Original article

Single- and multiple-dose pharmacokinetics of etravirine administered as two different formulations in HIV-1-infected patients

Thomas N Kakuda*, Monika Schöller-Gyüre, Cassy Workman, Keikawus Arasteh, Anton L Pozniak, Goedele De Smedt, Greet Beets, Monika Peeters, Kati Vandermeulen, Brian J Woodfall and Richard MW Hoetelmans

1Tibotec Inc, Yardley, PA, USA
2Tibotec BVBA, Mechelen, Belgium
3AIDS Research Initiative, New South Wales, Australia
4Epimed, Berlin, Germany
5Chelsea and Westminster Hospital, London, UK

*Corresponding author: E-mail: tkakuda@its.jnj.com

Background: An open-label, randomized, crossover study to evaluate the pharmacokinetics of two different formulations of etravirine after single and multiple dosing.

Methods: Treatment-experienced HIV-1-infected patients with viral load <50 copies/ml continued their current antiretroviral regimen and added etravirine twice daily for 7 days with a morning intake on day 8. Etravirine was administered following food as either 800 mg twice daily of the Phase II formulation or 100 mg or 200 mg twice daily of the Phase III formulation. A 12 h pharmacokinetic assessment was performed on days 1 and 8.

Results: After single- and multiple-dose administration, the exposure to etravirine was lower with 100 mg twice daily and higher with 200 mg twice daily compared with 800 mg twice daily. On day 8, the mean (±sd) area under the plasma concentration-time curve over 12 h (AUC0–12h) was 1,284 (±958) ng•h/ml when etravirine was administered as 100 mg twice daily (n=33), 3,713 (±2,069) ng•h/ml when administered as 200 mg twice daily (n=27) and 2,607 (±2,135) ng•h/ml when administered as 800 mg twice daily (n=32). Both formulations and all doses of etravirine tested were generally safe and well tolerated.

Conclusions: The range of exposure to etravirine was comparable between 200 mg twice daily dose and 800 mg twice daily. The Phase III formulation of etravirine significantly improves the bioavailability of etravirine over the Phase II formulation with reduced interpatient variability in etravirine pharmacokinetics.

Introduction

Non-nucleoside reverse transcriptase inhibitors (NNRTIs) are among the currently available antiretroviral drugs used to treat HIV infection [1]. However, a single mutation in the virus could lead to resistance to first-generation NNRTIs. Etravirine (formerly TMC125) is a second-generation NNRTI developed with the aim of having activity against both wild-type and NNRTI-resistant HIV-1 [2,3]. Due to its poor solubility and low to moderate permeability, etravirine is considered a Biopharmaceutics Classification System class IV compound. Etravirine is metabolized by cytochrome P450 (CYP) isozymes 3A, 2C9 and 2C19 with the metabolites undergoing glucuronidation via uridine diphosphogluconosyl transferase; etravirine is not a substrate for P-glycoprotein. Interaction data obtained to date suggest that etravirine is an inducer of CYP3A and a weak inhibitor of CYP2C9, CYP2C19 and P-glycoprotein [4,5]. A mass balance study determined 81.2–84.6% of etravirine is excreted unchanged in the faeces with minimal (<1.2%) renal excretion. The mean (±sd) terminal elimination half-life of etravirine was ~41 (±20) h [6]. Administration of etravirine after fasting significantly decreases exposure by 50% relative to administration following a meal [7]. Moderate inter- and intrapatient variability (60% and 40%, respectively) has been observed in HIV-infected patients treated with etravirine [8].

© 2008 International Medical Press 1359-6535
Etravirine has demonstrated antiviral activity in short-term (functional) monotherapy trials conducted in both treatment-naive and NNRTI treatment-experienced HIV-1-infected patients [9,10]. A subsequent Phase IIb dose-ranging trial with 199 HIV-1-infected patients with documented genotypic resistance to the NNRTI and protease inhibitor classes showed that etravirine with an investigator-selected optimized background regimen consisting of at least two antiretrovirals (nucleoside or nucleotide reverse transcriptase inhibitors and/or lopinavir/ritonavir and/or enfuvirtide) was effective after 24 weeks with both doses of etravirine tested (400 mg and 800 mg twice daily Phase II formulation) compared with a control group that received at least three investigator-selected antiretrovirals. Patients receiving 800 mg twice daily of etravirine showed a 1.18 log10 copies/ml reduction in HIV RNA versus a 0.19 log10 copies/ml reduction in the control group [11]. After 48 weeks, these values were 1.01 and 0.14 log10 copies/ml, respectively, demonstrating the sustained efficacy of etravirine [12]. Treatment with etravirine was generally safe and well tolerated [11,12]. Based on the results of the Phase IIb trials, the selected dose of etravirine was 800 mg twice daily, that is, four 200 mg tablets twice daily.

Concurrent to the Phase II trials, a process to reduce the pill burden and potentially improve adherence was undertaken. Solid dispersions using spray-drying technology is one method of increasing the bioavailability of poor water soluble drugs [13]. Using this process led to a new formulation of etravirine (Phase III formulation) with substantially improved bioavailability. On the basis of earlier clinical trials with single doses of the Phase III formulation, an increase in exposure of approximately ninefold was anticipated for the Phase III formulation compared with the Phase II formulation [14]. Therefore, a dose regimen of 100 mg twice daily of the Phase III formulation was expected to show comparable exposures to those obtained with 800 mg twice daily of the Phase II formulation. However, preliminary pharmacokinetic data obtained in the first two treatment periods of this trial failed to confirm this. After 8 days of treatment with etravirine, the least squares mean (LSM) ratio of the area under the plasma concentration-time curve over 12 h (AUC0–12h) for 100 mg twice daily of the Phase III formulation compared with 800 mg twice daily of the Phase II formulation was 0.54 instead of the expected 1. Thus, an optional additional treatment period with 200 mg twice daily of the Phase III formulation was added to this trial.

The primary objective of this multiple-dose crossover trial was to evaluate the pharmacokinetics of etravirine administered as two different formulations in HIV-1-infected patients. The short-term safety and tolerability of etravirine was also assessed.

Methods

Study population and study design
This was an open-label, randomized, crossover trial to evaluate the pharmacokinetics and relative oral bioavailability of etravirine after multiple dosing administered as the Phase III tablet formulation compared with the reference Phase II tablet formulation in HIV-1-infected patients. In addition to being HIV-1-infected, patients had to be between 18 and 55 years of age, have at least 3 months of documented NNRTI experience, a confirmed plasma viral load <50 HIV-1 RNA copies/ml and taking a current antiretroviral regimen that included lopinavir/ritonavir, saquinavir/ritonavir or lopinavir/saquinavir/ritonavir and at least one nucleoside or nucleotide reverse transcriptase inhibitor with or without enfuvirtide. The inclusion of lopinavir/ritonavir, saquinavir/ritonavir or lopinavir/saquinavir/ritonavir was based on previously conducted drug interaction trials showing no clinically relevant effect of these protease inhibitors on the pharmacokinetics of etravirine [15,16]. Patients were excluded if, in the opinion of the investigator, the patient had a history or suspicion of alcohol or illicit drug use that could compromise the patient’s safety and/or compliance with the trial procedures or any active clinically significant disease that may affect the outcome of the trial or could influence drug absorption or bioavailability. Patients continued to take their current antiretroviral regimen throughout the trial without interruption. Three clinical research sites (in Australia, Germany and the United Kingdom) participated in this trial.

A minimum of at least 28 patients completing all sessions was considered sufficient to allow for relevant conclusions. The estimated log-transformed intrapatient variability for etravirine AUC was 0.182 based on previous pharmacokinetic trials conducted in healthy patients with the Phase II formulation; among 28 patients the corresponding range of the 90% confidence interval (CI) of the ratio was estimated to be 0.82–1.21 (within the limits of bioequivalence).

The trial was conducted in three treatment sessions. In session 1, patients received etravirine as the Phase III formulation at a dose of 100 mg (taken as one 100 mg tablet) twice daily. In session 2, patients received the reference (Phase II) formulation of etravirine at a dose of 800 mg (taken as four 200 mg tablets) twice daily. Patients were randomized to start with either session 1 or 2 in a 1:1 ratio. Subsequently, in an optional third period, patients received etravirine as the Phase III formulation at a dose of 200 mg (taken as two 100 mg tablets) twice daily. As this third period was optional, there was no minimum to the number of patients to be included.
The three treatment sessions were separated by a washout period of at least 14 days. Patients were admitted to the testing facility on day 1 and day 8 of each treatment session. All doses of etravirine were to be taken following a meal, within 10 min after completion of a breakfast or dinner, with ~200 ml of water. On days 1 and 8 of all treatment sessions (pharmacokinetic sampling days) patients received a standard breakfast consisting of four slices of bread, two slices of ham or cheese, butter, jelly and two cups of decaffeinated coffee or tea with milk and/or sugar. The time of each intake of study medication and the start and stop times of meals served in the testing facility were recorded.

Pharmacokinetic assessments
In all treatment sessions, plasma samples for the measurement of etravirine plasma concentrations were taken on day 1 before dosing (within 2 h prior to drug intake) and at 1, 2, 3, 4, 6, 8, 10 and 12 h after dose administration, on days 6 and 7 before dosing and on day 8 before dosing and at 1, 2, 3, 4, 6, 8, 10 and 12 h after dose administration. Exact times of blood sampling for pharmacokinetics were recorded. Plasma samples were stored at ≤18°C until assayed. Plasma concentrations of etravirine were determined using a validated liquid chromatography with tandem mass spectrometry method (LC-MS/MS). For this assay, an aliquot of heparinized plasma (0.3 ml) was diluted with water and spiked with the stable isotope-standard: a mixture of 0.1% (v/v) formic acid and acetonitrile (20/80, v/v) was used as the elution mixture. Detection was carried out using MS/MS in the electropray-positive mode. The following transitions were monitored: m/z 440±0.5 to 209±0.5, m/z 435±0.5 to 164±0.5, and m/z 440±0.5 to 164±0.5, respectively. The effective linear range was 2–5,000 ng/ml for undiluted samples.

Pharmacokinetic and statistical methods
Pharmacokinetic analysis was carried out using SAS System for Windows® version 8.2 (SAS Institute Inc., Cary, NC, USA). A non-compartmental model with extravascular input was used for the pharmacokinetic analysis. For the calculation of the individual pharmacokinetic parameters, plasma concentrations below the lower limit of quantification (BLQ) were treated as being 0 in case of occurrence before the first measurable plasma concentration and treated as missing after the last measurable plasma concentration. Furthermore, no sample (missing sample) and intermediate not quantifiable (NQ) values were excluded from pharmacokinetic calculations. If more than one intermediate NQ value occurred, NQs and following values were excluded from the calculation. For statistical calculations, BLQ values were treated as 1 ng/ml (0.5× the lower limit of quantification). For graphical analysis, BLQ values were treated as being 0 in case of occurrence before the first measurable plasma concentration. In all other cases, they were not included in the graph.

The trough, minimum and the maximum plasma concentrations (C0 h, Cmin and Cmax, respectively) and time to reach Cmax (tmax) were obtained by inspection of the plasma concentration-time profiles. The Cmin was the lowest plasma concentration observed within a dosing interval. The AUC0–12 h was determined using the linear trapezoidal rule. Descriptive statistics were calculated for the plasma concentrations of etravirine at each time point and for all derived pharmacokinetic parameters.

Statistical analyses were performed using the Phase III formulation as test and the Phase II formulation as reference. The primary pharmacokinetic parameters were Cmax, AUC0–12 h and Cmin on the logarithmic scale. All available observations were included in the statistical analysis. The LSM of the primary parameters for each dose group were estimated with a linear mixed effects model, controlling for period and randomization group as fixed effects and patient (nested in a treatment group) as a random effect. A 90% CI was constructed around the difference between the LSM of the test and the reference group. Both the difference between the LSM and the 90% CIs were retransformed to the original scale. Pre-dose plasma concentrations on days 6, 7 and 8 were compared graphically to verify the achievement of steady-state conditions for etravirine in all treatment periods.

Safety evaluations
Safety and tolerability were assessed throughout the study until at least 30 days after the last study drug intake using adverse events, clinical laboratory evaluations, physical and skin examinations, vital signs and 12-lead electrocardiograms. For the determination of the toxicity grades of clinical laboratory and vital signs evaluations, the Division of AIDS grading scale was used. Safety parameters were evaluated by means of descriptive statistics and frequency tabulations.

Results
Patient characteristics
In total, 33 HIV-1-infected men were randomized in the study (32 Caucasian and 1 Black). The median age (range) was 42 years (30–55). The underlying protease inhibitor consisted of lopinavir/ritonavir (n=29),
saquinavir/ritonavir ($n=3$) or saquinavir/lopinavir/ritonavir ($n=1$). The most commonly used regimen was the combination of lopinavir/ritonavir, tenofovir disoproxil fumarate and lamivudine, which was used by 11 patients.

**Patient disposition**

Two patients discontinued before trial completion: one patient in the washout period after session 1 due to a serious adverse event (asthma; for more information see results for safety and tolerability below) and the other patient in the washout period after session 2 due to work-related commitments. A total of 33 patients completed session 1, 32 patients completed session 2 and 27 patients entered and completed the optional third period. Full pharmacokinetic profiles of etravirine were available for all patients completing a treatment session.

**Pharmacokinetics**

All pre-dose plasma concentrations taken on day 1 were BLQ and some plasma concentrations were BLQ at other timepoints, mostly at the very beginning or near the end of the sampling. The mean ($\pm$SD) pre-dose plasma concentration was 118 ($\pm$119), 67 ($\pm$59) and 191 ($\pm$123) ng/ml on day 6 for 32 patients administering 800 mg twice daily of the Phase II formulation, 33 patients administering 100 mg twice daily of the Phase III formulation and 27 patients administering 200 mg twice daily of the Phase III formulation, respectively; on day 7 these values were 133 ($\pm$110), 75 ($\pm$64) and 218 ($\pm$178), respectively. The mean ($\pm$SD) plasma concentration-time curves for etravirine administered on day 8 as either 800 mg twice daily of the Phase II formulation or 100 mg and 200 mg twice daily of the Phase III formulation are presented in Figure 1. A slight delay in absorption is observed within the first hour of administration irrespective of formulation. The mean pharmacokinetic parameters of etravirine after single-dose (Table 1; day 1) and multiple-dose (Table 2; day 8) administration were lower with the 100 mg twice daily dose of the Phase III formulation and higher with the 200 mg twice daily dose of the Phase III formulation when compared with the mean pharmacokinetic parameters observed with the 800 mg twice daily dose of the Phase II formulation. The mean ($\pm$SD) accumulation ratio for $C_{\text{max}}$ and $\text{AUC}_{0-12\text{h}}$ for etravirine when administered as 800 mg twice daily of the Phase II formulation was 6.49 ($\pm$4.80) and 8.27 ($\pm$5.29), respectively. Similar accumulation ratios were obtained with 100 mg twice daily (4.43 $\pm$2.15 and 5.65 $\pm$2.81, respectively) and 200 mg...
twice daily (5.04 [±2.97] and 6.62 [±3.21], respectively) of the Phase II formulation.

The median $t_{\text{max}}$ was 4 h after dosing for all treatments. After a single 100 mg dose of the Phase III formulation, the C$_{\text{max}}$ and AUC$_{0-12\,\text{h}}$ were 19% and 28% lower, respectively, relative to 800 mg of the Phase II formulation. These parameters increased 97% and 91%, respectively, when comparing a single 200 mg dose of the Phase III formulation to 800 mg of the Phase II formulation. After multiple dosing on day 8, AUC$_{0-12\,\text{h}}$ for C$_{\text{max}}$ and C$_{\text{min}}$ were, respectively, 46%, 39% and 53% lower with 100 mg twice daily of the Phase III formulation than after the administration of 800 mg twice daily of the Phase II formulation. When 200 mg twice daily of the Phase III tablet was given, all three pharmacokinetic parameters were 1.67-fold higher than those obtained with the administration of 800 mg twice daily of the Phase II tablet. The C$_{\text{max}}$ and AUC$_{0-12\,\text{h}}$ parameters showed considerable interpatient variability for all treatments with the highest interpatient variability for the Phase II formulation (coefficient of variation 77% and 82%, respectively); in contrast, the coefficient of variation with the Phase III formulation was 52–59% for C$_{\text{max}}$ and 56–75% for AUC$_{0-12\,\text{h}}$.

The Phase III formulation showed a trend towards a more than dose-proportional increase in pharmacokinetic parameters when comparing 100 mg with 200 mg twice daily dosing, which was more pronounced after multiple-dose administration than after single-dose administration. Further analysis of the data showed a trend for relatively high plasma concentrations of etravirine when administered as 200 mg twice daily of the Phase III formulation in patients with plasma concentrations of etravirine <3,000 ng•h/ml after the administration of the Phase II formulation (Figure 2).

Safety and tolerability assessments
One serious adverse event was reported during the trial and that was grade 2 asthma in the washout period after the patient received 100 mg (Phase III formulation) etravirine twice daily in period 1. The patient was a 42 year old man with a medical history of asthma. He was hospitalized with an exacerbation of asthma 19 days after the first intake of study medication and discontinued the trial. This event was considered by the investigator as unlikely to be related to the study medication and the symptoms resolved within 28 days after onset. No other patients permanently or temporarily discontinued from the trial due to an adverse event. No grade 3 (severe) or 4 (potentially life threatening) adverse events were reported. In total, 28 (84.8%) patients had at least one adverse event: 36.4%, 43.8% and 33.3% of patients when receiving treatment with, respectively, etravirine 100 mg twice daily (Phase III tablet), etravirine 800 mg twice daily (Phase II tablet) and etravirine 200 mg twice daily (Phase III tablet). The overall number of patients with one or more adverse events, which was considered at least possibly related to etravirine, was 17 (51.5%). With regards to these commonly reported adverse events, no relevant treatment differences were observed except for vomiting, which was only reported during treatment with the Phase II formulation (three patients; 9.1%). There were no cases of rashes or other cutaneous events of interest in this trial.

No clinically meaningful changes over time were observed in the laboratory parameters, including urinalysis. One patient had a grade 4 laboratory abnormality for elevated triglycerides during treatment with 100 mg twice daily of the Phase III formulation, but this patient also had a grade 4 triglyceride abnormality at screening. Seven patients had treatment-emergent

### Table 2. Pharmacokinetic parameters of etravirine after multiple-dose (day 8) administration of Phase III formulation at doses of 100 and 200 mg twice daily (test) and Phase II formulation at a dose of 800 mg twice daily (reference)

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Phase II formulation</th>
<th>Phase III formulation (test A)</th>
<th>Phase III formulation (test B)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Number of patients, n</td>
<td>32</td>
<td>33</td>
<td>27</td>
</tr>
<tr>
<td>Median $t_{\text{max},\text{h}}$ (range)</td>
<td>4 (0–6)</td>
<td>4 (0–6)</td>
<td>4 (2–8)</td>
</tr>
<tr>
<td>Mean C$_{\text{max},\text{ng/ml (±sd)}}$</td>
<td>148.8 (119.3)</td>
<td>86.3 (84.5)</td>
<td>235.9 (163.1)</td>
</tr>
<tr>
<td>Mean C$_{\text{max},\text{ng/ml (±sd)}}$</td>
<td>125.8 (116.4)</td>
<td>59.9 (63.8)</td>
<td>184.7 (128.1)</td>
</tr>
<tr>
<td>Mean C$_{\text{min},\text{ng/ml (±sd)}}$</td>
<td>318.8 (245.8)</td>
<td>170.9 (99.9)</td>
<td>4,513 (232.3)</td>
</tr>
<tr>
<td>Mean AUC$_{0-12,\text{h},\text{ng•h/ml (±sd)}}$</td>
<td>2,607 (2,135)</td>
<td>1,284 (958)</td>
<td>3,713 (2,069)</td>
</tr>
<tr>
<td>LSM C$_{\text{max}}$ (90% CI)</td>
<td>0.47 (0.38–0.59)</td>
<td>1.67 (1.37–2.04)</td>
<td></td>
</tr>
<tr>
<td>LSM C$_{\text{max}}$ (90% CI)</td>
<td>0.61 (0.50–0.75)</td>
<td>1.67 (1.37–2.04)</td>
<td></td>
</tr>
<tr>
<td>LSM AUC$_{0-12,\text{h}}$ (90% CI)</td>
<td>0.54 (0.44–0.65)</td>
<td>1.67 (1.38–2.02)</td>
<td></td>
</tr>
</tbody>
</table>
grade 3 laboratory abnormalities; these were mostly related to elevated total cholesterol (three patients) and triglycerides (five patients). There were no consistent or clinically relevant changes over time in median vital sign or electrocardiogram parameters.

Discussion

The selected dose of etravirine based on Phase IIb trials was 800 mg twice daily using a granulolayered tablet formulation. As a result of an intensive search for formulations with improved bioavailability, a spray-dried formulation was developed with reduced pill burden. On the basis of the results of previously conducted single-dose bioavailability trials in healthy volunteers and HIV-1-infected patients, this trial was designed to assess the pharmacokinetics of 100 mg twice daily of the spray-dried formulation and to compare this with the formulation used during Phase II trials. However, exposures to etravirine were lower than expected when it was dosed at 100 mg twice daily. After multiple dosing on day 8, the ratios of the LSM (90% CI) for AUC_0–12 h, C_max and C_min for the comparison of the 100 mg twice daily dose of the Phase III formulation versus the 800 mg twice daily dose of the reference Phase II formulation (that is, 1.67 [1.38–2.02], 1.67 [1.37–2.04] and 1.67 [1.37–2.04], respectively). Given the observed interpatient variability with the Phase II formulation and the non-proportional increase in exposures between the 100 mg and 200 mg twice daily doses of Phase III formulation, an exploratory analysis was performed to investigate whether this increase was homogenous across individual patients. For each individual, the relationship between individual steady-state AUC_0–12 h (day 8) administered at a dose of 800 mg twice daily as Phase II tablets and at a dose of 200 mg twice daily as Phase III tablets were portrayed graphically. This analysis indicated that the 200 mg twice daily dose of the Phase III formulation did not appear to increase the absolute exposures for those patients who achieved higher exposures with the 800 mg twice daily dose of Phase II formulation. Rather, for patients with lower exposures (<3,000 ng•h/ml) with the 800 mg twice daily dose of the Phase II formulation, treatment with 200 mg twice daily of the Phase III formulation substantially increased the absolute exposures. Consequently, there was a lower interpatient variability with the 200 mg twice daily dose. The mechanism behind this effect is likely related to the improved bioavailability of the spray-dried formulation.

These results show that the Phase III formulation at a dose of 200 mg twice daily provides a better pharmacokinetic profile compared with 800 mg twice daily of the Phase II formulation, because the plasma concentrations are in the same range with a reduced interindividual variability. On the basis of these observations, a dosage of etravirine 200 mg twice daily was selected as the dose for the Phase III trial and for all other subsequent trials with etravirine.

Recently, 24-week results from two large, randomized, placebo-controlled trials involving etravirine were published. In treatment-experienced patients with NNRTI resistance, patients randomized to receive etravirine 200 mg twice daily with an optimized background regimen achieved significantly better virological suppression at week 24 than patients receiving an optimized background regimen and placebo. These studies also confirmed that etravirine was generally safe and well tolerated [17,18].

No clinically relevant safety concerns were raised during this trial and there were no clinically relevant findings for laboratory and cardiovascular parameters. No
difference in severity or frequency of safety parameters was observed between the three treatments.

The range of exposures observed after the administration of 200 mg of the Phase III formulation was within the range of exposures observed after administration of 800 mg twice daily of the Phase II formulation, but overall, the 200 mg tablets of the Phase III formulation produced higher exposures in terms of Cmax, Cmin and AUC0–12 h. Etravirine was generally safe and well tolerated in all sessions of this study.

Acknowledgements

This study was financially supported by Tibotec. The authors are grateful to Iris Weimer for medical writing support. The study protocol was reviewed and approved by the appropriate Institutional Review Board (IRB) and health authorities and was conducted in accordance with the Declaration of Helsinki. Written informed consent was obtained from all patients.

Disclosure statement

TNK, MS-G, GDS, GB, MP, KV, BJW and RMWH are employees of Tibotec. The other authors declare no conflicts of interest.

References


