ORIGINAL ARTICLE

Oral Plasma Kallikrein Inhibitor for Prophylaxis in Hereditary Angioedema

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ABSTRACT

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BACKGROUND

Hereditary angioedema is a life-threatening illness caused by mutations in the gene encoding C1 inhibitor (also called C1 esterase inhibitor) that lead to overactivation of the kallikrein–bradykinin cascade. BCX7353 is a potent oral small-molecule inhibitor of plasma kallikrein with a pharmacokinetic and pharmacodynamic profile that may help prevent angioedema attacks.

METHODS

In this international, three-part, dose-ranging, placebo-controlled trial, we evaluated four doses of BCX7353 (62.5 mg, 125 mg, 250 mg, and 350 mg once daily) for the prevention of angioedema attacks over a 28-day period. Patients with type I or II hereditary angioedema with a history of at least two angioedema attacks per month were randomly assigned to BCX7353 or placebo. The primary efficacy end point was the number of confirmed angioedema attacks. Key secondary end points included angioedema attacks according to anatomical location and quality of life.

RESULTS

A total of 77 patients underwent randomization, 75 received BCX7353 or placebo, and 72 completed the trial. The rate of confirmed angioedema attacks was significantly lower among patients who received BCX7353 at daily doses of 125 mg or more than among those who received placebo, with a 73.8% difference at 125 mg (P<0.001). Significant benefits with respect to quality-of-life scores were observed in the 125-mg and 250-mg dose groups (P<0.05). Gastrointestinal adverse events, predominantly of grade 1, were the most commonly reported adverse events, particularly in the two highest BCX7353 dose groups.

CONCLUSIONS

Once-daily oral administration of BCX7353 at a dose of 125 mg or more resulted in a significantly lower rate of attacks of hereditary angioedema than placebo. Mild gastrointestinal symptoms were the principal side effect. (Funded by BioCryst Pharmaceuticals; APeX-1 ClinicalTrials.gov number, NCT02870972.)

HEREDITARY ANGIOEDEMA IS A GENETIC disorder characterized by recurrent episodes of swelling.[1,2] Oropharyngeal swelling can be life-threatening[3], and attacks in other sites, including the limbs, genitalia, face, and intestines, can be painful, disabling, and disfiguring and have a substantial effect on function and quality of life.[4] Most patients with hereditary angioedema have mutations in the regulatory or coding regions of the gene that encodes C1 inhibitor (*SERPING1*), which result in either a deficiency (type I) or dysfunction (type II) of C1 inhibitor.[5]

C1 inhibitor (also called C1 esterase inhibitor) is a serine protease inhibitor that normally prevents uncontrolled contact activation and bradykinin production by covalently binding to and inactivating plasma kallikrein.[6] Increased bradykinin generation leads to increased vascular permeability and swelling.

Plasma-derived purified C1 inhibitor is approved for the prophylactic treatment of hereditary angioedema through intravenous or subcutaneous injection. The only available effective drugs for oral prophylaxis against angioedema attacks are attenuated androgens, such as danazol, and tranexamic acid. Although administration of androgens is convenient, unacceptable adverse effects (e.g., virilizing androgenic hormonal effects, hypertension, and an increased risk of hepatocellular adenoma or carcinoma) and several contraindications (e.g., in growing children and pregnant women) limit their clinical use.[7-9]

BCX7353 is a potent, synthetic small molecule that inhibited plasma kallikrein with once-daily oral administration in healthy volunteers.[10] In contrast to parenterally administered options under development or commercially available for prophylaxis against angioedema attacks, inhibition of kallikrein with an orally bioavailable small molecule such as BCX7353 offers the distinct advantage of oral administration.

METHODS

TRIAL DESIGN

The Angioedema Prophylaxis 1 (APeX-1) trial was a phase 2, randomized, double-blind, placebo-controlled, parallel-group, three-part, dose-response trial that evaluated the safety, adverse-event profile, pharmacokinetics, pharmacodynamics, and efficacy of BCX7353 in patients with hereditary angioedema and C1 inhibitor deficiency. The trial was designed to establish proof of concept that a high dose of BCX7353, administered once daily for 28 days, was safe and effective in reducing the rate of angioedema attacks. Once proof of concept was established, additional doses were evaluated to characterize the dose response. On the basis of pharmacokinetic and pharmacodynamic modeling from a completed phase 1 study, the protocol was initially designed as a two-part trial with once-daily doses of 350 mg, 250 mg, and 125 mg of BCX7353. After the trial was initiated, the protocol was amended to include a lower dose of 62.5 mg of BCX7353 once daily in part 3. Eligible patients underwent central randomization to BCX7353 or placebo within each part of the trial. Patients participated in one trial part only. Further details of the trial design and randomization are provided in the protocol (available with the full text of this article at NEJM.org) and in Section S2 in the Supplementary Appendix (available at NEJM.org).

The trial was designed by the sponsor (Bio-Cryst Pharmaceuticals) in collaboration with the first, next-to-last, and last authors. The protocol was approved by regulatory authorities and independent ethics committees. All the patients provided written informed consent before the conduct of any trial-related procedures.

Investigators and site personnel collected data in collaboration with the sponsor, and the sponsor and PharStat (a contract research organization paid by the sponsor) analyzed the data. All the authors had access to the data (nondisclosure agreements were in place). The first, next-to-last, and last authors contributed to the analysis and interpretation of the data. All the authors reviewed and approved the manuscript for publication and vouch for the completeness and accuracy of the data and analyses and for the adherence of the trial to the protocol. Technical support in the drafting of the manuscript, including preparation of the first draft, was provided by J Anderson Solutions, funded by the sponsor.

A Quick Take is available at NEJM.org

PATIENTS AND PATIENT RECRUITMENT

Eligible male or female patients were 18 to 70 years of age with a clinical diagnosis of type I or type II hereditary angioedema. Patients were required to have a documented rate of angioedema attacks of at least two attacks per month for 3 consecutive months within the 6 months before the screening visit. Patients were excluded if they were suspected of having C1 inhibitor resistance (i.e., they were having regular attacks despite C1 inhibitor treatment for routine prophylaxis) or if they were using a C1 inhibitor, androgens, or tranexamic acid for prophylaxis of attacks within 7 days before screening. Treatment of acute attacks, including the use of a C1 inhibitor, was not excluded. Complete inclusion and exclusion criteria are provided in Table S3 in the Supplementary Appendix.

TRIAL REGIMENS

Blinded trial medication with matching placebo was supplied as capsules for daily oral administration for 28 days. Patients were permitted to use their usual medication (including icatibant and intravenous and subcutaneous C1 inhibitor) to treat any attacks that occurred at any time during the trial.

CLINICAL ASSESSMENTS

Patients were provided with paper diaries for daily recording of trial-regimen administration and angioedema attacks. If patients had an attack, they were to provide details of the symptoms, severity (using the Angioedema Activity Score),[11] triggers, and duration of the attack and medications administered to treat the attack. To assess the effect of the disease on the patient's quality of life, the Angioedema Quality of Life Questionnaire (AE-QoL)[12] was administered at the start and end of the intervention phase. Patients returned to the clinic for routine monitoring of vital signs, 12-lead electrocardiography, pulmonary diffusion testing, physical examinations, laboratory assessments of serum biochemical and hematologic values, liver-enzyme testing, urinalysis, and documentation of adverse events. Blood samples for pharmacokinetic and pharmacodynamic testing were obtained from patients before dose administration (on days 1, 14, and 29) and for up to 24 hours after the dose on day 14. Blood samples for measurement of functional levels of C1 inhibitor were obtained on days 1 and 29.

EFFICACY

The primary end point of the trial was the number of confirmed angioedema attacks during the effective dosing period (days 8 to 28, inclusive). All patient-reported attacks were adjudicated by an independent clinical end-point adjudication panel whose members were unaware of the trial-group assignments. Panel membership and details regarding the adjudication process are provided in Section S1 in the Supplementary Appendix. Attacks that were confirmed by the adjudication panel were included in efficacy analyses.

Key secondary end points of the trial included the number of angioedema attacks categorized according to anatomical location and the change from baseline in scores on the disease-specific and validated AE-QoL. The AE-QoL measures quality of life across four domains (functioning, fatigue and mood, fears and shame, and food) and derives a total score on a scale from 0 to 100, with higher scores indicating greater impairment. A minimal clinically important difference of 6 points has been described.[13] Other secondary efficacy end points are presented in Table S7 in the Supplementary Appendix.

SAFETY

The safety and adverse-event profile of BCX7353 were assessed by means of laboratory testing and monitoring of patients for adverse events that occurred from the time of the first dose to the time of the last dose plus 30 days. During the conduct of the trial, an independent data monitoring committee (Table S2 in the Supplementary Appendix) provided safety oversight by reviewing blinded data and advising the sponsor on safety-related findings.

PHARMACOKINETICS AND PHARMACODYNAMICS

Steady-state pharmacokinetic properties of BCX7353 — the maximum plasma concentration (C_{max}), the observed trough plasma concentration at the end of the dosing interval (C_{tau}), the time at which C_{max} occurred, and the area under the plasma concentration–time curve during the dosing interval (AUC_{tau}) — were additional secondary end points, as was the ex vivo

kallikrein inhibition activity of BCX7353 (analyzed as described previously[10]).

STATISTICAL ANALYSIS

In part 1 of the trial, we calculated that a sample of 18 patients per group would provide a power of 93% to detect a difference in the attack rate of 0.5 attacks per week between the 350-mg BCX7353 group and the placebo group at an alpha level of 0.05, assuming a mean baseline rate of 1 attack per week. The primary efficacy end point of the trial was the number of confirmed angioedema attacks in the intention-to-treat and per-protocol populations. Population definitions and numbers of patients included in each population are summarized in Section S4 in the Supplementary Appendix. The number of angioedema attacks was analyzed according to trial group with the use of appropriate descriptive statistics for the weekly attack rate, change and percentage change from baseline in attack rate, proportion of patients with no attacks, and number and percent of attack-free days. Efficacy analyses were conducted for angioedema attacks reported during the entire dosing interval (days 1 to 28, inclusive) and during the effective dosing period in which BCX7353 levels were at a steady state (days 8 to 28, inclusive).

Between-group comparisons (BCX7353 vs. placebo) for the weekly attack rate were performed with the use of an analysis-of-covariance model. The adjusted qualifying attack rate was included as a covariate in the model. The least-squares mean for each trial group and the relative difference and associated 95% confidence intervals in attack rate between the placebo group and each BCX7353 dose group were calculated.

Data for the primary end point are shown for the effective dosing period in the intention-to-treat population, since this would best assess the effectiveness of the trial medication after steady-state levels had been achieved in this small, phase 2 trial. Data are also presented for the per-protocol population and during the entire dosing period. The primary efficacy analysis was conducted without imputation for missing data (Tables S5 and S6 in the Supplementary Appendix).

Analyses of the primary end point of the number of confirmed attacks, followed by the secondary end point of AE-QoL scores, were performed with the use of post hoc hierarchical testing to preserve the prespecified level of significance of 0.05. Safety analyses were based on the safety population and included all randomly assigned patients who received a dose of a trial regimen. All statistical analyses were performed with the use of SAS software (version 9.3).

RESULTS

PATIENTS

The trial was initiated in August 2016, and the last patient observation occurred in August 2017. A total of 86 patients were screened from 26 sites across Europe, Canada, and Australia. Of the 77 patients who underwent randomization, 2 never received a trial regimen or entered any diary data and 3 discontinued the trial regimen owing to an adverse event or laboratory abnormality. Details regarding disposition of the patients and discontinuation are presented in Figure S2 and Table S4, respectively, in the Supplementary Appendix.

The trial groups were generally balanced with respect to age and qualifying attack rate, although the proportion of women was not evenly distributed across groups, and patients in the 350-mg dose group had a higher rate of previous androgen use than the other groups (Table 1). Baseline C1 inhibitor levels were low, with median values ranging from 19% to 34% of normal values. Sequential enrollment into parts 1, 2, and 3 may have contributed to the differences in demographic and clinical characteristics that were observed among the trial groups.

EFFICACY RESULTS

Primary End Point

The primary efficacy end point was the number of confirmed angioedema attacks. Significant and clinically meaningful differences in the rate of attacks were observed between patients who received BCX7353 at doses of 125 mg or more once daily and those who received placebo (Table 2). The least-squares mean rate of attacks among patients who received placebo (pooled across all three parts of the trial) was 0.95 per week, and the least-squares mean percent differences in the confirmed rate of attacks per week according to BCX7353 dose group were as follows: 350 mg, −45.5% (P=0.006); 250 mg, −44.6%

Table 1. Baseline Characteristics of the Patients (Intention-to-Treat Population).*

Characteristic	BCX7353				Placebo (N=23)
	62.5 mg (N=7)	125 mg (N=14)	250 mg (N=15)	350 mg (N=18)	
Age — yr	38.9±16.6	48.1±12.6	39.5±13.9	43.8±11.6	46.6±10.9
Female sex — no. (%)	6 (86)	10 (71)	7 (47)	11 (61)	13 (57)
Race — no. (%)†					
White	7 (100)	14 (100)	12 (80)	15 (83)	21 (91)
Asian	0	0	1 (7)	1 (6)	0
Other	0	0	2 (13)	2 (11)	2 (9)
Previous androgen use — no. (%)	3 (43)	4 (29)	8 (53)	15 (83)	13 (57)
Angioedema attacks/wk‡	1.15±0.52	0.99±0.52	0.93±0.45	0.90±0.44	1.03±0.57
Median C1 inhibitor function (IQR) — % of normal value	11 (7–47)	15 (11–35)	15 (6–28)	19 (5–28)	10 (4–37)
Alanine aminotransferase — U/liter	18.4±6.6	22.0±9.5	31.0±20.5	42.5±44.1	29.0±13.0

* Plus–minus values are means ±SD. The intention-to-treat population included all the patients who underwent randomization. IQR denotes interquartile range.
† Race was reported by the patient.
‡ The baseline attack rate was based on patient-reported attacks over a 3-month period.

(P=0.01); 125 mg, −73.8% (P<0.001); and 62.5 mg, −10.5% (P=0.64).

When attack rates were analyzed according to anatomical location, a differential response was observed for peripheral attacks as compared with abdominal attacks, particularly at the 250-mg and 350-mg doses of BCX7353 (Table 2). Although the rate of peripheral attacks was lower with BCX7353 than with placebo at all doses of 125 mg or more, the rate of abdominal attacks was lower with BCX7353 than with placebo at the 125-mg dose only.

Secondary End Points

The percent of attack-free days and the proportion of patients who were attack-free were higher with BCX7353 at doses of 125 mg or more than with placebo (Table 2). An important secondary end point in the trial was quality of life as measured by the AE-QoL. The least-squares mean change from baseline in the AE-QoL total score was −29.0 in the 125-mg dose group, as compared with −4.5 in the placebo group (difference, −24.5; P<0.001), indicating a significant benefit with the 125-mg dose. There were also significant differences favoring the 125-mg dose across three of the four domains of the AE-QoL (P≤0.006 vs. placebo) (Fig. 1). A significant difference between the 250-mg dose group and the placebo group was observed for the functioning domain (P=0.02), in favor of the 250-mg dose. Results for other secondary end points, including the rate of attacks resulting in treatment and the overall severity of illness, support the efficacy of BCX7353 at doses of 125 mg or more (Table S7 in the Supplementary Appendix). None of the patients discontinued the trial medication owing to lack of efficacy.

PHARMACOKINETICS AND PHARMACODYNAMICS

After administration of multiple daily doses of BCX7353, the C_{max} was reached at a median of 3 to 4 hours after dosing (Fig. 2A). At doses of 125 mg or more, geometric mean plasma trough concentrations (C_{tau}) were generally maintained above the minimum target concentration range (approximately 4 times the half-maximal effective concentration [EC_{50}]) throughout the dosing interval. There was a greater-than-dose-proportional increase in exposure (AUC_{tau} and C_{max}) across the dose range of 62.5 mg to 350 mg, with an increase in exposure of approximately 11 times with an increase in dose of 4.6 times (Table S8 in the Supplementary Appendix). A dose-dependent inhibition of kallikrein was observed with BCX7353 treatment across the dose range (Fig. 2B),
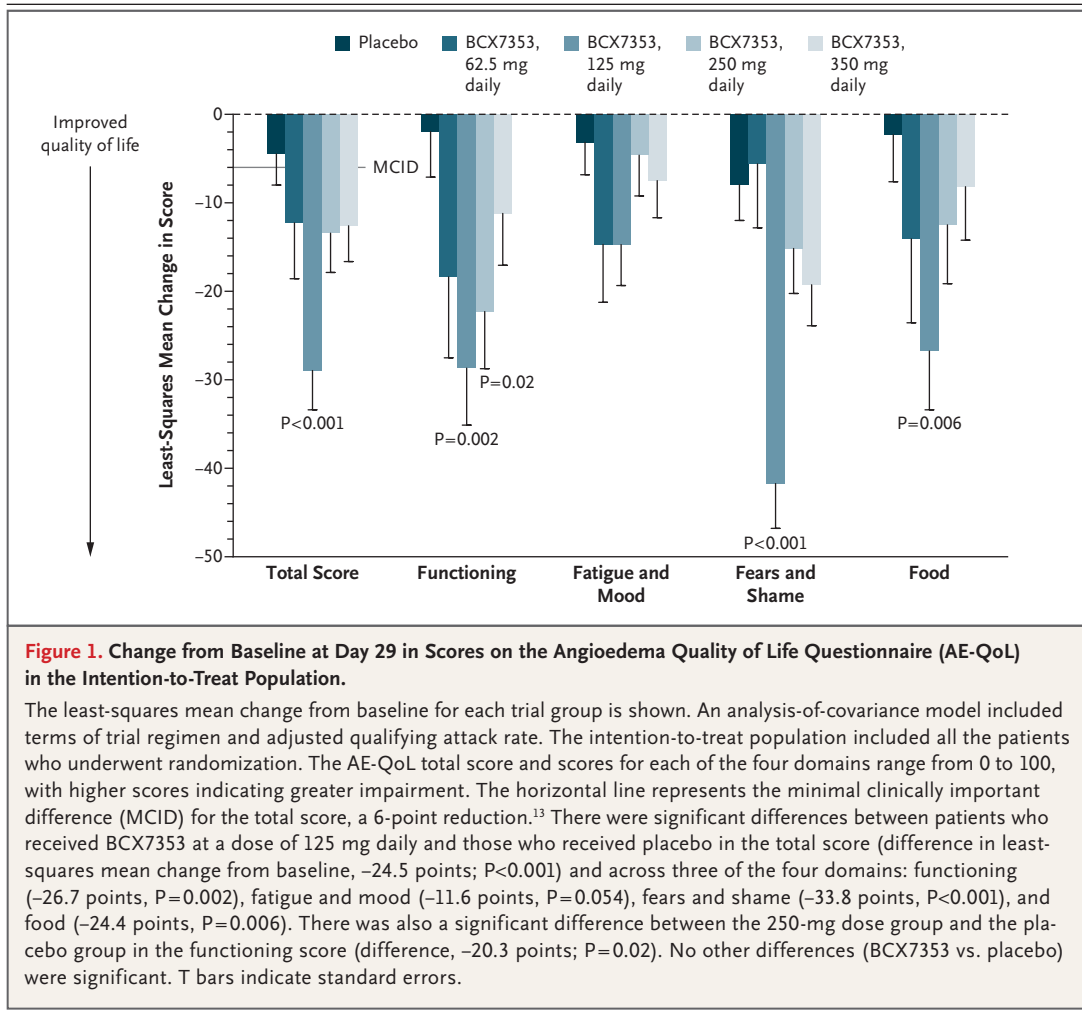
Table 2. Primary and Secondary Efficacy End Points (Intention-to-Treat Population, Effective Dosing Interval).*

End Point	BCX7353				Placebo (N=23)
	62.5 mg (N=7)	125 mg (N=14)	250 mg (N=15)	350 mg (N=18)	
Primary end point					
Least-squares mean no. of attacks/wk	0.85	0.25	0.53	0.52	0.95
Difference vs. placebo (95% CI)	−0.10 (−0.52 to 0.32)	−0.70 (−1.03 to −0.37)	−0.42 (−0.76 to −0.10)	−0.43 (−0.74 to −0.13)	
Percent difference vs. placebo	−10.5	−73.8	−44.6	−45.5	
P value vs. placebo	0.64	<0.001	0.01	0.006	
Secondary end points					
Least-squares mean no. of peripheral attacks/wk	0.44	0.11	0.19	0.15	0.60
Difference vs. placebo (95% CI)	−0.16 (−0.51 to 0.18)	−0.49 (−0.76 to −0.22)	−0.41 (−0.68 to −0.14)	−0.45 (−0.70 to −0.20)	
Percent difference vs. placebo	−27.4	−81.8	−68.3	−74.7	
Least-squares mean no. of abdominal attacks/wk†	0.42	0.14	0.34	0.37	0.35
Difference vs. placebo (95% CI)	0.07 (−0.24 to 0.37)	−0.21 (−0.45 to 0.03)	−0.01 (−0.25 to 0.22)	0.02 (−0.21 to 0.24)	
Percent difference vs. placebo	18.5	−60.2	−4.2	4.3	
Patients who were attack-free — %	0	43	21	39	9
Difference vs. placebo — percentage points (95% CI)	−9 (−21 to 3)	34 (5 to 62)	12 (−12 to 37)	30 (4 to 55)	
Least-squares mean percent of attack-free days‡	82.6	92.1	88.0	83.8	74.0
Difference vs. placebo (95% CI)	8.6 (−4.0 to 21.3)	18.1 (8.1 to 28.0)	14.0 (4.1 to 24.0)	9.8 (0.6 to 19.1)	
Percent difference vs. placebo	11.7	24.4	19.0	13.3	

* Confirmed attacks were adjudicated and confirmed by the independent clinical end-point adjudication panel. The effective dosing interval is the date of the first dose plus 7 days to the last dose on day 28 (or 24 hours after the last dose of the trial regimen, whichever was earlier). The number of confirmed attacks that occurred during the first 7 days that were not included in the effective dosing interval was 7 in the 62.5-mg group, 5 in the 125-mg group, 4 in the 250-mg group, 11 in the 350-mg group, and 17 in the placebo group. Differences in adjusted least-squares means are shown (active treatment minus placebo). Percent difference was calculated as (difference/placebo)×100. An analysis-of-covariance model included terms of trial regimen and adjusted qualifying attack rate. One patient each in the 250-mg and placebo groups underwent randomization but did not receive a trial regimen and did not enter any diary data. CI denotes confidence interval.
† Abdominal attacks included those with swelling in the stomach or gut or with any symptom of nausea, vomiting, or abdominal pain.
‡ The percent of attack-free days is the proportion of days during the intervention period without a confirmed attack.

Figure 1. Change from Baseline at Day 29 in Scores on the Angioedema Quality of Life Questionnaire (AE-QoL) in the Intention-to-Treat Population.

The least-squares mean change from baseline for each trial group is shown. An analysis-of-covariance model included terms of trial regimen and adjusted qualifying attack rate. The intention-to-treat population included all the patients who underwent randomization. The AE-QoL total score and scores for each of the four domains range from 0 to 100, with higher scores indicating greater impairment. The horizontal line represents the minimal clinically important difference (MCID) for the total score, a 6-point reduction.[13] There were significant differences between patients who received BCX7353 at a dose of 125 mg daily and those who received placebo in the total score (difference in least-squares mean change from baseline, –24.5 points; P<0.001) and across three of the four domains: functioning (–26.7 points, P=0.002), fatigue and mood (–11.6 points, P=0.054), fears and shame (–33.8 points, P<0.001), and food (–24.4 points, P=0.006). There was also a significant difference between the 250-mg dose group and the placebo group in the functioning score (difference, –20.3 points; P=0.02). No other differences (BCX7353 vs. placebo) were significant. T bars indicate standard errors.

a finding consistent with the exposure data. The drug effect on kallikrein inhibition was correlated with exposure ($r^2=0.75$) (data not shown).

SAFETY

Three patients who received BCX7353 at a dose of 350 mg discontinued the trial regimen owing to adverse events (Table 3). A total of 67% of all adverse events were mild in severity (grade 1). Analysis of the adverse-event profile identified a dose-related increase in the incidence of gastrointestinal events. Gastrointestinal events were most commonly reported in the 350-mg dose group (44%) and 250-mg dose group (50%), with considerably fewer in the 125-mg dose group (29%) and 62.5-mg dose group (14%); the incidence in the 62.5-mg dose group was similar to that in the placebo group (18%). Gastrointestinal events were generally mild (grade 1, 75% of treated patients with such events) to moderate

(grade 2, 20%) in severity, although one patient in the 350-mg dose group had severe (grade 3) abdominal pain that led to discontinuation of the trial regimen (Table 3). Only mild (grade 1) gastrointestinal events occurred in the 125-mg and 62.5-mg dose groups.

A total of three patients who received BCX7353 had mild (grade 1) or moderate (grade 2) liver-related adverse events (one patient in the 250-mg dose group and one in the 350-mg dose group had grade 1 events, and one patient in the 350-mg dose group had a grade 2 event). Two additional patients in the 350-mg dose group had a grade 3 or grade 4 liver-enzyme abnormality: one patient had a grade 3 elevation in the γ-glutamyltransferase level (3.4 times the upper limit of the normal range), and the other patient had a grade 3 elevation in the alanine aminotransferase level (3.9 times the upper limit of the normal range) and a grade 4 elevation in the γ-glutamyltransferase

Figure 2. Dose of BCX7353, Concentration–Time Profile at Steady State, and Plasma Kallikrein Inhibition.

On day 14, serial blood samples were drawn for pharmacokinetic analysis (Panel A) before dose administration and at the following time points: 1, 2, 3, 4, 5, 6, 8, and 24 hours after dose administration. For the 24-hour post-dose samples, patients returned to the clinic and had the blood sample drawn before dose administration on day 15. On day 29, a trough pharmacokinetic blood sample was drawn. However, if serial pharmacokinetic samples could not be obtained on day 14, steady-state serial pharmacokinetic samples were obtained on day 28, with a 24-hour pharmacokinetic sample drawn at the scheduled visit on day 29. The plasma samples for determination of BCX7353 concentrations were analyzed with the use of a validated liquid chromatography–mass spectrometry assay. In this trial, none of the patients who received 62.5 mg of BCX7353 met or exceeded the target of 4 times the EC_{50} across the 24-hour dosing interval, whereas 9 of 14 patients who received 125 mg met or exceeded the target, and all the patients who received 250 mg or 350 mg exceeded the target concentration. Kallikrein inhibition (Panel B) was measured in an ex vivo fluorogenic assay.[10] The assay uses ellagic acid to activate the contact pathway and convert prekallikrein to kallikrein. A fluorogenic substrate that is specific for the enzyme (N-carbobenzyloxy-phenylalanine-arginine-7-amino-4-methylcoumarin) was used to measure its amidolytic activity. Owing to the dilution in the sample preparation for the assay, the BCX7353 concentration in the ex vivo assay represents one quarter of the corresponding in vivo plasma concentration. The results of kallikrein activity were expressed as percent inhibition as compared with the activity present in the predose plasma sample on day 1. At doses of 250 mg or 350 mg of BCX7353, maximum kallikrein inhibition was approximately 90%, and inhibition of more than 75% was maintained throughout the 24-hour dosing period. Maximum kallikrein inhibition with 125 mg of BCX7353 was approximately 60%, with 45% inhibition at 24 hours, whereas at a dose of 62.5 mg, maximum inhibition was approximately 30%, with 8% kallikrein inhibition at 24 hours.
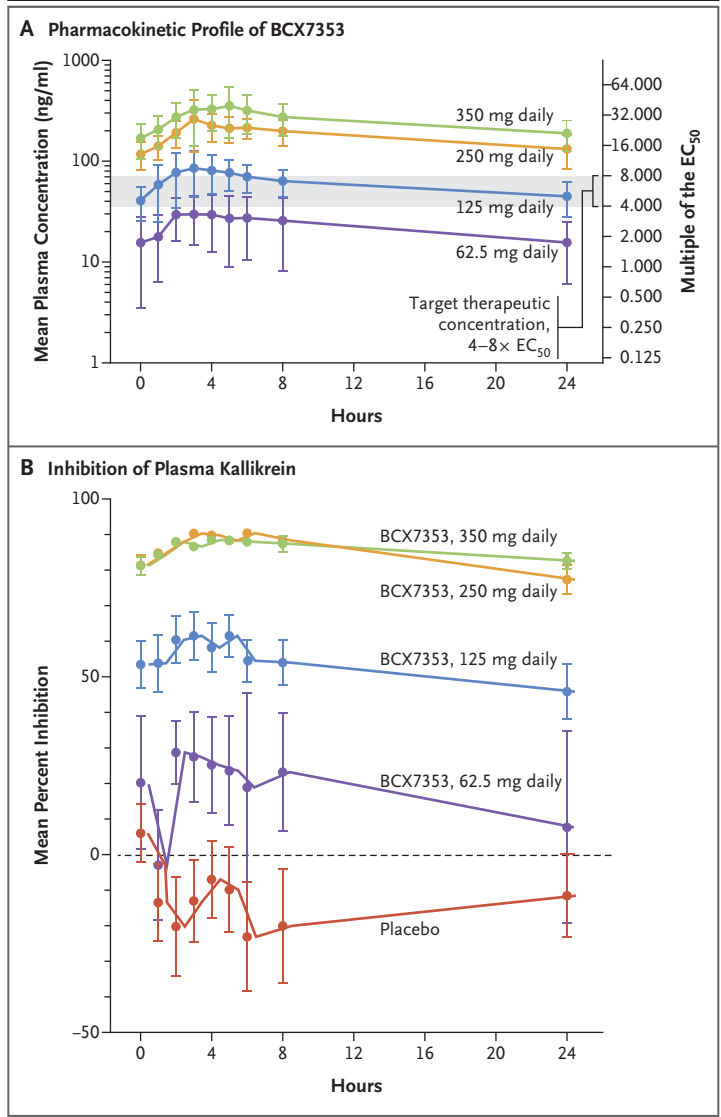
level (10.7 times the upper limit of the normal range). Both patients had elevated liver-enzyme levels at baseline.

All five of these patients had extensive histories of previous androgen use. These patients' laboratory values returned to normal or baseline without intervention once dosing was completed. No liver-related adverse events or grade 3 or 4 liver-enzyme abnormalities were observed at the 125-mg or 62.5-mg doses.

DISCUSSION

The APeX-1 trial showed that BCX7353 at doses of 125 mg or more administered orally once daily resulted in markedly lower rates of angioedema attacks than placebo. An apparent U-shaped dose response was observed in the primary end point, with the highest treatment effect observed at the 125-mg dose: the attack rate was 73.8% lower than with placebo, and 43% of patients were attack-free.

The data suggest that the efficacy of the BCX7353 doses of 250 mg and 350 mg was probably masked by gastrointestinal adverse events that may have been misattributed as early symptoms of abdominal angioedema attacks. Only the 125-mg dose group had a lower rate of abdominal attacks than the placebo group, whereas all groups that received BCX7353 at doses of 125 mg or more had lower rates of peripheral attacks than the placebo group (difference, 68%

Table 3. Summary of Adverse Events (Safety Population).*

Event	BCX7353				Placebo (N=22)
	62.5 mg (N=7)	125 mg (N=14)	250 mg (N=14)	350 mg (N=18)	
	number of patients (percent)				
Any adverse event	4 (57)	7 (50)	11 (79)	14 (78)	15 (68)
Any serious adverse event	0	0	1 (7)†	0	0
Grade 3 adverse event considered to be related to the trial regimen‡	0	0	0	1 (6)	0
Adverse event leading to discontinuation of the trial regimen	0	0	0	3 (17)§	0
Most commonly reported adverse events¶					
Any gastrointestinal adverse event	1 (14)	4 (29)	7 (50)	8 (44)	4 (18)
Abdominal pain‖	1 (14)	3 (21)	2 (14)	3 (17)	0
Diarrhea	0	0	2 (14)	4 (22)	2 (9)
Flatulence	0	0	0	2 (11)	0
Nausea	0	0	3 (21)	3 (17)	0
Vomiting	0	0	0	2 (11)	0
Fatigue	1 (14)	0	0	2 (11)	1 (5)
Nasopharyngitis	2 (29)	0	1 (7)	5 (28)	6 (27)
Liver-related event**	0	0	1 (7)	2 (11)	0
Headache	2 (29)	2 (14)	1 (7)	1 (6)	4 (18)

* The safety population included all randomly assigned patients who received a dose of a trial regimen. Shown are events that started after the date and time of the first dose to the last dose plus 30 days. Adverse events were graded according to the Division of Microbiology and Infectious Diseases Adult Toxicity Table (November 2007): grade 1 indicated mild, grade 2 moderate, grade 3 severe, and grade 4 life-threatening. No patient had a grade 4 adverse event.

† A grade 2 gastrointestinal infection was considered to be unrelated to the trial regimen. Abdominal symptoms of gastrointestinal infection were similar to those in several previous (non–hereditary angioedema attack) episodes occurring during the previous 3 years that resulted in severe vomiting and diarrhea and an overnight hospitalization for intravenous fluids and antiemetics.

‡ In addition to the adverse events that were considered by the investigator to be related to the trial regimen, two patients in the 350-mg dose group had grade 3 adverse events that were considered to be unrelated to the trial regimen. One patient had a persistent grade 3 elevation in the alanine aminotransferase (ALT) level that was present at baseline, before the first dose of the trial regimen. One patient had a grade 3 gastrointestinal infection 1 week after completion of the last dose of the trial regimen.

§ Reasons for discontinuation were grade 1 liver disorder (in one patient), grade 3 elevation in the ALT level (in one patient), and grade 3 abdominal pain and grade 2 vomiting (in one patient). The liver disorder was considered to be probably related to the trial regimen and occurred after a case of grade 1 gastroenteritis; the patient discontinued the trial regimen on day 18. Increases in levels of ALT (1.9 times the upper limit of the normal range [ULN] [grade 1]), γ-glutamyltransferase (5.4 times the ULN [grade 3]), and alkaline phosphatase (1.6 times the ULN [grade 1]) were observed, although levels of aspartate aminotransferase and bilirubin remained within the normal range. The grade 3 elevation in the ALT level was considered unlikely to be related to the trial regimen, and although the patient's ALT level was less than 2 times the ULN at screening (44 IU per liter, or 1.3 times the ULN [grade 1]) (i.e., the patient met the inclusion criteria), the patient entered the trial with an elevated ALT level (123 IU per liter, or 3.7 times the ULN [grade 3]) at baseline that remained at grade 2 or higher during the trial; the patient discontinued the trial regimen on day 23. The patient had received danazol (100 mg) before trial entry. The abdominal pain and vomiting were considered to be possibly related to the trial regimen and occurred two times before the third episode that resulted in discontinuation of the trial regimen on day 12.

¶ Shown are adverse events that occurred in at least 10% of the patients enrolled in any trial group. Events are listed according to preferred term.

‖ Abdominal pain included the following reported terms: abdominal pain, abdominal pain upper, abdominal pain lower, abdominal discomfort, and dyspepsia.

** Liver-related events were reported as liver disorder, elevation in the ALT level, and abnormal liver-function test. The elevation in the ALT level (in the 350-mg dose group) was mild in severity and was considered to be possibly related to the trial regimen. The ALT level increased from a normal baseline value (29 IU per liter) to grade 1 at day 29 (51 IU per liter) and at follow-up (86 IU per liter). The ALT level was within the normal range at day 65. The abnormal liver-function test (in the 250-mg dose group) was mild in severity, was considered to be possibly related to the trial regimen, and represented a 1-grade shift in the ALT level (grade 2 at baseline to grade 3 from day 7 through the end of the intervention). At follow-up on day 43, the ALT level was near baseline (107 IU per liter); approximately 1 month later, the ALT level had decreased to within the normal range. The liver disorder (in the 350-mg dose group) resulted in discontinuation of the trial regimen (discussed above).

to 82%). Gastrointestinal adverse events were more common at the doses of 250 mg and 350 mg than at lower doses, and a small number of liver abnormalities were observed at the highest doses in patients with extensive previous use of androgens. The side-effect profile in this trial was consistent with a trial involving healthy volunteers, in which gastrointestinal adverse events were more commonly reported in higher BCX7353 dose groups.[10]

The effectiveness of BCX7353 was further supported by secondary efficacy end points involving a post hoc hierarchical analysis, with substantial improvements observed in patients' quality of life at the 125-mg dose level, although post hoc P values should be interpreted with some caution. The mean improvement (change from baseline) in the AE-QoL total score was greater than 4 times the minimal clinically important difference of 6 points.[13] In addition, patients who received BXC7353 at doses of 125 mg or more had lower rates of attacks resulting in treatment and a lower overall severity of illness than those who received placebo.

An analysis of attack rates according to dose and exposure in this trial supports a hypothesis that drug exposures that are maintained above a threshold concentration at least 4 times the EC_{50} correlate with clinically meaningful reductions in attack rates. In this trial, only doses of 125 mg or more provided trough concentrations at least 4.0 times the EC_{50} with corresponding significant differences in the attack rate as compared with placebo. A 62.5-mg dose provided a mean C_{tau} of 1.8 times the EC_{50}, with no significant difference in attack rate as compared with placebo (P=0.64), whereas a 125-mg dose provided a mean C_{tau} of 5.0 times the EC_{50}, with a corresponding difference in the attack rate of 73.8% as compared with placebo (P<0.001).

These data are consistent with exposure-response results from a phase 3 trial of a subcutaneous preparation of C1 inhibitor (Clinical Study for Optimal Management of Preventing Angioedema with Low-Volume Subcutaneous C1-Inhibitor Replacement Therapy [COMPACT]).[14] In that trial, a clinically meaningful reduction in the frequency of attacks correlated with a trough plasma level of C1 inhibitor equal to 5.3 to 6.3 times the EC_{50} of C1 inhibitor on plasma kallikrein, which was determined by the same assay used to estimate the EC_{50} of BCX7353. In our trial, trough concentrations that were achieved at the 250-mg and 350-mg doses were 14 to 20 times the EC_{50}, which suggests that these doses are in excess of those required to achieve clinically meaningful reductions in the attack rate.

In a phase 3 study (OPuS-2) of avoralstat, a first-generation oral kallikrein inhibitor administered three times a day, efficacy in preventing angioedema attacks in patients with hereditary angioedema was not shown.[15] As compared with BCX7353, avoralstat had inferior pharmacokinetic properties, with low oral bioavailability, a short half-life, and a food effect on plasma exposure. Reported plasma concentrations of avoralstat from the OPuS-2 study were highly variable, with many time points having drug levels below the target therapeutic concentration of 4 times the EC_{50} for the drug. The results of the OPuS-2 trial further support the hypothesis that maintenance of drug levels above a threshold level for plasma kallikrein suppression is important in reducing the frequency of angioedema attacks.

In conclusion, this trial showed both proof of concept and dose response of orally administered BCX7353 in the prevention of angioedema attacks through short-lasting kallikrein inhibition. Longer studies will need to be performed to assess the safety profile of long-term dosing.

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