Tryptase levels in children presenting with anaphylaxis: Temporal trends and associated factors

Sarah De Schryver, MD,[a] Michelle Halbrich, MD,[b] Ann Clarke, MD, MSc,[c] Sebastien La Vieille, MD,[d] Harley Eisman, MD,[e] Reza Alizadehfar, MD,[a] Lawrence Joseph, PhD,[f] Judy Morris, MD, MSc,[g] and Moshe Ben-Shoshan, MD, MSc[a]

Montreal, Quebec, Winnipeg, Manitoba, Calgary, Alberta, and Ottawa, Ontario, Canada

Background: The diagnosis of anaphylaxis currently relies on suggestive clinical history after exposure to a potential triggering factor because no reliable diagnostic marker is available to confirm the diagnosis.

Objectives: We aimed to evaluate tryptase levels in children with anaphylaxis and to examine predictors of elevated tryptase level (defined as ≥11.4 μg/L during reaction and for those with a baseline level, defined as a reaction level of at least 2 ng/mL + 1.2 × [postreaction tryptase level]).

Methods: Children presenting with anaphylaxis to the Montreal Children's Hospital were recruited over a 4-year period. Symptoms, triggers, and management of anaphylaxis were documented. Levels during the reaction and approximately 9 months after the reaction were compared on the basis of paired means using the *t* distribution. Multivariate linear and logistic regressions were used to evaluate the association between tryptase levels and risk factors.

Results: Over a 4-year period, 203 children had serum tryptase levels measured. Among these, 39 children (19.2%; 95% CI, 14.1%-25.4%) had elevated levels. Only severe reactions were associated with reaction levels of 11.4 μg/L or more (odds ratio, 6.5; 95% CI, 2.2-19.0). Milk-induced anaphylaxis and severe reactions were more likely associated with increased tryptase levels (beta-adjusted, 4.0; 95% CI, 0.95-7.0, and 7.5; 95% CI, 4.8-10.3, respectively). Reaction levels exceeding the threshold level of 2 ng/mL + 1.2 × (postreaction tryptase level) detected most of the anaphylactic reactions, particularly if baseline levels were taken within 2 months of the reaction.

Conclusions: Tryptase levels are particularly useful for the diagnosis of severe and/or milk-induced anaphylaxis. Assessing the difference between reaction and postreaction tryptase levels may improve diagnostic sensitivity. (J Allergy Clin Immunol 2015;■■■:■■■-■■■.)

Key words: *Anaphylaxis, children, diagnosis, milk allergy, tryptase*

The diagnosis of anaphylaxis[1,2] currently relies on a suggestive clinical history after exposure to a potential triggering agent or event. However, the diagnosis may be challenging because it is not always possible to identify a clear trigger[2,3] and its presentation may mimic that of other serious medical conditions. Given that anaphylaxis affects at least 1.6% of the North American population,[4] it is crucial to identify biomarkers that will aid in the diagnosis of this important clinical condition.

Certain mast-cell mediators including tryptase, histamine, and platelet-activating factor have been purported to be elevated in anaphylaxis,[5] but practical challenges related to the timing and handling of samples may limit the usefulness of histamine and platelet-activating factor.[2] Tryptase levels within the first 3 hours of anaphylaxis are considered to be a selective marker of anaphylaxis and do not require specific handling of the sample.[6-8] However, large-scale studies establishing its role in anaphylaxis, particularly in pediatrics, are lacking.

We aimed to evaluate tryptase levels in children during and after anaphylaxis and to examine predictors of an elevated reaction tryptase level (defined hereafter as levels ≥11.4 μg/L)[9,10] and an increased difference between reaction and postreaction levels.

METHODS

As part of our Cross-Canada Anaphylaxis Registry, children presenting with anaphylaxis[1] (defined below) to the Montreal Children's Hospital emergency department between April 2011 and April 2015 were recruited. Data on reaction characteristics, triggers, patients' comorbidities, and management were collected either prospectively (at the time of presentation) or retrospectively through chart review as previously described.[11]

Anaphylaxis was defined as involvement of 2 organ systems and/or hypotension in response to a potential allergen,[1] and anaphylaxis severity was classified according to a modified grading system published by Brown.[12] Mild anaphylaxis was characterized by the presence of skin and subcutaneous tissue symptoms (urticaria, erythema, and angioedema) as well as oral pruritus, nausea (ie, gastrointestinal involvement and cutaneous) or nasal congestion, sneezing, rhinorrhea, or throat tightness (ie, respiratory involvement). Moderate anaphylaxis was characterized by the presence of any of the previous symptoms as well as crampy abdominal pain, diarrhea or recurrent vomiting, dyspnea, stridor, cough, wheeze, or "light headedness." Severe anaphylaxis was characterized by cyanosis, hypoxia (saturation <92%), respiratory arrest, hypotension, dysrhythmia, confusion, or loss of consciousness.[12]

Total tryptase levels were measured 30 to 120 minutes after the onset of symptoms at the discretion of the treating physician. All patients with anaphylaxis were referred for assessment and management to the Montreal Children's Hospital Paediatric Allergy clinics. In patients who consented to follow-up by the research team, postreaction tryptase levels were measured

From [a]the Division of Allergy and Clinical Immunology, Department of Pediatrics, Montreal Children's Hospital, Montreal; [b]the Division of Paediatric Allergy and Clinical Immunology, Department of Paediatrics, University of Manitoba, Winnipeg; [c]the Division of Rheumatology, Department of Medicine, University of Calgary, Calgary; [d]Food Directorate, Health Canada, Ottawa; [e]the Department of Emergency Medicine, Department of Pediatrics, Montreal Children's Hospital, Montreal; [f]the Department of Epidemiology and Biostatistics, McGill University, Montreal; and [g]the Department of Emergency Medicine, Hôpital du Sacré-Cœur, Montreal.

This study was supported by the Allergy, Genes, and Environment (AllerGen) Network of Centres of Excellence (grant no. 12canFAST3), Health Canada, and by Sanofi.

Disclosure of potential conflict of interest: S. De Schryver, M. Halbrich, R. Alizadehfar, J. Morris, and M. Ben-Shoshan have received grants from Allergy, Genes, and Environment Network of Centres of Excellence, Health Canada, and from Sanofi. The rest of the authors declare that they have no relevant conflicts of interest.

Received for publication May 15, 2015; revised August 11, 2015; accepted for publication September 3, 2015.

Corresponding author: Moshe Ben-Shoshan, MD, MSc, Montreal Children's Hospital, 1001 Blvd Décarie, Montreal, QC, Canada H4A 3J1. E-mail: moshebenshoshan@gmail.com.

0091-6749/$36.00

http://dx.doi.org/10.1016/j.jaci.2015.09.001

regardless of the level during anaphylaxis. Total tryptase level was measured using the UniCAP-Tryptase fluoroimmunoassay (Phadia, now Thermo Fisher Scientific, Uppsala, Sweden), following the manufacturer's instructions. A serum tryptase level of more than 11.4 μg/L was considered high.[10] As it has been suggested that the pathognomonic laboratory finding indicative of mast-cell degranulation is an acute total tryptase level (within 4 hours of the reaction) above 2 ng/mL + 1.2 × (baseline tryptase level) (drawn at least 24 hours after resolution of the event),[13,14] we also determined whether the tryptase level drawn during anaphylaxis in our cohort exceeded this threshold.

Descriptive statistics were used to assess demographic, clinical, and reaction characteristics, epinephrine use, and the levels of tryptase during and after the reaction. The demographic, clinical, and reaction characteristics of all participants presenting with anaphylaxis with and without measurement of tryptase levels were compared to address the potential for selection bias. These characteristics were also compared between those with tryptase level measured both during and after the reaction and measured only during the reaction. Tryptase levels during the anaphylactic reaction and postreaction were compared using CIs based on paired means using the *t* distribution. Multivariate logistic regressions were used to identify predictors of an elevated reaction tryptase level (beyond the threshold of 11.4 μg/L) as well as a reaction tryptase level exceeding the threshold of 2 ng/mL + 1.2 × (baseline tryptase level). Multivariate linear regressions were used to identify predictors of level of tryptase during the reaction and differences between tryptase levels during and after the reaction. Potential predictors included age, sex, reaction trigger, reaction severity, history of atopy, and interval between measurement of reaction and postreaction tryptase level (where applicable). All statistical analyses were conducted using R version 2.12.0 (October 15, 2010).

This study received ethics approval from the McGill Research Ethics Board.

RESULTS

Patient characteristics, anaphylaxis severity, management, and triggers

Over a period of 4 years, 965 children presented to the emergency department with anaphylaxis and of these, 203 had serum tryptase levels measured within 2 hours of the onset of the anaphylactic reaction. The 203 children with measurement of tryptase level were compared with the 762 children who were admitted to the emergency department with anaphylaxis over the same time period without measurement of tryptase level (Table I). The 2 groups were comparable regarding age, sex (predominantly male), reaction triggers, and known asthma. Most of the reactions occurred at home and during normal activity. In contrast, there were more children recruited prospectively who had tryptase level measured. Although food was the major trigger in both groups, tree nut was a more common precipitant in the group with tryptase level measurements. Although eczema was more commonly reported in children with tryptase level measurements, a known food allergy was more commonly reported in children without tryptase level measurements. Severe reactions were reported more frequently in children with tryptase level measurements, and the use of epinephrine inside the hospital was also higher in participants with tryptase level measurements, but the use outside the hospital was lower.

Serum tryptase level concentrations

Among the 203 children with serum tryptase level measurements, the mean level was 7.6 ± 6.2 μg/L and 39 cases (19.2%; 95% CI, 14.1% to 25.4%) had elevated tryptase levels (≥11.4 μg/L). Elevated levels were found in 9 of 18 or 50.0% (95% CI, 29.0% to 71.0%) of severe reactions, in 24 of 148 (95% CI, 10.8% to 23.4%) or 16.2% of moderate reactions, and in 6 of 37 or 16.2% (95% CI, 6.8% to 32.7 %) of mild reactions.

Among the 203 cases with tryptase level measurements within 2 hours of reaction, postreaction tryptase level was available on 68 (33.5%; 95% CI, 27.1% to 50.5%). The mean time interval between postreaction and reaction levels was 8.7 months. Tryptase levels during reaction were higher in subjects with postreaction levels. Other patient and reaction characteristics were comparable between those with and without postreaction levels (Table II). Most were recruited prospectively and were male. Most reactions were triggered by food and occurred most commonly at home and during normal activity.

Of the 68 children with postreaction tryptase levels, the mean tryptase level was 9.9 μg/L at the time of reaction and 3.6 μg/L postreaction, yielding a difference of 6.3 μg/L (95% CI, 4.7-7.8). The suspected trigger was confirmed in 76.5% (95% CI, 69.9% to 82.3%) of the 203 children who had reaction tryptase level measured and in all cases who had both reaction and postreaction measurements. In 41 of 68 children (60.3%; 95% CI, 47.7% to 71.7%), the reaction tryptase level exceeded the published threshold of 2 ng/mL + 1.2 × (baseline or postreaction tryptase level). Levels exceeding this threshold were found in 85.7% of severe reactions (95% CI, 42.0% to 99.2%), 54.2% of moderate reactions (95% CI, 39.3% to 68.4%), and 69.2% (95% CI, 38.9% to 89.6%) of mild reactions.

Specific risk factors associated with elevated tryptase level

The presence of a severe reaction was the only factor associated with elevated tryptase levels (≥11.4 μg/L) during the reaction, after adjusting for sociodemographic and reaction characteristics and comorbidities (adjusted odds ratio, 8.0; 95% CI, 5.2-10.8). A severe reaction and a milk trigger were associated with increased levels of tryptase during the reaction, regardless of previously published thresholds (beta-adjusted, 7.5; 95% CI, 4.8-10.3, and 4.0; 95% CI, 0.95-7.0, respectively). A severe reaction was also associated with an increased difference between reaction and postreaction tryptase levels (beta-adjusted, 9.7; 95% CI, 5.6-13.8). Interestingly, measurement of tryptase levels at least 3 months after the reaction was associated with a decreased difference (beta-adjusted, −3.7; 95% CI, −6.9 to −0.4).

The reaction tryptase levels was more likely to exceed 2 ng/mL + 1.2 × (baseline tryptase level) in severe reactions (adjusted odds ratio, 1.5; 95% CI, 1.1-2.1), whereas it was less likely to exceed this threshold with an increased time interval between reaction and postreaction levels (adjusted odds ratio, 0.99; 95% CI, 0.98-0.99). We did not detect a significant interaction between the type of food trigger (including milk) and age.

DISCUSSION

We have conducted the largest study assessing levels of tryptase during and after anaphylaxis in children and the first study to evaluate factors associated with an elevated level or a difference between postreaction and reaction levels. Our results reveal that the tryptase level during the reaction exceeded a previously published threshold in 50% cases of