–18.2)	5.8 (3.5–17.1)
Male sex, no. (%)	26 (74)	18 (69)
History of reaction, no. (%)	10 (29)	17 (65)
Peanut IgE, mean (range)	1.2 (0.4–21.1)	1.2 (0.4–6.3)

* One subject failed and then passed a food challenge at a later date and had a blood sample at both times.

TABLE II

Diagnostic characteristics of various thresholds for Ara h2-specific IgE

Threshold	Sensitivity, % (95% CI)	Specificity, % (95% CI)	Positive predictive value, % (95% CI)	Negative predictive value, % (95% CI)	Percent misclassified
ImmunoCap Ara h2					
0.23 kU _A /L	92.3 (74.9–99.1)	60 (42.1–76.1)	63.2 (46–78.2)	91.3 (72–98.9)	26.2
0.35 kU _A /L	88.5 (69.8–97.6)	71.4 (53.7–85.4)	69.7 (51.3–84.4)	89.3 (71.8–97.7)	21.3
2 kU _A /L	23.1 (9.0–43.6)	94.3 (80.8–99.3)	75 (34.9–96.8)	62.3 (47.9–75.2)	36.1
ISAC Ara h2					
0.3 ISU	80.8 (60.6–93.4)	77.1 (59.9–89.6)	72.4 (52.8–87.3)	84.4 (67.2–94.7)	21.3