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ORIGINAL ARTICLE

Pharmacokinetics and safety of neratinib during co-administration with loperamide in healthy subjects

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Abstract

Purpose To evaluate the effects of multiple doses of loperamide on the pharmacokinetics and safety of a single oral dose of neratinib.

Methods This was an open-label, two-period, fixed-sequence study. Twenty healthy adult subjects received an oral dose of neratinib 240 mg daily on Days 1–4 of Period 1 followed by a 7-day washout. In Period 2, oral neratinib 240 mg was administered with loperamide 4 mg followed by two further doses of loperamide 2 mg 8 and 16 h later on Days 1–4. Pharmacokinetic sampling was performed for 72 h following each neratinib dose. Safety was monitored throughout the study.

Results A median t_{max} of ~ 6 h was observed for neratinib during both periods. Apparent clearance and volume of distribution were similar for Periods 1 and 2: mean CL_{ss}/F 308.2 and 322.1 L/h; mean $V_{z\tau}/F$ 7995 and 10,318 L, respectively. The half-life of neratinib increased in the presence of loperamide from 18.0 to 22.2 h. Mean exposure was within the same range without and with loperamide administration: C_{max} 61.2 ng/mL and 49.5 ng/mL; AUC_{last} 1086 ng h/mL and 1153 ng h/mL, and AUC_{tau} 779 ng h/mL and 745 ng h/mL, respectively. Treatment-emergent adverse events were mainly mild in intensity, with the most frequent events being diarrhea (45%) and constipation (35%).

Conclusions Neratinib administered alone and concomitantly with multiple oral doses of loperamide is generally safe and well tolerated. Loperamide has minimal effects on neratinib pharmacokinetic parameters.

Keywords Drug interaction · Healthy · Volunteers · Loperamide · Neratinib · Pharmacokinetics · Safety

Introduction

Neratinib (Puma Biotechnology, Los Angeles, CA, USA) is an oral tyrosine kinase inhibitor of human epidermal growth factor receptors (HER or ERBB) 1, 2 and 4 [1]. In contrast to most other currently available tyrosine kinase inhibitors, the binding of neratinib to its target is irreversible, allowing sustained inhibition of receptor phosphorylation following drug clearance [2]. In vitro studies in cancer cell lines show that neratinib blocks downstream signal transduction and cell cycle regulatory pathways, which subsequently inhibit cell proliferation [2]. In vivo, neratinib has shown activity in HER2- and epidermal growth factor receptor (EGFR)-dependent tumor xenograft models when given orally on a once-daily schedule [2].

Neratinib has recently been approved by the US Food and Drug Administration for extended adjuvant treatment of early-stage HER2-positive metastatic breast cancer, having shown promising results both as a single agent and in combination with chemotherapeutic and targeted agents in clinical trials in this setting [3–9]. Clinical studies in women with HER2-positive breast cancer who had previously been treated with chemotherapy and trastuzumab showed overall response rates of 29–40% with neratinib monotherapy [3–5], while overall response rates of up to 75% have been observed for neratinib in combination with other chemotherapeutic agents [7, 9, 10]. In the phase III ExteNET trial in women with HER2-positive breast cancer previously treated with trastuzumab-based adjuvant therapy, 12 months' therapy with neratinib significantly

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improved 2-year invasive disease-free survival (hazard ratio vs placebo 0.67, 95% confidence interval (CI) 0.50–0.91; $p = 0.0091$) [11]. Analysis conducted after 5 years of ExteNET showed that extended adjuvant therapy resulted in significantly fewer clinically relevant invasive breast cancer relapses, i.e., those that might lead to death, such as distant and locoregional relapses outside the preserved breast (116 events with neratinib vs 163 events with placebo; stratified hazard ratio 0.73, 95% CI 0.57–0.92; $p = 0.0083$) [12]. Five-year invasive disease-free survival was 90.2% (95% CI 88.3–91.8%) with neratinib and 87.7% (85.7–89.4%) with placebo [12].

A once-daily dose of neratinib 240 mg on a continuous schedule is generally well tolerated with a low incidence of grade 3/4 adverse events (AEs) [13]. The most commonly reported AE associated with neratinib is diarrhea, which is a recognized class effect of EGFR tyrosine kinase inhibitors [13]. For example, in the ExteNET trial, without appropriate prophylaxis 40% of patients experienced grade 3 diarrhea, and one patient (< 1%) experienced grade 4 diarrhea [11]. Similar to other EGFR tyrosine kinase inhibitors, the incidence of diarrhea reported with neratinib administration is highest in the first months after treatment initiation and declines substantially thereafter [13, 14].

Diarrhea is an adverse effect associated with many cancer treatments and is well understood by oncologists, who manage its occurrence through early and aggressive treatment using patient education, diet, and antidiarrheal medications, such as the synthetic opiate loperamide [13]. Safety data indicate that intensive prophylaxis with loperamide can reduce the incidence of grade 3 diarrhea associated with neratinib from 30–50% to 0–17% [14]. As a result, an intensive prophylactic regimen is being implemented in ongoing and new clinical trials with neratinib [14]. Antidiarrheal prophylaxis is recommended to start with the first dose of neratinib and should continue through the first two cycles (56 days) of treatment.

In vitro and in vivo studies show that the transmembrane permeability glycoprotein, P-glycoprotein (P-gp), which is a member of the ATP binding cassette superfamily, plays a significant role in the disposition and absorption of drugs [15]. In humans, P-gp is expressed in a variety of cells including cells in the intestinal lumen, blood–brain barrier, renal tubules, and hepatocytes [15]. The profound effect of P-gp on the pharmacokinetics of drugs in humans makes it vital to screen new drugs for transport by this transmembrane protein [15]. In vitro studies of neratinib in Caco-2 cells, supported by clinical studies in combination with the sensitive P-gp substrate digoxin, have demonstrated inhibition of the P-gp transporter system by neratinib [16, 17]. Loperamide is a known P-gp substrate that is modulated by competitive and inhibitory effects of agents acting on the P-gp transporter system [18, 19]. It is therefore important to evaluate potential interaction between neratinib and loperamide if these agents are to be administered concomitantly.

The present study was designed to evaluate the effects of multiple doses of loperamide on the pharmacokinetics and safety of neratinib in healthy volunteers.

Materials and methods

Study design

This was a single-center, open-label, two-period, fixed-sequence study. During Period 1, an oral dose of neratinib 240 mg was administered daily on Days 1–4. During Period 2, the oral dose of neratinib 240 mg was administered with an oral dose of loperamide 4 mg on Days 1–4 (0 h). Two further doses of loperamide 2 mg were administered 8 and 16 h following the neratinib dose for a total of three daily doses (8 mg/day) on Days 1–4. Pharmacokinetic sampling was performed on Day 4 and continued until 96 h post-dose on that day. The washout period between the last dose of neratinib in Period 1 and the first dose in Period 2 was 7 days.

The primary objective of the study was to evaluate the effect of multiple doses of loperamide on the pharmacokinetics of neratinib in healthy subjects. The secondary objective was to study the safety and tolerability of neratinib when administered with loperamide.

The study protocol was approved by Chesapeake Research Review Inc. IRB. The study was performed in accordance with the ethical principles of the Declaration of Helsinki (1964) and subsequent amendments. Written informed consent was obtained from all subjects before enrollment.

Study participants

The study enrolled healthy men and women aged 19–55 years with a body mass index ≥ 18.5 and ≤ 32.0 kg/m^2 at screening. Participants were required to be medically healthy, with no clinically significant medical history based on physical examination, laboratory profiles, vital signs, and 12-lead electrocardiograms (ECGs). Non-smokers or moderate smokers (≤ 10 cigarettes/day for ≥ 3 months before screening) were eligible. Women of childbearing potential were ineligible. To be eligible, women must have undergone a sterilization procedure ≥ 6 months before the first dose of study drug or be postmenopausal for ≥ 1 year. Non-vasectomized men were required to use an acceptable form of contraception during the study and until 90 days after the last dose of study drug.

Consumption of foods and beverages containing xanthines/caffeine (from 24 h before neratinib administration through to the end of sample collection), alcohol (from 48 h before Day 1 of each period through to the end of

sample collection), and grapefruits and Seville oranges (from 14 days before Period 1 and throughout the study) was prohibited. Use of medications (including over-the-counter products), herbal products, and vitamin supplements was prohibited for 14 days before the first dose of study drug (or 28 days for cytochrome P450 and/or P-gp inducers) and throughout the study.

Treatments

During Period 1, all subjects received an oral dose of neratinib 240 mg (6 × 40 mg tablets; Excella GmbH, Feucht, Germany) within 30 min of a standard breakfast on Days 1–4. During Period 2, all subjects received oral neratinib as in Period 1. In addition, they received oral loperamide hydrochloride 4 mg (2 × 2 mg tablets; Major Pharmaceuticals Inc., Livonia, USA) administered at the 0-h time point and loperamide hydrochloride 2 mg (1 × 2 mg) at the 8-h and 16-h time points on Days 1–4.

Pharmacokinetics

Neratinib plasma concentration vs time data were calculated by noncompartmental methods using Phoenix® WinNonlin® Enterprise Version 6.3 (Certara Strategic Consulting, Montreal, Canada). Nominal sample times relative to the time of drug administration were used in all calculations. The maximum plasma concentration (C_{max}) was determined directly from the observed plasma concentration data; the time taken to reach C_{max} (T_{max}) was computed from blood draw time and date, and medication time and date. Area under the plasma concentration–time curve (AUC) from time 0 to the last measurable concentration (AUC_{last}) was calculated by the linear trapezoidal method. Apparent total plasma clearance at steady state (CL_{ss}/F) was calculated as dose/AUC_{tau}. The volume of distribution for dosing interval by fraction ($V_{z\tau}/F$) was calculated during the terminal elimination phase after oral (extravascular) administration using dose/($AUC_{tau} \times \lambda z$), where AUC_{tau} is the area under the plasma concentration–time curve during a dosing interval (τ) after multiple doses and λz is the elimination rate constant. A linear fixed-effects model including active treatment was used to compare natural log-transformed AUC_{tau}, AUC over the dosing interval, AUC_{last}, AUC from time zero to time of last measurable concentration, and C_{max} pharmacokinetic parameters of neratinib.

Sample collection and analytical methods

For quantification of neratinib plasma concentrations, venous blood samples (≤ 6 mL) were collected in blood collection tubes containing potassium ethylenediaminetetraacetic acid before neratinib dosing and at 0.5, 1, 2, 2.75, 3.5, 4.25, 5, 5.75, 6.5, 7.25, 8, 10, and 12 h after neratinib dosing on Day 4 of each study period. Further samples were collected on Days 5, 6, 7, and 8, corresponding to 24, 32, 48, 60, 72, and 96 h after the last neratinib dose.

Collection tubes were centrifuged within 30 min of collection at approximately 5 °C. Duplicate plasma aliquots were transferred into labeled storage tubes and stored within 60 min of collection at approximately 70 °C. Samples were shipped to Covance Bioanalytical Services LLC (Indianapolis, IN, USA) for analysis. Neratinib and the internal standard (neratinib) were extracted from samples using protein precipitation and analyzed using liquid chromatography with tandem mass spectrometric detection. The validated range of the assay was 3.00–250 ng/mL. The presence of loperamide did not affect the quantification of neratinib, and blank samples spiked with loperamide did not contain any significant peaks at the retention time of neratinib or the internal standard (data not shown). For the low- (9.00 ng/mL), medium- (45.0 ng/mL), and high-concentration (200 ng/mL) quality-control samples, the interassay accuracy was 100.1%, 100.4%, and 99.5%, respectively, the interassay precision (expressed as relative standard deviation) was 10.0%, 8.1%, and 7.1%, respectively, and the corresponding biases were 0.0%, 0.4%, and − 0.5%, respectively. All study samples ($n = 820$) were analyzed within the known neratinib stability period (703 days) for storage at − 60 °C to − 80 °C.

Safety

Safety end points included AEs, physical examinations, vital signs, 12-lead ECGs, and clinical laboratory tests (i.e., hematology, serum chemistry, and urinalysis). Safety was monitored throughout the study by repeated clinical and laboratory evaluations. All AEs were coded using the Medical Dictionary for Regulatory Activities (MedDRA®), version 20.0, and summarized by treatment for the number of subjects reporting the AE and the number of AEs reported. A treatment-emergent AE was defined as an event that started or worsened at the time of or after study drug administration, and up to 14 days after the last dose.

Statistical methods

Descriptive statistics were calculated for all pharmacokinetic parameters for neratinib with and without loperamide. To assess the effect of loperamide on the pharmacokinetics of neratinib, log-transformed neratinib parameters (i.e., AUC_{last}, AUC_{tau}, and C_{max}) were compared by an analysis of variance, which included treatment as a fixed effect and subject as a random effect in each model. The inferential results from these models (i.e., least-square means, differences between least-square means, and standard errors) were exponentiated to the original scale to obtain geometric least-square

means, geometric mean ratios, 90% CIs, and intrasubject coefficient of variations. Allowing for a Type I error of 5%, using a power of approximately 85%, and assuming the true geometric least-square mean ratio for the pharmacokinetic parameter C_{max} at steady state was within 95% or 105%, the estimated sample size to conclude no interaction between loperamide and neratinib was 16 subjects, as defined by the 90% CI for the mean ratio within the criterion of 80% and 125%. An additional four subjects were enrolled to allow for possible dropouts, giving a total study population of 20 subjects.

Results

The study was conducted from March 17, 2017, to April 17, 2017. A total of 20 subjects completed the screening assessments, met all eligibility criteria, and received the scheduled products. All subjects were evaluable for pharmacokinetic assessments for Period 1. One subject was discontinued from the study by the principal investigator on Day 1 of Period 2; another subject completed dosing but was considered lost to follow-up because they did not complete a required laboratory recheck. A summary of baseline characteristics is provided in Table 1. Most subjects were male (90%) with a median age of 37 (range 21–53) years and a mean ± standard deviation body mass index of 26.7 ± 2.7 kg/m^2.

Pharmacokinetics

Pharmacokinetic data were analyzed for 20 patients during Period 1 and 19 patients during Period 2. Plasma concentration–time profiles for neratinib 240 mg administered alone (Period 1) and in combination with loperamide (Period 2) are presented in Fig. 1. Neratinib concentrations generally declined in a mono-exponential fashion. A summary of pharmacokinetic parameters during the two treatment periods is presented in Table 2.

Following multiple administrations, T_{max} ranged from 2.75 to 7.25 h post-dose during Period 1 and from 3.50 to 10.0 h during Period 2, with a median T_{max} of approximately 6 h for both periods (neratinib 5.38 h; neratinib + loperamide 6.5 h). On average, clearance and volume of distribution were similar for neratinib following co-administration of loperamide compared with neratinib alone. Mean CL_{ss}/F was 308.2 and 322.1 L/h, and mean V_{zt}/F was 7995 and

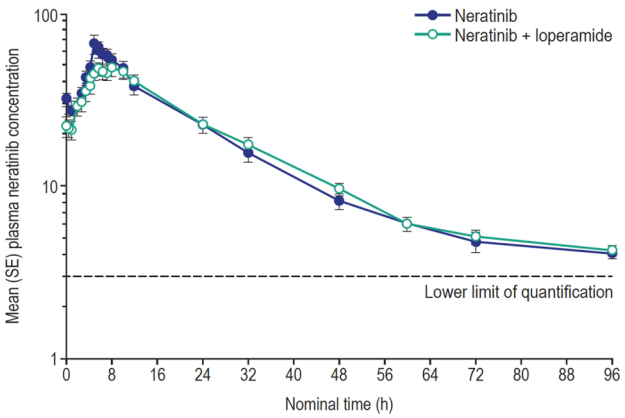

Fig. 1 Neratinib plasma concentrations over time following administration of an oral dose alone ($n = 20$) or in combination with loperamide ($n = 19$). *SE* standard error

Table 1 Demographic and baseline characteristics

Characteristic	All subjects ($n = 20$)
Median age, years (range)	37 (21–53)
Sex, *n* (%)	
Male	18 (90)
Female	2 (10)
Race, *n* (%)	
White	12 (60)
Black/African American	6 (30)
Other	2 (10)
Ethnicity, *n* (%)	
Hispanic/Latino	3 (15)
Other	17 (85)
Mean weight, kg (SD)	81.1 (11.1)
Mean height, cm (SD)	174.2 (7.8)
Mean BMI, kg/m^2 (SD)	26.7 (2.7)

BMI body mass index; *SD* standard deviation

Table 2 Neratinib pharmacokinetic parameters

Parameter	Neratinib (Period 1; $n = 20$)	Neratinib + loperamide (Period 2; $n = 19$)
AUC_{tau} (ng h/mL)	779 (63.5)	745.0 (55.8)
AUC_{last} (ng h/mL)	1086.4 (85.5)	1153.4 (69.7)
C_{max} (ng/mL)	61.2 (66.8)	49.5 (49.1)
C_{min} (ng/mL)	18.0 (66.3)	15.1 (81.1)
T_{max} (h)	5.38 (2.75, 7.25)	6.5 (3.50, 10.0)
$t_{1/2}$ (h)	18.0 (50.4)	22.2 (30.4)
CL_{ss}/F (L/h)	308 (63.5)	322 (55.8)
V_{zt}/F (L)	7995 (37.4)	10,318 (48.5)

Data are geometric mean (coefficient of variation % geometric mean), except for t_{max}, which is median (minimum, maximum)

AUC_{last} area under the plasma concentration–time curve from time zero to the time of last measurable concentration, AUC_{tau} area under the plasma concentration–time curve over dosing interval, CL_{ss}/F apparent total plasma clearance at steady state, C_{max} maximum plasma concentration, C_{min} minimum plasma concentration, $t_{1/2}$ half-life, T_{max} time taken to reach C_{max}, V_{zt}/F volume of distribution for dosing interval by fraction

10,318 L for neratinib alone and neratinib co-administered with loperamide, respectively. The half-life of neratinib increased in the presence of loperamide from 18.0 to 22.2 h. Mean neratinib exposure, as measured by C_{max}, AUC_{last}, and AUC_{tau}, was approximately within the same range without and with loperamide administration (Table 2; Fig. 2).

There were no statistically significant differences in exposure to neratinib following administration with loperamide vs neratinib alone (Table 3); however, the 90% CI for the ratio of geometric least-squares means for AUC_{last} and C_{max} were outside the range 80–125%.

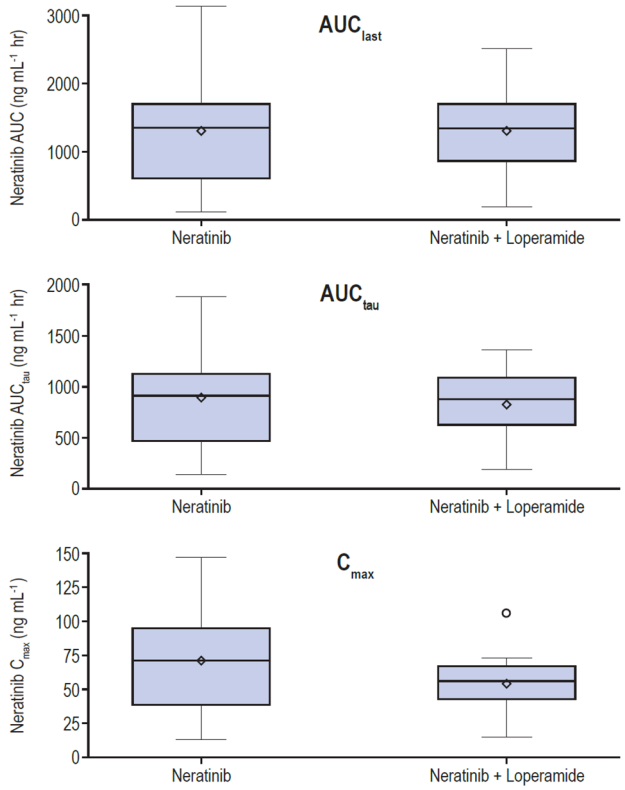

Fig. 2 Box plots of neratinib exposure parameters, where the mean is represented by diamond symbols, the median by the line inside the box, the 25% and 75% distribution by the bottom and top of the box, respectively, and the range by the whiskers. AUC_{last} AUC from time zero to last measurable concentration, AUC_{tau} AUC over the dosing interval, C_{max} maximum plasma concentration

Safety

Overall, 19 subjects received all doses of study drug during Periods 1 and 2. One subject was withdrawn after Day 1 of Period 2 due to an AE of vomiting. All subjects ($n = 20$) were included in the safety analysis.

In total, 122 treatment-emergent AEs were reported by 16 subjects (80%) during the study: 13 subjects during Period 1 (neratinib alone) and 12 subjects during Period 2 (neratinib + loperamide) (Table 4). The most frequently reported AEs were diarrhea (9 subjects; 45%), constipation (7 subjects; 35%), headache, decreased appetite (5 subjects each; 25%), nausea, fatigue, increased blood creatinine (4 subjects each; 20%), dyspepsia, vomiting, and somnolence (3 subjects each; 15%). All remaining AEs were reported by two or fewer subjects. Of the 122 AEs, 112 were mild in intensity (grade 1), 7 were moderate (grade 2), and 3 were severe (grade 3: diarrhea following neratinib alone; increased alanine aminotransferase, and increased aspartate aminotransferase following neratinib + loperamide). Of the 122 AEs, 101 were considered to be related to the study drug neratinib.

No deaths occurred during the study. One subject experienced three serious laboratory AEs: mild increased alkaline phosphatase, severe increased alanine transaminase, and severe increased aspartate transaminase. Onset of these events occurred approximately 3.3 days after the final dose of loperamide in Period 2. Levels of the three enzymes trended toward normal on the subsequent 3 days and were inside normal limits within 14 days. Four additional subjects experienced a laboratory AE of mild increased blood creatinine.

For the AE of diarrhea—reported 27 times by nine subjects overall (six subjects following neratinib alone, three subjects following both neratinib alone and neratinib + loperamide)—onset ranged from 13 min to 1.1 days after dosing and duration ranged from 1 min to approximately 23.5 h. Twenty-two diarrhea AEs were grade 1 in severity,

Table 3 Statistical comparison of neratinib plasma pharmacokinetic parameters for neratinib in combination with loperamide vs neratinib alone

Parameter	Geometric least-square mean		% Ratio of least-square means (90% CI)	*p* value
	Neratinib 240 mg + loperamide 4 mg ($n = 19$)	Neratinib 240 mg ($n = 20$)		
AUC_{tau} (ng h/mL)	745 (95.7)	771 (99.0)	96.6 (81.4–114.7)	0.7326
AUC_{last} (ng h/mL)	1153 (184)	1070 (171)	107.8 (90.2–128.9)	0.4734
C_{max} (ng/mL)	49.5 (6.24)	60.8 (7.67)	81.4 (65.8–100.8)	0.1117

AUC_{last} area under the plasma concentration–time curve from time zero to the time of last measurable concentration, AUC_{tau} area under the plasma concentration–time curve over dosing interval, *CI* confidence interval, C_{max} maximum plasma concentration

Table 4 Treatment-emergent adverse events occurring in ≥ 10% of subjects during either treatment period

Adverse event, *n* (%)[a]	Period 1 Neratinib (*n* = 20)	Period 2 Neratinib + loperamide (*n* = 20)	Overall
Subjects with treatment-emergent adverse events	13 (65)	12 (60)	16 (80)
Diarrhea	9 (45)	3 (15)	9 (45)
Constipation	3 (15)	5 (25)	7 (35)
Decreased appetite	3 (15)	3 (15)	5 (25)
Headache	1 (5)	4 (20)	5 (25)
Nausea	1 (5)	4 (20)	4 (20)
Fatigue	3 (15)	1 (5)	4 (20)
Blood creatinine increased	3 (15)	1 (5)	4 (20)
Somnolence	3 (15)	1 (5)	3 (15)
Dyspepsia	2 (10)	3 (15)	3 (15)
Vomiting	0 (0)	3 (15)	3 (15)
Dizziness	2 (10)	0 (0)	2 (10)
Rash macular	2 (10)	0 (0)	2 (10)
Dry skin	1 (5)	1 (5)	2 (10)
Abdominal pain	1 (5)	2 (10)	2 (10)

If a subject had ≥ 2 clinical adverse events, the subject was counted only once within a category. The same subject may appear in different categories

[a]Adverse events were coded using Medical Dictionary for Regulatory Activities (MedDRA®), Version 20.0

four were grade 2, and one was grade 3 (following neratinib alone). All diarrhea AEs resolved without treatment by study conclusion.

Grade 1 constipation was reported nine times by seven subjects (35%) overall (two subjects following neratinib alone, four subjects following neratinib + loperamide, and one subject following both neratinib alone and neratinib + loperamide). Most constipation events started within approximately 7 h after dosing, and most resolved within 5 days. Six subjects received prune juice for treatment of constipation. All constipation AEs resolved by study conclusion.

One AE—vomiting—led to discontinuation from the study. This subject was discontinued by the principal investigator on Day 1 of Period 2 due to the AE of mild vomiting. This subject had experienced diarrhea, which increased from moderate to severe during Period 1 (neratinib alone). Following the afternoon dose of loperamide on Day 1 of Period 2, the subject reported mild fatigue, moderate nausea, and headache, and two single episodes of mild vomiting. The subject vomited two more times approximately 10 h later. The nausea and headache resolved within 4.6 days and the fatigue within 7.6 days.

All mean vital sign parameters were within normal limits at all assessed time points, with minimal changes observed from baseline. Mean ECG parameters were also within normal limits at all time points, and no clinically important findings were observed during physical examinations.

Discussion

In this study, we evaluated the effects of multiple doses of loperamide on the pharmacokinetics of neratinib in healthy adults, and the safety of neratinib with or without loperamide. The need to understand any interactions between the two agents arose as a result of prophylactic loperamide being recommended to ameliorate diarrhea during neratinib treatment. The daily therapeutic doses of both neratinib and loperamide were used for the evaluation.

We observed that T_{max} values were similar for neratinib when administered alone or in combination with loperamide, while $t_{1/2}$, C_{max}, and AUC_{last} increased slightly when neratinib was administered with loperamide. The 90% CI for the geometric mean ratio of AUC_{tau} fit inside the 80–125% range, indicating that loperamide did not affect the exposure to neratinib at steady state after oral administration. The 90% CIs around the geometric mean ratio for AUC_{last} and C_{max} fell outside the 80–125% range; however, there were no statistically significant differences in exposure to neratinib following administration of neratinib either alone or in combination with loperamide. As a result, it is unlikely that loperamide will clinically affect the pharmacokinetics of neratinib.

Treatment-emergent AEs were observed during both study periods; however, the majority of AEs were mild in intensity. No unexpected AE occurred and those AEs that were considered to be serious resolved within 2 weeks of the end of the study. Diarrhea was the most frequently

reported AE, occurring in nine subjects overall (45% of the study population); however, the majority of diarrhea AEs were mild in severity. In general, the AEs observed during Period 1 were similar to those observed during Period 2. However, whereas diarrhea was documented in nine patients (45%) during Period 1 (neratinib alone), only three patients (15%) reported diarrhea during Period 2 when neratinib was administered with loperamide. Although the numbers of subjects included in this analysis were small, the findings are in line with the reduction in diarrhea observed with loperamide prophylaxis in other trials [14]. There were no treatment-related trends noted in vital sign, laboratory, ECG, or physical assessment data.

Preclinical studies suggest multifactor causes of neratinib-associated diarrhea, with inflammatory and secretory mechanisms potentially contributing to its occurrence [20]. Additional measures to reduce the incidence of diarrhea are currently being investigated, including concomitant use of the locally acting corticosteroid budesonide [20] and the bile-acid sequestrant colestipol [21]. Preliminary findings from the CONTROL study suggest that the addition of colestipol to loperamide prophylaxis may result in decreases in diarrhea incidence and severity, and may further diminish the duration of diarrhea vs administering loperamide prophylaxis without colestipol [21]. Concomitant colestipol may also improve overall tolerability, as reflected by fewer neratinib dose holds, dose reductions, and discontinuations in this ongoing cohort of patients [21]. Concomitant budesonide may be useful in the first cycle of treatment to decrease diarrhea severity, along with overall neratinib dose reductions and discontinuations during treatment. These preliminary findings with colestipol and budesonide warrant further investigation, as effective diarrhea prophylaxis may help to improve tolerability and long-term adherence to neratinib, thereby ensuring that maximum efficacy benefits are realized [20, 21].

In conclusion, multiple oral once-daily doses of neratinib administered alone or concomitantly with multiple oral doses of loperamide appeared generally safe and well tolerated by the healthy adult subjects in this study. The pharmacokinetic analysis suggests that loperamide minimally affects the neratinib pharmacokinetic parameters of $t_{1/2}$, C_{max}, and AUC, and these effects are unlikely to be clinically significant.

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Compliance with ethical standards

Conflict of interest At the time the research was conducted, Kiana Keyvanjah, Blaire Cooke, David Martin, Dan DiPrimeo, Jane Liang, Elizabeth Olek, and Alvin Wong were employees/stock holders of the study sponsor (Puma Biotechnology Inc.). Laura Sterling and Igor Rubets have no conflicts of interest to disclose.

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