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Results of a phase IIa study of VX-809, an investigational CFTR corrector compound, in subjects with cystic fibrosis homozygous for the *F508del-CFTR* mutation

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Abstract

Background—VX-809, a cystic fibrosis transmembrane conductance regulator (CFTR) modulator, has been shown to increase the cell surface density of functional *F508del-CFTR* in vitro.

Methods—A randomised, double-blind, placebo-controlled study evaluated the safety, tolerability and pharmacodynamics of VX-809 in adult patients with cystic fibrosis (n=89) who were homozygous for the *F508del-CFTR* mutation. Subjects were randomised to one of four VX-809 28 day dose groups (25, 50, 100 and 200 mg) or matching placebo.

Results—The type and incidence of adverse events were similar among VX-809- and placebo-treated subjects. Respiratory events were the most commonly reported and led to discontinuation by one subject in each active treatment arm. Pharmacokinetic data supported a once-daily oral dosing regimen. Pharmacodynamic data suggested that VX-809 improved CFTR function in at least one organ (sweat gland). VX-809 reduced elevated sweat chloride values in a dose-dependent manner (p=0.0013) that was statistically significant in the 100 and 200 mg dose groups. There was no statistically significant improvement in CFTR function in the nasal epithelium as measured by nasal potential difference, nor were there statistically significant changes in lung function or patient-reported outcomes. No maturation of immature *F508del-CFTR* was detected in the subgroup that provided rectal biopsy specimens.

Conclusions—In this study, VX-809 had a similar adverse event profile to placebo for 28 days in *F508del-CFTR* homozygous patients, and demonstrated biological activity with positive impact on CFTR function in the sweat gland. Additional data are needed to determine how improvements detected in CFTR function secondary to VX-809 in the sweat gland relate to those measurable in the respiratory tract and to long-term measures of clinical benefit. Clinical trial number NCT00865904

INTRODUCTION

Cystic fibrosis (CF) is the most common autosomal recessive lethal genetic disease in the Caucasian population, with an incidence of 1:3500 births in the USA. The most common cause of morbidity and mortality is lung disease, which is characterised by infection, inflammation and airway damage that leads to respiratory failure.[1]

CF is caused by mutations in the CF transmembrane conductance regulator (*CFTR*) gene,[2,3] which encodes the CFTR protein. CFTR is a member of the ATP-binding cassette protein family and functions as a chloride ion (Cl^-) channel and a key regulator of salt and water transport across a variety of epithelia.[4–8] The CFTR gene has >1600[9] reported disease-associated mutations, with *F508del-CFTR* being the most common. *F508del-CFTR* results from a 3 bp deletion that leads to the omission of phenylalanine at position 508 of the full-length protein.[10] The resulting F508del protein product is unstable and susceptible to rapid degradation in the 26S proteosome, with little if any *F508del-CFTR* at the plasma membrane.[11,12] The *F508del-CFTR* mutation is found in the majority of patients with CF,[13] and therefore the consequences of this mutation on the CFTR protein are important to address in therapeutic development for CF.

CFTR ‘correctors’ aim to increase the cell surface density of functional CFTR protein, resulting in improved chloride transport and decreased sodium reabsorption.[14–16] VX-809 restores *F508del-CFTR* processing and plasma membrane localisation in primary human bronchial epithelial (HBE) airway cells isolated from patients homozygous for the *F508del-CFTR* mutation, achieving ~15% of wild-type CFTR levels as measured by the amount of chloride channel function and the quantity of fully mature, C-Band CFTR.[17]

In this report, we describe the safety, tolerability, pharmaco-dynamics and pharmacokinetics (PK) of escalating doses of VX- 809 in patients with CF homozygous for the F508del-CFTR mutation compared with placebo.

METHODS

This study was a randomised, double-blind, placebo-controlled, multiple-dose, multicentre, phase IIa study. Institutional Review Board approvals and informed consents were obtained for all study subjects.

Subjects

Subjects enrolled in the study had a confirmed diagnosis of CF, accompanied by a sweat chloide value of ≥60 mmol/l. All subjects were ≥18 years of age, and were required to have the *F508del-CFTR* mutation on both alleles. At screening, subjects were required to have a forced expiratory volume in 1 s (FEV_1) of at least 40% of predicted normal for age, gender and height (Knudson standards).[18]

Study design

Subjects were enrolled into two cohorts, group A and group B. Group A subjects were randomised to receive VX-809 once daily at doses of 25 mg or 50 mg, or placebo in a 2:2:1 randomisation ratio. After 15 group A subjects completed 28 days of treatment, a safety review was conducted by an independent Data Monitoring Committee. Enrolment in group B began following the Data Monitoring Committee review. In group B, subjects were randomised to receive VX-809 at doses of 100 mg or 200 mg, or placebo in a 2:2:1 ratio for 28 days. All study sites, the patients, and the sponsor remained blinded to treatment assignment throughout the study.

End points

The primary objective, evaluation of safety and tolerability of VX-809, was assessed by adverse events (AEs), haematology, clinical chemistry, urinalysis, ECG, vital signs and physical examinations. Secondary objectives included evaluation of the pharmacodynamic impact of VX-809 on CFTR function. Measures of CFTR activity included sweat chloride and nasal potential difference (NPD).[19] The latter was considered optional. Other secondary end points included spirometry to measure pulmonary function (ie, FEV_1, forced expiratory flow 25−75% ($FEF_{25–75\%}$) and forced vital capacity (FVC)) and the CF Questionnaire-Revised (CFQ-R), a disease-specific patient-reported outcome.[20] The minimal clinically important difference in respiratory domain (MCID) is improvement ≥4.[21] The details of CFTR biomarkers and pharmacokinetic analysis are included in the Supplemental Methods section.

Statistical analyses

Safety data were analysed primarily using descriptive statistics, and efficacy data were analysed primarily using analysis of covariance (ANCOVA) models. The planned total sample size of 90 subjects provided a study power of >97% to detect a 20 mmol/l reduction in sweat chloride concentration from baseline, and a probability of 99% to observe at least 1 adverse event in the study. All subjects who received at least one dose of study drug were included in the analyses.

RESULTS

Subjects

A total of 109 adult subjects with CF who were homozygous for the *F508del-CFTR* mutation (based on screening medical history) were screened and 89 were randomised to one of four VX-809 dose groups (25 mg (n=18), 50 mg (n=18), 100 mg (n=17), 200 mg (n=19)) or to placebo (n=17). Confirmatory genotyping identified one subject randomised to the 50 mg VX-809 group who was heterozygous for *F508del-CFTR* despite a medical history indicating homozygosity. Safety and efficacy data obtained from this subject were included in the final analysis. The identity of this subject's non-F508del allele was not available (based on parameters of the informed consent document for that study site). Baseline characteristics are provided in table 1. Median sweat chloride values and FEV_1 percentage predicted were similar between the different groups and consistent with the values reported in the literature for patients homozygous for the *F508del-CFTR* mutation. The baseline median sweat chloride was 103.5 mmol/l and the median baseline FEV_1 was 71% predicted. The study groups were well matched except for trends towards less severe lung disease in the placebo and 25 mg dose groups.

Safety and AE profile

The incidence of AEs was similar between dose groups (table 2). Respiratory events were the most commonly reported type of AE, with cough occurring in 46% of VX-809-treated subjects and 41% of placebo-treated subjects. There was no difference in the incidence of physician-diagnosed pulmonary exacerbations between VX-809- and placebo-treated subjects (17% of VX-809 subjects compared with 12% of placebo subjects; p=0.62). AEs that occurred in more than one subject in any VX-809 treatment arm are included in table 2. Eight AEs were considered severe, including fatigue, sinus congestion, musculoskeletal discomfort, two events of cough and three events of acute pulmonary exacerbation. All pulmonary exacerbations were considered serious.

Four of 89 (5%) subjects discontinued study drug during the study, one subject in each of the VX-809 dose groups. No placebo-treated subjects withdrew from treatment. All discontinuations were due to the occurrence of respiratory AEs. There were no clinically significant changes in laboratory findings during the study.

PK results

Plasma concentrations of VX-809 were measured by a fully validated bioanalytical method. PK parameters estimated at the four dose levels of VX-809 on days 1 and 28 are summarised in table 3. VX-809 appeared to be slowly absorbed from the gut in subjects with CF with median time to reach maximum concentration (t_{max}) values ranging from 3 to 4 h across all doses on days 1 and 28. Based on predose PK samples collected from day 1 to day 28, plasma steady-state concentrations appeared to be reached by day 7 at all dose levels with a mean accumulation ratio ranging from 1.7 to 2.0 on day 28 (based on area under the cuve (AUC) over the 24 h dosing period, $AUC_{0–24\ h}$). The time to reach steady state and the extent of accumulation observed in this study were consistent with a terminal half-life approaching 24 h. The estimated mean values of this PK parameter in this study were slightly lower (range from 15 to 18 h) which may be explained by the low number of PK samples collected beyond 24 h on the last dosing day. The estimated oral VX-809 clearance (ie, volume of plasma purified per unit of time) at steady state was relatively low (<2% hepatic blood flow) and comparable for all doses. The estimated volume of distribution of VX-809 at steady state (V_z/F) suggested a potential diffusion of the drug into tissues.

Maximum (C_{max}) and total ($AUC_{0–24\ h}$) exposure to VX-809 increased proportionally with VX-809 dose increases from 25 to 200 mg over the 28 days of treatment. The intersubject variability observed for VX-809 in subjects with CF appeared to be moderate and comparable across all doses for C_{max} (30–40%) but with a trend to increase slightly with dose for $AUC_{0–24\ h}$ (from 40% to 60%) on day 28.

CFTR bioactivity

The effects of VX-809 on *F508del-CFTR* processing and function were evaluated throughout the trial. The assessments included measurements of sweat chloride concentration (in all subjects) and NPD (in 71 subjects). The relative amounts of the immature (B-Band) and mature (C-Band) forms of *F508del-CFTR*, which are markers of protein maturation beyond the endoplasmic reticulum and Golgi, were measured in biopsies of the rectal epithelium in 34 subjects.

Sweat chloride

Figure 1 shows changes in sweat chloride values, which were reduced in a dose-dependent manner (p=0.0013) in VX-809- treated subjects. The reduction in sweat chloride values was rapid and sustained, with measurable changes seen within 7 days of VX-809 dosing (figure 1A). The mean change from baseline in sweat chloride concentration (mmol/l) after 7 days was 2.2 in the placebo group, −0.5 in the 25 mg group, −3.7 in the 50 mg group (95% CI −7.1 to −0.28, p=0.03), −2.3 in the 100 mg group and −6.6 in the 200 mg group (95% CI −10.27 to −2.83, p=0.0008). At day 28, the mean treatment differences from baseline (−placebo) for the 25, 50, 100 and 200 mg groups were +0.10, −4.61, −6.13 (95% CI −12.25 to −0.01) and −8.21 mmol/l (95% CI −14.33 to −2.10), respectively (figure 1B). These differences were statistically significant versus placebo for the 100 mg (p<0.05) and 200 mg (p<0.01) groups. Following 7 days of drug washout, mean sweat chloride values returned to approximately pretreatment levels (figure 1A).

The number of subjects whose sweat chloride values were reduced by the threshold (‘responder’) values was evaluated using predefined and posthoc study criteria. Using a posthoc responder criterion of ≥10 mmol/l reduction, six subjects (38%) in the 200 mg group and six subjects (40%) in the 100 mg group responded compared with none of the placebo-treated patients (p=0.02 for both VX-809 treatment groups). In the 25 and 50 mg dose groups, there were no subjects classified as responders by either criterion. Using the predefined responder criterion, only one subject (6.3%, in the 200 mg dose group) had a ≥20 mmol/l response to VX-809 (p=NS).

Nasal potential difference

Seventy-one subjects underwent NPD measurements throughout the study. There was no significant change in CFTRdependent NPD parameters (chloride or sodium transport) in any of the dose groups. Supplementary figure 1 summarises the CFTR chloride ion transport parameters (change in chloride-free isoproterenol response) for the VX-809 treatment groups from baseline to day 28 after removing the change in the placebo group (treatment difference).

CFTR B- to C-Band maturation

Thirty-four subjects provided rectal biopsy tissue for evaluation of *F508del-CFTR* maturation. Among the 33 subjects who were homozygous for the F508del-CFTR allele, no mature C-Band was detected (supplementary figure 2). Only one subject in the highest dose group (VX-809 200 mg) provided rectal biopsy tissue.

Clinical outcomes

Spirometry—There were no significant changes in lung function (FEV_1, FVC, $FEF_{25–75\%}$) in any of the dose groups, including changes in percentage predicted values versus baseline and placebo, or raw measures of litre flow (data not shown). Figure 2 shows the percentage change relative to baseline FEV_1 (percentage predicted) for each study group across all study visits. After 28 days of treatment, the mean percentage change from baseline in FEV_1 percentage predicted was 0.07, −2.46, −2.15, 0.32 and 0.47 in the placebo, 25, 50, 100 and 200 mg dose groups, respectively (p=NS).

Patient-reported outcomes—Supplementary table 1 summarises the change in CFQ-R measures (day 28 vs baseline) across the placebo- and VX-809-treated subjects. There were no clear or sustained changes in the respiratory domain or in any other subdomains of the CFQ-R in any dose group. After 28 days, the respiratory domain score in the placebo group increased by 4.5. After 28 days of treatment in the 25, 50, 100 and 200 mg dose groups, the changes were −5.2, −6.3, −1.30 and +2.2, respectively.

DISCUSSION

These data provide evidence of a safety and tolerability profile sufficient to support further clinical evaluation of VX-809 in subjects with CF homozygous for the *F508del-CFTR* mutation, the most common CF mutation worldwide. Over a dosing range of 25–200 mg, study participants reported symptoms and AEs that were similar to those in the placebo group, and similar to those commonly found in adult patients with CF. One subject in each of the four dose groups developed a pulmonary exacerbation during the 28 day period of treatment, and the incidence of symptoms such as cough was similar across the dose groups and to that of the placebo group.

Sweat chloride concentration measurements demonstrated modest, statistically significant improvements in VX-809-treated subjects. This effect was dose dependent, sustained during the treatment period and rapidly reversed following discontinuation of VX-809. This suggests that VX-809 increased the chloride transport function of CFTR in the sweat gland, and supports the idea that VX-809 is bioactive in the sweat gland of patients homozygous for the *F508del-CFTR* mutation. These results also support the hypothesis that small molecule correction of *F508del-CFTR* is feasible. The results provide support for the use of previously obtained CFTR biomarker data sets for study planning of new CFTR modulators. Using this paradigm, the treatment effects on sweat chloride reported with the CFTR potentiator VX-770 in patients with CF with the *G551D-CFTR* mutation were successfully used to power the current study to detect improvements in sweat chloride (within-group comparisons and with placebo). It is not clear if the reductions in sweat chloride in this study achieved a peak effect (figure 1), and this raises the question of whether higher doses of VX-809 may produce greater effects on *F508del-CFTR*. The peak corrective effect of VX-809 on F508del-CFTR in primary HBE cells has been estimated at 3 μM (F. Van Goor, personal communication, 2011). Such concentrations of VX-809 were observed in the plasma of patients in the 100 and 200 mg dose cohorts at steady state. However, the drug exposure levels achieved in patient target tissues at these doses of VX-809 is not known.

The other biomarkers of CFTR activity (NPD, immunoblot from rectal biopsy tissue) failed to demonstrate changes over the course of VX-809 treatment. This may reflect the trial's lack of power to detect a predicted change in these measurements based on in vitro results with VX-809 in F508del/F508del primary HBE cells (maximum of ~15% of wild-type CFTR function).[17] The intrinsically greater variability of NPD measurement (potentially compounded by the large number of NPD sites used in this trial (n=19)) led to a greater likelihood of Type 2 error in analysing biomarker responses. To minimise a Type 2 error

and attain an 80% power to detect a chloride-free isoproterenol response approximating 30% of that seen with VX-770 in patients with CF with the *G551D-CFTR* mutation, the current study would have required >50 subjects per study group undergoing the NPD.[1722]

In vitro studies also suggest that measurement of the effect of VX-809 on CFTR may be below the lower limits of sensitivity of immunoblot assays performed on small biopsy samples.[17] Pretrial studies indicated that the assay developed here was capable of detecting ~10% of C-Band CFTR relative to wild-type levels (ex vivo dilution experiments using human rectal biopsy specimens obtained from non-CF study subjects; see online supplement, figure 2B). The sensitivity of this assay to detect *F508del-CFTR* correction relative to functional measurements in vivo is unknown, but in vitro studies in a variety of model systems suggest that biochemical detection of *F508del-CFTR* correction may be less robust than functional measures in intact epithelia.[1723] The failure to detect VX-809 effects on *F508del-CFTR* outside of the sweat gland could also reflect tissue-specific differences in VX-809 bioavailability and/or F508del-CFTR responsiveness to VX-809 treatment.

No improvements in clinical outcomes, including lung function and quality of life (CFQ-R), were observed over the course of this study. The trial was not powered to detect such improvements in these measures, and longer or larger trials of VX-809 (alone or in combination with CFTR potentiators) may be necessary to determine the clinical effect of these CFTR modulators.

Previous trials of systemically dosed CFTR modulators have frequently described discordant effects on CFTR-dependent biomarkers and pulmonary outcome measures. For example, PTC124 has been shown to have detectable bioactivity by NPD over 2 weeks of treatment in patients with CF possessing premature termination codons in CFTR, while sweat chloride measurements remained unchanged.[24] Improvements in lung function and cough frequency were not observed until months of treatment were completed.[25] Systemic gentamicin has also been shown to suppress PTCs[26] and improve NPD parameters in two pilot studies, but effects on sweat chloride were predominantly limited to a subset of patients with the Y122X mutation.[2728] Using a separate CFTR modulator strategy, Rubenstein and colleagues treated *F508del-CFTR* homozygous patients with CF with the *F508del-CFTR* modulator 4-phenyl butyrate. Improvements in NPD-dependent chloride secretion were seen, but there were no effects on sweat chloride.[29] The reasons for discrepancies in CFTR biomarkers across different CFTR modulator strategies are not clear, but suggest that organ effects may vary due to tissue drug availability, CFTR regulation in different cell types or the responsiveness of mutant CFTR across different tissue compartments and modulator strategies. These uncertainties should be considered in future study planning, as should the selection and continued development of CFTR biomarkers to demonstrate study drug bioactivity.

In conclusion, the results provide support for the continued evaluation of VX-809 in patients with CF with the *F508del-CFTR* mutation. This study demonstrated that VX-809 has an acceptable safety profile. It also provided evidence of the effect of VX-809 on improving CFTR function based on dose-dependent reductions in *F508del-CFTR* activity in the sweat gland using a CFTR corrector administered orally to subjects with CF, and further investigation is warranted to evaluate the potential of this therapeutic strategy to increase *F508del-CFTR* activity.

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

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References

1. Strausbaugh SD, Davis PB. Cystic fibrosis: a review of epidemiology and pathobiology. Clin Chest Med. 2007; 28:279–288. [PubMed: 17467547]
2. Kerem B, Rommens JM, Buchanan JA, et al. Identification of the cystic fibrosis gene: genetic analysis. Science. 1989; 245:1073–1080. [PubMed: 2570460]
3. Riordan JR, Rommens JM, Kerem B, et al. Identification of the cystic fibrosis gene: cloning and characterization of complementary DNA. Science. 1989; 245:1066–1073. [PubMed: 2475911]
4. Berger HA, Anderson MP, Gregory RJ, et al. Identification and regulation of the cystic fibrosis transmembrane conductance regulator-generated chloride channel. J Clin Invest. 1991; 88:1422–1431. [PubMed: 1717515]
5. Knowles M, Gatzy J, Boucher R. Relative ion permeability of normal and cystic fibrosis nasal epithelium. J Clin Invest. 1983; 71:1410–1417. [PubMed: 6853720]
6. Li C, Ramjeesingh M, Wang W, et al. ATPase activity of the cystic fibrosis transmembrane conductance regulator. J Biol Chem. 1996; 271:28463–28468. [PubMed: 8910473]
7. Quinton PM. Chloride impermeability in cystic fibrosis. Nature. 1983; 301:421–422. [PubMed: 6823316]

8. Poulsen JH, Fischer H, Illek B, et al. Bicarbonate conductance and pH regulatory capability of cystic fibrosis transmembrane conductance regulator. Proc Natl Acad Sci USA. 1994; 91:5340–5344. [PubMed: 7515498]
9. Report of a joint meeting of WHO/ECFTN/ICF(M)A/ECFS. Geneva: WHO; The Molecular Genetic Epidemiology of Cystic Fibrosis. Human Genetics Programme WHO/HGN/CF/WG/04.02. http://www.who.int/genomics/publications/en/
10. [accessed Jan 2011] Cystic Fibrosis Mutation Database. http://www.genet.sickkids.on.ca/cftr
11. Cheng SH, Gregory RJ, Marshall J, et al. Defective intracellular transport and processing of CFTR is the molecular basis of most cystic fibrosis. Cell. 1990; 63:827–834. [PubMed: 1699669]
12. Denning GM, Anderson MP, Amara JF, et al. Processing of mutant cystic fibrosis transmembrane conductance regulator is temperature-sensitive. Nature. 1992; 358:761–764. [PubMed: 1380673]
13. Bobadilla JL, Macek M, Fine JP, et al. Cystic fibrosis: a worldwide analysis of CFTR mutationsdcorrelation with incidence data and application to screening. Hum Mutat. 2002; 19:575–606. [PubMed: 12007216]
14. Van Goor F, Hadida S, Grootenhuis PD, et al. Rescue of CF airway epithelial cell function in vitro by a CFTR potentiator, VX-770. Proc Natl Acad Sci USA. 2009; 106:18825–18830. [PubMed: 19846789]
15. Pedemonte N, Lukacs GL, Du K, et al. Small-molecule correctors of defective DeltaF508-CFTR cellular processing identified by high-throughput screening. J Clin Invest. 2005; 115:2564–2571. [PubMed: 16127463]
16. Gelman MS, Kannegaard ES, Kopito RR. A principal role for the proteasome in endoplasmic reticulum-associated degradation of misfolded intracellular cystic fibrosis transmembrane conductance regulator. J Biol Chem. 2002; 277:11709–11714. [PubMed: 11812794]
17. Van Goor F, Hadida S, Grootenhuis PD. VX-809, a CFTR corrector, increases the cell surface density of functional F508del-CFTR in pre-clinical models of cystic fibrosis [abstract]. Pediatr Pulmonol. 2009; 44:154. Abstract No. S9.4.
18. Knudson RJ, Lebowitz MD, Holberg CJ, et al. Changes in the normal maximal expiratory flowevolume curve with growth and aging. Am Rev Respir Dis. 1983; 127:725–734. [PubMed: 6859656]
19. Rowe SM, Accurso FJ, Clancy JP. Detection of cystic fibrosis transmembrane conductance regulator activity in early-phase clinical trials. Proc Am Thorac Soc. 2007; 4:387–398. [PubMed: 17652506]
20. Quittner AL, Buu A, Messer MA, et al. Development and validation of The Cystic Fibrosis Questionnaire in the United States: a health-related quality-of-life measure for cystic fibrosis. Chest. 2005; 128:2347–2354. [PubMed: 16236893]
21. Quittner AL, Modi AC, Wainwright C, et al. Determination of the minimal clinically important difference scores for the Cystic Fibrosis Questionnaire-Revised respiratory symptom scale in two populations of patients with cystic fibrosis and chronic Pseudomonas aeruginosa airway infection. Chest. 2009; 135:1610–1618. [PubMed: 19447923]
22. Accurso FJ, Rowe SM, Clancy JP, et al. Effect of VX-770 in persons with cystic fibrosis and the G551D-CFTR mutation. N Engl J Med. 2010; 363:1991–2003. [PubMed: 21083385]
23. Bronsveld I, Mekus F, Bijman J, et al. Chloride conductance and genetic background modulate the cystic fibrosis phenotype of Delta F508 homozygous twins and siblings. J Clin Invest. 2001; 108:1705–1715. [PubMed: 11733566]
24. Kerem E, Hirawat S, Armoni S, et al. Effectiveness of PTC124 treatment of cystic fibrosis caused by nonsense mutations: a prospective phase II trial. Lancet. 2008; 372:719–727. [PubMed: 18722008]
25. Wilschanski M, Miller LL, Shoseyov D, et al. Chronic ataluren (PTC124) treatment of nonsense mutation cystic fibrosis. Eur Respir J. 2011; 38:59–69. [PubMed: 21233271]
26. Rowe SM, Clancy JP. Pharmaceuticals targeting nonsense mutations in genetic diseases: progress in development. BioDrugs. 2009; 23:165–174. [PubMed: 19627168]
27. Sermet-Gaudelus I, Renouil M, Fajac A, et al. In vitro prediction of stop-codon suppression by intravenous gentamicin in patients with cystic fibrosis: a pilot study. BMC Med. 2007; 5:5. [PubMed: 17394637]

28. Clancy JP, Bebok Z, Ruiz F, et al. Evidence that systemic gentamicin suppresses premature stop mutations in patients with cystic fibrosis. Am J Respir Crit Care Med. 2001; 163:1683–1692. [PubMed: 11401894]

29. Rubenstein RC, Zeitlin PL. A pilot clinical trial of oral sodium 4-phenylbutyrate (Buphenyl) in deltaF508-homozygous cystic fibrosis patients: partial restoration of nasal epithelial CFTR functiondeltaF508-homozygous cystic fibrosis patients: partial restoration of nasal epithelial CFTR function. Am J Respir Crit Care Med. 1998; 157:484–490. [PubMed: 9476862]

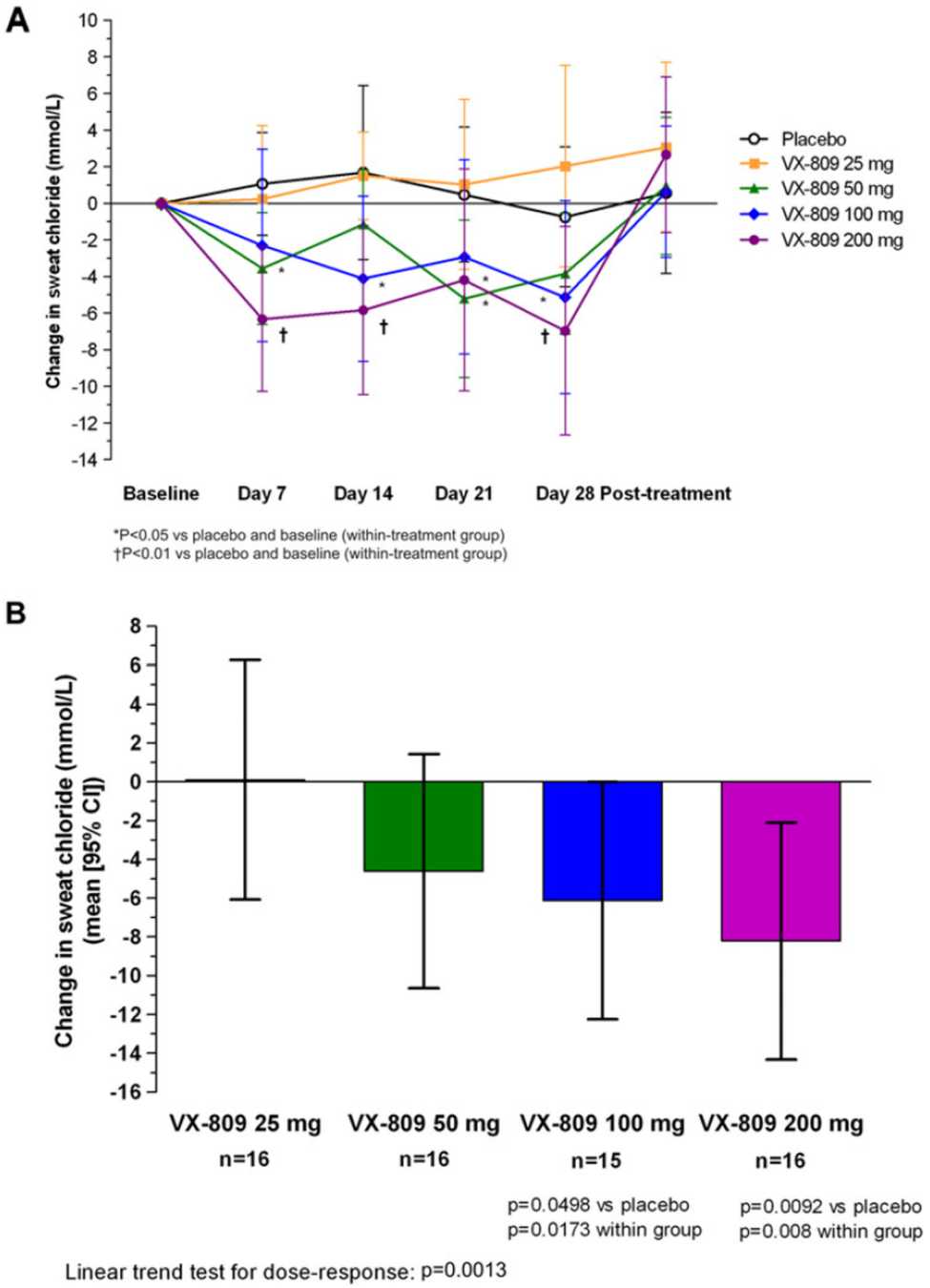

Figure 1.

(A)Change in sweat chloride measurements from baseline for placebo- and VX-809-treated subjects. Mean values are shown (±95% CI, based on analysis of covariance (ANCOVA) analysis) predose, at weekly intervals over the course of treatment, and 1 week following discontinuation of study drug. (B) Sweat chloride change from baseline to day 28 treatment, difference versus placebo (mean (95% CI, based on ANCOVA analysis)). Sweat Cl^- changes were seen as early as day 7 of treatment (data not shown), and reached statistical significance for the 100 and 200 mg dose groups ($p<0.05$ and $p<0.01$, respectively).

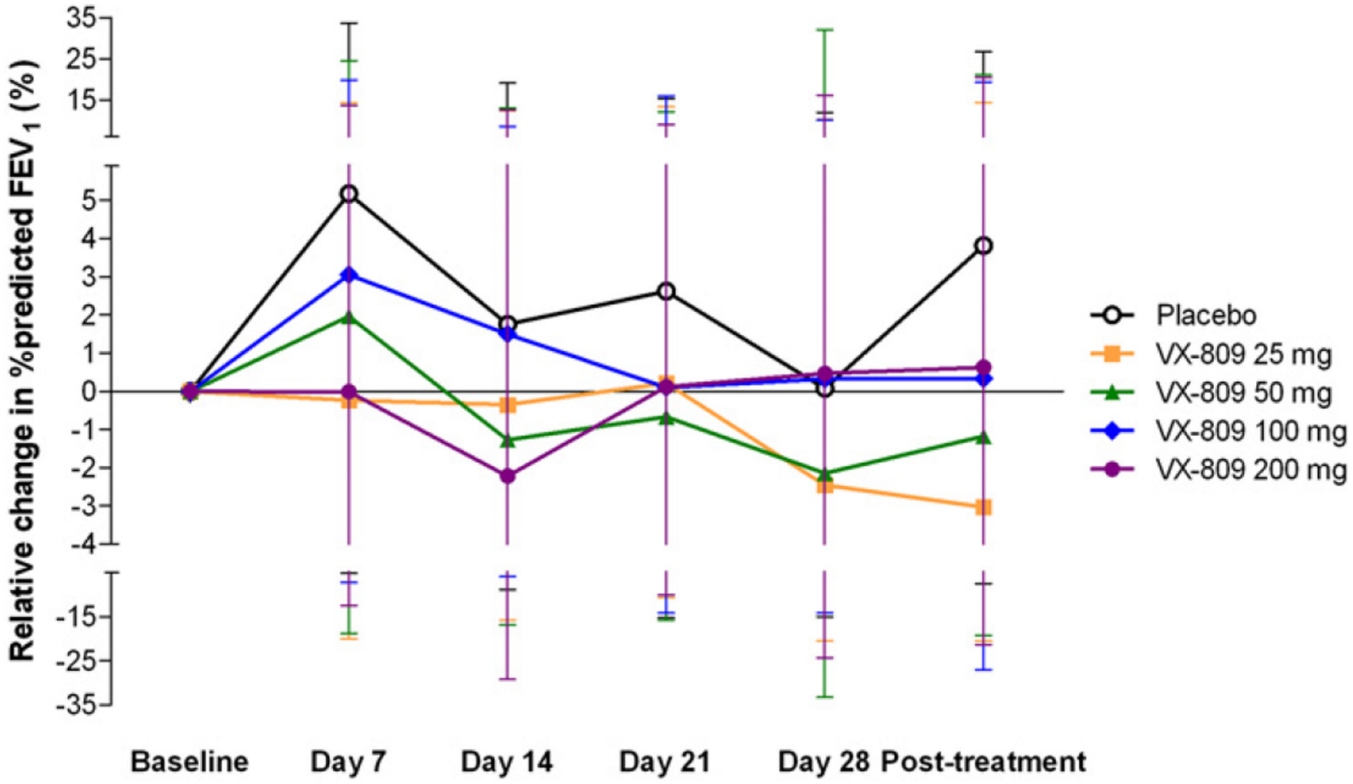

Figure 2.

Percentage change from baseline in forced expiratory volume in 1 s (FEV_1) % predicted (95% CI, based on analysis of covariance (ANCOVA) analysis). No significant changes in FEV_1 compared with baseline or placebo were seen for any of the VX-809 dose groups over the course of the study.

Table 1

Baseline demographic and clinical characteristics of the study subjects

		VX-809				
Characteristic	**Placebo n=17**	**25 mg n=18**	**50 mg n=18**	**100 mg n=17**	**200 mg n=19**	**Total n=89**
Male, n (%)	11 (65)	9 (50)	9 (50)	12 (71)	12 (63)	53 (60)
Caucasian, n (%)	17 (100)	18 (100)	18 (100)	17 (100)	19 (100)	89 (100)
Age, years, median (range)	28 (19–49)	25.5 (18–50)	24.5 (19–49)	26 (18–54)	25 (18–42)	26 (18–54)
BMI, kg/m^2, median (range)	23 (19–31)	22 (16–34)	22 (19–31)	23 (16–31)	21 (19–27)	22 (16–34)
FEV_1 %predicted, median (range)	78.4 (48.8–124.9)	78.4 (34.2–104.5)	61.5 (35.2–120.7)	61.5 (40.0–128.3)	68.2 (37.9–99.1)	71 (34.2–128.3)
Sweat chloride, mmol/l, median (range)	106.5 (80.0–125.5)	100 (86.0–109.0)	102.3 (76.0–120.0)	106 (66.0–129.0)	98.3 (72.0–122.5)	103.5 (66.0–129.0)
NPD—dchloride-free iso, mV, median (range)	1.55 (−10.6 * to 8.6)	1.6 (−3.7 to 9.5)	2.48 (−19.6 * to 11.1)	0.98 (−7.4 to 6.1)	1.33 (−7.4 to 6.3)	1.48 (−19.6 to 11.1)

* High confidence analysis removed two outliers but did not change efficacy analysis.

BMI, body mass index; FEV_1, forced expiratory volume in 1 s; NPD, nasal potential difference.

Table 2

Frequency of occurrence of adverse events occurring in more than one subject in any VX-809 treatment group (listed alphabetically by MedRA term)

Adverse event, n (%)	Placebo (n=17)	VX-809 25 mg (n=18)	VX-809 50 mg (n=18)	VX-809 100 mg (n=17)	VX-809 200 mg (n=19)	Total (n=45)
Cough	7 (41.2)	10 (55.6)	6 (33.3)	7 (41.2)	10 (52.6)	40 (88.9)
Headache	3 (17.6)	4 (22.2)	5 (27.8)	2 (11.8)	5 (26.3)	19 (42.2)
Rales	1 (5.9)	6 (33.3)	2 (11.1)	3 (17.6)	3 (15.8)	15 (33.3)
Productive cough	3 (17.6)	2 (11.1)	0	4 (23.5)	6 (31.6)	15 (17.8)
Dyspnoea	1 (5.9)	5 (27.8)	3 (16.7)	2 (11.8)	4 (21.1)	15 (33.3)
Pulmonary exacerbation *	2 (11.8)	4 (22.2)	2 (11.1)	2 (11.8)	4 (21.1)	14 (31.1)
Fatigue	2 (11.8)	3 (16.7)	3 (16.7)	2 (11.8)	3 (15.8)	13 (28.9)
Fever	2 (11.8)	2 (11.1)	1 (5.6)	1 (5.9)	5 (26.3)	11 (24.4)
Nasal congestion	3 (17.6)	2 (11.1)	1 (5.6)	2 (11.8)	2 (10.5)	10 (22.2)
Wheezing	3 (17.6)	1 (5.6)	4 (22.2)	1 (5.9)	0	9 (20)
Diarrhoea	3 (17.6)	3 (16.7)	1 (5.6)	2 (11.8)	0	9 (20)
Oropharyngeal pain	3 (17.6)	0	3 (16.7)	0	2 (10.5)	8 (17.8)
Upper respiratory tract infection	1 (5.9)	2 (11.1)	1 (5.6)	3 (17.6)	0	7 (15.6)
Sinus congestion	2 (11.8)	1 (5.6)	2 (11.1)	0	1 (5.3)	6 (13.3)
Respiration abnormal	0	1 (5.6)	1 (5.6)	0	4 (21.1)	6 (13.3)
Haemoptysis	2 (11.8)	1 (5.6)	1 (5.6)	0	2 (10.5)	6 (13.3)
Constipation	0	2 (11.1)	2 (11.1)	1 (5.9)	1 (5.3)	6 (13.3)
Abdominal pain	1 (5.9)	3 (16.7)	1 (5.6)	0	1 (5.3)	6 (13.3)
Myalgia	1 (5.9)	0	3 (16.7)	0	1 (5.3)	5 (11.1)
Post-tussive vomiting	0	0	2 (11.1)	1 (5.9)	1 (5.3)	4 (8.9)
Nausea	0	3 (16.7)	0	0	1 (5.3)	4 (8.9)
Nasopharyngitis	0	1 (5.6)	0	1 (5.9)	2 (10.5)	4 (8.9)
Dizziness	0	1 (5.6)	0	2 (11.8)	1 (5.3)	4 (8.9)
Back pain	0	2 (11.1)	1 (5.6)	0	1 (5.3)	4 (8.9)
Abdominal pain upper	1 (5.9)	0	0	1 (5.9)	2 (10.5)	4 (8.9)
Sputum abnormal	0	2 (11.1)	0	0	1 (5.3)	3 (6.7)
Epistaxis	1 (5.9)	0	0	0	2 (10.5)	3 (6.7)
C-reactive protein increased	0	1 (5.6)	0	2 (11.8)	0	3 (6.7)

Adverse event, n (%)	Placebo (n=17)	VX-809 25 mg (n=18)	VX-809 50 mg (n=18)	VX-809 100 mg (n=17)	VX-809 200 mg (n=19)	Total (n=45)
Paranasal sinus hypersecretion	0	2 (11.1)	0	0	0	2 (4.4)
Lung hyperinflation	0	0	0	2 (11.8)	0	2 (4.4)

*Physician-reported pulmonary exacerbation; coded as cystic fibrosis lung according to MedRA term.

Table 3

VX-809 Pharmacokinetic parameters following once-daily dosing of VX-809 for 28 days

		Day 1			Day 28						Day 28/day 1 AUC AR
Dose	**Parameter**	**C_{max} (μg/ml)**	**t_{max} (h)**	**$AUC_{0–24\ h}$ (μg×h/ml)**	**C_{max} (μg/ml)**	**t_{max}* (h)**	**$AUC_{0–24}$ h (μg×h/ml)**	**CL_{ss}/F (l/h)**	**V_z/F (litres)**	**$t_{1/2\lambda z}$ (h)**	
25 mg	n	18	18	18	17	17	17	17	17	17	17
	Mean	0.760	3.5*	7.20	1.10	4.0*	12.9	2.20	62.6	18.3	1.85
	SD	0.280	1.5–9.0†	2.19	0.44	2.9–12.0†	4.9	0.80	99.0	20.5	0.70
50 mg	n	18	18	18	17	17	17	17	17	17	17
	Mean	1.85	3.1*	16.6	2.66	3.1*	28.8	2.31	54.6	17.3	1.67
	SD	0.70	1.5–12.1†	6.3	1.01	0.8–9.0†	13.1	1.48	35.2	8.9	0.49
100 mg	n	17	17	17	16	16	16	16	16	16	16
	Mean	2.93	3.0*	29.0	4.62	3.1*	54.0	2.40	48.4	15.2	1.81
	SD	0.92	1.6–6.2†	11.5	1.84	1.5–6.0†	31.5	1.30	25.4	7.2	0.60
200 mg	n	19	19	19	18	18	18	18	18	18	18
	Mean	6.41	4.0*	59.6	10.3	3.0*	118	2.09	46.0	15.6	2.01
	SD	2.55	1.5–6.0†	26.3	4.5	1.5–6.1†	73	0.86	22.6	5.3	0.64

*Median.

†MineMax range.

AR, accumulation ratio; $AUC_{0–24\ h}$, area under the plasma concentration versus time curve from time 0 to 24 h; C_{max}, maximum observed drug concentration; CL_{ss}/F, oral clearance at steady state; $t_{1/2\lambda z}$, apparent terminal half-life; t_{max}, time to reach C_{max} after dosing; V_z/F, apparent volume of distribution.