



Letter

Apolipoprotein B: a possible new biomarker for anaphylaxis

There are few published studies describing biochemical markers that correlate with the severity of anaphylactic reactions during oral food challenges or accidental reactions to foods. Identifying patients with increased risk of severe reactions is a priority within the field of anaphylaxis. Previous studies have examined laboratory markers such as specific immunoglobulin E (IgE; to allergenic foods or components thereof), parameters of the basophil activation test, and platelet-activating factor (PAF).

It has been shown that PAF takes part in various inflammatory events and plays a key role in the life-threatening events that cause anaphylaxis.^{1,2} PAF has been shown to be more closely correlated to the severity of anaphylactic reactions than histamine or tryptase in humans.^{3,4} PAF mediates reactions causing increased vascular permeability, hypotension, and bronchoconstriction.^{5,6}

Anaphylactic responses are influenced by PAF acetylhydrolase (PAF-AH), which catalyzes the degradation of PAF to its inactive products. PAF-AH in plasma is probably produced by cells from hematopoietic lineage cells.⁷ Relative deficiency of PAF-AH has been shown to lead to an increased risk of life-threatening anaphylaxis.³ In other words, high levels of PAF-AH before an anaphylactic reaction will limit the production of PAF during the ongoing reaction and thus limit the severity of that reaction. However, the exact mechanism of this pathway is unknown.

In human plasma, PAF-AH circulates in complexes with low- and high-density lipoproteins, and the main protein component of low-density lipoprotein particles is apolipoprotein B-100 (ApoB). A recent study of a pediatric population allergic to peanuts showed a strong correlation between PAF-AH activity and the concentration of the more stable ApoB.⁸ Because of inactivation of PAF by PAF-AH, these data suggest a relation between the severity of allergic reactions and the plasma concentration of ApoB. Thus, the objective of this study was to examine serum ApoB as a possible biomarker for the occurrence and severity of anaphylactic reactions to foods.

Data and serum samples were collected from the food challenge unit data and serum bank at the University Medical Center Groningen (UMCG; 2002–2014). Serum was collected before the double-blinded, placebo-controlled food challenge (DBPCFC).

The study population consisted of children referred to the UMCG for a clinical suspicion of food allergy. No patients were excluded for a history of severe allergic reactions. Patient data were collected by history and laboratory analysis was performed as part of routine clinical care. Assenting children underwent a DBPCFC with the suspected food using previously described materials, methods, and protocols.⁹ The medical ethics committee of the UMCG deemed that formal medical ethical approval was not required for this study.

Cases without a matching available serum sample or having missing variables necessary for the analysis were excluded.

However, children with an unknown history of asthma, atopic dermatitis, or rhinoconjunctivitis were included in the analysis, and the inclusion of this patient group did not influence the results.

A symptom-based scoring system was used to determine the severity of the most severe accidental reaction as obtained from the history and the severity of the DBPCFC reaction as observed during the challenge test, as previously described.¹⁰ Briefly, the allergic symptoms received a score corresponding to the involved organ system. Skin symptoms were given a score of 1, upper airway symptoms (eyes, nose, and throat) were given a score of 3, lower airway symptoms (lungs) were given a score of 3, gastrointestinal symptoms were given a score of 2, and cardiovascular and/or neurological symptoms were given a score of 3. The scores of the involved organ systems were added together and the total severity score was calculated (range 0–12). Scores of 0 to 2 were considered mild allergic reactions, scores of 3 to 6 were considered moderate allergic reactions, and scores of 7 to 12 were considered severe allergic reactions.

The concentration of ApoB in the serum samples was determined by nephelometry. Linear regression analysis was used to examine the association between the concentration of ApoB and the severity of the DBPCFC reaction and the severity of the previous most severe accidental allergic reaction by history. Logistic regression analysis was used to study the relation between the concentration of ApoB and the food challenge outcome. A factor was considered a confounder when it changed the β coefficient of the effect of ApoB on the severity of anaphylactic reactions by at least 10%. For all tests, a 2-tailed significance level of a P value less than .05 was used.

Eight hundred thirty-seven cases were included in the final analysis. The study population consisted of children with a median age of 5.8 years (interquartile range 3.6–10.4), of which 62.7% were boys. A history of asthma was reported in 52.0% of children. Most of the study population had atopic dermatitis (88.4%) and 41.5% had rhinoconjunctivitis. Specific IgE was measured in all subjects and was positive in 82.1% of the children included (Table 1 presents additional patient characteristics).

The severity of the accidental reaction by history was inversely associated with the concentration of ApoB ($\beta = -0.09$, 95% confidence interval [CI] -2.53 to -0.33 , $P = .011$). This relation was not confounded by age, sex, type of food, level of specific IgE, eliciting dose, history of asthma, atopic dermatitis, and rhinoconjunctivitis. This significant association also was seen in the subgroup of patients with a positive history but a negative food challenge outcome ($\beta = -0.15$, 95% CI -3.75 to -0.66 , $P = .005$) but, remarkably, not for children with a positive history and a positive food challenge outcome ($\beta = -0.05$, 95% CI -2.24 to 0.72 , $P = .313$).

The concentration of ApoB was not associated with the food challenge outcome (adjusted odds ratio 0.90, 95% CI 0.41 to 1.98, $P = .790$) or the severity of the DBPCFC reaction ($\beta = -0.01$,

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Table 1
Characteristics of the Study Population

Patient characteristic	Frequency (Percent)	
Subjects	837 (100%)	
Girls	312 (37.3%)	
Boys	525 (62.7%)	
Food		
Peanut	244 (29.2%)	
Cow's milk	233 (27.8%)	
Egg	111 (13.3%)	
Hazelnut	84 (10.0%)	
Cashew nut	82 (9.8%)	
Soy milk	30 (3.6%)	
Walnut	19 (2.3%)	
Almond	15 (1.8%)	
Wheat	14 (1.7%)	
Pistachio	2 (0.2%)	
Beef	1 (0.1%)	
Lupine seed	1 (0.1%)	
Sesame seed	1 (0.1%)	
History of atopic dermatitis ^a		
Yes	740 (88.4 %)	
No	93 (11.1%)	
History of asthma ^a		
Yes	435 (52.0 %)	
No	393 (47.0 %)	
History of rhinoconjunctivitis ^a		
Yes	347 (41.5 %)	
No	468 (55.9 %)	
Food challenge outcome		
Positive	466 (55.7 %)	
Negative	371 (44.3 %)	
	N (range)	IQR (median)
Apolipoprotein B (g/l)	837 (0.24–1.31)	0.53–0.77 (0.65)
Age (months)	837 (6.00–215.00)	43.00–124.50 (70.00)
Specific IgE (kU/l)	837 (0.10–101.00)	0.56–16.80 (3.31)
Severity of accidental reaction	837 (0.00–12.00)	0.00–5.00 (3.00)
Severity of DBPCFC reaction ^b	466 (1.00–9.00)	2.00–5.00 (3.00)
Eliciting dose (mg protein) ^b	466 (0.58–1925.00)	14.00–350.00 (70.00)

Abbreviations: DBPCFC, double-blinded, placebo-controlled food challenge; IgE, immunoglobulin E; IQR, interquartile range.

^aMissing data not shown.

^bSeverity of DBPCFC reaction and eliciting dose only available for positive food challenge outcomes.

95% CI –1.17 to 1.02, $P = .894$). All association models used for the analysis met the assumptions of the test. To examine whether the outliers (the 10 cases outside the CI) had an effect on the outcome, the data were reanalyzed with these cases excluded. However, this analysis showed no difference in the results, and the inverse association between ApoB and the severity of the accidental reaction remained unchanged (data not shown).

This study has, for the first time, shown an association between ApoB and the severity of the accidental reaction in children with suspected food allergy. The results of this study suggest that ApoB might be a biomarker for the severity of anaphylactic reactions, probably by virtue of its association with the less stable PAF-AH. The finding that this marker is associated with severity of anaphylaxis in patients with negative food challenge outcomes is surprising but could be due to the fact that the severity of reactions to foods are governed to a larger extent by other factors such as type and dose of the ingested food, making the relation with ApoB more tenuous. A second conclusion suggested by this finding is that most patients with a negative food challenge outcome indeed experienced an anaphylactic event in the past. In view of the natural history of patients with negative food challenge outcomes, this finding suggests that such anaphylactic events do not seem to have an obvious allergic cause and do not tend to recur.

The association between ApoB and the severity of the accidental reaction is in agreement with the previous findings concerning the role of PAF and PAF-AH in anaphylactic reactions. In contrast to

PAF-AH, ApoB can be accurately determined from stored frozen serum samples and is more stable, convenient, and inexpensive to measure. Although measurement of ApoB explains only a relatively small part of the anaphylactic risk, serum samples obtained closer to the time of the anaphylactic reaction and from patients with other forms of anaphylaxis might be more informative. Further studies on this topic are indicated to evaluate this biomarker in food-induced anaphylaxis and other types of anaphylaxis.

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