

Abbreviations used

AE:	Treatment-emergent adverse event
C1-INH:	C1 esterase inhibitor
Dose B:	Additional dose of 30 mg of subcutaneous ecallantide
EDEMA:	Evaluation of DX-88's Effects in Mitigating Angioedema
EDEMA3-DB:	Evaluation of DX-88's Effects in Mitigating Angioedema Double-Blind
HAE:	Hereditary angioedema
MSCS:	Mean Symptom Complex Severity
SAE:	Treatment-emergent serious adverse event
SUAC:	Severe upper airway compromise
TOS:	Treatment Outcome Score

METHODS

The objectives of EDEMA3-DB and EDEMA4 were to assess the safety and efficacy of 30 mg of subcutaneous ecallantide versus placebo in the treatment of moderate-to-severe acute attacks of HAE. Both studies were randomized, double-blind, placebo-controlled trials of similar design.

Patients and interventions

The appropriate institutional review board approved the studies at each site. Informed consent was obtained from patients before study entry. Patients aged 10 years or older with a confirmed diagnosis of HAE who presented to the study site within 8 hours of onset of a moderate-to-severe HAE attack were randomized 1:1 to receive 30 mg of subcutaneous ecallantide or placebo. During randomization, patients were stratified by ecallantide treatment history and attack site.

After initial treatment, both protocols permitted administration of an additional open-label dose of 30 mg of subcutaneous ecallantide if there was risk of severe upper airway compromise (SUAC). In EDEMA4, but not in EDEMA3-DB, an additional dose of 30 mg of subcutaneous ecallantide (Dose B) was permitted if the patient had no response (failure to achieve beginning of improvement) or an incomplete response (failure to achieve significant overall improvement after experiencing initial improvement) within 4 hours or experienced a relapse (recurrence of attack symptoms between 4 and 24 hours after dosing after experiencing initial improvement).

Each study enrolled both patients who had received ecallantide previously (nonnaïve patients) and those who had never received ecallantide (naïve patients). For patients treated in both EDEMA3-DB and EDEMA4, this integrated analysis only includes results from their initial treatment episode to maintain treatment-group independence.

Efficacy and safety assessments

Treatment efficacy was assessed by using validated, HAE-specific, patient-reported outcome measures: the Mean Symptom Complex Severity (MSCS) score and the Treatment Outcome Score (TOS).¹⁶ For both scores, patients identified the location of attack symptoms based on 5 symptom complexes: internal head/neck (upper respiratory attacks, referred to as *laryngeal*); stomach/gastrointestinal (referred to as *gastrointestinal*); and genital/buttocks, external head/neck, or cutaneous (collectively referred to as *peripheral*).

Change in MSCS score, a comprehensive point-in-time measure of symptom severity, was the primary end point in EDEMA4 and a secondary end point in EDEMA3-DB. On presentation, patients identified all active symptom complexes and rated the severity of each on a 3-point scale (1, mild; 2, moderate; and 3, severe). At 4 and 24 hours after dosing, patients rated severity of all symptom complexes identified at baseline, as well as any symptom complexes that emerged since presentation (0, no symptoms [applicable only to symptoms present at baseline]; 1, mild; 2, moderate; and 3, severe). The ratings from all presenting and emerging symptom complexes were averaged to generate the MSCS score. A decrease in MSCS score from baseline reflects an improvement in symptoms; the minimally important

difference for change in MSCS score from baseline was estimated to be -0.30 .¹⁶

TOS was the primary end point in EDEMA3-DB and a secondary end point in EDEMA4. TOS provides a comprehensive measure of treatment response that integrates anatomic locations. At 1, 2, 3, 4, and 24 hours after dosing, the patient's assessment of response for each symptom complex compared with baseline was recorded as follows: significant improvement (+100), improvement (+50), same (0), worsening (−50), and significant worsening (−100). The response for each symptom complex was weighted by the baseline severity, and the TOS was generated by averaging responses at each time point. The minimally important difference for TOS was estimated to be +30.¹⁶

MSCS scores and TOSs were not determined at 4 and 24 hours for patients who received an SUAC dose or at 24 hours for patients who received Dose B. Patients were also asked to complete an overall response assessment every 15 minutes for the first 2 hours, every 30 minutes for hours 3 and 4, and again at 24 hours. Possible responses included "a lot better or resolved," "a little better," "same," "a little worse," or "a lot worse." Overall response within 4 hours of treatment was evaluated in 3 ways: "improvement in overall response" was defined as a response of "a little better" or "a lot better or resolved"; "sustained improvement in overall response" was defined as a response of "a little better" or "a lot better or resolved" for a continuous duration of at least 45 minutes; and "significant improvement in overall response" was defined as a response of "a lot better or resolved," reflecting complete or near-complete symptom resolution. Durable response was defined as achieving improvement in overall response at 4 hours with either no change or further improvement at 24 hours. Patients who received a SUAC dose or Dose B were considered nonresponders.

Safety analysis was based on evaluation of reported treatment-emergent adverse events (AEs), which were defined as adverse events with onset on or after study drug administration, as per individual study protocol. For placebo-treated patients who received open-label ecallantide, AEs experienced before ecallantide administration were counted in the placebo group; AEs experienced after ecallantide administration were counted in the ecallantide group. Presenting HAE attacks that resulted in hospitalization were captured as treatment-emergent serious adverse events (SAEs); new and untreated HAE attacks reported during the 28-day follow-up period were captured as either AEs or SAEs. Patients were also evaluated for the development of hypersensitivity reactions, including anaphylaxis, according to criteria developed by the National Institute of Allergy and Infectious Diseases.¹⁷

Other safety evaluations included posttreatment assessment of serum chemistries, hematologic parameters, and immunologic parameters (antibody status and seroconversion). Antibody status was evaluated by using a validated electrochemiluminescence assay, which uses bridging technology and detects anti-ecallantide antibodies of all immunoglobulin subclasses (IgG, IgA, IgM, and IgE); this methodology is unable to distinguish between non-IgE isotypes. If the initial screening assay result was positive, a confirmatory assay was conducted. Samples were considered positive only if both the screening and confirmatory assay results were positive at titers of greater than 5. Samples that were confirmed positive for anti-ecallantide antibodies were also tested for neutralizing antibodies to ecallantide. Seroconversion that occurred during treatment episodes in EDEMA3 (both the double-blind arm and the open-label extension) or EDEMA4 were captured in this analysis.

Statistical methods

Change from baseline in MSCS score was analyzed by using the nonparametric Wilcoxon rank sum test. The TOS was analyzed by using the exact Wilcoxon 2-sample test to account for the large number of ties in the TOS data; *P* values were estimated by using Monte Carlo simulation. The Fisher exact test was used to compare proportions of responders based on various threshold levels; patients who received an SUAC dose within 4 hours were imputed to be treatment failures in these analyses. Unless otherwise noted, *P* values of less than .05 (using a 2-sided significance level) indicate statistical significance. Interquartile range values, the 25th and the 75th percentiles of a data set, indicate the data spread.

By using the same assumptions that were used to power the individual studies (ie, TOS for EDEMA3-DB¹⁴ and change in MSCS score for

EDEMA4¹⁵), the pooling of the 2 studies provided the following power for analyses by attack location:

- abdominal: 82% power for TOS and 61% power for change in MSCS score;
- laryngeal: 34% power for TOS and 22% power for change in MSCS score; and
- peripheral: 76% power for TOS and 56% power for change in MSCS score.

RESULTS

Study population

EDEMA3-DB included 72 patients (36 per treatment group), and EDEMA4 included 96 patients (48 per treatment group); 25 patients enrolled in both studies ($n = 14$ ecallantide-treated patients and $n = 11$ placebo-treated patients). Thus the integrated efficacy analysis includes 143 unique patients ($n = 70$ ecallantide-treated patients and $n = 73$ placebo-treated patients). The safety analysis includes 100 ecallantide-treated patients and 81 placebo-treated patients. Figs E1 and E2 (available in this article's Online Repository at www.jacionline.org) show the derivation of the integrated efficacy and safety populations, respectively.

Baseline demographics are summarized in Table I. Treatment groups were generally well matched with respect to age, sex, and race. At baseline, gastrointestinal attacks (44.8%) were the most frequently reported primary attacks. Relative to the placebo group, the ecallantide group had more peripheral attacks (45.7% vs 31.5%, respectively) and laryngeal attacks (21.4% vs 12.3%, respectively) and fewer gastrointestinal attacks (32.9% vs 56.2%, respectively).

Clinical efficacy profile

In the integrated analysis the change from baseline MSCS score at 4 hours after dosing was significantly greater for the ecallantide group than for the placebo group (ecallantide [mean $\pm$ SD], -0.97 ± 0.78 ; placebo, -0.47 ± 0.71 ; $P < .001$, Wilcoxon rank sum test). By 4 hours, 74.3% of ecallantide-treated patients experienced a reduction in symptom severity at least as great as the minimally important difference (-0.30) compared with 49.3% of placebo-treated patients ($P = .003$). These results, along with the proportion of patients achieving additional higher thresholds with respect to changes in MSCS scores (which were sensitivity analyses corresponding to more stringent definitions of clinical relevance), are summarized in Table II.

The TOS at 4 hours after dosing was significantly higher for ecallantide-treated patients than for placebo-treated patients (ecallantide [mean $\pm$ SD], 55.5 ± 46.5 ; placebo, 20.0 ± 58.9 ; $P < .001$ for exact Wilcoxon 2-sample test). At 4 hours, 70.0% of ecallantide-treated patients experienced an improvement at least as great as the minimally important difference threshold (30.0) compared with 38.0% of placebo-treated patients ($P < .001$). TOS results, along with the proportion of patients achieving additional higher TOS thresholds (which were sensitivity analyses corresponding to more stringent definitions of clinical relevance), are summarized in Table III. For comparison purposes, the MSCS score and TOS results from the individual studies, EDEMA3-DB¹⁴ and EDEMA4,¹⁵ are presented in Table E1 (available in this article's Online Repository at www.jacionline.org).

The proportion of patients who achieved improvement in overall response within 4 hours after dosing was greater in the ecallantide-treated group (51/70 [72.9%]) than in the placebo-

TABLE I. Patients' demographics from integrated analysis of controlled phase 3 studies of ecallantide

Characteristic	Ecallantide (n = 70)	Placebo (n = 73)	Total (n = 143)
Age* (y)			
Mean (SD)	36.5 (13.3)	36.1 (13.5)	36.3 (13.4)
Median	34.8	37.1	35.9
Range	16-77	10-72	10-77
Sex, no. (%)			
Female	49 (70.0)	45 (61.6)	94 (65.7)
Male	21 (30.0)	28 (38.4)	49 (34.3)
Race, no. (%)			
White	58 (82.9)	65 (89.0)	123 (86.0)
Nonwhite	12 (17.1)	8 (11.0)	20 (14.0)
Black	4 (5.7)	6 (8.2)	10 (7.0)
Hispanic	6 (8.6)	1 (1.4)	7 (4.9)
Asian	1 (1.4)	1 (1.4)	2 (1.4)
Other	1 (1.4)	0	1 (0.7)
Prior exposure to ecallantide, no. (%)			
Naive	59 (84.3)	54 (74.0)	113 (79.0)
Nonnaive	11 (15.7)	19 (26.0)	30 (21.0)

*Age at time of informed consent for the HAE episode used in the integrated analysis.

treated group (42/73 [57.5%]), although this difference did not reach statistical significance ($P = .079$). A total of 48 (68.6%) of 70 ecallantide-treated patients compared with 30 (41.1%) of 73 placebo-treated patients achieved sustained improvement in overall response within 4 hours ($P = .001$). A total of 33 (47.1%) of 70 ecallantide-treated patients and 19 (26.0%) of 73 placebo-treated patients achieved significant improvement in overall response within 4 hours ($P = .010$). These results are summarized in Fig 1.

The durability of response was assessed by using change in MSCS score and TOS at 24 hours after dosing. For both change in MSCS score (Table II) and TOS (Table III), symptomatic improvement in the ecallantide group was significantly greater than in the placebo group at 24 hours (MSCS score, $P = .028$, Wilcoxon rank sum test; TOS, $P = .039$, exact Wilcoxon 2-sample). On the basis of patients' assessments of their overall response to treatment, a statistically significant difference in the proportion of patients with a durable response was observed between ecallantide-treated patients (34/65 [52.3%]) and placebo-treated patients (22/68 [32.4%]) at 24 hours after dosing ($P = .023$).

Table IV presents subpopulation analyses based on primary attack location (gastrointestinal, laryngeal, or peripheral). For patients experiencing gastrointestinal attacks, the change in MSCS score at 4 hours after dosing indicated significantly greater improvement for ecallantide-treated patients than for placebo-treated patients ($P = .001$). Similar trends were observed for patients experiencing peripheral and laryngeal attacks. With respect to TOS, improvement was significantly greater for ecallantide-treated patients than for placebo-treated patients for all attack locations (gastrointestinal, $P = .026$; laryngeal, $P = .041$; and peripheral, $P = .035$).

Clinical safety profile

The incidence of AEs was similar between ecallantide-treated (36.0%) and placebo-treated (34.6%) patients. The most commonly reported AEs in the ecallantide group were headache

TABLE II. MSCS scores at 4 and 24 hours after dosing

	Ecallantide (n = 70)	Placebo (n = 73)	P value
Change from baseline MSCS score at 4 h			
No.*	67	67	
Median	−1.00	−0.33	<.001†
IQR	−1.50 to −0.50	−1.00 to 0.00	
Mean (SD)	−0.97 (0.78)	−0.47 (0.71)	
Proportion of responders at 4 h based on threshold levels for change in MSCS score			
No.‡	70	71	
≤−0.3, no. (%)	52 (74.3)	35 (49.3)	.003§
≤−0.5, no. (%)	52 (74.3)	32 (45.1)	<.001§
≤−0.7, no. (%)	43 (61.4)	27 (38.0)	.007§
≤−1.0, no. (%)	42 (60.0)	27 (38.0)	.012§
Change from baseline MSCS score at 24 h			
No.	51	49	
Median	−1.50	−1.00	.028†
IQR	−2.00 to −1.00	−1.67 to −0.50	
Mean (SD)	−1.37 (0.78)	−1.04 (0.73)	

IQR, Interquartile range.

*Analysis included only patients with valid end point assessments. Three patients in the ecallantide group received an SUAC dose and thus had no 4-hour assessment. Six patients in the placebo group either received an SUAC dose (n = 4) or were missing 4-hour data (n = 2).

†Comparison between ecallantide and placebo, Wilcoxon rank sum test.

‡Number of evaluable patients: patients who received an SUAC dose within 4 hours were considered evaluable and were analyzed as nonresponders. Two patients in the placebo group who were missing 4-hour data were considered nonevaluable.

§Fisher exact test: P values of less than .0125 were considered significant to account for the high degree of dependence between the groups.

||Analysis included only patients with valid end point assessments. Nineteen patients in the ecallantide group either received Dose B (n = 11), an SUAC dose (n = 3), or were missing 24-hour data (n = 5). Similarly, 24 patients in the placebo group either received Dose B (n = 16), an SUAC dose (n = 4), or were missing 24-hour data (n = 5). One patient in the placebo group received both Dose B and an SUAC dose.

(8.0%), nausea (5.0%), diarrhea (4.0%), pyrexia (4.0%), and nasopharyngitis (3.0%) versus headache (7.4%), diarrhea (3.7%), and vomiting (3.7%) in the placebo group. A total of 3.0% of ecallantide-treated patients and 4.9% of placebo-treated patients reported an AE of HAE during the follow-up period. Three (3.0%) patients in the ecallantide group reported mild local injection site reactions versus 1 (1.2%) patient in the placebo group. Most AEs were of mild or moderate severity and unrelated to study drug administration. No patients withdrew from these studies because of an AE.

The incidence of SAEs was also similar between ecallantide-treated (3.0%) and placebo-treated (3.7%) patients. In the ecallantide group all 3 of the reported SAEs were worsening of the symptoms of the presenting HAE attack (1 moderate and 2 severe requiring hospitalization) considered unrelated to treatment. The 2 patients who experienced severe HAE attacks were both initially randomized to placebo; 1 patient received Dose B for incomplete resolution of gastrointestinal symptoms and 1 patient received both SUAC and Dose B for a severe laryngeal attack. Of the 3 SAEs reported in the placebo group, 2 were HAE attacks, and 1 was hospitalization for a moderately severe HAE attack.

Because of the potential for hypersensitivity reactions, including anaphylaxis, to ecallantide, a protein therapeutic, AEs were examined for cases suggestive of anaphylaxis and hypersensitivity. Each event was evaluated for temporal relationship, causal relationship, seriousness, and presence of other symptoms

TABLE III. TOS at 4 and 24 hours after dosing

	Ecallantide (n = 70)	Placebo (n = 73)	P value
TOS at 4 h			
No.*	67	67	
Median	50.0	0.0	<.001†
IQR	16.7 to 100.0	0.0 to 66.7	
Mean (SD)	55.5 (46.5)	20.0 (58.9)	
Proportion of responders at 4 h based on threshold levels for TOS			
No.‡	70	71	
≥30, no. (%)	49 (70.0)	27 (38.0)	<.001§
≥50, no. (%)	49 (70.0)	27 (38.0)	<.001§
≥70, no. (%)	30 (42.9)	16 (22.5)	.012§
≥100, no. (%)	26 (37.1)	15 (21.1)	.042§
TOS at 24 h			
No.	51	50	
Median	100.0	75.7	.039†
IQR	50.0 to 100.0	0.0 to 100.0	
Mean (SD)	75.5 (40.4)	51.4 (59.6)	

IQR, Interquartile range.

*Analysis included only patients with valid end point assessments. Three patients in the ecallantide group received an SUAC dose and thus had no 4-hour assessment. Six patients in the placebo group either received an SUAC dose (n = 4) or were missing 4-hour data (n = 2).

†Comparison between ecallantide and placebo, exact Wilcoxon 2-sample test with Monte Carlo simulation.

‡Number of evaluable patients: patients who received an SUAC dose within 4 hours were considered evaluable and were analyzed as nonresponders. Two patients in the placebo group who were missing 4-hour data were considered nonevaluable.

§Fisher exact test: P values of less than .0125 were considered significant to account for the high degree of dependence between the groups.

||Analysis included only patients with valid end point assessments. Nineteen patients in the ecallantide group either received Dose B (n = 11), an SUAC dose (n = 3), or were missing 24-hour data (n = 5). Similarly, 23 patients in the placebo group either received Dose B (n = 16), an SUAC dose (n = 4), or were missing 24-hour data (n = 4). One patient in the placebo group received both Dose B and a SUAC dose.

suggestive of hypersensitivity by using National Institute of Allergy and Infectious Diseases anaphylaxis criteria.¹⁷ There were no events reported in this patient population that were diagnostic of anaphylaxis or moderate-to-severe hypersensitivity.

Additional safety assessments revealed no clinically relevant changes in coagulation parameters (activated partial thromboplastin time, prothrombin time, and thrombin time), hepatic function, or renal function in ecallantide-treated patients. One placebo-treated patient experienced an increase in activated partial thromboplastin time levels 1.5 times greater than the upper limit of normal but did not experience a clinically significant hemorrhage.

Because patients with HAE are likely to receive multiple doses of ecallantide for the treatment of acute attacks over time, antibody status and seroconversion (development of anti-ecallantide antibodies) were part of the safety evaluation. A total of 133 patients who enrolled in EDEMA3 (including the open-label extension) or EDEMA4 received at least 1 dose of ecallantide during the overall EDEMA program (including the phase 2 studies). Of these patients, 127 had negative and 4 (all of whom had prior exposure to ecallantide) had positive results for anti-ecallantide antibodies at the time of their first treatment in either EDEMA3-DB or EDEMA4. After dosing, anti-ecallantide antibodies were detected at a titer of greater than 5 in 2 of the 127 patients who had negative results at baseline, yielding a seroconversion rate of 1.6%. One of these patients exhibited non-IgE antibodies to ecallantide, and the other patient exhibited both IgE

Percentage of patients achieving improvement in overall response within 4 hr

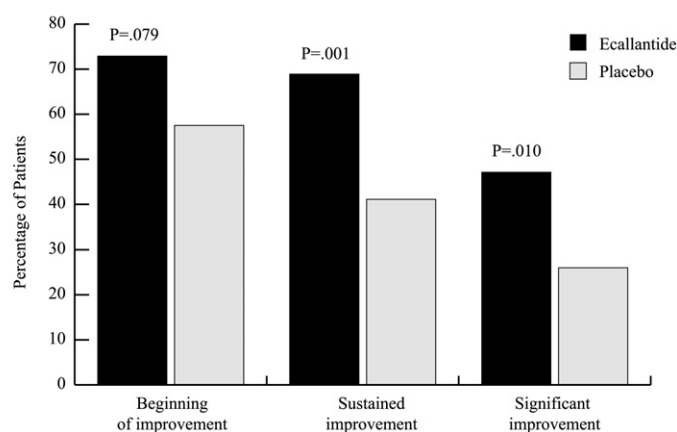


FIG 1. Proportion of patients achieving improvement in overall response by 4 hours after dosing. See the Methods section for definitions of beginning of improvement, sustained improvement, and significant improvement. These data reflect cumulative responses through 4 hours based on an overall response assessment conducted every 15 minutes for the first 2 hours and every 30 minutes for hours 3 and 4. *P* values are from the Fisher exact test.

TABLE IV. Change from baseline in MSCS score and TOS at 4 hours after dosing by primary attack location

Primary attack location*	Change in MSCS score at 4 h after dosing			TOS at 4 h after dosing		
	Ecallantide (n = 70)	Placebo (n = 73)	<i>P</i> value†	Ecallantide (n = 70)	Placebo (n = 73)	<i>P</i> value†
Gastrointestinal						
No.	23	39		23	39	
Median	-1.0	-0.5	.001	50.0	25.0	.026
IQR	-2.0 to -1.0	-1.0 to 0.0		50.0 to 100.0	0.0 to 100.0	
Mean (SD)	-1.4 (0.9)	-0.5 (0.8)		62.5 (40.7)	27.3 (60.9)	
Laryngeal						
No.	12	6		12	6	
Median	-1.0	-0.7	.335	100.0	-25.0	.041
IQR	-1.5 to -0.5	-1.0 to 0.0		66.7 to 100.0	-50.0 to 60.0	
Mean (SD)	-1.0 (0.7)	-0.6 (0.8)		73.6 (49.5)	-1.1 (67.9)	
Peripheral						
No.	32	22		32	22	
Median	-1.0	0.0	.111	50.0	0.0	.035
IQR	-1.0 to 0.0	-1.0 to 0.0		0.0 to 100.0	-33.3 to 50.0	
Mean (SD)	-0.7 (0.6)	-0.4 (0.6)		43.6 (47.4)	12.9 (53.2)	

IQR, Interquartile range.

*Primary attack location was determined in a hierarchical fashion: the primary attack was classified as laryngeal if a moderate-to-severe oropharyngeal head-neck symptom complex was identified at baseline (regardless of the presence of other symptom complexes). If the attack was not considered laryngeal, it was classified as gastrointestinal if a moderate-to-severe gastrointestinal symptom complex was identified at baseline. Otherwise, the attack was classified as peripheral if a moderate-to-severe nonoropharyngeal head-neck, genital, or cutaneous symptom complex was identified at baseline.

†Comparison between ecallantide and placebo, Wilcoxon rank sum test.

and non-IgE antibodies. Both patients seroconverted after treatment in the open-label extension of EDEMA3. Positive samples were assayed for the presence of neutralizing antibodies to ecallantide. In patients who had negative results at baseline for neutralizing antibodies, 2 (1.6%) of 127 demonstrated seroconversion to neutralizing antibodies.

In these double-blind studies, no differences in the overall incidence or type of AEs were observed between patients who had anti-ecallantide antibodies and those who did not. Furthermore, comparison of changes in MSCS scores and TOSs between seropositive and seronegative patients did not suggest any abrogation of treatment effect resulting from seroconversion.

DISCUSSION

During an HAE attack, unregulated activation of plasma kallikrein results in increased production of bradykinin, the key mediator of HAE attacks, through the contact pathway.^{18,19} This article describes the findings of an integrated analysis of pooled data sets from 2 placebo-controlled phase 3 studies of nearly identical design that evaluated the efficacy and safety of ecallantide, an inhibitor of plasma kallikrein, in the treatment of acute HAE attacks.

This analysis was conducted primarily to address the constraints on sample size inherent in clinical studies of HAE, the rarity of which makes enrollment of large patient numbers

difficult. For the primary end points (TOS and change in MSCS score at 4 hours), the pooled analysis achieved greater statistical significance for the ecallantide versus placebo comparison than either EDEMA3-DB or EDEMA4 (TOS: $P < .001$ vs $P = .004$ and $P = .003$, respectively; MSCS score: $P < .001$ vs $P = .014$ and $P = .010$, respectively).

Pooling the data from these studies also provided greater statistical power with respect to subset analyses and secondary end points. For most secondary measures of efficacy at 4 and 24 hours, P values for the integrated analysis were typically less than the smaller of the 2 P values from the individual studies, improving the degree of statistical confidence. The integrated analysis allowed for new ecallantide versus placebo comparisons based on attack location, a more robust examination of response durability, and greater insight into the response level that can be expected within 4 hours of treatment.

The study population was broadly representative of the overall HAE population: median age was 36 years, and 65% were female; in a recent survey of patients with HAE, median age was 40 years, and 60% were female.²⁰ Although small in absolute terms, the number of patients was substantial considering the low prevalence of HAE. Efficacy was measured by the MSCS score and the TOS, validated patient-reported outcomes that can be used to reliably assess variations in symptom complex severity, as well as the patient's response to administered therapy, respectively.¹⁶ The change from baseline in MSCS score and TOS demonstrated statistically robust, clinically meaningful between-group differences favoring ecallantide over placebo at 4 hours after dosing. Significantly more ecallantide-treated patients than placebo-treated patients achieved improvement at least as great as the minimally important differences in MSCS scores and TOSs. In addition, these between-group differences remained significant across more stringent improvement thresholds, confirming that a clinically meaningful benefit was observed.

The durability of response to ecallantide was demonstrated by significant differences favoring ecallantide over placebo in both the MSCS score and TOS at 24 hours after dosing. The majority of ecallantide-treated patients achieved a response by 4 hours and maintained it through 24 hours after a single dose, indicating the durability of response on an individual patient basis. No patient in the integrated analysis population experienced rebound, defined as symptom improvement at 4 hours followed by a global response assessment at 24 hours that was either at or worse than baseline.

The integrated analysis showed strong evidence for the efficacy of ecallantide at all attack locations based on TOS. In spite of pooling, not all attack location subgroups had sufficient size to be able to demonstrate statistically significant between-group differences, especially for the MSCS score, although the data trend in favor of ecallantide treatment. When assessed by MSCS score at 4 hours after dosing, symptom improvement was most robust for those patients with gastrointestinal attacks; when assessed based on TOS at 4 hours after dosing, symptom improvement after ecallantide treatment was statistically superior to placebo at all attack sites (gastrointestinal, laryngeal, and peripheral).

In this pooled analysis, ecallantide was found to be safe and generally well tolerated, with an incidence of AEs and SAEs comparable with that seen with placebo. Most AEs were mild to moderate in intensity and resolved without sequelae. Careful review of all potential hypersensitivity-related AEs demonstrated

no evidence of hypersensitivity reactions to ecallantide in this population.

The rate of seroconversion to both anti-ecallantide antibodies and to neutralizing antibodies was 1.6%. It is important to note that the electrochemiluminescence methodology used to test for anti-ecallantide antibodies is unable to differentiate between non-IgE isotypes. In this analysis, seroconversion was not associated with any observable effect on ecallantide efficacy or on AEs, although the number of seropositive patients was small, and these studies were not designed to assess the effect of seroconversion to anti-ecallantide antibodies. An ongoing, long-term, observational study should provide additional insight into the clinical significance of the presence of anti-ecallantide antibodies.

In conclusion, an integrated analysis of independent data sets from 2 placebo-controlled phase 3 studies provides statistically robust and new information about ecallantide for the treatment of acute attacks of HAE. Ecallantide demonstrated activity at all attack sites, sustained efficacy through 24 hours, and a low incidence of AEs. Additionally, in this population there were no reports of anaphylaxis or moderate-to-severe drug hypersensitivity reactions. Ecallantide reduces symptom severity during acute HAE attacks while offering ease of administration through the subcutaneous route. By blocking plasma kallikrein and reducing cleavage of high-molecular-weight kininogen and the subsequent release of bradykinin, ecallantide effectively treats HAE attacks. Thus ecallantide represents an important therapeutic option for a previously unmet medical need.

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Clinical implications: Ecallantide represents an important therapeutic option for a previously unmet medical need: the ability to reduce symptom severity during acute attacks of HAE with a drug offering ease of administration.

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The derivations of the efficacy analysis and safety analysis populations are illustrated schematically in Figs E1 and E2, respectively. A total of 72 patients (36 per treatment group) were enrolled in EDEMA3-DB, and 96 patients (48 per treatment group) were enrolled in EDEMA4. There were 25 patients enrolled in both studies ($n = 14$ ecallantide-treated patients and $n = 11$ placebo-treated patients).

Patients who participated in both EDEMA3-DB and EDEMA4 and received both placebo and ecallantide were included in both the ecallantide-treated and placebo-treated groups. The ecallantide-treated group consisted of 78 patients who were randomized to ecallantide (36 in EDEMA3-DB plus 48 in EDEMA4 minus 6 patients who were randomized to ecallantide in both studies and

were counted only once) plus 22 patients randomized to placebo who received open-label ecallantide as rescue medication (SUAC dose, Dose B, or both). The placebo-treated group consisted of 81 patients randomized to placebo (36 in EDEMA3-DB plus 48 in EDEMA4 minus 3 patients who were randomized to placebo in both studies and were counted only once).

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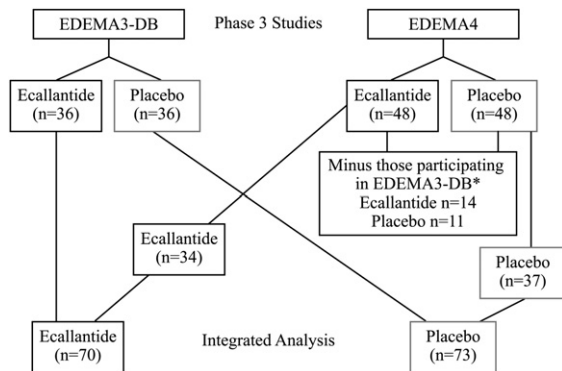


FIG E1. Derivation of the study population for integrated analysis of efficacy from ecallantide phase 3 studies. *Some patients participated in both EDEMA3-DB and EDEMA4; because the efficacy analysis was based only on the first treatment episode, EDEMA4 efficacy results from these patients were excluded.

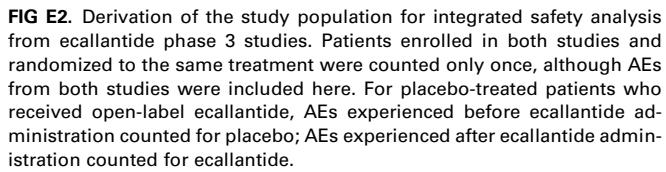


TABLE E1. Primary outcome data from the individual studies EDEMA3-DB and EDEMA4

MSCS score	EDEMA3-DB ^{E1}			EDEMA4 ^{E2}		
	Ecallantide (n = 36)	Placebo (n = 36)	P value	Ecallantide (n = 48)	Placebo (n = 48)	P value
Change from baseline at 4 h						
No.	36	36		47	42	
Mean (SD)	−0.88 (1.11)	−0.51 (0.68)	.014‡	−0.81 (0.63)	−0.37 (0.82)	.010‡
Proportion of patients with change in MSCS score ≤−0.3 at 4 h						
No.	36	36		48	45	
No. (%)	27 (75.0)	23 (63.9)	.443	33 (68.8)	18 (40.0)	.007†
Change from baseline at 24 h						
No.	33	33		32	24	
Mean (SD)	−0.87 (1.14)	−0.46 (1.07)	.041*†	−1.51 (0.63)	−1.07 (0.84)	.039*
TOS						
At 4 h						
No.	36	36		47	42	
Mean (SD)	46.8 (59.3)	21.3 (69.0)	.004*	53.4 (49.7)	8.1 (63.2)	.003*
Proportion of patients with TOS ≥30 at 4 h						
No.	36	36		48	45	
No. (%)	26 (72.2)	19 (52.8)	.144†	33 (68.8)	14 (31.1)	<.001†
At 24 h						
No.	33	34		32	24	
Mean (SD)	44.3 (70.4)	−0.5 (87.9)	.007*	88.8 (28.1)	55.1 (58.3)	.029*

*P value from Wilcoxon rank sum test blocked by the stratification variables of prior treatment and attack location.

†P value from Fisher exact test.