Tryptase: From Anaphylaxis to Mastocytosis

New Concepts in Mast Cell Mediators
WAO2011
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Virginia Commonwealth University

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Clinical Vignettes: Can a biomarker of mast cell involvement be clinically helpful?

56 y/o stung by an insect, underlying HBP (HCTZ, lisinopril), c/o dizziness, dyspnea and chest pain. ER: MI

24 y/o to OR for elective cholecystectomy, PCN allergy hx. During anesthesia induction: BP ↓ 120/60 to 60/30 & P ↑ 75 to 120, improved over ~30 min with iv fluids & epinephrine.

50 y/o male with osteoporosis, vertebral fx & flushing spells. When 20 y/o systemic anaphylaxis to wasp sting.

35 y/o M with prior urticaria response after an insect sting. DM, enalapril. Likelihood of systemic anaphylactic shock to a future insect sting?
**Definition of Systemic Anaphylaxis**

Systemic anaphylaxis is a form of immediate hypersensitivity arising when mast cells and/or basophils are provoked to secrete mediators with potent vasoactive and smooth muscle contractile activities that evoke a systemic response.

**Working Diagnosis of Anaphylaxis**

I. Acute onset of illness w/o apparent allergen involving:
   - Skin or Mucosa
     - Pruritis
     - Hives
     - Angioedema
   - Respiratory Compromise
     - Dyspnea
     - Wheeze
   - Hypotension or End Organ-System Dysfunction
     - Collapse
     - Syncope
     - Incontinence
   - Gastrointestinal Symptoms
     - Vomiting
     - Crampy abdominal pain
     - Diarrhea

II. Rapid onset after exposure to a likely allergen of ≥2 of the following:
   - Skin or Mucosa
   - Respiratory compromise
   - Hypotension

III. Rapid onset after exposure to a known allergen:
   - Hypotension or End Organ-System Dysfunction

**Differential Diagnosis of Systemic Anaphylaxis**

- Pulmonary/Cardiogenic Shock
- Flushing disorders (carcinoid syndrome, VIPoma)
- Vasovagal, Panic attacks, Vocal cord dysfunction
- Hereditary/Acquired Angioedema (bradykinin)
- Contact system activation (bradykinin, CH50 contaminant)
- Complement activation (C3a & C5a)
- Scombroidosis (histamine)
- Other shock syndromes (septic)
- Systemic mastocytosis (anaphylaxis)
Can a laboratory test provide objectivity to the clinical diagnosis of systemic anaphylaxis?

Resting Mast Cell | Activated Mast Cell

Preformed Granule Mediators:
- histamine, heparin, **tryptase**, chymase, carboxypeptidase A3

Newly-Generated Lipids, Cytokines, Chemokines:
- PGD₂, LTC₄, PAF (PAF acetyl hydrolase), S1P, IL-4/13

Degranulation: Externalization of Secretory Granule Contents

Mature Tryptase

Heparin

Protryptase(s)
Two Key Differences Between α- & β- Tryptases

<table>
<thead>
<tr>
<th>Processing</th>
<th>Catalysis</th>
</tr>
</thead>
<tbody>
<tr>
<td>α</td>
<td>α</td>
</tr>
<tr>
<td>β</td>
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</table>


Imunoassays for Total (pro + mature) & Mature Tryptases

Total Tryptase

- G4 mAb
- mature + pro
- B12 mAb

Mature Tryptase

- G5 mAb
- mature + pro
- B12 mAb

Phadia ImmunoCAP

VCU: S-Lab

Mature Tryptase & Histamine Levels in Plasma During Insect Sting-Induced Systemic Anaphylaxis

J Clin Invest 83:1551, 1989
### β-Tryptase Levels in Serum

**During Systemic Anaphylaxis from an Insect Sting**

5 min after Symptom Onset

\[ \Delta \log(\text{TRY}) = 0.2 - 0.03 \Delta \text{MAP}; r=0.86 \]

<table>
<thead>
<tr>
<th>2 ng/ml</th>
<th>-25</th>
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<tbody>
<tr>
<td>100</td>
<td>-57</td>
</tr>
<tr>
<td>1000</td>
<td>-89</td>
</tr>
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</table>

### Fatal Anaphylaxis

1. Local mast cell-mediated angioedema (laryngeal).
2. Mast cells with less tryptase (MC\(_v\) v MC\(_C\)).
3. Mast cells further from circulation (mucosal v perivascular).
4. Early (mast cell) v late (basophil/eosinophil) phase.
Serum Total Tryptase Levels Before → 60 min after Insect Sting: J Clin Immunol 14:190-204, 1994

Characteristics of the Total Tryptase & Mature Tryptase Immunoassays (ng/ml)

<table>
<thead>
<tr>
<th>Tryptase Type</th>
<th>Mature Tryptase</th>
<th>Total Tryptase</th>
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<tbody>
<tr>
<td>Normal Serum Baseline</td>
<td>&lt;1</td>
<td>1 – 15 (11.4)</td>
</tr>
<tr>
<td>Systemic Anaphylaxis (acute)</td>
<td>&gt;1</td>
<td>↑</td>
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Case 1

56 y/o stung by an insect, underlying HBP (HCTZ, lisinopril), c/o dizziness, dyspnea and chest pain. ER: MI

Acute:
- EKG: Inferior MI
- Troponin: elevated
- Tryptase: mature=6 ng/ml; total=15 ng/ml
- Venom IgE skin test: negative

Baseline (1 month later):
- Tryptase: mature tryptase <1; total tryptase =5
- Venom IgE skin test: positive

*Acute elevations in mature and total tryptase = diagnosis of systemic anaphylaxis to venom, which precipitated the MI.*
Case 2

24 y/o to OR for elective cholecystectomy with PCN allergy hx. Received fentanyl, lidocaine, midazolam, propofol, vancomycin, rocuronium prior to surgery → BP(P) from 120/60 (75) to 60/30(120), improved over ~30 min with iv fluids & epinephrine and procedure cancelled.

Acute trypatase: total = 13; mature =5.3 Allergy skin test negative to lidocaine, rocuronium, propofol Vancomycin (1g) infused over 5 min

*High peak [vancomycin] directly activates mast cells

~severe Red-Man Syndrome

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**Does the serum total tryptase level reflect:**

(1) The burden of mast cells in tissues &
(2) The effect of cytoreductive therapy?

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<tr>
<td>Systemic Mastocytosis (non-acute)</td>
<td>±↑</td>
<td>≥20* RATIO: Total/Mature &gt; 20</td>
</tr>
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*WHO minor criterion
Diagnosis of Systemic Mastocytosis

<table>
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<tr>
<th>Major Criterion</th>
<th>MC Granulomas (BM; &gt;15 MC)</th>
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<tbody>
<tr>
<td>Minor Criteria</td>
<td></td>
</tr>
<tr>
<td>1. Abnormal MC morphology (&gt;25% spindle-shaped)</td>
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<tr>
<td>2. Activating KIT mutation (e.g., D816V)</td>
<td></td>
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<tr>
<td>3. CD25⁺ or CD2⁺ MC</td>
<td></td>
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<tr>
<td>4. Baseline serum total tryptase &gt;20 ng/ml (≥11.4 insect sting anaphylaxis)</td>
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Diagnosis

1 major + 1 minor
≥3 minor

Systemic Mastocytosis

Urticaria Pigmentosa Bone Marrow

Differential Diagnosis of Elevated Total Tryptase Level in Serum

1. Systemic mastocytosis
2. Mast Cell Activation Syndrome
3. Hypereosinophilic syndrome: FIP1L1-PDGFRα
4. Acute Myelocytic Leukemia (~30%)
5. Myelodysplastic syndromes
6. SCF administration
7. End-stage kidney disease
8. ?Normal variant
9. ?Transient mastocytosis
Case 3

50 y/o male with osteoporosis, vertebral fx & flushing spells. When 20 y/o systemic anaphylaxis to wasp sting.

Cortisol, PTH, TSH, VS, Pi, creat, Ca WNL.
Baseline serum tryptase: 29 ng/ml
BM Bx: MC granulomas, CD25+ spindle-shaped MCs

Osteoporosis/vertebral fx may be a presenting manifestation of systemic mastocytosis

Systemic mastocytosis:
osteoporosis (30%)[40% vertebral fx], osteosclerosis (10%)
Osteoporosis: 1-2.5% systemic mastocytosis

Is an elevated serum total tryptase level an indicator for risk of severe systemic anaphylaxis?
Mast cell clonality in patients with systemic reactions to insect stings & ↑ serum baseline total tryptase levels (sBT)


3-year prospective study → 44/379 (12%) systemic reactors
sBT >11.4 ng/mL
BM bx 30/34 (88%) → clonal mast cell disorder (D816V Kit):
systemic mastocytosis (21/34); MCAS (9/34)
What % with sBT>11.4 have clonal mast cells?

1. Consider mast cell clonality: insect sting SA & sBT > 11.4 → BM bx
2. 12% of systemic reactors → 88% mast cell clonality
   (Epidemiology: 0.8-5% incidence systemic reactions)
3. sBT >11.4 ng/mL → OR=6 severe anaphylactic reaction

Implications of Constitutively Activated D816V Kit Tyrosine Kinase

Functionally:
1. Primes mast cell activation
2. Increases mast cell survival
3. Increases mast cell accumulation

Practically:
1. Minor criterion for diagnosis of systemic mastocytosis.
2. Presence indicates mast cell clonality.
3. Anaphylaxis to insect venom stings & IT, ?other allergens
4. Predisposes to spontaneous/primary MCAS

Odds ratio for severe systemic anaphylaxis to insect sting ~ baseline serum total tryptase level

Ruëff et al. JACI 124:1047-54, 2009
Case 4

35 y/o M with prior urticaria response after an insect sting. DM, enalapril. Likelihood of systemic anaphylactic shock to a future insect sting?

<table>
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<th>Clinical Feature</th>
<th>OR</th>
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<tbody>
<tr>
<td>Male</td>
<td>1.7</td>
</tr>
<tr>
<td>ACE-inhibitor</td>
<td>2.2</td>
</tr>
<tr>
<td>Prior systemic reaction</td>
<td>4.7</td>
</tr>
<tr>
<td>Tryptase = 30</td>
<td>6.0</td>
</tr>
</tbody>
</table>

The risk for a severe anaphylactic reaction to a future insect sting is substantial; venom immunotherapy and an action plan (Trendelenburg/EpiPen) to a future sting are indicated.

Diagnosis of Mast Cell Activation Syndrome

1. Typical clinical signs and symptoms
2. Clinically significant increase in serum total tryptase:
   > (baseline + 20% of baseline + 2 ng/ml)*
   ≤ 4 h after onset
3. Response of clinical symptoms to HR1 ± HR2 blockers or cromolyn

   * 1.0 → 1.0 + 0.2 + 2 → >3.2 ng/ml
   10 → 10 + 2 + 2 → >14 ng/ml
   20 → 20 + 4 + 2 → >26 ng/ml


Concluding Comments

Levels of serum tryptase can reflect

1. Mast cell activation during anaphylaxis
2. Mast cell number (mastocytosis and M-HES)
3. Risk of anaphylaxis severity to insect stings and IT
...thereby providing diagnostic and therapeutic guidance.
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