Insect anaphylaxis: addressing clinical challenges

James M. Tracy, Elena J. Lewis and Jeffrey G. Demain

Division of Allergy and Immunology and Allergy, Asthma & Immunology Associates, Omaha, Nebraska, USA

Correspondence to James M. Tracy, D.O., Creighton University School of Medicine, Allergy, Asthma & Immunology Associates, P.C. 2808 South 80th Ave, Suite 210, Omaha, NE 68124, USA
Tel: +1 402 391 1800; e-mail: jmtracy@cox.net

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Purpose of review

Few allergic reactions are as potentially life-threatening, or frightening to the patient, as anaphylaxis. Food, medications, and insect stings are the three most common triggers of anaphylaxis, but insect allergy provides the best opportunity to understand the biology of anaphylaxis. If the physician can establish a diagnosis of insect allergy, treatment with nearly 98% effectiveness can be initiated. However, sometimes patients have a compelling history of insect sting anaphylaxis, but negative skin and blood tests. This situation presents us with a fascinating opportunity to understand the biology of insect anaphylaxis.

Recent findings

Recent and ongoing work shows that occult mast cell disease may be critical in insect anaphylaxis. Mastocytosis, serum tryptase and basophil biology are key elements; genetic markers may potentially help us diagnose at-risk individuals and determine proper treatment. Understanding basophil activation may play an additional role both in diagnosis and knowing when therapy might be terminated.

Summary

Mast cell disease, serum tryptase and basophil biology are providing an opportunity to better understand and manage insect allergy. This evolving understanding should improve long-term management of insect anaphylaxis and help us to better understand the clinical dilemma of appropriate management of the history-positive patient in which testing is unable to detect venom-specific IgE. Furthermore, omalizumab's immunomodulatory effects may play a role in difficult-to-treat insect allergy and mastocytosis. Finally, unrelated to these, but still important as an ongoing risk factor, is the continued underutilization of epinephrine for both acute and long-term management of insect anaphylaxis.

Keywords

anaphylaxis, basophil, epinephrine, hymenoptera, mast cell, mastocytosis, tryptase

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Introduction

Of the three most common triggers of anaphylaxis – foods, medications, and insect allergies [1–4] – insect allergy provides us with unique opportunities to understand the biology of sensitivity and potentially fatal anaphylactic reactions. Insect allergy diagnosis is well established and can be treated with a 98% level of protection in sensitive individuals [5•,6–8]. In most cases the sting event is obvious, the nature of the reaction is clear, and specific testing can be accomplished, when appropriate [5•].

In clinical practice things are not always so straightforward. What if the physician believes that the patient has had a systemic reaction to an insect sting, but the highly reliable skin and blood tests are negative? [9–11] Since these tests are the basic criteria for initiating venom immunotherapy (VIT), without specific sensitivity established, VIT would not be started. As a result, this patient may remain at a greater risk for potentially fatal insect anaphylaxis [5•]. It is this biological enigma that has forced allergists to re-examine management of this vexing clinical dilemma and as a result expand our understanding of the biology of anaphylaxis. Herein, we examine the recent literature in the areas of mastocytosis, utility of serum tryptase, basophil activation, genetic expression, risk factors and management tools as they relate to insect anaphylaxis.

Insect allergy: tryptase level and mastocytosis

The appropriate management of patients with a compelling history of insect-induced anaphylaxis, yet are skin and blood-test negative, remains a clinical challenge [9,11]. Mastocytosis has emerged as a surprising link in this clinical quandary. A significant percentage of patients with severe systemic reactions following insect sting, who are demonstrated to have an elevated baseline tryptase level, indeed, may have mastocytosis. Consequently, in

patients with mastocytosis, the most common cause of anaphylaxis is insect sting [12].

A multicenter, European study [13••] of 962 Hymenoptera venom-sensitive patients looked at predictors of severe systemic anaphylaxis following a sting. Of the 962 patients, 202 (26%) had severe anaphylaxis following a field sting. The reported severity was either grade 3 (anaphylactic shock, loss of consciousness) or grade 4 (cardiac arrest, apnea). The identified risk predictors of sting-related anaphylaxis included patients taking angiotensin converting enzyme inhibitors; patients with vespid allergy; patients of male sex; and most interestingly, patients with baseline serum mast cell tryptase levels above 5 ng/l. In a separate study, Bonadonna *et al.* [14••] also reported a correlation between systemic reaction to Hymenoptera sting and mast cell tryptase. Of 379 patients with a history of systemic insect sting reactions, 11.6% had serum mast cell tryptase levels exceeding 11.4 ng/ml. Of this group, the rate of systemic (Muller grade IV) anaphylaxis was 70.5%. Thirty-four of the patients with elevated mast cell tryptase level underwent bone marrow biopsy; of those, 61.8% were ultimately diagnosed with indolent systemic mastocytosis. The opposite was true of food and drug allergies. A subsequent study [15] looked at patients with a history of severe systemic anaphylaxis induced either by a food or a drug. Of 137 patients, only 9 (6.6%) had a basal tryptase above 11.4 ng/ml. Ultimately, only two (1.5%) were identified with mastocytosis. In comparison to the prior study, the patients with Hymenoptera-induced anaphylaxis had a higher rate (11.6%) of basal tryptase level above 11.4 ng/ml. In addition, of the 379 insect-allergic patients, 21 (5.5%) were subsequently diagnosed with indolent systemic mastocytosis compared with 2 of 137 (1.5%) patients with severe food or drug-related reactions. Blum *et al.* [16] confirmed this connection. This study conducted a 5-year retrospective analysis on 868 patients referred for systemic reaction to Hymenoptera sting. Of those, 758 had both total IgE and baseline tryptase levels drawn. Baseline tryptase level (>11.4 ng/ml) was associated with severe systemic reactions ($P = 0.026$). In a German study [17], the correlation between severe systemic reaction to sting and elevated baseline tryptase ($P = 0.003$) was further confirmed as was an association with increasing age ($P = 0.001$). Because of the dramatic increase in severe sting-related anaphylaxis in patients with mastocytosis, physicians should consider clonal mast cell disease in anyone with unexplained anaphylaxis or sting-related anaphylaxis [18].

Not surprisingly, VIT in mastocytosis patients carries greater side effect risk than reported in the nonmastocytosis, venom-allergic patients. In a large multicenter, European study, Rueff *et al.* [19] studied 680 patients with established venom allergy to either honeybee or vespid. The objective was to determine if baseline tryptase level correlated with severe reactions occurring during the immunotherapy build-up phase. Fifty-seven patients, or 8.4%, experienced a significant enough adverse event as to warrant emergency intervention. The frequency of severe events correlated with higher basal tryptase levels with an odds ratio (OR) of 1.56 ($P < 0.005$). Interestingly, bee venom also correlated with higher risk of systemic reaction. Contrary to the current recommendation of 3–5-year duration of VIT, patients with mastocytosis have lower probability of long-term protection and consequently a greater risk for recurrent severe, even fatal anaphylaxis [20]. A thorough review published in this journal by Bonadonna *et al.* [21] recommends VIT for life in patients with mastocytosis and venom allergy. Since the efficacy of venom immunotherapy is less than optimal in mastocytosis patients, they should continue to carry two epinephrine injectors [22••]. In addition, baseline tryptase levels should be considered in all patients with anaphylaxis related to Hymenoptera sting. In this review, we have discussed several studies that have reported a high correlation with mastocytosis when the baseline tryptase exceeds 11.4 ng/ml, suggesting that this diagnosis be entertained in patients with severe anaphylaxis from Hymenoptera.

Key points

- Mast cell disease is key in understanding anaphylaxis.
- Basophil activation may aid in the diagnosis of insect allergy.
- Epinephrine is underutilized for the acute management of insect anaphylaxis.

Gene expression

Indolent or occult mastocytosis continues to play a role in the presence and severity of anaphylaxis and insect venom anaphylaxis as well as gene expression. Niedoszytko *et al.* [23•] accessed the difference in gene expression between patients with indolent mastocytosis and a history of venom anaphylaxis compared to patients with indolent mastocytosis without anaphylaxis. Their preliminary results suggest a difference in gene profiling between the two groups using whole-genome gene expression analysis [23•]. The same group also suggested that the angiotensinogen AGT p.M235T gene polymorphism may be responsible for the development of severe anaphylactic reactions to insect venom and looked at methods to determine if analysis of gene expression might be predictive of the effectiveness of VIT. Forty-six patients with 48 071 genetic transcripts demonstrated a genetic pattern characteristic of successful VIT [24]. This finding suggests a role for genetic profiling for successful long-term VIT [25]. In other work, Karakis *et al.* [26] looked at MCH associations between HLA class I and

HLA class II antigens. The purpose of the study was to look at broad HLA status as a predictor of those individuals who develop allergic reactions to insect stings, whereas others do not develop insect sting allergy. Twenty-one insect-venom-sensitive patients with life-threatening reactions were compared with 37 healthy individuals. Their finding indicated that HLA-B*18 and HLA-Cw*-7 alleles may be associated with susceptibility to venom allergy [26].

Basophils/basophil activation test

Mast cells and their mediators have been well studied. Our understanding of the role of the basophil also continues to expand. Since VIT is the principal management tool in insect anaphylaxis, an accurate diagnosis of venom hypersensitivity is crucial for its effective use. Current recommendations in the US dictate that skin prick testing, followed by serial increasing intradermal concentrations of specific venom and/or specific IgE in-vitro testing, be used to determine venom hypersensitivity. However, there remains a subset of patients with a history of systemic reaction after insect sting with negative testing in a range of 6–20% [27]. The basophil activation test (BAT) may provide an additional window for accurate diagnosis of insect venom hypersensitivity in the history-positive but venom test-negative patient. Furthermore, there is the issue of double positive results that may be secondary to true multiple venom allergies or cross-reactivity occurring in up to 50% of individuals [28]. In the case of cross-reactivity, knowing the true culprit is important when choosing components to use in VIT, since including components to which the patient is not truly allergic could lead to development of sensitization to other epitopes [27]. Lastly, there is currently no recommended testing to determine when and if patients should stop VIT [5•]. Testing to determine which patients are at an increased risk of reaction to a sting after VIT has been stopped would help clinicians determine which patients would benefit from continued lifelong VIT. The BAT has been studied as a surrogate test to assist in answering these questions.

The BAT measures CD63 activation on basophils when stimulated *in vitro* with specific IgE via flow cytometry. A large cohort study [29] of more than 1000 patients found 4% of patients with a history of systemic reaction after insect sting had negative skin testing (both skin prick and intradermal testing) and IgE testing. Sixty percentage of these negative patients were subsequently positive by BAT. There was only one patient reported as negative to BAT and positive via intradermal testing [29]. Testing using specific IgE is limited when low IgE levels causing results in a false-negative test. Skin testing is limited to intradermal testing concentrations maximum of 1 μg/ml due to irritant effects present at higher concentrations. BAT can be accomplished when low IgE is present and has wider concentration limits – again revealing the utility of this test. In a study [27] of BAT use in children, it was found to be less sensitive compared with adult studies (67–75 vs. 87–100%, respectively). The availability of the test decreases its utilization since there are only a few specialized laboratories where BAT is standardized and can be reliably performed. Furthermore, approximately 15–20% of the general population are considered BAT nonresponders or nonreleasers. However, this number can be decreased to 5% if the basophils are incubated with IL-3 [30••]. This test remains a viable option for patients who are otherwise negative via standard venom hypersensitivity testing. The limitations need to be further studied. BAT in patients with double positive venom hypersensitivity testing has recently been studied. Peternelj *et al.* [30••] classified 17% of double positive patients via IgE testing as positive for only a single venom on BAT. Kosnik *et al.* [28] were able to find 1/3 of patients in their cohort as single positive after BAT [29]. These studies reveal the potential benefit of BAT in more specifically characterizing patients with otherwise unclear testing thereby allowing more accurate VIT in these patients.

Next we examine the use of BAT in predicting a patient's response to future insect stings after completion of VIT. Literature is conflicting on this topic. Patients who had completed VIT for bee venom hypersensitivity were studied and sting challenge was performed as a gold standard method of determining reactivity. BAT in patients treated for at least 3.1 years of VIT correlated well with sting challenge. When tested with concentrations of 100 ng/ml of bee venom, 80% of sting challenge reactors had a positive BAT and 87.5% of nonreactors had a negative BAT [31]. Other authors have confirmed or rejected these findings in their studies, so further work is needed before recommendations for BAT and VIT can be applied [32–34].

The role of omalizumab in mast cell disease

The management of insect allergic individuals can be complicated by comorbid occult or indolent mastocytosis. Omalizumab has been reported to be a useful adjunctive therapy in individuals with severe anaphylaxis to bee venom immunotherapy [35,36]. Several recent cases report the usefulness of omalizumab in the management of insect anaphylaxis and indolent or occult mastocytosis. Kontou-Fill *et al.* [37•] reported the successful use of omalizumab mono-therapy as an effective strategy in a 48-year-old with occult mastocytosis who was venom-specific IgE negative and suffered three near-fatal bee sting reactions. Douglass *et al.* [38] reported the successful use of omalizumab in treating systemic mastocytosis in an atopic patent. In both Kontou-Fill *et al.*'s work and

that of Douglass *et al.*, the clinical effectiveness as measured by serum tryptase was directly attributed to omalizumab. This suggests that the effect of omalizumab may reside in its impact on mast cell stability. Further investigation is warranted before drawing long-range conclusions, but the observations are important.

Risk factors

Since venom allergy is one of the top three causes of anaphylaxis, it is necessary to have proper prevention and management strategies in place for these patients [39••]. It has been well established that patients are undertreated with epinephrine during anaphylaxis and strategies for prevention are overlooked at discharge [40,41]. A recent study [38] on epinephrine administration in the emergency department for stinging insect hypersensitivity found that only 18% of adult patients presenting with anaphylaxis received epinephrine. However, the same study reported 92% of children, defined as below 17 years of age, received epinephrine. Literature supports the use of epinephrine in cases of anaphylaxis as the first-line therapy with increased risk of death in patients who did not receive epinephrine or in cases when it was delayed in patient management. For patients presenting with systemic reactions, only 11% were referred to an allergist, only 3% had documentation of insect avoidance, and 68% received a prescription for self-injectable epinephrine at discharge [38]. Another study [42] in beekeepers found that only 66% of beekeepers who attended the emergency department for systemic reactions were reviewed by an allergist for further management. These findings may represent a lack of understanding among healthcare providers about the lifesaving benefits of VIT in insect venom hypersensitivity. It also stresses that education is needed since prevention and treatment are essential in preventing further reactions. VIT has been repeatedly shown to decrease risk of further reactions if re-stung and is now standard practice in individuals that have insect hypersensitivity [38].

The need for a second dose of epinephrine has been noted in patients with anaphylaxis based on limited data in food-induced anaphylaxis [43]. Risk factors associated with requiring a second epinephrine dose were observed in a recent study by Manivannan *et al.* [44]. This study included all causes of anaphylaxis (not only those specifically due to venom). Overall 13% of patients presenting with anaphylaxis received two doses of epinephrine. These patients were statistically significantly younger and presented more likely with respiratory, cardiac and gastrointestinal symptoms and were less likely to have urticaria. A history of asthma was not predictive of repeating epinephrine doses. Although 71.4% of patients who required a second dose of epinephrine were prescribed self-injectable epinephrine, the overall prescription rate was still low. Only 39.3% of patients treated with a single dose of epinephrine were discharged with a prescription for self-injectable epinephrine. Allergy referral was given for 51.9% of patients requiring repeated epinephrine doses and 40.3% of patients given single epinephrine dose [44].

Another important aspect focuses on who is at risk of anaphylaxis. A recent questionnaire was evaluated in a population of British beekeepers. It revealed some novel associations with the development of systemic reactions in this distinct population. Factors that were found to be associated with increased risk of systemic reactions included having a family member with bee venom allergy, having practiced beekeeping for more than 2 years, having premedication with antihistamine prior to sting, and being female [39••]. This study was performed on a selected group of individuals and may not be applied universally. In a large European work, Ruëff *et al.* [13••] reported that, of 962 insect-allergic patients, male sex was considered a risk factor for future anaphylaxis. Clearly, these are areas that need to be further studied prospectively before recommendations can be made.

Conclusion

Insect allergy is a common trigger for anaphylaxis and treatment provides a high level of protection. Improved understanding of the biology of various effector cells and their mediators will improve diagnosis, thereby enabling appropriate long-term treatment. Novel therapies such as omalizumab may also have a therapeutic role in difficult-to-manage insect-allergic individuals. Unfortunately, despite our recent observations in diagnosis management, the proper utilization of epinephrine in the outpatient arena and appropriate specialty referral remain as obstacles to effective long-term management of insect allergy and anaphylaxis.

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References and recommended reading

Papers of particular interest, published within the annual period of review, have been highlighted as:

- • of special interest
- •• of outstanding interest

Additional references related to this topic can also be found in the Current World Literature section in this issue (p. 388).

1 Simons FE. Anapylaxis. J Allergy Clin Immunol 2008; 121 (2 Suppl):S402–S407.

2 Simons FER, Frew AJ, Ansotegui IL, *et al.* Risk assessment in anaphylaxis: current and future approaches. J Allergy Clin Immunol 2007; l20:S2–S24.

3 Sampson HA, Mendclson L, Rosen JP. Fatal and near-fatal anaphylactic reactions to food in children and adolescents. N Engl J Med 1992; 327:380–384.

4 Simons PER, Sampson HA. Anaphylaxis epidemic: fact or fiction? J Allergy Clin Immunol 2008; 122:1166–1168.

5 • Golden DB, Moffitt JE, Nicklas RA, *et al.* Stinging insect hypersensitivity: a practice parameter update 2011. J Allergy Clin Immunol 2011; 127:852–854.
Most current and comprehensive review of insect allergy diagnosis and management available.

6 Valentine M. Insect venom allergy: diagnosis and treatment. J Allergy Clin Immunol 1984; 73:299–304.

7 Hunt KJ, Valentine MD, Sobotka AK, *et al.* A controlled trial of immunotherapy in insect hypersensitivity. N Engl J Med 1978; 299:157–161.

8 Reisman RE, Livingston A. Venom immunotherapy: 10 years of experience with administration of single venoms and 50 micrograms maintenance doses. J Allergy Clin Immunol 1992; 89:1189–1195.

9 Reisman RE. Insect sting allergy: the dilemma of the negative skin test reactor. J Allergy Clin Immunol 2001; 107:781–782.

10 Golden DBK, Kagey-Sobotka A, Hamilton RG, *et al.* Insect allergy with negative venom skin tests. J Allergy Clin Immunol 2001; 107:897–901.

11 Golden DBK, Tracy JM, Freeman TM, Hoffman DR. Negative venom skin test results in patients with histories of systemic reaction to a sting (Rostrum article). J Allergy Clin Immunol 2003; 112:495–498.

12 Brockow K, Jofer C, Behrendt H, Ring J. Anaphylaxis in patients with mastocytosis: a study on history, clinical features and risk factors in 120 patients. Allergy 2008; 63:226–232.

13 •• Ruëff F, Przybilla B, Biló MB, *et al.* Predictors of severe systemic anaphylactic reactions in patients with Hymenoptera venom allergy: importance of baseline serum tryptase-study of the European Academy of Allergology and Clinical Immunology Interest Group on Insect Venom Hypersensitivity. J Allergy Clin Immunol 2009; 124:1047–1054.
This observational, multicenter, European study of 962 Hymenoptera-sensitive patients demonstrates a direct relationship between increased basal serum tryptase levels and severe anaphylaxis following sting.

14 •• Bonadonna P, Perbellini O, Passalacqua G, *et al.* Clonal mast cell disorders in patients with systemic reactions to Hymenoptera stings and increased serum tryptase levels. J Allergy Clin Immunol 2009; 123:680–686.
A multicenter study demonstrating a correlation between severe insect anaphylaxis and mastocytosis.

15 Bonadonna P, Zanotti R, Pagani M, *et al.* How much specific is the association between hymenoptera venom allergy and mastocytosis? Allergy 2009; 64:1379–1382.

16 Blum S, Gunzinger A, Muller UR, Helbling A. Influence of total specific IgE, serum tryptase, and age on severity of allergic reactions to Hymenoptera stings. Allergy 2011; 66:222–228.

17 Guenova E, Volz T, Eichner M, *et al.* Basal serum tryptase as risk assessment for severe Hymenoptera sting reactions in elderly. Allergy 2010; 65:919–923.

18 Akin C. Anaphylaxis and mast cell disease: what is the risk? Curr Allergy Asthma Rep 2010; 10:34–38.

19 Rueff F, Przybilla B, Bilo MB, Muller U, *et al.* Predictors of side effects during build-up phase of venom immunotherapy for Hymenoptera venom allergy: the importance of baseline serum tryptase. J Allergy Clin Immunol 2010; 126:105–111.

20 Oude Elberink JNK, deMonchy JGR, Kors JW, *et al.* Fatal anaphylaxis after a yellow jacket sting, despite venom immunotherapy, in two patients with mastocytosis. J Allergy Clin Immunol 1997; 100:11–15.

21 Bonadonna P, Zanotti R, Muller U. Mastocytosis and insect venom allergy. Curr Opin Allergy Clin Immunol 2010; 10:347–353.

22 •• Niedoszytko M, de Monchy J, van Doormaal JJ, *et al.* Mastocytosis and insect venom allergy: diagnosis, safety and efficacy of venom immunotherapy. Allergy 2009; 64:1237–1245.
A thorough discussion of mastocytosis, insect allergy and a lower than expected efficacy of venom immunotherapy when compared with that of venom-sensitive patients without mastocytosis.

23 • Niedoszytko M, Bruinenberg M, Van Doormaal JJ, *et al.* Gene expression analysis predicts insect venom anaphylaxis in indolent systemic mastocytosis. Allergy 2010. doi: 10.1111/j.1398-9995.
In 22 patients, using gene ontogeny analysis, there was a significant difference in gene expression profiles between insect venom anaphylaxis with and without indolent systemic mastocytosis. The authors suggest that gene expression profiling may be useful in predicting anaphylaxis in insect-allergic patients with indolent systemic mastocytosis.

24 Niedoszytko M, Ratajska M, Chełmińska M, *et al.* The angiotensinogen AGT p.M235T gene polymorphism may be responsible for the development of severe anaphylactic reactions to insect venom allergens. Int Arch Allergy Immunol 2010; 153:166–172.

25 Niedoszytko M, Bruinenberg M, Van Doormaal JJ, *et al.* Gene expression analysis predicts insect venom anaphylaxis in indolent systemic mastocytosis. Allergy 2010. doi: 10.1111/j.1398-9995.

26 Karakis GP, Sin B, Tutkak H, *et al.* Genetic aspect of venom allergy: association with HLA class I and class II antigens. Ann Agric Environ Med 2010; 17:119–123.

27 Ott H, Tenbrock K, Baron J, *et al.* Basophil activation test for the diagnosis of hymenoptera venom allergy in childhood: a pilot study. Klin Padiatr 2011; 223:27–32.

28 Kosnik M, Korosec P. Importance of basophil activation testing in insect venom allergy. Allergy Asthma Clin Immunol 2011; 5:11.

29 Kruse P, Erzen R, Silar M, *et al.* Basophil responsiveness in patients with insect sting allergies and negative venom-specific immunoglobulin E and skin prick test results. Clinical Exp Allergy 2009; 39:1730–1737.

30 •• Peternelj A, Silar M, Bajrovic N, *et al.* Diagnostic value of the basophil activation test in evaluating Hymenoptera venom sensitization. Wien Klin Wochenschr 2009; 121:344–348.
Prospective study revealing that the basophil activation test is highly sensitive with a good positive predictive value when compared with the gold standard skin prick and IgE testing in diagnosis of venom hypersensitivity.

31 Kucera P, Cvackova M, Hulikova K, *et al.* Basophil activation can predict clinical sensitivity in patients after venom immunotherapy. J Investig Allergol Clin Immunol 2010; 20:110–116.

32 Ebo D, Hagendorens M, Schuerwegh A, *et al.* Flow-assisted quantification of in vitro activated basophils in the diagnosis of wasp venom allergy and follow-up of wasp venom immunotherapy. Cytometry B Clin Cytom 2007; 72:196–203.

33 Erdmann S, Sachs B, Kwiecien R, *et al.* The basophil activation test in wasp venom allergy: sensitivity, specificity and monitoring specific immunotherapy. Allergy 2004; 59:1102–1109.

34 Gober L, Eckman J, Sterba P, *et al.* Expression of activation markers on basophils in a controlled model of anaphylaxis. J Allergy Clin Immunol 2007; 119:1181–1188.

35 Kontou-Fili K. High omalizumab dose controls recurrent reactions to venom immunotherapy in indolent mastocytosis. Allergy 2008; 63:376–378.

36 Galera C, Soohun N, Zankar N, *et al.* Severe anaphylaxis to bee venom immunotherapy: efficacy of pretreatment and concurrent treatment with omalizumab. J Investig Allergol Clin Immunol 2009; 19:225–229.

37 • Kontou-Fill K, Fillis CI, Voulgari C, Panayiotidis PG. Omalizumab monotherapy for bee sting and unprovoked 'anaphylaxis' in a patient with systemic mastocytosis and undetectable specific IgE. Ann All Asthma Immunol 2010; 104:537–539.
A case report of successful management of insect anaphylaxis in an individual with a significant history of insect anaphylaxis and negative diagnostic testing using omalizumab.

38 Douglass JA, Carroll K, Voskamp A, *et al.* Omalizumab is effective in treating systemic mastocytosis in a nonatopic patient. Allergy 2009; 65:926–927.

39 •• Rudders SA, Banerji A, Katzman DP, *et al.* Multiple epinephrine doses for stinging insect hypersensitivity reactions treated in the emergency department. Ann Allergy Asthma Immunol 2010; 105:85–93.
16% of patients presenting to the emergency department between Jan 2001 and Dec 2006 with systemic stinging insect hypersensitivity required a second dose of epinephrine. Physicians should consider prescribing two doses of self-injectable epinephrine to patients at risk for systemic stinging insect hypersensitivity.

40 Demain JG, Minaei AA, Tracy JM. Anaphylaxis and insect allergy. Curr Opin Allergy Clin Immunol 2010; 10:318–322.

41 Simons FE, Clark S, Camargo CA Jr. Anaphylaxis in the community: learning from the survivors. J Allergy Clin Immunol 2009; 124:301–306.

42 Richter A, Nightingale P, Huissoon A, Krishna M. Risk factors for systemic reactions to bee venom in British beekeepers. Ann Allergy Asthma Immunol 2011; 106:159–163.

43 Järvinen KM, Sicherer SH, Sampson HA, Nowak-Wegrzyn A. Use of multiple doses of epinephrine in food-induced anaphylaxis in children. J Allergy Clin Immunol 2008; 122:133–138.

44 Manivannan V, Capbell R, Bellolio M, *et al.* Factors associated with repeated use of epinephrine for the treatment of anaphylaxis. Ann Allergy Asthma Immunol 2009; 103:395–400.