NCT05386823

Statistical Analysis Plan

Official Title: A Phase 1, Randomized, Double-Blind, Placebo-Controlled.Single Ascending Dose Study Following Intravenous Administration ofHF1K16 in Healthy Subjects to Evaluate the Safety, Tolerability andPharmacokinetics ofHF1K16

Document Date: June 20, 2021

Statistical Analysis Plan

Protocol No. HF1K16-101, Version 7.0, 01 April 2021

A PHASE 1, RANDOMIZED, DOUBLE-BLIND, PLACEBO-CONTROLLED, SEQUENTIAL PARALLEL GROUP, SINGLE ASCENDING DOSE STUDY FOLLOWING INTRAVENOUS ADMINISTRATION OF HF1K16 IN HEALTHY SUBJECTS TO EVALUATE THE SAFETY, TOLERABILITY AND PHARMACOKINETICS OF HF1K16

Sponsor:	HighField Biopharmaceutical Corporation 368 LiuHe Road, Building 1 BinJiang District, HangZhou, China
Prepared by:	Frontage Clinical Services, Inc. 200 Meadowlands Parkway Secaucus, NJ 07094, USA Tel: (201) 678-0288 Fax: (201) 552-2597
Document status:	Final v1.0
Release date:	11 June 2021

Signature Page

Author:	
Ruth E. Swanton, MPH Director of Biostatistics Frontage Clinical Services, Inc. 200 Meadowlands Parkway Secaucus, NJ 07094	Signature: Ruth E Swanton Date: 11 JUN 2021
Michael Willett, PharmD RAC Acting Director, Clinical Pharmacology Frontage Clinical Services, Inc. 200 Meadowlands Parkway Secaucus, NJ 07094	Signature: DocuSigned by: Michael S. Willett A31011AB9A964AE... Date: 11-Jun-2021
Frontage Representative:	
Annie Li, MD Associate Director, Clinical Operations and Project Management Frontage Clinical Services, Inc. 200 Meadowlands Parkway Secaucus, NJ 07094	Signature: Date: 16 Jun 2021
Sponsor Representative(s):	
Yuhong Xu, PhD CEOHighField Biopharmaceutical Corporation 368 LiuHe Road, Building 1 BinJiang District, HangZhou, China Email:	Signature: Date: 20 JUN 2021

Table of Contents

List of In-text Tables

Glossary and Abbreviations

Abbreviation	Term
AE	Adverse Event
ALP	Alkaline Phosphatase
ALT	Alanine Aminotransferase
AST	Aspartate Aminotransferase
AUC_{last}	AUC from Time Zero to Time of Last Measurable Concentration
AUC_{inf}	Area Under the Concentration-Time Curve from Zero Up to Infinity with Extrapolation of the Terminal Phase
BMI	Body Mass Index
BP	Blood Pressure
BQL	Below Quantitation Limit
CI	Confidence interval
C_{max}	Observed Maximum Serum Concentration
CL/F	Apparent Total Plasma Clearance of Drug
CRU	Clinical research unit
CTCAE	National Cancer Institute Common Terminology Criteria for Adverse Events
CV	Coefficient of Variation
D	Day
DLT	Dose limiting toxicity
eCRF	Electronic Case Report Form
ECG	Electrocardiogram
FCS	Frontage Clinical Services
FSH	Follicle Stimulating Hormone
λ_z	Terminal elimination rate constant
h or hr	Hour (s)
ICF	Informed consent form
ID	Identification
K_{el}	Terminal elimination rate constant
LLOQ	Lower limit of quantification
Max	Maximum
MedDRA	Medical Dictionary for Regulatory Activities
Min	Minimum
mg	Milligram(s)
mL	Milliliter(s)

Abbreviation	Term
MRT	Mean residence time
N	Sample size
NCA	Non-compartmental analysis
ng	Nanogram(s)
PK	Pharmacokinetic(s)
PP	Pharmacokinetic parameter
PR	Pulse rate
RR	Respiratory rate
SAD	Single Ascending Dose
SAE	Serious Adverse Event
SAP	Statistical Analysis Plan
SD	Standard deviation
SE	Standard error of the mean
SMC	Safety Monitoring Committee
T_{max}	Time to reach maximum concentration
$t_{1/2}$	Terminal half-life of elimination
TEAE	Treatment Emergent Adverse Event
TLF	Tables, Listings, Figures
V_zF	Volume of distribution during terminal phase
WHO	World Health Organization

1 Introduction

This study is designed to assess safety, tolerability, and pharmacokinetics (PK) of HF1K16 in healthy adult volunteers. This study is single ascending intravenous administration (SAD). The purpose of this study is to determine the safety, and tolerability of HF1K16 during dose escalation, and assess the PK of this drug with single dosing. Safety monitoring is utilized to ensure the safety of subjects.

The starting dose of Up to 3 mg/m^2 for the study represents about a 10-fold safety margin is selected based on the results from non-clinical toxicity studies. Refer to the protocol for further information on this starting dose.

Approximately 32 healthy subjects will be enrolled and randomized to receive a single intravenous administration of HF1K16 or matched placebo. A randomization 3 to 1 ratio will be applied to subjects who receive active or placebo. The escalated doses will be defined according to emerging safety and tolerability data and reviewed by the Safety Monitoring Committee (SMC). Different subjects will be enrolled into different dose cohorts to avoid repeated exposure of the investigational product (IP) to one individual.

This Statistical Analysis Plan (SAP) covers the detailed procedures for performing the Safety and PK statistical analyses and for producing the Tables, Listings, and Figures (TLFs) in the study. The templates in the TLF Shells are followed once the SAP is signed. Only minor changes to the TLF Shells will occur once the SAP is final (this includes footnotes due to data issues).

The safety measurements (laboratory analyses (serum chemistry, hematology, and urinalysis)) will be provided by BioReference Laboratories.

The bioanalytical pharmacokinetic work will be conducted by Frontage Laboratories.

2 Study Objectives and Endpoints

2.1 Primary objectives

- To evaluate the safety and tolerability of different doses of HF1K16 following single ascending doses by intravenous administration.

2.2 Secondary objectives

- To characterize the pharmacokinetics (PK) of HF1K16 following single ascending doses by intravenous administration.

2.3 Primary endpoints

- Incidence of Adverse Events (AEs) and Serious Adverse Events (SAEs)
- Injection Site Assessments

- Physical examination
- Laboratory abnormalities (hematology, serum chemistry, and urinalysis)
- Vital signs (blood pressure, respiratory rate, pulse rate, and oral body temperature)
- 12-lead electrocardiogram (ECG)

2.4 Secondary endpoints

- Pharmacokinetics (PK) in treated subjects, including:
    - Peak concentration (C_{max})
    - Time to peak (T_{max})
    - Area under the concentration-time curve (AUC_{inf}, AUC_{last})
    - Terminal elimination rate constant (K_{el})
    - Elimination half-life ($t_{½}$)
    - Apparent Clearance (CL/F)
    - Volume of distribution (VzF)
    - Mean Residence Time (MRT)

3 Study Design and Methods

This is a Phase 1, double blind, randomized, placebo controlled, single-ascending dose study. Approximately 32 healthy subjects will be randomized to either HF1K16 or matching placebo (3:1 ratio). The starting dose will be Up to 3 mg/m^2 with four (4) cohorts planned. The escalated doses will be defined according to emerging safety and tolerability data.

Each dose cohort will include 6 HF1K16 and 2 matching placebo subjects per the randomization schema. Dosing for each subsequent cohort will only proceed after the safety and tolerability data of the previous cohort has been reviewed.

The dose escalation scheme is provided in Table 1.

Table 1 Dose Escalation

Cohort (single infusion dosing)	**Dose (mg/ m^2) Once**
1	Up to 3 mg/m^2 (6 HF1K16 and 2 placebo)
2	Up to 6 mg/m^2 (6 HF1K16 and 2 placebo)
3	Up to 10 mg/m^2 (6 HF1K16 and 2 placebo)
4	Up to 13 mg/m^2 (6 HF1K16 and 2 placebo)

Note: the 'Up to…' will be removed and the final dose replaced in the TLF Shells..

Refer to the dosing escalation stopping rules (refer to the protocol for the complete description).

This study will include an up to 28-day screening period (Days -28 to -1), including a clinic check-in (Day -1), a treatment day (Day 1), and observation period refer to Table 2 below. Subjects will exit from the study after completing Follow-up telephone call on Day 8 (± 2 days) or early withdrawal evaluation(s). The expected duration of participation for each subject, therefore, will be approximately 38 days.

Eligible subjects will be admitted to the clinic on Day -1 for baseline evaluations. Safety assessments including physical examination, vital signs, ECGs (triplicate for specific time points), and standard clinical laboratory evaluations will be performed, and following enrollment, the subjects will be randomized. Subjects will be administered the study drug on Day 1, monitored according to the Schedule of Procedures Table 2 below during in-clinic confinement, and followed up during outpatient visits. Blood samples for PK analysis will be collected pre-dose and through post-dose (Day 8 (± 2 days)).

Refer to Table 2 for the Schedule of Procedures.

Table 2 Schedule of Procedures

	Screen		Subjects Resident in Clinical Research Unit														FU Tel Call
Day	-28 to -1	-1	Day 1											2		3*	Day 8 ± 2
Hour			0	0.25	0.5	0.75	1	1.5	2	4	6	9	12	24	36	48	
Informed Consent	X																
Eligibility Assessment	X	X															
Demographics	X																
Admission to CRU		X															
Randomization			X														
Medical History	X	X															
Physical Exam[1]	X	X														X	
Height, Weight, BMI	X																
Weight		X															
Resting 12-lead ECG[2]	X	X	X					X					X	X		X	
Vital signs (BP, PR, RR, Oral Temp)	X	X	X	X	X	X	X	X	X		X			X		X	
Clinical laboratory tests[3]	X	X														X	
HIV, Hep B&C screen	X																
Drug/alcohol screen	X	X															
Pregnancy test[4]	X	X															
FSH and estradiol[5]	X																
Dose administration[6]			X														
PK blood samples			X		X		X	X	X	X	X	X	X	X	X	X	
Injection site assessment			X							X				X			
AE & ConMeds	X																
Discharge from CRU																X	

NOTE: all timed procedures are with respect to START of study drug infusion.

AE= adverse event; BP = blood pressure; con med = concomitant medications; CRU = clinical research unit; ECG = electrocardiogram; FU = Follow-up; HIV = human immunodeficiency virus; Hep = Hepatitis; PR = pulse rate; PK = pharmacokinetics; RR = respiratory rate.

*Day 3 is the completion visit. Assessments to be performed on Day 3 prior to discharge from the CRU or at early termination; physical exam, vital sign assessments, clinical laboratory tests and ECG.

[1]A full physical exam will be conducted at screening and an abbreviated physical exam will be conducted on Day -1 and Day 3.

[2]Pre-dose and 1.5 h post infusion start ECGs will be triplicate ECGs (Triplicate ECGs will be performed approximately 1 minute apart).

[3]Clinical laboratory samples include: hematology, chemistry, coagulation parameters PT/INR/aPTT, and urinalysis.

[4]For female subjects – Serum pregnancy test at screening and urine pregnancy test at admission, both must be negative to enroll in the study.

[5]FSH and estradiol levels will be assessed in postmenopausal females to confirm status.

[6]IV infusion will occur at approximately 8:00 am, following completion of a low-fat breakfast. Infusion will over an approximately 60 minute period. Infusion stop time will have a time window of ±5 minutes.

Time windows: Vital signs and ECGs: Pre-dose: within 60 min prior dosing; post-dose to ≤ 12 h: (±15 min); ≥ 24 h: (±30 min).

PK sampling: pre-dose (-60 min), < 2 h (±5 min), 2 to <24 h (±10 min), 24, 36 and 48 h (±15 min).

Injection site assessments: pre-dose (-60 min), ≤ 24 h (±30 min).

Randomization

Each subject who signs an informed consent form (ICF) upon Screening will be assigned a unique subject identification (ID) number. The subject ID which will remain unchanged throughout the study. In case of screening failure, the subject ID will not be reused. If eligible for entry into the study, each subject will be assigned a randomization number and receive the study treatment assigned to the corresponding randomization number. Randomization subject numbers are pre-assigned on Day 1 based on order of arrival at the clinic for admission to the study.

This is a double-blind study with regard to treatment (HF1K16 or placebo) at each dose level cohort. The following personnel will have access to the randomization scheme: unblinded statistician who provides the randomization scheme, pharmacy personnel preparing IP on site, and Bioanalytical personnel analyzing the PK samples (refer to the Section 7 regarding blinding of treatment by Bioanalytical team prior to sending for the PK parameters to be estimated). The Bioanalytical team will only provide drug-treated subject concentrations in a blinded fashion. The randomization scheme will be kept in a secure location until the end of the study.

For this study, subjects will be randomized with a 3:1 ratio for HF1K16 and placebo. The escalated doses will be defined according to the emerging safety and tolerability data (refer to the protocol for further information).

A four-digit randomization number will include the first digit for the cohort number and the last 3 digits for the subject number. If a subject becomes ineligible and needs to be replaced, the replacement subject will have the same treatment to maintain the 3:1 ratio of active to placebo for the cohort.

At the initiation of the study, the study site will be instructed on the method for breaking the blind. Blinding codes should be broken only in emergency situations for reasons of subject safety.

3.1 Sample size justification

No formal sample size calculation has been performed. Sample sizes for this study are chosen based on feasibility of providing adequate information at each dose level.

3.2 Data handling

General:

Summaries for continuous variables will include the descriptive statistics for number of subjects (n), mean (arithmetic and geometric), standard deviation (SD), minimum (min), median, and maximum (max), coefficient of variation (CV, arithmetic, and geometric). Summaries for categorical (discrete) variables will include the descriptive statistics frequency and percentage.

Conventions for presentation of numerical data:

Minimum and maximum values will be presented to the same number of decimal places as the electronic case report form (eCRF) data. Means and medians will be presented to one more

decimal place than the eCRF data. Standard deviations will be presented to two more decimal places than the eCRF data.

Baseline

For comparison against baseline (e.g., for vital signs), baseline is considered as the last available assessment or value on or before the first treatment in the study.

Drug Concentrations and PK Parameter:

Plasma concentration of tretinoin and 4-oxotretinoin, will be displayed in listings as received from the bioanalytical laboratory.

In general, individual concentrations and PK parameters and summary values will be displayed using 3 significant figures regardless of the decimal point position as follows:

(1) Values $\geq$ 0.0001 and < 1 will be reported to 4 decimal places (e.g., 0.0123).

(2) Values $\geq$ 1 and < 10 will be reported to 2 decimal places (e.g., 1.02).

(3) Values $\geq$ 10 and < 100 will be reported to 1 decimal places (e.g., 10.2).

(4) Values $\geq$ 100 and < 1000 will be reported to as a whole integer (e.g., 100).

(5) Values $\geq$ 1000 or equal to 0 will be reported as a whole integer (e.g., 1000).

Values for T_{max} will be reported to 2 significant places.

3.3 Study Subjects

Subject disposition

Eligibility Status for the study will be listed for all subjects enrolled in the study.

Subject disposition will be summarized using number and percent of subjects who complete the study. Subject disposition and completion status will be listed for all enrolled subjects.

Protocol deviations

Key protocol deviations will be listed by subject and treatment.

Protocol deviations will be identified prior to database lock and may include but are not limited to significant violations of inclusion/exclusion criteria, noncompliance of the trial treatment taken, use of prohibited medications and not following clinical trial protocol procedures that may affect evaluation of the PK statistical analysis. Further programmatic edit checks on the time points and allowed windows may be conducted depending on the data.

4 Statistical Analysis Populations

4.1 Randomized Population:

All enrolled participants assigned a random number.

4.2 Safety Population

All randomized subjects who have received at least one dose of study drug.

4.3 Pharmacokinetic (PK) Population:

All subjects who receive active drug, have no major protocol violations, and have sufficient PK data to obtain reliable estimates of the key PK variables. Subjects in the PK population will be referred to as evaluable subjects.

4.4 Subject Demographics/other Baseline Characteristics

Demographic and baseline characteristics will be summarized for the Randomized Population. The demographic and baseline characteristics will consist of age, gender, race, ethnicity, height (cm), weight (kg), and BMI (kg/m^2). Demographic and baseline characteristics will be summarized by treatment. Demographic and baseline characteristics will be listed as well.

The age is a calculated parameter. Age at baseline of the entire study will be calculated using the subject's date of birth and the subject's informed consent date.

Continuous variables (e.g., age, height, weight, BMI) will be summarized by n, mean, standard deviation (SD), minimum, median, and maximum. Frequencies and percentages will be used to describe categorical (discrete) variables (e.g., gender, race, and ethnicity).

4.5 General Medical History and Non-Drug Therapies

The presence/absence of any current medical condition and/or other significant medical history will be coded using Medical Dictionary for Regulatory Activities (MedDRA) Version 24 or higher. Any non-drug therapies that are incurred during the course of the study will also be coded using MedDRA, as appropriate. Medical history will be listed by treatment and subject. Non-drug therapies will be listed by subject (unless these are not present in the database).

4.6 Prior and Concomitant Medications

All prescription medications taken in the 30 days prior to dosing or during the study period will be documented in the subject's eCRF.

Prior and concomitant medications will be coded using the current version of WHO Drug Enhanced Dictionary, version March 2021 or later, and listed by reported term and Anatomical Therapeutic Class classification. Medications will be listed by treatment and subject including the start and end dates and times, dose, dose unit, route, frequency, dose form, and indication.

5 Pharmacokinetic Analysis

Plasma samples for the PK analysis of HF1K16 (free tretinoin and liposome encapsulated tretinoin) will be collected relative to Day 1 dosing.

Blood samples for plasma tretinoin and 4-oxotretinoin PK analysis will be collected within 60 minutes prior to start of infusion, and 0.5, 1, 1.5, 2, 4, 6, 9, 12, 24, 36, and 48 hours relative to start of infusion.

A deviation within allowable time windows will not be considered as a protocol deviation as long as the actual sampling time is recorded, refer to the following Table 3 for allowed time window deviations.

Table 3 Plasma Concentration Time Point Deviations

Time Points	Deviation Window
Pre-dose, pre-infusion	Within 60 minutes prior to dosing
< 2h post-dose	± 5 minutes
2 hours to ≤ 24 hours	± 10 minutes
24, 36, and 48 hours	± 15 minutes

5.1 Plasma Concentrations

Plasma tretinoin and 4-oxotretinoin concentrations will be expressed in ng/mL, listed at each actual timepoint by subject and treatment, and summarized by treatment at each nominal timepoint using descriptive statistics (arithmetic mean, SD, coefficient of variation (CV%), geometric mean, median, min and max values and geometric CV%). All values below the quantification limit (BQL) will be labeled as such in the concentration listings and treated as zero for the concentration data summary statistics.

Linear and semi-log scale plots of mean concentration levels of plasma tretinoin and 4-oxotretinoin and individual subject concentrations versus time will be presented for the PK Population.

5.2 Pharmacokinetic Parameters

All PK calculations will be performed using Phoenix WinNonlin Version 8.1 (Certara USA, Inc., Princeton, USA). Plasma PK parameters for tretinoin and 4-oxotretinoin will be calculated with the IV infusion model non-compartmental analysis (NCA) with actual times of blood sample collection.

For PK parameter calculations, concentrations below BQL will be set to zero if they occur prior to the first or after the last quantifiable concentration and will be set to missing for data points in between.

Pharmacokinetic (PK) parameters AUC_{last}, AUC_{inf}, C_{max}, T_{max}, MRT, K_{el}, $t_{1/2}$, CL/F, and V_zF for tretinoin and 4-oxotretinoin will be listed by treatment, subject and summarized by treatment with descriptive statistics as listed above for concentration data for the PK Population.

AUC_{last} will be estimated using the linear trapezoidal method from time 0 hour to time t. AUC_{inf} will be calculated as the sum of AUC_{last} and $AUC_{t\text{-}inf}$, where t is the time corresponding to the last measurable serum concentration. $AUC_{t\text{-}inf}$ will be calculated as C_t/ λ_z, where C_t is the last measurable serum concentration and λ_z is the terminal phase rate constant. The value of λ_z will

be estimated by least-squares regression of the terminal log-linear serum concentration-time profile.

No value of K_{el}, AUC_{inf}, $t½$, V_zF, or CL/F will be reported for cases that do not exhibit a terminal log-linear phase in the concentration versus time profile with at least three timepoints. If the adjusted R-squared value (Rsq_adjusted) is < 0.80, the PK analyst may evaluate if other time points are more appropriate for the calculation of λ_z, otherwise no value of K_{el}, AUC_{inf}, $t_{1/2}$, V_zF and CL/F will be reported.

5.3 Dose Proportionality

For the dose cohorts Up to 3 mg/m^2, 6 mg/m^2, 10 mg/m^2, and 13 mg/m^2, a dose proportionality regression assessment for log-transformed C_{max}, $AUC_{0\text{-last}}$, and $AUC_{0\text{-inf}}$ and log-transformed dose will be analyzed using a power model[1,2]: ln (PK parameter) = $\mu + \beta$ x ln(dose) will be conducted for the free tretinoin and liposome encapsulated tretinoin. The 90% confidence intervals (CI) around the slopes (β) from each of these regression analyses will be obtained from the model and presented. Plots of the log-PK parameter by log-dose (AUC_{last}, AUC_{inf}, and C_{max}) will be produced for the dose cohorts for the PK Population.

6 Safety analysis

All summaries of safety data will be based on the Safety Population. Adverse events, 12-lead ECGs, vital signs (BP, respiratory rate, pulse rate, and oral body temperature), and safety laboratory (hematology, clinical chemistry, coagulation, and urinalysis) data will be reviewed and summarized on an ongoing basis during the study to evaluate the safety of subjects. Any clinical laboratory, ECG, BP, respiration, or pulse rate abnormalities of potential clinical concern will be described.

Medical history and physical examination information, as applicable, collected during the study will be captured for inclusion into the study database, unless otherwise noted. Any untoward findings identified on physical examination conducted after the administration of the first dose of study drug will be captured as an AE, if those findings meet the definition of an adverse event. Data collected at screening will be included in the study database. Physical examination (PE) will be performed at the visits as indicated in Table 2 above. The assessment should include, but not be limited to, the following body systems: general appearance and evaluation of dermatological system, head, eyes, ears/nose/throat, neck, lymph nodes, lungs, heart, abdomen, neurological, skin and musculoskeletal system. An abbreviated PE will include: heart, lungs, abdomen, skin and neuromuscular system. A symptom-directed PE may be performed at any time.

Safety evaluations will be based on the incidence, severity, and relatedness (Related, Probably Related, Probably Not Related, Not Related) of AEs and changes in subjects' physical examination findings, ECGs, vital signs, and clinical laboratory results. In the analysis of change from baseline for clinical laboratory parameters, 12 lead ECGs, and vital sign parameters, baseline is defined as the last value on or before the first dose of study drug.

6.1 Study product exposure

Exposure to study drug will be listed by subject and treatment, indicating dose date and time. Any deviations will be documented.

6.2 Adverse Events

All AEs (serious and non-serious) will be collected from the start of signing the ICF and until 30 days after the administration of the study drug. All reported AEs will be coded using Medical Dictionary for Regulatory Activities Version 24.0 or higher. All AEs will be graded using National Cancer Institute Common Terminology Criteria for Adverse Events (NCI CTCAE v5.0) refer to the protocol for further information. The causal relationship to study drug will be specified as Related, Probably Related, Probably not related, Not Related. For serious adverse events (SAE) that occur at any time following post dosing or end of the study and there is a reasonable possibility that the SAE is related to the study drug by the Investigator it must be promptly reported to the Sponsor. The incidence of treatment-emergent AEs (TEAE) – events with onset dates on or after the first dose date of the study drug and up to 30 days following the last dose of study drug – will be included in incidence tables. Events with missing onset dates will be considered treatment-emergent. All AEs will be summarized by severity and by relationship to study treatment. If a subject experiences more than 1 occurrence of the same AE (preferred term), the occurrence with the greatest severity and with association (Relationship to study treatment = 'Related' or 'Probably Related') to the study treatment will be used in the summary tables. Serious adverse events and AEs causing discontinuation will be tabulated. Treatment related SAEs and AEs causing discontinuation will also be summarized. All deaths during the study will be summarized and listed. All AEs will be listed by subject, System Organ Class (SOC) and Preferred Term (PT), along with information regarding onset, duration, CTCAE grade v5, and relationship to study treatment, action taken with study treatment, nature of event, and outcome. (Refer to the protocol for further information.) All tabulations will be presented by treatment. All AEs will be listed by subject and by treatment.

The injection site assessments will be assessed using a visual irritation scale (0-7): 0 = no evidence of irritation, 1 = minimal erythema, barely perceptible, 2 = definite erythema, readily visible; minimal edema or minimal papular response, 3 = erythema and papules, 4 = definite edema, 5 = erythema, edema and papules, 6 = vesicular eruption, or 7 = strong reaction spreading beyond application site. These will be listed along with other AEs and will be listed separately.

6.3 Clinical laboratory assessments

Clinical laboratory test parameters, with associated reference ranges provided by the laboratory, will be listed for individual subjects. Clinical laboratory test results outside the laboratory's reference ranges will be flagged with "L" for low and "H" for high and Investigator's assessment of clinical significance will be noted. Summary statistics at the visit and change from baseline, including n, mean, standard deviation (SD), minimum, maximum, and median values, will be

summarized for each quantitative laboratory parameter by treatment. Refer to Table 4Table 4 Clinical Laboratory Tests below for the laboratory tests performed and refer to the Schedule of Procedures (Table 2) for the timing of the tests.

Table 4 Clinical Laboratory Tests

Clinical Chemistry	**Hematology**	**Coagulation**	**Urinalysis**
Blood urea nitrogen (BUN) Creatinine Alkaline phosphatase (ALP) Aspartate transaminase (AST) Alanine transaminase (ALT) eGFR Gamma-glutamyl transferase (GGT) Bilirubin (total, direct, and indirect) Lactic dehydrogenase (LDH) Glucose Ferritin Albumin Total protein Bicarbonate Phosphate Sodium Magnesium Potassium Chloride Calcium Total Cholesterol Triglycerides HDL-C LDL-C Urate Uric acid Total Cholesterol Magnesium	Hemoglobin (Hgb) Hematocrit (Hct) Platelet count Red blood cell count White blood cell count with Differential	Prothrombin time (PT) Activated partial thromboplastin time (APTT) International normalized ratio (INR)	Specific Gravity pH Protein Glucose Ketones Bilirubin Blood Nitrites Leukocytes Urobilinogen Microscopic urine analysis
Urine/Saliva Drug Screen	**Serology Screen**		
Amphetamines, barbiturates, Cannabinoids, Cocaine metabolites, Opiates, Benzodiazepines, Marijuana, Methylenedioxymethamphetamine, Oxycodone, Phencyclidine, Tricyclic Antidepressants, Buprenorphine, Methamphetamine, Methadone, Alcohol breath test	Human immunodeficiency virus (HIV), Hepatitis B surface antigen (HBsAg) Hepatitis C antibody (IgG) **Other:** Serum pregnancy test (females, screening only) Urine pregnancy test (females, admission only) FSH, Estradiol (postmenopausal females, screening only)		

6.4 Vital signs assessments

Vital signs, including BP, respiratory rate, pulse rate, and oral temperature will be measured at times specified in the Schedule of Procedures (refer to above Table 2). Additional collection times or changes to these times will be permitted per the Investigator. All results will be listed by subject and treatment. Vital signs will be summarized using descriptive statistics including mean values and mean change from baseline values at each time point.

6.5 Physical examinations

Refer to the Table 2**Error! Reference source not found.**Schedule of Procedures for the times performed. Physical examination (PE) results will be listed by treatment and subject.

6.6 12-Lead ECG

To ensure safety of the subjects, a qualified individual at the investigative site will make comparisons to baseline measurements. For the time points of collection over the study, refer to Table 2. For the pre-dose and post-start of infusion 1.5 hr time points the ECG parameters will be collected using triplicates. For any summary presentations, the median of the triplicate observations at each of the applicable time points for each subject will be used. For further information on the consistent position of the subject during the ECG evaluations and other safety related matters, refer to the protocol. For all ECGs, details of rhythm, ventricular rate, PR, RR, QRS, QTc, QTcF, QTcB and QT intervals and an overall evaluation will be recorded.

A listing of the 12-Lead ECG data will be presented by subject, and treatment.

7 Interim and Final Analyses

No formal interim analysis will be conducted in this study. However, the Sponsor study team will conduct an ongoing blinded review of safety and PK data during the study for safety/PK assessment and to facilitate dose-escalation decisions.

8 Statistical programming and deliverables

All statistical analyses, tables and listings will be generated in SAS (version 9.4 or later) with appropriate documentation and programming validation. The table of contents for all tables, listings, and figures will be presented in a Tables, Listings and Figures shell supplemental document.

9 Changes from pre-specified analyses

There are no changes from the planned analyses for this study.

10 Changes to the planned analyses

Any deviation(s) of consequence from the statistical analysis plan (SAP) during the data analysis will be documented and justified in an amended SAP and/or in the final report or addressed in a separate document, as appropriate.

11 References

1. Gough K, Hutchinson M, Keene O, Byrom B, Ellis S, Lacey L, et al. Assessment of Dose Proportionality: Report from the statisticians in the pharmaceutical industry/pharmacokinetic UK Joint Working Party. Drug Info. J. 1995; 29:1039-1048.
2. Hummel J, McKendrick S, Brindley C and French, R. Exploratory assessment of dose proportionality: review of current approaches and proposal for a practical criterion. Pharmaceut. Statist. 2009; 8:38-49.

12 Revision history

Version	Date	Comments