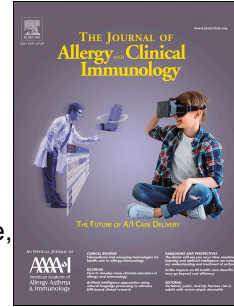


Journal Pre-proof

Prevalence of mastocytosis and hymenoptera venom allergy in the United States

Charles F. Schuler, IV, MD, Sofija Volertas, MD, Dilawar Khokhar, MD, Huseyin Yuce, PhD, Lu Chen, MS, MPH, Onur Baser, PhD, Jenny M. Montejo, MD, Cem Akin, MD



PII: S0091-6749(21)00652-7

DOI: <https://doi.org/10.1016/j.jaci.2021.04.013>

Reference: YMAI 15082

To appear in: *Journal of Allergy and Clinical Immunology*

Received Date: 11 January 2021

Revised Date: 26 March 2021

Accepted Date: 7 April 2021

Please cite this article as: Schuler IV CF, Volertas S, Khokhar D, Yuce H, Chen L, Baser O, Montejo JM, Akin C, Prevalence of mastocytosis and hymenoptera venom allergy in the United States, *Journal of Allergy and Clinical Immunology* (2021), doi: <https://doi.org/10.1016/j.jaci.2021.04.013>.

This is a PDF file of an article that has undergone enhancements after acceptance, such as the addition of a cover page and metadata, and formatting for readability, but it is not yet the definitive version of record. This version will undergo additional copyediting, typesetting and review before it is published in its final form, but we are providing this version to give early visibility of the article. Please note that, during the production process, errors may be discovered which could affect the content, and all legal disclaimers that apply to the journal pertain.

© 2021 Published by Elsevier Inc. on behalf of the American Academy of Allergy, Asthma & Immunology.

1 Prevalence of mastocytosis and hymenoptera venom allergy in the United States

2

3 Charles F. Schuler IV, MD*^{1,2}, Sofija Volertas, MD*³, Dilawar Khokhar, MD¹, Huseyin

4 Yuce, PhD⁴, Lu Chen, MS, MPH⁵, Onur Baser, PhD⁶, Jenny M. Montejo, MD⁷, Cem

5 Akin, MD¹

6 ¹ Division of Allergy and Clinical Immunology, Department of Internal Medicine,

7 University of Michigan, Ann Arbor, MI

8 ² Mary H. Weiser Food Allergy Center, University of Michigan, Ann Arbor, MI

9 ³ Division of Rheumatology, Allergy and Immunology, Department of Internal Medicine,

10 University of North Carolina, Chapel Hill, NC

11 ⁴ New York City College of Technology, New York, NY

12 ⁵ Columbia Data Analytics, LLC, New York, NY

13 ⁶ MEF University, Istanbul, Turkey

14 ⁷ Mayo Clinic, Rochester, MN

15 *Both authors contributed equally to this work.

16

17 Corresponding Author:

18 Charles Schuler

19 Address: Domino's Farms, 24 Frank Lloyd Wright Drive, Suite H-2100, Ann Arbor, MI

20 48106-0442, USA

21 Email: schulerc@med.umich.edu

22 Phone: 734-232-2154

23 Fax: 734-647-6263

24

25 Clinical Implications

26 Mastocytosis is less common among hymenoptera venom allergy patients in the United
27 States than in Europe. Basal serum tryptase elevations may predict venom
28 immunotherapy reactions.

29

30 Capsule Summary

31 Mastocytosis is less common in hymenoptera venom allergy patients in the United
32 States versus Europe. However, elevated basal serum tryptase may predict venom
33 immunotherapy reactions, supporting the recommendation to check this in venom
34 anaphylaxis patients.

35

36 Key words

37 Tryptase, venom allergy, venom immunotherapy, anaphylaxis, mastocytosis, mast cell
38 activation syndrome, mast cell disease

39

40 Abbreviations

41 Hymenoptera venom allergy (HVA)

42 United States (US)

43 Venom immunotherapy (VIT)

44 Mast Cell Disease (MCD)

45 American Academy of Allergy, Asthma, and Immunology (AAAAI)

46

47 **Word Count:** 3506

48

49 **Funding and Conflict of Interest**

50 This work described herein was not directly supported by any funding body.

51 Dr. Schuler reports salary and other support from the Mary H. Weiser Food Allergy

52 Center and the Taubman Innovation Institute at UM; he has also received sponsored

53 project support from Healgen Scientific and Access Bio Inc for unrelated projects. The

54 other authors report no conflicts of interest.

55 **Abstract**

56 **Background:** Mastocytosis is a risk factor for hymenoptera venom anaphylaxis (HVA).

57 Current guidelines recommend measuring tryptase in HVA patients and that those with
58 mastocytosis pursue lifelong venom immunotherapy (VIT). Available data on HVA and
59 mastocytosis largely derives from European single-center studies and the prevalence of
60 HVA with and without mastocytosis in the United States (US) is unknown.

61 **Objective:** We sought to determine the prevalence of HVA and mastocytosis in the US
62 using an insurance claims database and evaluate the impact of mastocytosis on VIT in
63 HVA patients in a US cohort.

64 **Methods:** The IBM Watson Database, consisting of insurance claims from
65 approximately 27 million US patients in 2018, was queried to identify patients with HVA
66 and/or mastocytosis. Further, a retrospective study of 161 patients undergoing VIT
67 between 2015 – 2018 at the University of Michigan (U-M) was conducted.

68 **Results:** In the IBM Watson Database, the prevalence of HVA was 167 per 100,000
69 (0.167%) and the prevalence of mastocytosis 10 per 100,000 (0.010%) overall and 97
70 per 100,000 (0.097%) among those with HVA. Mastocytosis showed a 9.7-fold increase
71 among HVA patients versus the general population. In the U-M cohort, 2.6% of VIT
72 patients had mastocytosis. Tryptase level did not correlate with venom reaction severity
73 but was higher in patients with systemic VIT reactions.

74 **Conclusions:** We observed a lower US HVA prevalence than previously reported.
75 Mastocytosis was more common in US HVA patients, though at lower rates than
76 previously reported. In VIT patients there was no correlation between tryptase level and
77 reaction severity.

78 Introduction

79 Hymenoptera venom allergy (HVA) constitutes an IgE-mediated anaphylactic
80 reaction with a prevalence from 0.5% to 3.3% in the United States (US) and 0.3% to
81 7.5% in Europe (1, 2). Patients are prescribed epinephrine as a rescue medicine and
82 may undergo prophylactic venom immunotherapy (VIT); VIT for honeybee, vespids, and
83 wasp reduces systemic reaction rates from 60% in untreated patients to as low as 0-5%
84 (3-5). VIT is recommended for patients with anaphylactic venom reactions (3, 5).

85 Prior work has shown a high prevalence among HVA patients of up to 11.6% (or
86 11,600 per 100,000) with elevated serum tryptase and up to 5.5% (or 5,500 per
87 100,000) for clonal mast cell (MC) disease (MCD), including systemic mastocytosis
88 (SM) (6, 7). Elevated serum tryptase has been linked to severe sting reactions and
89 reactions during VIT (8, 9). The diagnosis of SM also marks an increased risk for severe
90 sting reactions and adverse events during VIT (10, 11). Thus, updated AAAAI
91 guidelines for HVA patients have expanded recommendations for tryptase
92 measurement in this population (3). However, much of the work supporting high rates of
93 SM in HVA patients comes from European single-center studies many of which were
94 also referral centers for mastocytosis and may have carried a selection bias. In addition,
95 these studies were also performed prior to recognition of hereditary alpha tryptasemia
96 (6, 7, 12). A recent single-center study of 159 patients from Israel found a lower rate of
97 tryptase elevation and MC disease when compared to European data (3.8% as
98 compared to 10-15.9%), suggesting geographic differences in co-occurrence of MCD
99 and venom allergy may exist. (13). The rate of clonal MCD in the US HVA population
100 remains unknown.

101 We hypothesized that among HVA and VIT patients, the rate of MCD and
102 elevated tryptase levels would be lower than European data. We further sought to
103 evaluate the role of tryptase as a predictive marker in VIT patients given updated
104 guidelines.

105 **Methods**

106 **Database**

107 The IBM Watson MarketScan® Research Database was queried for patients with
108 a diagnosis of HVA (ICD-10 codes T63.44, T63.45, T63.46, Z91.030, Z91.038) and
109 Mastocytosis (ICD-10 codes D47.01, D47.02, D47.09, C96.21, C94.3). This database
110 consists of de-identified outpatient, inpatient, and pharmaceutical claims of
111 approximately 27 million privately insured patients in 2018. Complete claims originate
112 from over 150 large employer-sponsored health insurance plans with patient coverage
113 in all 50 states. Real world data from January 1st, 2018 through December 31st, 2018
114 was obtained for analysis. Eligible patients had 6-12 months of claims data in 2018.

115

116 **Allergy Clinic Patient Population**

117 We conducted a retrospective study of 161 patients with a history of systemic
118 venom reactions and received venom immunotherapy injections between 2015 and
119 2018 at the University of Michigan Allergy and Immunology clinic. The University of
120 Michigan Medical School Institutional Review Board on Human Subjects (IRB) reviewed
121 and cleared the protocol. Written informed consent was not required because of the
122 retrospective nature of the study.

123

124 **Venom Immunotherapy Protocol**

125 Patients were selected by the treating physician to undergo VIT based on
126 available practice parameter guidelines. A history of a venomous sting followed by an
127 allergic reaction, including a combination of diffuse urticaria, angioedema,

128 gastrointestinal distress (including nausea, vomiting, or diarrhea), respiratory symptoms
129 (including cough, wheeze, or dyspnea), loss of consciousness, or documented low
130 blood pressure was needed to begin venom immunotherapy (3, 5). Patients diagnosed
131 prior to the 2017 US guidelines may have started VIT based on a systemic cutaneous
132 reaction according to earlier guidelines and were not excluded from this study (14).
133 Positive skin or blood specific IgE testing to honeybee, yellow jacket, yellow hornet,
134 white-faced hornet, or wasp was required. Positive skin testing was defined as a wheal
135 of 3 mm greater than the negative control on skin prick or intradermal testing; we note
136 the guideline statements of uncertainty on the intradermal test cutoff, 3 mm over the
137 negative control was used in this study as it is the consensus among the U-M allergy
138 group where the study took place (3). Positive blood testing was defined as any serum
139 IgE level to a venom above the normal range (< 0.35 kU/L) (3). Patients were counseled
140 on risks and benefits by the treating physician in the course of standard clinical care.

141 Venom immunotherapy at the University of Michigan follows US guidelines (3). A
142 monthly maintenance dose of 1 mL of 100 mcg/mL concentrate for each venom, or 1
143 mL of 300 mcg/mL for mixed vespid (yellow jacket, yellow hornet, and white-faced
144 hornet), is used. 0.05 mL of dilutions down to 1:1000 of concentrate, or lower, are used
145 to build up weekly to maintenance.

146 Mastocytosis was diagnosed by the treating physician and the diagnosis was
147 verified by the study team after reviewing the bone marrow biopsy report and tryptase
148 values. Mastocytosis was defined according to the 2016 WHO diagnosis and
149 classification system (15).

150

151 Data Collection

152 A standardized approach was used to collect pre-specified variables from
153 patients' charts. Demographic data included age, gender, and race/ethnicity. Medical
154 history included a history of asthma, atopic sensitization, food allergy, atopic dermatitis,
155 family history of systemic venom allergy reactions, coronary artery disease, ACE
156 inhibitor use, and beta blocker use. A positive skin test (a wheal of 3 mm greater than
157 the negative control) to at least one allergen defined atopy (16). Allergens tested
158 included trees, grasses, weeds, molds, dust mite, cat, and dog. The original venom
159 reaction was coded according to the presence or absence of hives/rash, angioedema,
160 respiratory symptoms (cough, dyspnea, or wheezing), gastrointestinal symptoms
161 (nausea, vomiting, diarrhea), flushing, loss of consciousness, and low blood pressure.
162 All reactions were graded I – IV based on a modified Mueller anaphylaxis scale (17).

163 Venom skin testing was recorded. For both skin prick and intradermal testing,
164 wheal and flare in millimeters were recorded. For blood testing, total IgE and individual
165 venom IgE levels were recorded. Sensitivity to a venom was recorded as a positive skin
166 or blood test.

167 Venom immunotherapy characteristics was recorded, including the time to
168 maintenance, time on maintenance, total reactions, large local reactions and systemic
169 reactions (diagnosed by the treating physician).

170 Subsequent venom reactions in the field after starting VIT were recorded
171 identically as the original venom reaction. Tryptase laboratory order status, draw status,
172 date, and the value were recorded. If a bone marrow biopsy occurred, the pathologic

173 presence of mast cell disease (MCD), subtype, and number of major and minor criteria
174 for mastocytosis were recorded (18, 19).

175

176 **Statistical Analysis**

177 IBM SPSS version 22 (Armonk, NY) statistical software and GraphPad Prism
178 (San Diego, CA) were used to perform all statistical analysis. Potentially significant ($p <$
179 0.2) associations between patient characteristics and data of interest were initially
180 evaluated via bivariate correlation and chi-squared analysis. Linear or logistic
181 regression analysis was then performed as appropriate on all potentially significant
182 variables to create multivariate associations. A Cochran-Mantel-Haenszel (CMH) test
183 was used to compare prevalence of mastocytosis in patients with HVA compared to
184 those with mastocytosis without HVA in the database; this was stratified by adult vs
185 pediatric patients.

186 **Results**

187 **Database results**

188 The database query included 27,299,530 distinct patients in calendar year 2018.
189 This revealed a prevalence of 166.8 per 100,000 for HVA overall. Mastocytosis
190 prevalence was 10.1 per 100,000 overall and 96.7 per 100,000 (or 0.0967%) amongst
191 HVA patients in 2018; an odds ratio of 9.7 (95% CI 7.2 – 13.1, $p < 0.0001$) was noted
192 when comparing mastocytosis in patients with HVA compared to those without in 2018
193 (Figure 1). Among adult (age 18 years and older) patients, the odds ratio was 14.3 (95%
194 CI 10.5 – 19.6, $p < 0.0001$). Among pediatric (age < 18 years) patients, there was no
195 statistical significance (odds ratio 2.4 (95% CI 0.9 – 6.4, $p = 0.07$).

196

197 **Patient Characteristics**

198 Given the increased prevalence of mastocytosis in real world data, we
199 investigated the data from our allergy practice in patients with systemic reactions to
200 hymenoptera, undergoing VIT. University of Michigan is a referral center for MCD. Table
201 1 displays the demographic and disease-specific characteristics for the patients in the
202 University of Michigan cohort. The mean age was 47.6 and ranged from 7 – 81 years
203 old. 41% were female. 24% carried a diagnosis of asthma, 41% had a history of atopic
204 sensitization to a non-venom allergen. 8% had a family history of venom allergy.

205 The average Mueller anaphylaxis grade for the patients' original reactions was
206 2.90. This included 34% of patients with a documented low blood pressure and 26% of
207 patients who lost consciousness. Further, 66% experienced hives or rash, 61%
208 angioedema, 48% respiratory symptoms, and 12% gastrointestinal symptoms. 71% of

209 patients were sensitized to honeybee, 85% to yellow jacket, 68% to yellow hornet, 73%
210 to white-faced hornet, and 68% to wasp. During venom immunotherapy, 10%
211 experienced a systemic reaction.

212

213 **Impact of VIT on venom allergy course**

214 VIT was effective in this venom allergy population. Among these 161 patients
215 who began immunotherapy, 26 (16%) suffered a subsequent venom reaction (table 1).
216 The mean Mueller grade of these subsequent reactions was 0.96 (table 1), significantly
217 lower than the mean grade of initial reactions (Fig. 2A). This shows that venom
218 reactions after VIT were less frequent and less severe than before VIT started.

219 We sought to find variables associated with a subsequent venom reaction after
220 VIT initiation. Using logistic regression, the only significantly correlated variable was a
221 family history of a systemic venom reaction, with an odds ratio of 8.8 ($p = 0.049$) (table
222 2). 4% of patients who did not have subsequent venom reactions had a family history of
223 a systemic venom reaction, significantly fewer than the 25% rate in patients who did
224 have a subsequent venom reaction (Fig. 2B).

225

226 **Impact of guideline update on practice**

227 American practice guidelines published in January 2017 recommend measuring
228 tryptase levels for patients with cardiovascular compromise with venom reactions; these
229 guidelines suggest consideration of tryptase measurement for all patients with evidence
230 of venom anaphylaxis (3). We sought to evaluate the impact of these guidelines on
231 clinical practice in an academic allergy center. Among patients who did not lose

232 consciousness during the initial venom reaction, the rate of tryptase measurement was
233 52%, significantly lower than the 81% among patients who lost consciousness during
234 the initial reaction (Fig. 3A). Before 2017, the rate of tryptase measurement in VIT
235 patients was 59%; after 2017, the rate was 100%, significantly higher (Fig. 3B).

236

237 **Elevated tryptase and clonal MCD in VIT population**

238 We found 9 patients (5.6%) to have a baseline tryptase level greater than >11.5
239 ng/mL. Among these, 4 patients underwent bone marrow biopsy and 3 (1.8% of total)
240 had clonal MCD. Two of these had indolent SM and 1 had monoclonal mast cell
241 activation syndrome. These patients had significantly higher tryptase levels than
242 patients who had a tryptase measured but no known MCD (Fig. 4A).

243 We sought to evaluate whether tryptase levels correlates with other key features
244 of HVA. Among patients who had a tryptase level, tryptase did not correlate with the
245 initial reaction Mueller anaphylaxis grade (Fig. 4B). Elevated tryptase levels appeared in
246 patients throughout the grading scale. We also sought whether tryptase or other
247 variables might correlate with a low blood pressure, an indicator of cardiovascular
248 compromise, during the initial reaction. On logistic regression the only significantly
249 correlated variables were loss of consciousness, with an odds ratio of 12.9 ($p < 0.0001$),
250 and having a tryptase drawn, with an OR of 2.6 ($p = 0.039$) (table 2). Tryptase level,
251 whether analyzed as a continuous variable or when broken into discrete groups of 0 – 5
252 ng/mL, 5 – 11.4 ng/mL, and over 11.4 ng/mL, did not correlate with low blood pressure
253 during the initial reaction. However, tryptase levels divided according to the same levels
254 did correlate with loss of consciousness (LOC). Patients with tryptase levels between 5

255 – 11.4 ng/mL had the highest rate of LOC at 50%, significantly higher than the rate of
256 11% in those with a tryptase greater than 11.4 ng/mL (Fig. 4C).

257 Because systemic reactions during VIT are a key adverse effect and worse
258 reactions have been correlated with higher tryptase levels previously (9), we evaluated
259 whether this was true in our population. We found a significantly higher mean tryptase
260 level among patients who had a systemic VIT reaction versus patients who did not (Fig.
261 4D). Furthermore, to find variables correlated with having a systemic VIT reaction, we
262 used a multivariate logistic regression that involved all bivariate associations where p
263 was less than 0.2; the only significantly correlated variable was tryptase level, with an
264 odds ratio of 1.3 ($p = 0.027$) per unit increase in tryptase level (table 2). Notably, beta
265 blocker and ACE-inhibitor use did not correlate with the presence, number, or severity
266 of a systemic VIT reaction.

267 Discussion

268 VIT is an effective preventative therapy for HVA (3, 5). US guidelines recommend
269 considering a baseline tryptase level for all HVA patients undergoing VIT and outright
270 recommend to check in severe venom anaphylaxis (3). Given a high rate of MCD
271 amongst patients with HVA, up to 7-11%, some authors suggest measuring tryptase
272 values in all VIT patients (6, 7, 20). SM is associated with failure of VIT, prompting the
273 noted increased vigilance (10). Bonadonna et al found that among HVA patients, 11.6%
274 had tryptase levels >11.4 and clonal MCD was detected in 90.9% of patients with
275 elevated tryptase who underwent further evaluation; at a minimum, 5.5% of the total (a
276 rate of 5,500 per 100,000) were diagnosed with systemic mastocytosis (7). Further, one
277 study suggests that patients with severe hypotension and normal basal tryptase values
278 have rates of MCD up to 75% (21). However, a recent publication from Israel
279 demonstrated a MCD rate of 3.8% (or 3,800 per 100,000) amongst HVA patients (13),
280 lower than the figures cited in European data. The rate, to our knowledge, has not been
281 reported in a US population. In this work, we report a MCD rate among HVA patients of
282 0.097% in the US population. While this rate cannot be statistically compared directly to
283 the European or Israeli reports (7, 13) because the current estimate encompasses a
284 data set of millions versus prior reports involving hundreds patients, this reported rate is
285 qualitatively lower than prior reports by over an order of magnitude. One explanation for
286 the higher previously reported rate could be that prior studies occurred mainly within
287 referral centers for mastocytosis and may have carried a selection bias.

288 Prior work suggests 56-94% of adults report at least one lifetime hymenoptera
289 sting (22), and the rate of systemic reactions among those who are stung is 0.5-3.3% in

290 the United States (23) and 0.3-7.5% in Europe (5). We report a one-year prevalence of
291 0.167% in 2018 in a US cohort, lower than prior reports. Another explanation could be
292 that our data includes subjects with 6-12 months of data, which might lower the rate of
293 HVA detected here, as some patients may not have had claims data during this period.
294 This is a limitation shared with other claims database-based analyses. Furthermore, the
295 database used in this study does not necessarily include non-privately insured patients,
296 so the population may not fully represent the entire United States population, thereby
297 affecting prevalence. The rate of systemic reactions to insects in the US and Europe are
298 similar, suggesting similar rates of hymenoptera allergy; this could change with
299 variances in insect species' distribution. For example, climate change may promote
300 habitats favorable to invasive insects (24); as the number of insect stings changes by
301 region the incidence of HVA may also change as recurrent stings have been described
302 as a risk factor for HVA (23). Another factor that may affect the rate of MCD within the
303 HVA population is the total MCD burden by population; the prevalence of mastocytosis
304 in the general population is estimated to be 3 to 13 per 100,000 inhabitants (25). Our
305 finding of 7.7 per 100,000 falls within the previously reported range. Thus, there might
306 be a disproportionate change in the incidence of HVA relative to the incidence of mast
307 related diseases.

308 Another consideration is the etiology of an elevated tryptase. It has been
309 estimated that 4-6% of the population has an elevated tryptase (12). Most of these
310 patients do not have a clonal MCD. Hereditary alpha-tryptasemia (HAT) is a recently
311 described autosomal dominant trait caused by increased monoallelic α -tryptase copy
312 number at *TPSAB1* (12). Much of the prior work examining elevated serum tryptase in

313 HVA patients was performed before the recognition of HAT as a distinct entity, which
314 may have impacted mast cell disease estimates at that time. It has only been recently
315 that role of HAT in HVA has been examined. Cohort studies have suggested HAT is
316 associated with more severe HVA, but may not affect the rate of HVA as the rate of
317 HAT in some HVA patient cohorts is similar to that of the general population (26, 27).
318 HAT may be overrepresented in patients with clonal MCD and these may have a
319 synergistic effect on severity of anaphylaxis (26, 27). If HAT does affect anaphylaxis
320 severity this may explain why some patients with MCDs have more severe HVA versus
321 others and why elevated serum tryptase without clonal MCD has been at times
322 associated with HVA severity. The role of HAT in HVA is an area that requires further
323 investigation.

324 Race is increasingly recognized as a factor in many atopic diseases (28). It has
325 been implicated in not only outcomes disparities, but also in the immunology and
326 genetics of atopy. The US has a racially and ethnically diverse population. This
327 heterogeneity of this group could explain differences in MCD and HVA in the US
328 population compared to Europe. Role of ethnic background in mast cell related disease
329 and HVA are two areas that require further investigation.

330 The lower rate of MCD in this study may be due to various factors. The IBM
331 database relies on the proper coding of the diagnoses of hymenoptera allergy and
332 mastocytosis. The accuracy of coding can vary with literature suggesting rates of error
333 from 0-70% (29). This suggests that a significant number of HVA patients are not
334 screened for MCD. Another limitation is the method diagnosis for HVA, as the initial
335 reaction and allergy testing is not reported. Because up to 25% of adults may have

336 hymenoptera sensitization (30), it is possible that patients with non-anaphylactic insect
337 sting reactions and sensitization to hymenoptera were mislabeled as HVA.

338 The role of baseline tryptase in the course of VIT has been explored. Higher
339 basal tryptase levels have been proposed to predict a high risk of VIT side effects,
340 particularly during build-up (9). Elevated basal tryptase has also been associated with
341 severe sting reactions (8, 31, 32), including hypotension and fatality (33). In this work,
342 the tryptase level was not correlated with the severity of the initial reaction, though this
343 study involves only those on VIT, which may be a confounding factor. In fact, patients
344 with the tryptase levels over 11.4 ng/mL had less frequent loss of consciousness than
345 those with lower tryptase levels (Fig 4C). We do note that in patients with known
346 mastocytosis, patients with higher basal serum tryptase (e.g. 40 ng/ml or above) are
347 less likely to have anaphylaxis than those with mild to moderate elevations (34), so
348 perhaps a similar effect is occurring here in this broader population; more work would
349 be needed to address this further (11). As our cohort specifically examined VIT patients,
350 we cannot assess whether tryptase levels correlate with severity of initial reactions
351 among patients with anaphylactic reactions versus patients with non-anaphylactic
352 reactions (as that group does not typically undergo VIT). An elevated baseline tryptase
353 was associated here with systemic reactions to VIT, consistent with prior published
354 work.

355 Allergists may have increased suspicion of underlying mastocytosis when
356 particular characteristics of the sting reaction are present, such as hypotension without
357 hives, especially in males (35). This was reflected in our data, as the treating allergist
358 was more likely to draw a tryptase level in patients who presented with a low blood

359 pressure (Fig. 3A). Furthermore, the guideline changes in the US in 2017 (3) appeared
360 to impact practice patterns, as all VIT patients started after 2017 had a tryptase level
361 drawn, compared with before 2017 (Fig. 3B).

362 Prior authors have suggested no association between anaphylaxis severity and
363 comorbidities or cardiovascular medications (20, 36). Our data were supportive of this
364 as well; beta blocker use, ACE-inhibitor use, and cardiovascular disease were not
365 associated with initial reaction severity, subsequent reaction rate or severity, nor with
366 the rate of systemic reactions while patients were on VIT.

367 Prior literature has suggested an association between HVA and MCD based on
368 several observations. The prevalence of HVA in SM is higher than the general
369 population and HVA represents the most common anaphylaxis trigger in adult
370 mastocytosis patients; there is also more frequent clonal MCD in patients with systemic
371 HVA than the general population (37). This study supports a relationship between HVA
372 and mastocytosis with the prevalence of mastocytosis among patients with HVA being
373 12 fold higher than among the general population.

374 In conclusion, mastocytosis may be less common in the US population compared
375 to European reports with systemic venom reactions, but a strong association remains
376 between HVA and mastocytosis. In this population of VIT patients, serum tryptase
377 values do not correlate with severity of venom reactions; indeed, higher levels may
378 correlate with less frequent loss of consciousness. Baseline serum tryptase elevation
379 does correlate with more frequent systemic VIT reactions. Beta blocker and ACE-
380 inhibitor use in this population do not correlate with the frequency or severity of venom
381 or VIT reactions. Overall, these data suggest that while baseline serum tryptase may

382 help identify MCD patients amongst the HVA population and help predict systemic
383 reactions to VIT, the MCD rate may be lower in this US population than other
384 populations.

385

386 **Acknowledgements**

387 The authors acknowledge the patients who contributed data for this work and the
388 University of Michigan Allergy Clinic staff who perform venom immunotherapy and
389 clinical care with the excellence and consistency that allows this sort of work to proceed.

390 **References**

- 391 1. Bilo MB, Bonifazi F. The natural history and epidemiology of insect venom allergy: clinical
392 implications. *Clin Exp Allergy*. 2009;39(10):1467-76.
- 393 2. Golden DB, Marsh DG, Kagey-Sobotka A, Freidhoff L, Szkló M, Valentine MD, et al. Epidemiology
394 of insect venom sensitivity. *JAMA*. 1989;262(2):240-4.
- 395 3. Golden DB, Demain J, Freeman T, Graft D, Tankersley M, Tracy J, et al. Stinging insect
396 hypersensitivity: A practice parameter update 2016. *Ann Allergy Asthma Immunol*. 2017;118(1):28-54.
- 397 4. Hunt KJ, Valentine MD, Sobotka AK, Benton AW, Amodio FJ, Lichtenstein LM. A controlled trial
398 of immunotherapy in insect hypersensitivity. *N Engl J Med*. 1978;299(4):157-61.
- 399 5. Sturm GJ, Varga EM, Roberts G, Mosbech H, Bilo MB, Akdis CA, et al. EAACI guidelines on
400 allergen immunotherapy: Hymenoptera venom allergy. *Allergy*. 2018;73(4):744-64.
- 401 6. Bonadonna P, Zanotti R, Pagani M, Caruso B, Perbellini O, Colarossi S, et al. How much specific is
402 the association between hymenoptera venom allergy and mastocytosis? *Allergy*. 2009;64(9):1379-82.
- 403 7. Bonadonna P, Perbellini O, Passalacqua G, Caruso B, Colarossi S, Dal Fior D, et al. Clonal mast cell
404 disorders in patients with systemic reactions to Hymenoptera stings and increased serum tryptase
405 levels. *J Allergy Clin Immunol*. 2009;123(3):680-6.
- 406 8. Blum S, Gunzinger A, Muller UR, Helbling A. Influence of total and specific IgE, serum tryptase,
407 and age on severity of allergic reactions to Hymenoptera stings. *Allergy*. 2011;66(2):222-8.
- 408 9. Rueff F, Przybilla B, Bilo MB, Muller U, Scheipl F, Aberer W, et al. Predictors of side effects during
409 the buildup phase of venom immunotherapy for Hymenoptera venom allergy: the importance of
410 baseline serum tryptase. *J Allergy Clin Immunol*. 2010;126(1):105-11 e5.
- 411 10. Gonzalez de Olano D, Alvarez-Twose I, Esteban-Lopez MI, Sanchez-Munoz L, de Durana MD,
412 Vega A, et al. Safety and effectiveness of immunotherapy in patients with indolent systemic
413 mastocytosis presenting with Hymenoptera venom anaphylaxis. *J Allergy Clin Immunol*.
414 2008;121(2):519-26.
- 415 11. van Anrooij B, van der Veer E, de Monchy JG, van der Heide S, Kluin-Nelemans JC, van Voorst
416 Vader PC, et al. Higher mast cell load decreases the risk of Hymenoptera venom-induced anaphylaxis in
417 patients with mastocytosis. *J Allergy Clin Immunol*. 2013;132(1):125-30.
- 418 12. Lyons JJ, Yu X, Hughes JD, Le QT, Jamil A, Bai Y, et al. Elevated basal serum tryptase identifies a
419 multisystem disorder associated with increased TPSAB1 copy number. *Nat Genet*. 2016;48(12):1564-9.
- 420 13. Hershko AY, Shaharabani N, Rosman Y, Meir-Shafir K, Lachover-Roth I, Cohen Engler A, et al.
421 Hymenoptera venom allergy in a single-center Israeli cohort: Clinical and laboratory characteristics. *J*
422 *Allergy Clin Immunol Pract*. 2019.
- 423 14. Golden DB, Moffitt J, Nicklas RA, Freeman T, Graft DF, Reisman RE, et al. Stinging insect
424 hypersensitivity: a practice parameter update 2011. *J Allergy Clin Immunol*. 2011;127(4):852-4 e1-23.
- 425 15. Valent P, Akin C, Metcalfe DD. Mastocytosis: 2016 updated WHO classification and novel
426 emerging treatment concepts. *Blood*. 2017;129(11):1420-7.
- 427 16. Bernstein IL, Li JT, Bernstein DI, Hamilton R, Spector SL, Tan R, et al. Allergy diagnostic testing: an
428 updated practice parameter. *Ann Allergy Asthma Immunol*. 2008;100(3 Suppl 3):S1-148.
- 429 17. Mueller HL. Diagnosis and treatment of insect sensitivity. *J Asthma Res*. 1966;3(4):331-3.
- 430 18. Volertas S, Schuler CFT, Akin C. New Insights into Clonal Mast Cell Disorders Including
431 Mastocytosis. *Immunol Allergy Clin North Am*. 2018;38(3):341-50.
- 432 19. Valent P, Akin C, Hartmann K, Nilsson G, Reiter A, Hermine O, et al. Advances in the
433 Classification and Treatment of Mastocytosis: Current Status and Outlook toward the Future. *Cancer*
434 *Res*. 2017;77(6):1261-70.

- 435 20. Stoevesandt J, Hain J, Kerstan A, Trautmann A. Over- and underestimated parameters in severe
436 Hymenoptera venom-induced anaphylaxis: cardiovascular medication and absence of
437 urticaria/angioedema. *J Allergy Clin Immunol.* 2012;130(3):698-704 e1.
- 438 21. Zanotti R, Lombardo C, Passalacqua G, Caimmi C, Bonifacio M, De Matteis G, et al. Clonal mast
439 cell disorders in patients with severe Hymenoptera venom allergy and normal serum tryptase levels. *J*
440 *Allergy Clin Immunol.* 2015;136(1):135-9.
- 441 22. Bilo BM, Bonifazi F. Epidemiology of insect-venom anaphylaxis. *Curr Opin Allergy Clin Immunol.*
442 2008;8(4):330-7.
- 443 23. Golden DB. Anaphylaxis to insect stings. *Immunol Allergy Clin North Am.* 2015;35(2):287-302.
- 444 24. Vega A, Castro L. Impact of climate change on insect-human interactions. *Curr Opin Allergy Clin*
445 *Immunol.* 2019;19(5):475-81.
- 446 25. Brockow K. Epidemiology, prognosis, and risk factors in mastocytosis. *Immunol Allergy Clin*
447 *North Am.* 2014;34(2):283-95.
- 448 26. Lyons JJ, Chovanec J, O'Connell MP, Liu Y, Selb J, Zanotti R, et al. Heritable risk for severe
449 anaphylaxis associated with increased alpha-tryptase-encoding germline copy number at TPSAB1. *J*
450 *Allergy Clin Immunol.* 2020.
- 451 27. O'Connell MP, Lyons JJ. Hymenoptera venom-induced anaphylaxis and hereditary alpha-
452 tryptasemia. *Curr Opin Allergy Clin Immunol.* 2020;20(5):431-7.
- 453 28. Castells M. Race and allergy: Are we that different? *Ann Allergy Asthma Immunol.*
454 2019;122(5):439-40.
- 455 29. O'Malley KJ, Cook KF, Price MD, Wildes KR, Hurdle JF, Ashton CM. Measuring diagnoses: ICD
456 code accuracy. *Health Serv Res.* 2005;40(5 Pt 2):1620-39.
- 457 30. Abrams EM, Golden DBK. Approach to Patients with Stinging Insect Allergy. *Med Clin North Am.*
458 2020;104(1):129-43.
- 459 31. Rueff F, Przybilla B, Bilo MB, Muller U, Scheipl F, Aberer W, et al. Predictors of severe systemic
460 anaphylactic reactions in patients with Hymenoptera venom allergy: importance of baseline serum
461 tryptase-a study of the European Academy of Allergology and Clinical Immunology Interest Group on
462 Insect Venom Hypersensitivity. *J Allergy Clin Immunol.* 2009;124(5):1047-54.
- 463 32. Ludolph-Hauser D, Rueff F, Fries C, Schopf P, Przybilla B. Constitutively raised serum
464 concentrations of mast-cell tryptase and severe anaphylactic reactions to Hymenoptera stings. *Lancet.*
465 2001;357(9253):361-2.
- 466 33. Haeberli G, Bronnimann M, Hunziker T, Muller U. Elevated basal serum tryptase and
467 hymenoptera venom allergy: relation to severity of sting reactions and to safety and efficacy of venom
468 immunotherapy. *Clin Exp Allergy.* 2003;33(9):1216-20.
- 469 34. Gulen T, Ljung C, Nilsson G, Akin C. Risk Factor Analysis of Anaphylactic Reactions in Patients
470 With Systemic Mastocytosis. *J Allergy Clin Immunol Pract.* 2017;5(5):1248-55.
- 471 35. Golden DBK, Carter MC. Insect Sting Anaphylaxis-Or Mastocytosis-Or Something Else? *J Allergy*
472 *Clin Immunol Pract.* 2019;7(4):1117-23.
- 473 36. Smith MA, Newton LP, Barcena Blanch MA, Cuervo-Pardo L, Cho L, Newton D, et al. Risk for
474 Anaphylactic Reaction From Cardiac Catheterization in Patients Receiving beta-Adrenergic Blockers or
475 Angiotensin-Converting Enzyme-Inhibitors. *J Allergy Clin Immunol Pract.* 2019.
- 476 37. Bonadonna P, Scaffidi L, Boni E. Tryptase values in anaphylaxis and insect allergy. *Curr Opin*
477 *Allergy Clin Immunol.* 2019;19(5):462-7.

478

479

480 Figure Legends

481 **Figure 1:** In 2018, the prevalence of Mastocytosis among patients with hymenoptera
482 allergy was 9.7 times more than the prevalence among the US general population.

483 Note: OR = odds ratio, 95% confidence interval listed in parentheses.

484

485 **Figure 2:** A) Mueller grade of initial reaction (mean 2.90, n=158) compared to Mueller
486 grade of any subsequent reactions (mean 0.96, n=23). B) The proportion of patients
487 with a family history of a systemic venom reaction is stratified according to whether the
488 patient did (4%, n=126) or did not (25%, n=25) have a subsequent venom reaction after
489 a VIT course. Data represent mean +/- standard error of the mean (SEM). *** $p < 0.001$,
490 **** $p < 0.0001$.

491

492 **Figure 3:** A) Rate of tryptase measurement stratified by blood pressure drop presence
493 (52%, n=104) or absence (81%, n=54) at initial reaction. B) Rate of tryptase
494 measurement rate stratified by date, before 2017 (59%, n=150) or after the start of 2017
495 (100%, n=11). Data represent mean +/- standard error of the mean (SEM). ** $p < 0.01$,
496 *** $p < 0.001$.

497

498 **Figure 4:** A) Tryptase level stratified by whether a MCD was present (mean 18.0, n = 3)
499 or not present (mean 5.0, n = 93). B) Tryptase levels are plotted according to the
500 Mueller grade of the patient's initial venom reaction. Mean tryptase: Grade 1 = 7.0
501 (n=4), Grade 2 = 6.0 (n=15), Grade 3 = 5.2 (n=41), Grade 4 = 5.1 (n=34). C) The
502 proportion of patients with loss of consciousness (LOC) is plotted based on the tryptase

503 level, with ranges of < 5 ng/mL (34%, n=62), 5 – 11.4 ng/mL (50%, n=24), and > 11.4
 504 ng/mL (11%, n=9). D) Tryptase levels are plotted for patients who did (mean 8.8, n=7)
 505 or did not (mean 5.1, n=73) have systemic reactions during venom immunotherapy
 506 (VIT). Data represent mean +/- SEM. *p < 0.05, **p < 0.01, ****p < 0.0001.

507

508 Tables

Patient characteristic	All patients (n=161)	Adults (n=147)	Children (n=14)
Demographic			
Age (mean years [range])	47.6 (7-81)	50.8 (18-81)	13.2 (7-17)
Gender (n [%] female)	66 (41)	65 (44)	1 (7)
General disease history			
History of asthma (n [%])	39 (24)	35 (24)	4 (29)
History of other atopic sensitization (n [%])	66 (41)	63 (43)	3 (21)
History of atopic dermatitis (n [%])	15 (9)	10 (7)	5 (36)
History of food allergy (n [%])	8 (5)	7 (5)	1 (7)
Family history of venom allergy (n [%])	13 (8)	9 (6)	4 (29)
Beta blocker use (n [%])	7 (4)	7 (5)	0 (0)
ACE inhibitor use (n [%])	9 (6)	9 (6)	0 (0)
Original venom reaction			
Hives or rash (n [%])	106 (66)	96 (65)	10 (71)
Angioedema (n [%])	99 (61)	88 (60)	11 (79)
Respiratory symptoms (n [%])	78 (48)	71 (48)	8 (57)
Gastrointestinal symptoms (n [%])	19 (12)	15 (10)	4 (29)
Low blood pressure documented (n [%])	54 (34)	53 (36)	1 (7)
Loss of consciousness (n [%])	42 (26)	41 (28)	1 (7)
Mueller anaphylaxis grade (mean [SD])	2.90 (0.88)	2.92 (0.90)	2.71 (0.61)
Venom sensitization			
Honeybee (n [%])	115 (71)	107 (73)	8 (57)
Yellow jacket (n [%])	137 (85)	127 (86)	10 (71)
Yellow hornet (n [%])	110 (68)	101 (69)	9 (64)
White-faced hornet (n [%])	117 (73)	107 (73)	10 (71)
Wasp (n [%])	110 (68)	101 (69)	9 (64)
Venom IT course			
Systemic reaction to IT (n [%])	16 (10)	15 (10)	1 (7)
Subsequent venom reaction (n [%])	24 (15)	18 (12)	6 (43)
Subsequent venom reaction Mueller grade (mean [SD])	0.96 (0.98)	1.13 (0.97)	0.57 (0.98)
Tryptase measurement			

Tryptase drawn (n [%])	99 (61)	91 (62)	8 (57)
Tryptase value (mean ng/mL [range])	5.4 (1.5 - 20.9)	5.5 (1.5 - 20.9)	4.4 (2.3 - 8.4)
MCD (% all patients [% if tryptase measured])	1.9 (2.6)	2.0 (3.2)	0 (0)

509

510 **Table 1:** The baseline characteristics of the patient population are listed in aggregate
 511 and by adults (age 18 and up) and children (age < 18). SD = standard deviation. Mueller
 512 grading delineated in Methods section (17).

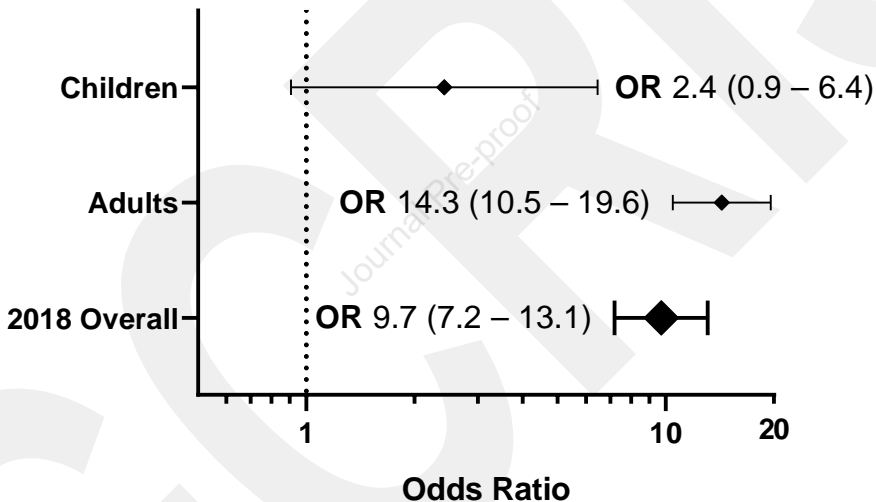
513

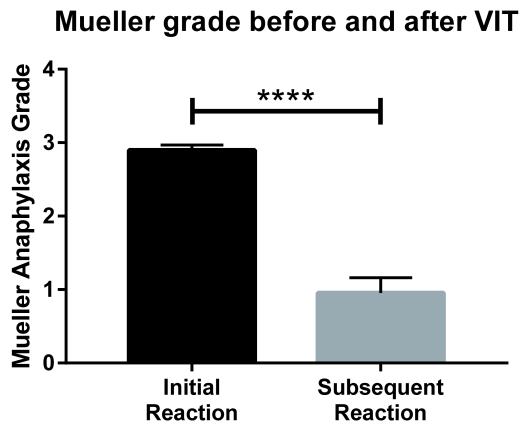
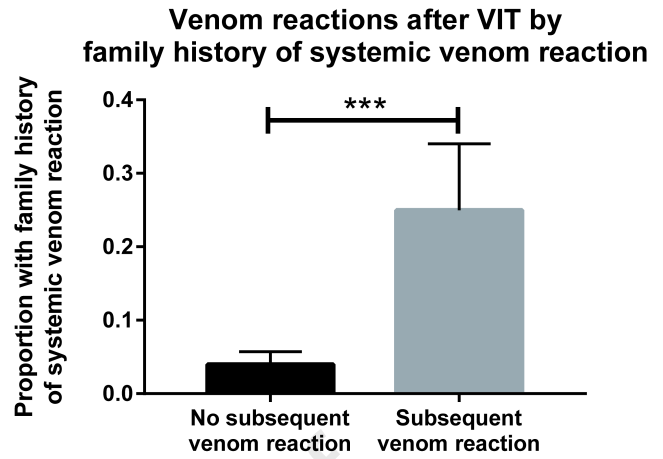
Dependent variable	Independent variables	Odds Ratio	p - value
Systemic IT reaction	Tryptase	1.3	0.027
Low blood pressure on first reaction	LOC on first reaction	12.9	<0.001
	Tryptase drawn	2.6	0.039
Subsequent venom reaction	Family history of systemic venom reaction	8.8	0.049

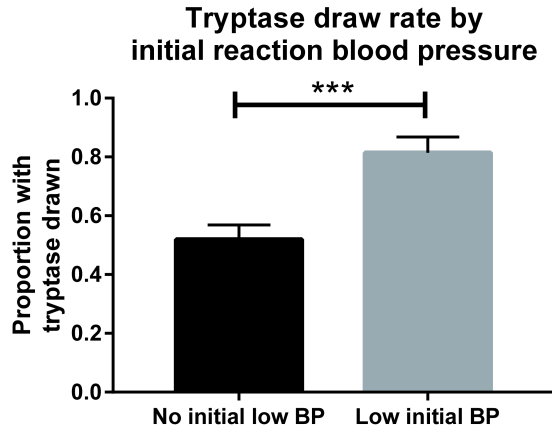
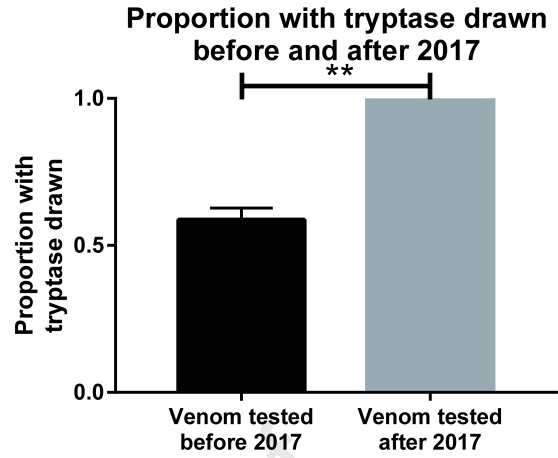
514

515 **Table 2:** Results of multivariate logistic regressions.

Mastocytosis in HVA



**A****B**

**A****B**

Journal Pre-proof

