Hereditary Angioedema

Joshua S. Jacobs, M.D

Allergy, Asthma and Immunology
Foundation of Northern California
February 9, 2013
Disclosures for:

Joshua Jacobs, MD

For the 12 months preceding this CME activity, I disclose the following types of financial relationships:

Honoraria received from: CSL Behring, Dyax Corporation, Shire HGT, ViroPharma

Consulted for: Dyax Corporation,

Held Common Stock in: None

Research, clinical trial, or drug study funds received from: Dyax Corporation, ViroPharma

I will be discussing products that are investigational or not labeled for use under discussion.
Hereditary Angioedema

Disease Epidemiology

- Rare, severely debilitating, life threatening, not linked to race or gender\(^1,2\)
- Prevalence estimated at 1:10,000 to 1:50,000\(^3\)
  - Multiple challenges with proper diagnosis\(^4\)
  - *Estimated that a large number remain undiagnosed*
  - *Delay in diagnosis: 13 to 21 years*\(^1\)
- Variable age of onset
  - Typically presents in childhood (2-3 years) or adolescence\(^5\)
  - Attacks begin in childhood, worsen in adolescence, and persist throughout life (with unpredictable severity)\(^5\)

History of HAE

Heinrich Quincke 1882

Sir William Osler 1888
Hereditary Angioedema

Disease Biology

- Inherited autosomal dominant trait\(^1\)
  - Genetic mutation causing a deficiency in C1-INH activity\(^1\)
  - Over 150 different mutations have been identified
  - Estimated that 25% of cases are new mutations\(^2\)
    - Type I – C1-INH deficiency (85%)\(^1\)
    - Type II – dysfunctional C1-INH (15%)\(^1\)
    - Type III – normal antigenic and functional C1-INH levels\(^1\)
      - Familial angioedema; indistinguishable except that
        majority of attacks
        are facial\(^1\)
- Considerable variation in the severity of HAE, even within a kindred\(^1\)

Inheritance and HAE

HAE Attacks
Variable and Heterogeneous

• Multiple precipitating factors\(^1,2\)
  – Stress, physical/emotional trauma, upper airway infection, GI disorders, oral contraceptives, menstruation/hormonal fluctuations, surgery (especially dental procedures)

• Acute attacks: unpredictable and can occur anywhere

• Swelling worsens slowly over the first 24 hours and gradually subsides in the next 48 to 72 hours\(^1\)

• Attacks may start in one location and then spread to another before resolving (typically lasting 5 days)\(^1\)

HAE Causes Disability
HAE: Abdominal Attacks


Courtesy of Dr. Marco Cicardi, personal archive.
HAE: Burden of Illness

The impact of HAE on patients’ work and activity functioning is comparable to impairment reported with severe asthma or Crohn’s disease.

Prodromal Symptoms

- Certain symptoms may accompany onset of an attack and could signal a need to begin treatment\(^1\)
- Significant variability in the expression, manifestation, prevalence, timing, and predictive reliability of prodromes\(^2\)

- Prodromal symptoms include\(^1\):
  - Mood changes
  - Abdominal discomfort
  - Nausea
  - Fatigue
  - Paresthesias
  - Decreased appetite
  - Hyperactivity
  - Mood changes
  - Thirst
  - Erythema marginatum-like nonpruritic rash
  - Irritability
  - Thirst
  - Malaise
  - Flu-like symptoms
  - Malaise

Laboratory Testing

<table>
<thead>
<tr>
<th></th>
<th>C4 Level</th>
<th>Antigenic C1-INH</th>
<th>Functional C1-INH</th>
<th>C1q Level</th>
<th>C3 Level</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Type I</strong></td>
<td>Low</td>
<td>Low</td>
<td>Low</td>
<td>Normal</td>
<td>Normal</td>
</tr>
<tr>
<td><strong>Type II</strong></td>
<td>Low</td>
<td>Normal or Elevated</td>
<td>Low</td>
<td>Normal</td>
<td>Normal</td>
</tr>
<tr>
<td><strong>Type III</strong></td>
<td>Normal</td>
<td>Normal</td>
<td>Normal</td>
<td>Normal</td>
<td>Normal</td>
</tr>
<tr>
<td><strong>Acquired C1INH Deficiency</strong></td>
<td>Low</td>
<td>Low</td>
<td>Low</td>
<td>Low</td>
<td>Normal or Low</td>
</tr>
<tr>
<td><strong>ACE-I Induced Angioedema</strong></td>
<td>Normal</td>
<td>Normal</td>
<td>Normal</td>
<td>Normal</td>
<td>Normal</td>
</tr>
<tr>
<td><strong>Allergic or Idiopathic Angioedema</strong></td>
<td>Normal</td>
<td>Normal</td>
<td>Normal</td>
<td>Normal</td>
<td>Normal</td>
</tr>
</tbody>
</table>

Diagnostic Algorithm for HAE

Adapted from Zuraw B
Idiopathic Angioedema

Facts on Histamine-Mediated (Allergic) Angioedema

• Often, but not always, associated with other signs and symptoms of histamine release
  – Urticaria, flushing, pruritus, bronchospasm, hypotension

• Usually increases over few hours, resolves in 24-48 hours

• May be caused by:
  – IgE-mediated reactions to allergen exposure
  – Non-IgE mast cell activation due to medications
  – Idiopathic mast cell activation
ACE-I Associated Angioedema

D. Winchester, Resident & Staff Physician. 2007;53(2);
Pathophysiology of ACE-I Associated Angioedema

Pregnancy and Attack Rate in Women With HAE

Only 6% of women had an attack of HAE within 48 hours of labor and delivery

C1 Esterase Inhibitor (C1-INH)

- Member of the serpin superfamily of proteases
- C1-INH regulates 3 proteolytic pathways that are activated during attacks, generating vasoactive substances
  - Coagulation (Contact System)
  - Fibrinolytic
  - Kallikrien-Kinin
- When C1-INH is limited, activation of other pathways depletes C1-INH leading to:
  - Inability to control bradykinin production
  - Excess bradykinin binding to bradykinin receptor (B2R) on endothelial cells causing angioedema
# Efficacy of On-Demand Therapies

<table>
<thead>
<tr>
<th>Drug</th>
<th>Dose</th>
<th>Treatment</th>
<th>Control</th>
<th>P value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Pd-C1-INH</td>
<td>20 U/kg</td>
<td>30 min</td>
<td>90 min</td>
<td>.0025</td>
</tr>
<tr>
<td>Ecallantide</td>
<td>30 mg</td>
<td>67 min</td>
<td>165 min</td>
<td>.08</td>
</tr>
</tbody>
</table>

Pd-C1-INH = plasma-derived C1 inhibitor
Rhucin is expressed in the mammary gland of transgenic rabbits. Rabbits were chosen because of ease of reproducibility and milk production.

The cloned human C1-INH gene was introduced into a mammary gland-specific expression vector under the control of an αS1 casein promoter. The construct was then introduced into fertilized New Zealand White rabbit oocytes by microinjection and a founder herd was generated that expressed high levels of secreted recombinant human C1-INH.

Each rabbit produced up to 10 l milk per annum from which Rhucin was produced, with average C1-INH levels of 12 mg/ml (as measured by ELISA).

Following collection and skimming of milk, Rhucin was purified by a process that included chromatography and filtration techniques, as well as screening for, and inactivation of, human and rabbit viruses. The purity was determined to be > 99% by sodium dodecylsulfate-polyacrylamide gel electrophoresis and > 99.95% by ELISA, which is a higher purity and more consistent than that of plasma-derived C1-INH (> 85%) and CE-1145 (approximately 95%). Batches were highly reproducible and N- and C-terminal sequence analysis of Rhucin and human plasma-derived C1-INH indicated that the two were identical.

Glycosylation patterns are necessary for correct protein formation, conformation and biological activity; therefore, disparity in these can affect efficacy, biodistribution and pharmacokinetics of the therapeutic. Rhucin consists of approximately 14% w/w carbohydrate when measured by mass spectroscopy, compared with 26% for human C1-INH; however, it was also reported that carbohydrate structures make up 21% of Rhucin and 28% of plasma-derived C1-INH. Monosaccharide analysis of Rhucin demonstrated that it contained fucose, galactose (Gal), N-acetyl-d-galactosamine, N-acetylgalactosamine (GlcNAc), mannose and N-acetyleneuraminic acid [789159]. Compared with plasma-derived C1-INH, the N-glycans of Rhucin were relatively undersialylated and its O-glycans oversialylated. Oligomannose-type glycosylation, sialylation and fucosylation decreased during lactation.

Longhurst H. Rhucin, a recombinant C1 inhibitor for the treatment of hereditary angioedema and cerebral ischemia. Current Opinion in Investigational Drugs 2008 9(3):310-323
rhC1INH for Acute HAE Attacks

rhC1INH Phase 3 Study

Phase III Study 1310

- **Study Design**
  - International, multicenter, randomized, placebo-controlled study
  - Single IV infusion of 50 U/kg rhC1INH vs. saline control
  - Primary endpoint time to beginning of symptom relief

- **Results:**
  - Statistically significant difference in time to beginning of symptom relief (n=75) between rhC1INH and placebo (p=0.031, log-rank test)
  - Median time to beginning of symptom relief was 90 minutes for rhC1INH patients (n=44) and 152 minutes for placebo patients (n=31)

- **Safety:**
  - Generally well tolerated and the frequency of patients experiencing at least one treatment emergent event was less than placebo
  - Thrombotic events, anaphylaxis or neutralizing antibodies to C1INH were not observed in any patient

www.pharming.com, accessed 1/10/2013
Nanofiltered C1 Inhibitor Concentrate for Treatment of Hereditary Angioedema

Results of Acute and Prophylactic Trial with nf-C1INH

Figure 1. Primary Outcome in the Trial of C1 Inhibitor Therapy for Acute Attacks of Angioedema.
Cumulative incidence estimates for the time to the onset of unequivocal relief (primary outcome) are shown for 35 subjects who received nanofiltered C1 inhibitor concentrate and 33 subjects who received placebo. The circles represent either subjects who received rescue therapy before 4 hours (competing events; 2 subjects in the placebo group received narcotic rescue at 15 and 146 minutes, respectively, and 1 subject in the C1 inhibitor group received open-label C1 inhibitor rescue at 110 minutes) or those who did not have an onset of unequivocal relief before 4 hours.

Figure 2. Normalized Rate of Angioedema Attacks during the Prophylaxis Trial.
The attack rates are shown for each of the 22 subjects during the 12-week period when either placebo or nanofiltered C1 inhibitor was being administered. Each pair of connected points represents the attack rates for a single subject during the two periods. The black horizontal lines indicate mean attack rates for the two treatments, which were 6.3 and 12.7 for the C1 inhibitor group and the placebo group, respectively.

Zuraw, et. al., NEJM, 2010, 363;6:513-522
Individual Results of Prophylactic Trial with nf-C1INH

Zuraw, et. al., NEJM, 2010, 363;6:513-522
Comparison of Efficacy Assessment Scales in Clinical Trials of Acute Therapy for HAE

<table>
<thead>
<tr>
<th>Agent</th>
<th>Study</th>
<th>Primary Endpoint/Timepoints</th>
</tr>
</thead>
<tbody>
<tr>
<td>C1 inhibitor concentrate (Berinert)</td>
<td>IMPACT1</td>
<td>• Onset of symptom relief (response to standardized question)</td>
</tr>
</tbody>
</table>
| C1 inhibitor concentrate (Cinryze)         | CHANGE 2  | • Time to unequivocal relief of symptoms at defining site (first of 3 consecutive reports of improvement)  
• Symptoms were assessed every 15 min posttreatment until unequivocal relief at the defining site                                                                                                                                                                                                                                               |
| C1 inhibitor concentrate, recombinant (Rhucin) | EDEMA 3   | • Time to beginning of relief of symptoms (first of 2 consecutive reports with VAS score reduced by ≥20 mm from baseline) at any eligible location  
• VAS was assessed at the time of evaluation (-1 h), at the start of infusion (baseline), 15 min, 30 min, and then at 1, 2, 4 h; consecutive assessment was preformed only if the patient was still hospitalized |
| Ecallantide (Kalbitor)                     | EDEMA 3/ 4| • TOS at 4 h posttreatment  
• Change in MSCS score at 4 h posttreatment                                                                                                                                                                                                                                                                                                           |
| Icatibant (Firyazyr)                       | FAST-1/2  | • Time to clinically significant symptom relief of index symptom (first of 3 consecutive reductions in VAS score of 20-30 mm depending on baseline severity)  
• VAS was assessed at 30-min intervals between 1 and 4 h, and then at 5, 6, 8, 10 and 12-15 h after study drug administration  
• Time to ≥50 % reduction symptom severity as defined by the VAS-3, the mean of VAS scores for cutaneous swelling, cutaneous pain, and abdominal pain  
• VAS was assessed before treatment, and then at 30-min intervals between 1 and 4 h and then at 5, 6, and 8-12 h after study drug administration |

SERPING1 Gene in genomic location

Start:  57,364,860 bp from pter
End:  57,382,326 bp from pter
Size:  17,467 bases
Orientation:  plus strand
Heterogeneity of Mutations in the C1NH Gene Causing C1Inh Deficiency and HAE in 59 Families

Distribution of disease-causing mutations in the SERPING1 gene in 25 Danish families with HAE

Distribution of 69 mutations found in 66 Type 1 HAE patients

Clinical Manifestations Associated with *C1INH* gene mutations

- **Study aim**
  - to investigate the factors influencing the heterogeneous clinical manifestations of HAE
- **Study design**
  - 106 caucasian subjects, including 39 unrelated patients and their family members diagnosed with Type 1 HAE
  - Disease characteristics documented: annual number of attacks, location, number of C1INH concentrate, age at occurrence of first attack
  - Screening for mutations in C1INH gene was done by sequencing after PCR amplification while relative quantification or exons was performed by quantitative PCR. The genotype of factor XIII polymorphism was determined by using PCR-RFLP.

Differences in the Clinical Course of Disease Between Patients with HAE Carrying Missense Mutations, or Other Mutations of the C1INH Gene

<table>
<thead>
<tr>
<th>Clinical property, median (interquartile range)</th>
<th>Genetically unrelated patients</th>
<th>All patients</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Missense (n = 7)</td>
<td>Nonmissense (n = 32)</td>
</tr>
<tr>
<td>Age at the onset of first symptoms (y)</td>
<td>18.0 (10.0-23.0)</td>
<td>14.3 (5.0-19.8)</td>
</tr>
<tr>
<td>Annual attack frequency</td>
<td>0.8 (0.2-5.2)</td>
<td>7.2 (2.5-10.4)</td>
</tr>
<tr>
<td>Subcutaneous attacks</td>
<td>0.6 (0.2-3.2)</td>
<td>4.1 (1.2-7.8)</td>
</tr>
<tr>
<td>Abdominal attacks</td>
<td>0.0 (0.0-0.4)</td>
<td>0.8 (0.2-3.1)</td>
</tr>
<tr>
<td>Severe attacks (abdominal and laryngeal)</td>
<td>0.0 (0.0-0.4)</td>
<td>1.5 (0.5-4.1)</td>
</tr>
<tr>
<td>Annual frequency of C1INH concentrate usage</td>
<td>0.0 (0.0-0.2)</td>
<td>0.5 (0.1-2.0)</td>
</tr>
</tbody>
</table>

Significant values are shown in boldface.

*Significance was estimated by applying the Mann-Whitney test.

†Significance was estimated considering correlation among family members by applying Poisson generalized estimating equations with independent correlation matrix and robust variance.

‡The number of symptom-free patients with C1INH deficiency in the group of subjects carrying missense and nonmissense mutations was 2 and 5, respectively.

Bors A et al. Less severe clinical manifestations in patients with hereditary angioedema with missense C1INH mutations. J Allergy Clin Immunol 2012; (Epub ahead of print)
Age at the Initial Onset of Symptoms in Patients Carrying Different Genotypes of the F12 Gene

Clinical Implications

• Assessment of mutations may be considered an essential component of the initial workup of new cases and potentially useful also for patient counseling.\(^1\)

\(^1\)Bors A et al. Less severe clinical manifestations in patients with hereditary angioedema with missense C1INH mutations. J Allergy Clin Immunol 2012;(Epub ahead of print)
Hypothesis: to determine a possible role for the exon 3-skipping isoform in the regulation of C1NH expression by comparing the mRNA splicing patterns found in healthy controls and HAE patients carrying different types of mutations. Additionally, we have tested the effect of androgen treatment on the relative levels of both transcripts.
Comparison of Total C1-Inh mRNA Expression Amongst Controls and Patients with Different Kinds of Mutations

Effect of the Androgen Treatment in Patients with Mutations

Missense Mutations

RNA copy number of splicing variants in controls (n: 18) and three treated and three untreated patients.

Mutations Affecting Splicing of Exon 3

RNA copy number of splicing variants in controls (n: 18) and two treated and two untreated patients.

Pathways Inhibited by C1 Inhibitor (C1INH) and New Drugs.

Come on... Come on... Throw the stick...