CLINICAL RESEARCH IN INFECTIOUS DISEASES

STATISTICAL ANALYSIS PLAN
for
DMID Protocol: 16-0058
Study Title:

A Phase 1 Safety and Intrapulmonary Pharmacokinetics Study of ZTI-01 (Intravenous Fosfomycin Disodium) in Healthy Adult Subjects

NCT03910673

Version 1.0

DATE: 20-FEB-2020

STUDY TITLE

Protocol Number Code:	**DMID Protocol: 16-0058**
Development Phase:	Phase 1
Products:	Intravenous (IV) fosfomycin disodium (ZTI-01)
Form/Route:	Intravenous infusion
Indication Studied:	Microbial Infection
Sponsor:	Division of Microbiology and Infectious Diseases National Institute of Allergy and Infectious Diseases National Institutes of Health
Clinical Trial Initiation Date:	11JUN2019
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This study was performed in compliance with Good Clinical Practice.

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LIST OF ABBREVIATIONS

AE	Adverse Event
ALP	Phosphorus, Alkaline Phosphatase
ATC	Anatomical Therapeutic Classification
ALT	Alanine Aminotransferase
AM	Alveolar Macrophage
aPTT	Partial Thromboplastin Time
AST	Aspartate Aminotransferase
$AUC_{(0\text{-}inf)}$	AUC Extrapolated To Infinity
$AUC_{(0\text{-}t)}$	AUC From Time Of Dosing To Time *T*.
AUMC	Area Under First Moment Curve
BAL	Bronchoalveolar Lavage
BLQ	Below Quantification Limit
BMI	Body Mass Index
BP	Blood Pressure
BSV	Between Subject Variability
BUN	Blood Urea Nitrogen
C	Degree(S) Celsius
CI	Confidence Interval
C_{max}	Maximum Measured Plasma Concentration
CPK	Creatine Phosphokinase
CRF	Case Report Form
CSR	Clinical Study Report
CV	Coefficient Of Variation
DCC	Data Coordinating Center
DMID	Division Of Microbiology And Infectious Diseases
DSMB	Data And Safety Monitoring Board
ECG	Electrocardiogram
ELF	Epithelial Lining Fluid
GM	Geometric Mean

List of Abbreviations *(continued)*

GSD	Geometric SD
h	Hour(S)
hCG	Human Chorionic Gonadotropin
HCV	Hepatitis C Virus
HIV	Human Immunodeficiency Virus
ICH	International Conference On Harmonisation
IRB	Institutional Review Board
ISM	Independent Safety Monitor
IV	Intravenous
L	Liter
LDH	Lactate Dehydrogenase
LLN	Lower Limit Of Normal
LLOQ	Lower Limit Of Quantification
max	Maximum
mcg	Microgram
MedDRA	Medical Dictionary For Regulatory Activities
mEq	Milliequivalent
mg	Milligram
MH	Medical History
min	Minimum
mL	Milliliter
MOP	Manual Of Procedures
N	Number (Typically Refers To Subjects)
NCA	Noncompartmental Analysis
NIH	National Institutes Of Health
PI	Principal Investigator
PK	Pharmacokinetics
PT	Prothrombin Time
RBC	Red Blood Cell
RCD	Reverse Cumulative Distribution
RV	Residual Variability

List of Abbreviations *(continued)*

SAE	Serious Adverse Event
SAP	Statistical Analysis Plan
SD	Standard Deviation
SDCC	Statistical And Data Coordinating Center
SDTM	Standard Data Tabulation Model
SMC	Safety Monitoring Committee
SOC	System Organ Class
SOP	Standard Operating Procedures
$t_{1/2}$	Terminal Elimination Half-Life
TEAE	Treatment Emergent Adverse Event
TFLs	Tables, Figures, And Listings
t_{max}	Time To Peak Concentration
U	Units
ULN	Upper Limit Of Normal
V_d	Volume Of Distribution
VPC	Visual Predictive Checks
WBC	White Blood Cell
WHO	World Health Organization
λ_z	Terminal-Phase Elimination Rate Constant
ZTI-01	Intravenous Fosfomycin Disodium

1. PREFACE

The Statistical Analysis Plan (SAP) for "A Phase 1 Safety and Intrapulmonary Pharmacokinetics Study of ZTI-01 (Intravenous Fosfomycin Disodium) in Healthy Adult Subjects" (DMID Protocol 16-0058) describes and expands upon the statistical information presented in the protocol.

This document describes all planned analyses and provides reasons and justifications for these analyses. It also includes sample tables, listings, and figures planned for the final analyses. Regarding the final analyses and Clinical Study Report (CSR), this SAP follows the International Conference on Harmonisation of Technical Requirements for Registration of Pharmaceuticals for Human Use (ICH) Guidelines, as indicated in Topic E3 (Structure and Content of Clinical Study Reports), and more generally is consistent with Topic E8 (General Considerations for Clinical Trials) and Topic E9 (Statistical Principles for Clinical Trials). The structure and content of the SAP provides sufficient detail to meet the requirements identified by the FDA and ICH, while all work planned and reported for this SAP will follow internationally accepted guidelines published by the American Statistical Association and the Royal Statistical Society for statistical practice.

Any deviation from this SAP will be described and justified in protocol amendments and/or in the CSR, as appropriate. The reader of this SAP is encouraged to also review the study protocol for details on conduct of the study and the operational aspects of clinical assessments.

2. INTRODUCTION

This is a Phase 1, open-label, multiple-dose trial conducted at a single center. The treatment period will consist of three 6 g doses of ZTI-01 as a 1 h IV infusion. A total of 30 enrolled subjects will be randomized. Six additional enrolled subjects will act as alternates. An evaluable subject is defined as a subject who receives all doses of ZTI-01, undergoes bronchoalveolar lavage (BAL) at the randomized sampling timepoint with BAL return volume adequate for testing, and undergoes at least the one blood sampling timepoint that is concurrent with the assigned BAL sampling timepoint, with blood sampling volume that is adequate for testing.

2.1. Purpose of the Analyses

These analyses will assess the safety and plasma and intrapulmonary pharmacokinetics (PK) of three 6 g doses of ZTI-01 administered as three 1 h IV infusions.

3. STUDY OBJECTIVES AND ENDPOINTS

3.1. Study Objectives

Safety:

- Determine the safety of multiple doses of ZTI-01 (6 g dose every 8 hours, administered as a 1-hour IV infusion, for a total of 18 g in three doses).

Pharmacokinetic:

- Determine the plasma and intrapulmonary PK (epithelial lining fluid (ELF) and alveolar macrophage (AM) concentrations of ZTI-01) after multiple doses of ZTI-01 (6 g dose every 8 hours, administered as a 1-hour IV infusion, for a total of 18 g in three doses).

3.2. Endpoints

Safety:

- The safety outcome measure will be the occurrence of adverse events (AEs), summary of physical exam findings, vital signs, clinical laboratory tests, and electrocardiogram (ECGs) at any time from the start of study drug administration through the end of subject follow-up.
- All subjects who receive at least one dose of study drug will be included in the safety outcome measure. AEs will be collected from the time of dosing on Day 1 through the telephone follow-up interview on Day 3. Treatment Emergent Adverse Events (TEAEs) will be assessed by severity and relationship to the study drug as determined by the investigator.

Pharmacokinetics:

- The pharmacokinetic outcome measures will be plasma and intrapulmonary concentrations of ZTI-01 by validated bioassays.
- Plasma PK parameters will be derived from the plasma concentration vs. time curves after the third dose of study drug. The parameters include, but are not limited to:
  - Maximum measured plasma concentration (C_{max})
  - Area under the concentration vs. time curve (AUC_{0-8} and AUC_{0-inf})
  - Time to peak concentration (t_{max})
  - Terminal elimination half-life ($t_{1/2}$)
  - Terminal-phase elimination rate constant (λ_z)
  - Volume of distribution (V_d)
  - Clearance (CL)

Intrapulmonary PK parameters will be derived from the ratios of drug concentrations in ELF and AMs to simultaneous drug concentrations in plasma for each subject and summarized for each sampling time. The median concentrations of fosfomycin from the BAL sampling times will be used to estimate the AUC_{0-8} of plasma, ELF, and AM. The ratios of AUC_{0-8} of ELF-to-plasma and AM-to-plasma will be calculated to determine the percent lung penetration.

3.3. Study Definitions and Derived Variables

3.3.1. Fosfomycin Concentrations in ELF and AM

Concentration of Fosfomycin in ELF

The volume of ELF recovered in the BAL samples will be estimated using the urea dilution method [1]. The estimated concentration of fosfomycin in ELF ($Fosfomycin_{ELF}$) will be calculated as follows:

$$Fosfomycin_{ELF} = Fosfomycin_{BAL} \times (Urea_{Plasma}/Urea_{BAL})$$

Where $Fosfomycin_{BAL}$ is the measured fosfomycin concentration in the BAL sample and $Urea_{Plasma}$ and $Urea_{BAL}$ are the measured concentrations of urea in plasma and BAL fluid, respectively.

3.3.2. Definition of AEs

Adverse Events

ICH E6 defines an AE as any untoward medical occurrence in a patient or clinical investigation subject administered a pharmaceutical product, and that does not necessarily have a causal relationship with this treatment. An AE can therefore be any unfavorable and unintended sign (including a clinically significant laboratory abnormality), symptom, or disease temporally associated with use of the study drug, whether or not it is deemed related to the study drug. The occurrence of an AE may come to the attention of study personnel during study visits and interviews of a study subject presenting for medical care, or upon review by a study monitor.

All AEs including local and systemic reactions not meeting the criteria for serious adverse events (SAEs) will be captured on the appropriate case report form (eCRF). Information to be collected includes event description, time of onset, clinician's assessment of severity, relationship to study drug (assessed only by those with the training and authority to make a diagnosis), outcome, action taken, and time of resolution/stabilization of the AE. All AEs occurring while on study will be documented appropriately regardless of relationship. All AEs will be followed until they adequately resolve or are determined by the PI to be medically stable (stable or improving over two assessments and no further intervention required). Any medical condition that is present at the time that the subject is screened will be considered as a baseline finding and not reported as an AE. However, if its Grade increases at any time during the trial such that it meets the AE definition specified above, it will be recorded as an AE.

All systemic and laboratory AEs will be graded using the Adverse Event Toxicity Grading Scale (See Protocol v6.0 Appendix B). Laboratory toxicity grading will be determined by using these tables as modified for the study to accommodate the reference range of the Central Laboratory and the acceptable ranges used in determining eligibility. If a subject was enrolled in the trial with a laboratory value that is outside the reference range, but within the acceptable range, an AE will be recorded if it otherwise meets the definition of AE in Protocol v6.0 Section 9.3 (occurred after the start of infusion of the first dose of study product) and the on-study value is higher (if initially high) or lower (if initially low) than the screening value.

Treatment Emergent Adverse Event (TEAE)

A TEAE is defined as an AE with an onset or increase in either severity or frequency after study drug administration. Since the reporting period for AEs in this protocol is from the time of dosing on Day 1 through the Telephone Follow-up Interview on Day 3, all recorded AEs are by definition TEAEs.

Severity of Event

All AEs will be assessed by the clinician using the protocol-defined grading system.

Changes in the severity of an AE will be documented to allow an assessment of the duration of the AE at each level of intensity to be performed. AEs characterized as intermittent require documentation of onset and duration of each episode.

Relationship to Study Products:

The clinician's assessment of an AE's relationship to the study drug is part of the documentation process, but it is not a factor in determining what is or is not reported in the trial. If there is any doubt as to whether a clinical observation is an AE, it will be reported. All AEs will have their relationship to the study drug assessed using the terms "related" or "not related." In a clinical trial, the study drug must always be suspect. To help assess relatedness, the following guidelines are used.

- Related: There is a reasonable possibility that the study drug caused the AE. Reasonable possibility means that there is evidence to suggest that the study drug caused the AE.
- Not Related: There is not a reasonable possibility that the study drug caused the AE.

Serious Adverse Events (SAE)

An AE or suspected AE is considered "serious" if, in the view of either the investigator or Sponsor, it results in any of the following outcomes:

- Death,
- a life-threatening AE,
- An adverse event is considered life-threatening if, in the view of either the site principal investigator or sponsor, its occurrence places the patient or subject at immediate risk of death. It does not include an adverse event that, had it occurred in a more severe form, might have caused death.
- inpatient hospitalization or prolongation of existing hospitalization,
- a persistent or significant incapacity or substantial disruption of the ability to conduct normal life functions, or
- a congenital anomaly/birth defect.
- Important medical events that may not result in death, be life-threatening, or require hospitalization may be considered serious when, based upon appropriate medical judgment they may jeopardize the subject and may require medical or surgical intervention to prevent one of the outcomes listed in this definition. Examples of such medical events include allergic bronchospasm requiring intensive treatment in an emergency room or at home, blood dyscrasias or convulsions that do not result in inpatient hospitalization, or the development of drug dependency or drug abuse.

All SAEs will be:

- Assessed for severity and relationship to the study drug and alternate etiology,
- Recorded on the appropriate SAE data collection form and eCRF,
- Followed through resolution,

- Reviewed and evaluated by the Independent Safety Monitor (ISM) (as deemed necessary), Safety Monitoring Committee (SMC) (periodic review unless related), DMID, and IRB.

4. INVESTIGATIONAL PLAN

4.1. Overall Study Design and Plan

This is a Phase 1, open-label, multiple-dose trial conducted at a single center. The treatment period will consist of three 6 g doses of ZTI-01 as a 1 h IV infusion. A total of 30 enrolled subjects will be randomized. Six additional enrolled subjects will act as alternates. An evaluable subject is defined as a subject who receives all doses of ZTI-01, undergoes BAL at the randomized sampling timepoint with BAL return volume adequate for testing, and undergoes at least the one blood sampling timepoint that is concurrent with the assigned BAL sampling timepoint, with blood sampling volume that is adequate for testing. Subjects who initiate the Treatment Phase but are not evaluable will be replaced with an alternate at the sample BAL sampling timepoint.

Each subject will complete Screening, Baseline, Treatment, and Follow-up Phases. A diagram of the overall study design is shown in Figure 1. The Screening Phase will be conducted on an outpatient basis within 30 days of the Baseline Phase. The Baseline Phase will consist of admission to DEPRU on the day before the first dose (Day -1) for pre-dosing assessments. The Treatment Phase (Day 1 to Day 2) will include dosing 6 g of ZTI-01, administered as a 1-hour IV infusion, every 8 hours for a total 18 g in 3 doses, followed by post-treatment safety assessments, and BAL and blood sample collections for PK measurements. Each subject will undergo a single standardized bronchoscopy with BAL after the start of the last IV infusion of ZTI-01. Blood samples for PK measurements will be collected before each dose, up to 8 hours after the start of the first IV infusion of ZTI-01, and up to 12 hours after the start of the third IV infusion.

Subjects will remain in DEPRU during the Treatment Phase and will be discharged at least 12 hours after the start of the third IV infusion (if safety parameters are acceptable to the investigator). Safety assessments will be conducted during the entire Treatment Phase. The Follow-Up Phase will consist of a telephone interview on Day 3 for symptom-driven safety assessments.

4.2. Discussion of Study Design, Including the Choice of Control Groups

As the main focus of this protocol is to evaluate the safety and pharmacokinetics of multiple doses of ZTI-01, there is no control group and all subjects receive the same dose of ZTI-01with the same PK blood collection timepoints. Subjects will be randomized to one of five BAL sampling timepoints with a total of 6 subjects assigned to each BAL sampling time.

4.3. Selection of Study Population

The study population for this trial is 36 healthy male and female adults aged 18-45 years, inclusive, with BMI 18-30 kg/m^2, inclusive, and body weight >50 kg (110 lbs.). Only subjects who meet all inclusion criteria and no exclusion criteria will be eligible for enrollment into the trial. No exemptions are granted on Inclusion/Exclusion Criteria in DMID-sponsored trials.

4.3.1. Subject Inclusion Criteria

To be considered for study enrollment, subjects must meet all of the following inclusion criteria:

1. Healthy[1] men and women aged 18-45 years with no clinically significant findings[2] at Screening and Baseline (Day -1 to Day 1)[2]

   [1]Healthy is defined by the absence of any medical condition described in the exclusion criteria in a subject with a normal physical exam including vital signs. If the subject has another current, ongoing

medical condition, the condition cannot meet any of the following criteria: 1) first diagnosed within 3 months of enrollment; 2) is worsening in terms of clinical outcome in the last 6 months; or 3) involves need for medication.
[2]Including findings on medical history, physical exam, vital signs, 12-lead ECG, or clinical laboratory tests.

2. Body Mass Index (BMI) = 18-30 kg/m^2, inclusive, and body weight >50 kg (110 lbs)
3. Females who have been surgically sterilized via bilateral oophorectomy and/or hysterectomy at least 90 days prior to Screening are considered lacking childbearing potential and will be eligible[3]

   [3]Postmenopausal females are not eligible, as the definition of menopause would require age >45, and all subjects in this study are age ≤45.
4. Females of childbearing potential must have a negative serum pregnancy test at Screening, a negative urine pregnancy test at Baseline (Day -1 to Day 1), and must use acceptable contraception[4]

   [4]Acceptable contraception methods are restricted to surgical sterilization (bilateral tubal ligation) or successful Essure placement (permanent, non-surgical, non-hormonal sterilization with documented radiological confirmation at least 90 days after the procedure), use of long-acting reversible contraceptive devices (progestin-releasing subdermal implants [Nexplanon and Implanon, Merck], copper intrauterine devices [Paragard, Teva], and levonorgestrel-releasing intrauterine devices [Mirena, Bayer; Skyla, Bayer; Liletta, Allergan/Medicines360]), licensed hormonal products such as injectables or oral contraceptives, barrier methods such as condoms or diaphragms with spermicidal agents, and abstinence from sexual intercourse with a male partner.
   Subjects must have used the above-listed method for a minimum of 30 days prior to the first dose of study drug and be willing to use the method for at least 30 days after the final dose of study drug.
5. Male subjects[5] whose partners are of childbearing age or pregnant must be willing to use condoms during the study and through the Day 3 follow-up call.

   [5]including men who have had vasectomies
6. Able to abstain from alcoholic beverages within 48 hours before Baseline (Day -1 to Day 1) and throughout the Treatment Phase
7. Able to abstain from caffeine use within 7 days before Baseline (Day -1 to Day 1) and throughout the inpatient period
8. Willing to remain in DEPRU during the Baseline and Treatment Phases
9. Have a high probability for compliance and completion of the trial
10. Sign a dated, witnessed, written ICF
11. Have adequate venous access for infusions and blood draws

4.3.2. Subject Exclusion Criteria

Subjects must meet <u>none</u> of the following exclusion criteria:

1. Any surgical or medical condition that in the opinion of the investigator could interfere with drug absorption, distribution, metabolism, or excretion.
2. Any surgical or medical condition that in the opinion of the investigator may place the subject at increased risk while participating in the trial.

3. History or presence of cardiovascular disease including coronary artery disease and chronic hypertension (systolic pressure >140 mmHg or diastolic pressure >90 mmHg)
4. Abnormal ECG at screening, as determined by the investigator to be clinically significant
5. History or presence of renal impairment or chronic renal disease
6. History or presence of liver disease (ALT, AST, or total bilirubin above the upper limit of normal)
7. History or presence of chronic pulmonary disease, including asthma, requiring use of medication in the year before screening
8. History of intolerance or hypersensitivity to phosphonic acid derivative antibiotics or any of its constituents (i.e., oral or IV fosfomycin)
9. Have cancer or have a history of cancer within the past 5 years, with the exception of non-melanomatous skin cancer, treated, without evidence of recurrence
10. Any medical condition that prevents a subject from undergoing bronchoscopy with BAL
11. Serum creatinine above the upper limit of normal, or estimated creatinine clearance (CrCl) <60 mL/min as determined by Cockcroft-Gault equation[6]

    [6]*Cockcroft-Gault equation where age is in years, weight is in kilograms, and serum Cr is in mg/dL units:*
    Males: CrCl (mL/min) = (140 – age) x Weight / (72 x Cr)
    Females: CrCl (mL/min) = [(140 – age) x Weight / (72 x Cr)] x 0.85
12. History of regular alcohol consumption within 6 months of Baseline (Day -1 to Day 1)[7]

    [7]*History of regular alcohol consumption is defined as an average weekly intake of >14 drinks for males or >7 drinks for females. One drink is equivalent to 12 g of alcohol: 12 ounces (360 mL) of beer, 5 ounces (150 mL) of wine, or 1.5 ounces (45 mL) of 80 proof distilled spirits.*
13. History of ≥10 pack-years smoking, or history of any nicotine use[8] in the 6 months before Baseline (Day -1 to Day 1) or positive urine cotinine screen at Baseline (Day -1 to Day 1)[9].

    [8]*Including cigarettes, pipe, cigar, chewing tobacco, nicotine patch*
    [9]*A positive urine cotinine at screening is allowed if negative at baseline*
14. History of illicit drug use *within* 6 months of Baseline (Day -1 to Day 1)[10]

    [10]*Use of cannabinoids with 1 month of Baseline (Day -1 to Day 1) is excluded, but other use of cannabinoids within 6 months is permitted*
15. Use of any prescription drugs, except acceptable contraception methods listed above, within 30 days of Baseline (Day -1 to Day 1)
16. Involvement in other investigational studies of any type (drugs, devices, procedures) within 30 days of Baseline (Day -1 to Day 1)
17. Blood or blood products donation within 30 days of Baseline (Day -1 to Day 1)
18. Planning egg or sperm donation any time before Day 3 follow-up call
19. Use of any non-prescription medications, vitamins, consumption >2 times/week of products containing genuine licorice, caffeine, or dietary or herbal supplements within 7 days of Baseline (Day -1 to Day 1).[11]

[11]*Excluded from this list is intermittent use of acetaminophen at doses ≤2 g/day. Herbal supplements must be discontinued 7 days before the initial dose of study drug on Day 1.*

20. Presence of any acute illness, including febrile illness with temperature >37.8°C (>100.0°F), within 7 days of Baseline (Day -1 to Day 1)
21. Currently pregnant or breastfeeding as determined by subject report
22. Positive tests for human immunodeficiency virus (HIV) 1 and 2 antibodies, hepatitis B surface antigen (HBsAg), or hepatitis C virus (HCV) antibody
23. Positive urine drug or positive breathalyzer test for alcohol at Screening or Baseline (Day -1 to Day 1) or positive cotinine at Baseline (Day -1 to Day 1)
24. Weight loss or gain of >10% within 30 days of Baseline (Day -1 to Day 1)
25. Any laboratory value at screening or enrollment that is Grade 2 or more (Protocol Appendix B). A laboratory value that is Grade 1 will be allowed if not considered to be clinically significant by the investigator.[12]

[12]*Excluded from this list of permissible Grade 1 laboratory values are ALT, AST, or Total bilirubin per Exclusion Criteria 6; serum creatinine per Exclusion Criteria 11; and serum electrolytes including sodium, potassium, calcium, phosphorus and magnesium.*

4.4. Treatments

4.4.1. Treatments Administered

Each subject will receive three 6 g doses of ZTI-01, administered as a 1-hour IV infusion, every 8 hours for a total 18 g in 3 doses.

4.4.2. Identity of Investigational Product(s)

The non-US commercial drug product that will be used in the trial is a single vial of ZTI-01 disodium powder (4 g presentations with recrystallized succinic acid, in a Type 1 glass vials).

Each Type 1, glass vial of ZTI-01 contains sterile white powder consisting of 5.28 g of fosfomycin disodium (equivalent to 4 g of fosfomycin and 1.28 g of sodium) and the inactive ingredient: recrystallized succinic acid. Each gram of ZTI-01 contains 330 mg of sodium. A total of two 4 g vials are required to prepare the 6 g dose.

Vials will be labeled with protocol number, product identity – 4 g of ZTI-01 (fosfomycin for injection), lot number, expiry date, and the IND caution statement "New Drug – Limited by Federal Law to Investigational Use Only," a statement to store at controlled room temperature, and a statement that each vial is for intravenous, single use only.

4.4.3. Method of Assigning Subjects to BAL Sampling Timepoint Groups (Randomization)

Subjects will be randomized to a single standardized bronchoscopy with BAL sampling timepoint upon study enrollment using an assignment schedule as follows: 30 subjects will be randomized 1:1:1:1:1 to one of the five BAL sampling timepoints (30 minutes, 75 minutes, 2 hours, 5 hours, or 8 hours after the start of the third IV infusion of ZTI-01). Up to six (6) subjects will be enrolled as alternates. Alternates will arrive at DEPRU on Day -1 to Day 1 and will adopt the planned BAL sampling timepoint of any other enrolled subject who fails to present for check-in or who fails to meet inclusion/exclusion criteria on Day -1 to Day 1. If all

scheduled subjects present for check-in and meet Day -1 to Day 1 inclusion/exclusion criteria, then alternates will not undergo any further study procedures at that time. In that case, alternates may be assigned to a new bronchoscopy time on a future date, alternate status for a future date, or as replacement subjects.

Subjects will be enrolled online using the enrollment module of AdvantageEDC^{SM} software. The randomization code will be prepared by DCC statisticians and included in the enrollment module for this study. AdvantageEDC will assign each subject to a sampling timepoint after the demographic and eligibility data have been entered.

4.4.4. Selection of Doses in the Study

The dosage selection was directly influenced by applying PK-PD principles of anti-infective agents. Recent studies have provided useful information for modernizing and optimizing fosfomycin dosing regimens. VanScoy et al found that the PK-PD parameter that most strongly associated with efficacy was the percentage of fosfomycin concentrations above the resistance inhibitory concentration (RIC) [% T>RIC] during the dosing interval [2]. The proposed magnitude % T>RIC associated with net bacterial stasis and a 1-$\log_{10}$ and 2-$\log_{10}$ colony forming unit (CFU) reduction in bacterial density over 24 hours was 11.8, 20.9, and 32.7%, respectively. Total daily fosfomycin doses of 8 g and 32 g resulted in a rapid reduction in bacterial density without amplification of a drug-resistant subpopulation. A subsequent study suggested that the lowest fosfomycin dosing regimen that did not amplify a drug-resistant bacterial subpopulation was 4 g administered every 8 hours [3]. An in-vivo study performed in a neutropenic murine thigh model [4] used dose fractionation to assess PK-PD targets and survival. This study demonstrated ZTI-01 potency against *E. coli*, *K. pneumoniae*, and *P. aeruginosa*, including ESBL-producing and carbapenem-resistant phenotypes *in vivo*. In this 24-hour model, the index AUC/MIC was most closely linked with efficacy. Notably, targets for AUC/MIC and T>Threshold were similar between organism groups. Enterobacteriaceae stasis was noted at AUC/MIC exposures of ~20 and maximal survival was observed at exposures similar to stasis endpoints.

4.4.5. Selection and Timing of Dose for Each Subject

This trial will evaluate a single dose sequence of ZTI-01: three 6-g doses of fosfomycin, administered as a 1-hour IV infusion, every 8 hours. Refer to Section 4.4.4 for additional details regarding the selection for the dose timing schedule.

4.4.6. Blinding

This is an open-label, unblinded study. Blinding (masking) is not needed.

4.4.7. Prior and Concomitant Therapy

Medications to treat any AEs the subject experiences during the trial are permitted. All information on concomitant medications will be recorded on the subject's eCRF and will include the name of the procedure or drug and duration of the treatment (start/stop times), dosages, and route. The intermittent intake of acetaminophen (maximum 2 g/day) is permitted throughout the trial if approved by the Site PI. Subjects are required to refrain from the use of all prescription medications for at least 30 days before administration of the first dose of study drug on Day 1 until the last PK sample is taken.

Herbal supplements and over-the-counter meds (except acetaminophen at doses ≤2 g/day) must be discontinued 7 days before the first dose of study drug until completion of the Treatment Phase. The use of illegal drugs is prohibited within the 6 months prior to Baseline (Day -1 to Day 1) and is not permitted while subjects are enrolled in this study. A urine drug screen will be performed at Screening. If a subject is unable to

comply with the restrictions described above, the subject's continued participation in the trial will be re-evaluated by the investigator.

4.4.8. Treatment Compliance

Study staff will be present for and during all study drug administrations. Visual inspection by study staff of the complete infusion will be appropriately recorded in the eCRF. All interruptions or premature terminations of infusion will be documented.

4.5. Safety and PK Variables

The following section describes the variables related to the safety and PK endpoints collected throughout the study. For a detailed schedule of procedures refer to Table 1.

4.5.1. Safety Variables

The type, incidence, relatedness, and severity of TEAEs and SAEs will be recorded from time of first dosing on day 1 through the follow-up call on Day 3. Refer to Section 3.3 for definitions of AEs, TEAEs, SAE, severity and relatedness.

The following safety laboratory parameters and vital signs will be summarized for baseline and each post-dose timepoint collected. Change from baseline will be summarized for all post dose timepoints. The most recent measurement prior to start of infusion of the first ZTI-01 dose will be regarded as baseline. For parameters with multiple pre-dose measurements, screening values prior to the baseline measurement will be listed only.

Serum chemistry:

- Albumin, glucose, blood urea nitrogen (BUN), potassium, magnesium, calcium, sodium, total protein, creatinine, triglycerides, total cholesterol, creatine phosphokinase (CPK), phosphorus, alkaline phosphatase (ALP), aspartate aminotransferase (AST), alanine aminotransferase (ALT), total bilirubin, direct bilirubin, and lactate dehydrogenase (LDH) will be collected at Screening and Day 2 before discharge. Albumin, random glucose, BUN, potassium, magnesium, calcium, sodium, phosphorus, total protein, creatinine, CPK, ALP, AST, ALT, and total bilirubin will be collected at Baseline (Day -1 to Day 1). Sodium, potassium, magnesium, calcium, and phosphorus will be collected approximately 2 hours prior to the assigned bronchoscopy timepoint.

Hematology:

- Hemoglobin, hematocrit, platelet count, white blood cell (WBC) count, neutrophils, lymphocytes, monocytes, eosinophils, basophils will be collected at Screening and Day 2 before discharge.

Coagulation:

- Prothrombin time (PT) and activated partial thromboplastin time (aPTT) will be collected at Screening and Day 2 before discharge.

Urinalysis:

- Leukocyte esterase, blood, pH, specific gravity, glucose, protein. A microscopic test will be performed if urinalysis is abnormal. The microscopic urinalysis result for the presence of blood will supersede the dipstick urinalysis result. Urinalyses performed at Screening and Day 2 before discharge.

Vital signs including heart rate, systolic blood pressure, diastolic blood pressure, temperature, respiratory rate, and peripheral oxygen saturation, are obtained for the following timepoints: Screening, Baseline (Day -1 to Day 1), immediately prior (within 10 minutes) to each of three doses of Study Drug, and at approximately 0.5, 1, 2, and 5 hours after the start of infusion for each dose, and before discharge from DEPRU. Change from baseline will be summarized for all post dose timepoints. The most recent measurement prior to start of infusion of the first ZTI-01 dose will be regarded as baseline.

ECGs including QTc interval, PR Interval, and QRS Duration will be obtained at Screening, Baseline (Day -1 to Day 1), and Day 2 within 2 hours of the third (last) dose of study drug. In case of premature discontinuation of study drug, ECG is to be performed before discharge.

Physical exam: overall physical assessment (to include vital signs [temperature, blood pressure, respiratory rate, heart rate, peripheral oxygen saturation], skin, head and neck, lungs, heart, liver, spleen, extremities, lymph nodes, and nervous system) and weight will be collected at Screening, Baseline (Day -1 to Day 1) and Day 2 prior to Discharge. Height will be measured and BMI will be calculated at screening only.

4.5.2. Pharmacokinetics Variables

Blood (plasma) for PK samples will be drawn for all subjects at the following timepoints relative to each dose:

- Dose 1: Within 10 minutes before starting the 1-hour infusion and at 30 minutes (±5 minutes), 1 hour (0 to 5 minutes from end of infusion), 1 hour 15 minutes (±5 minutes), 2 hours (±5 minutes), 5 hours (±15 minutes) from the start time of the first infusion of ZTI-01
- Dose 2: Within 10 minutes before starting the second 1-hour infusion.

  Note: This PK sample will be reported as the 8 hours post Dose 1 PK sample
- Dose 3: Within 10 minutes before starting the 1-hour infusion and at 30 minutes (±5 minutes), 1 hour (0 to 5 minutes from end of infusion), 1 hour 15 minutes (±5 minutes), 2 hours (±5 minutes), 5 hours (±15 minutes), 8 hour (±15 minutes), and 12 hours (±15 minutes) from the start time of the third (last) infusion of ZTI-01

A standardized bronchoscopy with BAL sampling will be performed at ±15 minutes of the single assigned timepoint based on each subject's schedule assignment: 30 minutes, 1 hour 15 minutes, 2, 5, or 8 hours after the start time of the third (last) infusion of ZTI-01.

5. SAMPLE SIZE CONSIDERATIONS

Since statistical analyses are not intended to serve as a basis of definitive conclusions, statistical power considerations for determination of sample size were not performed. The sample size for this study was selected as adequate for determining the safety and PK of ZTI-01. The sample size is based on clinical experience and judgment relative to study design and objectives.

6. GENERAL STATISTICAL CONSIDERATIONS

6.1. General Principles

All continuous variables will be summarized using the following descriptive statistics: N (non-missing sample size), mean, standard deviation (SD), median, maximum (max) and minimum (min). The frequency and percentages (based on the non-missing sample size) of observed levels will be reported for all categorical measures. When 95% confidence intervals (CIs) are given for a percent, exact (Clopper-Pearson) CIs will be used unless otherwise noted.

In general, all data will be listed. All summary tables will be annotated with the total population size relevant to that table, including any missing observations.

6.2. Timing of Analyses

The final analysis will be performed after database lock. There are no planned interim analyses

6.3. Analysis Populations

6.3.1. Safety Population

All subjects who receive any amount of ZTI-01 (start first infusion) will be included in the Safety Population

6.3.2. Pharmacokinetics Population

The pharmacokinetic analyses will be performed using the population of evaluable subjects. An evaluable subject is defined as a subject who receives all doses of ZTI-01, undergoes BAL at the randomized sampling timepoint with BAL return volume adequate for testing, and undergoes at least the one blood sampling timepoint that is concurrent with the BAL sampling timepoint, with blood sampling volume that is adequate for testing.

6.4. Covariates and Subgroups

The protocol does not define any formal subgroup analyses.

6.5. Missing Data

All attempts will be made to collect all data per protocol. No imputation will be performed for missing safety values and outliers will not be excluded from the safety analysis. The amount of missing data and reasons for missingness if available will be summarized in the body of the analysis report.

6.6. Interim Analyses and Data Monitoring

No interim analyses are planned. A safety summary report will be generated for review by the Safety Monitoring Committee (SMC) if a halting rule (see Protocol v6.0 Section 9.5) is met.

6.7. Multicenter Studies

This is a single site study.

6.8. Multiple Comparisons/Multiplicity

Safety and PK analyses will not be adjusted for multiple comparisons.

7. STUDY SUBJECTS

7.1. Disposition of Subjects

The disposition of subjects in the study will be tabulated for all subjects and by BAL sampling timepoint (Table 5). The table shows the total number of subjects screened, enrolled and randomized, enrolled as an alternate, received any study product, completed infusion of study product, completed all PK blood draws, completed bronchoscopy with BAL, completed all plasma urea samples, completed at least one PK blood draw concurrent with BAL timepoint, completed follow-up on Study Day 3 and the number of subjects determined to be PK evaluable.

A flowchart showing the disposition of study subjects will be included (Figure 2). This figure will present the number of subjects screened, enrolled, lost to follow-up, and analyzed. A listing of subjects who did not receive all three treatment infusions or terminated from study follow-up including the reason for treatment discontinuation or termination will be included in Listing 1.

The composition of the Safety and PK analysis populations, including reasons for subject exclusion, are presented in Table 6. Although there may be multiple reasons for exclusion, only one reason will be counted when summarizing reasons for exclusion from analysis populations in the table. The order that reasons will be considered are the same as the order shown in the table shell. Subjects who were excluded from any analysis population will be listed along with reason for exclusion (Listing 4).

Table 7 will present a summary of the reasons that subjects were screened but not enrolled summarized by inclusion and exclusion criteria.

7.2. Protocol Deviations

A summary of subject-specific protocol deviations will be presented by the reason for the deviation, the deviation category, and BAL Sampling Timepoint group for all subjects (Table 2). All deviations will be reviewed for possible subject exclusion from the PK population, including but not limited to incomplete infusion of study product, missing PK blood draws at PK timepoints, missing BAL at BAL sampling timepoint, and PK or BAL samples not processed per the MOP. All subject-specific protocol deviations and non-subject specific protocol deviations will be listed (Listing 2 and Listing 3, respectively).

8. EFFICACY EVALUATION

There are no efficacy endpoints for this protocol.

9. SAFETY EVALUATION

All safety analyses will be presented using the Safety Population.

When calculating the incidence of AEs and SAEs (i.e., on a per subject basis), each subject will be counted once and any repetitions within a subject will be ignored for events coded in the same category by the Medical Dictionary for Regulatory Activities (MedDRA®). The denominators for percent values will be indicated within the table or table header and denominators will consist of the maximal size of the Safety Population in the indicated observation period.

9.1. Demographic and Other Baseline Characteristics

Summaries of age, weight, height, BMI, sex, ethnicity, and race will be presented by BAL sampling timepoint and for all subjects (Table 8, Table 9). Ethnicity is categorized as Hispanic or Latino, not Hispanic or Latino, Not Reported, and Unknown. In accordance with NIH reporting policy, subjects may self-designate as belonging to more than one race or may refuse to identify a race, the latter reflected in the CRF as "No" to each racial option.

Individual subject listings will be presented for all demographics (Listing 5).

9.1.1. Prior and Concurrent Medical Conditions

All current illnesses and past pre-existing medical conditions will be coded using MedDRA dictionary version 20.1 or higher. Summaries of subjects' pre-existing medical conditions by MedDRA system organ class (SOC) and BAL sampling timepoints will be presented in Table 10. Individual subject listings will be presented for all medical conditions (Listing 6).

9.1.2. Prior and Concomitant Medications

Prior (30 days before administration of the first dose) and Concomitant medications will be coded to the Anatomical Therapeutic Classification (ATC) using the WHO Drug Dictionary. The use of prior or concomitant medications during the study will be summarized by ATC1 and ATC2 code and BAL sampling timepoint for the Safety Population (separately in Table 33 for prior medications and Table 34 for concomitant medications). Individual subject listings will be presented for all prior and concomitant medications (separately in Listing 18 for prior medications and Listing 19 for concomitant medications).

Drugs will be listed and summarized as prior medications when the end date for the medication is before the first dose of study product and listed and summarized as concomitant medications otherwise.

9.2. Measurements of Treatment Compliance

The infusion start and end dates and times for each dose of study product will be listed (Listing 7).

9.3. Adverse Events

TEAEs will be collected from the start of the first infusion on Day 1 through the Telephone Follow-Up Interview on Day 3. When calculating the incidence of AEs (i.e., on a per subject basis), each subject will only be counted once at the highest severity recorded and any repetitions of AEs within a subject will be ignored. Denominators for percentages are the number of subjects in the Safety Population.

9.3.1. Unsolicited Adverse Events

A brief overall summary of AEs will be shown in Table 11, including the number of subjects with at least 1 TEAE of any severity, the number of subjects with at least 1 related TEAE of each severity, and the number of subjects with at least 1 SAE.

The following summaries for unsolicited TEAEs will be presented by MedDRA system organ class (SOC) and Preferred Term:

- The total number of TEAEs and the number and percentage of subjects reporting at least one AE, regardless of severity or relationship to study product. 95% CI intervals will be presented for proportions (Table 12), following start of each infusions and for any time from the start of infusion for Dose 1 through the Telephone Follow-Up Interview on Day 3.
- The total number of TEAEs and the number and percentage of subjects reporting at least one AE by severity and relationship to study product (Table 13), for all AEs occurring from the start of infusion for Dose 1 through the Telephone Follow-Up Interview on Day 3.
- Bar chart displaying the incidence (number of subjects) of related TEAEs by severity and MedDRA SOC (Figure 3);
- Bar chart displaying the frequency (number of occurrences) of related TEAEs by severity and MedDRA SOC (Figure 4);

AEs by subject will be presented in Listing 8. A subject listing of non-serious AEs of moderate or greater severity will be reported (Table 15).

9.4. Deaths, Serious Adverse Events and other Significant Adverse Events

A listing of SAEs by subject will be presented in Table 14. The listing will include Subject ID, Adverse Event Description, Adverse Event Onset Date/End Date, Days Post Dose, Reason Reported as an SAE, Relationship to Treatment, Alternate Etiology if not Related, action taken with study treatment, whether subject discontinued due to AE, outcome, MedDRA SOC, and MedDRA Preferred Term.

9.5. Pregnancies

For any subjects in the Safety Population who became pregnant during the study, every attempt will be made to follow these subjects to completion of pregnancy to document the outcome, including information regarding any complications with pregnancy and/or delivery. If any pregnancies occur, listings of pregnancies and outcomes will be presented (Listing 20, Listing 21, Listing 22, Listing 23, and Listing 24).

9.6. Clinical Laboratory Evaluations

Chemistry, Hematology, Coagulation, and Urinalysis laboratory parameters will be collected at Screening, Baseline and Day 2 before discharge as described in Section 4.5.1. Sodium, potassium, magnesium, calcium, and phosphorus chemistry parameters will also be collected approximately 2 hours prior to the assigned bronchoscopy timepoint. The grading scale used for clinical laboratory evaluations is presented in Table 3.

Baseline will be defined as the most recent measurement prior to start of infusion of the first ZTI-01 dose. For parameters with multiple pre-dose measurements, screening values prior to the baseline measurement will be listed only.

The following parameters will be presented (in order):

- Chemistry: Sodium, Potassium, Magnesium, Calcium, Phosphorus, Albumin, Random Glucose, BUN, Total Protein, Creatinine, Triglycerides, Total Cholesterol, CPK, ALP, AST, ALT, Total Bilirubin, Direct Bilirubin, and LDH
- Hematology: Hemoglobin, Hematocrit, Platelet Count, WBC Count, Neutrophils, Lymphocytes, Monocytes, Eosinophils, Basophils
- Coagulation: PT and aPTT
- Urinalysis:
  - By dipstick: Leukocyte Esterase, Blood, pH, Specific Gravity, Protein, Glucose.
  - By microscopic evaluation: WBC in urine, Red Blood Cells (RBC) in urine, Bacteria, and Casts.

Laboratory results will be summarized by parameter and severity for baseline and all post-dose timepoints (including maximum severity post-baseline) and presented in Table 20, Table 22, Table 24, and Table 26. At baseline, measurements will be summarized according to the grading scale in Table 3. For post-dose measurements, if a value is outside the normal range but did not meet the definition for an AE due to the value being less than or equal to the measurement at baseline, it will be categorized as "Outside Normal Range." All other values will be summarized according to the grading scale in Table 3.

Laboratory parameters that have grading criteria for both decreases (result lower than normal range and baseline measurement) and increases (result higher than normal range and baseline measurement) will be summarized separately by direction of change. For example, sodium will be summarized separately as "Sodium, Decrease" and "Sodium, Increase." For the summary of the Urinalysis laboratory parameters, the denominator for WBC in urine and RBC in urine by microscopic evaluation will be the number of subjects with a urinalysis result (dipstick or microscopic evaluation) for the visit, not only the number of subjects with microscopic evaluation.

Abnormal laboratory results, Grade 1 severity or higher, will be listed in Table 16, Table 17, Table 18, and Table 19.

For continuous laboratory parameters, descriptive statistics including mean, SD, median, min and max values by timepoint and change from baseline all post-dose measures will be presented by laboratory parameter in Table 21, Table 23, Table 25, and Table 27.

Data will be visualized using box plots showing the change from baseline at each post dose timepoint.

- Chemistry Parameters: Beginning at Figure 5 and continuing through Figure 23
- Hematology Parameters: Beginning at Figure 24 and continuing through Figure 32
- Coagulation Parameters: Figure 33, Figure 34
- Urinalysis: Figure 35, Figure 36

Only laboratory parameters reported as continuous values will be included in tables of summary statistics and in box plots for change from baseline. Continuous parameters from the urine microscopic evaluation will not be visualized but will be summarized in the tables of summary statistics.

Listing 9, Listing 10, Listing 11, and Listing 12 will provide a complete listing of individual clinical laboratory results with applicable reference ranges.

Baseline serology screening results (Listing 13), urine toxicology and alcohol screening results (Listing 14) will be listed.

9.7. Vital Signs and Physical Evaluations

Vital sign measurements include systolic blood pressure, diastolic blood pressure, heart rate, respiratory rate, peripheral oxygen saturation, and temperature. Vital signs will be assessed at Screening and Baseline visits, immediately prior (within 10 minutes) to each of three doses of study drug, at approximately 0.5, 1, 2, and 5 hours after the start of infusion for each dose, and before discharge on Study Day 2.

The grading scale for vital sign evaluations is presented in Table 4. Summaries of vital signs by severity will be tabulated by timepoint and presented by parameter in Table 28. Vital signs that have grading criteria for both decreases (result lower than normal range and baseline measurement) and increases (result higher than normal range and baseline measurement) will be summarized separately by direction of change.

Descriptive statistics including mean, SD, median, min and max values by timepoint and change from baseline for each post dose timepoint will be presented by parameter in Table 29. The baseline measurement used for all post-dose timepoints will be the most recent measurement prior to start of infusion of the first ZTI-01 dose. Any pre-dose measurements from Screening or Baseline prior to the baseline value will be listed.

Data will be visualized using box plots showing the change from baseline at all post dose study timepoints with lines connecting the median, first quartile, and third quartile of the change from baseline at each timepoint (Figure 37, Figure 38, Figure 39, Figure 40, Figure 41 and Figure 42).

Vital signs results will be listed (Listing 15). Unplanned vital signs measurements will be included in the listing, but excluded from tabular and graphical summaries, except when calculating the maximum severity post-baseline.

The study protocol allows measurements of vital signs to be repeated in the case of an abnormal measurement. Repeat readings may be obtained up to twice more for a maximum of 3 measurements including the initial measurement. The following rules will be used to decide which measurement to include in summaries if more than one replicate was entered into the clinical database.

- If the first replicate is normal, then it will be used for analysis
- If the first replicate is abnormal, and the second replicate is also abnormal, then the first replicate will be used if has a severity equal to or greater to the second replicate, or the second replicate will be used if the second replicate has a higher severity than the first replicate.
- If the first replicate is abnormal, the second replicate is normal, and a third replicate measurement was not performed, then the first replicate will be used in the analysis.
- If the first replicate is abnormal, the second replicate is normal, and the third replicate is normal, then the second replicate will be used in the analysis.
- If the first replicate is abnormal, the second replicate is normal, and the third replicate is abnormal, then the first replicate will be used if has a severity equal to or greater to the third replicate, or the third replicate will be used if the third replicate has a higher severity than the first replicate.

Vital signs replicates not used in the analysis will not be listed.

A physical exam will be performed at Screening, Baseline (Day -1 to Day 1) and Day 2 (day of discharge) as described in Section 4.5, as well as the SAE follow-up as needed. Results of physical examinations, scheduled and unscheduled, will be presented in Listing 16.

9.8. ECG Assessments

An ECG will be performed at Screening, Baseline and within 1-2 hours of the last dose (3rd dose) of study drug and include the following parameters: QTc Interval, PR Interval, and QRS Duration. The grading scale for ECG evaluations is included in Table 4.

ECG change in overall interpretation from Baseline to Post Dose 3 will be shown in Table 30. Summaries of ECG results by severity for each parameter will be tabulated by timepoint and presented in Table 31.

Summary statistics of ECG results including mean, SD, median, min and max values by timepoint and change from baseline for the Post Dose 3 timepoint will be tabulated and presented by parameter for baseline and each post dose timepoint (Table 32). Data will be visualized using box plots showing the change from baseline at post Dose 3 (Figure 43, Figure 44, Figure 45).

Individual data listings of ECG results will be presented (Listing 17).

10. PHARMACOKINETICS

10.1. Analysis Population and Handling of Missing Timepoints

The pharmacokinetic analyses will be performed using the population of evaluable subjects as defined in Section 6.3.2.

Timepoints below the lower limit of quantification (LLOQ), referred to as below quantification limit (BQL), preceding the first PK concentration above the LLOQ will be imputed as 0. All other PK sample concentrations below the LLOQ will not be included in graphical displays or analyses.

All protocol deviations will be reviewed by the statistician. Major deviations such as receiving an incomplete dose, subject dosed but not meeting eligibility criteria, etc. may result in exclusion of a subject from PK analysis with approval from the sponsor. Statistical outlier concentrations will not be excluded from analysis unless associated with a protocol deviation that could plausibly explain the outlier.

Missing PK sample concentrations will not be imputed for the NCA.

10.2. Dosing and Pharmacokinetic Sampling Summary

Subject infusion start and end times (including interruptions) for all infusions received will be presented for each subject (Listing 7). If infusion of the drug is interrupted, the start and end times of the interruption will be recorded. Regardless of the amount of study product received, all scheduled blood draws will be attempted to be collected within a window based on the start of the administration. Special cases that potentially affect the analysis will be discussed in the final report. Actual infusion duration will be used for NCA analysis. If actual infusion duration time is missing, a duration of 1 h will be imputed.

Fosfomycin concentrations in plasma will be listed by subject and infusion dose number, with BQL and out of sample time window samples indicated (Listing 25). Fosfomycin concentrations in BAL and cell pellet samples concentrations will be listed by subject with out of sample time window samples indicated (Listing 27). The listings will also indicate the nominal and actual time associated with the sample.

Nominal time is defined as the time in hours since the start of infusion of either Dose 1 or Dose 3 as shown below:

Dose 1

- Timing of PK sample = “Pre-Dose for Dose 1”, Nominal Time = 0 h
- Timing of PK sample = “30 minutes after starting the first dose”, Nominal Time = 0.5 h
- Timing of PK sample = “1 hour after starting the first dose”, Nominal Time = 1 h
- Timing of PK sample = “1 hour 15 minutes after starting the first dose”, Nominal Time = 1.25 h
- Timing of PK sample = “2 hours after starting the first dose”, Nominal Time = 2 h
- Timing of PK sample = “5 hours after starting the first dose”, Nominal Time = 5 h
- Timing of PK sample = “Pre-Dose for Dose 2”, Nominal Time = 8 h

Dose 3

- Timing of PK sample = “Pre-Dose for Dose 3”, Nominal Time = 0 h
- Timing of PK sample = “30 minutes after starting the third dose”, Nominal Time = 0.5 h

- Timing of PK sample = "1 hour after starting the third dose", Nominal Time = 1 h
- Timing of PK sample = "1 hour 15 minutes after starting the third dose", Nominal Time = 1.25 h
- Timing of PK sample = "2 hours after starting the third dose", Nominal Time = 2 h
- Timing of PK sample = "5 hours after starting the third dose", Nominal Time = 5 h
- Timing of PK sample = "8 hours after starting the third dose", Nominal Time = 8 h
- Timing of PK sample = "12 hours after starting the third dose", Nominal Time = 12 h

Important protocol deviations related to dosing or PK sampling will be described. Possible bioanalytical errors and their effect on the PK analysis will be discussed.

10.3. Definition and Estimation of Individual PK Parameters

PK parameters will be estimated through a NCA using Phoenix® WinNonlin® version 8.0 or later (Pharsight Corporation, Cary, NC) using plasma concentrations collected after start of the first infusion. Actual post-dose time will be used for the estimation of PK parameters instead of nominal time.

Phoenix WinNonlin® NCA will use the following settings to compute parameters from serum and pulmonary PK data:

- Linear Up Log Down calculation method
- Uniform weighting
- IV Infusion
- Lambda Z Acceptance Criteria
  - Rsq_adjusted ≥ 0.90
  - Span ≥ 3.0 half-lives
  - Includes at least 3 timepoints after T_{max}

10.3.1. PK Parameters

C_{max}

Maximum concentration (C_{max}) for Dose 1 is defined as the maximum observed ZTI-01 concentration observed over all PK sample concentrations following Dose 1 and prior to Dose 2 and C_{max} for Dose 3 is defined as the maximum observed ZTI-01 concentration observed over all PK sample concentrations following Dose 3. It will be obtained from the **Cmax** parameter calculated by WinNonlin®. If there is no measurable concentration in the subject's PK profile, then C_{max} will be missing for that subject. C_{max} will be reported in units of µg/mL.

T_{max}

Time of maximum concentration (T_{max}) will be calculated separately for Dose 1 and for Dose 3. For Dose 1, T_{max} is defined as the time at which the maximum concentration occurs for PK sample concentrations following Dose 1 and prior to Dose 2. For Dose 3, T_{max} is defined as the time at which the maximum concentration occurs for PK sample concentrations following Dose 3. These will be obtained from the **Tmax** parameter calculated by WinNonlin®. If there is no measurable C_{max} in the subject's PK profile, then T_{max} will be missing for that subject. T_{max} will be reported in units of h.

λ_z

The elimination rate constant (λ_z) is defined as the first-order rate constant describing the rate of decrease of drug concentration in the terminal phase (defined as the region of the PK curve where absorption and distribution are no longer significant, and drug concentration follows first-order elimination kinetics). λ_z will be computed as the slope of a terminal region consisting of at least 3 successive points in the plot of log-transformed concentration data versus time. λ_z will be estimated using uniform weighting.

Time points used in the estimation of λ_z will be initially selected using the WinNonlin® automatic algorithm. Manually chosen time points may be used at the discretion of the PK analyst after examination of the automatically chosen points in the context of the semi-log profile. The set of points chosen will be that which maximizes the Pearson correlation (r) while satisfying the following. Computation of λ_z for a subject requires the use of at least three consecutive timepoints after T_{max} with evaluable concentrations above the LLOQ and for which the absolute value of the Pearson correlation (R^2) of log-transformed concentrations with time is greater than 0.8. If deemed necessary and appropriate, the observation collected at T_{max} may be used in the calculation of λ_z. The range of timepoints used to estimate λ_z must span at least 1.5 half-lives (definition below). Estimates of λ_z that use the timepoint at T_{max} will be clearly specified and justified in the analysis report. Drug concentrations used to calculate λ_z will be indicated in Listing 25. This parameter will be obtained from the **Lambda_z** parameter calculated by WinNonlin® and will be reported in units of /hours

t½

The apparent terminal elimination half-life (t½) is defined as the time required for the drug concentration to decrease by a factor of one-half in the terminal phase. The apparent terminal elimination half-life (t½) will be estimated as ln(2) / λ_z. It will be obtained from the **HL_Lambda_z** parameter calculated by WinNonlin®. Half-life will be reported in units of h.

AUC

$AUC_{(0-8)}$ for Dose 1 is defined as the area under the concentration-time curve from dosing for Dose 1 (Dose 1 Nominal Time=0 h) to 8 hrs post start of infusion, 12 hours post start of Dose 3 infusion, and the time of the last measured concentration, respectively.

$AUC_{(0-8)}$ and $AUC_{(0-12)}$ will be estimated using the Linear Up Log Down calculation method. $AUC_{(0-last)}$ will be estimated using the Linear Up Log Down calculation method and obtained from the **AUClast** parameter calculated by WinNonlin®.

$AUC_{(0-\infty)}$ for Dose 3 is defined as the total area under the concentration-time curve from start of infusion for Dose 3 (Nominal Time=0 h) taken to the limit as the end time becomes arbitrarily large. $AUC_{(0-\infty)}$ will be estimated by adding the Dose 3 $AUC_{(0-last)}$ to an extrapolated value equal to the last measured concentration greater than the LLOQ divided by λ_z:

$$AUC_{(0-\infty)} = AUC_{(0-last)} + \frac{C_{last}}{K_e},$$

where C_{last} is the last measured concentration ≥ LLOQ. $AUC_{(0-inf)}$ will be obtained from the **AUCINF_obs** parameter calculated by WinNonlin®. If the amount extrapolated portion of $AUC_{(0-\infty)}$ is >20%, the estimated $AUC_{(0-\infty)}$ value will be flagged when listed in the report (Listing 26) and will be excluded from statistical summaries of parameter estimates and downstream calculations.

All AUCs will be reported in units of h*µg/mL.

CL

Clearance (CL) is defined as the volume of plasma completely cleared of drug per unit time and is estimated in trials of an IV-administered drug as the dose divided by the $AUC_{(0-\infty)}$. It will be obtained from the **Cl_obs** parameter calculated by WinNonlin®. If the amount extrapolated portion of $AUC_{(0-\infty)}$ is >20%, the estimated CL value will be flagged when listed in the report and will be excluded from statistical summaries of parameter estimates and downstream calculations. CL will be reported in units of L/h.

V_d

Volume of distribution (V_d) will be estimated as the apparent volume of distribution at steady state (V_{ss}) in trials of an IV-administered drug. This value will be calculated using the AUC and area under the first moment curve (AUMC). It will be obtained from the **Vss_obs** parameter calculated by WinNonlin®. If the amount extrapolated portion of $AUC_{(0\text{-inf})}$ is >20%, the estimated V_{ss} value will be flagged when listed in the report and will be excluded from statistical summaries of parameter estimates and downstream calculations. V_d at steady state will be reported in units of L.

AR

The observed accumulation ratio (AR) is defined as the ratio of exposure parameters following multiple dosing. AR will be calculated using AUC_{0-8} and C_{max} for Dose 1 and Dose 3:

$$AR = \frac{AUC_{0\text{-}8,\ Dose\ 3}}{AUC_{0\text{-}8,\ Dose\ 1}}$$

$$AR = \frac{C_{max,\ Dose\ 3}}{C_{max,\ Dose\ 1}}$$

When λ_Z is estimable, AR will also be estimated using the elimination rate λ_z from Dose 3:

$$AR = \frac{1}{1-e^{-\lambda_Z \tau}}$$

where τ is the length of the dosing interval, 8 h.

Shift in T_{max}

Shift in T_{max} between Dose 1 and Dose 3 will be calculated by the subtraction of T_{max} for Dose 1 from T_{max} for Dose 3. Point estimates of the shift and associated 95% CIs derived from the inversion of the Sign Test will be reported. The 95% CIs will be calculated using the CIPCTLDF option in PROC Univariate.

10.3.2. Noncompartmental Analysis Population AUC and Percent Penetration Estimation

The median concentrations of fosfomycin from the BAL sampling times will be used to estimate the population average AUC_{0-8} of plasma, ELF, and AM will be estimated using the Linear Up Log Down calculation method.

The ratios of population average AUC_{0-8} of ELF-to-plasma and AM-to-plasma will be calculated to determine estimates of the percent penetration by dividing the AUC_{0-8} for ELF or AM by the geometric mean Plasma AUC_{0-8}.

10.4. Descriptive Statistics

Summary statistics for all continuous PK measures and parameters will include mean, SD, min, max, median, coefficient of variation as a percent (CV%), GM, and geometric SD (GSD), unless otherwise specified. CV% will not be calculated for T_{max} and T_{last} since those variables are not log-normal.

The CV% will be calculated using the method for log-normally distributed data.:

CV% = $\sqrt{\exp(\sigma^2) - 1} * 100\%$, where σ^2 is the variance of the log-transformed data [5].

10.4.1. Pharmacokinetic Parameter Summaries for Plasma

Plasma fosfomycin concentrations and summary statistics will be listed and summarized by nominal timepoint for Dose 1 (Table 35) and for Dose 3 (Table 36) and presented graphically:

- Figure 46 will plot all plasma PK profiles plots for Dose 1 and Dose 3 as linear plots and Figure 47 will plot all subject plasma PK profiles plots for Dose 1 and Dose 3 as semilogarithmic plots
- Linear plots of plasma mean concentration curves will be shown in Figure 48 for Dose 1 and Dose 3, with error bars representing +/- 1 standard SD.
- Semi-logarithmic plots of GM plasma concentration curves will be shown in Figure 49 Dose 1 and Dose 3.

PK Estimates for Dose 1 will include C_{max}, T_{max}, T_{last}, $AUC_{(0-8)}$ and will be presented in Table 37. PK Estimates for Dose 3 will include C_{max}, T_{max}, T_{last}, $AUC_{(0-8)}$, $AUC_{(0-last)}$ and $AUC_{(0-inf)}$, λ_z,$t_{1/2}$, CL, and V_d and will be presented in Table 38.

Shift in T_{max} will be summarized for Dose 3 compared to Dose 1 in Table 39 . AR for C_{max} and $AUC_{(0-8)}$ will be summarized for Dose 3 compared to Dose 1 in Table 40.

10.4.2. Pharmacokinetic Parameter Summaries for ELF and AM

ELF fosfomycin concentrations will be calculated as described in Section 3.3. Serum and BAL urea levels, volume of fosfomycin concentrations in BAL, the derived Fosfomycin concentrations in ELF, and fosfomycin concentrations in AM will be presented by BAL sampling timepoint and subject in Listing 27.

Statistical summaries by BAL sampling point for ELF fosfomycin concentrations and the concentration ratio of ELF to plasma fosfomycin concentrations will be presented in Table 41. Statistical summaries by BAL sampling point for AM fosfomycin concentrations and the concentration ratio of AM to Plasma fosfomycin concentrations will be presented in Table 42 respectively.

10.4.3. Analysis of Noncompartmental Analysis Parameters and Percent Penetration

The NCA population average AUC_{0-8} of plasma, ELF, and AM using the median concentrations of fosfomycin from the BAL sampling times will be presented in Table 43 along with the calculated percent penetration for ELF to plasma and AM to plasma.

Data will be presented graphically in the following figures:

- Box plot showing BAL sampling timepoint on the x-axis and the distribution of subject fosfomycin concentrations in ELF, AM, and plasma at matching BAL timepoint on the y-axis (Figure 50).
- Linear plots showing the population median fosfomycin concentrations by compartment (plasma, ELF, and AM) across each of the BAL sampling timepoints (Figure 51).

- Semi-logarithmic plots showing the population median fosfomycin concentrations by compartment (plasma, ELF, and AM) across each of the BAL sampling timepoints (Figure 52).

10.5. Exploratory Population PK Modeling

In addition to analysis of penetration of ZTI-01 into the lung using NCA, an exploratory analysis of penetration using the population PK approach will be performed as described in [6], where observed concentrations in both the central and peripheral compartment will be used simultaneously to fit a 2- or 3-compartment model. Between subject variability (BSV) for all PK parameters will be included using an exponential model, where the random errors have a N(0, ω^2) distribution.

If fitting both concentration types simultaneously does not produce an adequate model, then plasma PK and lung PK will be modeled sequentially as described in [7]. In this approach, plasma concentrations will first be modeled using a 2-compartment or 3-compartment model and ELF or AM concentrations will be subsequently modeled using approaches akin to treating ELF or AM concentrations as a response in a PK/PD analysis. Specifically, the following models would be considered for ELF or AM concentrations (referred to here generally as C_{lung}).

$$1)\ \text{Direct Response: } C_{lung} = \beta C_p$$

$$2)\ \text{Time} - \text{varying proportion: } C_{lung} = \beta_j C_p, j = 1,2,3,4,5$$

$$3)\ \text{Effect Site Equilibration: } \frac{dC_{lung}}{dt} = -Q \times C_{lung} + Q \times C_p$$

Model selection will be based on diagnostic plots and AIC. Other models such as use of an E_{max} link function to describe a nonlinear relationship between C_{lung} and C_p may be considered if none of the above models are adequate. BSV for all PK parameters will be included using an exponential model. The final model will include estimation of all off-diagonal estimates of the BSV covariance matrix, if feasible. Residual variability (RV) will be included using a combined multiplicative and additive model. The FOCE-ELS calculation method in Phoenix v8.0 or higher NLME will be used for model fitting. Model performance will be evaluated using visual predictive checks (VPCs). Parameter estimates for the final model will be presented with estimates of BSV (CV) where relevant.

Median values and 95% prediction intervals for penetration will be obtained by simulating 1,000 8-hour plasma and ELF/AM profiles with BSV and without RV, computing individual AUC0-8 values using the predicted concentrations via NCA with the Linear Up Log Down calculation method, computing individual penetration using AUC ratios, and taking the 2.5th and 97.5th percentiles of the ratio as the bounds for the prediction intervals.

11. IMMUNOGENICITY

There are no immunogenicity endpoints for this protocol.

12. OTHER ANALYSES

There are no other analyses.

13. REPORTING CONVENTIONS

P-values ≥0.001 and ≤0.999 will be reported to 3 decimal places; p-values less than 0.001 will be reported as "<0.001" The mean, standard deviation, and other statistics will be reported to 1 decimal place greater than the original data. The minimum and maximum will use the same number of decimal places as the original data. Proportions will be presented as 2 decimal places; values greater than zero but <0.01 will be presented as "<0.01". Percentages will be reported to the nearest whole number; values greater than zero but < 1% will be presented as "<1"; values greater than 99% but less than 100% will be reported as >99%. Estimated parameters, not on the same scale as raw observations (e.g. regression coefficients) will be reported to 3 significant figures.

For PK, AUCs will be reported as whole numbers (or using 3 significant digits if less than 100). $t_{1/2}$, T_{max}, CL, and V_d (V_{ss}) values will be reported to one decimal place (or to 2 significant digits if less than 1). λ_z values will be reported to 3 significant digits. C_{max} will be reported with the same number of significant digits as the measurement.

Listings of individual subject data include a Subject ID column. The subject identifiers assigned by site staff are replaced throughout this report with the SDTM variable USUBJID to protect the confidentiality of those who volunteered to participate in this protocol. USUBJID has been created as a composite of the 3-letter EDC platform code followed by a numeric identifier assigned chronologically to enrolled subjects as well as screening failures across all sites and protocols in the EDC platform. Any data sharing activities will include the USUBJID and not the subject identifiers assigned at the site.

14. TECHNICAL DETAILS

SAS version 9.4 or above will be used to generate all tables, figures, and listings.

15. SUMMARY OF CHANGES IN THE CONDUCT OF THE STUDY OR PLANNED ANALYSES

If there are changes to the planned analysis prior to final data lock and after finalization of the SAP, they may be added to the SAP as an addendum. The SAP will not be amended after final data lock.

16. REFERENCES

1. Rennard, S. I., et al. "Estimation of volume of epithelial lining fluid recovered by lavage using urea as marker of dilution." Journal of applied physiology 60.2 (1986): 532-538.
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3. VanScoy BD, McCauley J, Bhavnani SM, Ellis-Grosse EJ, Amborse PG. Relationship between fosfomycin exposure and amplification of Escherichia coli subpopulations with reduced susceptibility in a hollow-fiber infection model. Antimicrob Agents Chemother 2016;60:5141–5.
4. Lepak AJ, Zhao M, Vanscoy B, Taylor DS, Ellis-Grosse E, Ambrose PG, and Andes DR. In vivo Pharmacokinetics and Pharmacodynamics of ZTI-01 (Fosfomycin for Injection) in the Neutropenic Murine Thigh Infection Model against E. coli, K. pneumoniae, and P. aeruginosa. Antimicrob. Agents Chemother. AAC.00476-17; Accepted manuscript posted online 10 April 2017
5. Canchola, J. A., et al. "Correct use of percent coefficient of variation (% CV) formula for log-transformed data." MOJ Proteomics Bioinform 6.4 (2017): 316-317.
6. Lodise, Thomas P., et al. "Telavancin penetration into human epithelial lining fluid determined by population pharmacokinetic modeling and Monte Carlo simulation." Antimicrobial agents and chemotherapy 52.7 (2008): 2300-2304.
7. Dimelow, Richard, et al. "Population pharmacokinetic modelling of ceftazidime and avibactam in the plasma and epithelial lining fluid of healthy volunteers." Drugs in R&D 18.3 (2018): 221-230.

17. LISTING OF TABLES, FIGURES, AND LISTINGS

Table, figure, and listing shells are presented in Appendices 1, 2, and 3.

APPENDICES

APPENDIX 1. TABLE MOCK-UPS

LIST OF TABLES

9.1 Overall Study Design and Plan Description

9.5.1 Pharmacokinetic and Safety Measurements Assessed and Flow Chart

Table 1: Schedule of Study Procedures

Activities	Screening Visit	Baseline Visit	Treatment Phase	Discharge Visit	Telephone Follow-Up	Early Termination	Unscheduled Visit
	Day -30 to -1	Day -1 to 1	Days 1 to 2	Day 2	Day 3		
Informed Consent	X						
Review Eligibility Criteria	X	X					
Randomization		X					
Demographics	X						
Admission to DEPRU		X					
Medical History	X	X[a]					
Counsel on Birth Control	X			**X**		**X**	
Counsel on non-prescription medications	X						
Prior/Concomitant Medication Review	X	X	X	X	X	X	X
Collect and Record AEs/SAEs[b]			X	X	X	X	X
Height / BMI	X					X	
Vital Signs[c]	X	X	X	X		X	X
Physical Exam (including weight)	X	X		X		X	X[d]
Alcohol and Tobacco History[e]	X						
ECG (12-lead)[f]	X	X	X				X
HIV Antibody, Hepatitis B Ag, Hepatitis C Antibody	X						
Hem-Coag-UA[g]	X		X	X		X	
Chemistry[g]	X	X	X	X		X	
Lipids, LDH[g]	X		X	X		X	
Pregnancy Tests[h]	X	X					
Urine & Breath Tox Screens[i]	X	X					
Place Two Peripheral IVs		X					
Study Drug Administration[j]			X				
Blood for Urea Assay[k]			X				
Blood PK Samples[l]			X				
Bronchoscopy and BAL[m]			X				
Discharge from DEPRU				X			

[a] Update medical history as appropriate since screening including non-pharmacologic treatments/procedures within 72 hours before enrollment in the eCRF
[b] Collect and record AEs and SAEs from the time the start of treatment (Day 1) through Day 3.

[c] Vital signs (heart rate, blood pressure, temperature, respiratory rate, and peripheral oxygen saturation), measured with subject in the supine or semi-recumbent position, will be obtained at the following timepoints: Screening, Baseline (Day -1), immediately prior (within 10 minutes) to each of three doses of Study Drug, at approximately 0.5, 1, 2, and 5 hours (each ± 10 minutes) as after the start of infusion for each dose, and before discharge from DEPRU. If vital signs are abnormal, repeated readings will be obtained up to twice more, 5 to 10 minutes apart. If they remain abnormal, unscheduled vital signs will be repeatedly obtained until they normalize (at investigator's discretion). Since changes in vital signs are expected during bronchoscopy, when a vital sign timepoint is scheduled during a subject's bronchoscopy, that timepoint will be measured immediately before the start of bronchoscopy.

[d] At unscheduled visits, weight may be performed as part of the physical exam but is not required

[e] Alcohol history within past 6 months, lifetime tobacco history

[f] ECGs will be performed in the supine or semi-recumbent position (after rest for at least 10 min) at 3 timepoints: at Screening, Day 1 (before first dose of Study Drug), and within 1-2 hours of the last dose (3rd dose) of study drug. In case of premature discontinuation of study drug, ECG is to be performed before discharge.

[g] At Screening, and before discharge (Day 2), obtain hematology labs (white blood cell count, hemoglobin, hematocrit, and platelet count, neutrophils, lymphocytes, monocytes, eosinophils, basophils); coagulation tests (aPTT and PT), chemistry labs (albumin, glucose, blood urea nitrogen (BUN), potassium, magnesium, calcium, sodium, phosphorus, total protein, creatinine, triglycerides, total cholesterol, CPK, phosphorus, AST, ALT, total bilirubin, direct bilirubin, ALP, LDH), and urinalysis (leukocyte esterase, blood, glucose, protein, pH, and specific gravity via dipstick with addition of urine microscopy if dipstick abnormal). At Baseline (Day -1 to Day 1) obtain albumin, glucose, BUN, potassium, magnesium, calcium, sodium, phosphorus, total protein, creatinine, CPK, AST, ALT, total bilirubin, and ALP. Approximately two hours prior to bronchoscopy (Day 2) obtain sodium, potassium, magnesium, calcium, and phosphorus.

[h] Serum pregnancy test for women of childbearing potential at screening, and urine pregnancy test for women of childbearing potential at Baseline (Day -1 to Day 1)

[i] Alcohol Breathalyzer test, Urine cotinine, and Urine drug screen at Screening and Baseline (Day -1 to Day 1/admission to DEPRU). Cotinine will be reported qualitatively as positive/negative. Urine drug screen will consist of: barbiturates, benzodiazepines, THC, cocaine, opiates, and amphetamine/methamphetamine.

[j] Subjects will receive a total of three doses of ZTI-01 (fosfomycin disodium) 6g, administered every 8 hours, as 1-hour intravenous infusion

[k] Blood sample to determine plasma urea concentration will be obtained just before or during the scheduled bronchoscopy

[l] PK blood samples will be collected at 15 timepoints: before (within 10 minutes) the first, second, and third doses of study drug, at 30 minutes (during infusion), 1 hour (within 5 minutes after the end of infusion), 1 hour 15 minutes, 2, and 5 hours from the start time of the first dose of ZTI-01, and at 30 minutes (during infusion), 1 hour (within 5 minutes after the end of infusion), 1 hour 15 minutes, 2, 5, 8, and 12 hours from the start time of the third (last) infusion of ZTI-01. Blood PK samples will be drawn for all subjects within the following windows: -1 to -10 minutes for scheduled samples at pre-dose; +/- 5 minutes for scheduled samples at 30 minutes, 1 hour 15 minutes, and 2 hours; +0-5 minutes after end of infusion for the scheduled sample at 1 hour; +/- 15 minutes for scheduled samples at 5, 8, and 12 hours. Actual times for dosing and sample will be recorded.

[m] Bronchoscopy with BAL at 30 minutes (during the 1-hr infusion), 1 hr 15 minutes, 2, 5, or 8 hours from T0 (defined as the start time of the last [third] infusion of ZTI-01). BAL PK samples will be obtained within +/- 15 minutes of scheduled timepoint. Actual times for dosing and sample will be recorded.

10.2 Protocol Deviations

Table 2: Distribution of Protocol Deviations by Category, Type, and BAL Sampling Timepoint Group

Category	Deviation Type	30 minute BAL (N=X)		75 minute BAL (N=X)		2 hour BAL (N=X)		5 hour BAL (N=X)		8 hour BAL (N=X)		All Subjects (N=X)	
		No. of Subj.	No. of Dev.	No. of Subj.	No. of Dev.	No. of Subj.	No. of Dev.	No. of Subj.	No. of Dev.	No. of Subj.	No. of Dev.	No. of Subj.	No. of Dev.
Eligibility/enrollment	Any type	x	x	x	x	x	x	x	x	x	x	x	x
	Did not meet inclusion criterion												
	Met exclusion criterion												
	ICF not signed prior to study procedures												
	Other												
Treatment administration schedule	Any type												
	Out of window visit												
	Missed visit/visit not conducted												
	Missed treatment administration												
	Delayed treatment administration												
	Other												
Follow-up visit schedule	Any type												
	Out of window visit												
	Missed visit/visit not conducted												
	Other												
Protocol procedure/assessment	Any type												
	Incorrect version of ICF signed												
	Blood not collected												
	Urine not collected												
	BAL not collected												
	Other specimen not collected												
	Specimen result not obtained												

Category	Deviation Type	30 minute BAL (N=X)		75 minute BAL (N=X)		2 hour BAL (N=X)		5 hour BAL (N=X)		8 hour BAL (N=X)		All Subjects (N=X)	
		No. of Subj.	No. of Dev.	No. of Subj.	No. of Dev.	No. of Subj.	No. of Dev.	No. of Subj.	No. of Dev.	No. of Subj.	No. of Dev.	No. of Subj.	No. of Dev.
	Required procedure not conducted												
	Required procedure done incorrectly												
	Study product temperature excursion												
	Specimen temperature excursion												
	Other												
Treatment administration	Any type												
	Required procedure done incorrectly												
	Study product temperature excursion												
	Other												

12.2.2 Displays of Adverse Events

12.4.1 Individual Laboratory Measurements and Abnormal Laboratory Values

Table 3: Laboratory Adverse Event Grading Scale

LABORATORY ADVERSE EVENTS			
BLOOD, SERUM, PLASMA	**Mild (Grade 1)**	**Moderate (Grade 2)**	**Severe (Grade 3)**
Sodium – Hyponatremia mEq/L	132 – <LLN	130 – <132	<130
Sodium – Hypernatremia mEq/L	>ULN – 148	>148 – 150	>150
Potassium – Hyperkalemia mEq/L	>ULN – 5.2	>5.2 - 5.4	>5.4
Potassium – Hypokalemia mEq/L	3.1-<LLN	3.0-<3.1	<3.0
Glucose – Hypoglycemia mg/dL	65 - 69	55 - <65	<55
Glucose – Hyperglycemia Fasting – mg/dL	>ULN - 120	>120 - 130	>130
Glucose – Hyperglycemia Random – mg/dL	141 - 159	>159 – 200	>200
Blood Urea Nitrogen mg/dL	21-26	>26 – 31	> 31
Creatinine – mg/dL	>ULN – 1.7	>1.7 – 2.0	>2.0
Calcium – hypocalcemia mg/dL	8.0 – <LLN	7.5 – <8.0	<7.5
Calcium – hypercalcemia mg/dL	>ULN – 11.0	>11.0 – 11.5	>11.5
Magnesium – hypomagnesemia mg/dL	1.3 – 1.7	1.1 – <1.3	<1.1
Phosphorous – hypophosphatemia mg/dL	2.1 – 2.2	1.9 – <2.1	<1.9
CPK – mg/dL	221-1000	>1000-1500	>1500
Albumin – Hypoalbuminemia g/dL	2.8 – 3.4	2.5 – <2.8	< 2.5
Total Protein – Hypoproteinemia g/dL	5.2 – <LLN	5.0 – <5.2	< 5.0
Alkaline phosphatase (ALP) – U/L	111 - 240	>240– 360	>360
AST U/L	42 – 105	>105-175	>175
ALT U/L (Female)	>ULN - 105	>105-175	>175
ALT U/L (Male)	>ULN – 105	>105-175	>175
Bilirubin (serum total) - mg/dL	1.6 - 2.5	>2.5 - 3.0	> 3.0
Direct bilirubin – mg/dL	0.7-2.0	>2.0-3.5	>3.5
Total cholesterol – mg/dL	301 – 400	>400-500	>500
Triglycerides – mg/dL	>500 – 750	>750 – 1000	>1000
Lactate dehydrogenase – U/L	>200-500	>500-800	>800
Hemoglobin (Female) - g/dL	11.0 – 11.9	9.5 – <11.0	< 9.5
Hemoglobin (Male) - g/dL	12.0 – 13.6	10.0 – <12.0	<10.0
WBC Increase - cell/mm3	9900 – 15,000	>15,000 – 20,000	> 20,000
WBC Decrease - cell/mm3	2,500 – 3100	1,500 – <2,500	< 1500
Lymphocytes Decrease - cell/mm3	500 – <600	400 - <550	< 400
Neutrophils Decrease - cell/mm3	1,500 - < 2000	1000 - <1500	< 1000

LABORATORY ADVERSE EVENTS			
Eosinophils - cell/mm3	> 700 - 750	>750 - 1500	> 1500
Platelets Decreased - cell/mm3	120,000 – < 149,000	100,000 – <120,000	<100,000
PT – seconds (prothrombin time)	> ULN – 14.4	>14.4 – 15.7	> 15.7
aPTT – seconds (activated partial thromboplastin time)	> ULN – 42.1	>42.1 – 50.0	> 50.0
URINE	**Mild (Grade 1)**	**Moderate (Grade 2)**	**Severe (Grade 3)**
Protein	1+	2+	>2+
Glucose	1+	2+	>2+
Blood (microscopic) - red blood cells per high power field (rbc/hpf)	5-10*	>10-50*	> 50 and/or gross blood*

Table 4: Clinical Adverse Event Definition and Grading Scale

Clinical Adverse Events				
VITAL SIGNS	**Mild (Grade 1)**	**Moderate (Grade 2)**	**Severe (Grade 3)**	
Fever (°C) *	38.0 – 38.4	38.5 – 38.9	≥39.0	* Oral temperature; no recent hot or cold beverages or smoking.
(°F) *	100.4 – 101.1	101.2 – 102.0	≥102.1	
Tachycardia - beats per minute	101 – 115 and >25% change from baseline	116 – 130 and >25% change from baseline	> 130 or ventricular dysrhythmias and >25% change from baseline	
Bradycardia - beats per minute	50 – 54 or 45 - 49 bpm if baseline <60 bpm - Subject is asymptomatic; no treatment required	45 – 49 or 40 -44 if baseline <60 bpm - Subject is asymptomatic; no treatment required; Site PI/sub-investigator may confirm with ECG or 12-lead Holter monitors to rule out arrhythmia or advanced heart block.	< 45 or <40 bpm if baseline <60 bpm - Subject is symptomatic and/or requires treatment; Site PI/sub-investigator should confirm with ECG or 12-lead Holter monitors to rule out arrhythmia or advanced heart block	
Hypertension (systolic)- mm Hg	141-150	151-160	> 160	Assuming supine position, 10 min at rest conditions, not sleeping subjects measurements on the same arm and several concordant results.
Hypertension (diastolic) - mm Hg	91-95	96-100	> 100	
Hypotension (systolic) - mm Hg	85-89	80-84	< 80	
Tachypnea – breaths per minute	23-25	26-30	>30	
CARDIOVASCULAR	**Grade 1**	**Grade 2**	**Grade 3**	
Hemorrhage, Blood Loss	Estimated blood loss ≤ 100 mL	Estimated blood loss > 100 mL, no transfusion required	Transfusion required	
QTc interval (prolonged)	QTc 450-480 msec	QTc 481-500 msec	QTc >500 msec	
PR Interval (prolonged)	PR 200-250 msec and >25% change from baseline	PR >250 msec and >25% change from baseline	AV block 2nd degree Type II or higher OR ventricular pause >3 sec	
QRS Interval (prolonged)	QRS 120-150 msec and >25% change from baseline	QRS 150-180 msec and >25% change from baseline	New bundle branch block	
RESPIRATORY	**Grade 1**	**Grade 2**	**Grade 3**	
Cough	Transient- no treatment	Persistent cough;	Interferes with daily activities	
Bronchospasm, Acute	Transient; no treatment; 71% - 80% FEV1 of peak flow	Requires treatment; normalizes with bronchodilator; FEV1	No normalization with bronchodilator; FEV1 <60% of peak flow	

Clinical Adverse Events				
		60% - 70% (of peak flow)		
Dyspnea	Does not interfere with usual and social activities	Interferes with usual and social activities, no treatment	Prevents daily and usual social activity or requires treatment	
GASTROINTESTINAL	**Grade 1**	**Grade 2**	**Grade 3**	
Nausea	No interference with activity	Some interference with activity	Prevents daily activities	
Vomiting	No interference with activity or 1 - 2 episodes/24 hours	Some interference with activity or > 2 episodes/24 hours	Prevents daily activity or requires IV hydration	
Diarrhea	2 - 3 loose or watery stools or < 400 gm/24 hours	4 - 5 loose or watery stools or 400 - 800 gm/24 hours	6 or more loose or watery stools or > 800gms/24 hours or requires IV hydration	
REACTOGENICITY				
LOCAL REACTIONS	**Grade 1**	**Grade 2**	**Grade 3**	
Pain	Does not interfere with activity	Repeated use of non-narcotic pain reliever > 24 hours or interferes with activity	Any use of narcotic pain reliever or prevents daily activity	
Tenderness	Discomfort only to touch	Discomfort with movement	Significant discomfort at rest	
Erythema/Redness **	2.5 - 5 cm	5.1 - 10 cm	> 10 cm	** In addition to grading the measured local reaction at the greatest single diameter, the measurement should be recorded as a continuous variable.
Induration/Swelling ***	2.5 - 5 cm and does not interfere with activity	5.1 - 10 cm or interferes with activity	> 10 cm or prevents daily activity	***Induration/Swelling should be evaluated and graded using the functional scale as well as the actual measurement
SYSTEMIC	**Grade 1**	**Grade 2**	**Grade 3**	
Allergic Reaction	pruritus without rash	localized urticaria	generalized urticaria; angioedema or anaphylaxis	
Headache	No interference with activity	Repeated use of non-narcotic pain reliever > 24 hours or some interference with activity	Significant; any use of narcotic pain reliever or prevents daily activity	
Fatigue	No interference with activity	Some interference with activity	Significant; prevents daily activity	
Myalgia	No interference with activity	Some interference with activity	Significant; prevents daily activity	

Clinical Adverse Events				
ALL OTHER CONDITIONS	**Grade 1**	**Grade 2**	**Grade 3**	
Illness or clinical adverse event (as defined according to applicable regulations)	No interference with activity	Some interference with activity not requiring medical intervention	Prevents daily activity and requires medical intervention	

Table 5: Subject Disposition by BAL Sampling Timepoint Group

Subject Disposition	30 minute BAL (N=X)		75 minute BAL (N=X)		2 hour BAL (N=X)		5 hour BAL (N=X)		8 hour BAL (N=X)		All Subjects (N=X)	
	n	%	n	%	n	%	n	%	n	%	n	%
Screened	--	--	--	--	--	--	--	--	--	--	x	--
Total Enrolled	x	100	x	100	x	100	x	100	x	100	x	100
Enrolled and Randomized	x	xx	x	xx	x	xx	x	xx	x	xx	x	xx
Enrolled as Replacement	x	xx	x	xx	x	xx	x	xx	x	xx	x	xx
Began Infusion of Dose 1												
Completed Dose 1												
Completed Dose 2												
Completed Dose 3												
Completed All Scheduled Treatments[a]												
Completed All PK Blood Draws												
Completed Bronchoscopy with BAL												
Completed All Plasma Urea Samples												
At Least One PK Blood Draw Concurrent with BAL Timepoint												
Completed Follow-up (Study Day 3)[a]												
Evaluable[b,c]												

Note: N= number of subjects enrolled
[a] Refer to Listing 1 for reasons subjects discontinued or terminated early.
[b] Refer to Listing 4 for reasons subjects are excluded from the Analysis populations.
[c] An evaluable subject is defined as a subject who receives all doses of ZTI-01, undergoes BAL at the randomized sampling timepoint with BAL return volume adequate for testing, and undergoes at least the one blood sampling timepoint that is concurrent with the assigned BAL sampling timepoint, with blood sampling volume that is adequate for testing. Refer to Listing 4 for reasons subjects are not considered evaluable.

Table 6: Analysis Populations by BAL Sampling Timepoint Group

Analysis Populations	Reason Subjects Excluded [a]	30 minute BAL (N=X)		75 minute BAL (N=X)		2 hour BAL (N=X)		5 hour BAL (N=X)		8 hour BAL (N=X)		All Subjects (N=X)	
		n	%	n	%	n	%	n	%	n	%	n	%
Safety Population	Any Reason	xx	x	xx	xx	x	xx	xx	x	xx	xx	x	xx
	Did not receive any amount of study product												
PK Population	Any Reason												
	Did not receive all 3 doses of study product												
	Did not undergo BAL at Sampling Timepoint												
	BAL return volume not adequate for testing												
	Missing PK sample from blood sampling timepoint concurrent with BAL sampling timepoint												
	Blood sampling volume PK sample at timepoint concurrent with BAL sampling timepoint inadequate for testing												

Note: N= number of subjects enrolled
[a] Refer to Listing 4 for reasons subjects are excluded from the Analysis populations.

Table 7: Ineligibility Summary of Screen Failures

Inclusion/Exclusion Category	Inclusion/Exclusion Criterion	n[a]	%[b]
Inclusion and Exclusion	Number of subjects failing any eligibility criterion	x	100
Inclusion	Any inclusion criterion	x	xx
	[inclusion criterion 1]	x	xx
	[inclusion criterion 2]	x	xx
	[inclusion criterion 3]	x	xx
Exclusion	Any exclusion criterion	x	xx
	[exclusion criterion 1]	x	xx
	[exclusion criterion 2]	x	xx
	[exclusion criterion 3]	x	xx

[a] More than one criterion may be marked per subject.
[b] Denominator for percentages is the total number of screen failures.

Table 8: Summary of Categorical Demographic and Baseline Characteristics by BAL Sampling Timepoint Group

Variable	Characteristic	30 minute BAL (N=X)		75 minute BAL (N=X)		2 hour BAL (N=X)		5 hour BAL (N=X)		8 hour BAL (N=X)		All Subjects (N=X)	
		n	%	n	%	n	%	n	%	n	%	n	%
Sex	Male	x	xx	x	xx	x	xx	x	xx	x	xx	x	xx
	Female												
Ethnicity	Not Hispanic or Latino	x	xx	x	xx	x	xx	x	xx	x	xx	x	xx
Race	Hispanic or Latino												
	Not Reported												
	Unknown												
	American Indian or Alaska Native	x	xx	x	xx	x	xx	x	xx	x	xx	x	xx
	Asian												
	Native Hawaiian or Other Pacific Islander												
	Black or African American												
	White												
	Multi-Racial												
	Unknown												

Note: N=Number of subjects in the Safety Population

Table 9: Summary of Continuous Demographic and Baseline Characteristics by BAL Sampling Timepoint Group

Variable	Statistic	30 minute BAL (N=X)	75 minute BAL (N=X)	2 hour BAL (N=X)	5 hour BAL (N=X)	8 hour BAL (N=X)	All Subjects (N=X)
Age	Mean	xx	xx	xx	xx	xx	xx
	Standard Deviation	xx	xx	xx	xx	xx	xx
	Median	x	x	x	x	x	x
	Minimum	x	x	x	x	x	x
	Maximum	x	x	x	x	x	x
Height	Mean						
	Standard Deviation						
	Median						
	Minimum						
	Maximum						
Weight	Mean						
	Standard Deviation						
	Median						
	Minimum						
	Maximum						
BMI	Mean						
	Standard Deviation						
	Median						
	Minimum						
	Maximum						
Note: N=Number of subjects in the Safety Population							

Table 10: Summary of Subjects with Pre-Existing Medical Conditions by MedDRA System Organ Class and BAL Sampling Timepoint Group

MedDRA System Organ Class	30 minute BAL (N=X)		75 minute BAL (N=X)		2 hour BAL (N=X)		5 hour BAL (N=X)		8 hour BAL (N=X)		All Subjects (N=X)	
	n	%	n	%	n	%	n	%	n	%	n	%
Any SOC	x	xx	x	xx	x	xx	x	xx	x	xx	x	xx
[SOC 1]												
[SOC 2]												

Note: N=Number of subjects in the Safety Population

14.3 Safety Data

Table 11: Overall Summary of Adverse Events

	All Subjects (N = x)	
Subjects[a] with	**n**	**%**
At least one TEAE	x	x
At least one related TEAE	x	x
Mild (Grade 1)	x	x
Moderate (Grade 2)	x	x
Severe (Grade 3)	x	x
At least one severe (Grade 3) unsolicited TEAE	x	x
Related	x	x
Unrelated	x	x
At least one SAE[b]	x	x
At least one related, SAE	x	x
At least one adverse event leading to early termination[c]	x	x

Note: N = Number of subjects in the Safety Population
[a] Subjects are counted once for each category regardless of the number of events.
[b] A listing of Serious Adverse Events is included in Table 14.
[c] As reported on the Adverse Event eCRF.

14.3.1.2 Unsolicited Adverse Events

Table 12: Summary of Unsolicited Adverse Events by MedDRA System Organ Class and Preferred Term, and Timepoint

MedDRA System Organ Class	MedDRA Preferred Term	Start of Infusion 1 – Pre-Dose 2 (N=X)				Start of Infusion 2 – Pre-Dose 3 (N=X)				Start of Infusion 3 – Follow-Up (N=X)				Any Time Post Start of Infusion (N=X)			
		n	%	95% CI	Events	n	%	95% CI	Events	n	%	95% CI	Events	n	%	95% CI	Events
Any SOC	Any PT	x	xx	xx, xx	x	x	xx	xx, xx	x	x	xx	xx, xx	x	x	xx	xx, xx	x
[SOC 1]	Any PT																
	[PT 1]																
	[PT 2]																
[SOC 2]	Any PT																
	[PT 1]																
	[PT 2]																

Notes: N = Number of subjects in the Safety Population who started the specified infusion. This table presents number and percentage of subjects and the number of events. For the number of subjects (n) and corresponding percent (%), a subject is only counted once per Preferred Term and timepoint.

Table 13: Unsolicited Adverse Events by MedDRA System Organ Class and Preferred Term, Maximum Severity and Relationship

MedDRA System Organ Class	Preferred Term	Severity	All Subjects (N = X)					
			Related			Not Related		
			n	%	Events	n	%	Events
Any SOC	Any PT	Any Severity	x	x	x	x	x	x
		Mild	x	x	x	x	x	x
		Moderate	x	x	x	x	x	x
		Severe	x	x	x	x	x	x
SOC 1	PT 1	Any Severity	x	x	x	x	x	x
		Mild	x	x	x	x	x	x
		Moderate	x	x	x	x	x	x
		Severe	x	x	x	x	x	x
	PT 2	Any Severity	x	x	x	x	x	x
		Mild	x	x	x	x	x	x
		Moderate	x	x	x	x	x	x
		Severe	x	x	x	x	x	x

Note: N = Number of subjects in the Safety Population.

14.3.2 Listing of Deaths, Other Serious and Significant Adverse Events

Table 14: Listing of Serious Adverse Events

Adverse Event	Associated with Dose No.	No. of Hours Post Associated Dose (Duration)	Study Day Event Became Serious	Reason Reported as an SAE	Severity	Relationship to Study Treatment	If Not Related, Alternative Etiology	Action Taken with Study Treatment	Subject Discontinued Due to AE	Outcome	MedDRA System Organ Class	MedDRA Preferred Term
BAL Sampling Timepoint: , Subject ID: , AE Number:												
Comments:												
BAL Sampling Timepoint: ,Subject ID: , AE Number:												
Comments:												

Table 15: Listing of Non-Serious, Unsolicited, Moderate or Severe Adverse Events

Adverse Event	Associated with Dose No.	No. of Hours Post Associated Dose (Duration)	Severity	Relationship to Study Treatment	If Not Related, Alternative Etiology	Action Taken with Study Treatment	Subject Discontinued Due to AE	Outcome	MedDRA System Organ Class	MedDRA Preferred Term
BAL Sampling Timepoint: , Subject ID: , AE Number:										
Comments:										
BAL Sampling Timepoint: , Subject ID: , AE Number:										
Comments:										

14.3.3 Narratives of Deaths, Other Serious and Significant Adverse Events

(not included in SAP, but this is a placeholder for the CSR)

14.3.4 Abnormal Laboratory Value Listings (by Subject)

Table 16: Listing of Abnormal Laboratory Results – Chemistry

[Sort order: BAL Sampling Timepoint, Subject ID, Planned Timepoint, Laboratory Parameter (follow order specified in Section 9.6)]

BAL Sampling Timepoint	Subject ID	Sex	Age (years)	Planned Timepoint	Actual Study Day	Laboratory Parameter (Units)	Result (Severity)	Relationship to Treatment	If Not Related, Alternate Etiology	Action Taken with Study Treatment	Subject Discontinued Due to Result?

Table 17: Listing of Abnormal Laboratory Results – Hematology

[Sort order: BAL Sampling Timepoint, Subject ID, Planned Timepoint, Laboratory Parameter (follow order specified in Section 9.6)]

BAL Sampling Timepoint	Subject ID	Sex	Age (years)	Planned Timepoint	Actual Study Day	Laboratory Parameter (Units)	Result (Severity)	Relationship to Treatment	If Not Related, Alternate Etiology	Action Taken with Study Treatment	Subject Discontinued Due to Result?

Table 18: Listing of Abnormal Laboratory Results – Coagulation

[Sort order: BAL Sampling Timepoint, Subject ID, Planned Timepoint, Laboratory Parameter (follow order specified in Section 9.6)]

BAL Sampling Timepoint	Subject ID	Sex	Age (years)	Planned Timepoint	Actual Study Day	Laboratory Parameter (Units)	Result (Severity)	Relationship to Treatment	If Not Related, Alternate Etiology	Action Taken with Study Treatment	Subject Discontinued Due to Result?

Table 19: Listing of Abnormal Laboratory Results – Urinalysis

[Sort order: BAL Sampling Timepoint, Subject ID, Planned Timepoint, Laboratory Parameter (follow order specified in Section 9.6)]

BAL Sampling Timepoint	Subject ID	Sex	Age (years)	Planned Timepoint	Actual Study Day	Laboratory Parameter (Units)	Result (Severity)	Relationship to Treatment	If Not Related, Alternate Etiology	Action Taken with Study Treatment	Subject Discontinued Due to Result?

14.3.5 Displays of Laboratory Results

14.3.5.1 Chemistry Results

Table 20: Chemistry Laboratory Results by Parameter, Severity, and Timepoint

[Implementation Note: If there are no subjects with an early termination visit, then that row will not be included in the table. If no values are missing for any parameter then that column will not be included in the table. If no values meet the criteria for the "Outside Normal Range" column then exclude column and corresponding footnote. Only Sodium, potassium, magnesium, calcium, and phosphorus are collected at the 2 hours prior to the assigned bronchoscopy timepoint, summaries for all other parameters will not include the "2 hours prior to the assigned bronchoscopy" row]

Chemistry Parameter Timepoint	N	None		Outside Normal Range [a]		Mild/ Grade 1		Moderate/ Grade 2		Severe/ Grade 3		Missing	
		n	%	n	%	n	%	n	%	n	%	n	%
Chemistry - Any Parameter													
Baseline	x	x	x	NA	NA	x	x	x	x	x	x	x	x
2 hours prior to the assigned bronchoscopy[b]	x	x	x	x	x	x	x	x	x	x	x	x	x
Prior to Discharge (Day 2)													
Early Termination Visit													
Max Severity Post Baseline													
Sodium - Increase													
Baseline	x	x	x	NA	NA	x	x	x	x	x	x	x	x
2 hours prior to the assigned bronchoscopy[b]	x	x	x	x	x	x	x	x	x	x	x	x	x
Prior to Discharge (Day 2)													
Early Termination Visit													
Max Severity Post Baseline													
Sodium - Decrease													
Baseline	x	x	x	NA	NA	x	x	x	x	x	x	x	x
2 hours prior to the assigned bronchoscopy[b]	x	x	x	x	x	x	x	x	x	x	x	x	x
…													

Notes: N = Number of subjects in the Safety Population with clinical safety labs results at the respective study timepoint. Baseline = The most recent measurement prior to start of infusion of the first ZTI-01 dose. The "Max Post Baseline" rows indicate the maximum severity experienced by each subject at any timepoint post baseline, including unscheduled assessments.

[a] Post-dose measurements that were outside the normal range but did not meet the definition for an AE due to the value being less than or equal to the measurement at baseline will be categorized as "Outside Normal Range"

[b] Only Sodium, potassium, magnesium, calcium, and phosphorus are collected at the 2 hours prior to the assigned bronchoscopy timepoint.

Table 21: Chemistry Laboratory Summary Statistics by Parameter and Timepoint

		Measurement				Change from Baseline			
Timepoint	**N**	**Mean**	**SD**	**Median**	**Min, Max**	**Mean**	**SD**	**Median**	**Min, Max**
Sodium (mEq/L)									
Baseline	x	x.x	x.x	x.x	x.x, x.x	-	-	-	-
2 hours prior to the assigned bronchoscopy	x	x.x	x.x	x.x	x.x, x.x	x.x	x.x	x.x	x.x, x.x
Prior to Discharge (Day 2)									
Potassium (mEq/L)									
Baseline	x	x.x	x.x	x.x	x.x, x.x	-	-	-	-
2 hours prior to the assigned bronchoscopy	x	x.x	x.x	x.x	x.x, x.x	x.x	x.x	x.x	x.x, x.x
Prior to Discharge (Day 2)									
Magnesium (mg/dL)									
Baseline	x	x.x	x.x	x.x	x.x, x.x	-	-	-	-
2 hours prior to the assigned bronchoscopy	x	x.x	x.x	x.x	x.x, x.x	x.x	x.x	x.x	x.x, x.x
Prior to Discharge (Day 2)									
Calcium (mg/dL)									
Baseline	x	x.x	x.x	x.x	x.x, x.x	-	-	-	-
2 hours prior to the assigned bronchoscopy	x	x.x	x.x	x.x	x.x, x.x	x.x	x.x	x.x	x.x, x.x
Prior to Discharge (Day 2)									
Phosphorus (mg/dL)									
Baseline	x	x.x	x.x	x.x	x.x, x.x	-	-	-	-
2 hours prior to the assigned bronchoscopy	x	x.x	x.x	x.x	x.x, x.x	x.x	x.x	x.x	x.x, x.x
Prior to Discharge (Day 2)									
Albumin (g/dL)									
Baseline	x	x.x	x.x	x.x	x.x, x.x	-	-	-	-
Prior to Discharge (Day 2)	x	x.x	x.x	x.x	x.x, x.x	x.x	x.x	x.x	x.x, x.x
Random Glucose (mg/dL)									
Baseline	x	x.x	x.x	x.x	x.x, x.x	-	-	-	-
…									

Notes: N = Number of subjects in the Safety Population with clinical safety labs results at the respective study timepoint. Baseline = The most recent measurement prior to start of infusion of the first ZTI-01 dose.

14.3.5.2 Hematology Results

Table 22: Hematology Laboratory Results by Parameter, Severity, and Timepoint

[Implementation Note: If there are no subjects with an early termination visit, then that row will not be included in the table. If no values are missing for any parameter then that column will not be included in the table. If no values meet the criteria for the "Outside Normal Range" column then exclude column and corresponding footnote.]

Hematology Parameter Timepoint	N	None		Outside Normal Range [a]		Mild/ Grade 1		Moderate/ Grade 2		Severe/ Grade 3		Missing	
		n	%	n	%	n	%	n	%	n	%	n	%
Hematology - Any Parameter													
Baseline	x	x	x	NA	NA	x	x	x	x	x	x	x	x
Prior to Discharge (Day 2)	x	x	x	x	x	x	x	x	x	x	x	x	x
Early Termination Visit													
Max Severity Post Baseline													
Hemoglobin - Increase													
Baseline	x	x	x	NA	NA	x	x	x	x	x	x	x	x
Prior to Discharge (Day 2)	x	x	x	x	x	x	x	x	x	x	x	x	x
Early Termination Visit													
Max Severity Post Baseline													
Hematocrit - Increase													
Baseline	x	x	x	NA	NA	x	x	x	x	x	x	x	x
…	x	x	x	x	x	x	x	x	x	x	x	x	x

Notes: N = Number of subjects in the Safety Population with clinical safety labs results at the respective study timepoint. Baseline = The most recent measurement prior to start of infusion of the first ZTI-01 dose. The "Max Post Baseline" rows indicate the maximum severity experienced by each subject at any timepoint post baseline, including unscheduled assessments. NA = Not applicable.

[a] Post-dose measurements that were outside the normal range but did not meet the definition for an AE due to the value being less than or equal to the measurement at baseline will be categorized as "Outside Normal Range"

Table 23: Hematology Laboratory Summary Statistics by Parameter and Timepoint

		Measurement				Change from Baseline			
Timepoint	**N**	**Mean**	**SD**	**Median**	**Min, Max**	**Mean**	**SD**	**Median**	**Min, Max**
Hemoglobin (g/dL)									
Baseline	x	x.x	x.x	x.x	x.x, x.x	-	-	-	-
Prior to Discharge (Day 2)	x	x.x	x.x	x.x	x.x, x.x	x.x	x.x	x.x	x.x, x.x
Hematocrit (%)									
Baseline	x	x.x	x.x	x.x	x.x, x.x	-	-	-	-
…									

Notes: N= Number of subjects in the Safety Population with clinical safety labs results at the respective study timepoint. Baseline = The most recent measurement prior to start of infusion of the first ZTI-01 dose.

14.3.5.3 Coagulation

Table 24: Coagulation Laboratory Results by Parameter, Severity, and Timepoint

[Implementation Note: If there are no subjects with an early termination visit, then that row will not be included in the table. If no values are missing for any parameter then that column will not be included in the table. If no values meet the criteria for the "Outside Normal Range" column then exclude column and corresponding footnote.]

Coagulation Parameter Timepoint	N	None		Outside Normal Range [a]		Mild/ Grade 1		Moderate/ Grade 2		Severe/ Grade 3		Missing	
		n	%	n	%	n	%	n	%	n	%	n	%
Coagulation - Any Parameter													
Baseline	x	x	x	NA	NA	x	x	x	x	x	x	x	x
Prior to Discharge (Day 2)	x	x	x	x	x	x	x	x	x	x	x	x	x
Early Termination Visit													
Max Severity Post Baseline													
PT - Increase													
Baseline	x	x	x	NA	NA	x	x	x	x	x	x	x	x
Prior to Discharge (Day 2)	x	x	x	x	x	x	x	x	x	x	x	x	x
Early Termination Visit													
Max Severity Post Baseline													
aPPT - Increase													
Baseline	x	x	x	NA	NA	x	x	x	x	x	x	x	x
Prior to Discharge (Day 2)	x	x	x	x	x	x	x	x	x	x	x	x	x
Early Termination Visit													
Max Severity Post Baseline													

Notes: N = Number of subjects in the Safety Population with clinical safety labs results at the respective study timepoint. Baseline = The most recent measurement prior to start of infusion of the first ZTI-01 dose. The "Max Post Baseline" rows indicate the maximum severity experienced by each subject at any timepoint post baseline, including unscheduled assessments.

[a] Post-dose measurements that were outside the normal range but did not meet the definition for an AE due to the value being less than or equal to the measurement at baseline will be categorized as "Outside Normal Range"

Table 25: Coagulation Laboratory Summary Statistics by Parameter and Timepoint

		Measurement				Change from Baseline			
Timepoint	**N**	**Mean**	**SD**	**Median**	**Min, Max**	**Mean**	**SD**	**Median**	**Min, Max**
PT									
Baseline	x	x.x	x.x	x.x	x.x, x.x	-	-	-	-
Prior to Discharge (Day 2)	x	x.x	x.x	x.x	x.x, x.x	x.x	x.x	x.x	x.x, x.x
aPTT									
Baseline	x	x.x	x.x	x.x	x.x, x.x	-	-	-	-
Prior to Discharge (Day 2)	x	x.x	x.x	x.x	x.x, x.x	x.x	x.x	x.x	x.x, x.x

Notes: N = Number of subjects in the Safety Population with clinical safety labs results at the respective study timepoint. Baseline = The most recent measurement prior to start of infusion of the first ZTI-01 dose.

14.3.5.3 Urinalysis Results

Table 26: Urinalysis Laboratory Results by Parameter, Maximum Severity, and Timepoint

[Implementation Note: If there are no subjects with an early termination visit, then that row will not be included in the table. If no values are missing for any parameter then that column will not be included in the table. If no values meet the criteria for the "Outside Normal Range" column then exclude column and corresponding footnote.]

Urinalysis Parameter Timepoint	N	None		Outside Normal Range [a]		Mild/ Grade 1		Moderate/ Grade 2		Severe/ Grade 3		Missing	
		n	%	n	%	n	%	n	%	n	%	n	%
Urinalysis - Any Parameter													
Baseline	x	x	x	NA	NA	x	x	x	x	x	x	x	x
Prior to Discharge (Day 2)	x	x	x	x	x	x	x	x	x	x	x	x	x
Early Termination Visit													
Max Severity Post Baseline													
Protein													
Baseline	x	x	x	NA	NA	x	x	x	x	x	x	x	x
Prior to Discharge (Day 2)	x	x	x	x	x	x	x	x	x	x	x	x	x
Max Severity Post Baseline													
Glucose													
Baseline	x	x	x	NA	NA	x	x	x	x	x	x	x	x
Prior to Discharge (Day 2)	x	x	x	x	x	x	x	x	x	x	x	x	x
Early Termination Visit													
Max Severity Post Baseline													

Notes: N = Number of subjects in the Safety Population with clinical safety labs results at the respective study timepoint. Baseline = The most recent measurement prior to start of infusion of the first ZTI-01 dose. The "Max Post Baseline" rows indicate the maximum severity experienced by each subject at any timepoint post baseline, including unscheduled assessments.

[a] Post-dose measurements that were outside the normal range but did not meet the definition for an AE due to the value being less than or equal to the measurement at baseline will be categorized as "Outside Normal Range"

Table 27: Urinalysis Laboratory Summary Statistics by Parameter and Timepoint

[Implementation Note: If there are no subjects with microscopic evaluation then the rows for WBC in Urine, RBC in Urine, and Casts will not be included in table]

		Measurement				Change from Baseline			
Timepoint	**N**	**Mean**	**SD**	**Median**	**Min, Max**	**Mean**	**SD**	**Median**	**Min, Max**
pH									
Baseline	x	x.x	x.x	x.x	x.x, x.x	-	-	-	-
Prior to Discharge (Day 2)	x	x.x	x.x	x.x	x.x, x.x	x.x	x.x	x.x	x.x, x.x
Specific Gravity									
Baseline	x	x.x	x.x	x.x	x.x, x.x	-	-	-	-
Prior to Discharge (Day 2)	x	x.x	x.x	x.x	x.x, x.x	x.x	x.x	x.x	x.x, x.x
WBC in Urine									
Baseline	x	x.x	x.x	x.x	x.x, x.x	-	-	-	-
Prior to Discharge (Day 2)	x	x.x	x.x	x.x	x.x, x.x	x.x	x.x	x.x	x.x, x.x
RBC in Urine									
Baseline	x	x.x	x.x	x.x	x.x, x.x	-	-	-	-
Prior to Discharge (Day 2)	x	x.x	x.x	x.x	x.x, x.x	x.x	x.x	x.x	x.x, x.x
Casts									
Baseline	x	x.x	x.x	x.x	x.x, x.x	-	-	-	-
Prior to Discharge (Day 2)	x	x.x	x.x	x.x	x.x, x.x	x.x	x.x	x.x	x.x, x.x

Notes: N = Number of subjects in the Safety Population with clinical safety labs results at the respective study timepoint. Baseline = The most recent measurement prior to start of infusion of the first ZTI-01 dose.

14.3.6 Displays of Vital Signs

Table 28: Vital Signs by Assessment, Maximum Severity, and Timepoint

[Implementation Note: If there are no subjects with an early termination visit, then that row will not be included in the table. If no values are missing for any parameter then that column will not be included in the table]

Timepoint	N	None		Mild/ Grade 1		Moderate/ Grade 2		Severe/ Grade 3		Missing	
		n	%	n	%	n	%	n	%	n	%
Systolic Blood Pressure, Decrease											
Baseline	x	x	xx	x	xx	x	xx	x	xx	x	xx
Dose 1, 30 min Post Infusion Start											
Dose 1, 1 h Post Infusion Start											
Dose 1, 2 h Post Infusion Start											
Dose 1, 5 h Post Infusion Start											
Dose 2, ≤10 min Pre-Infusion											
Dose 2, 30 min Post Infusion Start											
Dose 2, 1 h Post Infusion Start											
Dose 2, 2 h Post Infusion Start											
Dose 2, 5 h Post Infusion Start											
Dose 3, ≤10 min Pre-Infusion											
Dose 3, 30 min Post Infusion Start											
Dose 3, 1 h Post Infusion Start											
Dose 3, 2 h Post Infusion Start											
Dose 3, 5 h Post Infusion Start											
Prior to Discharge (Day 2)											
Early Termination Visit											
Max Severity Post Baseline											
Systolic Blood Pressure, Increase											
Baseline											
Dose 1, 30 min Post Infusion Start											
Dose 1, 1 h Post Infusion Start											
…											

Notes: N = number of subjects in the Safety Population. Baseline = The most recent measurement prior to start of infusion of the first ZTI-01 dose. The "Max Post Baseline" rows indicate the maximum severity experienced by each subject at any timepoint post baseline, including unscheduled assessments.

Table 29: Vital Signs Summary Statistics and Change from Baseline by Assessment and Timepoint

		Measurement				Change from Baseline			
Timepoint	**N**	**Mean**	**SD**	**Median**	**Min, Max**	**Mean**	**SD**	**Median**	**Min, Max**
Systolic Blood Pressure (mmHg)									
Baseline	x	x.x	x.x	x.x	x.x, x.x	-	-	-	-
Dose 1, 30 min Post Infusion Start	x	x.x	x.x	x.x	x.x, x.x	x.x	x.x	x.x	x.x, x.x
Dose 1, 1 h Post Infusion Start									
Dose 1, 2 h Post Infusion Start									
Dose 1, 5 h Post Infusion Start									
Dose 2, ≤10 min Pre-Infusion									
Dose 2, 30 min Post Infusion Start									
Dose 2, 1 h Post Infusion Start									
Dose 2, 2 h Post Infusion Start									
Dose 2, 5 h Post Infusion Start									
Dose 3, ≤10 min Pre-Infusion									
Dose 3, 30 min Post Infusion Start									
Dose 3, 1 h Post Infusion Start									
Dose 3, 2 h Post Infusion Start									
Dose 3, 5 h Post Infusion Start									
Prior to Discharge (Day 2)									
Diastolic Blood Pressure (mmHg)									
Baseline	x	x.x	x.x	x.x	x.x, x.x	-	-	-	-
Dose 1, 30 min Post Infusion Start	x	x.x	x.x	x.x	x.x, x.x	x.x	x.x	x.x	x.x, x.x
…									

Notes: N = Number of subjects in the Safety Population with vital signs at the respective study timepoint. Baseline = The most recent measurement prior to start of infusion of the first ZTI-01 dose.

Table 30: ECG Overall Interpretations, Post Dose 3 Compared to Baseline

[Implementation Note: If there are no subjects with an early termination visit, then that row will not be included in the table. If no values are missing for any parameter then that column will not be included in the table]

	ECG Interpretation at Baseline (N = X)			
ECG Post Dose 3	**Normal n (%)**	**Abnormal, NCS n (%)**	**Abnormal, CS n (%)**	**Missing n (%)**
Normal	x (x)	x (x)	x (x)	x (x)
Abnormal, NCS	x (x)			
Abnormal, CS	x (x)			
Clinically significant change from baseline	x (x)			
Missing	x (x)			

Notes: N = number of subjects in the Safety Population. Baseline = The most recent measurement prior to start of infusion of the first ZTI-01 dose. CS = clinically significant; NCS = not clinically significant.

Table 31: ECG Results by Parameter, Severity, Timepoint and Assessment

[Implementation Note: If there are no subjects with an early termination visit, then that row will not be included in the table. If no values are missing for any parameter then that column will not be included in the table]

Timepoint	N	None		Mild/ Grade 1		Moderate/ Grade 2		Severe/ Grade 3		Missing	
		n	%	n	%	n	%	n	%	n	%
QTc Interval											
Baseline	x	x	xx	x	xx	x	xx	x	xx	x	Xx
Post Dose 3											
Early Termination Visit											
Max Severity Post Baseline											
PR Interval											
Baseline	x	x	xx	x	xx	x	xx	x	xx	x	Xx
Post Dose 3											
Early Termination Visit											
Max Severity Post Baseline											
QRS Duration											
Baseline	x	x	xx	x	xx	x	xx	x	xx	x	Xx
Post Dose 3											
Early Termination Visit											
Max Severity Post Baseline											

Notes: N = number of subjects enrolled that received any study product and ECG results at the respective study timepoint. Baseline = The most recent measurement prior to start of infusion of the first ZTI-01 dose. The "Max Post Baseline" rows indicate the maximum severity experienced by each subject at any timepoint post baseline, including unscheduled assessments.

Table 32: ECG Results - Summary Statistics by Parameter and Timepoint

[Implementation Note: If there are no subjects with an early termination visit, then that row will not be included in the table.]

		Measurement				Change from Baseline			
Timepoint	**N**	**Mean**	**SD**	**Median**	**Min, Max**	**Mean**	**SD**	**Median**	**Min, Max**
QTc Interval									
Baseline	x	x.x	x.x	x.x	x.x, x.x	-	-	-	-
Post Dose 3	x	x.x	x.x	x.x	x.x, x.x	x.x	x.x	x.x	x.x, x.x
Early Termination Visit									
PR Interval									
Baseline	x	x.x	x.x	x.x	x.x, x.x	-	-	-	-
Post Dose 3	x	x.x	x.x	x.x	x.x, x.x	x.x	x.x	x.x	x.x, x.x
Early Termination Visit									
QRS Duration									
Baseline	x	x.x	x.x	x.x	x.x, x.x	-	-	-	-
Post Dose 3	x	x.x	x.x	x.x	x.x, x.x	x.x	x.x	x.x	x.x, x.x
Early Termination Visit									

Notes: N= Number of subjects in the Safety Population with ECG results at the respective study timepoint. Baseline = The most recent measurement prior to start of infusion of the first ZTI-01 dose.

14.4 Summary of Concomitant Medications

Table 33: Number and Percentage of Subjects with Prior Medications by WHO Drug Classification and BAL Sampling Timepoint

WHO Drug Code Level 1, Anatomic Group	WHO Drug Code Level 2, Therapeutic Subgroup	30 minute BAL (N=X)		75 minute BAL (N=X)		2 hour BAL (N=X)		5 hour BAL (N=X)		8 hour BAL (N=X)		All Subjects (N=X)	
		n	%	n	%	n	%	n	%	n	%	n	%
Any Level 1 Codes	Any Level 2 Codes	x	xx	x	xx	x	xx	x	xx	x	xx	x	xx
[ATC Level 1 - 1]	Any [ATC 1 – 1]												
	[ATC 2 - 1]												
	[ATC 2 - 2]												
	[ATC 2 - 3]												
[ATC Level 1 – 2]	[ATC 2 - 1]												
	[ATC 2 - 2]												
	[ATC 2 - 3]												

Notes: N = Number of subjects in the Safety Population. n=Number of subjects reporting taking at least one medication in the specific WHO Drug Class.

Table 34: Number and Percentage of Subjects with Concurrent Medications by WHO Drug Classification and BAL Sampling Timepoint

WHO Drug Code Level 1, Anatomic Group	WHO Drug Code Level 2, Therapeutic Subgroup	30 minute BAL (N=X)		75 minute BAL (N=X)		2 hour BAL (N=X)		5 hour BAL (N=X)		8 hour BAL (N=X)		All Subjects (N=X)	
		n	%	n	%	n	%	n	%	n	%	n	%
Any Level 1 Codes	Any Level 2 Codes	x	xx	x	xx	x	xx	x	xx	x	xx	x	xx
[ATC Level 1 - 1]	Any [ATC 1 – 1]												
	[ATC 2 - 1]												
	[ATC 2 - 2]												
	[ATC 2 - 3]												
[ATC Level 1 – 2]	[ATC 2 - 1]												
	[ATC 2 - 2]												
	[ATC 2 - 3]												

Notes: N = Number of subjects in the Safety Population. n=Number of subjects reporting taking at least one medication in the specific WHO Drug Class.

14.4 Pharmacokinetics

Table 35: Individual Concentrations and Summary Statistics by for Plasma Fosfomycin Concentration (µg/mL) by Nominal Time - Dose 1

	Plasma Fosfomycin Concentration (µg/mL) by Nominal Time[a] After Dose 1						
Subject ID	**0 h**	**0.5 h**	**1[b] h**	**1.25 h**	**2 h**	**5 h**	**8[c] h**
SST.00101	x	x	x	x	x	x	x
SST.00102	x	x	x	x	x	x	x
SST.00102	x	x	x	x	x	x	x
…	x	x	x	x	x	x	x
Statistics	x	x	x	x	x	x	x
N[d]	x	x	x	x	x	x	x
Mean	x.x	x	x	x	x	x	x
SD	x.x	x	x	x	x	x	x
Min	x.x	x	x	x	x	x	x
Median	x.x	x	x	x	x	x	x
Max	x.x	x	x	x	x	x	x
CV%[e]	x	x	x	x	x	x	x
GM	x	x	x	x	x	x	x
GSD	x	x	x	x	x	x	x

[a] Times are relative to time of start of infusion of Dose 1.
[b] 0-5 minutes from end of infusion for Dose 1.
[c] Pre-dose for Dose 2
[d] Number of data points used to compute the summary statistics. For calculation of summary statistics, BQL values were imputed as 0 if they did not come after a PK sample concentration that is above the LLOQ and treated as missing otherwise.
[e] CV% = $\sqrt{(\exp(\sigma^2)-1)} * 100\%$, where σ^2 is the variance of the log-transformed data

Table 36: Individual Concentrations and Summary Statistics by for Plasma Fosfomycin Concentration (µg/mL) by Nominal Time - Dose 3

Subject ID	Plasma Fosfomycin Concentration (µg/mL) by Nominal Time[a] After Dose 3							
	0 h	**0.5 h**	**1 [b] h**	**1.25 h**	**2 h**	**5 h**	**8 h**	**12 h**
SST.00101	x	x	x	x	x	x	x	x
SST.00102	x	x	x	x	x	x	x	x
SST.00102	x	x	x	x	x	x	x	x
…	x	x	x	x	x	x	x	x
Statistics	x	x	x	x	x	x	x	x
N[c]	x	x	x	x	x	x	x	x
Mean	x	x	x	x	x	x	x	x
SD	x	x	x	x	x	x	x	x
Min	x	x	x	x	x	x	x	x
Median	x	x	x	x	x	x	x	x
Max	x	x	x	x	x	x	x	x
CV%	x	x	x	x	x	x	x	x
GM	x	x	x	x	x	x	x	x
GSD	x	x	x	x	x	x	x	x

[a] Times are relative to time of dosing. Times are relative to time of start of infusion of Dose 3. Blood PK samples will be drawn within the following windows: -10 to -1 minute for scheduled samples at 0h; ±5 minutes for scheduled samples at 0.5h, 1.25h, and 2h; +0-5 minutes from end of infusion for the scheduled sample at 1h; and ±15 minutes for scheduled samples at 5h, 8h and 12h.
[b] 0-5 minutes from end of infusion for Dose 3.
[c] Number of data points used to compute the summary statistics. For calculation of summary statistics, BQL values were imputed as 0 if they did not come after a PK sample concentration that is above the LLOQ and treated as missing otherwise.
[d] CV% = $\sqrt{(\exp(\sigma^2)-1)} * 100\%$, where σ^2 is the variance of the log-transformed data.

Table 37: Summary Statistics for Plasma Fosfomycin PK Parameters, Dose 1

Statistic	C_{max} (µg/mL)	T_{max} (h)	T_{last} (h)	$AUC_{(0-8)}$ (h*µg/mL)
N	x	x	x	x
Mean	x	x	x	x
SD	x	x	x	x
Min	x	x	x	x
Median	x	x	x	x
Max	x	x	x	x
CV%	x	NA	NA	x
GM	x	x	x	x
GSD	x	x	x	x

Notes: N=Number of data points used to compute the summary statistics. NA = Not applicable.

Table 38: Summary Statistics for Plasma Fosfomycin PK Parameters, Dose 3

Statistic	C_{max} (µg/mL)	T_{max} (h)	T_{last} (h)	$AUC_{(0-8)}$ (h*µg/mL)	$AUC_{(0-12)}$ (h*µg/mL)	$AUC_{(0-inf)}$ (h*µg/mL)	λ_z (/h)	$t_{1/2}$ (h)	CL (L/h)	Vss (L)
N	x	x	x	x	x	x	x	x	x	x
Mean	x	x	x	x	x	x	x	x	x	x
SD	x	x	x	x	x	x	x	x	x	x
Min	x	x	x	x	x	x	x	x	x	x
Median	x	x	x	x	x	x	x	x	x	x
Max	x	x	x	x	x	x	x	x	x	x
CV%	x	NA	NA	x	x	x	x	x	x	x
GM	x	x	x	x	x	x	x	x	x	x
GSD	x	x	x	x	x	x	x	x	x	x

Notes: N = Number of data points used to compute the summary statistics. NA = Not applicable.

Table 39: Summary of Shift in T_{max}, Comparing Dose 1 and Dose 3

Median T_{max} (h)		Median Difference in T_{max} (h)	95% CI of Estimated Median Difference (h)[a]
Dose 1	Dose 3		
x.x	x.x	x.x	(x.x, x.x)
[a] 95% CI derived from inversion of the Sign Test			

Table 40: Summary Statistics for Accumulation Ratios Comparing Dose 1 and Dose 3

Statistic	C_{max} AR	$AUC_{(0-8)}$ AR
N	x	x
Mean	x	x
SD	x	x
Min	x	x
Median	x	x
Max	x	x
GM	x	x
GSD		

Note: N=Number of data points used to compute the summary statistics.

Table 41: Summary Statistics for ELF Fosfomycin Concentration (µg/mL) and ELF to Plasma Concentration Ratio by BAL Sampling Timepoint

Statistic	ELF Fosfomycin Concentration (µg/mL) at BAL Sampling Timepoint					Concentration Ratio ELF to Plasma at BAL Sampling Timepoint				
	0.5 h	1.25 h	2 h	5 h	8 h	0.5 h	1.25 h	2 h	5 h	8 h
N	x	x	x	x	x	x	x	x	x	x
Mean	x	x	x	x	x	x	x	x	x	x
SD	x	x	x	x	x	x	x	x	x	x
Min	x	x	x	x	x	x	x	x	x	x
Median	x	x	x	x	x	x	x	x	x	x
Max	x	x	x	x	x	x	x	x	x	x
CV%	x	x	x	x	x	x	x	x	x	x
GM	x	x	x	x	x	x	x	x	x	x
GSD	x	x	x	x	x	x	x	x	x	x

Notes: times are relative to time of start of infusion of Dose 3. N = Number of data points used to compute the summary statistics.

Table 42: Summary Statistics for AM Fosfomycin Concentration (µg/mL) and AM to Plasma Concentration Ratio by BAL Sampling Timepoint

	AM Fosfomycin Concentration (µg/mL) at BAL Sampling Timepoint					Concentration Ratio AM to Plasma at BAL Sampling Timepoint				
Statistic	**0.5 h**	**1.25 h**	**2 h**	**5 h**	**8 h**	**0.5 h**	**1.25 h**	**2 h**	**5 h**	**8 h**
N	x	x	x	x	x	x	x	x	x	x
Mean	x	x	x	x	x	x	x	x	x	x
SD	x	x	x	x	x	x	x	x	x	x
Min	x	x	x	x	x	x	x	x	x	x
Median	x	x	x	x	x	x	x	x	x	x
Max	x	x	x	x	x	x	x	x	x	x
CV%	x	x	x	x	x	x	x	x	x	x
GM	x	x	x	x	x	x	x	x	x	x
GSD	x	x	x	x	x	x	x	x	x	x

Notes: times are relative to time of start of infusion of Dose 3. N = Number of data points used to compute the summary statistics.

Table 43: Noncompartmental Analysis Population Average AUC_{0-8} of Fosfomycin in Plasma, ELF, and AM and Percent Penetration

	N	$AUC_{(0-8)}$[a] (h*µg/mL)	Plasma Percent Penetration
Plasma	x	x	NA
ELF	x	x	x
AM	x	x	x

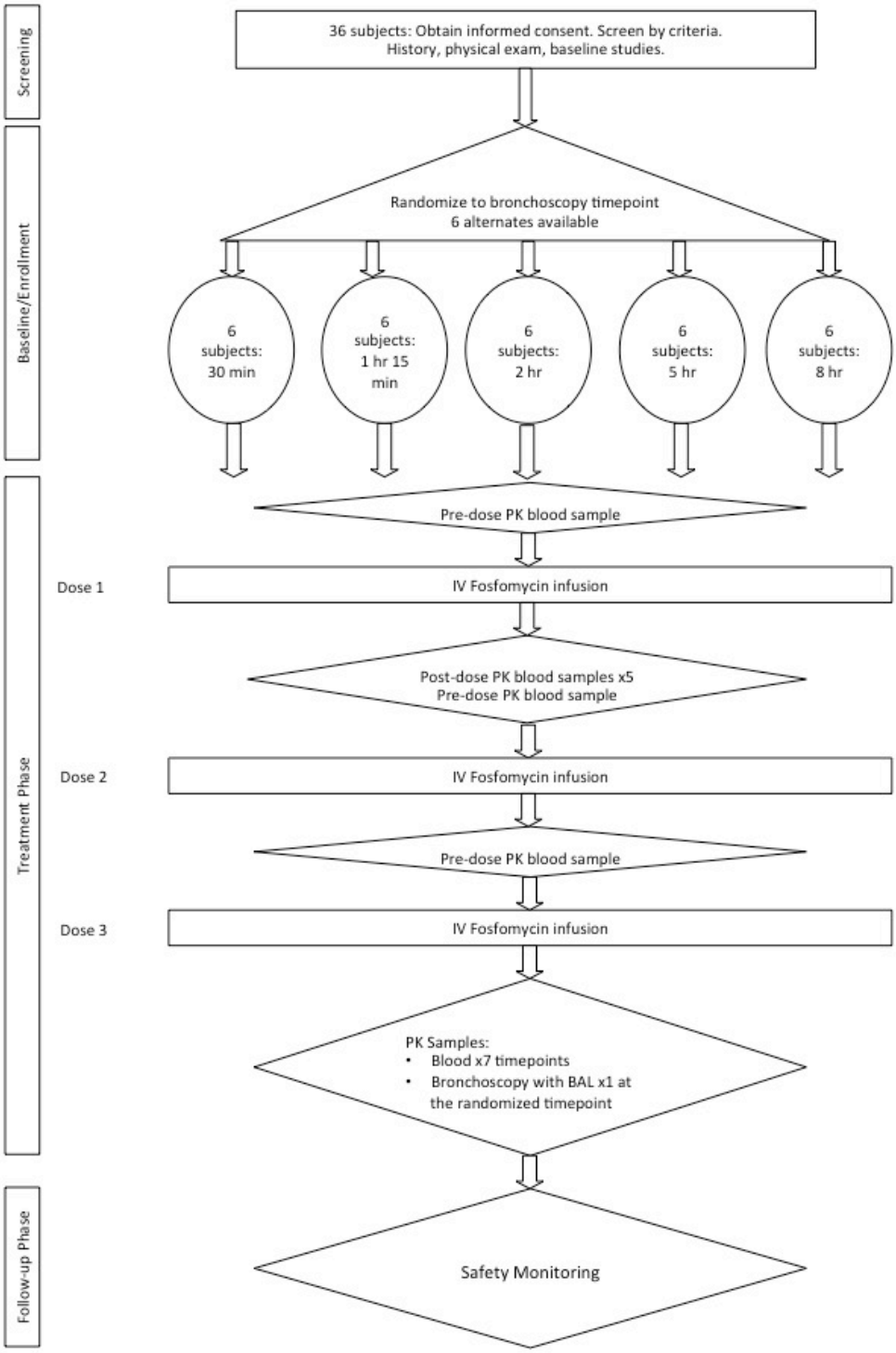
Note: N = number of data points used to compute used to derive $AUC_{(0-8)}$
[a] $AUC_{(0-8)}$ following Dose 3

APPENDIX 2. FIGURE MOCK-UPS

LIST OF FIGURES

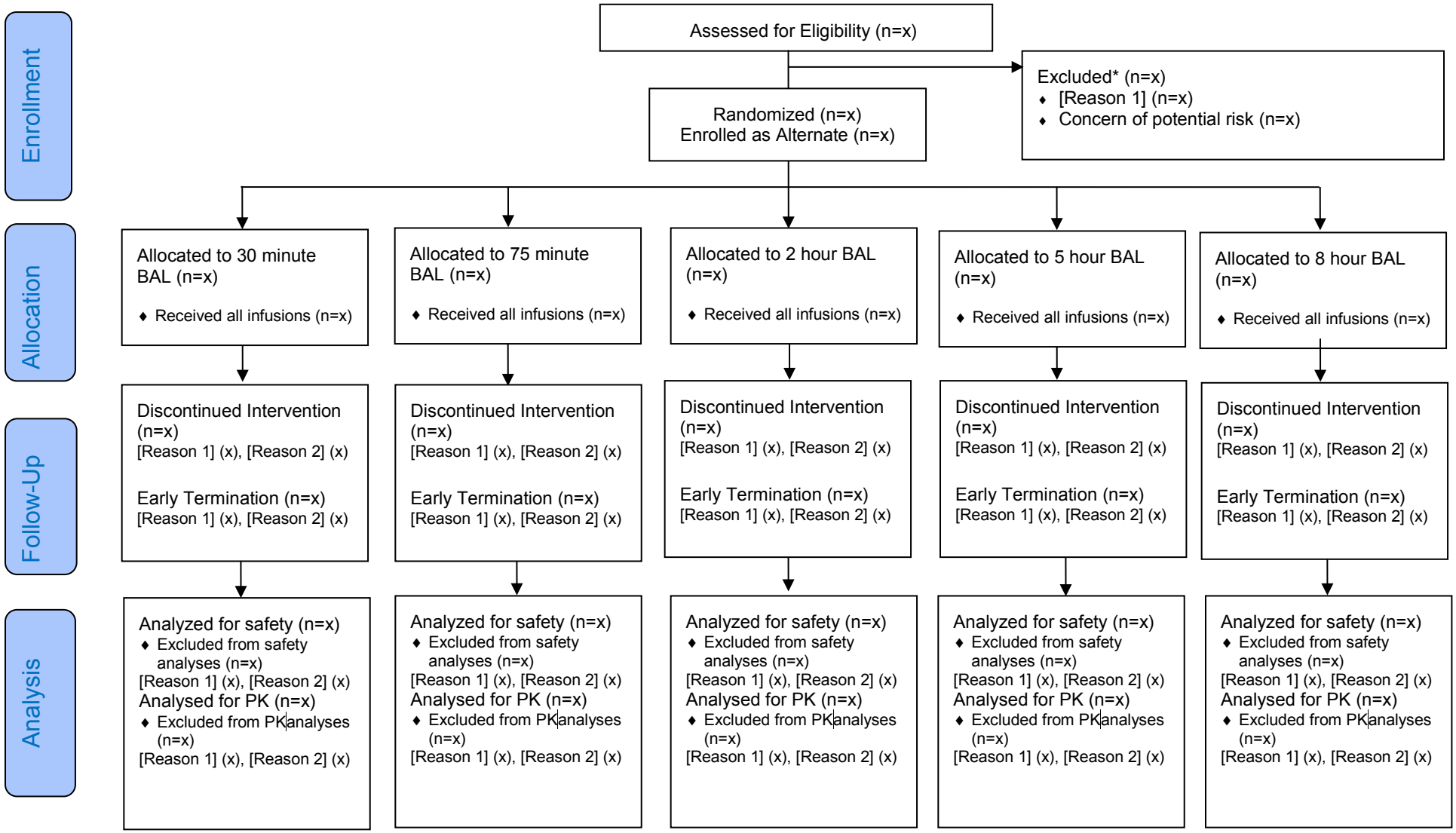
9.1 Overall Study Design and Plan Description

Figure 1: Study Design



10.1 Disposition of Subjects

Figure 2: CONSORT Flow Diagram



14.3.1.2 Unsolicited Adverse Events

Figure 3: Incidence of Related Adverse Events by MedDRA System Organ Class and Severity – All Subjects

[Implementation Note: Show one bar graph, include all subjects]

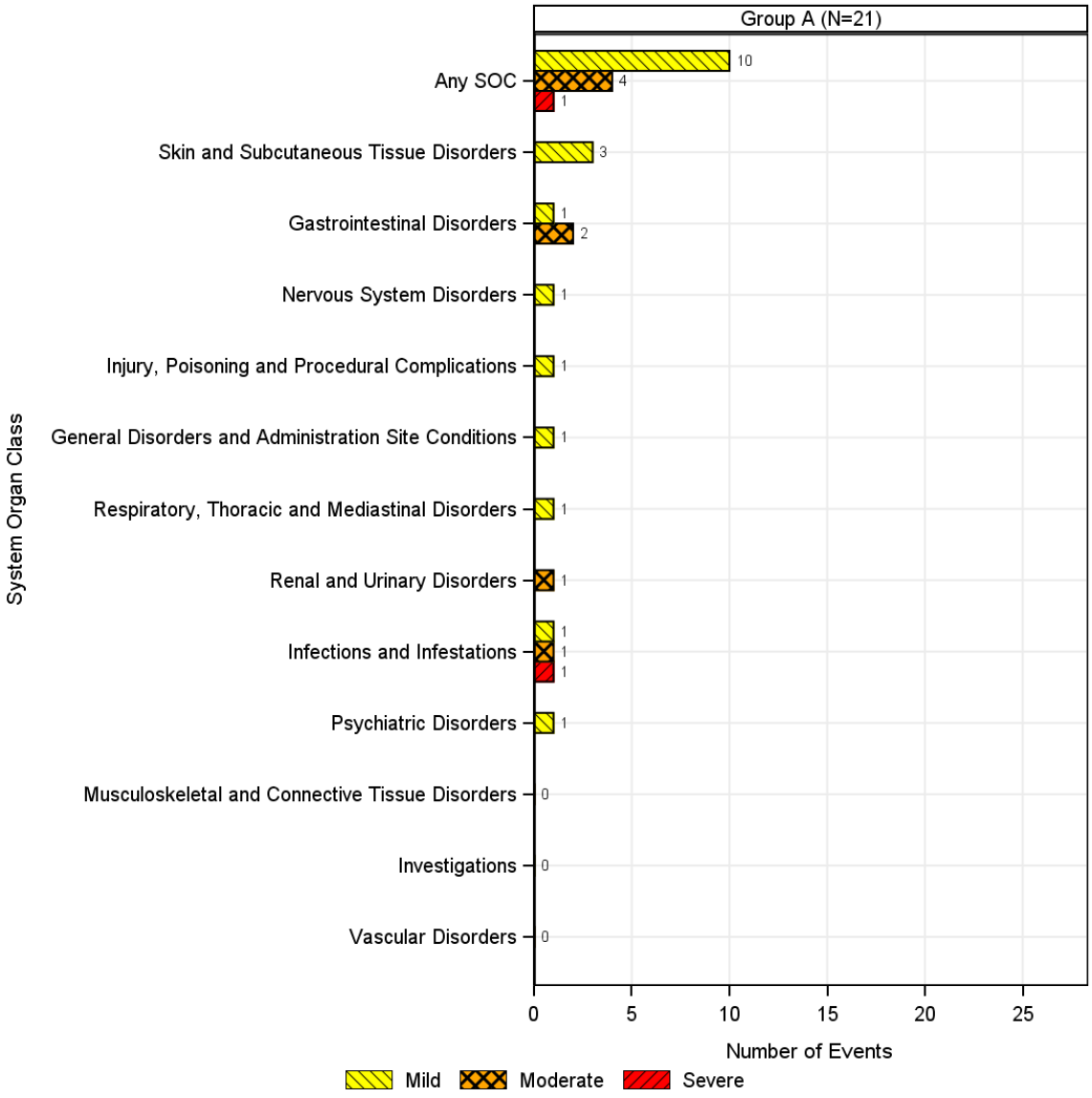


Figure 4: Frequency of Related Adverse Events by MedDRA System Organ Class and Maximum Severity – All Subjects

[Implementation Note: Show one bar graph, include all subjects]

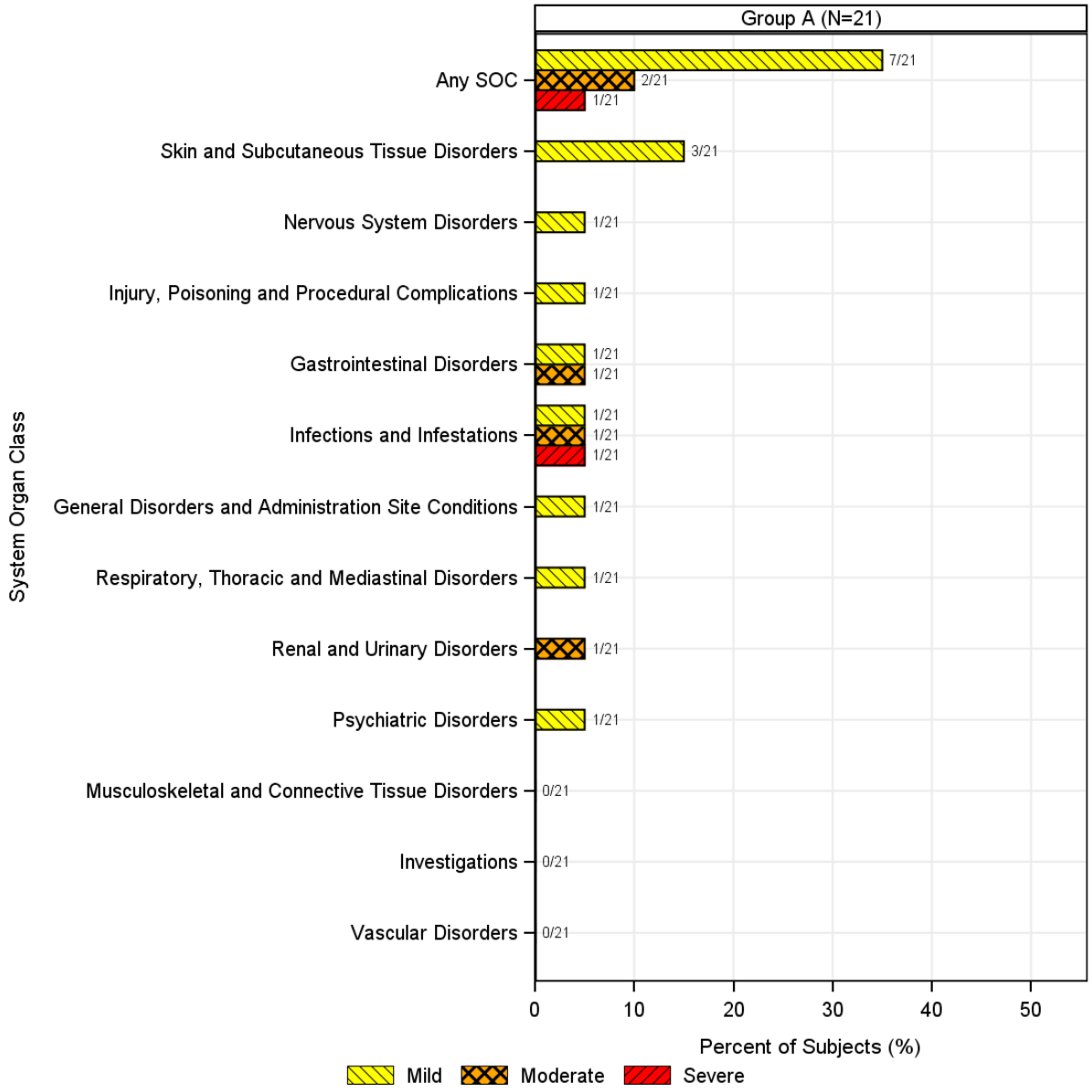
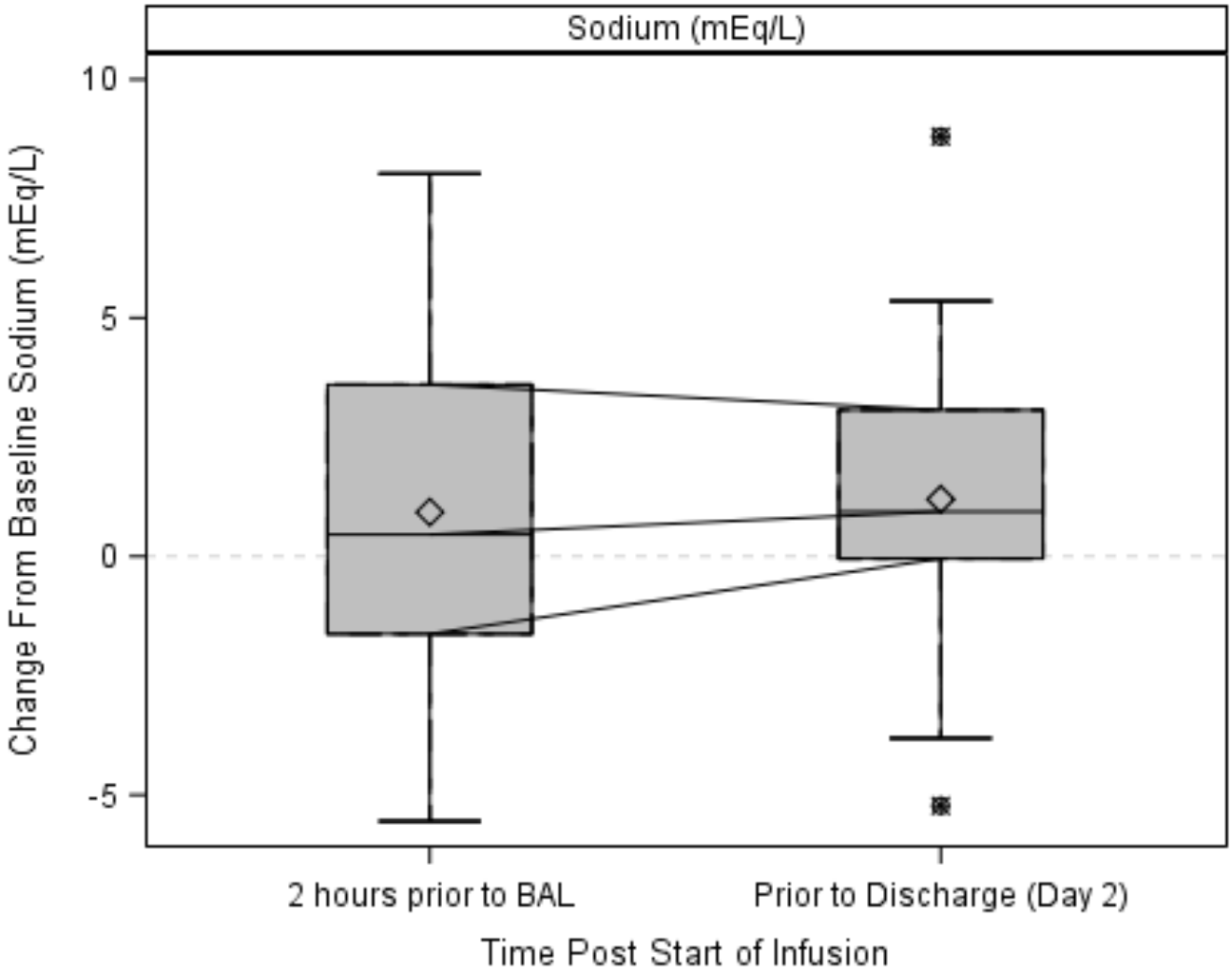


Figure 5: Chemistry Laboratory Results by Scheduled Timepoints: Box Plot of Change from Baseline by Laboratory Parameter – Sodium

[Implementation Note: For Laboratory Parameters with more than one timepoint, use lines to connect the 25th and 75th percentiles and median. Omit lines if Laboratory Parameter measured only one post dose timepoint]



Figures with Similar Format:

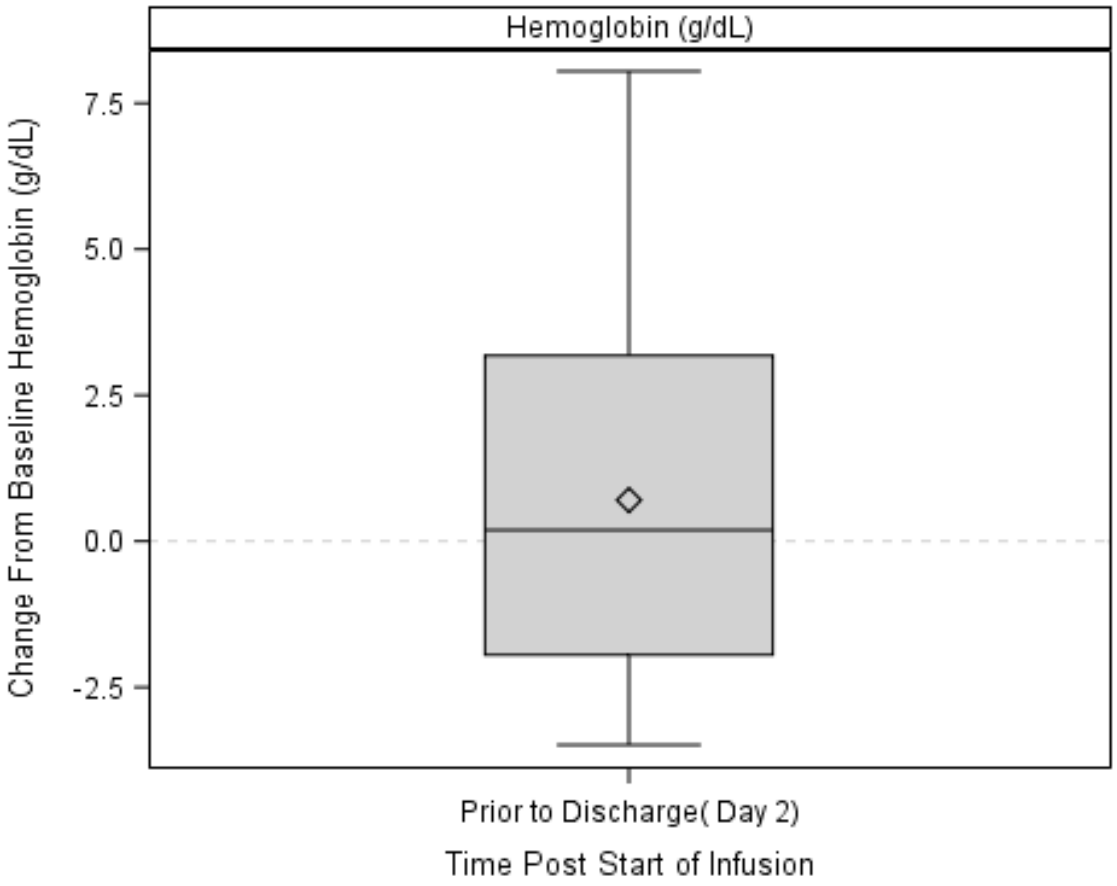
Figure 6: Chemistry Laboratory Results by Scheduled Timepoints: Box Plot of Change from Baseline by Laboratory Parameter – Potassium

Figure 7: Chemistry Laboratory Results by Scheduled Timepoints: Box Plot of Change from Baseline by Laboratory Parameter – Magnesium

Figure 8: Chemistry Laboratory Results by Scheduled Timepoints: Box Plot of Change from Baseline by Laboratory Parameter – Calcium

Figure 9: Chemistry Laboratory Results by Scheduled Timepoints: Box Plot of Change from Baseline by Laboratory Parameter – Phosphorus

Figure 24: Hematology Laboratory Results by Scheduled Timepoints: Box Plot of Change from Baseline by Laboratory Parameter – Hemoglobin



Figures with Similar Format:

Figure 25: Hematology Laboratory Results by Scheduled Timepoints: Box Plot of Change from Baseline by Laboratory Parameter – Hematocrit

Figure 26: Hematology Laboratory Results by Scheduled Timepoints: Box Plot of Change from Baseline by Laboratory Parameter – Platelet Count

Figure 27: Hematology Laboratory Results by Scheduled Timepoints: Box Plot of Change from Baseline by Laboratory Parameter – WBC Count

Figure 28: Hematology Laboratory Results by Scheduled Timepoints: Box Plot of Change from Baseline by Laboratory Parameter – Neutrophils

Figure 29: Hematology Laboratory Results by Scheduled Timepoints: Box Plot of Change from Baseline by Laboratory Parameter – Lymphocytes

Figure 30: Hematology Laboratory Results by Scheduled Timepoints: Box Plot of Change from Baseline by Laboratory Parameter – Monocytes

Figure 33: Coagulation Laboratory Results by Scheduled Timepoints: Box Plot of Change from Baseline by Laboratory Parameter – PT

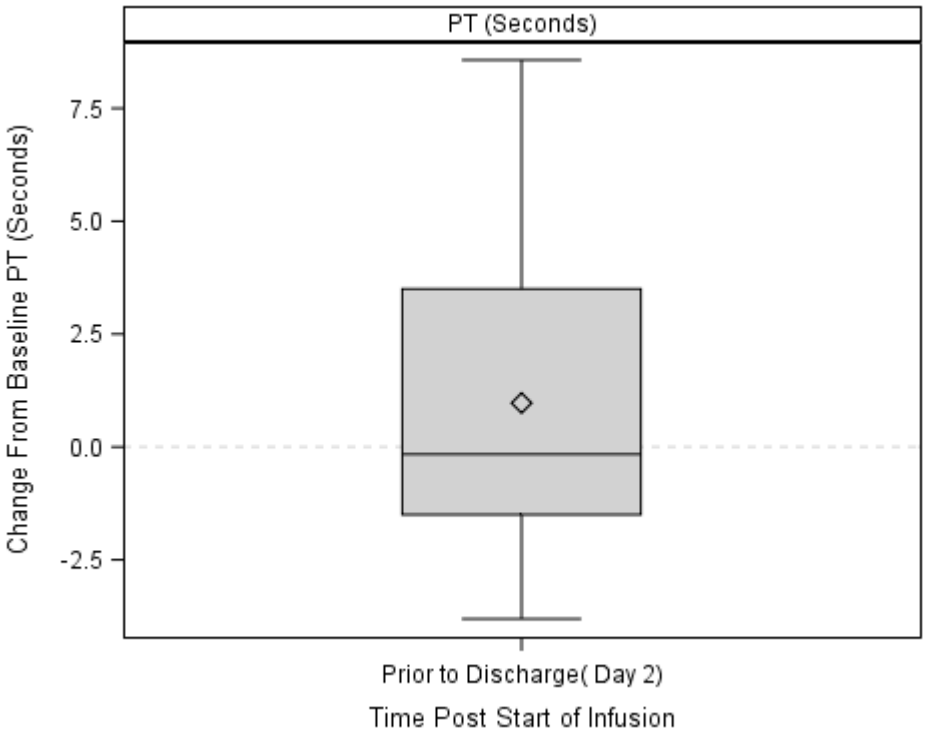
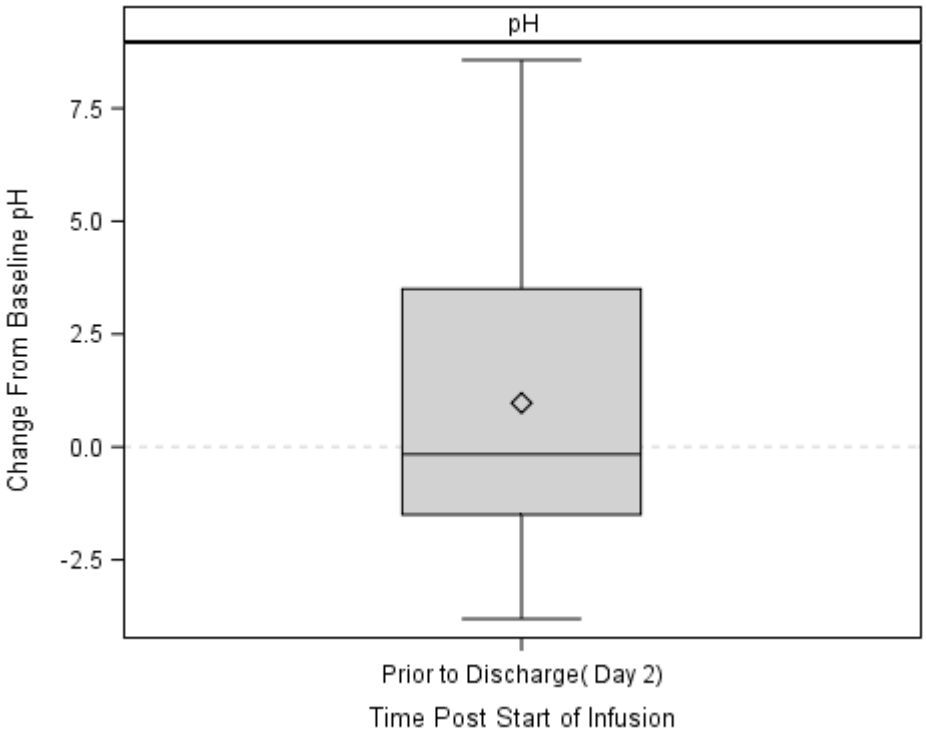


Figure with Similar Format:

Figure 34: Coagulation Laboratory Results by Scheduled Timepoints: Box Plot of Change from Baseline by Laboratory Parameter – aPTT

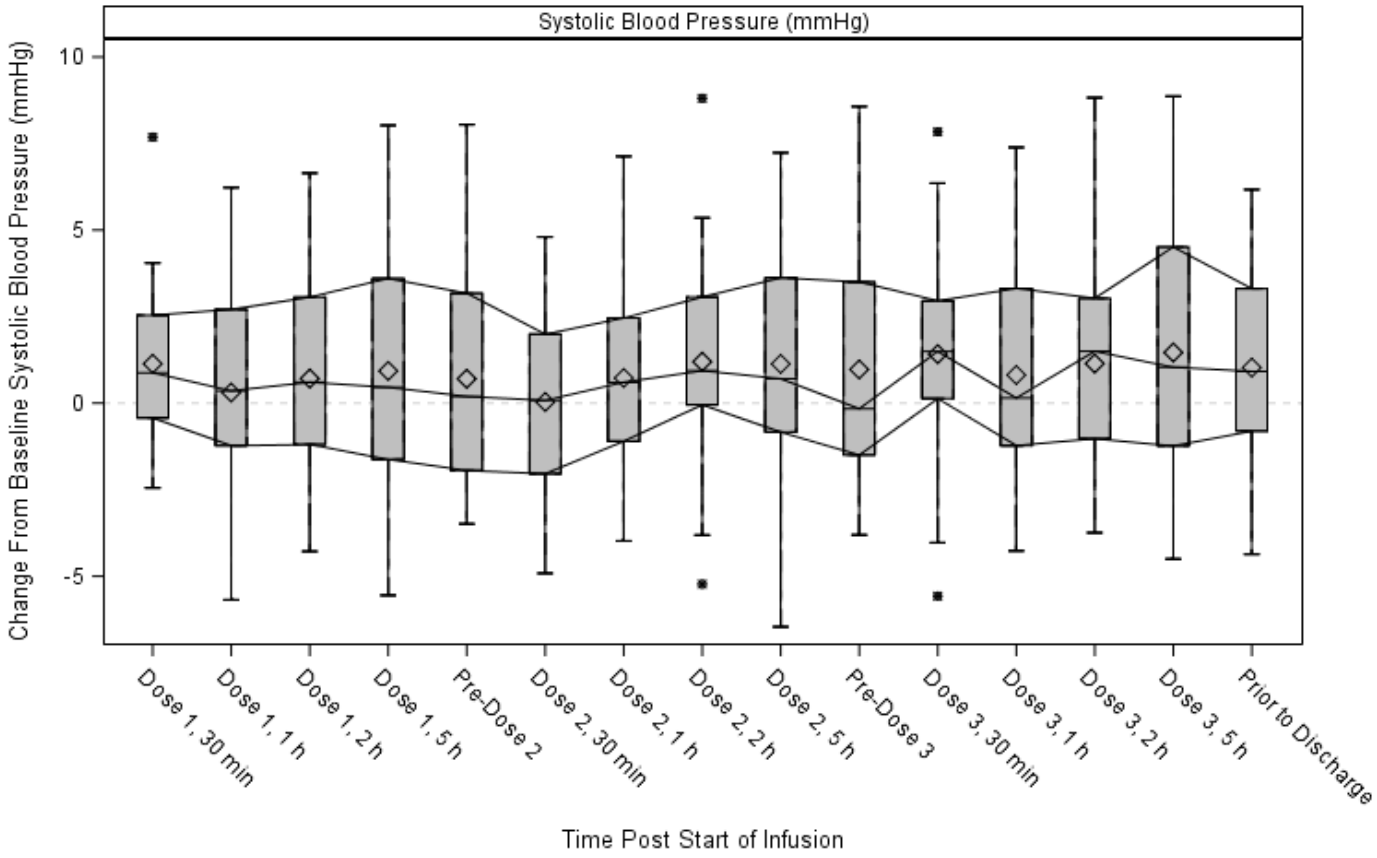
Figure 35: Urinalysis Laboratory Results by Scheduled Timepoints: Box Plot of Change from Baseline by Laboratory Parameter – pH



Figures with Similar Format:

Figure 36: Urinalysis Laboratory Results by Scheduled Timepoints: Box Plot of Change from Baseline by Laboratory Parameter – Specific Gravity

Figure 37: Vital Signs Change from Baseline by Scheduled Timepoint: Box Plot of Change from Baseline by Assessment – Systolic Blood Pressure



Figures with Similar Format:

Figure 38: Vital Signs Change from Baseline by Scheduled Timepoint: Box Plot of Change from Baseline by Assessment – Diastolic Blood Pressure

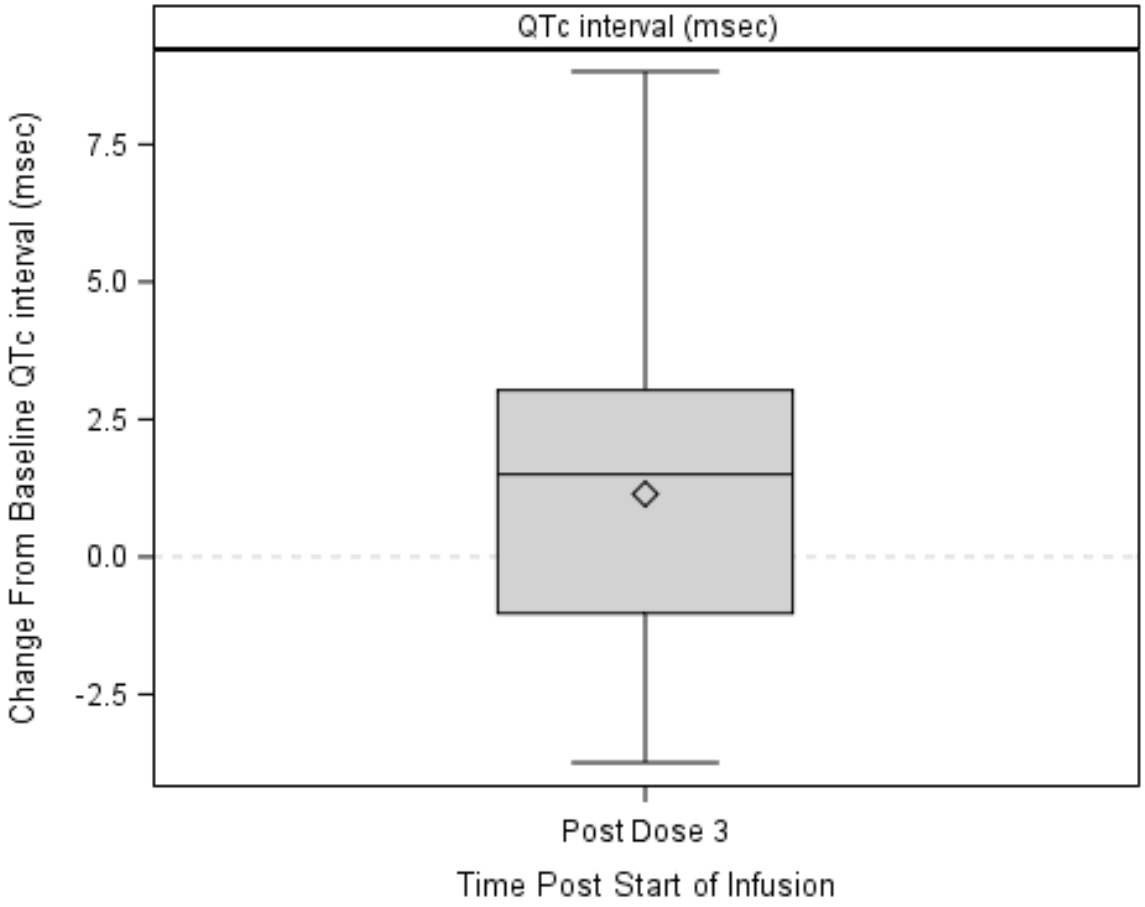
Figure 39: Vital Signs Change from Baseline by Scheduled Timepoint: Box Plot of Change from Baseline by Assessment – Heart Rate

Figure 40: Vital Signs Change from Baseline by Scheduled Timepoint: Box Plot of Change from Baseline by Assessment – Respiratory Rate

Figure 41: Vital Signs Change from Baseline by Scheduled Timepoint: Box Plot of Change from Baseline by Assessment – Peripheral Oxygen saturation

Figure 42: Vital Signs Change from Baseline by Scheduled Timepoint: Box Plot of Change from Baseline by Assessment – Temperature

Figure 43: ECG Change from Baseline by Scheduled Timepoint: Box Plot of Change from Baseline by Assessment – QTc Interval (msec)



Figures with Similar Format:

Figure 44: ECG Change from Baseline by Scheduled Timepoint: Box Plot of Change from Baseline by Assessment – PR Interval

Figure 45: ECG Change from Baseline by Scheduled Timepoint: Box Plot of Change from Baseline by Assessment – QRS Duration

Figure 46: Concentration Profiles for All Subjects by Time (h) – Dose 1 and Dose 3

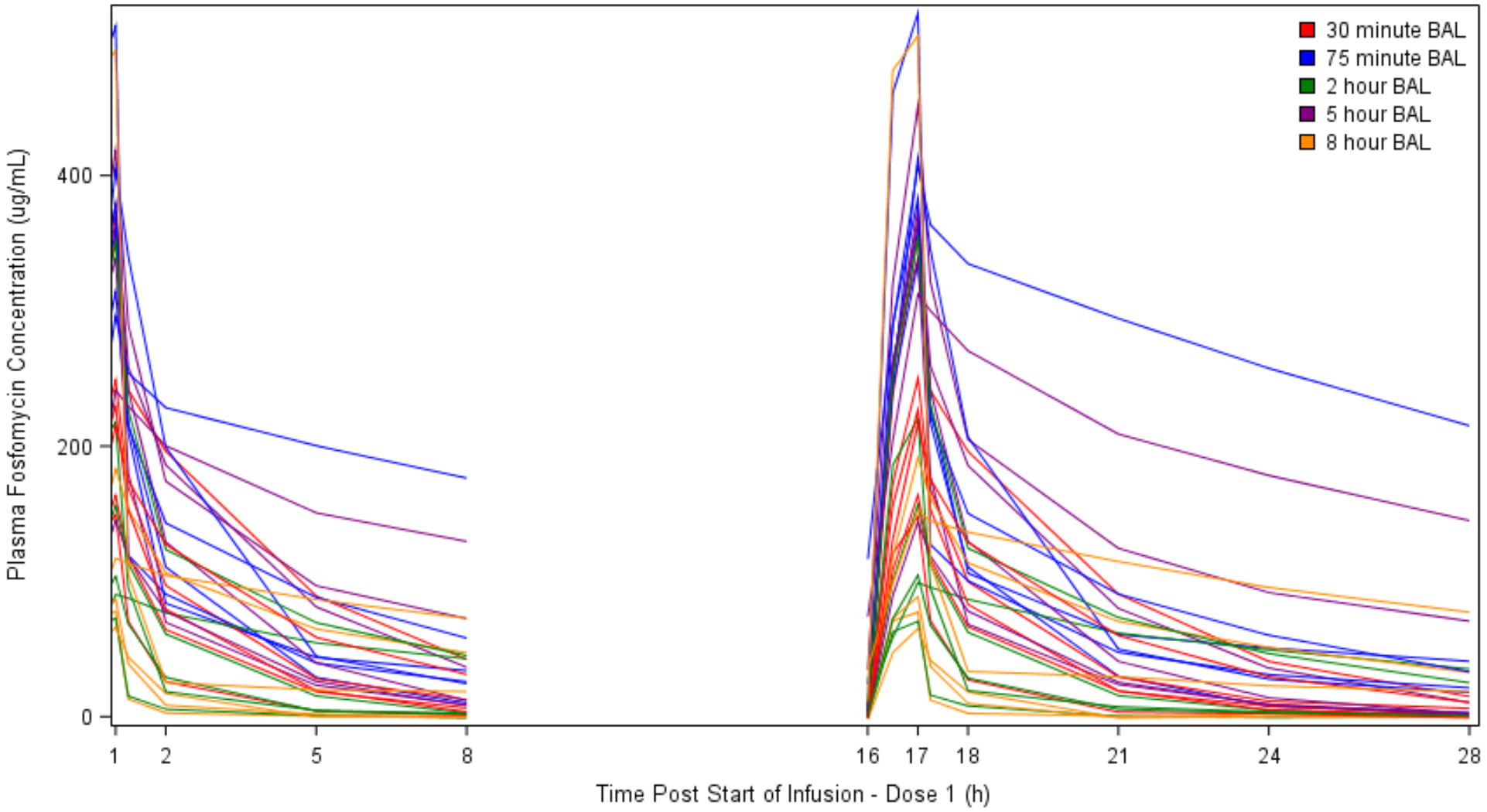


Figure 47: Semi-log Concentration Profiles for All Subjects by Time (h) – Dose 1 and Dose 3

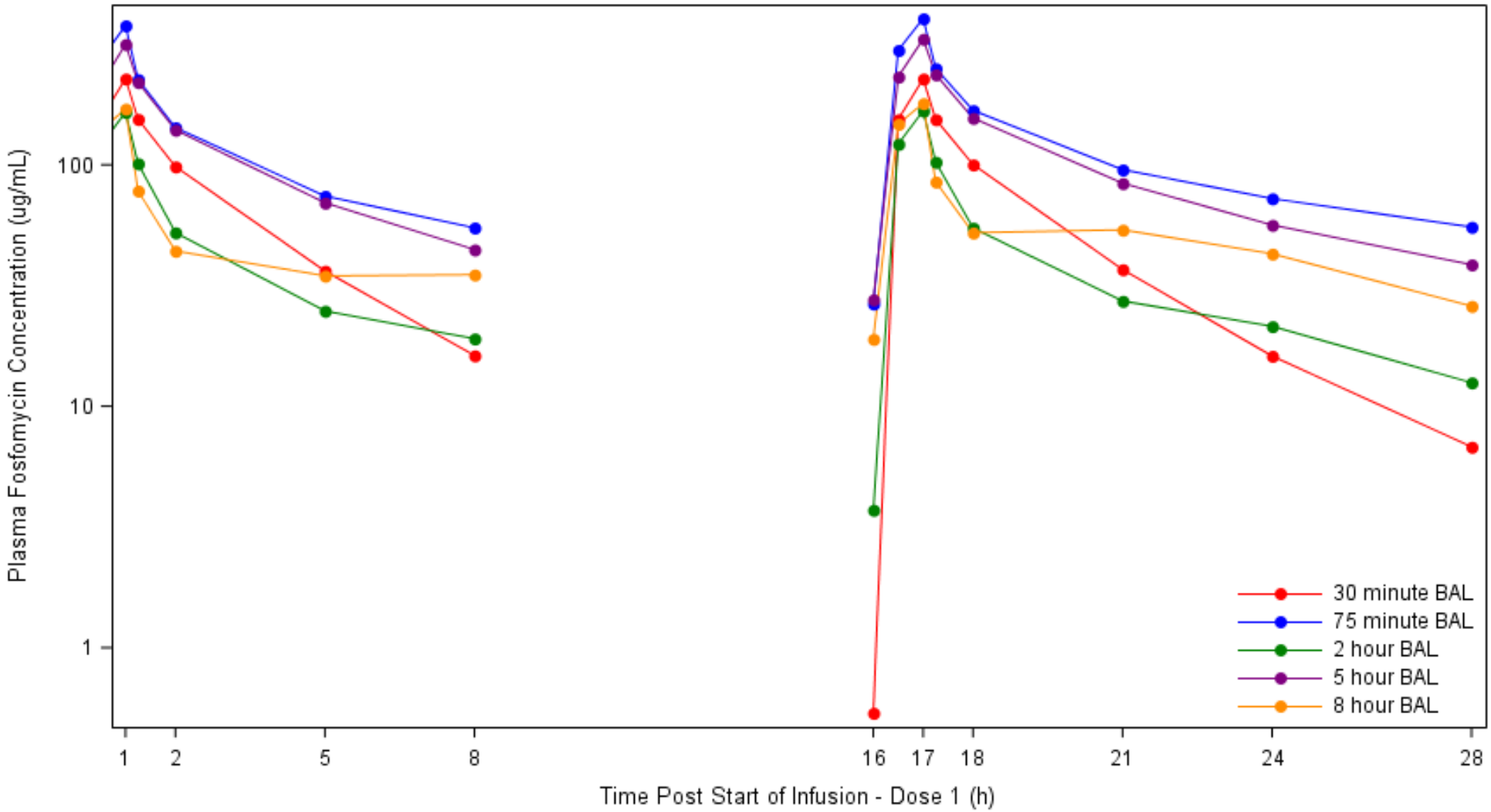
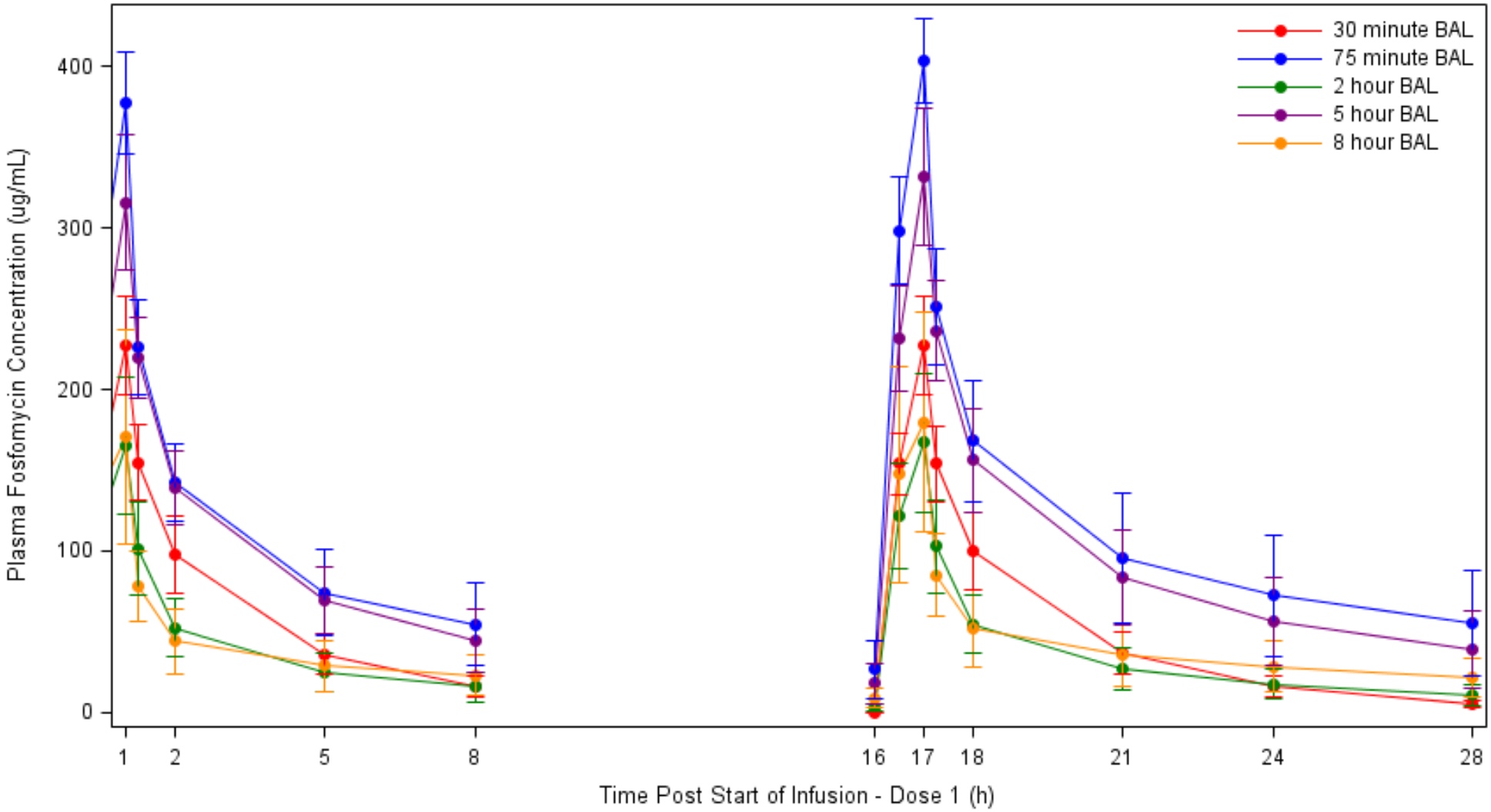


Figure 48: Mean Plasma Concentration by Nominal Time – Dose 1



Note: Error bars give +/- 1 standard deviation.

Figure 49: Semi-log Plot of GM Plasma Concentration by Nominal Time – Dose 1 and Dose 3

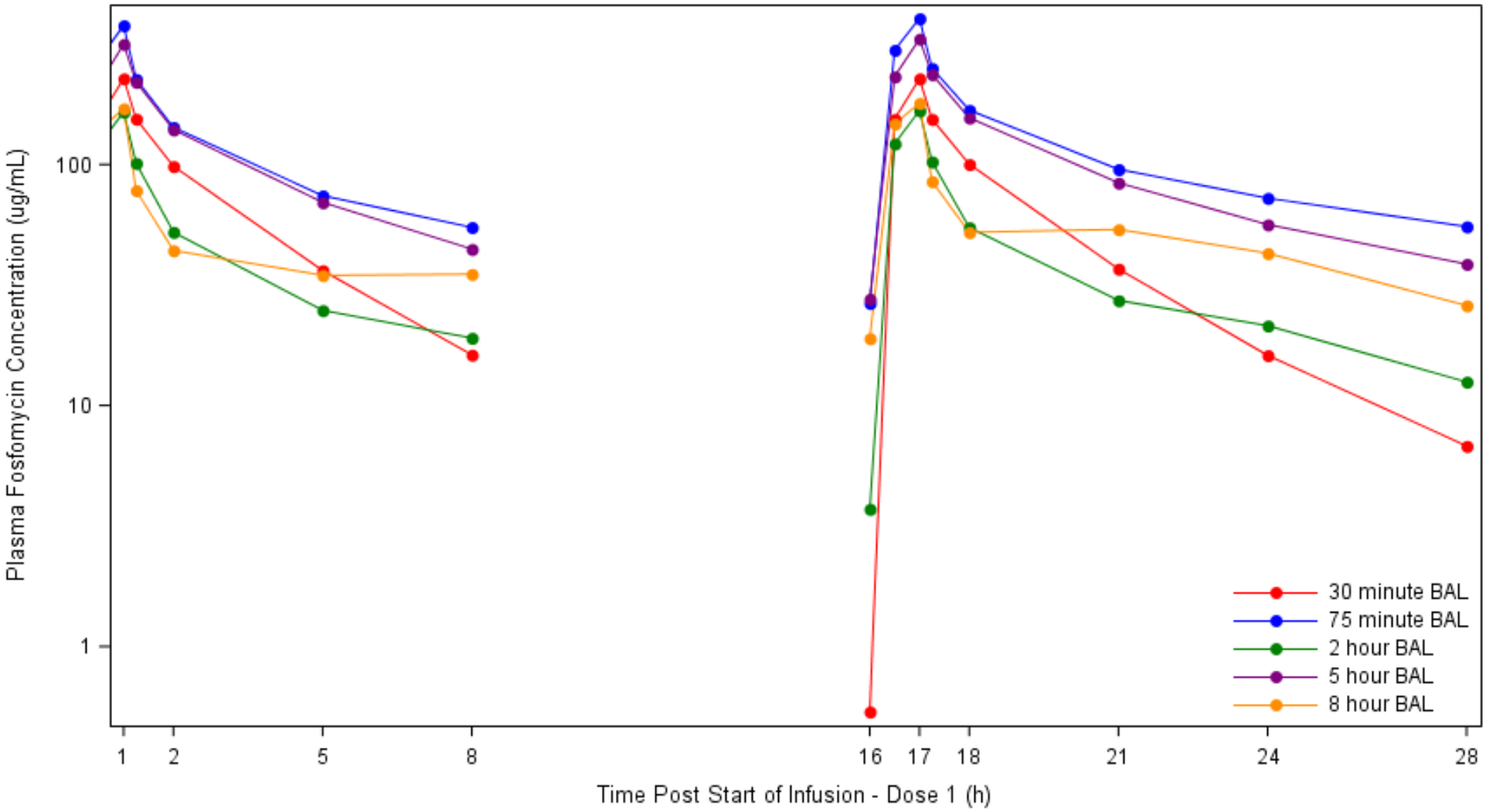


Figure 50: Fosfomycin Concentrations in Plasma, ELF, and AM by Subjects and Time(h) – Dose 3

[Implementation Note: Use different colors and symbols for Plasma, ELF and AM]

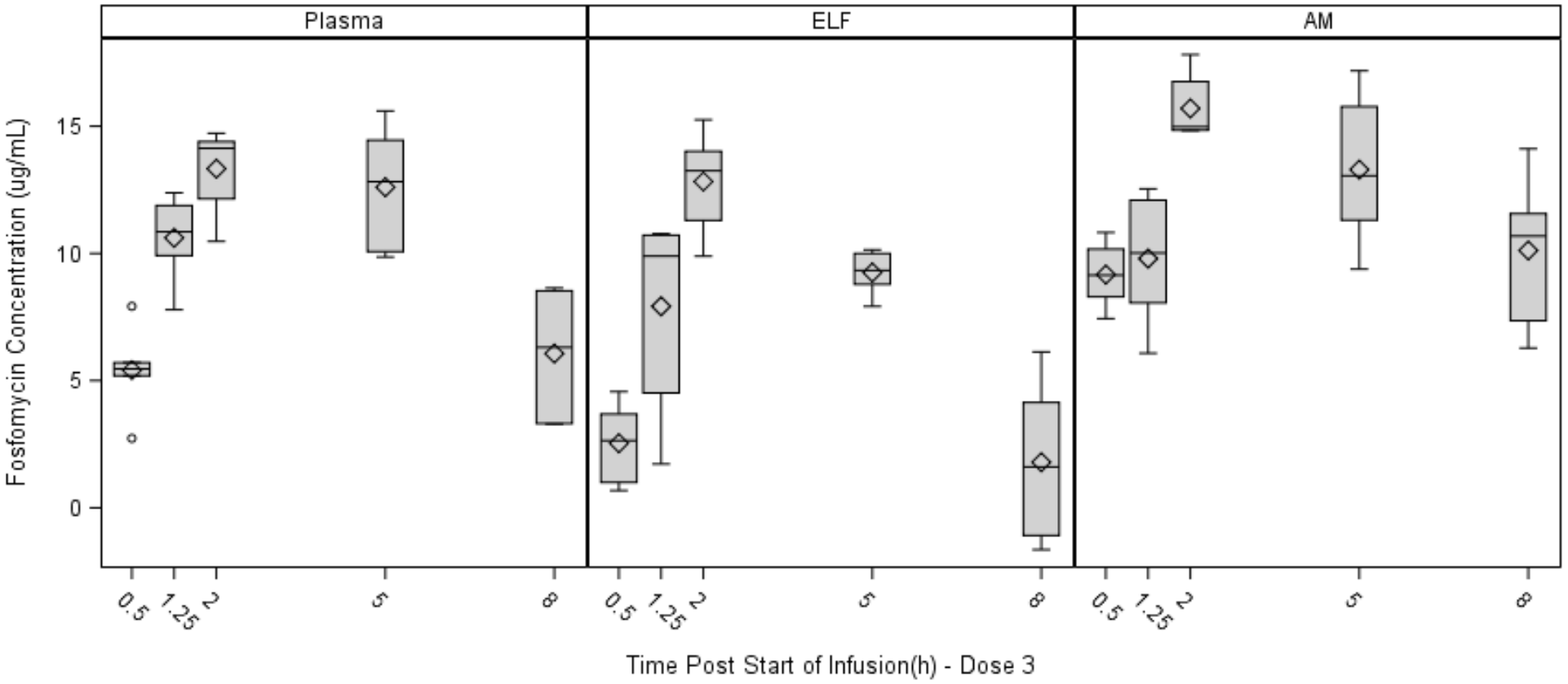


Figure 51: Population Median Concentrations in Plasma, ELF, and AM by Time (h) – Dose 3

[Implementation Note: Use different colors for Plasma, ELF and AM]

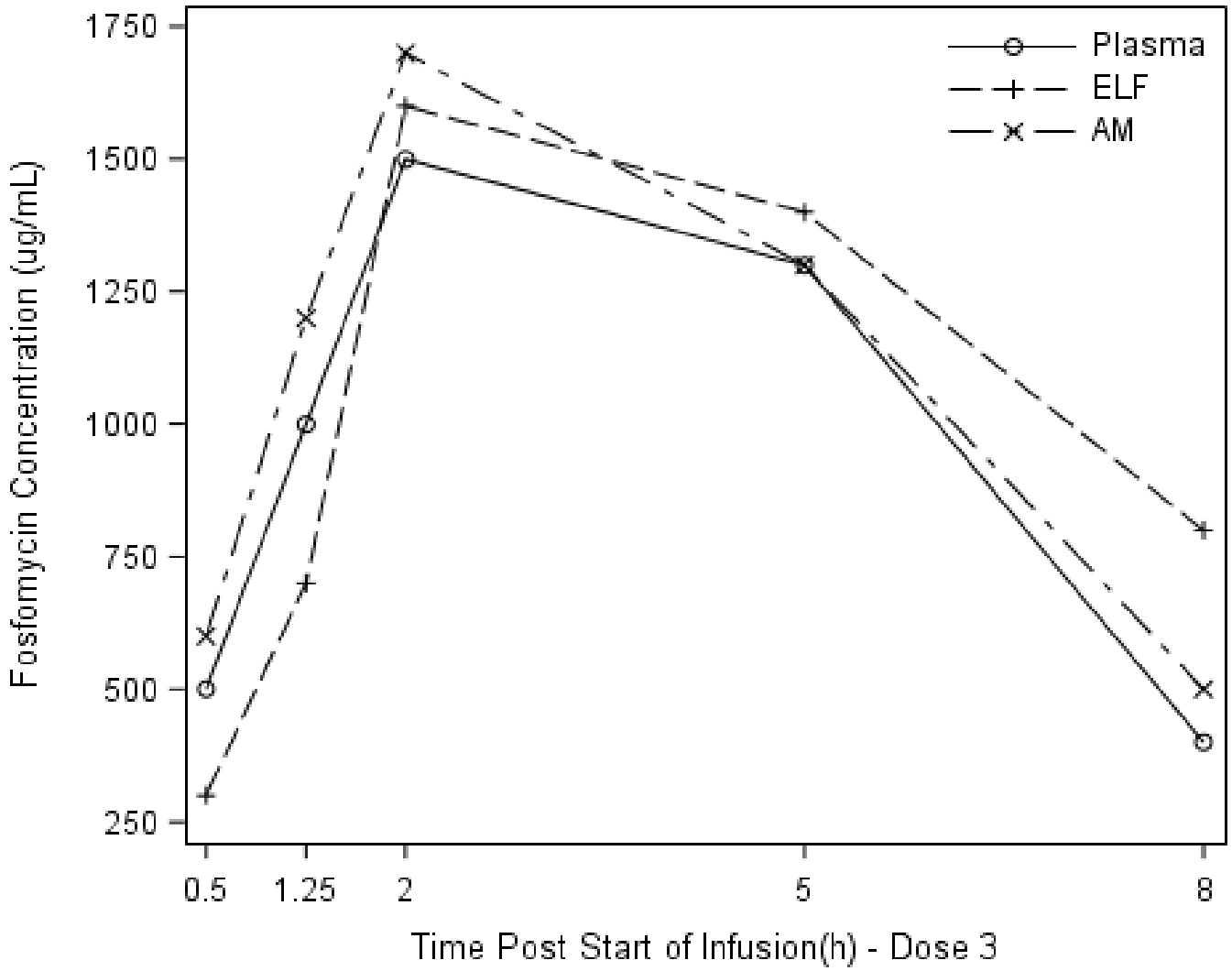
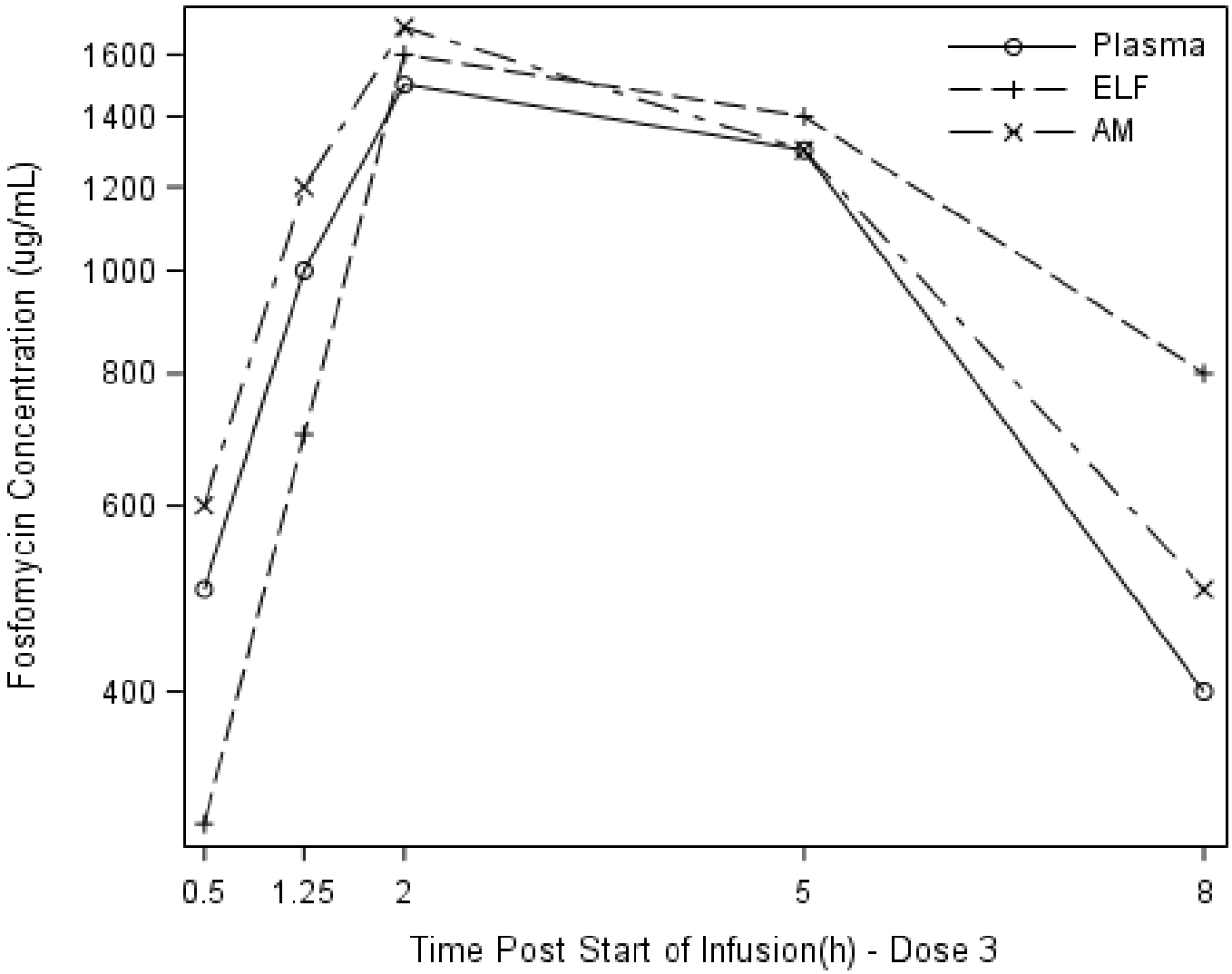


Figure 52: Semi-log Plot of Population Median Concentrations in Plasma, ELF, and AM by Time (h) – Dose 3

[Implementation Note: Use different colors for Plasma, ELF and AM]



APPENDIX 3. LISTINGS MOCK-UPS

LISTINGS

16.1.6 Listing of Subjects Receiving Investigational Product

(not included in SAP, but this is a placeholder for the CSR)

16.2 Database Listings by Subject

16.2.1 Discontinued Subjects

Listing 1: Early Terminations or Discontinued Subjects

[Implementation Note: Category will be either "Early Termination" or "Treatment Discontinuation." In the "Reason" column, concatenate any "specify" fields, including AE number and DV number. In the CSR, Subject ID should be USUBJID (not PATID) for purposes of de-identification. Sort order: BAL Sampling Timepoint, Subject ID, alphabetically by Category (in the case a subject both terminates early and discontinues treatment).]

BAL Sampling Timepoint	Subject ID	Category	Reason for Early Termination or Treatment Discontinuation	Study Day	Number of Doses Received	Replaced with Alternate

16.2.2 Protocol Deviations

Listing 2: Subject-Specific Protocol Deviations

[Implementation Note: In the "Deviation" column, concatenate any and all "specify" fields (including visit number, etc.). If "Reason for Deviation" is "Other," concatenate "specify" field, separate by a colon, e.g., "Other: Subject refusal." In the CSR, Subject ID should be USUBJID (not PATID) for purposes of de-identification. Sort order: BAL Sampling Timepoint, Subject ID, DV Number.]

BAL Sampling Timepoint	Subject ID	DV Number	Deviation	Deviation Category	Study Day	Reason for Deviation	Deviation Resulted in AE?	Deviation Resulted in Subject Termination?	Deviation Affected Product Stability?	Deviation Resolution	Comments

Listing 3: Non-Subject-Specific Protocol Deviations

[Implementation Note: In the "Deviation" column, concatenate any and all "specify" fields (including visit number, etc.). If "Reason for Deviation" is "Other," concatenate "specify" field, separate by a colon, e.g., "Other: Subject refusal." Sort order: Start Date.]

Start Date	Deviation	End Date	Reason for Deviation	Deviation Resulted in Subject Termination?	Deviation Affected Product Stability?	Deviation Category	Deviation Resolution	Comments

16.2.3 Subjects Excluded from the Efficacy Analysis

Listing 4: Subjects Excluded from Analysis Populations

[Implementation Note: This data in this listing should be congruent with the "Analysis Populations by Study Group" table. The reasons included here should match the SAP text that describes who will be excluded from analyses.]

BAL Sampling Timepoint	Subject ID	Analyses in which Subject is Included	Analyses from which Subject is Excluded	Results Available?	Reason Subject Excluded
		[e.g., Safety, PK]	[e.g., Safety, PK]		
Notes: "Yes" in the "Results available" column indicates that available data were removed from the analysis. "No" indicates that no data were available for inclusion in the analysis.					

16.2.4 Demographic Data

Listing 5: Demographic Data

[Implementation Note: If a subject is multi-racial, in "Race" column, note "Multiple: (list races, separated by a comma)." In the CSR, Subject ID should be USUBJID (not PATID) for purposes of de-identification. Height, weight, and BMI included in this listing will be from the initial screening visit

Sort order: BAL Sampling Timepoint, Subject ID]

BAL Sampling Timepoint	Subject ID	Sex	Age at Enrollment (years)	Ethnicity	Race	Height (cm)	Weight (Kg)	BMI (kg/m^2)

Listing 6: Pre-Existing and Concurrent Medical Conditions

[Sort order: BAL Sampling Timepoint, Subject ID]

BAL Sampling Timepoint	Subject ID	MH Number	Medical History Term	Condition Start Day	Condition End Day	MedDRA System Organ Class	MedDRA Preferred Term

16.2.5 Drug Concentration Data

Listing 7: Treatments Administered

[Sort order: BAL Sampling Timepoint, Subject ID]

BAL Sampling Timepoint	Subject ID	Drug	Dose Number	Planned Dose (mg)	Actual Dose (mg)	Infusion Start Date	Infusion Start Time (hh:mm)	Infusion End Date	Infusion End Time (hh:mm)	Interruptions in study product administration

Listing 8: Unsolicited Adverse Events

[Sort order: BAL Sampling Timepoint, Subject ID, AE Number]

Adverse Event	Associated with Dose No.	No. of hours Post Associated Dose (Duration)	Severity	SAE?	Relationship to Study Treatment	In Not Related, Alternative Etiology	Action Taken with Study Treatment	Subject Discontinued Due to AE	Outcome	MedDRA System Organ Class	MedDRA Preferred Term
BAL Sampling Timepoint: , Subject ID: , AE Number:											
Comments:											
BAL Sampling Timepoint: , Subject ID: , AE Number:											
Comments:											
Note: For additional details about SAEs, see Table: 14.											

16.2.8 Individual Laboratory Measurements

Listing 9: Clinical Laboratory Results – Chemistry

[Sort order: BAL Sampling Timepoint, Subject ID, Planned Timepoint]

BAL Sampling Timepoint	Subject ID	Planned Timepoint	Actual Timepoint	Sex	Age at Enrollment (years)	Laboratory Parameter (Units)	Result (Severity Grade)	Reference Range Low	Reference Range High

Listing 10: Clinical Laboratory Results – Hematology

[Sort order: BAL Sampling Timepoint, Subject ID, Planned Timepoint]

BAL Sampling Timepoint	Subject ID	Planned Timepoint	Actual Time Post Infusion Dose # (h)	Sex	Age at Enrollment (years)	Laboratory Parameter (Units)	Result (Severity Grade)	Reference Range Low	Reference Range High

Listing 11: Clinical Laboratory Results – Coagulation

[Sort order: BAL Sampling Timepoint, Subject ID, Planned Timepoint]

BAL Sampling Timepoint	Subject ID	Planned Timepoint	Actual Time Post Infusion Dose # (h)	Sex	Age at Enrollment (years)	Laboratory Parameter (Units)	Result (Severity Grade)	Reference Range Low	Reference Range High

Listing 12: Clinical Laboratory Results – Urinalysis

[Sort order: BAL Sampling Timepoint, Subject ID, Planned Timepoint]

BAL Sampling Timepoint	Subject ID	Planned Timepoint	Actual Time Post Infusion Dose # (h)	Sex	Age at Enrollment (years)	Laboratory Parameter (Units)	Result (Severity Grade)	Reference Range Low	Reference Range High

Listing 13: Screening Laboratory Results – Serology

[Sort order: BAL Sampling Timepoint, Subject ID, Planned Timepoint]

BAL Sampling Timepoint	Subject ID	Planned Timepoint	Actual Time Post Infusion Dose # (h)	HIV 1/2 antigen	HCV surface antibodies	HBsAg

Listing 14: Laboratory Results – Urine Toxicology and Alcohol Testing

[Sort order: BAL Sampling Timepoint, Subject ID, Planned Timepoint]

BAL Sampling Timepoint	Subject ID	Planned Timepoint	Actual Time Post Infusion Dose # (h)	Amphetamines	Methamphetamine	Barbiturates	Benzodiazepines	THC	Cocaine	Opiates	Cotinine	Alcohol

16.2.9 Vital Signs and Physical Exam Findings

Listing 15: Vital Signs

[Sort order: BAL Sampling Timepoint, Subject ID, Planned Timepoint]

BAL Sampling Timepoint	Subject ID	Planned Timepoint	Actual Time Post Infusion Dose # (h)	Temperature (°C)	Systolic Blood Pressure (mmHg)	Diastolic Blood Pressure (mmHg)	Heart Rate (beats/min)	Respiratory Rate (breaths/min)	Weight (kg)	Height (cm)

Listing 16: Physical Exam Findings

[Sort order: BAL Sampling Timepoint, Subject ID, Planned Timepoint]

BAL Sampling Timepoint	Subject ID	Planned Timepoint	Actual Time Post Infusion Dose # (h)	Body System	Abnormal Finding	Reported as an AE? (AE Description; Number)

Listing 17: Listing of ECG Interval Measurements

[Sort order: BAL Sampling Timepoint, Subject ID, Planned Timepoint]

					Interval Measurement			
BAL Sampling Timepoint	**Subject ID**	**Planned Timepoint**	**Actual Time Post Infusion Dose # (h)**	**Time of Assessment (hh:mm)**	**QRS duration (msec)**	**QTc interval (msec)**	**PR Interval (msec)**	**Overall Assessment**

16.2.10 Prior and Concomitant Medications

Listing 18: Prior Medications

[Sort order: BAL Sampling Timepoint, Subject ID, CM Number]

BAL Sampling Timepoint	Subject ID	CM Number	Medication	Medication Start Day	Medication End Day	Indication	Taken for a condition on Medical History? (MH Description; Number)	ATC Level 1 (ATC Level 2)

Listing 19: Concomitant Medications

[Sort order: BAL Sampling Timepoint, Subject ID, CM Number]

BAL Sampling Timepoint	Subject ID	CM Number	Medication	Medication Start Day	Medication End Day	Indication	Taken for an AE? (AE Description; Number)	Taken for a condition on Medical History? (MH Description; Number)	ATC Level 1 (ATC Level 2)

16.2.11 Pregnancy Reports

Listing 20: Pregnancy Reports – Maternal Information

[Sort order: BAL Sampling Timepoint, Subject ID, Pregnancy Number]

BAL Sampling Timepoint	Subject ID	Pregnancy Number	Study Day Corresponding to Estimated Date of Conception	Source of Maternal Information	Pregnancy Status	Mother's Pre-Pregnancy BMI	Mother's Weight Gain During Pregnancy	Tobacco, Alcohol, or Drug Use During Pregnancy?	Medications During Pregnancy?	Maternal Complications During Pregnancy?	Maternal Complications During Labor, Delivery, or Post-Partum?
Note: Maternal Complications are included in the Adverse Event listing. Medications taken during pregnancy are included in the Concomitant Medications Listing.											

Listing 21: Pregnancy Reports – Gravida and Para

			Live Births												
Subject ID	**Pregnancy Number**	**Gravida**	**Extremely PB[a]**	**Very Early PB[a]**	**Early PB[a]**	**Late PB[a]**	**Early TB[b]**	**Full TB[b]**	**Late TB[b]**	**Post TB[b]**	**Still Births**	**Spontaneous Abortion/ Miscarriage**	**Elective Abortions**	**Therapeutic Abortions**	**Major Congenital Anomaly with Previous Pregnancy?**

Note: Gravida includes the current pregnancy, para events do not.
a Preterm Birth
b Term Birth

Listing 22: Pregnancy Reports – Live Birth Outcomes

Subject ID	Pregnancy Number	Fetus Number	Pregnancy Outcome (for this Fetus)	Fetal Distress During Labor and Delivery?	Delivery Method	Gestational Age at Live Birth	Size for Gestational Age	Apgar Score, 1 minute	Apgar Score, 5 minutes	Cord pH	Congenital Anomalies?	Illnesses/ Hospitalizations within 1 Month of Birth?

Note: Congenital Anomalies are included in the Adverse Event listing.

Listing 23: Pregnancy Reports – Still Birth Outcomes

Subject ID	Date of Initial Report	Fetus Number	Pregnancy Outcome (for this Fetus)	Fetal Distress During Labor and Delivery?	Delivery Method	Gestational Age at Still Birth	Size for Gestational Age	Cord pH	Congenital Anomalies?	Autopsy Performed?	If Autopsy, Etiology for Still Birth Identified?

Listing 24: Pregnancy Reports – Spontaneous, Elective, or Therapeutic Abortion Outcomes

Subject ID	Date of Initial Report	Fetus Number	Pregnancy Outcome (for this Fetus)	Gestational Age at Termination	Abnormality in Product of Conception?	Reason for Therapeutic Abortion

Listing 25: Subject Level Fosfomycin Concentrations in Plasma

[Sort order: BAL Sampling Timepoint, Subject ID, Dose Number

If Fosfomycin Concentration level below LLOQ report as BQL and include footnote "BQL= Below Quantitative Limit" If no levels are BQL, footnote does not need to be included in table.]

BAL Sampling Timepoint	Subject ID	Dose Number	Nominal Time[a] (h)	Actual Time[a] (h)	Actual Dose (mg)	Fosfomycin Concentration (µg/mL)	Sample Within Time Window	Used in λ_z Calculations
							yes/no	

Note: BQL= Below Quantitative Limit
[a] Times are relative to start of study product infusion for associated dose. For actual time, out of window times are indicated by an asterisk.

Listing 26: Subject-Specific PK Parameters

BAL Sampling Timepoint	Subject ID	Dose Number	C_{max} (µg/mL)	T_{max} (h)	$AUC_{(0-8)}$ (h*µg/mL)	$AUC_{(0-12)}$ (h*µg/mL)	$AUC_{(0-inf)}$ (h*µg/mL)	λ_z (/h)	$t_{1/2}$ (h)	CL (L/h)	V_{ss} (L)

Listing 27: Subject Level Fosfomycin Concentrations in ELF and in AM Cell Pellet

[Sort order: BAL Sampling Timepoint, Subject ID]

BAL Sampling Timepoint	Subject ID	Nominal Time[a] (h)	Actual Time[a] (h)	BAL Fosfomycin Concentration (µg/mL) ($Fosfomycin_{BAL}$)	BAL Urea Concentration (µg/mL) ($Urea_{BAL}$)	Plasma Urea Concentration (µg/mL) ($Urea_{Plasma}$)	Calculated ELF Fosfomycin Concentration (µg/mL)[b] ($Fosfomycin_{ELF}$)	AM Fosfomycin Concentration (µg/mL)	Sample Within Time Window
									yes/no

[a]Times are relative to start of third dose of study product infusion. For actual time, out of window times are indicated by an asterisk.
[b] $Fosfomycin_{ELF} = Fosfomycin_{BAL} \times (Urea_{Plasma}/Urea_{BAL})$