CENTER FOR DRUG EVALUATION AND RESEARCH

APPLICATION NUMBER:

208610Orig1s000
208611Orig1s000

CLINICAL PHARMACOLOGY AND BIOPHARMACEUTICS REVIEW(S)

Office of Clinical Pharmacology Review

NDA Number	208610 (Oral Tablet) & 208611 (IV Injection)
Link to EDR	\\CDSESUB1\evsprod\NDA208610\0001 \\CDSESUB1\evsprod\NDA208611\0001
Submission Date	10/19/2016
Submission Type	New Molecular Entity; Priority
Brand Name	Baxdela
Generic Name	Delafloxacin
Dosage Form and Strength	• Injection: 300 mg of lyophilized powder in a single-use vial for reconstitution for intravenous infusion. • Oral tablets: 450 mg
Route of Administration	Oral or intravenous infusion
Proposed Indication	For the treatment of acute bacterial skin and skin structure infections (ABSSSI) caused by designated susceptible bacteria in adults
Applicant	Melinta Therapeutics Inc.
Associated IND	IND 62,772 (oral) IND 76,096 (IV)
OCP Review Team	Kunyi Wu, Pharm.D. Zhixia Yan, Ph.D. Luning Zhuang, Ph.D. Jeffry Florian, Ph.D. Anuradha Ramamoorthy, Ph.D. Christian Grimstein, Ph.D.
OCP Final Signatory	John Lazor, Pharm.D. / Office of Clinical Pharmacology, Division IV, Director

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1. EXECUTIVE SUMMARY

Delafloxacin is an anionic fluoroquinolone antibiotic proposed for the treatment of acute bacterial skin and skin structure infections (ABSSSI) caused by susceptible isolates of Gram-positive organisms: *Staphylococcus aureus* (including methicillin-resistant [MRSA] and methicillin-susceptible [MSSA] isolates), *Staphylococcus haemolyticus*, (b) (4) *Staphylococcus lugdunensis*, *Streptococcus agalactiae*, *Streptococcus anginosus* Group, (b) (4) *Streptococcus pyogenes*, and *Enterococcus faecalis*, and by the following Gram-negative organisms: *Escherichia coli*, *Enterobacter cloacae*, (b) (4), *Klebsiella pneumoniae*, (b) (4), and *Pseudomonas aeruginosa*. Delafloxacin can be administered by either an intravenous (IV) infusion route or orally as a tablet.

The key clinical pharmacology review questions focus on appropriateness of oral only dosing regimen and dose recommendation for delafloxacin in patients with renal impairment.

1.1 Recommendations

The Office of Clinical Pharmacology Divisions of Clinical Pharmacology IV, Pharmacometrics, and Genomics Group have reviewed the information contained in NDAs 208610 and 208611. The application is approvable from a clinical pharmacology perspective, provided that an agreement is reached between Applicant and the Agency on the dosing regimen and labeling (Table 1.1-1).

Table 1.1-1 Summary of OCP's Recommendations & Comments on Key Review Issues

Review Issue	Recommendations and Comments
Pivotal or Supportive evidence of effectiveness	The primary evidence of effectiveness for delafloxacin in patients with acute bacterial skin and skin structure infections (ABSSSI) comes from two Phase 3 trials (RX-3341-302 and RX-3341-303). Supportive evidence of effectiveness was provided by two Phase 2 trials, the Phase 1 pharmacokinetics (PK) studies demonstrating delafloxacin PK comparability between IV and oral formulations, and the exposure-response efficacy analysis.
General dosing instructions	The recommended dosage regimen is 300 mg every 12 hours (Q12h) administered by IV infusion over 60 minutes or 450 mg Q12h orally (with or without food) at the discretion of the physician for a total duration of 5 to 14 days.

<table>
<tr><td>Dosing in patient subgroups (intrinsic and extrinsic factors)</td><td>Due to accumulation of delafloxacin, dose adjustment for the IV formulation is recommended in patients with severe renal impairment (eGFR[a] 15-29 mL/min/1.73m^2).

Furthermore, due to accumulation of sulfobutylether-β-cyclodextrin (SBECD; intravenous vehicle), the lack of observed data in ESRD patients receiving oral delafloxacin, and the deficiency of the design of the renal impairment study, the use of delafloxacin (IV and oral) is not recommended in patients with ESRD. In addition, if the IV formulation is used in patients with severe renal impairment, routine monitoring of serum creatinine should be performed with consideration to changing to oral delafloxacin if increases in serum creatinine are observed.

<table>
<tr><th></th><th>FDA recommendation (Difference highlighted in Bold)</th><th>Applicant's proposal</th></tr>
<tr><td>Patients with Severe Renal Impairment (eGFR[a] 15-29 mL/min/1.73m^2)</td><td><u>IV</u>: 200 mg Q12h

<u>Oral</u>: 450 mg Q12h</td><td><u>IV</u>: 200 mg Q12h

Oral: (b) (4)</td></tr>
<tr><td>Patients with End Stage Renal Disease (ESRD) (eGFR< 15 mL/min/1.73m^2)</td><td><u>IV and Oral</u>: NOT RECOMMENDED</td><td>IV: (b) (4)
Oral: (b) (4)</td></tr>
</table>
</td></tr>
<tr><td>Labeling</td><td>Generally acceptable. The review team has specific content and formatting change recommendations.</td></tr>
</table>

[a] eGFR: estimate of GFR based on MDRD equation

1.2 Post-Marketing Requirements and Commitments

None.

2. SUMMARY OF CLINICAL PHARMACOLOGY ASSESSMENT

2.1 Pharmacology and Clinical Pharmacokinetics

Delafloxacin is an anionic fluoroquinolone antibiotic. Like other quinolone antibiotics, delafloxacin exerts its antibacterial activity by inhibiting DNA synthesis.

The following is a summary of the clinical pharmacokinetics of delafloxacin.

Absorption:
The absolute bioavailability for delafloxacin 450 mg oral tablet administered as a single dose was 58.8%. Delafloxacin exposure (AUC) following administration of a single 450 mg oral (tablet) dose was comparable to that following a single 300 mg IV dose. Peak plasma concentrations were achieved within about 1 hour after oral administration under a fasted condition. Total exposure (AUC) of delafloxacin is unchanged under fasted or fed conditions.

Distribution:
The plasma protein binding of delafloxacin ranged from 80% for the severe renal impairment group to 84% for the healthy group, while the ESRD group showed the lowest percentage of bound delafloxacin (75%). The steady state volume of distribution of delafloxacin was about 30-48 L.

Elimination:
The estimated mean half-life for delafloxacin was 3.7 hours and 4.2 to 8.5 hours following IV and oral administration of delafloxacin, respectively. Following administration of a single 300 mg IV dose of delafloxacin, the mean clearance (CL) of delafloxacin was 16.3 L/h, and the renal clearance (CLr) of delafloxacin accounts for 35-45% of the total clearance.

Metabolism:
Glucuronidation of delafloxacin is the primary metabolic pathway with oxidative metabolism representing about 1% of an administered dose. Unchanged parent drug is the predominant component in plasma. There are no significant circulating metabolites in humans.

Excretion:
After single intravenous dose of ^{14}C-labeled delafloxacin, 65% of the radioactivity is excreted in urine as unchanged delafloxacin and glucuronide metabolites and 28% is excreted in feces as unchanged delafloxacin. Following a single oral dose of ^{14}C-labeled delafloxacin, 50% of the radioactivity is excreted in urine as unchanged delafloxacin and glucuronide metabolites and 48% is excreted in feces as unchanged delafloxacin.

2.2 Dosing and Therapeutic Individualization

2.2.1 General dosing

Applicant's proposed dosage regimen for ABSSSI patients as shown below is supported by the PK, efficacy, and safety data from the clinical trials submitted in the application.

300 mg every 12 hours (Q12h) administered by IV infusion over 60 minutes or 450 mg Q12h orally (with or without food) for a total duration of 5 to 14 days at the discretion of the physician.

2.2.2 Therapeutic individualization

Renal Impairment

Applicant identified renal impairment status as the only intrinsic factor warranting dose adjustment. Specifically, for adult ABSSSI patients with severe renal impairment [redacted] Applicant proposes to decrease the dose to 200 mg IV Q12h [redacted] However, these dosing regimens have not been evaluated in clinical trials.

In a dedicated PK study in subjects with renal impairment, the AUC values for the mild, moderate, and severe renal impairment groups were 1.3, 1.6, and 1.8 fold higher, respectively, than the corresponding exposure in subjects with normal renal function after receiving a single 300 mg IV dose. Likewise, in ESRD subjects requiring dialysis as part of their clinical management, when on- and off-dialysis the AUC values were 2.1 and 2.6 fold higher, respectively, than the corresponding exposure in subjects with normal renal function after receiving a single 300 mg IV dose. The AUC values for the mild, moderate, and severe renal impairment groups were 1.1, 1.5, and 1.6 fold higher, respectively, than the corresponding exposure in subjects with normal renal function after receiving a single 400 mg oral dose.

The safety assessment for patients (N=37) with moderate renal impairment receiving the Applicant's proposed dose indicated that no additional Treatment-Emergent-Adverse-Events (TEAEs) were observed in this group compared to other patients in Phase 3 Safety analysis dataset. Therefore, it is acceptable to maintain the same dosing regimen for patients with mild and moderate renal impairment as the one for patients with normal renal function.

We agree with Applicant 's proposal of reducing IV dose to 200 mg Q12h in patients with severe renal impairment because it provides comparable delafloxacin exposure (AUC) to normal subjects based on simulation. However, we do not agree with Applicant's proposal [redacted] We recommend 450 mg Q12h oral dose in patients with severe renal impairment. Although simulation showed that the 450 mg Q12h oral dose achieved 48% higher mean AUC of delafloxacin compared to patients with normal renal function, the delafloxacin exposure at the recommended 450 mg Q12h oral dose is lower than that in patients receiving the 450 mg Q12h IV dose in one Phase 2 study (Study RX-3341-201) where no major safety concerns were identified with 450 mg Q12h IV.

We do not recommend the use of oral delafloxacin in patients with ESRD due to the unknown impact of ESRD on delafloxacin disposition following oral dosing as no observed data are

available in patients with ESRD receiving oral delafloxacin. In addition, the review team identified a deficiency in the design of the renal impairment study where administration of delafloxacin within one hour after completion of a hemodialysis session may not adequately describe delafloxacin exposure in subjects on an off-dialysis day.

The delafloxacin IV formulation contains sulfobutyl-ether-β-cyclodextrin (SBECD, Captisol®) as an intravenous vehicle, which is known to accumulate in patients with renal impairment and may cause pose a risk of nephrotoxicity. In the dedicated renal impairment study, SBECD was found to accumulate extensively in subjects with severe renal impairment and ESRD subjects receiving hemodialysis, suggesting potential safety concerns with the use of IV delafloxacin in these patients. There are several FDA approved IV drug products that contain SBECD, such as CARNEXIV™ (carbamazepine) injection and VFEND® (voriconazole) for injection. Both CARNEXIV and VFEND labels recommend to avoid using the IV formulation in patients with moderate and severe renal impairment and closely monitor renal function in these patients if IV formulation is used. Of note, both CARNEXIV and VFEND for injection products contain higher amounts of SBECD per dose than IV delafloxacin.

Based on the totality of data presented in this application together with the SBECD information from literature and FDA drug labeling, the review team recommends that for patients with severe renal impairment the IV dose should be reduced to 200 mg Q12h and the oral dose should remain unchanged (450 mg Q12h). If the IV formulation is used in patients with severe renal impairment, routine monitoring of serum creatinine should be performed with consideration to changing to oral delafloxacin if increases in serum creatinine are observed; for ESRD patients, the use of delafloxacin (IV and oral) is not recommended.

2.3 Outstanding Issues

Delafloxacin is shown to be a substrate of P-glycoprotein (P-gp) and breast cancer resistance protein (BCRP) in vitro. Applicant did not conduct a clinical drug-drug interaction (DDI) study to evaluate the in vivo DDI potential between delafloxacin and P-gp and/or BCRP inhibitors. The Clinical Pharmacology Review Team reviewed the rationale and clinical data provided by Applicant and concluded that the provided rationale does not rule out the possibility that the AUC of delafloxacin in patients coadministered IV or oral delafloxacin with P-gp and BCRP inhibitors may be greater than the exposure at the 450 mg Q12h IV dose shown to be tolerable in Study RX-3341-201. In addition, the in vivo DDI potential between delafloxacin and P-gp and/or BCRP inhibitors was not adequately addressed by the limited Phase 3 data. Therefore, the clinical relevance of coadministration of delafloxacin and P-gp and/or BCRP inhibitors is unknown (See Section 3.3.4).

2.4 Summary of Labeling Recommendations

The Office of Clinical Pharmacology recommends the following labeling concepts be included in the final package insert (Table 2.4-1).

Table 2.4-1 Summary of Labeling Issue Identification and Recommendations

Section/heading	Acceptable to OCP?			Comment
	A	AWE	NA	
Highlights/Section 2.1 Recommended Dosage Regimen	☐	☐	☒	• Revise the IV and/or oral dosing regimen in patients with severe renal impairment and ESRD
Section 5 / Warnings and Precautions	☐	☐	☒	Delete Section (b) (4) according to the FDA Guidance of Labeling for Human Prescription
Section 12.2 / Cardiac Electrophysiology	☐	☒	☐	Remove the description of (b) (4)
12.3 (b) (4)	☐	☒		(b) (4)
12.3/Specific Population	☐	☐	☒	• Revise classification of renal function based on actual eGFR cutoff values used in the dedicated renal impairment Study (RX-3341-110) • Revise study results based on Reviewer's independent analyses (see 3.3.3 for details)
12.3/DDI	☐	☒	☐	• Revise in vitro condition pertaining to delafloxacin as an inducer of CYP2C9 and CYP3A4 • Add the information regarding delafloxacin as a substrate of P-gp and BCRP based on results from in vitro studies

A = Acceptable; AWE=Acceptable with minor edits; NA=not acceptable/substantive disagreement (must provide comment)

3. COMPREHENSIVE CLINICAL PHARMACOLOGY REVIEW

3.1 Overview of the Product and Regulatory Background

Delafloxacin is a anionic fluoroquinolone anti-bacterial drug and has been developed as an oral tablet and an IV injection formulation. The indication being sought for delafloxacin is the treatment of ABSSSI caused by designated susceptible bacteria in adults. Two INDs are in effect in the Division of Anti-infective Products (DAIP) including INDs 62772 (oral tablet) established on 6/22/2001 and 76096 (IV solution) established on 3/20/2007. Qualified Infections Disease

Product (QIDP) and Fast Track status were granted to delafloxacin for development of ABSSSI indication on 09/08/2012 and 12/18/2012, respectively.

Applicant requested a full product-specific waiver for both IV and oral dosage forms from generating clinical data in pediatric patients aged 0 to <18 years old with ABSSSI in their iPSP submitted to INDs 062772 and 076096. The request for a full waiver is due to safety (i.e. quinolone products as a class have a known safety concern related to cartilage/tendon rupture). The FDA granted the full pediatric waiver on 03/07/2016.

Multiple milestone meetings have been held between the Division and Applicant throughout the development process, including the End of Phase 2 meeting (04/10/2010) and the Pre-NDA meeting (2/29/2016).

3.2 General Pharmacology and Pharmacokinetic Characteristics

Pharmacology	
Mechanism of Action	Delafloxacin, like other members of the fluoroquinolone class, inhibits DNA synthesis by targeting bacterial DNA gyrase and topoisomerase IV. Delafloxacin has a bulky shape, does not have basic properties, and is anionic in nature. The collaboration between a large N1 substitution and a weakly polar group at C8 results in increased potency against quinolone-resistant Gram-positive bacteria, a phenotype common among MRSA. The basicity at C7 leads to increased potency at acidic pH.
Active Moieties	Delafloxacin
QT Prolongation	Delafloxacin neither blocked hERG currents nor prolonged QT in animals at concentrations within the exposure range reached with the maximal tolerated dose in humans. Delafloxacin demonstrated no significant QTc prolongation effect after a single-dose IV (300 mg, 900 mg) administration in a thorough TQTc study with a moxifloxacin positive control (Study RX-3341-111). Although the QTc interval prolongation potential of 450 mg oral Q12h dosage has not been evaluated in a thorough TQTc study, no prolongation would be expected based on a similar metabolic profile after IV and oral administration and comparison of the peak plasma concentration from the IV TQTc study (900 mg supratherapeutic dose) and the C_{max} obtained after a 450-mg oral Q12h dosage of delafloxacin.
General Information	

<table>
<tr><td>Bioanalysis</td><td>

Validated LC/MS/MS methods to determine delafloxacin concentrations in human plasma and urine (See APPENDIX 4.1)
Validated LC-API/MS/MS methods to determine delafloxacin concentrations in human plasma, urine, and dialysate fluid (See APPENDIX 4.1)
Validated LC/MS/MS methods to determine sulfobutyl-ether-β-cyclodextrin (SBECD) concentrations in human plasma, urine, and dialysate fluid (See APPENDIX 4.1)

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<tr><td>Healthy vs. Patients</td><td>
In this application, PK samples were collected in both healthy volunteers and patients for the IV formulation. However, PK samples were only collected from healthy volunteers for the oral formulation.

For the IV formulation, under the same dosing regimen, the overall exposure (AUC) of delafloxacin was comparable between healthy volunteers (i.e. observed data from Phase 1 studies) and patients with ABSSSI (i.e., simulated data based on a population PK model).
For the oral formulation, no PK comparison is available between healthy volunteers and patients.

</td></tr>
<tr><td>Drug exposure at steady state following the therapeutic dosing regimen</td><td>
<table>
<tr><td></td><td>Oral tablet
(Study ML-3341-118)
450 mg Q12h, Day 7</td><td>IV injection
(Study RX-3341-104)
300 mg Q12h, Day 14</td></tr>
<tr><td></td><td>Mean (SD) N=22</td><td>Mean (SD) N=7</td></tr>
<tr><td>$AUC_{0\text{-}12h}$ (μg.h/mL)</td><td>30.8 (11.4)</td><td>23.4 (6.9)</td></tr>
<tr><td>C_{max} (μg/mL)</td><td>7.45 (3.16)</td><td>9.29 (1.83)</td></tr>
</table>
</td></tr>
<tr><td>Range of effective dose or exposure</td><td>

300 mg IV Q12h for 5 to 14 days (Studies RX-3341-302,RX3341-202, RX-3341-201)
300 mg IV Q12h for 3 days (6 doses), then 450 mg oral tablet Q12h for a total duration of treatment of 5 to 14 days (Study RX-3341-303)
450 mg IV Q12h for 5 to 14 days (Study RX-3341-201)

</td></tr>
<tr><td>Maximally tolerated dose (MTD) or exposure</td><td>

A single dose of 900 mg is the MTD for IV formulation (Study RX-3341-108). The highest IV multiple-dose evaluated in Phase 1 and 2 trials is 450 mg BID for 5-14 days (Studies ML-3341-118 and RX-3341-201).
No MTD was determined for oral formulation, although more GI side effects were observed at single dose > 1200 mg and multiple doses of 800 mg and 1200 mg Q24h for 5 days (Study M00-224).

</td></tr>
<tr><td>Dose Proportionality</td><td>

For IV administration, delafloxacin AUC increased

</td></tr>
</table>

	proportionally to dose over the dose range of 50 mg to 200 mg, but increased slightly more than dose proportionally at single doses ranging from 300 mg (24.7 µg·h/mL) to 1200 mg (156 µg·h/mL) (Study RX-3341-101 and Study RX-3341-108). • Delafloxacin total clearance was comparable from Day 1, Day 10, and Day 14 after IV doses ranging from 150 mg to 450 mg (Study RX-3341-102, Study RX-3341-103). • For Oral administration, the increase in delafloxacin AUC was approximately dose proportional at single doses ranging from 200 mg to 1600 mg for Phase 1 capsule (M00-224). The increase in delafloxacin AUC was approximately dose proportional at single doses ranging from 450 mg to 900 mg for the to-be-marketed tablet (Study RX-3341-115, Study RX-3341-116).
Accumulation	• The accumulation ratio for the dosing regimen of 450 mg tablet Q12h was 1.36 (Study ML-3341-118); • The accumulation ratio for the dosing regimen of 300 mg IV Q12h was 1.09 (ranged:1.01 to 1.14) (Studies RX-3341-102, RX-3341-103, RX-3341-104).
Variability	• For oral administration, the inter-subject variability (%CV) in C_{max} and AUC_t values for the 450-mg to-be-marketed tablet formulation in healthy subjects ranged from 24% to 42% and 27% to 37%, respectively (Studies RX-3341-115, 116, 118). • For IV administration, the inter-subject variability (%CV) in C_{max} and AUC_t values for the IV to-be-marketed formulation administered to healthy subjects ranged from 9% to 25% and 9% to 28%, respectively (Studies RX-3341-108, 110, 111, 112, 113).
Bioavailability	• The absolute bioavailability of the to-be-marketed 450 mg tablet was estimated to be 58.8% by comparing the AUC of delafloxacin following administration of a 450 mg oral tablet with the AUC of delafloxacin following administration of a single 300 mg IV dose corrected by the ratio of the 300 and 450 mg dose (Study RX-3341-115). • Similar to the bioavailability results from Study RX-3341-115, the absolute bioavailability of the 450 mg oral tablet was estimated to be 56.5% by comparing the AUC of delafloxacin following administration of a 450 mg oral tablet (Study RX-3341-115) with the AUC of delafloxacin following administration of a single 450 mg IV dose (Study RX-3341-108).
T_{max}	~ 1h
Food effect	**$AUC_{0-\infty}$** | **C_{max}** | **T_{max}**

(Fed/fasted) Geometric Mean % [90% CI]	104.996 (92.082, 119.720)	79.504 (73.126, 86.439)	Fasted: T_{max} (median) = 1.25; Fed: T_{max} (median) = 2.5;
	The total exposure ($AUC_{0-\infty}$ and AUC_{0-t}) of delafloxacin oral tablet was not affected by administration under fed condition (Study RX-3341-116). Since the PK/PD index for delafloxacin associated with antibacterial activity in animal models was AUC_{0-24}/MIC, a 20% lower C_{max} under fed condition is not expected to cause any clinically meaningful outcomes.		
Volume of Distribution	After a single IV dose, the volume of distribution of delafloxacin ranged from 30 to 48 L across clinical studies (i.e. Studies RX-3341-101, RX-3341-108, RX-3341-102, RX-3341-103, RX-3341-104, RX3341-110, RX-3341-111, ML-3341-112, RX-3341-113).		
Plasma Protein Binding	• At delafloxacin concentrations ranging from 0.1 to 50 µg/mL, the protein binding, primarily to albumin, is approximately 84%, as determined by the equilibrium-dialysis method (Study R&D/00/427 and Study R&D/00/428). • In a study in subjects with renal impairment (RX-3341-110), plasma protein binding of delafloxacin ranged from 80% for the severe renal impairment group to 84% for the healthy group, while the ESRD group showed the lowest percentage of bound delafloxacin (75%).		
Substrate transporter systems [*in vitro*]	• Delafloxacin is not a substrate of OAT1, OAT3, OCT1, OCT2, OATP1B1 or OATP1B3 according to the in vitro study results (Studies (b) (4) -2013-073, (b) (4) -2011-056). • Delafloxacin was shown to be a substrate of P-gp and BCRP in vitro (Study (b) (4) 148044). The clinical relevance of coadministration of delafloxacin and P-gp and/or BCRP inhibitors is unknown.		
Elimination			
Half-life	• In a mass balance study, the mean half-life for delafloxacin was 3.7 hours after a single dose IV administration (Study RX-3341-107). • The mean half-life values for delafloxacin ranged from 4.2 to 8.5 hours following multiple oral administration over the dose range of 100 mg QD to 450 mg BID (Study M00-224). • The reported half-lives of delafloxacin are consistent with the observation of the accumulation ratio of 1.09 and 1.36 following multiple-dose administration of IV and oral formulations, respectively, and the time to reach steady state by Day 3.		
Metabolism			

Fraction metabolized (% dose)	• No active metabolites have been identified. • After oral administration of [^{14}C]-delafloxacin (200 mg), three glucuronide conjugates M3, M5 and M6 representing 13.13%, 2.05%, 1.51%, and one oxidative metabolite (M7) representing 1.01% of administered dose were identified in urine; about 32.5% of the total delafloxacin dose was found as unchaged parent drug in urine and 48% as unchanged parent drug in feces (Study M01-292). • Following a single 300-mg IV dose of [^{14}C]-delafloxacin, about 20% of the dose was excreted as glucuronide metabolites in urine; about 45% of the total delafloxacin dose was found as unchanged parent drug in urine and 28% as unchanged parent drug in feces (Study RX-3341-107). • The metabolite profiling of plasma after an IV single dose showed that delafloxacin accounted for the majority of the radioactivity (87.1% to 91.7%) with delafloxacin glucuronide accounting for 8.0 to 11.0% (mean of 9.6%) of the radioactivity.
Primary metabolic pathway(s) [*in vitro*]	• Glucuronidation is the primary metabolic pathway for delafloxacin with oxidative metabolism representing about 1% of an administered dose. • The glucuronidation of delafloxacin is mediated mainly by UGT1A1, UGT1A3, and UGT2B15 (Study (b) (4) 134028). • Applicant performed an optional pharmacognetic sub-study using the data collected from two Phase 1 studies, M00-224 and M01-301. Applicant evaluated the effect of genetic polymorphisms in UGT1A1, UGT2B4, UGT2B15, and ABCB1 on the PK (e.g., C_{max}, C_{min}, AUC, $t_{1/2}$, CL/F, Vβ/F) of delafloxacin.The C_{min} data from Study M00-224 seems significantly correlated with UGT2B15*2 genotype. Due to the small sample size (N=27), Applicant concluded that further confirmation is required to elucidate the role of UGT2B15. However, no additional data are available from any of the subsequent Phase 2 or 3 studies to draw any conclusions on the impact of genetic polymorphisms on the PK of delafloxacin (see APPENDIX 4.5.18 Summary of Pharmacogenomics for Delafloxacin for details) .
Excretion	
Primary excretion pathways (% dose) ±SD	• After oral administration of [^{14}C]-delafloxacin (200 mg), a total 7-day recovery of radioactivity averaged 97.83% (range of 87.96%-100.61%). A mean of 50.19% (range of 44.43- 55.74%) of the dose was recovered in urine, and 47.65% (range of 39.78%-55.71%) was eliminated in feces. • Following a single 300-mg IV dose of [^{14}C]-delafloxacin, a mean of 92.85% of administered radioactivity was recovered from subjects with 64.50% in urine and 28.35% in feces.

In vitro interaction liability (Drug as perpetrator)	
Inhibition/Induction of metabolism	• Delafloxacin at clinically relevant concentrations does not inhibit the cytochrome P450 isoforms CYP1A2, CYP2A6, CYP2B6, CYP2C8, CYP2C9, CYP2C19, CYP2D6, CYP2E1 and CYP3A4/5 in vitro in human liver microsomes (Studies MWQ3034-10, R&D/01/136, and 418N-1301). • In human hepatocytes, delafloxacin showed no potential for in vitro induction of CYP1A2, 2B6, 2C19, or 2C8 (Studies (b) (4) 133033 and (b) (4) 142117), • Delafloxacin was a mild inducer (less than 2 fold) of CYP2C9 at a concentration of 100 µM and CYP3A4 at a clinically relevant concentration (Studies (b) (4) 133033 and (b) (4) 142117). • An in vivo study was conducted for the potential DDI with CYP3A4.The study results demonstrated that delafloxacin did not significantly affect the C_{max} and $AUC_{0\text{-}inf}$ of midazolam or 1-hydroxymidazolam when delafloxacin and midazolam were coadministered compared to administration of delafloxacin alone (Study ML-334-118). This result indicated that delafloxacin is not an inducer of CYP3A4 at clinically relevant concentration.
Inhibition/Induction of transporter systems	• Delafloxacin was not an inhibitor of the following transporters at clinical relevant concentrations: P-gp, MDR1, BCRP, OAT1, OAT3, OATP1B1, OATP1B3, BSEP, OCT1, or OCT2 (Studies (b) (4) -2011-055, (b) (4) -2013-072, and (b) (4) -2013-102). • Delafloxacin is not likely to inhibit UGT1A1 or UGT2B7 at clinically relevant concentrations (Study (b) (4) 134028).

a= Pharmacokinetic parameters are presented as mean ±standard deviation (SD) or median (minimum, maximum) unless otherwise noted; b= Approximately 150, 250, and 500 calories from protein, carbohydrate, and fat, respectively.

3.3 Clinical Pharmacology Review Questions

3.3.1 To what extent does the available clinical pharmacology information provide supportive evidence of effectiveness?

The primary evidence of efficacy of delafloxacin in the treatment of ABSSSI was provided by two pivotal Phase 3 studies (RX-3341-302 [IV only] and RX-3341-303 [IV to oral]).

Supportive evidence of effectiveness was provided by two supportive Phase 2 studies (RX-3341-201 [IV only] and RX-3341-202 [IV only]) along with Phase 1 pharmacokinetics (PK) studies demonstrating delafloxacin PK comparability between IV and oral formulations (see 3.3.2 for details), and an exposure-response relationship for efficacy.

The study design of the two pivotal Phase 3 trials and two supportive Phase 2 trials is summarized in Table 3.3.1-1.

Table 3.3.1-1 Summary of Study Designs for Key Studies in Support of ABSSSI Indication

Study No.	Design	Defloxacin Dosage Regimen[a]	Comparator Dosage Regimen[a]	Population Size
Phase 3				
RX-3341-302	Multicenter, randomized, double-blind, comparative study (ABSSSI)	300 mg IV Q12h	Vancomycin (15 mg/kg Q12h) and aztreonam (2 g Q12h)	Delafloxacin: N=331 Comparator: N=329
RX-3341-303		300 mg IV Q12h for 6 doses with switch to 450 mg oral Q12h		Delafloxacin: N=423 Comparator: N=427
Phase 2				
RX-3341-201	Multicenter, randomized, double-blind, comparative study (cSSSI)	300 mg Q12h (IV) 450 mg Q12h (IV)	Tigecycline 100 mg initially, followed by 50 mg Q12h	Delafloxacin: 300 mg N=49 450 mg N=51 Comparator: N=50
RX-3341-202	Multicenter, randomized, double-blind, comparative study (ABSSSI)	300 mg IV Q12h	Linezolid: 600 mg Q12h Vancomycin (15 mg/kg Q12h) With optional aztreonam (2 g Q12h)	Delafloxacin: N=81 Comparator: Linezolid N=77 Vancomycin N=98

a. Treatment Duration: 5-14 days

The primary efficacy endpoint used in Phase 3 studies was objective response, defined as ≥20% reduction in lesion area compared to baseline at 48 to 72 hours after initiation of treatment. Table 3.3.1-2 summarizes the objective response at 48 to 72 hours (primary endpoint) in two pivotal Phase 3 studies. Delafloxacin was noninferior to vancomycin + aztreonam (active comparator) in objective response at 48 to 72 hours after initiation of study drug in the 2 pivotal studies regardless of the analysis population.

Table 3.3.1-2 Objective Responder at 48 to 72 Hours – Study RX-3341-302, Study RX-3341-303, and Primary Analysis Pool 1 (Adapted from Table 18 in the Summary of Clinical Efficacy)

	RX-3341-302		RX-3341-303		Pool 1	
	Delafloxacin	Vancomycin (Aztreonam)	Delafloxacin)	Vancomycin (Aztreonam)	Delafloxacin	Vancomycin (Aztreonam)
ITT Analysis Set	**N = 331**	**N = 329**	**N = 423**	**N = 427**	**N = 754**	**N = 756**
Responder, n/N (%)	259 (78.2)	266 (80.9)	354 (83.7)	344 (80.6)	613 (81.3)	610 (80.7)
Difference (95% CI)	−2.6 (−8.8, 3.6)		3.1 (−2.0, 8.3)		0.8 (−3.2, 4.7)	
MITT Analysis Set	**N = 243**	**N = 247**	**N = 275**	**N = 277**	**N = 518**	**N = 524**
Responder, n/N (%)	197 (81.1)	207 (83.8)	241 (87.6)	228 (82.3)	438 (84.6)	435 (83.0)
Difference (95% CI)	−2.7 (−9.5, 4.0)		5.3 (−0.7, 11.4)		1.8 (−2.7, 6.3)	
CE72O Analysis Set	**N = 294**	**N = 297**	**N = 395**	**N = 387**	**N = 689**	**N = 684**
Responder, n/N (%)	250 (85.0)	257 (86.5)	346 (87.6)	327 (84.5)	596 (86.5)	584 (85.4)
Difference (95% CI)	−1.5 (−7.2, 4.2)		3.1 (−1.8, 8.0)		1.2 (−2.6, 4.9)	
ME72O Analysis Set	**N = 220**	**N = 225**	**N = 264**	**N = 250**	**N = 484**	**N = 475**
Responder, n/N (%)	190 (86.4)	199 (88.4)	235 (89.0)	216 (86.4)	425 (87.8)	415 (87.4)
Difference (95% CI)	−2.1 (−8.4, 4.2)		2.6 (−3.1, 8.4)		0.5 (−3.8, 4.7)	

Note: Results are based on data closest to and through 72 hours within a window of 46-74 hours which includes the +/− 2 hour window for each visit. A patient may have more than one reason for nonresponse and is counted once for each reason.

Difference = Difference in responder rates (Delafloxacin treatment group minus vancomycin + aztreonam treatment group).
Confidence intervals are calculated using Miettinen and Nurminen method without stratification for individual studies and stratified by studies for Pool 1 analysis.

*ITT: Intent to treat
*MITT: Microbiological intent to treat
*CE72O: clinically evaluable at 48 to 72 hours (± 2 hours) for the objective response
*ME72O: microbiologically evaluable at 48 to 72 hours (± 2 hours) for the objective response

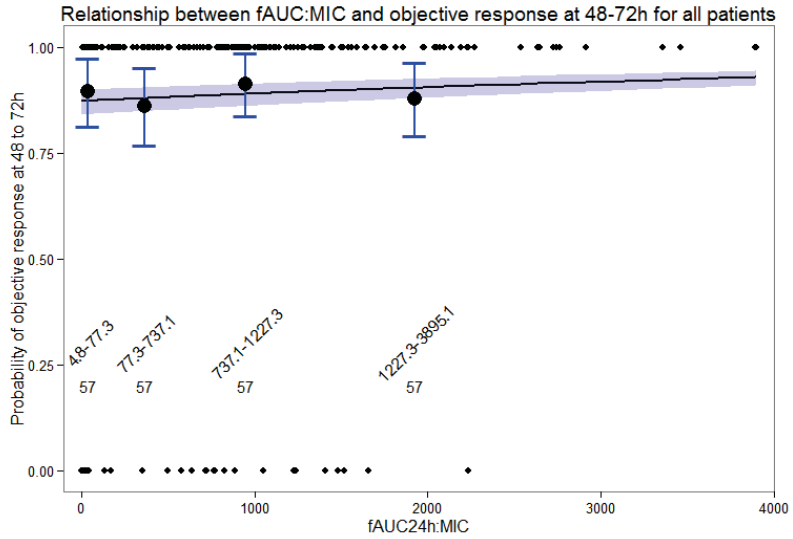
A population exposure-response analysis was conducted using data from delafloxacin treated patients in one Phase 2 study (RX-3341-202), and two Phase 3 studies (RX-3341-302 and RX-3341-303) (See APPENDIX 4.3). The following response endpoints were evaluated: objective response at 48 to 72 hours after initiation of treatment and at the end of treatment (Studies RX-3341-302 and RX-3341-303 only); investigator-assessed clinical response at end of treatment (EOT), follow-up (FU) and 30-days late follow-up (LFU); microbiological response at FU and LFU; and lesion size daily on days 1 to 4, EOT, FU and LFU. Reviewer also conducted an independent analysis for the relationship between free drug AUC_{24} and objective response at 48 to 72 h in microbiologically evaluable (ME) population.

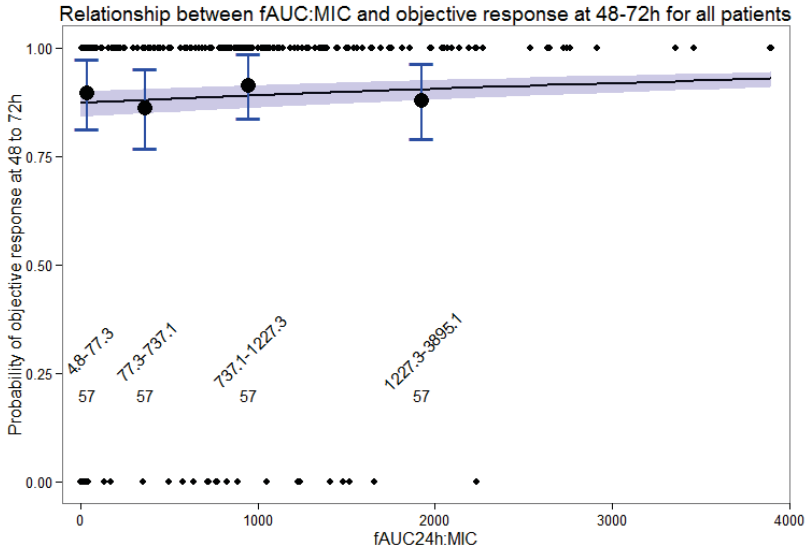
Applicant stated that a population exposure-response (E-R) analysis for all the efficacy endpoints failed to demonstrate relationships between $fAUC_{24}$/MIC and any response endpoints (flat E-R relationship), suggesting the delafloxacin exposure in both Phase 2 and Phase 3 studies may have reached a plateau of the exposure-response curve for efficacy for the organisms associated MIC values evaluated, therefore supporting the proposed dosing regimens in ABSSSI patients.

The pharmacometrics reviewer conducted an independent analysis for the relationship between free drug AUC_{24}/MIC ($fAUC_{24}$/MIC) and objective response at 48 to 72 h. A total of 228

patients were included in the exposure-response analysis with both exposure and efficacy endpoint information of objective response rate at 48 to 72 h. Among them, 159 patients were found to have *S. aureus* in their blood samples or infection sites. No significant relationship was identified for all the patients or patients with *S. aureus* (Figure 3.3.1-1). The results of reviewer's analyses are consistent with Applicant's conclusion.

Figure 3.3.1-1 Exposure-response analysis for objective response at 48 to 72h for all patients (upper) and patients with *S. aureus* (lower) using *f*AUC_{24}:MIC (Reviewer's Analysis)





3.3.2 Is the proposed dosing regimen appropriate for the general patient population for which the indication is being sought?

Yes, the proposed three dosing regimens (IV only, IV to oral, and oral only) as shown below are all acceptable for the general patient population with ABSSSI.

- 300 mg IV Q12h administered over 60 minutes by IV infusion for 5 to 14 days
- 300 mg IV Q12h administered over 60 minutes by IV infusion, then switch to 450 mg oral Q12h at the discretion of the physician for a total duration of 5 to 14 days
- 450 mg oral Q12h for 5 to 14 days

The IV only dosing regimen was used in two Phase 2 studies (Study RX-3341-201 and Study RX-3341-202) along with one Phase 3 study (Study RX-3341-302). The evidence of effectiveness for delafloxacin in the treatment of ABSSSI has been demonstrated in all three studies (see Section 3.3.1 for details). The IV switch to oral dosing regimen was studied in one Phase 3 study (Study RX-3341-303). In addition,the flat E-R relationship suggested that the delafloxacin exposure in Phase 2 and 3 studies may have reached a plateau for efficacy.

The oral only delafloxacin dosing regimen has not been evaluated in any clinical studies in this application. The evaluation of the appropriateness of the oral delafloxacin dosing regimen focused on comparison of delafloxacin PK between IV and oral formulations to ensure that adequate delafloxacin exposure can be achieved for the oral delafloxacin dosing regimen.

Applicant conducted a clinical study (Study RX-3341-115) to evaluate the bioavailability of oral delafloxacin (450-mg tablet) relative to intravenous (IV) delafloxacin (300 mg infused over 1 hour) in healthy subjects. The results indicated that the overall systemic exposure (AUC_{0-t} and AUC_{0-inf}) of delafloxacin following the oral administration of 450 mg tablet was comparable to the 300 mg IV infusion for 1 hour after a single dose (See APPENDIX 4.5.14).

The reviewer conducted a comparison of PK results across studies which used to-be-marketed IV and oral formulations in this application. The results from cross-study comparison indicated that the systemic overall exposure of delafloxacin (AUCs) after a single 300 mg IV dose was similar to that after a single 450 mg oral dose. Please refer to Table 3.3.2-1 for details.

Table 3.3.2-1 System Exposure (AUC) and C_{max} of Delafloxacin after a Single 300 mg IV Dose vs. a Single 450 mg Oral Dose (Reviewer's Analysis)

	Oral Tablet (to-be-marketed) 450 mg Single Dose		
Study	Mean AUC_{0-t} (µg.h/mL) (CV%)	Mean AUC_{0-inf} (µg.h/mL) (CV%)	Mean C_{max} (µg/mL) (CV%)
Study RX-3341-118 (N=22)	22.7 (27.4)	23.49 (26.1)	7.17 (28.1)
Study RX-3341-115 (N=55)	23.27 (29.7)	24.22 (26.6)	6.12 (32.0)
	IV (to-be-marketed captisol-L) 300 mg Single Dose		
Study	Mean AUC_{0-t} (µg.h/mL) (CV%)	Mean AUC_{0-inf} (µg.h/mL) (CV%)	Mean C_{max} (µg/mL) (CV%)
Study RX-3341-107 (N=6)	21.5 (38.0)	19.6 (34.2, N=4)	8.76 (24.5)
Study RX-3341-108 (N=12)	24.74 (20.0)	24.83 (20.0)	10.43 (19.0)
Study RX-3341-110 (N=8)	23.64 (23.2)	22.57 (20.0, N=6)	9.28 (25.3)
Study RX-3341-115 (N=55)	26.90 (21.5)	26.68 (22.6)	10.75 (21.3)
Study RX-3341-112 (N=18)	20.57 (21.3)	20.77 (21.9, N=17)	8.86 (18.9)

Taken together the results from Study RX-3341-115 and comparison of overall systemic exposure (AUCs) of delafloxacin across PK studies using to-be-marketed IV and oral formulations, the 450 mg oral tablet achieves comparable delafloxacin exposure to the 300 mg IV solution. Therefore, the proposed oral only dosing regimen 450 mg PO Q12h for 5-14 days is acceptable based on healthy volunteer PK data from a Clinical Pharmacology perspective.

Of note, the patients in both Phase 3 studies were exposed to sulfobutyl-ether-β-cyclodextrin (SBECD), an excipient in delafloxacin IV formulation (b) (4) SBECD may have potential nephrotoxicity based on animal studies. No adverse events in Phase 3 studies were found to be directly related to SBECD. Based on results from studies using IV delafloxacin formulations (with or without SBECD), SBECD did not appear to alter delafloxacin PK. In addition, SBECD has been used in several FDA approved IV drug products; there is no information collected by the Review Team that suggests that SBECD has an impact on the efficacy of delaflxoacin.

Because the proposed oral only dosing regimen was not studied in any patients with ABSSSI, an information request (IR) was issued to the Applicant on 11/30/2016 to clarify whether ABSSSI has any impact on the pharmacokinetics, specifically the absorption, of delafloxacin for the oral tablet formulation.

In their response to our IR, Applicant stated that since disease status was not identified as a statistically significant covariate in the Population PK analysis, the severity of ABSSSI disease did not impact the IV pharmacokinetics of delafloxacin; in addition, clinical outcomes by the key efficacy endpoints were comparable in ABSSSI patients who received only IV delafloxacin and patients who received IV followed by oral delafloxacin. Please refer to the clinical review by Dr. Caroline Jjingo for details.

Although the oral only dosing regimen was not studied in patients with ABSSSI, based on the rationale provided in this application and the reviewer's population PK analyses, it appears that the disease state of ABSSSI is not likely to alter the PK, specifically the absorption, of delafloxacin for the oral tablet formulation.

3.3.3 Is an alternative dosing regimen and/or management strategy required for subpopulations based on intrinsic factors?

Based on the population pharmacokinetic analysis, the pharmacokinetics of delafloxacin are not significantly impacted by age, gender, race, weight, and body mass index (See APPENDIX 4.2).

Pharmacogenomics

The primary metabolic pathway of delafloxacin is glucuronidation mediated mainly by UGT (UDP-glucuronosyltransferase)1A1, UGT1A3, and UGT2B15 (see Section 3.2 for details). Applicant evaluated the effect of genetic polymorphisms in UGT1A1, UGT2B4, UGT2B15, and ABCB1 on the PK (e.g., C_{max}, C_{min}, AUC, $t_{1/2}$, CL/F, V_{β}/F) of delafloxacin using DNA samples collected from Study M00-024 and Study M01-301.

Per Applicant's report of pooled data from the 2 studies, ABCB1 polymorphisms, UGT2B4*2, or UGT1A1*28 did not influence any of the PK parameters. Applicant's analysis concluded that day 5 dose-normalized trough mean plasma drug concentration (C_{min}) significantly correlated with UGT2B15*2 genotype (ANOVA F=3.8, p=0.036, N=27). However, due to the limited sample size, Applicant concluded that further confirmation is required to elucidate the role of UGT2B15. Unfortunately, no additional data are available from any of the subsequent studies (See APPENDIX 4.5.18). Since the PK/PD index for delafloxacin is *f*AUC/MIC, C_{min} is not expected to be critical to the clinical efficacy. In addition, the UGT2B15*2 genotype did not correlate with AUC.

Hepatic Impairment

It was observed that total (AUC_{0-t} and AUC_{0-inf}) and partial (AUC_{0-12} and AUC_{0-24}) exposures for delafloxacin in the plasma increased by approximately 1.1- to 1.4-fold for subjects with severe hepatic impairment (Child-Pugh Class C) compared with healthy subjects. Based on the acceptable safety findings in patients receiving 450 mg BID IV delafloxacin whose AUCs were approximately 1.6-fold higher than those in patients receiving 300 mg BID IV delafloxacin, dose adjustment in patients with hepatic impairment is not necessary (See APPENDIX 4.5.6).

Renal Impairment

We agree with Applicant on the proposed dosing regimen of delafloxacin for patients with mild and moderate renal impairment, 300mg IV Q12h or 450mg PO Q12h. However, we recommend the originally proposed dosing regimens for patients with severe renal impairment be revised as shown in the Table below. In addition, we do not recommend the use of delafloxacin (IV and oral) in patients with ESRD.

The dose adjustment in patients with renal impairment was based on the exposure of delafloxacin from IV and oral formulation along with the exposure of SBECD from the IV formulation.

	Applicant Proposed Dosing Regimen	FDA Recommended Dosing Regimen
Severe Renal Impairment (eGFR[a]: 15-29 mL/min/1.73m^2)	200 mg IV Q12h (b) (4)	200 mg IV Q12h or 450 mg PO Q12h Serum creatinine levels should be closely monitored in these patients receiving IV delafloxacin, and, if increases occur, consideration should be given to changing to oral delafloxacin.
End Stage Renal Disease (ESRD) (eGFR[a]< 15/min/1.73m^2 with or without hemodialysis)	(b) (4) (b) (4) (b) (4)	IV and Oral: NOT RECOMMENDED

[a] *eGFR: estimate of GFR based on MDRD equation*

Results from Renal Impairment Study

Based on Applicant's analysis of a PK study in subjects with renal impairment (Study RX-3341-110, APPENDIX 4.5.13), total delafloxacin exposure in plasma consistently increased with decreasing renal function. After a 1-hour 300 mg delafloxacin infusion, arithmetic mean AUC_{0-t} values for the severe renal impairment group and the ESRD group were 2.1-fold and 3.5- to 4.1-fold higher, respectively, than the corresponding exposure observed for the healthy group.

Of note, based on the description in the study report, Applicant only included patients requiring dialysis in the ESRD group and conducted crossover study by administering delafloxacin 1 hour before hemodialysis (with dialysis) and 1 hour after hemodialysis (without dialysis). Therefore, the subjects with dialysis and without dialysis are the same subjects.

After reviewing the raw plasma concentration data, the review team identified two outliers receiving 300 mg IV dose. The plasma concentrations of the first three time points for Subjects 1404 (severe renal impairment) and 1507 (ESRD) were unexpectedly high (approximately 10-fold higher than mean Cmax following administration of a single 300 mg IV dose) with no identified reasons. Therefore, the reviewer re-analyzed AUC_{0-t} by exclusion of these two subjects. Please refer to Table 3.3.3-1 for results.

Table 3.3.3-1 Comparison of Fold Change in AUC across Different Levels of Renal Impairment (Before vs. After Exclusion of the Two Subjects) for Subjects receiving 300 mg IV dose (Reviewer Analysis)

Observed Delafloxacin AUC fold increase	Mild	Moderate	Severe	ESRD with HD	ESRD w/o HD
Before exclusion	1.3	1.6	2.1	3.6	4.1
After exclusion	1.3	1.6	1.8	2.1	2.6

Based on the results after excluding the two subjects, the AUC values for the severe renal impairment group, ESRD with hemodialysis, and ESRD without hemodialysis, were 1.8, 2.1, and 2.6 fold higher, respectively, than the corresponding exposure in subjects with normal renal function after receiving 300 mg IV dose.

Similar to the IV dose, after a single 400 mg oral dose (4×100 mg delafloxacin capsules), total delafloxacin exposure in plasma also consistently increased with decreasing renal function but to a lesser extent (Table 3.3.3-2). There are no observed data in ESRD patients (with and without hemodialysis) receiving oral delafloxacin.

Table 3.3.3-2 Fold Change in AUC for both IV and Oral Formulations across Different Levels of Renal Function (The results for 300 mg IV were calculated after excluding Subject 1404 and Subject 1507)

Observed AUC fold increase	Mild	Moderate	Severe	ESRD with HD	ESRD w/o HD
300 mg IV	1.3	1.6	1.8	2.1	2.6
400 mg oral Phase 1 capsule	1.1	1.5	1.6		

It was also noted that the ratios of AUC_{0-t} of delafloxacin following oral and IV administration were comparable from normal renal function group to severe renal impairment. This result suggested that renal impairment (up to severe) does not appear to exert a significant impact on the absorption of delafloxacin oral capsules (Table 3.3.3-3). However, the impact of ESRD on

delafloxacin disposition following oral dosing is unknown as oral delafloxacin was not studied in patients with ESRD.

Table 3.3.3-3 Delafloxacin AUC ratio Following Oral and IV Administration across Renal Function Groups (The results for 300 mg IV were calculated after excluding Subject 1404 and Subject 1507)

	Normal (n=8)	**Mild (n=8)**	**Moderate (n=8)**	**Severe (n=8)**
Mean AUC_{0-t}, oral	25.75	26.75	36.91	37.85
Mean AUC_{0-t}, IV	23.64	31.04	39.29	43.19
AUC Ratio (oral/IV)	1.09	0.86	0.93	0.88

Results from Population PK Analyses

The plasma concentration values for the first three time points for Subject 1404 and 1507 were excluded from the population PK dataset. The fold change in patients with renal impairment vs. patients with normal renal function based on population PK model for both IV and oral formulations were lower than the observed results (See APPENDIX 4.2). Please refer to Tables 3.3.3-4 and 3.3.3-5 for details. This can be explained by the fact that the simulated exposure based on population PK model used the demographics of Phase 3 study, which is not exactly the same as that of dedicated renal impairment study. Also, the simulation was conducted on a basis of a much broader range of eGFR values than that in the dedicated renal impairment study with limited number of subjects in each arm. Of note, the simulation for patients with ESRD receiving oral delafloxacin was performed under the assumption that there is no impact of ESRD on delafloxacin absorption as no observed data are available in these patients.

Table 3.3.3-4 Simulated Mean Steady State AUC_{24h} of Delafloxacin – 300 mg IV Q12h in Patients with Renal Impairment

	Normal	**Mild**	**Moderate**	**Severe**	**ESRD w/o HD**
Simulated mean AUC_{24h} on Day 5 (mg h/L)	50.1	55.9	64.7	80.7	106.0
AUC ratio of impaired vs. normal		1.1	1.3	1.6	2.1

Table 3.3.3-5 Simulated Mean Steady State AUC_{24h} of Delafloxacin– 450 mg PO Q12h in Patients with Renal Impairment

	Normal	Mild	Moderate	Severe	ESRD w/o HD
Simulated mean AUC_{24h} on Day 5 (mg h/L)	47.5	51.8	59.5	70.3	87.0
AUC ratio of impaired vs. normal		1.1	1.3	1.5	1.8

There were 2 patients with ESRD treated with delafloxacin based on eGFR or 1 patient based on estimated CrCl in Phase 3 studies in this application. There were only 4 patients with severe renal impairment treated with delafloxacin based on eGFR or 1 patients based on estimated CrCl in Phase 3 studies. Therefore, only very limited safety and efficacy data were collected from these sub-populations. In addition, no patients received any dose adjustment based on their renal function. Please see Table 3.3.3-6 for details.

Table 3.3.3-6 Number of Patients in delafloxacin arm in clinical studies (Safety dataset for studies 302 and 303)

	Normal	Mild	Moderate	Severe	ESRD
CLcr (estimated by CG with actual body weight)	609	82	37	1	1
eGFR (estimated by MDRD)	460	216	48	4	2

**MDRD: Modification of Diet in Renal Disease equation*

**CG: Cockcroft-Gault equation*

Since the AUC was observed to be 1.6 fold higher in subjects with moderate renal impairment compared to normal renal function group in Study RX-3341-110, an analysis in patients with moderate renal impairment based on their estimated CrCl was conducted to evaluate whether the increased exposure will cause any additional safety concern. A total of 37 patients in Phase 3 studies and 4 patients in Phase 2 studies with moderate renal impairment, respectively, were identified. The results indicated that no additional Treatment-Emergent- Adverse-Events (TEAEs) were observed in patients with moderate renal impairment compared to other patients in the Phase 3 Safety analysis dataset. Since only 4 patients in Phase 2 studies were identified in moderate renal impairment group, no meaningful conclusions were able to drawn based on such a small sample size. Please refer to Table 3.3.3-7 for details.

Table 3.3.3-7 Comparison of Treatment-Emergent-Adverse-Events (TEAEs) between Patients with Moderate Renal Impairment vs. Pooled Phase 3 Patients in Delafloxacin Arm

Population	Percent of Patients with any TEAE (%)	Percent of patients with mild TEAEs (%)	Percent of patients with moderate TEAEs (%)	Percent of patients with severe TEAE (%)
pooled Phase 3 patients with moderate RI (N=37)	49	35.1	10.8	2.7
pooled Phase 3 patients in this submission (patients with moderate RI excluded) (N=704)	44.9	26.3	15.1	3.6

Moreover, the probability of target attainment (PTA) analysis results also supported the proposed dosing regimen for patients with mild or moderate renal impairment (See Section 3.3.4 and Appendices 4.4). The efficacy analysis also indicated that the objective response at 48-72 hours (i.e., primary clinical endpoint) in the two pivotal Phase 3 studies was comparable across renal function categories. Please refer to the statistics review by Dr. Janelle Charles.

Taken together, the AUC of delafloxacin, efficacy and safety data, and PTA analysis results indicated that the proposed dosing regimen, 300 mg IV Q12h or 450 mg oral Q12h, for patients with mild or moderate renal impairment is acceptable.

Based on the results from the PK study in patients with renal impairment and ESRD (Study RX-3341-110), Applicant proposed to reduce the dose in patients with severe renal impairment (b) (4) to 200 mg IV Q12h (b) (4) The proposed IV and oral dosing regimens in these populations were not studied in Phase 2 and 3 trials.

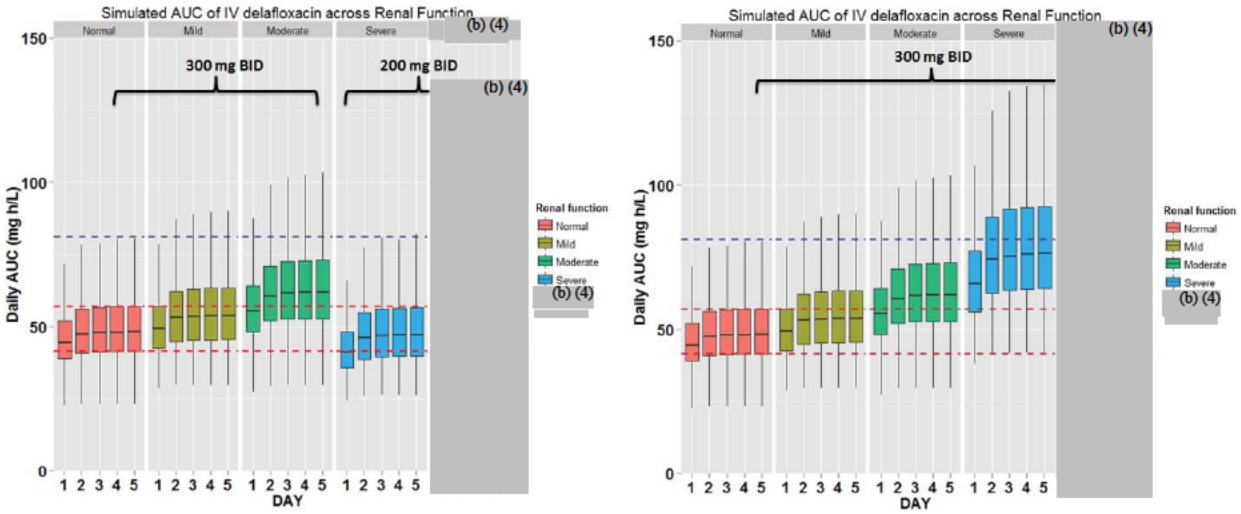
The simulated AUC of delafloxacin in patients with severe renal impairment using the proposed IV dose (200 mg IV Q12h) was comparable to that in patients with normal renal function receiving delafloxacin 300 mg IV Q12h (Table 3.3.3-8 and Figure 3.3.3-1). In addition, the PTA analysis results also supported the proposed IV dosing regimen for patients with severe renal impairment based on the AUC of delafloxacin (See Section 3.3.4 and APPENDIX 4.4).

Table 3.3.3-8 Simulated AUC_{24h} of Delafloxacin at Steady State after Receiving the Proposed IV dosing Regimen

	300 mg IV BID			200mg IV BID (b) (4)
Renal Function	Normal	Mild	Moderate	Severe
Simulated mean AUC_{24h} on Day 5 (mg h/L)	50.1	55.9	64.7	49.7

Note: (b) (4)

Figure 3.3.3-1 Simulated Daily AUC from Day 1 to 5 for IV Delafloxacin across Renal Function under Two Dosing Regimens (Proposed by the Applicant (Left) and no dose reduction (Right)) (Pharmacometrics Reviewer's analysis)



Blue dashed lines represent the mean daily AUC (81.1 mg·h/L) on Day 5 at the 450 mg IV Q12h dose based on population PK model and demographics information in Study RX-3341-201; Red dashed lines represent the 25th and 75th percentile of daily AUC on Day 5 at the 300 mg IV Q12h dose in normal renal function based on population PK model. Note (b) (4)

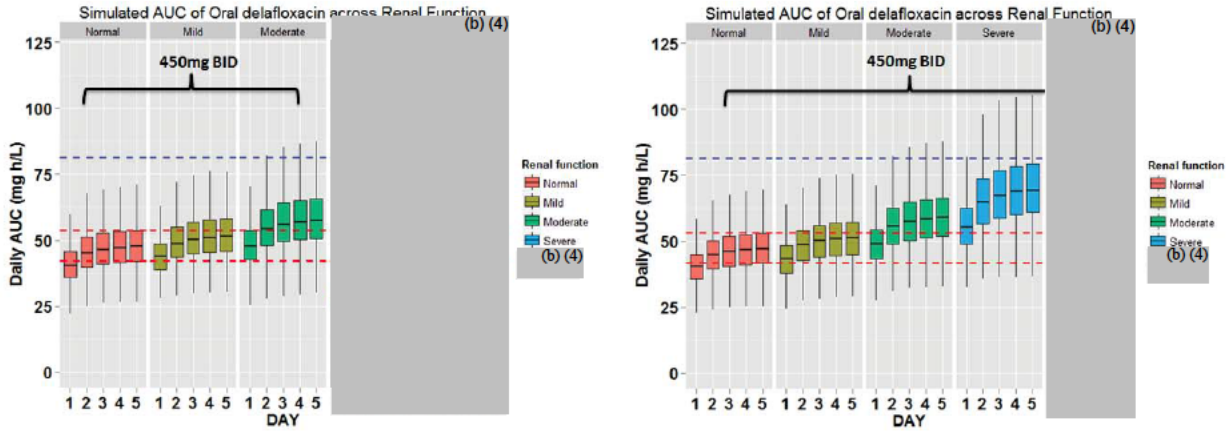
The simulated AUC of delafloxacin in patients with severe renal impairmen (b) (4) compared to the patients with normal renal function receiving delafloxacin 450 mg oral Q12h (Table 3.3.3-9 and Figure 3.3.3-2). In addition, results from the PTA analyses show that the (b) (4) µg/mL for *S. aureus* by using the target for net bacterial stasis. (See Section 3.3.4 and APPENDIX 4.4) Therefore, additional simulations were conducted to evaluate the appropriateness of the 450 mg Q12h oral dose (without dose reduction) in patients with severe renal impairment.

Table 3.3.3-9 Simulated AUC_{24h} of Delafloxacin at Steady State after Receiving the Proposed Oral dosing Regimen

	450mg PO BID			(b) (4)
Renal Function	Normal	Mild	Moderate	
Simulated mean AUC_{24h} on Day 5 (mg h/L)	48.2	52.3	58.7	

Note: (b) (4) *. The slight differences in the simulated AUC of delafloxacin in patients with normal renal function patients with mild and moderated renal impairment at the 450 mg Q12h oral dose shown in Table 3.3.3-5 and Table 3.3.3-9 are attributed to the separate simulations.*

Figure 3.3.3-2 Simulated Daily AUC from Day 1 to 5 for PO Delafloxacin across Renal Function under Two Dosing Regimens (Proposed by the Applicant (Left) and no dose reduction (Right)) (Pharmacometrics Reviewer's analysis)



Blue dashed lines represent the mean daily AUC (81.1 mg·h/L) on Day 5 at the 450 mg IV Q12h dose based on population PK model and demographics information in Study RX-3341-201; Red dashed lines represent the 25th and 75th percentile of daily AUC on Day 5 at the 450 mg PO Q12h dose in normal renal function based on population PK model. Note: (b) (4)

As shown in Table 3.3.3-5 and Figure 3.3.3-2, without dose reduction for the oral delafloxacin across renal function, the simulated steady state AUC of delafloxacin in patients with severe renal impairment receiving 450 mg oral Q12h (70.3 mg•h/L) was about 48% higher than the AUC in patients with normal renal function (47.5 mg•h/L), but was slightly lower than the simulated AUC in patients receiving 450 mg Q12h IV delafloxacin (81.1 mg•h/L) in one Phase 2 study (Study RX-3341-201). In this Phase 2 study, patients were treated in two delafloxacin dose arms, 300mg IV Q12h and 450mg IV Q12h, for 5-14 days. More AEs were experienced by patients in the 450 mg treatment group compared to patients in the 300 mg treatment group, the most common AEs overall were nausea, diarrhea, vomiting, headache, and infusion site pain; all of these AEs were more frequent in the 450-mg delafloxacin group than in the 300-mg delafloxacin group. However, most of these AEs were mild or moderate in severity and did not lead to treatment discontinuation. No patients died during the study. A total of 7 patients experienced 8 serious adverse events (SAEs) during the study, of which 3 were in the 450 mg

delafloxacin group (osteomyelitis, convulsion, and bradycardia). Based on the safety evaluation, no major safety concerns were identified with 450 mg Q12h IV. Please refer to the clinical review by Dr. Caroline Jjingo for details.

In conclusion, based on the simulated overall delafloxacin exposure and safety analysis results, Applicant proposed IV dosing regimen for patients with severe renal impairment, 200 mg IV Q12h, seems reasonable. (b) (4) in patients with severe renal impairment, we recommend no dose reduction (i.e., 450 mg oral Q12h) in these patients. This 450 mg Q12h oral dosing regimen in patients with severe renal impairment is also supported by 1) the lower AUC between patients with severe renal impairment receiving delafloxacin 450 mg oral Q12h and patients receiving delafloxacin 450 mg IV Q12h, and 2) no major safety concerns were identified with 450 mg Q12h IV (RX-3341-201).

Sulfobutyl-ether-β-cyclodextrin (SBECD, Captisol®)

Sulfobutyl-ether-β-cyclodextrin (SBECD, Captisol®) is a cyclodextrin excipient used to improve drug solubility, stability, and bioavailability. Delafloxacin for IV injection is formulated with SBECD (b) (4) In humans, SBECD is eliminated unchanged almost entirely by renal filtration. Therefore, SBECD pharmacokinetics can be markedly altered in subjects with impaired renal function, and SBECD exposures was reported to increase with decreased renal function in Study RX-3341-110 (See APPENDIX 4.5.13). Applicant's analysis results are summarized in Table 3.3.3-10. The reviewer's analysis results after excluding the two subjects (Subject 1404 and Subject 1507) whose plasma concentrations in the first three time points were unexpectedly high are summarized in Table 3.3.3-11.

Table 3.3.3-10 SBECD exposure following a 300mg IV Single Dose (Study RX-3341-110, Applicant's results)

Renal Function Group (eGFR)	normal	mild	moderate	severe	ESRD with HD	ESRD w/o HD
Mean SBECD AUC 0-t(μg*hr/mL) (CV%)	374.55 (11.9)	493.39 (31.8)	815.87 (26.5)	1975.35 (30.7)	3183.89 (62.0)	11164.52 (31.7)
AUC fold increase compared to patients with normal renal function	1	1.3	2.2	5.2	8.5	30

Table 3.3.3-11 SBECD Exposure Following a 300mg IV Single Dose (Study RX-3341-110, Reviewer's results)

Renal Function Group (eGFR)	normal	mild	moderate	severe	ESRD with HD	ESRD w/o HD
Mean SBECD AUC 0-t(μg*hr/mL) (CV%)	374.55 (11.9)	493.39 (31.8)	815.87 (26.5)	2010.4 (32.1)	2807.9 (55.8)	10048 (34.7)
AUC fold increase compared to patients with normal renal function	1	1.3	2.2	5.4	7.5	26.8

The potential risk of nephrotoxicity caused by SBECD has been identified in animal studies, specifically cytoplasmic vacuolation in the epithelium of the renal tubules, renal pelvis and urinary bladder (APPENDIX 4.5.17). There are two animal studies included in this application using the IV formulation containing SBECD. One is a 14-day rat intravenous comparison with delafloxacin in Captisol vs (b) (4). Captisol is the sulfobutylether-beta-cyclodextrin and (b) (4) The dose of SBECD in this study was 80 mg/kg Q24h, the parenteral permitted daily exposure (PDE) recommended by European Medicine Agency (EMA)[1]. It appears that delafloxacin formulated in either solvent showed no real evidence of toxicity (other than infusion site histopathology), and no real difference between formulations. The other study was a 28-day IV toxicology study in dogs administered delafloxacin in SBECD. In this study, Applicant did observe minimal to mild "renal tubule vacuolation" in both treated and vehicle control animals that they linked to SBECD use. Therefore, it seems that the toxicity caused by SBECD is treatment duration related. Please refer to the Pharmacology/Toxicology review by Dr. Amy Nostrandt for details.

Data of adverse events associated with cyclodextrins in humans are limited and conflictory (See APPENDIX 4.5.17), especially in patients with renal impairment. In addition, only 4 patients (renal function estimated based on CrCl) with severe renal impairment were enrolled in clinical studies in this application. As a consequence, the safety of SBECD in patients with severe renal impairment cannot be evaluated adequately by the limited Phase 2 and 3 clinical data.

The IV formulation of voriconazole (VFEND®) contains SBECD. In a study of IV voriconazole, three out of six subjects with moderate renal impairment receiving voriconazole had > 25% increase in serum creatinine compared to baseline[2]. The VFEND label recommends that IV voriconazole should be avoided in patients with moderate or severe renal impairment (creatinine clearance < 50 mL/min), unless an assessment of the benefit/risk to the patient justified the use of IV voriconazole. Serum creatinine levels should be closely monitored in these patients, and, if increases occur, consideration should be given to changing to oral voriconazole therapy. We agree with Applicant's proposal of reducing IV dose to 200 mg Q12h in patients with severe renal impairment because it not only provides comparable delafloxacin exposure to normal subjects, but also decreases the SBECD amount from 2400 to 1600 mg per dose. At the 200 mg Q12h IV dose in patients with severe renal impairment, the AUC_{τ} of SBECD is estimated to be 1340 µg•hr/mL assuming a linear dose-exposure relationship of SBECD (Table 3.3.3-12), which is still 64% higher than that in patients with moderate renal impairment receiving the 300 mg Q12h IV dose. In addition, given uncertainties about SBECD safety margin in humans, we recommend serum creatinine levels should be closely monitored in patients with severe renal

[1] http://www.ema.europa.eu/docs/en_GB/document_library/Report/2014/12/WC500177936.pdf
[2] Samantha Abel, et al., "Pharmacokinetics, Safety and Tolerance of Voriconazole in Renally Impaired Subjects-Two Prospective, Multicentre, Open-Label, Parallel-Group Volunteer Studies", Clin Drug Invest 2008; 28 (7): 409-420

impairment if receiving IV delafloxacin, and, if increases occur, consideration should be given to changing to oral delafloxacin therapy.

Table 3.3.3-12 Comparison of SBECD Exposures between IV Voriconazole and IV Delafloxacin

	VFEND (3 mg/kg IV BID)	Delafloxacin[a] (300 mg BID)	Delafloxacin[a] (200 mg BID)
SBECD / Dose (mg)	3936 (BW: 82 kg)[b]	2400	1600
Mean AUC_τ (µg*hr/mL)	2128 (moderate RI)	815 (moderate RI)	1340 (severe RI)

[a] *$AUC\tau$ of delafloxacin is estimated based on single dose SBECD PK*
[b] *Actual mean body weight of subjects in the voriconazole study*

(b) (4)

(b) (4)

(b) (4) Because hemodialysis is usually performed intermittently (every 2 or 3 days) in ESRD patients, SBECD exposure is expected to accumulate extensively on interdialytic days. Although SBECD can be removed by hemodialysis, it is basically impractical to restrict admistration of IV delafloxacin only during hemodialysis.

Patients with End Stage Renal Disease (ESRD)

(b) (4)

(b) (4) the unknown risk of extensive accumulation of SBECD raises a safety concern in patients with ESRD receiving IV delafloxacin. In addition the dedicated renal impairment study included subjects where delafloxacin was administered within one hour after completion of a hemodialysis session. While such information can inform dosing with respect to the timing of dialysis on dialysis days, this information may not adequately describe delafloxacin and SBECD exposure in subjects on an off-dialysis day. Furthermore, no observed data are available in patients with ESRD receiving oral delafloxacin. Therefore, the impact of ESRD on delafloxacin disposition following oral dosing is unknown. Taken together, we do not recommend the use of delafloxacin (IV and oral) in patients with ESRD.

3.3.4 Are there clinically relevant food-drug or drug-drug interactions and what is the appropriate management strategy?

Food Effect

Applicant conducted a food effect study for the to-be-marketed delafloxacin tablet (Study RX-3341-116). Results from this study showed that total delafloxacin exposure (AUC_{0-t} and AUC_{0-inf}) of delafloxacin was not affected by administration under fasted or fed (high-fat, high-calorie) conditions (Table 3.3.5-1). Although the peak exposure (C_{max}) of delafloxacin was reduced by 20.5% when administered under fed conditions versus fasted conditions, it is not expected to affect the clinical efficacy outcomes since the *f*AUC/MIC was shown to be the PK/PD index for delafloxacin associated with antibacterial activity in animal studies. Please refer to APPENDIX 4.5.15 for details. Therefore, delafloxacin (oral tablet) may be administered with or without food.

Table 3.3.5-1 Mean ± SD (CV) Plasma Pharmacokinetic Parameters of Delafloxacin by Treatment (Pharmacokinetic Population) (Adapted from Table 11-2 in the Study report of Study RX-3341-116)

	Treatment		
Parameter (Unit)	**Delafloxacin 900 mg (Fasted) (A) (N = 28)**	**Delafloxacin 900 mg (Fed) (B) (N = 29)**	**Delafloxacin 900 mg (Fasted, with a meal 2 hours after dosing) (C) (N = 25)**
AUC_{0-12} (µg•h/mL)	52.12 ± 14.44 (27.7)	49.91 ± 12.19 (24.4)	44.51 ± 10.86 (24.4)
AUC_{0-inf} (µg•h/mL)[a]	55.15 ± 13.68 (24.8)	58.21 ± 11.41 (19.6)	52.21 ± 12.55 (24.0)
AUC_{0-t} (µg•h/mL)	56.21 ± 15.21 (27.1)	57.09 ± 14.15 (24.8)	49.77 ± 12.01 (24.1)
AUC_{0-24} (µg•h/mL)	54.52 ± 15.01 (27.5)	55.08 ± 13.94 (25.3)	47.64 ± 11.74 (24.6)
AUC_{ext} (µg•h/mL)[a]	0.91 ± 0.85 (93.1)	0.71 ± 0.64 (90.7)	0.64 ± 0.44 (68.9)
C_{max} (µg/mL)	11.52 ± 2.76 (24.0)	9.14 ± 2.81 (30.8)	11.75 ± 2.64 (22.5)[b]
T_{max} (h)[c]	1.25 (0.50, 4.00)	2.50 (1.00, 6.00)	1.50 (0.50, 2.50)[b]

a. n = 9, 16, and 14 for Treatments A, B, and C, respectively

Of note, the dose used in the food effect study was higher (900 mg, 450 mg×2 tablets) than the strength of the oral formulation (450 mg). However, since the increase in delafloxacin AUC was approximately dose proportional at the single dose ranging from 450 mg to 900 mg for the to-be-marketed tablet (See Dose Proportionality in Section 3.2 for details),the use of 900 mg in the food effect study for delafloxacin is acceptable.

Drug-Drug Interactions (DDIs)

UGT or CYP Enzyme Mediated DDIs

In vitro studies showed that multiple UGTs (UGT1A1, UGT1A3, UGT2B15) are involved in delafloxacin metabolism. Because metabolism is only responsible for ⩽ 20% of the elimination of delafloxacin, inhibition of UGTs are unlikely to impact delafloxacin PK.

In vitro studies showed that delafloxacin was not an inhibitor of UGT enzymes (UGT1A1, 1A3, 1A4, 1A6, 1A9, 2B7, or 2B15). Delafloxacin does not inhibit CYP1A2, 2A6, 2B6, 2C8, 2C9, 2C19, 2D6, 2E1, or 3A4/5 activities at drug concentrations (300–500 μM) that are well above the C_{max} achieved at a 300 Q12h IV and 450 mg Q12h oral dosages of delafloxacin. Therefore, clinically significant DDIs due to inhibition or activation of CYP-mediated biotransformation of co-administered drugs by delafloxacin seem unlikely in humans. Please refer to APPENDIX 4.5.16 for details.

In vitro, delafloxacin was shown to be a weak inducer of CYP3A, but not of CYP1A2 or CYP2B6 expression. An in vivo DDI study (Study ML-3341-118) of multiple oral doses of delafloxacin (450 mg Q12h) on a single oral dose of midazolam (5 mg) demonstrated no impact on midazolam PK, and minimal impact on 1-hydroxymidazolam PK (Tables 3.3.5-2 and 3.3.5-3). These results indicate that delafloxacin is not a CYP3A inducer at clinically relevant concentrations.

Table 3.3.5-2 Statistical Analysis of Plasma Pharmacokinetic Parameters of Midazolam (Midazolam Pharmacokinetic Population) (Adapted from Table 11-3 in the study report of Study ML-3341-118)

Parameter (Unit)	Treatment	N	Geometric LS Means	90% CI of Geometric LS Means	Treatment Comparison	Ratio (%) of Geometric LS Means	90% CI of the Ratio (%)
AUC_{0-t} (h•ng/mL)	Midazolam Alone	22	61.767	(55.436, 68.820)			
	Midazolam + Delafloxacin	22	55.216	(49.557, 61.522)	Midazolam + Delafloxacin vs Midazolam Alone	89.40	(83.196, 96.056)
AUC_{0-24} (h•ng/mL)	Midazolam Alone	22	62.074	(55.778, 69.081)			
	Midazolam + Delafloxacin	22	55.584	(49.946, 61.858)	Midazolam + Delafloxacin vs Midazolam Alone	89.54	(83.404, 96.137)
AUC_{0-inf} (h•ng/mL)	Midazolam Alone	22	63.080	(56.620, 70.276)			
	Midazolam + Delafloxacin	22	56.394	(50.619, 62.828)	Midazolam + Delafloxacin vs Midazolam Alone	89.40	(83.226, 96.035)
C_{max} (ng/mL)	Midazolam Alone	22	25.706	(22.309, 29.622)			
	Midazolam + Delafloxacin	22	24.052	(20.873, 27.715)	Midazolam + Delafloxacin vs Midazolam Alone	93.56	(83.701, 104.589)

Abbreviations: CI, confidence interval; h, hours; LS, least squares.
Note: A linear mixed-effect model was performed on the natural log-transformed values of AUC_{0-t}, AUC_{0-24}, AUC_{0-inf}, and C_{max} with treatment as a fixed effect and subject as a random effect.

Table 3.3.5-3 Statistical Analysis of Plasma Pharmacokinetic Parameters of 1-Hydroxymidazolam (Midazolam Pharmacokinetic Population) (Adapted from Table 11-5 in the study report for Study ML-3341-118)

Parameter (Unit)	Treatment	N	Geometric LS Means	90% CI of Geometric LS Means	Treatment Comparison	Ratio (%) of Geometric LS Means	90% CI of the Ratio (%)
AUC_{0-t} (h•ng/mL)	Midazolam Alone	22	28.864	(25.136, 33.144)			
	Midazolam + Delafloxacin	22	30.754	(26.783, 35.315)	Midazolam + Delafloxacin vs Midazolam Alone	106.55	(98.790, 114.919)
AUC_{0-24} (h•ng/mL)	Midazolam Alone	22	29.410	(25.656, 33.714)			
	Midazolam + Delafloxacin	22	31.052	(27.088, 35.596)	Midazolam + Delafloxacin vs Midazolam Alone	105.58	(97.990, 113.767)
AUC_{0-inf} (h•ng/mL)	Midazolam Alone	20	29.610	(25.797, 33.987)			
	Midazolam + Delafloxacin	22	31.294	(27.310, 35.859)	Midazolam + Delafloxacin vs Midazolam Alone	105.69	(97.691, 114.337)
C_{max} (ng/mL)	Midazolam Alone	22	13.326	(11.008, 16.132)			
	Midazolam + Delafloxacin	22	15.466	(12.776, 18.722)	Midazolam + Delafloxacin vs Midazolam Alone	116.05	(101.700, 132.430)

Abbreviations: CI, confidence interval; h, hours; LS, least squares.
Note: A linear mixed-effect model was performed on the natural log-transformed values of AUC_{0-t}, AUC_{0-24}, AUC_{0-inf}, and C_{max} with treatment as a fixed effect and subject as a random effect.

Transporter-Mediated DDIs

Based on results from in vitro studies, delafloxacin is not an inhibitor of human P-gp, BCRP, OAT1, OAT3, OCT1, OCT2, OATP1B1, OATP1B3, and BSEP-mediated transport; delafloxacin is not a substrate of the transporters OAT1, OAT3, OCT1, OCT2, OATP1B1 and OATP1B3 but is considered to be a substrate for breast cancer resistance protein (BCRP) and a potential substrate of P-gp as the observed efflux ratios were 8.86 and 2.1, respectively (APPENDIX 4.5.16). Applicant provided the following rationale for not conducting an in vivo DDI study with delafloxacin and inhibitors of BCRP and P-gp: the absolute bioavailability of delafloxacin is 58.8% (to-be-marketed), so any interaction with BCRP or P-gp at the enterocyte would likely not increase peak exposures beyond those exposures shown to be tolerable at doses of 450 mg IV Q12h. (Table 3.3.5-4). In addition, safety data are available for 450 mg IV Q12h administered over multiple days which provide reassurance that any increase in exposure resulting from BCRP or P-gp inhibition would likely be tolerated.

Table 3.3.5-4 Mean Systemic Exposure of Delafloxacin After IV and Oral Administration in Clinical Studies With Highest Tolerable Doses (Adapted from Table 57 in Summary of Clinical Pharmacology)

Study No.	Delafloxacin Treatment				AUC_∞ or AUC_τ (µg·h/mL)	C_{max} or $C_{max,ss}$ (µg/mL)
	Route	Formulation	Dose (mg)	Days		
ML-3341-118	Oral	To-be-marketed	450 Q12h	5	22.7	7.45
RX-3341-104	IV	To-be-marketed	300 Q12h	12	23.36	9.29
M00-224	Oral	Phase 1	1600	1	81.7	16.6
	Oral	Phase 1	1200	5	54.3	11.5
RX-3341-108	IV	To-be-marketed	1200	1	67.1	13.2
RX-3341-103	IV	(b) (4)	450 Q12h	14	42.3	15.2

Q12h = every 12 hours.

Note: AUC_τ is 12 hours.

P-gp and BCRP are also expressed in liver and kidney and may be involvded in delafloxacin excretion; DDI at these organs could also increase delafloxacin exposure. Therefore, an information request (IR) was sent to Applicant on March 6th, 2017, requesting Applicant to provide further rationales regarding other organs (i.e., liver and kidney) for not conducting an in vivo DDI study.

In their response to the IR (submitted on March 9th, 2017), Applicant stated:

1. There are metabolic pathways for elimination of delafloxacin that do not involve the hepatic or renal efflux transporters, P-gp or BCRP. These pathways include acyl and ether glucuronidation, and oxidative metabolism (M7) which will provide an alternative pathway of elimination.
2. The general tolerability of delafloxacin has been characterized at exposures higher than generated with the proposed IV (300 mg Q12h) or oral (450 mg Q 12h) dosages. Delafloxacin has been tolerated with oral doses up to 1600 mg as a single dose and 1200 mg/day orally for 5 days. Single intravenous doses of up to 1200 mg are also tolerated. Patients also have received 450 mg IV q12h for up to 14 days in clinical trials. The most common adverse events in these higher exposures are gastrointestinal in nature, primarily diarrhea and nausea.
3. Delafloxacin will be administered for a short duration from 5 to 14 days.

A follow-up IR was sent to Applicant on March 13th, 2017, requesting Applicant to provide delafloxacin systemic exposure (i.e. AUC, C_{max}) and safety profiles for patients receiving P-gp

and/or BCRP inhibitors relative to patients receiving no P-gp and/or BCRP inhibitors in two Phase 3 studies (RX-3341-302 and RX-3341-303).

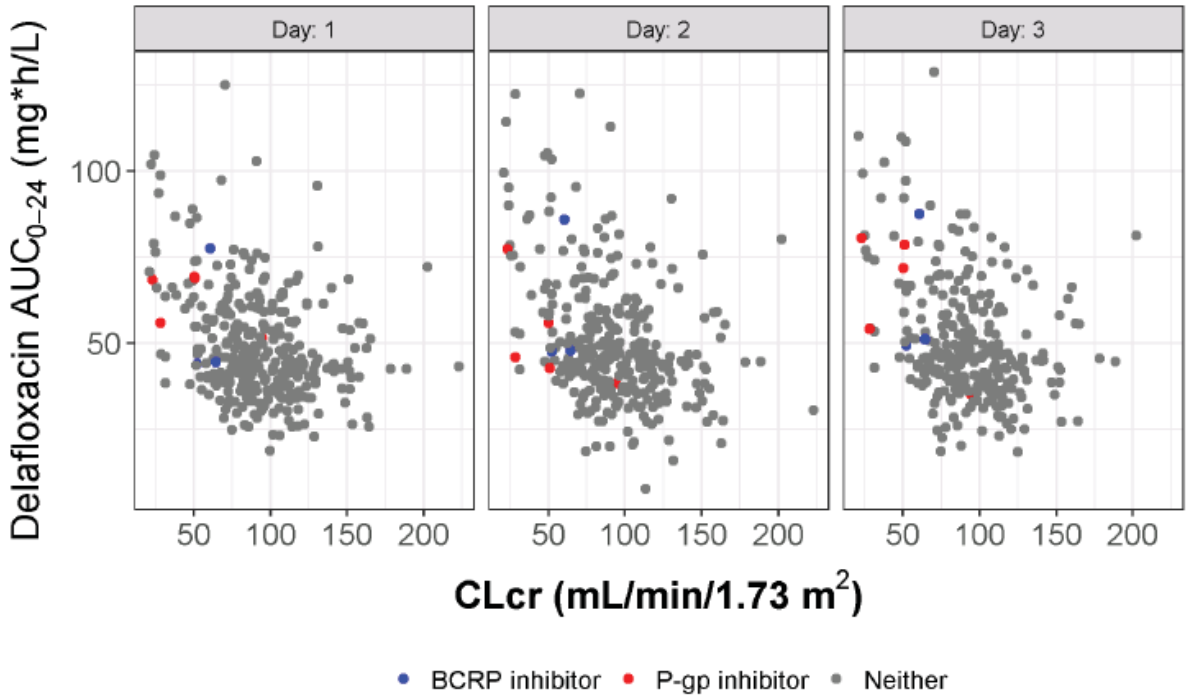
In Applicant's response to the IR (submitted on March 16th, 2017), Applicant provided additional rationale for not conducting an in vivo DDI study with delafloxacin and inhibitors of P-gp or BCRP as shown below.

- The efflux ratio of delafloxacin in BCRP-expressing MDCK-II cells is 8.86. This ratio is about 10-fold lower than corresponding efflux ratios of the proposed BCRP sensitive substrates, sulfasalazine and rosuvastatin. Recently, the role of BCRP in clinical pharmacokinetics and drug-drug interactions have been reviewed . The reported systemic exposure increase of sulfasalazine and rosuvastatin in the presence of BCRP inhibitors in in-vivo studies is ⩽2-fold. Because (1) the efflux ratio of delafloxacin is 10-fold lower than sensitive substrates, (2) the effects of BCRP inhibition on systemic exposure is ⩽ 2-fold for sensitive BCRP substrates, and (3) the good safety profile for delafloxacin, Melinta believes that the risk of clinically relevant drug-drug interaction of delafloxacin with inhibitors of P-gp/BCRP are minimal.

Applicant also identified a total of 28 of the 33 patients who received a P-gp/BCRP inhibitor at some time during the trial had received the medication concomitantly (from start of delafloxacin to EOT), 8 of which had delafloxacin concentration data available. However, none of these 8 patients received a P-gp and BCRP dual inhibitor.

Delafloxacin AUC_{0-24} values from patients who received P-gp/BCRP inhibitors along with delafloxacin are overlayed with the values from patients who received no P-gp/BCRP inhibitors. Please refer to Figure 3.3.4-1 for details.

Figure 3.3.4-1 Comparison in Delafloxacin AUC_{0-24} vs. CLcr between patients who received P-gp/BCRP inhibitors and patients who did not receive P-gp/BCRP inhibitors



Of the 33 patients that reported taking a P-gp/BCRP inhibitor during the course of the trial, a total of 21 of those patients had reported AEs/SAEs. The sponsor identified 18 patients that had an AE/SAE start occurring on study days 1-14. A total of 4/18 patients had SAEs, none of which were considered by the investigator to be treatment related. There was a total of 7/18 (38.8%) patients that reported an AE that the investigator believed might be treatment-related. Of note, the percentage of this patients with mild or moderate treatment related AE in pooled Phase 3 studies was 41.5%.

The Clinical Pharmacology Review Team reviewed the information provided by Applicant. The provided rationale does not rule out the possibility that the AUC of delafloxacin in patients coadministered IV or oral delafloxacin with P-gp and BCRP inhibitors may be greater than the exposure at the 450 mg Q12h IV dose shown to be tolerable in Study RX-3341-201. In addition, the in vivo DDI potential between delafloxacin and P-gp and/or BCRP inhibitors was not adequately addressed by the limited Phase 3 data. Therefore, the clinical relevance of coadministration of delafloxacin and P-gp and/or BCRP inhibitors is unknown.

3.3.5 Are the proposed susceptibility breakpoints acceptable?

Yes, the results of the probability of target attainment analyses (PTA) support the proposed susceptibility breakpoints for delafloxacin (Table 3.3.5-1). As for the susceptibility breakpoints of other pathogens (not listed in Table 3.3.5-1) proposed in the delafloxacin label, the reviewers defer to Clinical/Clinical Microbiology reviewers because there were no relevant clinical pharmacology data for these pathogens.

Table 3.3.5-1 Applicant Proposed Susceptibility Breakpoints for Delafloxacin

Susceptibility Test Interpretive Criteria for BAXDELA	**Minimum Inhibitory Concentrations (µg/mL)**		
Pathogen	**S**	**I**	**R**
Staphylococcus aureus (methicillin-resistant and methicillin-susceptible isolates)	(b) (4)		
Escherichia coli	≤ 0.25	0.5	≥ 1
Pseudomonas aeruginosa	≤ 0.5	1	≥ 2

1. Animal Models to Determine the PK/PD Target

The PK/PD targets associated with the activity against *S. aureus, E. coli* , and *P. aeruginosa* were determined in neutropenic murine thigh model of infection. We defer to the Clinical Microbiology reviewer for the evaluation of the bacterial strain selections. Similar to other quinolone antibiotics, the *f*AUC_{24}/MIC ratio was determined to be the PK/PD index most closely associated with delafloxacin's activity. The results from PK/PD studies in the murine neutropenic thigh model using 5 strains of *S. aureus* against which delafloxacin has a range of MICs indicated median values of *f*AUC_{24}/MIC ratios of 9.3 [range: 2.0-12.8] for bacterial stasis and 14.3 [range: 6.3-18.5] for 1-$\log_{10}$ CFU reduction, respectively. The results from a PK/PD study in the murine neutropenic thigh model using 3 strains of *E. coli* against which delafloxacin has a range of MICs indicated mean *f*AUC_{24}/MIC ratios of 14.5 for bacterial stasis, and 26.2 for a 1-$\log_{10}$ CFU reduction, respectively. Applicant did not provide ranges of *f*AUC_{24}/MIC ratios for *E. coli* in their study report. The results from a PK/PD study in the murine neutropenic thigh model using 3 strains of *P. aeruginosa* against which delafloxacin has a range of MICs indicated mean *f*AUC_{24}/MIC ratios of 3.81 [range: 3.2-5.2] for bacterial stasis, and 5.02 [range: 4.2-6.0] for a 1-$\log_{10}$ CFU reduction, respectively (See APPENDIX 4.5.1)

2. Probability of Target Attainment (PTA) for *S. aureus, E. coli,* and *P. aeruginosa*

The Pharmacometrics Reviewer conducted independent analysis for assessing the probability of target attainment in patients based on simulation using population PK model. For the indication of ABSSSI, the PK/PD target for bacterial stasis was used for the PTA analysis. For *S. aureus,* more than 90% patients receiving the proposed IV or oral dosing regimens would achieve the

PK/PD target (*f*AUC/MIC) associated with net bacterial stasis at MIC equal to or lower than 0.5 µg/mL across different levels of renal function except the patients with severe renal impairment [redacted] (b) (4) (Tables 3.3.5-2 and 3.3.5-3). Similarly for *E. coli* and *P. aeruginosa,* more than 90% of patients would achieve the respective PK/PD target for stasis at MICs equal to or lower than the respective proposed breakpoint, 0.25 and 0.5 µg/mL (See APPENDIX 4.4).

Table 3.3.4-2 Percent probabilities of PK-PD target attainment by MIC for IV delafloxacin using the Applicant's proposed dosing regimen. Results are shown for net bacterial stasis and 1-$\log_{10}$ CFU reduction based on *f*AUC_{24}:MIC for *S. aureus* among simulated patients

MIC	*Normal renal function*		*Mild renal impairment*		*Moderate renal impairment*		*Severe renal impairment*		[redacted] (b) (4)
	Stasis	*1-log*	*Stasis*	*1-log*	*Stasis*	*1-log*	*Stasis*	*1-log*	
0.004	*1*	*1*	*1*	*1*	*1*	*1*	*1*	*1*	
0.008	*1*	*1*	*1*	*1*	*1*	*1*	*1*	*1*	
0.015	*1*	*1*	*1*	*1*	*1*	*1*	*1*	*1*	
0.03	*1*	*1*	*1*	*1*	*1*	*1*	*1*	*1*	
0.06	*1*	*1*	*1*	*1*	*1*	*1*	*1*	*1*	
0.12	*1*	*1*	*1*	*1*	*1*	*1*	*1*	*1*	
0.25	*1*	*0.99*	*1*	*1*	*1*	*1*	*1*	*0.99*	
0.5	*0.93*	*0.56*	*0.97*	*0.68*	*0.99*	*0.82*	*0.92*	*0.53*	
1	*0.26*	*0.03*	*0.38*	*0.07*	*0.54*	*0.13*	*0.26*	*0.04*	
2	*0.01*	*0*	*0.01*	*0*	*0.04*	*0*	*0.01*	*0*	
4	*0*	*0*	*0*	*0*	*0*	*0*	*0*	*0*	

Table 3.3.4-3 Percent probabilities of PK-PD target attainment by MIC for PO delafloxacin using the Applicant's proposed dosing regimen. Results are shown for net bacterial stasis and 1-$\log_{10}$ CFU reduction based on *f*AUC_{24}:MIC for *S. aureus* among simulated patients

MIC	*Normal renal function*		*Mild renal impairment*		*Moderate renal impairment*		*Severe renal impairment*		[redacted] (b) (4)
	Stasis	*1-log*	*Stasis*	*1-log*	*Stasis*	*1-log*	*Stasis*	*1-log*	
0.004	*1*	*1*	*1*	*1*	*1*	*1*	*1*	*1*	
0.008	*1*	*1*	*1*	*1*	*1*	*1*	*1*	*1*	
0.015	*1*	*1*	*1*	*1*	*1*	*1*	*1*	*1*	
0.03	*1*	*1*	*1*	*1*	*1*	*1*	*1*	*1*	
0.06	*1*	*1*	*1*	*1*	*1*	*1*	*1*	*1*	
0.12	*1*	*1*	*1*	*1*	*1*	*1*	*1*	*1*	
0.25	*1*	*0.98*	*1*	*0.99*	*1*	*1*	*0.99*	*0.88*	
0.5	*0.92*	*0.54*	*0.95*	*0.66*	*0.97*	*0.77*	*0.65*	*0.13*	
1	*0.24*	*0.01*	*0.31*	*0.03*	*0.47*	*0.06*	*0.02*	*0*	
2	*0*	*0*	*0*	*0*	*0.01*	*0*	*0*	*0*	
4	*0*	*0*	*0*	*0*	*0*	*0*	*0*	*0*	

For comparison, additional PTA analyses were conducted for PO delafloxacin without dose adjustment in patients with severe renal impairment and the results for *S. aureus* are listed in Table 4. The results show that 450 mg Q12h administered to patients with severe renal impairment would result in a PTA >90% at an MIC of 0.5 µg/mL for *S. aureus*. These results support the oral dose of 450 mg Q12h in patients with severe renal impairment.

Table 3.3.4-4 Percent probabilities of PK-PD target attainment by MIC for PO delafloxacin using dosing regimen without dose adjustment. Results are shown for net bacterial stasis and 1-$\log_{10}$ CFU reduction based on $fAUC_{24}$:MIC for *S. aureus* among simulated patients

MIC	*Normal renal function*		*Mild renal impairment*		*Moderate renal impairment*		*Severe renal impairment*		(b) (4)
	Stasis	*1-log*	*Stasis*	*1-log*	*Stasis*	*1-log*	*Stasis*	*1-log*	
0.004	1	1	1	1	1	1	1	1	
0.008	1	1	1	1	1	1	1	1	
0.015	1	1	1	1	1	1	1	1	
0.03	1	1	1	1	1	1	1	1	
0.06	1	1	1	1	1	1	1	1	
0.12	1	1	1	1	1	1	1	1	
0.25	1	0.98	1	0.99	1	1	1	1	
0.5	0.92	0.53	0.95	0.65	0.98	0.78	0.99	0.90	
1	0.21	0.01	0.30	0.02	0.49	0.06	0.67	0.18	
2	0	0	0	0	0	0	0.04	0	
4	0	0	0	0	0	0	0	0	

Regarding determination of susceptibility breakpoints , the MIC distributions in clinical trials and surveillance programs, nonclinical PK-PD target attainment data, and clinical data should all be taken into consideration. Please refer to the clinical microbiology review by Dr. Jalal Sheikh and clinical review by Dr. Caroline Jjingo for details on the breakpoints determination.

3 . Clinical isolates from two Phase 3 studies of *S. aureus*, *E. coli*, and *P. aeruginosa*

The distribution of *S. aureus* and Gram-negative pathogens from ABSSSI site or blood in Phase 3 Studies by geographic region are summarized in Table 3.3.4-5 and Table 3.3.4-6, respectively.

Table 3.3.4-5 Distribution of Gram-positive Target Pathogens From ABSSSI Site or Blood During Phase 3 Studies Overall and by Geographic Region (MITT) (Data derived from Table 172 in the Summary of Clinical Pharmacology)

MITT:	North America - n (%; n/N)		Europe - n (%; n/N)		Overall - n (%; n/N)	
Organism	**Delafloxacin (N = 359)**	**Vancomycin (aztreonam) (N = 358)**	**Delafloxacin (N = 139)**	**Vancomycin (aztreonam) (N = 144)**	**Delafloxacin (N = 518)**	**Vancomycin (aztreonam) (N = 524)**
Staphylococci						
S. aureus	246 (68.5)	250 (69.8)	60 (43.2)	61 (42.4)	319 (61.6)	324 (61.8)
MRSA	136 (37.9)	137 (38.3)	6 (4.3)	1 (0.7)	144 (27.8)	141 (26.9)
MSSA	113 (31.5)	113 (31.6)	54 (38.8)	60 (41.7)	178 (34.4)	183 (34.9)

Table 3.3.4-6 Distribution of Gram-negative Target Pathogens From ABSSSI Site or Blood During Phase 3 Studies Overall and by Geographic Region (MITT) (Adapted partially from Table 173 in the Summary of Clinical Pharmacology)

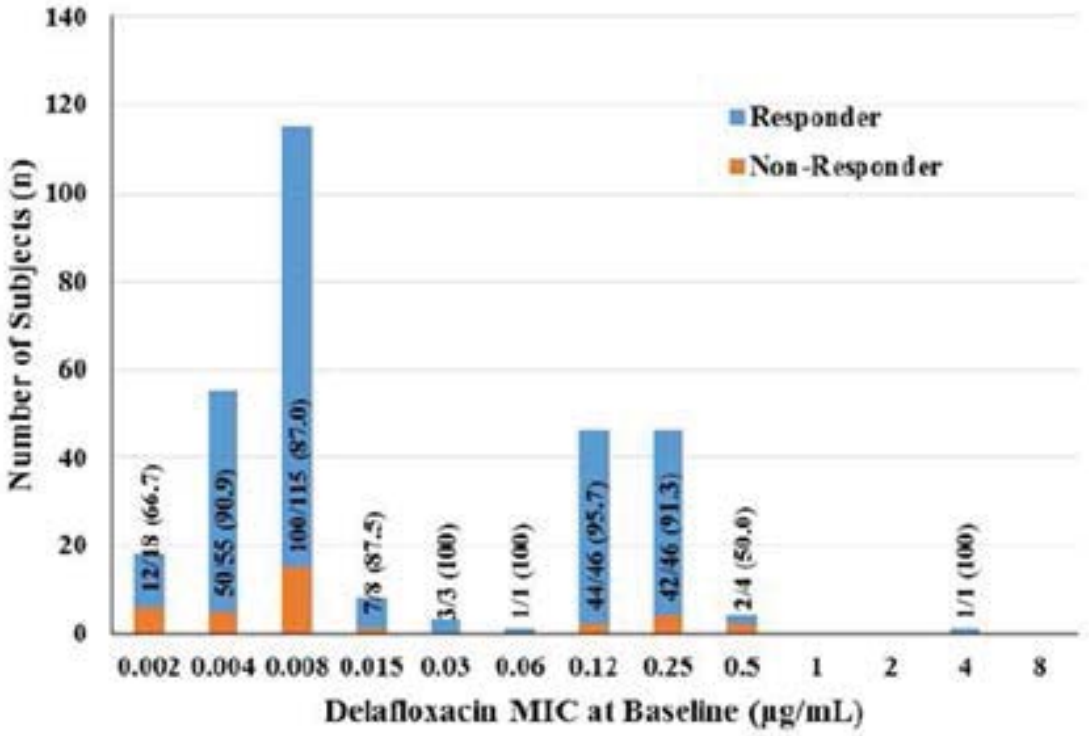
MITT:	North America - n (%; n/N)		Europe - n (%; n/N)		Overall - n (%; n/N)	
Organism	**Delafloxacin (N = 359)**	**Vancomycin (aztreonam) (N = 358)**	**Delafloxacin (N = 139)**	**Vancomycin (aztreonam) (N = 144)**	**Delafloxacin (N = 518)**	**Vancomycin (aztreonam) (N = 524)**
Enterobacteriaceae						
K. pneumoniae	18 (5.0)	20 (5.6)	4 (2.9)	3 (2.1)	22 (4.2)	23 (4.4)
E. coli	4 (1.1)	5 (1.4)	10 (7.2)	11 (7.6)	14 (2.7)	20 (3.8)
E. cloacae	10 (2.8)	3 (0.8)	4 (2.9)	8 (5.6)	14 (2.7)	11 (2.1)
P. mirabilis	2 (0.6)	2 (0.6)	6 (4.3)	6 (4.2)	8 (1.5)	8 (1.5)
K. oxytoca	4 (1.1)	1 (0.3)	2 (1.4)	4 (2.8)	6 (1.2)	5 (1.0)
Non-Enterobacteriaceae						
P. aeruginosa	6 (1.7)	4 (1.1)	4 (2.9)	8 (5.6)	11 (2.1)	12 (2.3)
H. parainfluenzae	6 (1.7)	7 (2.0)	0	0	6 (1.2)	7 (1.3)

ABSSSI = acute bacterial skin and skin structure infection; MITT = microbiological intent-to-treat; N = total number of patients; n = total number of patients with positive cultures at baseline from the primary infection site or blood for the indicated pathogen

As expected for ABSSSI, Gram-positive pathogens constituted the majority of pathogens recovered during the course of the Phase 3 studies. *S. aureus* was the most prevalent pathogen isolated from patients during the course of the Phase 3 studies. Overall, 61.6% of delafloxacin treated patients and 61.8% of vancomycin/aztreonam treated patients had *S. aureus* at baseline. Based on the microbiologically evaluable (ME) population and objective response at 48 to 72 hours (primary clinical endpoint), 88.2% (262/297) of delafloxacin-treated patients with *S. aureus* at baseline were responders; In addition, the MICs of most of patients (296/297) were ⩽ 0.5 µg/mL; based on microbiologic response at Follow-up (FU), *S. aureus* was eradicated or

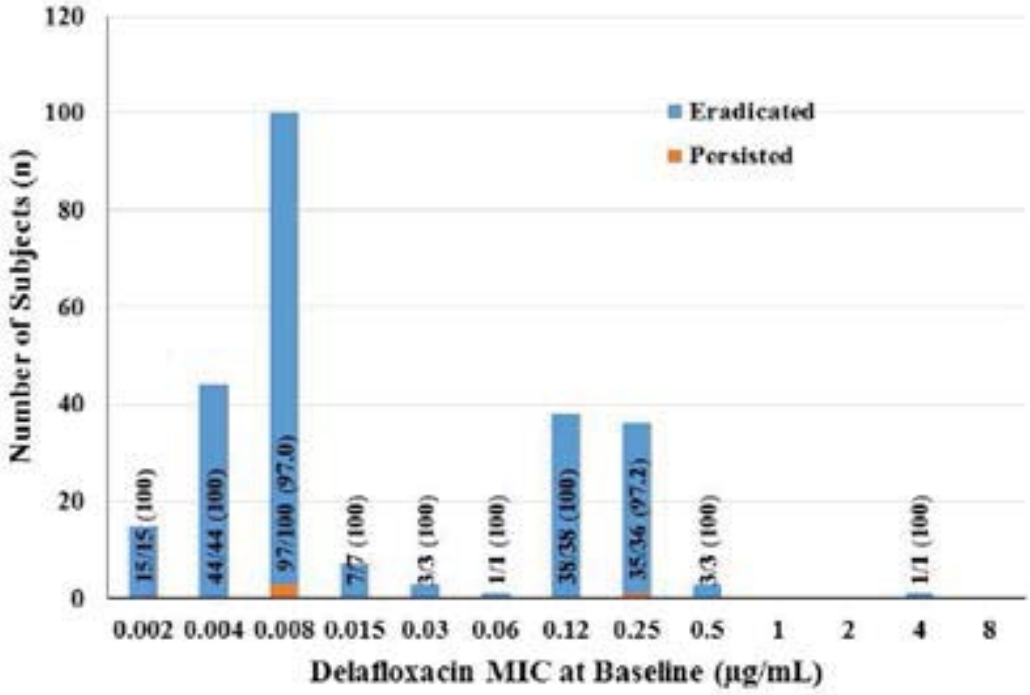
presumed eradicated in 98.4% (244/248) of patients; The MICs of most of these patients (microbiologic response at FU) (247/248) were ≤ 0.5 μg/mL. However, of note, only 4 clinical isolates (MIC= 0.5 μg/mL) were evaluated as objective response at 48-72 hours; 3 clinical isolates (MIC=0.5 μg/mL) were evaluated as microbiological response at follow-up visits. In addition, the objective response rate at MIC of 0.5 μg/mL at 48 to 72 hours was only 50%. The relationship of delafloxacin MIC value to objective response and microbiologic response for *S. aureus* in the ME population is summarized in Figure 3.3.4-1 and Figure 3.3.4-2, respectively.

Figure 3.3.4-1 Correlation of Delafloxacin MIC Against *S. aureus* to Objective Response at 48-72 hours (ME72O) (Adapted from Figure 80 in the Summary of Clinical Pharmacology)



ME72O = microbiologically evaluable at 48 to 72 hours (± 2 hours) for the objective response; MIC = minimum inhibitory concentration
Responder rate shown as number of subjects that were responders/total number of subjects and the corresponding percentage in parenthesis at each MIC

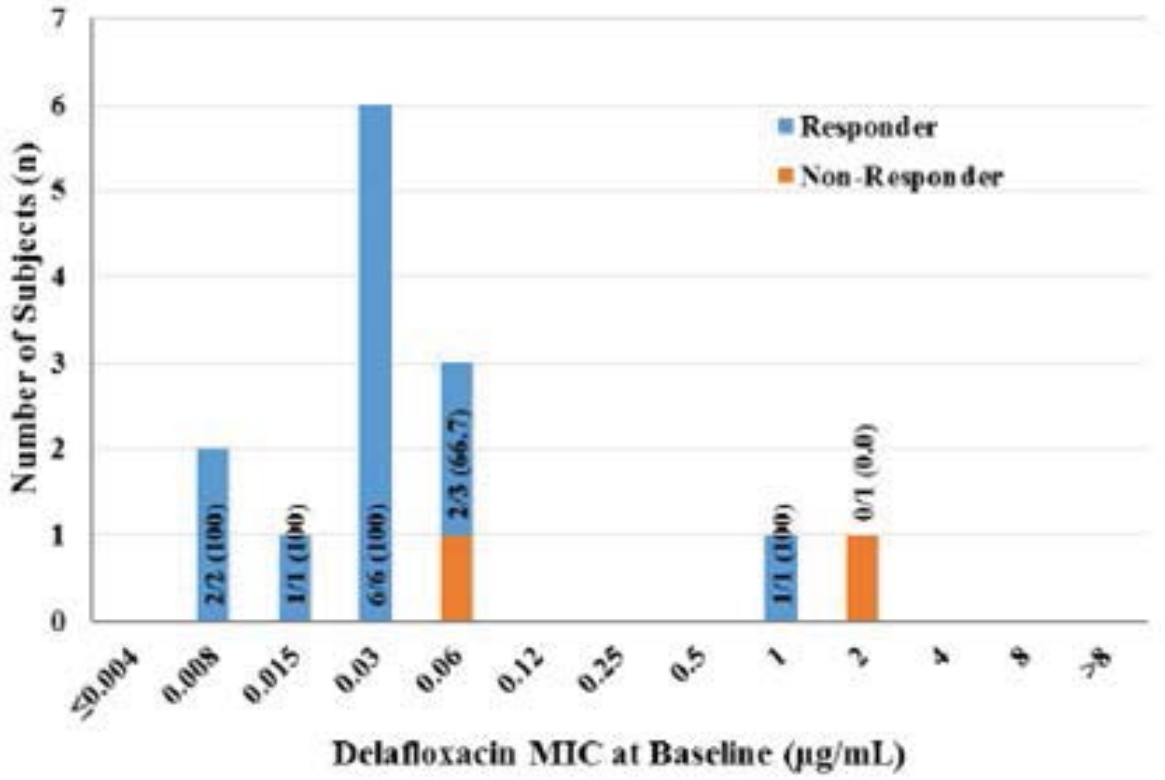
Figure 3.3.4-2 Correlation of Delafloxacin MIC Against *S. aureus* to Microbiologic Response at Follow-up (MEFUI)



MEFUI = microbiologically evaluable at follow-up for the investigator-assessed response; MIC = minimum inhibitory concentration
Microbiologic eradication rate shown as number of subjects that were microbiologically eradicated or presumed eradicated/total number of subjects and the corresponding percentage in parenthesis at each MIC

Only 14 clinical isolates of *E. coli* were identified in the ME delafloxacin treatment arm for the evaluation at 48 to 72 hours (primary clinical endpoint). Please refer to Figure 3.3.4-3 for the results.

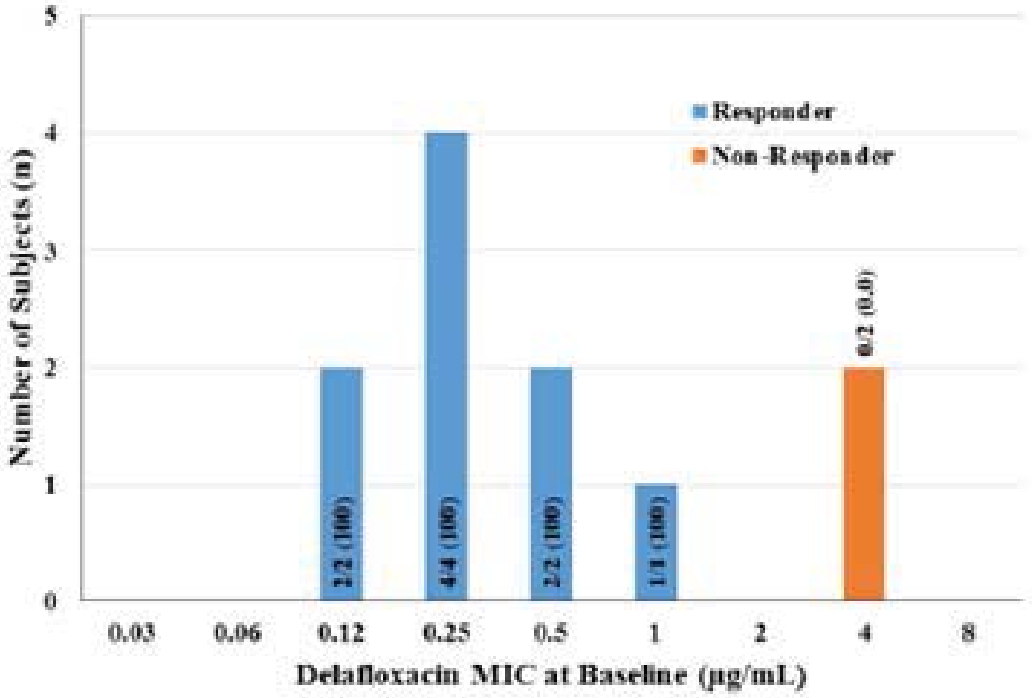
Figure 3.3.4-3 Correlation of Delafloxacin MIC Against *E. coli* to Objective Response at 48-72 hours (ME72O)



ME72O = microbiologically evaluable at 48 to 72 hours (± 2 hours) for the objective response; MIC = minimum inhibitory concentration
Responder rate shown as number of subjects that were responders/total number of subjects and the corresponding percentage in parenthesis at each MIC

Similar to *E. coli*, only 11 clinical isolate of *P. aeruginosa* were identified in ME delafloxacin treatment arm for the evaluation at 48 to 72 hours (primary clinical endpoint) in two Phase 3 studies. Please refer to Figure 3.3.4-4 for the results. Of note, the MICs of 5 out of 11 isolates were greater than or equal to 0.5 µg/mL.

Figure 3.3.4-4 Correlation of Delafloxacin MIC Against *P. aeruginosa* to Objective Response at 48-72 hours (ME72O)



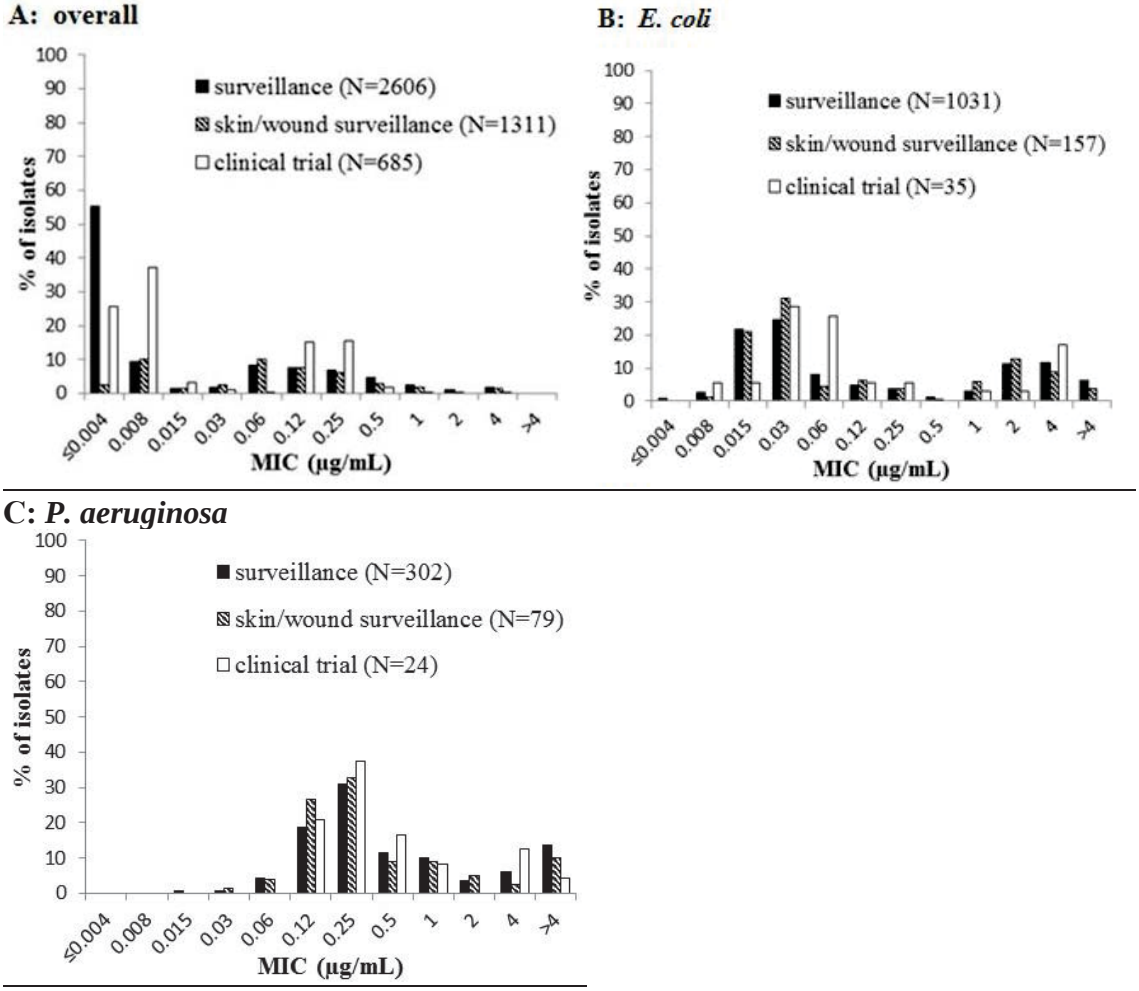
ME72O = microbiologically evaluable at 48 to 72 hours (± 2 hours) for the objective response; MIC = minimum inhibitory concentration
Responder rate shown as number of subjects that were responders/total number of subjects and the corresponding percentage in parenthesis at each MIC

In conclusion, the clinical data appear insufficient to support the breakpoint of [redacted] µg/mL for *S. aureus* based on the clinical response in microbiologically–evaluable patients. Due to the limited number of clinical isolates for *E coli* and *P. aeruginosa* in Phase 3 clinical studies, clinical evidence appears insufficient to determine the breakpoints for *E coli* and *P. aeruginosa*.

4. The MIC distribution in Phase 3 trials and surveillance programs for *S. aureus, E. coli* and *P. aeruginosa*

Overall, the delafloxacin MIC distribution as observed with clinical study isolates was similar to that observed during recent surveillance (2014-2015) for *S. aureus, E. coli* and *P. aeruginosa* (Figure 3.3.4-5). Based on the surveillance data provided by Applicant, the MIC_{90} for S. aureus to delafloxacin was 0.25 µg/mL [redacted] However, the MIC_{90} for *E.coli* and P. aeruginosa [redacted] than the proposed breakpoints 0.25 µg/mL for *E.coli* and 0.5 µg/mL for *P. aeruginosa*.

Figure 3.3.4-5. MIC Distribution of Delafloxacin Against (A) *S. aureus* (overall), (B) *E. coli*, and (C) *P. aeruginosa* During Phase 3 Clinical Studies and Recent Surveillance



Taken together, the Applicant's proposal of a susceptibility breakpoint of (b) (4) µg/mL for *S. aureus*, 0.25 µg/mL for *E. coli*, and 0.5 µg/mL for *P. aeruginosa*, is supported by the PTA analysis results. It should be noted that the determination of breakpoints involves multiple disciplines including clinical and microbiological perspectives in addition to the nonclinical and clinical PK-PD considerations. The ultimate determination of the delafloxacin breakpoint will depend on the totality of information provided by each discipline and continues to be assessed at the time of the completion of this review.

4. APPENDIX

4.1 Summary of Bioanalytical Method Validation and Performance

Multiple validated assays as listed below were used for quantification of delafloxacin and sulfobutyl-ether-β-cyclodextrin (SBECD) in various matrices. See Table 4.1-1 (delafloxacin in human plasma), Table 4.1-2 (delafloxacin urine and dialysate), and Table 4.1-3 (SBECD in human plasma, urine, and dialysate) for details.

- Validated LC/MS/MS methods to determine delafloxacin concentrations in human plasma and urine
- Validated LC-API/MS/MS methods to determine delafloxacin concentrations in human plasma, urine, and dialysate fluid
- Validated LC/MS/MS methods to determine sulfobutyl-ether-β-cyclodextrin (SBECD) concentrations in human plasma, urine, and dialysate fluid

When concentrations exceeded the standard curve range, samples were diluted, then assayed. Dilution integrity was verified within each clinical pharmacology study when sample dilutions were performed. The sample preparation, stability, analysis accuracy, and precision in each clinical pharmacology study were reported in its individual bioanalytical report and reviewed by the Clinical Pharmacology reviewer. Please refer to individual clinical pharmacology study report (See APPENDIX 4.5) for details.

Table 4.1-1 Summary of Analytical Methods for Quantification of Delafloxacin in Human Plasma (Adapted from Appendix 2 in the Summary of Biopharmaceutical and Associated AM)

Method						(b) (4)
Report No.	R&D/01/005 and Addendum A	(b) (4) 3341C and Addendum 1	(b) (4) 3341G	(b) (4) 3341F	OZZ2	(b) (4) 06411QB02
Method No.	Report No. 8	BP3341C	BP3341G	BP3341F	LCMSC634 and Version 1. 01 and 1.02, 1.03	NA
Method	LC-MS/MS	LC-API-MS/MS	LC-MS/MS	LC-API-MS/MS	LC-MS/MS	LC-MS/MS
Matrix	Plasma	Plasma	Plasma	Plasma and PBS	Plasma	Plasma
Clinical Studies Supported	M00-224 M01-284 M01-298 M01-301 M01-344 M01-365 M02-422 M02-463	RX-3341-101 RX-3341-102 RX-3341-103 RX-3341-104 RX-3341-106 RX-3341-108 RX-3341-109 RX-3341-110 RX-3341-111 RX-3341-201 RX-3341-202	RX-3341-113	RX-3341-110	ML-3341-112 ML-3341-118 RX-3341-114 RX-3341-115 RX-3341-116 RX-3341-302 RX-3341-303	RX-3341-107
Standard Curve Range (LLOQ, ULOQ)	7.7 – 5.366.3 µg/mL (7.7; 5366.3)	5.00 – 5000 ng/mL	0.005 – 2.50 µg/mL	0.00500 – 2.50µg/mL	5.00 – 5000 ng/mL	5.00 – 5000 ng/mL
QC Samples	17.68, 884.23, and 4421.17 ng/mL	15.0, 2000, and 4000 ng/mL	0.00500, 0.0150, 1.00, and 2.00 µg/mL	0.00500, 0.150, 1.00, and 2.00 µg/mL	5.00, 15.0, 40.0, 150, 600, and 3750 ng/mL	5.00, 12.5, 750, and 3500 ng/mL
Linearity	r = 0.9972 (range = 0.9966-0.9982)	r^2 = 0.9848-0.9993	r^2 > 0.9986-0.9997	r^2 > 0.9978-0.9992	Ave122rage r^2 = 0.9996 (0.9993-0.9998)	Typical curve r = 0.9954 (0.9944-0.9956)

Method						(b) (4)
Accuracy Intra-day Inter-day	For LLOQ 94.0% ND	For LLOQ 91.4%	For LLOQ 2.44 to -10.44%[a] -3.85%[a]	For LLOQ -2.00-12.4%[a] 5.22%[a]	For LLOQ -0.888-3.91%[a] 0.872%[a]	For LLOQ 6.2 - 17.8%[a] 12.2%[a]
Precision Intra-day Inter-day	For LLOQ 9.1% ND	For LLOQ 5.13%	For LLOQ 1.31-6.35% 7.56%	For LLOQ 5.09-5.16% 8.68%	For LLOQ 3.36-5.36% 4.49%	For LLOQ 9.7 -19.8% 15.1%
Freeze-thaw stability	3 cycles	4 cycles	4 cycles	3 cycles	5 cycles	3 cycles
Table-top stability	15 hours at room temperature	40 hours at room temperature	122 hours at room temperature	122 hours at room temperature	24 hours at room temperature	4 hours at room temperature
Long-term stability	634 days at -20°C	374 days at -20°C	374 days at -20°C	217 days at -20 C	150 days at -20°C	35 days at -20°C

LC-API-MS/MS = liquid chromatography atmospheric pressure ionization tandem mass spectrometry; LC-MS/MS = liquid chromatography tandem mass spectrometry; NA = not applicable; ND = not determined; PBS = phosphate-buffered saline

[a]Values are presented as percent deviation.

Table 4.1-2 Summary of Analytical Methods for Quantification of Delafloxacin in Human Urine or Dialysate(Adapted from Appendix 3 in the Summary of Biopharmaceutical and Associated Analytical Methods)

Method	[redacted]			
Report No.	R&D/01/005	(b) (4)3341D and Addendum 1	(b) (4)3341H	(b) (4)3341E
Method No.	Report No. 8	BP3341D	BP3341H	BP3341E
Method	LC-MS/MS	LC-API-MS/MS	LC-API-MS/MS	LC-MS/MS
Matrix	Urine	Urine	Urine	Dialysate
Clinical Studies Supported	M00-224	RX-3341-101 RX-3341-102 RX-3341-103 RX-3341-104 RX-3341-106 RX-3341-108 RX-3341-109	RX-3341-110	RX-3341-110
Standard Curve Range (LLOQ, ULOQ)	7.7 – 5366.3 ng/mL (7.7; 5366.3)	50.0 – 10000 ng/mL	50.0 – 10000 ng/mL	0.00500 – 2.50 µg/mL
QC Samples	22.06, 551.41, and 4595.09 ng/mL	150, 4000, and 8000 ng/mL	50, 150, 4000, 8000, 20000 ng/mL	0.00500, 0.0150, 2.00, and 10.0 µg/mL
Linearity		r^2 = 0.9926 – 0.9981	r^2 = 0.9990 – 0.9996	r^2 = 0.9944-0.9992
Accuracy Intra-day Inter-day	For LLOQ ND	For LLOQ 94.5% ND	For LLOQ[a] 10.8 to 19.4%[a] 15.5%[a]	For LLOQ[a] 4.53 to 10.8% 7.91%[a]
Precision Intra-day Inter-day	For LLOQ ND	For LLOQ 18.4% ND	For LLOQ 1.49 to 2.03%[a] 4.26%[a]	For LLOQ 2.42% to 12.2% 7.61%
Freeze-thaw stability	4 cycles	4 cycles		4 cycles
Table-top stability	20 hours at room temperature	22 hours		26 hours
Long-term stability	98 days at -20°C	168 days at -20°C; 229 days at -70°C	743 days at –70°C	31 days at -20°C

LC-API-MS/MS = liquid chromatography atmospheric pressure ionization tandem mass spectrometry; LC-MS/MS = liquid chromatograph tandem mass spectrometry; NA = not applicable; ND = not determined

[a]Values are presented as percent deviation.

Table 4.1-3 Summary of Analytical Methods for Quantification of Sulfobutyl-Ether-β-Cyclodextrin in Human Plasma,Urine, and Dialysate(Adapted from Appendix 4 in the Summary of Biopharmaceutical and Associated Analytical Methods)

Method	(b) (4)		
Report No.	(b) (4) 64743, Amendments 1 and 2	(b) (4) 67598	(b) (4) 67600 and Amendment 1
Method	LC-MS/MS	LC- MS/MS	LC-MS/MS
Matrix	Plasma	Urine	Dialysate
Clinical Studies Supported	RX-3341-110	RX-3341-110	RX-3341-110
Standard Curve Range (LLOQ)	2.00 – 200 µg/mL (2.00 µg/mL)	5.00 – 200 µg/mL (5.00 µg/mL)	2.00 to 200 µg/mL
QC Samples	7.50, 80.0, and 190 µg/mL	6.00, 40.0, and 150 µg/mL	6.00, 40.0, and 150 µg/mL
Linearity	$r^2 \geq 0.9993$-0.9971	$r^2 = 0.9987$ and-0.9992.	$r^2 \geq 0.990$
Accuracy Intra-day Inter-day	For LLOQ 84.7 -104% 93.7%	For LLOQ 97.5%	For LLOQ 95.9%
Precision Intra-day Inter-day	For LLOQ 16.9% 15.3	For LLOQ 9.3%	For LLOQ 8.1%
Freeze-thaw stability	4 cycles	3 cycles	3 cycles
Table-top stability	3.5 hours	8 hours	5 hours
Long-term stability	383 days at ≤ -70°C 129 days at – 20 C	190 days at ≤ -70°C	185 days at ≤ -70°C

LC-MS/MS = liquid chromatograph tandem mass spectrometry; NA = not applicable; ND = not determined

4.2 Population PK Analysis

Two population PK models were developed separately by Applicant to describe the PK of delafloxacin: one model was solely based on intravenous (IV) administration with a wide range of doses (from 300 mg to 1200 mg) in healthy volunteers and patients and another model based on oral (PO) doses ranging from 400 to 900 mg plus an IV dose of 300 mg (all in healthy volunteers). The two population PK models utilized a similar model structure (three-compartment model with mixed linear and non-linear elimination) except that a parallel immediate and delayed absorption was included for PO administration. As a combined model (IV data in healthy volunteers and patients combined with PO data in healthy volunteers) was not available or developed, the exposures of delafloxacin administered by IV and PO were simulated using the appropriate population PK model.

4.2.1 Population PK analysis for delafloxacin with IV administration only

A population PK model for IV delafloxacin was developed using pooled data from healthy subjects and patients enrolled in Studies RX-3341-104 (Study 104), RX-3341-108 (Study 108), RX-3341-110 (Study 110), RX-3341-111 (Study 111), RX-3341-202 (Study 202), RX-3341-302 (Study 302), and RX-3341-303 (Study 303).

Study 104: The study was conducted to determine the PK of two IV formulations of delafloxacin. A total of 24 healthy male and female subjects were enrolled. Subjects received single or multiple 1-hour IV infusions of delafloxacin 300 mg. Intensive blood and urine samples were collected for PK evaluation (N=20).

Study 108: This study was conducted to determine the relative PK of two IV formulations of delafloxacin, as well as the bioavailability of an oral formulation relative to an IV formulation, and to determine the maximum tolerated dose of delafloxacin. A total of 62 healthy male and female subjects were enrolled. Subjects received single IV infusions of delafloxacin 300, 450, 600, 750, 900, or 1200 mg and a single oral dose of delafloxacin 300 mg. Intensive blood and urine samples were collected for PK evaluation (N=40).

Study 110: This study was conducted to assess the PK of delafloxacin in healthy subjects with normal and varying degrees of renal insufficiencies. Subjects received single oral doses of 400 mg, single IV doses of 300 mg infusions. An additional treatment group included patients with end-stage renal disease (ESRD) who received 300 mg delafloxacin IV infusion approximately 1 hour prior to initiation of dialysis, a 300 mg delafloxacin infusion approximately 1 hour after the completion of dialysis. Intensive blood and urine samples were collected for PK evaluation. For patients with ESRD, only the data obtained when drug was administered after dialysis were included (N=34).

Study 111: This study was conducted to assess the electrocardiogram effects of IV delafloxacin following single dose administration of 300 and 900 mg IV delafloxacin, as well as to evaluate

the safety and PK of delafloxacin. A total of 52 healthy male and female subjects were enrolled. Intensive blood samples were collected for PK evaluation (N=52).

Study 202: A total of 256 adult male and female subjects who were diagnosed with ABSSSI were enrolled and randomized 1:1:1 to receive IV therapy with either delafloxacin 300 mg every Q12h, linezolid 600 mg Q12h, or vancomycin 15 mg/kg (based on actual body weight) Q12h. Treatment continued for 5 to 14 days. Blood samples for PK evaluations were collected from all patients on Day 3 at pre-dose and at 1, 2, 3, 5, and 12 hours after the start of the infusion (N=75).

Study 302: A total of 660 male and female subjects who were diagnosed with ABSSSI were enrolled and randomized 1:1 to receive either delafloxacin 300 mg IV Q12h or vancomycin 15 mg/kg IV Q12h based on actual body weight. Duration of all treatment was 10 to 28 doses. Blood samples for PK evaluations were collected at select sites on Day 3 at pre-dose and at 1, 2, 3, and 5 hours after the start of the infusion (N=227).

Study 303: Approximately 850 male and female subjects who were diagnosed with ABSSSI were enrolled and randomized 1:1 to receive either delafloxacin 300 mg IV Q12h or vancomycin 15 mg/kg IV Q12h. Duration of all treatment was 10 to 28 doses. Blood samples for PK evaluations were collected at select sites on Day 3 at pre-dose and at 1.5 and 3 hours after the start of the infusion (N=110).

Table 4.2.1-1 Summary statistics or counts of the subject demographic characteristics of analysis population

Variable	Phase 1 studies		Phase 2 study	Phase 3 studies	Total	
	N = 146[a]	N = 154[b]	N = 75	N = 322	N = 543[a]	N = 551[b]
	Median (Min. – Max.)	Median (Min. – Max.)	Median (Min. – Max.)	Median (Min. – Max.)	Median (Min. – Max.)	Median (Min. – Max.)
Age (yr)	33.5 (18.0 – 70.0)	34.0 (18.0 – 71.0)	36.0 (19.0 – 81.0)	46.5 (18.0 – 88.0)	43.0 (18.0 – 88.0)	43.0 (18.0 – 88.0)
Weight (kg)	74.6 (44.3 – 140)	74.9 (44.3 – 140)	86.5 (42.0 – 133)	80.9 (41.9 – 170)	80.0 (41.9 – 170)	80.0 (41.9 – 170)
BSA (m^2)	1.90 (1.40 – 2.60)	1.87 (1.35 – 2.60)	2.10 (1.40 – 2.60)	1.93 (1.24 – 2.86)	1.93 (1.24 – 2.86)	1.93 (1.24 – 2.86)
BMI (kg/m^2)	26.4 (19.2 – 40.3)	26.5 (18.4 – 40.3)	28.3 (15.4 – 49.3)	28.0 (15.0 – 59.0)	27.0 (15.0 – 59.0)	27.1 (15.0 – 59.0)
CLcr (mL/min/1.73 m^2)	90.7 (9.70 – 160)	88.9 (4.00 – 160)	116 (55.8 – 244)	89.9 (21.0 – 223)	93.6 (9.70 – 244)	93.2 (4.00 – 244)
Gender						
Male	82 (56%)	87 (56%)	44 (59%)	199 (62%)	325 (60%)	330 (60%)
Female	64 (44%)	67 (44%)	31 (41%)	123 (38%)	218 (40%)	221 (40%)
Race						
White	97 (66%)	99 (64%)	59 (79%)	286 (89%)	442 (81%)	444 (81%)
Black	46 (32%)	52 (34%)	8 (11%)	22 (7%)	76 (14%)	82 (15%)
Asian	1 (1%)	1 (1%)	0 (0%)	5 (2%)	6 (1%)	6 (1%)
Other	2 (1%)	2 (1%)	8 (11%)	9 (3%)	19 (3%)	19 (3%)

a. Model development population (excluding ESRD).
b. Final, pooled population (including ESRD).

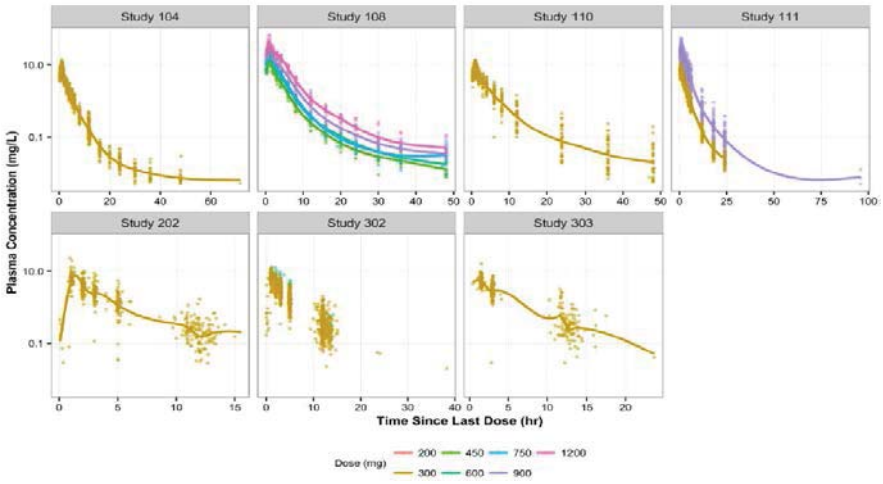
Source: Applicant's population PK report for IV delafloxacin, Page 35, Table 3

A total of 570 subjects and 5792 plasma concentration records were available from the seven studies used for population PK model development. A total of 96 subjects and 489 urine concentration records were available from three of the studies (104,108 and 110). ESRD patients

from Study 110 and urine samples from Study 110 were excluded during model development but were added to the pooled dataset after the covariate analysis. Summary statistics of baseline subject descriptors for the analysis population are presented in Table 4.2.1-1.

Semilog scatterplots of delafloxacin plasma concentrations versus time, stratified by study and dose, are provided in Figure 4.2.1-1. After IV administration, delafloxacin plasma concentrations appeared to decline in a poly-phasic manner. Exploratory analysis suggested that delafloxacin exposure increased more than dose-proportionally in healthy subjects after IV administration in Studies 104, 108, and 111, suggesting that delafloxacin exhibits nonlinear disposition in plasma.

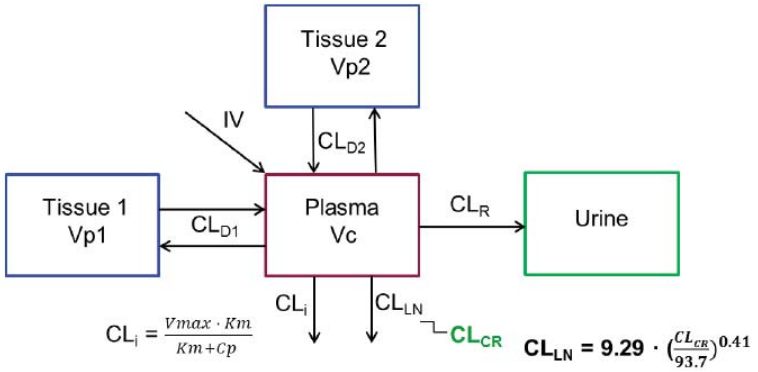
Figure 4.2.1-1 Semi-log scatterplots of delafloxacin plasma concentrations versus time, stratified by study, and dose



Source: Applicant's population PK report for IV delafloxacin, Page 38, Figure 4

A three-compartment PK model was utilized to describe delafloxacin pooled IV data in healthy subjects as well as patients with ABSSSI (Figure 4.2.1-2). A mixed linear (CL_{LN}) and nonlinear (CL_i; saturable) elimination combined with linear renal clearance (CL_R) was fit to the data with a good prediction.

Figure 4.2.1-2 Structural population PK model diagram

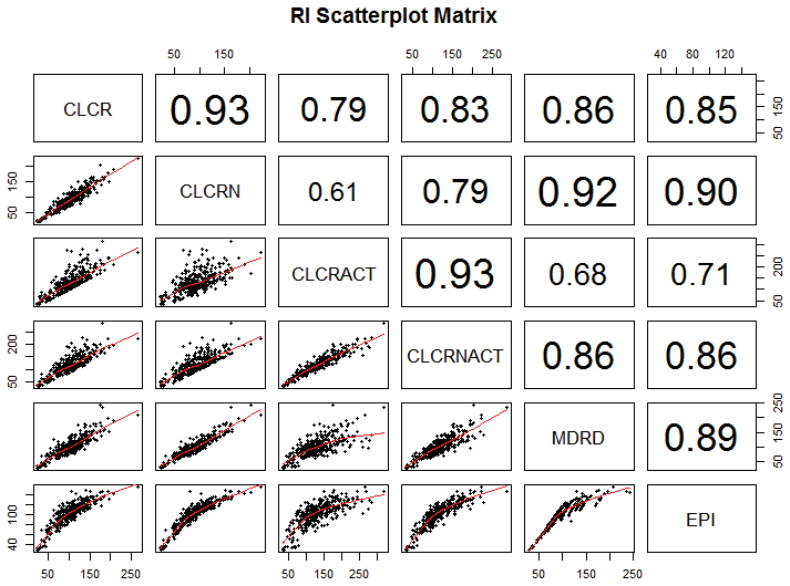


Source: Applicant's population PK report for IV delafloxacin, Page 39, Figure 5

The following subject demographics were evaluated as potential covariates of PK variability: 1) age; 2) gender; 3) height and/or weight; 4) race; and 5) creatinine clearance (CLcr). Subject CLcr was calculated from serum creatinine, age, and body weight using the Cockcroft and Gault equation (C-G) and then normalized to body surface area (BSA). For the sponsor's analysis, creatinine clearanace calculated by the C-G equation and normalized by BSA was used at it removes some of the colinearity between body weight and age for the unadjusted C-G equation. Note that the serum creatinine that was observed closest to the time of PK sampling was used for the calculation (i.e., not necessarily the baseline value, depending on timing of serum creatinine sampling). With regard to use of body weight for the calculation of CLcr, the lesser of ideal body weight (IBW) and actual body weight (WTKG) for a given subject was utilized.

Reviewer's comments: Subjects in the dedicated renal impairment study were grouped using eGFR (eGFR calculated by MDRD method), and the population PK model used CLCRN (CLcr calculated by C-G method using lesser of ideal body weight and actual body weight and normalized by BSA). The correlation between these two renal function parameters is high (coefficient~92%) as shown in Figure 4.2.1-3, suggesting the conclusion would be similar by using either method.

Figure 4.2.1-3 Correlation of all renal function parameters based on Phase 3 data (Study 302 and 303)



CLCR is creatinine clearance calculated by C-G using the lesser of ideal body weight and actual body weight; CLCRN is CLCR normalized by BSA; CLCRACT is creatinine clearance calculated by C-G using actual body weight; CLCRACTN is CLCRACT normalized by BSA; MDRD is eGFR calculated by MDRD; EPI is creatinine clearance calculated by CKD-EPI

Source: Reviewer's independent analysis

However, it should be noted that the correlation between CLCRACT (CLcr calculated by C-G method using actual body weight) and MDRD is relatively poor (~68%), while CLCRACT is the basis of dose recommendations in the labeling of drugs in the same class such as levofloxacin and moxifloxacin. This finding suggests that the covariate used in Applicant's analysis may be similar to that resulting from MDRD. However, in order to have the assessment performed using the same measure as was used in the dedicated renal impairment study, Reviewer reevaluated Applicant's population PK model using MDRD as the covariate for renal function.

Ultimately, five covariate-parameter relationships were added into the model: relationships between body weight and linear CL (CL_{LN}), volume of distribution (Vc), and volume of distribution for peripheral compartment 2 (Vp2), a relationship between sex and CL_{LN}, and a relationship between CLcr and CL_{LN}. The population PK parameter estimates and associated standard errors for the model are provided in Table 4.2.1-2.

Table 4.2.1-2 Final population PK model parameter estimates and standard errors

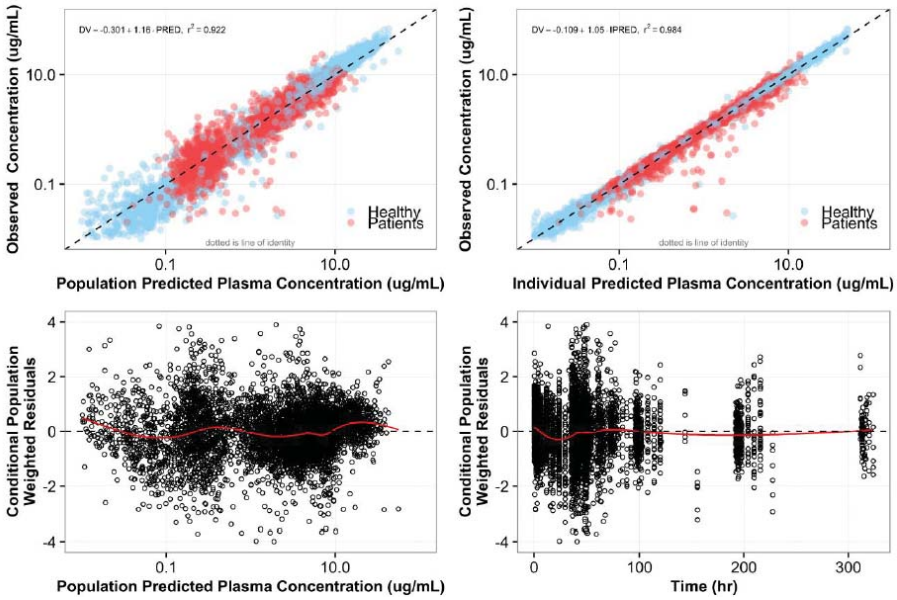
Parameter	Population mean		Magnitude of interindividual variability (%CV)	
	Estimate	% SEM	Estimate	%SEM
CL_{LN} (L/hr)	10.2	2.33	30.0	8.86
Vc (L)	18.4	2.98	38.9	8.34
Vmax (L/hr)	6.0	Fixed	45.4	17.7
Km (µg/mL)	3.0	Fixed	60.7	67.4
CL_{D1} (L/hr)	1.59	4.40	52.4	15.5
Vp1 (L)	17.5	3.89	55.8	14.7
CL_{D2} (L/hr)	24.9	3.08	NE	NA
Vp2 (L)	18.6	2.49	27.5	10.2
CL_R (L/hr)	4.12	6.67	45.9	16.3
Power coefficient of CLcr on CL_{LN}	0.45	8.64	NE	NA
Power coefficient of WT on CL_{LN}	0.57	13.5	NE	NA
Proportion of CL_{LN} for female	0.80	3.43	NE	NA
Power coefficient of WT on Vc	1.01	10.8	NE	NA
Power coefficient of WT on Vp2	0.69	12.1	NE	NA
Power coefficient of CLcr on CL_R	1.45	4.32		
Residual variability				
Plasma proportional error	0.026	1.12	NE	NA
Plasma additive error	0.00004	10.6	NE	NA
Urine proportional error	0.145	8.97	NE	NA

Source: Applicant's population PK report for IV delafloxacin, Page 45, Table 5

The goodness-of-fit plots for the final population PK model are provided in Figure 4.2.1-4. These plots demonstrate the adequacy of the model fit across healthy subjects and patients with ABSSSI. Additionally, the VPC plots of delafloxacin plasma concentrations after administration of two to seven doses of delafloxacin 300 mg IV Q12h are provided in Figure 4.2.1-5. As shown by these data, there was good agreement between simulated delafloxacin plasma concentrations

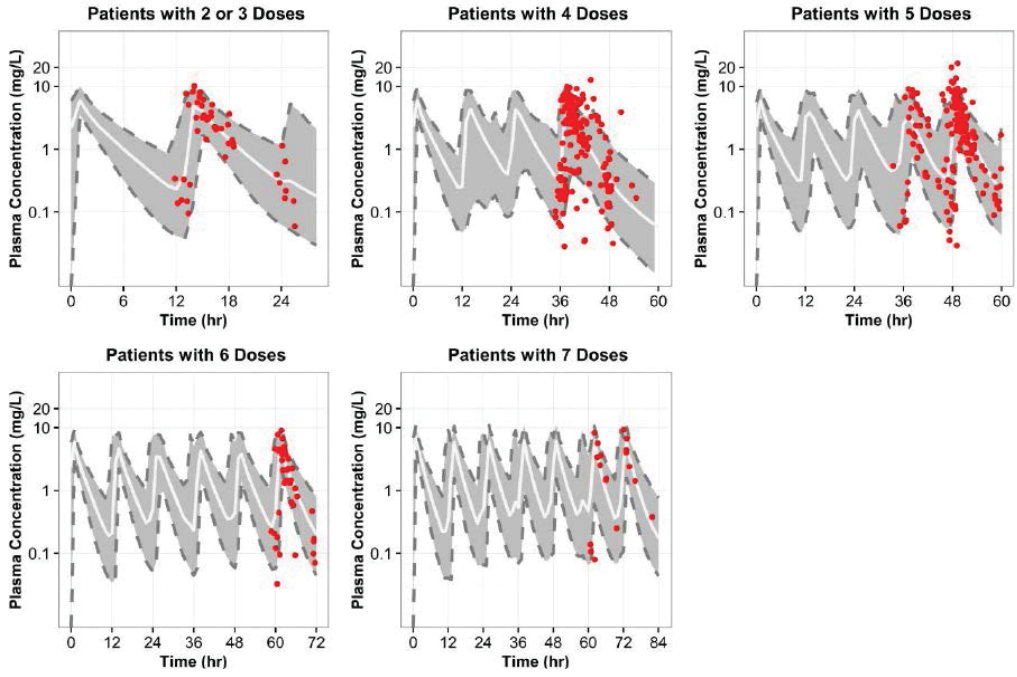
based on the final population PK model and observed delafloxacin plasma concentrations from patients with ABSSSI from Study 202 after multiple IV dosing.

Figure 4.2.1-4 Standard goodness-of-fit plots for the final population PK model



Source: Applicant's population PK report for IV delafloxacin, Page 47, Figure 8

Figure 4.2.1-5 Visual predictive check of delafloxacin in patients with ABSSSI from Study 202 following multiple IV doses stratified by number of doses received



Source: Applicant's population PK report for IV delafloxacin, Page 49, Figure 9

The impact of subject demographics on delafloxacin exposure was evaluated.

1) Renal impairment

The importance of renal impairment on delafloxacin was evaluated using the final population PK model and Monte Carlo simulation to predict exposure in a group of 7500 patients, 1500 in each renal impairment category. These summary statistics (Table 4.2.1-3) show that a dose of 200 mg IV Q12h in patients with severe renal impairment (b) (4) is expected to result in delafloxacin exposures that are comparable to those expected in patients with normal, mild or moderate renal impairment with a dose of 300 mg IV Q12h.

Table 4.2.1-3 Summary statistics [geometric mean (CV%)] for model-derived delafloxacin plasma exposure on treatment Day 3 using the Monte Carlo simulation stratified by renal impairment category

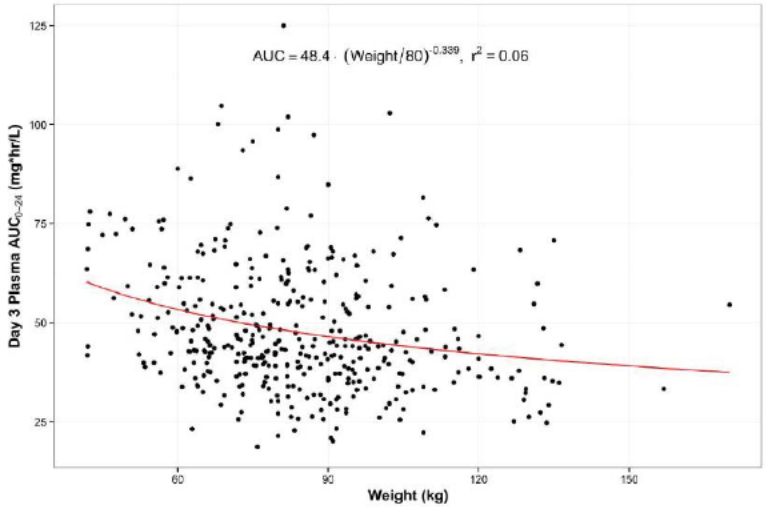
Renal function	N	Dose regimen	AUC_{0-24} (µg•hr/mL)	C_{max} (µg/mL)
Normal renal function	1500	300 mg IV Q12h for 3 days	39.7 (33.9)	7.26 (25.3)
Mild renal impairment	1500		50.1 (33.6)	7.83 (26.2)
Moderate renal impairment	1500		60.0 (33.6)	8.32 (25.8)
Severe renal impairment	1500	200 mg IV Q12h	46.8 (34.0)	5.83 (25.8)
(b) (4)				

Source: Applicant's population PK report for IV delafloxacin, Page 54, Table 7

2) Body weight

Body weight was identified as a significant covariate for CL_{LN}, Vc, and Vp2. The relationship between body weight and AUC_{24} on Day 3 (Figure 4.2.1-6) shows that, despite the trend for decreasing exposure with increasing body weight, the variability in PK results in a high degree of overlap across the weight, suggesting dose adjustment is not warranted based on body weight.

Table 4.2.1-6 Scatterplot of Bayesian post-hoc AUC0-24 on Day 3 versus body weight



Source: Applicant's population PK report for IV delafloxacin, Page 55, Figure 12

3) Sex

Patient sex was identified as a statistically significant predictor of the inter-subject variability (IIV) in delafloxacin CL_{LN}. Females are expected to have slightly higher exposure than males (Table 4.2.1-4). Based on the relatively small difference in exposures between males and females, dose adjustments are not needed based on gender.

Table 4.2.1-4 Summary statistics [geometric mean (CV%)] for model-derived delafloxacin plasma exposure on treatment Day 3 using Monte Carlo simulation stratified by sex

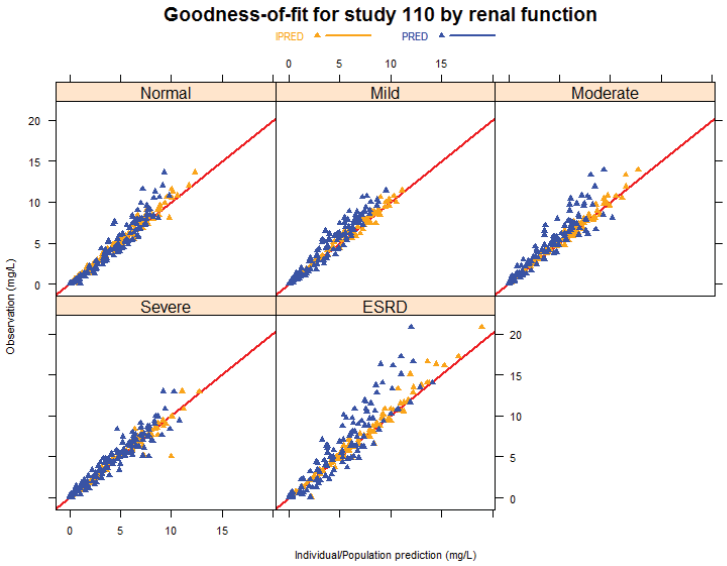
PK parameters	Sex	
	Male N = 4379	Female N = 3121
AUC_{0-24} (µg•hr/mL)	46.3 (37.0)	57.4 (37.5)
C_{max} (µg/mL)	6.73 (27.9)	7.58 (28.9)

Source: Applicant's population PK report for IV delafloxacin, Page 59, Table 9

To conclude, the developed model provided an adequate fit to the delafloxacin concentration-time data pooled from healthy volunteers and patients with ABSSSI after administration of IV delafloxacin.

Reviewer's comments: Although a high correlation between CLCRN and eGFR was identified, the reviewer conducted an independent population PK analysis for IV delafloxacin using eGFR calculated by MDRD method as a covariate instead of CLCRN calculated by C-G method. The parameter estimates were consistent with Applicant's result.

Figure 4.2.1-7 Diagnostic Plots for Study 110 by Renal Function

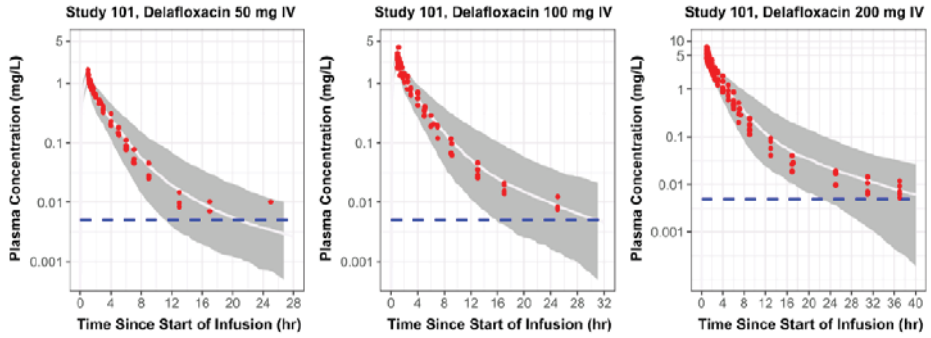


Source: Reviewer's independent analysis.

As renal function is a significant covariate for PK of IV delafloxacin and dose reduction was proposed for patients with severe renal impairment (b) (4) *the goodness-of-fit for PK profile in study RX-3341-110 (Dedicated renal impairment study) was independently evaluated (Figure 4.2.1-7) to ensure that the population PK model accurately describes exposures in patients with reduced renal function. The plots showed that the population PK model can generally describe the data and can be used to predict the exposure in patients with different levels of renal function. Note that the patients labeled as ESRD in Figure 4.2.1-7 are ESRD patients on dialysis who were administered drug within 1-hr of their most recent dialysis session. Throughout the rest of the review this group will be referred to as ESRD patients without hemodialysis.*

To ensure that the dose proposed by the Applicant (200 mg BID for IV delafloxacin) could be predicted by the model given the nonlinear clearance, the reviewer evaluated what doses were included in the Applicant's population PK analysis. However, in the population PK model, the lowest dose included was 300 mg IV. The reviewer identified PK data with lower single doses of 50, 100, and 200 mg IV from a Phase 1 study (RX-3341-101) and an IR letter was sent to the Applicant requesting them to update the population PK model by including those PK data. The Applicant performed an external validation based on data from Study RX-3341-101 to assess the population PK model's ability to describe exposures with these doses. The provided visual predictive check diagnostic plots shown below verify that the original population PK model can reasonably describe the PK data for doses of 50, 100, and 200 mg IV, respectively (Figure 4.2.1-8).

Figure 4.2.1-8 Visual Predictive Check Plots for IV Delafloxacin Administered as a Single Dose Ranging from 50 to 200 mg (Study RX-3341-101)



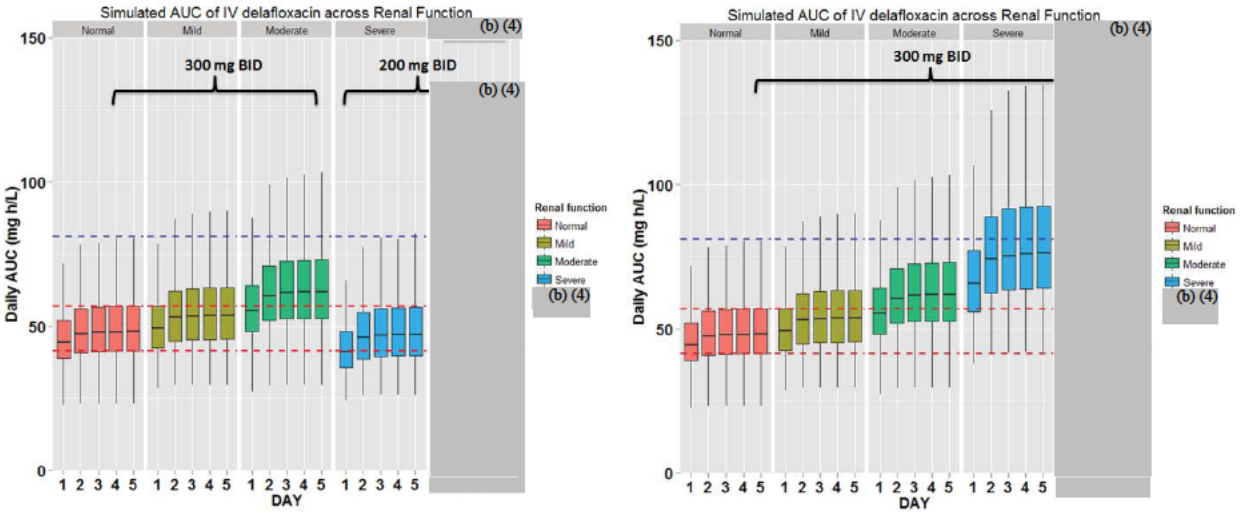
Source: Applicant's response to IR letter dated on Dec 15, 2016, Page 4, Figure 1

Based on the information above, the Reviewer simulated exposure on Day 1 to 5 for IV delafloxacin across different renal function categories based on the patient demographics in the two Phase 3 studies under two dosing regimens (i.e., proposed by Applicant and no dose reduction). The results illustrate that the proposed IV dosing regimen in patients with severe renal impairment (b) (4) *would result in comparable exposure to patients with normal renal function and mild or moderate renal impairment.* (b) (4)

(b) (4)

The simulated mean and 95% confidence intervals of AUC values are listed in Table 4.2.1-5.

Figure 4.2.1-9 Simulated Daily AUC from Day 1 to 5 for IV Delafloxacin across Renal Function under Two Dosing Regimens (Proposed by Applicant (Left) and no dose reduction (Right))



Blue dashed lines represent the mean daily AUC (81.1 mg·h/L) on Day 5 for 450 mg IV BID based on population PK model and demographics information in Study RX-3341-201; Red dashed lines represent the 25th and 75th percentile of daily AUC on Day 5 for 300 mg IV BID in normal renal function based on population PK model.

Source: Reviewer's independent analysis.

Table 4.2.1-5 Summary of Simulated Exposure of IV delafloxacin across Renal Function based on Population PK Model

Parameter eGFR (mL/min/1.73m^2)	*Proposed dosing by Applicant*		*No dose reduction*	
	Dose	*Daily AUCss(mg·h/L) (mean, 95% CI)*	*Dose*	*Daily AUC_{ss}(mg·h/L) (mean, 95% CI)*
Normal (⩾90)	*300 BID*	*50.1 (49.3-50.9)*	*300 BID*	*50.1 (49.3-50.9)*
Mild (60 (b) (4)	*300 BID*	*55.9 (54.9-56.9)*	*300 BID*	*55.9 (54.9-56.9)*
Moderate (30 (b) (4)	*300 BID*	*64.7 (63.6-65.8)*	*300 BID*	*64.7 (63.6-65.8)*
Severe (15- (b) (4)	*200 BID*	*49.7 (48.5-50.9)*	*300 BID*	*80.7 (78.7-82.8)*

(b) (4)

Source: Reviewer's independent analysis

Other than renal impairment, the impact of body weight and sex on exposure was evaluated in patients with normal renal function and is summarized in Table 4.2.1-6. In patients with normal renal function, neither body weight nor sex would result in significant differences on exposure and no dose adjustments are proposed based on these covariates.

Table 4.2.1-6 Summary of Simulated Exposure across Body Weight and Sex in Patients with Normal Renal Function based on Population PK model

Parameter	*Category/Quantile*	*Daily AUCss (mg·h/L) (mean, 95% CI)*
Sex	*Male*	*44.8 (44.2-45.5)*
	Female	*59.0 (57.7-60.4)*
Body Weight (kg)	*1st quantile (41.9-67.8)*	*58.1 (56.3-59.9)*
	2nd quantile (67.8-80.6)	*51.1 (49.7-52.6)*
	3rd quantile (80.6-94.0)	*48.5 (47.3-49.8)*
	4th quantile (94.0-156.9)	*42.7 (41.6-43.8)*

Source: Reviewer's independent analysis

On the other hand, sulfobutylether-β-cyclodextrin (SBECD), (b) (4) *was included in IV delafloxacin formulation. Administration of this compound, which is primarily renally eliminated, results in higher exposures in patients with reduced renal function. As the exposure of SBECD in mild and moderate renal impairment patients was around or less than 2-fold higher than that of healthy volunteers, no dose adjustment is needed. The dedicated renal impairment study shows that the AUC_{inf} of severe renal impairment group was 5-fold higher than that of healthy volunteers while the AUCinf of ESRD patients without hemodialysis was 30-fold higher. For patients with severe renal impairment, the total daily dose of SBECD to be delivered with a 300 mg Q12h IV delafloxacin infusion is 4800 mg, which is lower than that delivered with current products on the market (VFEND, 5760 mg/day with maintenance dose for a 60-kg person). It appears that 200 mg BID dosing would be acceptable for patients with severe renal impairment as one means to reduce SBECD exposure while still maintaining delafloxacin exposures at or exceeding that of patients with normal renal function. It is worth noting that in the VFEND label, IV formulation has cautionary language with regards to use in patients with moderate to severe renal impairment* (b) (4) *. For ESRD patients, whether with or without hemodialysis, the accumulation of SBECD would be a potential safety concern. Therefore, IV delafloxacin should be avoided in this population.*

4.2.2 Population PK analysis for delafloxacin with IV and PO administration

As no PK samples of PO delafloxacin were collected from ABSSSI patients, the population PK analysis for PO delafloxacin was developed based solely on PK data from healthy volunteers. The population PK data were obtained from four Phase 1 studies, Studies RX-3341-110 (Study 110), RX-3341-114 (Study 114), RX-3341-115 (Study 115) and RX-3341-116 (Study 116). A brief description of each study, as well as the subject populations, doses administered, and

sampling strategies are provided in Table 4.2.2-1. Study 110 was conducted in healthy subjects as well as subjects with renal impairment, while the other three studies were conducted in healthy volunteers. Study 116 employed PO administration under both fasted and fed conditions, and the other studies employed PO administration under fasted conditions. Delafloxacin doses from 300 to 900 mg were administrated in the studies.

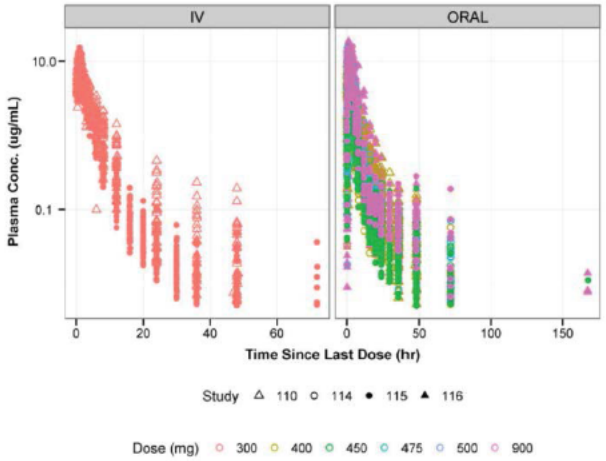
A total of 157 subjects and 6497 plasma concentration records were available from the four studies used for the population PK model development dataset. Summary statistics of baseline patient descriptors for the analysis population are presented in Table 4.2.2-1. The model development population was 51.6% female, included a range of renal function (CLCR (normalized by BSA) ranged from 17.2 to 175.4 mL/min/1.73 m^2) and a range of body weight from 51 to 140.4 kg.

Table 4.2.2-1Summary of studies included in the delafloxacin population PK analyses

Protocol number	Study design	Dose regimens	Scheduled plasma PK samples collection times
RX3341-110	Randomized, placebo-controlled, crossover, single dose study in healthy subjects or subjects with renal impairment	IV arm: 300 mg 1-hr infusion PO arm: 400 mg orally administrated	IV arm: pre-dose and 0.33, 066, 1, 1.083, 1.167, 1.33, 1.5, 2, 3, 4, 6, 8, 12, 24, 36, and 48 hr post-dose PO arm: pre-dose and 0.5, 1, 1.5, 2, 3, 4, 6, 8, 12, 16, 20, 24, 30, 36, and 48 hr post-dose
RX3341-114	Randomized, crossover, single dose study in healthy volunteers	400, 450, 500 mg of (b) (4) layer tablets or 475 mg (b) (4) tablets orally administrated	Pre-dose and 0.25, 0.5, 0.75, 1, 1.5, 2, 3, 4, 6, 8, 12, 16, 20, 24, 36, and 48 hr post-dose on Days 1, 4, 7, and 10
RX3341-115	Part 1: randomized, 2-period, 2-sequence, crossover, single dose study in healthy subjects Part 2: randomized, 1-period single dose study in healthy subjects	Part 1: 300 mg IV 1-hr infusion or 450 mg orally administrated Part 2: 900 mg orally administrated	Pre-dose and 0.25, 0.5, 0.75, 1, 1.25, 1.5, 2, 3, 4, 6, 8, 12, 16, 20, 24, 30, 36, 48, and 72 hr post-dose
RX3341-116	Randomized, 3-period, 6-sequence, crossover, single dose study in healthy subjects	900 mg orally administrated under fasting or fed conditions	Pre-dose and 0.25, 0.5, 0.75, 1, 1.25, 1.5, 2, 2.5, 3, 3.5, 4, 6, 8, 12, 16, 20, 24, 36, and 48 hr post-dose in Periods 1, 2, and 3

Source: Applicant's population PK report for PO delafloxacin, Page 20, Table 2

Figure 4.2.2-1 Semilog scatterplots of delafloxacin plasma concentrations versus time since start of last dose, stratified by route of administration and dose



Source: Applicant's population PK report for PO delafloxacin, Page 31, Figure 3

Semilog scatterplots of delafloxacin plasma concentrations versus time since last dose stratified by the route of administration and dose are provided in Figure 4.2.2-1. In general, delafloxacin plasma concentrations appeared to decline in a poly-phasic manner following completion of one-hour IV infusion.

Subject demographic and disease characteristics collected prior to administration of study drug were used to characterize the analysis population and to evaluate their ability to explain a portion of the IIV for selected PK parameters.

Table 4.2.2-2 Summary statistics or counts of the subject demographic characteristics of analysis population

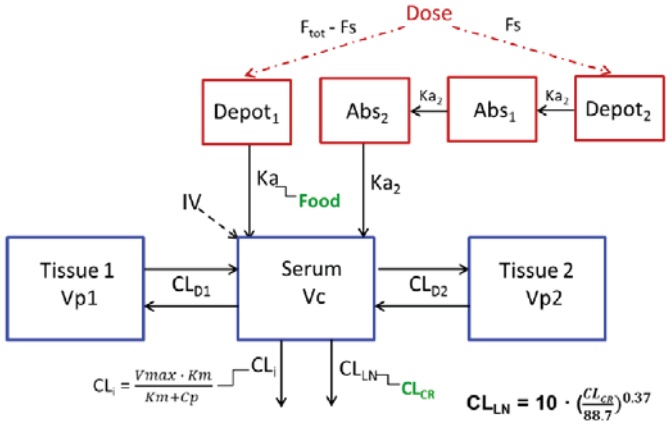
Variable	N	Mean	Median	Minimum	Maximum
Age (yr)	157	39.6	39	19	70
Weight (kg)	157	76.9	74.8	51	140.4
Height (cm)	157	168.5	168.7	147.5	198.1
BSA (m^2)	157	1.9	1.9	1.4	2.6
BMI (kg/m^2)	157	26.9	26.8	18.7	40.3
CLCR (mL/min/1.73 m^2)	157	104.7	105.6	17.2	175.4
Gender					
Male	76 (48.4%)	—	—	—	—
Female	81 (51.6%)	—	—	—	—

Source: Applicant's population PK report for PO delafloxacin, Page 28, Table 4

Reviewer's comment: As eGFR was used in the dedicated renal impairment study, Reviewer repeated the population PK analysis by using eGFR instead of CLcr. In addition, the eGFR was calculated using serum creatinine concentration closest to the time of PK sampling to keep the consistency with the population PK data for IV delafloxacin.

The final population PK model was a three-compartment model with mixed linear and nonlinear (saturable) elimination, two parallel first-order absorption processes, and an absorption delay for the second absorption process occurring through multiple transit compartments (Figure 4.2.2-2).

Figure 4.2.2-2 Diagram of the structural PK model diagram



Source: Applicant's population PK report for PO delafloxacin, Page 37, Figure 7

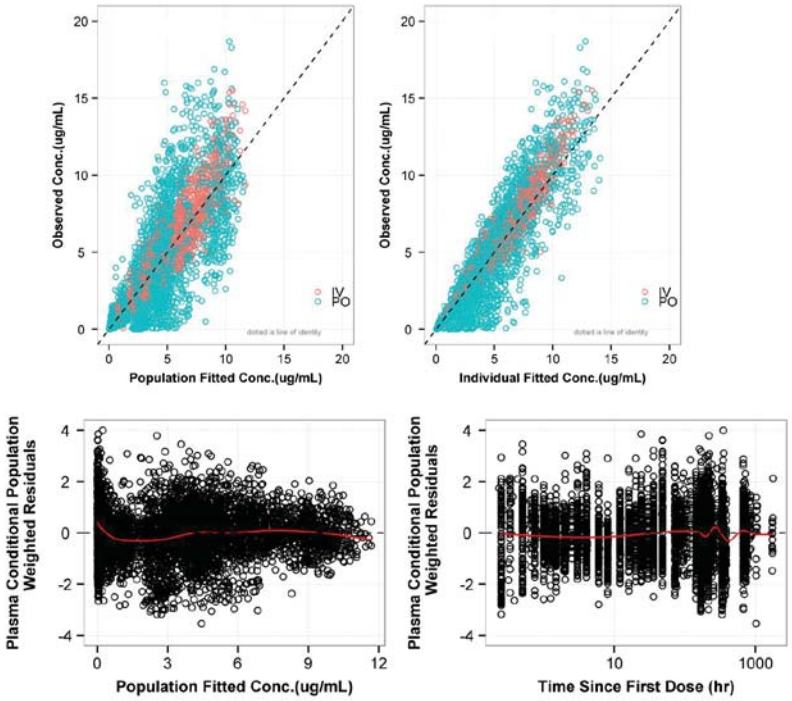
The relationships between CLcr and CL_{LN}, and WTKG and Vc were described using power functions. The population PK parameter estimates and associated standard errors for the model are provided in Table 4.2.2-3.

Table 4.2.2-3 Final population PK model parameter estimates and standard errors

Parameter	Population mean		Magnitude of interindividual variability (%CV)	
	Final estimate	%SEM	Final estimate	%SEM
CL_{LN} (L/hr)	9.84	2.89	23.5	14.5
VC (L)	17.5	3.42	13.4	52.2
Vmax (L/hr)	4.16	Fixed	14.5	Fixed
Km (µg/mL)	0.96	Fixed	19.5	Fixed
CL_{D1} (L/hr)	16.1	12.4	43.6	52.6
Vp1 (L)	12.3	6.52	34.6	24.5
CL_{D2} (L/hr)	1.18	4.83	37.4	27.9
Vp2 (L)	17.0	5.98	48.0	23.0
$K_{a\ (fasted)}$ (hr^{-1})	0.88	6.31	52.0	13.3
$K_{a\ (fed)}$ (hr^{-1})	0.41	5.88	52.0	13.3
Ka_2 (hr^{-1})	0.06	6.81	52.0	13.3
F_{TOT}	0.64	2.15	38.7	33.3
FS	0.06	8.64	76.2	20.7
Coefficient of power relationship between CLcr and CL_{LN}	0.35	13.9	NE	NA
Coefficient of power relationship between WTKG and Vc	1.04	12.8	NE	NA
Inter-occasion variability of F_{TOT} (%)	37.4	20.5	NE	NA
Residue variability				
Proportional error	0.09	0.99	NE	NA
Minimum value of the objective function = -8097				

Source: Applicant's population PK report for PO delafloxacin, Page 42, Table 7

Figure 4.2.2-3 Standard goodness-of-fit plots for the final population PK model

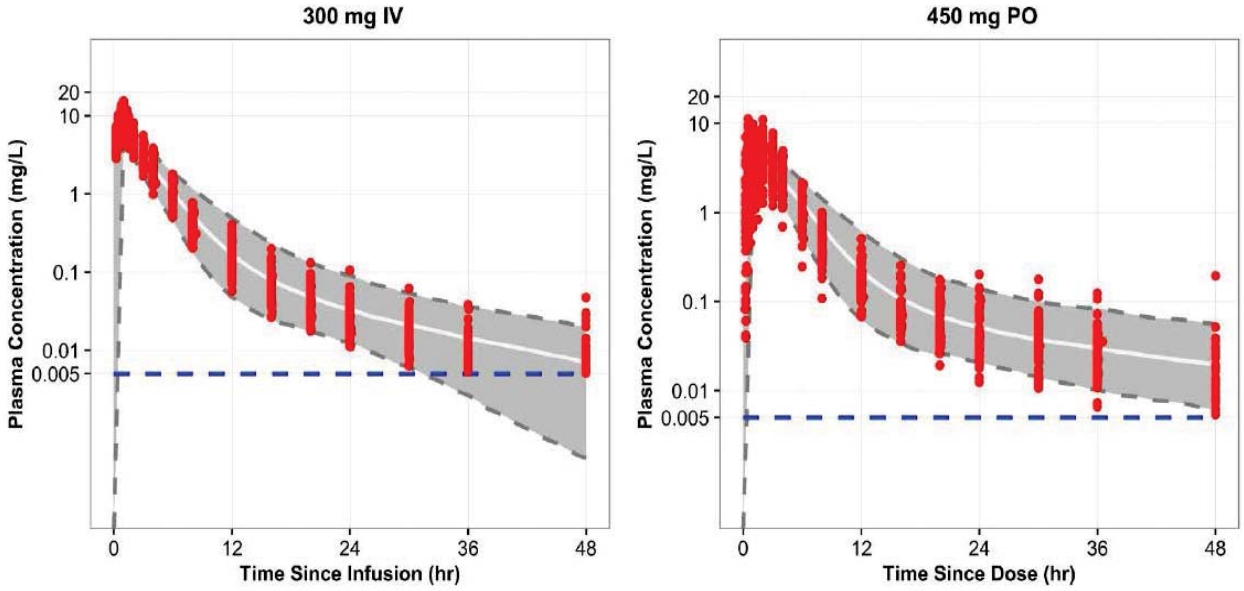


Source: Applicant's population PK report for PO delafloxacin, Page 44, Figure 10

The primary goodness-of-fit plots for the final population PK model are provided in Figure 4.2.2-3. These plots demonstrate the adequacy of the model fit across dosing regimens and routes of administration.

Additionally, the VPC plots of simulated and observed delafloxacin plasma concentrations after administration of delafloxacin 300 mg IV or 450 mg PO are provided in Figure 4.2.2-4. As shown by these data, there was good agreement between simulated delafloxacin plasma concentrations based on the final population PK model and observed delafloxacin plasma concentrations from healthy subjects from Study 115 after administration of single doses of delafloxacin 300 mg IV or 450 mg PO. The results of this assessment provided support for the adequacy of current model for predicting delafloxacin exposure after IV and PO administration in healthy subjects.

Figure 4.2.2-4 Visual predictive check of simulated and observed delafloxacin plasma concentrations from healthy subjects of Phase 1 Study 115 after administration of delafloxacin 300 mg IV or 450 mg PO, stratified by route of administration



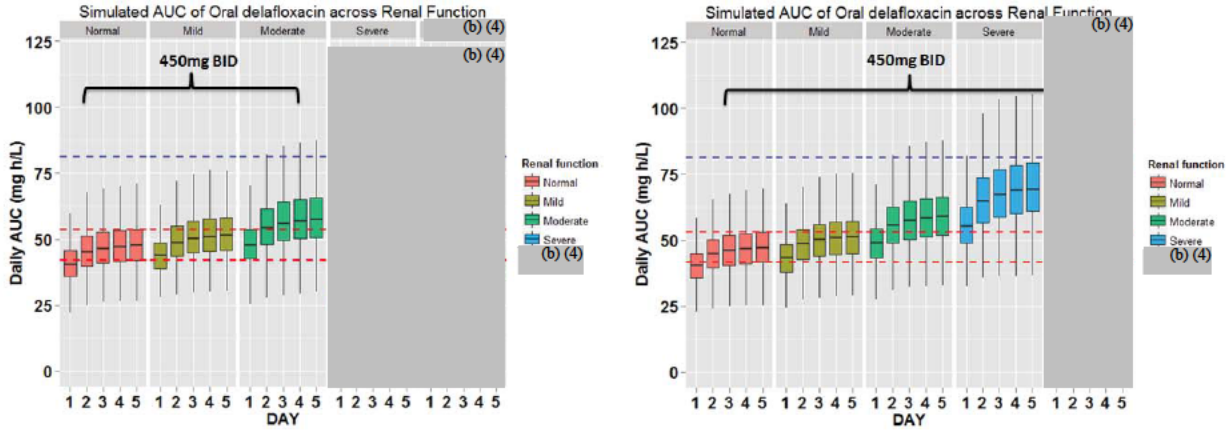
Blue dashed lines: Lower limit of quantification of plasma analytical assay.

Source: Applicant's population PK report for PO delafloxacin, Page 47, Figure 11

Reviewer's comments: Similar to the population PK analysis for IV delafloxacin, an independent population PK analysis for PO delafloxacin using eGFR instead of CLCRN was performed by Reviewer. The parameter estimates were consistent with Applicant's results. The population PK model was able to describe the PK data of delafloxacin and can be used to predict exposure of delafloxacin after PO administration. It should be noted that there were no PK data for PO delafloxacin in ESRD subjects with or without hemodialysis, nor were there PO data in patients. Based on observations from the IV population PK model that no difference in the PK was

observed based on disease status (ABSSSI vs healthy), the Reviewer considers the developed population PK model for PO dosing to be acceptable for simulating exposures in ABSSSI patients. However, the population PK model is unable to simulate the exposure of PO delafloxacin in ESRD patients with or without hemodialysis as the absorption of PO delafloxacin is unknown in this population. The Reviewer simulated the exposure on Day 1 to 5 for PO delafloxacin across renal function categories based on demographics of patients in the two Phase 3 studies under two dosing regimens (i.e., Proposed by Applicant and no dose reduction) (Figure 4.2.2-5). The simulated mean, 95% CI AUC values are listed in Table 4.2.2-4. The results illustrate that the Applicant's proposed dosing regimen (b) (4) (b) (4) (b) (4) *Therefore, a dose of 450 mg BID is recommended for patients with severe renal impairment and no dose is recommended for ESRD patients with or without hemodialysis.*

Figure 4.2.2-5 Simulated Daily AUC from Day 1 to 5 for PO Delafloxacin across Renal Function under Two Dosing Regimens (Proposed by Applicant (Left) and no dose reduction (Right))



Blue dashed lines represent the mean daily AUC (81.1 mg·h/L) on Day 5 under dosing of 450 mg IV BID based on population PK model and demographics information in Study RX-3341-201; Red dashed lines represent the 25th and 75th percentile of daily AUC on Day 5 under dosing of 450 mg PO BID in normal renal function based on population PK model. The simulated daily

(b) (4)

Source: Reviewer's independent analysis

Table 4.2.2-4 Summary of Simulated Exposure of PO delafloxacin across Renal Function based on Population PK Model

Parameter eGFR (mL/min/1.73m^2)	*Proposed dosing by Applicant*		*No dose reduction*	
	Dose	*Daily AUCss(mg·h/L) (mean, 95% CI)*	*Dose*	*Daily AUC_{ss}(mg·h/L) (mean, 95% CI)*
Normal (⩾90)	*450 BID*	*48.2 (47.6-48.8)*	*450 BID*	*47.5 (47.0-48.1)*
Mild (60- (b) (4)	*450 BID*	*52.3 (51.7-53.0)*	*450 BID*	*51.8 (51.1-52.4)*
Moderate (30- (b) (4)	*450 BID*	*58.7 (58.0-59.4)*	*450 BID*	*59.5 (58.8-60.2)*
Severe (15- (b) (4)	(b) (4)		*450 BID*	*70.3 (69.1-71.5)*
(b) (4)				

Source: Reviewer's independent analysis

4.3 Exposure-response Analysis

Exposure-response analysis for efficacy was conducted by Applicant based on one Phase 2 (Study 202) and two Phase 3 studies (Study 302 and 303). These analyses are based on the microbiologically evaluable (ME) population, which is a subset of the overall population PK population (see Section 4.2).

Study 202: Subjects were randomly assigned in a 1:1:1 ratio and received intravenous therapy with either delafloxacin 300 mg q12h (n = 80) or linezolid, 600 mg q12h (n = 80), or vancomycin (n = 80), 15 mg/kg. Treatment continued for 5 to 14 days. Scheduled study visits occurred at Screening, daily on Days 1 through 14, at Follow-up (FU), and at Late Follow-up (LFU). Clinical response was determined at the Follow-up and Late Follow-up visits, based upon the investigator's subjective assessment of the subject's signs and symptoms of infection. Microbiological response was determined for subjects in the ME population at the Follow-up and Late Follow-up visits at both the subject and pathogen levels. Blood samples were taken for PK assessments in all subjects. Multiple blood samples for analysis were obtained on Day 3 of treatment. Samples were taken within 2 hours before the first study drug administration, and at 1, 2, 3, 5, and 12 hours after the start of infusion (N=34).

Study 302: Approximately 660 male and female patients with ABSSSI were enrolled. Patients were randomly assigned to one of two treatment arms in a 1:1 ratio and received either delafloxacin 300 mg IV q12h or vancomycin 15 mg/kg IV q12h based on actual body weight with concomitant aztreonam therapy. Duration of treatment was 10 to 28 doses. Clinical response was determined at 48 to 72 hours after initiation of treatment, FU, and LFU. Microbiological response was determined at the FU and LFU assessment at both the patient and pathogen levels. Samples for PK analysis were collected from a minimum of 50% of patients on Day 3 of treatment within 2 hours before first study drug administration and at 1, 2, 3, 5, and 12 hours after start of the first infusion (N=162).

Study 303: Approximately 850 male and female patients with ABSSSI were enrolled. Patients were randomly assigned in a 1:1 ratio and received either delafloxacin 300 mg IV q12h for 6 doses with a mandatory switch to 450 mg orally q12h for the remaining does or vancomycin 2 g q12h. Duration of treatment was 10 to 28 doses. Samples for PK analysis were collected from patients at select sites on Day 3 of treatment within 30 minutes before first study drug administration and at 1.5 and 3 hours after the first study drug administration. The objective response was determined at 48 to 72 hours after initiation of treatment, at Follow up, and at Late Follow up (N=72).

Objective response at 48 to 72 hours after initiation of treatment was assessed in Studies 302 and 303. A patient was considered a responder if there was a $\geqslant$ 20% reduction of the ABSSSI lesion spread of erythema area, and the patient experienced none of the criteria for the failures. A patient was considered a non-responder if one of the following conditions was met: 1) A < 20% reduction of the ABSSSI lesion spread of erythema area at the assessment made at 48 to 72 hours

after initiation of study drug; 2) Administration of rescue antibacterial drug therapy or administration of non-study antibacterial drug therapy for treatment of the ABSSSI before the primary efficacy endpoint assessment at 48 to 72 hours; 3) Need for unplanned surgical intervention except for limited bedside debridement and standard wound care before the primary efficacy endpoint assessment at 48 to 72 hours; or 4) Death by 74 hours after initiation of study drug.

PK/PD relationships for delafloxacin efficacy described were evaluated using ratio of the free-drug plasma area under the concentration-time curve from 0 to 24 hours (*f*AUC24) to the minimum inhibitory concentration (MIC) (*f*AUC24:MIC ratio) for delafloxacin. It was demonstrated that *f*AUC:MIC ratio was the PK/PD index most predictive of bacterial reduction in animal studies for delafloxacin. In addition to Day 1 *f*AUC24:MIC ratio, Day 1 *f*AUC24 and MIC were evaluated. Delafloxacin exposures from 0 to 24 h were generated using population PK model (*See section 4.2*). The *f*AUC24 values were adjusted using a free-fraction of 0.16. This corresponds to the mean estimate for protein binding of 84% determined over a concentration range of 0.1 to 50 mg/L based on the results of *in vitro* protein binding study.

Univariable analyses were conducted to identify a relationship between the probability of achieving objective response at 48 to 72 hours and delafloxacin *f*AUC24:MIC ratio, fAUC24, and MIC value. A total of 268 patients who received delafloxacin and who were included in the ME Populations had PK data and at least one pathogen with MIC data isolated at baseline. Of these 268 patients, 195 had *S. aureus* isolated at baseline. A summary of the analysis population and the subset of patients with *S. aureus* at baseline is shown in Table 4.3-1.

Table 4.3-1 Summary of the number of patients by Phase 2 and 3 study who were included in the exposure-resposne analysis

Patient description	Study 202*	Study 302	Study 303	Total
Patients in the ME population who received delafloxacin with appropriate source pathogen and MIC data with PK	34	162	72	268
Patients in the ME populations with *S. aureus* isolated at baseline who received delafloxacin	31	119	45	195

**Objective response at 48 to 72 hours was not available in Study 202. The reason to include study 202 in the table is other efficacy endpoints included in the exposure-response analysis were collected by Applicant in this study.*
Source: Applicant's exposure-response analysis report, Page 43, Table 6

Summary statistics for categorical and continuous patient characteristics for all patients and patients with *S. aureus* are provided in Table 4.3-2 and Table 4.3-3.

Table 4.3-2 Summary for categorical patient characteristics for all patients and patients with *S. aureus*

Baseline characteristic	All patients (N=268)	*S. aureus* patients (N=195)
	n (%)	n (%)
Race		
White	232 (86.6)	170 (87.2)
Black or African American	23 (8.6)	14 (7.2)
Asian	4 (1.5)	3 (1.5)
American Indian or Alaska Native	4 (1.5)	4 (2.1)
Native Hawaiian or Other Pacific Islander	4 (1.5)	3 (1.5)
Other	1 (0.37)	1 (0.51)
Sex (female)	110 (41.0)	80 (41.0)
ABSSSI category		
Burn infection	2 (0.75)	2 (1)
Cellulitis/Erysipelas	69 (25.7)	42 (21.5)
Major cutaneous abscess	67 (25.0)	58 (29.7)
Wound infection	130 (48.5)	93 (47.7)
MRSA	98 (36.6)	98 (50.3)
Polymicrobial infection	57 (21.3)	33 (16.9)
Gram-negative pathogen at baseline	37 (13.8)	17 (8.7)
Prior antibiotic therapy	42 (15.7)	28 (14.4)
Presence of diabetes	22 (8.3)	11 (5.7)

Source: Applicant's exposure-response analysis report, Page 45, Table 7

Table 4.3-3 Summary for continuous patient characteristics for all patients and patients with *S. aureus*

Baseline characteristic	All patients (N=268)		*S. aureus* patients (N=195)	
	Mean (%CV)	Median (Min. – Max.)	Mean (%CV)	Median (Min. – Max.)
Age (yr)	44.7 (31.2)	46.0 (18 – 87)	42.7 (30.9)	44.0 (18 – 87)
Baseline CLcr (mL/min/1.73 m^2)	97.2 (34.5)	95.3 (21 – 244)	102 (30.5)	101 (28.3 – 244)
BMI (kg/m^2)	28.3 (23.7)	27.95 (15 – 59.0)	28.0 (23.0)	27.0 (15 – 59)
Body surface area (m^2)	1.94 (12.2)	1.94 (1.33 – 2.60)	1.93(12.1)	1.92 (1.33 – 2.51)
Body weight (kg)	82.5 (23.5)	81.3 (42.1 – 157)	81.3 (23.0)	79.6 (42.1 – 157)
Height (cm)	170 (5.7)	170 (142 – 196)	170 (5.8)	170 (142 – 196)
Ideal body weight	64.36 (16.1)	65 (36.3 – 89.1)	64.27 (16.3)	65 (36.3 –89.1)

** The unit for ideal body weight is kg*

Source: Applicant's exposure-response analysis report, Page 46, Table 8

Summary statistics for free-drug plasma AUC24, baseline MIC, and free-drug plasma AUC24:MIC ratio based on data from all patients and patients with *S. aureus* are provided in Table 4.3-4.

Table 4.3-4 Summary statistics for free-drug plasma AUC24, baseline MIC, and free-drug plasma AUC24:MIC ratio for all patients and patients with *S. aureus*

Analysis population	Variable	Free-drug plasma AUC (mg•h/L)	MIC (µg/mL)	Free-drug plasma AUC:MIC ratio
All patients (N=268)	Mean (%CV)	7.74 (30.5)	—	906 (109)
	Median or $MIC_{50/90}$ (Min. – Max.)	7.22 (3.35 – 16.8)	0.008/0.25 (0.001 - 2)	733 (4.85 – 5442)
Patients with *S. aureus* (N=195)	Mean (%CV)	7.68 (28.9)	—	979 (107)
	Median or $MIC_{50/90}$ (Min. – Max.)	7.25 (3.35 – 15.8)	0.008/0.25 (0.001 - 0.5)	825 (11.5 – 5442)

Source: Applicant's exposure-response analysis report, Page 50, Table 12

A summary of the percentages of patients with successful responses for objective response at 48 to 72 h by visit based on data from all patients and patients with *S. aureus* is shown in Table 4.3-5. Only the 228 patients with information of objective response at 48 to 72 h (159 patients with *S. aureus*) were included.

Table 4.3-5 Summary of successful responses for objective response at 48 to 72 h by visit for all patients and patients with *S. aureus*

Analysis population	Visit	% success (n/N)
All patients	48-72 h	88.6 (202/228)
Patients with *S. aureus*	48-72 h	89.3 (142/159)

Source: Applicant's exposure-response analysis report, Page 47, Table 9

A summary of p-values for univariable relationships between the probability of achieving objective response at 48 to 72 h and *f*AUC24:MIC ratio, *f*AUC or MIC variables is shown in Table 4.3-6.

Table 4.3-6 Summary of p-values for univariable relationships between the probability of achieving objective response at 48 to 72 h and *f*AUC24:MIC ratio, *f*AUC, and MIC on Day 1 based on data from all patients and patients with *S. aureus*

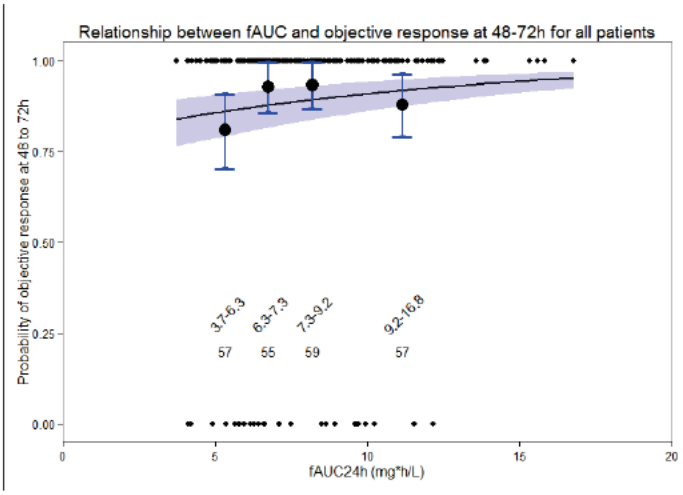
	Continuous	Quartiles
Free-drug plasma AUC:MIC ratio (*f*AUC:MIC)		
All patients	0.54	0.83
Patients with *S. aureus*	0.82	1.00
free-drug plasma AUC (*f*AUC)		
All patients	0.27	0.13
Patients with *S. aureus*	0.032	0.0013
MIC		
All patients	0.97	-
Patients with *S. aureus*	0.42	-

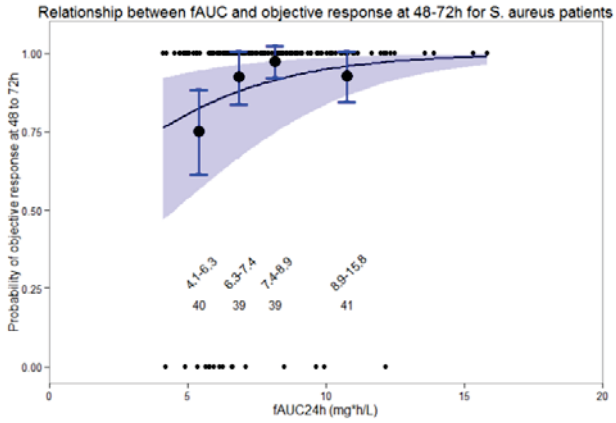
Source: Applicant's exposure-response analysis report, Page 120, Appendix 3.1-3.3

The univariable relationships for *f*AUC24:MIC ratio and objective response at 48 to 72 h was not significant at the 0.05 level for all the patients. A similar relationship was identified for *f*AUC and objective response at 48-72 h. The results for patients with *S. aureus* are not consistant for *f*AUC.

Reviewer's comment: Reviewer conducted an independent analysis for the relationship between free drug AUC24 and objective response at 48 to 72 h in ME population. Similar analysis was performed for fAUC24:MIC, as free drug AUC/MIC is the most relevant PK/PD index based on animal study and previous experience with fluoroquinolones. The ME population was used due to the fact that MIC is only available in this population, which is required to use fAUC/MIC for exposure-response analyses. In total 228 patients were included in the exposure-response analysis with both exposure and efficacy endpoint information of objective response rate at 48 to 72 h. Among them, 159 patients were found to have S. aureus in their blood samples or from the ABSSSI infection site. No significant relationship was identified for all the patients (p=0.29) and a nominal relationship was identified for patients with S. aureus (p=0.0497) (Figure 4.3-1). The result of reviewer's analyses is consistent with that of Applicant's conclusion. A comparable result was found that no significant relationship was identified between AUC24:MIC and objective response at 48 to 72 h (Figure 4.3-2). Overall, the exposure-response relationship for efficacy support the proposed dose of 300 mg BID IV (or its comparable 450 mg BID PO).

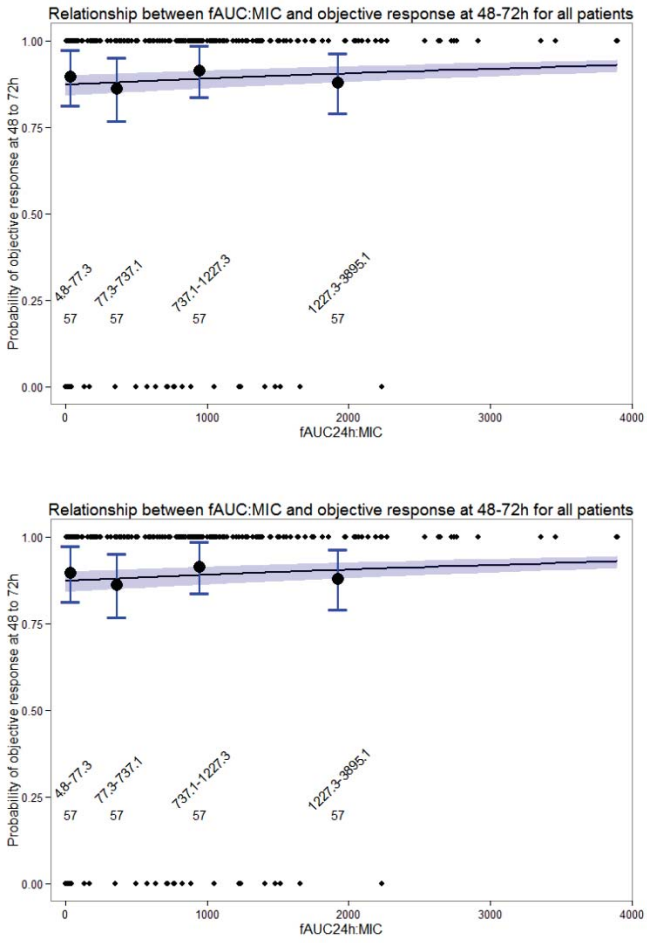
Figure 4.3-1 Exposure-response analysis for objective response at 48 to 72h for the ME patient population (top) and patients in the ME population with S. aureus (bottom) using fAUC24





Source: Reviewer's independent analysis

Figure 4.3-2 Exposure-response analysis for objective response at 48 to 72h for all patients in the ME population (top) and patients in the ME population with S. aureus (bottom) using fAUC24:MIC



Source: Reviewer's independent analysis

4.4 Target Attainment Analysis

The PK/PD target derived from three animal infection models with *S. aureus*, *E. coli*, and *P. aeruginosa* are summarized in Table 4.4-1.

Table 4.4-1The PK/PD target of fAUC24/MIC by endpoints and pathogens

	Net Bacterial Stasis	1-log10 decline in CFU
S. aureus	9.3 (range: 2.0-12.8)	14.3 (range: 6.3-18.5)
E. coli	14.5	26.2
P. aeruginosa	3.8 (range: 3.2-5.2)	5.0 (range: 4.2-6.0)

Source: Summary from Applicant's summary of clinical pharmacology and PK-3341-020 report.

The evaluation of PK-PD target attainment in simulated patients receiving delafloxacin IV followed by PO was carried out using a previously-developed population PK model, free-drug plasma AUC24:MIC ratio targets for efficacy of delafloxacin against *S. aureus* based on a neutropenic murine-thigh infection model, *in vitro* surveillance data, and Monte Carlo simulation. The simulated populations, which differed by ranges of creatinine clearance (CLcr) included those with normal renal function (90 ≤ CLcr < 200 mL/min/1.73 m^2) or mild (60 ≤ CLcr < 90 mL/min/1.73 m^2), moderate (30 ≤ CLcr < 60 mL/min/1.73 m^2), or severe renal impairment (15 ≤ CLcr < 30 mL/min/1.73 m^2) or with end-stage renal disease (ERSD; 5 ≤ CLcr < 15 mL/min/1.73 m^2) on non-dialysis day. In each group, CLcr was simulated using a uniform probability distribution. In addition, a population of simulated patients with similar demographics to those patients in Studies 202, 302 and 303 in the population PK dataset were generated.

Simulated patients with normal renal function, and either mild or moderate renal impairment received delafloxacin 300 mg IV q12h on Days 1-3 followed by 450 mg PO q12h on Day 4. Simulated patients with severe renal impairment [redacted] (b) (4) received delafloxacin 200 mg IV q12h on Days 1-3 followed by [redacted] (b) (4) on Day 4. Delafloxacin free-drug plasma AUC24:MIC ratio targets associated with net bacterial stasis and a 1-log10 colony forming unit (CFU) reduction from baseline for *S. aureus* were 9.3 and 14.3, respectively.

Table 4.4-2 Delafloxacin MIC distributions for *S. aureus* based on in vitro surveillance data

Isolate group (sample size)[a]	Number of occurrences n (%) by MIC (µg/mL)[b]												
	≤0.004	0.008	0.015	0.03	0.06	0.12	0.25	0.5	1	2	4	MIC_{50}	MIC_{90}
All (n=1311)	731 (55.8)	131 (65.8)	19 (67.2)	34 (69.8)	134 (80.0)	101 (87.7)	78 (93.7)	36 (96.4)	23 (98.2)	5 (98.6)	19 (100.0)	≤0.004	0.25
MRSA (n=563)	164 (29.1)	31 (34.6)	5 (35.5)	26 (40.1)	118 (61.1)	85 (76.2)	64 (87.6)	29 (92.7)	18 (95.9)	5 (96.8)	18 (100.0)	0.06	0.5
MSSA (n=748)	567 (75.8)	100 (89.2)	14 (91.0)	8 (92.1)	16 (94.3)	16 (96.4)	14 (98.3)	7 (99.2)	5 (99.9)	0 (99.9)	1 (100.0)	≤0.004	0.015

Source: Applicant's exposure-response analysis report, Page 41, Table 5

Using the free-drug plasma AUC24 values on Days 1 and 4 following the first day of IV therapy and first day of therapy after the PO switch, respectively, for simulated patients, percent probabilities of attaining the above-described nonclinical free-drug plasma AUC24:MIC ratio targets were determined. Percent probabilities of PK-PD target attainment on Days 1 and 4 were assessed at fixed MIC values ranging from 0.004 to 4 µg/mL and weighted over MIC distributions for delafloxacin against all *S. aureus* isolates, MRSA, and MSSA based on recent *in vitro* surveillance data (Table 4.4-2).

The results are shown in Table 4.4-3. In the population of simulated patients resembling the clinical trial population, percent probabilities of PK-PD target attainment at the MIC90 value for *S. aureus* isolates of 0.25 µg/mL and based on the MIC distributions for MSSA and MRSA ranged from 91.0 to 100% on Days 1 or 4 for simulated patients across renal function groups. Percent probabilities of PK-PD target attainment at a MIC value of 0.5 µg/mL based on the free-drug plasma AUC24:MIC ratio target associated with net bacterial stasis were 90.8 and 86.8% on Days 1 and 4, respectively.

Table 4.4-3 Percent probabilities of PK-PD target attainment by MIC on Days 1 and 4 for delafloxacin associated with net bacterial stasis and 1-log10 CFU reduction based on the free-drug plasma AUC:MIC ratio targets for *S. aureus* efficacy among simulated patients

Endpoints for free-drug plasma AUC:MIC ratio targets	MIC (µg/mL)	Percent probabilities of PK-PD target attainment by MIC on Days 1 or 4 among simulated patients								(b) (4)	All patients[e]	
		Normal renal function		Mild renal impairment		Moderate renal impairment		Severe renal impairment[d]				
		Day 1	Day 4	Day 1	Day 4	Day 1	Day 4	Day 1	Day 4		Day 1	Day 4
Net bacterial stasis[a]	0.25	99.9	99.5	100	100	100	100	100	98.3		100	99.8
	0.5	81.6	77.8	95.1	92.5	98.4	97.1	89.2	66.1		90.8	86.7
	1	10.3	12.4	24.8	31.1	43.9	49.7	13.2	8.20		17.5	22.5
	Overall[c]											
	All	96.1	96.0	96.7	96.8	97.1	97.2	96.3	95.5		96.5	96.4
	MRSA	92.1	91.9	93.3	93.3	94.0	94.2	92.6	91.0		92.8	92.7
	MSSA	99.1	99.1	99.3	99.3	99.5	99.5	99.2	98.9		99.2	99.2
1-$\log_{10}$ CFU reduction[b]	0.25	96.9	93.1	99.7	98.5	99.7	99.4	98.3	87.9		98.5	96.2
	0.5	31.9	33.4	58.4	58.3	75.8	75.1	38.9	22.5		47.4	48.6
	1	0.33	1.07	1.00	4.40	5.33	11.8	0.67	0.73		1.18	3.19
	Overall[c]											
	All	94.4	94.2	95.3	95.3	95.8	95.9	94.7	93.6		94.9	94.8
	MRSA	88.9	88.5	90.6	90.5	91.6	91.7	89.4	87.4		89.9	89.7
	MSSA	98.5	98.5	98.8	98.8	99.0	99.0	98.6	98.3		98.7	98.7

Source: Applicant's exposure-response analysis report, Page 21, Table 2

Similar analysis was performed for *P. aeruginosa*. Delafloxacin MIC values were determined for skin and soft tissue isolates of *P. aeruginosa*, collected from North American patients in 2014 to 2015 (MB-3341-050 and MB-3341-061). MIC values for P. aeruginosa are given in Table 4.4-4. The majority of isolates (35 of 40 total) exhibited MICs $\leq$ 1 µg/mL.

The simulations for *P. aeruginosa* predicted target attainment probabilities of 97% for stasis and 86% for 1-log10 bacterial reduction at an MIC of 1 µg/mL (Table 4.4-5 and Figure 4.4-1).

Table 4.4-4 MIC Frequencies (Cumulative Percentage) of *P. aeruginosa* Skin and Soft Tissue Isolates From 2014-2015 North American Delafloxacin Surveillance

MIC (µg/mL)					
0.008	0.015	0.03	0.06	0.12	0.25
--	0 (0.0%)	1 (2.5%)	1 (5.0%)	15 (42.5%)	10 (67.5%)

MIC (µg/mL)							
0.5	1	2	4	>4	Total	MIC_{50}	MIC_{90}
3 (75.0%)	5 (87.5%)	3 (95.0%)	1 (97.5%)	1 (100.0%)	40	0.25	2

Source: Applicant's PK-3341-020 report, Page 16, Table 5

Table 4.4-5 Percent Probability of Stasis or 1-Log10 Bacterial Reduction Target Attainment in Monte Carlo Simulations Based On Clinical Isolate Distributions

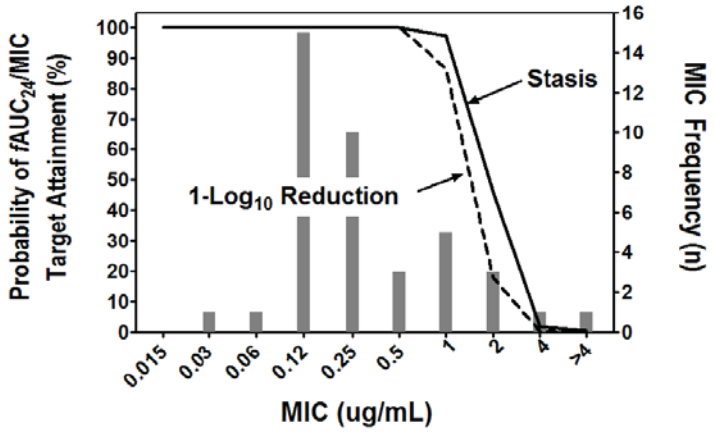
Each value is the percent probability of target attainment at the indicated MIC
Values in parentheses are the overall percent probability of target attainment at MIC values up to and including the indicated MIC

	MIC (µg/mL)								
Target	0.03	0.06	0.12	0.25	0.5	1	2	4	>4
Stasis	100 (100)	100 (100)	100 (100)	100 (100)	100 (100)	97.3 (99.6)	45.9 (95.5)	1.7 (93.1)	0.5 (90.7)
1-Log_{10} CFU Reduction	100 (100)	100 (100)	100 (100)	100 (100)	100 (100)	86.5 (98.1)	17.8 (91.9)	0.3 (89.6)	0.1 (87.3)

Source: Applicant's PK-3341-020 report, Page 17, Table 6

Figure 4.4-1 Probability of Target Attainment in Humans Based on Monte Carlo Simulations Using the MIC Distribution *for P. aeruginosa* Skin and Soft Tissue Isolates from Delafloxacin North American Surveillance

Solid Line: Probability (%) of *f*AUC_{24}/MIC target attainment for stasis (3.81)
Dashed Line: Probability (%) of *f*AUC_{24}/MIC target attainment for 1-log_{10} CFU reduction (5.02)
Grey Bars: MIC Frequencies for *P. aeruginosa* skin and soft tissue isolates from 2014-2015 delafloxacin surveillance (n=40)



Source: Applicant's PK-3341-020 report, Page 17, Figure 8

Reviewer's comment: The Reviewer conducted an independent analysis for assessing the probability of target attainment in patients based on simulation using the delafloxacin population PK models and the PK/PD target reported in Table 4.4-1. The Reviewer used fixed MIC values from 0.004 to 4 µg/mL for all simulated patients with eGFR ranging from 5 to 120 mL/min/1.73m^2 and demographics from the Phase 3 studies in the population PK dataset. The populations were grouped by different levels of renal function including patients with normal renal function (90 ≤ eGFR<120 mL/min/1.73 m^2), mild (60 ≤ eGFR < 90 mL/min/1.73 m^2), moderate (30 ≤ eGFR < 60 mL/min/1.73 m^2), severe renal impairment (15 ≤ eGFR < 30 mL/min/1.73 m^2), and end-stage renal disease (ERSD; 5 ≤ eGFR < 15 mL/min/1.73 m^2) without dialysis. The steady state AUC (AUC on day 5) and protein binding of 84% was used to calculate fAUC24:MIC for patients with normal renal function and mild, moderate, and severe renal impairment. (b) (4). The results for IV delafloxacin are listed in Tables 4.4-6, 4.4-7, and 4.4-8 separated by renal function for S. aureus, E. coli, and P. aeruginosa, respectively. The dosing regimens for IV delafloxacin used in the Reviewer's PTA analysis were 300 mg BID for patients with normal renal function, mild renal impairment, and moderate renal impairment and 200 mg BID for patients with severe renal impairment (b) (4) which are the proposed dosing regimens by Applicant.

Table 4.4-6 Percent probabilities of PK-PD target attainment by MIC for IV delafloxacin using the proposed dosing regimen. Results are shown for net bacterial stasis and 1-log10 CFU reduction based on fAUC24:MIC for S. aureus among simulated patients

MIC	Normal renal function		Mild renal impairment		Moderate renal impairment		Severe renal impairment		(b) (4)
	Stasis	1-log	Stasis	1-log	Stasis	1-log	Stasis	1-log	
0.004	1	1	1	1	1	1	1	1	
0.008	1	1	1	1	1	1	1	1	
0.015	1	1	1	1	1	1	1	1	
0.03	1	1	1	1	1	1	1	1	
0.06	1	1	1	1	1	1	1	1	
0.12	1	1	1	1	1	1	1	1	
0.25	1	0.99	1	1	1	1	1	0.99	
0.5	0.93	0.56	0.97	0.68	0.99	0.82	0.92	0.53	
1	0.26	0.03	0.38	0.07	0.54	0.13	0.26	0.04	
2	0.01	0	0.01	0	0.04	0	0.01	0	
4	0	0	0	0	0	0	0	0	

Source: Reviewer's independent analysis

Table 4.4-7 Percent probabilities of PK-PD target attainment by MIC for IV delafloxacin using proposed dosing regimen. Results are shown for net bacterial stasis and 1-log10 CFU reduction based on fAUC24:MIC for E. coli among simulated patients

MIC	Normal renal function		Mild renal impairment		Moderate renal impairment		Severe renal impairment		(b) (4)
	Stasis	1-log	Stasis	1-log	Stasis	1-log	Stasis	1-log	
0.004	1	1	1	1	1	1	1	1	
0.008	1	1	1	1	1	1	1	1	
0.015	1	1	1	1	1	1	1	1	
0.03	1	1	1	1	1	1	1	1	
0.06	1	1	1	1	1	1	1	1	
0.12	1	1	1	1	1	1	1	1	
0.25	0.99	0.66	1	0.78	1	0.88	0.98	0.64	
0.5	0.54	0.05	0.67	0.10	0.81	0.18	0.51	0.06	
1	0.03	0	0.06	0	0.12	0	0.03	0	
2	0	0	0	0	0	0	0	0	
4	0	0	0	0	0	0	0	0	

Source: Reviewer's independent analysis

Table 4.4-8 Percent probabilities of PK-PD target attainment by MIC for IV delafloxacin using proposed dosing regimen. Results are shown for net bacterial stasis and 1-log10 CFU reduction based on fAUC24:MIC for P. aeruginosa among simulated patients

MIC	Normal renal function		Mild renal impairment		Moderate renal impairment		Severe renal impairment		(b) (4)
	Stasis	1-log	Stasis	1-log	Stasis	1-log	Stasis	1-log	
0.004	1	1	1	1	1	1	1	1	
0.008	1	1	1	1	1	1	1	1	
0.015	1	1	1	1	1	1	1	1	
0.03	1	1	1	1	1	1	1	1	
0.06	1	1	1	1	1	1	1	1	
0.12	1	1	1	1	1	1	1	1	
0.25	1	1	1	1	1	1	1	1	
0.5	1	1	1	1	1	1	1	1	
1	0.99	0.90	1	0.95	1	0.98	0.98	0.89	
2	0.49	0.20	0.62	0.30	0.76	0.46	0.46	0.21	
4	0.02	0	0.04	0.01	0.10	0.02	0.03	0	

Source: Reviewer's independent analysis

The results of PTA analysis are consistent with Applicant's conclusion. For S. aureus more than 90% patients would achieve the PK/PD target associated with net bacterial stasis for MIC equal to or lower than (b) (4) *µg/mL across different levels of renal function. The proposed breakpoint appears to be acceptable. However, less than 90% patients would achieve the PK/PD target associated with 1-log10 CFU reduction for MIC equal to* (b) (4) *µg/mL across different levels of*

renal function. For E. coli, the MIC needs to be equal to or lower than 0.25 µg/mL, while for P. aeruginosa, the MIC needs to be equal to or lower than 1 µg/mL, respectively, for more than 90% of patients to achieve the PK/PD target of 9.3, 14.5 and 3.8.

A similar PTA analysis was conducted for PO delafloxacin across renal function categories. The dosing regimen for PO delafloxacin used in Reviewer's PTA analysis was 450 mg BID for patients with normal renal function, mild renal impairment and moderate renal impairment, and (b) (4) *for patients with severe renal impairment* (b) (4) *which are proposed dosing regimens by Applicant. The steady state AUC (AUC on day 5) was simulated based on population PK model and protein binding of 84% was used to calculate fAUC24:MIC for patients with normal renal function and mild, moderate, and severe renal impairment.* (b) (4) *. The results for PO delafloxacin are listed in Tables 4.4-9, 4.4-10, and 4.4-11 separated by renal function for S. aureus, E coli, and P. aeruginosa, respectively. The results show that the proposed dose reduction in patients with severe renal impairment would result in relatively lower PTA compared to that in other patients. The breakpoint would be lower in patients with severe renal impairment for all three strains. Therefore, a higher dose may be needed in patients with severe renal impairment.* (b) (4) (b) (4) *due to concerns stated in Section 4.2 about the ability of the population PK model to predict exposures in ESRD patients with or without hemodialysis, it is unsure if the predicted exposures would be precise and the predicted PTA would be achieved. As such, no dosing is proposed in this population.*

Table 4.4-9 Percent probabilities of PK-PD target attainment by MIC for PO delafloxacin using proposed dosing regimen. Results are shown for net bacterial stasis and 1-log10 CFU reduction based on fAUC24:MIC for S. aureus among simulated patients

MIC	Normal renal function		Mild renal impairment		Moderate renal impairment		Severe renal impairment		(b) (4)
	Stasis	1-log	Stasis	1-log	Stasis	1-log	Stasis	1-log	
0.004	1	1	1	1	1	1	1	1	
0.008	1	1	1	1	1	1	1	1	
0.015	1	1	1	1	1	1	1	1	
0.03	1	1	1	1	1	1	1	1	
0.06	1	1	1	1	1	1	1	1	
0.12	1	1	1	1	1	1	1	1	
0.25	1	0.98	1	0.99	1	1	0.99	0.88	
0.5	0.92	0.54	0.95	0.66	0.97	0.77	0.65	0.13	
1	0.24	0.01	0.31	0.03	0.47	0.06	0.02	0	
2	0	0	0	0	0.01	0	0	0	
4	0	0	0	0	0	0	0	0	

Source: Reviewer's independent analysis

Table 4.4-10 Percent probabilities of PK-PD target attainment by MIC for PO delafloxacin using proposed dosing regimen. Results are shown for net bacterial stasis and 1-log10 CFU reduction based on fAUC24:MIC for E. coli among simulated patients

(b) (4)

MIC	*Normal renal function*		*Mild renal impairment*		*Moderate renal impairment*		*Severe renal impairment*	
	Stasis	*1-log*	*Stasis*	*1-log*	*Stasis*	*1-log*	*Stasis*	*1-log*
0.004	1	1	1	1	1	1	1	1
0.008	1	1	1	1	1	1	1	1
0.015	1	1	1	1	1	1	1	1
0.03	1	1	1	1	1	1	1	1
0.06	1	1	1	1	1	1	1	1
0.12	1	0.99	1	1	1	1	1	0.93
0.25	0.98	0.65	0.99	0.76	1	0.84	0.87	0.22
0.5	0.52	0.03	0.65	0.05	0.76	0.10	0.12	0
1	0.01	0	0.02	0	0.06	0	0	0
2	0	0	0	0	0	0	0	0
4	0	0	0	0	0	0	0	0

Source: Reviewer's independent analysis

Table 4.4-11 Percent probabilities of PK-PD target attainment by MIC for PO delafloxacin using proposed dosing regimen. Results are shown for net bacterial stasis and 1-log10 CFU reduction based on fAUC24:MIC for P. aeruginosa among simulated patients

(b) (4)

MIC	*Normal renal function*		*Mild renal impairment*		*Moderate renal impairment*		*Severe renal impairment*	
	Stasis	*1-log*	*Stasis*	*1-log*	*Stasis*	*1-log*	*Stasis*	*1-log*
0.004	1	1	1	1	1	1	1	1
0.008	1	1	1	1	1	1	1	1
0.015	1	1	1	1	1	1	1	1
0.03	1	1	1	1	1	1	1	1
0.06	1	1	1	1	1	1	1	1
0.12	1	1	1	1	1	1	1	1
0.25	1	1	1	1	1	1	1	1
0.5	1	1	1	1	1	1	1	0.98
1	0.97	0.88	0.99	0.92	0.99	0.96	0.84	0.57
2	0.46	0.17	0.58	0.23	0.70	0.38	0.09	0.01
4	0.01	0	0.02	0	0.04	0	0	0

Source: Reviewer's independent analysis

For comparison, additional PTA analyses were conducted for PO delafloxacin in patients with severe renal impairment. The results show that 450 mg BID administered to patients with severe renal impairment would result in a PTA >90% at an MIC similar to that for patients with normal renal function, mild renal impairment, and moderate renal impairment.

Table 4.4-12 Percent probabilities of PK-PD target attainment by MIC for PO delafloxacin using dosing regimen of 450 mg BID. Results are shown for net bacterial stasis and 1-log10 CFU reduction based on fAUC24:MIC for S. aureus among simulated patients

MIC	*Normal renal function*		*Mild renal impairment*		*Moderate renal impairment*		*Severe renal impairment*	
	Stasis	*1-log*	*Stasis*	*1-log*	*Stasis*	*1-log*	*Stasis*	*1-log*
0.004	1	1	1	1	1	1	1	1
0.008	1	1	1	1	1	1	1	1
0.015	1	1	1	1	1	1	1	1
0.03	1	1	1	1	1	1	1	1
0.06	1	1	1	1	1	1	1	1
0.12	1	1	1	1	1	1	1	1
0.25	1	0.98	1	0.99	1	1	1	1
0.5	0.92	0.53	0.95	0.65	0.98	0.78	0.99	0.90
1	0.21	0.01	0.30	0.02	0.49	0.06	0.67	0.18
2	0	0	0	0	0	0	0.04	0
4	0	0	0	0	0	0	0	0

Source: Reviewer's independent analysis

Table 4.4-13 Percent probabilities of PK-PD target attainment by MIC for PO delafloxacin using dosing regimen of 450 mg BID. Results are shown for net bacterial stasis and 1-log10 CFU reduction based on fAUC24:MIC for E. coli among simulated patients

MIC	*Normal renal function*		*Mild renal impairment*		*Moderate renal impairment*		*Severe renal impairment*	
	Stasis	*1-log*	*Stasis*	*1-log*	*Stasis*	*1-log*	*Stasis*	*1-log*
0.004	1	1	1	1	1	1	1	1
0.008	1	1	1	1	1	1	1	1
0.015	1	1	1	1	1	1	1	1
0.03	1	1	1	1	1	1	1	1
0.06	1	1	1	1	1	1	1	1
0.12	1	0.99	1	0.99	1	1	1	1
0.25	0.98	0.64	0.99	0.73	1	0.85	1	0.93
0.5	0.51	0.03	0.63	0.05	0.77	0.12	0.89	0.27
1	0.01	0	0.02	0	0.06	0	0.17	0
2	0	0	0	0	0	0	0	0
4	0	0	0	0	0	0	0	0

Source: Reviewer's independent analysis

Table 4.4-14 Percent probabilities of PK-PD target attainment by MIC for PO delafloxacin using dosing regimen of 450 mg BID. Results are shown for net bacterial stasis and 1-log10 CFU reduction based on fAUC24:MIC for P. aeruginosa among simulated patients

MIC	*Normal renal function*		*Mild renal impairment*		*Moderate renal impairment*		*Severe renal impairment*	
	Stasis	*1-log*	*Stasis*	*1-log*	*Stasis*	*1-log*	*Stasis*	*1-log*
0.004	1	1	1	1	1	1	1	1
0.008	1	1	1	1	1	1	1	1
0.015	1	1	1	1	1	1	1	1
0.03	1	1	1	1	1	1	1	1
0.06	1	1	1	1	1	1	1	1
0.12	1	1	1	1	1	1	1	1
0.25	1	1	1	1	1	1	1	1
0.5	1	1	1	1	1	1	1	1
1	0.97	0.88	0.98	0.92	1	0.96	1	0.98
2	0.44	0.15	0.58	0.22	0.72	0.40	0.86	0.60
4	0.01	0	0.01	0	0.04	0	0.14	0.02

Source: Reviewer's independent analysis

To summarize, the Reviewer's PTA analysis results are consistent with those from Applicant's results as shown in Table 4.4-15. The Applicant only conducted PTA analyses for S. aureus and P. aeruginosa but not for E. coli. The Reviewer conducted a PTA analysis for all three organisms. In Applicant's analysis, the exposure of IV delafloxacin was AUC24 on the first day and the exposure of oral delafloxacin was AUC24 on fourth day after IV dosing was given for first three days.The same protein binding was used for all the patients across renal function. In the Reviewer's analysis, the exposure of IV and PO delafloxacin was AUC24 on Day 5 after five days of dosing as steady state AUC is considered the most relavant PK parameter associated with delafloxacin antibacterial activity. The same protein binding (84%) was used for patients with normal renal function, and mild, moderate, and severe renal impairment, (b) (4)

Table 4.4-15 Breakpoint summary based on applicant and reviewer's PTA analysis results

Strains	*Applicant proposed breakpoint*	*Reviewer's breakpoint*
S. aureus	(b) (4)	
E. coli	*0.25*	*0.25*
P. aeruginosa	(b) (4)	

Source: Reviewer's independent analysis

4.5 Individual Clinical Pharmacology Report Reviews

	Study No.	Study information
4.5.1		Animal models to determine the PK/PD target
4.5.2	M00-224	Safety and Pharmacokinetics of Rising Single and Multiple Oral Doses of ABT-492 (Delafloxacin Oral Capsule) with Evaluation of Effects of Food, Age, and Gender
4.5.3	M01-284	Effect of ABT-492 (Delafloxacin Oral Capsule) on Photosensitivity Levels in Healthy Male and Female Subjects
4.5.4	M01-292	Metabolism and Disposition of [14C]ABT-492 (Delafloxacin Oral Capsule) in Man Following a Single Oral Administration
4.5.5	ML-334-118	A Phase 1 Study to Evaluate The Effect of Repeated Oral Doses of Delafloxacin (To-Be-Marketed Tablet) on the Pharmacokinetics of a Single Oral Dose of Midazolam in Healthy Subjects
4.5.6	ML-3341-112	An Open-label Evaluation of The Single-dose Pharmacokinetics of Delafloxacin (To-Be-Marketed IV formulation [Captisol Lyophilized]) in Subjects with and Without Hepatic Impairment
4.5.7	RX-3341-101	A Double-Blind, Placebo-Controlled Single Ascending Dose Tolerabilty and Pharmacokinetic Study of RX-3341 (Delafloxacin (b) (4) Solution) in Healthy Subjects
4.5.8	RX-3341-102	A Randomized, Double-Blind, Placebo-Controlled, Safety,Tolerability, and Pharmacokinetic Study of RX-3341 (Delafloxacin (b) (4) Solution) in Healthy Subjects following Twice-Daily Ontravenous Administration
4.5.9	RX-3341-103	A Phase 1, Single and Multiple Intravenous Dose Safety, Tolerability, and Pharmacokinetic Study of RX-3341 (Delafloxacin (b) (4) Solutions) in Healthy Subjects
4.5.10	RX-3341-104	A Phase 1, Single and Multiple Intravenous Dose Safety, Tolerability, and Pharmacokinetic Study of Two Formulations of Delafloxacin ((b) (4) Solutions) in Healthy Subjects
4.5.11	RX-3341-107	An Open Label, Single IV Dose Study in Healthy Male Subjects Designed to Assess the Mass Balance Recovery, Metabolite Profile and Metabolite Identification of [14C]-RX-3341 (Delafloxacin To-Be-Marketed IV formulation [Captisol Lyophilized])
4.5.12	RX-3341-108	A Phase 1, Single-dose, Safety, Tolerability, and Pharmacokinetic Study of Intravenous and Oral Formulations, and A Maximum Tolerated Dose Study of Delafloxacin (To-Be-Marketed IV formulation [Captisol Lyophilized]) in Healthy Subjects

4.5.13	RX-3341-110	A Phase 1, Multicenter, Open-Label, Parallel-Group Crossover Study to Assess the Effect of Renal Impairment on the Single-Dose Pharmacokinetics and Safety of Oral (Defloxacin Oral Capsule) and IV RX-3341 (Delafloxacin To-Be-Marketed IV formulation [Captisol Lyophilized]) and IV Placebo
4.5.14	RX-3341-115	A Phase 1, Open-Label, Randomized, Single-Dose Crossover Study to Determine the Pharmacokinetics and Relative Bioavailability of Intravenous and Oral Formulations of Delafloxacin (To-Be-Marketed IV and Oral formulations) in Healthy Subjects
4.5.15	RX-3341-116	A Phase 1, Open-Label, Crossover Study to Determine the Effect of Food on the Pharmacokinetics of a Single Dose of Oral Delafloxacin (To-Be-Marketed Tablet) in Healthy Subjects
4.5.16		In Vitro Study Reports
4.5.17		Literature Review for Sulfobutyl-Ether-β- Cyclodextrin (SBECD, Captisol) by the Reviewer
4.5.18		Pharmacogenomics Summary

4.5.1 Animal Models to Determine the PK/PD Target

Experimental S. aureus Thigh Infection Model in Neutropenic Mice

It has been shown that fluoroquinolones exhibit concentration-dependent killing, and $fAUC_{24}$/MIC is the PK/PD index that correlates with antibacterial activity in animal models[3]. The efficacy of delafloxacin was studied in a neutropenic mouse thigh infection model against 5 strains of *S. aureus* (Table 4.5.1-1). Two out of five were methicillin-sensitive *S. aureus* (MSSA) and the rest were methicillin-resistant *S. aureus* (MRSA).

To induce the thigh infection, each mouse received 0.1 mL of the inoculum (~10^6 CFU/mouse) in the caudal thigh muscle under light anesthesia. The bacterial burden in tissues was determined 26 hours after inoculation using standard techniques. The single-dose PK evaluation for delafloxacin following subcutaneous (SC) administration (0.2 mL/dose) was performed in thigh-infected neutropenic mice at 1.25, 20, 80, and 320 mg/kg. Each dose was administered 2 hours post-inoculation. For each subcutaneous dose examined, 3 mice per time point were sampled at the following time points: 0.5, 1, 2, 4, 6, and 8 hours. PK parameters including AUC, C_{max}, T_{max}, and $t_{1/2}$ were determined via noncompartmental analysis. Nonlinear least-squares regression was used to determine the correlation between antibacterial activity and *f*AUC/MIC. Please refer to Figure 4.5.1-1 for details. The *f*AUC_{24}/MIC ratios associated with achieving net bacterial stasis and with a 1-$\log_{10}$ reduction in CFU are summarized in Table 4.5.1-2. Based on

[3] Jacobs MR. Optimization of antimicrobial therapy using pharmacokinetic and pharmacodynamics parameters. Clin Microbiol Infect. 2001;7: 589-596

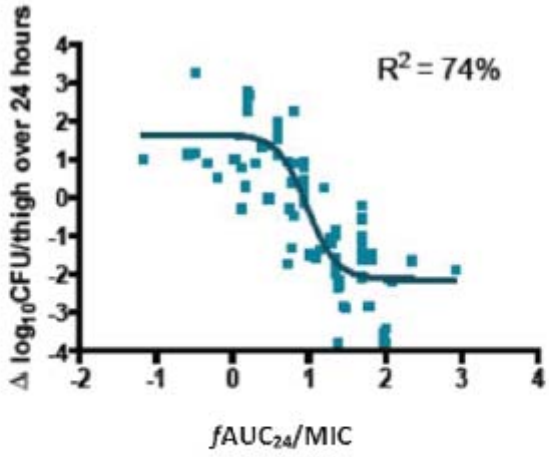
the results shown in Table 4.5.1-2, the *f*AUC$_{24}$/MIC ratios of 9.3 (net bacterial stasis) and 14.3 (1-log$_{10}$ reduction) were used, in part, to aid dose justification for Phase 3 clinical studies and breakpoint determination for *S. aureus*.

Table 4.5.1-1 MIC Values for *S. aureus* Strains Used in Experimentally-Induced Soft-Tissue Infection in Neutropenic Mice (Adapted from Table 159 in the Summary of Clinical Pharmacology)

Organism	MIC (µg/mL)			
Strain	Delafloxacin	Oxacillin	Levofloxacin	Clindamycin
S. aureus Smith ATCC 13709	0.004	0.5	0.125	0.125
S. aureus ATCC 29213	0.006[a]	0.5	0.25	0.125
S. aureus MRSA 11512	0.5	> 8	16	ND
S. aureus MRSA 11540 (USA 300)	0.8[a]	> 8	> 8	0.125
S. aureus HA-MRSA 2926 (USA 100)	0.016	> 8	0.25	> 8

ATCC = American Type Culture Collection, HA = Hospital-associated, MIC = Minimum inhibitory concentration, MRSA = Methicillin-resistant *S. aureus*, ND = Not determined
[a]Geometric means of replicates

Figure 4.5.1-1 Change in Bacterial Burden as a Function of *f*AUC$_{24}$/MIC With 5 Strains of *S. aureus*



CFU = colony-forming unit; *f*AUC$_{24}$/MIC = ratio of free delafloxacin area under the mean concentration-time curve from time 0 to 24 hours to pathogen minimum inhibitory concentration

**The X-axis is log scaled.*

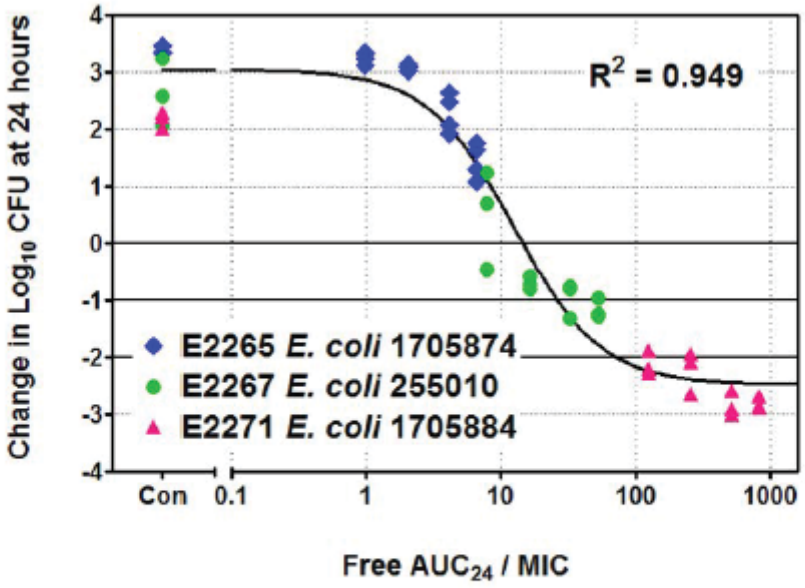
Table 4.5.1-2 Delafloxacin *f*AUC$_{24}$/MIC Values for Bacterial Stasis and 1-log$_{10}$ CFU Reduction for 5 *S. aureus* Strains

***S. aureus* Strain**	**MIC (µg/mL)**	**Delafloxacin *f*AUC$_{24}$/MIC Ratio Associated with Activity against *S. aureus***	
		Net Bacterial Stasis	**1-log$_{10}$ CFU Reduction**
ATCC 13709	0.004	3.8	
11540 (USA300)	0.8	9.3	14.3
2926 (USA100)	0.016	2.0	6.3
ATCC 29213	0.006	12.8	18.5
MRSA 11512	0.8	9.5	17.9
	Median	9.3	14.3
	Min, Max	2.0, 12.8	6.3, 18.5

Experimental E. coli Thigh Infection Model in Neutropenic Mice
The antibacterial activity of delafloxacin in a neutropenic mouse thigh infection model was evaluated against 3 strains of *E. coli*, with delafloxacin MIC values of 0.016, 0.25 and 2 µg/mL. The strains used in this model included *E. coli* 1705874 (an ESBL-positive, MDR strain), *E. coli* 1705884, and *E. coli* 255010.

Each mouse received an intramuscular inoculation (ranging from 10^5 to 10^6 CFU/mouse) in both caudal thigh muscles. Treatment with delafloxacin was administered SC at 2 and 9 hours after inoculationat doses of 25, 50, 100, and 160 mg/kg for each *E. coli* strain. The antibacterial activity was determined by enumerating the bacterial burdens in thighs 24 hours after initiation of therapy. Similar to the neutropenic thigh infection model for *S. aureus*, a nonlinear least-squares regression was used to determine the correlation between antibacterial activity and *f*AUC/MIC. Please refer to Figure 4.5.1-2 for details. For this set of strains, the results indicate that the *f*AUC24/MIC ratio for bacterial stasis is 14.5, and that for 1-log$_{10}$ CFU reduction is 26.2.

Figure 4.5.1-2 Delafloxacin PK/PD vs. 3 *E. coli* Strains in Mouse Neutropenic Thigh Infection Model (Adapted from Figure 77 in the Summary of Clinical Pharmacology)



CFU = colony-forming unit; *f*AUC_{24}/MIC = ratio of free delafloxacin area under the mean concentration-time curve from time 0 to 24 hours to pathogen minimum inhibitory concentration; PK/PD = pharmacokinetic/pharmacodynamic
Closed symbols: individual values for change in log CFU vs. estimated *f*AUC_{24}/MIC based on the mean percent free delafloxacin in mouse plasma
MIC values were 2 µg/mL for *E. coli* 1705874, 0.25 µg/mL for *E. coli* 255010, and 0.016 µg/mL for *E. coli* 1705884.

Applicant also conducted similar experiments in neutropenic mouse murine lung infection models using three respiratory pathogens: *S. aureus, S. pneumoniae,* and *K. pneumonia*. However, given that the infection site in these lung infection models is not relavant to the indication of ABSSSI, the reviewer did not review any animal studies in the lung infection murine model.

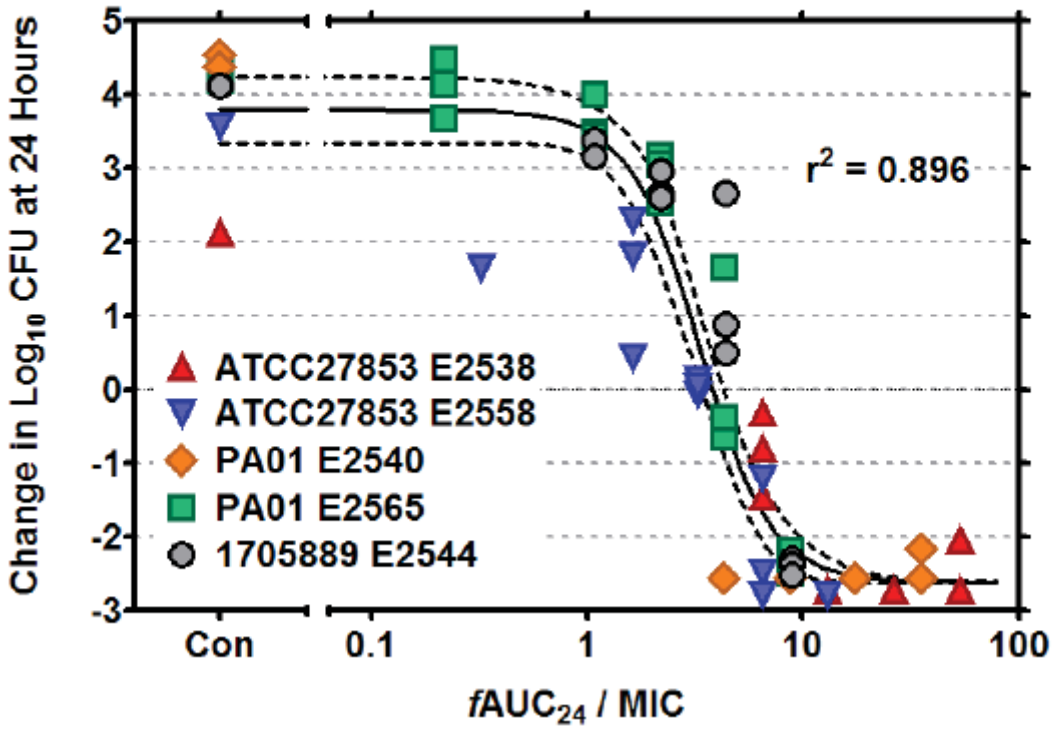
Experimental P. aeruginosa Thigh Infection Model in Neutropenic Mice
On 1/13/2017, Applicant submitted an update (SDN 011) including a PK-PD neutropenic mouse study report with *P. aeruginosa* and and subsequent human target attainment simulations (PK-3341-20) to support the clinical breakpoint for *P . aeruginosa*.

The activity of delafloxacin against three strains of *P. aeruginosa* was tested using a neutropenic CD-1 female mouse thigh infection model. The three strains were ATCC27853, PA01, and 1705889, with delafloxacin MICs of 0.25, 0.25 to 0.5, and 1 to 2 µg/mL, respectively. On Day 0, animals were inoculated with a preparation of the bacteria and then either untreated, or dosed with delafloxacin at 1, 5, 10, 20, 40, 80, or 160 mg/kg/dose, at 2 hr and 9 hours after inoculation, by subcutaneous administration. CFUs in thigh tissue were quantitated at 2 hr (untreated controls only) and 24 hr (controls and delafloxacin-dosed) after inoculation. The changes in log CFU relative to the 2 hr control group were determined. For each of the three strains, the change in *P. aeruginosa* levels (change in log_{10} CFU at 24 hr) were plotted against estimated *f*AUC_{24}/MIC.

Please refer to Figure 4.5.1-3 for details. The curve indicated mean *f*AUC*24*/MIC ratios of 3.81 [range: 3.2-5.2] for bacterial stasis, and 5.02 [range: 4.2-6.0] for a 1-log10 bacterial reduction.

Figure 4.5.1-3 Delafloxacin PK/PD vs Combined Results from Three *P. aeruginosa* Strains in Mouse

Closed Symbols: Individual values for change in $\log_{10}$ CFU vs estimated *f*AUC_{24}/MIC
Solid Line: Sigmoid curve with variable Hill slope
Dashed Lines: 95% Confidence Intervals



In summary, the animal models, study design and data analysis methods look reasonable for the PK/PD target determination in all three animal studies. We defer to the Clinical Microbiology reviewer for the evaluation of the bacterial strain selections.

Similar to other quinolone antibiotics, the *f*AUC_{24}/MIC ratio was determined to be the PK/PD index associated with delafloxacin's antibacteral activity.

PK/PD studies in the murine neutropenic thigh model using 5 strains of *S. aureus* against which delafloxacin has a range of MICs resulted in *f*AUC_{24}/MIC ratios of 2 to 12.8 to achieve bacterial stasis and 6.3 to 18.5 for 1-$\log_{10}$ CFU reduction. The median values for bacterial stasis and 1-log10 CFU reduction over the 5 *S. aureus* strains in this model are 9.3 and 14.3, respectively.

The results from a PK/PD study in the murine neutropenic thigh model using 3 strains of *E. coli* against which delafloxacin has a range of MICs indicated mean *f*AUC_{24}/MIC ratios of 14.5 for bacterial stasis, and 26.2 for a 1-log_{10} CFU reduction, respectively.

The results from a PK/PD study in the murine neutropenic thigh model using 3 strains of *P. aeruginosa* against which delafloxacin has a range of MICs indicated mean *f*AUC_{24}/MIC ratios of 3.81 [range: 3.2-5.2] for bacterial stasis, and 5.02 [range: 4.2-6.0] for a 1-log_{10} CFU reduction, respectively.

4.5.2 Study No.: M00-224

Safety and Pharmacokinetics of Rising Single and Multiple Oral Doses of ABT-492 (Delafloxacin Oral Capsule) with Evaluation of Effects of Food, Age, and Gender

Date(s): 27 October 2000 to 30 March 2001
Sponsor: Melinta Therapeutics, Inc, 300 George Street, Suite 301, New Haven, CT 06511
Clinical Site: Single site: Guys Drug Research Unit, 6 Newcomen Street, London SE1 1YR, United Kingdom
Analytical Site: Drug Analysis Department (R-46W) at Abbott Laboratories, Abbott Park, IL

OBJECTIVE(S):

- Single dose escalation (Part 1 and 2): To determine the safety, tolerability, and pharmacokinetics of ABT-492 under fasted conditions, following oral administration of single doses of ABT-492 in normal, healthy adult male volunteers and to assess the effect of food on the bioavailability of ABT-492. The safety, tolerability, and pharmacokinetics of ABT-492 were also to be assessed in subpopulations of healthy adult female volunteers and healthy elderly volunteers.
- Multiple dose escalation (Part 3): To determine the safety, tolerability, and pharmacokinetics of multiple doses of ABT-492 in normal, healthy, adult male and female volunteers.

METHODS

Study Design: This was a Phase 1, single-center study consisting of three parts:

- Part 1 was a single, rising-dose trial of ABT-492 (50, 100, 200,400, 800, 1200, or 1600 mg) or matching placebo in fasting, healthy adult (age 18 to 40 years) male volunteers. Fifty-six subjects were enrolled in parallel dosing groups of 8 subjects each. In each dosing group, subjects were randomly assigned to ABT-492 or placebo in a 3: 1 ratio. The dose was administered after a fast of at least 8 hours.
- The dose for Part 2 (250 mg) was selected based on the results of Part 1. Part 2 consisted of testing in three subpopulations. Part 2 (Food Effect), an evaluation of the effect of food on ABT-492 bioavailability, was a single-dose, two-period crossover trial in which healthy male subjects received ABT-492 (n=8) or placebo (n=2), fed in one period and fasted in the other period. Twenty subjects were randomly assigned to the four combinations of study drug (ABT-492 or placebo) and fasting or nonfasting administrations. The subjects were randomly assigned to active ABT-492 doses and placebo in a 4: 1 ratio, and received the same study drug in both periods of the study. Part 2 (Women) and Part 2 (Elderly) were single-dose trials in which sixteen healthy (age 18 to 40 years) female volunteers and sixteen elderly (age $\geqslant$65 years) male and female volunteers, respectively, received in a 3: 1 ratio a single dose (250 mg) of ABT-492 or placebo under fasting conditions.
- Part 3 was a multiple, rising-dose trial of ABT-492 (100, 200, 400, 800, or 1200 mg) or placebo once daily for 5 days in fasting, healthy adult male volunteers. Sixty subjects were assigned to parallel dosing groups of twelve subjects each. In each dosing group, subjects were randomly assigned to ABT-492 or placebo in a 2:1 ratio.

In all three parts of the study, blood and urine samples were collected on specific schedules for evaluation of ABT -492 concentrations and pharmacokinetic parameters. Safety was evaluated throughout the study by physical examinations, vital signs, laboratory tests, electrocardiograms (ECGs), and monitoring of adverse events.

Please refer to Table 1 below for the number of subjects enrolled and analyzed in each part of the study.

Table 1. Number of Subjects Planned and Analyzed in Study M00-224 (*Adapted from Synopsis in the study report)*

	ABT-492		
Number of Subjects (planned and analyzed):	Part 1	Part 2	Part 3
Number of Subjects Planned	56	52	60
Number of Subjects Randomized and Dosed	56	52	60
Subjects Included in the Pharmacokinetic Analyses (excludes placebo subjects and subjects that did not complete the study).	42	40	37
Subjects Included in the Safety Analyses	56	52	60

Note: *All subjects who received at least one dose of study drug were included in the safety analyses.*

Drug Product: Please refer to Table 2 below for the test products.

Table 2. Test Product, Dose, and Batch Number

Test Product, Dose and Mode of Administration, Batch Number:			
NPRO	**9773**	**9864**	**0013**
Finishing Lot Numbers: ABT-492 50 mg capsules ABT-492 100 mg capsules Matching placebo capsules	 70-098-S2 70-099-S2 70-100-S2	 73-346-S2 73-347-S2 73-348-S2	 N/A 74-466-S2 N/A
The test product was ABT-492 in 50 or 100 mg capsules for oral administration. In Part 1, the dosing groups were 50, 100, 200, 400, 800, 1200, or 1600 mg. Dosing in Part 2, determined based on the results from Part 1, was 250 mg. In Part 3, the multiple-dose phase, the dosing began at 100 mg and was escalated at a factor no greater than 3.33X the previous dose; the next successive dose groups received 200, 400, and 800 mg. A fifth dose was determined based on the results of the previous doses (1200 mg); however, maximum dose in Part 3 was not to exceed the maximum tolerated dose of the single-dose phase.			
Duration of Treatment: Single dose in Parts 1 and 2; 5 days in Part 3			

PK Sample Collection:
Please refer to Table 3 for the schedule of plasma and urine sample collection for Part 1 and 2 of the study and Table 4 for Part 3 of the study.

Table 3. Post-Dosing Procedures for Parts 1 and 2, Single-Dose *(Adapted from Table 9.5.b of the study report)*

Post-dosing procedures	Hours After Dosing																				
	Day 1															Day 2				Day 3	Day 4
	0.25	0.5	1	1.5	2	2.5	3	3.5	4	5	6	7	8	10	12	16	20	24	30	36	48	72
Blood Collection for Assay	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	
Urine Collection Intervals (Part 1 only)	X (0-2 hr)					X (2-4 hr)				X (4-8 hr)				X (8-12 hr)		X (12-24 hr)				X (24-48 hr)				
12-Lead ECG					X				X				X					X			X	
Physical Examination																		X			X	
Blood Pressure, Pulse Rate, Respiration Rate			X		X				X				X		X			X			X	
Oral Temperature															X							
Clinical Labs: *																						
Serum Chemistry									X									X				X**
Hematology									X									X				X**
Urinalysis (Dipstick)					X				X				X		X			X			X	
Micro Analysis - Urine					X				X				X		X			X			X	

* Any clinically significant laboratory result was repeated for verification.

** Subjects returned to the unit 72 hours post-dose for phlebotomy.

Table 4. Schedule of Assessments for Part 3, Multiple-Dose

Post-dosing Procedures	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7	Day 9-12
Blood Collection for all Subjects	X 0hr & 0.5hr, 1hr, 2hr, 3hr, 4hr, 6hr, 8hr, 10hr, 12hr, Post Dose	X Pre-Day 2 Dose) 18hr and 24hr Post-Day 1 Dose	X Pre-Day 3 Dose (48hr Post-Day 1 Dose)	X Pre-Day 4 Dose	X 0hr & 0.5hr, 1hr, 2hr, 3hr, 4hr, 6hr, 8hr, 10hr, 12hr, Post Dose	X 18hr, 24hr and 36hr Post-Day 5 Dose	X 48hr Post-Day 5 Dose	
Urine Collection Intervals	X Prior to dose				X (0-24hr)			
12-Lead ECG	X 2hrs post-dose				X 2hr post-dose		X	X
Physical Examination	X						X	
Blood Pressure, Pulse Rate, Respiration Rate	X[a]	X[a]	X[a]	X[a]	X[a]	X	X	X
Oral Temperature	X Prior to dose				X Prior to dose			
Clinical Labs:[b]								
Serum Chemistry			X Prior to dose		X Prior to dose		X	X
Hematology					X Prior to dose		X	X
Urinalysis (Dipstick)	X Prior to dose				X Prior to dose		X	X

[a] Predose and 2 hours post-dose

[b] Any clinically significant laboratory result was repeated for verification.

Analytical Methods:

The detail of the analytical method used for this study can be found in the method validation report "An analytical method for the determination of Abbott-319492 concentrations in human

plasma and urine using liquid chromatography with mass spectral detection" in this submission. Please refer to Table 5 for the dates of sample collection and dates of sample analysis.

Table 5. Dates of Sample Collection and Dates of Sample Analysis

	Dates of Sample Collection	Dates of Sample Analyses
Part I Single-Dose Escalation	03 Nov 2000 – 17 Dec 2000	08 Nov 2000 – 15 Jan 2001
Part II Females and Elderly	12 Jan 2001 – 04 Feb 2001	05 Feb 2001 – 26 Feb 2001
Part II Food Effect	12 Jan 2001 – 28 Jan 2001	06 Feb 2001 – 28 Feb 2001
Part III Multiple-Dose Escalation	22 Feb 2001 – 28 Mar 2001	06 Mar 2001 – 04 May 2001

Please refer to Table 6 for the summary of analytical method for quantification of delafloxacin in plasma and urine.

Table 6 Analytical Method for Plasma and Urine Samples *(Summarized based on the bioanalytical report provided on page 1261-91)*

	Delafloxacin in Plasma	**Delafloxacin in Urine**
Method Validation #	RD-01-005 for Abbott 319492	RD-01-005 for Abbott 319492
Range	8.0 - 5576.0 ng/mL	8.0 - 5576.0 ng/mL
LLOQ	8.0 ng/mL	8.0 ng/mL
Linearity	linear (correlation coefficent $\geq$ 0.9896)	linear (correlation coeffienct $\geq$ 0.99)
Accuracy	85-115%	85-115%
Precision	$\leq$ 15%	$\leq$ 15%
Recovery over range	93.8 - 111.8 %	96.5 - 108%
Stability	stable at room temperature for at least 6 days; stable for 144 days at -20°C;stable through 3 freeze and thaw cycles	stable at room temperature for at least 6 days;stable for 98 days at -20°C;stable through 4 freeze and thaw cycles
Quality Control (QC)	Analytical batches were accepted if at least 2/3 of all prepared QC samples met the acceptance criteria. Two QC sets(high, middle, and low) were made for human plasma samples. Set 1: 4389.5 ng/mL (QC High), 526.7 ng/mL (QC Mid), 21.1 ng/mL (QC low); Set 2: 4410.4 ng/mL (QC Hight), 529.2 ng/mL (QC Mid), 21.2 ng/mL (QC low)	Analytical batches were accepted if at least 2/3 of all prepared QC samples met the acceptance criteria. One QC set (high, middle, and low) was made for human urine samples. Set 1: 4785.3 ng/mL (QC High), 574.2 ng/mL (QC Mid), 23.0 ng/mL (QC low)

Reviewer's Comment: The bioanalytical method and analytical run in this study meet the acceptance criteria specified in the FDA Guidance for Industry Bioanalytical Method Validation.

Pharmacokinetic Assessment:
The pharmacokinetic parameters estimated included: peak plasma concentration (C_{max}), time to reach the peak concentration (T_{max}), minimum plasma concentration (C_{min} – multipledose phase only), area under the plasma concentration-time curve (AUC), terminal elimination half-life ($t_{1/2}$), fraction excreted in the urine (fe). and the apparent oral clearance (CL/F).

Statistical Methods:
Pharmacokinetics:
Part 1: For each dose level, descriptive statistics were provided for the pharmacokinetic variables. To address the question of linear kinetics and dose proportionality, an analysis of covariance (ANCOVA) was performed on dose-normalized C_{max}, T max, terminal elimination rate constant, and dose-normalized AUC. Observations were classified by dose level. Body weight was a covariate; if other variables were found to have explanatory values for a pharmacokinetic variable, they might also have been included as covariates. If the probability distribution of a variable appeared to be nonsymmetric and a transformation had an approximately symmetric distribution, then the transformation was employed. In particular, a transformation of C_{max} and AUC, such as the logarithm, was considered. To test the hypothesis of invariance with dose, two tests were performed within the framework of the ANCOVA. The hypothesis of no difference between the highest and lowest doses was tested. The second test was performed on a contrast in the dose level effects, with the contrast chosen so that there was good power for a trend with dose. Other analyses could have been performed if appropriate.

Part 2: (Food effect, woman, elderly)
The effect of non-fasting conditions was assessed from the data of younger males who received the dose under both fasting and non-fasting conditions in a crossover fashion. An analysis of variance (ANOVA) was performed on the logarithm of Cmax T max and the logarithm of AUC. The model had effects for sequence, subject nested within sequence, period, and dosing condition (fasting or non-fasting). Although this was a largely exploratory study, the two one-sided tests procedure was performed to assess the effect of nonfasting on C_{max} and AUC. The two one-sided tests procedure was carried out via a 90% confidence interval for relative bioavailability as defined by the exponentiation of the difference of logarithm means. The range of equivalence for relative bioavailability for AUC was 0.80 to 1.25, and the corresponding range of equivalence for C_{max} was 0.70 to 1.43. If a variable had a seriously non-normal probability distribution, then alternative methodology was used.

Reviewer's Comment: The food effect was not reviewed since this dosage form is not the to-be-marketed one and the Sponsor has conducted another food effect study for to-be-marketed oral dosage form.

To explore the possibility of gender and age effects, two analyses were performed for pharmacokinetic variables. In the first, a two-way ANOVA was performed on the data of the

younger males under fasting conditions and data of the younger females. The factors were sex and period, and interaction was included. In the second analysis, the data of all doses administered under fasting conditions in Part 2 were included. A two-way ANOVA with classification by gender and age category and with the interaction of the two factors was used. However, the potential for confounding size and sex effects in such an analysis must be realized.

Part 3: Methodology described for Part 1 was employed for the Day 5 variables of the multiple-dose portion of the study. In addition to an analysis of the variables identified for Part 1, an analysis of C_{min} was performed.

A repeated measures analysis was performed on the pre-dose concentrations of Days 3, 4, and 5. The model included effects for dose level, day, the interaction of dose level and day, and, if useful, other explanatory variables.

Safety: No formal statistical analyses were conducted for the safety outcome.

Safety Assessment: Safety was evaluated throughout the study by physical examinations, vital signs, laboratory tests, electrocardiograms (ECGs), and monitoring of adverse events.

RESULTS

Study Population:
No statistically significant differences in demographic characteristics were observed among the dose groups in Part 1 of the study. All subjects were male. Please refer to Table 7 for details.

Table 7. Demographic Characteristics of Subjects in Part 1 of the Study *(Adapted from Table 11.2.a in the study report)*

		ABT-492 Dose and Group						
	Placebo	50 mg: Group A	100 mg: Group B	200 mg: Group C	400 mg: Group D	800 mg: Group E	1200 mg: Group F	1600 mg: Group G
All Randomized	**14**	**6**	**6**	**6**	**6**	**6**	**6**	**6**
Sex								
Male	14 (100%)	6 (100%)	6 (100%)	6 (100%)	6 (100%)	6 (100%)	6 (100%)	6 (100%)
Race								
White	13 (93%)	6 (100%)	4 (67%)	6 (100%)	6 (100%)	6 (100%)	5 (83%)	6 (100%)
Black	1 (7%)	0 (0%)	1 (17%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)
Asian	0 (0%)	0 (0%)	1 (17%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)
Other	0 (0%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)	1 (17%)	0 (0%)
Age (years)								
Mean (SD)	26.4 (6.88)	22.3 (3.01)	26.8 (8.35)	24.5 (3.39)	25.0 (7.21)	23.2 (2.14)	27.2 (5.27)	24.5 (3.39)
Range	19 – 40	19 – 26	19 – 37	21 – 31	20 – 39	20 – 26	20 – 35	20 – 29
Nicotine Use								
Ex-User	3 (21%)	0 (0%)	1 (17%)	2 (33%)	1 (17%)	1 (17%)	0 (0%)	1 (17%)
Non-User	11 (79%)	6 (100%)	5 (83%)	4 (67%)	5 (83%)	5 (83%)	6 (100%)	5 (83%)
Alcohol Use								
User	13 (93%)	6 (100%)	6 (100%)	6 (100%)	6 (100%)	4 (67%)	4 (67%)	6 (100%)
Non-User	1 (7%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)	2 (33%)	2 (33%)	0 (0%)

SD = standard deviation

No statistically significant differences in demographic characteristics were observed in Part 2 of the study among Food Effect, Women, or Elderly subjects. Please refer to Table 8 for the demographic characteristics of subjects in Part 2 of the study.

Table 8. Demographic Characteristics of Subjects in Part 2 of the Study *(Adapted from Table 11.2.b in the study report)*

	Food Effect			Women		Elderly	
		ABT-492 250 mg			ABT-492		ABT-492
	Placebo	AB	BA	Placebo	250 mg	Placebo	250 mg
All Randomized	4	8	8	4	12	4	12
Sex							
Female	0 (0%)	0 (0%)	0 (0%)	4 (100%)	12 (100%)	2 (50%)	6 (50%)
Male	4 (100%)	8 (100%)	8 (100%)	0 (0%)	0 (0%)	2 (50%)	6 (50%)
Race							
White	4 (100%)	8 (100%)	6 (75%)	4 (100%)	12 (100%)	4 (100%)	12 (100%)
Black	0 (0%)	0 (0%)	2 (25%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)
Age (years)							
Mean (SD)	24.5 (3.79)	23.75 (6.27)	22.13 (4.97)	26.8 (8.50)	23.6 (3.80)	77 (5.72)	71.6 (5.05)
Range	22 - 30	19 - 37	18 - 34	21 - 39	18 - 30	71 - 84	65 - 82
Nicotine Use							
Ex-User	0 (0%)	1 (13%)	1 (13%)	2 (50%)	4 (33%)	2 (50%)	6 (50%)
Non-User	4 (100%)	7 (88%)	7 (88%)	2 (50%)	8 (67%)	2 (50%)	6 (50%)
Alcohol Use							
User	4 (100%)	8 (100%)	7 (88%)	2 (50%)	9 (75%)	4 (100%)	11 (92%)
Non-User	0 (0%)	0 (0%)	1 (13%)	2 (50%)	3 (25%)	0 (0%)	1 (8%)

AB: Regimen A in Period 1, Regimen B in Period 2; BA: Regimen B in Period 1, Regimen A in Period 2; Regimen A (single 250 mg dose of ABT-492 under fasted condition); Regimen B (single 250 mg dose of ABT-492 under fed condition)

No statistically significant differences in demographic characteristics were observed among the dose groups in Part 3 of the study. Please refer to Table 9 for the demographic characteristics of subjects in Part 3 of this study.

Table 9. Demographic Characteristics of Subjects in Part 3 of the Study

		ABT-492 Dose and Group				
	Placebo	**100 mg: Group A**	**200 mg: Group B**	**400 mg: Group C**	**800 mg: Group D**	**1200 mg: Group E**
All Randomized	**20**	**8**	**8**	**8**	**8**	**8**
Sex						
Male	20 (100%)	8 (100%)	8 (100%)	8 (100%)	8 (100%)	8 (100%)
Race						
White	19 (95%)	8 (100%)	7 (88%)	7 (88%)	8 (100%)	7 (88%)
Black	1 (5%)	0 (0%)	0 (0%)	1 (13%)	0 (0%)	1 (13%)
Other	0 (0%)	0 (0%)	1 (13%)	0 (0%)	0 (0%)	0 (0%)
Age (years)						
Mean (SD)	25.1 (3.52)	26.8 (6.09)	23.6 (5.78)	26.1 (3.44)	25.8 (1.98)	27.5 (6.16)
Range	20 – 33	21 – 38	20 – 37	21 – 32	23 – 29	19 – 39
Nicotine Use						
Ex-User	4 (20%)	1 (13%)	2 (25%)	1 (13%)	0 (0%)	1 (13%)
Non-User	16 (80%)	7 (88%)	6 (75%)	7 (88%)	8 (100%)	7 (88%)
Alcohol Use						
User	15 (75%)	6 (75%)	7 (88%)	5 (63%)	6 (75%)	5 (63%)
Ex-User	1 (5%)	0 (0%)	1 (13%)	0 (0%)	0 (0%)	0 (0%)
Non-User	4 (20%)	2 (25%)	0 (0%)	3 (38%)	2 (25%)	3 (38%)
SD = standard deviation						

Pharmacokinetics:

The mean± SD pharmacokinetic parameters of ABT-492 after single dose administration of 50 mg to 1600 mg of ABT-492 under fasting conditions are shown in Table 10 below.

Table 10. Summary of Pharmacokinetics of ABT-492 Following Single Rising Doses of ABT - 492 (Part 1 of Study M00-224) *(Adapted from Table 11.5.a of the study report)*

Pharmacokinetic	Dose of ABT-492 (mg)			
Parameters	**50**	**100**	**200**	**400**
N	**6**	**6**	**6**	**6**
T_{max} (h)	1.3 ± 0.3	2.3 ± 0.8	1.3 ± 0.9	1.0 ± 0.0
C_{max} (ng/mL)	949 ± 181	1122 ± 303	2385 ± 620	7220 ± 1263
C_{max}/MIC Ratio†	10.1 ± 1.9	12.0 ± 3.2	25.4 ± 6.6	77.0 ± 13.5
AUC_{∞} (ng•h/mL)	1823 ± 342	3778 ± 954	9342 ± 2007	20910 ± 2963
AUC/MIC Ratio†	19.4 ± 3.6	40.3 ± 10.2	99.6 ± 21.4	223.0 ± 31.6
$t_{1/2}$ (h)‡,$	1.5	2.5	5.9	6.2
CL/F (L/h)†	28.2 ± 5.1	28.1 ± 8.3	22.3 ± 4.8	19.5 ± 3.0
V_{β}/F (L/h)†	66.0 ± 19.4	130.0 ± 82.9	203.3 ± 85.2	207.2 ± 143.6
f_e£,†	35.6 ± 7.0	28.6 ± 4.9	29.2 ± 6.8	23.6 ± 3.7
CL_r (L/h)†	10.1 ± 2.6	8.0 ± 1.6	6.6 ± 2.0	4.7 ± 1.0
	800	1200	1600	
N	6	6	6	
T_{max} (h)	1.8 ± 0.9	1.9 ± 0.8	2.1 ± 0.9	
C_{max} (ng/mL)	9567 ± 2141	13172 ± 2496	16567 ± 3828	
C_{max}/MIC Ratio†	102.0 ± 22.8	140.5 ± 26.6	176.7 ± 40.8	
AUC_{∞} (ng•h/mL)	41415 ± 10765	67149 ± 17161	81674 ± 18516	
AUC/MIC Ratio†	441.8 ± 114.8	716.3 ± 183.1	871.2 ± 197.5	
$t_{1/2}$ (h)‡	7.7	5.5	7.2	
CL/F (L/h)†	20.4 ± 5.0	18.9 ± 4.8	20.4 ± 4.4	
V_{β}/F (L/h)†	285.9 ± 176.3	148.8 ± 25.2	269.3 ± 182.9	
f_e£,†	16.8 ± 2.5	16.7 ± 2.4	12.8 ± 4.1	
CL_r (L/h)†	3.5 ± 0.9	3.0 ± 0.8	2.7 ± 0.8	

† Parameter was not tested statistically.

‡ Harmonic mean; evaluations of $t_{1/2}$ were based on statistical tests for β.

Reviewer's comment:

- *The mean T_{max} of delafloxacin ranged from 1.0 to 2.3 hours following single oral administration of delafloxacin capsule ranging from 50 mg to 1600 mg.*

- *The increase in delafloxacin AUC was approximately dose proportional at single doses ranging from 200 mg to 1600 mg*
- *It was also observed that C_{max} did not increase proportionally with the dose at 50mg and 100 mg.*
- *Delafloxacin was excreted as unchanged drug in the urine following administration of single doses of 50 mg to 1600 mg. The fraction of the dose recovered in the urine (fe) decreased with increasing dose from a median value of 35.6% at the 50 mg dose to 12.8% at the 1600 mg dose.*

Food Effect:
The 90% confidence intervals for relative bioavailability are summarized in Table 11. The confidence intervals were defined by the exponentiation of the difference of logarithm means for AUC and C_{max} of ABT-492 from the analyses of log-transformed data, with fasting conditions as the reference.

Table 11. Relative Bioavailability under Nonfasting vs. Fasting Conditions

Regimens	Pharmacokinetic	Central Value#		Relative Bioavailability	
				Point	90% Confidence
Test *vs.* Reference	Parameter	Test	Reference	Estimate	Interval
B *vs.* A	C_{max}	1781	3527	0.505	0.441 – 0.579
(Nonfasting vs.	AUC_t	10973	12826	0.856	0.795 – 0.921
Fasting)	AUC_∞	11279	13281	0.849	0.795 – 0.907

Antilogarithm of the least squares means for logarithms.

Effect of Age and Sex on ABT-492 Pharmacokinetic Parameters

The PK parameters of ABT-492 after single dose administration of 250 mg ABT-492 to the elderly male and female subjects and to the young female subjects are summarized in Tables 12.

Table 12. The mean± SD pharmacokinetic parameters of ABT-492 after single dose administration of 250 mg of ABT-492 to young males and females and elderly males and females

Pharmacokinetic Parameters (units)	Young Males	Young Females	Elderly Males	Elderly Females
N	16	12	6	6
T_{max} (h)	1.5 ± 1.0	1.4 ± 0.9	1.2 ± 0.4	1.1 ± 0.2
C_{max} (ng/mL)	3582 ± 602*	3890 ± 1265*	4621 ± 793	5297 ± 1057
AUC_{∞} (ng•h/mL)	13592 ± 2849*	14355 ± 2633*	19279 ± 4841	18973 ± 3547
$t_{½}$ (h)‡	8.3	9.0	5.9	7.1
CL/F (L/h)†	19.3 ± 4.8	17.8 ± 2.6	13.7 ± 3.4	13.6 ± 2.5
V_{β}/F (L)†	444.4 ± 605.2	550.6 ± 730.8	158.3 ± 146.6	164.6 ± 84.7

* Statistically significantly different from elderly subjects (ANOVA, $p < 0.05$).
‡ Harmonic mean; evaluations of $t_{½}$ were based on statistical tests for β.
† Parameter was not tested statistically.

Reviewer's comment: It seems that there was little effect of gender on delafloxacin pharmacokinetics in either the young or the elderly populations. However, the mean C_{max} and AUC_{inf} values were statistically lower after oral administration of 250 mg delafloxacin to young subjects compared to elderly subjects ($p<0.05$, ANOVA, Table 12). No other demographic difference was identified by the reviewer between the young and elder groups (e.g. weight, nicotine, alcohol). The 46% increase in AUC_{inf} and 38% increase in C_{max} in the older subjects may be explained by a decrease in estimated creatinine clearance due to aging.

Multiple-Dose Escalation/Administration

The mean PK parameters of ABT-492 after single- and multiple-dose administration of 100, 200,400, 800 and 1200 mg of ABT-492 are summarized in Table 13 below.

Table 13. The mean (SD) pharmacokinetic parameters of ABT-492 after single (Day I) and multiple dose (Day 5) administration of 100 mg to 1200 mg of ABT-492

Day 1					
Pharmacokinetic	Dose of ABT-492 (mg)				
Parameters	100	200	400	800	1200
N	8	8	8	8	8
T_{max} (h)	1.1 ± 0.8	1.0 ± 0.7	1.1 ± 0.4	1.1 ± 0.4	1.7 ± 1.1
C_{max} (ng/mL)	1364 ± 452	3168 ± 699	4492 ± 965	8184 ± 2357	11467 ± 2961
C_{max}/MIC Ratio†	14.5 ± 4.8	33.8 ± 7.5	47.9 ± 10.3	87.3 ± 25.1	122.3 ± 31.6
AUC_{∞} (ng•h/mL)	4057 ± 576	11241 ± 1260	14807 ± 2133	27977 ± 8133	50618 ± 8918
AUC/MIC Ratio†	43.3 ± 6.1	119.9 ± 13.4	157.9 ± 22.8	298.4 ± 86.7	539.9 ± 95.1
$t_{½}$ (h)‡	3.2	3.8	4.1	4.3	4.5
CL/F (L/h)†	25.1 ± 3.7	18.0 ± 2.0	27.5 ± 4.0	30.5 ± 7.9	24.4 ± 4.4
V_{β}/F (L)†	121.1 ± 32.2	110.2 ± 39.8	210.4 ± 139.6	211.1 ± 89.1	171.8 ± 77.4
Day 5					
Pharmacokinetic	QD Dose of ABT-492 (mg)				
Parameters	100	200	400	800	1200
N	8	8	8	8	5
T_{max} (h)	1.1 ± 0.7	1.8 ± 1.4	1.1 ± 0.4	1.1 ± 0.4	1.4 ± 0.5
C_{max} (ng/mL)	1325 ± 460	3069 ± 810	4954 ± 947	8174 ± 2156	11482 ± 2024
C_{max}/MIC Ratio†	14.1 ± 4.9	32.7 ± 8.6	52.8 ± 10.1	87.2 ± 23.0	122.5 ± 21.6
C_{min} (ng/mL)	10.5 ± 10.5	39.5 ± 14.1	52.3 ± 18.8	96.3 ± 30.1	178.7 ± 63.4
AUC_{24} (ng•h/mL)	4456 ± 822	11507 ± 1848	17770 ± 3403	31406 ± 8576	54289 ± 8038
AUC/MIC Ratio†	47.5 ± 8.8	122.7 ± 19.7	189.5 ± 36.3	335.0 ± 91.5	579.1 ± 85.7
$t_{½}$ (h)‡	4.2	7.4	8.4	8.5	7.6
CL/F (L/h)†	23.1 ± 4.1	17.7 ± 2.6	23.3 ± 4.8	27.1 ± 7.2	22.5 ± 3.6
V_{β}/F (L)†	154.1 ± 60.6	235.9 ± 142.2	347.0 ± 222.5	386.6 ± 197.5	315.1 ± 186.4
AR£,†	1.10 ± 0.07	1.03 ± 0.12	1.24 ± 0.33	1.15 ± 0.13	1.18 ± 0.10
f_e¢,†	29.3 ± 13.5	29.4 ± 11.3	23.9 ± 14.4	18.9 ± 25.9	23.9 ± 9.3
CL_r (L/h)†	5.9 ± 3.2	5.9 ± 2.3	6.2 ± 4.0	7.1 ± 8.1	4.2 ± 2.0

† Parameter was not tested statistically.
‡ Harmonic mean; evaluations of $t_{½}$ were based on statistical tests for β.
£ Day 5 AUC_{24} / Day 1 AUC_{24}.
¢ Median ± SD values presented.

Reviewer's comments:

- *Following multiple-dose (QD) administration of delafloxacin capsules, the increase in delafloxacin AUC_{24} on Day 5 was approximately dose proportional from 400 mg to 1200 mg.*
- *Little accumulation of delafloxacin was observed following oral administration of delafloxacin once daily for 5 days. The mean C_{max} values were similar on Days 1 and 5. The mean of Day 5 AUC_{24}/Day 1 AUC_{24} ranged from 1.03 to 1.24 (AR in Table 13).*
- *However, it was observed that the harmonic mean half-lives of delafloxacin almost doubled from Day 1 to Day 5. The Sponsor stated that it may be due to the enterohepatic cycling of delafloxacin as secondary peak plasma concentrations were observed in many subjects. These secondary peaks added uncertainty to the determination of the terminal log-linear portion of the plasma concentration-time profile.*
- *The total urinary recoveries of delafloxacin over the 24-hour dosing interval on Day 5 after multiple dosing of 100 mg to 1200 mg QD of delafloxacin remained approximately*

constant across this dose range. The Sponsor stated that the equilibrium solubility of ABT -492 in pH 5.0 buffer at 25°C is 3.0 ng/mL. The solubility increases markedly with increasing pH, reaching 66.9 ng/mL in pH 6.8 buffer and 283.6 ng/mL in pH 7.4 buffer. The individual urinary pH values for the subjects in this study were typically 5 to 7. Therefore, the observed decrease in urinary recovery with increasing dose may be due to the pH variability.

Visual inspection of the mean plasma trough concentrations of ABT-492 suggests that steady state for oral capsule was achieved after approximately 3 days of multiple dosing. Statistical analyses showed that the mean trough concentrations on Day 3 and 4 were not statistically different from those on Day 5 ($p > 0.1527$).

Safety:

Part 1

- The incidence of all treatment-emergent adverse events was highest in the ABT-492 1200 mg (Group F; 5/6, 83%) and 1600 mg (Group G; 3/6, 50%) dose groups.
- Overall, the most commonly reported treatment -emergent adverse events in the ABT-492 dose groups were headache, nausea and diarrhea;
- All cases of diarrhea and all but one case of nausea (800 mg group) were reported by subjects who received the two highest doses of ABT-492 (1200 mg and 1600 mg).
- No serious adverse events were reported during Part I of the study and no subject prematurely discontinued the study due to adverse events.

Part 2

- In Part 2 (Food Effect), one or more treatment-emergent adverse events were reported by 5 of 16 (31 %) subjects during Regimen A (fasted condition) and 4 of 16 (25%) subjects during Regimen B (fed condition); no placebo-treated subjects (Regimens C and D) reported adverse events.
- Among both Women and Elderly subjects, 5 of 12 (42%) subjects who took ABT-492 and I of 4 (25%) subjects who took placebo reported at least one treatment-emergent adverse event.
- Overall, the most frequently occurring study drug-related adverse events in Part 2 of the study were diarrhea and headache.

Part 3

- The incidence of all treatment-emergent adverse events was highest in the ABT-492 800 mg (Group D; 7/8, 88%) and 1200 mg (Group E; 7/8, 88%) dose groups.
- A clear dose response was not observed, although 50% of subjects in both the 800 mg and 1200 mg dose groups reported diarrhea.

SPONSOR'S CONCLUSIONS

- ABT-492 was safe and generally well tolerated when administered across a wide dosing interval, although gastrointestinal side effects were observed more commonly at doses of 1200 mg and above. Feeding status, gender, and age did not appear to significantly affect the safety profile.
- In Part 1 (single-dose) of the study, the increases in the AUC~ values were proportional to dose for doses of 200 mg or more. The mean AUC/MIC ratio exceeded 30 at a dose of 100 mg and approached 100 at a dose of 200 mg ABT-492.

- In Part 2 (Food Effect), ABT-492 was better absorbed when administered under fasting conditions than when administered with a high fat meal. In Part 2 (Female and Elderly), there were no significant differences between males and females in any of the ABT-492 pharmacokinetic parameters; in the elderly subjects, the mean ABT-492 C_{max} and AUC values were about 35% greater than those observed for the young subjects.
- In Part 3 (multiple-dose) of the study, little accumulation of ABT-492 was apparent following administration of ABT-492 once daily for 5 days. The mean C_{max} values were similar on Days 1 and 5, and the mean C_{min} values were less than 2% of the corresponding mean C_{max} values. Steady state was achieved by the morning of Day 3. On Day 5, the mean AUC/MIC ratios for quinolone-sensitive Streptococcus pneumoniae approached 50 for the 100 mg QD regimen and exceeded 120 for the 200 mg QD regimen.

REVIEWER ASSESSMENT:

- Based on data from Part 1 and Part 3, it seems that the increase in AUC of delafloxacin following administration of the oral capsule was approximately dose proportional in the dose range of 200 mg to 1600 mg for single dose, and 400 mg to 1200 mg for QD multiple dose.
- There were no significant differences between males and females in any of the delafloxacin pharmacokinetic parameters; in the elderly subjects, the mean delafloxacin C_{max} and AUC values were about 35% and 46% greater than those observed for the young subjects, respectively.
- Little accumulation of delafloxacin was observed following administration of delafloxacin once daily for 5 days.

4.5.3 Study No.: M01-284 4.5.3

Effect of ABT-492 (Delafloxacin) on Photosensitivity Levels in Healthy Male and Female Subjects

Date(s): 12 July 2001 to 04 October 2001
Sponsor: Abbott Laboratories, 200 Abbott Park Rd., Abbott Park, IL 60064-6145
Clinical Site: Single site: DDS Medicines Research, LTD, Ninewells Hospital and Medical School, Dundee, Scotland DD1 9SY
Analytical Site: Abbott Laboratories, Abbott Park, IL.

Objective:
The objective of this study was to evaluate if ABT-492 had any photosensitizing potential in healthy male and female subjects

Study Design:
This was a Phase 1, single-blind, placebo- and positive-controlled, randomized, parallel-group study in 52 healthy male and female volunteers. The study was designed to demonstrate the photosensitizing potential and wavelength dependency of ABT-492, by comparing the response of the skin to ultraviolet A (UVA), ultraviolet B (UVB) and visible radiation prior to and during administration of ABT-492, positive control (lomefloxacin), or negative control (placebo).

Within 3 weeks prior to dosing, subjects were screened to verify compliance with the inclusion/exclusion criteria and underwent baseline photosensitivity testing to UV and visible radiation (by means of a standard monochromator technique) over a period of 3 days; the endpoint used at each wavelength tested was the minimal erythema dose (MED). Eligible subjects were admitted to an ultraviolet- and blue light-screened clinical unit (DDS Medicines Research Limited) and randomly assigned to study drug in a 1: 1: 1: 1 ratio (as shown in the table below), to receive ABT-492 (200 mg/day or 400 mg/day), placebo, or lomefloxacin 400 mg/day for 6 days. On Study Days 5-6, phototesting was repeated during which each subject had his/her back exposed to selected wavelengths of UVA, UVB and visible light. On Study Day 7, exposed areas were read and the MED was determined. If an abnormal response (reduction in MED of>40% from baseline) was revealed on Study Day 7, phototesting was to be repeated on Study Days 7-9; subjects remained in the DDS until any drug induced photosensitivity had resolved. Safety was assessed by 12-lead electrocardiograms (ECGs), physical examination, laboratory parameters, vital signs, and monitoring of adverse events.

Number of Subjects (Planned and Analyzed):	ABT-492 200 mg/day	ABT-492 400 mg/day	Lomefloxacin 400 mg/day	Placebo
Number of Subjects Planned	12	12	12	12
Number of Subjects Randomized and Dosed	13	13	13	13
Number of Subjects Who Completed the Study	12	11	10	12

Drug Product:
ABT-492 capsules; 200 mg QD (100 mg capsule x 2) or 400 mg QD (100 mg capsule x 4); administered under fasting conditions (no food for 2 hours before and 2 hours after study drug administration); Bulk Lot Number: 68-398-AR

Statistical Methods:
The primary endpoint was the change in MED within subject/group comparing their baseline with on drug/placebo value. A secondary analysis calculated the Phototoxic Index (Pl), obtained by dividing the baseline MED value for each individual, and the median MED value for each group, by the post-dose MED value. Data for each waveband tested were analyzed separately; the maximum PI indicated the phototoxic potential of the study drug.

Safety:
The number and percentage of subjects having treatment-emergent adverse events were tabulated by COST ART term and body system. Laboratory test values outside the normal range were flagged in the data listings and evaluated for clinical significance by the investigator. Vital signs and ECG data were summarized.

Results:
A total of 52 subjects were randomized, and 45 subjects completed the study. A total of 4 subjects prematurely terminated due to AEs, 2 subjects withdrew consent, and 1 subject did not return for a study visit. The majority of subjects was male (65%) and White (100%). The mean age was 33.7 years (ranging from 18 to 54 years). The mean subject body weight was 74.8 kg (ranging from 51 to 97 kg). There were no statistically significant differences among the dosing groups with regard to sex, race, age, or weight.

As shown in Table 1, ABT-492 at 200 and 400 mg/day did not demonstrate clinically significant phototoxic potential at any wavelengths tested (295 - 430 nm), while the active comparator, lomefloxacin, demonstrated a moderate degree of phototoxicity at UVA wavelengths 335 nm and 365 nm. At these same wavelengths, statistically significant differences in percent change from baseline in MED were also seen when lomefloxacin was compared to both ABT-492 dosing groups and the placebo group. Substantially higher mean and median phototoxic index values were also demonstrated by the lomefloxacin group compared to the other 3 dosing groups at wavelengths of 335 nm and 365 nm; the maximum PI in the lomefloxacin group at these wavelengths (6.8 and 10.0, respectively) greatly exceeded those in the other 3 dosing groups (1.4 and 1.4, respectively in the ABT-492 200 mg dosing group, 2.1and1.5, respectively, in the ABT-492 400 mg dosing group, and 1.8 and 1.5, respectively, in the placebo group). No statistically significant differences in percent change from baseline in MED were observed at each

wavelength tested within either ABT-492 dosing group or between the ABT-492 dosing groups and the placebo group, using the monochromator or solar simulator.

Table 1. Mean Percent Change From Baseline to Day 7 in Minimal Erythema Dose by Monochromator Wavelength and Solar Simulator (Adapted from Table 43 in the Summary of Clinical Pharmacology)

Treatment Group	Mean Percent Change (SD)	P-value Comparing MED Within Group[a]	P-value vs. Placebo[b]	P-value vs. Lomefloxacin[b]
Wavelength: 295 ± 5 nm				
Delafloxacin 200 mg (N = 12)	–0.4 (17.43)	0.492	> 0.999	0.612
Delafloxacin 400 mg (N = 11)	11.9 (18.37)	0.094	0.245	0.328
Lomefloxacin (N = 12)	5.8 (18.35)	0.313	0.665	
Placebo (N = 12)	0.7 (14.46)	0.438		
Wavelength: 300 ± 5 nm				
Delafloxacin 200 mg (N = 12)	–6.0 (8.86)	0.125	0.973	0.561
Delafloxacin 400 mg (N = 11)	–7.9 (11.74)	0.125	0.885	0.475
Lomefloxacin (N = 12)	–3.9 (15.22)	0.750	0.561	
Placebo (N = 12)	–7.1 (11.11)	0.125		
Wavelength: 305 ± 5 nm				
Delafloxacin 200 mg (N = 12)	–4.8 (14.43)	0.375	0.715	0.903
Delafloxacin 400 mg (N = 11)	–5.8 (20.65)	0.406	0.614	0.614
Lomefloxacin (N = 12)	–3.2 (21.06)	0.984	0.954	
Placebo (N = 12)	–1.2 (18.47)	0.711		

Treatment Group	Mean Percent Change (SD)	P-value Comparing MED Within Group[a]	P-value vs. Placebo[b]	P-value vs. Lomefloxacin[b]
Wavelength: 335 ± 30 nm				
Delafloxacin 200 mg (N = 12)	–1.4 (18.89)	0.723	0.351	< 0.001[c]
Delafloxacin 400 mg (N = 11)	0.0 (31.52)	> 0.999	0.419	< 0.001[c]
Lomefloxacin (N = 12)	–64.0 (17.11)	< 0.001[c]	< 0.001[c]	
Placebo (N = 12)	–11.4 (20.08)	0.184		
Wavelength: 365 ± 30 nm				
Delafloxacin 200 mg (N = 12)	–6.2 (16.66)	0.516	0.703	< 0.001[c]
Delafloxacin 400 mg (N = 11)	–7.1 (14.81)	0.281	0.875	< 0.001[c]
Lomefloxacin (N = 12)	–76.0 (12.94)	< 0.001[c]	< 0.001[c]	
Placebo (N = 12)	–7.2 (20.71)	0.422		
Wavelength: 400 ± 30 nm				
Delafloxacin 200 mg (N = 12)	–2.8 (6.57)	0.500	0.166	1.000
Delafloxacin 400 mg (N = 11)	–9.1 (17.43)	0.250	0.066	0.523
Lomefloxacin (N = 12)	–4.0 (9.88)	0.500	0.166	
Placebo (N = 12)	0.0 (0.00)			
Wavelength: 430 ± 30 nm				
Delafloxacin 200 mg (N = 12)	0.0 (0.0)		N/A	N/A
Delafloxacin 400 mg (N = 11)	0.0 (0.0)		N/A	N/A
Lomefloxacin (N = 12)	0.0 (0.0)		N/A	
Placebo (N = 12)	0.0 (0.0)			
Solar Simulator				
Delafloxacin 200 mg (N = 12)	5.3 (12.89)	0.250	0.664	0.014[c]
Delafloxacin 400 mg (N = 11)	–0.2 (20.48)	> 0.999	0.177	0.119
Lomefloxacin (N = 12)	–15.3 (19.69)	0.039[c]	0.012[c]	
Placebo (N = 12)	6.5 (15.34)	0.078		

MED = minimum erythema dose; MED = minimal erythema dose; NA = not available; SD = standard deviation

[a] P-value comparing MED to baseline within treatment groups using Wilcoxon signed rank test.

[b] P-value comparing MED between treatment groups using Wilcoxon rank sum test.

[c] Statistically significant different ($p \leq 0.05$).

ABT-492 at dosages of 200 and 400 mg/day failed to demonstrate a significant phototoxic effect. In particular, the classical pattern of fluoroquinolone phototoxicity as detected in previous phototoxicity studies (i.e., a UV A phenomenon maximal at 24 hours) was not seen. Lomefloxacin, however, revealed phototoxicity as expected within the moderate phototoxic index group at 335 and 365 ± 30 nm test groups, maximal at 24 hours, with susceptibility clearing within 48 hours after drug cessation. Phototoxicity was not demonstrated in the placebo group.

One or more treatment-emergent adverse events were reported by 7 (54%) subjects in the ABT-492 200 mg group, 9 (69%) subjects in the ABT-492 400 mg group, 4 (31 %) subjects in the lomefloxacin group, and 10 (77%) subjects in the placebo group. Excluding events judged not related or probably not related to study drug, 23% (3/13) of subjects in the ABT-492 200 mg group, 62% (8/13) subjects in the ABT-492 400 mg group, 15% (2/13) of subjects in the lomefloxacin group, and 38% (5/13) of subjects in the placebo group reported at least one adverse event considered possibly or probably related to study drug therapy. In the ABT-492 400 mg group, drug-related diarrhea was reported by 5 (38%) subjects and abdominal pain was reported by 3 (23%) subjects; all other drug-related adverse events were reported by 2 (16.7%) subjects in any dosing group.

No deaths or serious adverse events were reported during the conduct of this study. Four (4) subjects (1 ABT-492 200 mg, 2 ABT-492 400 mg, 1 placebo) were prematurely discontinued from study drug due to the occurrence of at least one adverse event; 3 of these subjects withdrew due to adverse events considered possibly or probably related to study drug (headache in an ABT-492 400 mg subject; diarrhea and abdominal pain in an ABT-492 400 mg subject; migraine, myasthenia and dizziness in a placebo subject).

Conclusions:
Both doses of ABT-492 were safe and well tolerated in healthy adult volunteers. The phototoxicity potential of ABT-492 was similar to that of placebo.

Reviewer's Comment:

- No PK samples were collected for this study and no bioassay was conducted.
- The delafloxacin doses used in this study were lower than the therapeutic dose. However, since no photosensitivity cases were observed in clinical studies in this application (One patient reported symptoms of photosensitivity after completing the study. The Sponsor concluded that it was not treatment related. Please refer to the clinical review by Dr. Caroline Jjingo for details.), combining with results from this study result, the photosensitivity risk appears to be low for delafloxacin.

4.5.4 STUDY NO.: M01-292

Metabolism and Disposition of [14C]ABT-492 (Delafloxacin Oral Capsule) in Man Following a Single Oral Administration

Date(s): 04 June, 2001 to 16 July 2001
Sponsor: Abbott Laboratories, 200 Abbott Park Rd, Abbott Park, IL 60064-6145
Clinical Site: Single site: Janet Dickson, M.D., Inveresk Research, Research Park, Riccarton, Edinburgh EH14 4AP, Scotland
Analytical Site: (b) (4)

OBJECTIVE(S):

- The primary objective of this study was to investigate the metabolism and disposition of [14C]ABT-492 in healthy adult male subjects following a single oral dose.

METHODS

Study Design: This was an open-label, single-center study in 6 subjects to investigate the metabolism and disposition of [14C]ABT-492. Each subject received a single oral administration of [14C]ABT-492 at a dose level of 200 mg (two capsules of 100 mg) in a formulation comprised of bulk drug (N-methyl glucamine salt). The radioactive dose level was 100 µCi of [14C]ABT-492 per subject. To allow measurement of metabolism and disposition, excreta and blood were collected to 168 hours post-dose.

Drug Product:
[14C]ABT-492 (100 mg of ABT-492 [delafloxacin]/capsule and 50 µCi/capsule) capsules; 200 mg (100 mg capsule x 2); administered under fasting conditions (no food for 12 hours before and 2.5 hours after study drug administration); Bulk Lot Number: 75-573-S2

PK Sample Collection:
Please refer to Table 1 for the plasma, urine, feces sampling schedule.

Table 1. Schedule of Assessments

Study Procedure	Screening	Study Day											
	Up to 2 weeks prior to Day -1	-1	1	2	3	4	5	6	7	8[a]	9	10	11
Confinement		x	x	x	x	x	x	x	x	x	x	x	x
Informed consent	x												
Medical history	x												
Physical examination	x	x[b]											x[b,c]
Vital signs	x	x	x[d]	x	x	x	x	x	x	x	x	x	x
12-lead ECG	x	x	x[e]	x[f]									x[c]
Blood chemistry and hematology	x	x								x			x[c]
Coagulation	x		x[g]										x[c]
Dose (after 12 hour fast)			x										
Urinalysis	x	x								x			x[c]
Urine drug and alcohol screen	x	x											
Hepatitis B and C screen	x												
Blood collection for ABT-492 Assay[h,i]			x	x	x	x	x	x	x	x	x	x	x
Urine collection[i,j]		x	x	x	x	x	x	x	x	x	x	x	x
Fecal collection[i,k]		x	x	x	x	x	x	x	x	x	x	x	x

[a] Final study day and end of collection interval if 90% of the administered radioactivity had been recovered at Study Day 8 (168 hours).
[b] Brief physical exam including weight.
[c] Assessments could have been performed at Study Day 8 (168 hours) if 90% of the administered radioactivity had been recovered.
[d] Pre-dose and at 3 hours post-dose.
[e] At 1.5 hours post-dose.
[f] At 24 hours post-dose.
[g] Pre-dose and at 30-60 minutes post-dose.
[h] Pre-dose and at 0.25, 0.5, 0.75, 1, 1.5, 2, 3, 4, 6, 8, 10, 12, 24, 48, 72, 96, 120, 144, and 168 hours post dose.
[i] Samples were not collected after Study Day 8 (168 hours) if 90% of the administered radioactivity had been recovered.
[j] Pre-dose and for the following intervals: 0-2, 2-4, 4-6, 6-8, 8-12, 12-24, 24-48, 48-72, 72-96, 96-120, 120-144, and 144-168 hours post-dose.
[k] Pre-dose, and at intervals of 0-24, 24-48, 48-72, 72-96, 96-120, 120-144 and 144-168 hours post-dose.

Analytical Methods:
The Sponsor stated that the analytical phases of the study were conducted in accordance with the OECD Principles of Good Laboratory Practice as set forth by the United Kingdom Department of Health and as accepted by Regulatory Authorities throughout the European Community, United States of America (FDA and EPA), and Japan (MHW, MAFF, and MITI). The clinical phase was conducted in accordance with ICH Guideline for Good Clinical (Research) Practice and applicable regulatory requirements outlined in the Dec1aration of Helsinki (2000 version). Because this work was conducted in support of a clinical study, no formal claim of GLP compliance could be made.

The detail analytical method for the metabolism and disposition of [14C] ABT-492 was reported in Method for the Determination of the Radiochemical Purity and Radioactivity of Abbott-319492 Capsule (100 mg) with 14C-Labeled Abbott-319492 (50 μCi) in the appendix of this study report.

Mass Balance and Pharmacokinetic Assessment:
Plasma parameters included the following:

- AUC_{0-t}: Area under the plasma concentration versus time curve from time 0 to the time of the last quantifiable concentration
- AUC_{0-inf}: Area under the plasma concentration versus time curve from time 0 extrapolated to infinity
- AUC_{0-t}: Area under the concentration vs time curve from time zero to 't' hours, where 't' = the time point of the last sample on the PK profile at which the analyte was quantifiable.
- C_{max}: Maximum observed plasma concentration
- $t_{1/2}$: Terminal elimination half-life
- T_{max}: Time to achieve maximum observed plasma concentration
- CL: clearance

Statistical Methods:
No formal statistical analysis was performed.

Safety Assessment: Safety was assessed by analysis of adverse events (AEs), laboratory parameters, vital signs, physical examinations and electrocardiograms (ECGs).

RESULTS

Study Population:
All subjects received 1 dose of Investigational medicinal product (IMP) as planned and were included in the safety analysis population. Please refer to Table 2 for the demographic characteristics of the study population.

Table 2. Demographic Characteristics: All subjects (*Adapted from Table 14.1-1.1 in the study report)*

VARIABLE	N	MEAN	SE	SD	MEDIAN	MIN	MAX
AGE (YEARS)	6	44.3	2.57	7.12	44.5	36.0	52.0
HEIGHT (CM)	6	173.7	2.57	2.34	173.5	170.0	177.0
WEIGHT# (KG)	6	77.3	2.57	7.94	75.5	68.0	92.0

SEX				RACE							
FEMALE		MALE		BLACK		WHITE		ASIAN		OTHER	
N	(%)	N	(%)	N	(%)	N	(%)	N	(%)	N	(%)
0	(0)	6	(100)	0	(0)	6	(100)	0	(0)	0	(0)

Pharmacokinetics in Plasma:
The metabolism and disposition of ABT-492 were studied in 6 fasted adult male human subjects following oral administration of a single 200 mg dose of [14C]ABT-492. Radioactivity was rapidly absorbed after drug administration, with peak concentrations in the plasma occurring within 45 minutes in 5 of the 6 subjects (Tmax range = 0. 7 5-1.0 hour; mean= 0.79 hour). Mean levels of radioactivity in the plasma achieved a peak concentration of 5.57 µg eq/mL (C_{max} range

= 2.80-7.30 µg eq/mL) and then rapidly declined to 3.58, 2.14, and 0.65 µg eq/mL at 1.5, 3, and 6 hours, respectively. At 12 hours after dosing, mean plasma radioactivity concentrations had declined to 0.11 µg eq/mL and by 48 hours post-dose, all concentrations were below the limit of quantification. Total plasma radioactivity $AUC_{0-\infty}$ values averaged 17.01 µg eq•h/mL (range of 14.79-21.58 µg eq•h/mL), while the plasma half-life ranged from 3.71-4.07 hours (mean 3.85 hours).

Unchanged parent drug was the predominant plasma component after oral administration of [14C]ABT-492. ABT-492 represented a mean of 90.41 % of the plasma radioactivity 0.5 hours post-dose, which decreased to 79.87% after 3 hours. The major plasma metabolite was M3, an ester glucuronide of parent, which typically accounted for the remaining radioactivity. Small amounts of minor metabolites, M5 and M6A, were occasionally detected in some subjects.

Total radioactivity was rapidly eliminated, with an average of 92.05% of the administered dose excreted within 3 days. Total 7-day recoveries averaged 97 .83%, with an average of 50.19% of the radioactive dose recovered in the urine and 47.65% eliminated in the feces.

Urinary metabolite profiles indicated that unchanged ABT-492 was the major component, representing a mean of 69.79% of the total urinary radioactivity 24 hours post-dose. The remaining radioactivity was accounted for as the metabolites, M3 (ester glucuronide), M5 (ester glucuronide), M6A (ether glucuronide), and M7 (unknown), which represented 26.36%, 4.04%, 3.09%, and 2.02% of the urinary radioactivity, respectively. The metabolite profiles observed from feces clearly indicated that unchanged parent drug was the only component. This study demonstrated that unchanged parent drug is the predominant component in the plasma of subjects given ABT-492 and that greater than 80% of the administered dose is eliminated as unchanged parent drug (35.16% and 47.65%, urine and feces, respectively). The major metabolites in the urine resulted from direct glucuronidation of parent drug to form ester glucuronides.

Although these metabolites were not observed in the feces, previous in vivo animal metabolism indicated that, following biliary secretion, hydrolysis of the ester glucuronide of ABT-492 in the gut would result in the excretion of parent drug. This hydrolysis may be spontaneous and/or enzymatic but also may be due to the analytical methodology and the inherent instability of the acyl glucuronide. Hence, following a single oral administration (200 mg) of ABT-492 to human subjects, glucuronidation is considered the major pathway of metabolism.

Safety:
Three of the 6 subjects had 1 adverse event each. One subject had increased SGOT and 2 subjects had increased SGPT; elevations were approximately 2 x the upper limit of normal (ULN). All adverse events were mild in intensity and resolved. No deaths, serious adverse events, or discontinuations due to adverse events were reported.

SPONSOR'S CONCLUSIONS
In conclusion, following a single oral administration (200 mg) of ABT-492 to human subjects, absorption and elimination were rapid, with the dose being equally excreted via the urine and

feces. Glucuronidation was considered the major pathway of metabolism. [14C]ABT-492 was well tolerated in all subjects.

REVIEWER ASSESSMENT:
The Sponsor did not describe their findings, especially the identified metabolites of delafloxacin, in detail, in this study report probably because this study was conducted in the early stage of delafloxacin development (2002). However, the overall findings in this study are consistent with the results of mass balance study after IV infusion.

4.5.5 Study No.: ML-3341-118

A PHASE 1 STUDY TO EVALUATE THE EFFECT OF REPEATED ORAL DOSES OF DELAFLOXACIN ON THE PHARMACOKINETICS OF A SINGLE ORAL DOSE OF MIDAZOLAM IN HEALTHY SUBJECTS

Date(s): 09 June 2015 to 20 July 2015
Sponsor: Melinta Therapeutics, Inc, 300 George Street, Suite 301, New Haven, CT 06511
Clinical Site: Single site: PPD Phase I Clinic, located at 7551 Metro Center Drive, Suite 200, Austin, TX 78744
Analytical Site: (b) (4)

OBJECTIVE(S):

- The primary objective was to evaluate the effect of repeated doses of oral delafloxacin on the pharmacokinetic (PK) profile of a single oral dose of midazolam.
- The secondary objectives were to evaluate the pharmacokinetics, safety, and tolerability of repeated oral doses of delafloxacin in healthy male and female subjects and to obtain a steady-state PK profile for oral delafloxacin.

METHODS

Study Design: This was a Phase 1, nonrandomized, open-label study designed to evaluate the effect of repeated oral doses of delafloxacin on the PK profile of a single oral dose of midazolam in healthy subjects.

This study was a single sequence, single group study in which 22 male and female subjects were enrolled. Each subject received a single 5-mg oral dose of midazolam on Day 1, oral delafloxacin (450-mg) twice-daily (every 12 hours [q12h]) doses on Days 3 to 8, and coadministered a single 5-mg oral dose of midazolam in the morning on Day 8.

Serial blood samples for PK analysis were drawn from all subjects before dosing and for up to 24 hours after dosing on Days 1, 3, 7, and 8. Blood samples were also collected just before the morning dose of delafloxacin on Days 4, 5, and 6.

Safety and tolerability were assessed by monitoring and recording of adverse events (AEs); clinical laboratory results including hematology, coagulation, serum chemistry, and urinalysis; vital sign (systolic and diastolic blood pressures, heart rate, respiratory rate, and body temperature) and pulse oximetry measurements, 12-lead electrocardiogram (ECG) results; and physical examination findings.

Drug Product: Delafloxacin 450 mg tablets; Dose: 450 mg (q12h);

Analytical Methods:
The method used to measure delafloxacin in human plasma by HPLC with MS/MS detection was validated under Project code "OZZ2" (Table 1).

Table 1. Validation Method for Quantification of Delafloxacin

(b) (4)			
Project Code	OZZ2		
Method ID	LCMSC 634		
Analytes	Delafloxacin		
Matrix	Human Plasma		
Anticoagulant	Dipotassium EDTA		
Method Description	Supported liquid phase extraction		
Sample Volume (µL)	100-µL		
Sample Storage Temperature	-20 °C or colder		
Internal Standard (IS)	Delafloxacin-d_5		
Regression, Weighting	Linear, 1/concentration^2		
Average Recovery of Drug (%)	71.9%		
Average Recovery of IS (%)	70.7%		
Standard Curve Concentrations	5.00 to 5000 ng/mL		
QC Concentrations	5.00, 15.0, 40.0, 150, 600, and 3750 ng/mL		
QC Intra-assay Statistics (%)	Conc. (ng/mL)	Precision	Accuracy
	5.00	3.36 to 5.36%	-0.888 to 3.91%
	15.0	1.83 to 2.18%	-1.90 to 2.98%
	40.0	0.794 to 1.74%	-0.267 to 1.80%
	150	1.42 to 2.56%	-1.11 to 2.69%
	600	1.21 to 1.93%	0.821 to 2.20%
	3750	1.28 to 4.13%	0.192 to 2.02%
QC Inter-assay Statistics (%)	Conc. (ng/mL)	Precision	Accuracy
	5.00	4.49%	0.872%
	15.0	2.79%	0.730%
	40.0	1.54%	0.747%
	150	2.49%	0.593%
	600	1.71%	1.60%
	3750	2.58%	1.02%
Thawed Matrix Stability (hrs)	24 hours at room temperature		
Solution Stability (days)	Delafloxacin 35 days at 2 to 8 °C in standard diluent* Delafloxacin-d_5 35 days at 2 to 8 °C in standard diluent*		
Solution Stress Stability (hours)	Delafloxacin 7 hours at room temperature in 50:50 Acetonitrile / Water* Delafloxacin-d_5 7 hours at room temperature in 50:50 Acetonitrile / Water*		
Extract Stability (hrs)	167 hours at 2 to 8 °C		
Freeze-thaw Stability (cycles)	Five cycles thawed at room temperature		
Frozen Matrix Storage Stability (days)	29 days at -20 °C and -70 °C		
Whole Blood Stability	N/A		
Dilutional Linearity	150 ng/mL diluted four-fold 10000 ng/mL diluted ten-fold		
Selectivity	No significant interfering peaks noted in blank plasma samples		
Hemolysis	No effect from hemolysis on the quantitation of delafloxacin		
Lipemia	No effect from lipemia on the quantitation of delafloxacin		

* Established prior to/following the validation

Reviewer's Comment: Based on the method validation and description of the Run Acceptance Criteria, both analytical method and runs met the acceptable criteria specified in the FDA Guidance to Industry: Bioanalytical Method Validation.

Project samples were analyzed according to (b) (4) Method LCMSC 97 V 1.00, entitled "Quantitation of Midazolam and 1'-Hydroxymidazolam in Human Plasma via HPLC with MS/MS Detection," which was validated under Project Code "XZX2" (Table 2). All samples were analyzed within the 852 days demonstrated long-term storage stability in human plasma containing sodium heparin at -20 °C.

Table 2. Analytical Method for Quantification of Midazolam and 1'-Hydroxymidazolam

	Midazolam	**1-Hydroxylmidazolam**
Range	0.3 - 75 ng/mL	0.3 – 37.5 ng/mL
LLOQ	0.1ng/mL	0.1 ng/mL
Linearity	linear (correlation coefficient ⩾ 0.990)	linear (correlation coefficient ⩾ 0.990)
Accuracy	85-115%	85-115%
Precision	⩽ 15%	⩽ 15%
Selectivity	no interference	No interference
Stability	Sponsor only stated that "All samples were analyzed within 854 days, at -20°C.	Sponsor only stated that "All samples were analyzed within 854 days, at -20°C.

Statistical Methods:

Pharmacokinetics:
To assess the effect of delafloxacin on the PK of midazolam and 1-hydroxymidazolam, an analysis of variance was performed on the natural log-transformed AUC_{0-t}, AUC_{0-24}, AUC_{0-inf}, and C_{max} of midazolam and 1-hydroxymidazolam to estimate the ratio of geometric least-squares (LS) means between the treatments and their 90% confidence interval (CI). The analysis of variance model included treatment as a fixed effect and subject as a random effect.

Absence of the effect of delafloxacin on the PK of midazolam was concluded if the 90% CI for the test-to-reference ratio (midazolam + delafloxacin/midazolam alone) of geometric means were entirely contained within the criterion interval of 80% to 125% for AUC_{0-inf} and C_{max}.

Delafloxacin steady state was assessed based on the predose concentrations from Days 4, 5, 6, 7, and 8. A linear mixed model using day as fixed effects and subject as a random effect on the natural log-transformed predose values was performed evaluating whether steady state was achieved using the Helmert transformation approach. The comparison began with Day 4 versus Days 5 through 8. The ratio of geometric LS means and its 95% CI was presented for the comparison.

Safety: No formal statistical analysis of safety parameters was performed. All TEAEs were presented in a summary table by treatment (Treatment A, B, and C) and overall, with the number and percentage of subjects who experienced at least 1 TEAE. Treatment-emergent AEs were also presented in a summary table by treatment and overall, relationship to study drug, severity, TEAEs that led to study drug discontinuation, and treatment-emergent serious AEs.

Safety Assessment:
Adverse events were coded using the Medical Dictionary for Regulatory Activities.

Safety assessments included adverse event (AE) reporting, clinical laboratory results, vital sign measurements (systolic and diastolic blood pressure, pulse rate, respiratory rate, and oral body temperature), 12-lead electrocardiogram (ECG) measurements, and physical examination findings.

RESULTS

Study Population:
Please refer to Table 3 for the summary of subject demographic and baseline characteristics.

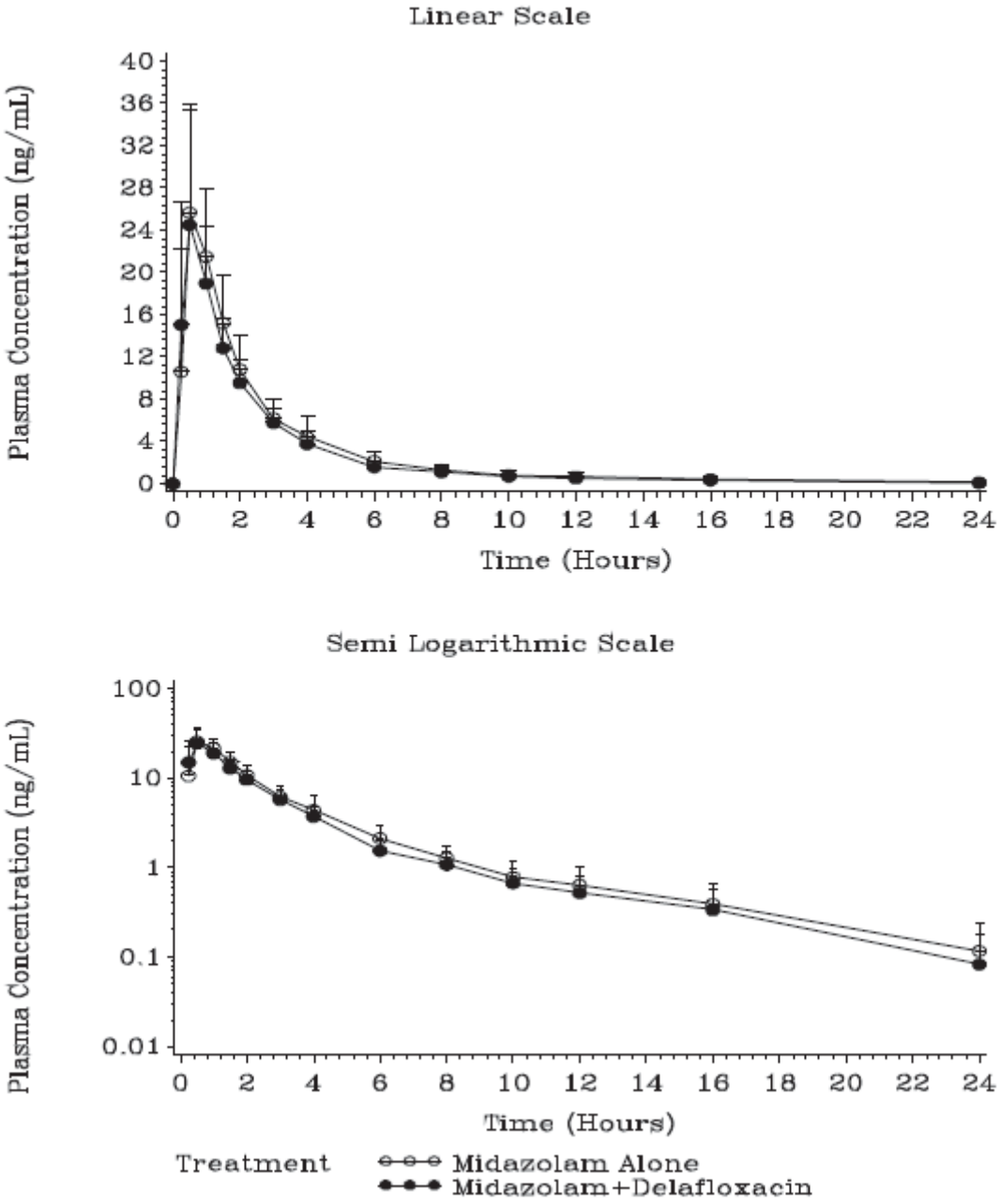
Table 3. Summary of Subject Demographic and Baseline Characteristics (All Subjects) *(Adapted from Table 11-1 in the study report)*

	Overall (N=22)
Age (years)	
Mean (SD)	37.1 (10.37)
Minimum, maximum	18, 50
Sex, n (%)	
Female	8 (36.4)
Male	14 (63.6)
Race, n (%)	
White	12 (54.5)
Black or African American	10 (45.5)
Ethnicity, n (%)	
Hispanic or Latino	11 (50.0)
Not Hispanic or Latino	11 (50.0)
Height (cm)	
Mean (SD)	170.18 (11.990)
Minimum, maximum	148.8, 192.8
Weight (kg)	
Mean (SD)	74.73 (14.372)
Minimum, maximum	50.1, 97.1
Body mass index (kg/m^2)	
Mean (SD)	25.61 (2.816)
Minimum, maximum	19.9, 30.7

Abbreviation: SD, standard deviation.
Note: Percentages were based on the number of subjects overall.

Pharmacokinetics:
The mean (±SD) concentration versus time profile for midazolam is presented in Figure 1 below.

Figure 1. Mean (+SD) Plasma Concentrations of Midazolam Versus Time (Pharmacokinetic Population, Linear and Semilogarithmic Scales) *(Adapted from Figure 11-1 in the study report)*



Please refer to Table 4 for the Mean ± SD (CV) values for selected PK parameters of Midazolam,

Table 4. Mean ± SD (CV) Plasma Pharmacokinetic Parameters of Midazolam by Treatment (Pharmacokinetic Population) *(Adapted from Table 11-2 in the study report)*

Parameter (unit)	Midazolam Alone (N=22)	Midazolam + Delafloxacin (N=22)
AUC_{0-t} (h•ng/mL)	64.44 (31.2)	57.56 (29.4)
AUC_{0-24} (h•ng/mL)	64.72 (31.0)	57.87 (28.9)
AUC_{0-inf} (h•ng/mL)	65.85 (31.7)	58.76 (29.3)
C_{max} (ng/mL)	27.51 (35.7)	25.76 (37.9)
T_{max} (h)[a]	0.50 (0.25, 1.00)	0.50 (0.25, 1.50)
$t_{1/2}$ (h)	4.67 (28.5)	4.63 (26.3)
CL/F (L/h)	82.66 (30.6)	92.58 (31.7)
V_z/F (L)	535.4 (31.0)	597.6 (29.7)

Abbreviations: CV, coefficient of variation; h, hours.
[a] For T_{max}, the median (minimum, maximum) values are presented.

Please refer to Table 5 for statistical analysis of plasma PK parameters of Midazolam.

Table 5. Statistical Analysis of Plasma Pharmacokinetic Parameters of Delafloxacin (Pharmacokinetic Population) (*Adapted from Table 11-3 in the study report)*

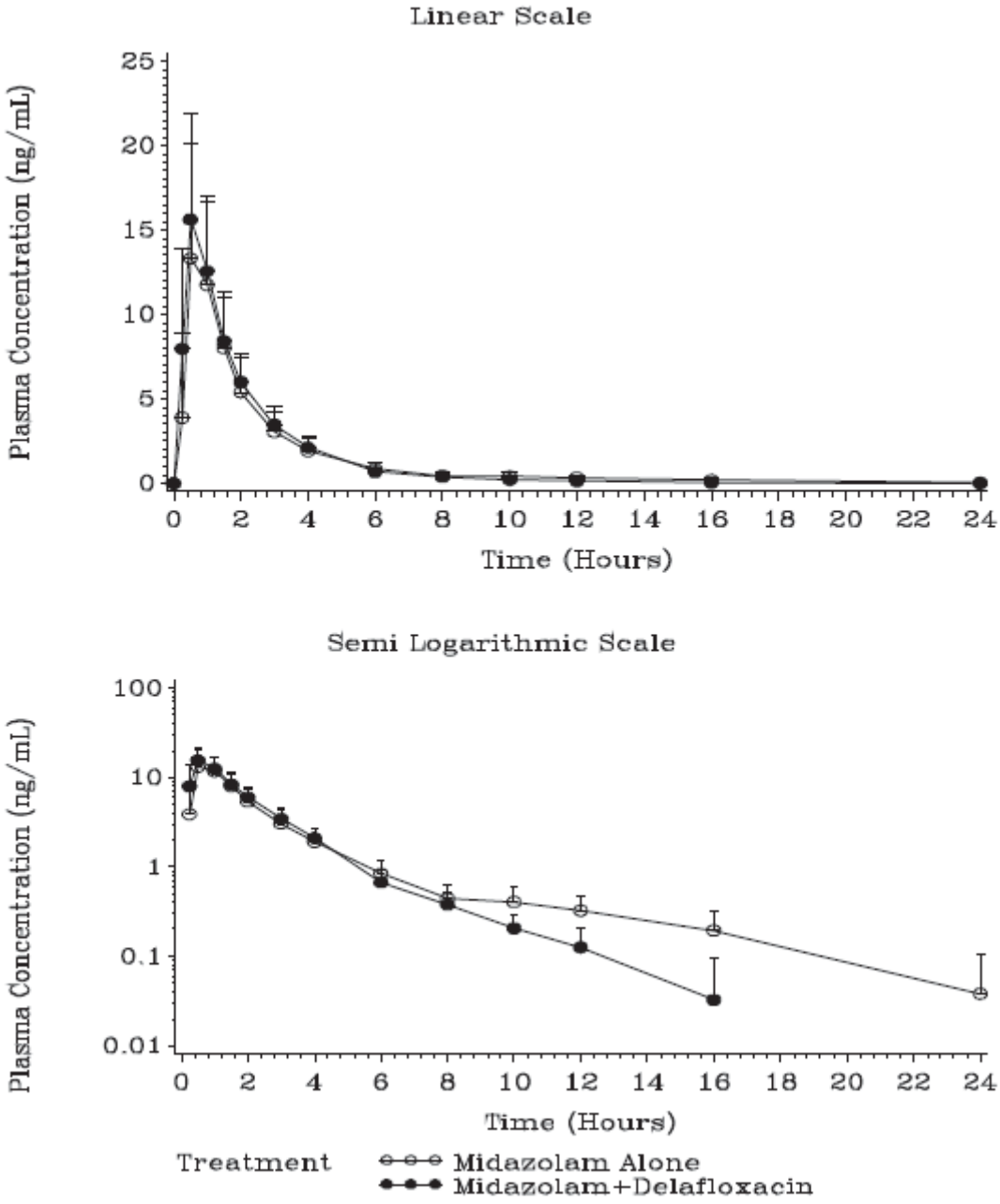
Parameter (Unit)	Treatment	N	Geometric LS Means	90% CI of Geometric LS Means	Treatment Comparison	Ratio (%) of Geometric LS Means	90% CI of the Ratio (%)
AUC_{0-t} (h•ng/mL)	Midazolam Alone	22	61.767	(55.436, 68.820)			
	Midazolam + Delafloxacin	22	55.216	(49.557, 61.522)	Midazolam + Delafloxacin vs Midazolam Alone	89.40	(83.196, 96.056)
AUC_{0-24} (h•ng/mL)	Midazolam Alone	22	62.074	(55.778, 69.081)			
	Midazolam + Delafloxacin	22	55.584	(49.946, 61.858)	Midazolam + Delafloxacin vs Midazolam Alone	89.54	(83.404, 96.137)
AUC_{0-inf} (h•ng/mL)	Midazolam Alone	22	63.080	(56.620, 70.276)			
	Midazolam + Delafloxacin	22	56.394	(50.619, 62.828)	Midazolam + Delafloxacin vs Midazolam Alone	89.40	(83.226, 96.035)
C_{max} (ng/mL)	Midazolam Alone	22	25.706	(22.309, 29.622)			
	Midazolam + Delafloxacin	22	24.052	(20.873, 27.715)	Midazolam + Delafloxacin vs Midazolam Alone	93.56	(83.701, 104.589)

Abbreviations: CI, confidence interval; h, hours; LS, least squares.
Note: A linear mixed-effect model was performed on the natural log-transformed values of AUC_{0-t}, AUC_{0-24}, AUC_{0-inf}, and C_{max} with treatment as a fixed effect and subject as a random effect.

Reviewer's Comment: Based on the analysis results presented in Table 5, it appears that delafloxacin did not affect the AUCs or C_{max} of midazolam, the 90% CIs of these PK parameters of midazolam were all within the 80-125%.

Please refer to Figure 2 for the plasma concentrations of 1-hydroxymidazolam versus time.

Figure 2. Mean (+SD) Plasma Concentrations of 1-Hydroxymidazolam versus Time on Linear and Semilogarithmic Scales (Midazolam Pharmacokinetic Population) *(Adapted from Figure 11-2 in the study report)*



Please refer to Table 6 for the plasma PK parameter of 1-hydroxymidazolam.

Table 6. Mean (CV) Plasma Pharmacokinetic Parameters of 1-Hydroxymidazolam by Treatment (Midazolam Pharmacokinetic Population) *(Adapted from Table 11-4 in the study report)*

Parameter (unit)	Midazolam Alone (N=22)	Midazolam + Delafloxacin (N=22)
AUC_{0-t} (h•ng/mL)	31.07 (36.9)	32.29 (28.2)
AUC_{0-24} (h•ng/mL)	31.57 (36.2)	32.60 (28.2)
AUC_{0-inf} (h•ng/mL)	31.58 (37.9)[a]	32.84 (28.1)
C_{max} (ng/mL)	15.13 (42.3)	16.69 (32.8)
T_{max} (h)[b]	0.50 (0.50, 1.00)	0.50 (0.50, 1.50)
$t_{1/2}$ (h)	4.95 (34.1)[a]	2.68 (33.2)

Abbreviations: CV, coefficient of variation; h, hours.
[a] n = 20.
[b] For T_{max}, the median (minimum, maximum) values are presented.

Please refer to Table 7 for the statistical analysis of the plasma PK parameters of 1-hydroxymidazolam.

Table 7. Statistical Analysis of Plasma Pharmacokinetic Parameters of 1-Hydroxymidazolam (Midazolam
Pharmacokinetic Population) *(Adapted from Table 11-5 in the study report)*

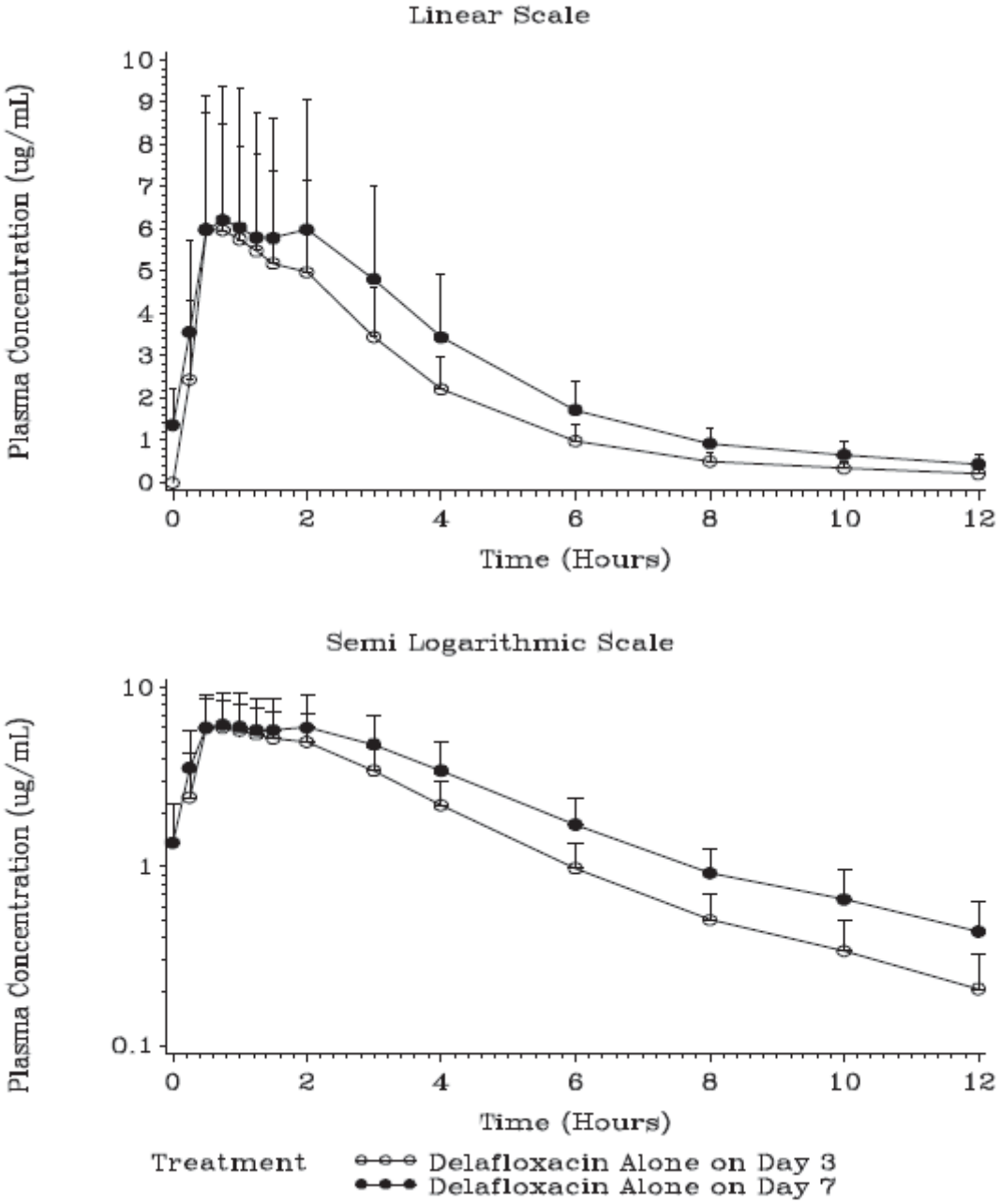
Parameter (Unit)	Treatment	N	Geometric LS Means	90% CI of Geometric LS Means	Treatment Comparison	Ratio (%) of Geometric LS Means	90% CI of the Ratio (%)
AUC_{0-t} (h•ng/mL)	Midazolam Alone	22	28.864	(25.136, 33.144)			
	Midazolam + Delafloxacin	22	30.754	(26.783, 35.315)	Midazolam + Delafloxacin vs Midazolam Alone	106.55	(98.790, 114.919)
AUC_{0-24} (h•ng/mL)	Midazolam Alone	22	29.410	(25.656, 33.714)			
	Midazolam + Delafloxacin	22	31.052	(27.088, 35.596)	Midazolam + Delafloxacin vs Midazolam Alone	105.58	(97.990, 113.767)
AUC_{0-inf} (h•ng/mL)	Midazolam Alone	20	29.610	(25.797, 33.987)			
	Midazolam + Delafloxacin	22	31.294	(27.310, 35.859)	Midazolam + Delafloxacin vs Midazolam Alone	105.69	(97.691, 114.337)
C_{max} (ng/mL)	Midazolam Alone	22	13.326	(11.008, 16.132)			
	Midazolam + Delafloxacin	22	15.466	(12.776, 18.722)	Midazolam + Delafloxacin vs Midazolam Alone	116.05	(101.700, 132.430)

Abbreviations: CI, confidence interval; h, hours; LS, least squares.
Note: A linear mixed-effect model was performed on the natural log-transformed values of AUC_{0-t}, AUC_{0-24}, AUC_{0-inf}, and C_{max} with treatment as a fixed effect and subject as a random effect.

Reviewer's comment: The results presented in Table 7 indicated that delafloxacin did not affect the AUCs of 1-hydroxymidazolam; however, it shows some minor effects on C_{max} of 1-hydroxymidazolam.

Please refer to Figure 3 for the plasma concentration of delafloxacin.

Figure 3. Mean (+SD) Plasma Concentrations of Delafloxacin on Day 3 and Day 7 versus Time on Linear and Semilogarithmic Scales (Delafloxacin Pharmacokinetic Population) *(Adapted from Figure 11-3 in the study report)*



Please refer to Table 8 for the plasma PK parameters of delafloxacin.

Table 8. Mean (CV) Plasma Pharmacokinetic Parameters of Delafloxacin by Treatment (Delafloxacin Pharmacokinetic Population) *(Adapted from Table 11-6 for the study report)*

Parameter (unit)	Delafloxacin (N=22)
Day 3 (Single-dose)	
AUC_{0-t} (h•µg/mL)	22.70 (27.4)
AUC_{0-12} (h•µg/mL)	22.70 (27.4)
AUC_{0-inf} (h•µg/mL)	23.49 (26.1)
C_{max} (µg/mL)	7.17 (28.1)
T_{max} (h)[a]	0.75 (0.50, 4.00)
$t_{1/2}$ (h)	2.46 (20.9)
CL/F (L/h)	20.61 (29.4)
V_z/F (L)	74.4 (41.0)
Day 7 (Steady State)	
$AUC_{0-t,ss}$ (h•µg /mL)	30.75 (37.1)
$AUC_{0-12,ss}$ (h•µg /mL)	30.75 (37.1)
$C_{max,ss}$ (µg/mL)	7.45 (42.4)
$T_{max,ss}$ (h)[a]	1.00 (0.50, 6.00)
$C_{trough,ss}$ (µg/mL)	1.36 (63.5)
$t_{1/2}$ (h)	2.92 (25.5)[b]
CL_{ss}/F (L/h)	16.81 (38.9)
V_z/F (L)	70.8 (47.9)[b]

Abbreviations: CV, coefficient of variation; h, hours.
Note: Values below the limit of quantification were set to zero for summary statistics.
[a] For T_{max} and $T_{max,ss}$, the median (minimum, maximum) values are presented.
[b] n = 21

Please refer to the statistical analysis of PK parameter of delafloxacin in Table 9.

Table 9. Steady State Statistical Analysis of Delafloxacin (Delafloxacin Pharmacokinetic Population) *(Adapted from Table 11-7 in the study report)*

Parameter (Unit)	Treatment	Day	Geometric LS Mean	Comparison	Ratio of Geometric LS Mean	95% CI of the Ratio	p-value
$C_{trough,ss}$ (µg/mL)	1	4	0.723	Day 4 versus Days 5, 6, 7, 8	82.77	(70.713, 96.877)	0.0191
		5	0.722	Day 5 versus Days 6, 7, 8	77.54	(65.907, 91.229)	0.0025
		6	0.695	Day 6 versus Days 7, 8	64.52	(54.298, 76.657)	<.0001
		7	1.149	Day 7 versus Day 8	113.72	(93.186, 138.768)	0.2028
		8	1.011				

Abbreviations: CI, confidence interval; LS, least squares.
Note: A linear mixed-effect model using day as fixed effects and subject as a random effect on the natural log-transformed predose values was performed to evaluate if steady state was achieved using the Helmert transformation approach.

Reviewer's Comment: The results presented in Table 9 indicated that delafloxacin reaches steady state by Day 7 after initiating dosing on Day 4. The results in Table 8 indicated that delafloxacin has some accumulation (AR=1.149/.732=1.56) in this study by comparing the C_{trough}.

SPONSOR'S CONCLUSIONS

PHARMACOKINETIC

- Delafloxacin did not affect the C_{max} and $AUC_{0\text{-}inf}$ of midazolam.
- Delafloxacin did not affect the $AUC_{0\text{-}inf}$ of 1-hydroxymidazolam. There was a minimal effect indicated for the C_{max} of 1-hydroxymidazolam.
- Steady state for almost all subjects appeared to have reached by the Day 7 dosing of delafloxacin.

REVIEWER ASSESSMENT:

- Based on the in vivo study result, delafloxacin does not seem to induce CYP3A4 at a clinically relevant concentration.

APPEARS THIS WAY ON ORIGINAL

4.5.6 Study No.: ML-3341-112

AN OPEN-LABEL EVALUATION OF THE SINGLE-DOSE PHARMACOKINETICS OF DELAFLOXACIN IN SUBJECTS WITH AND WITHOUT HEPATIC IMPAIRMENT

Date(s): 04 September 2014 to 1 16 February 2015
Sponsor: Melinta Therapeutics, Inc 300 George Street, Suite 301, New Haven, CT 06511
Clinical Site: Site 1, Orlando Clinical Research Center (Orlando, FL); Site 2, University of Miami (Miami, FL 33136)
Analytical Site: (b) (4)

OBJECTIVE(S):

- The primary objective was to evaluate the pharmacokinetic (PK) profile of a single intravenous (IV) dose of delafloxacin in normal healthy subjects and subjects with mild, moderate, or severe hepatic impairment.
- The secondary objective was to evaluate the safety and tolerability of a single IV dose of delafloxacin in normal healthy subjects and subjects with mild, moderate, or severe hepatic impairment.

METHODS

Study Design: This was a Phase 1, multicenter, open-label, single-dose, PK study. A total of 36 subjects were planned to be stratified into groups based on hepatic function as defined by the Child-Pugh classification system as follows:

- Group A: 6 subjects with mild hepatic impairment (Child-Pugh Class A)
- Group B: 6 subjects with moderate hepatic impairment (Child-Pugh Class B)
- Group C: 6 subjects with severe hepatic impairment (Child-Pugh Class C)
- Group D: 18 healthy subjects

Groups A and B (mild and moderate hepatic impairment) were enrolled first. An interim analysis was performed after completion of Groups A and B and the matching healthy subjects. Enrollment of Group C (severe hepatic impairment) began after the interim analysis. Each matching healthy subject (Group D) was recruited after the subject with hepatic impairment had been enrolled in the study in order to match them as closely as possible with respect to age (±10 years), weight (±20%), and gender. In addition, a reasonable attempt was made to match healthy subjects to the subjects with hepatic impairment in terms of smoking status and alcohol use.

This was a single dose study. Subjects entered the clinical site the day before dosing (Day –1). On Day 1, a single 1-hour IV infusion of delafloxacin 300 mg was administered in the morning. Blood samples were collected for the PK analysis of delafloxacin concentrations through 72 hours after dosing.

Reviewer Comment: The study design meets the criteria outlined in the FDA "Guidance for Industry Pharmacokinetics in Patients with Impaired Hepatic Function: Study Design, Data Analysis, and Impact on Dosing and Labeling".

Drug Product: Delafloxacin supplied as a sterile, light yellow to tan colored lyophilized cake in (b) (4) glass vials; Dose: 300 mg; Mode of Administration: IV infusion for 1 hour; Lot Number: 12DEL1

Inclusion Criteria: Male and female subjects between 18 and 80 years of age, inclusive, with a body mass index of 18 to 40 kg/m^2, inclusive, with either normal hepatic function or hepatic impairment. Subjects with hepatic impairment had a clinical diagnosis of cirrhosis classified as mild, moderate, or severe as defined by the Child-Pugh classification system. Subjects with normal hepatic function were matched to the age (±10 years), weight (±20%), and gender of subjects with hepatic impairment.

PK Sample Collection: Blood samples for the determination of plasma concentrations of delafloxacin were collected within 15 minutes before dosing, at 20, 40, and 60 minutes (end of infusion); 65 minutes after the start of infusion (SOI); and at 0.25, 0.5, 0.75, 1, 1.5, 2, 3, 4, 6, 8, 12, 16, 24, 36, 48, 60, and 72 hours after the end of the infusion. Pharmacokinetic blood samples were collected from the opposite arm as the one used for the study drug infusion.

Analytical Methods: Project samples were analyzed according to (b) (4) Method LCMSC 634 V.1.02, entitled "Quantitation of Delafloxacin in Human Plasma via HPLC with MS/MS Detection," which was validated under Project Code "OZZ2". Please see the summary of method validation in Table 1.

Table 1. Method Validation Summary

(b) (4) Project Code	OZZ2		
Method ID	LCMSC 634		
Analytes	Delafloxacin		
Matrix	Human Plasma		
Anticoagulant	Dipotassium EDTA		
Method Description	Supported liquid phase extraction		
Sample Volume (µL)	100-µL		
Sample Storage Temperature	-20 °C or colder		
Internal Standard (IS)	Delafloxacin-d_5		
Regression, Weighting	Linear, 1/concentration^2		
Average Recovery of Drug (%)	71.9%		
Average Recovery of IS (%)	70.7%		
Standard Curve Concentrations	5.00 to 5000 ng/mL		
QC Concentrations	5.00, 15.0, 40.0, 150, 600, and 3750 ng/mL		
QC Intra-assay Statistics (%)	Conc. (ng/mL)	Precision	Accuracy
	5.00	3.36 to 5.36%	-0.888 to 3.91%
	15.0	1.83 to 2.18%	-1.90 to 2.98%
	40.0	0.794 to 1.74%	-0.267 to 1.80%
	150	1.42 to 2.56%	-1.11 to 2.69%
	600	1.21 to 1.93%	0.821 to 2.20%
	3750	1.28 to 4.13%	0.192 to 2.02%
QC Inter-assay Statistics (%)	Conc. (ng/mL)	Precision	Accuracy
	5.00	4.49%	0.872%
	15.0	2.79%	0.730%
	40.0	1.54%	0.747%
	150	2.49%	0.593%
	600	1.71%	1.60%
	3750	2.58%	1.02%
Thawed Matrix Stability (hrs)	24 hours at room temperature		
Solution Stability (days)	Delafloxacin 35 days at 2 to 8 °C in standard diluent*		
	Delafloxacin-d_5 35 days at 2 to 8 °C in standard diluent*		
Solution Stress Stability (hours)	Delafloxacin 7 hours at room temperature in 50:50 Acetonitrile / Water*		
	Delafloxacin-d_5 7 hours at room temperature in 50:50 Acetonitrile / Water*		
Extract Stability (hrs)	167 hours at 2 to 8 °C		
Freeze-thaw Stability (cycles)	Five cycles thawed at room temperature		
Frozen Matrix Storage Stability (days)	29 days at -20 °C and -70 °C		
Whole Blood Stability	N/A		
Dilutional Linearity	150 ng/mL diluted four-fold		
	10000 ng/mL diluted ten-fold		
Selectivity	No significant interfering peaks noted in blank plasma samples		
Hemolysis	No effect from hemolysis on the quantitation of delafloxacin		
Lipemia	No effect from lipemia on the quantitation of delafloxacin		

* Established prior to/following the validation

Reviewer's Comment: The bioanalytical method and analytical run in this study meet the acceptance criteria specified in the FDA Guidance for Industry Bioanalytical Method Validation.

Pharmacokinetic Assessment: Noncompartmental PK analysis was performed using Phoenix® WinNonlin® version 6.2.1 (Pharsight, St Louis, Missouri). The individual plasma concentration versus actual time data were used to derive the PK parameters and were presented by subject

number and group in a data listing. The following PK parameters were calculated for each subject: AUC_{0-t}, AUC_{0-inf}, AUC_{0-12}, AUC_{0-24}, C_{max}, T_{max}, $t_{1/2}$, CL, Vss,

Statistical Methods:
Pharmacokinetics: Plasma PK parameters were summarized by group using descriptive statistics: number of subjects, mean, SD, CV, minimum, median, and maximum. The geometric mean and geometric CV were also presented for the AUCs and Cmax. Box-and-whiskers plots of PK parameters versus group were also presented.

The geometric least squares (LS) mean ratios of the hepatic impairment groups to the corresponding healthy matching group (A:D [mild match], B:D [moderate match], and C:D [severe match]) for AUC_{0-t}, AUC_{0-inf}, AUC_{0-12}, AUC_{0-24}, Cmax, and CL were calculated by using the antilog of the LS mean difference of the natural log-transformed values. A 90% confidence interval (CI) for each ratio was constructed as the antilog of the 90% CI of the LS mean difference. No adjustment was made for multiplicity. In addition, the geometric LS means and corresponding 90% CIs were computed for AUC_{0-t}, AUC_{0-inf}, AUC_{0-12}, AUC_{0-24}, C_{max}, and CL by taking the antilog of the LS means and corresponding 90% CI from the linear mixed-effect model on the natural logarithm of the corresponding PK parameters. A similar analysis was performed for the geometric LS mean ratios of the hepatic impairment groups to the pooled healthy subjects group (A:D [pooled subjects], B:D [pooled subjects], and C:D [pooled subjects]).

Safety: No formal statistical analysis of safety parameters was performed.

Safety Assessment: Safety assessments included evaluation of AEs, clinical laboratory results (serum chemistry, coagulation, hematology, and urinalysis), vital sign measurements (blood pressure, heart rate, and body temperature), 12-lead ECG measurements, and physical examination findings.

RESULTS

Study Population: Thirty-nine subjects received the single 1-hour IV infusion of delafloxacin 300 mg. Thirty-six subjects (92.3%) had evaluable PK data and were included in the PK analysis population. Subject 1101 (mild hepatic impairment group), Subject 1113 (healthy subject group, mild hepatic impairment match), and Subject 2211 (healthy subject group, moderate hepatic impairment match) were excluded from the PK analysis population due to study discontinuation caused by TEAEs. One subject (Subject 2311) was erroneously enrolled twice into the study. The subject was first enrolled as Subject 2111 (mild impairment match group) and later as Subject 2311 (severe impairment match group). Subject 2311 was excluded from the PK analysis population. The data for subject 2311 were presented in the PK and safety listings, but were excluded from the PK analysis, as well as PK and safety summary tables.

The demographics and baseline characteristics of the study population were summarized in Table 2. Overall, demographic and baseline characteristics were similar across groups. The majority of subjects were white (31 of 39 subjects [79.5%]), male (35 of 39 subjects [89.7%]), and Hispanic

or Latino (24 of 39 subjects [61.5%]), with a mean age of 54.5 years (range: 42 to 68 years) and a mean BMI of 29.68 kg/m^2 (range: 21.8 to 39.2 kg/m^2).

Table 2. Summary of Subject Demographics and Baseline Characteristics (All Subjects) (*Adapted from Table 11-1 from the study report*)

	Hepatic Function Group[a]							
	Group A Mild (n = 7)	Group D Mild Match[b] (n = 7)	Group B Moderate (n = 6)	Group D Moderate Match (n = 7)	Group C Severe (n = 6)	Group D Severe Match[b] (n = 6)	Group D Overall[b] (n = 20)	Overall[b] (N = 39)
Age (years)[a]								
Mean (SD)	54.4 (6.75)	51.7 (6.52)	56.8 (7.33)	56.4 (6.95)	54.3 (5.43)	53.7 (7.69)	54.0 (6.95)	54.5 (6.58)
Minimum, maximum	48, 68	42, 60	45, 67	44, 64	47, 62	45, 67	42, 67	42, 68
Gender, No. (%)								
Male	6 (85.7)	6 (85.7)	5 (83.3)	6 (85.7)	6 (100.0)	6 (100.0)	18 (90.0)	35 (89.7)
Female	1 (14.3)	1 (14.3)	1 (16.7)	1 (14.3)	0	0	2 (10.0)	4 (10.3)
Race, No. (%)								
White	7 (100.0)	5 (71.4)	5 (83.3)	4 (57.1)	5 (83.3)	5 (83.3)	14 (70.0)	31 (79.5)
Black or African American	0	2 (28.6)	1 (16.7)	3 (42.9)	0	1 (16.7)	6 (30.0)	7 (17.9)
Multiracial	0	0	0	0	1 (16.7)	0	0	1 (2.6)
Ethnicity, No. (%)								
Hispanic or Latino	4 (57.1)	5 (71.4)	4 (66.7)	3 (42.9)	4 (66.7)	4 (66.7)	12 (60.0)	24 (61.5)
Not Hispanic or Latino	3 (42.9)	2 (28.6)	2 (33.3)	4 (57.1)	2 (33.3)	2 (33.3)	8 (40.0)	15 (38.5)
Height (cm)								
Mean (SD)	169.43 (9.515)	172.26 (8.636)	175.70 (12.056)	171.14 (7.983)	172.45 (6.712)	174.60 (7.118)	172.57 (7.687)	172.47 (8.500)
Minimum, maximum	156.0, 181.5	158.0, 180.0	159.2, 190.0	156.0, 178.0	165.0, 182.0	165.1, 184.0	156.0, 184.0	156.0, 190.0
Weight (kg)								
Mean (SD)	79.54 (7.081)	82.30 (11.596)	93.07 (17.548)	89.69 (7.145)	96.55 (25.827)	90.95 (20.761)	87.48 (13.692)	88.31 (16.086)
Minimum, maximum	70.0, 91.0	66.0, 96.0	73.0, 115.1	78.3, 100.6	65.0, 124.2	70.1, 114.2	66.0, 114.2	65.0, 124.2

	Hepatic Function Group[a]							
	Group A Mild (n = 7)	Group D Mild Match[b] (n = 7)	Group B Moderate (n = 6)	Group D Moderate Match (n = 7)	Group C Severe (n = 6)	Group D Severe Match[b] (n = 6)	Group D Overall[b] (n = 20)	Overall[b] (N = 39)
Body mass index (kg/m^2)								
Mean (SD)	28.10 (5.213)	27.73 (3.453)	30.35 (5.965)	30.71 (2.962)	32.07 (6.513)	29.53 (4.688)	29.32 (3.739)	29.68 (4.801)
Minimum, maximum	22.6, 35.5	22.4, 31.4	21.8, 38.9	27.2, 35.4	23.6, 39.2	23.4, 35.4	22.4, 35.4	21.8, 39.2
Child-Pugh Class Score								
Mean (SD)	5.7 (0.49)	NA	8.5 (0.55)	NA	10.8 (0.98)	NA	NA	8.2 (2.27)
Minimum, maximum	5, 6		8, 9		10, 12			5, 12

Abbreviation: NA, not applicable; SD, standard deviation.

Note: Percentages were based on the number of subjects in the all subjects population within each group and overall.

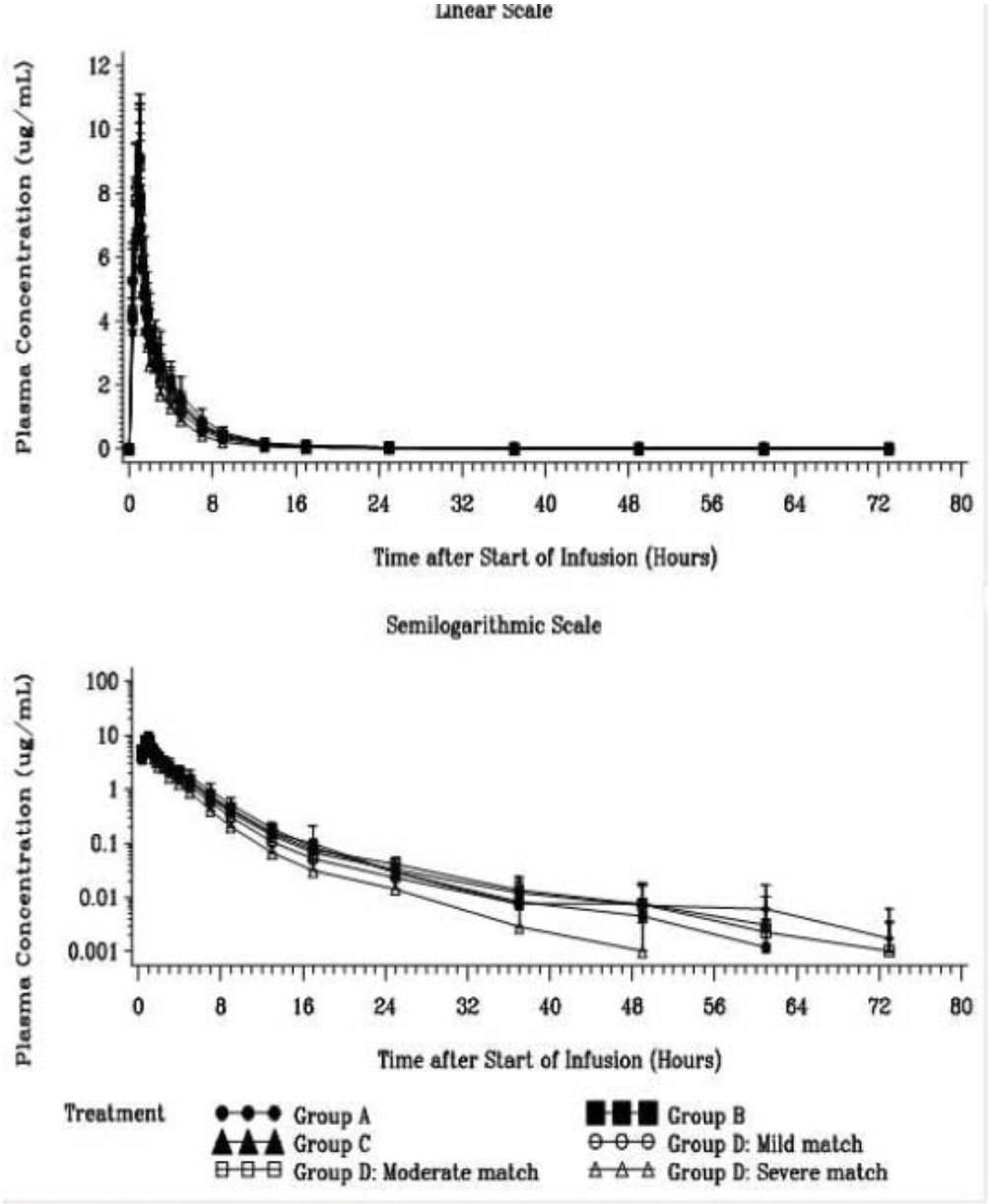
[a] Group A Mild: Subjects with mild hepatic impairment (Child-Pugh Class A);
Group B Moderate: Subjects with moderate hepatic impairment (Child-Pugh Class B);
Group C Severe: Subjects with severe hepatic impairment (Child-Pugh Class C);
Group D Healthy subjects.

[b] Subject 2311 was the same subject as 2111 and was erroneously enrolled twice into the study. Subject 2311 was excluded from the PK population and the safety summary tables.

Pharmacokinetics:

The mean (±SD) concentration versus time profile for delafloxacin is presented in Figure 1 below.

Figure 1. Mean (±SD) Concentrations of Delafloxacin (µg/mL) Versus Time by Hepatic Function Group (Linear and Semilogarithmic Scales) (Pharmacokinetic Population) (*Adapted from Figure 11-1 in the study report*)



Abbreviation: SD, standard deviation.
Note: Subjects 1101, 1113, and 2211 did not have complete dosing information since the infusion was stopped due to adverse events and were excluded from the PK population. Subject 2311 was the same as Subject 2111 and was erroneously enrolled twice into the study. Subject 2311 was excluded from the PK population and the safety summary tables.
Group A Mild: Subjects with mild hepatic impairment (Child-Pugh Class A);
Group B Moderate: Subjects with moderate hepatic impairment (Child-Pugh Class B);
Group C Severe: Subjects with severe hepatic impairment (Child-Pugh Class C);
Group D Healthy subjects.

Mean (CV) plasma PK parameters of delafloxacin are summarized in Table 3 below.

Table 3. Mean (CV) Plasma Pharmacokinetic Parameters of Delafloxacin by Hepatic Function Group
(Pharmacokinetic Population) (*Adapted from Table 11-2 in the study report*)

	Hepatic Function Group[a]						
Parameter (unit)	Group A Mild (n = 6)	Group B Moderate (n = 6)	Group C Severe (n = 6)	Group D Mild Match (n = 6)	Group D Moderate Match (n = 6)	Group D Severe Match (n = 6)	Group D Overall (n = 18)
AUC_{0-t} (h•µg/mL)	23.35 (16.3)	23.29 (9.2)	24.34 (28.8)	21.15 (16.3)	23.48 (19.9)	17.10 (15.2)	20.57 (21.3)
AUC_{0-inf} (h•µg/mL)	23.52 (16.3)	23.40 (9.2)	24.41 (28.7)	21.72 (17.3)[b]	23.58 (19.9)	17.18 (15.2)	20.77 (21.9)[c]
AUC_{0-12} (h•µg/mL)	21.89 (13.8)	21.80 (6.1)	22.91 (29.1)	20.07 (14.3)	21.87 (20.4)	16.56 (15.5)	19.50 (20.1)
AUC_{0-24} (h•µg/mL)	22.83 (15.0)	22.96 (8.4)	24.10 (28.6)	20.80 (15.5)	22.94 (20.1)	17.00 (15.0)	20.25 (20.7)
C_{max} (µg/mL)	9.22 (15.5)	7.92 (9.8)	8.42 (30.6)	9.26 (17.3)	9.01 (19.9)	8.33 (21.3)	8.86 (18.9)
T_{max} (h)[d]	1.00 (1.00, 1.08)	1.00 (1.00, 1.02)	1.00 (1.00, 1.08)	1.00 (0.67, 1.00)	1.00 (0.67, 1.00)	1.00 (0.67, 1.00)	1.00 (0.67, 1.00)
$t_{1/2}$ (h)	13.45 (58.7)	9.09 (92.5)	5.26 (24.4)	8.23 (62.5)[b]	9.30 (50.9)	7.09 (80.1)	8.20 (60.4)[c]
CL (L/h)	13.07 (17.9)	12.91 (9.1)	13.57 (41.3)	14.09 (14.3)[b]	13.22 (22.7)	17.85 (17.4)	15.11 (22.3)[c]
V_{ss} (L)	50.88 (20.2)	48.74 (20.2)	49.68 (42.5)	44.75 (18.6)[b]	51.51 (25.3)	47.92 (23.5)	48.25 (22.5)[c]
V_z (L)	253.27 (60.9)	166.87 (88.5)	100.28 (36.2)	159.83 (58.3)	169.00 (41.8)	175.04 (66.5)	168.43 (53.1)[c]

Abbreviations: CV, coefficient of variation; h, hours.
Note: Subjects 1101, 1113, and 2211 did not have complete dosing information since the infusion was stopped due to adverse events and were excluded. Subject 2311 was the same as Subject 2111 and was erroneously enrolled twice into the study. Subject 2311 was excluded from the PK population and the safety summary tables.
[a] Group A Mild: Subjects with mild hepatic impairment (Child-Pugh Class A);
Group B Moderate: Subjects with moderate hepatic impairment (Child-Pugh Class B);
Group C Severe: Subjects with severe hepatic impairment (Child-Pugh Class C);
Group D Healthy subjects.
[b] n = 5
[c] n = 17
[d] For T_{max}, the median (minimum, maximum) values are presented.

Statistical analysis of the plasma PK parameters of delafloxacin versus matched healthy subjects is summarized in Table 4 below.

Table 4. Statistical Analysis of Plasma Pharmacokinetic Parameters of Delafloxacin by Hepatic Function Group Versus Matched Healthy Subject Group (Pharmacokinetic Population) (*Adapted from Table 11-3 in the study report*)

Parameter (Unit)	Group[a]	N	Geometric LS Mean	90% CI of Geometric LS Mean	Comparison	Ratio (%) of Geometric LS Means	90% CI of the Ratio
AUC_{0-t} (h•µg/mL)	A: mild	6	23.080	(20.025, 26.602)	A: mild/D: mild match	110.20	(90.146, 134.710)
	D: mild match	6	20.945	(18.172, 24.141)			
	B: moderate	6	23.208	(20.135, 26.749)	B: moderate/D: moderate match	100.65	(82.334, 123.036)
	D: moderate match	6	23.058	(20.006, 26.577)			
	C: severe	6	23.286	(20.203, 26.840)	C: severe/D: severe match	137.65	(112.599, 168.263)
	D: severe match	6	16.917	(14.678, 19.499)			
AUC_{0-inf} (h•µg/mL)	A: mild	6	23.246	(20.135, 26.836)	A: mild/D: mild match	108.15	(87.395, 133.825)
	D: mild match	5	21.495	(18.365, 25.157)			
	B: moderate	6	23.315	(20.195, 26.916)	B: moderate/D: moderate match	100.71	(82.194, 123.389)
	D: moderate match	6	23.151	(20.253, 26.727)			
	C: severe	6	23.376	(20.249, 26.987)	C: severe/D: severe match	137.50	(112.225, 168.472)
	D: severe match	6	17.001	(14.726, 19.627)			
AUC_{0-12} (h•µg/mL)	A: mild	6	21.700	(18.897, 24.919)	A: mild/D: mild match	108.92	(89.566, 132.446)
	D: mild match	6	19.923	(17.350, 22.879)			
	B: moderate	6	21.767	(18.955, 24.996)	B: moderate/D: moderate match	101.40	(83.387, 123.309)
	D: moderate match	6	21.466	(18.693, 24.650)			
	C: severe	6	21.910	(19.080, 25.160)	C: severe/D: severe match	133.72	(109.965, 162.612)
	D: severe match	6	16.384	(14.268, 18.815)			
AUC_{0-24} (h•µg/mL)	A: mild	6	22.601	(19.660, 25.982)	A: mild/D: mild match	109.64	(90.023, 133.533)
	D: mild match	6	20.614	(17.932, 23.697)			
	B: moderate	6	22.895	(19.916, 26.320)	B: moderate/D: moderate match	101.62	(83.440, 123.768)
	D: moderate match	6	22.530	(19.598, 25.900)			
	C: severe	6	23.080	(20.077, 26.532)	C: severe/D: severe match	137.11	(112.576, 166.987)
	D: severe match	6	16.833	(14.643, 19.351)			

Parameter (Unit)	Group[a]	N	Geometric LS Mean	90% CI of Geometric LS Mean	Comparison	Ratio (%) of Geometric LS Means	90% CI of the Ratio
C_{max} (µg/mL)	A: mild	6	9.132	(7.909, 10.542)	A: mild/D: mild match	99.92	(81.546, 122.428)
	D: mild match	6	9.139	(7.916, 10.551)			
	B: moderate	6	7.885	(6.830, 9.104)	B: moderate/D: moderate match	88.92	(72.568, 108.949)
	D: moderate match	6	8.868	(7.681, 10.238)			
	C: severe	6	8.051	(6.974, 9.295)	C: severe/D: severe match	98.52	(80.403, 120.713)
	D: severe match	6	8.173	(7.079, 9.435)			
CL (L/h)	A: mild	6	12.906	(11.179, 14.899)	A: mild/D: mild match	92.47	(74.724, 114.423)
	D: mild match	5	13.957	(11.925, 16.335)			
	B: moderate	6	12.867	(11.146, 14.855)	B: moderate/D: moderate match	99.30	(81.044, 121.664)
	D: moderate match	6	12.958	(11.225, 14.960)			
	C: severe	6	12.833	(11.116, 14.816)	C: severe/D: severe match	72.73	(59.357, 89.107)
	D: severe match	6	17.646	(15.285, 20.372)			

Abbreviations: CI, confidence interval; LS, least squares.

Note: An analysis of variance was performed on the natural log-transformed parameters with hepatic impairment group as a fixed effect. Subjects 1101, 1113, and 2211 did not have complete dosing information since the infusion was stopped due to adverse events and were excluded. Subject 2311 was the same as Subject 2111 and was erroneously enrolled twice into the study. Subject 2311 was excluded from the PK population and the safety summary tables.

[a] Group A Mild: Subjects with mild hepatic impairment (Child-Pugh Class A);
Group B Moderate: Subjects with moderate hepatic impairment (Child-Pugh Class B);
Group C Severe: Subjects with severe hepatic impairment (Child-Pugh Class C);
Group D Healthy subjects.

Reviewer's Comment:

1) *Higher variability in estimated PK parameters was observed in patients with severe hepatic impairment (Table 3). However, the absolute variability in patients with severe hepatic impairment was still acceptable (CV = 28.8%).*
2) *It was observed that there was a numerical increase in the geometric LS mean ratio for the overall exposure (AUCs) in patients with severe hepatic impairment compared to the respective controls (about 1.37 fold). The 90% CIs (%) of the geometric LS mean ratio for C_{max} for the two aforementioned groups were within 80-125%. However, it was also noticed that the AUCs in severe match group were overall lower compared to other match groups (i.e. mild match and moderate match) and the absolute AUC values in patients with severe hepatic impairment group were similar to the ones in other groups with different hepatic function. The reviewer did not identify any PK parameter outliers in patients with severe hepatic impairment in raw dataset for individual subjects.*
3) *With 300 mg IV single dose, the overall exposure (AUCs) of delafloxacin was similar in patients with normal, mild, and moderate hepatic function.*

Safety: As mentioned in the Study Population before, three out of thirty nine subjects were discontinued infusion due to TEAEs. Subject 1101 (mild hepatic impairment group) was discontinued due to a mild, probably related TEAE of drug hypersensitivity, Subject 1113 (healthy subject, mild hepatic impairment match group) was discontinued due to a mild, possibly related TEAE of presyncope, and Subject 2211 (healthy subject, moderate hepatic impairment match group) was discontinued due to a moderate, probably related TEAE of drug hypersensitivity.

Overall, 11 TEAEs were reported and 10 of 39 subjects (25.6%) reported at least 1 TEAE after receiving the delafloxacin 300 mg IV dose. With the exception of 2 TEAEs of moderate severity, all TEAEs reported during this study were of mild severity. Moderate TEAEs reported in 1 subject each included headache and drug hypersensitivity. There were no deaths or SAEs reported during this study.

Treatment-emergent AEs were reported by 1 of 7 subjects (14.3%) in the mild hepatic impairment group, no subjects in the moderate hepatic impairment group, 2 of 6 subjects (33.3%) in the severe hepatic impairment group, and 7 of 20 subjects (35.0%) overall in the healthy subjects group (3 of 7 subjects [42.9%] in the mild hepatic impairment match group, 3 of 7 subjects [42.9%] in the moderate hepatic impairment match group, and 1of 6 subjects [16.7%] in the severe hepatic impairment match group).

Reviewer's Comment: No trend of increasing TEAEs was observed with the decreasing of hepatic function in this single dose study.

SPONSOR'S CONCLUSIONS

PHARMACOKINETIC

- Overall, the effects of hepatic impairment on the systemic exposures for delafloxacin did not display a clear trend across the hepatic impairment groups.
- Total (AUC_{0-t} and AUC_{0-inf}) and partial (AUC_{0-12} and AUC_{0-24}) exposures for plasma delafloxacin were slightly increased by approximately 1.1-fold for subjects with mild hepatic impairment compared with healthy subjects; whereas, there was no significant difference in peak (C_{max}) exposure between each group.
- Clearance for delafloxacin was slightly decreased for subjects with mild hepatic impairment compared with healthy subjects.
- Total (AUC_{0-t} and AUC_{0-inf}) and partial (AUC_{0-12} and AUC_{0-24}) exposures for plasma delafloxacin were increased by approximately 1.1- to 1.2-fold for subjects with moderate hepatic impairment only when compared with the pooled healthy subject group; whereas, peak (C_{max}) exposure was slightly decreased for subjects with moderate hepatic impairment when compared with the matched healthy subjects and the pooled healthy subject group, as the C_{max} values for subjects with moderate hepatic impairment were reduced by approximately 10% compared with Cmax values for healthy subjects.
- Clearance for delafloxacin was decreased for subjects with moderate hepatic impairment only when compared the pooled healthy subject group.
- Total (AUC_{0-t} and AUC_{0-inf}) and partial (AUC_{0-12} and AUC_{0-24}) exposures for plasma delafloxacin were increased by approximately 1.1- to 1.4-fold for subjects with severe hepatic impairment compared with healthy subjects; whereas, peak (C_{max}) exposure was slightly decreased for subjects with severe hepatic impairment only when compared with the pooled healthy subject group, as the C_{max} values for subjects with severe hepatic impairment were reduced by approximately 8% compared with Cmax values for healthy subjects.
- Clearance for delafloxacin was decreased for subjects with severe hepatic impairment compared with healthy subjects.

SAFETY

- Overall, the delafloxacin 300 mg 1-hour IV infusion was safe and generally well tolerated by the healthy subjects and subjects with hepatic impairment in this study.
- At least 1 TEAE was reported by 25.6% of subjects. In subjects who reported TEAEs, the majority had TEAEs that were considered mild and not related to study drug.by the healthy subjects and subjects with hepatic impairment in this study.

- The most commonly reported TEAE was drug hypersensitivity and was reported by 2 subjects (5.1%). Majority had TEAEs that were considered mild and not related to study drug.
- Three subjects (7.7%) were discontinued due to TEAEs. Subject 1101 (mild hepatic impairment group) was discontinued due to a mild, probably related TEAE of drug hypersensitivity, Subject 1113 (healthy subject, mild hepatic impairment match group) was discontinued due to a mild, possibly related TEAE of presyncope, and Subject 2211 (healthy subject, moderate hepatic impairment match group) was discontinued due to a moderate, probably related TEAE of drug hypersensitivity. All TEAEs resolved by the end of the study.
- There were no deaths or SAEs reported during this study.
- There were no apparent treatment-related trends or clinically significant findings observed in the clinical laboratory assessments, vital sign measurements, ECG results, or physical examination findings.

REVIEWER ASSESSMENT:

- It was observed that total (AUC_{0-t} and AUC_{0-inf}) and partial (AUC_{0-12} and AUC_{0-24}) exposures for plasma delafloxacin were increased by approximately 1.1- to 1.4-fold for subjects with severe hepatic impairment compared with healthy subjects receiving a single IV dose of 300 mg delafloxacin. Based on the acceptable safety findings in patients receiving 450 mg BID IV delafloxacin whose AUCs were approximately 1.5 fold higher than those in patients receiving 300 mg BID IV delafloxacin, dose adjustment in patients with hepatic impairment does not deem necessary.
- The Reviewer noted that only the IV injection formulation was evaluated in the dedicated hepatic impairment study. Since delafloxacin was primarily metabolized in the liver by glucuronidation and the fractions of dose metabolized following IV and oral administration were similar (IV: 15-23%, oral: about 18%), the effect of hepatic impairment on delafloxacin PK is expected to be similar between the IV injection and oral tablet formulations.

4.5.7 Study No.: RX-3341-101

A DOUBLE-BLIND, PLACEBO-CONTROLLED, SINGLE ASCENDING DOSE TOLERABILITY AND PHARMACOKINETIC STUDY OF RX-3341 IN HEALTHY SUBJECTS

Date(s): 19 April 2007 to 07 June 2007
Sponsor: Melinta Therapeutics, Inc, 300 George Street, Suite 301, New Haven, CT 06511
Clinical Site: Single site: PPD Phase I Clinic, located at 7551 Metro Center Drive, Suite 200, Austin, TX 78744
Analytical Site: (b) (4)

OBJECTIVE(S):

- The primary objective was to determine the safety, tolerability, and pharmacokinetics of single doses of an intravenous formulation of RX-3341.
- The secondary objective was to investigate the effect of infusion duration on the safety, tolerability, and pharmacokinetics of RX-3341.

METHODS

Study Design: This was a double-blind, randomized, placebo-controlled, single dose study conducted in 43 subjects. Dose ranging covered 5 doses of RX-3341 (50, 100, 200, 300, and 400 mg). The study was conducted in 2 phases. In the Ascending Dose Phase, a single dose of RX-3341 (50, 100, 200, 300 or 400 mg) or corresponding placebo was administered intravenously over 1 hour. Subsequent dosing groups received the next highest dose in the dosage schedule. Decisions to escalate to the next higher dose were based on the results of the safety and tolerability of the previous dose.

After a 2-week break, the Increased Infusion Rate Phase was begun. A dose of 300 mg was chosen to assess an infusion rate of 30 minutes. The dose and infusion rate were based on safety and pharmacokinetic (PK) assessments of data from the Ascending Dose Phase.

Reviewer's Comment: The increased infusion rate phase will not be reviewed since the 30 minute infusion time was not the one used in Phase 3 trials and also the IV formulation used in this study was not the to-be-marketed IV dosage form. The results of this part will not be included in this review.

For the 1-hour infusion rate, the 50-mg and 100-mg groups contained 6 subjects each (4 active and 2 placebo), the 200-mg and 400-mg dosing contained 8 subjects each (6 active and 2 placebo), and the 300-mg dosing group contained 9 subjects (7 active and 2 placebo). The 300-mg infusion over 30 minutes was performed in 1 group of 6 subjects (4 active, 2 placebo).

Drug Product:

Test Product: RX-3341 (delafloxacin meglumine salt; containing [redacted] provided as a 3% sterile solution in 10-mL glass vials

Dose: 50 mg/100 mL, 100 mg/100 mL, 200 mg/100 mL, 300 mg/250 mL, and 400 mg/250 mL (doses were brought to correct infusion volume using sterile 0.9% saline)

Mode of Administration: Intravenous

Batch Number: LO106036

PK Sample Collection:
For the 1-hour infusion, plasma samples were collected at 0 (before infusion), 20 and 40 minutes (during infusion), and 60 minutes (end of infusion). After the infusion, additional blood samples were collected at the following times: 2, 5, 10, 15, 20, 30, and 45 minutes, and 1, 1.5, 2, 3, 4, 5, 6, 8, 12, 16, 24, 30, and 36 hours.

For all infusion regimens, urine was collected immediately before dosing (–2 to 0 hour), and at intervals of 0 to 4, 4 to 8, 8 to 12, 12 to 24, and 24 to 36 hours after the start of the infusion.

Analytical Methods:

- The LC-API/MS/MS method used to measure the delafloxacin concentration in human plasma was validated in the validation report [redacted]3341C.
- The LC-API/MS/MS method used to measure the delafloxacin concentration in human urine was validated in the validation report [redacted]3341D.

	Delafloxacin in Plasma	**Delafloxacin in Urine**
Method Validation #	[redacted]3341C	[redacted]3341D
Range	5.00- 5000 ng/mL	50.0 – 10000 ng/mL
LLOQ	5.0 ng/mL	50.0 ng/mL
Linearity	linear	linear
Accuracy	85-115%	85-115%
Precision	≤ 15%	≤ 15%
Stability	Long-term storage stability has been established for RX-3341 in human plasma for 174 days at -70°C (±10°C). A total of 30 days of long-term storage stability is required to cover the storage of the human plasma study samples at -70°C (±10°C).Stable through three freeze-thaw cycles.	Long-term storage stability has been established for RX-3341 in human urine for 168 days at -70°C (±10°C). A total of 51 days of long-term storage stability is required to cover the storage of the human urine study samples at -70°C (±10°C).Stable through three freeze/thaw cycles.

Pharmacokinetic Assessment:
Plasma parameters included the following:

- AUC_{0-t}: Area under the plasma concentration versus time curve from time 0 to the time of the last quantifiable concentration
- AUC_{0-inf}: Area under the plasma concentration versus time curve from time 0 extrapolated to infinity
- AUC_{0-24}: Area under the plasma concentration versus time curve from time 0 to 24 hours after dosing
- C_{max}: Maximum observed plasma concentration
- K_{el}: Terminal elimination rate constant
- $t_{1/2}$: Terminal elimination half-life
- T_{max}: Time to achieve maximum observed plasma concentration
- CL: clearance
- V_d: apparent volume of distribution

Urine:

- Ae_{0-4}, Ae_{4-8}, Ae_{8-12}, Ae_{12-24}, Ae_{24-36}, Ae_{0-36}: total amount excreted unchanged in urine from 0-12, 12-24, 24-36, or 0-36 hours
- $Fe\%_{0-36}$: fraction of the dose excreted unchanged in urine from 0-36 hours after dosing calculated as (Ae_{0-36}/Dose) x 100
- CLr: renal clearance calculated as Ae_{0-36}/AUC_{0-t}

Statistical Methods:
Pharmacokinetics:
Concentrations of RX-3341 in plasma and urine samples were listed, and summary statistics were presented by time point. All the individual PK parameters were listed, and summary statistics were presented in tables.

For each escalating dosing group, dose proportionality analyses using a power model were performed on AUC_{0-t}, AUC_{0-inf}, and C_{max}. Plots of $\ln(AUC_{0-t}/Dose)$, $\ln(AUC_{0-inf}/Dose)$, $\ln(C_{max}/Dose)$ versus lnDose were presented. The model was defined as: ln(PK parameter) = $\beta 0 + \beta 1$ lnDose, where the PK parameter was AUC_{0-t}, AUC_{0-inf}, and C_{max}. The null hypothesis being tested was that the AUC_{0-t}, AUC_{0-inf}, and C_{max} values were dose proportional, or $\beta 1 = 1$.

Safety: No formal statistical analysis of safety parameters was performed.

Safety Assessment:

- Clinical laboratory evaluations (hematology [including coagulation assays], serum chemistry, and urinalysis)
- Vital signs
- Physical examination findings
- 12-lead electrocardiograms (ECGs)
- Adverse events (AEs)

RESULTS

Study Population:
Please refer to Table 1 for the summary of subject demographics.

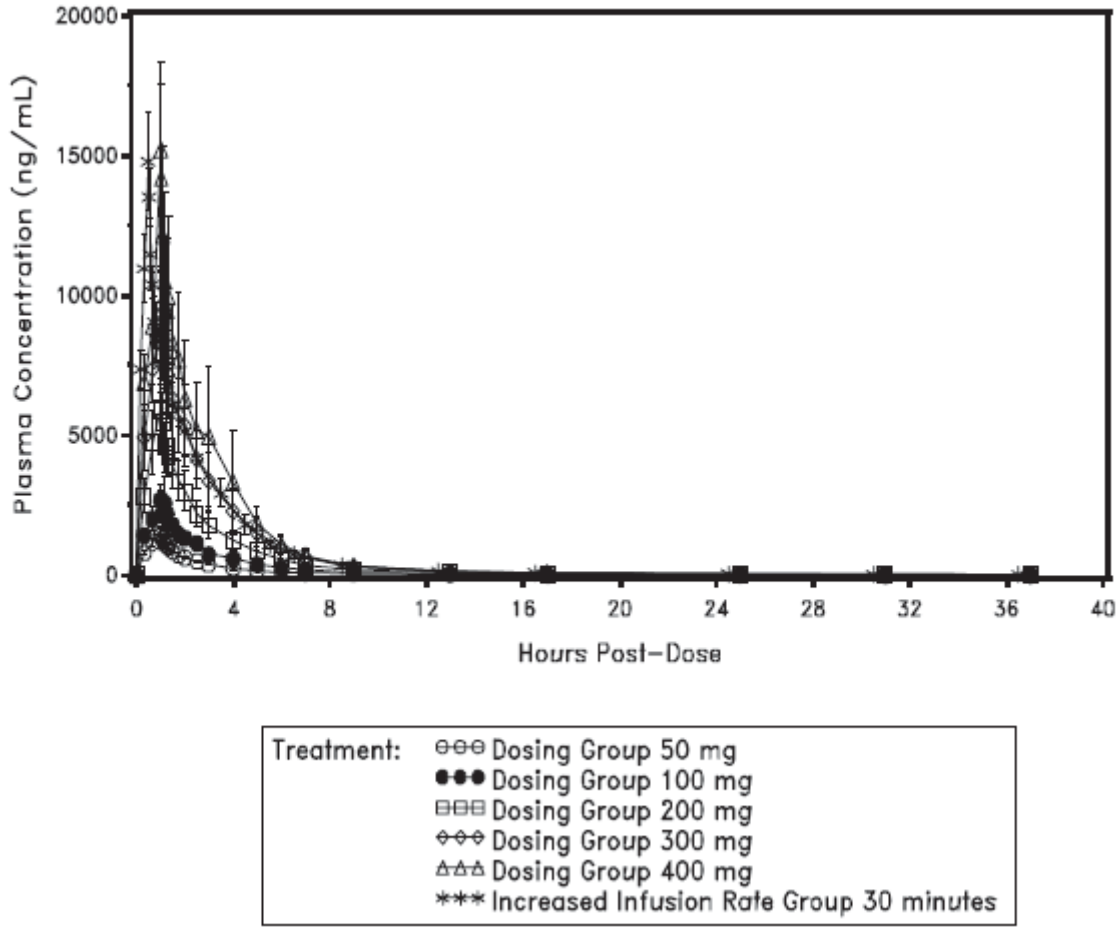
Table 1. Summary of Subject Demographics (Safety Population) *(Adapted from Table 4 in the study report)*

	Group							
Number of Subjects	50 mg (n=4)	100 mg (n=4)	200 mg (n=6)	300 mg (n=7)	400 mg (n=6)	Increased Infusion Rate Group (300 mg Over 30 Minutes) (n=4)	Placebo (n=12)	Overall (N=43)
Age (y)								
Mean (SD)	20.8 (0.50)	24.8 (5.74)	29.8 (8.38)	30.9 (10.95)	28.0 (7.43)	30.0 (11.34)	25.5 (6.19)	27.2 (7.94)
Minimum, maximum	20, 21	20, 33	21, 44	21, 47	20, 37	18, 45	19, 42	18, 47
Sex								
Male	2 (50.0%)	2 (50.0%)	1 (16.7%)	3 (42.9%)	2 (33.3%)	2 (50.0%)	5 (41.7%)	17 (39.5%)
Female	2 (50.0%)	2 (50.0%)	5 (83.3%)	4 (57.1%)	4 (66.7%)	2 (50.0%)	7 (58.3%)	26 (60.5%)
Race								
Caucasian	4 (100.0%)	4 (100.0%)	5 (83.3%)	6 (85.7%)	6 (100.0%)	4 (100.0%)	11 (91.7%)	40 (93.0%)
Black	0	0	1 (16.7%)	1 (14.3%)	0	0	1 (8.3%)	3 (7.0)
Ethnicity								
Hispanic	1 (25.0%)	2 (50.0%)	2 (33.3%)	2 (28.6%)	1 (16.7%)	3 (75.0%)	3 (25.0%)	14 (32.6%)
Not Hispanic	3 (75.0%)	2 (50.0%)	4 (66.7%)	5 (71.4%)	5 (83.3%)	1 (25.0%)	9 (75.0%)	29 (67.4%)
Height (cm)								
Mean (SD)	170.18 (11.762)	168.55 (8.569)	166.42 (4.873)	166.70 (8.416)	168.35 (5.150)	170.53 (8.403)	166.37 (7.047)	167.65 (7.187)
Minimum, maximum	156.0, 180.1	157.3, 177.9	161.5, 172.9	157.2, 181.6	161.7, 175.2	161.5, 179.6	159.1, 179.0	156.0, 181.6
Weight (kg)								
Mean (SD)	70.95 (14.559)	73.65 (9.541)	73.90 (10.149)	74.47 (11.648)	68.45 (11.693)	78.35 (7.630)	65.15 (8.325)	70.91 (10.564)
Minimum, maximum	56.7, 90.1	62.8, 82.3	59.1, 88.9	56.3, 94.0	55.7, 87.0	68.9, 86.2	54.7, 85.3	54.7, 94.0

	Group							
Number of Subjects	50 mg (n=4)	100 mg (n=4)	200 mg (n=6)	300 mg (n=7)	400 mg (n=6)	Increased Infusion Rate Group (300 mg Over 30 Minutes) (n=4)	Placebo (n=12)	Overall (N=43)
Body mass index (kg/m^2)								
Mean (SD)	24.35 (3.196)	25.98 (3.185)	26.63 (3.053)	26.74 (3.218)	24.08 (3.497)	26.88 (0.512)	23.51 (2.191)	25.17 (2.975)
Minimum, maximum	20.8, 27.9	21.5, 28.7	22.4, 30.2	21.9, 30.2	21.2, 29.2	26.4, 27.6	20.7, 26.8	20.7, 30.2

Pharmacokinetics:
Mean plots of plasma concentration versus time by treatment are presented in Figure 1.

Figure 1. Mean (SD) Plasma Concentrations of RX-3341 Over Time by Treatment (Pharmacokinetic Population) *(Adapted from Figure 2 in study report)*



The arithmetic means and CVs for selected PK parameters for all the dose groups are presented in Table 2.

Table 2. Mean (CV%) Plasma Pharmacokinetic Parameters of RX-3341 Infused Over 1-Hour (Pharmacokinetic Population) *(Adapted from Table 6 in the study report)*

	Group									
Parameter	**n**	**50 mg**	**n**	**100 mg**	**n**	**200 mg**	**n**	**300 mg**	**n**	**400 mg**
$AUC_{0-\infty}$ (ng·h/mL)	3	3102.2 (7%)	4	6886.8 (13%)	5	15300.4 (23%)	5	28127.0 (14%)	3	37740.8 (34%)
AUC_{0-t} (ng·h/mL)	4	3185.0 (9%)	4	6803.6 (13%)	6	14948.2 (21%)	6	26042.8 (23%)	6	33838.6 (28%)
C_{max} (ng/mL)	4	1507.5 (9%)	4	3045.0 (21%)	6	6103.3 (16%)	6	9836.7 (17%)	6	15079.2 (25%)
T_{max} (h)[a]	4	1.00 (1.00, 1.00)	4	1.03 (1.00, 1.17)	6	1.00 (1.00, 1.03)	6	1.01 (1.00, 1.25)	6	1.02 (1.00, 1.03)
$t_{1/2}$ (h)	3	2.8 (18%)	4	5.7 (25%)	5	9.4 (8%)	5	8.2 (58%)	3	13.6 (65%)
CL (L/h)	3	16.2 (7%)	4	14.7 (14%)	5	13.7 (25%)	5	10.8 (14%)	3	11.3 (29%)
V_d (L)	3	66.2 (21%)	4	120.5 (30%)	5	184.1 (23%)	5	128.3 (60%)	3	237.7 (87%)

Please refer to Table 3 for the statistical analysis of dose proportionality.

Table 3. Analysis of Dose Proportionality (Pharmacokinetic Population) *(Adapted from Table 7 in the study report)*

Parameter	Estimated Slope for ln(Dose)	90% Confidence Interval for the Slope	P-value (Slope=1)
AUC_{0-t} (ng·h/mL)	1.142	1.048, 1.236	0.017
$AUC_{0-\infty}$ (ng·h/mL)	1.208	1.103, 1.314	0.003
C_{max} (ng/mL)	1.085	0.999, 1.171	0.104

Source: End-of-Text Table 14.2.5.

Note: The power model, ln (parameter) = $\beta_0 + \beta_1$*ln(dose) + error, was used to estimate the slope, corresponding 90% confidence interval, and he *P* value testing dose proportionality (β_1=1).

Reviewer's Comment: Based on results in Table 3, it seems that the increase in delafloxacin AUC was slightly greater than dose proportional over the dose range of 50 mg to 400 mg. However, C_{max} appeared to be dose proportional (p=0.104).

Please refer to Table 4 for the urinary pharmacokinetics.

Table 4. Mean (CV%) Urinary Pharmacokinetic Parameters of RX-3341 (Pharmacokinetic Population)

	Group										
Parameter	n	50 mg	n	100 mg	n	200 mg	n	300 mg	n	400 mg	n	Increased Infusion Rate Group (300 mg over 30 Minutes) (n=4)
$Ae_{0 \text{ to } 36}$ (mg)	4	20.4 (19%)	4	45.7 (19%)	6	64.3 (30%)	6	104.3 (21%)	6	89.4 (44%)	4	102.9 (19%)
CLr (L/h)	3	7.2 (3%)	4	6.8 (27%)	5	4.8 (51%)	5	3.9 (30%)	3	2.5 (14%)	4	3.6 (23%)
$Fe\%_{0 \text{ to } 36}$ (%)	4	40.8 (19%)	4	45.7 (19%)	6	32.2 (30%)	6	34.8 (21%)	6	22.3 (44%)	4	34.3 (19%)

Reviewer's Comment: It seems that the mean fraction of drug excreted unchanged in urine during the first 36 hours after dosing slightly decreased with the increasing dose.

Safety:

Please refer to Table 5 below for the summary of Treatment-Emergent Adverse Events (TEAEs).

Table 5. Summary of Treatment-Emergent Adverse Events: Number of Subjects Reporting at Least 1 Adverse Event, Relationship to Study Drug, and Adverse Events Reported by 2 or More Subjects (Safety Population)

	Number (%) of Subjects by Treatment Group							
	50 mg (n=4)	100 mg (n=4)	200 mg (N=6)	300 mg (n=7)	400 mg (n=6)	Increased Infusion Rate Group (300 mg Over 30 Minutes) (n=4)	Placebo (n=12)	Overall (N=43)
System Organ Class Preferred Term								
Subjects with at least 1 adverse event	0	2 (50.0)	4 (66.7)	1 (14.3)	4 (66.7)	2 (50.0)	4 (33.3)	17 (39.5)
Subjects with at least 1 treatment-related adverse event								
Possibly related	0	0	4 (66.7)	1 (14.3)	4 (66.7)	0	3 (25.0)	12 (27.9)
Probably related	0	1 (25.0)	0	0	0	0	0	1 (2.3)
Very likely/certainly related	0	0	0	0	0	0	0	0
General disorders and administration site conditions	0	0	3 (50.0)	0	4 (66.7)	0	0	7 (16.3)
Infusion site pain	0	0	3 (50.0)	0	4 (66.7)	0	0	7 (16.3)

Reviewer's Comment: It seems that 400 mg IV had more TEAEs, all of which are infusion site pain.

SPONSOR'S CONCLUSIONS

PHARMACOKINETIC

- For each of the AUC parameters (AUC_{0-t}, AUC_{0-inf}), dose proportionality analysis indicated an increase that was slightly greater than proportional, reflected in the trend to lower clearance values at the highest doses, 300 and 400 mg. The analysis for C_{max} resulted in acceptance of the dose proportionality of this parameter over the range of doses studied.
- For the 300-mg dose, which was evaluated at both 30-minute and 1-hour infusions, the exposure measures (AUCs) were independent of the infusion duration. The reduction in T_{max} was accompanied by a marked increase in mean C_{max} for the group with the shorter 30-minute infusion.
- The mean fraction of drug excreted unchanged in urine during the first 36 hours after dosing was lowest for the highest dose (22% at 400 mg); this value was approximately half the mean fractions of drug excreted (~40%) for the 2 lowest doses, 50 mg and 100 mg.
- At all doses, the variability among subjects was modest, with the coefficients of variation for C_{max} and the AUC measures under 30% (with 1 exception at 34%).

SAFETY

- Intravenous infusions of 50, 100, 200, 300, and 400 mg of RX-3341 given over 1 hour, and 300 mg given over 30 minutes, were safe and well tolerated by the healthy subjects in this study based on analysis of AEs, clinical laboratory results, vital sign measurements, physical examination findings, and ECG results.

REVIEWER ASSESSMENT:

- Following single dose administration of IV delafloxacin from 50 mg to 400 mg, the increase of AUC_{0-t} and AUC_{0-inf} of delafloxacin was slightly greater than dose proportional. The increase of C_{max} was approximately dose proportional.
- It seems that the major TEAEs were infusion site pain and the frequency of TEAEs increased with the increase of dose.

4.5.8 Study No.: RX-3341-102

A RANDOMIZED, DOUBLE-BLIND, PLACEBO-CONTROLLED, SAFETY, TOLERABILITY, AND PHARMACOKINETIC STUDY OF RX-3341 (DELAFLOXACIN [redacted] SOLUTION) IN HEALTHY SUBJECTS FOLLOWING TWICE-DAILY INTRAVENOUS ADMINISTRATION

Date(s): 03 August 2007 to 15 November 2007
Sponsor: Melinta Therapeutics, Inc, 300 George Street, Suite 301, New Haven, CT 06511
Clinical Site: Single site: PPD Phase I Clinic, located at 7551 Metro Center Drive, Suite 200, Austin, TX 78744
Analytical Site: [redacted]

OBJECTIVE(S):
The primary objective of this study was to determine the safety, tolerability, and pharmacokinetics (PK) of delafloxacin after single- and multiple-day intravenous dosing.

METHODS
Study Design:
This was a double-blind, randomized, placebo-controlled study of delafloxacin administered intravenously on a single day or over multiple days to healthy subjects. There were 2 treatment phases: Part 1 and Part 2. Each part of the study included a 21-day screening period, a check-in day, a treatment phase, a check-out day, and a follow-up period. After subjects completed their screening and enrollment procedures, they checked into the clinic on Day –1 for baseline assessments and then proceeded to the treatment phase on Day 1.

Treatment Phase in Part 1: During this 1-day treatment phase, subjects were sequentially assigned to the following treatment groups:
• 300 mg of delafloxacin or placebo infused over 1 hour
• 600 mg of delafloxacin or placebo infused over 1 hour
A total of 16 subjects were planned for Part 1, with each treatment group consisting of 8 subjects randomly assigned to either delafloxacin or placebo (6 active and 2 placebo). On Day 1, subjects received two 1-hour continuous intravenous infusions of either delafloxacin (300 or 600 mg) or placebo at 0 hours and at 12 hours.

Treatment Phase in Part 2: During this 10-day treatment phase, subjects were sequentially assigned to the following treatment groups:
• 150 mg of delafloxacin or placebo infused over 1 hour
• 300 mg of delafloxacin or placebo infused over 1 hour
• 450 mg of delafloxacin or placebo infused over 1 hour

A total of 36 subjects were planned for Part 2, with each treatment group consisting of 12 subjects randomly assigned to receive either delafloxacin or placebo (8 active and 4 placebo). On Day 1, subjects received a single 1-hour continuous intravenous infusion of either delafloxacin (150, 300, or 450 mg) or placebo at 0 hours. On Days 2 through 10, subjects received twice-daily intravenous infusions of delafloxacin or placebo 24 hours after the start of the infusion

administered on Day 1, with the second infusion administered 12 hours later. Thereafter, infusions occurred every 12 hours until the last infusion on Day 10.

During both treatment phases of the study, blinded safety data were reviewed by the sponsor and principal investigator before progression to the next dose group. Higher doses were administered only after the review of blinded safety data suggested that it was safe to do so.

Reviewer's Comment: According to the Sponsor, the 300-mg, multiple-day dose group of the study was stopped prematurely because of significant irritation at the infusion site experienced by all 8 subjects in the 300-mg, multiple-day dose group. Therefore, no subjects were administered the planned 450-mg, multiple-day dose component of the study. As a result, 40 subjects were enrolled rather than the planned 52 subjects.

PK Sample Collection:
For treatment groups in Part 1, plasma and urine samples were collected at the following time points (all sampling times were relative to the start of the first infusion): before dosing and at 0.5, 1 (end of infusion), 1.083, 1.167, 1.33, 1.5, 2, 3, 4, 6, 8, 12, 12.5, 13, 13.083, 13.167, 13.33, 13.5, 14, 16, 20, 24, 30, and 36 hours. Urine samples were collected over the intervals of –2 to 0 hours, 0 to 12 hours, 12 to 24 hours, and 24 to 36 hours after the start of the first infusion.

For treatment groups in Part 2, blood samples were obtained according to the following schedule (all sampling times are relative to the start of the first infusion):
Day 1: before dosing and at 0.5, 1 (end of infusion), 1.083, 1.167, 1.33, 1.5, 2, 3, 4, 6, 8, 12, 16, and 24 hours after infusion.
Days 3, 8, and 9: –2 to 0 hours (before the morning dosing).
Day 10: before the morning dosing and at 0.5, 1 (end of infusion), 1.083, 1.167, 1.33, 1.5, 2, 3, 4, 6, 8, and 12 hours after the start of the first infusion.

Urine samples were collected over the intervals of –2 to 0 hours, 0 to 12 hours, and 12 to 24 hours after the start of the first infusion on Day 1 and from –2 to 0 hours and 0 to 12 hours after the start of the first infusion on Day 10.

Analytical Methods:
- The LC-API/MS/MS method used to measure the delafloxacin concentration in human plasma was validated in the validation report (b) (4) 3341C.
- The LC-API/MS/MS method used to measure the delafloxacin concentration in human urine was validated in the validation report (b) (4) 3341D.

	Delafloxacin in Plasma	**Delafloxacin in Urine**
Method Validation #	(b) (4) 3341C	(b) (4) 3341D
Range	5.00- 5000 ng/mL	50.0 – 10000 ng/mL
LLOQ	5.0 ng/mL	50.0 ng/mL
Linearity	linear	linear
Accuracy	85-115%	85-115%

Precision	⩽ 15%	⩽ 15%
Stability	Long-term storage stability has been established for RX-3341 in human plasma for 374 days at -20°C (±10°C) and -70°C (±10°C). A total of 55 days (time from first sample drawn to the last sample analyzed) of long-term stability is required to cover the storage of the human plasma study samples at -70°C (±10°C). Stable through three freeze-thaw cycles.	Long-term storage stability has been established for RX-3341 in human urine for 227 days at -20°C (±10°C) and -70°C (±10°C). A total of 55 days (time from first sample drawn to the last sample analyzed) of long-term stability is required to cover the storage of the human urine study samples at -70°C (±10°C).Stable through three freeze/thaw cycles.

Pharmacokinetic Assessment:
Pharmacokinetic parameters included the following:
Group 1
Plasma:

- AUC_{0-t}: Area under the plasma concentration versus time curve from time 0 to the time of the last quantifiable concentration
- AUC_{0-tau}: area under the concentration versus time curve over the dosing interval of 12 hours
- AUC_{0-inf}: Area under the plasma concentration versus time curve from time 0 extrapolated to infinity
- $AUC_{0-24:}$ Area under the plasma concentration versus time curve from time 0 to 24 hours after dosing
- C_{max}: Maximum observed plasma concentration
- Kel: Terminal elimination rate constant
- $t_{1/2}$: Terminal elimination half-life
- T_{max}: Time to achieve maximum observed plasma concentration

Urine:

- Ae_{0-12}, Ae_{12-24}, Ae_{24-36}, Ae_{0-36}: total amount excreted unchanged in urine from 0-12, 12-24, 24-36, or 0-36 hours
- $Fe\%_{0-36}$: fraction of the dose excreted unchanged in urine from 0-36 hours after dosing calculated as (Ae_{0-36}/Dose) x 100
- CLr: renal clearance calculated as Ae_{0-36}/AUC_{0-t}

Group 2:
Plasma PK parameters Day 1 of Part 2 were similar to those PK parameters in Part 1 and included the following additional parameters:

- CL: clearance calculated as Dose/$AUC_{0-\infty}$
- MRT: mean residence time
- Vss: Volume of distribution under steady-state conditions, calculated as CLxMRT

Statistical Methods:

Pharmacokinetics:

Summary statistics for all the PK parameters by treatment group were presented in tables and included n, mean, geometric mean (for only $AUC_{0-\tau}$, AUC_{0-t}, $AUC_{0-\infty}$, and C_{max}), median, SD, CV, and range (minimum, maximum). Pharmacokinetic parameters were also presented for individual subjects in a data listing. If possible, the PK parameters were determined from the plasma and urine concentrations of delafloxacin each subject using actual sampling times by noncompartmental methods.

Safety: Adverse events were summarized by the number of events and by the number of subjects who experienced them. Adverse events were also summarized by system organ class, relationship, severity, and seriousness. Clinical laboratory (hematology, serum chemistry, and urinalysis) and vital sign measurements were summarized by time point and change from Baseline. Electrocardiogram results were listed and summarized by time point. Physical examination findings were listed.

RESULTS

Study Population:

Please refer to Table 1 for the summary of subject demographics and baseline characteristics.

Table 1. Summary of Demographics (Safety Population) *(Adapted from Table 4 in the study report)*

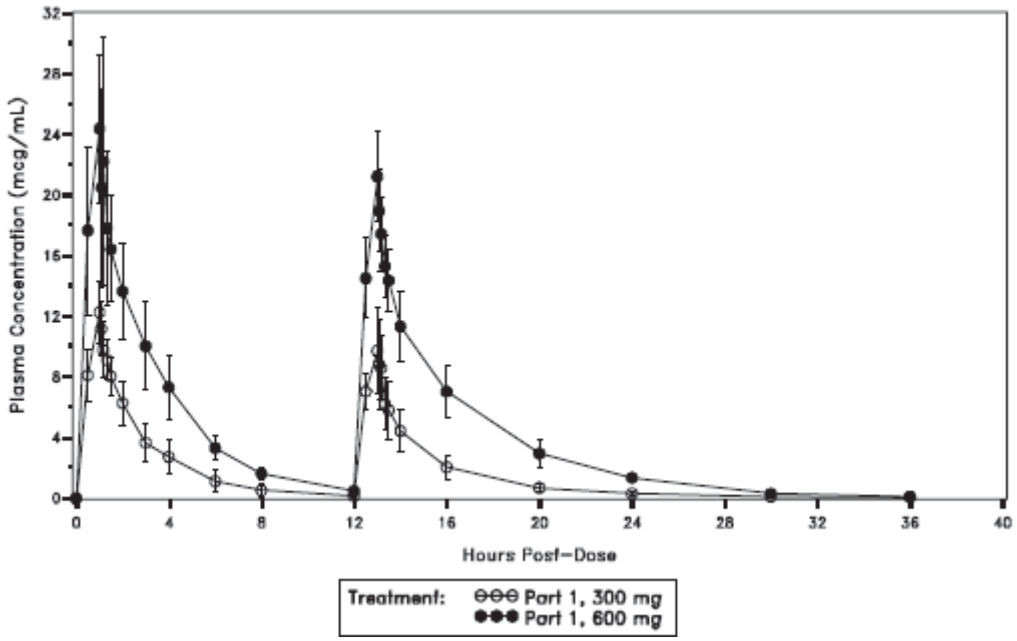
	Group					
Number of Subjects	300 mg Single Day (n=6)	600 mg Single Day (n=6)	150 mg Multiple Day (n=8)	300 mg Multiple Day (n=8)	Placebo (n=12)	Overall (N=40)
Age (years)						
Mean (SD)	26.5 (11.04)	27.0 (9.94)	33.5 (16.51)	32.1 (12.78)	36.7 (12.34)	32.2 (12.85)
Minimum, maximum	18, 48	18, 43	18, 62	21, 59	21, 57	18, 62
Sex						
Male	1 (16.7%)	4 (66.7%)	7 (87.5%)	4 (50.0%)	10 (83.3%)	26 (65.0%)
Female	5 (83.3%)	2 (33.3%)	1 (12.5%)	4 (50.0%)	2 (16.7%)	14 (35.0%)
Race						
Caucasian	5 (83.3%)	3 (50.0%)	7 (87.5%)	8 (100.0%)	10 (83.3%)	33 (82.5%)
Black	0	2 (33.3%)	1 (12.5%)	0	2 (16.7%)	5 (12.5%)
Asian	0	1 (16.7%)	0	0	0	1 (2.5%)
Other	1 (16.7%)	0	0	0	0	1 (2.5%)
Ethnicity						
Hispanic	4 (66.7%)	0	1 (12.5%)	1 (12.5%)	4 (33.3%)	10 (25.0%)
Not Hispanic	2 (33.3%)	6 (100.0%)	7 (87.5%)	7 (87.5%)	8 (66.7%)	30 (75.0%)
Height (cm)						
Mean (SD)	163.58 (9.826)	170.33 (11.810)	173.10 (10.086)	171.48 (8.711)	173.69 (6.918)	171.11 (9.345)
Minimum, maximum	154.5, 181.5	150.0, 183.0	155.0, 190.5	158.5, 183.0	154.0, 181.0	150.0, 190.5
Weight (kg)						
Mean (SD)	64.90 (5.047)	70.67 (14.322)	76.55 (14.384)	73.24 (13.723)	76.38 (11.025)	73.21 (12.320)
Minimum, maximum	58.9, 72.7	55.8, 95.7	56.2, 95.6	51.5, 92.1	62.5, 94.6	51.5, 95.7
Body mass index (kg/m^2)						
Mean (SD)	24.33 (2.318)	24.60 (5.659)	25.40 (3.170)	24.76 (3.241)	25.31 (3.288)	24.97 (3.420)
Minimum, maximum	20.6, 27.4	18.7, 31.6	20.9, 30.7	18.7, 27.5	20.8, 30.5	18.7, 31.6

Note: Percentages were based on the number of subjects in the safety population in each group.
Categorical data were presented as n (%).

Pharmacokinetics:

In Part 1, the PK population consisted of those subjects who received 2 infusions of 300 mg or 600 mg of delafloxacin 12 hours apart. Mean plots (linear scale) of plasma concentration versus time for each Part 1 treatment are presented in Figure 1.

Figure 1. Mean (±SD) Plasma Concentrations of Delafloxacin Over Time by Treatment (Part 1 Pharmacokinetic Population, Linear Scale) *(Adapted from Figure 2 in the study report)*



The arithmetic means and coefficients of variation for selected PK parameters for both dose groups in Part 1 are presented in Table 2.

Table 2. Mean (CV) Plasma Pharmacokinetic Parameters of Delafloxacin (Part 1) (Pharmacokinetic Population) (Adapted from Table 5 in the study report)

		Group		
Parameter (units)	**n**	**300 mg Single Day**	**n**	**600 mg Single Day**
$AUC_{0-\infty}$ (µg·h/mL)	6	59.296 (26%)	6	154.615 (19%)
AUC_{0-t} (µg·h/mL)	6	58.959 (26%)	6	153.985 (20%)
$AUC_{0-\tau}$ [a] (µg·h/mL)	6	30.649 (27%)	6	72.706 (23%)
C_{max} (µg/mL)	6	12.272 (17%)	6	25.383 (24%)
$t_{1/2}$ (h)	6	3.9 (17%)	6	3.4 (11%)

[a] τ = 12 hours.

Based on visual inspection, the increase in C_{max} is approximately proportional to increases in dose. However, the increase in AUC is disproportionately greater to increases in dose. At both doses, the variability among subjects was modest with the CVs for both C_{max} and AUC under 30%. Mean terminal elimination half-life ($t_{1/2}$) was comparable between the 2 doses.

Please refer to Table 3 for the urinary PK parameters.

Table 3. Mean (CV) Urinary Pharmacokinetic Parameters of Delafloxacin (Part 1) (Pharmacokinetic Population) *(Adapted from Table 6 in study report)*

		Group		
Parameter (units)	**n**	**300 mg Single Day**	**n**	**600 mg Single Day**
$Ae_{0\text{-}36}$ (mg)	6	217.0 (22%)	6	399.9 (24%)
CL_r (L/h)	6	3.86 (28%)	6	2.72 (37%)
$Fe\%_{0\text{-}36}$ (%)	6	36.2 (22%)	6	33.4 (24%)

In Part 2, the PK population consisted of those subjects who received either 150-mg or 300-mg infusions of delafloxacin with 1 infusion on Day 1 and 2 infusions 12 hours apart on Days 2 through 10. Subjects in the PK population received the infusions of delafloxacin at one of the following dose levels: 150 mg or 300 mg. Subjects at the 300-mg dose were actually dosed for only 8 days instead of the planned 10 days. For ease of reference, the general label "Day 10" will be used with the understanding that for the 300-mg group it is actually Day 8.

Please refer to Figure 2 and Figure 3 for the mean plots of plasma concentration versus time for each Part 2 treatment.

Figure 2. Mean (±SD) Plasma Concentrations of Delafloxacin Over Time by Treatment, Day 1 (Part 2 Pharmacokinetic Population, Linear Scale) *(Adapted from Figure 3 in the study report)*

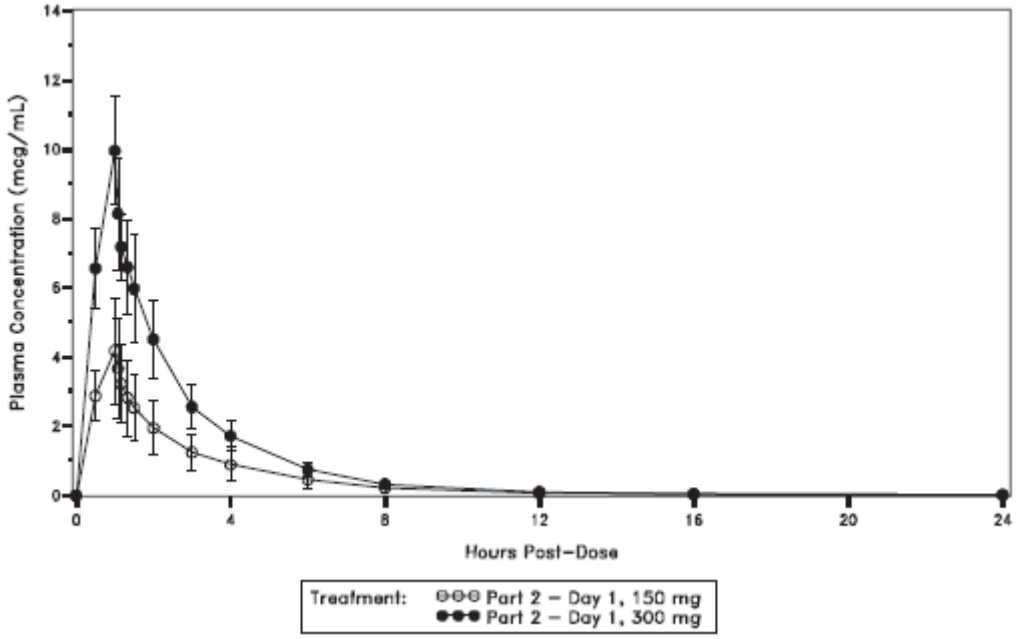
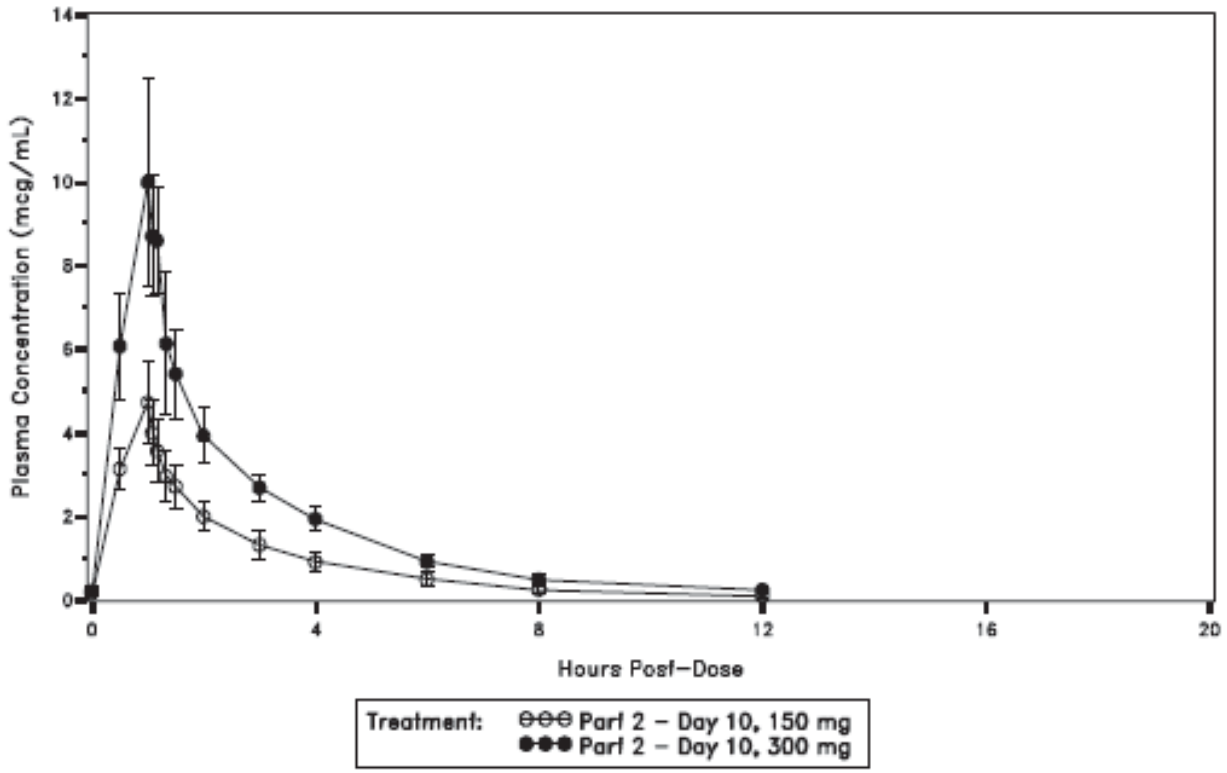


Figure 3. Mean (±SD) Plasma Concentrations of Delafloxacin Over Time by Treatment, Day 10 (Part 2 Pharmacokinetic Population, Linear Scale)



Please refer to Table 4 for the arithmetic means and coefficients of variation for selected PK parameters for Days 1 and 10 for both dose groups.

Table 4. Mean (CV) Plasma Pharmacokinetic Parameters of Delafloxacin (Part 2) (Pharmacokinetic Population)

	Group							
	Day 1				Day 10			
Parameter (units)	n	150 mg Multiple Day	n	300 mg Multiple Day	n	150 mg Multiple Day	n	300 mg Multiple Day
$AUC_{0-\tau}$[a] (µg·h/mL)	7	11.540 (27%)	8	22.168 (21%)	7	11.359 (20%)	4	24.317 (16%)
AUC_{0-inf} (µg·h/mL)	7	12.098 (27%)	7	21.831 (18%)	-	-	-	-
C_{max} (µg·h/mL)	7	4.650 (18%)	8	9.990 (16%)	7	4.733 (20%)	4	9.993 (25%)
$t_{1/2}$ (h)	7	5.3 (25%)	7	5.6 (27%)	-	-	-	-
CL (L/h)	7	13.1 (23%)	7	14.2 (19%)	7	13.6 (18%)	4	12.6 (16%)
V_{ss} (L)	7	39.8 (27%)	7	35.7 (17%)	-	-	-	-
R_{ac}[b]	-	-	-	-	7	1.01 (18%)	4	1.04 (6%)

Note: Subject 2004 in the 150-mg, multiple-day group was excluded from summary statistics because he received only 108 mL of the 250 mL intended for infusion.
Note: All subjects in Group D (300-mg, multiple-day) had concentration samples collected after the evening dosing on Day 8 instead of on Day 10 as originally planned.
[a] τ = 12 hours.
[b] $R_{ac} = AUC_{0-\tau\ (Day\ 10)} / AUC_{0-\tau\ (Day\ 1)}$.

Reviewer's Comment: It appears the increase in AUC and C_{max} of delafloxacin are approximately dose proportional on both Day 1 and Day 10 following IV administration of 150 and 300 mg BID delafloxacin (b) (4) *solution.*

Please refer to Table 5 for the urinary PK parameters.

Table 5 Mean (CV) Urinary Pharmacokinetic Parameters of Delafloxacin (Part 2) (Pharmacokinetic Population)

	Group							
	Day 1				Day 10			
Parameter (units)	n	150 mg Multiple Day	n	300 mg Multiple Day	n	150 mg Multiple Day	n	300 mg Multiple Day
Ae_{0-12} (mg)	7	56.8 (17%)	8	121.1 (15%)	7	63.2 (25%)	5	101.0 (33%)
CL_r (L/h)	7	5.25 (33%)	8	5.74 (29%)	7	5.72 (32%)	4	4.00 (41%)
$Fe\%_{0-24}$ (%)	7	38.9 (16%)	8	41.4 (15%)	-	-	-	-
$Fe\%_{0-12}$ (%)	-	-	-	-	7	42.2 (25%)	5	33.7 (33%)

Note: All subjects in the 300-mg, multiple-day group had concentration samples collected after the evening dosing on Day 8 instead of on Day 10 as originally planned.
Subject 2004 in the 150-mg, multiple-day group was excluded from Day 1 summary statistics because he received only 108 mL of the 250 mL intended for infusion.

Safety:
Overall, 30 subjects (75.0%) reported 1 or more TEAEs. The highest percentage of subjects with at least 1 TEAE and the highest percentage of subjects with AEs considered study drug related were reported for the 600-mg, single-day (100.0%) and the 300-mg, multiple-day (100.0%) dose groups. The percentage of AEs reported for the active study drug groups other than the 600-mg, single-day and 300-mg, multiple-day dose groups was similar across the treatment groups and comparable to the placebo group.

The majority of AEs were classified as possibly related to study drug and mild in severity.

The majority of AEs resolved without treatment, and no deaths or SAEs were reported. Eight subjects discontinued because of AEs.

SPONSOR'S CONCLUSIONS
PHARMACOKINETIC

- After multiple days of dosing (Part 2), dose proportionality for both peak (C_{max}) and total exposure (AUC) was demonstrated for twice-daily, 150- and 300-mg doses of delafloxacin.
- After 1 day of twice-daily dosing (Part 1), visual inspection indicated that, while increases in Cmax were approximately proportional to increases in dose, increases in AUC were disproportionately greater for twice-daily, 300- and 600-mg doses of delafloxacin.
- No drug accumulation occurred after multiple days of twice-daily dosing of delafloxacin.
- The mean elimination half-life ($t_{1/2}$) was between 3.4 and 5.6 hours.

- Under steady state conditions, a little over one-third of the administered dose was excreted unchanged in urine during the 12-hour dosing interval. Mean renal clearance was about 5 L/h.

SAFETY

- Vital sign measurements and electrocardiogram results showed that intravenous infusions of delafloxacin administered on a single day and over multiple days were well tolerated. Based on analysis of AEs, clinical laboratory results, and physical examination findings, intravenous infusions of 600 mg of delafloxacin administered on a single day and of 300 mg of delafloxacin administered over multiple days were less well tolerated by the healthy subjects in this study than intravenous infusions of 300 mg of delafloxacin administered on a single day and of 150 mg of delafloxacin administered over multiple days.

REVIEWER ASSESSMENT:

- The increase of AUC were greater than dose proportional for single dose 300 mg and 600 mg, but approximately dose proportional for multiple doses of 150 mg and 300 mg BID for 10 days.

APPEARS THIS WAY ON ORIGINAL

4.5.9 Study No.: RX-3341-103

A PHASE 1, SINGLE AND MULTIPLE INTRAVENOUS DOSE SAFETY, TOLERABILITY, AND PHARMACOKINETIC STUDY OF RX-3341 IN HEALTHY SUBJECTS

Date(s): 09 April 2008 to 17 June 2008
Sponsor: Melinta Therapeutics, Inc, 300 George Street, Suite 301, New Haven, CT 06511
Clinical Site: Single site: PPD Phase I Clinic, located at 7551 Metro Center Drive, Suite 200, Austin, TX 78744
Analytical Site: (b) (4)

OBJECTIVE(S):

- The primary objectives were to determine the relative intravenous (IV) pharmacokinetics of 2 formulations of delafloxacin and to determine the safety and tolerability of multiple IV doses of delafloxacin in healthy male and female subjects.
- The secondary objective was to determine the pharmacokinetics of multiple IV doses of delafloxacin, administered as Formulation B, in healthy male and female subjects.

METHODS

Study Design: This was a randomized, double-blind study conducted in 32 subjects who were assigned to either Group 1 or Group 2. In the first part of this study, the relative pharmacokinetics and safety of 2 formulations (A and B) of delafloxacin were evaluated in a crossover design. On Day 1, 12 subjects in Group 1 received a single, 1-hour, continuous IV infusion of either delafloxacin Formulation A (Treatment A) or delafloxacin Formulation B (Treatment B), according to the treatment assignment, each at a dose of 300 mg. Six subjects received Formulation A, and 6 received Formulation B. On Day 3, subjects received a second dose of the alternate formulation in a crossover fashion.

In the second part of this study, the pharmacokinetics and safety of multiple doses of delafloxacin Formulation B were evaluated. Two dose levels were studied; the low dose was 300 mg and the high dose was 450 mg. Blinded safety data and unblinded PK data from Group 1 were reviewed by the sponsor and principal investigator before dosing in Group 2 was permitted.

On Day 1, 20 subjects in Group 2 received a single, 1-hour, continuous IV infusion of either low-dose Formulation B (300 mg), high-dose Formulation B (450 mg), or placebo according to treatment assignment. On Days 2 through 14, subjects received 2 daily IV infusions (BID).

Drug Product:
Group 1:
Test Product:
Delafloxacin Formulation A: (b) (4)
Delafloxacin Formulation B:

Dose: Single 300-mg dose
Mode of Administration: IV infusion

Batch Number: Formulation A: L0200272
Formulation B: L0202168

Group 2:
Test Product: Delafloxacin Formulation B
Doses: Delafloxacin 300 mg
Delafloxacin 450 mg
Mode of Administration: IV infusion
Batch Number: L0202168

PK Sample Collection:
For Group 1, blood samples were collected for pharmacokinetic (PK) assessments before dosing and at 0.33, 0.66, 1 (end of infusion), 1.083, 1.167, 1.33, 1.5, 2, 3, 4, 5, 6, 8, 12, 16, 20, 24, 30, and 36 hours after start of the infusion on Days 1 and 3. Urine was collected before dosing from –2 to 0 hours and at 0 to 12, 12 to 24, and 24 to 36 hours after start of the infusion on Days 1 and 3.

For Group 2, blood samples were collected for PK assessments at the following times:
Day 1: Before dosing and at 0.5, 1 (end of infusion), 1.083, 1.167, 1.33, 1.5, 2, 3, 4, 5, 6, 8, 12, 16, and 24 hours
after start of the infusion.
Days 3, 7, and 10: Before the morning and evening infusions.
Day 14: Before morning dosing and at 0.5, 1 (end of morning infusion), 1.083, 1.167, 1.33, 1.5, 2, 3, 4, 5, 6, 8, and 12 hours after the morning infusion

For Group 2, urine samples were collected at the following times:
Day 1: Before dosing from –2 to 0 hours and at 0 to 12, 12 to 24 hours after start of the infusion.
Day 14: Before dosing from –2 to 0 hours and at 0 to 12 hours after the start of the morning infusion.

Analytical Methods:
- The LC-API/MS/MS method used to measure the delafloxacin concentration in human plasma was validated in the validation report (b) (4) 3341C.
- The LC-API/MS/MS method used to measure the delafloxacin concentration in human urine was validated in the validation report (b) (4) 3341D.

	Delafloxacin In Plasma	**Delafloxacin in Urine**
Method Validation #	(b) (4) 3341C	(b) (4) 3341D
Range	5.00- 5000 ng/mL	50.0 – 10000 ng/mL
LLOQ	5.0 ng/mL	50.0 ng/mL
Linearity	linear	linear
Accuracy	85-115%	85-115%
Precision	⩽ 15%	⩽ 15%

Stability	Long-term storage stability has been established for RX-3341 in human plasma for 374 days at -20°C (±10°C) and -70°C (±10°C). A total of 55 days (time from first sample drawn to the last sample analyzed) of long-term stability is required to cover the storage of the human plasma study samples at -70°C (±10°C). Stable through three freeze-thaw cycles.	Long-term storage stability has been established for RX-3341 in human urine for 168 days at -20°C (±10°C) and -70°C (±10°C). A total of 55 days (time from first sample drawn to the last sample analyzed) of long-term stability is required to cover the storage of the human urine study samples at -70°C (±10°C).Stable through three freeze/thaw cycles.

Pharmacokinetic Assessment:
Pharmacokinetic parameters included the following:
Group 1
Plasma:

- AUC_{0-t}: Area under the plasma concentration versus time curve from time 0 to the time of the last quantifiable concentration
- AUC_{0-tau}: area under the concentration versus time curve over the dosing interval of 12 hours
- AUC_{0-inf}: Area under the plasma concentration versus time curve from time 0 extrapolated to infinity
- $AUC_{0-24:}$ Area under the plasma concentration versus time curve from time 0 to 24 hours after dosing
- C_{max}: Maximum observed plasma concentration
- K_{el}: Terminal elimination rate constant
- $t_{1/2}$: Terminal elimination half-life
- Tmax: Time to achieve maximum observed plasma concentration

Urine:

- Ae_{0-4}, Ae_{4-8}, Ae_{8-12}, Ae_{12-24}, Ae_{24-36}, Ae_{0-36}: total amount excreted unchanged in urine from 0-4, 4-8, 8-12, 12-24, 24-36, or 0-36 hours
- $Fe\%_{0-36}$: fraction of the dose excreted unchanged in urine from 0-36 hours after dosing calculated as (Ae_{0-36}/Dose) x 100
- CLr: renal clearance calculated as Ae_{0-36}/AUC_{0-t}

Group 2:
Day 1 Plasma:

- Similar to Group 1
- MRT: mean residence time
- Vss: Volume of distribution under steady-state conditions, calculated as CLxMRT

Day 1 Urine:

- Same as Group 1

Day 14 Plasma:

- Similar to Group 1
- Rac: accumulation ratio calculated as AUC $_{0\text{-tau Day 14}}$/ AUC $_{0\text{-tau Day 1}}$

Day 14 Urine:

- Same as Group 1

Statistical Methods:

Pharmacokinetics:

For Group 1, log-transformed PK parameters $AUC_{0\text{-tau}}$, $AUC_{0\text{-t}}$, $AUC_{0\text{-inf}}$, and C_{max} were analyzed by a mixed-effects model using treatment, sequence, and period as fixed effects and subject within sequence as random effect. The 90% confidence interval of the ratio of the geometric least squares means of Formulation B to Formulation A was provided.

For Group 2, dose-proportionality analyses using a power model were performed on the AUC0-tau, AUC0-tt, AUC0-inf, and Cmax. Mean and individual plots of AUC0-tau, AUC0-t, AUC0-inf, and Cmax versus dose were presented. The model was defined as:

PK parameter = $\beta 0 + \beta 1$ logeDose,

Where the PK parameter was logeAUC or logeCmax. The null hypothesis being tested was that the AUC and Cmax values were dose proportional, or $\beta 1 = 1$.

Reviewer's Comment: The study design and statistical approach to evaluate a bioequivalence between two formulations (Formulation A vs. Formulation B) are in line with the requirement specified in the "Draft Guidance for Industry: Bioavailability and Bioequivalence Studies Submitted in NDAs and INDs" and "FDA guidance for industry Statistical Approaches to Establishing Bioequivalence".

Safety: No formal statistical analysis of safety parameters was performed.

Safety Assessment: All safety assessments, including AEs, clinical laboratory assessments (hematology [including coagulation assays], serum chemistry, and urinalysis), vital sign measurements, physical examination findings, and 12-lead ECG measurements, were summarized with descriptive statistics, where appropriate, and presented in the data listings. The date and time of study drug administration were also presented in a data listing.

RESULTS

Study Population:

Please refer to Table 1 for the summary of subject demographics and baseline characteristics.

Table 1. Summary of Subject Demographics and Baseline Characteristics –Part 1 (Safety Population) *(Adapted from Table 6 in the study report)*

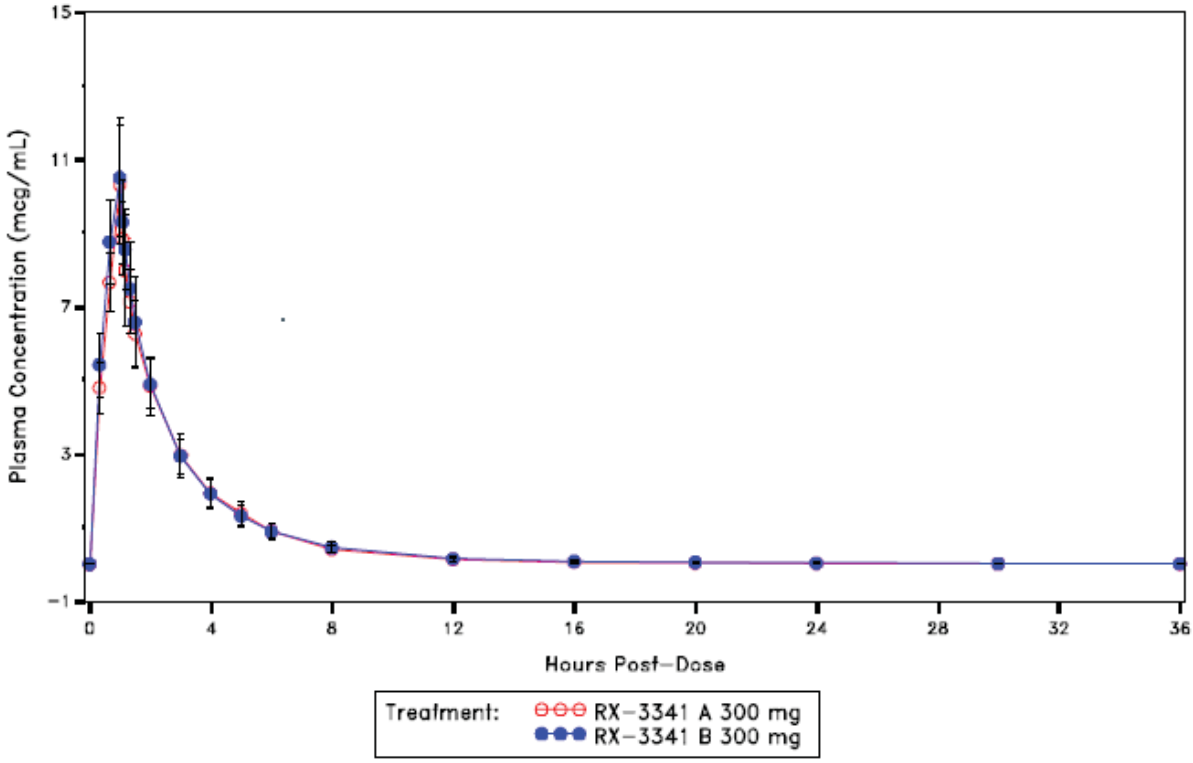
	Group 1: Sequence[a]		Group 2: Treatment[b]				
Number of Subjects	AB (n=6)	BA (n=6)	C (n=8)	D (n=8)	E (n=2)	F (n=2)	Overall (N=32)
Age (y)							
Mean (SD)	23.5 (2.88)	27.0 (8.51)	27.6 (6.95)	26.4 (4.00)	41.0 (16.97)	21.0 (2.83)	26.8 (7.39)
Minimum, maximum	20, 27	19, 43	20, 42	21, 32	29, 53	19, 23	19, 53
Sex							
Male	0	0	6 (75.0%)	7 (87.5%)	2 (100.0%)	1 (50.0%)	16 (50.0%)
Female	6 (100.0%)	6 (100.0%)	2 (25.0%)	1 (12.5%)	0	1 (50.0%)	16 (50.0%)
Race							
Caucasian	3 (50.0%)	5 (83.3%)	8 (100.0%)	6 (75.0%)	2 (100.0%)	2 (100.0%)	26 (81.3%)
Black	1 (16.7%)	1 (16.7%)	0	2 (25.0%)	0	0	4 (12.5%)
Asian	1 (16.7%)	0	0	0	0	0	1 (3.1%)
Other	1 (16.7%)	0	0	0	0	0	1 (3.1%)
Ethnicity							
Hispanic	1 (16.7%)	1 (16.7%)	5 (62.5%)	3 (37.5%)	0	2 (100.0%)	12 (37.5%)
Not Hispanic	5 (83.3%)	5 (83.3%)	3 (37.5%)	5 (62.5%)	2 (100.0%)	0	20 (62.5%)
Height (cm)							
Mean (SD)	163.55 (7.090)	160.88 (4.400)	172.88 (9.497)	174.00 (9.012)	178.75 (5.303)	164.00 (7.778)	168.97 (9.453)
Minimum, maximum	155.0, 173.5	155.5, 166.8	161.0, 192.0	158.5, 187.0	175.0, 182.5	158.5, 169.5	155.0, 192.0
	Group 1: Sequence[a]		Group 2: Treatment[b]				
Number of Subjects	AB (n=6)	BA (n=6)	C (n=8)	D (n=8)	E (n=2)	F (n=2)	Overall (N=32)
Weight (kg)							
Mean (SD)	64.40 (7.124)	65.97 (5.299)	77.70 (11.273)	78.18 (11.566)	77.25 (13.930)	63.35 (10.253)	72.20 (11.148)
Minimum, maximum	55.2, 72.6	57.5, 71.6	64.5, 101.2	58.2, 95.2	67.4, 87.1	56.1, 70.6	55.2, 101.2
Body mass index (kg/m^2)							
Mean (SD)	24.13 (2.633)	25.52 (2.418)	25.93 (2.295)	25.76 (2.910)	24.30 (5.798)	23.45 (1.626)	25.22 (2.653)
Minimum, maximum	21.0, 27.8	22.3, 29.6	23.0, 29.8	21.2, 28.3	20.2, 28.4	22.3, 24.6	20.2, 29.8

Note: Percentages were based on the number of subjects in the safety population in each sequence (for Group 1) or treatment (for Group 2).
Categorical data were presented as n (%).
[a]A = delafloxacin Formulation A 300 mg.
B = delafloxacin Formulation B 300 mg.
[b]C = delafloxacin Formulation B 300 mg.
D = delafloxacin Formulation B 450 mg.
E = delafloxacin Formulation B placebo low dose.
F = delafloxacin Formulation B placebo high dose.

Pharmacokinetics:

Mean plots (linear scale) of plasma concentration versus time for each Group 1 formulation are presented in Figure 1.

Figure 1. Mean (±SD) Plasma Concentrations of Delafloxacin Over Time by Formulation (Group 1 Pharmacokinetic Population, Linear Scale)



The arithmetic means and CVs for selected PK parameters for both formulations in Group 1 are presented in Table 2.

Table 2. Mean (CV) Plasma Pharmacokinetic Parameters of Delafloxacin (Group 1, N=12) (Pharmacokinetic Population)

Parameter (units)	Formulation A 300 mg	Formulation B 300 mg
$AUC_{0 to \infty}$ (µg·h/mL)	26.1 (11%)[a]	26.5 (16%)[a]
$AUC_{0 to t}$ (µg·h/mL)	25.1 (12%)	26.1 (15%)
$AUC_{0 to \tau}$[b] (µg·h/mL)	24.2 (12%)	25.0 (15%)
C_{max} (µg/mL)	10.5 (15%)	10.6 (14%)
$t_{1/2}$ (h)	6.9 (36%)[a]	8.6 (34%)[a]

CV = coefficient of variation.

[a] N=9 for Formulation A and N=11 for Formulation B.

[b] τ = 12 hours.

Please refer to Table 3 for the statistical analysis of relative bioavailability.

Table 3. Analysis of Relative Bioavailability of Delafloxacin (Group 1) (Pharmacokinetic Population)

Parameter (units)	Ratio of Geometric Least Squares Means [B/A]	90% Confidence Interval of the Ratio [B/A]
$AUC_{0\ to\ \tau}$ (µg·h/mL)	1.0316	(0.9833, 1.0823)
$AUC_{0\ to\ t}$ (µg·h/mL)	1.0369	(0.9903, 1.0856)
$AUC_{0\ to\ \infty}$ (µg·h/mL)	1.0356	(0.9730, 1.1023)
C_{max} (µg/mL)	1.0090	(0.9476, 1.0743)

Note: Ratio of geometric least squares means and 90% confidence interval for the ratio of geometric least squares means were determined from a linear mixed effect model for natural log-transformed pharmacokinetic parameters with treatment, sequence, and period as fixed effects, and subject (sequence) as a random effect.
Treatment: A = delafloxacin Formulation A 300 mg (reference), B = delafloxacin Formulation B 300 mg (test).

Reviewer's Comment: Based on results in Table 3, it seems that Formulation A is bioequivalent to Formulation B based on both the ratio of geometric LS mean of C_{max} and AUC.

Please refer to Figure 2 and Figure 3 for the mean plots (linear scale) of plasma concentration versus time for each Group 2 treatment on Day 1 and Day 14, respectively.

Figure 2. Mean (±SD) Plasma Concentrations of Delafloxacin Over Time by Treatment, Day 1 (Group 2 Pharmacokinetic Population, Linear Scale)

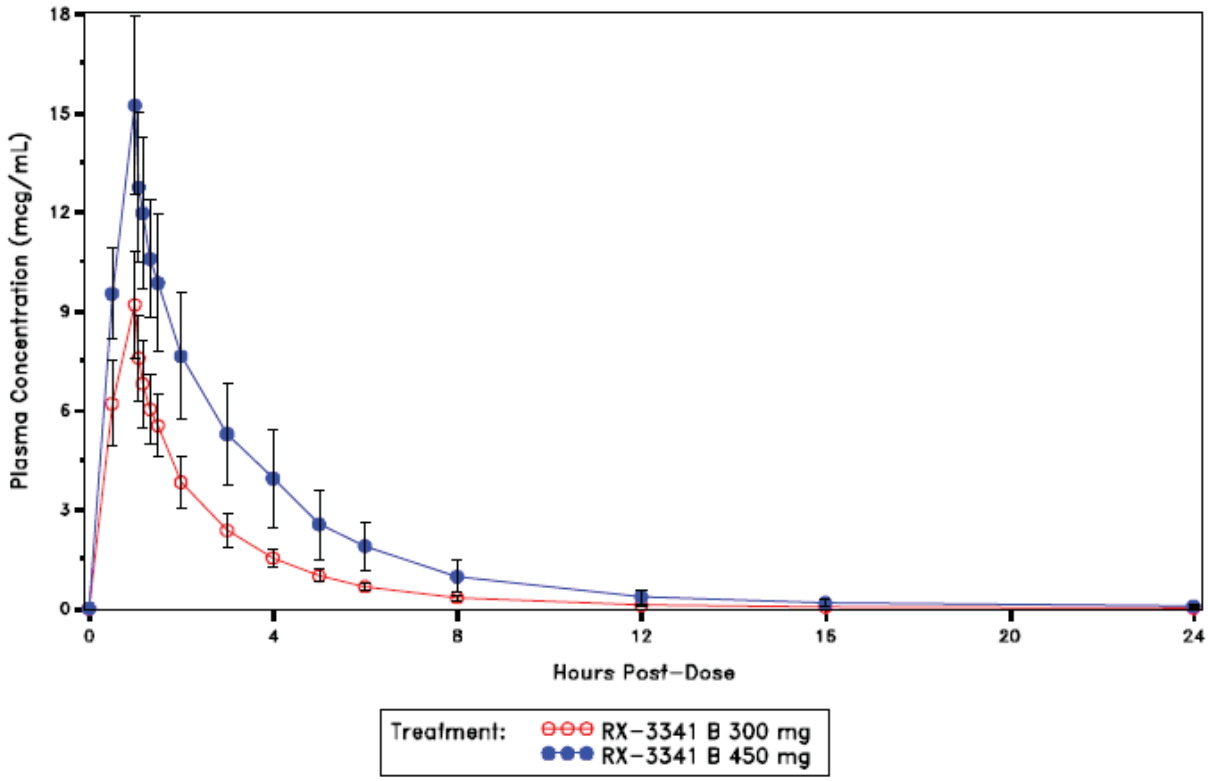
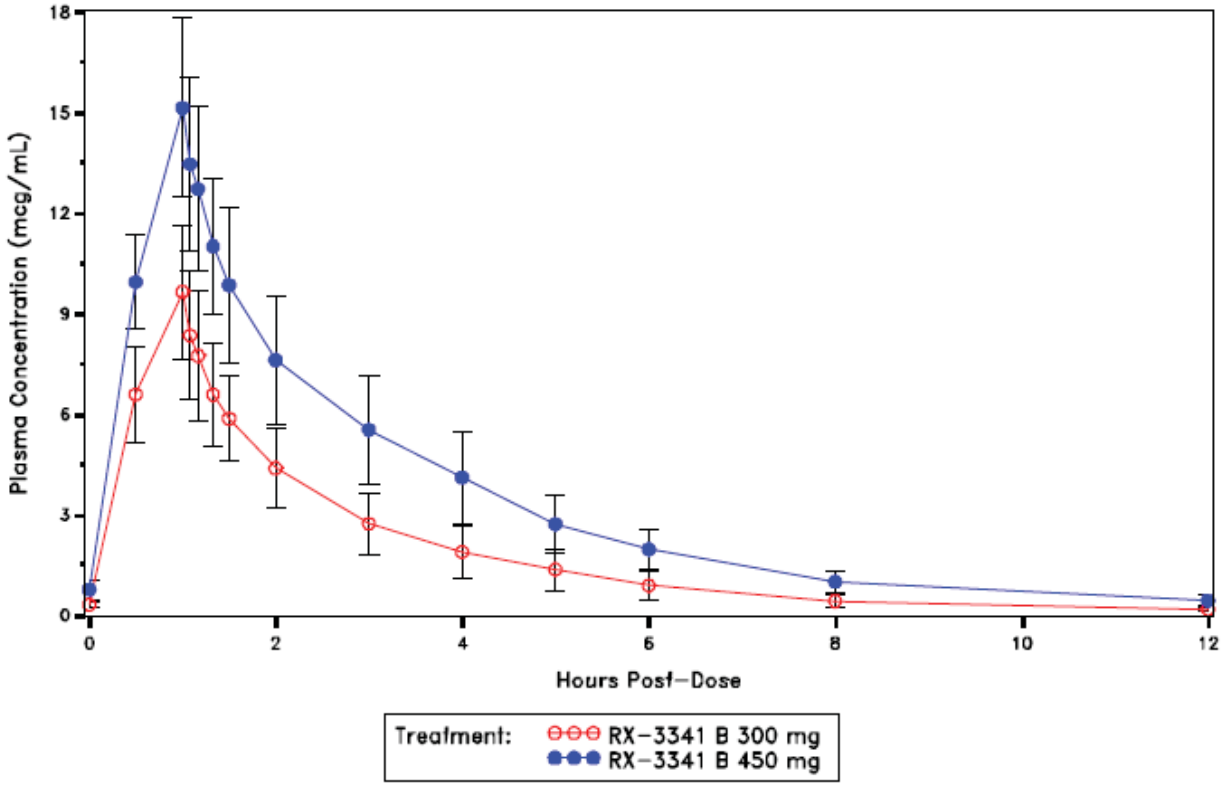


Figure 3. Mean (±SD) Plasma Concentrations of Delafloxacin Over Time by Treatment, Day 14 (Group 2 Pharmacokinetic Population, Linear Scale)



Please refer to Table 4 for the plasma PK parameters of delafloxacin in Group 2.

Table 4. Mean (CV) Plasma Pharmacokinetic Parameters of Delafloxacin (Group 2) (Pharmacokinetic Population)

	Formulation B Dosing Group							
	Day 1				Day 14			
Parameter (units)	n	300 mg BID 14 Days[a]	n	450 mg BID 14 Days[a]	n	300 mg BID 14 Days[a]	n	450 mg BID 14 Days[a]
$AUC_{0 to \tau}$[b] (µg·h/mL)	8	20.1 (17%)	8	40.6 (26%)	6	23.2 (29%)	8	42.3 (23%)
$AUC_{0 to \infty}$ (µg·h/mL)	7	20.5 (18%)	7	43.3 (29%)	-	-	-	-
C_{max} (µg/mL)	8	9.2 (18%)	8	15.2 (18%)	6	9.7 (21%)	8	15.2 (18%)
$t_{1/2}$ (h)	7	5.1 (20%)	7	5.3 (24%)	-	-	-	-
CL (L/h)	7	15.1 (20%)	7	11.4 (36%)	6	13.9 (28%)	8	11.3 (28%)
V_{ss} (L)	7	39.2 (25%)	7	35.6 (20%)	-	-	-	-
R_{ac}[c]	-	-	-	-	6	1.14 (21%)	8	1.05 (9%)

BID = twice a day, CV = coefficient of variation.

[a] Dosing regimen was single dose on Day 1 and BID dosing on Days 2 through 14. [b] τ = 12 hours.

[c] $R_{ac} = AUC_{0 to \tau \text{ Day } 14}/AUC_{0 to \tau \text{ Day } 1}$.

Reviewer's Comment: By visual inspection, the increase of delafloxacin AUC on Day 1 and Day 14 appears to be approximately dose proportional following IV administration of delafloxacin formulation B at 300 mg and 450 mg BID for 14 days. No accumulation was observed after either 300 mg BID or 450 mg BID for 14 days.

Please refer to Table 5 for the statistical analysis results of dose proportionality in Group 2.

Table 5. Analysis of Dose Proportionality of Delafloxacin (Group 2) (Pharmacokinetic Population)

Study Day	Parameter (units)	Estimated Slope for ln(Dose)	90% Confidence Interval for the Slope	*P* value (slope = 1)
1	$AUC_{0 to \tau}$ (µg·h/mL)	1.687	(1.161, 2.214)	0.037
	$AUC_{0 to t}$ (µg·h/mL)	1.728	(1.185, 2.270)	0.033
	$AUC_{0 to \infty}$ (µg·h/mL)	1.772	(1.140, 2.404)	0.050
	C_{max} (µg/mL)	1.246	(0.833, 1.660)	0.312
14	$AUC_{0 to \tau}$ (µg·h/mL)	1.507	(0.869, 2.145)	0.182
	C_{max} (µg/mL)	1.123	(0.662, 1.583)	0.644

Note: The power model, ln(parameter) = a + b × ln(dose) + error, was used to estimate the slope, corresponding 90% confidence interval, and the *P* value testing dose proportionality (b=1).

Please refer to Table 6 for the mean urinary PK parameters of delafloxacin.

Table 6. Mean (CV) Urinary Pharmacokinetic Parameters of Delafloxacin (Group 2) (Pharmacokinetic Population)

	Formulation B Dosing Group							
	Day 1				Day 14			
Parameter (units)	n	300 mg BID 14 Days[a]	n	450 mg BID 14 Days[a]	n	300 mg BID 14 Days[a]	n	450 mg BID 14 Days[a]
$Ae_{0 \text{ to } 12}$ (mg)	8	125.7 (15%)	8	154.1 (17%)	6	87.5 (71%)	8	140.5 (18%)
CLr (L/h)	8	6.50 (32%)	8	4.18 (42%)	6	4.31 (87%)	8	3.48 (26%)
$Fe\%_{0 \text{ to } 24}$ (%)	8	42.8 (15%)	8	35.7 (16%)	-	-	-	-
$Fe\%_{0 \text{ to } 12}$ (%)	-	-	-	-	6	29.2 (71%)	8	31.2 (18%)

BID = twice a day, CV = coefficient of variation.

[a] Dosing regimen was single dose on Day 1 and BID dosing on Days 2 through 14.

Safety:
Overall, 24 of 32 subjects (75.0%) reported 1 or more TEAEs. In Group 1, 5 of 12 subjects (41.7%) who received Formulation A and 4 of 12 subjects (33.3%) who received Formulation B reported at least 1 TEAE. In Group 2, infusion site pain was reported by 4 of 8 subjects (50.0%) who received high-dose Formulation B versus 2 of 8 subjects (25.0%) who received low-dose Formulation B. Three of the 4 subjects (75.0%) who received placebo also reported infusion site pain.

Overall in Group 2, 8 of 8 subjects (100%) who received low-dose Formulation B and 5 of 8 subjects (62.5%) who received high-dose Formulation B reported at least 1 TEAE. In the placebo groups, 1 of 2 subjects (50.0%) who received low-dose Formulation B placebo and 2 of 2 subjects (100%) who received high-dose Formulation B placebo reported at least 1 TEAE. The second most commonly reported TEAE after infusion site pain was headache (8 of 32 subjects [25.0%]). The third most commonly reported TEAE was diarrhea (5 of 32 subjects [15.6%]).

No subject had a TEAE that was considered to have a probable, very likely, or certain relationship to the study drug. All TEAEs were mild (97.0%) or moderate (3.0%) in severity, resolved by the end of the study, and there were no deaths or serious AEs. One subject discontinued because of an AE of joint sprain unrelated to study drug.

SPONSOR'S CONCLUSIONS
PHARMACOKINETIC

- Formulations A and B showed similar bioavailability based on analysis of C_{max} and $AUC_{0\text{-inf}}$.
- For Formulation B, the increase in AUC on Day 1 was greater than dose proportional as the dose increased from 300 mg to 450 mg. The increase in C_{max} on both Days 1 and 14 was dose proportional.
- There was little or no drug accumulation after 14 days of twice-daily dosing of delafloxacin as Formulation B (1.14 for the 300-mg dose and 1.05 for the 450-mg dose).
- Estimates of the mean $t_{1/2}$ of delafloxacin were between 5.1 hours and 8.6 hours.

- Under steady-state twice-daily dosing of Formulation B, approximately 30% of the dose was excreted unchanged in urine during the 12-hour dosing interval; mean renal clearance was between 3.48 L/h and 4.31 L/h.

SAFETY

- Based on analysis of AEs, clinical laboratory results, vital sign measurements, physical examination findings, and ECG results, IV infusions of 2 formulations of delafloxacin at a dose of 300 mg and Formulation B at a dose of 450 mg were safe and well tolerated by the healthy subjects in this study.

REVIEWER ASSESSMENT:

- Delafloxacin C_{max} and $AUC_{0\text{-}inf}$ were similar following a single 300 mg IV administration of delafloxacin in (b) (4) formulations.
- For (b) (4) formulation, the increase in delafloxacin AUC appears to be approximately dose proportional following 300 and 450 mg BID doses for 14 days.
- (b) (4) formulation seems well tolerated for both 300 mg and 450 mg BID up to 14 days.

APPEARS THIS WAY ON ORIGINAL

4.5.10 Study No.: RX-3341-104

A PHASE 1, SINGLE AND MULTIPLE INTRAVENOUS DOSE SAFETY, TOLERABILITY, AND PHARMACOKINETIC STUDY OF TWO FORMULATIONS OF DELAFLOXACIN IN HEALTHY SUBJECTS

Date(s): 14 April 2009 to 24 June 2009
Sponsor: Melinta Therapeutics, Inc, 300 George Street, Suite 301, New Haven, CT 06511
Clinical Site: Single site: PPD Phase I Clinic, located at 7551 Metro Center Drive, Suite 200, Austin, TX 78744
Analytical Site: (b) (4)

OBJECTIVE(S):

- The primary objectives were to determine the relative pharmacokinetics of 2 intravenous (IV) formulations of delafloxacin, as well as the safety and tolerability of multiple IV doses of a new formulation of delafloxacin (Formulation B) in healthy subjects.
- The secondary objective was to determine the pharmacokinetics of multiple IV doses of delafloxacin (Formulation B) in healthy subjects.

Formulation A is (b) (4) based formulation. Formulation B is captisol (sulfobutyl ether-β-cyclodextrin) based formulation.

Reviewer's Comment: Of note, Formulation B is not the to-be-marketed IV formulation. It contains SBECD (b) (4) Study RX-3341-108 compared the PK profile of Formulation B in this study to the to-be-marketed delafloxacin IV formulation.

METHODS

Study Design: This was a randomized, double-blind, placebo-controlled (Part 2 only) study in 24 male and female subjects who were assigned sequentially to Parts 1 and 2. In Part 1, the relative pharmacokinetics and safety of 2 formulations (A and B) of delafloxacin were evaluated in a crossover design. On Day 1, twelve subjects received a single, 1-hour continuous IV infusion of either delafloxacin Formulation A or delafloxacin Formulation B, each at a dose of 300 mg, according to the treatment assignment. Six subjects received Formulation A and 6 subjects received Formulation B. On Day 4, subjects received a second dose of the alternate formulation in a crossover fashion.

In Part 2, the pharmacokinetics and safety of multiple IV doses of delafloxacin Formulation B were evaluated. The dose studied was 300 mg. Blinded safety and PK data from Part 1 were reviewed by the sponsor and principal investigator before progression to Part 2. Dosing in Part 2 was permitted to begin only after the review of blinded safety data from Part 1 suggested that it was safe to do so.

On Day 1, twelve subjects received a single, 1-hour, continuous IV infusion of delafloxacin Formulation B 300 mg, or matching delafloxacin Formulation B placebo, according to treatment assignment in a 2:1 ratio. On Days 2 through 14, subjects received 2 daily IV infusions. On Day

2, the first infusion started 24 hours after the start of the infusion on Day 1, and the second infusion started 12 hours later. Thereafter, infusions occurred every 12 hours until the afternoon infusion on Day 14.

Drug Product:
The components of delafloxacin Formulation A 300 mg IV were:
• RX-3341 meglumine salt
(b) (4)
• (b) (4)
• Meglumine
• (b) (4)

The components of the delafloxacin Formulation B 300 mg IV were:
• Delafloxacin meglumine salt
• Sulfobutyl ether-β-cyclodextrin (Captisol)
• Meglumine
(b) (4)

PK Sample Collection:
In Part 1, blood samples for PK assessments were collected before dosing and at 0.33 and 0.66 (during infusion), 1, 1.083, 1.167, 1.33, 1.5, 2, 3, 4, 5, 6, 8, 12, 16, 20, 24, 30, 36, and 48 hours after the start of the first infusion on Days 1 and 4. Urine was collected before dosing from –2 to 0 hours, and at 0 to 12, 12 to 24, 24 to 36, and 36 to 48 hours after dosing on Days 1 and 4.

In Part 2, blood samples for PK assessment were collected at the following time (all sampling times were relative to the start of the first infusion):

Day 1: Before dosing and at 0.5 (during infusion), 1, 1.083, 1.167, 1.33, 1.5, 2, 3, 4, 5, 6, 8, 12, 16, and 24 hours after dosing.
Day 3, 7, and 10: Before the morning and evening dosing.
Day 14: Before morning dosing, and at 0.5 (during infusion), 1, 1.083, 1.167, 1.33, 1.5, 2, 3, 4, 5, 6, 8, and 12 hours after dosing.

Urine samples:
Day 1: Before dosing from –2 to 0 hours, and at 0 to 12 and 12 to 24 hours after the start of the first infusion on Day 1.
Day 14: Before dosing from –2 to 0 hours and at 0 to 12 hours after the start of the first infusion on Day 14.

Analytical Methods:
- The LC-API/MS/MS method used to measure the delafloxacin concentration in human plasma was validated in the validation report (b) (4) 3341C.
- The LC-API/MS/MS method used to measure the delafloxacin concentration in human urine was validated in the validation report (b) (4) 3341D.
-

	Delafloxacin in Plasma	Delafloxacin in Urine
Method Validation #	(b) (4) 3341C	(b) (4) 3341D
Range	5.00 – 5000 ng/mL	50.0 – 10000 ng/mL
LLOQ	5.0 ng/mL	50.0 ng/mL
Linearity	linear	linear
Accuracy	85-115%	85-115%
Precision	⩽ 15%	⩽ 15%
Stability	Long-term storage stability has been established for RX-3341 in human plasma for 374 days at -20°C (±10°C) and -70°C (±10°C). A total of 55 days (time from first sample drawn to the last sample analyzed) of long-term stability is required to cover the storage of the human plasma study samples at -70°C (±10°C). Stable through three freeze-thaw cycles.	Long-term storage stability has been established for RX-3341 in human urine for 168 days at -20°C (±10°C) and -70°C (±10°C). A total of 55 days (time from first sample drawn to the last sample analyzed) of long-term stability is required to cover the storage of the human urine study samples at -70°C (±10°C). Stable through three freeze/thaw cycles.

Pharmacokinetic Assessment:
Plasma parameters included the following:

- AUC_{0-t}: Area under the plasma concentration versus time curve from time 0 to the time of the last quantifiable concentration
- AUC_{0-inf}: Area under the plasma concentration versus time curve from time 0 extrapolated to infinity
- AUC_{0-tau}: area under the concentration versus time curve from time 0 (beginning of infusion) to 12 hours from initiation of infusion, calculated using the linear trapezoidal rule for the first dose.
- C_{max}: Maximum observed plasma concentration
- K_{el}: Terminal elimination rate constant
- $t_{1/2}$: Terminal elimination half-life
- Tmax: Time to achieve maximum observed plasma concentration
- MRT: mean residence time
- Vss: volume of distribution under steady-state conditions
- Rac: accumulation ratio calculated as AUC_{0-tau} Day14/AUC_{0-tau} Day1

Urine:

- Ae_{0-4}, Ae_{4-8}, Ae_{8-12}, Ae_{12-24}, Ae_{24-36}, Ae_{36-48}, Ae_{0-48}: total amount excreted unchanged in urine from 0-4, 4-8, 8-12, 12-24, 24-36, 36-48, or 0-48 hours
- $Fe\%_{0-48}$: fraction of the dose excreted unchanged in urine from 0-48 hours after dosing calculated as (Ae_{0-48}/Dose) x 100
- Ae_{0-12}: total amount excreted unchanged in urine from 0 to 12 hours after dosing

- Fe%$_{0\text{-}12}$: fraction of the dose excreted unchanged in urine from 0-12 hours after dosing calculated as ($Ae_{0\text{-}12}$/Dose) x 100
- CLr: renal clearance calculated as $Ae_{0\text{-}48}/AUC_{0\text{-}t}$

Statistical Methods:
Pharmacokinetics:
Concentrations of RX-3341 in plasma and urine samples were listed, and summary statistics were presented by time point. All the individual PK parameters were listed, and summary statistics were presented in tables.

For Part 1, log-transformed PK parameters, $AUC_{0\text{-}tau}$, $AUC_{0\text{-}t}$, $AUC_{0\text{-}inf}$, and the C_{max}, were analyzed by a mixed-effects model using treatment, sequence, and period as fixed effects and subject within sequence as random effect. The 90% confidence interval of the ratio of the means of Treatments B and A was provided.

Safety: No formal statistical analysis of safety parameters was performed.

Safety Assessment:
- Clinical laboratory evaluations (hematology [including coagulation assays], serum chemistry, and urinalysis)
- Vital sign measurements (oral temperature, systolic and diastolic blood pressure, heart rate, and respiratory rate)
- 12-lead electrocardiograms (ECGs)
- Adverse events (AEs)
- Physical examination findings

RESULTS

Study Population:
Please refer to Table 1 for the summary of subject demographics and baseline characteristics.

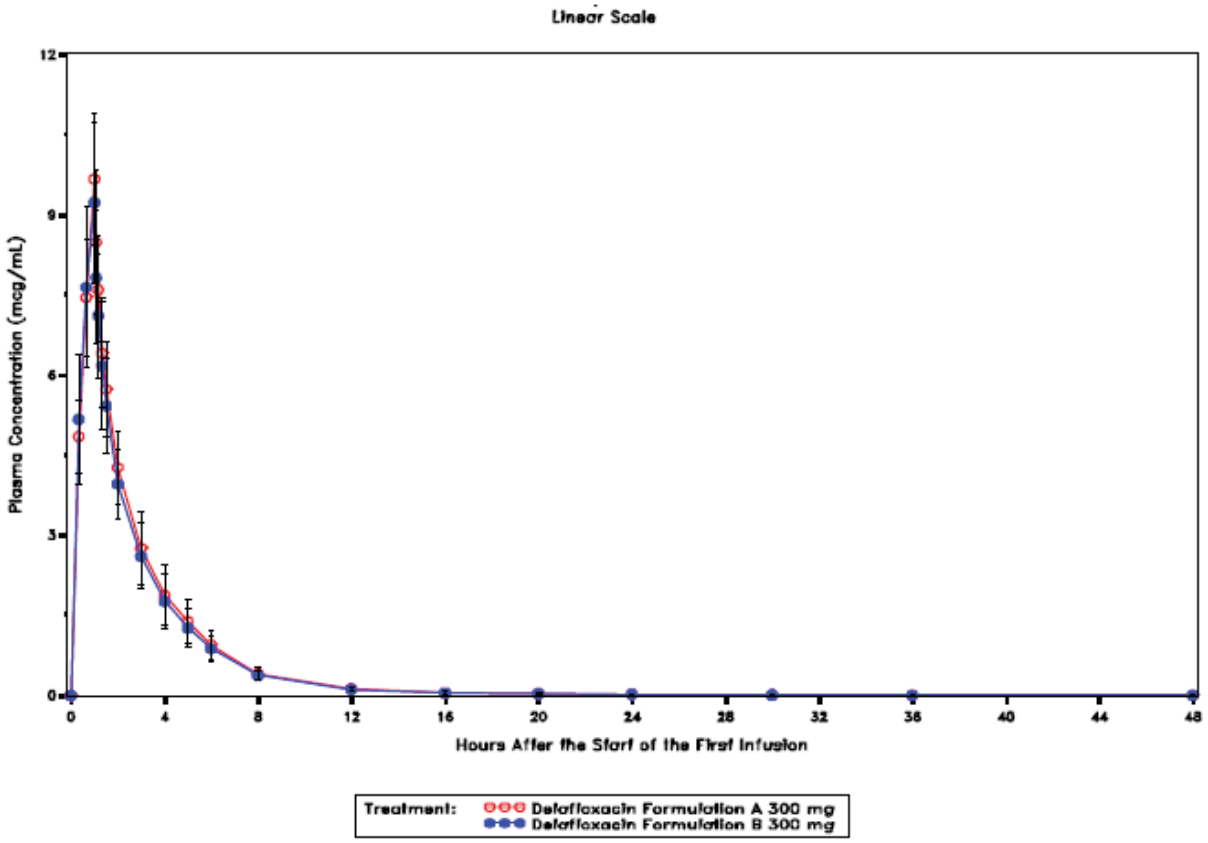
Table 1. Summary of Subject Demographics and Baseline Characteristics (Safety Population)
(Adapted from Table 6 in the study report)

	Part 1: Treatment Sequence		Part 2: Treatment		
Number of Subjects	Formulation A[a]/ Formulation B[a] (n=6)	Formulation B[a]/ Formulation A[a] (n=6)	Formulation B[a] (n=8)	Placebo (n=4)	Overall (N=24)
Age (years)					
Mean	29.5	27.2	37.8	30.3	31.8
SD	10.65	9.52	10.39	7.63	10.22
Minimum, maximum	20, 47	20, 46	18, 50	23, 41	18, 50
Sex, n (%)					
Male	4 (66.7)	3 (50.0)	5 (62.5)	2 (50.0)	14 (58.3)
Female	2 (33.3)	3 (50.0)	3 (37.5)	2 (50.0)	10 (41.7)
Race, n (%)					
White	6 (100.0)	6 (100.0)	4 (50.0)	4 (100.0)	20 (83.3)
Black	0	0	4 (50.0)	0	4 (16.7)
Ethnicity, n (%)					
Hispanic or Latino	4 (66.7)	1 (16.7)	1 (12.5)	2 (50.0)	8 (33.3)
Not Hispanic or Latino	2 (33.3)	5 (83.3)	7 (87.5)	2 (50.0)	16 (66.7)
Height (cm)					
Mean	172.5	169.5	168.56	167.38	169.58
SD	2.449	10.927	12.373	6.787	9.127
Minimum, maximum	168.0, 174.5	157.0, 188.0	150.5, 186.0	161.5, 176.5	150.5, 188.0
Weight (kg)					
Mean	79.45	74.18	71.74	72.23	74.36
SD	6.705	11.457	10.169	8.987	9.501
Minimum, maximum	67.4, 85.8	60.7, 88.2	52.2, 86.2	60.4, 82.0	52.2, 88.2
Body mass index (kg/m^2)					
Mean	26.72	25.92	25.26	25.83	25.88
SD	2.209	4.022	2.823	3.117	2.930
Minimum, maximum	22.3, 28.3	21.2, 31.7	22.1, 29.6	21.3, 28.2	21.2, 31.7

Note: Percentages were based on the number of subjects in the safety population within each treatment sequence (Part 1) or Treatment (Part 2), and overall.
Categorical data were presented as n (%).
[a] 300 mg of delafloxacin.

Pharmacokinetics:
Part 1:
Mean plots of plasma concentration versus time by formulations are presented in Figure 1.

Figure 1. Mean (SD) Plasma Concentrations of RX-3341 Over Time by Formulations (Pharmacokinetic Population) *(Adapted from Figure 2 in study report)*



The arithmetic means and CVs for selected PK parameters for Formulation A and B are presented in Table 2.

Table 2. Mean (CV) Plasma Pharmacokinetic Parameters of Delafloxacin (Pharmacokinetic Population) *(Adapted from Table 7 in the study report)*

Parameter (unit)	Formulation A 300 mg (n=12)	Formulation B 300 mg (n=12)
$AUC_{0\text{-tau}}$ (µg•h/mL)[a]	22.91 (18)	21.86 (18)
$AUC_{0\text{-t}}$ (µg•h/mL)	23.77 (18)	22.67 (18)
$AUC_{0\text{-inf}}$ (µg•h/mL)	23.60 (21)[b]	22.87 (17)[b]
C_{max} (µg/mL)	9.67 (13)	9.23 (16)
$t_{1/2}$ (h)	11.12 (49)[b]	9.17 (35)[b]

CV = coefficient of variation.
[a] tau = 12 hours.
[b] n = 9.

Please refer to Table 3 for the statistical analysis of the plasma pharmacokinetics parameters of delafloxacin.

Table 3. Statistical Analysis of Plasma Pharmacokinetic Parameters (Pharmacokinetic Population) *(Adapted from Table 8 in the study report)*

Parameter (unit)	Treatment Comparison	n		Ratio of Geometric Least Squares Means (B/A)	90% Confidence Interval of the Ratio
		A	B		
$AUC_{0\text{-}tau}$ (µg•h/mL)	B-A	12	12	0.954	(0.906, 1.004)
$AUC_{0\text{-}t}$ (µg•h/mL)	B-A	12	12	0.954	(0.905, 1.006)
$AUC_{0\text{-}inf}$ (µg•h/mL)	B-A	9	9	0.880	(0.856, 0.904)
C_{max} (µg/mL)	B-A	12	12	0.950	(0.891, 1.013)

Note: Ratio of geometric least squares means and 90% confidence interval for the ratio of geometric least squares means were estimated from a mixed-effects model with log-transformed pharmacokinetic parameter as the dependent variable, treatment, sequence, and period as fixed effects, and subject within sequence as a random effect.
Treatment: A = delafloxacin Formulation A 300 mg, B = delafloxacin Formulation B 300 mg.

Reviewer's Comment: Based on results in Table 3, it seems that the PK profile of delafloxacin for Formulation A [redacted] *(b) (4)* *) was comparable to that for Formulation B (captisol).*

Please refer to Table 4 for the urinary pharmacokinetics.

Table 4. Mean (CV) Urinary Pharmacokinetic Parameters of Delafloxacin (Pharmacokinetic Population) *(Adapted from Table 9 in the study report)*

Parameter (unit)	Formulation A 300 mg (n=12)	Formulation B 300 mg (n=12)
$Ae_{0\text{-}12}$ (mg)	120.96 (21)	131.11 (24)
$Ae_{0\text{-}48}$ (mg)	126.24 (19)	135.98 (23)
$Fe\%_{0\text{-}48}$ (%)	42.08 (19)	45.33 (23)
CL_r (L/h)	5.53 (29)	6.24 (32)

CV = coefficient of variation.

Part 2:
Mean (±SD) plasma concentrations of delafloxacin versus time for Day 1 and 14 are presented on a linear scale in Figure 2 and Figure 3, respectively.

Figure 2. Mean (±SD) Plasma Concentrations of Delafloxacin (Formulation B 300 mg) Versus Time, Day 1 (Pharmacokinetic Population, Linear Scale) *(Adapted from Figure 3 in the study report)*

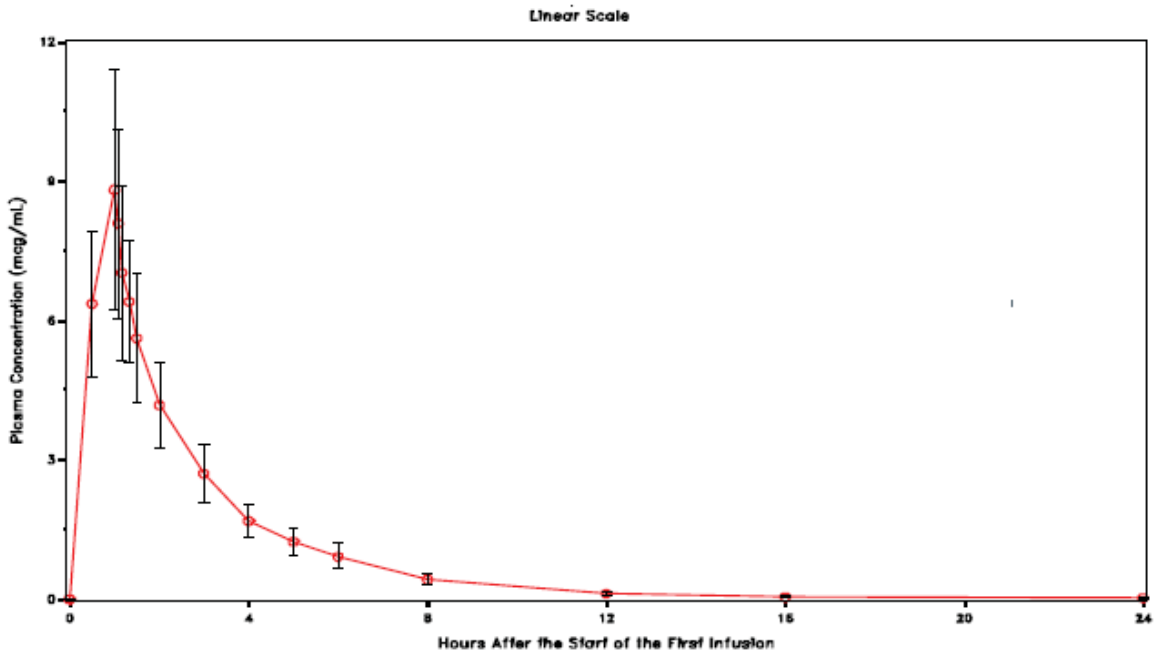
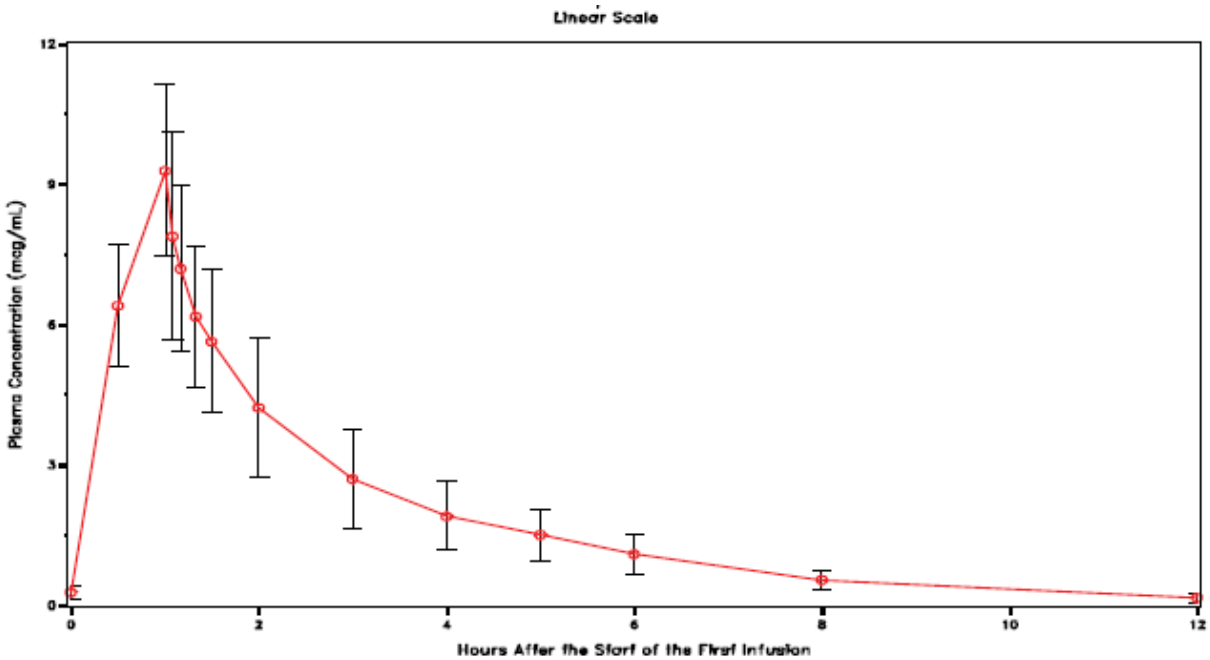


Figure 3. Mean (±SD) Plasma Concentrations of Delafloxacin (Formulation B 300 mg) Versus Time, Day 14 (Pharmacokinetic Population, Linear Scale) *(Adapted from Figure 4 in the study report)*



Please refer to Table 5 for the arithmetic means and CVs for selected PK parameters for Day 1 and Day 14.

Table 5. Mean (CV) Plasma Pharmacokinetic Parameters of Delafloxacin on Days 1 and 14 (Pharmacokinetic Population) *(Adapted from Table 10 in the study report)*

Parameter (unit)	Delafloxacin Formulation B 300 mg	
	Day 1 (n=8)	Day 14 (n=7)
$AUC_{0\text{-}tau}$ (µg•h/mL)[a]	21.80 (21)	23.36 (30)
$AUC_{0\text{-}t}$ (µg•h/mL)	22.47 (20)	ND
$AUC_{0\text{-}inf}$ (µg•h/mL)	22.05 (20)[b]	ND
C_{max} (µg/mL)	8.94 (28)	9.29 (20)
CL (L/h)	14.08 (20)[b]	13.80 (29)
$t_{1/2}$ (h)	5.61 (30)[b]	ND
MRT (h)	2.94 (13)[b]	ND
V_{ss} (L)	41.77 (26)[b]	ND
R_{ac}	ND	1.09 (11)

CV = coefficient of variation; ND = not determined.
[a] tau = 12 hours.
[b] n = 7.

Reviewer's comment: Based on the results shown in Figure 3 and 4 and Table 5, 1) there was no accumulation of delafloxacin after BID dose for 14 days; 2) no change in CL from Day 1 to Day 14.

Please refer to Table 6 for the urinary pharmacokinetic parameters of delafloxacin on Day 1 and Day 14.

Table 6. Mean (CV) Urinary Pharmacokinetic Parameters of Delafloxacin on Days 1 and 14 (Pharmacokinetic Population) *(Adapted from Table 11 in the study report)*

Parameter (unit)	Delafloxacin Formulation B 300 mg	
	Day 1 (n=8)	Day 14 (n=7)
$Ae_{0\text{-}12}$ (mg)	123.77 (11)	145.10 (13)
$Ae_{12\text{-}24}$ (mg)	3.27 (21)	ND
$Ae_{0\text{-}24}$ (mg)	127.04 (11)	ND
$Fe\%_{0\text{-}12}$ (%)	ND	48.37 (13)
$Fe\%_{0\text{-}24}$ (%)	42.35 (11)	ND
CL_r (L/h)	5.89 (26)	6.69 (33)

CV = coefficient of variation; ND = not determined.

Reviewer's comment: It appears that the mean renal clearance of delafloxacin after single and multiple doses for formulation B on Days 1 and 14 in Part 2 were similar to the mean renal clearance after single-dose administration for formulation A and B in Part 1.

Safety Summary

Overall, 10 of 24 subjects (41.7%) reported 1 or more TEAEs. In Part 1, TEAEs were not reported in any subjects during the Formulation A treatment period, and only 1 of 12 subjects reported TEAEs during the Formulation B treatment period. In Part 2, six of 8 subjects (75.0%) who received Formulation B and 3 of 4 subjects (75.0%) who received placebo reported TEAEs.

Overall, the TEAEs most commonly reported were gastrointestinal disorders (9 of 24 subjects [37.5%]), nervous system disorders (8 of 24 subjects [33.3%]), and general disorders and administration site conditions (6 of 24 subjects [25.0%]). In Part 2, the percentage of subjects who reported TEAEs in these system organ classes were similar regardless of whether they received active study drug or placebo. Similarly, the percentage of subjects who reported TEAEs that were considered possibly related to study drug were similar regardless of whether they received active study drug or placebo. No subject had a TEAE that was probably related, very likely, or certainly related to study drug.

All but 1 subject reported TEAEs that were mild in severity. Only 1 subject (Subject 0014 [300-mg delafloxacin Formulation B group, Part 2]) reported TEAEs (nausea and vomiting) considered moderate in severity, and this was the only subject who discontinued because of a TEAE. There were no deaths or SAEs, and all TEAEs resolved by the end of the study.

SPONSOR'S CONCLUSIONS

PHARMACOKINETIC

- Total and peak exposure indicate that Formulations A and B were similar in bioavailability.
- No appreciable drug accumulation was observed after multiple days of twice-daily dosing of Formulation B.
- More than 40% of the drug was excreted unchanged in urine following single and twice daily multiple dosing, with most of the drug excretion occurring within the first 12 hours after dosing. The overall mean renal clearance throughout both parts of the study was approximately 6 L/h.

SAFETY

- There was no difference in the percentage of subjects who reported TEAEs following multiple IV infusions of 300 mg delafloxacin Formulation B compared with placebo.
- Single IV infusions of 300 mg delafloxacin Formulations A and B and multiple IV infusions of 300 mg delafloxacin Formulation B were safe and generally well tolerated by the healthy subjects in this study.

REVIEWER ASSESSMENT:

- It seems that the PK profiles of delafloxacin between Formulation A and B were comparable.
- No accumulation of delafloxacin was observed after multiple-dose administration (BID for 14 days) of Formulation B.

4.5.11 Study No.: RX-3341-107

An Open Label, Single IV Dose Study in Healthy Male Subjects Designed to Assess the Mass Balance Recovery, Metabolite Profile and Metabolite Identification of [14C]-Delafloxacin (RX-3341)

Date(s): 21 Jul 2011 to 25 Aug 2011
Sponsor: Melinta Therapeutics, Inc, 300 George Street, Suite 301, New Haven, CT 06511
Clinical Site: Single site: Quotient Clinical, Mere Way, Ruddington, Nottingham, UK
Analytical Site: (b) (4)

OBJECTIVE(S):

- The primary objectives of the study were:
  - To determine the mass balance after a single intravenous (IV) dose of carbon-14-radiolabelled RX-3341 ([^{14}C]-RX-3341)
  - To determine the routes of IV [14C]-RX-3341 metabolism and excretion
  - To provide plasma, urine and feces samples for metabolite profiling (results of which are to be reported separately)
  - To assess the IV pharmacokinetics (PK) of [14C]-RX-3341 in plasma
- The secondary objective of the study was to provide additional safety and tolerability information for IV [14C]-RX-3341.

METHODS

Study Design: This was an open-label, non-randomized, single-dose study in 6 healthy male subjects of 30 to 65 years of age. Subjects attended a screening appointment within 28 days before drug administration and were admitted to the clinical unit on the morning of the day before dosing (Day -1). All subjects received a single IV dose of investigational medicinal product (IMP) on Day 1 in the fasted state. Blood samples were taken at specified time points and urine and feces were collected over pre-defined time intervals for evaluation of the mass balance, PK and metabolism of the IMP. Subjects were discharged after 6 days (i.e., on Day 7) if a mass balance cumulative recovery of >95% had been achieved, or if a mass balance cumulative recovery of >90% had been achieved and <1% had been collected within 2 separate, consecutive 24 h periods. If insufficient radioactivity was recovered by Day 7, subjects could have been required to collect samples in clinic for up to 2 further days.

It was planned to enroll 6 subjects. Six subjects were enrolled; all 6 completed the study and were included in the safety and PK analysis populations.

Drug Product:
IV infusion containing approximately 300 mg [14C]-RX-3341 (as the meglumine salt, Captisol solution containing sulfobutyl ether-β-cyclodextrin, to-be-marketed IV formulation) in approximately 250 mL (batch number 106411/C/02), infused over approximately 60 min. The target radiation dose was 3.7 MBq (100 µCi); the maximum permitted dose was 5.3 MBq (143 µCi).

PK Sample Collection:
Please refer to Table 1 for the plasma, urine, feces sampling schedule.

Table 1. Schedule of Assessments

Study Day	-28 to -2	-1	1												
				Time after dosing (min)					Time after dosing (h)						
	Screening	Admission	Pre-dose	0	5	15	30	45	1 (end of infusion)	1.08	1.25	1.5	2	3	4	5
Informed Consent	x															
Inclusion/Exclusion criteria	x															
Medical/Surgical History	x															
Body Weight and Height	x															
Physical Examination	x															
Virology	x															
Urine Drug Screen	x	x														
Breath Alcohol and CO	x	x														
ECG	x	x	x[a]	[a]	[a]	[a]	[a]	[a]	x[a]	[a]	[a]		x			
Vital Signs	x	x	x			x	x		x				x			
Haematology, Clinical Chemistry and Coagulation[b]	x[c]	x														
Urinalysis	x	x														
IMP Administration[d]				◄					►							
Whole Blood Sampling for TR			x			x	x		x			x	x	x	x	x
Plasma Sampling for TR, Parent Drug and Metabolite Profiling and Identification			x		x	x	x	x	x	x[e]	x[e]	x	x	x	x	x
Plasma Sampling for Protein Binding									x					x		
Urine Collection[f]			◄													►
Faeces Collection[g]			◄													►
Adverse Events	◄															►

CO: carbon monoxide ECG: electrocardiogram IMP: investigational medicinal product TR: total radioactivity
[a] In addition to ECGs, continuous ECG monitoring was carried out from 5 min before the start of the RX-3341 IV infusion until 15 min after the end of the infusion.
[b] All safety bloods were taken in the fasted state.
[c] Clinical chemistry at screening included creatinine clearance.
[d] [^{14}C]-RX-3341 IV infusion was administered over 60 min.
[e] Plasma samples for the measurement of RX-3341 parent drug only.
[f] Urine was collected for determination of total radioactivity during the following periods: pre-dose (-12–0 h), and 0–4, 4–8, 8–12, 12–24, 24–48, 48–72, 72–96, 96–120 and 120–144 h post-dose.
[g] Faeces were collected for determination of total radioactivity during the following periods: pre-dose (-24–0 h), and 0–24, 24–48, 48–72, 72–96, 96–120 and 120–144 h post-dose.

Study Day	1 (continued)			2		3		4	5	6	7
	Time after dosing (h)										
	6	8	12	24	36	48	60	72	96	120	144
Physical Examination											x[h]
ECG											x[h]
Vital Signs				x							x[h]
Haematology, Clinical Chemistry and Coagulation[i]											x[h]
Urinalysis											x[h]
Whole Blood Sampling for TR	x	x	x	x	x	x	x	x	x	x	x[h]
Plasma Sampling for TR, Parent Drug and Metabolite Profiling and Identification	x	x	x	x	x	x	x	x	x	x	x[h]
Plasma Sampling for Protein Binding			x	x		x					
Urine Collection[j]	►										
Faeces Collection[k]	►										
Adverse Events	►										

CO: carbon monoxide ECG: electrocardiogram IMP: investigational medicinal product TR: total radioactivity
[h] Procedures were to be done before discharge from the clinical unit. [i] All safety bloods were taken in the fasted state.
[j] Urine was collected for determination of total radioactivity during the following periods: pre-dose (-12–0 h), and 0–4, 4–8, 8–12, 12–24, 24–48, 48–72, 72–96, 96–120 and 120–144 h post-dose.
[k] Faeces were collected for determination of total radioactivity during the following periods: pre-dose (-24–0 h), and 0–24, 24–48, 48–72, 72–96, 96–120 and 120–144 h post-dose.

Analytical Methods:
Whole blood, plasma, urine and feces samples were dispatched daily to Quotient Metabolism for analysis of total radioactivity by liquid scintillation counting (LSC). The limits of detection were 0.044 µg equiv/mL in plasma and 0.164 µg equiv/g in whole blood. A mass spectrometry method incorporating a full scan and multiple reaction monitoring (MRM) was used to confirm the presence of delafloxacin and any additional drug-related components.

Plasma RX-3341 concentrations were measured by [redacted] (b) (4) after protein precipitation sample preparation, using a validated liquid chromatography with tandem mass spectrometry (LC-MS/MS) method [redacted] (b) (4) 106411). The calibration range of the assay was 5 to 5000 ng/mL. Data were reported as RX-3341 free acid in µg/mL after being corrected for the ^{14}C component. Please see table below for the parameters of the validated method (Adapted from validation report Study number [redacted] (b) (4) 106411QB02).

	RX-3341
Validated Range	5– 5000 ng/mL
Calibration Model	Linear weighted $1/x^2$
Precision (%CV) Intra-assay	≤19.8% (LLOQ)
Precision (%CV) Inter-assay	≤15.1% (LLOQ)
Accuracy (%RE) Intra-assay	≤±17.8% (LLOQ)
Accuracy (%RE) Inter-assay	≤±12.2% (LLOQ)
Accuracy (%RE) and Precision (%CV) following 1/10 dilution	≤±1.1% RE ≤4.2% CV
Stability in Acetonitrile:water=50:50 (v/v)	1 month at +4°C
Stability in human plasma	4 hours at room temperature 35 days at -20°C
Freeze-Thaw Stability	3 cycles at -20°C
Processed Extract Stability	1 week at +4°C
Instrument Validated	API5000

Mass Balance and Pharmacokinetic Assessment:
Plasma parameters included the following:

- AUC_{0-t}: Area under the plasma concentration versus time curve from time 0 to the time of the last quantifiable concentration
- AUC_{0-inf}: Area under the plasma concentration versus time curve from time 0 extrapolated to infinity
- AUC_{0-t}: Area under the concentration vs time curve from time zero to 't' hours, where 't' = the time point of the last sample on the PK profile at which the analyte was quantifiable.
- C_{max}: Maximum observed plasma concentration
- $t_{1/2}$: Terminal elimination half-life
- T_{max}: Time to achieve maximum observed plasma concentration
- CL: clearance

Statistical Methods:
No formal statistical analysis was performed.

Safety Assessment: Safety was assessed by analysis of adverse events (AEs), laboratory parameters, vital signs, physical examinations and electrocardiograms (ECGs).

RESULTS

Study Population:
All subjects received 1 dose of Investigational medicinal product (IMP) as planned and were included in the safety analysis population. All infusions were given as a volume of 240 mL over 60 min. All subjects also had sufficient urinary and fecal concentration data to demonstrate >90% mass balance recovery and were included in the PK analysis population. Please refer to Table 2 for the demographic characteristics of the study population.

Table 2. Demographic Characteristics: Safety Population (*Adapted from Table 2 in the study report)*

		300 mg [^{14}C]-RX-3341 (N = 6)
Age (years)	Mean (SD)	51.2 (9.2)
Height (cm)	Mean (SD)	172.92 (7.38)
Weight (kg)	Mean (SD)	80.58 (14.46)
BMI (kg/m^2)	Mean (SD)	26.7 (3.2)
Race	White (%)	6 (100)
Sex	Male (%)	6 (100)

Pharmacokinetics in Plasma:
Please refer to Table 3 for the key PK parameters of total radioactivity and delafloxacin.

Table 3. Geometric Mean (Geometric CV%) Values of Key Pharmacokinetic Parameters for Total Radioactivity and RX-3341: Pharmacokinetic Population (*Adapted from Table 3 in the study report*)

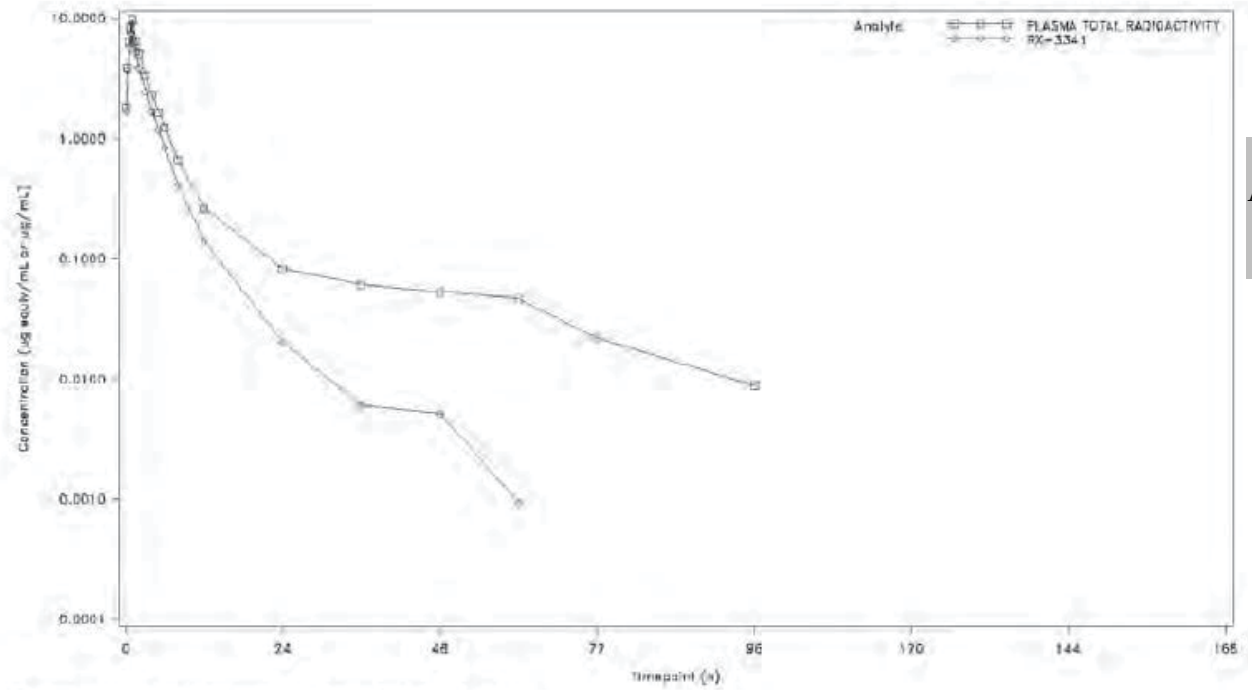
Parameter	Total Radioactivity in Plasma	Total Radioactivity in Whole Blood	RX-3341 in Plasma
C_{max} (µg/mL)[a]	9.73 (26.5%)	7.57 (27.0%)	8.76 (24.5%)
t_{max} (h)[b]	1.00 (0.75–1.00)	1.00 (1.00–1.00)	1.00 (1.00–1.00)
AUC_{0-t} (µg.h/mL)[c]	30.4 (32.8%)	20.9 (23.6%)	21.5 (28.0%)
AUC_{0-inf} (µg.h/mL)[c]	NC	21.6 (8.70%) [n = 3]	19.6 (24.2%) [n = 4]
$t_{1/2}$ (h)	NC	2.93 (8.0%) [n = 3]	3.65 (18.6%) [n = 4]
CL (mL/min)[d]	NC	245 (10.6%) [n = 3]	266 (22.4%) [n = 4]
CLr (mL/min)[d]	111 (33.9%)	154 (25.4%)	NC

NC: not calculated
[a] units are (µg equiv/mL) for total radioactivity in plasma and (µg equiv/g) for total radioactivity in whole blood
[b] median (range)
[c] units are (µg equiv.h/mL) for total radioactivity in plasma and (µg equiv.h/g) for total radioactivity in whole blood
[d] units are (g/min) for total radioactivity in whole blood

*Reviewer's Comment: The AUC_{0-t} (30.4 µg*h/mL) of total radioactivity in plasma was higher than the AUC_{0-t} (21.5 µg.h/mL) of RX-3341 in plasma. Based on AUC_{0-t} Table 3, it seems that the parent drug (RX-3341) only accounted for about 69% of the systemic exposure and metabolites or breakdown products accounted to about 31% of the systemic exposures. It was also observed that the AUC_{0-inf} of total radioactivity in whole blood was lower than the one of total radioactivity in plasma. This observation suggests that there may be some distribution of total radioactivity into the cellular component of whole blood.*

Please refer to Figure 1 for the mean plasma RX-3341 and total radioactivity concentrations.

Figure1. Mean Plasma RX-3341 and Total Radioactivity Concentrations (Semi-Log Scale): Pharmacokinetic Population *(Adapted from Figure 8 in the study report)*



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Reviewer's Comment: According to Figure 1, it seems that there was a slower decline in the total plasma radioactivity in the apparent terminal elimination phase compared to RX-3341.

The plasma concentration vs time profiles of RX-3341 were consistent with a 1 h IV infusion of [14C]-RX-3341. After the end of the infusion, concentrations of RX-3341 were lower than those for total radioactivity in all plasma samples where both analytes could be quantified. Based on geometric mean plasma concentrations, the percentage of plasma total radioactivity accounted for by plasma RX-3341 decreased from 92% at the end of the infusion to 52% at 12 h after the start of the infusion. Comparison of AUC_{0-t} estimates for total radioactivity and RX-3341 suggested that RX-3341 accounted for between 66% and 80% of systemic exposure to total radioactivity.

At time points where radioactivity was detectable in both whole blood and plasma, total radioactivity concentrations were lower in whole blood than in plasma. At the end of the infusion the mean total radioactivity in whole blood was 21% lower than the geometric mean value for plasma total radioactivity and this difference remained consistent up to 8 h after the start of the infusion (geometric mean radioactivity in whole blood was 23% lower than the geometric mean value for plasma total radioactivity at the 8 h time point). Comparisons of AUC_{0-t} estimates for plasma and whole blood were confounded by longer times to last quantifiable concentrations (t) for plasma. The geometric mean estimate AUC_{0-t} for total radioactivity in whole blood was 31% lower than that for plasma total radioactivity.

The Sponsor stated that measurement of total radioactivity in plasma and whole blood following the administration of [14C]-RX-3341 did not discriminate between radiolabelled parent drug and radiolabelled metabolites and breakdown products. Accordingly, the PK findings for total radioactivity should be interpreted with some caution, in light of possible heterogeneity of the components contributing to these data.

An average of 93% of administered radioactivity was recovered during the study: 65% from the urine and 28% from the feces. Approximately half the radioactivity administered as [14C]-RX-3341 was recovered from urine and feces within 24 h of the start of the infusion. The extent of plasma protein binding of [14C]-RX-3341-related material in plasma ranged from 71.07% to 84.62% over the measurable time points (1, 3 and 12 h), indicating that [14C]-RX-3341 was moderately bound to plasma proteins. Please refer to Table 4 below for details.

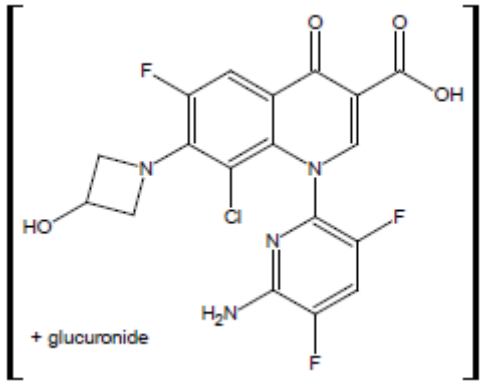
Table 4. Degree of Protein Binding (%) *(Adapted from Table 14.2.10.1)*

Dose of [14C]-RX-3341	Time point	Mean	Median	SD	Min	Max	n
300 mg (N=6)	1 H	73.172	72.885	1.510	71.26	75.79	6
	3 H	72.382	71.590	1.639	71.07	74.94	6
	12 H	78.580	78.600	4.177	73.81	84.62	5

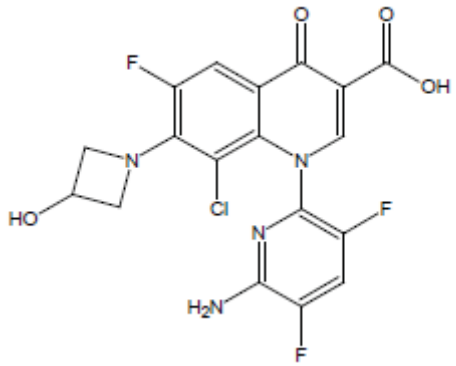
Metabolite Identification (Report Study RX-3341-107-03)
Metabolite identification using full scan and product ion analyses was carried out on the preliminary samples to screen for the presence of 'typical' or 'predicted' metabolites. Samples were screened for components corresponding to Delafloxacin and potential hydroxylated and/or conjugated metabolites. Full scan techniques were used to attempt to identify any additional unassigned components by direct comparison with the [14C]-radiochromatogram.

Components corresponding to greater than 5% sample radioactivity in the plasma extracts and urine samples and greater than 10% sample radioactivity in the feces extracts are discussed below.

U1 / P1 - Glucuronide of Delafloxacin – retention time 24.2 minutes (M3)



U2 / P2 / F1 - Delafloxacin – retention time 54.9 minutes



Please refer to Table 5, 6, 7, respectively, for the summary of the components identified in human plasma, urine, and feces, respectively.

Table 5 Summary of multiple reaction monitoring (MRM) results from analysis of pooled human plasma samples (1-12 hours) *(Adapted from Table 4 in the study report)*

MET No	Component	Molecular Weight (Da)	Retention Time (min)[a]	Subject					
				001	002	003	004	005	006
P1	Glucuronide of Delafloxacin	616	24.5	√	√	√	√	√	√
N/A	Tentative glucuronide of unassigned metabolite of Delafloxacin	588	25.3	√	√	√	√	√	√
N/A	Glucuronide of Delafloxacin	616	25.4	√	√	√	√	√	√
N/A	Glucuronide of Delafloxacin	616	34.5	√	√	√	√	√	√
N/A	Di-hydroxy Delafloxacin	472	35.6	√	√	√	√	√	√
N/A	Acetic acid conjugate of Delafloxacin	498	Component Not Present in Sample						
N/A	M+18 amu metabolite of Delafloxacin	458	41.9	√	√	√	√	√	√
N/A	Methyl ester of Delafloxacin	454	46.3	√	√	√	√	√	√
P2	Delafloxacin	440	54.6	√	√	√	√	√	√

√ Component detected by LC-MS/MS
a Retention times taken from subject 001 1-12 hour sample
N/A No MET No. assigned – component accounting for less than 5% sample radioactivity

Table 6 Summary of MRM results from analysis of human urine samples (0-4 and 4-24 hours) *(Adapted from Table 5 in the study report)*

MET No	Component	Molecular Weight (Da)	Retention Time (min)[a]	Subject / Timepoint											
				001 0-4h	001 4-24h	002 0-4h	002 4-24h	003 0-4h	003 4-24h	004 0-4h	004 4-24h	005 0-4h	005 4-24h	006 0-4h	006 4-24h
U1	Glucuronide of Delafloxacin	616	24.3	√	√	√	√	√	√	√	√	√	√	√	√
N/A	Tentative glucuronide of unassigned metabolite of Delafloxacin	588	25.1	√	√	√	√	√	√	√	√	√	√	√	√
N/A	Glucuronide of Delafloxacin	616	25.3	√	√	√	√	√	√	√	√	√	√	√	√
N/A	Acetic acid conjugate of Delafloxacin	498	31.5	√[t]	√[t]	X	X	X	√[t]	X	√[t]	√	√[t]	√	√
N/A	Glucuronide of Delafloxacin	616	34.2	√	√	√	√	√	√	√	√	√	√	√	√
N/A	Di-hydroxy Delafloxacin	472	35.5	√	√	√	√	√	√	√	√	√	√	√	√
N/A	M+ 18 amu metabolite of Delafloxacin	458	41.9	√	√	√	√	√	√	√	√	√	√	√	√
N/A	Methyl ester of Delafloxacin	454	46.5	X	X	X	X	X	X	X	X	√[t]	X	√[t]	X
U2	Delafloxacin	440	54.7	√	√	√	√	√	√	√	√	√	√	√	√

√ Component present by LC-MS/MS
√[t] Component tentatively present by LC-MS/MS (2 out of 3 (or 4) confirmatory MRM transitions present)
X Component not detected by LC-MS/MS or below limit of detection
a Retention times taken from subject 006 0-4 hour sample
N/A No MET No. assigned – component accounting for less than 5% sample radioactivity

Table 7 Summary of MRM results from analysis of human feces samples *(Adapted from Table 6 in the study report)*

MET No	Component	Molecular Weight (Da)	Retention Time (min)[a]	Subject / Timepoint											
				001 24-48h	001 48-96h	002 24-48h	002 72-96h	003 24-48h	003 48-96h	004 0-48h	004 48-72h	005 24-48h	005 48-96h	006 24-72h	006 72-144h
N/A	Glucuronide of Delafloxacin	616	21.1[b]	√	√	X	X	X	√	√[t]	√[t]	X	X	X	X
N/A	Glucuronide of Delafloxacin	616	21.4[b]	√	√	X	X	X	√	√[t]	√[t]	X	X	X	X
N/A	Glucuronide of Delafloxacin	616	23.5[c]	√	√	X	X	X	√	X	√	X	X	X	X
N/A	Glucuronide of Delafloxacin	616	24.0[c]	√	√	X	X	X	√	X	√[t]	X	X	X	X
N/A	Glucuronide of Delafloxacin	616	24.4[c]	√	√	X	√	X	√	X	√	X	X	X	√[t]
N/A	Tentative glucuronide of unassigned metabolite of Delafloxacin	588	Component Not Present in Any Sample												
N/A	Glucuronide of Delafloxacin	616	25.3	√	√	X	X	X	√	√	√	X	X	X	X
N/A	Acetic acid conjugate of Delafloxacin	498	31.5	X	√[t]	√[t]	X	X	X	√[t]	√[t]	X	√[t]	X	X
N/A	Glucuronide of Delafloxacin	616	34.2	√	√	X	X	√	√	√	X	X	X	X	X
N/A	Di-hydroxy Delafloxacin	472	35.3	√	√	√	√	√	√	√	√	√	√	√	√
N/A	M+ 18 amu metabolite of Delafloxacin	458	41.9	√	√	√	√	√	√	√	√	√	√	√	√
N/A	Methyl ester of Delafloxacin	454	46.5	X	√	√[t]	X	√[t]	√[t]	√[t]	X	X	√[t]	X	X
F1	Delafloxacin	440	54.7	√	√	√	√	√	√	√	√	√	√	√	√

√ Component present by LC-MS/MS
√[t] Component tentatively present by LC-MS/MS (2 out of 3 (or 4) confirmatory MRM transitions present)
X Component not detected by LC-MS/MS or below limit of detection
a Retention times taken from subject 001 48-96 hour sample
N/A No MET No. assigned – component accounting for less than 10% sample radioactivity

Please refer to Table 8, 9, 10, 11 for the proportions of radioactive components present in the 1-12 hour pooled plasma extracts, 0-4 hour urine samples, 4-24 hour urine samples, and feces extracts, respectively.

Table 8 Proportions of radioactive components present in the 1-12 hour pooled plasma extracts *(Adapted from Table 7 in the study report)*

Component	Subject					
	001	002	003	004	005	006
P1- Glucuronide of Delafloxacin	0.268 (8.0)	0.180 (8.3)	0.410 (8.5)	0.411 (11.1)	0.353 (11.0)	0.353 (10.5)
P2- Delafloxacin	3.025 (90.4)	1.990 (91.7)	4.355 (90.4)	3.263 (88.1)	2.829 (88.1)	2.927 (87.1)

Results expressed as µg equivalents/g and, in parentheses % sample radioactivity
µg equivalents/g calculated using data from RBX/02

Table 9 Proportions of radioactive components present in the 0-4 hour urine samples *(Adapted from Table 8 in the study report)*

Component	Subject					
	001	002	003	004	005	006
U1- Glucuronide of Delafloxacin	13.77 (25.8)	13.78 (26.1)	14.22 (31.5)	13.78 (34.5)	11.84 (25.9)	6.63 (21.2)
U2- Delafloxacin	37.62 (70.5)	38.49 (72.9)	29.66 (65.7)	25.60 (64.1)	31.59 (69.1)	22.94 (73.4)

Results expressed as % administered radioactivity and, in parentheses % sample radioactivity
% administered radioactivity calculated using data from RBX/02

Table 10 Proportions of radioactive components present in the 4-24 hour urine samples *(Adapted from Table 9 in the study report)*

Component	Subject					
	001	002	003	004	005	006
U1- Glucuronide of Delafloxacin	7.26 (39.7)	6.16 (41.5)	8.06 (48.1)	9.39 (50.5)	6.67 (41.1)	8.72 (26.1)
U2- Delafloxacin	9.86 (53.9)	7.95 (53.6)	7.34 (43.8)	7.91 (42.5)	8.44 (52.0)	21.38 (64.0)

Results expressed as % administered radioactivity and, in parentheses % sample radioactivity
% administered radioactivity calculated using data from RBX/02

Table 11 Proportions of radioactive components present in the faeces extracts *(Adapted from Table 10 in the study report)*

	Subject											
	001		002		003		004		005		006	
Time point (hour) / Component	24-48	48-96	24-48	72-96	24-48	48-96	0-48	48-72	24-48	48-96	24-72	72-144
F1- Delafloxacin	9.32 (>99.9)	10.00 (>99.9)	24.53 (>99.9)	2.71 (>99.9)	11.45 (94.9)	19.28 (>99.9)	29.32 (>99.9)	4.17 (>99.9)	13.21 (>99.9)	15.01 (>99.9)	13.16 (95.8)	11.66 (92.0)

Results expressed as % administered radioactivity and, in parentheses % sample radioactivity
% administered radioactivity calculated using data from RBX/02

Safety:
No AEs were recorded during the study.

SPONSOR'S CONCLUSIONS

- An average of 93% of administered radioactivity was recovered during the study: 65% from the urine and 28% from the feces. Approximately half the radioactivity

administered as [14C]-RX-3341 was recovered from urine and feces within 24 h of the start of the infusion.

- Plasma RX-3341 and total radioactivity concentration vs time data were consistent with an 1 h IV infusion of [14C]-RX-3341.
- Based on individual estimates of AUC_{0-t}, between 66% and 80% of total radioactivity in the plasma was parent RX-3341, suggesting that formation of, and therefore systemic exposure to, metabolites or breakdown products was much less than to parent RX-3341.
- Total radioactivity appeared to be distributed to some extent to the cellular components of whole blood.
- [14C]-RX-3341 was well tolerated when a dose equivalent to approximately 300 mg. [14C]-RX-3341 was administered as an IV infusion, and no AEs were reported.
- There were no clinically significant findings in clinical laboratory assessments, vital signs parameters, ECG measurements or physical examinations.

REVIEWER ASSESSMENT:

It appears that delafloxacin is the predominant component in plasma after IV infusion, at least up to 6 hours. The identified major metabolite (M3) is through glucuronidation. This metabolite has been identified in the previous animal studies in this application. Please refer to the Pharm/Tox review by Dr. Amy Nostrandt.

4.5.12 Study No.: RX-3341-108

A PHASE 1, SINGLE-DOSE, SAFETY, TOLERABILITY, AND PHARMACOKINETIC STUDY OF INTRAVENOUS AND ORAL FORMULATIONS, AND A MAXIMUM TOLERATED DOSE STUDY OF DELAFLOXACIN IN HEALTHY SUBJECTS

Date(s): 22 December 2010 to 09 April 2011
Sponsor: Melinta Therapeutics, Inc, 300 George Street, Suite 301, New Haven, CT 06511
Clinical Site: Single site: PPD Phase I Clinic, located at 7551 Metro Center Drive, Suite 200, Austin, TX 78744
Analytical Site: (b) (4)

OBJECTIVE(S):

- The primary objectives were to determine the relative pharmacokinetics of 2 intravenous (IV) formulations of delafloxacin (Formulations A and B) in healthy male and female subjects, to determine the bioavailability of oral Formulation C relative to IV Formulation B, and to determine the maximum tolerated dose of delafloxacin Formulation B.
- The secondary objective was to determine the safety and tolerability of single IV doses of delafloxacin Formulations A and B and of a single oral dose of delafloxacin Formulation C in healthy male and female subjects.

METHODS

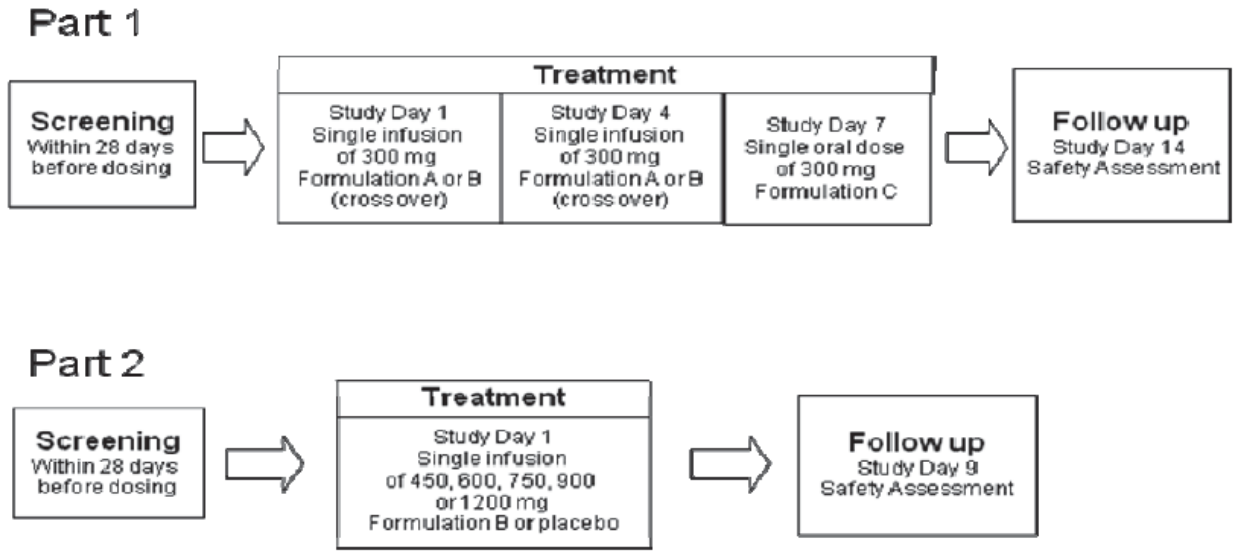
Study Design: This was a 2-part study in healthy male and female subjects.

- Part 1 was a double-blind, randomized, single IV dose, 2-treatment (Formulations A and B) crossover study followed by an additional single oral dose (Formulation C). Please refer to Figure 1 for details. On Day 1 of Part 1, 12 subjects (1:1) received a single IV infusion of 300-mg delafloxacin as either Formulation A or Formulation B according to the randomization schedule. On Day 4, subjects crossed over to receive the alternate formulation. On Day 7, all subjects received a single oral dose of delafloxacin Formulation C. There was a minimum of 72 hours between dose administrations.
- Part 2 was a double-blind, randomized, placebo-controlled, single IV maximum tolerated dose study with Formulation B. Please refer to Figure 1 for details. On Day 1 of Part 2, 10 subjects per group received a single IV infusion of 450-, 600-, 750-, 900-, or 1200-mg delafloxacin Formulation B or placebo (8:2), according to the randomization schedule. The 450-mg and 600-mg dose levels were administered to assigned subjects concurrently. Before sequential administration of the 750-mg and 900-mg dose levels, a minimum 7-day interval allowed the sponsor and investigator to conduct a blinded review of the safety data and an unblinded review of the PK data before proceeding to the next dose level. The 1200-mg dose group was added by protocol Amendment 2 after it was established that the maximum tolerated dose was indeterminable at the 900-mg dose level. A review of the safety and PK data from all previous groups suggested no concerns with increasing

the dose to 1200 mg. The stopping criterion for dose progression was the occurrence of 1 or more SAEs or any other medically important adverse event (AE) that impacted subject safety.

- Both Part 1 and Part 2 were conducted under the fasted condition.

Figure 1. Flowchart of Study Conduct *(Adapted from Figure 9.1 in the study report)*



Drug Products:

Delafloxacin Formulation A: captisol solution (b) (4)

Delafloxacin Formulation B: captisol solution with EDTA (to-be-marketed IV formulation)

Delafloxacin Formulation C: Phase 1 capsule used in most Phase 1 studies in this submission

Dose: 300 mg

Mode of Administration: 1-hour IV Infusion (Formulation A and Formulation B)
Oral (Formulation C)

PK Sample Collection:

In Parts 1 and 2, blood for plasma samples were collected before dosing and at 0.25, 0.5, 0.75, 1, 1.5, 2, 3, 4, 6, 8, 12, 16, 20, 24, 30, 36, and 48 hours after initiation of dosing. Urine samples were collected from –2 to 0 hours before dosing and at the following intervals after initiation of dosing: 0 to 4 hours, 4 to 8 hours, 8 to 12 hours, 12 to 24 hours, and 24 to 48 hours.

Analytical Methods:

The samples were analyzed for RX-3341 using LC-API/MS/MS detection. The analytical method for plasma samples is validated and detailed in (b) (4) Procedure BP3341C in this submission. A total of 22 analytical runs were required to analyze the plasma samples. One (1) analytical run was rejected due to low QC failure. The analytical method used to analyze the urine samples was validated and detailed in (b) (4) Procedure BP3341D in this submission. A total of 10 analytical runs were required to analyze the urine samples. One (1)

analytical run was rejected due to suspected curve preparation error. Please refer to the table below for details.

	Delafloxacin in Plasma	**Delafloxacin in Urine**
Method Validation #	(b) (4) 3341C	(b) (4) 3341D
Range	5.00- 5000 ng/mL	50.0 – 10000 ng/mL
LLOQ	5.0 ng/mL	50.0 ng/mL
Linearity	linear	linear
Accuracy	85-115%	85-115%
Precision	⩽ 15%	⩽ 15%
Stability	Long-term storage stability has been established for RX-3341 in human plasma for 374 days at -20°C (±10°C) and -70°C (±10°C). A total of 83 days (time from first sample drawn to the last sample analyzed) of long-term stability is required to cover the storage of the human plasma study samples at -70°C (±10°C). Stable through three freeze-thaw cycles.	Long-term storage stability has been established for RX-3341 in human urine for 168 days at -20°C (±10°C) and 229 days at -70°C (±10°C). A total of 95 days (time from first sample drawn to the last sample analyzed) of long-term stability is required to cover the storage of the human urine study samples at -70°C (±10°C). Stable through three freeze/thaw cycles.

Reviewer's Comment: The bioanalytical method and analytical run in this study meet the acceptance criteria specified in the FDA Guidance for Industry Bioanalytical Method Validation.

Pharmacokinetic Assessment:
The following PK parameters were calculated for delafloxacin from plasma concentration data for each subject using noncompartmental analysis:
Plasma:

- $AUC_{0-\tau}$: area under the plasma concentration versus time curve from time 0 to 12 hours from initiation of dosing
- AUC_{0-t}: area under the plasma concentration versus time curve from time 0 (initiation of dosing) to the time of the last quantifiable concentration
- $AUC_{0-\infty}$: area under the plasma concentration versus time curve from time 0 (initiation of dosing) extrapolated to infinityAUC_{0-24}: Area under the plasma concentration versus time curve from time 0 to 24 hours after dosing
- %F: bioavailability, calculated by dividing $AUC_{0-\infty}$ following oral administration by $AUC_{0-\infty}$ following IV administration × 100 (Part 1 only)
- C_{max}: Maximum observed plasma concentration
- CL: total body clearancet1/2: Terminal elimination half-life
- T_{max}: Time to achieve maximum observed plasma concentration
- CL/F: Apparent total clearance

- Vd/F: apparent volume of distribution
- K_{el}: apparent terminal elimination rate constant obtained from the concentration data
- V_z volume distribution

Urine:

- Ae_{0-4} total amount excreted unchanged in urine from time 0 to 4 hours after dosing
- Ae_{0-48} total amount excreted unchanged in urine from time 0 to 48 hours after dosing
- Ae_{4-8} total amount excreted unchanged in urine from time 4 to 8 hours after dosing
- Ae_{8-12} total amount excreted unchanged in urine from 8 to 12 hours after dosing
- Ae_{12-24} total amount excreted unchanged in urine from 12 to 24 hours after dosing
- Ae_{24-48} total amount excreted unchanged in urine from 24 to 48 hours after dosing
- $Fe\%_{0-12}$ fraction of the dose excreted unchanged in urine from 0 to 12 hours after dosing
- $Fe\%_{0-24}$ fraction of the dose excreted unchanged in urine from 0 to 24 hours after dosing
- $Fe\%_{0-48}$ fraction of the dose excreted unchanged in urine from 0 to 48 hours after dosing
- CLr renal clearance calculated as Ae_{0-48}/AUC_{0-t}

Statistical Methods:

Pharmacokinetics:

For Part 1, log-transformed PK parameters $AUC_{0-\tau}$, AUC_{0-t}, $AUC_{0-\infty}$, and C_{max} were analyzed by a mixed-effects model using formulation (Formulations A and B), period, and sequence as fixed effects and subject within sequence as a random effect. The 90% confidence interval of the ratio of the geometric means for the PK parameters for Formulations B and A was provided to evaluate relative pharmacokinetics of the 2 IV formulations. An analysis of variance model was used to evaluate the relative bioavailability of Formulation C to Formulation B. The 90% confidence interval of the ratio of the geometric means for dose-normalized $AUC_{0-\infty}$ for Formulations C and B was provided.

Bioavailability for each subject was calculated by dividing $AUC_{0-\infty}$ following oral administration of Formulation C by $AUC_{0-\infty}$ following IV administration of Formulation B.

Dose proportionality analyses using a power model was performed on AUC_{0-t}, $AUC_{0-\infty}$, and C_{max} for all subjects in the PK population who received Formulation B in Parts 1 and 2.

Safety: No formal statistical analysis of safety parameters was performed.

Safety Assessment:

- Adverse event (AE) reporting
- Clinical laboratory results (hematology [including coagulation assays], serum chemistry, and urinalysis)
- Vital sign measurements (oral temperature, systolic and diastolic blood pressure, pulse rate, and respiratory rate)
- 12-lead electrocardiogram (ECG) results
- Physical examination findings

RESULTS

Study Population:
Overall, demographic characteristics were similar in Part 1 across treatment sequences and in Part 2 across treatment groups. Please refer to Table 1 and Table 2 for details.

Table 1. Demographic and Baseline Characteristics (Safety Population) - Part 1 *(Adapted from Table 11-1 in the study report)*

	Treatment Sequence[a]		
	Delafloxacin		
	Formulation ABC (n=6)	Formulation BAC (n=6)	Overall (N=12)
Age (years)			
Mean	31.3	44.5	37.9
SD	7.97	15.15	13.43
Minimum, maximum	25, 47	22, 58	22, 58
Sex, n (%)			
Male	4 (66.7)	2 (33.3)	6 (50.0)
Female	2 (33.3)	4 (66.7)	6 (50.0)
Race, n (%)			
White	6 (100.0)	3 (50.0)	9 (75.0)
Black or African American	0	3 (50.0)	3 (25.0)
Ethnicity, n (%)			
Hispanic or Latino	0	2 (33.3)	2 (16.7)
Not Hispanic or Latino	6 (100.0)	4 (66.7)	10 (83.3)
Height (cm)			
Mean	170.32	164.27	167.29
SD	7.660	9.878	9.000
Minimum, maximum	156.3, 177.0	153.3, 178.5	153.3, 178.5
Weight (kg)			
Mean	71.17	74.45	72.81
SD	12.189	15.066	13.178
Minimum, maximum	56.3, 91.2	56.5, 96.4	56.3, 96.4
Body mass index (kg/m^2)			
Mean	24.40	27.22	25.81
SD	2.684	2.676	2.948
Minimum, maximum	21.9, 29.6	24.0, 30.3	21.9, 30.3

Note: Percentages were based on the number of subjects in the safety population within each treatment sequence and overall.

[a] Formulation A: delafloxacin 300 mg infused intravenously; Formulation B: delafloxacin 300 mg infused intravenously; Formulation C: delafloxacin 300 mg administered orally.

Table 2. Demographic and Baseline Characteristics (Safety Population) - Part 2 *(Adapted from Table 11-2 in the study report)*

	Treatment[a]						
		Delafloxacin					
	Placebo (n=10)	450 mg (n=8)	600 mg (n=8)	750 mg (n=8)	900 mg (n=8)	1200 mg (n=8)	Overall (N=50)
Age (years)							
Mean	31.2	38.5	38.9	39.4	32.9	38.3	36.3
SD	9.65	14.62	10.33	12.02	13.60	14.92	12.37
Minimum, maximum	21, 52	21. 59	25, 55	22, 53	19, 52	24, 64	19, 64
Sex, n (%)							
Male	6 (60.0)	5 (62.5)	5 (62.5)	4 (50.0)	5 (62.5)	4 (50.0)	29 (58.0)
Female	4 (40.0)	3 (37.5)	3 (37.5)	4 (50.0)	3 (37.5)	4 (50.0)	21 (42.0)
Race, n (%)							
White	9 (90.0)	6 (75.0)	7 (87.5)	7 (87.5)	6 (75.0)	5 (62.5)	40 (80.0)
Black or African American	1 (10.0)	1 (12.5)	1 (12.5)	1 (12.5)	2 (25.0)	3 (37.5)	9 (18.0)
Asian	0	1 (12.5)	0	0	0	0	1 (2.0)
Ethnicity, n (%)							
Hispanic or Latino	4 (40.0)	4 (50.0)	3 (37.5)	1 (12.5)	3 (37.5)	4 (50.0)	19 (38.0)
Not Hispanic/Latino	6 (60.0)	4 (50.0)	5 (62.5)	7 (87.5)	5 (62.5)	4 (50.0)	31 (62.0)
Height (cm)							
Mean	170.74	169.53	170.39	165.60	172.90	165.15	169.12
SD	7.169	7.577	10.586	8.818	6.730	9.031	8.404
Minimum, maximum	156.8, 179.0	159.7, 181.9	152.9, 180.8	154.0, 183.0	164.4, 182.2	153.2, 178.5	152.9, 183.0
Weight (kg)							
Mean	78.79	70.06	73.71	73.86	79.33	72.19	74.82
SD	14.417	8.800	8.539	10.730	10.487	14.911	11.631
Minimum, maximum	60.1, 100.6	56.5, 83.1	66.4, 90.7	60.4, 95.1	67.9, 98.6	50.8, 99.2	50.8, 100.6
Body mass index (kg/m^2)							
Mean	26.87	24.34	25.44	26.94	26.54	26.29	26.10
SD	3.383	2.033	2.326	3.183	3.102	3.751	3.024
Minimum, maximum	20.4, 31.4	21.5, 26.6	21.7, 28.4	20.9, 31.1	21.8, 30.2	19.9, 31.1	19.9, 31.4

Note: Percentages were based on the number of subjects in the safety population within each treatment and overall.
[a] Formulation B was used for all delafloxacin dose groups.

Pharmacokinetics:

Part 1

The mean (±SD) concentration versus time profile for delafloxacin formulation A versus B is presented in Figure 2 and formulation C versus B is in Figure 3 below.

Figure 2. Mean (±SD) Plasma Concentrations of Delafloxacin Versus Time by Treatment - Part 1 (A versus B) *(Adapted from Figure 11-1 in the study report)*

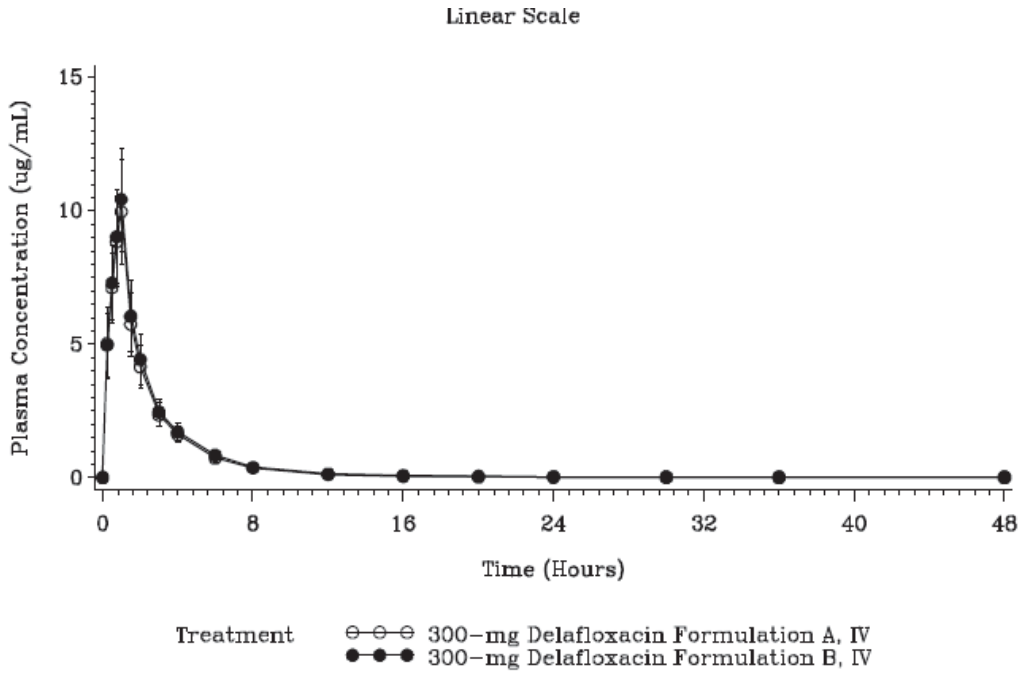
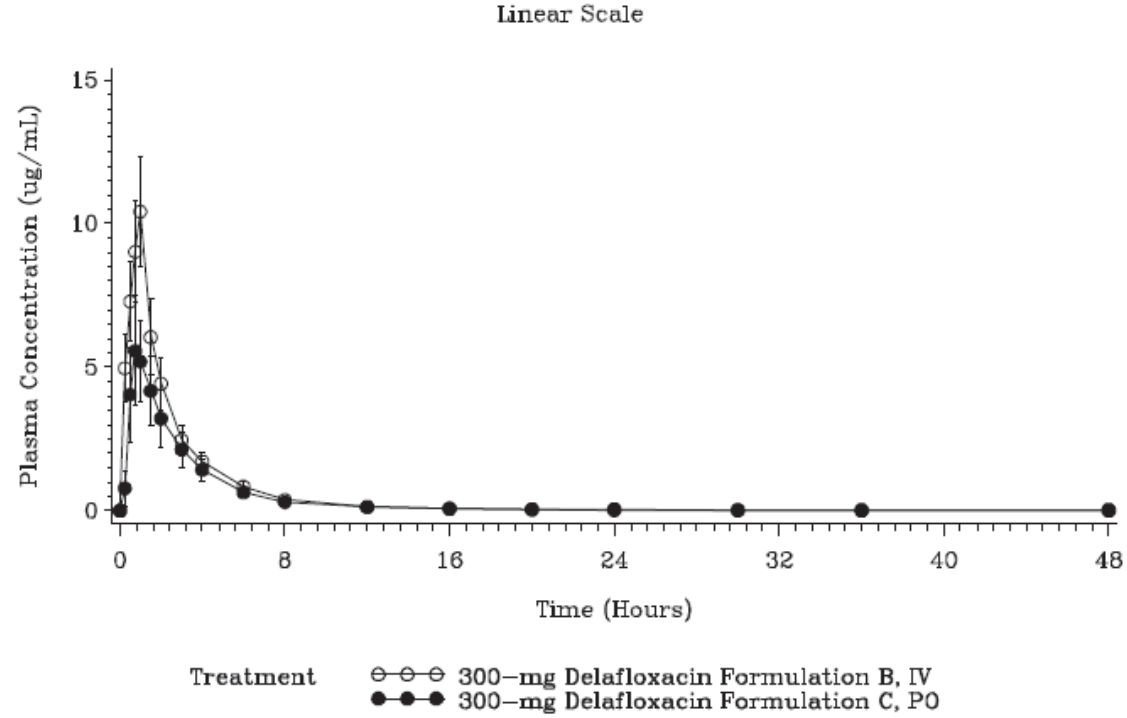


Figure 3. Mean (±SD) Plasma Concentrations of Delafloxacin Versus Time by Treatment - Part 1 (B versus C) *(Adapted from Figure 11-1.2 in the study report)*



The PK parameters for delafloxacin in plasma are summarized in Table 3.

Table 3. Mean (CV) Plasma Pharmacokinetic Parameters of Delafloxacin (Pharmacokinetic Population) *(Adapted from Table 11-3 in the study report)*

Parameter (unit)	Formulation A 300 mg, IV (n=12)	Formulation B 300 mg, IV (n=12)	Formulation C 300 mg, Oral (n=12)
$AUC_{0-\tau}$ (µg•h/mL)[a]	22.54 (19)	23.63 (19)	15.84 (24)
AUC_{0-t} (µg•h/mL)	23.51 (20)	24.74 (20)	17.31 (24)
$AUC_{0-\infty}$ (µg•h/mL)	23.61 (23)[b]	24.83 (20)[b]	17.76 (25)[d]
C_{max} (µg/mL)	10.11 (18)	10.43 (19)	5.78 (31)
T_{max} (h)[c]	1.0 (0.75,1.05)	1.0 (0.97, 1.08)	0.75 (0.50, 1.02)
CL (L/h) or CL/F (L/h)	13.32 (23)[b]	12.53 (21)[b]	17.85 (24)[d]
Vss (L)	38.0 (17)[b]	34.18 (20)[b]	ND
Vz (L) or Vd/F (L)	177.14 (39)[b]	146.13 (34)[b]	277.48 (30)[d]
$t_{1/2}$ (h)	9.90 (53)[b]	8.21 (33)[b]	10.86 (29)[d]

Abbreviation: CV, coefficient of variation; IV, intravenous; ND, not done
[a] tau = 12 hours
[b] n = 9
[c] Median (Minimum, Maximum)
[d] n = 11

A statistical analysis of the plasma PK parameters is presented in Table 4.

Table 4. Statistical Analysis of Plasma Pharmacokinetic Parameters (Pharmacokinetic Population) *(Adapted from Table 11-4 in the study report)*

Parameter (unit)	Treatment Comparison	n A	n B	n C	Ratio of Geometric Least Squares Means	90% Confidence Interval of the Ratio
$AUC_{0-\tau}$ (µg•h/mL)	B/A	12	12	–	1.0479	(1.0069, 1.0906)
AUC_{0-t} (µg•h/mL)	B/A	12	12	–	1.0522	(1.0045, 1.1022)
$AUC_{0-\infty}$ (µg•h/mL)	B/A	9	9	–	1.0246	(0.9552, 1.0990)
C_{max} (µg/mL)	B/A	12	12	–	1.0292	(0.9855, 1.0749)
$AUC_{0-\infty}$/Dose (µg•h/mL/mg)	C/B	–	9	11	0.7089	(0.6389, 0.7866)

Note: Ratio of geometric least squares means and 90% confidence interval for the ratio of geometric least squares means were estimated from a mixed-effects model with log-transformed pharmacokinetic parameter as the dependent variable, treatment, sequence, and period as fixed effects, and subject within sequence as a random effect.
Treatments: A = delafloxacin Formulation A 300 mg, IV; B = delafloxacin Formulation B 300 mg, IV; C= delafloxacin Formulation C 300 mg, oral.

Reviewer's Comment: Based on the results shown in Table 3 and Table 4, the two IV formulations demonstrated bioequivalence. The bioavailability for the Phase 1 capsule is around 70% by using the IV to-be-marketed formulation as a reference.

Please refer to Table 5 for the urinary PK parameters of delafloxacin.

Table 5. Mean (CV) Urinary Pharmacokinetic Parameters of Delafloxacin (Pharmacokinetic Population) *(Adapted from Table 11-5 in the study report)*

Parameter (unit)	Formulation A, IV 300 mg (n=12)	Formulation B, IV 300 mg (n=12)	Formulation C, Oral 300 mg (n=12)
Ae_{0-4} (mg)	88.97 (36)	94.56 (23)	53.63 (24)
Ae_{4-8} (mg)	14.31 (44)	13.39 (64)	13.52 (38)
Ae_{8-12} (mg)	3.50 (44)	4.79 (37)	3.52 (52)
Ae_{12-24} (mg)	2.29 (44)	2.28 (45)	3.20 (51)
Ae_{24-48} (mg)	1.49 (52)	1.72 (89)	2.56 (46)
Ae_{0-48} (mg)	110.55 (34)	116.74 (23)	76.43 (20)
$Fe\%_{0-12}$ (%)	35.59 (34)	37.58 (23)	23.56 (21)
$Fe\%_{0-24}$ (%)	36.35 (34)	38.34 (23)	24.62 (21)
$Fe\%_{0-48}$ (%)	36.85 (34)	38.91 (23)	25.48 (20)
CL_r (L/h)	4.98 (42)	4.98 (33)	4.64 (28)

Abbreviation: CV, coefficient of variation; IV, intravenous.

More than 36% of the drug was excreted unchanged in urine within 48 hours after dosing for both intravenous formulations, with most of the excretion occurring within the first 12 hours after dosing. Urinary drug excretion and renal clearance (CLr) for both formulations were similar.

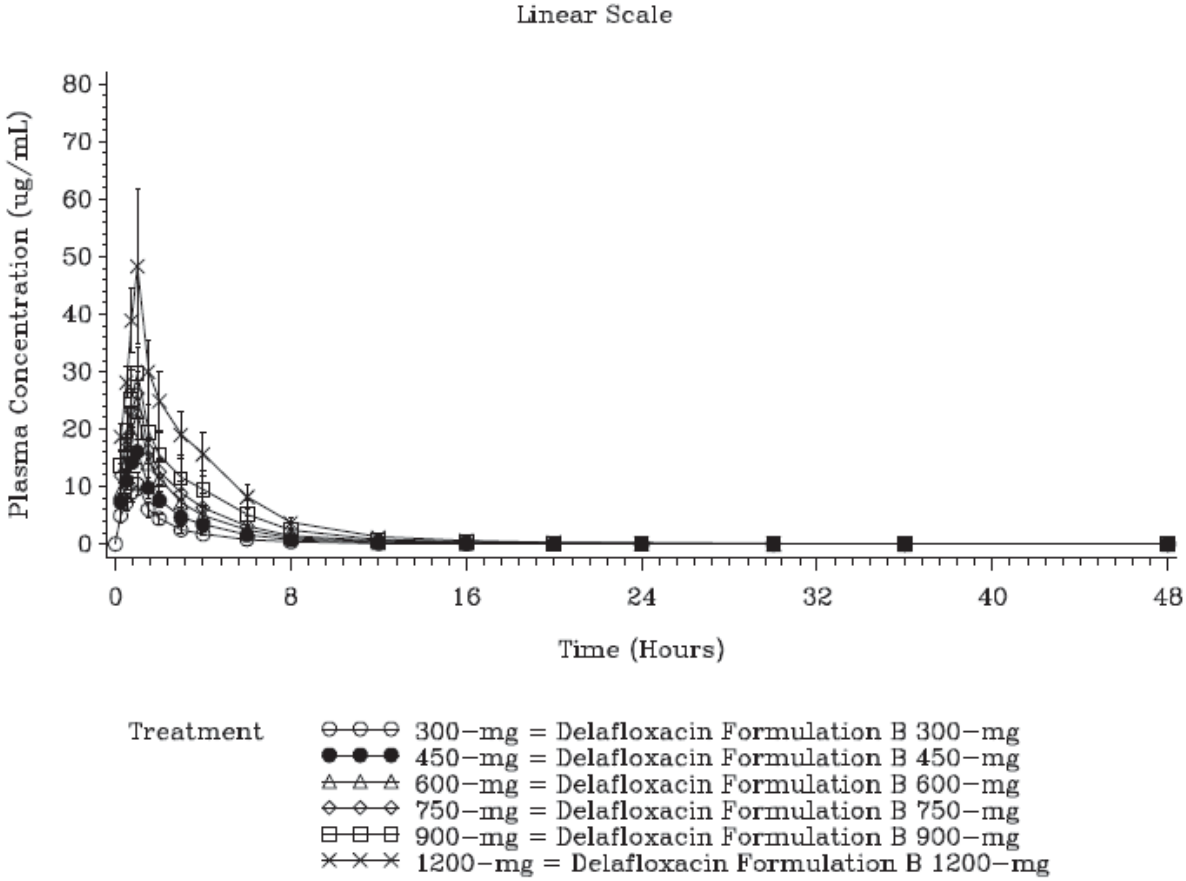
Likewise, after oral administration more than 25% of the drug was excreted unchanged in urine within 48 hours, with most of the excretion occurring within the first 12 hours after dosing. Renal clearance after oral administration was similar to that of Formulation B which was administered intravenously.

Part 2

In Part 2 of the study, 40 subjects were dosed with Formulation B with delafloxacin dose ranging from 450 mg to 1200 mg. The 12 subjects dosed with delafloxacin 300 mg (Formulation B) in Part 1 were also included in the PK analysis presented in this section.

Please refer to Figure 4 for the plasma concentration and time curve after administration of various dose levels of Formulation B.

Figure 4. Mean (±SD) Plasma Concentrations of Delafloxacin Over Time by Treatment - Formulation B *(Adapted from Figure 11-3 in the study report)*



The arithmetic means and CVs for selected PK parameters of delafloxacin (Part 2) are presented in Table 6.

Table 6. Mean (CV) Plasma Pharmacokinetic Parameters of Delafloxacin-Formulation B (Pharmacokinetic Population) *(Adapted from Table 11-6 in the study report)*

	Delafloxacin					
Parameter (unit)	**300 mg (n=12)**	**450 mg (n=8)**	**600 mg (n=8)**	**750 mg (n=8)**	**900 mg (n=8)**	**1200 mg (n=8)**
$AUC_{0-\tau}$ (µg•h/mL)[a]	23.63 (19)	40.56 (17)	58.59 (26)	70.61 (25)	92.42 (28)	146.93 (18)
AUC_{0-t} (µg•h/mL)	24.74 (0)	42.61 (17)	61.48 (24)	74.04 (27)	98.13 (29)	155.56 (18)
$AUC_{0-\infty}$ (µg•h/mL)	24.83 (20)[b]	42.87 (17)	59.03 (23)[d]	74.43 (5)[e]	99.34 (26)[f]	160.02 (12)[f]
C_{max} (µg/mL)	10.43 (19)	16.08 (13)	23.00 (21)	26.15 (15)	29.66 (16)	49.06 (25)
T_{max} (h)[c]	1.00 (0.97, 1.08)	1.00 (1.00, 1.00)	1.02 (1.00, 1.03)	1.00 (1.00, 1.15)	1.00 (1.00, 1.03)	1.02 (0.80, 1.05)
CL (L/h)	12.53 (21)[b]	10.78 (18)	10.63 (22)[d]	10.09 (5)[e]	9.65 (29)[f]	7.59 (13)[f]
Vss (L)	34.18 (20)[b]	36.63 (26)	38.46 (33)[d]	32.89 (13)[e]	36.40 (18)[f]	30.21 (14)[f]
Vz (L)	146.13 (34)[b]	197.87 (56)	193.63 (66)[d]	257.21 (9)[e]	147.61 (51)[f]	131.37 (44)[f]
$t_{1/2}$ (h)	8.21 (33)[b]	12.45 (44)	11.92 (45)[d]	17.74 (14)[e]	11.64 (67)[f]	11.71 (34)[f]

Abbreviation: CV, coefficient of variation.
[a] tau = 12 hours
[b] n = 9
[c] Median (Minimum, Maximum)
[d] n = 7
[e] n = 3
[f] n = 6

Please refer to Table 7 for the statistical analysis result of dose proportionality of formulation B.

Table 7. Analysis of Dose Proportionality - Formulation B *(Adapted from Table 11-7 in the study report)*

Parameter (unit)	**Estimated Slope for Ln (Dose)**	**90 % Confidence Interval for the Slope**	***P*-value (slope=1)**
AUC_{0-t} (µg•h/mL)	1.2790	(1.1700, 1.3880)	<0.0001
$AUC_{0-\infty}$ (µg•h/mL)	1.3064	(1.1969, 1.4158)	<0.0001
C_{max} (µg/mL)	1.0520	(0.9615, 1.1426)	0.3402

Note: The power model, ln(parameter) = a + b*ln(dose) + error, was used to estimate the slope, corresponding 90% confidence interval, and the P value testing dose proportionality (b=1).

Dose proportionality of delafloxacin was evaluated for the IV dose range of 300 mg to 1200 mg. The mean slopes and 90% confidence intervals were 1.31 (1.20, 1.42) and 1.28 (1.17, 1.39) for $AUC_{0-\infty}$, and AUC_{0-t}, respectively (pvalues <0.0001). The mean slope and 90% confidence intervals for C_{max} were 1.05 (0.96, 1.14) (p value = 0.3402). These results indicated that the AUCs increased in a greater than dose proportional manner over the 300- to 1200-mg dose range after a single dose of delafloxacin was administered intravenously in Part 2 of this study. However, C_{max} increased in a dose proportional manner over the 300- to 1200-mg dose range. When doses increased in ratios of 1.5 (450 mg/300 mg), 2 (600 mg/300 mg), 2.5 (750 mg/300 mg), 3 (900 mg/300 mg) and 4 (1200 mg/300 mg), the corresponding arithmetic mean ratios for $AUC_{0-\infty}$ were approximately 1.7, 2.4, 3.0, 4.0, and 6.4, respectively; the corresponding C_{max} ratios were 1.5, 2.2, 2.5, 2.8, and 4.7, respectively.

Please refer to Table 8 for the mean urinary PK parameters.

Table 8. Mean (CV) Urinary Pharmacokinetic Parameters of Delafloxacin-Formulation B (Pharmacokinetic Population) *(Adapted from Table 11-8 in the study report)*

	Delafloxacin[a]					
Parameter (unit)	300 mg (n=12)	450 mg (n=8)	600 mg (n=8)	750 mg (n=8)	900 mg (n=8)	1200 mg (n=8)
Ae_{0-4} (mg)	94.56 (23)	124.38 (26)	175.73 (16)	175.60 (29)	211.54 (33)	246.74 (25)
Ae_{4-8} (mg)	13.39 (64)	27.30 (15)	27.78 (47)	35.56 (51)	43.63 (68)	92.81 (44)
Ae_{8-12} (mg)	4.79 (37)	5.87 (53)	12.62(33)	11.64 (76)	24.88 (47)	39.92 (41)
Ae_{12-24} (mg)	2.28 (45)	4.57 (28)	6.72 (43)	7.25 (58)	16.16 (61)	18.01 (22)
Ae_{24-48} (mg)	1.72 (89)	2.79 (43)	3.47 (44)	5.20 (44)	8.25 (50)	12.15 (48)
Ae_{0-48} (mg)	116.74 (23)	164.91 (21)	226.31 (13)	235.26 (28)	304.46 (22)	409.65 (25)
$Fe\%_{0-12}$ (%)	37.58 (23)	35.01 (22)	36.02 (14)	29.71 (29)	31.12 (26)	31.62 (26)
$Fe\%_{0-24}$ (%)	38.34 (23)	36.03 (22)	37.14 (13)	30.67 (28)	32.91 (23)	33.12 (25)
$Fe\%_{0-48}$ (%)	38.91 (23)	36.65 (21)	37.72 (13)	31.37 (28)	33.83 (22)	34.14 (25)
CL_r (L/h)	4.98 (33)	4.04 (34)	3.95 (34)	3.24 (25)	3.47 (51)	2.64 (19)

Abbreviation: CV, coefficient of variation.
[a] Formulation B was used for all delafloxacin doses.

Reviewer's Comment:

- *AUCs of Delafloxacin increased slightly greater than proportionally over the dose range of 300 to 1200 mg (single dose).*
- *Total CL decreased with the increasing dose.*
- *The mean renal clearance (CLr) decreased with the increasing dose.*
- *It seems that Ae_{0-4} (total amount excreted unchanged in urine from time 0 to 4 hours after dosing) almost reached a plateau when doses increased to 1200 mg.*

Safety:
Overall, in Part 1 (single dose 300 mg IV), 9 TEAEs were reported and 4 of 12 subjects (33.3%) experienced at least 1 TEAE. Similar percentages of subjects experienced TEAEs across formulations. The most frequently reported TEAEs were dizziness and headache reported by 2 subjects each (16.7%) overall. No TEAE in any treatment group was reported by more than 1 subject. All TEAEs were either unlikely or possibly related to study drug. No subject had a TEAE that was probably or very likely/certainly related to study drug. All TEAEs were mild in severity.

Overall, in Part 2, 38 TEAEs were reported and 16 of 50 subjects (32.0%) experienced at least 1 TEAE. For the groups receiving delafloxacin, the number of TEAEs increased with increasing dose. The most frequently reported TEAEs overall were classified as gastrointestinal disorders (26.0%) and nervous system disorders (10.0%). The highest percentage of subjects (100.0%) reported TEAEs after receiving delafloxacin 1200 mg. Similar percentages of subjects reported TEAEs after receiving delafloxacin 750 mg (37.5%), delafloxacin 900 mg (37.5%), and placebo (20.0%). No TEAEs were reported by subjects after receiving delafloxacin 450 mg or 600 mg. The majority of TEAEs were of mild severity. Ten moderate TEAEs were reported after delafloxacin 1200 mg. Overall, the majority of TEAEs were possibly or probably related to study drug. All TEAEs considered probably related to study drug were reported after delafloxacin 1200 mg. No subject had a TEAE that was very likely or certainly related to study drug. Because of the occurrence of vomiting in 75% of subjects receiving the 1200-mg dose, the 900-mg dose was selected as the MTD.

There were no deaths, SAEs, or AEs leading to study discontinuation, and all TEAEs resolved by the end of the study.

Please refer to Table 9 and Table 10 for the TEAEs in part 1 and 2 of the study.

Table 9. Treatment-Emergent Adverse Events – Part 1*(Adapted from Table 12-1 in the study report)*

System Organ Class Preferred Term	Number (%) of Subjects by Treatment Group Treatment[a] Delafloxacin Formulation A 300 mg IV (n=12)	Delafloxacin Formulation B 300 mg IV (n=12)	Delafloxacin Formulation C 300 mg oral (n=12)	Overall (N=12)
Total number of adverse events	4	3	2	9
Subjects with at least 1 TEAE	3 (25.0)	2 (16.7)	2 (16.7)	4 (33.3)
Subjects with at least 1 treatment-related AE				
Possibly related	1 (8.3)	1 (8.3)	2 (16.7)	2 (16.7)
Probably related	0	0	0	0
Very likely/certainly related	0	0	0	0
Gastrointestinal disorders	2 (16.7)	1 (8.3)	0	2 (16.7)
Abdominal distension	0	1 (8.3)	0	1 (8.3)
Anal haemorrhage	1 (8.3)	0	0	1 (8.3)
Nausea	1 (8.3)	0	0	1 (8.3)
Infections and infestations	0	0	1 (8.3)	1 (8.3)
Vulvovaginal candidiasis	0	0	1 (8.3)	1 (8.3)
Nervous system disorders	2 (16.7)	1 (8.3)	1 (8.3)	2 (16.7)
Dizziness	1 (8.3)	1 (8.3)	0	2 (16.7)
Headache	1 (8.3)	0	1 (8.3)	2 (16.7)
Respiratory, thoracic and mediastinal disorders	0	1 (8.3)	0	1 (8.3)
Nasal congestion	0	1 (8.3)	0	1 (8.3)

Abbreviation: IV, intravenous; TEAE, treatment-emergent adverse event.

Note: The total number of adverse events counts all TEAEs in the safety population. Subjects may have had more than 1 TEAE per system organ class and preferred term. At each level of subject summarization, a subject was counted once if he or she reported 1 or more events. The TEAEs were summarized by treatment at onset of the event. Adverse events were coded using MedDRA Version 13.1. Percentages were based on the number of subjects in the safety population who received the specified treatment.

[a] Formulation A: delafloxacin 300 mg infused intravenously; Formulation B: delafloxacin 300 mg infused intravenously; Formulation C: delafloxacin 300 mg administered orally.

Table 10. Treatment-Emergent Adverse Events – Part 2

System Organ Class Preferred Term	Number (%) of Subjects by Treatment Group[a] Placebo (n=10)	Delafloxacin 450 mg IV (n=8)	Delafloxacin 600 mg IV (n=8)	Delafloxacin 750 mg IV (n=8)	Delafloxacin 900 mg IV (n=8)	Delafloxacin 1200 mg IV (n=8)	Overall (N=50)
Total number of adverse events	4	0	0	5	11	18	38
Subjects with at least 1 TEAE	2 (20.0)	0	0	3 (37.5)	3 (37.5)	8 (100.0)	16 (32.0)
Subjects with at least 1 treatment-related AE							
Possibly related	1 (10.0)	0	0	2 (25.0)	3 (37.5)	1 (12.5)	7 (14.0)
Probably related	0	0	0	0	0	7 (87.5)	7 (14.0)
Very likely/certainly related	0	0	0	0	0	0	0
Gastrointestinal disorders	1 (10.0)	0	0	2 (25.0)	3 (37.5)	7 (87.5)	13 (26.0)
Nausea	1 (10.0)	0	0	2 (25.0)	3 (37.5)	4 (50.0)	10 (20.0)
Vomiting	0	0	0	0	0	6 (75.0)	6 (12.0)
Diarrhoea	0	0	0	1 (12.5)	1 (12.5)	2 (25.0)	4 (8.0)
Abdominal pain	0	0	0	1 (12.5)	0	0	1 (2.0)
Retching	0	0	0	0	0	1 (12.5)	1 (2.0)
General disorders and administration site conditions	1 (10.0)	0	0	0	1 (12.5)	1 (12.5)	3 (6.0)
Feeling hot	0	0	0	0	1 (12.5)	1 (12.5)	2 (4.0)
Asthenia	0	0	0	0	1 (12.5)	0	1 (2.0)
Thirst	0	0	0	0	1 (12.5)	0	1 (2.0)
Vessel puncture site swelling	1 (10.0)	0	0	0	0	0	1 (2.0)
Nervous system disorders	1 (10.0)	0	0	1 (12.5)	2 (25.0)	1 (12.5)	5 (10.0)
Headache	1 (10.0)	0	0	0	2 (25.0)	0	3 (6.0)
Dizziness	0	0	0	0	1 (12.5)	1 (12.5)	2 (4.0)
Presyncope	0	0	0	1 (12.5)	0	0	1 (2.0)
Somnolence	1 (10.0)	0	0	0	0	0	1 (2.0)

System Organ Class Preferred Term	Number (%) of Subjects by Treatment Group[a] Placebo (n=10)	Delafloxacin 450 mg IV (n=8)	Delafloxacin 600 mg IV (n=8)	Delafloxacin 750 mg IV (n=8)	Delafloxacin 900 mg IV (n=8)	Delafloxacin 1200 mg IV (n=8)	Overall (N=50)
Respiratory, thoracic and mediastinal disorders	0	0	0	0	0	1 (12.5)	1 (2.0)
Nasal congestion	0	0	0	0	0	1 (12.5)	1 (2.0)
Rhinorrhoea	0	0	0	0	0	1 (12.5)	1 (2.0)
Skin and subcutaneous tissue disorders	0	0	0	0	1 (12.5)	1 (12.5)	2 (4.0)
Hyperhidrosis	0	0	0	0	1 (12.5)	0	1 (2.0)
Pruritus	0	0	0	0	0	1 (12.5)	1 (2.0)

Abbreviation: IV, intravenous; TEAE, treatment-emergent adverse event.
Note: The total number of adverse events counts all TEAEs in the safety population. Subjects may have had more than 1 TEAE per system organ class and preferred term. At each level of subject summarization, a subject was counted once if he or she reported 1 or more events. The TEAEs were summarized by treatment at onset of the event. Adverse events were coded using MedDRA Version 13.1. Percentages were based on the number of subjects in the safety population who received the specified treatment.
[a] Treatments are delafloxacin Formulation B infused intravenously.

Overall, mean values and changes from Baseline in hematology, serum chemistry, and urinalysis results were similar across all treatment groups at all time points. There were no clinically significant abnormal laboratory results and no apparent treatment sequence- or dose-related trends in changes from Baseline in either Part 1 or Part 2.

Reviewer's Comment:

- *MTD for to-be-marketed IV dosage form was 900 mg due to the occurrence of vomiting in 75% of subjects receiving the 1200 mg dose.*
- *The frequency of TEAEs increased with increasing dose.*

SPONSOR'S CONCLUSIONS

PHARMACOKINETIC

- Formulations A and B showed equivalent bioavailability.

- The oral formulation (Formulation C) yielded approximately 70% of the total exposure of the IV Formulation B.
- Delafloxacin total exposure (AUC) increased greater than proportionally over the 300- to 1200-mg dose range.
- Total body clearance (CL) decreased with increasing dose.
- More than 30% of the drug was excreted unchanged in urine following single dose administration, with most of the drug excretion occurring within the first 12 hours after dosing.
- Mean renal clearance was approximately 5 L/h after administration of delafloxacin 300 mg and decreased to approximately 2.6 L/h as dose increased to 1200 mg.

SAFETY

- In Part 1, the number of TEAEs was similar across the 3 formulations, and no TEAE in any treatment group was reported by more than 1 subject.
- Overall in Part 2, 16 subjects (32.0%) reported a total of 38 TEAEs; the majority of these TEAEs were mild and considered possibly related to delafloxacin. Overall, the largest number of subjects experienced TEAEs in the gastrointestinal disorders SOC (primarily nausea, vomiting, and diarrhea).
- Compared with the other delafloxacin dose groups and the placebo group in Part 2, subjects in the 1200-mg treatment group reported the largest number of TEAEs, with most of these TEAEs of moderate severity and probably related to delafloxacin.
- Single IV infusions of delafloxacin Formulation B at doses of 450, 600, 750, and 900 mg appeared to be safe and well tolerated by the healthy subjects in this study. A single IV infusion of 1200 mg delafloxacin Formulation B was not as well tolerated. Because of the occurrence of vomiting in 75% of subjects receiving the 1200-mg dose, the 900-mg dose was selected as the MTD.
- There were no clinically significant findings noted or TEAEs reported resulting from clinical laboratory assessments, vital sign measurements, ECG results, or physical examination findings in either Part 1 or Part 2.

REVIEWER ASSESSMENT:

- Based on Part 1 results, formulations A and B showed comparable PK profiles after a 300 mg IV single dose.
- The oral formulation (Formulation C, Phase 1 capsule) yielded approximately 70% of AUC of the IV Formulation B (to-be-marketed IV dosage form).
- The MTD for the single IV dose of delafloxacin was 900 mg based on the results of Part 2 study.

4.5.13 Study No.: RX-3341-110

A PHASE I, MULTICENTER, OPEN-LABEL, PARALLEL-GROUP CROSSOVER STUDY TO ASSESS THE EFFECT OF RENAL IMPAIRMENT ON THE SINGLE-DOSE PHARMACOKINETICS AND SAFETY OF ORAL AND IV DELAFLOXACIN (RX-3341) AND IV PLACEBO

Date(s): 28 July 2011 to 21 April 2013
Sponsor: Melinta Therapeutics, Inc 300 George Street, Suite 301, New Haven, CT 06511
Clinical Site: Single site: DaVita Clinical Research, 825 South Eighth Street, Suite 300, Minneapolis, MN 55404.
Analytical Site:

- Bioanalytical Laboratory (for analysis of delafloxacin [RX-3341] and determination of delafloxacin plasma protein binding):
  - (b) (4)
- Bioanalytical Laboratory (for analysis of Captisol®):
  - (b) (4)
- (b) (4) served as the contract research organization for this study and was responsible for program management, clinical and medical monitoring, clinical data management, pharmacokinetic (PK) and biostatistical analysis, and final reporting.

OBJECTIVE(S):

- The primary objective was to compare the pharmacokinetics of delafloxacin administered as a 400-mg oral dose (four 100-mg capsules) and a 300-mg IV infusion with Captisol to normal healthy subjects and subjects with mild, moderate, and severe renal impairment, and a 300-mg delafloxacin/Captisol IV infusion in dialysis subjects before and after hemodialysis sessions.
- The secondary objective was to compare the safety and pharmacokinetics of a placebo/Captisol IV infusion with a 300-mg delafloxacin/Captisol IV infusion in normal healthy subjects and subjects with mild, moderate, and severe renal impairment.

METHODS

Study Design: This was a Phase 1, open-label, parallel-group, single-dose study. A total of 40 subjects were planned (5 groups of 8 subjects each) as follows:

- Group A: healthy subjects (estimated glomerular filtration rate [eGFR] of >80 mL/min/1.73 m^2)
- Group B: subjects with mild renal impairment (eGFR >50-80 mL/min/1.73 m^2)
- Group C: subjects with moderate renal impairment (eGFR >30-50 mL/min/1.73 m^2)
- Group D: subjects with severe renal impairment (eGFR $\leq$30 mL/min/1.73 m^2)
- Group E: subjects with end-stage renal disease (ESRD) on hemodialysis

Healthy subjects (Group A) were matched to the renally impaired subjects in terms of gender distribution, mean age (±10 years), and mean body mass index (±20%)

Subjects in Groups A through D participated in 3 treatment periods, with dosing in these periods separated by a washout period of at least 14 days. Subjects in Groups A through D entered the clinic the day before each study drug administration (Day –1). On Day 1 of each period, these subjects received a single dose of study drug as follows:

- Period 1: 300-mg delafloxacin/Captisol 1-hour IV infusion
- Period 2: placebo/Captisol 1-hour IV infusion
- Period 3: 400-mg oral delafloxacin (four 100-mg capsules)

Subjects fasted for 4 hours before and after dosing (or after the end of the infusion for the IV treatments). Samples of blood and urine were collected for pharmacokinetic (PK) analysis of delafloxacin and Captisol concentrations (delafloxacin only in Period 3) through approximately 48 hours after dosing.

Subjects in Group E participated in 2 treatment periods, with dosing in these periods separated by a washout period of at least 14 days. Subjects in Group E entered the clinic the day before each study drug administration (Day –1). On Day 1 of each period, subjects in Group E received a single dose of study drug as follows:

- Period 1: 300-mg delafloxacin/Captisol 1-hour IV infusion starting approximately 1 hour before initiation of the last hemodialysis session of the week
- Period 2: 300-mg delafloxacin/Captisol 1-hour IV infusion starting within 1 hour after completion of the last hemodialysis session of the week

Subjects fasted for 4 hours before the start of the infusion and for 4 hours after the end of the infusion. Samples of blood and urine (if available) were collected for PK analysis of delafloxacin and Captisol concentrations before dosing, before hemodialysis, during hemodialysis, and, in Period 2, up to approximately 65 to 69 hours after dosing (ie, the last sample was collected before the next hemodialysis session). In addition, dialysate samples were collected for determination of delafloxacin and Captisol concentrations.

Subjects in Group A to E were discharged on Day 3 after all scheduled PK and safety assessments had been completed except the subjects of Period 2 in Group E were discharged on Day 4. All these subjects returned to the clinic for a follow-up visit approximately 14 days ((±2 days) after the last dose was administered.

Drug Product: Delafloxacin infusion was supplied as a lyophilized powder in (b) (4) glass vials (to-be-marketed IV formulation). Lot Number: 098-101123;
Oral delafloxacin was supplied as active pharmaceutical ingredient in 100-mg capsules. Lot number: L0305619;

Inclusion Criteria: Male and female subjects between 18 and 80 years of age, inclusive, with either normal renal function or with renal impairment. Subjects with normal renal function were matched to the body mass index, age, and gender of subjects with renal impairment.

PK Sample Collection:
Serial blood, urine, and dialysate sampling was collected for the determination of concentrations of delafloxacin and Captisol in plasma, urine, and dialysate.

Blood
For the IV treatments, blood for plasma samples was collected before dosing and at 0.33, 0.66, 1 (end of infusion), 1.083, 1.167, 1.33, 1.5, 2, 3, 4, 6, 8, 12, 24, 36, and 48 hours after the start of the infusion. Subjects in Group E had an additional blood sample collected between 65 and 69 hours after dosing in Period 2 only.

For the oral treatment, blood for plasma samples was collected before dosing and at 0.5, 1, 1.5, 2, 3, 4, 6, 8, 12, 16, 20, 24, 30, 36, and 48 hours after dosing.

Subjects in Group E had additional arterial and venous blood samples collected during dialysis in Period 1. These pre and post-dialyzer PK plasma samples were collected hourly during the hemodialysis session at 1, 2, and 3 hours after initiation of dialysis, and at dialysis completion if less than or equal to 4 hours. If dialysis exceeded 4 hours, the last sample was obtained at 4 hours.

Urine
For the IV and oral treatments, urine samples were collected in intervals from –2 to 0 hours, and after dosing from 0 to 12, 12 to 24, 24 to 36, and 36 to 48 hours. Subjects in Group E had an additional urine sample collected during the 48- to 69-hour interval after dosing in Period 2 only. It was noted that subjects in Group E may not have been able to produce urine samples due to their medical condition.

Dialysate
Subjects in Group E had dialysate fluid collected on Day 1 after dosing (Period 1 only) over each 1-hour collection interval of the hemodialysis session (1, 2, and 3 hours after the start of the hemodialysis and at the completion) to determine the amount of delafloxacin and Captisol present in the fluid during hemodialysis.

Analytical Methods:
The Sponsor provided three bioanalytical reports as listed below.

- Bioanalylical Report for the Analysis of RX-3341 in Human Samples (completed on June 19, 2013)
- Bioanalytical Final Report: Quantitation of Captisol in Human Plasma, Urine and Dialysate Samples Collected Under Rib-X Pharmaceuticals Protocol RX-3341-110, completion date of 06 August 2013
- Final Report Amendment No. 1, Report Title: Partial Validation of an LC-MS/MS Method for the Determination of Captisol in Human Dialysate, Report Amendment date of 06 August 2013

Delafloxacin in human plasma, urine, and dialysate samples

	Delafloxacin in Plasma	**Delafloxacin in Urine**	**Delafloxacin in Dialysate**
Method Validation #	(b) (4) 3341C	(b) (4) 3341D	(b) (4) 3341E
Range	5.00- 5000 ng/mL	50.0 – 10000 ng/mL	5.00-2500 ng/mL
LLOQ	5.00 ng/mL	50 ng/mL	5.00 ng/mL
Linearity	linear	linear	linear
Accuracy	85-115%	85-115%	85-115%
Precision	⩽ 15%	⩽ 15%	<15%
Recovery over range	low, mid, and high QC samples were 83.3%, 87.7%, and 89.9%, respectively	low, mid, and high QC samples were 96.5%, 94.5%, and 92.3%, respectively	Low, Mid and High QC samples were 87.6%, 86.9% and 87.7%, respectively
Selectivity	no interference	no interference	no interference
Stability	Stable at room temperature for 23 and 49 hours and through three freeze/thaw cycles	Stable at room temperature for at least 22 hours and through three freeze/thaw cycles.	Stable at room temperature for at least 22 hours and through three freeze/thaw cycles.
Quality Control (QC)	QC samples were prepared at concentrations of 15, 100, and 2000 ng/mL.	QC samples were prepared at concentrations of 150, 4000, and 8000 ng/mL,	QC samples were prepared at concentrations of 15, 100, and 2000 ng/mL.

Protein binding assay for delafloxacin

This bioanalytical method validation report describes the results of the analysis of RX-3341 in 1:1 PBS:human plasma. A full validation was conducted in human plasma (K2EDTA), Study (b) (4) 3341F, in module 5.3.1.4. A reversed-phase LC-MS/MS method was validated with a range of 0.00500 - 2.50 μg/mL.

Please refer to the table below for a summary of the validation data for the analyte.

Analyte:	RX-3341
Matrix:	1:1 PBS:Human Plasma (K_2EDTA)
Analytical Procedure:	LC-MS/MS
Sample Preparation:	Liquid-liquid extraction
Assay Aliquot Volume:	50 µL
Assay Range:	0.00500 - 2.50 µg/mL
Regression:	$1/x^2$, linear
Injector Carryover:	None detected
[a]**Validation Batch Acceptance** (Acceptable / Total Primary Runs):	2/2
[b]**Intra-Assay Accuracy** (%DEV):	0.167 to 12.4
[b]**Intra-Assay Precision** (%CV):	2.25 to 5.16
Inter-Assay Accuracy (%DEV):	-0.0417 to 5.22
Inter-Assay Precision (%CV):	2.12 to 8.68
Dilutions Established:	1:20
Short-Term Matrix Stability (RT):	4 hours
Freeze/Thaw Stability (-70°C):	3 cycles
Extract Stability (RT):	3 hours
Autoinjector Stability (RT), Original Curve Quantification:	Not established
Autoinjector Stability (RT), Re-Injected Curve Quantification:	70 hours
Largest Validation Batch Size	115 (29902_001)

RT = Room Temperature

[a] Primary runs were 29902_001 and 29902_002.

[b] Run 29902_001.

Captisol in human plasma, urine, and dialysate samples

	Captisol in Plasma	**Captisol in Urine**	**Captisol in Dialysate**
Method Validation #	64743	67598	67600
Range	5.00 to 200 µg/mL	5.00 to 200 µg/mL	2.00 to 200 µg/mL
LLOQ	5 µg/mL	5 µg/mL	2 µg/mL
Linearity	linear (correlation coefficient ⩾0.9933)	linear (correlation coefficient ⩾0.990)	linear (correlation coefficient ⩾0.990)
Accuracy	85-115%	85-115%	85-115%
Precision	⩽ 15%	⩽ 15%	⩽ 15%
Selectivity	no interference	no interference	no interference

Stability	Stable at room temperature for at least 29 hours and through three freeze/thaw cycles	190 days at ⩽ -70 ° C;179 days at -20°C; 29 hr at ambient temperature; and through three freeze/thaw cycles	185 days at ⩽ -70 ° C; 5 hours at ambient temperature; and through three freeze/thaw cycles
Quality Control (QC)	QC samples were prepared at concentrations of 6, 40, and 150 µg/mL.	QC samples were prepared at concentrations of 6, 40, 150 µg/mL.	QC samples were prepared at concentrations of 6, 40, 150 µg/mL.

Pharmacokinetic Assessment:
The following PK parameters were estimated for delafloxacin and Captisol using noncompartmental analysis:

Plasma: $AUC_{0\text{-}inf}$, $AUC_{0\text{-}t}$, CL, CL/F, CL_{NR}, CL_u (clearance after IV administration corrected for plasma protein binding (delafloxacin only)), CL_u/F, C_{max}, F_u (fraction of delafloxacin unbound to plasma protein (delafloxacin only)), $t_{1/2}$, T_{max}

Urine: $Ae_{0\text{-}12}$, $Ae_{12\text{-}24}$, $Ae_{24\text{-}36}$, $Ae_{36\text{-}48}$, $Ae_{48\text{-}69}$ (for Group E, Period 2 only), $Ae_{0\text{-}48}$, $Ae_{0\text{-}69}$ (for Group E, Period 2 only), CLr, $Fe\%_{0\text{-}69}$ (of the dose excreted unchanged in urine from 0 to 69 hours after dosing (Group E, Period 2 only));

Dialysate (Group E subjects in Period 1): Arem0-1 (amount of drug removed by hemodialysis from time 0 to 1 hour after the start of hemodialysis), Arem1-2, Arem2-3, Arem3-4, Arem0-4, CLd, $Frem\%_{0\text{-}4}$ (fraction (%) of the dose removed by hemodialysis from 0 to 4 hours after the start of hemodialysis (or to the end of dialysis if less than 4 hours));

Statistical Methods:
Pharmacokinetics:
Plasma and urine concentrations of delafloxacin and Captisol and the associated PK parameters were listed and summarized using descriptive statistics (number of subjects, mean, median, SD, minimum, maximum, and coefficient of variation). Mean and individual plasma concentration-time profiles were presented graphically on linear and semilogarithmic scales.

The natural log-transformed $AUC_{0\text{-}t}$, $AUC_{0\text{-}inf}$, and C_{max} values for delafloxacin and Captisol in the renally impaired and healthy groups were compared using an analysis of variance with group as a fixed effect. Geometric mean ratios between renally impaired and healthy groups and 90% confidence intervals (CIs) for the ratios of $AUC_{0\text{-}t}$, $AUC_{0\text{-}inf}$, and C_{max} of both delafloxacin and Captisol were presented. These statistical analyses were performed with subjects classified by both the Cockroft-Gault and modification of diet in renal disease (MDRD) equations.

The effect of delafloxacin on the pharmacokinetics of Captisol as measured by $AUC_{0\text{-}t}$, $AUC_{0\text{-}inf}$, and C_{max} was also evaluated using a linear mixed model with treatment (300-mg IV or placebo) as a fixed effect and subject as a random effect. Interaction between group and treatment was

evaluated first. If the interaction was statistically significant, then the analysis of the delafloxacin effect on Captisol was performed separately for each group.

Linear regression analysis was used to evaluate the relationships between estimated renal function and relevant PK parameters (AUC_{0-t}, AUC_{0-inf}, and C_{max}, and CL or CL/F) of delafloxacin. The possible relationships were explored using both the Cockroft-Gault and MDRD equations as measures of renal function. If the data permitted the identification of linear functions that fit the data well, the resulting slopes were to provide a comparison of any difference in the response of the oral and IV delafloxacin treatments as renal function changed. These estimates of the mean slopes and their 95% CIs were summarized.

Safety: No formal statistical analysis of safety parameters was performed.

Safety Assessment: Safety assessments included evaluation of AEs, clinical laboratory results (hematology [including coagulation], serum chemistry [including liver function parameters], and urinalysis), vital sign measurements (blood pressure, heart rate, and body temperature), 12-lead ECG measurements, and physical examination findings.

RESULTS

Study Population:
Forty-four subjects were enrolled into this study. Forty subjects received delafloxacin and Captisol, had evaluable PK data, and were included in the PK analysis. Subjects 0001102 (healthy group), 0001406 (severe renal impairment group), 0001502 (ESRD group), and 0001504 (ESRD group) were excluded from the PK analysis population due to subject replacement or because the subject did not have evaluable PK concentrations.

The subject demographic and baseline characteristics are summarized in Table 1.

Table 1. Summary of Subject Demographics and Baseline Characteristics (Safety Population)

	Group[a]					
	Group A Normal Renal Function (n = 9)	Group B Mild Renal Impairment (n = 8)	Group C Moderate Renal Impairment (n = 8)	Group D Severe Renal Impairment (n = 9)	Group E ESRD (n = 10)	Overall (N = 44)
Age (years)						
Mean	51.6	55.9	56.8	53.8	51.2	53.7
SD	4.00	9.76	9.30	8.63	16.57	10.44
Minimum, maximum	46, 59	42, 70	42, 70	40, 66	26, 71	26, 71
Sex, No. (%)						
Male	4 (44.4)	5 (62.5)	5 (62.5)	4 (44.4)	7 (70.0)	25 (56.8)
Female	5 (55.6)	3 (37.5)	3 (37.5)	5 (55.6)	3 (30.0)	19 (43.2)
Race, No. (%)						
White	8 (88.9)	4 (50.0)	2 (25.0)	4 (44.4)	3 (30.0)	21 (47.7)
Black or African American	1 (11.1)	4 (50.0)	5 (62.5)	4 (44.4)	7 (70.0)	21 (47.7)
American Indian or Alaska Native	0	0	1 (12.5)	0	0	1 (2.3)
Multi-racial	0	0	0	1 (11.1)	0	1 (2.3)
Ethnicity, No. (%)						
Not Hispanic or Latino	9 (100.0)	8 (100.0)	8 (100.0)	9 (100.0)	10 (100.0)	44 (100.0)
Height (cm)						
Mean	174.14	173.56	175.43	170.18	170.56	172.65
SD	12.876	8.070	8.899	9.399	11.476	10.134
Minimum, maximum	160.0, 198.1	160.7, 182.8	160.7, 186.7	159.4, 182.9	152.4, 185.4	152.4, 198.1
Weight (kg)						
Mean	83.72	93.96	98.23	91.92	82.83	89.70
SD	12.904	14.818	24.951	18.123	24.163	19.692
Minimum, maximum	66.3, 107.9	71.8, 117.1	68.4, 140.4	64.0, 116.2	45.9, 118.2	45.9, 140.4
Body mass index (kg/m^2)						
Mean	27.52	31.05	31.78	31.66	27.89	29.87
SD	2.061	3.395	6.762	5.261	5.755	5.085
Minimum, maximum	25.0, 31.7	27.8, 37.8	23.6, 40.3	23.9, 38.4	18.4, 39.4	18.4, 40.3

	Group[a]					
	Group A Normal Renal Function (n = 9)	Group B Mild Renal Impairment (n = 8)	Group C Moderate Renal Impairment (n = 8)	Group D Severe Renal Impairment (n = 9)	Group E ESRD (n = 10)	Overall (N = 44)
eGFR (mL/min/1.73 m^2)						
Mean	91.9	62.8	38.6	22.3	6.4	43.3
SD	11.43	7.78	4.90	6.18	1.78	31.99
Minimum, maximum	83, 119	53, 78	31, 45	9, 29	5, 10	5, 119
CLcr (mL/min)						
Mean	121.2	86.8	57.6	34.8	10.3	60.5
SD	19.13	15.78	14.77	10.43	4.14	42.36
Minimum, maximum	106, 163	60, 110	38, 79	20, 55	6, 20	6, 163

Abbreviations: CLcr, creatinine clearance; eGFR, estimated glomerular filtration rate; ESRD, end-stage renal disease; SD, standard deviation.

Note: For re-enrolled subjects, data collected from the original enrollment were not included in this table. Percentages were based on the number of subjects in the safety population within each group.

[a] Group A: healthy subjects (eGFR >80 mL/min/1.73 m^2).
Group B: subjects with mild renal impairment (eGFR >50-80 mL/min/1.73 m^2).
Group C: subjects with moderate renal impairment (eGFR >30-50 mL/min/1.73 m^2).
Group D: subjects with severe renal impairment (eGFR ≤30 mL/min/1.73 m^2).
Group E: subjects with ESRD on hemodialysis.

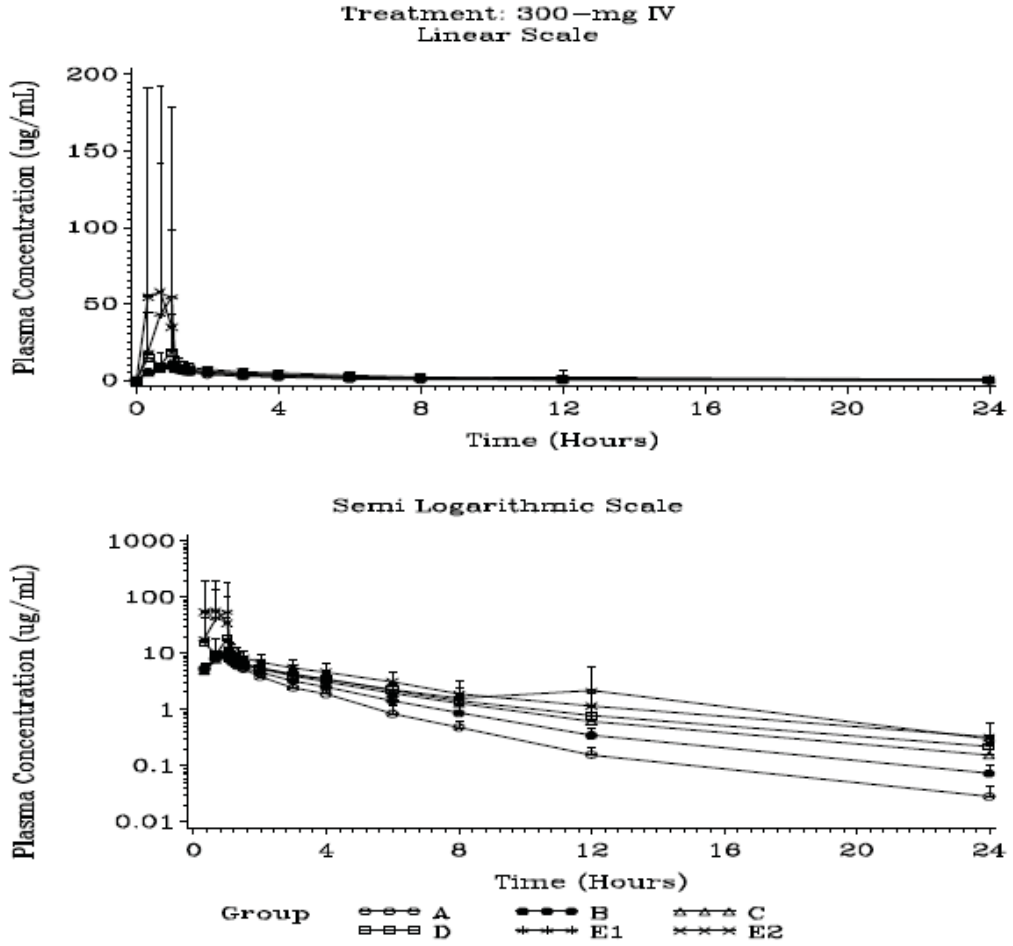
With the exception of race and renal status, overall mean demographic characteristics were similar across the groups. The majority of subjects were white (21 of 44 subjects; 47.7%) or black (21 of 44 subjects; 47.7%). More male subjects (25 of 44 subjects; 56.8%) were enrolled than female subjects (19 of 44 subjects; 43.2%). Subjects ranged in age from 26 to 71 years and had a mean BMI of 29.87 kg/m^2. No infusion was discontinued in any subject.

Pharmacokinetics:

Plasma:

Mean (±SD) plasma concentrations of delafloxacin and Captisol versus time on linear and semilogarithmic scales are presented in Figure 1 and Figure 2, respectively.

Figure 1. Mean (±SD) Plasma Concentrations of Delafloxacin Versus Time (Linear and Semilogarithmic Scales) (Pharmacokinetic Population) (*Adapted from Figure 11-1 in the study report*)



Abbreviations: 300-mg IV, 300-mg delafloxacin/Captisol IV; IV, intravenous.

Note: For re-enrolled subjects, data collected from the original enrollment were not included in this figure.

Group A: healthy subjects (estimated glomerular filtration rate [eGFR] >80 mL/min/1.73 m^2).

Group B: subjects with mild renal impairment (eGFR >50-80 mL/min/1.73 m^2).

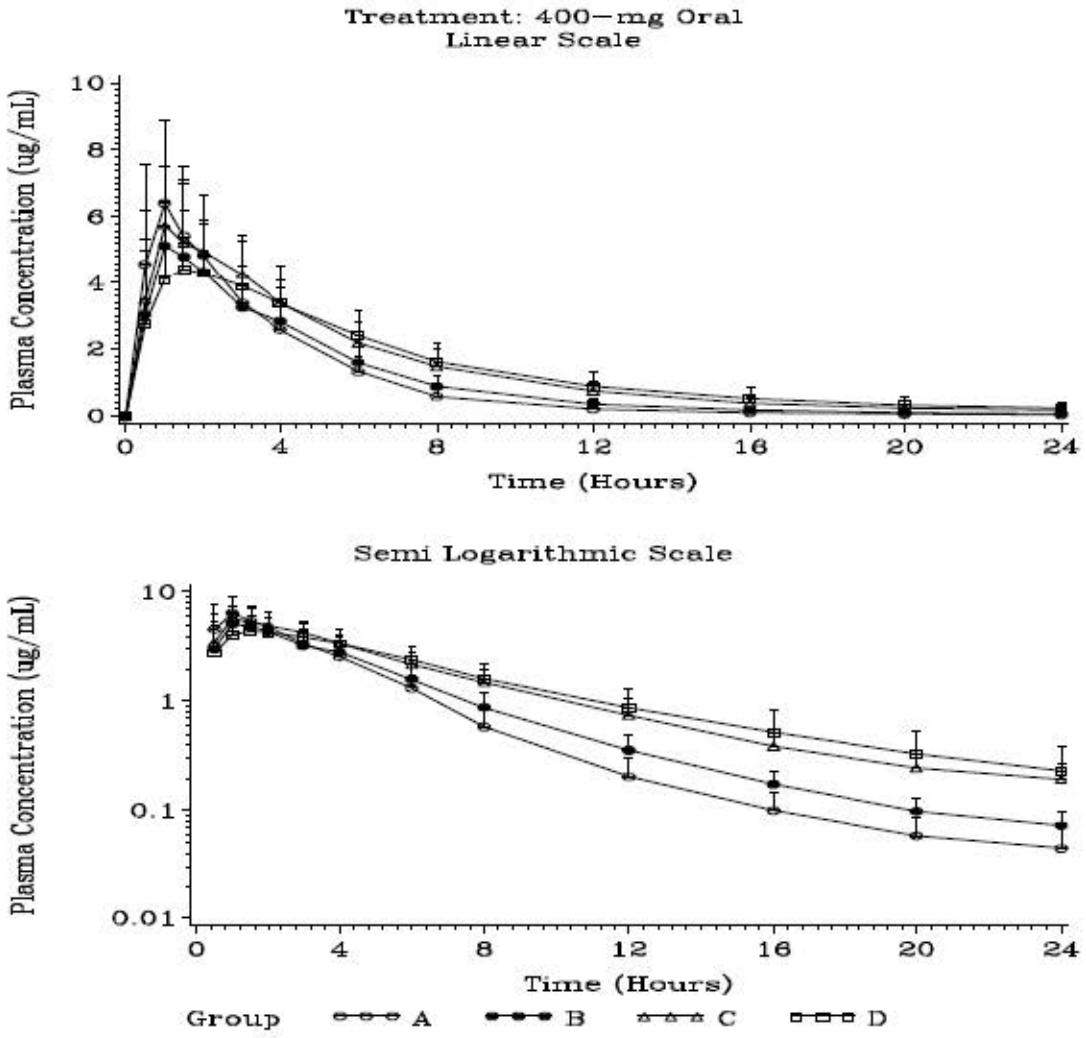
Group C: subjects with moderate renal impairment (eGFR >30-50 mL/min/1.73 m^2).

Group D: subjects with severe renal impairment (eGFR ≤30 mL/min/1.73 m^2).

Group E1: subjects with end-stage renal disease on hemodialysis; 300-mg delafloxacin/Captisol IV starting 1 hour before initiation of the hemodialysis.

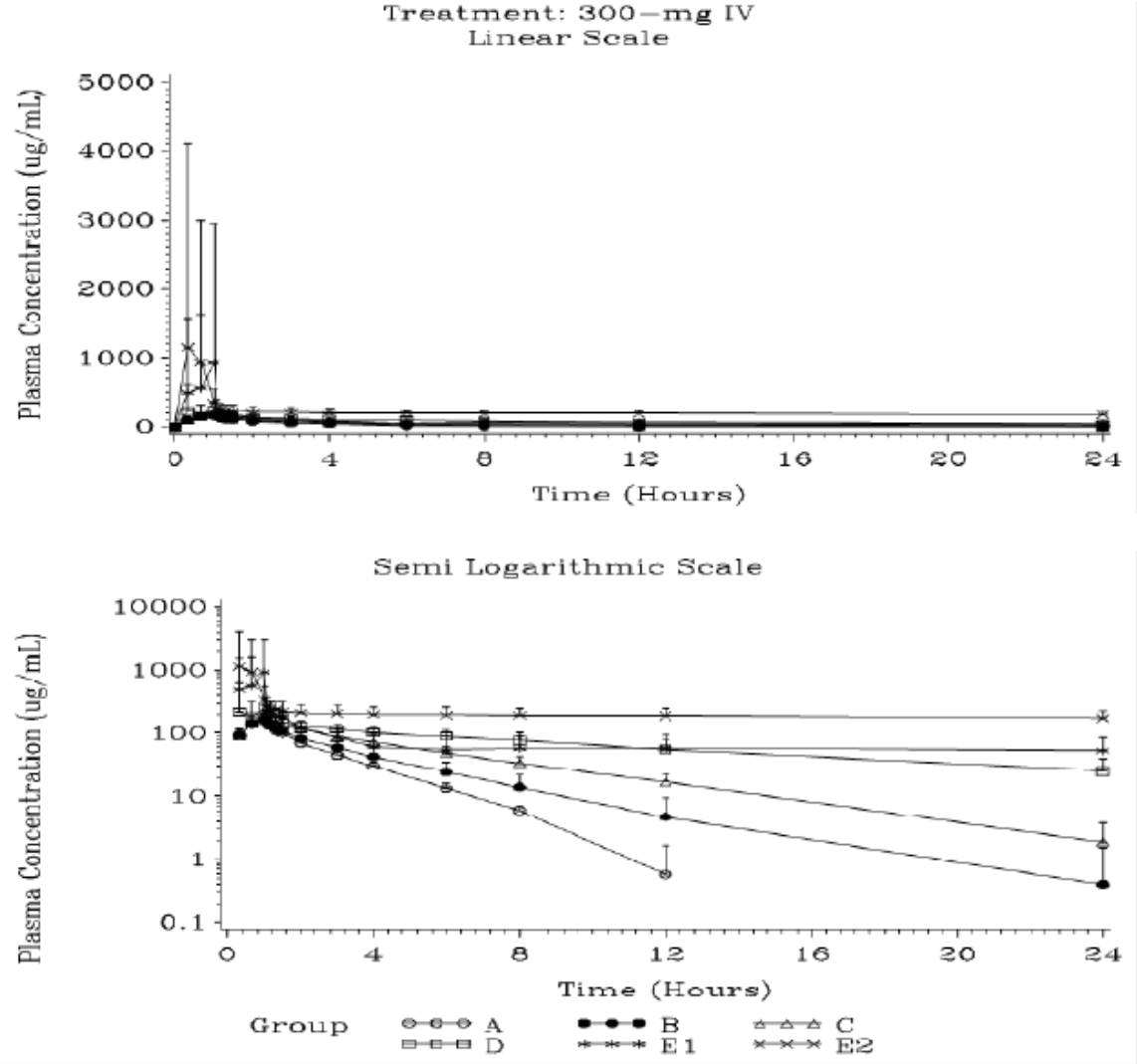
Group E2: subjects with end-stage renal disease on hemodialysis; 300-mg delafloxacin/Captisol IV starting within 1 hour after completion of the hemodialysis.

Figure 1 (continue).



Abbreviation: 400-mg Oral, 400-mg oral delafloxacin (four 100-mg capsules).
Note: For re-enrolled subjects, data collected from the original enrollment were not included in this figure.
Group A: healthy subjects (estimated glomerular filtration rate [eGFR] >80 mL/min/1.73 m^2).
Group B: subjects with mild renal impairment (eGFR >50-80 mL/min/1.73 m^2).
Group C: subjects with moderate renal impairment (eGFR >30-50 mL/min/1.73 m^2).
Group D: subjects with severe renal impairment (eGFR ≤30 mL/min/1.73 m^2).

Figure 2. Mean (±SD) Plasma Concentrations of Captisol Versus Time (Linear and Semilogarithmic Scales) (Pharmacokinetic Population) (*Adapted from Figure 11-2 in the study report*)



Abbreviations: 300-mg IV, 300-mg delafloxacin/Captisol IV; IV, intravenous.
Note: For re-enrolled subjects, data collected from the original enrollment were not included in this figure.
Group A: healthy subjects (estimated glomerular filtration rate [eGFR] >80 mL/min/1.73 m^2).
Group B: subjects with mild renal impairment (eGFR >50-80 mL/min/1.73 m^2).
Group C: subjects with moderate renal impairment (eGFR >30-50 mL/min/1.73 m^2).
Group D: subjects with severe renal impairment (eGFR ≤30 mL/min/1.73 m^2).
Group E1: subjects with end-stage renal disease on hemodialysis; 300-mg delafloxacin/Captisol IV starting 1 hour before initiation of the hemodialysis.
Group E2: subjects with end-stage renal disease on hemodialysis; 300-mg delafloxacin/Captisol IV starting within 1 hour after completion of the hemodialysis.

The arithmetic means and CVs for plasma PK parameters of delafloxacin are presented in Table 2.

Table 2. Mean (CV) Plasma Pharmacokinetic Parameters of Delafloxacin (Pharmacokinetic Population)

Parameter (unit)	Group[a] A Normal Renal Function (n = 8)	Group B Mild Renal Impairment (n = 8)	Group C Moderate Renal Impairment (n = 8)	Group D Severe Renal Impairment (n = 8)	Group E1 ESRD (n = 8)	Group E2 ESRD (n = 8)
300-mg IV						
AUC_{0-t} (µg•h/mL)	23.64 (23.2)	31.04 (19.3)	39.29 (26.3)	49.56 (41.3)	83.29 (120.7)	96.74 (104.9)
AUC_{0-inf} (µg•h/mL)	22.57[b] (20.0)	31.30 (19.0)	38.41[c] (27.9)	51.07 (40.9)	84.31 (119.2)	97.46 (104.2)
C_{max} (µg/mL)	9.28 (25.3)	9.80 (11.1)	9.86 (25.6)	19.53 (140.0)	54.40 (228.5)	59.85 (225.0)
T_{max}[d] (h)	1.00 (0.66, 1.00)	1.00 (1.00, 1.08)	1.00 (1.00, 1.08)	1.00 (0.33, 1.10)	1.00 (1.00, 1.17)	1.00 (0.33, 1.02)
$t_{1/2}$ (h)	9.28[b] (46.7)	10.71 (22.90)	8.90[c] (33.5)	14.15 (42.5)	10.57 (47.4)	15.02 (14.3)
CL (L/h)	13.72[b] (19.1)	9.92 (20.3)	8.25[c] (22.9)	6.59 (31.5)	6.58 (66.0)	5.09 (57.9)
CL_u (L/h)	2.19[b] (23.1)	1.87 (28.0)	1.44[c] (30.5)	1.30 (28.1)	1.70 (75.5)	1.30 (66.8)
CL_{NR} (L/h)	7.37[b] (25.6)	6.95 (23.5)	6.90[c] (26.5)	6.19 (30.8)	3.08[e] (70.0)	3.04[f] (21.8)
400-mg Oral						
AUC_{0-t} (µg•h/mL)	25.75 (29.3)	26.75 (31.4)	36.91 (18.8)	37.85 (26.9)	–	–
AUC_{0-inf} (µg•h/mL)	25.36[c] (31.6)	28.32[c] (28.9)	37.34 (18.8)	39.52 (27.9)	–	–
C_{max} (µg/mL)	7.16 (34.9)	5.67 (34.2)	6.00 (29.7)	5.35 (24.9)	–	–
T_{max}[d] (h)	1.00 (0.50, 1.50)	1.00 (0.50, 2.00)	1.00 (0.50, 3.00)	1.50 (0.50, 6.00)	–	–
$t_{1/2}$ (h)	15.37[c] (43.3)	12.47[c] (21.9)	10.49 (40.5)	15.46 (33.8)	–	–
CL/F (L/h)	17.63[c] (40.2)	15.92[c] (47.5)	11.04 (18.4)	10.78 (25.7)	–	–
CL_u/F (L/h)	2.87[c] (41.1)	2.97[c] (47.9)	1.93 (26.7)	2.19 (30.5)	–	–

Abbreviations: 300-mg IV, 300-mg delafloxacin/Captisol IV; 400-mg Oral, 400-mg oral delafloxacin (four 100-mg capsules); CV, coefficient of variation; ESRD, end-stage renal disease; IV, intravenous.

Note: For re-enrolled subjects, data collected from the original enrollment were not included in this table.

[a] Group A: healthy subjects (estimated glomerular filtration rate [eGFR] >80 mL/min/1.73 m^2).
Group B: subjects with mild renal impairment (eGFR >50-80 mL/min/1.73 m^2).
Group C: subjects with moderate renal impairment (eGFR >30-50 mL/min/1.73 m^2).
Group D: subjects with severe renal impairment (eGFR ≤30 mL/min/1.73 m^2).
Group E1: subjects with ESRD on hemodialysis; 300-mg delafloxacin/Captisol IV starting 1 hour before initiation of hemodialysis.
Group E2: subjects with ESRD on hemodialysis; 300-mg delafloxacin/Captisol IV starting within 1 hour after completion of the hemodialysis.

[b] n = 6.

[c] n = 7.

[d] For T_{max}, the median (minimum, maximum) values are presented.

[e] n = 3.

[f] n = 2.

After 300-mg delafloxacin/Captisol IV dosing, the arithmetic mean total exposure (AUC0-t) of delafloxacin increased consistently as the degree of renal impairment increased. Mean AUC0-t for the severe renal impairment group was 2.1-fold higher than the exposure observed for the healthy group. Mean AUC0-t for the ESRD group without hemodialysis after dosing (Group E2) was 4.1-fold higher than the exposure observed for the healthy group. Peak exposure (Cmax) was similar for the healthy group and the mild and moderate renal impairment groups. Arithmetic mean Cmax values for the severe renal impairment group and the ESRD group without hemodialysis after dosing (Group E2) were 2.1-fold and 6.4-fold higher, respectively, than the corresponding value observed for the healthy group. The t1/2 was also increased for the severe renal impairment group and the ESRD group without hemodialysis after dosing (Group E2). The mean $t_{1/2}$ was 9.3 hours for the healthy group and increased to 15.0 hours for the ESRD group without hemodialysis after dosing (Group E2).

Reviewer's Comment: After 300 mg delafloxacin/Captisol IV single dose,

- *The arithmetic mean ratios for AUC_{0-t} in patients with mild, moderate, severe renal impairment, ESRD patients with hemodialysis, and without hemodialysis compared to the control group (patients with normal renal function) were 1.31, 1.66, 2.10, 3.52, and 4.10, respectively.*
- *The mean AUC_{0-t} for the ESRD group after 4 hours of hemodialysis (83.29 µg*h/mL) was only slightly lower compared to the one without hemodialysis (96.74 µg*h/mL) but much higher than the one for patients with severe renal impairment (49.56 µg*h/mL). This observation indicated that the dialysis clearance (CLd) may not be high for delafloxacin. According to the Sponsor, based on the results from group E1, mean CLd = 4.21 L/hr. The mean fraction of administer delafloxacin recovered in the dialysate (Frem%0-4) was 19.2%.*
- *The arithmetic mean ratios for C_{max} in patients with mild, moderate, server renal impairment, patients with hemodialysis, and patients without hemodialysis compared to the control group (patients with normal renal function) were 1.06, 1.06, 2.10, 5.86 and 6.45, respectively.*
- *It was observed that higher variability in the PK parameter estimates in ESRD patients with and without hemodialysis. The high CV was partially due to one outlier (subject 0001507) in both E1 and E2 group (List 16.2.6.4 on page 2317-19 of this study report).*
- *The review team identified two outliers receiving 300 mg IV dose. The plasma concentrations of first three time points for Subjects 1404 (severe renal impairment) and 1507 (ESRD) were unexpectedly high with no identified reasons. Therefore, the reviewer re-analyzed AUC0-t by exclusion of these two subjects. Please refer to the table below for details. Based on the results after excluding the two subjects, the AUC values for the severe renal impairment group, ESRD with hemodialysis, and without hemodialysis, were 1.8, 2.1, and 2.6 fold higher, respectively, than the corresponding exposure in subjects with normal renal function after receiving 300 mg IV dose.*

Comparison of Fold Change in AUC and Cmax across Different Levels of Renal Impairment (Before vs. After Exclusion of the Two Subjects) for Subjects receiving 300 mg IV dose (Reviewer Analysis)

Variables	normal	mild	moderate	severe	ESRD w/ hemodialysis	ESRD w/o hemodialysis
AUC0-inf	1	1.38	1.7	2.26	3.77	4.3
AUC0-t	1	1.31	1.66	2.08	3.57	4.09
Cmax	1	1.06	1.06	2.01	5.86	6.44
After exclusion	•	•	•	•	•	•
AUC0-inf	1	1.38	1.7	1.99	2.23	2.78
AUC0-t	1	1.31	1.66	1.83	2.1	2.62
Cmax	1	1.06	1.06	1.06	1.12	1.32

After the 400-mg oral delafloxacin dose, the healthy group had nearly the same total exposure as observed following the 300-mg delafloxacin/Captisol IV dose. For the healthy group, the arithmetic mean AUC_{0-t} was 23.64 µg•h/mL after IV dosing and 25.75 µg•h/mL after oral dosing. Mean AUC_{0-t} values for the moderate and severe renal impairment groups were approximately 1.5-fold higher than the corresponding value for the healthy group; however, mean C_{max} varied little across the various renal function groups after oral dosing, with a slightly higher mean Cmax of 7.2 µg/mL for the healthy group.

Reviewer's Comment: After 400 mg delafloxacin single oral dose,

- *No ESRD patients with or without hemodialysis received oral dose of delafloxacin.*
- *The mean AUC_{0-t} after 400 mg oral delafloxacin single dose (25.75 µg*h/mL) is similar to the one after 300 mg IV delafloxacin single dose (23.64 µg*h/mL).*
- *The arithmetic mean ratios for AUC_{0-t} in patients with mild, moderate, server renal impairment, compared to the control group (patients with normal renal function) were 1.03, 1.43, and 1.47, respectively.*
- *The arithmetic mean ratios for C_{max} in patients with mild, moderate, and server renal impairment were 0.79, 0.83, and 0.74, respectively.*
- *Most inter-individual variability (CV%) in the estimated PK parameters after oral dose were around or under 30%.*

The plasma protein binding in different renal function groups were summarized in Table 3 below.

Table 3. Summary of Plasma Protein Binding of Delafloxacin PK Population *(Adapted from Table 14.2.3 in the study report)*

		Group				
Parameter	Statistics	A (N=8)	B (N=8)	C (N=8)	D (N=8)	E (N=8)
Fu (%)	n	8	8	8	8	8
	Mean	15.94	18.80	17.29	20.23	24.74
	SD	1.62	3.76	1.89	3.51	4.01
	CV%	10.2	20.0	10.9	17.4	16.2
	Geometric Mean	15.87	18.53	17.19	19.95	24.46
	Median	15.55	17.60	17.55	20.20	23.40
	Minimum	13.8	15.8	14.6	14.0	19.9
	Maximum	19.5	27.6	19.2	27.0	29.9

Note: For re-enrolled subjects, data collected from the original enrollment is not included in this table.
Fu = fraction unbound.
Group A: healthy subjects (eGFR >80 mL/min/1.73 m^2);
Group B: subjects with mild renal disease (eGFR >50-80 mL/min/1.73 m^2);
Group C: subjects with moderate renal disease (eGFR >30-50 mL/min/1.73 m^2);
Group D: subjects with severe renal disease (eGFR <=30 mL/min/1.73 m^2);
Group E: subjects with end-stage renal disease (ESRD) on hemodialysis.

For the healthy group and the mild, moderate, and severe renal impairment groups, protein binding ranged from 80% for the severe renal impairment group to 84% for the healthy group. The ESRD group showed the lowest percentage of bound delafloxacin (75%).

Reviewer's Comment: With the decrease of protein binding in severe renal impairment and ESRD patients, the percent of unbound fraction of delafloxacin increased to 20% and 25% in Group D and E, respectively. Delafloxacin is a moderately protein binding drug. The significance in the pharmacological effect of delafloxacin on decreasing of the protein binding may depend on its therapeutic index.

The mean (CV) plasma pharmacokinetic parameters of captisol were summarized in the Table 4 below.

Table 4. Mean (CV) Plasma Pharmacokinetic Parameters of Captisol (Pharmacokinetic Population)

Parameter (unit)	Group[a]					
	Group A Normal Renal Function (n = 8)	Group B Mild Renal Impairment (n = 8)	Group C Moderate Renal Impairment (n = 8)	Group D Severe Renal Impairment (n = 8)	Group E1 ESRD (n = 8)[b]	Group E2 ESRD (n = 8)
300-mg IV						
AUC_{0-t} (µg•h/mL)	374.55 (11.9)	493.39 (31.8)	815.87 (26.5)	1975.35 (30.7)	3183.89 (62.0)	11164.52 (31.7)
AUC_{0-inf} (µg•h/mL)	386.82 (11.6)	508.10 (31.5)	851.68 (25.4)	2083.13 (31.6)	–	29565.30[c] (.)
C_{max} (µg/mL)	176.88 (14.0)	167.13 (10.1)	190.75 (26.9)	322.63 (111.0)	946.38 (211.8)	1321.75 (217.9)
T_{max}[d] (h)	1.00 (1.00, 1.08)	1.00 (1.00, 1.08)	1.00 (1.00, 1.13)	1.00 (0.33, 1.10)	1.00 (1.00, 1.17)	1.05 (0.33, 1.17)
$t_{1/2}$ (h)	1.76 (8.5)	2.51 (33.0)	3.99 (19.3)	10.27 (29.1)	–	65.57[c] (.)
CL (L/h)	6.28 (12.5)	5.08 (26.0)	3.00 (27.3)	1.26 (30.1)	–	0.08[c] (.)
CL_{NR} (L/h)	0.92 (40.3)	0.42 (289.7)	0.09 (683.4)	0.11 (176.4)	–	–
Placebo IV						
AUC_{0-t} (µg•h/mL)	358.55 (13.6)	426.55 (20.1)	760.15 (27.3)	1878.17 (38.9)	–	–
AUC_{0-inf} (µg•h/mL)	371.12 (13.2)	446.44 (23.1)	802.50 (25.8)	1989.31 (40.4)	–	–
C_{max} (µg/mL)	173.50 (14.9)	160.50 (10.5)	181.25 (20.9)	190.63 (19.8)	–	–
T_{max}[d] (h)	1.00 (0.33, 1.00)	1.00 (1.00, 1.08)	1.00 (1.00, 1.08)	1.00 (1.00, 1.08)	–	–
$t_{1/2}$ (h)	1.74 (9.6)	2.38 (26.6)	3.96 (18.8)	9.69 (30.6)	–	–
CL (L/h)	6.57 (13.1)	5.60 (20.0)	3.15 (23.2)	1.38 (36.2)	–	–
CL_{NR} (L/h)	1.13 (102.1)	0.00[e] (0.0)	0.28 (202.7)	0.25 (108.8)	–	–

Abbreviations: 300-mg IV, 300-mg delafloxacin/Captisol IV; Placebo IV, placebo/Captisol IV; CV, coefficient of variation; ESRD, end-stage renal disease; IV, intravenous.

Note: For re-enrolled subjects, data collected from the original enrollment were not included in this table.

[a] Group A: healthy subjects (estimated glomerular filtration rate [eGFR] >80 mL/min/1.73 m^2).
Group B: subjects with mild renal impairment (eGFR >50-80 mL/min/1.73 m^2).
Group C: subjects with moderate renal impairment (eGFR >30-50 mL/min/1.73 m^2).
Group D: subjects with severe renal impairment (eGFR ≤30 mL/min/1.73 m^2).
Group E1: subjects with ESRD on hemodialysis; 300-mg delafloxacin/Captisol IV starting 1 hour before initiation of hemodialysis.
Group E2: subjects with ESRD on hemodialysis; 300-mg delafloxacin/Captisol IV starting within 1 hour after completion of the hemodialysis.

[b] The $t_{1/2}$ could not be estimated for the ESRD group. Several dependent parameters could not be generated.

[c] n = 1.

[d] For T_{max}, the median (minimum, maximum) values are presented.

[e] CL_{NR} for Captisol placebo Group B was –0.94 (–625.0). Values presented as zeros in the table.

The 300-mg IV dose of delafloxacin was delivered with 2400 mg of Captisol. Since Captisol plasma clearance is known to be reduced with impaired renal function, the primary interest in Captisol for this study was to assess any potential interaction with delafloxacin. Much greater total exposure of Captisol was observed in the severe renal impairment and ESRD groups than in the healthy group. Arithmetic mean AUC_{0-t} for the severe renal impairment group was 5-fold higher than the AUC_{0-t} of the healthy group while the increase for the ESRD group without hemodialysis after dosing (Group E2) was 30-fold higher. However, for the healthy group and the mild, moderate, and severe renal impairment groups that received both the delafloxacin/Captisol IV infusion and the placebo/Captisol IV infusion, the arithmetic mean AUC_{0-t} values appeared to be similar.

Reviewer's Comment:

- *For the potential interaction between captisol and delafloxacin:*
  - *It seems that delafloxacin did not change the exposure of captisol (i.e. The mean AUCs and C_{max} of Captisol with or without delafloxacin were comparable across different levels of renal impairment (Table 4).)*
  - *Captisol did not change the overall exposure of delafloxacin (i.e. The mean AUCs of delafloxacin in healthy volunteers were comparable for oral (no Captisol) and IV (with Captisol) formulations.*
- *The arithmetic mean ratios for AUC_{0-t} after a single dose of 2400 mg of Captisol in patients with mild, moderate, server renal impairment, and ESRD patients with or without hemodialysis compared to the control group (patients with normal renal function) were 1.19, 1.78, 5.24, 8.50, and 29.80, respectively.*
- *The C_{max} of Captisol in patients with severe renal impairment after a single 300 mg IV dose of delafloxacin appeared to be about 2 fold of the one in subjects with normal renal function. The observation may be due to one subject (ID0001404) whose Cmax was 1205.00 µg/mL, about 10 folds of others. However, all other PK parameters for this subject were similar to the ones in same group.*

Urine

The arithmetic means and CVs for urine PK parameters of delafloxacin and Captisol were summarized in Table 5 and Table 6, respectively.

Table 5. Mean (CV) Urine Pharmacokinetic Parameters of Delafloxacin (Pharmacokinetic Population) *(Adapted from Table 11-4 of the study report)*

	Group[a]			
Parameter (unit)	Group A Normal Renal Function (n = 8)	Group B Mild Renal Impairment (n = 8)	Group C Moderate Renal Impairment (n = 8)	Group D Severe Renal Impairment (n = 8)
300-mg IV				
Ae_{0-12} (mg)	131.3 (17.6)	75.7 (66.2)	43.8 (45.3)	14.0 (77.0)
Ae_{12-24} (mg)	2.7 (30.0)	5.6 (83.7)	3.2 (66.5)	1.7 (73.8)
Ae_{0-48} (mg)	135.9 (16.8)	84.2 (58.6)	49.2 (40.3)	16.6 (75.6)
$Fe\%_{0-48}$	45.3 (16.8)	28.1 (58.6)	16.4 (40.3)	5.5 (75.6)
CLr (L/h)	6.03 (26.8)	2.96 (72.7)	1.30 (49.0)	0.40 (90.2)
400-mg Oral				
Ae_{0-12} (mg)	108.6 (25.9)	66.5 (80.0)	32.1 (70.7)	8.7 (58.2)
Ae_{12-24} (mg)	4.2 (29.5)	6.3 (46.6)	4.8 (82.9)	1.9 (73.6)
Ae_{0-48} (mg)	116.2 (21.8)	75.5 (70.3)	38.5 (64.6)	11.5 (56.0)
$Fe\%_{0-48}$	29.1 (21.8)	18.9 (70.3)	9.6 (64.6)	2.9 (56.0)
CLr (L/h)	5.09 (46.3)	2.95 (54.2)	1.03 (57.4)	0.29 (41.4)

Abbreviations: 300-mg IV, 300-mg delafloxacin/Captisol IV; 400-mg Oral, 400-mg oral delafloxacin (four 100-mg capsules); CV, coefficient of variation; IV, intravenous.

Note: For re-enrolled subjects, data collected from the original enrollment were not included in this table. The end-stage renal disease group was not summarized in this table because the majority of subjects did not have urinary output.

[a] Group A: healthy subjects (estimated glomerular filtration rate [eGFR] >80 mL/min/1.73 m^2).
Group B: subjects with mild renal impairment (eGFR >50-80 mL/min/1.73 m^2).
Group C: subjects with moderate renal impairment (eGFR >30-50 mL/min/1.73 m^2).
Group D: subjects with severe renal impairment (eGFR ≤30 mL/min/1.73 m^2).

After the 300-mg delafloxacin/Captisol IV dosing, the fraction of the delafloxacin dose excreted in urine during the 48 hours after dosing declined from 45.3% for the healthy group to 5.5% for the severe renal impairment group. The vast majority of the urinary excretion of delafloxacin occurred in the first 12 hours after dosing. The CLr decreased from 6.03 L/h for the healthy group to 0.40 L/h for the severe renal impairment group. The CLr after oral dosing was similar to the values and pattern observed for the IV dosing and decreased from 5.09 L/h for the healthy group to 0.29 L/h for the severe renal impairment group. The ESRD groups were not included in Table 5 because the majority of those subjects did not have urinary output.

Table 6. Mean (CV) Urine Pharmacokinetic Parameters of Captisol (Pharmacokinetic Population) *(Adapted from Table 11-5 of the study report)*

Parameter (unit)	Group[a]			
	Group A Normal Renal Function (n = 8)	Group B Mild Renal Impairment (n = 8)	Group C Moderate Renal Impairment (n = 8)	Group D Severe Renal Impairment (n = 8)
300-mg IV				
Ae_{0-12} (mg)	1946 (9.0)	2012 (26.60)	2029 (25.8)	1357 (43.6)
Ae_{12-24} (mg)	24 (32.7)	113 (154.7)	191 (57.1)	424 (17.9)
Ae_{0-48} (mg)	1970 (8.7.)	2152 (24.5)	2250 (21.3)	1994 (23.8)
$Fe\%_{0-48}$	82.1 (8.7)	89.7 (24.5)	93.8 (21.3)	83.1 (23.8)
CLr (L/h)	5.36 (18.9)	4.65 (36.4)	2.91 (29.7)	1.14 (47.7)
Placebo IV				
Ae_{0-12} (mg)	1911 (19.4)	2020 (32.2)	1744 (28.2)	1126 (28.0)
Ae_{12-24} (mg)	22 (65.6)	944 (266.9)	251 (49.8)	486 (19.0)
Ae_{0-48} (mg)	1932 (19.4)	2983 (100.0)	2026 (23.4)	1857 (19.6)
$Fe\%_{0-48}$	80.5 (19.4)	124.3 (100.0)	84.4 (23.4)	77.4 (19.6)
CLr (L/h)	5.43 (20.4)	6.54 (78.5)	2.87 (36.2)	1.13 (44.7)

Abbreviations: 300-mg IV, 300-mg delafloxacin/Captisol IV; Placebo IV, placebo/Captisol IV; CV, coefficient of variation; IV, intravenous.

Note: For re-enrolled subjects, data collected from the original enrollment were not included in this table. The end-stage renal disease group was not summarized in this table because the majority of subjects did not have urinary output.

[a] Group A: healthy subjects (estimated glomerular filtration rate [eGFR] >80 mL/min/1.73 m^2).
Group B: subjects with mild renal impairment (eGFR >50-80 mL/min/1.73 m^2).
Group C: subjects with moderate renal impairment (eGFR >30-50 mL/min/1.73 m^2).
Group D: subjects with severe renal impairment (eGFR ≤30 mL/min/1.73 m^2).

Dialysate

The arithmetic means and CVs for dialysate PK parameters of delafloxacin and Captisol for subjects with ESRD receiving hemodialysis are presented in Table 7 and Table 8, respectively.

Table 7. Mean (CV) Dialysate Pharmacokinetic Parameters of Delafloxacin *(Adapted from Table 11-6 in the study report)*

Parameter (unit)	Group E1[a] ESRD (n = 8)
$Arem_{0-1}$ (mg)	25.2 (18.5)
$Arem_{1-2}$ (mg)	16.5 (19.8)
$Arem_{2-3}$ (mg)	11.4 (32.7)
$Arem_{3-4}$ (mg)	7.2 (7.7)[b]
$Arem_{0-4}$ (mg)	57.6 (18.9)
CL_d (L/h)	4.21 (36.6)
$Frem\%_{0-4}$	19.2 (18.9)

Abbreviations: CV, coefficient of variation; ESRD, end-stage renal disease.
Note: For re-enrolled subjects, data collected from the original enrollment were not included in this table.
[a] Group E1: subjects with ESRD on hemodialysis; 300-mg delafloxacin/Captisol IV starting 1 hour before initiation of hemodialysis.
[b] n = 5.

Table 8. Mean (CV) Dialysate Pharmacokinetic Parameters of Captisol *(Adapted from Table 11-7 in the study report)*

Parameter (unit)	Group E1[a] ESRD (n = 8)
$Arem_{0-1}$ (mg)	624 (47.6)
$Arem_{1-2}$ (mg)	367 (38.6)
$Arem_{2-3}$ (mg)	254 (35.3)
$Arem_{3-4}$ (mg)	163 (33.0)[b]
$Arem_{0-4}$ (mg)	1347 (44.0)
CL_d (L/hr)	4.74 (53.8)
$Frem\%_{0-4}$ (%)	56.1 (44.0)

Abbreviations: CV, coefficient of variation; ESRD, end-stage renal disease.
Note: For re-enrolled subjects, data collected from the original enrollment were not included in this table.
[a] Group E1: subjects with ESRD on hemodialysis; 300-mg delafloxacin/Captisol IV starting 1 hour before initiation of hemodialysis.
[b] n = 5.

For delafloxacin, when hemodialysis occurred 1 hour after the delafloxacin/Captisol IV infusion in the ESRD group, the mean fraction of administered delafloxacin recovered in the dialysate (Frem%0-4) was 19.2%. Mean CLd was 4.21 L/h.

For Captisol, when hemodialysis occurred 1 hour after the delafloxacin/Captisol IV infusion in the ESRD group, the mean fraction of administered Captisol recovered in the dialysate (Frem%0-4) was 56.1%. Mean CLd was 4.74 L/h.

Statistical Analysis of Pharmacokinetic Parameters

Delafloxacin

Statistical analyses of the plasma PK parameters of delafloxacin based on renal classification by the MDRD and Cockroft-Gault equations are presented in Table 9 and Table 10, respectively.

Table 9. Statistical Analysis of Plasma Pharmacokinetic Parameters of Delafloxacin Based on Renal Classification by the MDRD Equation (Pharmacokinetic Population) *(Adapted from Table 11-8 from the study report)*

Parameter (Unit)	Group[a]	N	Geometric LS Means	Ratio of Geometric LS Means (B/A or C/A or D/A or E2/A)	90% CI of the Ratio
300-mg IV					
AUC_{0-t} (µg•h/mL)	A	8	23.12		
	B	8	30.51	1.320	(0.930, 1.873)
	C	8	38.20	1.652	(1.165, 2.345)
	D	8	46.57	2.014	(1.420, 2.858)
	E2	8	71.10	3.076	(2.168, 4.364)
AUC_{0-inf} (µg•h/mL)	A	8	22.21		
	B	8	30.78	1.386	(0.940, 2.044)
	C	8	37.30	1.680	(1.126, 2.506)
	D	8	47.98	2.161	(1.465, 3.186)
	E2	8	71.84	3.235	(2.194, 4.770)
C_{max} (µg/mL)	A	8	9.05		
	B	8	9.75	1.078	(0.597, 1.944)
	C	8	9.60	1.062	(0.588, 1.915)
	D	8	12.75	1.410	(0.781, 2.543)
	E2	8	17.98	1.988	(1.102, 3.587)
400-mg Oral					
AUC_{0-t} (µg•h/mL)	A	8	24.58		
	B	8	25.32	1.030	(0.799, 1.329)
	C	8	36.36	1.480	(1.147, 1.908)
	D	8	36.78	1.497	(1.161, 1.930)
AUC_{0-inf} (µg•hr/mL)	A	8	24.09		
	B	8	26.94	1.118	(0.850, 1.472)
	C	8	36.77	1.526	(1.170, 1.991)
	D	8	38.27	1.588	(1.218, 2.072)
C_{max} (µg/mL)	A	8	6.69		
	B	8	5.39	0.806	(0.602, 1.079)
	C	8	5.74	0.858	(0.641, 1.148)
	D	8	5.21	0.779	(0.582, 1.043)

Abbreviations: 300-mg IV, 300-mg delafloxacin/Captisol IV; 400-mg Oral, 400-mg oral delafloxacin (four 100-mg capsules); CI, confidence interval; IV, intravenous; LS, least squares; MDRD, modification of diet in renal disease.

Note: For re-enrolled subjects, data collected from the original enrollment were not included in this table. An analysis of variance with group as a fixed effect was performed on the natural log-transformed parameters AUC_{0-t}, AUC_{0-inf}, and C_{max}.

[a] Group A: healthy subjects (estimated glomerular filtration rate [eGFR] >80 mL/min/1.73 m^2).
Group B: subjects with mild renal impairment (eGFR >50-80 mL/min/1.73 m^2).
Group C: subjects with moderate renal impairment (eGFR >30-50 mL/min/1.73 m^2).
Group D: subjects with severe renal impairment (eGFR ≤30 mL/min/1.73 m^2).
Group E2: subjects with end-stage renal disease on hemodialysis; 300-mg delafloxacin/Captisol IV starting within 1 hour after completion of the hemodialysis.

Table 10. Statistical Analysis of Plasma Pharmacokinetic Parameters of Delafloxacin Based on Renal Classification by the Cockroft-Gault Equation (Pharmacokinetic Population) *(Adapted from Table 11-9 from the study report)*

Parameter (Unit)	Group[a]	N	Geometric LS Means	Ratio of Geometric LS Means (B/A or C/A or D/A or E2/A)	90% CI of the Ratio
300-mg IV					
AUC_{0-t} (µg•h/mL)	A	14	25.55		
	B	7	34.97	1.369	(0.991, 1.890)
	C	8	48.39	1.894	(1.390, 2.580)
	D	3	39.87	1.560	(1.001, 2.432)
	E2	8	71.10	2.783	(2.043, 3.791)
AUC_{0-inf} (µg•h/mL)	A	12	25.57		
	B	6	33.37	1.305	(0.912, 1.866)
	C	8	49.79	1.947	(1.404, 2.699)
	D	3	40.60	1.587	(1.000, 2.520)
	E2	8	71.84	2.809	(2.026, 3.894)
C_{max} (µg/mL)	A	14	9.22		
	B	7	9.55	1.036	(0.600, 1.788)
	C	8	12.82	1.391	(0.825, 2.345)
	D	3	10.29	1.116	(0.527, 2.364)
	E2	8	17.98	1.951	(1.157, 3.290)
400-mg Oral					
AUC_{0-t} (µg•h/mL)	A	14	24.45		
	B	7	32.73	1.338	(1.060, 1.690)
	C	8	38.84	1.589	(1.271, 1.986)
	D	3	34.36	1.405	(1.020, 1.936)
AUC_{0-inf} (µg•h/mL)	A	12	24.93		
	B	7	33.14	1.329	(1.041, 1.697)
	C	8	39.75	1.594	(1.261, 2.016)
	D	3	36.17	1.451	(1.041, 2.022)
C_{max} (µg/mL)	A	14	6.13		
	B	7	5.07	0.826	(0.628, 1.087)
	C	8	5.77	0.940	(0.723, 1.223)
	D	3	5.45	0.888	(0.608, 1.295)

Abbreviations: 300-mg IV, 300-mg delafloxacin/Captisol IV; 400-mg Oral, 400-mg oral delafloxacin (four 100-mg capsules); CI, confidence interval; IV, intravenous; LS, least squares.
Note: For re-enrolled subjects, data collected from the original enrollment were not included in this table. An analysis of variance with group as a fixed effect was performed on the natural log-transformed parameters AUC_{0-t}, AUC_{0-inf}, and C_{max}. For the analysis in this table, subjects were re-assigned to renal groups based on the Cockroft-Gault Equation as follows:

[a] Group A: healthy subjects (creatinine clearance [CLcr] >80 mL/min).
Group B: subjects with mild renal impairment (CLcr >50-80 mL/min).
Group C: subjects with moderate renal impairment (CLcr >30-50 mL/min).
Group D: subjects with severe renal impairment (CLcr ≤30 mL/min).
Group E2: subjects with end-stage renal disease on hemodialysis; 300-mg delafloxacin/Captisol IV starting within 1 hour after completion of the hemodialysis.

The Sponsor stated that since subjects were enrolled in this study based on their renal function as measured by the MDRD method, assignment of renal status by Cockcroft-Gault method resulted in more subjects in the healthy group and fewer in the severe renal impairment group.

Reviewer's Comment: Although there are some discrepancies in the number of subjects in each renal function group due to the different methods used during enrollment for renal function estimation, the overall trend, increasing delafloxacin plasma exposure with decreasing renal function, was observed by using both renal function estimation methods.

It was also noticed that the renal function for patients in two Phase 2 and two Phase 3 clinical studies was measured by the Cockcroft-Gault method when enrolled.

Captisol

Statistical analyses of the plasma PK parameters of Captisol based on renal classification by the MDRD and Cockroft-Gault equations are summarized in Table 11 and Table 12, respectively.

Table 11. Statistical Analysis of Plasma Pharmacokinetic Parameters of Captisol Based on Renal Classification by the MDRD Equation (Pharmacokinetic Population) (*(Adapted from Table 11-10 from the study report)*

Parameter (Unit)	Group[a]	N	Geometric LS Means	Ratio of Geometric LS Means (B/A or C/A or D/A or E2/A)	90% CI of the Ratio
300-mg IV					
AUC_{0-t} (µg•h/mL)	A	8	372.1		
	B	8	474.4	1.275	(1.011, 1.608)
	C	8	790.1	2.123	(1.683, 2.678)
	D	8	1896.8	5.097	(4.040, 6.430)
	E2	8	10653.3	28.627	(22.692, 36.114)
AUC_{0-inf} (µg•h/mL)	A	8	384.5		
	B	8	489.0	1.272	(1.021, 1.585)
	C	8	826.4	2.149	(1.725, 2.678)
	D	8	1994.9	5.189	(4.164, 6.465)
	E2	8	29565.3	76.902	(48.230, 122.618)
C_{max} (µg/mL)	A	8	175.3		
	B	8	166.4	0.949	(0.552, 1.630)
	C	8	184.5	1.053	(0.613, 1.808)
	D	8	243.6	1.390	(0.809, 2.387)
	E2	8	445.2	2.540	(1.478, 4.363)
Placebo IV					
AUC_{0-t} (µg•h/mL)	A	8	355.7		
	B	8	419.4	1.179	(0.949, 1.466)
	C	8	738.3	2.076	(1.670, 2.580)
	D	8	1762.4	4.955	(3.986, 6.159)
AUC_{0-inf} (µg•h/mL)	A	8	368.3		
	B	8	437.0	1.187	(0.948, 1.485)
	C	8	781.0	2.120	(1.695, 2.653)
	D	8	1857.7	5.044	(4.031, 6.311)
C_{max} (µg/mL)	A	8	171.8		
	B	8	159.8	0.930	(0.807, 1.071)
	C	8	177.8	1.035	(0.899, 1.192)
	D	8	187.7	1.092	(0.948, 1.258)

Abbreviations: 300-mg IV, 300-mg delafloxacin/Captisol IV; Placebo IV, placebo/Captisol IV; CI, confidence interval; IV, intravenous; LS, least squares; MDRD, modification of diet in renal disease.
Note: For re-enrolled subjects, data collected from the original enrollment were not included in this table. An analysis of variance with group as a fixed effect was performed on the natural log-transformed parameters AUC_{0-t}, AUC_{0-inf}, and C_{max}.

[a] Group A: healthy subjects (estimated glomerular filtration rate [eGFR] >80 mL/min/1.73 m^2).
Group B: subjects with mild renal impairment (eGFR >50-80 mL/min/1.73 m^2).
Group C: subjects with moderate renal impairment (eGFR >30-50 mL/min/1.73 m^2).
Group D: subjects with severe renal impairment (eGFR ≤30 mL/min/1.73 m^2).
Group E2: subjects with end-stage renal disease on hemodialysis; 300-mg delafloxacin/Captisol IV starting within 1 hour after completion of the hemodialysis.

Table 12. Statistical Analysis of Plasma Pharmacokinetic Parameters of Captisol Based on Renal Classification by the Cockroft-Gault Equation (Pharmacokinetic Population) *(Adapted from Table 11-11 from the study report)*

Parameter (Unit)	Group[a]	N	Geometric LS Means	Ratio of Geometric LS Means (B/A or C/A or D/A or E2/A)	90% CI of the Ratio
300-mg IV					
AUC_{0-t} (µg•h/mL)	A	14	401.5		
	B	7	641.6	1.598	(1.323, 1.930)
	C	8	1332.1	3.318	(2.769, 3.975)
	D	3	2673.9	6.659	(5.137, 8.632)
	E2	8	10653.3	26.532	(22.145, 31.788)
AUC_{0-inf} (µg•h/mL)	A	14	414.3		
	B	7	675.0	1.629	(1.372, 1.934)
	C	8	1380.5	3.332	(2.827, 3.926)
	D	3	2843.2	6.862	(5.422, 8.685)
	E2	8	29565.3	71.355	(48.636, 104.686)
C_{max} (µg/mL)	A	14	168.7		
	B	7	172.3	1.021	(0.620, 1.682)
	C	8	243.3	1.442	(0.894, 2.325)
	D	3	218.3	1.294	(0.652, 2.568)
	E2	8	445.2	2.639	(1.637, 4.254)
Placebo IV					
AUC_{0-t} (µg•h/mL)	A	14	373.0		
	B	7	587.9	1.576	(1.352, 1.838)
	C	8	1210.6	3.246	(2.802, 3.760)
	D	3	2615.0	7.011	(5.677, 8.660)
AUC_{0-inf} (µg•h/mL)	A	14	388.1		
	B	7	617.4	1.591	(1.353, 1.871)
	C	8	1271.0	3.275	(2.804, 3.825)
	D	3	2785.0	7.176	(5.743, 8.967)
C_{max} (µg/mL)	A	14	164.5		
	B	7	163.9	0.996	(0.886, 1.120)
	C	8	185.5	1.128	(1.008, 1.262)
	D	3	219.0	1.332	(1.134, 1.565)

Abbreviations: 300-mg IV, 300-mg delafloxacin/Captisol IV; Placebo IV, placebo/Captisol IV; CI, confidence interval; IV, intravenous; LS, least squares.

Note: For re-enrolled subjects, data collected from the original enrollment were not included in this table. An analysis of variance with group as a fixed effect was performed on the natural log-transformed parameters AUC_{0-t}, AUC_{0-inf}, and C_{max}. For the analysis in this table, subjects were re-assigned to renal groups based on the Cockroft-Gault Equation as follows:

[a] Group A: healthy subjects (creatinine clearance [CLcr] >80 mL/min).
Group B: subjects with mild renal impairment (CLcr >50-80 mL/min).
Group C: subjects with moderate renal impairment (CLcr >30-50 mL/min).
Group D: subjects with severe renal impairment (CLcr ≤30 mL/min).
Group E2: subjects with end-stage renal disease on hemodialysis; 300-mg delafloxacin/Captisol IV starting within 1 hour after completion of the hemodialysis.

Reviewer's Comment: Although there are some discrepancies in the number of subjects in each renal function group due to the different methods used during enrollment for renal function

estimation, the overall trend, increasing Captisol plasma exposure with decreasing renal function, was observed by using both renal function estimation methods.

Safety

- For subjects excluded from PK analyses:
    - Subject 0001102 (healthy group) received the 300-mg delafloxacin/Captisol IV infusion in Period 1 and the placebo/Captisol IV infusion in Period 2. The subject was discontinued from the study on Day 4 of Period 2, due to a severe TEAE of C difficile colitis that was considered unrelated to study drug by the investigator.
    - Subject 0001406 (severe renal impairment group) received a single, 300-mg delafloxacin/Captisol IV infusion in Period 1. The subject's participation in Period 2 was delayed due to follow up of a moderate bursitis TEAE. The subject was discontinued from the study approximately 18 days later because of a moderate TEAE of musculoskeletal pain in the left shoulder that was considered unrelated to study drug by the investigator.

- Overall, 83 TEAEs were reported and 28 of 44 subjects (63.6%) experienced at least 1 TEAE after receiving at least 1 dose of study drug. Among the 83 TEAEs, one was a severe TEAE and 31 were moderate. All others were mild severity. The severe TEAE was *C difficile* colitis in 1 subject (Subject 0001102). This subject was in normal renal function group.
- Treatment-emergent AEs were reported by 2 of 9 subjects (22.2%) in the healthy group, 5 of 8 subjects (62.5%) in the mild renal impairment group, 3 of 8 subjects (37.5%) in the moderate renal impairment group, and 5 of 9 subjects (55.6%) in the severe renal impairment group after the 300-mg delafloxacin/Captisol IV infusion. For the ESRD group, TEAEs were reported by 8 of 10 subjects (80.0%) when the 300-mg delafloxacin/Captisol IV infusion was administered 1 hour before hemodialysis and by 7 of 10 subjects (70.0%) when administered 1 hour after hemodialysis.
- Treatment-emergent AEs were reported by 1 of 9 subjects (11.1%) in the healthy group, 4 of 8 subjects (50.0%) in the mild renal impairment group, 3 of 8 subjects (37.5%) in the moderate renal impairment group, and no subjects in the severe renal impairment group after the placebo/Captisol IV infusion.
- Treatment-emergent AEs were reported by no subjects in the healthy and mild renal impairment groups, 3 of 8 subjects (37.5%) in the moderate renal impairment group, and 2 of 8 subjects (25.0%) in the severe renal impairment group after 400-mg oral delafloxacin.

SPONSOR'S CONCLUSIONS

Pharmacokinetics:

- Total delafloxacin exposure in plasma consistently increased with decreasing renal function. After a 1-hour 300-mg delafloxacin/Captisol infusion, arithmetic mean AUC0-t values for the severe renal impairment group and the ESRD group were 2.1-fold and 4.1-fold higher, respectively, than the corresponding exposure observed for the healthy group.

- For the IV dosing of delafloxacin, peak exposure increased for the severe renal impairment group and the ESRD group. Arithmetic mean Cmax values were 2.1-fold and 6.4-fold higher, respectively, than the corresponding value observed for the healthy group.
- For the oral dosing of delafloxacin, peak exposure varied modestly across the various renal function groups, with a slightly higher mean C_{max} of 7.2 μg/mL for the healthy group.
- The delafloxacin $t_{1/2}$ was longer only for the groups with severe renal impairment and ESRD. The mean $t_{1/2}$ following IV dosing of delafloxacin was 9.3 hours for the healthy group and increased to 15.0 hours for the ESRD group.
- For healthy subjects, the 300-mg delafloxacin/Captisol IV dose and the 400-mg oral delafloxacin dose had a similar total exposure of delafloxacin.
- Protein binding of delafloxacin ranged from 80% for the severe renal impairment group to 84% for the healthy group, while the ESRD group showed the lowest percentage of bound delafloxacin (75%).
- Following either IV or oral dosing, the CLr of delafloxacin decreased with decreasing renal function. For IV dosing, the CLr decreased from 6.03 L/h for the healthy group to 0.40 L/h for the severe renal impairment group.
- When hemodialysis occurred 1 hour after the delafloxacin/Captisol IV infusion in the ESRD group, the mean CLd was 4.21 L/h, and the mean fraction of administered delafloxacin recovered in the dialysate (Frem%0-4) was 19.2%.
- For the IV administration, delafloxacin clearance and CLcr were related by the following linear regression equation: CL (L/h) = 4.151 + 0.072 × CLcr (mL/min), which had an R2 = 0.666.
- Delafloxacin did not significantly affect the total or peak plasma exposure of Captisol.

Safety

- Overall, 300-mg delafloxacin/Captisol IV infusion, placebo/Captisol IV infusion, and 400-mg oral delafloxacin were safe and well tolerated by the healthy subjects and renally impaired subjects in this study.
- At least 1 TEAE was reported by 63.6% of subjects. In subjects who reported treatment-related TEAEs, the majority had TEAEs that were considered mild and possibly related to study drug.
- The most commonly reported TEAEs were diarrhea and headache with 7 subjects (15.9%) reporting each across all groups.
- Two subjects (4.5%) were discontinued due to TEAEs that were considered unrelated to study drug. Subject 0001102 (healthy group) was discontinued due to a severe, unrelated TEAE of C difficile colitis and Subject 0001406 (severe renal impairment group) was discontinued due to a moderate, unrelated TEAE of musculoskeletal pain in the left shoulder.
- There were no deaths or SAEs reported during this study.
- With the exception of 2 clinical laboratory values (elevated lipase and potassium) and 1 physical examination finding (lower abdomen tenderness) considered clinically significant during this study, there were no clinically significant findings noted resulting from clinical laboratory assessments, vital sign measurements, ECG results, or physical examination findings.

REVIEWER'S ASSESSMENT

- According to the FDA Draft Guidance for Industry: Pharmacokinetics in Patients with Impaired Renal Function — Study Design, Data Analysis, and Impact on Dosing and Labeling[4], there are two commonly used serum-creatinine based equations used to estimate renal function. One is Cockcroft-Gault (C-G) equation to estimate creatinine clearance (Clcr) by using unit of mL/min. The other one is Modification of Diet in Renal Disease (MDRD) to estimate glomerular filtration rate (eGFR) by using the unit of mL/min/1.73m^2). The Sponsor did not specify the method to estimate renal function in their proposed label. However, based on the unit used for renal function estimation in the label (eGFR), it seems that they refer to MDRD equation.
- In the draft label, the Sponsor proposed "Dosing in patients with severe renal impairment (b) (4) (b) (4) should be decreased to 200 mg IV (b) (4)
    - No ESRD patients were studied in Phase 2 and Phase 3 studies.
    - No patients with severe renal impairment received 200 mg IV dose in this submission.
    - The dose selection in patients with renal impairment will depend on the simulated delafloxacin exposure based on the population PK model in patients with renal impairment, the safety and efficacy margin for delafloxacin and the safety margin for SBECD.

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[4] http://www.fda.gov/downloads/Drugs/GuidanceComplianceRegulatoryInformation/Guidances/UCM204959.pdf

4.5.14 Study No.: RX-3341-115

A PHASE 1, OPEN-LABEL, RANDOMIZED, SINGLE-DOSE CROSSOVER STUDY TO DETERMINE THE PHARMACOKINETICS AND RELATIVE BIOAVAILABILITY OF INTRAVENOUS AND ORAL FORMULATIONS OF DELAFLOXACIN IN HEALTHY SUBJECTS

Date(s): 29 July 2013 to 06 September 2013
Sponsor: Melinta Therapeutics, Inc, 300 George Street, Suite 301, New Haven, CT 06511
Clinical Site: Single site: PPD Phase I Clinic, located at 7551 Metro Center Drive, Suite 200, Austin, TX 78744
Analytical Site: (b) (4)

OBJECTIVE(S):

- The primary objective of this study was to demonstrate bioavailability of oral delafloxacin (450-mg tablet) relative to intravenous (IV) delafloxacin (300 mg infused over 1 hour) in healthy subjects.
- The secondary objectives were:
  - To evaluate the relative safety and tolerability of 2 formulations of delafloxacin in healthy adult subjects;
  - To determine the exposure of a 900-mg oral dose in healthy adult subjects

METHODS

Study Design: This study consisted of 2 parts. Part 1 was a Phase 1, single-dose, open-label, randomized, 2-period, 2-sequence crossover study in 56 healthy subjects. Part 2 was a Phase 1 single-dose, open-label, 1-period study in 20 healthy subjects.

Part 1

In Part 1, 56 subjects who met all of the eligibility criteria were randomly assigned in a 1:1 ratio to a single dose of oral delafloxacin (450-mg tablet; Treatment A) and IV delafloxacin (300 mg infused over 1 hour; Treatment B) in 1 of 2 treatment sequences. On Day 1, subjects received a single dose of the assigned study drug after an overnight fasting period of at least 8 hours. Subjects continued fasting for at least 4 hours after study drug administration. In Period 2, subjects crossed over to receive a single dose of the alternate assigned formulation of study drug under fasting conditions. There was a minimum 7-day washout interval between doses in each period.

Part 2

In Part 2, 20 subjects who met all of the eligibility criteria received a single 900-mg oral dose of delafloxacin (two 450-mg tablets) on Day 1.

For both Parts 1 and 2, serial blood samples for the pharmacokinetic analysis of delafloxacin were collected up to 72 hours after each dose of study drug (Periods 1 and 2).

Drug Product: Delafloxacin 450 mg tablet (to be marketed oral formulation), Lot # 13DE009A; Delafloxacin 300 mg IV injection (to be marketed IV formulation), Lot# 12DEL1

PK Sample Collection:
For Part 1, serial blood samples for the determination of plasma concentrations of delafloxacin were collected before dosing and at 0.25, 0.5, 0.75, 1 (end of infusion for IV dosing and 1 hour after administration for oral dosing), 1.25, 1.5, 2, 3, 4, 6, 8, 12, 16, 20, 24, 30, 36, 48, and 72 hours after dosing in each period (Periods 1 and 2)**.**

For Part 2, serial blood samples for the determination of plasma concentrations of delafloxacin were collected before dosing and at 0.25, 0.5, 0.75, 1, 1.25, 1.5, 2, 3, 4, 6, 8, 12, 16, 20, 24, 30, 36, 48, and 72 hours after dosing.

Analytical Methods:
Project samples were analyzed according to (b) (4) Method LCMSC 634, entitled "Quantitation of Delafloxacin in Human Plasma via HPLC with MS/MS Detection," which was validated under Project Code "OZZ2." All samples were analyzed within the 150 days demonstrated long-term storage stability in human plasma containing dipotassium EDTA at -20 °C or colder.

Table 1. Summary of Method Validation

(b) (4) Project Code	OZZ2		
Method ID	LCMSC 634		
Analytes	Delafloxacin		
Matrix	Human Plasma		
Anticoagulant	Dipotassium EDTA		
Method Description	Supported liquid phase extraction		
Sample Volume (µL)	100-µL		
Sample Storage Temperature	-20 °C or colder		
Internal Standard (IS)	Delafloxacin-d_5		
Regression, Weighting	Linear, 1/concentration^2		
Average Recovery of Drug (%)	71.9%		
Average Recovery of IS (%)	70.7%		
Standard Curve Concentrations	5.00 to 5000 ng/mL		
QC Concentrations	5.00, 15.0, 40.0, 150, 600, and 3750 ng/mL		
QC Intra-assay Statistics (%)	Conc. (ng/mL)	Precision	Accuracy
	5.00	3.36 to 5.36%	-0.888 to 3.91%
	15.0	1.83 to 2.18%	-1.90 to 2.98%
	40.0	0.794 to 1.74%	-0.267 to 1.80%
	150	1.42 to 2.56%	-1.11 to 2.69%
	600	1.21 to 1.93%	0.821 to 2.20%
	3750	1.28 to 4.13%	0.192 to 2.02%
QC Inter-assay Statistics (%)	Conc. (ng/mL)	Precision	Accuracy
	5.00	4.49%	0.872%
	15.0	2.79%	0.730%
	40.0	1.54%	0.747%
	150	2.49%	0.593%
	600	1.71%	1.60%
	3750	2.58%	1.02%
Thawed Matrix Stability (hrs)	24 hours at room temperature		
Solution Stability (days)	Delafloxacin 35 days at 2 to 8 °C in standard diluent*		
	Delafloxacin-d_5 35 days at 2 to 8 °C in standard diluent*		
Solution Stress Stability (hours)	Delafloxacin 7 hours at room temperature in 50:50 Acetonitrile / Water*		
	Delafloxacin-d_5 7 hours at room temperature in 50:50 Acetonitrile / Water*		
Extract Stability (hrs)	167 hours at 2 to 8 °C		
Freeze-thaw Stability (cycles)	Five cycles thawed at room temperature		
Frozen Matrix Storage Stability (days)	29 days at -20 °C and -70 °C		
Whole Blood Stability	N/A		
Dilutional Linearity	150 ng/mL diluted four-fold		
	10000 ng/mL diluted ten-fold		
Selectivity	No significant interfering peaks noted in blank plasma samples		
Hemolysis	No effect from hemolysis on the quantitation of delafloxacin		
Lipemia	No effect from lipemia on the quantitation of delafloxacin		

* Established prior to/following the validation

	Delafloxacin Plasma
Range	5.00- 5000 ng/mL
LLOQ	5.00 ng/mL
Linearity	linear (correlation coefficient ≥ 0.990)
Accuracy	85-115%
Precision	≤ 15%
Selectivity	no interference

Stability	Sponsor only stated that "All samples werer analyzed within 241 days, demonstrated long term storage stabililty in human plasma containing dipotassium EDTA at -20C. No freeze and thaw stability was specified.

Reviewer's Comment: Based on the method validation and description of the Run Acceptance Criteria, both analytical method and runs met the acceptable criteria specified in the FDA Guidance to Industry: Bioanalytical Method Validation.

Pharmacokinetic Assessment:
The following PK parameters were calculated for delafloxacin from plasma concentration data for each subject using noncompartmental analysis:

- AUC_{0-t}: Area under the plasma concentration versus time curve from time 0 to the time of the last quantifiable concentration
- AUC_{0-tau}: area under the concentration versus time curve over the dosing interval of 12 hours
- AUC_{0-inf}: Area under the plasma concentration versus time curve from time 0 extrapolated to infinity
- AUC_{0-24}: Area under the plasma concentration versus time curve from time 0 to 24 hours after dosing
- AUC_{ext}: Area under the plasma concentration versus time curve extrapolated from the time of the last measurable concentration (tlast) to infinite time
- C_{max}: Maximum observed plasma concentration
- Kel: Terminal elimination rate constant
- $t_{1/2}$: Terminal elimination half-life
- T_{max}: Time to achieve maximum observed plasma concentration

Statistical Methods:
Pharmacokinetics:
For Part 1, to assess the relative exposures of the test dose form (Treatment A; oral delafloxacin 450 mg) to the reference dose form (Treatment B; IV delafloxacin 300 mg), a linear mixed-effect model was performed on the natural logarithm (ln)-transformed values of AUC_{0-t}, AUC_{0-inf}, and C_{max} with sequence, treatment, and period as fixed effects and subject nested within sequence as a random effect. The geometric least squares (LS) mean ratio of the 2 treatments for AUC_{0-t}, AUC_{0-inf}, and C_{max} were calculated by the antilog of the LS mean difference of the log-transformed values. A 90% confidence interval (CI) for the ratio was constructed as the antilog of the confidence limits of the LS mean difference. No adjustment was made for multiplicity. Statistical analysis of relative bioavailability was also assessed in the same manner on the dose-normalized parameters.

In addition, the geometric LS means and corresponding 90% CI were computed for AUC_{0-t}, AUC_{0-inf}, and C_{max} by taking the antilog of the LS means from the analysis of variance on the natural logarithm of the corresponding PK parameters.

Equivalence in exposure was concluded if the 90% CI for the test-to-reference ratios (A/B) of geometric means was entirely contained within the criterion interval of 80% to 125% for AUC_{0-t} and AUC_{0-inf}.

Safety: No formal statistical analysis of safety parameters was performed.

Safety Assessment: Safety and tolerability were assessed by monitoring and recording of AEs, clinical laboratory results (hematology [including coagulation parameters], serum chemistry, and urinalysis), vital sign measurements, 12-lead ECG results, and physical examination findings.

RESULTS

Study Population:
Please refer to Table 2 for the summary of subject demographics and baseline characteristics.

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Table 2. Summary of Subject Demographics and Baseline Characteristics –Part 1 (Safety Population) *(Adapted from Table 11-1 in the study report)*

	Treatment Sequence[a]		
	AB	**BA**	**Overall**
	(n = 28)	**(n = 28)**	**(N = 56)**
No. of subjects (%)			
Age (years)[b]			
Mean (SD)	35.0 (11.17)	37.5 (10.60)	36.2 (10.87)
Minimum, maximum	20, 56	22, 61	20, 61
Gender, No. (%)			
Male	15 (53.6)	13 (46.4)	28 (50.0)
Female	13 (46.4)	15 (53.6)	28 (50.0)
Race, No. (%)			
White	19 (67.9)	21 (75.0)	40 (71.4)
Black or African American	8 (28.6)	7 (25.0)	15 (26.8)
Asian	1 (3.6)	0 (0.0)	1 (1.8)
Ethnicity, No. (%)			
Hispanic or Latino	9 (32.1)	9 (32.1)	18 (32.1)
Not Hispanic or Latino	19 (67.9)	19 (67.9)	38 (67.9)
Height (cm)			
Mean (SD)	169.16 (10.015)	167.69 (10.237)	168.43 (10.061)
Minimum, maximum	152.3, 189.7	148.3, 186.5	148.3, 189.7
Weight (kg)			
Mean (SD)	75.58 (12.839)	75.09 (11.185)	75.33 (11.933)
Minimum, maximum	52.7, 97.7	53.5, 95.1	52.7, 97.7
Body mass index (kg/m^2)			
Mean (SD)	26.32 (3.214)	26.68 (3.024)	26.50 (3.097)
Minimum, maximum	19.2, 31.0	21.6, 31.6	19.2, 31.6

Abbreviation: SD, standard deviation.

Note: Percentages were calculated based on the number of subjects in the safety population.

[a] Treatment A = delafloxacin 450-mg tablet.
Treatment B = delafloxacin 300 mg intravenously infused over 1 hour.

[b] Age in years was calculated as the integer part of (date of informed consent – date of birth)/365.25.

Please refer to Table 3 for the summary of subject demographic and baseline characteristics for Part 2 study.

Table 3. Summary of Subject Demographic and Baseline Characteristics- Part 2 (Safety Population) *(Adapted from Table 11-2 in the study report)*

	Overall (N = 20)
No. of subjects (%)	
Age (years)[b]	
Mean (SD)	30.6 (8.29)
Minimum, maximum	19, 49
Gender, No. (%)	
Male	10 (50.0)
Female	10 (50.0)
Race, No. (%)	
White	12 (60.0)
Black or African American	6 (30.0)
Asian	1 (5.0)
Native Hawaiian or Other Pacific Islander	1 (5.0)
Ethnicity, No. (%)	
Hispanic or Latino	9 (45.0)
Not Hispanic or Latino	11 (55.0)
Height (cm)	
Mean (SD)	166.39 (9.677)
Minimum, maximum	147.5, 180.3
Weight (kg)	
Mean (SD)	72.47 (14.649)
Minimum, maximum	53.3, 98.2
Body mass index (kg/m^2)	
Mean (SD)	25.96 (3.283)
Minimum, maximum	20.0, 30.6

Abbreviation: SD, standard deviation.

Note: Percentages were calculated based on the number of subjects in the safety population.

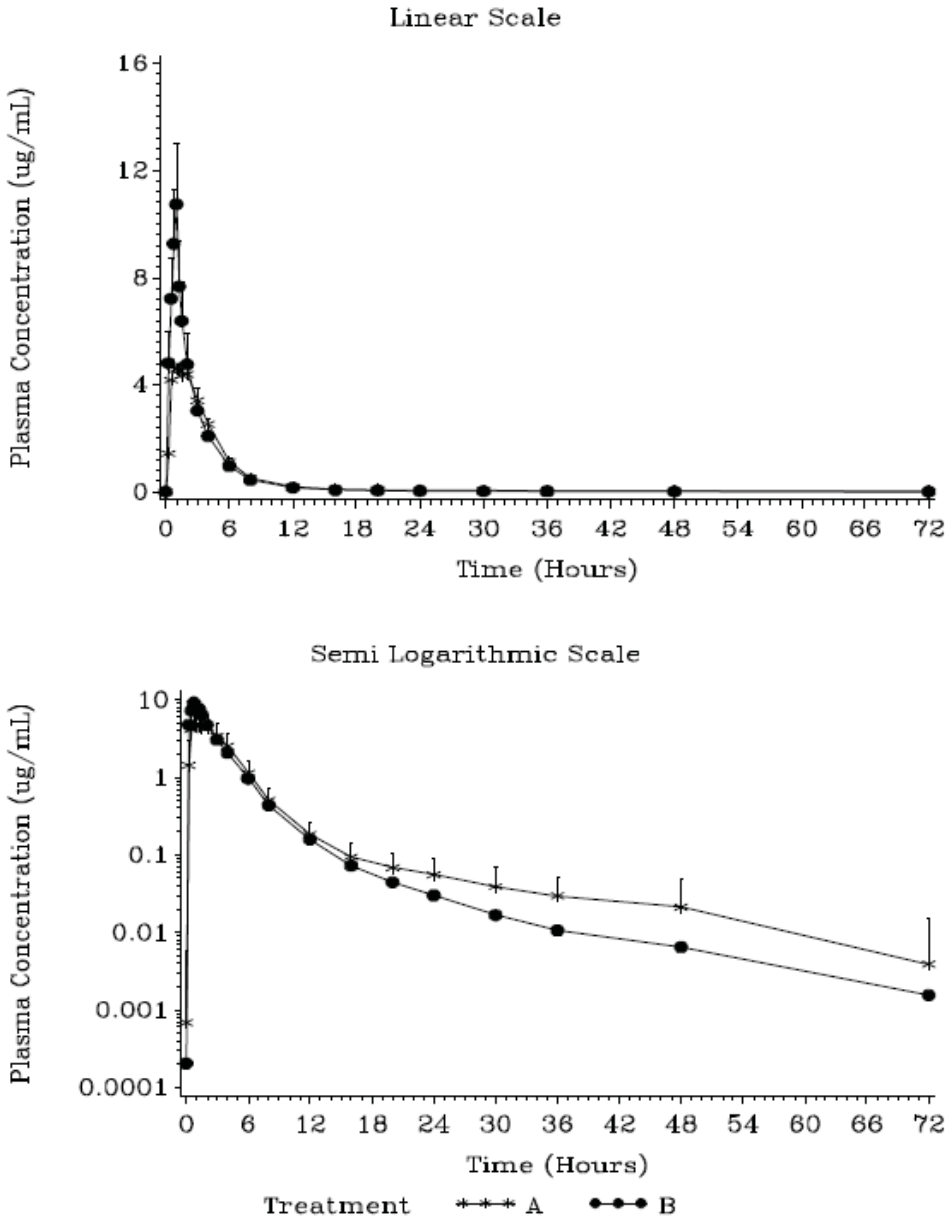
All subjects received 900 mg (2 × 450 mg) of delafloxacin.

[a] Age in years was calculated as the integer part of (date of informed consent – date of birth)/365.25.

Pharmacokinetics:

In Part 1, subjects received an oral dose of delafloxacin as a 450-mg tablet (Treatment A) and an IV dose of delafloxacin 300 mg infused over 1 hour (Treatment B). Mean (±SD) plasma concentrations of delafloxacin over time are presented on linear and semilogarithmic scales in Figure 1.

Figure 1. Mean (+SD) Plasma Concentrations of Delafloxacin (450-mg Oral and 300-mg IV Treatments) Versus Time (Pharmacokinetic Population, Linear and Semilogarithmic Scales) *(Adapted from Figure 11-1 in the study report)*



Treatment A = delafloxacin 450-mg tablet
Treatment B = delafloxacin 300 mg intravenously infused over 1 hour

Please refer to Table 4 for arithmetic mean (CV) for selected PK parameters of delafloxacin in Part 1.

Table 4. Mean (CV) Plasma Pharmacokinetic Parameters of Delafloxacin by Treatment (Pharmacokinetic Population) – Part 1

	Treatment	
	Delafloxacin 450-mg Tablet (A)	Delafloxacin 300-mg IV Infusion (B)
Parameter (unit)	**(n = 55)**	**(n = 55)**
$AUC_{0\text{-}tau}$ (µg•h/mL)	21.181 (29.7)	25.694 (21.5)
$AUC_{0\text{-}inf}$ (µg•h/mL)[a]	24.216 (26.6)	26.676 (22.6)
$AUC_{0\text{-}t}$ (µg•h/mL)	23.271 (30.1)	26.896 (21.5)
C_{max} (µg/mL)	6.115 (32.0)	10.745 (21.3)
T_{max} (h)[b]	0.817 (0.50, 4.00)	1.000 (0.75, 1.13)
K_{el} (1/h)[a]	0.0588 (43.0)	0.0804 (48.2)
$t_{1/2}$ (h)[a]	14.06 (42.0)	10.90 (54.6)
$AUC_{0\text{-}tau}$/Dose (µg•h/mL/mg)	0.04707 (29.7)	0.08565 (21.5)
$AUC_{0\text{-}inf}$/Dose (µg•h/mL/mg)[a]	0.05381 (26.6)	0.08892 (22.6)
$AUC_{0\text{-}t}$/Dose (µg•h/mL/mg)	0.05171 (30.1)	0.08965 (21.5)
C_{max}/Dose (µg/mL/mg)	0.013589 (32.0)	0.035816 (21.3)

Abbreviation: CV, coefficient of variation.

[a] n = 42 for Treatment A and n = 49 for Treatment B. Corresponding parameters were not calculated for some of the subjects because a linear regression could not be fitted through the terminal phase.

[b] For T_{max}, the median (minimum, maximum) values are presented.

The statistical analysis of plasma PK parameters of delafloxacin in Part 1 is presented in Table 5.

Table 5. Statistical Analysis of Plasma Pharmacokinetic Parameters of Delafloxacin Following 450-mg Oral and 300-mg IV Treatments (Pharmacokinetic Population)

Parameter (unit)	Treatment[a]	N	Geometric LS Means[c]	90% CI of the Geometric LS Means (A,B)	Ratio (%) of Geometric LS Means (A/B)[d]	90% CI of the Ratio (A/B)[e]
$AUC_{0\text{-}inf}$ (µg•h/mL)[b]	A	42	22.970	(21.614, 24.412)	87.680	(83.562, 92.001)
	B	49	26.198	(24.708, 27.778)		
$AUC_{0\text{-}t}$ (µg•h/mL)	A	55	22.239	(20.987, 23.567)	84.447	(80.898, 88.151)
	B	55	26.335	(24.852, 27.908)		
C_{max} (µg/mL)	A	55	5.797	(5.443, 6.173)	55.155	(51.496, 59.075)
	B	55	10.510	(9.869, 11.193)		

Abbreviations: CI, confidence interval; CV, coefficient of variation; IV, intravenous; LS, least square.

Note: A linear mixed-effect model on the natural logarithms (ln) of $AUC_{0\text{-}inf}$, $AUC_{0\text{-}t}$ and C_{max} was performed with treatment, sequence, and period as fixed effects and subject nested within sequence as a random effect.

[a] Treatment A = delafloxacin 450-mg tablet.
Treatment B = delafloxacin 300 mg infused over 1 hour.

[b] n = 42 for Treatment A and n = 49 for Treatment B. Corresponding parameters were not calculated for some of the subjects because a linear regression line could not be fitted through the terminal phase.

[c] Least squares mean from analysis of variance. Least squares means were calculated by transforming the ln means back to linear scale, ie, geometric means.

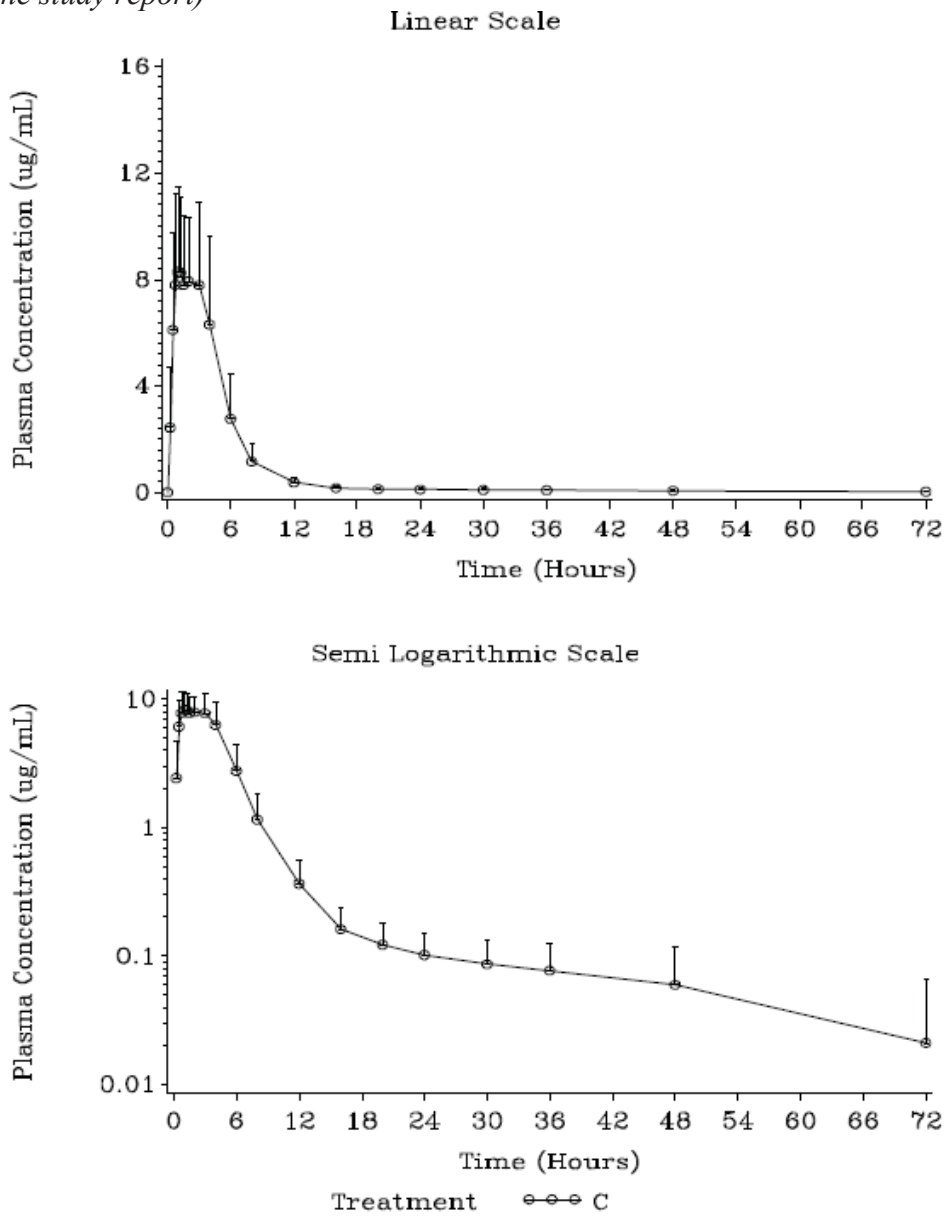
[d] Ratio of least square means (expressed as a percentage). Ln-transformed difference was transformed back to linear scale to obtain the ratio of geometric least square means.

[e] The 90% CI for the ratio of metric means (expressed as a percentage). Ln-transformed confidence limits were transformed back to linear scale.

Reviewer's Comment: The overall systemic exposure of delafloxacin (AUC) following administration of a 450 mg oral tablet was comparable to that following 300 mg IV infusion. Of note, AUC is the PK/PD index associated with antibacterial activity for delafloxacinin in animal studies.

In Part 2, subjects received an oral dose of 900 mg of delafloxacin (2 × 450-mg tablets [Treatment C]). Mean (+SD) plasma concentrations of delafloxacin over time are presented on linear and semilogarithmic scales in Figure 2.

Figure 2. Mean (+SD) Plasma Concentrations of Delafloxacin (900 mg Oral) Versus Time (Pharmacokinetic Population, Linear and Semilogarithmic Scales) *(Adapted from Figure 11-2 in the study report)*



Treatment C = delafloxacin 900 mg (2 × 450-mg tablet)

Please refer to Table 6 for the Mean (CV) values for Plasma Pharmacokinetic Parameters of Delafloxacin in Part 2.

Table 6. Mean (CV) Plasma Pharmacokinetic Parameters of Delafloxacin by Treatment (Pharmacokinetic Population) – Part 2

	Treatment
	Delafloxacin Tablet (2 × 450 mg) (C)
Parameter (unit)	**(n = 20)**
$AUC_{0\text{-}tau}$ (µg•h/mL)	44.127 (35.5)
$AUC_{0\text{-}inf}$ (µg•h/mL)[a]	44.137 (31.1)
$AUC_{0\text{-}t}$ (µg•h/mL)	48.762 (35.9)
C_{max} (µg/mL)	10.384 (32.7)
T_{max} (h)[b]	1.375 (0.50, 4.02)
K_{el} (1/h)[a]	0.0497 (42.3)
$t_{1/2}$ (h)[a]	17.82 (58.8)

Abbreviation: CV, coefficient of variation.
[a] n = 14. Corresponding parameters were not calculated for some of the subjects because a linear regression could not be fitted through the terminal phase.
[b] For T_{max}, the median (minimum, maximum) values are presented.

Safety:
In Part 1, all TEAEs were considered either possibly related (6 subjects, 10.7%) or unrelated (5 subjects, 8.9%) to study drug. No TEAE was considered probably or definitely related to study drug. With the exception of 2 TEAEs considered moderate in severity, all TEAEs were mild. Moderate, possibly related TEAEs of nausea and vomiting were reported by 1 subject after delafloxacin 300 mg infused over 1 hour. All TEAEs resolved by the end of the study. There were no deaths, SAEs, or TEAEs leading to study discontinuation.

In Part 2, 3 TEAEs were reported and 3 of 20 subjects (15.0%) experienced at least 1 TEAE. The TEAEs of diarrhea, nausea, and epistaxis were experienced by 1 subject each. Diarrhea and epistaxis were considered unrelated to study drug and nausea was considered possibly related to study drug. No TEAE was considered probably or definitely related to study drug. All TEAEs were mild and resolved by the end of the study. There were no deaths, SAEs, or TEAEs leading to study discontinuation.

SPONSOR'S CONCLUSIONS
PHARMACOKINETIC

- Equivalence in total exposure of delafloxacin was concluded since the 90% CI of the geometric mean ratios for the 450-mg tablet relative to 300 mg infused over 1 hour (A/B) was entirely contained within the predefined criterion interval of 80% to 125% for $AUC_{0\text{-}t}$ and $AUC_{0\text{-}inf}$.
- Median T_{max} of delafloxacin occurred at 0.82 hour following the administration of the 450-mg tablet and at 1.00 hour following the 300 mg infused over 1 hour.

- Mean total exposure ($AUC_{0\text{-}inf}$) and mean peak plasma concentration (C_{max}) of delafloxacin were 44.14 µg•h/mL and 10.38 µg/mL, respectively, following the administration of two 450-mg tablets.
- Median T_{max} of delafloxacin occurred at 1.38 hours following the administration of the two 450-mg tablets.

SAFETY

- Single delafloxacin doses of 1 × 450-mg tablet and 2 × 450-mg tablets, and 300 mg infused over 1 hour were safe and well tolerated by the healthy adult subjects in this study.
- In Part 1, 16 TEAEs were reported and 11 of 56 subjects (19.6%) experienced at least 1 TEAE. Treatment-emergent AEs were reported by 7 of 55 subjects (12.7%) after delafloxacin 300 mg infused over 1 hour and by 4 of 55 subjects (7.3%) after delafloxacin 450-mg tablet.
- In Part 1, the most frequently reported TEAE overall was headache followed by diarrhea.
- All TEAEs were considered either possibly related (6 subjects, 10.7%) or unrelated (5 subjects, 8.9%) to study drug. Most TEAEs were mild and all TEAEs resolved by the end of the study. There were no deaths, SAEs, or TEAEs leading to study discontinuation.
- In Part 2, 3 TEAEs were reported and 3 of 20 subjects (15.0%) experienced at least 1 TEAE. The TEAEs of diarrhea, nausea, and epistaxis were experienced by 1 subject each. Two TEAEs were considered unrelated to study drug and 1 TEAE was considered possibly related to study drug. All TEAEs were mild and resolved by the end of the study. There were no deaths, SAEs, or TEAEs leading to study discontinuation.
- There were no clinically significant findings observed or treatment-related AEs reported resulting from clinical laboratory assessments, vital sign measurements, ECG results, or physical examination findings.

REVIEWER ASSESSMENT:

The results of this study showed that the overall exposure (AUCs) of 450 mg oral tablet appeared to be comparable to that of 300 mg IV injection. However, it was also noticed that the overall exposure following oral administration of 450 mg tablet was numerically lower than that after 300 mg IV infusion.

4.5.15 Study No.: RX-3341-116

A PHASE 1, OPEN-LABEL, CROSSOVER STUDY TO DETERMINE THE EFFECT OF FOOD ON THE PHARMACOKINETICS OF A SINGLE DOSE OF ORAL DELAFLOXACIN IN HEALTHY SUBJECTS

Date(s): 05 March 2014 to 08 May 2014
Sponsor: Melinta Therapeutics, Inc, 300 George Street, Suite 301, New Haven, CT 06511
Clinical Site: Single site: PPD Phase I Clinic, located at 7551 Metro Center Drive, Suite 200, Austin, TX 78744
Analytical Site: (b) (4)

OBJECTIVE(S):

- The primary objective was to compare the pharmacokinetic (PK) profile of single-dose oral administration of delafloxacin in the fed and fasted states in healthy adult subjects.
- The secondary objective was to evaluate the safety and tolerability of a single oral dose of delafloxacin in healthy adult subjects.

METHODS

Study Design: This was a Phase 1, single-dose, randomized, open-label, 3-period, 6-sequence crossover study in 30 healthy subjects. Subjects who met all of the eligibility criteria were randomly assigned to 1 of 6 treatment sequences (see Table 1 below for details). Each sequence comprised 3 treatments of various fed and fasted states as follows:

- Treatment A: A single oral dose of 900 mg delafloxacin under fasting conditions (overnight fast of at least 10 hours)
- Treatment B: A single oral dose of 900 mg delafloxacin under fed conditions (overnight fast of at least 10 hours, followed by a standardized Food and Drug Administration high-fat breakfast 30 minutes before dosing)
- Treatment C: A single oral dose of 900 mg delafloxacin under fasting conditions (overnight fast of at least 10 hours, with a high-fat meal 2 hours after dosing)

This study consisted of a screening period (Days –28 to –2), 3 check-in periods (Day –1 of each period), 3 treatment periods (Days 1 to 3 of each period), and end-of-study/early termination assessments (Day 3 of Period 3).

Table 1. Treatment Sequences *(Adapted from Table 9-1 in the study report)*

Sequence	Period 1	Period 2	Period 3
ABC	A	B	C
ACB	A	C	B
BAC	B	A	C
BCA	B	C	A
CAB	C	A	B
CBA	C	B	A

Thirty subjects were planned for this study. Twenty-six subjects (86.7%) completed the study and 4 subjects (13.3%) were discontinued for reasons classified as other. All 30 subjects (100.0%) were analyzed for safety and pharmacokinetics. Serial blood samples for the PK analysis of delafloxacin were collected before dosing and up to 48 hours after dosing.

Drug Product: Delafloxacin 900 mg (two 450 mg tablets, to-be-marketed oral dosage form), Lot # 13DE009A.

PK Sample Collection:
Please refer to Table 2 for the schedule of plasma PK sample collection.

Table 2. Schedule of Events *(Adapted from Table 9-2)*

		Period 1, 2, or 3																					
			Day and Time Time after study drug administration (hours)																				
			Day 1																Day 2				Day 3[a]
Procedure	Screening Days −28 to −2	Check-in Day −1	Predose	0	0.25	0.5	0.75	1	1.25	1.5	2	2.5	3	3.5	4	6	8	12	16	20	24	36	48
Informed consent	X																						
Inclusion/Exclusion criteria	X	X																					
Medical history	X	X																					
Physical examination[b]	X	X																					X
Vital sign measurements[c]	X	X	X																				X
12-Lead electrocardiogram[d]	X	X	X								X												X
Serology[e]	X																						
Clinical laboratory testing[f]	X	X	X																				X
Urine drug and alcohol screen	X	X																					
Pregnancy test (all female subjects)	X	X																					
Admission to clinical unit		X																					
Randomization[g]			X																				
Study drug administration[h]				X																			
Pharmacokinetic sample collection[i]			X		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
Adverse event monitoring[j]		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
Concomitant medications	X	X	X		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
Study discharge																							X

[a] A minimum 7-day washout interval between doses in each period was held after Day 3 of Periods 1 and 2.

[b] Physical examination measurements included height and weight at Screening for body mass index calculation. A complete physical examination was performed at Screening and before discharge (Day 3, Period 3) or early termination. A brief physical examination was performed at Check-in (Day −1, Periods 1, 2, and 3).

[c] Vital signs (systolic and diastolic blood pressure, pulse rate, respiratory rate, and oral body temperature) were measured at Screening, Check-in (Day −1), before dosing, and before discharge or early termination.

[d] A standard 12-lead electrocardiogram was performed at Screening, Check-in (Day −1), before dosing, 2 hours after initiation of dosing, and before discharge or early termination.

[e] Serology testing included hepatitis B surface antigen, hepatitis C virus antibody, and human immunodeficiency virus (types 1 and 2) antibodies.

[f] Clinical laboratory testing (hematology [including coagulation parameters], serum chemistry, and urinalysis) was performed at Screening, Check-in (Day −1), before dosing, and before discharge or early termination.

[g] Randomization to 1 of 6 treatment sequences was performed on Day 1 of Period 1.

[h] All subjects received oral delafloxacin (two 450-mg tablets).

[i] Blood samples for pharmacokinetic analysis were collected before dosing (0 hour) and at 0.25, 0.5, 0.75, 1, 1.25, 1.5, 2, 2.5, 3, 3.5, 4, 6, 8, 12, 16, 20, 24, 36, and 48 hours after dosing.

[j] Adverse events were monitored from Check-in through 7 days after discharge from the study. Any serious adverse events occurring within 30 days after the last dose were collected.

Analytical Methods:

	Delafloxacin Plasma
Range	5.00- 5000 ng/mL
LLOQ	5.00 ng/mL
Linearity	linear (correlation coefficient ⩾ 0.990)
Accuracy	85-115%
Precision	⩽ 15%
Selectivity	no interference
Stability	Sponsor only stated that "All samples were analyzed within 241 days, demonstrated long term storage stabililty in human plasma containing dipotassium EDTA at -20°C. No freeze and thaw stability was specified.

Reviewer's Comment: The Sponsor should specify the validated method used for this assay. The bioanalytical report is not well prepared and with a lot of spelling errors.

Pharmacokinetic Assessment:
The following PK parameters were calculated for delafloxacin from plasma concentration data for each subject using noncompartmental analysis:

- AUC_{0-t}: Area under the plasma concentration versus time curve from time 0 to the time of the last quantifiable concentration
- AUC_{0-inf}: Area under the plasma concentration versus time curve from time 0 extrapolated to infinity
- AUC_{0-12}: Area under the plasma concentration versus time curve from time 0 to 12 hours from initiation of dosing
- AUC_{0-24}: Area under the plasma concentration versus time curve from time 0 to 24 hours after dosing
- AUC_{ext}: Area under the plasma concentration versus time curve extrapolated from the time of the last measurable concentration (tlast) to infinite time
- C_{max}: Maximum observed plasma concentration
- K_{el}: Terminal elimination rate constant
- $t_{1/2}$: Terminal elimination half-life
- T_{max}: Time to achieve maximum observed plasma concentration
- CL/F: Apparent total clearance
- V_z/F: Apparent volume of distribution

Statistical Methods:
Pharmacokinetics:

The geometric least squares (LS) mean ratios of the 2 test treatments to the reference treatment (B/A and C/A) for AUC_{0-t}, AUC_{0-inf}, and C_{max} were calculated by the antilog of the LS mean difference of the natural log-transformed values. A 90% confidence interval (CI) for each ratio was constructed as the antilog of the 90% CI of the LS mean difference. No adjustment was made for multiplicity. In addition, the geometric LS means and corresponding 90% CIs were computed for AUC_{0-t}, AUC_{0-inf}, and Cmax by taking the antilog of the LS means and corresponding 90% CIs from the linear mixed-effect model on the natural logarithm of the corresponding PK parameters.

The absence of a food effect was concluded if the 90% CI for the test-to-reference ratios (C/A or B/A) of geometric LS means was entirely contained within the criterion interval of 80% to 125% for AUC_{0-t}, AUC_{0-inf}, and C_{max}.

The Wilcoxon signed-rank test was performed on T_{max}. A p value less than or equal to 0.05 was considered a significant difference between treatments.

Reviewer's Comment: Per the FDA Guidance for Industry: Food-effect bioavailability and Fed Bioequivalence Studies, the highest strength of the oral formulation should be used in the food effect study. However, two tablets (450 mg × 2) instead of one 450 mg tablet were administered to subjects enrolled in this study. Based on results from Study M00-224, Study RX-3341-115, and this study, the increase in delafloxacin AUC appeared approximately dose proportional over the dose range of 450 to 900 mg. Therefore, the use of 900 mg is acceptable in this food effect study.

Safety: No formal statistical analysis of safety parameters was performed. All TEAEs were presented in a summary table by treatment (Treatment A, B, and C) and overall, with the number and percentage of subjects who experienced at least 1 TEAE. Treatment-emergent AEs were also presented in a summary table by treatment and overall, relationship to study drug, severity, TEAEs that led to study drug discontinuation, and treatment-emergent serious AEs.

Safety Assessment: Safety assessments included adverse event (AE) reporting, clinical laboratory results (hematology [including coagulation parameters], serum chemistry, and urinalysis), vital sign measurements (systolic and diastolic blood pressure, pulse rate, respiratory rate, and oral body temperature), 12-lead electrocardiogram (ECG) measurements, and physical examination findings.

RESULTS

Study Population:
Please refer to Table 3 for the summary of subject disposition.

Table 3. Summary of Subject Disposition (All Subjects) *(Adapted from Table 10-1 in the study report)*

	Treatment Sequence[a]						
	ABC (n = 5)	ACB (n = 5)	BAC (n = 5)	BCA (n = 5)	CAB (n = 5)	CBA (n = 5)	Overall (N = 30)
Total number of subjects, No. (%)							
Completed	4 (80.0)	4 (80.0)	5 (100.0)	3 (60.0)	5 (100.0)	5 (100.0)	26 (86.7)
Discontinued	1 (20.0)	1 (20.0)	0	2 (40.0)	0	0	4 (13.3)
Study populations							
Safety population	5 (100.0)	5 (100.0)	5 (100.0)	5 (100.0)	5 (100.0)	5 (100.0)	30 (100.0)
PK population	5 (100.0)	5 (100.0)	5 (100.0)	5 (100.0)	5 (100.0)	5 (100.0)	30 (100.0)
Reason for discontinuation							
Other	1 (20.0)	1 (20.0)	0	2 (40.0)	0	0	4 (13.3)

Abbreviation: PK, pharmacokinetic.

Note: Percentages were based on the number of all subjects within each treatment sequence and overall. The safety population included all subjects who received at least 1 dose of study drug. The PK population included all subjects who received at least 1 dose of delafloxacin and had sufficient concentration data to support accurate estimation of at least 1 PK parameter.

[a] Treatment A = oral delafloxacin 900 mg, fasted.
Treatment B = oral delafloxacin 900 mg, fed.
Treatment C = oral delafloxacin 900 mg, fasted, with a meal 2 hours after dosing.

Thirty subjects were enrolled and 26 subjects (86.7%) completed the study. Four subjects (13.3%) discontinued the study as follows:

- Subject 112 did not check-in for Period 3 and was lost to follow-up.
- Subject 123 did not check-in to Period 2 due to a family emergency.
- Subject 125 was excluded due to a positive urine drug screen.
- Subject 127 was excluded due to ECG Bazett-corrected QT interval per investigator discretion.

There were no admission criteria deviations. Deviations in PK sampling times and 2 missing time points for 1 subject occurred during the study; however, because calculation of the PK parameters was based on actual sampling times, these differences did not affect the results.

Please refer to Table 4 for the summary of subject demographic and baseline characteristics.

Table 4. Summary of Subject Demographic and Baseline Characteristics (All Subjects) *(Adapted from Table 11-1 in the study report)*

	Treatment Sequence[a]						
No. of subjects (%)	ABC (n = 5)	ACB (n = 5)	BAC (n = 5)	BCA (n = 5)	CAB (n = 5)	CBA (n = 5)	Overall (N = 30)
Age (years)							
Mean (SD)	36.6 (11.10)	30.0 (9.62)	36.8 (9.36)	35.6 (14.15)	29.8 (8.01)	33.4 (10.99)	33.7 (10.19)
Minimum, maximum	25, 49	22, 45	24, 46	22, 55	25, 44	22, 50	22, 55
Gender, No. (%)							
Female	2 (40.0)	3 (60.0)	5 (100.0)	2 (40.0)	2 (40.0)	5 (100.0)	19 (63.3)
Male	3 (60.0)	2 (40.0)	0	3 (60.0)	3 (60.0)	0	11 (36.7)
Race, No. (%)							
White	4 (80.0)	2 (40.0)	5 (100.0)	3 (60.0)	5 (100.0)	5 (100.0)	24 (80.0)
Black or African American	1 (20.0)	3 (60.0)	0	2 (40.0)	0	0	6 (20.0)
Ethnicity, No. (%)							
Hispanic or Latino	3 (60.0)	2 (40.0)	3 (60.0)	3 (60.0)	4 (80.0)	3 (60.0)	18 (60.0)
Not Hispanic or Latino	2 (40.0)	3 (60.0)	2 (40.0)	2 (40.0)	1 (20.0)	2 (40.0)	12 (40.0)
Height (cm)							
Mean (SD)	167.44 (6.750)	168.30 (7.077)	161.16 (2.587)	171.90 (8.407)	162.06 (8.580)	157.46 (8.448)	164.72 (8.290)
Minimum, maximum	157.0, 175.2	161.6, 178.9	158.2, 165.1	161.5, 181.0	150.0, 173.5	149.0, 169.8	149.0, 181.0
Weight (kg)							
Mean (SD)	74.28 (9.275)	72.56 (9.691)	68.54 (3.745)	73.22 (20.168)	62.52 (9.236)	62.94 (14.567)	69.01 (12.147)
Minimum, maximum	65.8, 89.0	62.9, 83.0	63.3, 73.7	51.3, 93.4	55.0, 77.5	51.0, 84.6	51.0, 93.4
Body mass index (kg/m^2)							
Mean (SD)	26.46 (1.727)	25.56 (2.341)	26.40 (1.485)	24.40 (4.555)	23.76 (2.641)	25.12 (3.295)	25.28 (2.793)
Minimum, maximum	24.2, 29.0	22.9, 29.1	24.6, 28.1	18.7, 28.9	20.4, 25.9	21.1, 29.3	18.7, 29.3

Abbreviation: SD, standard deviation.

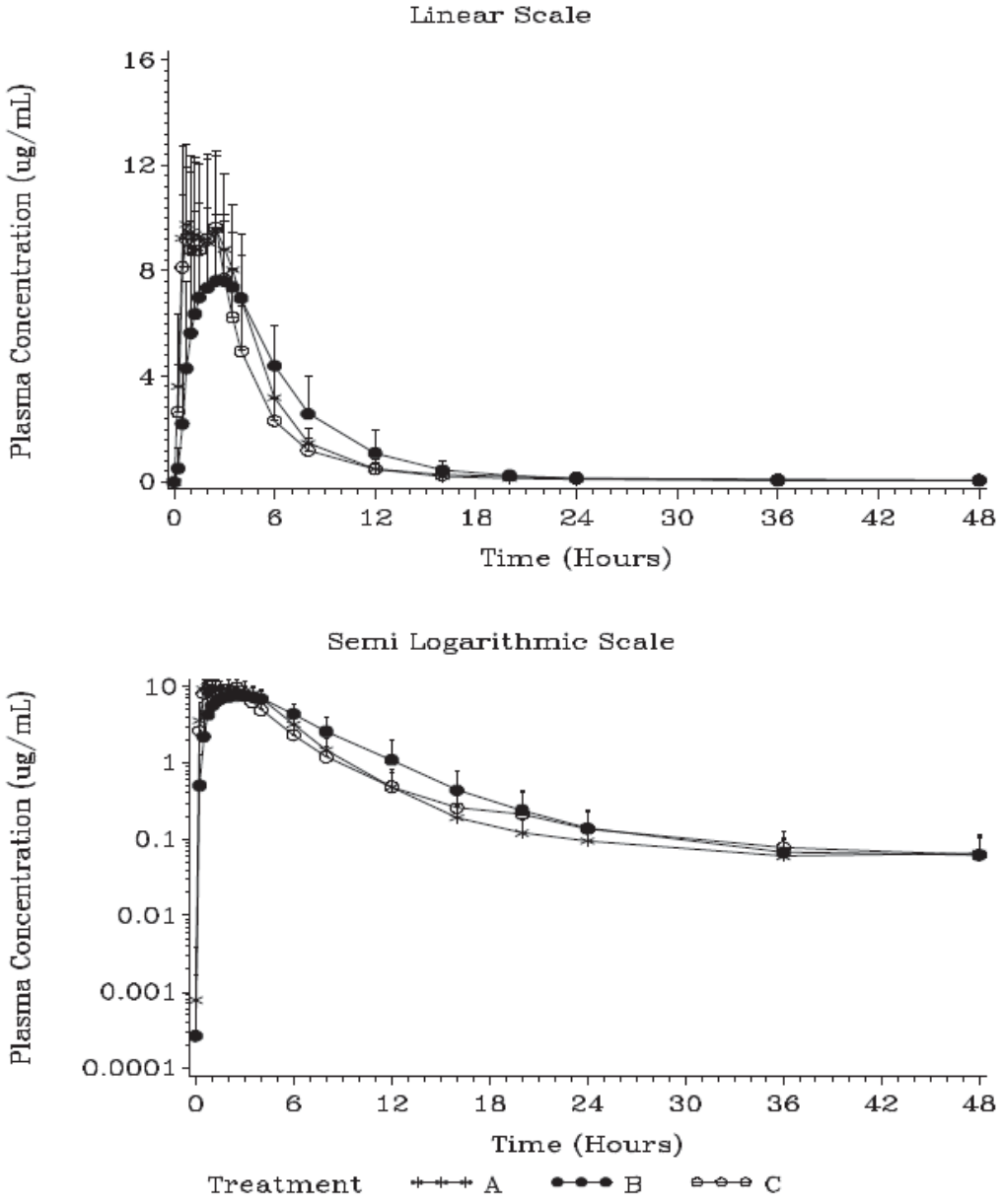
Note: Percentages were calculated based on the number of subjects in the safety population within each treatment sequence and overall.

[a] Treatment A = oral delafloxacin 900 mg, fasted.
Treatment B = oral delafloxacin 900 mg, fed.
Treatment C = oral delafloxacin 900 mg, fasted, with a meal 2 hours after dosing.

Pharmacokinetics:

The mean (±SD) concentration versus time profile for delafloxacin is presented in Figure 1 below.

Figure 1. Mean (+SD) Plasma Concentrations of Delafloxacin Versus Time (Pharmacokinetic Population, Linear and Semilogarithmic Scales) *(Adapted from Figure 11-1 in the study report)*



Please refer to Table 5 for the Mean ± SD (CV) values for selected PK parameters of delafloxacin,

Table 5. Mean ± SD (CV) Plasma Pharmacokinetic Parameters of Delafloxacin by Treatment (Pharmacokinetic Population) *(Adapted from Table 11-2 in the study report)*

	Treatment		
Parameter (Unit)	**Delafloxacin 900 mg (Fasted) (A) (N = 28)**	**Delafloxacin 900 mg (Fed) (B) (N = 29)**	**Delafloxacin 900 mg (Fasted, with a meal 2 hours after dosing) (C) (N = 25)**
AUC_{0-12} (µg•h/mL)	52.12 ± 14.44 (27.7)	49.91 ± 12.19 (24.4)	44.51 ± 10.86 (24.4)
AUC_{0-inf} (µg•h/mL)[a]	55.15 ± 13.68 (24.8)	58.21 ± 11.41 (19.6)	52.21 ± 12.55 (24.0)
AUC_{0-t} (µg•h/mL)	56.21 ± 15.21 (27.1)	57.09 ± 14.15 (24.8)	49.77 ± 12.01 (24.1)
AUC_{0-24} (µg•h/mL)	54.52 ± 15.01 (27.5)	55.08 ± 13.94 (25.3)	47.64 ± 11.74 (24.6)
AUC_{ext} (µg•h/mL)[a]	0.91 ± 0.85 (93.1)	0.71 ± 0.64 (90.7)	0.64 ± 0.44 (68.9)
C_{max} (µg/mL)	11.52 ± 2.76 (24.0)	9.14 ± 2.81 (30.8)	11.75 ± 2.64 (22.5)[b]
T_{max} (h)[c]	1.25 (0.50, 4.00)	2.50 (1.00, 6.00)	1.50 (0.50, 2.50)[b]
$t_{1/2}$ (h)[a]	14.11 ± 8.37 (59.3)	12.86 ± 6.31 (49.0)	11.95 ± 4.26 (35.6)
CL/F (L/h)[a]	17.19 ± 4.06 (23.6)	16.08 ± 3.45 (21.4)	18.15 ± 4.23 (23.3)
V_z/F (L)[a]	369.86 ± 282.22 (76.3)	307.03 ± 179.60 (58.5)	313.37 ± 133.36 (42.6)

Abbreviations: CV, coefficient of variation.

Note: Subject 129 had emesis in Period 1/Treatment C, and Subject 130 had emesis in Period 3/Treatment C. These subjects were excluded from the pharmacokinetic analysis for the respective periods. Collection samples were missing for Subject 118 Treatment C and Subject 128 Treatment C around the median T_{max} of the treatment group. These subjects' observed C_{max} and T_{max} were excluded from the pharmacokinetic analysis since these were not true parameters.

[a] n = 9, 16, and 14 for Treatments A, B, and C, respectively, for parameters AUC_{0-inf}, AUC_{ext}, $t_{1/2}$, CL/F, and V_z/F.

[b] n = 23 for C_{max} and T_{max} following Treatment C.

[c] For T_{max}, the median (minimum, maximum) values are presented.

Please refer to Table 6 for statistical analysis of plasma PK parameters of delafloxacin.

Table 6. Statistical Analysis of Plasma Pharmacokinetic Parameters of Delafloxacin (Pharmacokinetic Population) (*Adapted from Table 11-3 in the study report)*

Parameter (Unit)	Treatment[a]	N	Geometric LS Means	90% Confidence Interval of Geometric LS Means	Treatment Comparison	Ratio (%) of Geometric LS Means	90% Confidence Interval of the Ratio (%)
AUC_{0-t} (µg•h/mL)	A	28	51.419	(47.052, 56.190)			
	B	29	52.337	(47.953, 57.123)	B/A	101.787	(95.706, 108.253)
	C	25	45.913	(41.914, 50.293)	C/A	89.292	(83.818, 95.124)
AUC_{0-inf} (µg•h/mL)	A	9	49.479	(43.696, 56.029)			
	B	16	51.951	(47.082, 57.324)	B/A	104.996	(92.082, 119.720)
	C	14	49.629	(44.601, 55.225)	C/A	100.303	(86.730, 116.001)
C_{max} (µg/mL)	A	28	10.309	(9.413, 11.290)			
	B	29	8.196	(7.500, 8.955)	B/A	79.504	(73.126, 86.439)
	C	23	10.546	(9.562, 11.631)	C/A	102.301	(93.644, 111.758)

Abbreviation: LS, least squares.

Note: A linear mixed-effect model was performed on the natural log-transformed values of AUC_{0-t}, AUC_{0-inf}, and C_{max} with sequence, treatment, and period as fixed effects and subject nested within sequence as a random effect.

Subject 129 had emesis in Period 1/Treatment C, and Subject 130 had emesis in Period 3/Treatment C. These subjects were excluded from pharmacokinetic analysis for the respective periods. Collection samples were missing for Subject 118 Treatment C and Subject 128 Treatment C around the median T_{max} of the treatment group. These subjects' observed C_{max} and T_{max} were excluded from the pharmacokinetic analysis since these were not true parameters.

[a] Treatment A = oral delafloxacin 900 mg, fasted.
Treatment B = oral delafloxacin 900 mg, fed.
Treatment C = oral delafloxacin 900 mg, fasted, with a meal 2 hours after dosing.

Reviewer's Comment: When comparing the delafloxacin exposures following oral 900 mg delafloxacin under fed conditions (Treatment B) to delafloxacin 900 mg under fasted conditions (Treatment A), the geometric LS mean ratios (90% CI) of AUC_{0-t}, AUC_{0-inf}, and C_{max} were 101.8% (95.7, 108.3),105.0% (92.1, 119.7), and 79.5% (73.1, 86.4), respectively. According to the FDA Guidance for Industry: Food-Effect Bioavailability and Fed Bioequivalence Studies, an absence of food effect on bioavailability can be established based on AUC_{0-t} and AUC_{0-inf} since the 90% CI for the ratio of population geometric mean of these two PK parameters were contained in the equivalence limits of 80-125%. However, 90% CI for the ratio of population geometric means between fed and fasted treatments was not contained in the equivalence limits of 80-125% for C_{max} (79.5% (73.1, 86.4)). There was a 20.5% decrease in C_{max} under fed condition. As the PK-PD index for delafloxacin is AUC/MIC, the slightly decrease in C_{max} in fed condition does not seem to affect the overall efficacy outcome of delafloxacin.

Please refer to Table 7 for statistical analysis of the difference in median T_{max} for delafloxacin.

Table 7. Statistical Analysis of the Difference in Median T_{max} for Delafloxacin (Pharmacokinetic Population) *(Adapted from Table 11-4 in study report)*

Parameter (Unit)	Treatment	N	Median	Treatment Comparison	*P* Value
T_{max} (h)	A	28	1.250		
	B	29	2.500	B-A	<0.001
	C	23	1.500	C-A	0.805

Note: The Wilcoxon signed-rank test was performed to estimate the difference in median T_{max}.
Treatment A = oral delafloxacin 900 mg, fasted.
Treatment B = oral delafloxacin 900 mg, fed.
Treatment C = oral delafloxacin 900 mg, fasted, with a meal 2 hours after dosing.

Reviewer's Comment: Median T_{max} was delayed by 1.25 hours following oral administration of 900 mg of delafloxacin under fed conditions compared to fasted conditions (B-A) (P<0.001).

Safety:
Overall, 28 TEAEs were reported and 14 of 30 subjects (46.7%) reported at least 1 TEAE. Treatment-emergent AEs were reported by 7 subjects (25.9%) after receiving delafloxacin under fasted conditions with a meal 2 hours after dosing, 6 subjects (20.7%) after delafloxacin under fed conditions, and 4 subjects (14.3%) after delafloxacin under fasted conditions. The most frequently reported TEAE overall was diarrhea (5 subjects, 16.7%). Diarrhea was reported by 4 subjects (13.8%) after receiving delafloxacin under fed conditions, 2 subjects (7.4%) after delafloxacin under fasted conditions with a meal 2 hours after dosing, and 1 subject (3.6%) after delafloxacin under fasted conditions.

With the exception of 3 TEAEs of moderate severity occurring in 2 subjects, all TEAEs reported during this study were mild. One subject reported a moderate TEAE of nausea and 2 subjects reported a moderate TEAE of vomiting, each after receiving delafloxacin under fasted conditions with a meal 2 hours after dosing. Subject 129 reported a moderate, possibly related TEAE of vomiting and Subject 130 reported moderate, possibly related TEAEs of nausea and vomiting. All TEAEs resolved by the end of the study. There were no deaths, SAEs, or TEAEs leading to study discontinuation.

SPONSOR'S CONCLUSIONS
PHARMACOKINETIC

- Total exposure (AUC_{0-t} and AUC_{0-inf}) of delafloxacin was not affected by administration under fed conditions.
- Peak exposure (C_{max}) of delafloxacin was reduced by 20.5% when administered under fed conditions versus fasted conditions
- Median T_{max} of delafloxacin administered under fed conditions was delayed by 1.25 hours compared to administration under fasted conditions.
- Meals fed 2 hours after dosing did not have a statistically significant effect on delafloxacin plasma AUC_{0-inf}, C_{max}, or T_{max} following a 900-mg dose.

SAFETY

- Overall, a single oral dose of 900 mg delafloxacin administered under fasted and fed conditions was safe and well tolerated by the healthy subjects in this study.
- Treatment-emergent AEs were reported by 7 subjects (25.9%) after receiving 900 mg delafloxacin under fasted conditions with a meal 2 hours after dosing, 6 subjects (20.7%) after delafloxacin under fed conditions, and 4 subjects (14.3%) after delafloxacin under fasted conditions. The most frequently reported TEAE overall was diarrhea (5 subjects, 16.7%).
- In subjects who reported TEAEs, the majority had TEAEs that were considered mild and possibly related to study drug.
- With the exception of localized skin reaction (antecubital) in 1 subject that was unrelated to study drug, there were no clinically significant findings noted or TEAEs reported resulting from clinical laboratory assessments, vital sign measurements, physical examination findings, or ECG results.

REVIEWER ASSESSMENT:

After oral administration, food did not seem to affect the overall systemic exposure (AUC_{0-t}, AUC_{0-inf}) of delafloxacin. Therefore, delafloxacin may be administered with or without food.

4.5.16 In Vitro Study Reports

Metabolism

Study R&D/00/552: In Vitro Metabolism of [^{14}C] Abbott-319492 (Delafloxacin) by Rat, Dog, Monkey and Human Liver Microsomes and Hepatocytes

The in vitro metabolism of [^{14}C]Abbott-319492 (delafloxacin) was examined in hepatic subcellular fractions (liver microsomes) and whole cell preparations (hepatocytes) of rat, dog, monkey and human livers.

Experiment designs

Liver microsomes:

Rat liver microsomes were prepared from a male Sprague-Dawley rat (b) (4) Dog liver microsomes were prepared from a male beagle dog (b) (4) . Monkey liver microsomes were prepared from a male cynomolgus monkey (b) (4) Human liver microsomes were prepared from the following donors: 814961, 1110962, 1211961, H#3-96, H#4-96, FRX710, EIX345, GJ 5562 and pooled in equal milligram quantities. All the liver microsomes used in this study were from males.

In all cases, the livers were rapidly chopped into small (6.0 g) pieces and finely minced in a blender. The minced tissues were homogenized in ice-cold 1.15% potassium chloride containing 10 mM potassium phosphate buffer (pH 7.4) using a tissue homogenizer. Microsomes were prepared by differential centrifugation and were stored at -70°C in 0.1 M potassium phosphate buffer (pH 7.4) containing 20% (v/v) glycerol and 1.0 mM EDTA. Microsomal protein concentrations were determined using a bicinchoninic acid (BCA) assay kit procedure (Pierce Chemical) with bovine serum albumin as the standard.

A standard 200 µL incubation mixture contained liver microsomal protein (1 mg/mL) in 50 mM phosphate buffer (pH 7.4), 50 µM EDTA, 2 mM NADPH and Abbott-319492. Following a 5 minute pre-incubation, reactions were started by the addition NADPH and incubated at 37°C in a water bath for 0, 30, and 60 minutes. The reaction were stopped by the addition of 100 µL of a mixture of methanol and acetonitrile (1: 1, v/v), followed by vigorous mixing. For control incubations, at all time points, buffer was substituted for the NADPH and again for microsomal protein. The metabolic marker, [^{14}C] testosterone (200 µM), was incubated with each group of microsomes as positive metabolic controls.

Hepatocyte:

Male Sprague-Dawley rat (DH0200-24, Lot No. 012400), male beagle dog (DH0200-24,Lot No. 011000) and male cynomolgus monkey (MH0200-24, Lot No. 011700) hepatocytes cultured on a collagen substratum and preserved in a solid matrix (Vidacyte Gel)™ were obtained from (b) (4)

(b) (4) Cells were allowed to incubate in the culture medium at 37°C for at least 30 minutes prior to initiating the reaction. [^{14}C]Abbott-319492 (final concentration 5 μM, 0.28 μCi/mL and 25 μM, 0.34 μCi/mL) was added to each well. Cryopreserved human hepatocytes were obtained from the (b) (4) and came from a 34-year old Caucasian male donor (hepatocyte ID HH 110). [^{14}C]Abbott-319492 (final concentrations 5 μM, 0.28 μCi and 25 μM, 0.38 μCi/mL) in 5 μL of DMSO were added. Cell-free control incubations contained substrate in the medium without the hepatocytes. The culture plates were incubated at 37°C with gentle shaking under 5% carbon dioxide/95% air atmosphere for 0, 2, 4, 6 and 24 hours. The reaction was stopped by aspirating the contents of the well into a microcentrifuge tube containing 250 μL of a mixture of methanol and acetonitrile (1: l, v/v) and followed by vigorous mixing. All the samples were kept frozen until HPLC analysis. The metabolic markers, [^{14}C]testosterone (200 μM) and hydroxycoumarin (20 μM), were incubated with each group of hepatocytes as positive metabolic controls.

Results

The results show extensive NADPH-dependent metabolism of [^{14}C]testosterone but little or no metabolism of [^{14}C]Abbott-319492 from rat, dog or human microsomes. [^{14}C]Abbott-3 l 9492 was metabolized by monkey liver rnicrosomes in a NADPH-dependent manner, forming trace amounts of metabolites (M7, M9 and MIO). After a 60 minute incubation M7, M9, and MIO accounted for 1.35, 0.0 and 3.9% and 1.31, 0.8 and 3.71 % of the turnover of [^{14}C]Abbott-319492 at 5 and 25 μM, respectively.

[^{14}C]Abbott-319492 formed several metabolites in the various hepatocytes studied but only at the highest concentration (25 μM) and after a 24 hour incubation period (see Table 1). After incubating for six hours, only two metabolites, M3 and M5 were observed at both concentrations and for all species, except for human. The rate of metabolism was consistently slow across species, except dog, with percentage turnover values of 0, 2.70, 5.32 and 20.37% for human, rat, monkey and dog, respectively at 5 μM for 6 hours (data not shown). Turnover values for Abbott-319492 at 25 μM for 6 hours were similar (0, 1.37, 5.24 and 16.03%) to those seen at the lower substrate concentration. Incubation of [14C]testosterone (200 μM) and hydroxycoumarin (20 μM) with hepatocytes from all species indicated that both oxidative and conjugative reactions were viable.

Table 1. Metabolism of [^{14}C]Abbott-319492 (5 and 25 µM) by Isolated Human Hepatocytes

Conc. (µM)	Incubation Time (h)	Percentage of Total Radioactivity					
		M1	M2	M3	M5	ABT-492	Impurity
5	0	-	-	-	-	100.00	-
5	2	-	-	-	-	100.00	-
5	4	-	-	-	-	100.00	-
5	6	-	-	-	-	100.00	-
5	24	-	-	1.02	-	98.75	-
25	0	-	-	-	-	100.00	-
25	2	-	-	-	-	100.00	-
25	4	-	-	-	-	100.00	-
25	6	-	-	-	-	100.00	-
25	24	-	-	0.94	0.96	98.10	-

-; not detected

Based upon LC/MS fragmentation, metabolites M3 and M5 have been tentatively identified as glucuronide conjugated of Abbott-319492.

Reviewer's comment: It seems that the turnover rates for delafloxacin in both human liver microsomes and hepatocytes were very low. The metabolism of delafloxacin in humans was predominated by the formation of glucuronic acid conjugates of the parent drug. The molecular weight of delafloxacin is 440.764 g/mol. The highest concentration of delafloxacin tested in this study was 25 µM, which is equivalent to about 11 µg/mL. Of note, the free C_{max} of delafloxacin after receiving 300 mg IV single dose was about 1.6 µg/mL (total C_{max} of 10 µg/mL times 16% unbound fraction in plasma).

Transporter Characterization

Study (b) (4)-2011-056: Assessment of Delafloxacin (RX-3341) permeability across Caco-2 monolayer

The permeability of delafloxacin in Caco-2 cells was evaluated. The apparent permeability of delafloxacin in the B→A and A→B directions were 87.6 and 5.06 nm/sec, respectively, leading to an efflux ratio of 17.3. In this study, [3H]-genistein was used as a reference compound. The

mean efflux ratio of genistein transport in Caco-2 cells was 4.20 which was within the criteria for acceptable control values.

Additionally, RX-3341 did not have any effect on the monolayer integrity as indicated by the percentage of inulin appearing in the receiver chamber in the presence of 30, 100 and 300 μM of RX-3341.

Study (b) (4)-2013-073: Assessment of RX-3341 as a substrate of human P-gp, BCRP, OAT1, OAT3, OCT1, OCT2, OATP1B1 and OATP1B3 mediated transport

The purpose of the study was to determine whether RX-3341 is transported by human P-gp, BCRP, OAT1, OAT3, OCT1, OCT2, OATP1B1 and OATP1B3.

Test System

The uptake test system for the transporters represented by the solute carrier (SLC) family, OAT1, OAT3, OCT1, OCT2, OATP1B1 and OATP1B3 was comprised of a polarized monolayer of MDCK-II cells grown on permeable supports. The MDCK-II cells were treated to express the transporter of interest using the (b) (4) transient expression technology or treated with a control vector. For BCRP, the experimental system was comprised of a polarized monolayer of Caco-2 cells grown on permeable supports. The system relied on the endogenous expression of BCRP in the Caco-2 cells. The test system for P-gp transport was comprised of a polarized monolayer of MDCK-MDR1 cells grown on permeable supports.

Statistical analysis

Statistical analysis was performed using an unpaired t-test. A p value of <0.05 was considered statistically significant.

Assay Acceptance

Criteria for acceptable control values for transport:

- OAT1-mediated p-aminohippurate uptake: $\geqslant$0.667 pmol/min/cm^2
- OAT3-mediated p-aminohippurate uptake: $\geqslant$0.287 pmol/min/cm^2
- OCT1-mediated MPP+ uptake: $\geqslant$17.6 pmol/min/cm^2
- OCT2-mediated metformin uptake: $\geqslant$2.03 pmol/min/cm^2
- OATP1B1-mediated estradiol-17β-d-glucuronide uptake: $\geqslant$0.505 pmol/min/cm^2
- OATP1B3- mediated CCK-8 uptake: $\geqslant$0.733 pmol/min/cm^2
- Mean efflux ratio of digoxin transport in MDCK-MDR1 cells: $\geqslant$10.9
- Mean efflux ratio of genistein transport in Caco-2 cells: $\geqslant$4.20

Results

There were no statistically significant differences in uptake of RX-3341 between OAT1, OAT3, OCT1, OCT2, OATP1B1, or OATP1B3 expressing cells and the corresponding controls cells, indicating RX-3341 is not a substrate for these transporters under the study conditions. For Pgp, the observed efflux ratio was 2.10, barely above than the 2.0 threshold for P-gp substrates (Table 2), indicating that RX-3341 may be a P-gp substrate under these conditions. For BCRP, the observed efflux ratio was 8.86 (Table 2), above the threshold of 2.0 for BCRP substrates, suggesting that RX-3341 is a BCRP substrate. Recoveries in P-gp and BCRP assays are between 98.4% -117%, indicating no issues in non-specific binding in these assays.

Table 2. In vitro assay data for the transport of RX-3341 by P-gp and BCRP (Adapted from Table 2 of the study report)

Test Conditions	Papp B->A ($x10^{-6}$ cm/s)	Papp A->B ($x10^{-6}$ cm/s)	Mean Net B->A flux (pmol/hr/cm^2)	Efflux ratio (Papp B->A)/(Papp A->B)
BCRP - 25 genistein	19.2 ± 0.851	2.95 ± 0.115	1.46 ± 0.0766	6.50 ± 0.288
BCRP - 23 µM RX-3341	7.54 ± 0.595	0.851 ± 0.00249	554 ± 49.3	8.86 ± 0.700
P-gp - 100 nM digoxin	6.49 ± 0.0262	0.149 ± 0.00345	2.27 ± 0.00940	43.7 ± 0.176
P-gp - 23 µM RX-3341	1.60 ± 0.0999	0.762 ± 0.125	69.5 ± 8.27	2.10 ± 0.131

Data represent the mean and standard deviation of triplicate samples.

Study (b) (4)148044: In Vitro Evaluation of RX-3341 as a Substrate of Human P-gp and BCRP Transporters

The objective of this study was to evaluate if RX-3341 is a substrate of human efflux transporters (namely, P-gp and BCRP).

P-gp and BCRP are members of the ATP-binding cassette superfamily of transporters and are expressed on the apical membrane of a number of tissues. P-gp and BCRP are expressed in the luminal membrane of enterocytes, endothelial cells in the brain, the brush border membrane of renal proximal tubules and the canalicular membrane of
hepatocytes where they limit the intestinal absorption, blood-brain barrier penetration and facilitate excretion into the bile and urine.

Study Design:
The potential of RX-3341 to be a substrate of human efflux transporters was evaluated as outlined in test system as shown in the following table. MDCKII cells (Madin-Darby Canine Kidney cells over-expressing human MDR1 [P-gp/ABCB1] and BCRP) were used in experiments to determine if RX-3341 is a substrate of P-gp and BCRP. MDCKII-MDR1, MDCKII-BCRP and control MDCKII cells were purchased from the (b) (4)

Transporter	Test system	Experimental design
P-gp	MDCKII-MDR1	Bidirectional permeability of test article in MDCKII-MDR1 and control cells
BCRP	MDCKII-BCRP	Bidirectional permeability of the test article in MDCKII-BCRP and control cells

Drug transport assays were conducted using the reagents listed in the table below.

Transporter	Reagent use	Reagent	Manufacturer
P-gp	Substrate	Digoxin	(b) (4)
	Inhibitor	Valspodar	
BCRP	Substrate	Prazosin	
	Inhibitor	Ko143	

Results

At 1 and 10 µM RX-3341, the permeability (Papp) of RX-3341 was similar in the B-A and A-B directions resulting in an efflux ratio less than 2. At 100 µM RX-3341, the efflux ratio was 2.25 and was reduced to 1.08 in the presence of the P-gp inhibitor valspodar which suggests RX-3341 is possibly a substrate of P-gp. The permeability of RX-3341 ranged from 1.20 to 3.21 × 10−6 cm/sec suggesting RX-3341 is a low permeability compound. On control cells, the permeability and efflux ratio of 10 µM RX-3341 was similar to the permeability and efflux ratio on MDCKII-MDR1 cells.

The efflux ratio of digoxin (10 µM) on MDCKII-MDR1 cells was 22.9 and reduced to −0.808 in the presence of valspodar (complete inhibition) indicating the test system worked as expected.

The permeability (Papp) of RX-3341 was higher in the B-A direction than the A-B direction resulting in efflux ratios of 2.62, 3.86 and 6.41 at 1, 10 and 100 µM, respectively. In the presence of the BCRP inhibitor Ko143, the efflux ratios were reduced to 0.668, 1.53 and 1.50, respectively. The results indicate RX-3341 is a substrate of BCRP. On control cells, the efflux ratio of 10 µM RX-3341 was close to 1 indicating BCRP is responsible for the transport of RX-3341 across MDCKII-BCRP cells.

The efflux ratio of prazosin (1 µM) on MDCKII-BCRP cells was 7.89 and reduced to 0.966 in the presence of Ko143 (complete inhibition) indicating the test system worked as expected.

Reviewer's Comment: Taking together the results of Study (b) (4) 148044, (b) (4) -2013-073, and (b) (4) -2011-056, it seems that delafloxacin may be a weak substrate of P-gp and a substrate of BCRP. Delafloxacin does not seem to be the substrates of OAT1, OAT3, OCT1, OCT2, OAP1B1, and OATP1B3. According to the FDA Draft Guidance for Industry Drug Interaction Studies, additional data are needed to establish clinical relevance of the in vitro data to justify if it is necessary to complete an assessment of nonclinical and clinical information to determine whether an in vivo DDI study is warranted for P-gp and BCRP. In the Summary of Clinical Pharmacology, the Sponsor stated that the absolute bioavailability of delafloxacin is good at 58.8% (to-be-marketed), so any interaction with BCRP or P-gp at the enterocyte would likely not increase peak exposures beyond those exposures shown to be tolerable at doses higher than the proposed 300-mg and 450-mg Q12h doses. Also, safety data are available for higher doses administered over multiple days which provide reassurance that any increase in exposure resulting from BCRP or P-gp inhibition would likely be tolerated. Therefore, a drug-drug

interaction study with delafloxacin and inhibitors of BCRP and P-gp are not warranted and were not conducted. Please see the table below for details.

Mean Systemic Exposure of Delafloxacin After IV and Oral Administration in Clinical Studies With Highest Tolerable Doses (Adapted from Table 57 in Summary of Clinical Pharmacology)

Study No.	Delafloxacin Treatment				AUC_∞ or AUC_τ (µg·h/mL)	C_{max} or $C_{max,ss}$ (µg/mL)
	Route	Formulation	Dose (mg)	Days		
ML-3341-118	Oral	To-be-marketed	450 Q12h	5	22.7	7.45
RX-3341-104	IV	To-be-marketed	300 Q12h	12	23.36	9.29
M00-224	Oral	Phase 1	1600	1	81.7	16.6
	Oral	Phase 1	1200	5	54.3	11.5
RX-3341-108	IV	To-be-marketed	1200	1	67.1	13.2
RX-3341-103	IV	(b) (4)	450 Q12h	14	42.3	15.2

Q12h = every 12 hours.

Note: AUC_τ is 12 hours.

The rationales and clinical results provided by the Sponsor to exempt from conducting in vivo DDI studies for P-gp and BCRP appear reasonable when P-gp and BCRP mediated DDIs occur at the GI tract. However, these two transporters are also expressed in other organs (i.e., liver and kidney), and delafloxacin is eliminated primarily by renal excretion, with biliary excretion as a minor pathway, DDI between delafloxacin and inhibitors of P-gp and BCRP could also occur in these organs. The Sponsor should provide further rationale for not conducting in vivo DDI studies. In addition, the Sponsor should state that BAXDELA was a substrate of the P-gp and BCRP according to the in vitro study results in the proposed labeling.

Study (b) (4)-2011-055: Assessment of RX-3341as a potential inhibitor of human P-gp, BCRP, OAT1, OAT3, OCT1, OCT 2, OATP1B1 and OATP1B3-mediated transport

The purpose of the study was to determine whether RX-3341 inhibits the transport of substrate by P-gp, BCRP, OAT1, OAT3, OCT1, OCT2, OATP1B1 or OATP1B3.

Test Systems

The uptake test system for the transporters represented by the solute carrier (SLC) family, OAT1, OAT3, OCT1, OCT2, OATP1B1, and OATP1B3, was comprised of a polarized monolayer of MDCK-II cells grown on permeable supports. The MDCK-II cells were treated to express the transporter of interest using the (b) (4) transient expression technology or treated with a control vector. For BCRP, the experimental system was comprised of a polarized monolayer of Caco-2 cells grown on permeable supports. The system relied on the endogenous expression of BCRP in the Caco-2 cells and chemical inhibition by 100 µM chrysin. The test system for P-gp transport was comprised of a polarized monolayer of MDCK-MDR1 cells grown

on permeable supports. Chemical inhibition with 50 μM ketoconazole was used to determine the background flux of substrate.

Assay Acceptance

Criteria for acceptable control values for transport:

- OCT1 mediated metformin uptake: >3.70 pmol/min/cm^2.
- OCT2 mediated metformin uptake: >4.38 pmol/min/cm^2.
- OAT1 mediated p-aminohippurate uptake: >0.714 pmol/min/cm^2.
- OAT3 mediated estrone-3-sulfate uptake: >0.620 pmol/min/cm^2.
- OATP1B1 mediated estradiol-17β-d-glucuronide uptake: >0.87 pmol/min/cm^2.
- OATP1B3 mediated bromosulfophthalein uptake: >1.17 pmol/min/cm^2.
- Net B→A flux of digoxin in MDCK-MDR1 cells: >2.39 pmol/hr/cm^2 (with ketoconazole correction)
- Mean efflux ratio of genistein transport in Caco-2 cells: >4.20

Criteria for acceptable control values for reference inhibition:

- Percent inhibition of human OCT1 mediated metformin uptake by 100 μM quinidine: 91.3±15%.
- Percent inhibition of human OCT2 mediated metformin uptake by 100 μM quinidine: 90.9±15%.
- Percent inhibition of human OAT1 mediated p-aminohippurate uptake by 100 μM probenecid: >70%.
- Percent inhibition of human OAT3 mediated estrone-3-sulfate uptake by 100 μM probenecid: 90.9±15%.
- Percent inhibition of human OATP1B1 mediated estradiol-17β-d-glucuronide uptake by 100 μM rifampicin: 9.1±15%.
- Percent inhibition of human OATP1B3 mediated bromosulfophthalein uptake by 100 μM rifampicin: 91.8±15%.
- Percent inhibition of P-gp transport of digoxin by 100 μM verapamil: 93.8±15%.
- Percent inhibition of human BCRP transport of genistein by 100 μM chrysin: 97.3±15%.

Results

The inhibitory effects of RX-3341 on transport of substrate by P-gp, BCRP, OAT1, OAT3, OCT1, OCT2, OATP1B1 and OATP1B3 were investigated. The transport of substrate in the presence of the vehicle control (0.5% DMSO) was compared to the uptake in the presence of 23 μM RX-3341 or a reference inhibitor.

The presence of 23 μM RX-3341 did not significantly decrease the transport of the probe substrates of OAT3, OATP1B1, OATP1B3, P-gp and BCRP (Tables 3, 4, and 5). RX-3341 decreased transport of probe substrates of OCT1, OCT2 and OAT1, with inhibition of 22.6% ($p<0.05$), 58.3% ($p<0.001$), and 11.2% ($p<0.05$), respectively (Table 3).

The positive control for transport assays and the positive control for the inhibition assays satisfied the respective control criteria set by [redacted] (b) (4)

Table 3. In vitro assay data for the inhibition of OCT1, OCT2, OAT1, OAT3, OATP1B1, and OATP1B3-mediated transport by RX-3341 (Adapted from Table 1 in the study report)

Name	Cellular Accumulation (transporter) (pmol/min/cm^2)	Cellular Accumulation (control) (pmol/min/cm^2)	Net Transporter Mediated Cellular Accumulation (pmol/min/cm^2)	Inhibition (%)
OCT1 - vehicle	5.65 ± 0.358	1.94 ± 0.158	3.72 ± 0.358	0.00 ± 9.64
OCT1 - 23 µM RX-3341	5.02 ± 0.205	2.15 ± 0.020	2.87 ± 0.205	22.6 ± 5.53
OCT1 - 100 µM Quinidine	2.73 ± 0.287	2.38 ± 0.245	0.35 ± 0.287	90.6 ± 7.72
OCT2 - vehicle	10.3 ± 0.326	3.46 ± 0.306	6.87 ± 0.326	0.00 ± 4.74
OCT2 - 23 µM RX-3341	6.24 ± 0.684	3.38 ± 0.199	2.86 ± 0.684	58.3 ± 9.96
OCT2 - 100 µM Quinidine	3.98 ± 0.391	3.07 ± 0.353	0.911 ± 0.391	86.7 ± 5.69
OAT1 - vehicle	1.68 ± 0.0500	0.332 ± 0.0200	1.35 ± 0.0500	0.00 ± 3.70
OAT1 - 23 µM RX-3341	1.52 ± 0.0709	0.321 ± 0.0117	1.20 ± 0.071	11.2 ± 5.25
OAT1 - 100 µM Probenecid	0.392 ± 0.0438	0.314 ± 0.0303	0.0776 ± 0.0438	94.3 ± 3.24
OAT3 - vehicle	1.16 ± 0.0473	0.315 ± 0.0100	0.846 ± 0.0473	0.00 ± 5.59
OAT3 - 23 µM RX-3341	1.12 ± 0.0736	0.366 ± 0.0434	0.75 ± 0.0736	11.4 ± 8.70
OAT3 - 100 µM Probenecid	0.472 ± 0.0694	0.323 ± 0.0159	0.1490 ± 0.0694	82.4 ± 8.21
OATP1B1 - vehicle	1.72 ± 0.0833	0.322 ± 0.00757	1.40 ± 0.0833	0.00 ± 5.97
OATP1B1 - 23 µM RX-3341	1.48 ± 0.128	0.312 ± 0.0540	1.16 ± 0.128	16.5 ± 9.17
OATP1B1 - 100 µM rifampicin	0.384 ± 0.0414	0.309 ± 0.0289	0.0752 ± 0.0414	94.6 ± 2.96
OATP1B3 - vehicle	3.07 ± 0.0751	1.69 ± 0.136	1.38 ± 0.0751	0.00 ± 5.44
OATP1B3 - 23 µM RX-3341	3.02 ± 0.0879	1.78 ± 0.117	1.24 ± 0.0879	10.3 ± 6.37
OATP1B3 - 100 µM rifampicin	1.72 ± 0.0726	1.79 ± 0.055	-0.0727 ± 0.0726	105 ± 5.26

Probe substrate for each transporter is 10 µM metformin (OCT1 and OCT2), 2 µM p-aminohippurate (OAT1), 750 nM estrone-3-sulfate (OAT3), 2 µM estradiol-17β-d-glucuronide (OATP1B1), 2 µM bromosulfophthalein (OATP1B3).

Table 4. In vitro assay data for the inhibition of P-gp-mediated transport by RX-3341 (Adapted from Table 2 in the study report)

Name	Papp B->A (nm/s)	Papp A->B (nm/s)	Efflux ratio (B->A)/(A->B)	Mean Net B->A flux (with background subtracted) (pmol/hr/cm^2)	Inhibition (%)
MDCK-MDR1					
100 nM digoxin	74.6 ± 2.74	4.19 ± 0.0813	17.8	2.39 ± 0.100	0.00 ± 4.19
100 nM digoxin + 23 µM RX-3341	69.4 ± 3.61	4.23 ± 0.250	16.4	2.20 ± 0.121	7.79 ± 5.06
100 nM digoxin + 100 µM verapamil	22.7 ± 0.345	18.2 ± 4.75	1.25	0.0211 ± 0.182	99.1 ± 7.62
100 nM digoxin + 50 µM ketoconazole	23.5 ± 2.15	19.6 ± 0.594	1.20	0.00 ± 0.0976	100 ± 4.09

Table 5. In vitro assay data for the inhibition of BCRP-mediated transport by RX-3341

Name	Papp B->A (nm/s)	Papp A->B (nm/s)	Efflux ratio (B->A)/(A->B)	Mean Net B->A flux (pmol/hr/cm^2)	Inhibition (%)
Caco2 (BCRP)					
25 nM genistein + vehicle	593 ± 28.1	91.1 ± 3.37	6.51	4.52 ± 0.265	0.00 ± 5.86
25 nM genistein + 23 µM RX-3341	557 ± 28.1	79.2 ± 2.83	7.03	4.30 ± 0.268	4.88 ± 5.93
25 nM genistein + 100 µM chrysin	364 ± 19.0	340 ± 13.6	1.07	0.210 ± 0.293	95.4 ± 6.47

Reviewer's comment: The concentration of delafloxacin tested in this study was 23 µM (equivalent to about 10 µg/mL), which is approximately 6-fold higher than the free C_{max} of delafloxacin after receiving 300 mg IV single dose (1.6 µg/mL = total C_{max} of 10 µg/mL times 16% unbound fraction in plasma).

According to the FDA Draft Guidance, the criteria to decide whether the investigational drug is an inhibitor of OAT1, OCT2, or OAT3 is whether the uptake of model substrates decreases with increased concentrations of the investigational drug. However, only one concentration was used in this study.

Study (b) (4) 2013-072: Assessment of RX-3341 as a potential inhibitor of human OCT 2 and BSEP mediated transport

The purpose of the study was to determine whether RX-3341 inhibits the transport of substrate by BSEP, and determine the IC_{50} value of RX-3341 against the transport of substrate by OCT2.

Test Systems

The uptake test system for the transporters represented by the solute carrier (SLC) family, OCT2 was comprised of a polarized monolayer of MDCK-II cells grown on permeable supports. The MDCK-II cells were treated to express the transporter of interest using the (b) (4) transient expression technology or treated with a control vector. For BSEP, the experimental system was comprised of Sf9 membrane vesicles containing human BSEP. The vesicles were applied as a suspension onto a 96-well flat-bottom assay plate, and were separated from the reaction mixture by a 96-well glass-fiber filtration plate.

Assay Acceptance

Criteria for acceptable control values for transport:

- OCT2 mediated metformin uptake: ⩾2.03 pmol/min/cm^2.
- BSEP mediated taurocholate transport (ATP dependent) >7.45 pmol/min/mg protein.

Criteria for acceptable control values for reference inhibition:

- Percent inhibition of OCT2 mediated metformin uptake by 100 μM quinidine: ⩾65.4%.
- Percent inhibition of BSEP mediated taurocholate transport by 300 μM rifampicin: ⩾ 82.1%.

Results:

The presence of 23 μM RX-3341 did not significantly decrease the transport of the probe substrate of BSEP (Table 6). RX-3341 was studied in the concentration range of 1 - 400 μM against OCT2-mediated transport of probe substrate. Due to lack of sufficient inhibition, IC_{50} value cannot be determined (Table 7).

The positive control for transport assays and the positive control for the inhibition assays satisfied the respective control criteria set by (b) (4)

Table 6. In vitro assay data for the inhibition of BSEP-mediated transport by RX-3341

Test Conditions	Vesicular Accumulation (ATP) (pmol/min/mg)	Vesicular Accumulation (AMP) (pmol/min/mg)	Net Transporter Mediated Vesicular Accumulation (pmol/min/mg)	Inhibition (%)
vehicle	8.55 ± 0.335	0.838 ± 0.0902	7.71 ± 0.335	-
300 µM rifampicin	0.781 ± 0.0498	0.613 ± 0.0177	0.168 ± 0.0498	97.8 ± 0.645
23 µM RX-3341	8.13 ± 0.0202	0.763 ± 0.0241	7.36 ± 0.0202	4.52 ± 0.262

Probe substrate was 1 µM taurocholate.
Data represent the mean and standard deviation of triplicate samples.

Table 7. In vitro assay data for the determination of IC_{50} value of RX-3341 against OCT2-mediated transport of probe substrate.

RX-3341 (µM)	Cellular Accumulation (transporter) (pmol/min/cm^2)	Cellular Accumulation (control) (pmol/min/cm^2)	Net Transporter Mediated Cellular Accumulation (pmol/min/cm^2)	Inhibition (%)
0	4.89 ± 0.738	0.524 ± 0.0683	4.37 ± 0.738	-
1	4.34 ± 0.402	0.398 ± 0.102	3.94 ± 0.402	9.74 ± 9.21
3	4.12 ± 0.235	0.430 ± 0.0683	3.69 ± 0.235	15.6 ± 5.38
20	4.77 ± 0.253	0.514 ± 0.014	4.26 ± 0.253	2.53 ± 5.78
30	3.92 ± 0.491	0.421 ± 0.121	3.50 ± 0.491	19.9 ± 11.2
100	3.43 ± 0.233	0.369 ± 0.0504	3.06 ± 0.233	30.0 ± 5.33
400	3.69 ± 0.301	0.361 ± 0.00700	3.33 ± 0.301	23.9 ± 6.90
100 µM Quinidine	1.69 ± 0.156	0.510 ± 0.0699	1.18 ± 0.156	73.0 ± 3.57

Probe substrate was 10 µM metformin.
Data represent the mean and standard deviation of triplicate samples.

Study [(b) (4)]-2013-102: Assessment of RX-3341 as a potential inhibitor of human OCT2 mediated transport

The purpose of the study was to determine the IC_{50} value of RX-3341 against the transport of substrate by OCT2.

Test Systems

The uptake test system for the transporters represented by the solute carrier (SLC) family, OCT2 was comprised of a polarized monolayer of MDCK-II cells grown on permeable supports. The MDCK-II cells were treated to express the transporter of interest using the [(b) (4)] transient expression technology or treated with a control vector.

Assay Acceptance

- Criteria for acceptable control values for transport (24-well): OCT2 mediated metformin uptake: ⩾4.38 pmol/min/cm^2.
- Criteria for acceptable control values for reference inhibition (24-well): Percent inhibition of OCT2 mediated metformin uptake by 100 µM quinidine: ⩾75.9%.

Results

RX-3341 was studied in the concentration range of 1 - 400 µM against OCT2-mediated transport of probe substrate with or without pre-incubation of RX-3341 with cells. The study was performed in 24-well format.

- With the preincubation (15 min) of RX-3341 with cells, the maximum inhibition observed was 92.7% at 400 µM RX-3341 (Table 8). The IC_{50} value was determined to be 80.7 µM (Figure 1).
- Without the preincubation of RX-3341 with cells, the maximum inhibition observed was 46.3% at 400 µM RX-3341 (Table 9). Due to lack of sufficient inhibition, IC_{50} value cannot be determined (Figure 2).

The positive control for transport assays and the positive control for the inhibition assays satisfied the respective control criteria set by [redacted] (b) (4)

Table 8. In vitro assay data for the inhibition of OCT2-mediated transport by RX-3341 (with 15-min preincubation of cells with RX-3341) (Adapted from Table 1 in the study report)

RX-3341 (µM)	Cellular Accumulation (transporter) (pmol/min/cm^2)	Cellular Accumulation (control) (pmol/min/cm^2)	Net Transporter Mediated Cellular Accumulation (pmol/min/cm^2)	Inhibition (%)
0	8.85 ± 0.218	1.71 ± 0.195	7.14 ± 0.218	0.00 ± 3.05
1	7.80 ± 0.705	1.86 ± 0.460	5.93 ± 0.705	16.9 ± 9.88
3	7.34 ± 1.27	1.79 ± 0.145	5.55 ± 1.27	22.2 ± 17.8
20	8.00 ± 1.50	1.99 ± 0.0820	6.01 ± 1.50	15.8 ± 21.00
30	5.75 ± 0.339	1.60 ± 0.216	4.15 ± 0.339	41.8 ± 4.75
100	4.66 ± 0.437	1.59 ± 0.492	3.07 ± 0.437	57.0 ± 6.13
400	2.54 ± 0.548	2.02 ± 0.284	0.519 ± 0.548	92.7 ± 7.68
100 µM Quinidine	2.60 ± 0.208	2.35 ± 0.337	0.248 ± 0.208	96.5 ± 2.92

Probe substrate was 10 µM metformin.
Data represent the mean and standard deviation of triplicate samples.

Table 9. In vitro assay data for the inhibition OCT2-mediated transport by RX-3341 (without preincubation of cells with RX-3341) (Adapted from Table 2 in the study report)

RX-3341 (µM)	Cellular Accumulation (transporter) (pmol/min/cm^2)	Cellular Accumulation (control) (pmol/min/cm^2)	Net Transporter Mediated Cellular Accumulation (pmol/min/cm^2)	Inhibition (%)
0	7.31 ± 1.50	1.40 ± 0.215	5.91 ± 1.50	0.00 ± 25.3
1	6.99 ± 1.14	1.20 ± 0.305	5.79 ± 1.14	2.08 ± 19.3
3	7.94 ± 0.978	1.29 ± 0.253	6.65 ± 0.978	-12.5 ± 16.5
20	6.97 ± 0.238	1.54 ± 0.208	5.42 ± 0.238	8.30 ± 4.03
30	6.59 ± 1.70	1.41 ± 0.0812	5.17 ± 1.70	12.5 ± 28.7
100	5.32 ± 0.524	0.950 ± 0.246	4.37 ± 0.524	26.0 ± 8.86
400	4.72 ± 0.461	1.55 ± 0.314	3.17 ± 0.461	46.3 ± 7.80
100 µM Quinidine	5.04 ± 0.151	1.62 ± 0.102	3.42 ± 0.151	42.2 ± 2.56

Probe substrate was 10 µM metformin.
Data represent the mean and standard deviation of triplicate samples.

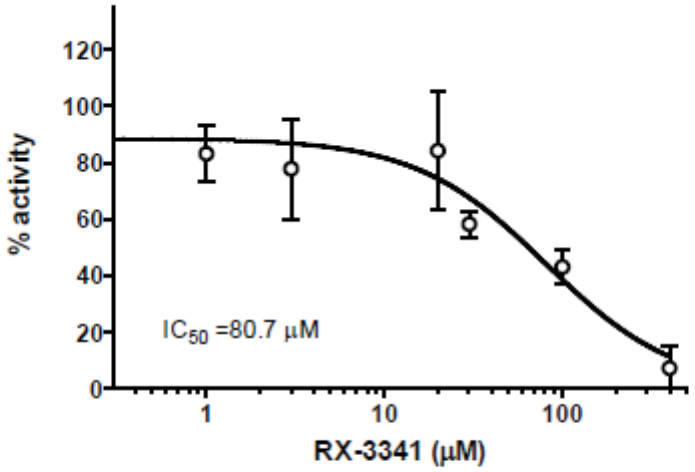


Figure 1 **Determination of IC_{50} of RX-3341 against OCT2-mediated transport of probe substrate (with preincubation of RX-3341 with cells).** RX-3341 was tested in the concentration range of 1 – 400 µM. The probe substrate was 10 µM metformin. The IC_{50} was determined to be 80.7 µM . Data represent the mean and standard deviation of triplicate samples.

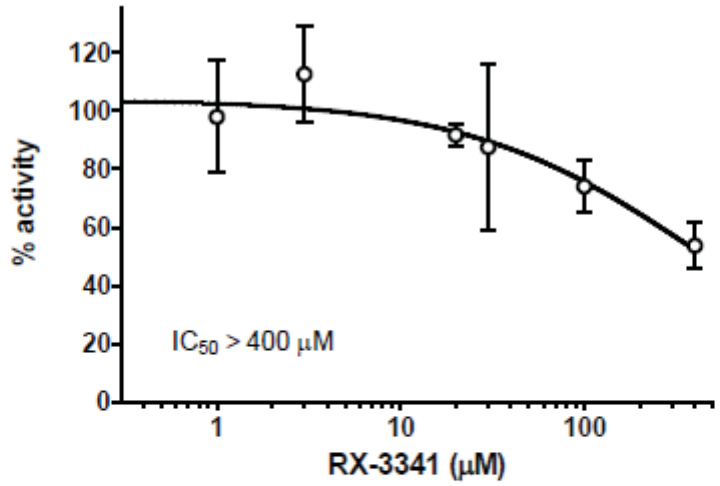


Figure 2 **Determination of IC_{50} of RX-3341 against OCT2-mediated transport of probe substrate (without preincubation of RX-3341 with cells).** RX-3341 was tested in the concentration range of 1 – 400 µM. The probe substrate was 10 µM metformin. Due to lack of sufficient inhibition, the IC_{50} value cannot be determined. Data represent the mean and standard deviation of triplicate samples.

Reviewer's Comment: Taken the results from Study (b) (4)-2011-055, (b) (4)-2013-072, and (b) (4)-2013-102 together, it seems that delafloxacin is not the inhibitor of P-gp, BCRP, OATP1B1,

OATP1B3 but decreased transport of probe substrates of OCT1, OCT2 and OAT1, with inhibition of 22.6% ($p<0.05$), 58.3% ($p<0.001$), and 11.2% ($p<0.05$), respectively, at the concentration of 10 µg/mL free drug. Delafloxacin also decreased transport of probe substrate of OAT3, with inhibition of 11.4% without statistical significance ($p>0.05$). The IC_{50} of OCT2 was 87 µM (38 µg/mL). Of note, delafloxacin $C_{max,ss}$ following 300 mg BID IV dose is approximately 9.3 µg/mL and the unbound fraction is 16%. Therefore, the unbound C_{max}/IC_{50} = 1.5/38=0.04<0.1. Therefore, in vivo DDI study is not needed based on the FDA DDI guidance. However, the Sponsor did not determine the IC_{50} for OCT1 and OAT1, although their IC_{50} may be higher than the one of OCT2 based on study (b) (4) *-2011-055.* Note: The Sponsor stated that "BAXDELA was not an inhibitor of the following hepatic and renal transporters in vitro at clinically relevant concentrations: MDR1, BCRP, OAT1, OAT3, OATP1B1, OATP1B3, BSEP, OCT1 and OCT2."

CYP inhibition and Induction

Study MWQ3034-10: The effect of WQ-3034 (Delafloxacin) and ciprofloxacin on the cytochrome P450 1A2 dependent monooxygenase activity in human liver microsomes

The purpose of this study was to examine the effect of WQ-3034 and ciprofloxacin on CYP 1A2 dependent monooxygenase activity in human liver microsomes.

Test System:

Transplant quality human liver tissue was obtained from the (b) (4) The liver was rapidly chopped into small (6.0 g) pieces and finely minced with a hand held razor blade or in a blender. The minced tissue was homogenized in ice-cold 1.15% potassium chloride containing 10 mM potassium phosphate buffer (pH 7.4) using a tissue homogenizer. Microsomes were prepared by differential centrifugation and were stored at -70°C in 0.1 M potassium phosphate buffer (pH 7.4), 20% (v/v) glycerol and
1.0 mM EDT A. Microsomal protein concentrations were determined using a bicinchoninic acid (BCA) assay kit procedure (Pierce Chemical), with bovine serum albumin as the standard.

Results

WQ-3034 at 0.5, 5, 50 and 500 µM did not inhibit CYP 1A2 (Table 10).

Table 10 CYP 1A2 activity with WQ-3034 and ciprofloxacin

CYP Isoform	Assay	% Control Activity* WQ-3034 Conc. 0.5 µM	5 µM	50 µM	500 µM
1A2	Phenacetin *O*-deethylation	101.7 ± 9	104.7 ± 8	101.3 ± 11	110.3 ± 6
		% Control Activity* Ciprofloxacin Conc. 0.5 µM	5 µM	50 µM	500 µM
1A2	Phenacetin *O*-deethylation	99.7 ± 3	100.7 ± 3	80.6 ± 1	70.2 ± 9

* = Mean ± SD; n = 3

Study R&D/01/136: The Effect of Abbott-319492 (Delafloxacin) on the Cytochrome P450-Dependent Monooxygenase Activities in Human Liver Microsomes

Test System

Transplant quality human liver tissue was obtained from the (b) (4) . The liver was rapidly chopped into small (6.0 g) pieces and finely minced with a hand-held razor blade or in a blender. The minced tissue was homogenized in ice-cold 1.15% potassium chlori.de containing 10 mM potassium phosphate buffer (pH 7.4) using a tissue homogenizer. Microsomes were prepared by differential centrifugation and were stored at -70°C in 0.1 M potassium phosphate buffer (pH 7.4), 20% (v/v) glycerol or 250 mM sucrose and 1.0 mM EDTA. Microsomal protein concentrations were determined using a bicinchoninic acid (BCA) assay kit procedure (Pierce Chemical), with bovine serum albumin as the standard.

Please refer to Table 11 below for the substrates used in this study.

Table 11. Isoform-Specific CYP Assays - Assay Conditions and Analytical Methods

Isoform	Activity	Substrate µM	Protein mg/mL	Incubation (mins)	Known inhibitor	Sample preparation	Analyte	Analytical method
CYP2A6	Coumarin 7-hydroxylase	5	0.1	Continuous	8-Methoxy-psoralen; 5 µM	Continuous assay Ex: 360 nm Em: 450 nm	7-Hydroxy-coumarin	Microtitre plate fluorescence assay
CYP2C9	Tolbutamine hydroxylase	100	0.6	60	Sulfaphenazole 2 µM	Protein precipitation with in acetonitrile/ methanol	Hydroxy tolbutamide	Radio-HPLC
CYP2C19	S-Mephenytoin 4'-hydroxylase	73.4	1.0	45	Tranylcypromine 30 µM	Protein precipitation with acetonitrile/ methanol	4'-Hydroxy S-mephenytion	Radio-HPLC
CYP2D6	Dextromethorphan *O*-demethylase	20	0.5	20	Quinidine 0.5 µM	Basification; charcoal adsorption	[^{14}C]HCHO	Liquid scintillation
CYP2E1	Chlorzoxazone 6-hydroxylase	100	1.0	10	DDC 20 µM	Protein precipitation with acetonitrile/ methanol	6-Hydroxy-chlorzoxazone	Radio-HPLC
CYP3A4	Benzyloxyquinolone *O*-dealkylation	40	0.3	Continuous	Ketoconazole	Continuous assay Ex: 409 nm Em: 550 nm	7-Hydroxy-quinoline	Microtitre plate fluorescence assay

Results

The effect of Abbott-319492 on isoform-specific cytochrome P450-dependent monooxygenase activities in human liver microsomes was examined. Over the range of concentrations studied (0.5 to 500 µM), Abbott-319492 resulted in <16% inhibition ofCYP1A2, CYP2C9, CYP2C19, CYP2D6 and CYP3A. Significant inhibition ofCYP2A6 (76%) and activation ofCYP2El (368%) were observed at a concentration of 500 µM (Table 12). The Sponsor stated that the concentration of 500 µM is way higher than the Cmax with therapeutic dose. Hence, clinically significant drug-drug interactions due to inhibition or activation of CYP-mediated biotransformations of co-administered drugs by Abbott-319492 are unlikely in humans.

Table 12. Activity of CYP Isoforms with Abbott-319492

		% Control Activity*			
		Abbott-319492			
CYP Isoform	Assay	0.5 µM	5.0 µM	50 µM	500 µM
1A2	Phenacetin *O*-deethylation	101.7 ± 9.8	104.7 ± 8.0	101.3 ± 11.1	110.3 ± 6.0
2A6¶	Coumarin 7-hydroxylation	95.7 ± 2.0	90.4 ± 1.6	77.0 ± 0.5	24.3 ± 0.9
2C9	Tolbutamide hydroxylation	108.3 ± 7.9	110.9 ± 8.9	105.1 ± 9.9	91.3 ± 5.4
2C19	*S*-Mephenytoin 4'-hydroxylation	103.8 ± 10.2	107.2 ± 6.5	96.9 ± 7.8	85.0 ± 4.5
2D6	Dextromethorphan *O*-demethylation	101.1 ± 6.9	100.3 ± 6.7	107.5 ± 9.0	133.8 ± 10.5
2E1	Chlorozoxazone 6-hydroxylation	83.9 ± 3.8	118.7 ± 14.8	124.9 ± 7.4	368.3 ± 22.3
3A†	Benzyloxyquinolone *O*-dealkylation	97.4 ± 14.4	105.5 ± 13.6	110.5 ± 5.8	120.6 ± 17.6

* = Mean ± SD; (n = 3), ¶; (n=6), †; (n=8)

Study 418N-1301: Evaluation of the Inhibitory Potential of Delafloxacin Towards CYP2B6, CYP2C8, and CYP2A6 Using Human Liver Microsomes

The objective of this study was to evaluate the inhibitory potential of delafloxacin towards CYP2A6, CYP2B6, and CYP2C8. For CYP2B6 and CYP2C8, the concentration of delafloxacin that inhibits activity by 50% (IC_{50}) was to be determined and the inhibition constant (Ki) was to be determined for CYP2A6.

Test System

Pooled human hepatic microsomes (Lot No. 1210223, [redacted] were purchased from [redacted] Upon receipt, the liver microsomes were stored at approximately –70 °C. Product data sheets are included in the study file. Positive control inhibitors (tranylcypromine and quercetin), substrates (coumarin, bupropion, and amodiaquine), NADPH (β-Nicotinamide Adenine Dinucleotide Phosphate), antipyrine, 2-benzoxazolinone, and the reagents needed to prepare buffers were purchased from [redacted] Metabolite standards 7-hydroxycoumarin, hydroxybupropion, and N-desethylamodiaquine were purchased from [redacted] U-bottom assay plates (96-well) were purchased from [redacted]

Please refer to Table 13 for the summary of probes, positive controls used in this study.

Table 13 Summary of CYP450 Substrates, Metabolites, and Positive Controls

CYP Isoform	Substrate	Final Concentration of Substrate	Metabolite Analyzed	Positive Control	Final Highest Concentration of Positive Control
2A6	Coumarin	0.4 µM	7-Hydroxycoumarin	Tranylcypromine	10 µM
2B6	Bupropion	80 µM	Hydroxybupropion	Tranylcypromine	100 µM
2C8	Amodiaquine	20 µM	N-desethylamodiaquine	Quercetin	10 µM

Results

The inhibitory activity was measured using 10 concentrations of delafloxacin in triplicate (0, 0.0457, 0.137, 0.410, 1.23, 3.70, 11.1, 33.3, 100 and 300 µM) for CYP2B6 and CYP2C8 IC_{50} determination. There was no change in activity when compared to the vehicle control for CYP2B6 and CYP2C8 (Table 14).

The inhibitory activity was measured using eight concentrations of delafloxacin in triplicate (0, 0.410, 1.23, 3.70, 11.1, 33.3, 100 and 300 µM) and at three different concentrations of the substrate, coumarin (0.25× Km, at Km and 4× Km) for CYP2A6 Ki determination. There was no change in activity when compared to the vehicle control for CYP2A6 and therefore the Ki value could not be determined (Table 15).

The mean IC_{50} values for the positive controls, tranylcypromine (for CYP2A6 and CYP2B6) and quercetin (for CYP2C8) were 0.02, 4.63, and 1.79 µM for CYP2A6, CYP2B6, and CYP2C8, respectively. The positive controls were determined to be potent CYP450 inhibitors using selected probe substrates. The calculated IC_{50} values for positive controls were consistent with reported literature values.

Please refer to Tables 16 and 17 for the effects of positive controls on CYP2B6, CYP2C8, and CYP2A6.

Table 14 Summary of the Effect of Delafloxacin on CYP2B6 and CYP2C8 Isozyme Activities (Adapted from Table 2 in the study report)

	CYP450 Activity Expressed as % Vehicle Control	
Delafloxacin Concentration (µM)	**CYP2B6**	**CYP2C8**
300	95	94
100	91	98
33.3	98	97
11.1	97	97
3.70	98	104
1.23	97	109
0.410	98	98
0.137	106	102
0.0457	100	102
0 (VC)	100	100
IC50 (µM)	>300	>300

VC = Vehicle Control
CYP450 activity was defined as rate of metabolite formed (pmol/min/mg protein) at each delafloxacin concentration, expressed as a percent of vehicle control (0 µM).

Table 15. Summary of the Effect of Delafloxacin on CYP2A6 Isozyme Activities (Adapted from Table 3 in the study report)

	CYP450 Activity Expressed as % Vehicle Control		
Delafloxacin Concentration (µM)	**CYP2A6 (substrate - 0.1 µM)**	**CYP2A6 (substrate - 0.4 µM)**	**CYP2A6 (substrate - 1.6 µM)**
300	101	101	97
100	97	98	101
33.3	101	99	106
11.1	98	99	102
3.70	104	101	102
1.23	100	97	106
0.410	102	97	107
0 (VC)	100	100	100
IC50 (µM)	>300	>300	>300

VC = Vehicle Control
CYP450 activity was defined as rate of metabolite formed (pmol/min/mg protein) at each delafloxacin concentration, expressed as a percent of vehicle control (0 µM).

Table 16. Summary of Inhibition of CYP450 Activity by Positive Controls (Concentration 0 to 10 µM) for CYP2A6 and CYP2C8 (Adapted from Table 4 in the study report)

	CYP450 Activity Expressed as % Vehicle Control	
Positive Control Concentration (µM)	**CYP2A6**	**CYP2C8**
10	0	20
3.33	0	28
1.11	0	66
0.37	3	93
0.123	12	97
0.042	34	108
0.0137	63	100
0.00456	NA	102
0.0015	NA	104
0 (VC)	100	100
IC_{50} (µM)	0.02	1.79
Inhibitor	Tranylcypromine	Quercetin

VC = Vehicle control
CYP450 activity was defined as rate of the metabolite formed (pmol/min/mg protein) at each inhibitor concentration, expressed as a percent of vehicle control (0 µM).

Table 17. Summary of Inhibition of CYP450 Activity by Positive Controls (Concentration 0 to 100 μM) for CYP2B6

	CYP450 Activity Expressed as % VC
Positive Control Concentration (μM)	**CYP2B6**
100	2
33.3	10
11.1	26
3.7	53
1.23	79
0.42	94
0.137	93
0.046	93
0.015	93
0 (VC)	100
IC50 (μM)	4.63
Inhibitor	Tranylcypromine

VC = Vehicle control
CYP450 activity was defined as the rate of the metabolite formed (pmol/min/mg protein) at each inhibitor concentration, expressed as a percent of vehicle control (0 μM).

Reviewer's Comment: Taken the results from Studies MWQ3034-10, R&D/01/136, and 418N-1301 together, it seems that delafloxacin does not inhibit CYP1A2, CYP2A6, CYP2B6, CYP2C8, CYP2C9, CYP2C19, CYP2D6, CYP2E1 and CYP3A4/5 in vitro in human liver microsomes at clinically relevant concentration. However, at the concentration of 500 μM, delafloxacin showed inhibition towards CYP2E1.

Study (b) (4) 133033: In Vitro Evaluation of Delafloxacin (RX-3341) as an Inducer of Cytochrome P450 Expression in Cultured Human Hepatocytes

Test System

Three preparations of cryopreserved human hepatocytes (hereafter, referred to as HC10-1, HC3-22 and HC5-25, respectively) supplied internally by (b) (4) were treated in this study.

Please refer to the table below for the positive control and vehicles.

Chemical	Catalog number	Lot number	Storage conditions [a]	Vehicle	Purity	Supplier
DMSO	D2650	RNBC1326 RNBB9702 RNBC5969	Room temperature	Not applicable	Not applicable	(b) (4)
Flumazenil	F6300	031M1334V	−20 ± 5 °C	DMSO	100%	
Omeprazole	O104	BCBF7621V	2 to 8 °C	DMSO	99.7%	
Phenobarbital	P5178	061M1313V	Room temperature	DMSO	99.8%	
Rifampin	R3501	011M1189V	−20 ± 5 °C	DMSO	98%	

a Storage conditions of neat compound.

Result

The effect of RX-3341 on human CYP1A2 activity, mRNA levels and EC_{50} and E_{max}

Determination of phenacetin O-dealkylation (CYP1A2 activity): In cultured human hepatocytes, phenacetin O-dealkylation is catalyzed by CYP1A2, which is the major omeprazole-inducible CYP enzyme. The effects of treating cultured human hepatocytes with RX-3341 on phenacetin O-dealkylase (CYP1A2) activity are shown in Table 18. Treatment of cultured human hepatocytes once daily for three consecutive days with omeprazole caused increases ranging from 5.09- to 42.1-fold change in phenacetin O-dealkylation (CYP1A2 activity). Treatment with the non-inducer, flumazenil, had little or no effect on CYP1A2 activity with changes ranging from 1.00- to 1.41-fold change.

Table 18 CYP1A2 activity fold increase: The effect of treating cultured human hepatocytes with RX-3341 or prototypical inducers on the rate of phenacetin O-dealkylase activity (Adapted from Figure 3 in the study report)

Activity Fold Increase		HC10-1	HC3-22	HC5-25
Phenacetin *O*-dealkylation (CYP1A2)	0.1% DMSO	0	0	0
	1 µM RX-3341	-0.0481	NT	-0.0557
	5 µM RX-3341	-0.0625	NT	-0.0579
	10 µM RX-3341	-0.115	0.0424	-0.0413
	25 µM RX-3341	-0.213	-0.00955	-0.141
	50 µM RX-3341	-0.304	-0.0637	-0.167
	100 µM RX-3341	-0.486	-0.249	-0.101
	250 µM RX-3341	-0.642	-0.623	-0.349
	500 µM RX-3341	NT	-0.720	NT
	1000 µM RX-3341	NT	-0.830	NT
	25 µM Flumazenil	0.406	0.00240	0.217
	50 µM Omeprazole	4.09	9.18	41.1

NT Not tested

Fold increase = fold change − 1

Determination of CYP1A2 mRNA levels: The effects of treating cultured human hepatocytes with RX-3341 on CYP1A2 mRNA expression are shown in Table 19. Treatment of cultured human hepatocytes once daily for three consecutive days with omeprazole caused increases

ranging from 40.7- to 56.1-fold change in CYP1A2 mRNA levels. Treatment with the non-inducer, flumazenil, had little or no effect on CYP1A2 mRNA levels with changes ranging from 1.15- to 3.05-fold change.

Table 19. CYP1A2 mRNA fold increase: The effect of treating cultured human hepatocytes with RX-3341 on CYP1A2 mRNA levels (Adapted from Figure 7 in the study report)

mRNA Fold Increase		HC10-1	HC3-22	HC5-25
CYP1A2	0.1% DMSO	0	0	0
	1 µM RX-3341	0.338	NT	-0.112
	5 µM RX-3341	0.0920	NT	-0.0450
	10 µM RX-3341	-0.0850	0.0440	-0.227
	25 µM RX-3341	-0.476	-0.254	-0.223
	50 µM RX-3341	-0.658	-0.478	-0.349
	100 µM RX-3341	-0.477	-0.733	-0.473
	250 µM RX-3341	-0.480	-0.789	-0.888
	500 µM RX-3341	NT	-0.669	NT
	1000 µM RX-3341	NT	-0.709	NT
	25 µM Flumazenil	2.05	0.150	0.534
	50 µM Omeprazole	39.7	49.4	55.1

NT Not tested
mRNA fold increase = fold change − 1

The effect of RX-3341 on human CYP2B6 activity, mRNA levels and EC50 and Emax
Determination of bupropion hydroxylation (CYP2B6 activity): In cultured human hepatocytes, bupropion hydroxylation is catalyzed by CYP2B6, which is inducible by phenobarbital. The effects of treating cultured human hepatocytes with RX-3341 on bupropion hydroxylase (CYP2B6) activity are shown in Table 20. Treatment of cultured human hepatocytes once daily for three consecutive days with phenobarbital caused increases ranging from 4.78- to 8.09-fold change in bupropion hydroxylation (CYP2B6 activity). Treatment with the non-inducer, flumazenil, had little or no effect on CYP2B6 activity with changes ranging from 1.04- to 1.21-fold change.

Table 20 CYP2B6 activity fold increase: The effect of treating cultured human hepatocytes with RX-3341 or prototypical inducers on the rate of bupropion hydroxylase activity (Adapted from Figure 11 from study report)

Activity Fold Increase		HC10-1	HC3-22	HC5-25
Bupropion hydroxylation (CYP2B6)	0.1% DMSO	0	0	0
	1 µM RX-3341	-0.0910	NT	0.0457
	5 µM RX-3341	-0.183	NT	0.101
	10 µM RX-3341	-0.161	0.0702	0.120
	25 µM RX-3341	-0.327	-0.0184	0.0409
	50 µM RX-3341	-0.296	0.138	0.0149
	100 µM RX-3341	-0.544	0.297	-0.103
	250 µM RX-3341	-0.716	-0.481	-0.636
	500 µM RX-3341	NT	-0.709	NT
	1000 µM RX-3341	NT	-0.886	NT
	25 µM Flumazenil	0.209	0.0600	0.0409
	750 µM Phenobarbital	3.78	7.09	6.37

NT Not tested

Fold increase = fold change − 1

Determination of CYP2B6 mRNA levels: The effects of treating cultured human hepatocytes with RX-3341 on CYP2B6 mRNA expression are shown in Table 21. Treatment of cultured human hepatocytes once daily for three consecutive days with phenobarbital caused increases ranging from 4.81- to 13.7-fold change in CYP2B6 mRNA levels. Treatment with the non-inducer, flumazenil, had little or no effect on CYP2B6 activity with changes ranging from 1.11- to 1.67-fold change.

Table 21 CYP2B6 mRNA fold increase: The effect of treating cultured human hepatocytes with RX-3341 on CYP2B6 mRNA levels (Adapted from Figure 15 from study report)

mRNA Fold Increase		HC10-1	HC3-22	HC5-25
CYP2B6	0.1% DMSO	0	0	0
	1 µM RX-3341	0.122	NT	0.0620
	5 µM RX-3341	0.0990	NT	0.191
	10 µM RX-3341	0.152	0.146	0.200
	25 µM RX-3341	-0.111	0.119	0.128
	50 µM RX-3341	-0.365	0.00900	-0.0130
	100 µM RX-3341	-0.467	0.0620	-0.227
	250 µM RX-3341	-0.544	-0.572	-0.922
	500 µM RX-3341	NT	-0.359	NT
	1000 µM RX-3341	NT	-0.572	NT
	25 µM Flumazenil	0.668	0.109	0.148
	750 µM Phenobarbital	3.81	9.43	12.7

NT Not tested

mRNA fold increase = fold change − 1

The effect of RX-3341 on human CYP3A4/5 activity, CYP3A4 mRNA levels and CYP3A4 EC_{50} and E_{max}

Determination of midazolam 1′ -hydroxylation (CYP3A4/5 activity): In cultured human hepatocytes, midazolam 1′ -hydroxylation is catalyzed by CYP3A4/5. CYP3A4 is the major rifampin-inducible CYP enzyme and is also moderately inducible by phenobarbital. The effects of treating cultured human hepatocytes with RX-3341 on midazolam 1′ -hydroxylase (CYP3A4/5) activity are shown in Table 22. Treatment of cultured human hepatocytes once daily for three consecutive days with rifampin caused increases ranging from 3.03- to 7.95-fold change in midazolam 1′ -hydroxylation (CYP3A4/5 activity). Treatment with the non-inducer, flumazenil, had little or no effect on CYP3A4/5activity with changes ranging from 0.942-to 1.03-fold change.

Table 22 CYP3A4/5 activity fold increase: The effect of treating cultured human hepatocytes with RX-3341 or prototypical inducers on the rate of midazolam 1′ -hydroxylase activity (Adapted from Figure 19 in the study report)

Activity Fold Increase		HC10-1	HC3-22	HC5-25
Midazolam 1′-hydroxylation (CYP3A4/5)	0.1% DMSO	0	0	0
	1 µM RX-3341	0.00447	NT	0.0134
	5 µM RX-3341	0.0145	NT	0.203
	10 µM RX-3341	0.104	0.529	0.520
	25 µM RX-3341	0.0685	0.786	1.06
	50 µM RX-3341	0.215	1.26	1.71
	100 µM RX-3341	-0.0783	1.20	1.23
	250 µM RX-3341	-0.440	-0.0452	-0.221
	500 µM RX-3341	NT	-0.523	NT
	1000 µM RX-3341	NT	-0.883	NT
	25 µM Flumazenil	-0.0109	-0.0582	0.0331
	20 µM Rifampin	2.03	3.52	6.95

NT Not tested

Fold increase = fold change − 1

Determination of CYP2B6 mRNA levels: The effects of treating cultured human hepatocytes with RX-3341 on CYP2B6 mRNA expression are shown in Table 23. Treatment of cultured human hepatocytes once daily for three consecutive days with phenobarbital caused increases ranging from 4.81- to 13.7-fold change in CYP2B6 mRNA levels. Treatment with the non-inducer, flumazenil, had little or no effect on CYP2B6 activity with changes ranging from 1.11- to 1.67-fold change.

Table 23 CYP3A4 mRNA fold increase: The effect of treating cultured human hepatocytes with RX-3341 on CYP3A4 mRNA levels (Adapted from Figure 23 from the study report)

mRNA Fold Increase		HC10-1	HC3-22	HC5-25
CYP3A4	0.1% DMSO	0	0	0
	1 µM RX-3341	0.392	NT	0.236
	5 µM RX-3341	0.885	NT	0.550
	10 µM RX-3341	1.64	0.939	0.954
	25 µM RX-3341	2.44	1.15	1.40
	50 µM RX-3341	1.68	1.61	2.54
	100 µM RX-3341	1.48	1.57	1.27
	250 µM RX-3341	5.00	0.750	-0.241
	500 µM RX-3341	NT	0.654	NT
	1000 µM RX-3341	NT	-0.490	NT
	25 µM Flumazenil	1.17	-0.0880	0.156
	20 µM Rifampin	15.1	5.31	12.3

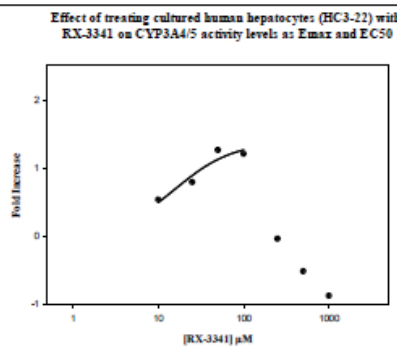
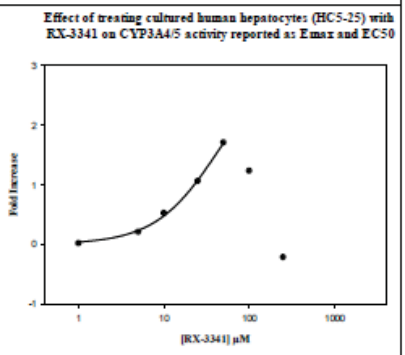
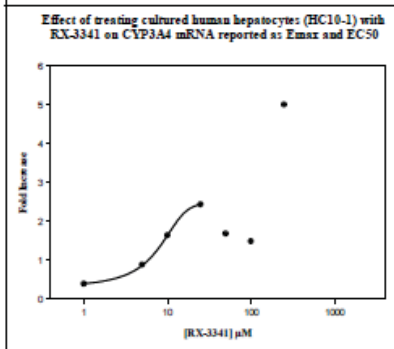
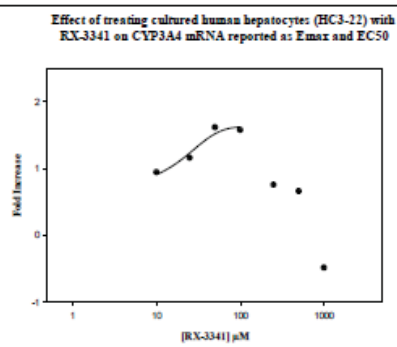
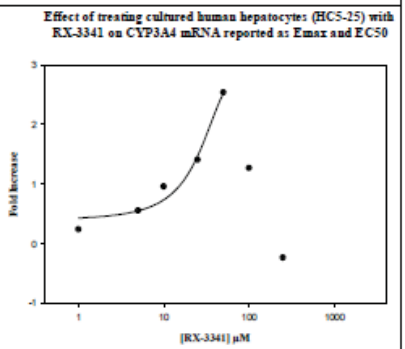
NT Not tested

mRNA fold increase = fold change − 1

The data suggests that RX-3341 did not induce CYP3A4/5 activity in hepatocyte preparation HC10-1 (⩽ 2.0-fold change, the comparison of the test article effects to the prototypical inducer was less than 20% and there were no concentration-dependent increases), therefore E_{max} and EC_{50} calculations were not performed. For hepatocyte preparations HC3-22 and HC5-25, when plotted as a response (fold increase) versus concentration profile the E_{max} values determined for HC3-22 and HC5-25 CYP3A4/5 activity were 1.38- and 3.17-fold increase (i.e., 2.38- and 4.17-fold change) and the EC_{50} values were determined to be 15.8 and 44.0 µM RX-3341, respectively (Figure 3).

For hepatocyte preparations HC10-1, HC3-22 and HC5-25, when plotted as a mRNA response (fold increase) versus concentration profile the E_{max} values were 2.46-, 1.62- and 2.91-fold increase (i.e., 3.46-, 2.62- and 3.91-fold change) and the EC_{50} values were determined to be 7.29, 6.18 and 24.6 µM RX-3341, respectively (Figure 3).

Figure 3. CYP3A4/5 and CYP3A4 EC_{50} and E_{max}: The effect of treating cultured human hepatocytes with RX-3341 on CYP3A4/5 activity and CYP3A4 mRNA levels reported as E_{max} and EC_{50}

HC10-1		HC3-22		HC5-25	
NA		 250, 500 and 1000 µM RX-3341 were not used to calculate EC_{50} and E_{max}		 100 and 250 µM RX-3341 were not used to calculate EC_{50} and E_{max}	
Parameter	**Value (SE)**	**Parameter**	**Value (SE)**	**Parameter**	**Value (SE)**
E_{max} (fold)	NA	E_{max} (fold)	1.38 (0.46)	E_{max} (fold)	3.17 (0.87)
EC_{50} (µM)	NA	EC_{50} (µM)	15.8 (10.7)	EC_{50} (µM)	44.0 (20.7)
Equation	NA	Equation	Sigmoid Hill 3-Parameter	Equation	Sigmoid Hill 3-Parameter
HC10-1		**HC3-22**		**HC5-25**	
 50, 100 and 250 µM RX-3341 were not used to calculate EC_{50} and E_{max}		 250, 500 and 1000 µM RX-3341 were not used to calculate EC_{50} and E_{max}		 100 and 250 µM RX-3341 were not used to calculate EC_{50} and E_{max}	
Parameter	**Value (SE)**	**Parameter**	**Value (SE)**	**Parameter**	**Value (SE)**
E_{max} (fold)	2.46 (0.02)	E_{max} (fold)	E_{max} (fold)	E_{max} (fold)	2.91 (0.62)
EC_{50} (µM)	7.29 (0.10)	EC_{50} (µM)	EC_{50} (µM)	EC_{50} (µM)	24.6 (8.7)
Equation	Sigmoid 3-Parameter	Equation	Sigmoid 3-Parameter	Equation	Sigmoid 3-Parameter

SE Standard error, rounded to the same degree of precision
NA Not applicable

Conclusion

In conclusion, under the conditions of this study, where the positive controls caused anticipated and appropriate increases in CYP enzyme activity and mRNA levels, treatment of hepatocytes with RX-3341 at concentrations of 250 µM or above caused visible cell toxicity resulting in decreases in CYP activity and mRNA expression. Treatment of cultured human hepatocytes with up to 1000 µM RX-3341 in hepatocyte preparation HC3-22 or up to 250 µM RX-3341 in HC5-

25 and HC10-1 did not induce CYP1A2 or CYP2B6 enzyme activity or mRNA levels. Treatment of cultured human hepatocytes with up to 50 µM RX-3341 caused dose-dependent increases in CYP3A4/5 activity and CYP3A4 mRNA levels followed by dose-dependent decreases at higher concentrations (in two of three cultures). The presence of this bell-shaped curve in activity and mRNA expression may be indicative of the morphological changes observed following increased exposure of RX-3341 to the hepatocyte test systems. EC_{50} and E_{max} values were calculated for CYP3A4/5 activity in two of three cultures and CYP3A4 mRNA expression in three cultures.

Reviewer's Comment: Based on the effect of delafloxacin on CYP450 3A4, the Sponsor conducted an in vivo study to further investigate the potential induction effect of delafloxacin on CYP3A4.

Study (b) (4) 142117: In Vitro Evaluation of Delafloxacin as an Inducer of Cytochrome P450 Expression in Cultured Human Hepatocytes

Test System

Three preparations of cryopreserved human hepatocytes (hereafter, referred to as HC10-8, HC4-18 and HC7-5, respectively) supplied internally by (b) (4) were treated in this study. Hepatocyte cultures were treated once daily for three consecutive days and cultured according to SOP L5021.02. Please refer to the table below for the positive, negative controls, and vehicle.

Chemical	Catalog number	Lot number	Storage conditions [a]	Vehicle	Purity	Supplier
DMSO	D2650	RNBC9660 RNBC9659 RNBD2515 RNBD8969	Room temperature	Not applicable	Not applicable	(b) (4)
Flumazenil	F6300	SLBG5144V	2 to 8 °C	DMSO	99%	
Rifampin	R3501	011M1159V	−20 ± 5 °C	DMSO	98%	

a Storage conditions of neat compound.

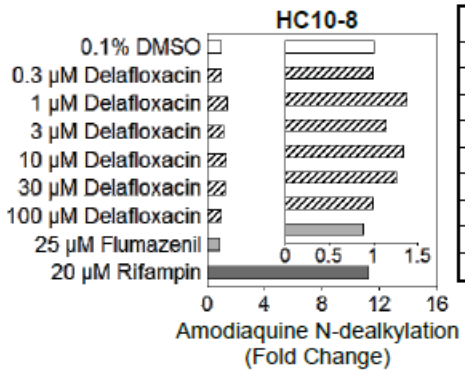
Results

Determination of amodiaquine N-dealkylation (CYP2C8 activity)

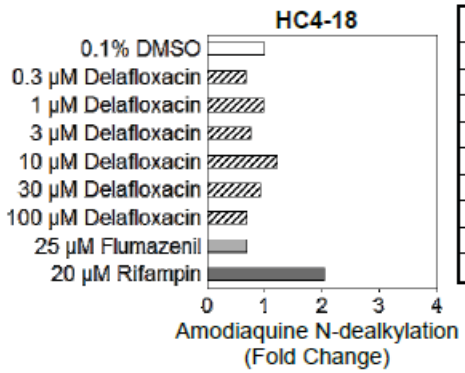
In cultured human hepatocytes, the N-dealkylation of amodiaquine is catalyzed by CYP2C8, which is inducible by rifampin. The effects of treating cultured human hepatocytes with delafloxacin on amodiaquine N-dealkylase (CYP2C8) activity are shown in Figure 4. Treatment of cultured human hepatocytes once daily for three consecutive days with rifampin caused increases ranging from 2.04-to 11.2-fold change in amodiaquine N-dealkylation (CYP2C8 activity). Treatment with the non-inducer, flumazenil, had little or no effect on CYP2C8 activity with changes ranging from 0.703- to 0.985-fold change.

Treatment of cultured human hepatocytes with up to 100 µM delafloxacin had little or no effect (< 2-fold change and < 20% as effective as the positive control, rifampin) on CYP2C8 activity, ranging from 0.991 to 1.38-fold change, 0.672- to 1.23-fold change and 0.526- to 1.23-fold change in HC10-8, HC4-18 and HC7-5, respectively.

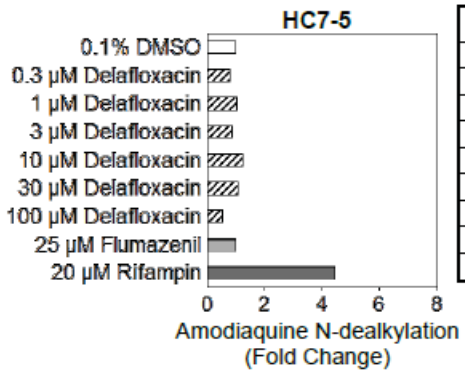
Figure 4 CYP2C8 activity: The effect of treating cultured human hepatocytes with delafloxacin on the rate of amodiaquine N-dealkylase activity



Fold Change	Rate ± Std Dev	n
1.00	0.978 ± 0.080	3
0.995	0.973 ± 0.145	3
1.38	1.35	2
1.14	1.12 ± 0.11	3
1.34	1.31 ± 0.15	3
1.26	1.23 ± 0.16	3
0.991	0.969 ± 0.109	3
0.883	0.863 ± 0.058	3
11.2	11.0 ± 0.6	3



Fold Change	Rate ± Std Dev	n
1.00	2.71 ± 0.27	3
0.672	1.82 ± 0.48	3
0.970	2.63 ± 0.33	3
0.767	2.08 ± 0.27	3
1.23	3.34	2
0.930	2.52 ± 0.49	3
0.688	1.86 ± 0.03	3
0.703	1.90 ± 0.51	3
2.04	5.53 ± 1.13	3

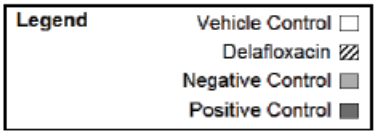


Fold Change	Rate ± Std Dev	n
1.00	0.857 ± 0.097	3
0.800	0.686 ± 0.063	3
1.03	0.886 ± 0.246	3
0.866	0.743 ± 0.069	3
1.23	1.06 ± 0.09	3
1.07	0.921 ± 0.118	3
0.526	0.451 ± 0.058	3
0.985	0.845 ± 0.074	3
4.44	3.80 ± 0.27	3

n Number of replicates

Std Dev represents standard deviation.

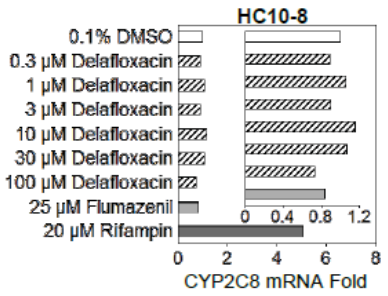
Fold change = Activity of test article treated cells / Activity of vehicle control



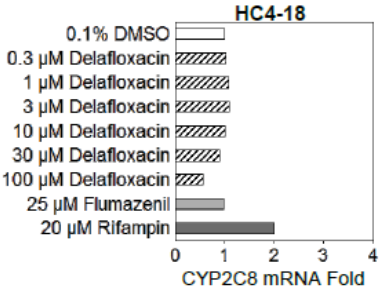
Determination of CYP2C8 mRNA levels: The effects of treating cultured human hepatocytes with delafloxacin on CYP2C8 mRNA expression are shown in Figure 5. Treatment of cultured human hepatocytes once daily for three consecutive days with rifampin caused increases ranging from 2.01- to 8.25-fold change in CYP2C8 mRNA levels. Treatment with the non-inducer, flumazenil, had little or no effect on CYP2C8 mRNA levels with changes ranging from 0.839- to 1.18-fold change.

Treatment of cultured human hepatocytes with up to 100 µM delafloxacin had little or no effect (< 2-fold change and < 20% as effective as rifampin) on CYP2C8 mRNA levels, ranging from 0.735- to 1.16-fold change, 0.563- to 1.09-fold change and 0.852- to 1.28-fold change in HC10-8, HC4-18 and HC7-5, respectively.

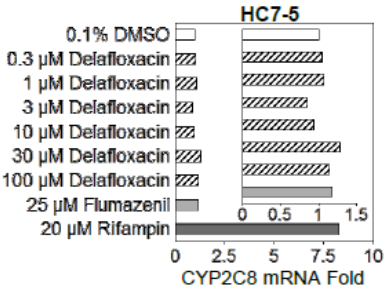
Figure 5. CYP2C8 fold change: The effect of treating cultured human hepatocytes with delafloxacin on CYP2C8 mRNA levels



mRNA Fold Change	n
1.00	3
0.885	3
1.06	3
0.898	3
1.16	3
1.07	3
0.735	3
0.839	3
5.07	3



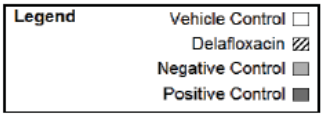
mRNA Fold Change	n
1.00	3
1.03	3
1.08	3
1.09	3
1.01	3
0.896	3
0.563	3
0.970	3
2.01	3



mRNA Fold Change	n
1.00	3
1.05	3
1.06	3
0.852	3
0.941	3
1.28	3
1.15	3
1.18	3
8.25	3

n Number of replicates

Fold change values are relative to vehicle control, normalized to GAPDH.



The effect of delafloxacin on human CYP2C9 activity and mRNA levels

Determination of diclofenac 4′ -hydroxylation (CYP2C9 activity): In cultured human hepatocytes, the 4′ -hydroxylation of diclofenac is catalyzed by CYP2C9, which is inducible by rifampin. The effects of treating cultured human hepatocytes with delafloxacin on diclofenac 4′ -hydroxylase activity are shown in Figure 6. Treatment of cultured human hepatocytes once daily for three consecutive days with rifampin caused increases ranging from 2.39- to 4.15-fold change in diclofenac 4′ -hydroxylation (CYP2C9 activity). Treatment with the non-inducer, flumazenil, had little or no effect on CYP2C9 activity with changes ranging from 0.590- to 1.16-fold change.

In hepatocyte cultures HC10-8 and HC4-18, delafloxacin had little or no effect (< 2-fold change and < 20% as effective as rifampin) on CYP2C9 activity at all concentrations tested, ranging from 0.930- to 1.34-fold change and 0.971- to 1.22-fold change, respectively. In Hepatocyte culture HC7-5, however, there were concentration-dependent increases in CYP2C9 activity, ranging from 1.04- to 1.63-fold change. Although the increases did not reach 2-fold change, they were up to 20.0% as effective as rifampin.

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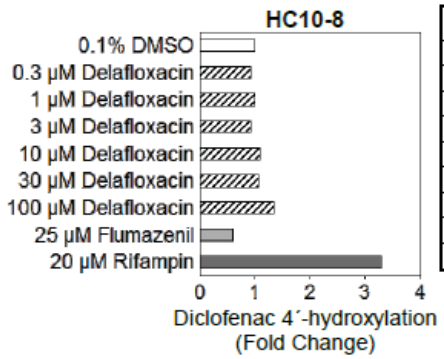
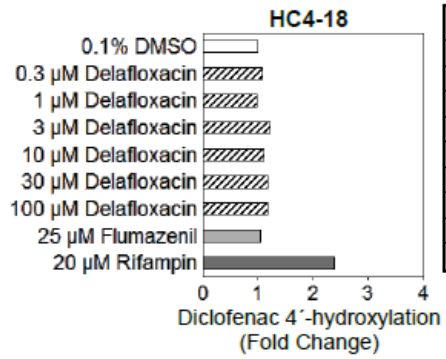
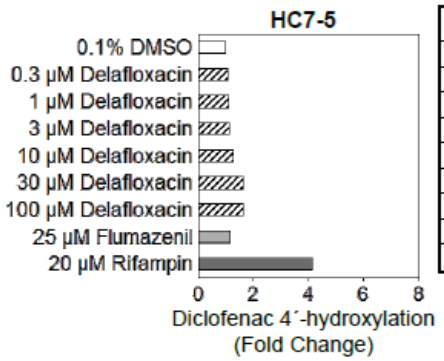
Figure 6 CYP2C9 activity: The effect of treating cultured human hepatocytes with delafloxacin on the rate of diclofenac 4′ -hydroxylase activity



Fold Change	Rate ± Std Dev	n
1.00	5.68 ± 0.24	3
0.930	5.28 ± 1.06	3
0.991	5.63 ± 0.32	3
0.934	5.31 ± 0.68	3
1.09	6.18 ± 0.43	3
1.06	6.05 ± 0.77	3
1.34	7.63 ± 0.56	3
0.590	3.35	2
3.29	18.7 ± 1.5	3



Fold Change	Rate ± Std Dev	n
1.00	12.1 ± 0.5	3
1.08	13.1 ± 1.3	3
0.971	11.7 ± 1.1	3
1.22	14.7 ± 2.6	3
1.11	13.4 ± 0.8	3
1.17	14.2 ± 1.6	3
1.17	14.1 ± 0.6	3
1.04	12.6 ± 0.5	3
2.39	28.8 ± 0.9	3

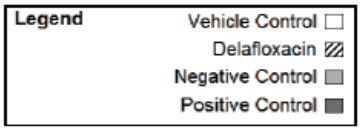


Fold Change	Rate ± Std Dev	n
1.00	6.79 ± 0.25	3
1.04	7.09	2
1.08	7.33 ± 0.44	3
1.12	7.63 ± 1.26	3
1.25	8.51 ± 0.68	3
1.62	11.0 ± 1.0	3
1.63	11.1 ± 0.2	3
1.16	7.88 ± 1.38	3
4.15	28.2 ± 1.6	3

n Number of replicates

Std Dev represents standard deviation.

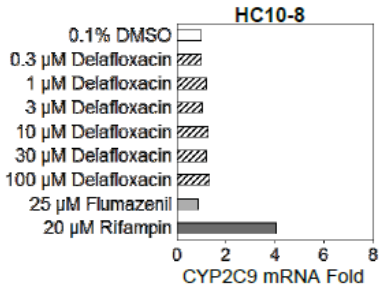
Fold change = Activity of test article treated cells / Activity of vehicle control



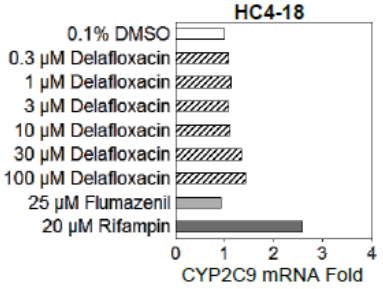
Determination of CYP2C9 mRNA levels: The effects of treating cultured human hepatocytes with delafloxacin on CYP2C9 mRNA expression are shown in Figure 7. Treatment of cultured human hepatocytes once daily for three consecutive days with rifampin caused increases ranging from 2.59- to 6.20-fold change in CYP2C9 mRNA levels. Treatment with the non-inducer, flumazenil, had little or no effect on CYP2C9 mRNA levels with changes ranging from 0.858- to 1.08-fold change.

Delafloxacin had little or no effect on CYP2C9 mRNA levels in hepatocyte cultures HC10-8 and HC7-5 at all concentrations tested. Changes ranged from 0.991- to 1.32-fold change (HC10-8) and 0.901- to 1.93-fold change (HC7-5). In hepatocyte culture HC4-18, treatment with up to 100 µM delafloxacin caused concentration-dependent increases in CYP2C9 mRNA levels that ranged from 1.08- to 1.42-fold change. These were < 2-fold change but were up to 26.3% as effective as rifampin. Of note, compared to hepatocyte cultures HC10-8 and HC7-5 where rifampin caused 4.03-and 6.20-fold change increases, respectively, rifampin caused an increase of 2.59-fold change in CYP2C9 mRNA levels. Therefore, the reported effectiveness of delafloxacin, > 20%, relative to rifampin may likely be as a result of the relatively low response of rifampin to CYP2C9 mRNA levels.

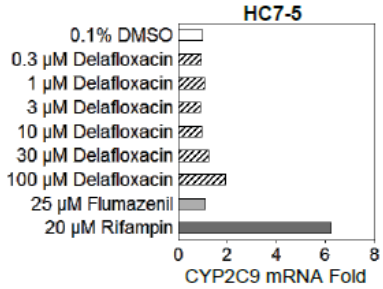
Figure 7. CYP2C9 fold change: The effect of treating cultured human hepatocytes with delafloxacin on CYP2C9 mRNA levels



mRNA Fold Change	n
1.00	3
0.991	3
1.18	3
1.04	3
1.26	3
1.18	3
1.32	3
0.858	3
4.03	3



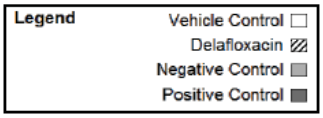
mRNA Fold Change	n
1.00	3
1.08	3
1.13	3
1.08	3
1.11	3
1.34	3
1.42	3
0.929	3
2.59	3



mRNA Fold Change	n
1.00	3
0.916	3
1.07	3
0.901	3
0.961	3
1.24	3
1.93	3
1.08	3
6.20	3

n Number of replicates

Fold change values are relative to vehicle control, normalized to GAPDH.



The effect of delafloxacin on human CYP2C19 activity and mRNA levels

Determination of S-mephenytoin 4′ -hydroxylation (CYP2C19 activity): In cultured human hepatocytes, S-mephenytoin 4′ -hydroxylation is catalyzed by CYP2C19, which is inducible by rifampin. The effects of treating cultured human hepatocytes with delafloxacin on S-mephenytoin 4′ -hydroxylase (CYP2C19) activity are shown in Figure 8. Treatment of cultured human hepatocytes once daily for three consecutive days with rifampin caused increases ranging from 3.12-to 19.3-fold change in S-mephenytoin 4′ -hydroxylation (CYP2C19 activity). Treatment with the non-inducer, flumazenil, had little or no effect on CYP2C19 activity with changes ranging from 0.843- to 1.19-fold change.

Treatment with up to 100 μM delafloxacin caused concentration-dependent increases in CYP2C19 activity in hepatocyte culture HC10-8. These increases, however, were < 2-fold change (a range of 0.833- to 1.76-fold change). Delafloxacin was > 20% as effective as rifampin (up to 27.2%) at 100 μM. In hepatocyte cultures HC4-18 and
HC7-5, treatment with delafloxacin had little or no effect on CYP2C19 activity, except for an elevation of 2.09-fold change in HC7-5 at 30 μM. This increase, however, was neither concentration-dependent nor > 20% of rifampin. Therefore, this was not likely an artifact of delafloxacin.

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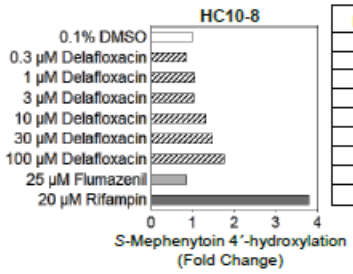
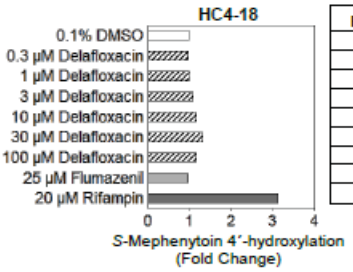
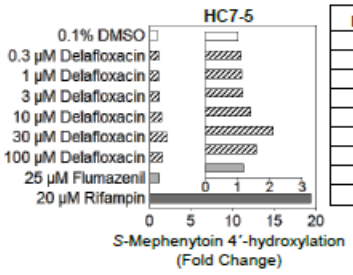
Figure 8. CYP2C19 activity: The effect of treating cultured human hepatocytes with delafloxacin on the rate of S-mephenytoin 4′ - hydroxylase activity (Adapted from Figure 10 in the study report)



Fold Change	Rate ± Std Dev	n
1.00	0.0484 ± 0.0056	3
0.833	0.0403 ± 0.0126	3
1.04	0.0503 ± 0.0040	3
1.03	0.0497 ± 0.0057	3
1.30	0.0630 ± 0.0041	3
1.47	0.0714 ± 0.0013	3
1.76	0.0852 ± 0.0056	3
0.843	0.0408 ± 0.0093	3
3.79	0.184 ± 0.031	3

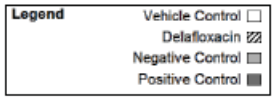


Fold Change	Rate ± Std Dev	n
1.00	0.0665 ± 0.0102	3
0.953	0.0633 ± 0.0073	3
0.990	0.0658 ± 0.0027	3
1.08	0.0716 ± 0.0054	3
1.16	0.0769 ± 0.0065	3
1.30	0.0865 ± 0.0099	3
1.16	0.0770 ± 0.0031	3
0.961	0.0639 ± 0.0059	3
3.12	0.207 ± 0.024	3



Fold Change	Rate ± Std Dev	n
1.00	0.0376 ± 0.0014	3
1.09	0.0412 ± 0.0016	3
1.11	0.0419 ± 0.0032	3
1.15	0.0433 ± 0.0043	3
1.40	0.0528 ± 0.0026	3
2.09	0.0787 ± 0.0101	3
1.57	0.0591 ± 0.0004	3
1.19	0.0447 ± 0.0060	3
19.3	0.727 ± 0.149	3

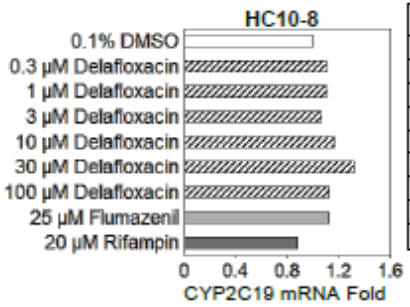
n Number of replicates
Std Dev represents standard deviation.
Fold change = Activity of test article treated cells / Activity of vehicle control



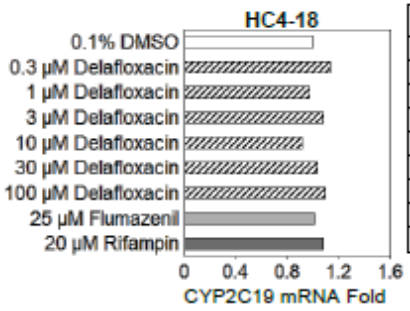
Determination of CYP2C19 mRNA levels: The effects of treating cultured human hepatocytes with delafloxacin on CYP2C19 mRNA expression are shown in Figure 9. Treatment of cultured human hepatocytes once daily for three consecutive days with rifampin caused increases ranging from 0.879- to 1.61-fold change in CYP2C19 mRNA levels. Treatment with the non-inducer, flumazenil, had little or no effect on CYP2C19 mRNA levels with changes ranging from 1.01- to 1.13-fold change.

Delafloxacin had little or no effect on CYP2C19 mRNA levels at all concentrations tested. Changes ranged from 1.06- to 1.32-fold change, 0.912- to 1.13-fold change and 0.918- to 1.19-fold change in HC10-8, HC4-18 and HC7-5, respectively. The effect of delafloxacin on CYP2C19 mRNA levels relative to rifampin was not determined because rifampin caused < 2-fold change in CYP2C19 mRNA levels. The low response of rifampin to CYP2C19 mRNA levels is expected in secondary enzymes, such as CYP2C19.

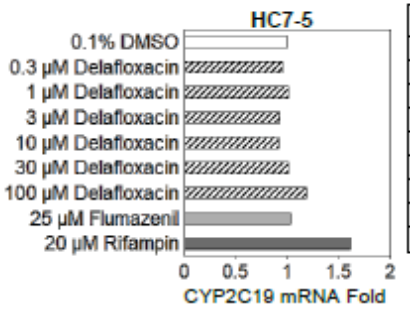
Figure 9. CYP2C19 fold change: The effect of treating cultured human hepatocytes with delafloxacin on CYP2C19 mRNA levels (Adapted from Figure 12 in the study report)



mRNA Fold Change	n
1.00	3
1.11	3
1.10	3
1.06	3
1.16	3
1.32	3
1.12	3
1.13	3
0.879	3



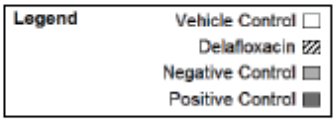
mRNA Fold Change	n
1.00	3
1.13	3
0.970	3
1.08	3
0.912	3
1.03	3
1.09	3
1.01	3
1.08	3



mRNA Fold Change	n
1.00	3
0.952	3
1.01	3
0.925	3
0.918	3
1.01	3
1.19	3
1.04	3
1.61	3

n Number of replicates

Fold change values are relative to vehicle control, normalized to GAPDH.



Reviewer's Comment: Take the results from Study (b) (4) 133033 and Study (b) (4) 142117 together, it seems that delafloxacin was not likely to induce CYP1A2, CYP2B6, CYP2C19, or CYP2C8. However, delafloxacin was a mild inducer for CYP2C9 at the concentration of 100 µM (less than 2 fold) and CYP3A4 at a clinical relevant concentration.

UGT Substrate and Inducer

Study (b) (4) 134028: In Vitro UDP-Glucuronosyltransferase (UGT) Reaction Phenotyping of Delafloxacin (RX-3341)

The aim of this study was to determine the role of human UDP-glucuronosyltransferase enzymes in the metabolism of delafloxacin (RX-3341) by UDPGA-fortified human liver microsomes.

Test System:

Human liver microsomes from a pool of 200 individuals (catalog number: H2620, lot number: 1210057) were used for this study. These microsomes were prepared at the Testing Facility according to the applicable SOP and characterized with respect to the activities of various CYP enzymes.

Recombinant human UGT enzymes (Supersomes) and the corresponding control microsomes (from insect cells transfected with wild type baculovirus but no human UGT enzyme) were purchased from (b) (4) and characterized by the manufacturer with respect to UGT enzyme activity.

Recombinant human UGT enzymes: Delafloxacin (RX-3341) (10 µM) was incubated with recombinant human UGT enzymes (UGT1A1, UGT1A3, UGT1A4, UGT1A6, UGT1A9, UGT2B7 and UGT2B15, 1 mg protein/mL) according to SOP L7060.09.

Chemical inhibition: Incubation of delafloxacin (RX-3341) in the presence and absence of direct-acting chemical inhibitors known to inhibit UGT enzymes was conducted to evaluate the role of these enzymes in the metabolism of delafloxacin (RX-3341) according to SOP L7040.09.

An LC/MS/MS method for the analysis of delafloxacin (RX-3341) was developed for concentrations of delafloxacin (RX-3341) ranging from 0.1 to 12.5 µM.

Results

Recombinant human UGT enzymes: Formation of the glucuronide metabolite was observed after 120 min incubation of delafloxacin (RX-3341) (10 µM) with rUGT1A1, rUGT1A3 and rUGT1A9. The metabolism of delafloxacin (RX-3341) based on substrate loss was also evaluated. After 120 minute incubation with delafloxacin (RX-3341) the percent substrate loss ranged from no loss to approximately 9%. Control incubations of 4-methylumbelliferone and imipramine (1 mM and 100 µM, respectively) with recombinant human UGT enzymes were performed concurrent with the test article incubations.

Chemical inhibition: After 120 minutes of incubation the formation of the glucuronide metabolite was completely inhibited by the UGT1A1 and UGT1A3 inhibitors, erlotinib and ritonavir, respectively. This observation is consistent with the result of the recombinant human

UGT enzyme experiment. After 120 minutes of incubation, no inhibition of glucuronide formation was observed in incubations of the UGT1A4 inhibitor hecogenin. Niflumic acid did not inhibit formation of the glucuronide which suggests that UGT1A9 is not involved in the metabolism of delafloxacin.

Conclusions

In conclusion, incubations of delafloxacin with recombinant human UGT enzymes and chemical inhibitors indicate that UGT1A1 and UGT1A3 are involved in the metabolism of delafloxacin (RX-3341).

Study (b) (4)135024: In Vitro Evaluation of Delafloxacin (RX-3341) as an Inhibitor of UDP-Glucuronosyltransferase Enzymes in Human Liver Microsomes

This study was designed to evaluate the ability of delafloxacin to inhibit, in vitro, select UGT enzymes in human liver microsomes (namely UGT1A1 and UGT2B7) with the aim of ascertaining the potential of delafloxacin to inhibit the metabolism of concomitantly administered drugs.

Test System

A mixed-gender pool of sixteen individual human liver microsomal samples was used for this study (b) (4) sample code numbers 286, 290, 312, 313, 315, 333, 334, 335, 336, 339, 348, 359, 364, 383, 389 and 390). The kinetic constants (Km or S50) used to select marker substrate concentrations and incubation conditions were determined previously.

Result

Under the experimental conditions examined, there was some evidence of direct inhibition of UGT1A1 and UGT2B7 as approximately 26% and 20% inhibition, respectively, was observed at the highest concentration of delafloxacin evaluated (100 μM) and the IC_{50} values were reported as greater than 100 μM.

Reviewer's Comment: Due to the high IC_{50}, it is not likely that delafloxacin will inhibit UGT1A1 or UGT2B7 at clinically relevant concentrations.

Protein Binding

Study R&D/00/427: In Vitro Protein Binding of [14C]Abbott-319492 in Mouse, Rat, Dog, Monkey and Human Plasma

Test System

Blood was obtained from male and female CD-1 mice, Sprague-Dawley rats, beagle dogs and cynomolgus monkeys (at least two ·of each sex), which were fasted overnight. Blood was also obtained from adult human volunteers (at least two of each sex), who had fasted at least 8 hours,

had taken no medication except aspirin during the preceding seven days and had taken no salicylates during the preceding 48 hours. The heparinized blood samples were centrifuged to separate the plasma which was then separated into aliquots of 3-5 mL.

Appropriate aliquots of the stock solutions were added to the plasma aliquots to give initial [14C]Abbott-3 l 9492 concentrations of approximately 0.1, 1, 5, 10, 25 and 50 µg/mL. The spiked plasma aliquots were equilibrated for 15 minutes at 37°C before being loaded into the dialysis cells. At least 2 mL of spiked plasma from each species was incubated at 37°C for 2 hours during the course of the equilibrium dialysis.

Preliminary experiments, using male dog plasma at two concentrations (10 and 50 µg /mL) determined that 2 hours was sufficient to attain equilibrium. Plasma samples (not extracted) characterized by high-performance liquid chromatography showed that [14C]Abbott-319492 was stable (>99% purity) in plasma under the conditions of this study.

Results

The percent of [14C]Abbott-319492 bound ranged from 95.52-97.74% in mouse, 90.73-92.27% in rat, 74.18-75.90% in dog, 77.66-79.52% in monkey and 82.98-84.54% in human plasma. The in vitro plasma protein binding results obtained for [14C]Abbott-319492 in human plasma are listed in Tables 24, while the mean protein binding values for these species are compared in Table 25.

The extent of protein binding of [14C]Abbott-319492 varied considerably over the five species studied (75 to 97%). However for each species studied, the extent of binding was consistent over the entire drug concentration range of 0.1-50 µg acid/mL.

Reviewer's Comment: It seems that there were no sex related differences in plasma protein binding among the species examined. Delafloxacin was shown to be 84% bound to proteins in human plasma at clinically relevant concentrations.

Table 24 In Vitro Protein Binding of [14C]Abbott-319492 in Human Plasma (Adapted from Table 5 in the study report)

Drug Concentration (µg/mL)		Sample Number				
		1 Male	2 Male	3 Female	4 Female	Mean ± SD
0.1	% Bound	82.95	87.89	81.45	83.63	83.98 ± 2.76
	% Free	17.05	12.11	18.55	16.37	16.02 ± 2.76
1.0	% Bound	84.41	88.76	81.05	83.94	84.54 ± 3.18
	% Free	15.59	11.24	18.95	16.06	15.46 ± 3.18
5.0	% Bound	82.52	86.81	81.19	83.24	83.44 ± 2.40
	% Free	17.48	13.19	18.81	16.76	16.56 ± 2.40
10.0	% Bound	83.50	84.81	80.92	82.69	82.98 ± 1.63
	% Free	16.50	15.19	19.08	17.31	17.02 ± 1.63
25.0	% Bound	84.60	85.86	81.44	83.38	83.82 ± 1.88
	% Free	15.40	14.14	18.56	16.62	16.18 ± 1.88
50.0	% Bound	84.07	86.36	81.05	82.74	83.56 ± 2.24
	% Free	15.98	13.64	18.95	17.26	16.44 ± 2.24

Table 25 Mean In Vitro Protein Binding of [14C]Abbott-319492 in Mouse, Rat, Dog, Monkey and Human Plasma (Adapted from Table 6 in the study report)

Drug Concentration (µg/mL)	% Bound					
	Sex	Mouse	Rat	Dog	Monkey	Human
0.1	Male	97.28	90.97	77.84	75.85	85.42
	Female	96.57	91.83	73.95	79.46	82.54
	Mean	96.92	91.40	75.90	77.66	83.98
1.0	Male	97.85	91.48	76.78	77.81	86.59
	Female	97.64	92.81	73.40	82.15	82.50
	Mean	97.74	92.15	75.09	79.98	84.54
5.0	Male	97.78	91.43	77.20	76.65	84.67
	Female	97.71	93.10	73.37	80.86	82.21
	Mean	97.74	92.27	75.28	78.75	83.44
10.0	Male	97.11	90.96	77.30	78.08	84.16
	Female	97.39	91.56	73.01	80.95	81.81
	Mean	97.25	91.26	75.15	79.52	82.98
25.0	Male	97.16	90.98	76.91	77.90	85.23
	Female	97.02	91.43	72.83	79.67	82.41
	Mean	97.09	91.21	74.87	78.79	83.82
50.0	Male	96.48	90.37	76.23	79.65	85.22
	Female	94.56	91.09	72.12	77.86	81.90
	Mean	95.52	90.73	74.18	78.76	83.56
Mean (0.1-50.0 µg /mL)	M + F	97.04	91.50	75.08	78.91	83.72

Study R&D/00/428: In vitro Binding of [14c]Abbott-319492 to Human a1-Acid Glycoprotein and Albumin

The purpose of this study was to determine the binding of ['4C]Abbott-319492 to human a.1-acid glycoprotein (AAG) and human serum albumin (HSA), the principal drug binding proteins in human plasma.

Test System

A 0.067 M phosphate buffer was prepared and adjusted to pH 7.4. HSA and AAG, obtained from (b) (4) were dissolved in the phosphate buffer at a concentration of 40 and 0.8 mg/mL, respectively.

Six radiolabeled stock solutions having final concentrations of 0.01, 0.1, 0.5, 1.0, 2.5 and 5 mg/mL were prepared in a solution of sodium hydroxide (1: 1.1 molar equivalent). Appropriate amounts of non-radiolabeled Abbott-319492 (Lot No. 981112) and radiolabeled Abbott-319492 were combined to prepare these stock solutions.

The binding affinity of [14C]Abbott-319492 was determined in a Spectrum Equilibrium Dialysis System (b) (4) using 1 mL cells and a Spectra/Por 2 membrane with a molecular weight cut-off of 12000-14000 daltons.

Results

The in vitro binding of [14C]Abbott-319492 in human serum albumin (HSA) and a 1-acid glycoprotein (AAG) was determined. The percentages of [I4C]Abbott-319492 present as bound and free fractions in HSA and AAG are presented in Tables 26 and 27, respectively.

Table 26. Binding of [14C]Abbott-319492 in Human Serum Albumin

Drug Concentrations (µg/mL)		Abbott-319492
0.1	% Bound	84.89
	% Free	15.11
1.0	% Bound	84.48
	% Free	15.52
5.0	% Bound	84.09
	% Free	15.91
10.0	% Bound	83.93
	% Free	16.07
25.0	% Bound	84.21
	% Free	15.79
50.0	% Bound	84.98
	% Free	15.02

Table 27. Binding of [14C]Abbott-319492 in Human α1-Acid Glycoprotein

Drug Concentrations (µg/mL)		Abbott-319492
0.1	% Bound	3.68
	% Free	96.32
1.0	% Bound	2.40
	% Free	97.60
5.0	% Bound	4.49
	% Free	95.51
10.0	% Bound	2.57
	% Free	97.43
25.0	% Bound	1.12
	% Free	98.88
50.0	% Bound	3.24
	% Free	96.76

Reviewer's Comment: The results of this study indicate that [14C]delafloxacin has higher affinity for human serum albumin which probably contributes to the majority of protein binding of delafloxacin in human plasma.

APPEARS THIS WAY ON ORIGINAL

4.5.17 Literature Review for Sulfobutyl-Ether-β-Cyclodextrin (SBECD, Captisol) by the Reviewer

BACKGROUND

Sulfobutylether cyclodextrin (SBECD, Captisol®) is a cyclodextrin excipient used to improve drug solubility, stability, and bioavailability. Delafloxacin for injection is formulated with SBECD (b) (4). In humans, SBECD is eliminated unchanged almost entirely by renal filtration . Therefore, SBECD pharmacokinetics can be markedly altered in subjects with impaired renal function, and SBECD exposures have been reported to increase with decreased renal filtration[3]. The same phenomenon was observed in a PK study in subjects with renal impairment (Study RX-3341-110) in this submission. Since the systemic exposure of delafloxacin was increased with decrease of renal function, the Sponsor proposed to reduce the delafloxacin IV dose to 200 mg BID for patients with severe renal impairment (b) (4). However, the Sponsor only measured the systemic exposure of SBECD after one single dose (300 mg delafloxacin with 2400 mg SBECD) in patients with different levels of renal function in this submission.

The reviewer conducted an independent literature review for SBECD. The objectives for this review are to 1) find more pharmacokinetics information about SBECD, especially the linear PK range of SBECD if identified in any publications; 2) compare the SBECD systemic exposure in subjects receiving proposed delafloxacin dose to subjects receiving other IV drug products containing SBECD; 3) find more clinical safety data for SBECD in IV drug products.

According to David Luke et al[6]., at the time of the development of SBECD, its clinical pharmacokinetics were characterized in healthy male volunteers with [^{14}C]-SBECD. Subjects received 100 mg/kg SBECD twice daily (BID) on Day 1, then 50 mg/kg SBECD BID on Days 2–9, and then a single 50 mg/kg SBECD dose on Day 10. The principal pharmacokinetics of this study are summarized in Table 1 below.

Table 1. Pharmacokinetics of [C^{14}]-SBECD in Healthy Volunteers (Mean±SD)

Parameter Dose	Day 1, 100 mg/kg BID[a]	Day 10, 50 mg/kg BID
C_{max} (μg equiv/mL)	458 ± 44	223 ± 28
AUC (μg equiv h/mL)	919 ± 82	462 ± 64
$t_{1/2}$ (h)	1.4	1.6
CL_T (mL/min/kg)	1.9 ± 0.2	1.8 ± 0.2
CL_r (mL/min)	113 ± 21	118 ± 19
V_{ss} (mL/kg)	185 ± 19	208 ± 25

[a]BID, twice daily dosing.

[5] David R. Luke et al. Review of the Basic and Clinical Pharmacology of Sulfobutylether-beta-Cyclodextrin (SBECD) Journal of Pharmaceutical Sciences, Vol.99 No.8: 3291-3301 2010

Based on data in Table 1, it seems that both clearance (CL) and volume of distribution (Vss) of SBECD remained same after Day 1 and at steady state in dose range 50 mg/kg (3500 mg per dose for a subject weighing 70 kg) to 100 mg/kg (7000 mg per dose for a subject weighing 70 kg).

There are six FDA-approved NDAs whose IV formulations containing SBECD identified in FDA database. Please refer to Table 2 below for details. Only two NDAs collected human data for SBECD in subjects with renal impairment (IV VFEND [Voriconazole] and CARNEXIV [Carbamazepine]).

Table 2. IV Formulations with Sulfobutyl Ether β-Cyclodextrin (SBECD)

Brand Name	Generic Name	Human data for SBECD in Subjects with Renal Impairment	NDA
Nexterone	Amiodarone	No	22325 (2008)
Carnexiv	Carbamazepine	Yes	206030 (2016)
Kyprolis	Carfilzomib	No	202714 (2012)
Evomela	Melphalan	No	207155 (2016)
Noxafil	posaconazole	No	205596(2014)
Vfend	voriconazole	Yes	21267 (2002)

The IV VFEND labeling state (b) (4) The mean AUC and peak plasma concentrations (Cmax) of SBECD were increased 4 -fold and almost 50%, respectively, in the moderately impaired group compared to the normal control group. Samantha Abel et al.[6] treated male subjects with no (n = 6) or moderate (n = 7) renal impairment with multiple doses of IV voriconazole containing SBECD (6 mg/kg voriconazole twice daily [day 1] then 3 mg/kg voriconazole twice daily [days 2–6] followed by a final dose of 3 mg/kg on the morning of day 7) at an infusion rate of 3 mg/kg/h. The Steady state PK parameters were summarized in Table 3 below. In this study, three out of seven subjects with moderate renal impairment had increases in serum creatinine (>25%) compared to baseline after 7 days of IV voriconazole. Therefore, the VFEND labeling recommends that IV voriconazole should be avoided in patients with moderate or severe renal impairment (creatinine clearance < 50 mL/min), unless an assessment of the benefit/risk to the patient justified the use of IV voriconazole.

[6] Samantha Abel et al. Pharmacokinetics, Safety and Tolerance of Voriconazole in Renally Impaired Subjects, Clin Drug Invest 2008; 28 (7):409-420

Table 3. Steady-state pharmacokinetic parameters following multiple intravenous doses of voriconazole

Parameter	Degree of renal impairment		Ratio or difference between groups
	none	moderate	(95% CI)[a]
Voriconazole			
n	5	6	
C_{max} (ng/mL)[b]	5416	3096	57.2% (27.2, 120.3)
AUC_{τ} (ng • h/mL)[b]	33 284	21 182	63.6% (21.0, 192.6)
t_{max} (h)[c]	1.0	1.0	–
CL (L/h)[b]	7.4	11.3	151.6% (51.4, 447.5)
SBECD			
n	6	6	
C_{max} (ng/mL)[b]	251	371	148.1% (114.7, 191.2)
AUC_{τ} (ng • h/mL)[b]	441	2128	483.1% (328.8, 709.6)
t_{max} (h)[c]	1.0	1.1	0.074 (–0.030, 0.179)
CL (L/h)[b]	8.5	1.8	21.1% (14.8, 30.2)
$t_{½\beta}$ (h)[c]	1.8	8.9	1.59 (1.24, 1.95)
Vd (L)[c]	23.0	24.8	–

a Ratio for C_{max}, AUC, CL; difference for t_{max}, $t_{½}$.

b Geometric mean.

c Arithmetic mean.

AUC = area under the concentration-time curve; **AUC_{τ}** = AUC from zero to time τ; **CI** = confidence interval; **CL** = total clearance; **C_{max}** = maximum plasma concentration; **SBECD** = sulphobutylether-β-cyclodextrin; **t_{max}** = time to first occurrence of C_{max}; **$t_{½\beta}$** = terminal elimination half-life; **Vd** = apparent volume of distribution; – indicates not calculated.

According to the VFEND labeling[7], for IV injection, lyophilized powder contains 200 mg voriconazole and 3200 mg of SBECD; after reconstitution, it yields 10 mg/mL of voriconazole and 160 mg/mL of SBECD. According to the label recommended dosing regimen, adult patients will take loading doses of 6 mg/kg q12h for the first 24 hours and maintenance dose of 4 mg/kg q12 hours for at least 14 days.

A vial of delafloxacin for injection contains 300 mg of delafloxacin and 2400 mg of SBECD. For the treatment of ABSSSI, the proposed dosing regimen for IV delafloxacin is 300 mg, BID (q12hr), for up to 14 days. Therefore, patients receiving IV delafloxacin therapy would receive 2400 mg SBECD per dose, or 4800 mg per day. The proposed dosing regimen for IV delafloxacin in patients with severe renal impairment (b) (4) is 200 mg BID. Therefore, these patients would receive 1600 mg SBECD per dose, or 3200 mg per day. Please refer to Table 4 for the comparison of daily SBECD doses in patients with severe renal impairment (b) (4) between delafloxacin and IV VFEND.

[7] http://www.accessdata.fda.gov/drugsatfda_docs/label/2015/021266s038,021267s047,021630s028lbl.pdf

Table 4. Daily SBECD Doses for Delafloxacin and Voriconazole IV Dosage Form

SBECD dose/per day	
Delafloxacin IV (200 mg BID)	3200 mg
VFEND IV (Maintenance dose: 4 mg/kg BID)	7680 mg (based on body weight of 60 kg)
VFEND IV (Loading dose: 6 mg/kg BID in first 24 hours)	11520 mg (based on body weight of 60 kg)

It was observed that the geometric mean of AUCτ for SBECD in subjects with normal renal function in the study above (Table 3) was slightly higher than the one observed in Study RX-3341-110 (355.7 µg*h/mL). The AUCτ for patients with moderate renal impairment was also slightly higher than the one observed in subjects with severe renal impairment in Study RX-3341-110 after 300 mg IV single dose (1975.35 µg*h/mL).

The potential risk of nephrotoxicity caused by SBECD has been identified in animal studies, specifically cytoplasmic vacuolation in the epithelium of the renal tubules, renal pelvis and urinary bladder[6]. However, the data of adverse events associated with cyclodextrins in humans, especially the nephrotoxicity, are limited and controversial. According to Turner's review[8] of seven original articles, receipt of IVV (intravenous voriconazole) in those with baseline renal impairment was not a risk factor for worsening renal function. The incidence of worsening renal function was similar in those receiving IVV compared with those receiving POV (oral voriconazole) or other antifungals.

Hafner et al.[9] characterized the disposition of SBECD in patients with ESRD in different hemodialysis systems after a single IV dose of voriconazole. SBECD can be removed by hemodialysis. SBECD concentrations declined with a half-life ranging from 2.6 ±0.6 h (Genius dialysis) to 2.4 ± 0.9 h (hemodialysis) and 2.0 ± 0.6 h (hemodiafiltration) ($P < 0.01$ for Genius dialysis versus hemodiafiltration).

Luke et al.[10] assessed the disposition and safety of SBECD in seven end-stage renal disease patients on hemodialysis vs. six subjects with normal renal function up to five days. All subjects received twice-daily IV voriconazole at the standard voriconazole dose [6 mg/kg (96 mg/kg SBECD) every 12 h (Q12h) on Day 1 followed by 3 mg/kg (48 mg/kg SBECD) Q12h on Days 2–4, with a single IV dose on the morning of Day 5]. Hemodialysis in renal dysfunction subjects

[8] R. Brigg Turner, Jay L. Martello, Ashim Malhotra Worsening renal function in patients with baseline renal impairment treated with intravenous voriconazole: A systematic review International Journal of Antimicrobial Agents 46(2015) 362-66

[9] Verena Hafner et al. Pharmacokinetics of Sulfobutylether-Beta-Cyclodextrin and Voriconazole in Patients with End-Stage Renal Failure during Treatment with Two Hemodialysis Systems and Hemodiafiltration. Antimicrobial Agent and Chemotherapy, June 2010 : 2596-2602

[10] David R. Luke et al. Pharmacokinetics of sulfobutylether-β-cyclodextrin (SBECD) in subjects on hemodialysis. Nephrol Dial Transplant (2012) 27:1207-1212

was performed after the morning IV infusions of voriconazole formulation containing SBECD on Days 2 and 4. Subjects were sampled at selected pre-dose trough times, at selected times after infusions and intensively on Day 3 (non-dialysis) and Day 4 (dialysis with high-flux membranes). In those patients, the mean half-life of SBECD for these patients off-dialysis was 79 hours ($t_{1/2}$ of SBECD in subjects with normal renal function = 2.1 hours). Two subjects withdrew due to adverse events. These were both dialysis subjects and the events were assessed by the investigator as being related to the active compound, voriconazole. Treatment-emergent all-causality adverse events were reported for five normal subjects (13 events) and six dialysis subjects (31 events) and were mild or moderate in severity. Treatment-emergent and treatment-related adverse events were reported for five normal subjects (8 events) and four dialysis subjects (11 events). One dialysis subject experienced five serious adverse events (SAE) after three doses of voriconazole/SBECD, recorded as mental confusion, visual hallucination, chest pain, slurred speech and altered gait; all of these SAE were considered related to the active drug, voriconazole, by the investigator and were moderate in severity. This subject withdrew from the study on Day 2 due to mental confusion and hallucinations, which had resolved by Day 3.

Based on the limited literature information collected, 1) it seems that the systemic exposure of SBECD will decrease with dose reduction; 2) the systemic exposure of SBECD in patients with severe renal impairment after receiving a single dose of IV 300 mg delafloxacin was similar to the one in patients with moderate renal impairment after receiving 7 days of therapeutic dose of IV voriconazole; 3) the data of adverse events associated with SBECD in humans, especially the nephrotoxicity, are limited and controversial.

4.5.18 Summary of Pharmacogenomics for Delafloxacin (Prepared by Dr. Anuradha Ramamoorthy)

The primary metabolic pathway of delafloxacin is glucuronidation mediated mainly by UDP-glucuronosyltransferase (UGT)1A1, UGT1A3, and UGT2B15 (see metabolism in Section 3.2 for details on metabolism and metabolites). The UGT genes are highly polymorphic and these polymorphisms can affect enzyme activity [Reference: UGT Alleles Nomenclature Home Page http://www.ugtalleles.ulaval.ca]. Additionally, racial/ethnic differences in the frequencies of some UGT gene polymorphisms are common.

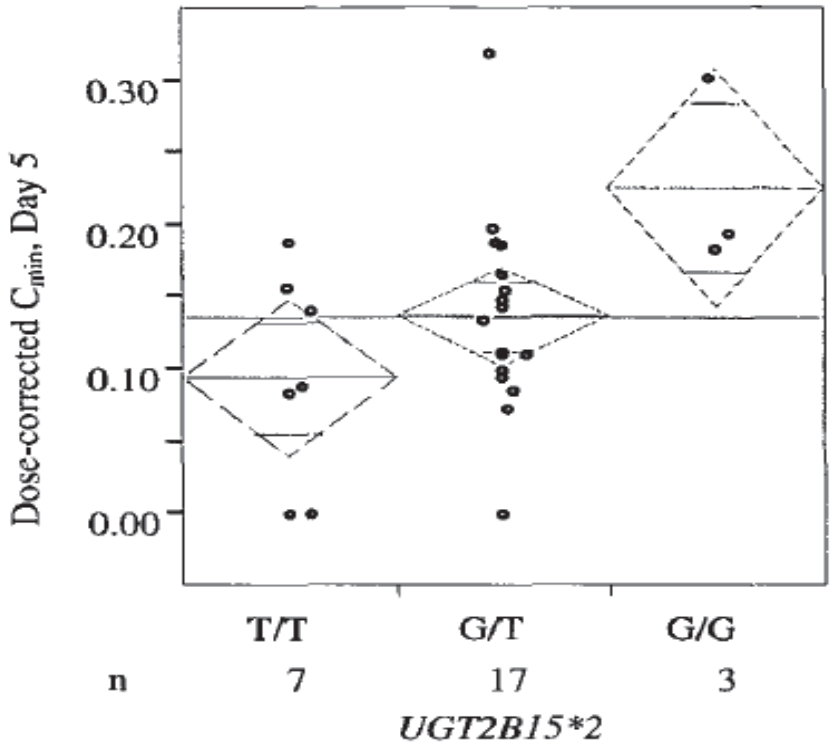
The Applicant performed an optional pharmacogenetic sub-study in two Phase 1 trials, M00-224 (pharmacokinetics of rising single and multiple oral doses of delafloxacin and the effects of food, sex and age; N=168; see 4.5.2 and 4.5.16 for details) and M01-301 (comparison of bioavailability of two capsule formulations of delafloxacin in healthy subjects; N=18;) to evaluate the effect of genetic polymorphisms in UGT1A1, UGT2B4, UGT2B15, and ABCB1 on the PK (e.g., C_{max}, C_{min}, AUC, $t_{1/2}$, CL/F, Vβ/F) of delafloxacin. DNA was collected from subjects who provided additional consent for a blood sample, and genotypes were determined using pyrosequencing at 7 polymorphic sites in ABCB1 and 1 polymorphic site each in UGT1A1, UGT2B4, and UGT2B15. The Applicant did not provide information regarding the ABCB1 polymorphisms that were assessed. In subjects treated with delafloxacin, samples for DNA analysis were reportedly available for 97/122 in M00-224 and 14/18 in M01-301.

Per the Applicant's report of pooled data from the 2 studies, ABCB1 polymorphisms, UGT2B4*2, or UGT1A1*28 did not influence any of the PK parameters. However, it is not clear how many subjects with both genotype and PK data were included in this analysis. The Applicant's analysis concluded that in the multiple rising dose segment of M00-224 in N=27/30 subjects with both genotype and PK data, day 5 dose-normalized trough mean plasma drug concentration (C_{min}) significantly correlated with UGT2B15*2 genotype (ANOVA F=3.8, p=0.036) (Figure 1). Delafloxacin day 5 $t_{1/2}$ reportedly trended in the same direction as C_{min}, but was not statistically significant. No difference by UGT2B15*2 genotype group was reported for single-dose PK parameters.

The C_{min} data may suggest a role for UGT2B15 polymorphism in the metabolism of delafloxacin, though the sample size is limited and may not be representative of the entire trial population. The Applicant concluded that further confirmation is required to elucidate the role of UGT2B15; however, no additional data are available from any of the subsequent Phase 2 or 3 studies to draw any conclusions on the impact of genetic polymorphisms on the PK of delafloxacin.

Figure 1: Relationship between UGT2B15 genotype and delafloxacin trough levels at steady state in multiple dose portion of M00-224.

Source: Figure 1 from the Study report R&D/02/260



The observation in Study M00-224 that delafloxacin C_{min} on day 5 is lowest in UGT2B15*2 T/T individuals, intermediate in G/T individuals and highest in G/G individuals may suggest a role for UGT2B15 protein in the clearance of ABT-492. However, due to the limited sample size (i.e. only three individuals for G/G genotype), further studies are needed to confirm this finding and its clinical significance.

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/s/

KUNYI WU
03/27/2017

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03/27/2017

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CLINICAL PHARMACOLOGY FILING FORM

Application Information

NDA/BLA Number	208610 & 208611	**SDN**	0001
Applicant	Melinta Therapeutics Inc.	**Submission Date**	10/19/2016
Generic Name	Delafloxacin Meglumine	**Brand Name**	Baxdela
Drug Class	Fluoroquinolone		
Indication	Treatment of Acute Bacterial Skin and Skin Structure Infection (ABSSSI)		
Dosage Regimen	The proposed dose is 300 mg IV every 12 hours (Q12h) or 450-mg oral tablet Q12h for 5 to 14 days total. Dosing in patients with severe renal impairment (eGFR of 15-29 mL/min/1.73 m^2) (b) (4) should be decreased to 200 mg IV BID (b) (4)		
Dosage Form	Oral tablet (NDA 208610) IV infusion (NDA 208611)	**Route of Administration**	Oral & IV
OCP Division	DCP 4	**OND Division**	DAIP

OCP Review Team	**Primary Reviewer(s)**	**Secondary Reviewer/ Team Leader**
Division	Kunyi Wu, Pharm.D.	Zhixia Yan, Ph.D.
Pharmacometrics	Luning Zhuang, Ph.D.	Jeffery Florian, Ph.D.
Genomics		

Review Classification	☐ Standard ☑ Priority ☐ Expedited		
Filing Date	12/18/2016	**74-Day Letter Date**	1/17/2017
Review Due Date	3/22/2017	**PDUFA Goal Date**	6/19/2017

Application Fileability

Is the Clinical Pharmacology section of the application fileable?

☑ Yes

☐ No

If no list reason(s)

Are there any potential review issues/ comments to be forwarded to the Applicant in the 74-day letter?

☐ Yes

☑ No

If yes list comment(s)

Is there a need for clinical trial(s) inspection?

☐ Yes

☑ No

If yes explain

Clinical Pharmacology Package

Tabular Listing of All Human Studies	☑ Yes ☐ No	Clinical Pharmacology Summary	☑ Yes ☐ No
Bioanalytical and Analytical Methods	☑ Yes ☐ No	Labeling	☑ Yes ☐ No

Clinical Pharmacology Studies

Study Type	Count (to-be-reviewed/total)	Comment(s) (Underlined: to-be-reviewed)
In Vitro Studies		
☑ Metabolism Characterization	1	R&D/00/552: Metabolism by rat, dog, monkey and human liver microsomes and hepatocytes;
☑ Transporter Characterization	6	(b) (4)-2011-056: Permeability across Caco-2 monolayer (b) (4)-2013-073: Substrate of P-gp, BCRP, OAT1, OAT3, OCT1, OCT2, OAP1B1 and OATP1B3; (b) (4)148044: Effect of inhibitors of P-gp and BCRP on the bidirectional permeability of delafloxacin across DKII-MDR1 and MDCKII-BRCP cells; (b) (4)-2011-055: Inhibitor of P-gp, BCRP, OAT1, OAT3, OCT1, OCT2, OATP1B1 and OATP1B3 (b) (4)-2013-072: Inhibitor of OCT2 and BSEP (b) (4)-2013-102: Inhibitor of OCT2
☑ Distribution	2	R&D /00/427: plasma bindings; R&D/00/428: Albumin and α1-acid glycoprotein bindings;
☑ Drug-Drug Interaction	8	(b) (4)134028: Substrate of UGT1A1, 1A3, 1A4, 1A6, 1A9, 2B7, and 2B15; (b) (4)135024: Inhibitor of UGT1A1 and 2B7; MWQ3034-10, R&D/01/136, and 418N-1301: Inhibitor of CYP1A2, 2A6, 2B6, 2C8, 2C9, 2C19, 2D6, 2E1 and 3A; (b) (4)133033: Inducer of CYP1A2, 2B6, and 3A4/5; (b) (4)143117: Inducer of CYP2C8, CYP2C9, and CYP2C19 R&D/000/260: Correlation of PK parameters with polymorphic sites in ABCB1, UGT2B4, UGT2B15, and UGT1A1
In Vivo Studies		
Biopharmaceutics		
☑ Absolute Bioavailability	2	RX-3341-115: absolute BA to-be-marketed tablet; RX-3341-108 (Part 1): absolute BA Phase 1 capsule;
☑ Relative Bioavailability	4/5	M01-301: BA Phase 1 and 2 capsule; M02-463: BA Phase 1,2 and potential 3 oral formulations RX-3341-106: BA of capsule formulations (Formulation A and Formulation B); RX-3341-103: BA of (b) (4) RX-3341-104: BA of Captisol (b) (4)
☐ Bioequivalence		
☑ Food Effect	4	M00-224 (Part 2): Food effect Phase 1 capsule, single dose; M02-422: Food effect Phase 2 capsule; RX-3341-109: Food effect Phase 1 capsule, multiple dose; RX-3341-116 (to-be-marketed tablet): 900 mg oral SD
☐ Other		
Human Pharmacokinetics		
Healthy	☑ Single Dose	3	M00-224 (Part 1) (oral): 50, 100, 200, 400, 800, 1200, 1600

Subjects			mg SD; RX-3341-101 (IV [redacted] solution): 50, 100, 200, 300, 400 mg SD; RX-3341-108 (Part 2) (IV captisol lyophilized): 450, 600, 750, 900, 1200 mg SD;
	☑ Multiple Dose	4	M00-224 (Part 3) (oral): 100, 200, 400, 800, 1200 mg QD for 5 days; RX-3341-102 (IV [redacted] solution): 300, 600 mg Q12h 2 doses; 150, 300, or 450 mg Q12h 10 days; RX-3341-103 (Part 2) (IV [redacted] solution): 300, 450 mg Q12h 14 days; RX-3341-104 (Part 2) (IV captisol solution): 300 mg Q12h 14 days
Patients	☐ Single Dose		
	☐ Multiple Dose		
☑ Mass Balance Study		2	M01-292 (oral): Radiolabeled ADME Following Single Oral Administration with Phase 1 capsule; RX-3341-107 (IV): Radiolabeled ADME Following Single IV Administration with captisol solution; protein binding was also measured at the time point of 1,3, and 12 h.
☐ Other (e.g. dose proportionality)			
Intrinsic Factors			
☐ Race			
☑ Sex		1	M00-224 (part 2) : 250 mg SD
☑ Geriatrics		1	M00-224 (part 2) : 250 mg SD
☐ Pediatrics		0	The Sponsor submitted "Request for waiver of pediatric studies" for both the oral and IV dosage forms (SDN #0002) on 10/26/2016.
☑ Hepatic Impairment		1	ML-3341-112 (IV: captisol lyophilized): Hepatic impairment 300 mg SD in subjects with mild, moderate, and severe hepatic impairment;
☑ Renal Impairment		1	RX-3341-110 (IV and oral): in subjects with different levels of renal impairment (mild, moderate, severe, and ESRD); protein binding was also measured;
☐ Genetics			
Extrinsic Factors			
☐ Effects on Primary Drug			
☑ Effects of Primary Drug		2	ML-3341-118: oral midazolam DDI M01-284: Photosensitivity
Pharmacodynamics			
☐ Healthy Subjects			
☐ Patients			
Pharmacokinetics/Pharmacodynamics			
☐ Healthy Subjects			
☐ Patients			
☑ QT		0/2	M01-365 (oral): 200, 800, 1200 mg (This study does not have

		a positive control.); RX-3341-111 (IV): 300, 900 mg SD; a thorough TQTc study
Pharmacometrics		
☑ Population Pharmacokinetics	3	PK-3341-009: Delafloxicin IV formulation; PK-3341-018: Delafloxicin oral formulation; PK-3341-010: Population Modeling and Simulation of sulfobutylether Cyclodextrin Pharmacokinetics in Subjects with Moderate and Severe Renal Impairment
☑ Exposure-Efficacy	1	PK-3341-015: PK-PD and PK-PD Target Attainment Analyses for Delafloxacin Based on Data from Patients with Acute Bacterial Skin and Skin Structure Infections Enrolled in Phase 2 and 3 Studies
☐ Exposure-Safety	0	

Total Number of Studies	**In Vitro**	17	**In Vivo**	27
Total Number of Studies to be Reviewed		17		24

Criteria for Refusal to File (RTF)		
RTF Parameter	**Assessment**	**Comments**
1. Did the applicant submit bioequivalence data comparing to-be-marketed product(s) and those used in the pivotal clinical trials?	☐Yes ☐No ☑N/A	
2. Did the applicant provide metabolism and drug-drug interaction information? (Note: RTF only if there is complete lack of information)	☑Yes ☐No ☐N/A	
3. Did the applicant submit pharmacokinetic studies to characterize the drug product, or submit a waiver request?	☑Yes ☐No ☐N/A	
4. Did the applicant submit comparative bioavailability data between proposed drug product and reference product for a 505(b)(2) application?	☐Yes ☐No ☑N/A	
5. Did the applicant submit data to allow the evaluation of the validity of the analytical assay for the moieties of interest?	☑Yes ☐No ☐N/A	
6. Did the applicant submit study reports/rationale to support dose/dosing interval and dose adjustment?	☑Yes ☐No ☐N/A	
7. Does the submission contain PK and PD analysis datasets and PK and PD parameter datasets for each primary study that supports items 1 to 6 above (in .xpt format if data are submitted electronically)?	☑Yes ☐No ☐N/A	The Sponsor only included the pdf file for SBECD plasma concentration dataset in Study PK-3341-010. The Sponsor should submit .xpt format data for this dataset.
8. Did the applicant submit the module 2 summaries (e.g. summary-clin-pharm, summary-biopharm, pharmkin-written-summary)?	☑Yes ☐No ☐N/A	
9. Is the clinical pharmacology and biopharmaceutics section of the submission legible, organized, indexed and paginated in a manner to allow substantive review to begin? If provided as an electronic submission, is the electronic submission searchable, does it have appropriate hyperlinks and do the hyperlinks work leading to appropriate sections, reports, and appendices?	☑Yes ☐No ☑N/A	
Complete Application **10.** Did the applicant submit studies including study reports, analysis datasets, source code, input files and key analysis output, or justification for not conducting studies, as agreed to at the pre-NDA or pre-BLA meeting? If the answer is 'No', has the sponsor submitted a justification that was	☑Yes ☐No ☐N/A	

previously agreed to before the NDA submission?		
Criteria for Assessing Quality of an NDA (Preliminary Assessment of Quality) Checklist		
Data		
1. Are the data sets, as requested during pre-submission discussions, submitted in the appropriate format (e.g., CDISC)?	☑Yes ☐No ☐N/A	
2. If applicable, are the pharmacogenomic data sets submitted in the appropriate format?	☐Yes ☐No ☑N/A	
Studies and Analysis		
3. Is the appropriate pharmacokinetic information submitted?	☑Yes ☐No ☐N/A	
4. Has the applicant made an appropriate attempt to determine reasonable dose individualization strategies for this product (i.e., appropriately designed and analyzed dose-ranging or pivotal studies)?	☑Yes ☐No ☐N/A	
5. Are the appropriate exposure-response (for desired and undesired effects) analyses conducted and submitted as described in the Exposure-Response guidance?	☑Yes ☐No ☐N/A	
6. Is there an adequate attempt by the applicant to use exposure-response relationships in order to assess the need for dose adjustments for intrinsic/extrinsic factors that might affect the pharmacokinetic or pharmacodynamics?	☑Yes ☐No ☐N/A	
7. Are the pediatric exclusivity studies adequately designed to demonstrate effectiveness, if the drug is indeed effective?	☐Yes ☐No ☑N/A	
General		
8. Are the clinical pharmacology and biopharmaceutics studies of appropriate design and breadth of investigation to meet basic requirements for approvability of this product?	☑Yes ☐No ☐N/A	
9. Was the translation (of study reports or other study information) from another language needed and provided in this submission?	☐Yes ☐No ☑N/A	

Filing Memo

Information Requests listed below were conveyed to the Sponsor on 12/25/2016 and 12/1/2016, respectively.

1. As you are using the model to predict exposures for doses less than 300 mg, we request that you include studies with doses lower than 300 mg in your population PK analysis for IV delafloxacin. Specifically, please include study RX-3341-101 in the population PK model for IV delafloxacin and update the population PK parameter estimates for IV delafloxacin based on inclusion of this study.
2. Please provide datasets, modeling control streams, and a define file for the sulfobutylether cyclodextrin population PK model. Datasets should be submitted as SAS transport files (*.xpt). A description of each data item should be provided in a define.pdf file. Please provide the modeling control stream for the complete model mentioned on page 19 of PK-3341-010 final report. If available, provide additional PK data from sulfobutylether cyclodextrin from studies other than RX-3341-110 and update the population PK model.
3. If available, provide an updated population PK model for delafloxacin that uses all available IV and oral PK data (including all the available dose levels) or provide a justification why the IV and oral PK for delafloxacin cannot be characterized using the same population PK model.
4. Provide tables and/or plots comparing the simulated exposures (AUC and Cmax) of IV delafloxacin, sulfobutylether cyclodextrin and oral delafloxacin based on the appropriate pop PK model for the following patient groups using the dosing proposed in labeling: patients with normal renal function, patients with mild renal impairment, patients with moderate renal impairment, patients with severe renal impairment, (b) (4) .
5. According to the proposed dosing regimen, one option is to treat patients with 450 mg oral tablet Q12 hours for 5 to 14 days. However, this proposed oral only dosing regimen was not evaluated in any ABSSSI patients. In addition, systemic exposure of delafloxacin resulted from the to-be-marketed tablet was only measured in healthy subjects, but not in ABSSSI patients. Please clarify whether ABSSSI has any impact on the pharmacokinetics, especially absorption, of delafloxacin for the oral tablet.

APPENDIX

Table 1. Listing of Delafloxacin Clinical Pharmacology Studies

Study Number	Route	Description	Delafloxacin Dose	Formulation	No. Subjects Enrolled (Treated[a]/Placebo)
Phase 1 Studies with Clinical Pharmacology Data Summarized in Section 2.7.2					
M01-292	Oral	Radiolabeled ADME	200 mg SD	Phase 1 Capsule	6 (6/0)
M00-224	Oral	SAD & MAD, Sex, Age	Part 1: 50, 100, 200, 400, 800, 1200, 1600 mg SD Part 2: 250 mg SD (age and sex) Part 3: 100, 200, 400, 800, 1200 mg QD for 5 days	Phase 1 Capsule	Part 1: 56 (42/14) Part 2: 52 (40/12) Part 3: 60 (40/20)
RX-3341-110 (Oral)[b]	Oral	Renal Impairment	400 mg SD	Phase 1 Capsule	42 (42/42)
M01-365	Oral	QTc	200, 800, 1200 mg	Phase 2 Capsule	68 (68/68)
M01-284	Oral	Photosensitivity	200, 400 mg QD for 6 days	Phase 1 Capsule	52 (26/13)
RX-3341-107	IV	Radiolabeled ADME	300 mg SD	Captisol Solution	6 (6/0)
RX-3341-101	IV	SAD	50, 100, 200, 300, 400 mg SD	(b) (4) Solution	43 (30/12)
RX-3341-108 (Part 2)	IV	SAD	Part 2: 450, 600, 750, 900, 1200 mg SD	Captisol Lyophilized	Part 2: 50 (40/10)
RX-3341-102	IV	MD	300, 600 mg Q12h 2 doses 150, 300, or 450 mg, Q12h 10 days	(b) (4) olution	32 (24/8)
RX-3341-103 (Part 2)	IV	MAD	300, 450 mg Q12h 14 days	Solution	20 (16/4)
RX-3341-104 (Part 2)	IV	MAD	300 mg Q12h 14 days	Captisol Solution	12 (8/4)
RX-3341-110 (IV) [b]	IV	Renal Impairment	300 mg SD	Captisol Lyophilized	44 (44/44)
RX-3341-111	IV	TQTc	300, 900 mg SD	Captisol Lyophilized	52 (52/52)

Study Number	Route	Description	Delafloxacin Dose	Formulation	No. Subjects Enrolled (Treated[a]/Placebo)
ML-3341-112	IV	Hepatic Impairment	300 mg SD	Captisol Lyophilized	36 (36/0)
ML-3341-118	Oral	Midazolam DDI	450 mg Q12h 6 days	To-be-marketed Tablet	22 (22/0)
Phase 1 Studies with Biopharmaceutics Data Summarized in Section 2.7.1					
RX-3341-115	IV and Oral	Absolute BA To-be-marketed Tablet	450, 900 mg oral SD 300 mg IV SD	Captisol Lyophilized To-be-marketed Tablet	76 (76/0)
RX-3341-116	Oral	Food Effect To-be-marketed Tablet	900 mg oral SD	To-be-marketed Tablet	30 (30/0)
M01-301	Oral	BA Phase 1 and 2 Capsule	200 mg oral SD	Phase 1 and Phase 2 Capsules	18 (18/0)
RX-3341-108 (Part 1)	IV and Oral	Absolute BA Phase 1 Capsule	300 mg oral SD 300 mg IV SD	Phase 1 Capsule and Captisol Solution	12 (12/0)
M00-224 (Part 2 Food Effect)	Oral	Food Effect Phase 1 Capsule	250 mg SD	Phase 1 Capsule	20 (16/4)
RX-3341-109	Oral	Food Effect Phase 1 Capsule	400 mg QD on Day 1; Q12h Days 2–10	Phase 1 Capsule	105 (70/0)
M02-422	Oral	Food Effect Phase 2 Capsule	200 mg SD	Phase 2 Capsule	21 (21/0)
M02-463	Oral	BA Phase 1, 2 and Potential Phase 3 Formulations	200 mg SD	Phase 1 Capsule, Phase 2 Capsule, (b) (4) (b) (4) Capsule Formulations	24 (24/0)
RX-3341-106	Oral	BA of Capsule Formulations	400 mg SD	Phase 1 Capsule, (b) (4) Capsule	16 (16/0)

Study Number	Route	Description	Delafloxacin Dose	Formulation	No. Subjects Enrolled (Treated[a]/Placebo)
RX-3341-113	IV and Oral	Salt Formulation Development	400 mg Oral SD 300 mg IV SD	Phase 1 Capsule Captisol Lyophilized 3 Other Capsule Formulations[c]	100 (100/0)
RX-3341-114	Oral	Tablet Formulation Development	400, 450, 475, 500 mg	(b) (4) (b) (4) Tablet and (b) (4) Tablet	20 (20/0)
RX-3341-103 (Part 1)	IV	BA of Solutol and (b) (4)	300 mg	(b) (4)	12 (12/0)
RX-3341-104 (Part 1)	IV	BA of Captisol and (b) (4)	300 mg	Captisol (b) (4)	12 (12/0)

ADME = absorption, distribution, metabolism, and excretion; IV = intravenous; DDI = drug-drug interaction; MAD = multiple ascending dose; MD = multiple dose; QD = once daily; QTc = corrected QT interval; SAD = single ascending dose; SD = single dose; Q12h = every 12 hours; TQTc = thorough QTc study.

[a] Delafloxacin-treated subjects.

[b] The renal impairment study was a crossover design. A total of 44 were enrolled and received 3 treatments (300 mg IV delafloxacin, 400 mg oral delafloxacin and Captisol vehicle). Two subjects in RX-3341-110 received IV delafloxacin (total of 44 subjects) but discontinued prior to receiving oral delafloxacin (total of 42 subjects).

(b) (4)

Table 2. Phase 2 and Phase 3 Trials for Delafloxacin in ABSSSI

Study No.	Design	Delafloxacin Regimen	Comparator Regimen	No. of Subjects
Phase 2				
RX-3341-201	Multicenter, randomized, double-blind comparative study (tigecycline)	300 or 450 mg Q12h IV	Tigecycline: 100 loading and 50 mg Q12h IV	150
RX-3341-202	Multicenter, randomized, double-blind, comparative study (vancomycin or linezolid)	300 mg Q12h IV	Vancomycin: 15 mg/kg Q12h or local standard of care Linezolid: 600 mg Q12h IV (with concomitant aztreonam in linezolid and vancomycin patients allowed if Gram-negative cultures were positive)	256
Phase 3				
RX-3341-302	Multicenter, randomized, double-blind, active-controlled (vancomycin and aztreonam)	300 mg Q12h IV	Vancomycin: 15 mg/kg (actual body weight) Q12h (with concomitant aztreonam)	660
RX-3341-303	Multicenter, randomized, double-blind, active-controlled (vancomycin and aztreonam)	300 mg Q12h IV for 6 doses then 450 mg oral Q12h	Vancomycin: 15 mg/kg Q12h (actual body weight) or local standard (with concomitant aztreonam)	850

IV = intravenous; Q12h = every 12 hours

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/s/

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12/15/2016

ZHIXIA YAN
12/15/2016