BACKGROUND
The next-generation cystic fibrosis transmembrane conductance regulator (CFTR) corrector VX-659, in triple combination with tezacaftor and ivacaftor (VX-659–tezacaftor–ivacaftor), was developed to restore the function of Phe508del CFTR protein in patients with cystic fibrosis.

METHODS
We evaluated the effects of VX-659–tezacaftor–ivacaftor on the processing, trafficking, and function of Phe508del CFTR protein using human bronchial epithelial cells. A range of oral VX-659–tezacaftor–ivacaftor doses in triple combination were then evaluated in randomized, controlled, double-blind, multicenter trials involving patients with cystic fibrosis who were heterozygous for the Phe508del CFTR mutation and a minimal-function CFTR mutation (Phe508del–MF genotypes) or homozygous for the Phe508del CFTR mutation (Phe508del–Phe508del genotype). The primary end points were safety and the absolute change from baseline in the percentage of predicted forced expiratory volume in 1 second (FEV₁).

RESULTS
VX-659–tezacaftor–ivacaftor significantly improved the processing and trafficking of Phe508del CFTR protein as well as chloride transport in vitro. In patients, VX-659–tezacaftor–ivacaftor had an acceptable safety and side-effect profile. Most adverse events were mild or moderate. VX-659–tezacaftor–ivacaftor resulted in significant mean increases in the percentage of predicted FEV₁ through day 29 (P<0.001) of up to 13.3 points in patients with Phe508del–MF genotypes; in patients with the Phe508del–Phe508del genotype already receiving tezacaftor–ivacaftor, adding VX-659 resulted in a further 9.7-point increase in the percentage of predicted FEV₁. The sweat chloride concentrations and scores on the respiratory domain of the Cystic Fibrosis Questionnaire–Revised improved in both patient populations.

CONCLUSIONS
Robust in vitro activity of VX-659–tezacaftor–ivacaftor targeting Phe508del CFTR protein translated into improvements for patients with Phe508del–MF or Phe508del–Phe508del genotypes. VX-659 triple-combination regimens have the potential to treat the underlying cause of disease in approximately 90% of patients with cystic fibrosis. (Funded by Vertex Pharmaceuticals; VX16-659-101 and VX16-659-001 ClinicalTrials.gov numbers, NCT03224351 and NCT03029455.)
Cystic fibrosis is a serious, autosomal-recessive, multisystem disease that affects approximately 80,000 people worldwide and is associated with early death due to progressive lung disease.\(^1,2\) Cystic fibrosis is caused by diminished quantity and defective function of an epithelial anion channel, the cystic fibrosis transmembrane conductance regulator (CFTR), as a consequence of mutations in CFTR.\(^3,4\) For approximately 60% of patients with cystic fibrosis (i.e., those with two Phe508del alleles or a gating or residual-function CFTR mutation), small-molecule CFTR modulators provide therapeutic benefit by restoring CFTR activity.\(^5\) However, high levels of chloride-transport restoration have not been achieved for the majority of patients with cystic fibrosis, approximately 90% of whom are either homozygous or heterozygous for the Phe508del CFTR mutation.\(^6\) There are currently no approved CFTR modulators to treat the estimated 30% of patients who are heterozygous for the Phe508del CFTR mutation and a minimal-function CFTR mutation (Phe508del–MF genotypes), which leaves a substantial portion of the patient population without a treatment addressing the basic defect.\(^7,9\)

The Phe508del CFTR mutation causes severe processing and trafficking defects, which result in decreased quantity and function of CFTR protein at the cell surface,\(^4\) and has proven to be a challenging molecular target. Among patients who have at least one Phe508del CFTR mutation, approximately half have a second Phe508del allele (Phe508del–Phe508del genotype), and one third have a minimal-function CFTR mutation as the second allele. Minimal-function mutations, which produce either no protein or a defective protein that is unresponsive to CFTR modulators, include nonsense mutations, insertion and deletion mutations, canonical splicing mutations, and certain severe protein misfolding mutations.\(^1\) A CFTR modulator regimen that could achieve high levels of functional CFTR in patients with a Phe508del allele, regardless of whether the second CFTR allele is also responsive to CFTR modulator therapy (e.g., a second Phe508del allele) or is incapable of response (i.e., the allele contains a minimal-function mutation), would be of value.

On the basis of the well-established understanding of the molecular defects caused by the Phe508del CFTR mutation, two complementary approaches have been developed to restore chloride transport by increasing the quantity and function of Phe508del CFTR protein at the cell surface. CFTR correctors, such as lumacaftor and tezacaftor, bind the Phe508del CFTR protein, augmenting intracellular processing and trafficking and thereby increasing the amount of mature CFTR available at the cell surface. The CFTR potentiator ivacaftor increases the channel-gating activity of Phe508del CFTR protein that is delivered to the cell surface, to augment anion transport.\(^10\) With differing mechanisms of action, the combination of CFTR correctors and a CFTR potentiator is required to increase both the amount and function of Phe508del CFTR protein and is more effective than either approach alone.\(^10\)

For patients with the Phe508del–Phe508del genotype, dual combinations of a first-generation CFTR corrector (lumacaftor or tezacaftor) and ivacaftor provide both short-term and long-term clinical benefit and are now accepted as the standard of care.\(^11\) Nonetheless, these dual combinations do not fully restore function to Phe508del CFTR protein and are not sufficiently active to improve outcomes in patients with Phe508del–MF genotypes. As a consequence, there remains a need for highly effective CFTR modulation that will more adequately treat the underlying cause of disease in most patients with cystic fibrosis.\(^12,14\)

We have previously shown in vitro that a combination of two correctors with distinct binding sites on CFTR and complementary mechanisms of action can increase the amount of Phe508del CFTR protein at the cell surface to a greater extent than either corrector alone.\(^15\) On the basis of these findings, we developed next-generation CFTR correctors for use in combination with tezacaftor–ivacaftor to increase both the amount and function of Phe508del CFTR protein to a greater extent than a dual combination of a corrector and a potentiator. VX-659, a next-generation corrector, has both a different structure and a different mechanism of action. Its distinct mechanism of action is based on clear additivity on functional and biochemical assays of CFTR processing, trafficking, and function in vitro. Additivity of VX-659 to tezacaftor and ivacaftor in vitro increases Phe508del CFTR protein processing within the cell and trafficking to the cell surface. VX-659 shares some structural similarity and a mechanism of action with the next-generation corrector VX-445, described in a companion report also appearing in the *Journal*.\(^16\) Because tezacaftor and VX-659 work through different mechanisms, we hypothesized that the
combination of VX-659 and tezacaftor would increase the amount of Phe508del CFTR protein at the cell surface more than either compound alone, an effect that could be potentiated by ivacaftor to further increase chloride transport.

We evaluated the effect of the VX-659–tezacaftor–ivacaftor triple combination on the processing, trafficking, and function of Phe508del CFTR protein in airway epithelial cells, which are an important and predictive preclinical model in cystic fibrosis.10 These cells were obtained from patients with Phe508del–MF or Phe508del–Phe508del genotypes. We then conducted a small, phase 1 trial involving patients, followed by a larger, phase 2, proof-of-concept trial to evaluate the safety and efficacy of a range of VX-659 triple-combination doses in patients with one or two Phe508del alleles. For patients with Phe508del–MF genotypes, there is no available CFTR modulator, whereas for patients with the Phe508del–Phe508del genotype the standard of care includes a two-drug CFTR modulator combination; as a consequence, the trial designs differed according to patient genotype.

**METHODS**

**PRECLINICAL DEVELOPMENT**

The in vitro pharmacologic characteristics of VX-659 were evaluated in biochemical and functional studies alone and in dual and triple combination with tezacaftor–ivacaftor with the use of human bronchial epithelial cells derived from patients with cystic fibrosis and Phe508del–MF genotypes (four donors) or the Phe508del–Phe508del genotype (three donors). For details, see the Methods section in the Supplementary Appendix (available with the full text of this article at NEJM.org).

**CLINICAL DEVELOPMENT**

**Trial Design and Oversight**

A randomized, placebo-controlled, double-blind, multicenter, phase 1 trial (VX16-659-001) was conducted to evaluate preliminary pharmacokinetics and initial safety in a small group of patients with cystic fibrosis (Fig. 1). This trial enrolled patients 18 years of age or older with cystic fibrosis and Phe508del–MF CFTR genotypes and was conducted at nine sites in the United Kingdom from April 2017 through July 2017. Patients were randomly assigned, in a 2:1 ratio, to receive 14 days of active treatment with oral VX-659 at a dose of 120 mg every 12 hours in combination with tezacaftor (100 mg once daily) and ivacaftor (150 mg every 12 hours) or to receive oral triple placebo in parallel cohorts. Triple placebo was used as the control in this and subsequent trials involving patients with Phe508del–MF genotypes because these patients currently lack an approved standard-of-care CFTR modulator regimen.

After the phase 1 trial, a randomized, parallel-track, placebo- or active-controlled, double-blind, multicenter, dose-ranging, phase 2 trial (VX16-659-101) was conducted with multiple dose levels of VX-659 (Fig. 1). This trial enrolled patients 18 years of age or older with cystic fibrosis and Phe508del–MF or Phe508del–Phe508del CFTR genotypes. The phase 2 trial was conducted at 48 sites in the United States, the United Kingdom, Ireland, and Israel from August 2017 through February 2018.

The VX16-659-101 trial had three parts. In the first part, patients with Phe508del–MF genotypes were assigned to receive 4 weeks of active treatment with oral VX-659 at doses of 80, 240, or 400 mg once daily in triple combination with tezacaftor (100 mg once daily) and ivacaftor (150 mg every 12 hours) or to receive oral triple placebo in parallel cohorts. A randomization ratio of 1:2:2:1 was used to decrease the variability and thus increase the accuracy of the within-group efficacy estimate for the intermediate- and high-dose levels, at which the efficacy was predicted to plateau. This intervention period was followed by 4 days of only tezacaftor–ivacaftor (washout of VX-659) for those who had received active triple-combination therapy.

In the second part of the trial, patients with the Phe508del–Phe508del genotype were recruited. Because dual-combination CFTR modulators are the standard of care for these patients, tezacaftor–ivacaftor was used as the active control. Patients received 4 weeks of oral tezacaftor–ivacaftor during a run-in period, followed by randomization (in a 2:1 ratio) to 4 weeks of either oral VX-659 (400 mg once daily) or matched placebo in combination with tezacaftor–ivacaftor. This intervention period was followed by 4 weeks of tezacaftor–ivacaftor only (washout of VX-659). Baseline status for efficacy end points was assessed at the time of randomization, when all patients with the Phe508del–Phe508del genotype had been receiving daily tezacaftor–ivacaftor for at least 28 days.
A

**Phase 1 (Phe508del–minimal function)**

- **Screening period (4 wk)**
  - VX-659 + TEZ–IVA: N=9
  - Triple placebo: N=3

- **Intervention period (2 wk)**
  - VX-659 + TEZ–IVA: N=9
  - Triple placebo: N=3

- **Safety follow-up period (3–5 wk)**
  - VX-659 + TEZ–IVA: N=9
  - Triple placebo: N=3

**Phase 2 (Phe508del–minimal function)**

- **Run-in period (4 wk)**
  - VX-659, 400 mg, +TEZ–IVA: N=22
  - VX-659, 240 mg, +TEZ–IVA: N=20
  - VX-659, 80 mg, +TEZ–IVA: N=11

- **Period 1 (4 wk)**
  - VX-659, 400 mg, +TEZ–IVA: N=22
  - TEZ–IVA: N=10

- **Period 2 (4 wk)**
  - VX-659, 400 mg, +TEZ–IVA: N=19
  - TEZ–IVA: N=9

**Phase 2 (Phe508del–Phe508del)**

- **Run-in period (4 wk)**
  - TEZ–IVA: N=18
  - Placebo + TEZ–IVA: N=11

- **Period 1 (4 wk)**
  - VX-659, 400 mg, +TEZ–VX-651: N=19
  - TEZ–IVA: N=9

**Phase 2 (Phe508del–minimal function: VX-561)**

- **Screening period (4 wk)**
  - VX-659, 400 mg, +TEZ–VX-561: N=19
  - TEZ–IVA: N=9

- **Intervention period (2 wk)**
  - VX-659, 400 mg, +TEZ–VX-561: N=19

- **Safety follow-up period (4 wk)**
  - VX-659, 400 mg, +TEZ–VX-561: N=19

B

**Phase 1 (Phe508del–minimal function)**

- 12 Patients were enrolled and underwent randomization
  - 3 Were assigned to placebo
  - 9 Were assigned to VX-659 + TEZ–IVA
  - 3 Received ≥1 dose
  - 9 Received ≥1 dose
  - 3 Completed 2 wk of trial regimen
  - 9 Completed 2 wk of trial regimen

**Phase 2 (Phe508del–minimal function)**

- 63 Patients were enrolled and underwent randomization
  - 10 Were assigned to placebo
  - 10 Received ≥1 dose
  - 10 Completed 4 wk of trial regimen

**Phase 2 (Phe508del–Phe508del)**

- 29 Patients were enrolled and underwent randomization
  - 11 Were assigned to placebo + TEZ–IVA
  - 18 Were assigned to VX-659, 400 mg, +TEZ–IVA
  - 11 Received ≥1 dose
  - 18 Received ≥1 dose
  - 11 Completed 4 wk of trial regimen
  - 18 Completed 4 wk of trial regimen

**Phase 2 (Phe508del–minimal function: VX-561)**

- 25 Patients were enrolled and underwent randomization
  - 6 Were assigned to placebo
  - 6 Received ≥1 dose
  - 6 Completed 4 wk of trial regimen

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The third part of the VX16-659-101 trial involved patients with Phe508del–MF genotypes who received VX-659–tezacaftor–VX-561 or triple placebo. VX-561 is a deuterated form of the CFTR potentiator ivacaftor that is administered orally once daily; the nondeuterated form of ivacaftor is administered twice daily. In the phase 2 trial, the dose of VX-659 was 120 mg every 12 hours; tezacaftor–ivacaftor (TEZ–IVA) was administered at a dose of 100 mg of TEZ once daily and 150 mg of IVA every 12 hours. In the phase 2 trial, the dose of VX-659 shown is the dose that was administered once daily; TEZ–IVA was administered at the same dose as in the phase 1 trial. A subgroup of patients with the Phe508del–minimal function genotype received VX-561 once daily instead of IVA every 12 hours. In the phase 2 trial, period 1 is the 4-week intervention period; period 2 is a washout of VX-659, as applicable. Panel B shows the number of patients in each intervention group who received at least one dose of the study regimen and the number who completed the intervention period of 2 weeks (phase 1 trial) or 4 weeks (phase 2 trial).

The primary end points were safety and side-effect profile, based on the assessment of adverse events, clinically significant laboratory-test results, standard 12-lead electrocardiograms, vital signs, and spirometric measurements. 

**Phase 1 Trial Assessments**

The primary end points were safety, side-effect profile, and the absolute change in the percentage of predicted FEV₁ from baseline through day 29. Safety assessments included adverse events, clinical laboratory values, electrocardiograms, and vital signs. Summarized safety data were based on events observed during the interval from the first dose of the trial regimen to 28 days after the last dose of the trial regimen or the end of the trial, whichever occurred first. Secondary end points included the absolute change in sweat chloride concentration from baseline through day 29 and the absolute change from baseline in Cystic Fibrosis Questionnaire–Revised (CFQ-R) respiratory domain score at day 29. Scores range from 0 to 100, with higher scores indicating a higher patient-reported quality of life with re-
spect to respiratory status (minimal clinically important difference, 4 points).

**STATISTICAL ANALYSIS**

For both the phase 1 and 2 trials, the intention-to-treat population, which included all the patients who received at least one dose of the trial regimen and had the intended CFTR mutation, was used to analyze the efficacy outcomes. The safety population, which included all the patients who received at least one dose of the trial regimen, was used for all safety analyses. For the phase 1 trial, efficacy end points, including the absolute change from baseline in the percentage of predicted FEV₁ and sweat chloride concentration, were summarized with the use of descriptive statistics.

For the phase 2 trial, safety data from patients receiving VX-659–tezacaftor–ivacaftor who had Phe508del–MF or Phe508del–Phe508del genotypes were pooled for the safety analysis. The primary end point of the absolute change from baseline in the percentage of predicted FEV₁ was analyzed separately for each part of the trial with the use of a mixed-effects model for repeated measures, with the change from baseline in the percentage of predicted FEV₁ at days 15 and 29 as the dependent variable. The within-group estimate along with the 95% confidence intervals and the corresponding P values were calculated as the primary analysis. Secondary end points, including sweat chloride concentration and CFQ-R respiratory domain score, were analyzed similarly, and the within-group estimate and 95% confidence intervals were calculated. The widths of the confidence intervals have not been adjusted for multiple comparisons and the intervals should not be used to infer definitive treatment effects. The primary analysis for all efficacy end points was the within-group comparison. (Additional details of the statistical analysis are provided in the Methods section and Table S1 in the Supplementary Appendix.)

**RESULTS**

**IN VITRO EFFICACY IN PRIMARY HUMAN BRONCHIAL EPITHELIAL CELLS**

VX-659 improved the processing and trafficking of Phe508del CFTR protein in human bronchial epithelial cells derived from patients with Phe508del–MF or Phe508del–Phe508del genotypes, as shown by higher steady-state levels of mature CFTR protein than in controls (Fig. 2A and 2B). The combination of VX-659–tezacaftor without or with ivacaftor resulted in significantly higher steady-state levels of mature CFTR protein than either compound alone. Ivacaftor slightly reduced the magnitude of CFTR processing correction by VX-659–tezacaftor in human bronchial epithelial cells from patients with the Phe508del–Phe508del genotype; however, correction by the triple combination was greater than that observed after treatment with dual combinations of either tezacaftor–ivacaftor or VX-659–ivacaftor.

Untreated human bronchial epithelial cells from patients with Phe508del–MF or Phe508del–Phe508del genotypes showed minimal CFTR-mediated chloride transport, a finding consistent with little to no functional CFTR at the cell surface (Fig. 2C). Treatment with VX-659 alone produced a modest increase in chloride transport, which was further augmented by combination with tezacaftor, ivacaftor, or both. The greatest increase in chloride transport was observed with the triple combination of VX-659–tezacaftor–ivacaftor (Fig. 2C). Together, these in vitro studies provided the rationale for investigating a triple-combination regimen of two correctors, VX-659 and tezacaftor, with the CFTR potentiator ivacaftor in patients with cystic fibrosis with either Phe508del–MF or Phe508del–Phe508del genotypes.

**VX-659–TEZACAFTOR–IVACAFTOR IN PATIENTS WITH CYSTIC FIBROSIS**

**Patient Population**

As part of a phase 1 trial (VX16-659-001) in which safety and pharmacokinetics were initially shown in healthy volunteers, we evaluated preliminary safety and pharmacokinetics of VX-659–tezacaftor–ivacaftor in a cohort of 12 patients with cystic fibrosis with Phe508del–MF genotypes. Patients were randomly assigned to receive either VX-659–tezacaftor–ivacaftor (9 patients) or triple placebo (3 patients) (Fig. S1 and Tables S2 through S5 in the Supplementary Appendix).

In the subsequent phase 2 trial, 117 patients with cystic fibrosis and Phe508del–MF genotypes (88 patients) or the Phe508del–Phe508del genotype (29 patients) were randomly assigned to receive either a VX-659 triple-combination regimen containing ivacaftor (71 patients) or VX-561 (19 patients) or a control regimen (27 patients)
Figure 2. In Vitro Effect of VX-659 Alone or in Combination with TEZ, IVA, or TEZ–IVA.

Panels A and B show the effect of VX-659 on the processing and trafficking of Phe508del CFTR protein in human bronchial epithelial (HBE) cells derived from patients with Phe508del–minimal function (Phe508del–MF) or Phe508del–Phe508del genotypes. Panel A shows the results of representative immunoblotting from three independent experiments. Panel B shows the quantitative assessment of that data through densitometry findings pooled from three independent experiments, with six replicates in each experiment. Data are presented as mean relative intensities normalized to calnexin, a control for protein loading. The compound concentrations used were as follows: 1.4 μM of VX-659, 18 μM of TEZ, and 1 μM of IVA in the presence of 10 mg per milliliter of human serum albumin. The letter a represents P<0.05 for the comparison with vehicle, b P<0.05 for the comparison with TEZ–IVA, and c P<0.05 for the comparison with VX-659 in unpaired t-tests.

Panel C shows an assessment of chloride transport in HBE cells treated with various combinations of TEZ (18 μM), IVA (1 μM), and VX-659 (1 μM) by means of an Ussing chamber. Data represent the mean of three or four donor bronchi, with three or four replicate experiments per donor. The letter x represents P<0.05 for the comparison with vehicle, y P<0.05 for the comparison with TEZ–IVA, and z P<0.05 for the comparison with VX-659–IVA in paired t-tests. In Panels B and C, T bars indicate standard errors.

(Fig. 1). A total of 115 patients completed the 4-week intervention period.

Baseline age, sex, percentage of predicted FEV1, and sweat chloride concentration were generally well balanced across genotype and intervention groups (Table 1, and Table S6 [VX-659–tezacaftor–ivacaftor] and Table S7 [VX-659–tezacaftor–VX-561] in the Supplementary Appendix). For patients with Phe508del–MF genotypes who received VX-659–tezacaftor–ivacaftor or triple placebo, the mean baseline CFQ-R respiratory domain score was numerically higher in the placebo group than in the active-treatment groups (difference, 12.6 to 14.1 points). Consequently, analysis of the change in the CFQ-R respiratory domain score was performed both with and without adjustment for the baseline score (see the Methods section in the Supplementary Appendix for details).

Phase 1 Trial

The most common adverse events with VX-659–tezacaftor–ivacaftor were cough, infective pulmonary exacerbation of cystic fibrosis, and productive cough (each occurred in two patients). Oral candidiasis occurred in one patient in the VX-659–tezacaftor–ivacaftor group and one in the placebo group. Most adverse events were mild or moderate, and none led to interruption or discontinuation of the trial regimen. Two serious adverse events of infective pulmonary exacerbation of cystic fibrosis occurred in the VX-659–tezacaftor–ivacaftor group, one mild and one moderate in severity; both events occurred after the end of dosing (Table S3 in the Supplementary Appendix).
Table 1. Baseline Demographic and Clinical Characteristics of the Phe508del–Minimal Function and Phe508del–Phe508del Cohorts.*

<table>
<thead>
<tr>
<th>Characteristic</th>
<th>Phe508del–Minimal Function</th>
<th>Phe508del–Phe508del†</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Triple Placebo (N = 10)</td>
<td>VX-659, 80 mg, + TEZ–IVA (N = 11)</td>
</tr>
<tr>
<td>Male sex — no. (%)</td>
<td>6 (60)</td>
<td>4 (36)</td>
</tr>
<tr>
<td>Age — yr</td>
<td>26.6±6.0</td>
<td>32.0±11.7</td>
</tr>
<tr>
<td>Percentage of predicted FEV₁</td>
<td>53.9±12.0</td>
<td>57.9±10.8</td>
</tr>
<tr>
<td>Sweat chloride — mmol/liter</td>
<td>98.2±13.3</td>
<td>102.7±7.0</td>
</tr>
<tr>
<td>CFQ-R respiratory domain score‡</td>
<td>77.2±15.1</td>
<td>63.1±18.5</td>
</tr>
</tbody>
</table>

* Plus–minus values are means ±SD. Patients with Phe508del–minimal function genotypes were heterozygous for the Phe508del CFTR mutation and a minimal-function CFTR mutation, and those with the Phe508del–Phe508del genotype were homozygous for the Phe508del CFTR mutation. All baseline characteristics were tested for balance between groups with the use of Fisher’s exact test for categorical variables and the F test for all other variables. There were no significant differences between the intervention groups. The dose of VX-659 is the dose that was administered once daily. Tezacaftor–ivacaftor (TEZ–IVA) was administered at a dose of 100 mg of TEZ once daily and 150 mg of IVA every 12 hours. FEV₁ denotes forced expiratory volume in 1 second.

† The baseline characteristics of patients with the Phe508del–Phe508del genotype were assessed after a 4-week run-in with TEZ–IVA.

‡ Scores on the respiratory domain of the Cystic Fibrosis Questionnaire–Revised (CFQ-R) range from 0 to 100, with higher scores indicating a higher patient-reported quality of life with respect to respiratory status (minimal clinically important difference, 4 points).

VX-659–tezacaftor–ivacaftor therapy resulted in substantial improvements in the percentage of predicted FEV₁ and sweat chloride concentration (Fig. S1 and Table S5 in the Supplementary Appendix). These improvements were not seen in patients who received triple placebo.

Phase 2 Safety

Most patients who received VX-659–tezacaftor–ivacaftor (57 of 71 patients [80%]), triple placebo (9 of 10 [90%]), or tezacaftor–ivacaftor (9 of 11 [82%]) reported having at least one adverse event (Table 2, and Table S8 in the Supplementary Appendix). Among those receiving VX-659–tezacaftor–ivacaftor who had an adverse event, the maximum severity was mild or moderate for the majority of patients (53 of 57 patients [93%]). The most commonly observed adverse events (>10% occurrence in the pooled VX-659–tezacaftor–ivacaftor groups) were cough, infective pulmonary exacerbation of cystic fibrosis, headache, oropharyngeal pain, and increased sputum production. Four adverse events in patients receiving VX-659–tezacaftor–ivacaftor therapy were considered to be severe by site investigators (Table 2). Serious adverse events were reported in 7 patients (10%) who received VX-659–tezacaftor–ivacaftor, 3 patients (30%) who received triple placebo, and 2 patients (18%) who received tezacaftor–ivacaftor control. Most serious adverse events were infective pulmonary exacerbations of cystic fibrosis requiring hospitalization. No deaths occurred during the trial.

There were no adverse events leading to discontinuation of the trial regimen in any patients who received VX-659–tezacaftor–ivacaftor. One patient interrupted VX-659–tezacaftor–ivacaftor because of a rash. The rash resolved, and the patient resumed the trial regimen and completed the trial without recurrence of the rash.

Two patients who received VX-659–tezacaftor–ivacaftor had elevations of alanine aminotransferase or aspartate aminotransferase levels (>3 to ≤5 times the upper limit of the normal range in one patient and >5 to ≤8 times the upper limit of the normal range in one patient) without concurrent elevation in the bilirubin level (Table S9 in the Supplementary Appendix). The elevations resolved during the trial without interruption of the trial regimen. There was no evidence of bronchoconstriction after VX-659 triple-combination dosing in any genotype or lung-function category as assessed by post-dose spirometry. The safety profile of VX-659–tezacaftor–VX-561 was similar to that of VX-659–tezacaftor–ivacaftor (Tables S10 and S11 in the Supplementary Appendix).

Phase 2 Efficacy

All doses of VX-659–tezacaftor–ivacaftor resulted in significant improvement in the percentage of predicted FEV₁, as compared with the respec-
Table 2. Summary of Adverse Events.

<table>
<thead>
<tr>
<th>Event</th>
<th>Phe508del–Minimal Function</th>
<th>Phe508del–Phe508del</th>
<th>Any VX-659</th>
<th>N=71*</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>VX-659, 80 mg, + TEZ–IVA</td>
<td>VX-659, 240 mg, + TEZ–IVA</td>
<td>VX-659, 400 mg, + TEZ–IVA</td>
<td>Placebo, + TEZ–IVA</td>
</tr>
<tr>
<td></td>
<td>(N = 11)</td>
<td>(N = 20)</td>
<td>(N = 22)</td>
<td>(N = 11)</td>
</tr>
<tr>
<td>Any adverse event†</td>
<td>9 (90)</td>
<td>10 (91)</td>
<td>15 (75)</td>
<td>17 (77)</td>
</tr>
<tr>
<td>Maximum severity of adverse event‡</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Mild</td>
<td>5 (56)</td>
<td>7 (70)</td>
<td>6 (35)</td>
<td>2 (22)</td>
</tr>
<tr>
<td>Moderate</td>
<td>4 (44)</td>
<td>3 (30)</td>
<td>8 (59)</td>
<td>4 (44)</td>
</tr>
<tr>
<td>Severe</td>
<td>0</td>
<td>0</td>
<td>1 (6)</td>
<td>3 (33)</td>
</tr>
<tr>
<td>Serious adverse event</td>
<td>3 (30)</td>
<td>1 (9)</td>
<td>4 (20)</td>
<td>1 (5)</td>
</tr>
<tr>
<td>Adverse event leading to interruption of the trial regimen</td>
<td>0</td>
<td>0</td>
<td>1 (5)</td>
<td>0</td>
</tr>
<tr>
<td>Adverse event leading to discontinuation of the trial regimen</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
</tbody>
</table>

The denominator is the number of patients with at least one adverse event.

* Shown are patients with Phe508del–minimal function or Phe508del–Phe508del genotypes who received VX-659.
† A patient with multiple events within a category was counted only once in that category.
‡ No events were considered life-threatening.
tive within-group baseline values in patients with Phe508del–MF or Phe508del–Phe508del genotypes (Table 3). These improvements were observed at the first assessment on day 15 and were sustained at day 29 (Fig. 3). Changes indicating a beneficial effect of VX-659–tezacaftor–ivacaftor were observed for the secondary end points of sweat chloride concentration and the CFQ-R respiratory domain score, findings consistent with the magnitude of improvement in the percentage of predicted FEV1 (Fig. 3 and Table 3). Improvement in the CFQ-R respiratory domain score at day 29 was observed with or without adjustment for the baseline score (Table 3 [without adjustment], and Table S12 in the Supplementary Appendix). Data on the absolute and relative changes in FEV1, as a percentage of the predicted value and in liters are provided in Table S12 in the Supplementary Appendix. Individual patient data for the absolute change from baseline in the percentage of predicted FEV1 and sweat chloride concentration at day 29 are included in Figure S2 in the Supplementary Appendix. Similar improvements in all these end points were observed in patients with Phe508del–MF genotypes who received VX-659–tezacaftor–VX-561 (Table S13 and Fig. S3 in the Supplementary Appendix).

### DISCUSSION

In our in vitro to in vivo studies, the next-generation CFTR corrector VX-659 in combination with...
VX-659–Tezacaftor–Ivacaftor in Cystic Fibrosis

Absolute change in percentage of predicted FEV$_1$

**A** Phe508del–Minimal Function

<table>
<thead>
<tr>
<th>Group</th>
<th>Day 1</th>
<th>Day 15</th>
<th>Day 29</th>
</tr>
</thead>
<tbody>
<tr>
<td>TC, 400 mg</td>
<td>22</td>
<td>22</td>
<td>21</td>
</tr>
<tr>
<td>TC, 240 mg</td>
<td>20</td>
<td>18</td>
<td>18</td>
</tr>
<tr>
<td>TC, 80 mg</td>
<td>11</td>
<td>11</td>
<td>11</td>
</tr>
<tr>
<td>Placebo</td>
<td>10</td>
<td>9</td>
<td>10</td>
</tr>
</tbody>
</table>

Absolute change in sweat chloride (mmol/liter)

**A** Phe508del–Minimal Function

<table>
<thead>
<tr>
<th>Group</th>
<th>Day 1</th>
<th>Day 15</th>
<th>Day 29</th>
</tr>
</thead>
<tbody>
<tr>
<td>TC, 400 mg</td>
<td>10</td>
<td>-10</td>
<td>-20</td>
</tr>
<tr>
<td>TC, 240 mg</td>
<td>9</td>
<td>-10</td>
<td>-20</td>
</tr>
<tr>
<td>TC, 80 mg</td>
<td>9</td>
<td>-10</td>
<td>-20</td>
</tr>
<tr>
<td>Placebo</td>
<td>9</td>
<td>-10</td>
<td>-20</td>
</tr>
</tbody>
</table>

Absolute change in CFQ-R Respiratory Domain Score

**A** Phe508del–Minimal Function

<table>
<thead>
<tr>
<th>Group</th>
<th>Day 1</th>
<th>Day 15</th>
<th>Day 29</th>
</tr>
</thead>
<tbody>
<tr>
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</tr>
<tr>
<td>TC, 240 mg</td>
<td>20</td>
<td>18</td>
<td>19</td>
</tr>
<tr>
<td>TC, 80 mg</td>
<td>11</td>
<td>11</td>
<td>11</td>
</tr>
<tr>
<td>Placebo</td>
<td>10</td>
<td>10</td>
<td>10</td>
</tr>
</tbody>
</table>

**B** Phe508del–Phe508del

<table>
<thead>
<tr>
<th>Group</th>
<th>Day 1</th>
<th>Day 15</th>
<th>Day 29</th>
</tr>
</thead>
<tbody>
<tr>
<td>TC, 400 mg</td>
<td>18</td>
<td>18</td>
<td>18</td>
</tr>
<tr>
<td>TEZ–IVA</td>
<td>11</td>
<td>11</td>
<td>11</td>
</tr>
</tbody>
</table>

Absolute change in sweat chloride (mmol/liter)

**B** Phe508del–Phe508del

<table>
<thead>
<tr>
<th>Group</th>
<th>Day 1</th>
<th>Day 15</th>
<th>Day 29</th>
</tr>
</thead>
<tbody>
<tr>
<td>TC, 400 mg</td>
<td>19</td>
<td>19</td>
<td>19</td>
</tr>
<tr>
<td>Placebo</td>
<td>9</td>
<td>9</td>
<td>9</td>
</tr>
</tbody>
</table>

Absolute change in CFQ-R Respiratory Domain Score

**B** Phe508del–Phe508del

<table>
<thead>
<tr>
<th>Group</th>
<th>Day 1</th>
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<td>TEZ–IVA</td>
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<td>11</td>
<td>11</td>
</tr>
</tbody>
</table>
percentage of predicted FEV1 of 9.7 points. The resulted in an average absolute increase in the percentage of predicted FEV1 were accompanied with administration of VX-659. Most notably, patients with Phe508del–MF genotypes, who have only one responsive Phe508del allele and no currently available CFTR modulator therapy, had improvements in lung function, sweat chloride concentration, and CFQ-R respiratory domain score. In addition, patients with the Phe508del–Phe508del genotype who were already receiving tezacaftor–ivacaftor had additional improvements in these outcome measures after the administration of VX-659. Thus, these trials of VX-659–tezacaftor–ivacaftor served as the way for larger and longer trials of the clinical usefulness of triple-combination CFTR modulator regimens for patients with at least one Phe508del allele was predicted to be similar to to that of ivacaftor in patients with gating mutations, and the subsequent clinical-trial results substantiated this prediction.

These trials of relatively short duration revealed no concerning safety signals across the genotype groups and range of VX-659 triple-combination doses studied. The majority of patients in cystic fibrosis and Phe508del–MF or Phe508del–Phe508del genotypes. An additive response was seen for these events. Overall, the safety profile of VX-659 triple-combination therapy was similar to that seen in previous studies of CFTR modulators. In addition, two other components of the triple combination, tezacaftor and ivacaftor, have well-characterized safety profiles in short-term and long-term studies.

In conclusion, we found that VX-659 triple-combination therapy led to improvements in all evaluated efficacy outcomes in patients with cystic fibrosis and Phe508del–MF or Phe508del–Phe508del genotypes. An additive response was seen for VX-659 triple-combination therapy in comparison with the dual combination of tezacaftor–ivacaftor in patients with the Phe508del–Phe508del genotype. These trials provide proof of the concept that targeting the Phe508del CFTR protein with a triple-combination corrector–potentiator regimen can restore CFTR function and has the potential to represent a clinical advance for patients with cystic fibrosis who harbor either one or two Phe508del alleles, approximately 9 of every 10 patients with the disease.
Supported by Vertex Pharmaceuticals, which received funding from the Cystic Fibrosis Foundation for the development of VX-659. The National Institute for Health Research provided support to the Respiratory Clinical Research Facility at the Royal Brompton and Harefield NHS Foundation Trust and Imperial College London, London, and to the Manchester Clinical Research Facility at Wythenshawe Hospital, Manchester, United Kingdom. The National Institutes of Health provided grant support to the University of Alabama at Birmingham (P30DK079401, K25HL13996, and US4TR001368).

Disclosure forms provided by the authors are available with the full text of this article at NEJM.org.

APPENDIX

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REFERENCES


