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ORIGINAL ARTICLE

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Lack of effect of ketoconazole-mediated CYP3A inhibition on sorafenib clinical pharmacokinetics

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Abstract Sorafenib is a novel, small-molecule anticancer compound that inhibits tumor cell proliferation by targeting Raf in the Raf/MEK/ERK signalling pathway, and inhibits angiogenesis by targeting tyrosine kinases such as vascular-endothelial growth factor receptor (VEGFR-2 and VEGFR-3) and platelet-derived growth factor receptor (PDGFR). In vitro microsomal data indicate that sorafenib is metabolized by two pathways: phase I oxidation mediated by cytochrome P450 (CYP) 3A4; and phase II conjugation mediated by UGT1A9. Approximately 50% of an orally administered dose is recovered as unchanged drug in the feces, due to either biliary excretion or lack of absorption. The aim of this study was to evaluate the effect of CYP3A inhibition by ketoconazole on sorafenib pharmacokinetics. This was an open-label, non-randomized, 2-period, one-way crossover study in sixteen healthy male subjects. A single 50 mg dose of sorafenib was administered alone (period 1) and in combination with ketoconazole 400 mg once daily (period 2) (ketoconazole was given for 7 days, and a single 50 mg sorafenib dose was administered concomitantly on day 4). No clinically relevant change in pharmacokinetics of sorafenib and no clinically relevant adverse events or laboratory abnormalities were observed in this study upon co-administration of the two drugs. Plasma concentrations of the main CYP3A4 generated metabolite, sorafenib N-oxide, decreased considerably upon ketoconazole co-administration. This effect is in accordance with the in vitro finding that CYP3A4 is the primary enzyme for sorafenib N-oxide formation. Further, these data indicate that blocking sorafenib metabolism by the CYP3A4 pathway will not lead to an increase in sorafenib exposure. This is consistent with data from a clinical mass-balance study that showed 15% of the administered dose was eliminated by glucuronidation, compared to less than 5% eliminated as oxidative metabolites. Since there was no increase in sorafenib exposure following concomitant administration of the highly potent CYP3A4 inhibitor ketoconazole with low dose sorafenib, it is postulated that higher therapeutic doses of sorafenib may be safely co-administered with ketoconazole, as well as with other inhibitors of CYP3A.

Keywords Sorafenib · Ketoconazole · CYP3A · Drug interaction · Anticancer · Raf Kinase inhibitor · Angiogenesis inhibitor · Pharmacokinetics

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Introduction

Sorafenib (4-{4-[3-(4-chloro-3-trifluoromethyl-phenyl) ureido]phenoxy} pyridine 2-carboxylic acid methylamide 4-methylbenzenesulfonate) is a novel oral Raf kinase and vascular-endothelial growth factor receptor (VEGFR) inhibitor that prevents tumor growth by combining two anticancer activities: inhibition of tumor cell proliferation and tumor angiogenesis [9]. Sorafenib inhibits tumor cell proliferation by targeting the Raf/MEK/ERK signaling pathway at the level of Raf kinase, and exerts an anti-angiogenic effect by targeting the receptor tyrosine kinases VEGFR-2, VEGFR-3 and platelet-derived growth factor receptor (PDGFR), and their associated signaling cascades [9].

Sorafenib has a mean half-life ranging from approximately 25 to 48 h. Following oral administration of [^{14}C]-sorafenib tosylate to healthy volunteers, approximately 19% of the dose is excreted in urine, almost exclusively as glucuronide conjugates of parent drug or metabolites; and 77% in feces (50% as unchanged drug). Approximately 17% of circulating radioactivity in plasma was in the form of sorafenib N-oxide. Sorafenib

undergoes glucuronidation by the UGT1A9 pathway and Phase I oxidative metabolism by the cytochrome P4503A4 (CYP3A4) enzyme system (data on file). The CYP3A4 isoform is responsible for the oxidation of more than half of all marketed drugs [8] and has been identified as the major CYP isoform involved in the oxidative metabolism of sorafenib (data on file). The metabolic scheme for sorafenib is shown in Fig. 1. Sorafenib N-oxide was the major Phase I metabolite formed in vitro and also the main circulating metabolite in human plasma. In addition, N-methylhydroxylation, N-demethylation and combinations of all three metabolic reactions led to minor metabolites both in vitro and in vivo. In addition to oxidative metabolism, ~15% of the administered dose of sorafenib was eliminated by glucuronidation, a Phase II metabolic pathway.

The potential for drug-drug interactions between concurrently administered CYP3A4 substrates and CYP3A4 inhibitors is high due to the contribution of the CYP3A4 enzyme to the metabolism of a wide range of drugs [15]. Further, like many new cytostatic anticancer agents, sorafenib has a relatively wide therapeutic index and is administered to patients with cancer who may be taking other concomitant medications. At doses that are above the recommended Phase III dose, there is an increased incidence of dose limiting toxicities such as hand-foot syndrome, fatigue and diarrhea. In vitro studies suggest that CYP3A4 is the primary Phase I oxidative pathway in the metabolism of sorafenib.. However, the relative contribution of the CYP3A4 pathway to the overall elimination of sorafenib was not known. Therefore, it was considered important to assess the extent to which modulation of CYP3A4 activity by other drugs may alter sorafenib exposure.

Ketoconazole is an imidazole antifungal drug that potently inhibits CYP3A enzymes both in vitro and in vivo [15, 16]. As a result, ketoconazole is widely used as a prototypical CYP3A inhibitor [1]. The primary objective of this study was to determine the effect of ketoconazole administration on the single-dose pharmacokinetics of 50 mg sorafenib in healthy volunteers. A sub-therapeutic dose of sorafenib was used in this study for safety reasons, because this was the first study to assess the interaction of sorafenib with a potent CYP3A inhibitor in healthy human volunteers and there was a concern about the potential magnitude of interaction. The secondary objective was to assess the clinical implications of co-administering sorafenib with CYP3A inhibitors based on the exposure and safety information obtained from this study.

Methods

Study design

This was an open-label, non-randomized, 2-period, one-way crossover study conducted in healthy male volunteers, in which sorafenib was administered alone (period 1) and concomitantly with ketoconazole (period 2) to determine the effect of potent CYP3A enzyme inhibition on sorafenib pharmacokinetics. A one-way crossover design was used for safety reasons to ensure that the subjects tolerated the single dose of sorafenib, prior to administration of the combination treatment. The study protocol was approved by the Heartland Institutional Review Board, Lenexa, KS and the study was conducted at Quintiles Phase I Services, Lenexa, KS in accordance with FDA guidelines for Good Clinical Practice (GCP). All subjects gave written informed consent prior to participating in this single-center study.

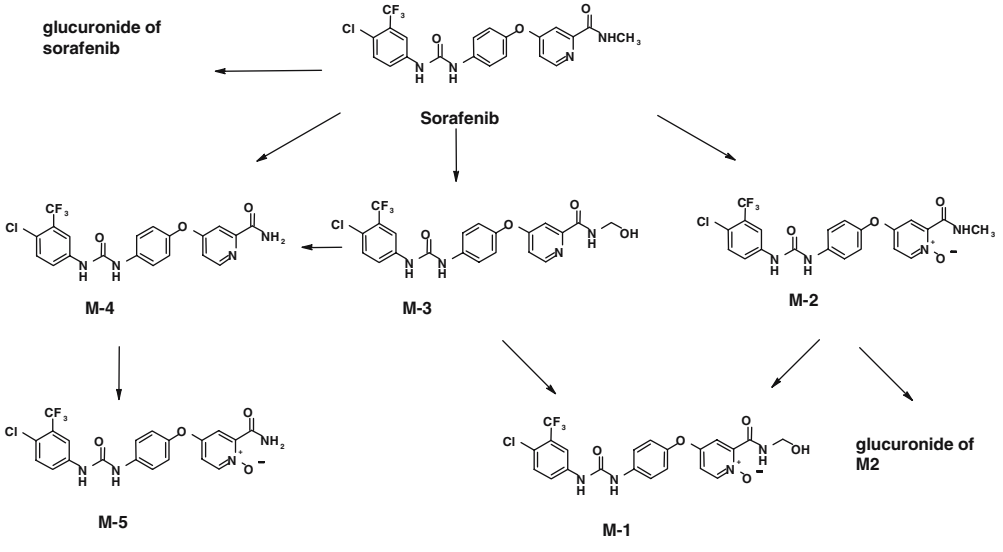

Fig. 1 Structure of the drug substance sorafenib (BAY 43-9006) which was administered as the tosylate salt (BAY 54-9085)

On day 1 of period 1, all subjects received a single oral dose of sorafenib tosylate 50 mg tablet (calculated as free base sorafenib). After at least a 10-day washout, 400 mg ketoconazole (Nizoral, Janssen Pharmaceutica, Titusville, NJ; Batch 91P0982E) was administered for three consecutive days in period 2. On day 4 in period 2, a single dose of sorafenib tosylate 50 mg tablet (calculated as free base sorafenib) from the same batch as in period 1 was co-administered with 400 mg ketoconazole. Sorafenib doses were administered in the fasted state with 240 ml of water. Ketoconazole was administered for three additional days in period 2 (days 5, 6 and 7) to maintain CYP3A inhibition for the duration of blood sampling. Blood samples (5 ml) for the determination of sorafenib and its metabolite concentrations in plasma were collected before and 0.5, 1, 2, 3, 4, 6, 8, 12, 16, 24, 48, 72, 96, 120, and 168 h after drug administration. Samples were prepared by centrifugation to obtain plasma, which was frozen at −20° C until analysis.

During period 1, subjects remained in the clinic from the evening of Day-1 until the morning of day 5. The final sorafenib dose was given on day 4. Subjects returned to the clinic for a brief visit on the mornings of days 6 (48 h) and 8 (96 h)to have blood drawn for sorafenib pharmacokinetics. During period 2, subjects remained in the clinic from the evening of Day-1 until the morning of day 8. They returned to the clinic on the mornings of days 9 (120 h) and 11 (168 h) for sorafenib pharmacokinetic sample collection.

Sample size

From an earlier BAYER sorafenib study, the within-subject standard deviation for the log-transformed $AUC_{(0-\infty)}$ was estimated to be 0.36. For the current study, with 16 subjects, and assuming a within-subject SD for log-transformed $AUC_{(0-\infty)}$ of 0.36, a 90% confidence interval for the ratio of least squares geometric means $AUC_{(0-\infty)}$ of the two treatments (i.e., sorafenib administered with or without ketoconazole) would have a lower/upper limit equal to the observed ratio of means divided/multiplied by 1.251. If the observed ratio of means was 1.0, the 90% confidence interval for the ratio of least squares geometric means $AUC_{(0-\infty)}$ of the two treatments would be (0.799, 1.251).

Although the 90% confidence interval calculated above approximately met the standard bioequivalence criteria of the ''no effect'' bounds 80–125%, this study was considered an observational trial conducted to determine the magnitude of change in sorafenib pharmacokinetics with concomitant ketoconazole administration. This is consistent with the FDA's Guidance for Industry entitled, ''In Vivo Drug Metabolism/Drug Interaction Studies—Study Design, Data Analysis, and Recommendations for Dosing and Labeling'' [4]. This study, with 16 subjects, provided adequate information on the effect of ketoconazole on sorafenib pharmacokinetics.

Subjects

Sixteen healthy male volunteers participated in the study. Subjects were determined to be healthy on the basis of a physical examination and laboratory screening prior to study initiation. Subjects were required to test negative for a urine drug screen and also for HIV, Hepatitis B and Hepatitis C. Subjects with a history of major medical or organ disorder, recent illness, malignancy, psychiatric or mental illness, chronic dermatitis or skin condition, clinically significant allergic disease, abnormal laboratory values, drug hypersensitivity or allergies to investigational agents, or those who had undergone treatment with prescription medication, cimetidine or herbal products within 14 days of day 1, period 1 were excluded from the study. Further, use of these products during the study was prohibited. Subjects who had a history of drug or alcohol abuse, current smoking (ten cigarettes or equivalent per day) or excessive consumption of caffeine containing products (>5 eight ounce cups of coffee or caffeine containing beverages) or had donated blood or been treated with an investigational agent within 30 days prior to study commencement were excluded from the study. Over-the-counter medications were not allowed for 48 h before and 48 h after dosing in each period. Consumption of grapefruit or grapefruit juice or products containing grapefruit juice was not allowed for 72 h before and 72 h after sorafenib or ketoconazole administration.

Safety assessment

A 12-lead ECG (supine) recording was assessed as part of the screening process and at the last visit to the clinic (period 2, day 11). Safety and tolerability were assessed by clinical laboratory analyses (hematology, chemistry, and urinalysis), vital sign measurements and wellness assessment. Hematology and chemistry were performed as part of the pre-screening (no more than 21 days before study start) and/or on period 1, Day 1 (pre-dose) and period 2, Days 1 (pre-dose), 4 (pre-dose), 7 (pre-dose) and 11 (end of study). Urinalysis was performed as part of the screening process and at the last visit to the clinic (period 2, day 11). Vital sign assessments were performed on period 1, day 1 through 5 and period 2, day 1 through 7. Wellness assessments were performed on each visit. All adverse events were documented in detail.

Bioanalytical methods

A validated LC/MS/MS method, specific for the measurement of sorafenib and its metabolite concentrations in human plasma was used. In brief, an aliquot of plasma (0.2 mL) was transferred into a screw cap culture tube (13×100 mm), internal standard solution (40 μL) containing 40 ng of [$^{2}H_{3}$, ^{15}N]-sorafenib tosylate were

added and the sample was mixed. Samples were extracted with methyl-t-butyl ether (2.5 mL) on an automatic shaker at 200 rpm for 15 min and then centrifuged at 3,000 rpm (3,200 *g*) for 5 min at 4°C. After centrifugation, each test tube was immersed in a dry ice and acetone bath to flash-freeze the aqueous layer. The organic solvent was decanted and evaporated to dryness at 50°C under a stream of nitrogen for about 12 min. The samples were reconstituted in acetonitrile (100 µL) and transferred to an autosampler vial. An aliquot (10 µL) of this solution was injected into the LC/MS/MS system. The mobile phase consisted of 10 mM ammonium acetate (pH = 3.0) : acetonitrile (10:90, v/v) and was delivered at a flow rate of 0.7 ml/min (Perkin Elmer Series 200 LC pump with Series 200 autosampler). The chromatography column (Zorbax SB-C8 Column, 4.6×150 mm, 3.5 µm) was maintained at 30°C. Detection was by tandem mass spectrometry using a Sciex 365 mass spectrometer with a heated nebulizer interface operating in the multiple reaction monitoring, positive ion mode. The calibration range for the method was 0.01 to 12 mg/L for sorafenib and 0.01 to 2.5 mg/L for sorafenib N-oxide. The lower limit of quantitation (LOQ) for sorafenib and its metabolite was 0.01 mg/L and the intra- and inter-day coefficients of variation were less than 8%. The accuracy and precision at the lowest calibrator (0.01 mg/L) were 99.4% and 4.2% respectively for sorafenib and 98.7% and 2.62% respectively for sorafenib N-oxide. Quality control samples were analyzed along with the samples of analytes of interest.

Pharmacokinetic and statistical analysis

Pharmacokinetic parameters were calculated using standard non-compartmental methods. The following parameters were determined from the plasma concentration-time profiles of sorafenib and its N-oxide metabolite: maximum observed plasma concentration (C_{max}); the area under the plasma concentration-time curve (AUC) from time 0 to the last quantifiable concentration at time t ($AUC_{(0-t)}$), determined by the log-linear trapezoidal rule; and AUC extrapolated to infinity ($AUC_{(0-\infty)}$), calculated as $AUC_{(0-t)} + C_n/\lambda_z$, where λ_z is the terminal elimination rate constant determined by regression analysis, and C_n is the last measurable concentration. The half-life ($t_{1/2}$) was calculated as $0.693/\lambda_z$.

Descriptive statistics of plasma concentrations and the derived pharmacokinetic parameters were calculated for sorafenib given alone and in combination with ketoconazole. The natural logarithms of $AUC_{(0-\infty)}$ and C_{max} and other derived pharmacokinetic variables were analyzed using analysis of variance (ANOVA) with terms for treatment and subject. A 90% two-sided confidence interval (CI) for the ratio of geometric means of the two treatment conditions was calculated. Equality of means was evaluated at the 0.05 significance level. Statistical analyses were performed using SAS, version 8.0 (SAS Institute, Cary, NC, USA).

Results

Patient baseline statistics/demographics

The study was conducted in healthy male subjects with a mean (± SD) age of 28 ± 9 years (range 21–45 years). The mean (± SD) weight was 81 ± 12.6 kg (range 56–101 kg); the mean (± SD) body mass index was 25.8 ± 3.2 (range 19.8–31.6) and the mean (± SD) height was 176.2 ± 5.9 cm (range 167–183 cm). There were 12 Caucasians and four African Americans. One subject who discontinued prematurely was an African American male, who weighed 101 kg, and was 183 cm tall.

Effect of ketoconazole on sorafenib pharmacokinetics

Fifteen of the 16 subjects completed both treatment periods and were valid for the pharmacokinetic analysis. The mean plasma concentration-time profiles of sorafenib and sorafenib N-oxide with and without ketoconazole are shown in Fig. 2 and the individual $AUC_{(0-\infty)}$ and C_{max} data are shown in Fig. 3. The pharmacokinetic parameter estimates by treatment are presented in Table I. Two other oxidative metabolites seen in in vitro studies and described in the introduction section were below quantitation limits in this study. Overall, the mean concentration-time profiles of sorafenib were similar with and without ketoconazole (Fig. 2) and co-administration of ketoconazole (400 mg per day) did not affect the single-dose pharmacokinetics of sorafenib 50 mg. The geometric means for $AUC_{(0-\infty)}$ and C_{max} were marginally smaller in the presence of ketoconazole. The geometric mean $AUC_{(0-\infty)}$ values for sorafenib were 11.04 and 9.82 mg*h/L, respectively (Table 1), when administered alone and in combination with ketoconazole. The corresponding geometric mean C_{max} values for sorafenib were 0.46 and 0.34 mg/L respectively when administered alone and in combination with ketoconazole. The terminal elimination $t_{1/2}$ of sorafenib was approximately 30 h in each period. The geometric mean ratio (GMR) (with corresponding 90% CI in parentheses) for sorafenib $AUC_{(0-\infty)}$ was 0.89 (0.69–1.14) and the GMR (with corresponding 90% CI in parenthesis) for sorafenib C_{max} was 0.74 (0.56–0.97).

Although ketoconazole administration did not influence sorafenib pharmacokinetics, it did affect the pharmacokinetic profile of the metabolite, sorafenib N-oxide, as shown in Fig. 2. Whereas sorafenib N-oxide was measurable following administration of sorafenib alone, in the presence of ketoconazole, plasma metabolite concentrations were measurable in only one sample

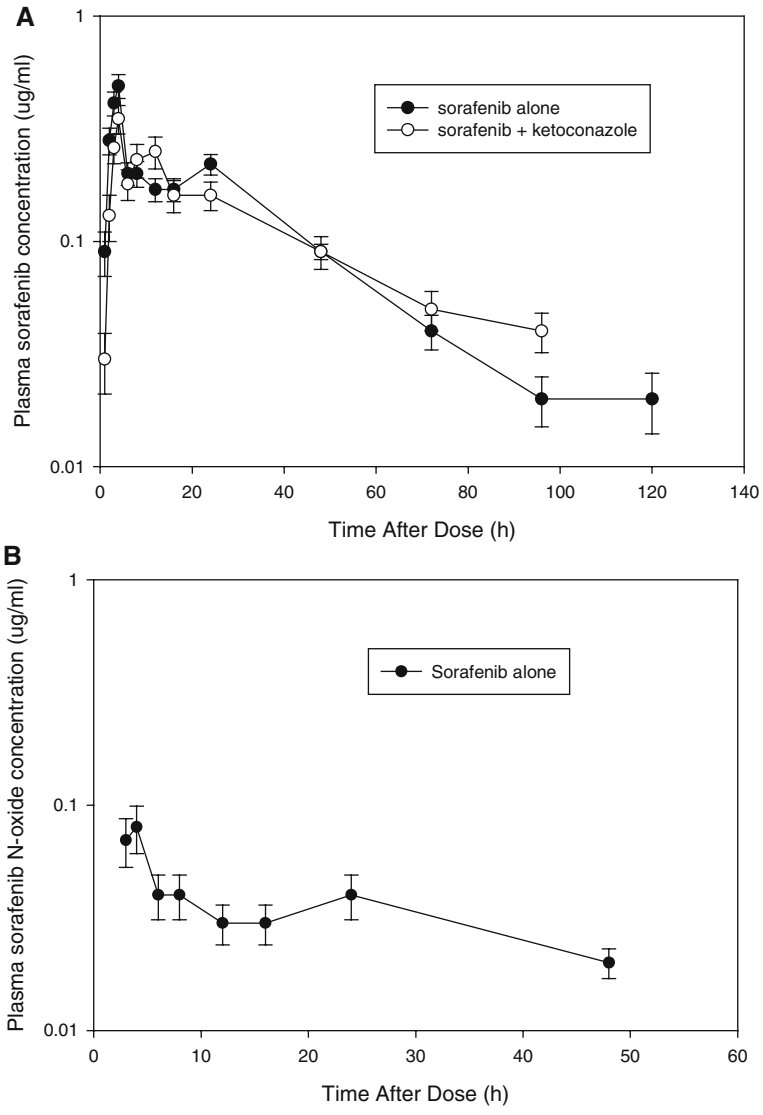

Fig. 2 Mean (± SE) plasma concentration* versus time plots for **(A)** Sorafenib and **(B)** Sorafenib N-oxide following a single oral dose of sorafenib 50 mg alone and in combination with ketoconazole 400 mg once daily for 7 days. *Sorafenib N-oxide concentrations were below limit of quantitation during the sorafenib-ketoconazole combination treatment, and beyond 48 h following dosing with sorafenib alone

from one subject, indicating inhibition of formation of this metabolite.

Safety and tolerability

Fifteen subjects completed both periods of the study. One subject did not return to the clinic for period 2 for reasons unrelated to the drug treatment. All 16 subjects were valid for the safety evaluation. There were no clinically relevant changes in vital signs, ECG recordings, or laboratory safety parameters for any subject. Four subjects of sixteen (25%) reported at least one treatment-emergent adverse event during administration of sorafenib alone (period 1), compared to five subjects of fifteen (33%) dosed with the combination of sorafenib and ketoconazole (period 2). Two subjects of fifteen (13%) reported at least one treatment-emergent event during the ketoconazole treatment preceding sorafenib dosing in period 2. One subject of fifteen (7%) reported drug-related adverse events during the sorafenib and ketoconazole combination treatment. The adverse events included nausea, tremor, and hypotension. No serious adverse events were reported in this study. Sporadic laboratory abnormalities were transient and slight. None of these were deemed to be clinically relevant. No changes or trends with respect to vital signs were observed in association with sorafenib dosing across the two periods. There was no individual QT interval greater than 450 ms. Thus, sorafenib was considered safe and well tolerated both with and without ketoconazole administration when administered at a low dose.

Discussion

Ketoconazole is a potent inhibitor of CYP3A activity [15, 16]. Potent inhibition of CYP3A in vitro has been demonstrated, with an average competitive inhibition con-

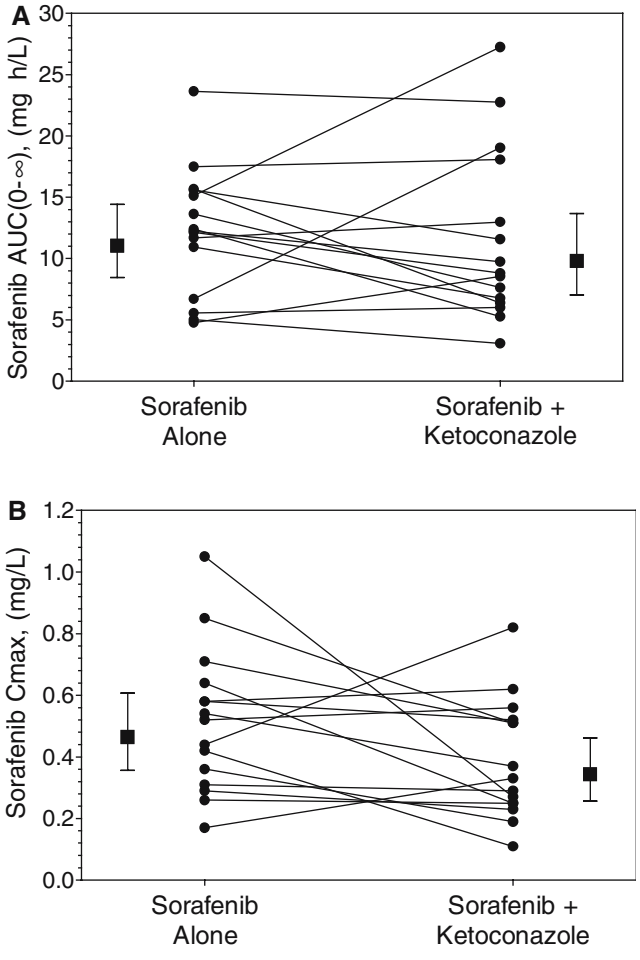

Fig. 3 Sorafenib **(A)** $AUC_{0-\infty}$ and **(B)** C_{max} after administration alone and during co-administration of ketoconazole 400 mg per day ($n = 15$). The solid square represents the geometric mean and 95% confidence interval

stant (K_i) for 1′-hydroxymidazolam formation of 14.9 nM in human liver microsomes and 26.7 nM in cDNA-expressed CYP3A4 [5]. Ketoconazole is classified as a strong in vivo CYP3A inhibitor on the basis of its effects on oral midazolam AUC [1]. Ketoconazole 400 mg daily for four days caused a 16-fold increase in the AUC of midazolam after oral administration [10]. Ketoconazole administration has also been shown to markedly affect the pharmacokinetics of a number of other CYP3A substrates including alprazolam, cisapride, felodipine, lovastatin, nifedipine, simvastatin, terfenadine, and triazolam leading to clinically relevant drug-drug interactions [15]. Therefore, ketoconazole was chosen as the inhibitor in this study to assess the effect of CYP3A4 inhibition on the pharmacokinetics of sorafenib.

Sorafenib is a novel Raf kinase and VEGFR inhibitor that is being investigated for use in a variety of cancers [3, 6, 7, 11, 13]. Sorafenib is eliminated by a combination of CYP3A4-mediated oxidative metabolism, Phase II glucuronidation, and (possibly) biliary secretion, with glucuronidated metabolites accounting for approximately 19% of an oral dose. After co-administration of a single dose of sorafenib on a background of daily dosing with ketoconazole, there was no increase in sorafenib AUC and C_{max} mean values, and no change in terminal elimination half-life, compared to administration of sorafenib alone. There was a small decrease in mean sorafenib AUC when co-administered with ketoconazole. Because of sorafenib's intra-subject variability, some subjects showed an increase in $AUC_{(0-\infty)}$ whereas other subjects showed a decrease in $AUC_{(0-\infty)}$ upon co-administration of sorafenib with ketoconazole. The maximum increase in exposure, in one subject, was a 3-fold increase in AUC. There were no adverse events as a result of this increase. Sorafenib is well tolerated over a 10-fold range of exposures in clinical studies in cancer patients. The change in exposure seen in these subjects is well within this range and is not likely to lead to safety or tolerability issues in the clinical setting [14].

The formation of sorafenib N-oxide was decreased by ketoconazole, consistent with ketoconazole's known activity as a CYP3A inhibitor. However, this decrease did not result in an increase in sorafenib plasma con-

Table 1 Pharmacokinetic parameters (geometric mean, % CV) of sorafenib and sorafenib N-oxide following administration of a single 50 mg dose either alone or in combination with ketoconazole

Parameter	Sorafenib only		Sorafenib + ketoconazole		Geometric mean ratio (90% CI) (combination: sorafenib alone)
	N	Geometric mean (% CV)	*N*	Geometric mean (% CV)	
Sorafenib					
$AUC_{0-\infty}$ (mg*h/L)	15	11.04 (48%)	15	9.82 (60%)	0.89 (0.69–1.14)
$AUC_{(0-t)}$ (mg*h/L)	15	10.28 (50%)	15	8.81 (66%)	0.86 (0.66–1.11)
C_{max} (mg/L)	15	0.46 (48%)	15	0.34 (54%)	0.74 (0.56–0.97)
$t_{1/2}$ (h)	15	29.0 (35%)	14	30.2 (30%)	1.01 (0.80–1.29)
Sorafenib N-oxide					
$AUC_{0-\infty}$ (mg*h/L)	11	3.30 (63%)	0	ND[a]	ND[a]
$AUC_{(0-t)}$ (mg*h/L)	13	1.11 (152%)	1	—[b]	ND[a]
C_{max} (mg/L)	13	0.07 (97%)	1	0.01[b]	ND[a]
$t_{1/2}$ (h)	5	30.0 (28%)	0	ND[a]	ND[a]

[a]ND-Not determined since levels of sorafenib N-oxide were below the limit of quantitation (LOQ)
[b]Sorafenib N-oxide concentrations were detected in a single sample from 1 subject only

centrations. The effect of ketoconazole is more pronounced on high clearance drugs [2] that are exclusively metabolized by the CYP3A4 pathway. This is consistent with sorafenib either being a low clearance drug and/or a drug that is not predominantly eliminated by CYP3A4 metabolism. In vitro metabolism studies with human hepatocytes have shown that sorafenib is metabolized to form an N-oxide by the CYP3A4 pathway. Additionally, sorafenib is metabolized by the Phase II glucuronidation pathway. In vivo mass balance studies in humans using radiolabeled sorafenib, have demonstrated that ~19% of the administered dose is excreted in the urine, primarily as sorafenib glucuronide (~15% of dose), and ~77% of the drug dose is eliminated in the feces. Neither the N-oxide metabolite nor the glucuronide metabolite of sorafenib are detected in feces. This may be due to instability of these metabolites in the presence of bacterial reductases and bacterial glucuronidases present in the GI tract. Therefore, the actual fraction of administered dose excreted in feces by the oxidative or glucuronidation pathway may have been underestimated in the mass balance study. However, available urinary excretion data indicates that glucuronidation plays an important role in the elimination of sorafenib. Data from this study showing that blocking the CYP3A4 based oxidative pathway does not lead to an increase in sorafenib exposure, affirms the importance of the glucuronidation pathway in sorafenib's elimination. Glucuronidation is not considered a limiting pathway under normal physiologic conditions and is not saturated under normal circumstances. Therefore, even though observed data indicates that sorafenib glucuronide elimination accounts for only 15% of the dose when the drug is given alone, more sorafenib could be eliminated by this pathway if the CYP3A4 based oxidative pathway is blocked. Given the importance of the glucuronidation pathway in the elimination of sorafenib and that it is not saturated under normal circumstances, the likelihood of an increase in sorafenib exposure following inhibition of the CYP3A pathway is low.

Since the effect of co-administration of ketoconazole on the pharmacokinetics of sorafenib was unknown, the 50 mg dose was intentionally selected for safety reasons to avoid potential over-exposure of the compound in healthy volunteers. Higher sorafenib doses have been used in the clinic with 400 mg bid being the dose used in Phase III trials. Throughout the dosing range, up to clinically evaluated doses of 800 mg bid, the ratio's of C_{max} and AUC of CYP3A4 mediated oxidative metabolites to the C_{max} and AUC of sorafenib are similar, indicating linearity of CYP3A4 mediated metabolism at and above the therapeutic dose (unpublished data). If the CYP3A4 pathway were blocked by ketoconazole, when administering therapeutic doses of sorafenib, more sorafenib would likely get metabolized by the glucuronidation pathway. Since glucuronidation is a high capacity pathway, it is not expected to saturate at higher sorafenib doses and therefore a large CYP3A4-mediated interaction would not be expected following administration of CYP3A4 inhibitors. Data from this study indicate that a low dose of sorafenib can be safely co-administered with drugs known to inhibit CYP3A-mediated metabolism (e.g., azole antifungals including ketoconazole). The interaction between the therapeutic dose (400 mg bid) of sorafenib with ketoconazole has not been investigated. The fraction of sorafenib that is metabolized by the Phase I oxidative pathway appears to be low, i.e., $< 30\%$, (data on file). It is also metabolized by Phase II glucuronidation. Therefore, the likelihood of a clinically significant increase in sorafenib exposure, at the therapeutic dose, when administered with a CYP3A4 inhibitor is low [12].

Consistent with the comparable sorafenib plasma concentrations seen in the presence and absence of ketoconazole, no differences in the side effect profile were observed in the two treatment periods. No clinically relevant drug-related adverse events or laboratory abnormalities were observed in this study. Based on the pharmacokinetic and safety data obtained from this study, the results suggest that sorafenib may be safely administered at low dose with drugs known to inhibit CYP3A-mediated metabolism and that no dose adjustment may be necessary during concomitant therapy. Additionally, the likelihood of a clinically significant increase in sorafenib exposure, at the therapeutic dose, is low. In a recently completed clinical trial, patients who received 400 mg bid sorafenib chronically with CYP3A4 inhibitors had a similar safety profile to those who did not receive CYP3A4 inhibitors.

Conclusions

This study demonstrated that co-administration of the strong CYP3A inhibitor ketoconazole had no effect on the $AUC_{(0-\infty)}$ and C_{max} of sorafenib at a single 50 mg dose. Consistent with ketoconazole's known activity as an inhibitor of CYP3A, the formation of the Phase I oxidative metabolite, sorafenib N-oxide, was decreased by ketoconazole. However, this decrease did not result in a corresponding increase in plasma concentrations of sorafenib, possibly because other elimination pathways (e.g., glucuronidation) may also play a significant role in sorafenib elimination. Upon co-administration of a single 50 mg dose of sorafenib with ketoconazole, there was no apparent change in sorafenib exposure or its side effect profile. This study indicates that no dose adjustment of sorafenib is necessary upon co-administration with ketoconazole. Although this study investigated a 50 mg sorafenib dose, rather than the currently proposed therapeutic dose of 400 mg, the results are considered relevant to the higher dose because (a) the relative contribution of CYP3A4 to sorafenib elimination is fairly stable over the dose range of 50 to 400 mg,and (b) sorafenib has alternate elimination pathways that account for a significant fraction of the administered dose. It is considered unlikely that ke-

toconazole would produce a clinically significant increase in sorafenib exposure.

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