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# A Phase 1 Drug-Drug Interaction Study Between Brigatinib and the CYP3A Substrate Midazolam in Patients With *ALK*-Positive or *ROS1*-Positive Solid Tumors

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**Abstract**

Brigatinib is a next-generation anaplastic lymphoma kinase (ALK) inhibitor approved for the treatment of patients with *ALK*-positive (*ALK*+) non–small cell lung cancer (NSCLC). A phase 1 drug-drug interaction study was conducted to evaluate the effect of multiple-dose administration of brigatinib on the single-dose pharmacokinetics of midazolam, a sensitive cytochrome P450 3A substrate. In cycle 1, patients with *ALK*+ or *ROS1*+ solid tumors, including NSCLC, received a single 3-mg dose of midazolam as an oral solution alone on day 1 and then coadministered with brigatinib on day 21 (brigatinib 90 mg once daily on days 2-8; 180 mg once daily on days 9-28). After cycle 1, patients could continue to receive brigatinib in 28-day treatment cycles. The primary study objective was to characterize the effect of brigatinib 180 mg once daily on midazolam pharmacokinetics. The secondary objective was to assess safety. Exploratory efficacy endpoints included objective response rate and progression-free survival. Brigatinib was generally well tolerated, and safety data were consistent with the known safety profile. Among the 10 patients with *ALK*+ NSCLC, the confirmed objective response rate was 30% and median progression-free survival was 7.2 months. Coadministration of brigatinib reduced midazolam maximum observed plasma concentration by ≈16% (geometric least-squares mean ratio, 0.836 [90%CI, 0.662-1.056]) and area under the plasma concentration–time curve from time 0 to infinity by ≈26% (geometric least-squares mean ratio, 0.741 [90%CI, 0.600-0.915]). Thus, brigatinib is a weak inducer of cytochrome P450 3A in vivo.

**Keywords**

anaplastic lymphoma kinase, brigatinib, cytochrome P450 (CYP) 3A, drug-drug interaction, midazolam, non–small cell lung cancer, protein kinase inhibitors

Anaplastic lymphoma kinase gene (*ALK*) rearrangements are key drivers of oncogenesis in several cancers.[1] In metastatic non–small cell lung cancer (NSCLC), *ALK* fusions are found to occur in ≈5% of tumors.[2] Brigatinib is a next-generation ALK tyrosine kinase inhibitor that was initially approved for the treatment of patients with *ALK*+ NSCLC who had disease progression on, or were intolerant to, crizotinib based on results of the phase 2 ALTA trial (NCT02094573).[3] More recently, brigatinib was granted approval as a first-line treatment for patients with advanced *ALK*+ NSCLC based on results from the phase 3 ALTA-1L trial (NCT02737501), in which brigatinib exhibited superior efficacy versus crizotinib (hazard ratio [HR] for progression-free survival [PFS]: 0.48 [95%CI, 0.35-0.66]; $P < .0001$) in patients with ALK inhibitor-naive advanced *ALK*+ NSCLC, with tolerability consistent with previous studies.[4] In preclinical studies, brigatinib was also demonstrated to be a potent inhibitor of ROS1.[5] However, the clinical activity of brigatinib in *ROS1*+ solid tumors has not been extensively studied.

Brigatinib is approved at a dose of 90 mg orally once daily for the first 7 days of treatment, and the dose is then increased to 180 mg once daily thereafter.[6] This regimen provides a favorable benefit-risk profile because the lower starting dose (90 mg once daily) reduces the frequency of rare early-onset pulmonary

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events, while escalation to the higher maintenance dose (180 mg once daily) allows patients the opportunity to derive additional clinical benefit. After both single- and multiple-dose administration, systemic exposures of brigatinib increased in a dose-proportional manner over the dose range of 60-240 mg once daily.[3,7–11] The plasma elimination half-life ($t_{1/2}$) of brigatinib was 25 hours following administration of 180 mg once daily in patients with advanced malignancies.[12] In a study of healthy participants administered a single dose of 180 mg of brigatinib after an overnight fast or after consumption of a high-fat meal, brigatinib geometric mean maximum observed plasma concentration ($C_{max}$) was reduced by 13% and median time to first occurrence of $C_{max}$ ($t_{max}$) was increased from 2 to 5 hours under fed conditions; however, a high-fat meal had no effect on total systemic exposure (ie, area under the plasma concentration–time curve [AUC]).[13] Therefore, brigatinib can be administered with or without food. Based on results from population pharmacokinetic (PK) analyses, age, sex, race, body weight, and mild or moderate renal impairment have no clinically meaningful effect on brigatinib PK.[14] However, a dose reduction is recommended for patients with severe renal impairment or severe hepatic impairment (Child-Pugh class C) based on results from dedicated renal impairment and hepatic impairment studies.[15]

Following oral administration of a single 180-mg dose of [$^{14}$C]-brigatinib to healthy participants, the majority of the administered dose was recovered in the feces (65%), and 25% of the dose was recovered in the urine. The major circulating radioactive component was brigatinib (92%). Brigatinib is metabolized in vitro primarily by cytochrome P450 (CYP) 2C8 and CYP3A.[16] As a result, a clinical, 3-arm, drug-drug interaction (DDI) study was conducted in healthy participants to evaluate the single-dose PK of brigatinib in the presence of a strong CYP2C8 inhibitor (gemfibrozil), a strong CYP3A inhibitor (itraconazole), and a strong CYP3A inducer (rifampin).[16] Gemfibrozil had no clinically meaningful effect on the PK of brigatinib (geometric least-squares mean [LSM] AUC from time 0 to infinity [$AUC_\infty$] ratio, 0.88 [90%CI, 0.83-0.94]). Accordingly, CYP2C8 does not appear to be a meaningful determinant of brigatinib clearance in vivo, and no dose adjustment is recommended during coadministration with CYP2C8 inhibitors.[16] In contrast, brigatinib systemic exposure increased by 101% in the presence of itraconazole (geometric LSM $AUC_\infty$ ratio, 2.01 [90%CI, 1.84-2.20]) and decreased by 80% in the presence of rifampin (geometric LSM $AUC_\infty$ ratio, 0.20 [90%CI, 0.18-0.21]). Consequently, these results indicate that the concomitant use of brigatinib with strong CYP3A inhibitors or strong CYP3A inducers should be avoided. If concomitant use of strong CYP3A inhibitors cannot be avoided, the dose of brigatinib should be reduced by ≈50%.[16] Dose modifications are also recommended during concomitant use with moderate CYP3A inhibitors or moderate CYP3A inducers based on the results of physiologically based PK analyses.[17]

At clinically relevant concentrations, brigatinib did not inhibit CYP1A2, 2B6, 2C8, 2C9, 2C19, 2D6, or 3A4/5 activity in human liver microsomes (data on file). However, in vitro studies using human hepatocytes indicate that brigatinib, at clinically relevant concentrations of 1-2.5 $\mu$M, caused an increase in CYP3A4 messenger RNA levels (data on file). This suggested that brigatinib may have the potential for PK drug interactions with substrates of CYP3A through induction of the expression of this enzyme, which may lead to decreased systemic exposures of its substrates. Thus, a clinical DDI study was conducted to evaluate the effect of multiple-dose administration of brigatinib on the single-dose PK of midazolam, a sensitive CYP3A probe substrate, to determine whether brigatinib, at therapeutic doses, produces clinically meaningful CYP3A induction in vivo.

## Methods

### Patients

The clinical study protocol and patient consent forms were reviewed and approved by the independent ethics committees of the participating study sites before study initiation (Table S1). All patients provided written informed consent. The study was performed at 10 study sites in Europe in accordance with the requirements of the Declaration of Helsinki, the International Council for Harmonisation, Good Clinical Practice, and all applicable local regulations.

Eligible patients were 18 years of age or older with locally advanced or metastatic solid tumors who had met 1 of the following 4 criteria: (1) patients with *ALK*+ NSCLC who had disease progression on or were intolerant to treatment with at least 1 other ALK inhibitor; (2) patients with *ALK*+ nonlung solid tumors for whom no standard, nonexperimental therapy was available; (3) patients with *ROS1*+ NSCLC who had disease progression on or were intolerant to crizotinib; or (4) patients with *ROS1*+ nonlung solid tumors for whom no standard, nonexperimental therapy was available. Patients had at least 1 target lesion per Response Evaluation Criteria in Solid Tumors (RECIST) version 1.1, had an Eastern Cooperative Oncology Group performance status of 0 or 1, and had recovered (ie, grade ≤1) from toxicities related to prior anticancer therapy (of note, irreversible treatment-related grade >1 alopecia and peripheral neuropathy were allowed). Patients were also required to have adequate organ function,

including total bilirubin ≤1.5 times the upper limit of the normal range (ULN), alanine aminotransferase and aspartate aminotransferase ≤2.5 times ULN (≤5 times ULN if liver metastases were present), and an estimated glomerular filtration rate ≥30 mL/min/1.73 $m^2$.

Patients were excluded if they had received prior brigatinib therapy; systemic treatment with strong or moderate CYP3A inhibitors or inducers within 14 days before study enrollment; any investigational systemic anticancer agent, chemotherapy, or radiation therapy within 14 days before the first dose of study drug, or prior ALK inhibitor therapy within 7 days before the first dose of study drug; had received antineoplastic monoclonal antibodies or undergone major surgery within 30 days before the first dose of study drug; had symptomatic central nervous system metastases at screening; had significant, uncontrolled, active cardiovascular disease including uncontrolled hypertension; were pregnant or breastfeeding; had a history or presence of pulmonary interstitial disease, drug-related pneumonitis, or radiation pneumonitis; had an ongoing or active infection; or had a known gastrointestinal condition that could potentially interfere with the oral absorption or tolerance of midazolam or brigatinib.

Approximately 20 patients were planned to be enrolled to achieve ≈15 PK-evaluable patients. The sample size calculation was based on the expected 2-sided 90%CI for the difference in the paired, log-transformed $AUC_\infty$ means of midazolam in the absence and in the presence of brigatinib. The within-patient percentage coefficient of variation (%CV) for midazolam $AUC_\infty$ was estimated to be 28% based on a pooled analysis of data from 5 previously reported DDI studies with midazolam conducted in patients with cancer.[18–22] Assuming that the $AUC_\infty$ ratio for midazolam in the presence versus absence of brigatinib was 1, with a sample size of 15, the 90%CI for the $AUC_\infty$ ratio was expected to be 0.84-1.19 on the basis of the variance assumptions.

### Study Design

This was an open-label, multicenter, phase 1, DDI study (NCT03420742) conducted in patients with *ALK*+ or *ROS1*+ solid tumors, including NSCLC. The study comprised 2 parts: part A (cycle 1, PK cycle) and part B (cycle 2 and beyond, additional treatment cycles). Each treatment cycle was 28 days. The primary objective of the study was to characterize the effect of repeat-dose administration of brigatinib 180 mg once daily on the single-dose PK of oral midazolam. The secondary objective was to assess the safety and tolerability of brigatinib in patients with *ALK*+ or *ROS1*+ solid tumors. The study also included an exploratory objective to assess the response to brigatinib treatment in each of the 4 tumor subgroups: (1) *ALK*+ NSCLC, (2) *ROS1*+ NSCLC, (3) all other *ALK*+ solid tumors, and (4) all other *ROS1*+ solid tumors.

Part A used a fixed-sequence design over a single 28-day treatment cycle (cycle 1, PK cycle) as shown in Figure 1. Patients received a single 3-mg dose of midazolam as an oral solution on day 1, with serial PK sampling performed over the 24 hours following dosing to characterize the PK of midazolam in the absence of brigatinib. Brigatinib 90 mg once daily was then orally administered on days 2-8. If the 90-mg brigatinib dose was tolerated by the patient, the brigatinib dose was increased to 180 mg once daily on day 9 and then continued through day 28 (in accordance with the recommended posology for patients with *ALK*+ NSCLC).[6] For patients escalating to brigatinib 180 mg once daily, a single 3-mg dose of midazolam as an oral solution was administered on day 21 of part A, with serial PK sampling performed over the 24 hours after dosing to characterize midazolam PK in the presence of brigatinib. Predose blood samples were also collected on days 21 and 22 of part A and on day 1 of cycle 2 (part B) to measure trough plasma concentrations of brigatinib.

After completion of part A, patients could continue to part B of the study to receive the potential therapeutic benefits of brigatinib. The starting dose of brigatinib in part B was the dose that the patient tolerated at the end of part A. Part B of the study included 28-day treatment cycles in which brigatinib was administered once daily at the highest tolerated dose (up to 180 mg once daily) for up to 23 cycles of treatment or until progressive disease, intolerable toxicity, or another discontinuation criterion was met.

### Assessments

In part A (cycle 1), venous blood samples were collected at 0 (predose), 0.25, 0.5, 1, 1.5, 2, 4, 6, 8, 10, and 24 hours postdose to measure plasma concentrations of midazolam after administration on days 1 and 21. Plasma concentrations of midazolam were determined by a validated sensitive and specific liquid chromatography–tandem mass spectrometry–based method. A 150-$\mu$L plasma aliquot was fortified with midazolam-$d_4$ internal standard working solution. Analytes were isolated through supported liquid extraction. The eluate was evaporated, and the remaining residue was then reconstituted. The final extract was analyzed using a Nexera liquid chromatograph (Shimadzu, Kyoto, Japan) and an API 4000 triple quadrupole mass spectrometer (SCIEX, Framingham, Massachusetts) equipped with a Turbo-V electrospray ion source. Chromatographic separation was achieved using a Pursuit XRs 5 C18, 50 × 2 mm, 5-$\mu$m column. Mobile phase A was a mixture of water : acetonitrile : ammonium formate : 1M formic acid

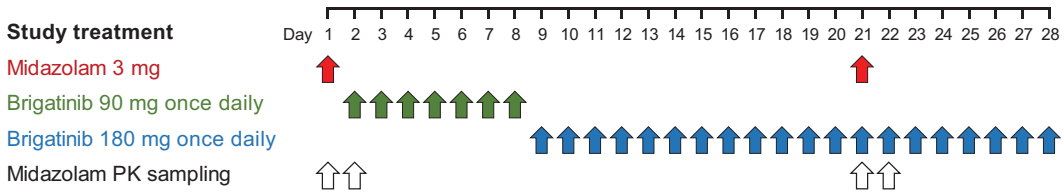


**Figure 1.** Drug-drug interaction study design for part A (cycle 1, PK cycle). PK, pharmacokinetic.

(900:100:10:1; v:v:v:v), and mobile phase B comprised acetonitrile : formic acid (1000:1; v:v). The flow rate was 0.25 mL/min. For tandem mass spectrometry detection, the (parent/product) transitions of m/z 326.1 → 291.2 and m/z 332.0 → 295.2 were used for midazolam and midazolam-$d_4$, respectively. Calibration curves for midazolam were established using standards, and the peak area ratios of midazolam versus the isotopically labeled internal standard were used to quantify samples. Linearity was achieved in the midazolam concentration range of 0.100 to 100 ng/mL, with quality control samples ranging from 0.300 to 75.0 ng/mL. Interassay precision (%CV) ranged from 3.76% to 5.77% with a bias of 1.62% to 5.36%. Incurred sample reanalysis of plasma samples met acceptance criteria.

Blood samples were also collected predose on days 21 and 22 of part A (cycle 1), and on day 1 of cycle 2 (part B) to measure plasma trough concentrations of brigatinib. Concentrations of brigatinib in human plasma were determined by a validated sensitive and specific liquid chromatography–tandem mass spectrometry–based method. A 50-$\mu$L plasma aliquot was added to a 96-well plate followed by 20 $\mu$L of a brigatinib-$d_4$ internal standard solution (150 ng/mL). Plasma proteins were precipitated by the addition of 150 $\mu$L of acetonitrile. The plate was covered, vortexed, and centrifuged. A 50-$\mu$L aliquot was then transferred, reconstituted in 150 $\mu$L of water and mixed for analysis using an API-5500 mass spectrometer (SCIEX) equipped with dual Agilent 1312B pumps (Agilent Technologies, Santa Clara, California) and a PAL autosampler (Thermo Fisher Scientific, Waltham, Massachusetts). A reverse-phase gradient method running at a flow rate of 0.6 mL/min on an ACE 3 C18, 2.1 × 50 mm, 3-$\mu$m column (Agilent) provided brigatinib and brigatinib-$d_4$ retention times of 0.82 and 0.79 minutes, respectively. The mobile phases used were (5:95:0.1; v:v:v) acetonitrile : water : formic acid (mobile phase A), and (50:50:0.1; v:v:v) acetonitrile : methanol : formic acid (mobile phase B). Brigatinib and its internal standard (brigatinib-$d_4$) were ionized under a positive ion spray mode and detected through multiple reaction monitoring of mass transition pairs of m/z 584.3 → 484.3 for brigatinib and 588.3 → 484.2 for brigatinib-$d_4$. Calibration curves for brigatinib were established using standards, and the peak area ratios of brigatinib versus brigatinib-$d_4$ were used to quantify samples. Linearity was achieved in the brigatinib concentration range of 5.00 to 2500 ng/mL with quality control samples ranging from 15.0 to 2000 ng/mL. Interassay precision (%CV) ranged from 3.2% to 6.1% with a bias of −8.5% to −7.3%. Incurred sample reanalysis of plasma samples met acceptance criteria.

Safety and tolerability evaluations included adverse event (AE) monitoring, clinical laboratory tests, vital signs measurements, physical examinations, and 12-lead electrocardiograms performed during the study. AEs were coded according to the Medical Dictionary for Regulatory Activities version 23.0 and were graded according to the National Cancer Institute Common Terminology Criteria for Adverse Events version 4.03. All patients were followed up for AEs and serious AEs (SAEs) from the signing of informed consent through 30 days after administration of the last dose of study drug.

Radiological assessments of tumor response, using RECIST version 1.1 and evaluated by computed tomography and/or magnetic resonance imaging, were performed at screening (as close as possible to day 1 of cycle 1), at 8-week intervals during treatment through cycle 14 and every 12 weeks thereafter (ie, every 3 cycles), and at the end of treatment.

### Pharmacokinetic Analyses

The PK-evaluable population was used for all PK analyses and was defined as patients who met all of the following 3 criteria: (1) received the protocol-specified dosing regimen during part A (including escalation to brigatinib 180 mg once daily) without dose reductions or interruptions through the completion of PK sampling; (2) did not receive any excluded concomitant medications through the completion of PK sampling; and (3) had sufficient midazolam plasma concentration–time data to permit the reliable estimation of PK parameters. Plasma PK parameters for midazolam in the absence and in the presence of brigatinib were calculated using noncompartmental analysis methods with Phoenix WinNonlin version 8.1 (Certara, Princeton, New Jersey) and included the $C_{max}$, AUC from time 0 to the time of the last quantifiable concentration ($AUC_{last}$), $AUC_{\infty}$, apparent oral clearance,

$t_{max}$, the terminal disposition phase half-life ($t_{1/2z}$), and the apparent volume of distribution during the terminal disposition phase. Midazolam PK parameters in the absence and in the presence of brigatinib were summarized using descriptive statistics (SAS version 9.4; SAS Institute, Cary, North Carolina).

For the estimation of the effect of brigatinib on midazolam PK, the ratios of geometric mean midazolam $AUC_{last}$, $AUC_{\infty}$, and $C_{max}$ (with vs without brigatinib coadministration) and the associated 2-sided 90%CIs were derived on the basis of the within-patient variance calculated via a mixed-effects analysis of variance fitting terms for treatment (ie, midazolam with or without brigatinib coadministration). Patient was treated as a random effect in the model. After log transformation, midazolam $AUC_{last}$, $AUC_{\infty}$, and $C_{max}$ were separately analyzed. Point estimates and adjusted 90%CIs for the difference in treatment were calculated and then exponentially back-transformed to provide point and CI estimates for the ratios of interest.

### Safety and Exploratory Efficacy Analyses

All patients who received at least 1 dose of brigatinib or midazolam were included in the safety population. Treatment-emergent AEs (TEAEs) were summarized descriptively.

For the exploratory efficacy analyses, the response-evaluable population was defined as all patients who had measurable disease at baseline, received at least 1 dose of brigatinib, and had at least 1 postbaseline response assessment. The efficacy endpoints evaluated included objective response rate (ORR; defined as best overall response of complete response or partial response per RECIST version 1.1), duration of response (DOR), and progression-free survival (PFS). ORR was summarized descriptively; DOR and PFS were estimated using Kaplan-Meier analysis.

## Results

### Patients

A total of 24 patients were enrolled in the study and continued from part A to part B. All 24 patients received at least 1 dose of study drug and were included in the safety and efficacy analyses. Of these 24 patients, 15 patients were considered PK evaluable and therefore included in the PK analyses. The remaining 9 patients were excluded from the PK analyses because they did not receive the protocol-specified dosing regimen of midazolam and brigatinib during part A (8 patients) or received an excluded concomitant medication during part A (1 patient). Demographic and baseline disease characteristics of the safety and PK-evaluable populations are presented in Table 1. In the *ALK*+ NSCLC cohort, all 10 patients received at least 1 prior line of therapy with alectinib, and 7 patients received ≥2 prior lines of ALK inhibitor therapy. All patients had stage IV disease.

**Table 1.** Patient Demographics and Baseline Characteristics

| Characteristic | Safety Population (n = 24) | PK-Evaluable Population (n = 15) |
|---|---|---|
| Age (y) | | |
| Median | 57 | 57 |
| Range | 26-75 | 37-73 |
| Sex, n (%) | | |
| Male | 11 (46) | 5 (33) |
| Female | 13 (54) | 10 (67) |
| Race, n (%) | | |
| White | 23 (96) | 14 (93) |
| Not reported | 1 (4) | 1 (7) |
| Weight (kg) | | |
| Median | 67.5 | 64.0 |
| Range | 44.4-129.0 | 44.4-129.0 |
| Disease type, n (%) | | |
| *ALK*+ NSCLC | 10 (42) | 8 (53) |
| *ALK*+ nonlung solid tumors[a] | 7 (29) | 3 (20) |
| *ROS1*+ NSCLC | 5 (21) | 3 (20) |
| *ROS1*+ nonlung solid tumors[b] | 2 (8) | 1 (7) |
| Prior solid tumor therapy, n (%) | 24 (100) | 15 (100) |
| Radiotherapy | 13 (54) | 8 (53) |
| Surgery | 12 (50) | 6 (40) |
| Site of cancer involvement, n (%) | | |
| Bone | 8 (33) | 6 (40) |
| Brain/CNS | 9 (38) | 7 (47) |
| Kidney | 1 (4) | 0 (0) |
| Liver | 11 (46) | 6 (40) |
| Lungs | 13 (54) | 10 (67) |
| Lymph nodes | 10 (42) | 7 (47) |
| Skin | 1 (4) | 0 (0) |
| Other | 13 (54) | 7 (47) |

*ALK*, anaplastic lymphoma kinase gene; CNS, central nervous system; NSCLC, non–small cell lung cancer; PK, pharmacokinetic; *ROS1*, ROS proto-oncogene.
[a] Includes breast, esophageal, and prostate adenocarcinoma, ovarian germ cell tumor, Mullerian tumor, mesothelioma, and thymus carcinoma (1 patient each).
[b] Cholangiocarcinoma and colon cancer (1 patient each).

### Pharmacokinetics

Mean midazolam plasma concentration–time plots following administration of midazolam alone (day 1) and in the presence of brigatinib (day 21) are shown in Figure 2. Mean plasma concentrations of midazolam were lower throughout the entire 24-hour postdose period when midazolam was administered with brigatinib compared with when midazolam was administered alone.

A summary of midazolam plasma PK parameters in the presence and in the absence of brigatinib is presented in Table 2. Midazolam was rapidly absorbed with and without coadministration of brigatinib with a median $t_{max}$ of 0.5 hours postdose on both days 1 and 21. The geometric mean (geometric %CV) midazolam $C_{max}$ with and without brigatinib coadministration was

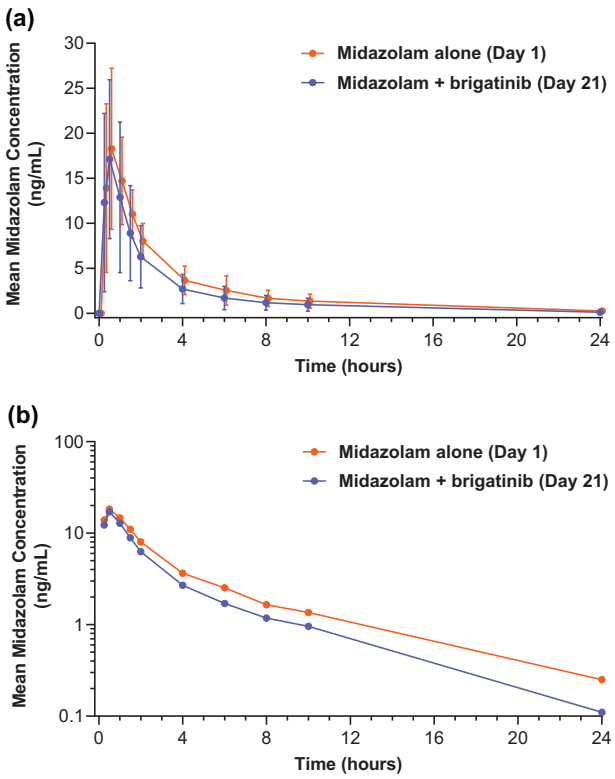


**Figure 2.** Mean midazolam plasma concentration–time plots following administration of a single oral dose of 3 mg of midazolam alone (day 1) and in the presence of brigatinib (day 21) on a linear (a) and semi-log (b) scale. In panel a, error bars represent standard deviation.

16.5 (49.9) and 19.7 (42.6) ng/mL, respectively. The corresponding values for midazolam $AUC_\infty$ with and without brigatinib coadministration were 42.1 (54.2) and 57.2 (30.3) ng • h/mL, respectively. Thus, systemic exposures of midazolam were slightly reduced in the presence of brigatinib. In contrast, the geometric mean midazolam $t_{1/2z}$ was generally comparable with (4.42 hours) and without (4.80 hours) brigatinib coadministration.

For the estimation of the effect of brigatinib on midazolam PK, the geometric LSM ratios and associated 90%CIs were calculated for midazolam $C_{max}$, $AUC_{last}$, and $AUC_\infty$ with (day 21) versus without (day 1) brigatinib coadministration. As shown in Table 3, the geometric LSM ratios for these midazolam PK parameters were 0.836 (90%CI, 0.662-1.057), 0.697 (90%CI, 0.545-0.891), and 0.741 (90%CI, 0.600-0.915), respectively. Therefore, brigatinib reduced midazolam $C_{max}$, $AUC_{last}$, and $AUC_\infty$ by ≈16%, ≈30%, and ≈26%, respectively.

Plasma trough concentrations of brigatinib were generally similar on cycle 1 day 21, cycle 1 day 22, and cycle 2 day 1, thereby indicating that steady state was achieved (Figure 3).

**Table 2.** PK Parameters of Midazolam Following Administration of a Single Oral Dose of Midazolam 3 mg Alone (Day 1) and in the Presence of Brigatinib (Day 21)

| Midazolam PK Parameter | Day 1 Midazolam Alone[a] (n = 15) | Day 21 Midazolam + Brigatinib[a] (n = 15) |
|---|---|---|
| $t_{max}$ (h) | 0.500 (0.220-1.93) | 0.500 (0.250-1.00) |
| $C_{max}$ (ng/mL) | 19.7 (42.6) | 16.5 (49.9) |
| $AUC_{last}$ (ng • h/mL) | 53.8 (29.8) | 37.5 (58.5) |
| $AUC_\infty$ (ng • h/mL) | 57.2 (30.3) | 42.1 (54.2)[b] |
| CL/F (L/h) | 52.4 (30.3) | 71.3 (54.2)[b] |
| $V_z$/F (L) | 363 (43.0) | 455 (68.4)[b] |
| $t_{1/2z}$ (h) | 4.80 (64.8) | 4.42 (45.2)[b] |

%CV, percentage coefficient of variation; $AUC_{last}$, area under the plasma concentration–time curve from time 0 to the time of the last quantifiable concentration; $AUC_\infty$, area under the plasma concentration–time curve from time 0 to infinity; CL/F, apparent oral clearance; $C_{max}$, maximum observed plasma concentration; PK, pharmacokinetic; $t_{1/2z}$, terminal disposition phase half-life; $t_{max}$, time of first occurrence of $C_{max}$; $V_z$/F, apparent volume of distribution during the terminal disposition phase.
[a] Parameters are presented as geometric mean (geometric %CV), except for $t_{max}$, which is presented as median (range).
[b] n = 14.

**Table 3.** Geometric Least-Squares Mean Ratios and 90%CIs for Midazolam $C_{max}$, $AUC_{last}$, and $AUC_\infty$ With (Day 21) Versus Without (Day 1) Brigatinib Coadministration

| Midazolam PK Parameter | Geometric LSM Ratio (Day 21 vs Day 1) | 90%CI |
|---|---|---|
| $C_{max}$ (ng/mL) | 0.836 | 0.662-1.057 |
| $AUC_{last}$ (ng • h/mL) | 0.697 | 0.545-0.891 |
| $AUC_\infty$ (ng • h/mL) | 0.741 | 0.600-0.915 |

$AUC_{last}$, area under the plasma concentration–time curve from time 0 to the time of the last quantifiable concentration; $AUC_\infty$, area under the plasma concentration–time curve from time 0 to infinity; $C_{max}$, maximum observed plasma concentration; LSM, least-squares mean; PK, pharmacokinetic.

## Safety

The median duration of exposure to treatment was 68.5 days (range, 5-595 days). Table 4 summarizes the overall incidence of TEAEs among the 24 patients in the safety population. TEAEs of any grade were reported in all 24 patients; the most common (≥25% of patients) were increased blood creatine phosphokinase (46%), nausea (46%), dyspnea (42%), diarrhea (38%), increased aspartate aminotransferase (33%), increased alanine aminotransferase (29%), vomiting (29%), anemia (29%), cough (25%), and increased lipase (25%). Grade 3 or 4 TEAEs deemed to be study drug–related were reported in 6 patients (25%); the most common (>1 patient) were increased amylase and hypertension (2 patients each, 8%). Four patients (17%) discontinued treatment because of TEAEs, all in part B of the study. Two patients discontinued due to the progression of NSCLC, 1 patient because of general health deterioration, and 1 patient after experiencing respiratory failure. None of the discontinuations were considered to

**Table 4.** Summary of TEAEs

| TEAE[a], n (%) | Brigatinib 90 mg → 180 mg Once Daily (n = 24) |
|---|---|
| Any TEAE | 24 (100) |
| Grade ≥3 TEAE | 20 (83) |
| Drug-related | 6 (25) |
| Investigations | 4 (17) |
| Amylase increased | 2 (8) |
| Alanine aminotransferase increased | 1 (4) |
| Blood creatine phosphokinase increased | 1 (4) |
| Lipase increased | 1 (4) |
| Vascular disorders | 2 (8) |
| Hypertension | 2 (8) |
| Gastrointestinal disorders | 1 (4) |
| Pancreatitis | 1 (4) |
| Serious TEAE | 17 (71) |
| Drug-related | 1 (4) |
| TEAE leading to discontinuation | 4 (17) |
| On-study deaths | 8 (33) |
| Drug-related | 0 (0) |

TEAE, treatment-emergent adverse event.
[a]Patients can be counted in >1 category.

be study drug–related. Any-grade treatment-emergent SAEs were reported in 17 (71%) patients, with only 1 patient having an SAE that was considered treatment-related (pancreatitis). Eight patients died on-study; however, none of the deaths were considered related to study drug. TEAEs leading to death were reported for 4 patients during part B of the study; 3 were due to disease progression, and 1 was due to invasive ductal breast carcinoma; none were considered study drug–related.

### Exploratory Efficacy

All 24 enrolled patients were eligible for the efficacy analyses. For the overall population, the confirmed ORR was 12.5% (95%CI, 3%-32%; 3 partial responses) and the median PFS was 2.8 months (95%CI, 1.6-7.6 months). In the 10 patients with *ALK*+ NSCLC, confirmed ORR was 30% (95%CI, 7%-65%; 3 partial responses). Two of the 3 responders had documented progressive disease. The DOR for these 2 patients was 5.5 months and 17.5 months, respectively. The remaining patient had been in partial response status until initiation of alternate subsequent anticancer therapy. The DOR for this patient was 1.8 months when censoring initiation of anticancer therapy by the last valid assessment or 2.3 months when treating subsequent anticancer therapy as an event. Median PFS was 7.2 months (95%CI, 1.1 months-not calculable) in the *ALK*+ NSCLC cohort. No patients in the other tumor cohorts had a confirmed response. Four (80%) patients with *ROS1*+ NSCLC had a best response of stable disease and median PFS of 4.8 months (95%CI, 1.5 months-not calculable). Two patients (29%) with other *ALK*+ solid tumors and 1 patient (50%) with other *ROS1*+ solid tumors had a best response of stable disease and had a short median PFS (1.8 months).

## Discussion

During development, in vitro studies using human hepatocytes demonstrated an increase in CYP3A4 messenger RNA levels in the presence of clinically relevant concentrations of brigatinib. Consequently, this phase 1 DDI study was conducted to assess whether therapeutic doses of brigatinib cause clinically meaningful induction of CYP3A in vivo. Brigatinib was administered at its recommended dose of 180 mg once daily (following a 7-day lead-in of 90 mg once daily) for the treatment of *ALK*+ NSCLC[6] and midazolam was used as the sensitive CYP3A probe substrate. Because multiple-dose administration of brigatinib was required for this

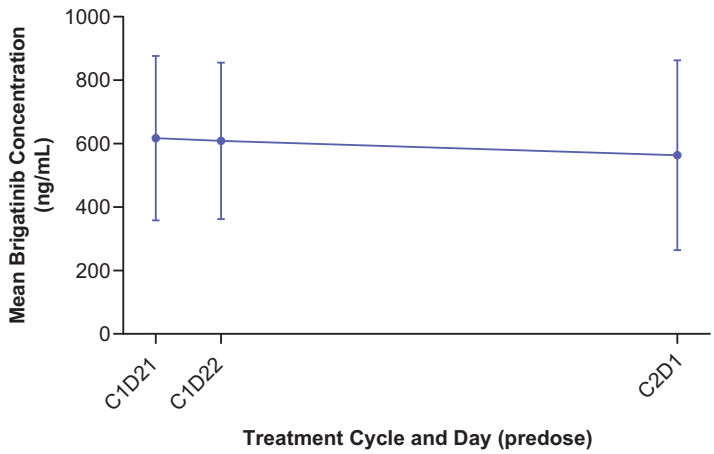


**Figure 3.** Mean brigatinib trough plasma concentrations on cycle 1 day 21, cycle 1 day 22, and cycle 2 day 1 following multiple-dose administration of brigatinib. Error bars represent standard deviation. C, cycle; D, day.

DDI assessment, the study was conducted in patients with cancer.

Administration of a single oral dose of midazolam in the presence of brigatinib resulted in lower mean plasma concentrations of midazolam throughout the entire 24-hour postdose period compared with midazolam administered alone. Coadministration with brigatinib reduced midazolam $C_{max}$ and $AUC_{\infty}$ by ≈16% (geometric LSM ratio, 0.836 [90%CI, 0.662-1.057]) and ≈26% (geometric LSM ratio, 0.741 [90%CI, 0.600-0.915]), respectively. The observed 26% reduction in midazolam $AUC_{\infty}$ is consistent with weak induction of CYP3A by brigatinib in vivo, as it just exceeded the ≥20% threshold used for defining a weak inducer.[23,24] Consequently, brigatinib has limited potential to cause clinically meaningful DDIs with CYP3A substrates (ie, only with CYP3A substrates where an ≈25% reduction in systemic exposure would be clinically meaningful).

Other ALK inhibitors have also been evaluated for their effects on CYP3A activity in vivo. Crizotinib is a moderate inhibitor of CYP3A as coadministration of 250 mg twice daily for 28 days increased oral midazolam $AUC_{\infty}$ by 3.7-fold compared with midazolam administered alone.[22] Coadministration of ceritinib 750 mg once daily under fasted conditions for 3 weeks increased oral midazolam $AUC_{\infty}$ by 5.42-fold versus midazolam administered alone; thus, ceritinib is a strong CYP3A inhibitor.[25] In contrast, alectinib has no clinically meaningful effect on CYP3A activity, as multiple-dose administration of 600 mg twice daily reduced oral midazolam $AUC_{\infty}$ by ≈3%.[18] Finally, lorlatinib is a moderate inducer of CYP3A; administration of a single oral dose of midazolam following lorlatinib 150 mg once daily for 15 days decreased midazolam $AUC_{\infty}$ by ≈60% compared with midazolam administered alone.[26]

Brigatinib was generally well tolerated in patients with *ALK*+ or *ROS1*+ NSCLC or other nonlung solid tumors enrolled in this study. However, it should be noted that the median treatment duration was relatively short at 68.5 days (range, 5-595 days). The TEAEs reported were consistent with the known safety profile of brigatinib in patients with NSCLC, and no new safety signals were observed.[3,4,7,11,27,28] TEAEs were mostly low grade in severity, with study drug–related grade ≥3 TEAEs occurring in 6 patients. Four patients discontinued treatment due to TEAEs, 2 of which were related to progression of NSCLC; 1 patient experienced pancreatitis that was considered a treatment-related SAE.

Evidence of brigatinib efficacy was observed, particularly in the *ALK*+ NSCLC cohort (n = 10) that demonstrated a 30% ORR with 3 partial responses. Notably, 1 of the responders in this cohort exhibited a DOR of 17.5 months. None of the patients in the other tumor cohorts (ie, *ROS1*+ NSCLC, *ROS1*+ nonlung solid tumors, or *ALK*+ nonlung solid tumors) had a confirmed response. Although the ORR reported in the *ALK*+ NSCLC cohort in this study was lower than the ORR reported for brigatinib in the ALTA (56%)[3] and ALTA-1L (74%)[28] trials, this finding is likely explained by the small sample size for the *ALK*+ NSCLC cohort (n = 10) in this drug interaction study, as well as the fact that all 10 patients received at least 1 prior line of therapy with a second-generation ALK inhibitor and that 7 of the 10 patients received ≥2 prior lines of therapy with an ALK inhibitor.

## Conclusions

In conclusion, this clinical DDI study demonstrated that brigatinib is a weak inducer of CYP3A, thereby indicating a limited potential for clinically meaningful DDIs with CYP3A substrates. The prescribing information for brigatinib has been updated to reflect the results from this clinical study.

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## Conflicts of Interest

Michael J. Hanley: employment with Takeda. Manolo D'Arcangelo: The author has no disclosures to report. Enriqueta Felip: consultancy fees and honoraria: AbbVie, Amgen, AstraZeneca, Bayer, Boehringer Ingelheim, Bristol Myers Squibb, Daiichi Sankyo, Eli Lilly, F. Hoffman-La Roche, GlaxoSmithKline, Ipsen, Janssen, Medical Trends, Medscape, Merck KGaA, MSD, Novartis, PeerVoice, Peptomyc, Pfizer, Sanofi, Springer, Takeda, and TouchTime; grants for research: Fundación Merck Salud, Grant for Oncology Innovation and Merck, Healthcare KGaA; and independent board member: Grifols. Pilar Garrido: consulting and advisory services: AbbVie, Amgen, AstraZeneca, Bayer, Bristol Myers Squibb, Boehringer Ingelheim, GlaxoSmithKline, Janssen, Eli Lilly, MSD, Novartis, Pfizer, Roche, and Takeda; speaking and public presentations: AstraZeneca, Bristol Myers Squibb, Boehringer Ingelheim, Janssen, MSD, Medscape, Novartis, Pfizer, Roche, Takeda, and TouchTime. Jiaxi Zhu, Meng

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## Data Sharing Statement

The data sets, including the redacted study protocol, redacted statistical analysis plan, and individual participant data supporting the results reported in this article, will be made available within 3 months from initial request to researchers who provide a methodologically sound proposal. The data will be provided after deidentification, in compliance with applicable privacy laws, data protection, and requirements for consent and anonymization.

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## Supplemental Information

Additional supplemental information can be found by clicking the Supplements link in the PDF toolbar or the Supplemental Information section at the end of web-based version of this article.