Title of study: A Randomized, Double-blind, Placebo-controlled, First-in-human Study Designed to Evaluate the Safety, Tolerability, and Pharmacokinetics of EIDD-2801 Following Oral Administration to Healthy Volunteers

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A Randomized, Double-Blind, Placebo-Controlled, First-in-Human Study Designed to Evaluate the Safety, Tolerability, and Pharmacokinetics of EIDD-2801 Following Oral Administration to Healthy Volunteers

(EIDD-2801-1001-UK)

Protocol Version	1.1 (United Kingdom)
Version Date	02 April 2020

Sponsor Team:

Ridgeback Biotherapeutics	3162 Commodore Plaza, Suite 3E Miami, FL 33133-5815 United States
Medical Officer	[redacted]
EudraCT Number	2020-001407-17

STATEMENT OF COMPLIANCE

The study will be carried out in accordance with Good Clinical Practice (GCP) as required by the following:

- Consensus ethical principles derived from international guidelines including the Declaration of Helsinki and Council for International Organizations of Medical Sciences International Ethical Guidelines.
- Applicable International Conference on Harmonisation (ICH) GCP Guidelines.
- United States Code of Federal Regulations (CFR) applicable to clinical studies (45 CFR Part 46; 21 CFR Part 50, 21 CFR Part 56, and 21 CFR Part 312).
- Applicable laws and regulations.

SIGNATURE PAGE

The signature below constitutes the approval of this protocol and the attachments and provides the necessary assurances that this trial will be conducted according to all stipulations of the protocol, including all statements regarding confidentiality, and according to local legal and regulatory requirements and applicable regulations and ICH guidelines.

Principal Investigator:

Signed: [redacted] Date: 06 APR 2020

Name

Title EXECUTIVE MEDICAL DIRECTOR

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ABBREVIATIONS

AE	adverse event
AGP	alpha1-acid glycoprotein
ALT/SGPT	alanine aminotransferase/serum glutamic pyruvic transaminase
AST/SGOT	aspartate aminotransferase/serum glutamic oxaloacetic transaminase
AUC	area under the curve
BID	twice-daily
BMI	body mass index
CDC	Centers for Disease Control and Prevention
CEM	a cell line of lymphoblastic cells derived from a child with leukemia
CFR	Code of Federal Regulations
CHKV	chikungunya virus
CI	confidence interval
CoV	coronavirus
CRF	case report form
CTP	cytosine triphosphate cytidine triphosphate
CYP	cytochrome P450
DMSO	dimethylsulfoxide
DRF	dose range finding
DSS	Drug Safety Services
EBOV	Ebola virus
EC	Ethics Committee
EC_{50}	half maximal effective concentration
ECGs	electrocardiogram
EEEV	eastern equine encephalitis virus
EIDD	Emory Institute for Drug Development
EOS	end of study
FDA	Food and Drug Administration
FE	food-effect
FIH	first-in- human
FSFV	first-subject-first-visit
FSH	follicle stimulating hormone
GCP	Good Clinical Practice
GGT	gamma glutamyl transferase
GI	gastrointestinal
GLP	good laboratory practice

HAE	human airway epithelium
HBV	hepatitis b virus
HCV	hepatitis c virus
HED	human equivalent dose
HIV	human immunodeficiency virus
HLGT	high level group term
HPMC	hydroxypropyl methylcellulose
HSA	human serum albumin
IAV	influenza a virus
IB	Investigator's Brochure
IBV	influenza b virus
IC_{50}	half maximal inhibitory concentration
ICF	informed consent form
ICH	International Council for Harmonisation
IND	Investigational New Drug
LDH	lactate dehydrogenase
LSLV	last-subject-last-visit
MAD	multiple ascending dose
MCH	mean corpuscular hemoglobin
MCHC	mean corpuscular hemoglobin concentration
MCV	mean cell volume
MDCK	Madin-Darby Canine Kidney Cells
MERS	Middle East Respiratory Syndrome
MRSD	maximum recommended starting dose
MTD	maximum tolerated dose
NOAEL	no-observed-adverse-effect-level
NSAID	nonsteroidal anti-inflammatory drug
OTC	over-the-counter
PBMC	peripheral blood mononuclear cells
PBO	placebo
PI	principal investigator
[redacted]	[redacted]
PK	pharmacokinetic
RDW	red cell distribution width
RNA	ribonucleic acid
RSV	respiratory syncytial virus

SAD	single ascending dose
SAE	serious adverse event
SAP	statistical analysis plan
SARS	Severe Acute Respiratory Syndrome
SI	selectivity index
SMP	safety management plan
SOP	standard operating procedure
SUSAR	suspected unexpected serious adverse event
UTP	uridine 5'-triphosphate
VEEV	Venezuelan equine encephalitis virus
ZIKV	Zika virus

PROTOCOL SYNOPSIS

Sponsor: Ridgeback Biotherapeutics	
Title: A Randomized, Double-Blind, Placebo-Controlled, First-in-Human Study Designed to Evaluate the Safety, Tolerability, and Pharmacokinetics of EIDD-2801 Following Oral Administration to Healthy Volunteers (EIDD-2801-1001-UK)	
Short Title: EIDD-2801-1001-UK	**Development Phase:** Phase 1
Description of Study Drugs, Dose and Mode of Administration: EIDD-2801 and matching placebo (PBO) will be supplied [redacted] **Part 1:** A single oral dose of EIDD-2801 or PBO will be administered to subjects enrolled in Part 1 (P1; single ascending dose [SAD]) cohorts. The starting dose in the first SAD cohort will be 50 mg. The doses will be administered in an escalating manner. Dose levels will not increase by more than 3-fold for predicted nonpharmacologically active dose levels and by 2-fold for predicted pharmacologically active dose levels. [redacted] **Part 2:** Subjects in Part 2 (P2)/food-effect (FE) will receive 2 single doses of EIDD-2801 with a 14-day (minimum) washout period between doses. The dose for the P2/FE cohort is planned to be 100 mg but may be adjusted based on safety, tolerability, and/or pharmacokinetic (PK) data from P1. In any case, the dose assessed in the P2/FE cohort will have been given previously to subjects in a P1/SAD cohort successfully (i.e., no halting rules were met following dosing).	
Treatment Duration: **Part 1:** Subjects enrolled into P1/SAD cohorts will receive a single dose of study drug (EIDD-2801 or matching PBO). **Part 2:** Subjects enrolled into the P2/FE cohort will receive 2 single doses of EIDD-2801, with a washout period between doses.	
Subject Duration: **Part 1:** The maximum possible study duration for participants enrolled in P1/SAD cohorts will be approximately 43 days. **Part 2:** The maximum possible study duration for participants enrolled in the P2/FE cohort will be approximately 58 days.	
Objectives and Endpoints: **Part 1: SAD Cohorts** Primary: • Objective: To determine the safety and tolerability of single ascending doses of EIDD-2801 o Endpoints: ▪ Results of safety evaluations including safety laboratory assessments, physical examination (PE), electrocardiograms (ECGs), vital signs, and adverse events (AEs).	

Secondary:

- Objective: To define PK of EIDD-2801 and EIDD-1931 in plasma and urine following single doses administered to healthy volunteers
    - Endpoints:
        - Plasma PK parameters, including C_{max}, t_{max}, $t_{1/2}$, CL/F, λz, Vz/F, $AUC_{0\text{-}inf}$ and AUC_{last} as appropriate
        - Urinary excretion of EIDD-2801 and EIDD-1931 following single-dose administration

Part 2: Single-Dose Food-Effect

Primary:

- Objective: To assess the effect of food on the PK of EIDD-2801 and EIDD-1931 following a single oral dose
    - Endpoints:
        - Plasma PK parameters, including C_{max}, t_{max}, $t_{1/2}$, CL/F, λz, Vz/F, $AUC_{0\text{-}inf}$ and AUC_{last} as appropriate
        - Urinary excretion of EIDD-2801 and EIDD-1931 following single-dose administration

Secondary:

- Objective: To determine the safety and tolerability of single doses of EIDD-2801.
    - Endpoints:
        - Results of safety evaluations, including safety laboratory assessments, PE, ECG, vital signs, and AEs

Population:

Part 1: P1/SAD cohorts will include 8 subjects each, 2 randomized to receive PBO and 6 randomized to receive EIDD-2801. Initially, 4 SAD cohorts are planned for P1/SAD with the option to add an additional 3 cohorts based on study results.

Part 2: Ten subjects will be enrolled into the P2/FE cohort; all subjects enrolled in the P2/FE cohort will receive EIDD-2801. One P2/FE cohort is planned for the study. If PK results obtained are equivocal, additional subjects may be enrolled into the P2/FE cohort at the previously tested dose or an additional P2/FE cohort may be added at a different dose. The site is strongly encouraged to ensure that women are represented in each cohort.

Inclusion and exclusion criteria for study participation are as follows:

Inclusion Criteria: subjects who meet all of the criteria at Screening will be eligible for study participation

1. Male or female between the ages of 18 and 60, inclusive.
2. Female subjects must be of non-childbearing potential. Non-childbearing potential is defined as women who:
    - are physiologically incapable of becoming pregnant including those who are surgically sterile, via hysterectomy, bilateral salpingectomy, and/or bilateral oophorectomy by reported medical history and/or medical records. Surgical sterilization to have occurred a minimum of 6 weeks, or at the Principal Investigator's (PI's; or designee) discretion, prior to Screening.

- are post-menopausal (at least 45 years of age with amenorrhea for 12 months without an alternative medical reason with confirmatory follicle stimulating hormone (FSH) levels of ≥ 40 mIU/mL. The amenorrhea should not be induced by a medical condition such as anorexia nervosa, hypothyroid disease or polycystic ovarian disease, or by extreme exercise. It should not be due to concomitant medications that may have induced the amenorrhea such as oral contraceptives, hormones, gonadotropin-releasing hormones, anti-estrogens, or selective estrogen receptor modulators.)

3. Male participants must agree to the use of effective contraception for study duration, i.e., from Check-in until 90 days following the last dose of study drug.
4. Is in generally good health as determined by medical history, PEs (at Screening and/or Check-in), vital signs, and other screening procedures.
5. Has a body mass index (BMI) of 18 to 30 kg/m^2.
6. Is willing and able to comply with all study procedures including providing informed consent.

Exclusion Criteria: subjects who meet any of the following criteria at Screening (unless otherwise stated) will not be eligible for participation

1. Females who are pregnant, planning to become pregnant, or breastfeeding.
2. Has a clinically relevant acute or chronic medical condition or disease of the cardiovascular, gastrointestinal, hepatic, renal, endocrine, pulmonary, neurologic, or dermatologic systems as determined by the PI (or designee).
3. Has any current or historical disease or disorder of the hematological system or significant liver disease or family history of bleeding/platelet disorders.
4. Has a history of cancer (other than basal cell or squamous cell cancer of the skin), rheumatologic disease or blood dyscrasias.
5. Has a history of gastrointestinal surgery or other condition that may interfere with absorption of study drug, in the option of the PI (or designee).
6. Has a history of febrile illness within the 14 days prior to the first dose of study drug.
7. Has a positive alcohol or drug screen at Screening or the Day -1 visit or has a history of alcohol or drug abuse within the past 5 years.
8. Currently uses tobacco, nicotine or tobacco products, e-cigarettes or has stopped using tobacco products within the past 3 months and/or has a positive cotinine test at Screening or Check-in.
9. Has any abnormal laboratory value of Grade 2 or higher, which is considered to be clinically significant by the PI (or designee).
10. Has a total white cell count or absolute lymphocyte count outside the normal range, or hemoglobin or platelet levels below the lower limit of normal, at Screening of Day -1.
11. Has values above the upper limit of normal for the following laboratory analytes: alanine aminotransferase (ALT/SGPT), alkaline phosphatase (serum), aspartate aminotransferase (AST/SGOT), at Screening or Day -1
12. Positive test result for human immunodeficiency virus (HIV), hepatitis B virus (HBV), or hepatitis C virus (HCV).
13. Has an autoimmune disease, is immunosuppressed or is in any way

immunocompromised.

14. Has any of the following:
    - QTcF >450 ms confirmed by repeat measurement
    - QRS duration >110 ms confirmed by repeat measurement
    - PR interval >220 ms confirmed by repeat measurement
    - findings which would make QTc measurements difficult or QTc data uninterpretable
    - History of additional risk factors for torsades de pointes (e.g., heart failure, hypokalemia, family history of long QT syndrome).
15. Except as noted, has used prescription drugs (other than hormone replacement therapy) within 14 days prior to the first dose of study drug unless, in the opinion of the PI (or designee), the drug will not interfere with study assessments. The following prescription drugs will be allowed:
    - Statins: Subjects who have been on a stable dose of statins for a minimum of 6 months and who have normal creatine kinase at Screening.
    - Antihypertensives: Subjects who have been on a stable dose of antihypertensives for a minimum of 6 months and have controlled blood pressure and no active heart disease.
16. Has received an experimental agent (vaccine, drug, biologic, device, blood product or medication) within 3 months prior to the first dose of study drug and agrees not to receive another experimental agent during the duration of this trial.
17. Has donated blood within 30 days, plasma within 2 weeks, or platelets within 6 weeks prior to Screening or refuses to agree to not donate blood until 3 months after the End of Study (EOS) visit.
18. Uses over-the-counter (OTC) medications or nutritional supplements (including, e.g., gingko biloba, aspirin at any dose, and nonsteroidal anti-inflammatory drugs [NSAIDs]) on a routine/scheduled basis, and/or is unwilling to abstain from such use for the duration of the study.
19. Is unwilling to abstain from quinine containing products including quinine water, tonic water, bitter lemon, jello shots, any quinine preparations or Cinchona tree bark preparations (e.g., herbal medications containing Cinchona tree bark).
20. Has any condition that would, in the opinion of the PI (or designee), put the subject at increased risk for participation in a clinical trial or confinement in a clinical research unit.

Retesting for Inclusion/Exclusion Criteria: In the case where a parameter is outside of the acceptable limits for enrollment, measurement of the parameter can be repeated one time if both the following are met:

- In the opinion of the PI (or designee), the parameter value does not represent a chronic condition that otherwise will preclude the volunteer from enrolling in the study; and
- In the opinion of the PI (or designee), the abnormality is not likely to recur.

General Investigational Plan: For all potential subjects, volunteers who express interest in the study will report to the clinic for informed consent. The study will be explained to the subject

and the Ethics Committee (EC)-approved Informed Consent Form (ICF) will be presented. The subject will be given the chance to review the document and ask any questions he/she may have. If, after reviewing the consent, the subject would like to participate in the study, then he/she will sign the ICF and begin screening for study entry; those satisfying all criteria will be enrolled into the study and admitted to the clinic on Day -1. Retesting will be allowed as described above.

Part 1: For P1/SAD, the first cohort will be divided into 2 groups. The first group, a sentinel group, will include 2 subjects.

On Day 1, one subject in the sentinel group will be administered PBO and one will be administered EIDD-2801. Analysis of the safety results for the sentinel group will be reviewed 24 hours postdose. If no halting criteria have been met, the remainder of the subjects in Cohort 1 (the second group) will be dosed. There will be no sentinel groups for the remaining cohorts.

Following dosing on Day 1, subjects will remain in the clinic through completion of study assessments on Day 4, returning to the clinic on Day 9 for study assessments, and Day 15 for the EOS visit.

Doses will be administered in an escalating manner following satisfactory review by the Sponsor and PI of the blinded safety and tolerability data (up to 72 hours post final dose). Dose-escalation will only occur if data from a minimum of 6 subjects have been reviewed from the previous lower dose group, such that data from a minimum of 4 subjects who have received EIDD-2801 will be used to make the dose-escalation decision. Dose levels will not increase by more than 3-fold for predicted nonpharmacologically active dose levels and by 2-fold for predicted pharmacologically active dose levels. The highest dose to be studied under this protocol will not exceed 200 mg.

Part 2: For P2/FE, one cohort assessing the effect of food on EIDD-2801 and EIDD-1931 PK parameters will be enrolled; it is planned that the FE cohort will be at a dose of 100 mg EIDD-2801, although higher or lower doses may be selected based on safety and available PK data from P1.

In addition to assessing the effect of food on dosing, peripheral blood mononuclear cells (PBMCs) will be collected from subjects following each dose; the PBMCs will be stored for analysis of EIDD-2061 levels at some point in the future.

The 10 subjects enrolled in the FE cohort will all receive EIDD-2801; subjects will be randomized to treatment sequence; i.e., to receive drug in the fed then fasted state (Sequence 1) versus fasted then fed state (Sequence 2). Half of the subjects in the cohort will be randomized to Sequence 1 and the other half will be randomized to Sequence 2.

Following dosing on Day 1, subjects will remain in the clinic through completion of study assessments on Day 4, returning to the clinic on Day 9 for study assessments, and Day 14 to begin the second dosing sequence on Day 15. Subjects will then be discharged from the clinic on Day 18, returning to the clinic on Day 23 for study assessments, and Day 30 for the EOS visit. There will be a 14-day (minimum) washout period between doses.**Additional parts may be added to this study, including a SAD part in elderly healthy subjects, a multiple-ascending dose (MAD) part in healthy subjects, and a study of the safety, tolerability, and PK in Japanese subjects. These additional parts are not described in this protocol but may be added via a protocol amendment. It is planned that the MAD part will include 4 dose-escalation cohorts (with the option to include an additional 3 cohorts), with subjects receiving BID doses for 7 days. It is planned that each MAD cohort will comprise 8**

subjects, with 6 subjects receiving EIDD-2801 and 2 subjects randomized to PBO.
Safety Monitoring and Potential Unblinding: Safety for this study will be continually monitored by the PI and Sponsor.
Blinding: All study personnel will remain blinded to treatment assignment (i.e., EIDD-2801 or PBO) in P1, except for personnel at the bioanalytical laboratory, and the unblinded pharmacy staff and pharmacokineticist. If unblinding is required to manage subject safety or to support dose-escalation decisions, the decision to unblind lies solely with the PI. If possible and providing that it does not interfere with or delay any decision in the best interest of the subject, the PI will discuss the intended code-break with the Sponsor.
Dose-Escalation Halting Rules: For Group 1 in P1/SAD, sentinel dosing will occur whereby 2 subjects (1 active and 1 PBO) will be dosed on 1 day and, providing no safety concerns arise, the remaining 6 subjects will be dosed after at least 24 hours. The study will be halted if one or more subjects experience a serious adverse event (SAE) that is considered to be related to EIDD-2801 or 2 or more subjects in the same group experience severe AEs that are considered to be related to EIDD-2801. Dose-escalation will stop if clinically relevant signs or symptoms of similar nature occur in 2 or more subjects in a group that, in the opinion of the PI, warrant stopping of dose-escalation.
Data Monitoring, Safety Reporting and Unblinding: Data will be monitored throughout the course of the study by experienced clinical monitors according to the clinical monitoring plan. As data will be entered into an electronic case report form (CRF), data will be monitored to ensure accuracy as well as adherence to data entry instructions provided in the CRF guidelines. Procedures for reporting any SAE will be detailed in the protocol and forms and instructions provided to the site.
Statistical Considerations: A complete description of all statistical analyses and methods will be presented in the Statistical Analysis Plan (SAP). The SAP will be reviewed and approved by the Sponsor and will be finalized prior to database lock. Plans for PK analyses will be included in the SAP. **Determination of Sample Size:** The sample sizes for the P1/SAD cohorts are typical for a Phase 1 first in human (FIH) study. The sample size for the P2/FE cohort is in accordance with FDA guidelines for sample size in food-effect studies.

Study Populations:

Safety Population: All subjects who receive at least one dose of study drug.

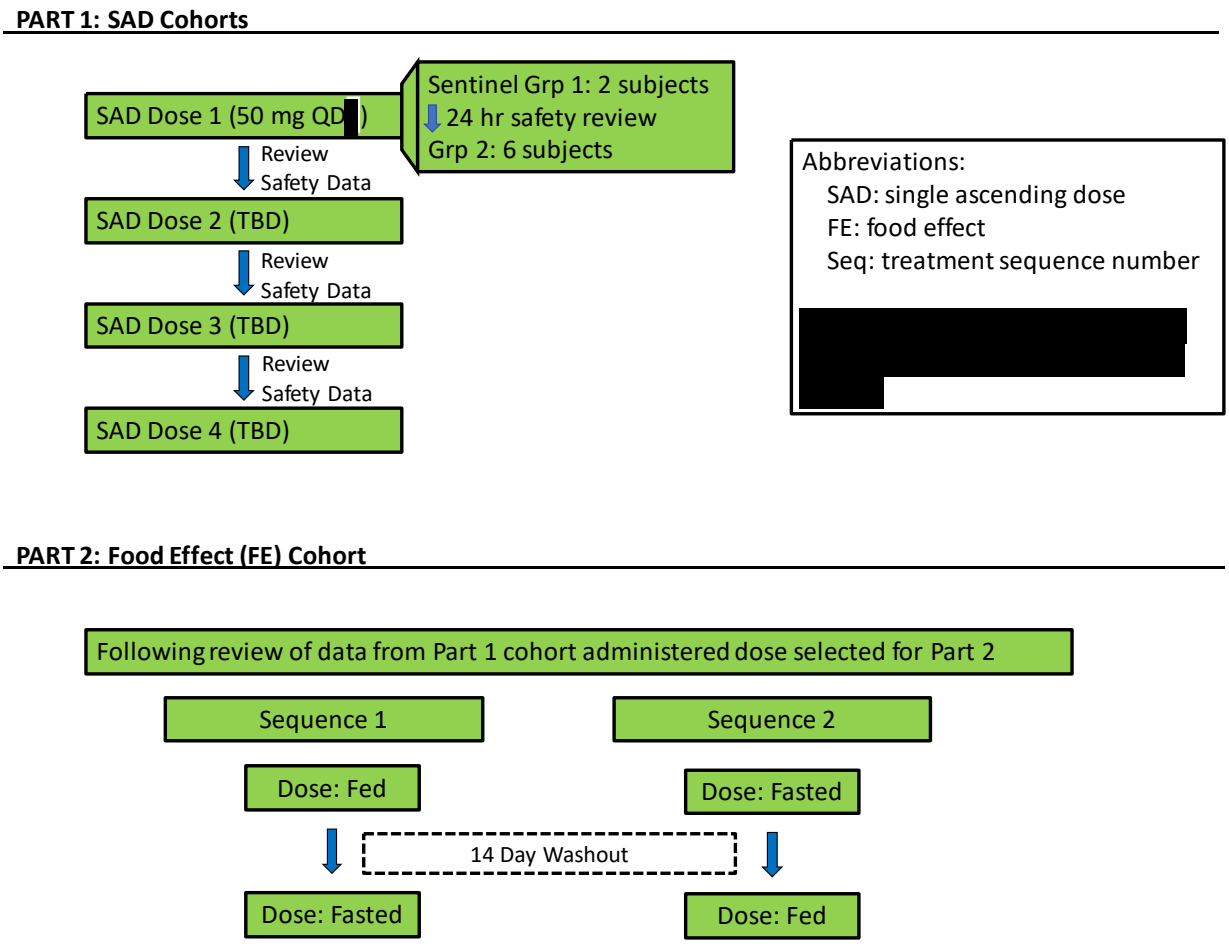
Pharmacokinetic Population: All subjects receiving study drug who comply sufficiently with protocol requirements (i.e., no major protocol deviations) and for whom a sufficient number of analyzable PK samples have been obtained to permit the determination of the PK profile for EIDD-2801/EIDD-1931 and the statistical analysis of the results.

Safety Analyses: Statistical methods for the safety analyses will be descriptive in nature. Safety data, including AEs, clinical laboratory data, vital signs, ECG parameters, and PEs. All appropriate AEs will be graded using the DMID toxicity scale (March 2014). Change from baseline will be included in summary tables for laboratory parameters. All laboratory data will be included in the data listings and all test values outside the normal range will be flagged.

Pharmacokinetic Analyses: Plasma PK parameters for each dose level will be calculated from the concentrations of EIDD-2801 and EIDD-1931 measured in predose and postdose plasma samples. For each dose level, descriptive statistics will be presented. Figures will be created to display mean and individual subject EIDD-2801 and EIDD-1931 concentration versus time. Urine PK parameters will be calculated whenever possible for each subject based on the urine concentrations of EIDD-2801 and EIDD-1931. The following PK parameters will be calculated:

Plasma PK Parameter	**Description**
AUC_{last}	The area under the plasma concentration-time curve, from time 0 to the last measurable non-zero concentration, as calculated by the linear up/log down trapezoidal method.
$AUC_{0\text{-}inf}$	The area under the plasma concentration-time curve from time 0 extrapolated to infinity. $AUC_{0\text{-}inf}$ is calculated as the sum of AUC_{last} plus the ratio of the last measurable plasma concentration to the elimination rate constant (λz).
C_{max}	Maximum observed concentration.
t_{max}	Time to reach C_{max}. If the maximum value occurs at more than one time point, t_{max} is defined as the first time point with this value.
λz	Apparent terminal elimination rate constant; represents the fraction of medication eliminated per unit time.
t½	Apparent terminal elimination half-life of medication, calculated as 0.693/λz.
Vz/F	Apparent volume of distribution (EIDD-2801 only).
CL/F	Apparent oral drug clearance (EIDD-2801 only).
Urine PK Parameter	**Description**
A_e	Amount excreted in urine over the sampling interval.
F_e	Fraction of drug excreted in the urine over sampling interval.
CL_R	Renal clearance, $CL_R = A_e/AUC$ where both A_e and AUC are determined over co-incident time ranges after dosing.
Dose proportionality for PK parameters of EIDD-2801 and EIDD-1931 ($AUC_{0\text{-}inf}$, AUC, and C_{max}) will be assessed by the power model. The mean slope and the associated 90% confidence interval (CI) based on the power model will be reported. To assess the effect of food on the PK of EIDD-2801 and EIDD-1931, natural log transformed PK parameters from P2 ($AUC_{0\text{-}inf}$, AUC_{last}, and C_{max}) will be analyzed using a mixed effects model with fixed effect terms for treatment (with or without food), period, and sequence and with subject as a random effect. Point estimates and their associated 90% CIs for the natural log transformed PK parameters for the difference between EIDD-2801 administered under fed relative to fasted conditions will be constructed. The point estimates and interval estimates will be back transformed to give estimates of the ratio and 90% CI for the geometric least squares mean ratio for fed/fasted to assess the effect of food.	
Interim Analyses: No formal interim analyses are planned for this study. Data from P1 cohorts may be locked, unblinded, and analyzed prior to conducting P2.	

Figure 1: Study Schema



1. INTRODUCTION

This study is a FIH study designed to assess the safety, tolerability and PK of EIDD-2801 in healthy human volunteers. EIDD-2801 is a ribonucleoside analog with broad-spectrum activity against many RNA viruses. It is currently being developed by Ridgeback Biotherapeutics as a treatment of infections caused by highly pathogenic coronaviruses (CoV), including COVID-19. In addition, EIDD-2801 is being developed in parallel as a treatment of uncomplicated influenza caused by all subtypes of circulating and emerging (drifted and shifted) influenza A virus (IAV) and influenza B virus (IBV), including seasonal, epidemic and pandemic strains.

1.1. Background

EIDD-2801 is the 5'-isopropyl ester prodrug of the broadly active, direct-acting antiviral ribonucleoside analog EIDD-1931. After oral delivery, the prodrug (EIDD-2801) is rapidly hydrolyzed by circulating esterases to produce high circulating (plasma) levels of EIDD-1931. In cell culture systems, EIDD-1931 has been shown to inhibit replication of multiple viral pathogens from multiple RNA virus families including pathogenic CoV (e.g., Middle East respiratory syndrome [MERS], severe acute respiratory syndrome [SARS]-CoV-1 and SARS-CoV-2), influenza viruses (seasonal, pandemic, and avian subtypes), respiratory syncytial virus (RSV), alphaviruses (e.g., Eastern equine encephalitis virus [EEEV], Venezuelan equine encephalitis virus [VEEV], and chikungunya virus [CHKV]), Filoviruses (e.g., Ebola virus [EBOV]), and Zika virus (ZIKV). In addition, EIDD-2801 is active against orthopoxviruses (tested against vaccinia virus) probably because orthopoxviruses encode their own unique RNA polymerase. [redacted] The primary mechanism of action of EIDD-2801 is inhibition of viral RNA replication by incorporation of the EIDD-1931 monophosphate metabolite into the viral RNA genome resulting in induction of viral error catastrophe.

1.2. Rationale for Development

EIDD-2801 is being developed for the treatment of infections caused by RNA viruses, specifically for COVID-19 and other CoV infections, influenza, and VEEV. The FIH study is being conducted under an FDA Investigational New Drug (IND) for the treatment of uncomplicated influenza caused by all subtypes of circulating and emerging IAV and IBV, and under a Clinical Trial Application for infections with COVID-19. During conduct of the FIH study, the Sponsor intends to submit an IND for the treatment of COVID-19 patients in a MAD trial to help define a dose that may be efficacious in treating SARS-CoV-2 and MERS-CoV infections.

EIDD-2801 has a unique dual mechanism of action against RNA viruses, including COVID-19 and other CoV infections. The compound acts as a competitive, alternative substrate for the virally encoded RNA-dependent RNA-polymerase that upon incorporation into nascent chain RNA induces increased mutational frequency in the viral genome. Incorporation quickly results in the production of non-viable virus. Additionally, the active metabolite EIDD-1931-5'-triphosphate may act directly as a chain terminator and arrest replication by

exerting a next nucleoside effect. It is anticipated that the high barrier to resistance observed during in vitro passaging studies will translate to slow, if any, emergence of viral resistance. Resilience to viral escape is a distinguishing feature of EIDD-2801.

Currently, there is no approved antiviral therapeutic for the treatment of SARS-CoV-2. An antiviral drug is urgently needed. [redacted]

1.3. Nonclinical Overview

1.3.1. Mechanism of Action

The mechanism of antiviral activity of EIDD-2801 is "lethal mutagenesis"; a concept that is predicated on increasing the viral mutation rate beyond a biologically-tolerable threshold, resulting in impairment of viral fitness and leading to viral extinction.

The specifics of the mechanism are as follows. EIDD-2801 is rapidly taken up by cells and the 5'-isopropylester cleaved to liberate EIDD-1931, which is in turn phosphorylated to EIDD-2061 by host kinases (Hernandez-Santiago et al., 2004; Painter et al., 2019). The 5'-triphosphate, EIDD-2061, acts as a competitive alternative substrate for virally encoded RNA-directed RNA polymerases and EIDD-2061 is incorporated into nascent viral RNA. Owing to the ability of the N^4-hydroxycytosine base of EIDD-1931 to tautomerize, EIDD-2061 can pair with either guanosine or adenosine, and consequently can substitute for either CTP or UTP, respectively (Flavell et al., 1974). This results in an accumulation of mutations that increases with each cycle of viral replication. The process whereby the mutation rate is increased by exposure to a drug is referred to as Viral Decay Acceleration (Mullins et al., 2011) and results in viral ablation.

Significant work has gone into validating this mechanism of action for EIDD-2801/1931, and it has been shown for MERS-CoV, VEEV, and IAV that viruses grown in the presence of EIDD-1931 have significantly increased levels of transition mutations (Agostini et al., 2019; Toots et al., 2019; Urakova et al., 2018). Multi-log decreases in virus yields were observed after treatment with EIDD-1931. Additionally, it was demonstrated for VEEV that the infectivity of virions formed in the presence of EIDD-1931 decreases from ~20% to less than 0.2%, and that the infectious virions are significantly Impaired in their replication ability (Urakova et al., 2018). As a consequence of this mechanism of action, the generation of drug-resistant escape mutants is practically impossible. This same effect was demonstrated for CoV (Agostini et al., 2019) and influenza virus (Toots et al., 2019). Furthermore, given the unique mechanism of action, EIDD-2801 is expected to be active against viruses resistant to other antiviral agents which have a different mechanism of action. The only data generated to date regarding the activity of EIDD-1931 against viruses resistant to other nucleoside analogs found that EIDD-1931 was

active against CoV resistant to remdesivir in cell culture assays (T. Sheahan et al, preprint available at https://www.biorxiv.org/content/10.1101/2020.03.19.997890v1).

As an alternative or additional mechanism of action, it has been theorized that incorporation of EIDD-2061 into viral genomic RNA can change the thermodynamics of RNA secondary structure and thus decrease the efficiency of the promoter regions involved in RNA genome replication (Stuyver et al., 2003).

1.3.2. *In Vitro* Pharmacology

1.3.2.1. Antiviral Activity in Tissue Culture and in Human Airway Epithelium

The ribonucleoside analog EIDD-1931 is the parent of the prodrug EIDD-2801. EIDD-1931 shows specific antiviral activity in different tissue culture cells and in the differentiated organoid model of human airway epithelium (HAE) with a selectivity index (SI) ranging from 21 to >100 for all influenza viral isolates tested. It is active against IAV (pandemic and seasonal) and IBV strains, as well as against highly pathogenic H5N1 and H7N9 strains (Table 1).

Table 1: EIDD-1931 Antiviral Activity Against Influenza A and B Viruses in Tissue Culture and Primary Human Bronchial/Tracheal Epithelial Cells

Virus	Strain	Cell line	EC_{50}* (µM)	CC_{50} (µM)	SI	Reference
IAV H1N1	Ca/07/2009	MDCK	1.24	68	55	NIAID Antiviral Testing Program
IAV H1N1	WSN/33	MDCK	1.1	299.8	275	Yoon et al., 2018
IAV H1N2	WSN/33	primary hBTEC	5.4	-	-	Yoon et al., 2018
IAV H2N3	Perth/16/2009	MDCK	0.88	52	59	NIAID Antiviral Testing Program
IAV H2N3	Ohio/sw-10-132/2010	MDCK	3.2	299.8	94	Yoon et al., 2018
IAV H5N1	Duck/MN/1525/81	MDCK	1.28	27	21	NIAID Antiviral Testing Program
IAV H5N1	Vietnam/1203/2004	MDCK	0.14	299.8	2100	Yoon et al., 2018
IAV H7N9	Anhui/1/2013	MDCK	0.13	299.8	2300	Yoon et al., 2018
IBV	Florida/4/2006	MDCK	<0.4	76	>190	NIAID Antiviral Testing Program
IBV	Brisbane/60/08	MDCK	0.006	299.8	50000	Yoon et al., 2018
IAV H1N1	Ca/07/2009	HAE-3D*	0.08	50	625	Toots et al., 2019
IAV H1N1	WSN/33	HAE-3D*	0.08	50	625	Toots et al., 2019
IBV	Brisbane/60/08	HAE-3D*	0.06	50	833	Toots et al., 2019

* Human Airway Epithelium organoid model.

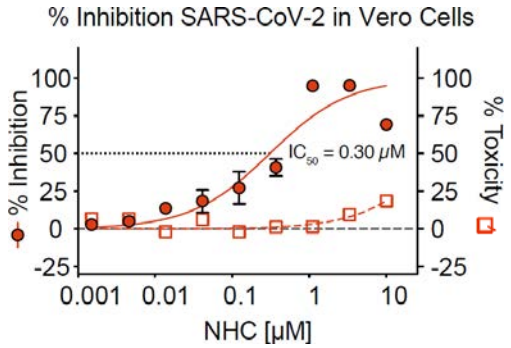
EIDD-1931 also showed specific antiviral activity against human SARS-CoV-1, MERS-CoV (Table 2) and SARS-CoV-2 (Figure 2), against togaviruses VEEV, EEEV and Chikungunya virus (Table 3) [redacted]

Table 2: EIDD-1931 Antiviral Activity Against SARS and MERS Coronaviruses in Tissue Culture and Primary Human Bronchial/Tracheal Epithelial Cells

Virus	Strain	Cell line	EC_{50} (µM)	CC_{50} (µM)	SI	Reference
SARS-CoV-1	Urbani	Vero76	<0.4	144	>360	NIAID Antiviral Testing Program
SARS-CoV-1	SARS-CoV-GFP(†)	HAE-3D(*)	<1	>100	>100	Tech. Report 25.038
MERS-CoV	GenBank Ac.No JX869059**	DBT-9	0.56	>200	>357	Agostini et al., 2019
MERS-CoV	Human ß-CoV C, Novel 2912	Vero E6	<0.8	20	>25	NIAID Antiviral Testing Program

* Human Airway Epithelium organoid model; ** cDNA Derived clone

Figure 2: Inhibition of SARS-CoV-2 by EIDD-1931



EIDD-1931 (NHC) antiviral activity (closed circles) and cytotoxicity (open squares) in Vero Cells infected with SARS- CoV-2. Vero cells were infected in duplicate with SARS-CoV- 2 clinical isolate virus at a multiplicity of infection (MOI) of 0.05 in the presence of a dose response of EIDD-2801 for 48 hours after which replication was measured through quantitation of cell viability by Cell-Titer-Glo assay. Cytotoxicity was measured in similarly treated but uninfected cultures. Reproduced from Sheahan et al 2020.

Table 3: EIDD-1931 Antiviral Activity Against Togaviruses in Tissue Culture

Virus	Strain	Cell line	EC_{50} (µM)	CC_{50} (µM)	SI	Reference
VEEV	TC-83	Vero	0.43	>200	>930	Urakova et al., 2018
VEEV	TC-83	Vero76	1.92	32	17	NIAID Antiviral Testing Program
EEEV	FL93-939	Vero76	1.08	84	78	NIAID Antiviral Testing Program
CHKV	S27 (VR-64)	Vero76	1.8	96	53	NIAID Antiviral Testing Program

[redacted]

1.3.2.2. Cytotoxicity of EIDD-1931 in Tissue Culture Utilizing Cells from Different Organs and Species

EIDD-1931 was tested for cytotoxicity in human hepatic origin Huh7 and HepG2 cells, in human lymphoid CEM, human pancreatic BxPC-3, human prostate cancer PC-3, human muscle A204, human lung A549, human epithelial hEp-2, rat heart muscle H9c2, monkey kidney Vero, and canine kidney MDCK cell lines (Table 5). The compound exhibits low cytotoxicity in the majority of cells tested (half maximal effective concentration [EC_{50}] values are in the range of 40 to >100 µM) except in lymphoid origin CEM cells where the compound shows a 7.5 µM EC_{50} value (Sticher et al., 2020; Urakova et al., 2018; Yoon et al., 2018).

Table 5: Cytotoxicity (CC50) of EIDD-1931 in Mammalian Cell Lines

Cell Line	CEM	HepG2	PC-3	A204	A549	BxPC-3	Huh-7	H9c-2	Vero	hEp-2	MDCK
CC50 (µM)	7.5	42.3	267.1	84	46	48	165.5	81	53	272.4	299.8

Sources: Sticher et al., 2020, Yoon et al., 2018

EIDD-2801 typically showed 2-4× lower activity and cytotoxicity than EIDD-1931 due to slightly slower uptake and anabolism in tissue culture.

1.3.2.3. Assessment of Mitochondrial Toxicity

Since EIDD-1931 is a nucleoside analog, additional investigations were performed to analyze whether observed cytotoxicity of EIDD-1931 is caused by mitochondrial toxicity. It was demonstrated that the prolonged treatment (14 days) with the compound does not result in selective killing of mitochondria or in mitochondrial dysfunction in CEM and HepG2 cells (Sticher et al., 2020).

1.3.3. *In Vivo* Pharmacology

The prodrug EIDD-2801 or its parent EIDD-1931 have been tested in animal models of RNA viral infection. An overview of results from the animal studies in indications to be pursued are described below. Additional detail is provided in the Investigator's Brochure (IB).

1.3.3.1. Coronavirus: SARS-CoV-1 and MERS-CoV

In mouse models of SARS and MERS infection and disease, coronaviral disease was assessed by changes in body weight, measured daily, and lung hemorrhage, assessed in the large left lobe of the lung following sacrifice on Day 5 (SARS-CoV) or Day 6 (MERS-CoV). To assess production of infectious virions, virus was isolated from the lower left lobe of the lung following sacrifice on Day 5 (SARS-CoV) or Day 6 (MERS-CoV) and quantified using a plaque assay.

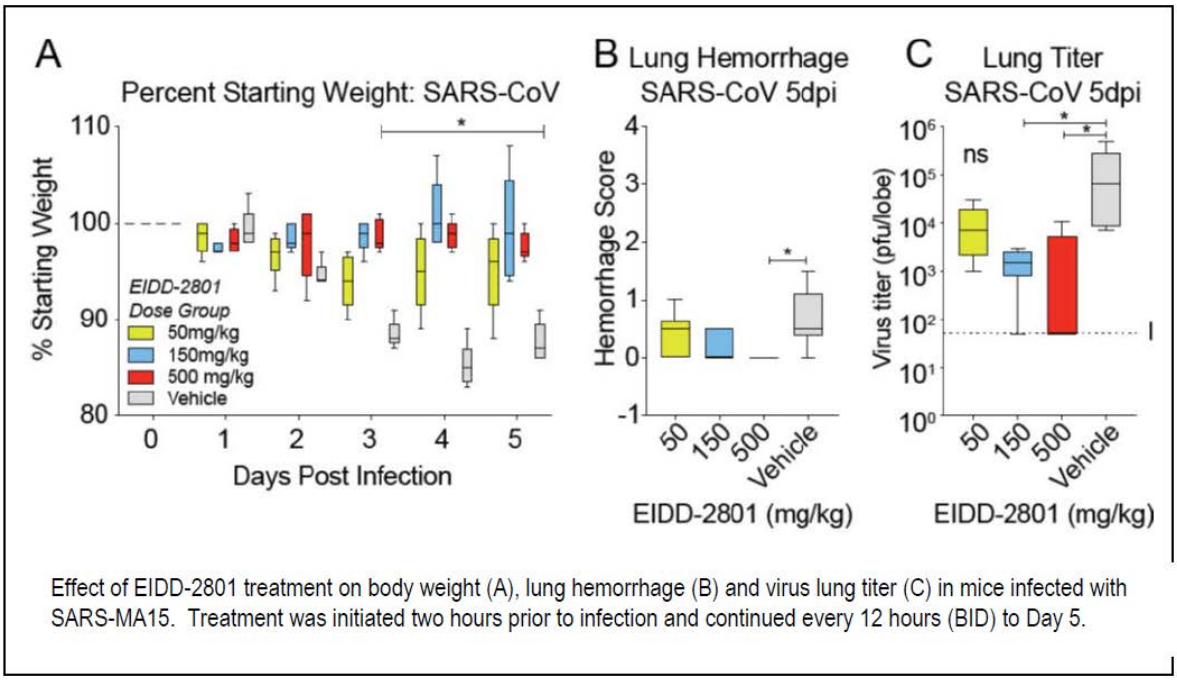
The results demonstrated that, in mice infected with either SARS- or MERS-CoV, both prophylactic and therapeutic treatment with EIDD-2801 resulted in a reduction in virus replication, improvements in pulmonary function, and improvements in maintaining body weight

(i.e., reduced body weight loss). While EIDD-2801 doses of 50, 150 and 500 mg/kg BID were assessed in the CoV mouse experiments, 500 mg/kg BID yielded the most consistent therapeutic effect.

A prophylactic, dose-escalation study was conducted in C57BL/6 mice infected with mouse-adapted SARS-CoV (SARS-MA15). Prophylactic oral treatment with EIDD-2801 was initiated 2 hours before intranasal infection and continued every 12 hours thereafter through the end of the study (Day 5; Figure 3).

In mice treated BID with EIDD-2801, body weight loss observed with vehicle treatment was diminished in the 50 mg/kg treatment group, beginning on Day 3 post-infection. No body weight loss was seen in the 150 and 500 mg/kg treatment groups (Figure 3, Panel A). Lung hemorrhage was also significantly reduced on Day 5 post-infection, following treatment with 500 mg/kg EIDD-2801 (Figure 3, Panel B). When compared to vehicle control, a dose-dependent reduction in SARS-CoV lung titers at Day 5 was seen across all 3 treatment groups (Figure 3, Panel C) with significant differences among the vehicle, 150 mg/kg and 500 mg/kg groups. Thus, prophylactic treatment with EIDD-2801 resulted in a robust antiviral effect that was able to prevent SARS-CoV replication and disease.

Figure 3: EIDD-2801 Prophylactic Treatment of SARS-CoV Infected Mice

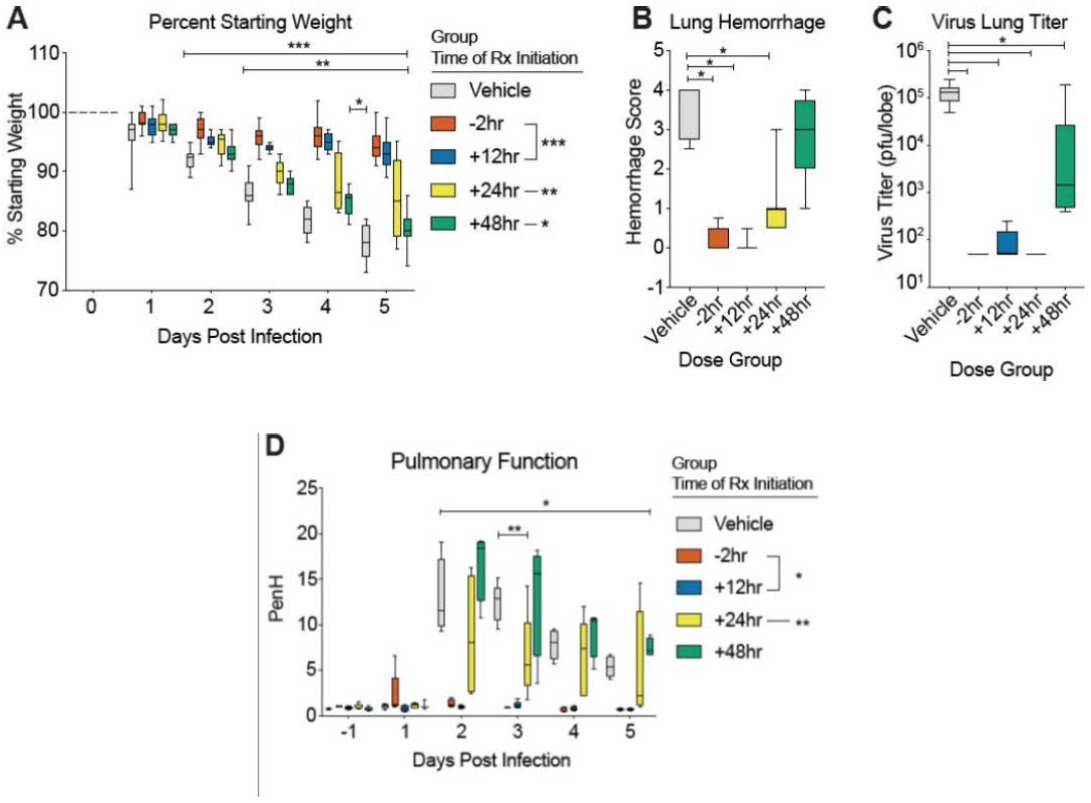


Effect of EIDD-2801 treatment on body weight (A), lung hemorrhage (B) and virus lung titer (C) in mice infected with SARS-MA15. Treatment was initiated two hours prior to infection and continued every 12 hours (BID) to Day 5.

The antiviral activity of EIDD-2801 against SARS-CoV was compared when treatment was initiated at -2 hours (pre-infection) and 12, 24, or 48 hours post-infection. After initiation of treatment, all groups were dosed every 12 hours for the duration of the study (Figure 4). For SARS-challenged mice, initiating treatment at 12 hours post-infection significantly prevented body weight loss beginning on Day 2, a result similar to that seen when dosing prophylactically

(i.e., beginning at 2 hours pre-infection). Initiation of treatment with EIDD-2801 at 24 hours post-infection also significantly reduced body weight loss on Days 3 through 5 post-infection. When EIDD-2801 treatment was initiated at 48 hours post-infection, body weight loss was only statistically different from vehicle on Day 4 post-infection (Figure 4, Panel A). Significant reductions in lung hemorrhage were seen when EIDD-2801 treatment was initiated before (-2 hours) and up to 24 hours after infection; a result that mirrored body weight loss data (Figure 4, Panel B). All mice treated with EIDD-2801 had significantly reduced viral loads in the lungs, even in the group where treatment was initiated 48-hour post-infection (Figure 4, Panel C). Pulmonary function, measured via whole body plethysmography (WPB), was assessed using the PenH metric which is a surrogate marker for bronchoconstriction or pulmonary obstruction. The administration of EIDD-2801 prior to infection (-2 hours) and at 12 hour post-infection completely abrogated the loss of pulmonary function observed in vehicle-treated animals (Figure 4, Panel D). Improved pulmonary function was also seen in the group where treatment was initiated 24 hours after virus challenge.

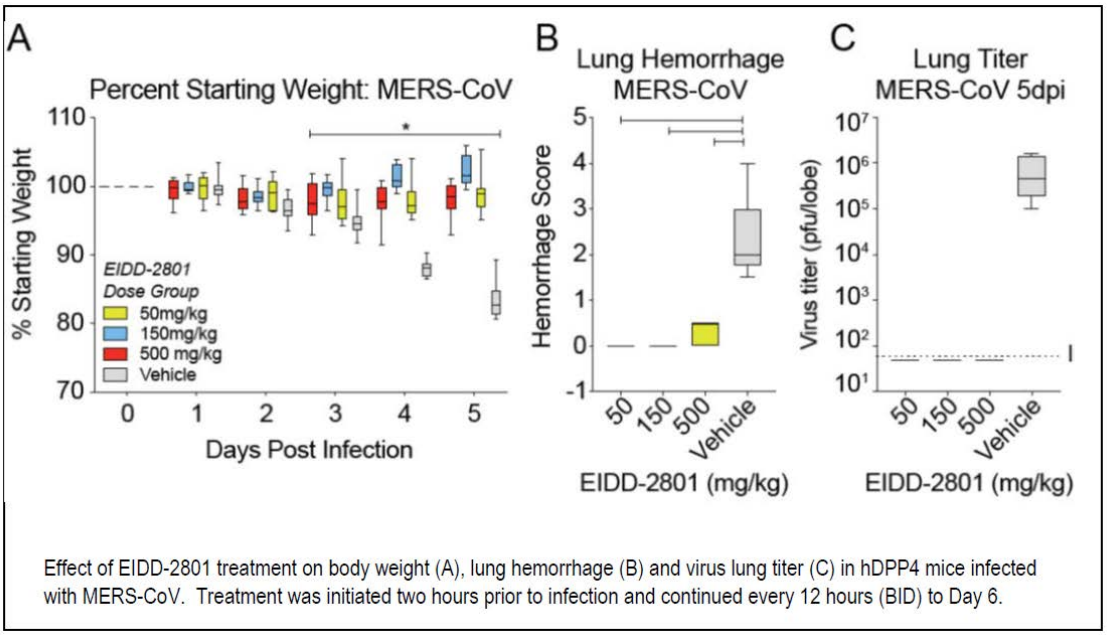
Figure 4: EIDD-2801 Treatment of SARS-CoV Infected Mice



EIDD-2801 was also tested to determine if it is active *in vivo* against MERS-CoV as described by Sheahan et. al (draft manuscript). The murine ortholog of the MERS-CoV receptor, dipeptidyl peptidase 4 (DPP4), does not support viral binding and entry. Thus, all *in vivo* studies described below were performed in genetically modified hDPP4 mice permissive for MERS infection. Prophylactic treatment starting at 2 hours before viral challenge with either 50, 150, or

500 mg/kg EIDD-2801 prevented body weight loss on Days 2 through 6 post-infection (Figure 5, Panel A), prevented lung hemorrhage measured on Day 6 (Figure 5, Panel B), and reduced virus lung titer on Day 6 to the limit of detection (Figure 5, Panel C).

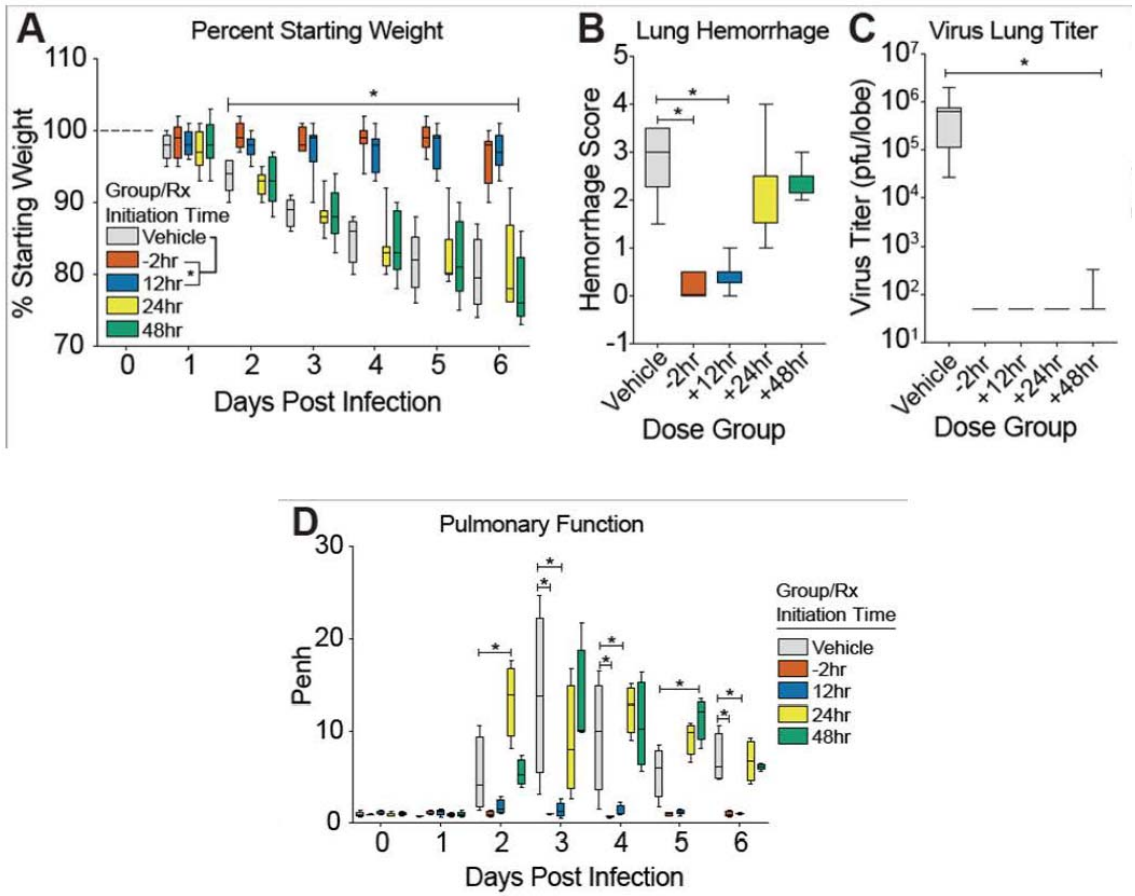
Figure 5: EIDD-2801 Prophylactic Treatment of MERS-CoV Infected Mice



Effect of EIDD-2801 treatment on body weight (A), lung hemorrhage (B) and virus lung titer (C) in hDPP4 mice infected with MERS-CoV. Treatment was initiated two hours prior to infection and continued every 12 hours (BID) to Day 6.

The effect of EIDD-2801 treatment on MERS-CoV infected mice is shown in Figure 6. When EIDD-2801 treatment was initiated 12 hours post-infection, there was no loss in body weight from Days 2 through 6 post-infection (Figure 6, Panel A) and no evidence of lung hemorrhage on Day 6 post-infection (Figure 6, Panel B). However, protection was not observed in groups where treatment was initiated either 24- or 48-hours post-infection. Conversely, virus lung titer on Day 6 post-infection was significantly reduced to the limit of detection in all treatment groups, regardless of the time treatment began (Figure 6, Panel C). To gauge the effect of the timing of EIDD-2801 treatment initiation on physiologic measures of lung disease, pulmonary function was assessed by whole body plethysmography. As expected, a loss of pulmonary function, as determined by measuring the PenH metric, was observed in vehicle-treated animals infected with MERS-CoV beginning on Day 2 post-infection (Figure 6, Panel D). Mirroring the body weight loss data, normal pulmonary function was observed in groups where treatment was initiated prior to or at 12 hours post-infection (Figure 6, Panel D).

Figure 6: EIDD-2801 Treatment of MERS-CoV Infected Mice



1.3.3.2. Influenza Virus

EIDD-2801 was tested in a ferret model of influenza virus infection and disease. Ferrets recapitulate hallmarks of human influenza infection, providing a clinically relevant animal model to investigate therapeutic intervention. Therapeutic oral dosing of influenza virus-infected ferrets reduced shed levels of pandemic and seasonal IAV by multiple orders of magnitude, and alleviated fever and inflammation while improving airway epithelium histopathology. Post-exposure prophylactic dosing was sterilizing (Toots et al., 2019).

Ferrets infected with pandemic IAV and treated with EIDD-2801 at a dose of 7 mg/kg demonstrated significantly less viral shedding and inflammatory cellular infiltrates in nasal lavages, but mounted a normal humoral antiviral response (Toots et al., 2019).

When examining the effect of delayed dosing, Toots et. al. (2019) demonstrated that treatment with 20 mg/kg of EIDD-2801 initiated 24 hours after challenge with IAV (H1N1) resulted in a rapid decrease in both shed virus and fever relative to control, with fever returning to baseline at 24 hours. When oseltamivir (20 mg/kg) was dosed prophylactically as a positive control, a modest suppression of fever was observed; however, there was no reduction in shed virus titer.

1.3.3.3. Venezuelan Equine Encephalitis Virus

Treatment with EIDD-1931 was evaluated in a mouse model of lethal VEEV infections. To be truly effective as a therapeutic agent for VEEV infection, a drug must penetrate the blood brain barrier and arrest virus replication in the brain. High plasma levels of EIDD-1931 are rapidly achieved in mice after oral dosing. Once in the plasma, EIDD-1931 is efficiently distributed into organs important in the pathology of VEEV infection, including the brain, where it is rapidly converted to its active 5'-triphosphate (EIDD-2061). EIDD-1931 showed a good safety profile in mice after 7 days of dosing with up to 1,000 mg/kg/day. In mouse model studies of VEEV infection, EIDD-1931 was 90-100% effective in protecting mice against lethal intranasal infection when therapeutic treatment was started as late as 24 hours post-infection, and partial protection was achieved when treatment was delayed for 48 hours post-infection (Painter et al., 2019).

[redacted]

1.4. Safety and Secondary Pharmacology

The standard battery of safety pharmacology studies including studies assessing the cardiovascular, respiratory and central nervous systems have been conducted. The studies are discussed in the IB; results indicated that there were no adverse pharmacologic effects of EIDD-2801 on the cardiovascular, respiratory or central nervous systems.

1.5. Nonclinical Pharmacokinetics and Metabolism

1.5.1. Overview

The uptake, metabolism and protein binding of EIDD-2801 and EIDD-1931 have been studied in plasma, microsomes, and non-hepatic cells from several species as outlined below. The PK and tissue distribution of EIDD-2801 and its metabolite EIDD-1931 have been studied extensively in rats, dogs and ferrets. Key results from these studies are presented below; additional detail can be found in the IB.

1.5.2. Absorption

EIDD-1931 is parent of the prodrug EIDD-2801. The appearance of EIDD-1931 is dependent on the absorption of EIDD-2801 and the rate of its conversion to EIDD-1931.

EIDD-2801 PK studies have been completed in dog, rat, mouse, ferret, and monkey. EIDD-2801 was efficiently absorbed and rapidly converted to EIDD-1931 in each species. The t_{max} for EIDD-2801 (not observed in rodents) occurred at 0.5-1 hours, while the t_{max} for EIDD-1931 occurred in 1-2 hours.

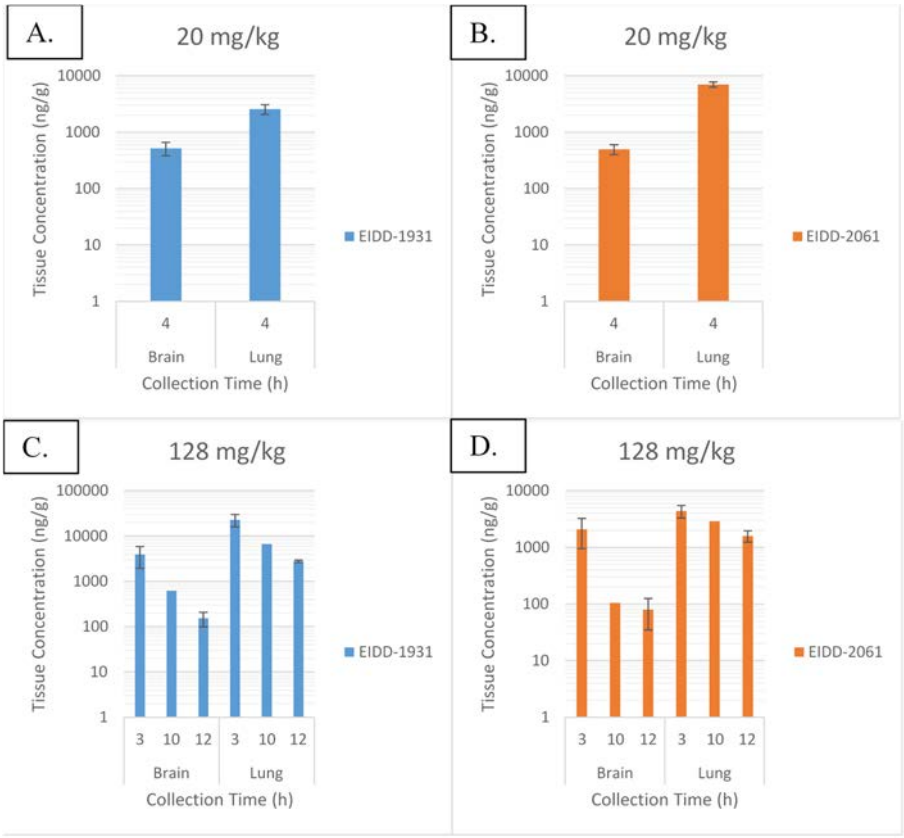
1.5.3. Distribution

[redacted]

1.5.3.2. Tissue Distribution Studies

EIDD-2801 is rapidly absorbed in the gut and converted to EIDD-1931 reaching C_{max} in 1-3 hours in mice, rats, ferrets, dogs and monkeys. EIDD-1931 is then widely distributed to tissues including lungs and brain, where it is rapidly taken up into cells and converted to EIDD-2061. Figure 7 shows the concentration of EIDD-1931 and EIDD-2061 in ferret brain and lung following single doses of 20 (Panels A and B) and 128 (Panels C and D) mg/kg.

Figure 7: Tissue Distribution of EIDD-1931 and EIDD-2061 in Ferret Brain and Lung



1.5.4. Metabolism

1.5.4.1. Metabolic Stability of EIDD-1931 and EIDD-2801

EIDD-2801 was designed to be converted to EIDD-1931 by esterases in plasma or in cells. Stability has been assessed in plasma and liver microsomes from mouse, rat, dog, monkey and humans. The stability of EIDD-2801 in mouse, rat and monkey plasma is relatively short (≤0.4 hours) while the stability is longer in human and dog plasma (1-3 hours). EIDD-2801 stability in mouse, rat, dog and monkey liver microsomes is very short, ranging from 0.02 to 0.08 hours while the stability in human liver microsomes is 1.2 hours (Table 6).

Table 6: Metabolic Stability of EIDD-2801 in Plasma and Liver Microsomes

	Plasma stability	LM stability
Species	t1/2 (h)	t1/2 (h)
Mouse	0.017	0.033
Rat	0.033	0.017
Dog	3.2	0.083
Monkey	0.40	0.017
Human	1.05	1.22

EIDD-2801 is stable in simulated gastric and intestinal fluids (Table 7).

Table 7: Metabolic Stability of EIDD-2801 in Simulated Gastric and Intestinal Fluids and in Buffered Saline with Fetal Bovine Serum

Matrix	t1/2 (hr)
Simulated Gastric Fluid	>24
Simulated Intestinal Fluid	>24
Phosphate Buffered Saline plus 10% Fetal Bovine Serum*	>24

EIDD-1931 was found to be stable when incubated with all tested plasmas, whole blood, liver microsomes and liver S9 extracts and intestinal microsomes (Table 8).

Table 8: Metabolic Stability of EIDD-1931 in Plasma, Whole Blood and Liver and Intestinal Microsomes

Medium	Plasma	Whole Blood	Liver Microsomes	Liver S9 Stability	Intestinal Microsomes
Species	t1/2 (h)	t1/2 (h)	t1/2 (h)	t1/2 (h)	t1/2 (h)
Mouse		>24	>24		>24
Rat	17		>24	>24	
Dog	>24			>24	
Monkey	6.5	>24	>24	>24	>24
Human	10	7	>24	20	>24

1.5.4.2. Uptake and Anabolism of EIDD-1931 in Tissue Culture and Primary Cells

EIDD-1931 is efficiently taken up by tissue culture cells and converted to its pharmacologically active metabolite EIDD-2061 (EIDD-1931-5'-triphosphate). Intracellular EIDD-2061 accumulates dose-dependently, with C_{max} levels ~200-2000 pmol/10^6 cells (at 10-20 µM dose) in different cell lines. It reaches high levels relatively quickly, typically within 1-3 hours, though the t_{max} values vary widely between 1 and 24 hours depending on the cell line and dose concentration tested. Detailed data on the uptake and anabolism of EIDD-2801 is presented in the IB.

EIDD-2801 is also taken up by tissue culture cells and is converted to EIDD-1931 and then to EIDD-2061, but the process is slightly delayed compared to dosing with EIDD-1931. EIDD-1931 is also taken up and metabolized to EIDD-2061 by primary cells. EIDD-2061 is accumulated in all primary cells tested except in mouse primary hepatocytes where EIDD-1931 is apparently extensively metabolized to cytidine and uridine which, in turn, quickly metabolize into CTP and UTP. The quick metabolism of EIDD-1931 consequently results in low levels of EIDD-2061 in mouse hepatocytes. The intracellular stability ($t_{1/2}$) of EIDD-2061 is 4-5 hours in human astrocytes and hBTEC and is significantly shorter (0.2-1.1 hours) in primary hepatocytes.

1.5.5. Excretion

Currently, there is no data on excretion of EIDD-2801 or EIDD-1931. Excretion will be measured in urine during this study.

1.5.6. Pharmacokinetic Drug Interactions

1.5.6.1. Cytochrome P450 (CYP) Inhibition

The purpose of this non-GLP *in vitro* study was to determine the time-dependent inhibitory potential of EIDD-2801 and EIDD-1931 on human cytochromes P450 (CYP) 1A2, 2B6, 2C8, 2C9, 2C19, 2D6, and 3A4 (midazolam and testosterone substrates) enzyme activity, using pooled human liver microsomes in an half maximal inhibitory concentration (IC_{50}) shift assay.

Neither EIDD-2801 nor EIDD-1931 demonstrated inhibition greater than 31.4% for any of the CYP isozymes tested nor could the data for each assay condition be curve fit to determine time-dependent inhibition by these compounds. Full dose-response curves were not achieved at concentrations ranging from 0.00545 to 50.0 µM indicating EIDD-2801 and EIDD-1931 have no CYP inhibition potential at concentrations ranging from 0.00545 to 50.0 µM. Assay performance was acceptable based on the results for the positive control inhibitors.

1.5.6.2. CYP Induction

An assay was performed to determine the induction potential of EIDD-2801 on human CYP isoenzyme (1A2, 2B6, and 3A4) activity using 3 single-donor lots of inducible, cryopreserved human hepatocytes. Both enzyme activity and mRNA results demonstrated that EIDD-2801 did not show induction for any of the CYP isozymes.

[redacted]

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[redacted]	[redacted]	[redacted]	[redacted]	[redacted]
[redacted]	[redacted]	[redacted]	[redacted]	[redacted]
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1.7. Potential Risks and Benefits

1.7.1. Potential Benefits

As this is a FIH study in healthy volunteers, there is no direct benefit to subjects enrolled in the study. However, given the current pandemic situation and *in vitro* antiviral activity of EIDD-2801 against SARS-CoV-2, and the activity against several other viruses of public health concern, it is possible that participants may benefit from future availability of the drug.

1.7.2. Potential Risks

EIDD-2801 has never been administered to humans; therefore, the risks from EIDD-2801 to subjects participating in this trial are unknown. Although toxicology studies have been done, unexpected AEs may occur. [redacted]. Subjects will also be monitored for other end-organ effects through a range of safety assessments.

2. OBJECTIVES

2.1. Study Objectives

2.1.1. Primary Objectives

The primary objective of Part 1 of the study is to determine the safety and tolerability of single ascending doses of EIDD-2801.

The primary objective of Part 2 of the study is to assess the effect of food on the PK on EIDD-2801 and EIDD-1931 following a single oral dose.

2.1.2. Secondary Objective

The secondary objective of Part 1 of the study is to define the PK of EIDD-2801 and EIDD-1931 in plasma and urine following singledoses administered to healthy volunteers.

The secondary objective of Part 2 of the study is to determine the safety and tolerability of single doses of EIDD-2801.

2.2. Study Outcome Measures

2.2.1. Primary Outcome Measures

The primary outcome measures for Part 1 of are results of safety evaluations including safety laboratory assessments, PEs, ECGs, vital signs, and AEs.

The primary outcome measures for Part 2 of the study are plasma PK parameters including C_{max}, t_{max}, $t_{1/2}$, CL/F, λz, Vz/F, $AUC_{0\text{-}inf}$ and AUC_{last} as appropriate.

2.2.2. Secondary Outcome Measures

The secondary outcome measures are as follows:

- Single-dose plasma PK parameters, including C_{max}, t_{max}, $t_{1/2}$, CL/F, λz, Vz/F, $AUC_{0\text{-}inf}$ and AUC_{last} as appropriate
- Urinary excretion of EIDD-2801 and EIDD-1931 following single-dose administration.
- Results of safety evaluations including safety laboratory assessments, PEs, ECGs, vital signs, and AEs (Part 2).

3. STUDY DESIGN

3.1. Overview

EIDD-2801-1001-UK is a Phase 1, randomized, double-blind, placebo-controlled, FIH, SAD study of the safety, tolerability and PK of EIDD-2801 and EIDD-1931 following oral administration of singledoses of EIDD-2801 to healthy volunteers. In addition, for a minimum of one cohort, the effect of food on the single-dose EIDD-2801 and EIDD-1931 PK parameters will be assessed in subjects taking open-label EIDD-2801. The overall objective of the study is to identify a starting dose for future safety and therapeutic intervention trials.

The study is composed of 2 parts; Part 1 (P1) is the SAD study and Part 2 (P2) is the FE cohort study.

3.1.1. Part 1

A single oral dose of EIDD-2801 or PBO will be administered to subjects. Subjects will be randomized in a 3:1 ratio to receive either EIDD-2801 or PBO.

Following dosing on Day 1, subjects will remain in the clinic through completion of study assessments on Day 4, returning to the clinic on Day 9 for study assessments, and Day 15 for the EOS visit.

The first cohort will be divided into 2 groups. The first group, a sentinel group, will include 2 subjects. On Day 1, one subject in the sentinel group will be administered PBO and one will be administered EIDD-2801. Analysis of the safety results for the sentinel group will be reviewed 24 hours postdose. If no halting criteria (Section 9) have been met, the remainder of the subjects in Cohort 1 (the second group) will be dosed. There will be no sentinel groups for the remaining cohorts.

After completion of each dosing cohort, safety and tolerability data will be reviewed to determine if any of the halting rules have been met. If not, then the subsequent cohort may be dosed. As PK data become available, these data may be used for dose-escalation decisions.

The proposed dose-escalation scheme is shown in Figure 1, however, planned dose escalations will be determined based on ongoing review of the safety, tolerability, and available PK data. The starting dose in the first SAD cohort will be 50 mg. Dose levels will not increase by more than 3-fold for predicted nonpharmacologically active dose levels and by 2-fold for predicted pharmacologically active dose levels. [redacted]

Four cohorts are initially planned for P1; however, up to an additional 3 cohorts may be enrolled.

3.1.2. Part 2

Two single oral doses of EIDD-2801 will be administered to subjects, in an open-label manner. The dose for the P2/FE cohort is planned to be 100 mg but may be adjusted based on safety,

tolerability, and/or PK data from P1. The dose assessed in the P2/FE cohort will have been given previously to subjects in a P1/SAD cohort successfully (i.e., no halting rules were met following dosing).

Subjects will be randomized to treatment sequence; i.e., to receive drug in the fed then fasted state (Sequence 1) versus fasted then fed state (Sequence 2; Figure 1). Half of the subjects in the cohort will be randomized to Sequence 1 and the other half will be randomized to Sequence 2.

Following dosing on Day 1, subjects will remain in the clinic through completion of study assessments on Day 4, returning to the clinic on Day 9 for study assessments, and Day 14 to begin the second dosing sequence on Day 15. Subjects will then be discharged from the clinic on Day 18, returning to the clinic on Day 23 for study assessments, and Day 30 for the EOS visit. There will be a 14-day (minimum) washout period between doses.

One cohort of 10 subjects is planned for P2. However, if PK results obtained are equivocal, additional subjects may be enrolled into the P2/FE cohort at the previously tested dose or an additional P2/FE cohort may be added at a different dose.

Additional parts may be added to this study, including a SAD part in elderly healthy subjects, a MAD part in healthy subjects, and a study of the safety, tolerability, and PK in Japanese subjects. These additional parts are not described in this protocol, but may be added via a protocol amendment. It is planned that the MAD part will include 4 dose-escalation cohorts (with the option to include an additional 3 cohorts), with subjects receiving BID doses for 7 days. It is planned that each MAD cohort will comprise 8 subjects, with 6 subjects receiving EIDD-2801 and 2 subjects randomized to PBO.

3.2. Rationale and Justification

3.2.1. Justification of Design

The FIH study is a typical dose-escalation study designed to provide the maximum amount of data in the minimum number of subjects. The cohort size in P1 is planned to be 8 subjects (6 active:2 PBO). This number of subjects allows for a sufficient PK analysis, considered to be important because dose extrapolation from efficacious animal models to humans will be based on exposure. The duration of participation for each subject following dosing well exceeds 5 drug half-lives (up to 9.1 hours in dogs; 5 hours in ferrets) [redacted] The FE cohort is considered to be important to maximize exposure based on fed vs. fasted condition and obtaining this information early in Phase 1 will minimize study drug dose in all future studies. This design follows the FDA guidance document on assessing FE in clinical studies.

3.2.2. Justification of Starting Dose

[redacted]

[redacted] The FDA guidance document *("Estimating the Maximum Safe Starting Dose in Initial Clinical Trials for Therapeutics in Adult Healthy Volunteers")* provides guidance for when a safety factor smaller than 10 may be used to calculate the starting dose:

- A smaller safety factor might also be used when toxicities produced by the therapeutic are easily monitored, reversible, predictable, and exhibit a moderate-to-shallow dose-response relationship with toxicities that are consistent across the tested species (both qualitatively and with respect to appropriately scaled dose and exposure).

- A safety factor smaller than 10 could be justified when the NOAEL was determined based on toxicity studies of longer duration compared to the proposed clinical schedule in healthy volunteers. In this case, a greater margin of safety should be built into the NOAEL, as it was associated with a longer duration of exposure than that proposed in the clinical setting. This assumes that toxicities are cumulative, are not associated with acute peaks in therapeutic concentration (e.g., hypotension), and did not occur early in the repeat dose study.

EIDD-2801 satisfies both of these criteria as follows:

[redacted]

Importantly, given the activity of EIDD-2801 versus SARS-CoV-2 (cause of COVID-19), the Sponsor thinks it is most prudent to start with a dose that is predicted to be a safe starting dose and is also as close as possible to a potential therapeutic dose that would allow the Sponsor to

move into COVID-19 patients as safely and expeditiously as possible. Based on modeling to animal data, a dose of 100 mg BID is projected to be an active dose in humans.

3.2.3. Justification of Study Population

Healthy volunteers are considered to be the appropriate population for conduct of the FIH study. Healthy volunteers without confounding medical conditions that may obscure the interpretation of AEs or affect absorption, distribution, metabolism and excretion of study drug will provide the most valuable data regarding the tolerability, safety, and plasma exposures observed and expected following single doses. EIDD-2801 is intended for eventual study in patients with potential CoV-2 infection as defined by the Centers for Disease Control and Prevention (CDC) in whom a range of AEs are expected based on the disease under study. Understanding the safety and PK profile in a normal population will better inform the use of EIDD-2801 in disease settings where complications are frequent, and AEs will need to be interpreted in context. While EIDD-2801 is being developed for the treatment of highly pathogenic CoV, there are plans underway to develop protocols for the treatment of influenza. Without safety data from healthy individuals, it will be more difficult to extrapolate the interpretation of AEs from one disease state to another.

4. STUDY POPULATION

This study will enroll healthy volunteers; 8 subjects will be enrolled into each SAD cohort in P1 and 10 subjects will be enrolled into the FE cohort in P2.

The site is strongly encouraged to ensure that women are represented in each cohort.

4.1. Subject Inclusion Criteria

Subjects who meet all of the criteria at Screening will be eligible for study participation

1. Male or female between the ages of 18 and 60, inclusive.

2. Female subjects must be of non-childbearing potential. Non-childbearing potential is defined as women who:

    - are physiologically incapable of becoming pregnant including those who are surgically sterile, via hysterectomy, bilateral salpingectomy, and/or bilateral oophorectomy by reported medical history and/or medical records. Surgical sterilization to have occurred a minimum of 6 weeks, or at the PI's (or designee's) discretion, prior to Screening.

    - are post-menopausal (at least 45 years of age with amenorrhea for 12 months without an alternative medical reason with confirmatory FSH levels of ≥ 40 mIU/mL. The amenorrhea should not be induced by a medical condition such as anorexia nervosa, hypothyroid disease or polycystic ovarian disease, or by extreme exercise. It should not be due to concomitant medications that may have induced the amenorrhea such as oral contraceptives, hormones, gonadotropin-releasing hormones, anti-estrogens, or selective estrogen receptor modulators).

3. Male participants must agree to the use of effective contraception for study duration, i.e., from Check-in until 90 days after the EOS visit.

4. Is in generally good health as determined by medical history, PEs (at Screening and/or Check-in), vital signs, and other screening procedures.

5. Has a BMI of 18 to 30 kg/m^2.

6. Is willing and able to comply with all study procedures including providing informed consent.

4.2. Subject Exclusion Criteria

Subjects who meet any of the following criteria at Screening (unless otherwise stated) will not be eligible for participation:

1. Females who are pregnant, planning to become pregnant, or breastfeeding.
2. Has a clinically relevant acute or chronic medical condition or disease of the cardiovascular, gastrointestinal, hepatic, renal, endocrine, pulmonary, neurologic, or dermatologic systems as determined by the PI (or designee).
3. Has any current or historical disease or disorder of the hematological system or significant liver disease or family history of bleeding/platelet disorders.
4. Has a history of cancer (other than basal cell or squamous cell cancer of the skin), rheumatologic disease or blood dyscrasias.
5. Has a history of gastrointestinal surgery or other condition that may interfere with absorption of study drug, in the option of the PI (or designee).
6. Has a history of febrile illness within the 14 days prior to the first dose of study drug.
7. Has a positive alcohol or drug screen at Screening or the Day -1 visit or has a history of alcohol or drug abuse within the past 5 years.
8. Currently uses tobacco, nicotine or tobacco products or e-cigarettes or stopped using tobacco products within the past 3 months and/or has a positive cotinine test at Screening or Check-in.
9. Has any abnormal laboratory value of Grade 2 or higher, which is considered to be clinically significant by the PI (or designee).
10. Has a total white cell count or absolute lymphocyte count outside the normal range, or hemoglobin or platelet levels below the lower limit of normal, at Screening or Day -1.
11. Has values above the upper limit of normal for the following laboratory analytes: ALT/SGPT, alkaline phosphatase (serum), AST/SGOT, at Screening or Day -1.
12. Positive test result for HIV, HBV, or HCV.
13. Has an autoimmune disease, is immunosuppressed or is in any way immunocompromised.
14. Has any of the following:
    - QTcF >450 ms confirmed by repeat measurement

- QRS duration >110 ms confirmed by repeat measurement
- PR interval >220 ms confirmed by repeat measurements
- findings which would make QTc measurements difficult or QTc data uninterpretable
- history of additional risk factors for torsades de pointes (e.g., heart failure, hypokalemia, family history of long QT syndrome).

15. Except as noted, has used prescription drugs (other than hormone replacement therapy) within 14 days prior to the first dose of study drug unless, in the opinion of the PI (or designee), the drug will not interfere with study assessments. The following prescription drugs will be allowed:
    - Statins: Subjects who have been on a stable dose of statins for a minimum of 6 months and who have normal creatine kinase at Screening.
    - Antihypertensives: Subjects who have been on a stable dose of antihypertensives for a minimum of 6 months and have controlled blood pressure and no active heart disease.
16. Has received an experimental agent (vaccine, drug, biologic, device, blood product or medication) within 3 months prior to the first dose of study drug and agrees not to receive another experimental agent during the duration of this trial.
17. Has donated blood within 30 days, plasma within 2 weeks, or platelets within 6 weeks prior to Screening or refuses to agree to not donate blood until 3 months after the EOS visit.
18. Uses OTC medications or nutritional supplements (including, e.g., gingko biloba, aspirin at any dose, and NSAIDs) on a routine/scheduled basis and/or is unwilling to abstain from such use for the duration of the study.
19. Is unwilling to abstain from quinine containing products including quinine water, tonic water, bitter lemon, jello shots, any quinine preparations or Cinchona tree bark preparations (e.g., herbal medications containing Cinchona tree bark).
20. Has any condition that would, in the opinion of the PI (or designee), put the subject at increased risk for participation in a clinical trial or confinement in a clinical research unit.

5. STUDY MEDICATION, RANDOMIZATION AND DOSE ADMINISTRATION

5.1. Study Drug Description

EIDD-2801 and matching PBO will be supplied [redacted].

[redacted]

[redacted]

[redacted].

5.1.1. Acquisition, Formulation, Packaging and Labeling

[redacted]

All study drug will be labeled according to the regulatory requirements for investigational product.

5.1.2. Product Storage and Stability

Study drug should be stored at controlled room temperature defined as [redacted]. If excursions occur which are outside of this range, the pharmacy staff should contact Sponsor to determine the course of action. Additional stability data may be available which would allow continued use of the study drug, or study drug may need to be replaced.

5.2. Randomization

Unmasked study drug (EIDD-2801 and matching PBO) will be supplied to the study site pharmacy. The pharmacy staff will be unmasked with regards to treatment assignment. A randomization list will be provided to the pharmacy staff who will use that list to dispense masked study drug for administration to each study participant. In P1, 6 subjects will receive

EIDD-2801 while 2 subjects will be randomized to PBO. In P2, all 10 subjects in the FE cohort will receive EIDD-2801. The pharmacy staff will maintain the security of the randomization list ensuring that no study personnel outside of the pharmacy have access to identify treatment assignment. In the case that it becomes necessary to know a subject's treatment assignment, unmasking procedures will be followed as discussed in Section 8.4.

5.3. Dosage, Preparation and Administration of Study Drug

Detailed instructions for extemporaneous compounding (as necessary), dispensing and administering study drug can be found in the pharmacy manual.

[redacted]

5.4. Drug Accountability

The site pharmacy must maintain records of receipt and disposition of all study drug supplied to the site by the Sponsor. The records must be maintained according to site standard operating procedures (SOPs) and should include at a minimum, receipt date, lot or batch number, amount and formulation received, [redacted], date, time and amount dispensed, subject identifier for each aliquot of study drug dispensed. In addition, the clinic staff must ensure that the time of dosing is identified for each subject.

Monitors must verify drug accountability/dispensing records during the monitoring visit.

Unused study drug must be disposed of according to the procedures described in the pharmacy manual.

5.5. Concomitant Medication/Treatments

[redacted] In this FIH study, limited types of concomitant medications are permitted. Adjustment of routine medications taken by subjects should be avoided during study participation except when subject safety could be affected by lack of

adjustment. There are no restrictions on treatment medications prescribed by the PI (or designee) to be used for AEs that occur during study participation. For additional restrictions, see Section 6.1.2.

6. STUDY CONDUCT AND VISIT SCHEDULE

All study assessments will be conducted according to the Time and Events Schedule.

6.1. Study Conduct

6.1.1. Study Windows and Rounding Principles

The time schedule described in the protocol for each scheduled activity should be followed as closely as possible. However, if it is not possible and if it is not otherwise specifically contraindicated per protocol, then the time windows detailed in Table 10 are allowed without incurring a protocol deviation.

Table 10: Allowable Time Windows for Study Assessments/Visits

Protocol Specified Time	Allowable Window
PK Samples and Associated Assessments	
Predose	within 2 hours prior to dosing
<2 hours	± 5 minutes
≥2 hours to 24 hours	± 15 minutes (± 2 hours for urinalysis)
>24 hours to 48 hours	± 30 minutes
>48 hours	± 1 hour (PK) and ± 2 hours (safety)
Study Visits - Parts 1 and 2	
Day −1 to Day 4 (Part 1)	must occur on scheduled day
Day −1 to Day 4 and Day 14 to Day 18 (Part 2)	must occur on scheduled day
Day 9 (Part 1) and Days 9 and 23 (Part 2)	± 1 day
End-of-Study (EOS) Visit	± 2 days

6.1.2. Restrictions

Prior to arriving at the clinic for the Day -1 visit, subjects must abstain from consumption of alcoholic beverages for a minimum of 72 hours prior to Check-in. Subjects must continue to abstain from consumption of alcoholic beverages throughout clinic confinement. Subjects enrolled in P2/FE must continue to refrain from consuming alcoholic beverages from discharge on Day 4 through Check-in on Day 14, and then through the second clinic confinement to Day 18. After discharge on Day 4 (P1) or Day 18 (P2) subjects must minimize consumption of alcoholic beverages (i.e., limit of up to one serving per day) until the EOS procedures have been completed.

All subjects must refrain from the following:

- consuming quinine containing products from 72 hours prior to Check-in through to completion of the EOS procedures.
- using nutraceuticals and nutritional/vitamin supplements (e.g., gingko biloba,

multivitamins) from 72 hours prior to Check-in through to completion of the EOS procedures. However, vitamin supplements required by a physician are exempt from this restriction.

- taking OTC analgesics including aspirin (any dose) and NSAIDS from 72 hours before Check-in until completion of EOS study procedures unless prescribed by the PI (or designee).

- use of tobacco, nicotine or tobacco products, or e-cigarettes from 3 months prior to Screening until the EOS visit.

- strenuous exercise from 7 days before Check-in until the EOS visit. Subjects will otherwise maintain their normal level of physical activity during this time (i.e., will not begin a new exercise program nor participate in any unusually strenuous physical exertion).

- Female subjects must not donate eggs/ovum from the time of Check-in until 3 months after the EOS visit.

All subjects in P1 should be dosed in the fasted state. Subjects should fast overnight for a minimum of 10 hours prior to dosing in the morning on Day 1. Following dosing, subjects may have water after 2 hours and food beginning 4 hours postdose. For subjects to be dosed in the fed state, subjects should have a high-fat breakfast as defined in the FDA guidance. Subjects must complete the meal within 30 minutes of starting the meal and should be dosed after 30 minutes of starting the meal.

6.2. Screening

6.2.1. Screening Visit

Subjects who meet preliminary pre-screening criteria (as defined by the site) and are interested in participating in the study will arrive at the study site for administration of informed consent according to site SOPs. After the subject has signed and dated the ICF, screening procedures can begin. Assessments and procedures should be conducted as shown in the Time and Events Schedule Screening may be conducted as early as 28 days prior to dosing.

6.2.2. Retesting Procedures

In the case where a parameter is outside of the acceptable limits for enrollment, measurement of the parameter can be repeated one time if both the following are met:

- In the opinion of the PI (or designee), the parameter value does not represent a chronic condition that otherwise will preclude the volunteer from enrolling in the study; and

- In the opinion of the PI (or designee), the abnormality is not likely to recur.

6.2.3. Study Visits

Subjects who satisfy entry criteria will return to the clinic for the Day -1 visit. Following review of I/E criteria, subjects who still qualify will be checked into the clinic and enrolled into the study. Following enrollment, clinical chemistry, hematology and urine samples will be collected to determine baseline values. Based on site standard practices, alternate subjects will also be enrolled into the study in case one of the selected subjects cannot be dosed. If all subjects can be dosed on the morning of Day 1, the alternates will be released and may be enrolled into subsequent cohorts. All assessments and procedures will be performed according to the Time and Events Schedule.

6.2.4. Part 1

Subjects will remain in the clinic through completion of study procedures on Day 4, returning to the clinic for procedures on Day 9.

6.2.5. Part 2

Subjects will remain in the clinic through completion of study procedures on Day 4, returning to the clinic on Day 9 for study assessments, and Day 14 to begin the second dosing sequence on Day 15. Subjects will then be discharged from the clinic on Day 18, returning to the clinic on Day 23 for study assessments.

6.3. Safety Follow-up and End-of-Study Visit

Subjects in P1 will return to the clinic for the EOS visit on Day 15. Subjects in the FE cohort (P2) will return for the EOS visit on Day 30. Subjects with drug-related AEs at the EOS visit will be followed as discussed in Section 8.2.1.

6.4. Subject Withdrawal and Replacement

As this is a small study with a limited number of subjects per cohort, it is critical that all subjects complete the study including postdose study assessments. Site personnel should emphasize this to study subjects at the time of informed consent so that subjects will understand this fact before agreeing to participate in the study.

Subjects may voluntarily withdraw their consent for study participation at any time without penalty or loss of benefits to which they are otherwise entitled. An PI (or designee) may also withdraw a subject from receiving study drug or participation in the study for any reason. Subjects who withdraw or are withdrawn from the study should undergo withdrawal procedures as discussed below. These procedures would include follow-up safety evaluations.

6.4.1. Reasons for Withdrawal

If a subject withdraws or is withdrawn from the study, the primary consideration must be the health and welfare of the subject. The reasons for withdrawal might include but are not limited to the following:

- Subject no longer meets eligibility criteria including subject withdraws consent from study participation (with or without a reason)
- Subject becomes noncompliant
- Medical disease or condition, or new clinical finding(s) for which continued participation, in the opinion of the PI (or designee) might compromise the safety of the subject, interfere with the subject's successful completion of this study, or interfere with the evaluation of responses
- Subject Lost-to-Follow-up
- Subject becomes pregnant, if applicable
- Determined by a physician's discretion to require additional therapy not indicated in the protocol to ensure subject's health and well-being (or treatment failure, if applicable)

The PI should be explicit regarding study follow-up (e.g. safety follow-up) that might be carried out. If the subject consents, every attempt will be made to follow all AEs through resolution, return to baseline, or until stabilized with sequelae for a maximum of 30 days following discontinuation. The procedures that collect safety data for the purposes of research must be inclusive in the original ICF or the PI may seek subsequent informed consent using an EC-approved ICF with the revised procedures.

The PI will inform the subject that already collected data will be retained and analyzed even if the subject withdraws from this study.

6.4.2. Handling of Withdrawals

Subjects who withdraw from the study prior to receiving study drug (i.e., on Day -1 or before dosing on Day 1) will be discharged from the clinic and followed only if AEs are present which occurred due to participation in the study (e.g., AE resulting from a study procedure). In this case, the subject should be followed until the AE resolves or the PI determines that the AE has stabilized.

Subjects who withdraw from the study after receiving study drug should have EOS assessments at the time of withdrawal or as quickly thereafter as possible.

Subjects who do not return for follow-up procedures on Days 9/23 or 15/30 (P1 and P2) will be contacted by the site at least 3 times using the subject’s preferred method of communication (as determined at Check-in). If the site is unable to contact the person, then a certified letter will be sent. If the subject still cannot be reached or refuses to come back to the clinic after all attempts, the subject will be considered Lost-to-Follow-up and withdrawn from the study.

6.4.3. Documentation of Withdrawals

If a subject withdraws or is withdrawn prior to completion of the study, the reason for this decision must be recorded in the CRF. If the subject is Lost-to-Follow-up, the site should document the attempts to contact the subject in the source documents. If the subject has an ongoing AE at the time of withdrawal, then the AE should be followed as detailed in Section 8.2.1.

6.4.4. Subject Replacement

If a subject withdraws from the study prior to receiving study drug, the subject will be replaced. In this case, designated alternate subjects, if available, will be first in line to replace the withdrawn subject.

If a subject withdraws from the study after receiving study drug, then the decision to replace the subject will be made by the PI (or designee) in consultation with the Sponsor. Factors to consider will be the timing postdose of withdrawal and the number of safety and PK assessments completed prior to withdrawal. Subjects who are withdrawn because of an AE related to the study drug will not be replaced.

6.5. Unscheduled Visit(s)

If a subject experiences an AE after discharge from the clinic but prior to the EOS visit, the subject should be instructed to call the site. Based on the issue, the PI (or designee) may request that the subject return to the site for an unscheduled visit. In this case, procedures/assessments should be conducted as deemed appropriate for the situation by the PI (or designee). The visit should be recorded in the unscheduled visit page of the CRF.

7. STUDY PROCEDURES/EVALUATIONS

7.1. Demography and Medical History

Demographics including age, gender, race, ethnicity, and medical history will be recorded for each subject. All significant medical history should be recorded. In general, significant medical history should include all ongoing events and all events occurring within the last 6 months. Clinically relevant or clinically significant events occurring greater than 6 months ago should be recorded. All surgeries occurring in adulthood should be recorded. If surgeries occurred more than 2 years ago, then only the year needs to be recorded on the CRF.

7.2. Clinical Evaluations

7.2.1. Physical Examinations

The PE will be performed by the PI or a designee that is licensed to perform a PE per local requirements. The initial PE performed at Screening and the final PE conducted at the EOS visit will include examination of all pertinent body systems as defined by the site standard PE body systems (general appearance, HEENT, lymphatic, cardiovascular, respiratory, gastrointestinal musculoskeletal, neurological, dermatological).

Subsequent PEs will be performed as shown in the Time and Events Schedule and will be targeted to any new signs or symptoms, any change from the previous status or any body system deemed pertinent to that subject by the PI (or designee). Clinically significant abnormalities should be recorded in the CRF; those occurring prior to dosing will be included in medical history unless the abnormality was the direct result of study participation.

7.2.2. Vital Sign Measurements and ECGs

Vital signs include pulse rate, respiratory rate, blood pressure, and body temperature. Vital signs should be measured after the subject has been supine for a minimum of 5 minutes. Site standard ranges will be used for determining any out-of-range values.

Height and weight should be measured, and BMI calculated at Screening as indicated in the Time and Events Schedule.

Resting 12-lead ECGs should be recorded at the visits indicated in the Time and Events Schedule after the subject has been supine for a minimum of 5 minutes. The PI (or designee) will evaluate the ECG tracings to determine if there are out-of-range values; if out-or-range values are detected, the PI (or designee) will determine if they are clinically significant. Site standard ranges will be used to determine if any parameters are considered out-of-range. At the discretion of the PI (or designee), the ECG may be repeated if erroneous readings are suspected.

7.2.3. Adverse Events and Concomitant Medications

Adverse Events: The PI is responsible for identifying and documenting events meeting the definition of an AE or SAE (Section 8.1). Once each day while the subject is in the clinic and once during each out-patient visit, the PI (or designee) should inquire about the occurrence of AEs. The following are examples of open-ended questions that may be used to obtain this information: *"How are you feeling?"; "Have you had any medical problems recently?"; "Have you taken any new medicines since your last visit/assessment?"*

All AEs and SAEs must be documented in the source documents and recorded in the CRF.

Concomitant Medications: All medications (prescription or over-the-counter), nutritional supplements, and nutraceuticals taken by the subject from 30 days prior to dosing through the EOS visit must be recorded in the CRF. Medication information should include indication, dose, frequency, and route of administration. Any medication taken for an AE/SAE should be documented as such. Refer to Section 5.5 for additional information.

7.3. Laboratory Evaluations

The laboratory will perform standard routine testing, and processing of all blood samples. For the entire study, the amount of blood collected from any one subject will not exceed 500 mL.

7.3.1. Routine Laboratory Panels

Blood and urine samples will be collected at the times indicated in the Time and Events Schedule. The analytes shown in the table below (Table 11) will be assessed.

If a method to determine COVID-19 status becomes readily available, subjects may be tested at Screening and Check-in to confirm they are not positive for COVID-19 prior to dosing.

Table 11: Laboratory Evaluations

CHEMISTRY PANEL Alanine Aminotransferase (ALT/SGPT) Albumin, Serum Albumin/Globulin (A/G) Ratio (calculation) Alkaline Phosphatase, Serum Amylase Aspartate Aminotransferase (AST/SGOT) Bilirubin, Total and Direct BUN BUN/Creatinine Ratio (calculation) Calcium, Serum Creatinine, Serum Creatinine Kinase (CK) Gamma Glutamyl Transferase (GGT) Lactate Dehydrogenase (LDH) Uric Acid Electrolyte Panel (Na+, K+, Cl-, Bicarb.) Phosphorus Globulin, Total Glucose, Serum Lipase Protein, Total, Serum	**HEMATOLOGY PANEL** Hemoglobin Hematocrit White Blood Cell Count with differential (absolute and percentage) Red Blood Count Prothrombin Time (PT)/Partial Prothrombin Time (PTT) and International Normalized Ratio (INR) Mean cell hemoglobin (MCH), mean cell hemoglobin concentration (MCHC), mean cell volume (MCV), red cell distribution width (RDW; may be a Grade 1 abnormality) Platelets
	ADDITIONAL ASSESSMENTS Virology: Human Immunodeficiency Virus (HIV) serology, Hepatitis B Virus (HBV; Surface Antigen [HBsAg]), Hepatitis C Virus (HCV) Follicle-Stimulating Hormone (FSH; as applicable)
	PREGNANCY TEST Serum Pregnancy Test Urine Dipstick (optional blood follow-up)
	DRUG SCREENING Serum/urinalysis (per site SOP) Cotinine Urine Dipstick Alcohol Breathalyzer
	ROUTINE URINALYSIS Bilirubin Color and appearance Glucose Ketones Leukocytes Microscopic (including red blood cells [RBCs] and white blood cells [WBCs]) Nitrite Occult blood pH Protein Specific Gravity Urobilinogen

7.3.2. Pharmacokinetic Sampling

Samples for PK analysis will be collected as shown in the Time and Events Schedule. All samples will be analyzed to define the PK parameters for EIDD-2801 and the active metabolite, EIDD-1931. In order to preserve EIDD-2801, special handling procedures will be put in place. These procedures will be documented in the Laboratory Manual for the study.

Depending on ongoing review of the PK data, timepoints may be modified or removed, and up to 3 additional samples (in each period for the food-effect) may be added.

7.3.3. Peripheral Blood Mononuclear Cell Collection

Peripheral blood mononuclear cells will be collected in P2. Blood will be collected into specialized tubes and processed according to procedures described in the Laboratory Manual.

Depending on ongoing review of the data, timepoints may be modified or removed, and up to 2 additional samples may be added.

7.3.4. Urine Collection

Urine will be collected over the time periods noted in the Time and Events Schedule. Samples will be collected for routine urinalysis and PK analysis according to site standard practices and as described in the Laboratory Manual.

8. SAFETY MONITORING, MANAGEMENT AND REPORTING

8.1. Definitions

8.1.1. Adverse Events

An AE is 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 an abnormal laboratory finding), symptom, or disease temporally associated with the use of a medicinal (investigational) product, whether or not related to the medicinal (investigational) product.

Abnormal clinical laboratory findings (e.g. clinical chemistry, hematology) or other abnormal assessments that are judged by the PI (or designee) as clinically significant will be recorded as AEs or SAEs if they meet the definitions of an AE or an SAE as defined in this Section 8.1.2. Disease specific signs and symptoms which were ongoing prior to study entry will not be considered AEs unless they worsen (e.g. increase in frequency or severity) unexpectedly during the course of the trial.

8.1.2. Serious Adverse Events

An AE or suspected adverse reaction is considered "serious" if, in the view of either the PI (or designee) or Sponsor, it results in any of the following outcomes:

- Death,
- A life-threatening AE: An AE or suspected adverse reaction is considered "life-threatening" if, in the view of either the PI or Sponsor, its occurrence places the patient or subject at immediate risk of death. It does not include an AE or suspected adverse reaction 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 hospitalizations may be considered serious when, based upon appropriate medical judgment they may jeopardize the patient or 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.

Suspected adverse reaction means any adverse event for which there is a reasonable possibility that the drug caused the adverse event. “Reasonable possibility” means there is evidence to suggest a causal relationship between the drug and the adverse event. Suspected adverse reaction implies a lesser degree of certainty about causality than adverse reaction, which means any adverse event caused by a drug.

An AE or suspected adverse reaction is considered “unexpected” if

- It is not listed in the IB or is not listed at the specificity or severity that has been observed; or, if an IB is not required or available,
- Is not consistent with the risk information described in the general investigational plan or elsewhere in the current application as amended. For example, under this definition, hepatic necrosis would be unexpected (by virtue of greater severity) if the IB referred only to elevated hepatic enzymes or hepatitis. Similarly, cerebral thromboembolism and cerebral vasculitis would be unexpected (by virtue of greater specificity) if the IB listed only cerebral vascular accidents.
- “Unexpected” as used in this definition, also refers to AE or suspected adverse reactions that are mentioned in the IB as occurring with a class of drugs or as anticipated from the pharmacological properties of the drug, but are not specifically mentioned as occurring with the particular drug under investigation.

As of the date of this protocol, there are no expected events listed in the current version of the IB; therefore, all AEs will be considered unexpected until such a time that the reference safety information in the IB is updated with any identified, expected events.

8.2. Documenting Adverse Events

8.2.1. Timeframe for Collection and Follow-up of AEs/SAEs

All AEs/SAEs will be collected from the time of the first study drug administration until the subject has completed the EOS visit and been discontinued from the study. This includes subjects who discontinue early. Events considered related to study drug will be followed as noted:

- AEs that are related to study drug and continue beyond the normal collection period (i.e., are ongoing at the time a subject exits the study) will be followed until resolution, return to baseline or until stabilized with sequelae for a maximum of 30 days following discontinuation. After 30 days, the AE will be closed, and the outcome noted (see Table 12).
- SAEs that are related to study drug and continue beyond the normal collection period

(i.e., are ongoing at the time a subject exits the study) will be followed until resolution, return to baseline or until stabilized with sequelae.

- SAEs that are reported to the site within 30 days after the subject has been discontinued from the study (i.e., completed the EOS visit) will be recorded. Those that are considered related to study drug will be followed as noted in the bullet above.

Note that all events which occurred prior to dosing with study drug should be recorded as medical history unless the event is directly related to study procedures.

8.2.2. Recording of Adverse Events/Serious Adverse Events

AEs/SAEs must be recorded in the CRF as indicated in the CRF completion instructions. Information to be collected includes event description, time of onset, clinician's assessment of severity (Section 8.2.3), relationship to study drug (Section 8.2.4), outcome (Section 8.2.5), and time of resolution/stabilization of the event. All AEs occurring while on study must be documented appropriately regardless of relationship and will be followed to adequate resolution as described above (Section 8.2.1). All SAEs will be recorded as noted above; SAEs reported to the site within 30 days following the EOS visit will also be recorded. All SAEs must be entered onto the SAE form and reported as discussed below (Section 8.3.1).

If an AE changes in severity, the highest severity will be recorded in the CRF. AEs characterized as intermittent require documentation of onset and duration of each episode.

8.2.3. Assessing Severity of Adverse Events

All AEs/SAEs will be assessed by the PI or those with the training and authority to make a medical judgment. AEs/SAEs will be graded according to the DMID Toxicity Grading Scale. For any AEs not specifically listed in the tables, the following guidelines should be used to grade severity:

- **Mild** (Grade 1); asymptomatic or mild symptoms; clinical or diagnostic observations only; intervention not indicated.
- **Moderate** (Grade 2); minimal, local or noninvasive intervention indicated; limiting age-appropriate instrumental activities of daily living.
- **Severe** (Grade 3); medically significant but not immediately life-threatening; hospitalization or prolongation of hospitalization indicated; disabling; limiting self-care activities of daily living.
- **Life-threatening** (Grade 4) Life-threatening consequences; urgent intervention indicated.
- **Death** (Grade 5) Death related to AE.

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

8.2.4. Relationship to Study Drug

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

- **Definite –** The AE is clearly related to the study drug**.**
- **Probable –** The AE is likely related to the study drug.
- **Possible –** The AE may be related to the study drug.
- **Unlikely –** The AE is doubtfully related to the study drug.
- **Unrelated –** The AE is clearly NOT related to the study drug.

8.2.5. Classifying Adverse Event Outcome

All AEs/SAEs in the study must be assigned an outcome by site staff. The outcome will be included on the AE CRF. Possible outcomes are shown below:

Table 12: Adverse Event Outcomes

Outcome	Description
Recovered / Resolved	AE resolved with no residual signs or symptoms; an event is considered resolved if it returns to baseline (pretreatment) values.
Recovered / Resolved with sequelae	AE stabilized but residual signs or symptoms remain; this includes stabilization of an event/condition with the expectation that it will remain chronic.
Not Recovered / Not Resolved	AE remains ongoing AND no or only minimal improvement has occurred.
Ongoing	AE has not yet resolved, but continues to improve/resolve and complete resolution is expected over time.
Fatal	Outcome of the AE is death.
Unknown	AE outcome is not known; usually because the subject has been Lost-to-Follow-up.

8.3. Reporting Procedures

8.3.1. Serious Adverse Events

The PI or clinical site personnel should notify Covance Drug Safety Services (DSS) of all SAEs, regardless of relationship to the investigational drug, within 24 hours of clinical site personnel becoming aware of the event. The PI (or designee) will provide the initial notification by sending a completed "SAE Notification Form," which must include the PI's (or designee's) assessment of the relationship of the event to investigational drug and must be signed by the PI. Follow-up information, or new information regarding an ongoing SAE, must be provided promptly to Covance DSS.

The responsibilities of Covance DSS include the following:

- Prepare an AE reporting plan prior to the start of the study. Where this plan differs from the applicable site standard operating procedure on SAE reporting, the AE reporting plan will always take precedence.

- Receive and review SAE report forms from the site and inform the Sponsor of the SAE within 1 working day of the initial notification to DSS. Drug Safety Services will delete any information from the SAE report forms that may identify the subject.

- Write case narratives and enter the case into Covance's safety database as defined in the AE reporting plan.

- Produce appropriate reports of all Suspected Unexpected Serious Adverse Reactions and forward to the EC, Medicines and Healthcare Products Regulatory Agency, PI's (or designee's), and the Sponsor within the timeframes stipulated in the Clinical Trials Directive Guideline (ENTR/CT 3).

The responsibility for reporting SAEs will be transferred to the Sponsor 28 days after the end of the study.

8.3.2. Pregnancy

Subjects in this study must be of non-childbearing potential. However, should a pregnancy occur any time from informed consent to the EOS visit, subjects must immediately report the event to the clinical site which in turn must immediately report the pregnancy to the Sponsor or their designee. The subject will be followed until the end of the pregnancy. A separate ICF will be used for consenting for follow-up pregnancy activities. Pregnancy will not be considered an AE unless deemed likely related to study drug. Pregnancy will not be considered an SAE unless there is an associated SAE. A spontaneous abortion (miscarriage) or abnormal outcome (including congenital anomalies) will be reported as an SAE.

8.4. Unmasking Treatment Assignment

The following controls will be employed to maintain the double-blind status of the study:

- Placebo will be identical in appearance to the EIDD-2801.
- The PI and other members of staff involved with the study will remain blinded to the treatment randomization code in P1.
- Interim bioanalytical data will be provided in a blinded manner.

To maintain the blind, the PI will be provided with a sealed randomization code for each subject in P1, containing details of their treatment. These individual sealed envelopes will be kept in a limited access area that is accessible 24 hours a day. In order to manage subject safety or to support dose-escalation decisions (in the event of possibly treatment-related SAEs or severe AEs), the decision to unblind resides solely with the PI. Whenever possible and providing it does not interfere with or delay any decision in the best interest of the subject, the PI will discuss the intended code-break with the Sponsor. If it becomes necessary to break the code during the study, the date, time, and reason will be recorded in the subject's source data and on the individual envelope and will be witnessed by a second person.

The pharmacy and bioanalytical lab will have access to the treatment randomization and will be unblinded. Pharmacokinetic personnel may be unblinded to perform interim PK analysis and to ensure that PK data are provided in a blinded manner for dose-escalation decisions.

9. DOSE-ESCALATION AND HALTING RULES

9.1. Guidelines for Dose-Escalation

Doses will be administered in an escalating manner following satisfactory review by the Sponsor and PI of the blinded safety and tolerability data up to 72 hours post final dose. [redacted] As PK data become available, these data will be reviewed and may be used to assist in dose selection.

Doses may be reduced and may be lower than the starting dose. There will be a minimum of 4 days between dose escalations to allow sufficient time for an adequate safety review.

Dose-escalation in P1 will only occur if data from a minimum of 6 subjects have been reviewed from the previous lower dose group, such that data from a minimum of 4 subjects who have received EIDD-2801 will be used to make the dose-escalation decision.

The justification for this is as follows:

- The study treatment is of a known pharmacological class for which the on-target effects in humans are well characterized. Based upon nonclinical data, no clinically important off target effects are expected within the proposed dose range.

- A minimum of 4 subjects receiving the active drug is considered sufficient to characterize the safety profile of EIDD-2801.

Between each dose-escalation, the PI will review all available data in a blinded manner to ensure it is safe to proceed with the planned dose-escalation. The results from all available safety assessments will be sent to the Sponsor prior to the start of each successive group/treatment period. Any clinically significant results will be discussed with the Sponsor before dose-escalation continues. Interim PK data may also be reviewed in terms of dose-escalation and to confirm that the study design remains appropriate. In the event of a disagreement between Sponsor and PI on the dose-escalation decision, the most conservative decision will be upheld.

Dose levels will not increase by more than 3-fold for predicted nonpharmacologically active dose levels and by 2-fold for predicted pharmacologically active dose levels. [redacted]

9.2. Dose-Escalation Halting Rules

For Group 1 in P1/SAD, sentinel dosing will occur whereby 2 subjects (1 active and 1 PBO) will be dosed on 1 day and, providing no safety concerns arise, the remaining 6 subjects will be dosed after at least 24 hours.

The study will be halted (i.e., no further dosing will occur) if one or more subjects experience an SAE that is considered to be related to EIDD-2801 or 2 or more subjects in the same group experience severe AEs that are considered to be related to EIDD-2801. If, following an internal safety review, the Sponsor deems it appropriate to restart the study, this can be done following approval of a substantial protocol amendment.

Dose-escalation will stop if clinically relevant signs or symptoms of similar nature occur in 2 or more subjects in a group that, in the opinion of the PI, warrant stopping of dose-escalation.

10. STATISTICAL CONSIDERATIONS

10.1. General Considerations

All summaries will be provided by study part and treatment. Continuous variables will be summarized using descriptive statistics including number of observations (n), mean, standard deviation, minimum (min), median (med), and maximum (max). Categorical variables will be summarized using frequency counts and percentages. Note that PBO data from each dose level in Part 1 will be combined into one PBO group.

No missing data imputation will be performed.

Subject listings will be provided for all the data collected during the study period.

Specific information about the statistical analysis will be provided in a SAP that will be reviewed and approved by the Sponsor and will be finalized before final database lock. If there is a discrepancy between the methods described in the protocol and final approved SAP, the SAP will take precedence.

10.2. Sample Size Considerations

No formal sample size calculation was conducted. The sample size of 8 per cohort (6 active: 2 PBO) for the SAD cohorts is considered adequate for a Phase 1 FIH study. The sample size of 10 subjects (all administered EIDD-2801) is in accordance with FDA guidelines for sample size in FE studies.

10.3. Analysis Populations

Safety Population: All subjects who receive at least one dose of study drug.

Pharmacokinetic Population: All subjects receiving study drug who comply sufficiently with protocol requirements (i.e., no major protocol violations) and for whom a sufficient number of analyzable PK samples have been obtained to permit the determination of the PK profile for EIDD-2801/EIDD-1931 and the statistical analysis of the results. Subjects who experience an AE of vomiting before 2× the median t_{max} of the group may be excluded from the PK population.

10.4. Analysis of Safety Data

All safety analyses will be performed on the Safety Population as defined in Section 10.3. Safety will be assessed on the basis of AEs, clinical laboratory data, vital signs, ECG parameters, and PEs.

10.4.1. Extent of Exposure

Dosing data will be listed by study part.

10.4.2. Adverse Events

Adverse events will be coded using Medical Dictionary for Regulatory Activities (MedDRA) preferred term and system organ classification. Any events reported after the initiation of study treatment and through the EOS are defined as treatment-emergent. The occurrence of treatment-emergent AEs will be summarized using MedDRA preferred terms, system organ classifications, and severity. Separate summaries of treatment-emergent SAEs and AEs considered related to study treatment and AEs leading to study treatment discontinuation will be generated. All AEs will be listed for individual subjects showing both verbatim and preferred terms.

10.4.3. Clinical Laboratory Results

Laboratory abnormalities will be graded according to the DMID Toxicity Grading Scale. Any graded abnormality that occurs following the initiation of study treatment and represents at least a one-grade increase from the baseline assessment is defined as treatment-emergent. The number and percentage of subjects experiencing treatment-emergent graded toxicities will be summarized. Raw values and mean changes from baseline in clinical laboratory measures will be summarized.

Listings of the clinical laboratory test results will be provided. Abnormal laboratory values will be flagged in the listings.

10.4.4. Other Parameters

Individual data for ECG parameters and vital sign measurements will be listed by subject and time point and summarized for each treatment. Individual data for PE will be listed by subject and time point.

Concomitant medications will be coded using the World Health Organization-Drug Dictionary (WHO-DD) and summarized.

10.5. Analysis of Pharmacokinetic Data

Pharmacokinetic analysis as defined in the SAP will be conducted using the PK population defined in Section 10.3. In the event of discrepancies between analyses described in the SAP and this clinical study protocol, the SAP will supersede the protocol.

- All samples will be analyzed and all concentrations listed.
- Descriptive statistics will be performed for all time points available, with the exclusion of subjects who had any significant protocol deviation.
- Pharmacokinetic parameters will be derived where possible for all subjects. Data from subjects with incomplete profiles (missed blood draws, lost samples, samples unable to be quantified) may be used if PK parameters can be estimated using the remaining data points.

- Descriptive statistics will be performed on all parameters available, and any missing parameters will be flagged.

Plasma concentration data for EIDD-2801 and EIDD-1931 will be listed for individual subjects and summarized by study part and treatment. Individual and mean plasma concentration versus time plots for EIDD-2801 and EIDD-1931 will be provided. Urine concentration data for EIDD-2801 and EIDD-1931 will be listed.

Plasma PK parameters of EIDD-2801 and EIDD-1931 for each subject will be estimated over the sampling interval using noncompartmental analysis and summarized by study part and treatment using descriptive statistics. Actual blood sampling times will be used for plasma PK analysis.

Urine PK parameters of EIDD-2801 and EIDD-1931 will also be analyzed and summarized when possible. The PK parameters that will be estimated are listed in the table below.

Table 13: Pharmacokinetic Parameters

Plasma PK Parameter	Description
AUC_{last}	The area under the plasma concentration-time curve, from time 0 to the last measurable non-zero concentration, as calculated by the linear up/log down trapezoidal method.
$AUC_{0\text{-}inf}$	The area under the plasma concentration-time curve from time 0 extrapolated to infinity. $AUC_{0\text{-}inf}$ is calculated as the sum of AUC_{last} plus the ratio of the last measurable plasma concentration to the elimination rate constant (λz).
C_{max}	Maximum observed concentration.
t_{max}	Time to reach C_{max}. If the maximum value occurs at more than one time point, t_{max} is defined as the first time point with this value.
λz	Apparent terminal elimination rate constant; represents the fraction of medication eliminated per unit time.
t½	Apparent terminal elimination half-life of medication, calculated as 0.693/λz.
Vz/F	Apparent volume of distribution (EIDD-2801 only).
CL/F	Apparent oral drug clearance (EIDD-2801 only).
Urine PK Parameter	**Description**
A_e	Amount excreted in urine over the sampling interval.
F_e	Fraction of drug excreted in the urine over sampling interval.
CL_R	Renal clearance, $CL_R = A_e/AUC$ where both A_e and AUC are determined over co-incident time ranges after dosing.

Additional PK parameters may be analyzed as appropriate.

10.5.1. Statistical Analysis of Pharmacokinetic Data

10.5.1.1. Dose Proportionality

Dose proportionality for PK parameters of EIDD-2801 and EIDD-1931 ($AUC_{0\text{-}inf}$, AUC_{last}, and C_{max}) will be assessed by the power model. The mean slope and the associated 90% CI based on the power model will be reported.

10.5.1.2. Food-Effect

To assess the effect of food on the PK of EIDD-2801 and EIDD-1931, natural log transformed PK parameters from P2 ($AUC_{0\text{-}inf}$, AUC_{last}, and C_{max}) will be analyzed using a mixed effects model with fixed-effect terms for treatment (with or without food), period, and sequence and with subject within sequence as a random effect. Point estimates and their associated 90% CIs for the natural log transformed PK parameters for the difference between EIDD-2801 administered under fed relative to fasted conditions will be constructed. The point estimates and interval estimates will be back transformed to give estimates of the ratio and 90% CI for the geometric least squares mean ratio for fed/fasted to assess the effect of food.

10.6. Peripheral Blood Mononuclear Cells

PBMCs will be collected from subjects following each dose in Part 2.The PBMCs will be stored for analysis of EIDD-2061 levels at some point in the future and reported separately.

10.7. Interim Analyses

There are no formal interim analyses planned for this study. However, interim analyses may be implemented at the discretion of the Sponsor or health authority request. In addition, data from P1 cohorts may be locked, unblinded, and analyzed prior to conducting P2.

11. STUDY ADMINISTRATION

11.1. Regulatory and Ethical Considerations

11.2. Ethical Standard

This study will be conducted in accordance with the protocol and with the following:

- Consensus ethical principles derived from international guidelines including the Declaration of Helsinki and Council for International Organizations of Medical Sciences International Ethical Guidelines.
- Applicable ICH GCP Guidelines.
- United States Code of Federal Regulations applicable to clinical studies (45 CFR Part 46; 21 CFR Part 50, 21 CFR Part 56, and 21 CFR Part 312).
- Applicable laws and regulations.

11.2.1. Ethics Committee Approval

The PI (or designee) must ensure that all required study-specific documents and/or information are submitted to the EC for review and approval as appropriate including but not limited to:

- the protocol and any future protocol amendments
- ICF and any other documents (electronic or paper) given to the subject
- IB

Any substantial protocol amendments, likely to affect the safety of the subjects or the conduct of the study, will require EC and regulatory authority (as locally required) approval before implementation of changes made to the study design, except for changes necessary to eliminate an immediate hazard to study subjects or any nonsubstantial changes, as defined by regulatory requirements.

11.2.2. Informed Consent

Informed consent is a process that is initiated prior to the individual's agreeing to participate in the study and continuing throughout the individual's study participation. Extensive discussion of risks and possible benefits of this therapy will be provided to the subjects and, if applicable, their families. Consent forms describing in detail the study interventions/products, study procedures, and risks are given to the subject and written documentation of informed consent is required prior to starting any screening procedures or intervention/administering study product. Consent forms will be EC-approved and the subject will be asked to read and review the document.

Upon reviewing the document, the PI (or designee) will explain the research study to the subject and answer any questions that may arise. The subjects will sign the ICF prior to any procedures being done specifically for the study. The subjects should have the opportunity to discuss the study with their surrogates or think about it prior to agreeing to participate. The subjects may withdraw consent at any time throughout the course of the trial. A copy of the ICF will be given to the subjects for their records. The rights and welfare of the subjects will be protected by emphasizing to them that the quality of their medical care will not be adversely affected if they decline to participate in this study.

11.3. Financing and Insurance

Financing and insurance will be addressed in a separate agreement.

11.4. Source Documentation and Access

The site will maintain appropriate medical and research records in compliance with ICH E6, Section 4.9 and regulatory and institutional requirements for the protection of confidentiality of subjects. The site will permit authorized representatives of the Sponsor, its designees, and appropriate regulatory agencies to examine (and when required by applicable law, to copy) clinical records for the purposes of quality assurance reviews, audits, and evaluation of the study safety and progress. These representatives will be permitted access to all source data and source documents, which include, but are not limited to, clinical and office charts, laboratory notes, memoranda, subjects' memory aid or evaluation checklists, pharmacy dispensing records, recorded data from automated instruments, copies or transcriptions certified after verification as being accurate and complete, photographic negatives, microfilm or magnetic media, x-rays, and subject files and records kept at the pharmacy, at the laboratory, and medico-technical departments involved in the clinical trial.

11.5. Data Collection and Record Keeping

11.5.1. Data Collection

Data collection and data entry are the responsibility of the clinical trial staff at the site. The PI is responsible for ensuring the accuracy, completeness, legibility, and timeliness of the data reported. All source documents should be completed in a neat, legible manner to ensure accurate interpretation of data.

A CRF must be completed for every subject who signs the ICF and has at least one protocol-specified assessment conducted. The CRF must be completed and processed according to the CRF guidelines and the SOPs of the site. All data should be entered into the CRF, where possible, within 3 days after each visit for any one subject. After the subject has completed the study, the PI must review and sign the signature page of the CRF indicating that he has reviewed the completed CRF and pertinent clinical data for that subject and that, to the best of his knowledge, all data recorded in the CRF accurately reflects the subject's clinical performance in the study

11.5.2. Study Records Retention

Study documents should be retained for a minimum of 5 years after the end of the study. These documents should be retained for a longer period; however, if required by local regulations. No records will be destroyed without the written consent of the Sponsor, if applicable. It is the responsibility of the Sponsor to inform the PI when these documents no longer need to be retained.

11.5.3. Protocol Deviations

A protocol deviation is any noncompliance with the protocol or study procedures detailed in the Laboratory or Pharmacy Manuals. The noncompliance may have been the result of action by the PI, site staff, or subject. All deviations should be handled in accordance with site SOPs.

It is the responsibility of the site to use continuous vigilance to identify and report deviations within 5 working days of identification of the protocol deviation, or within 5 working days of the scheduled protocol-required activity.

11.6. Clinical Monitoring

Site monitoring is conducted to ensure that the human subjects' protections, study and laboratory procedures, study intervention administration, and data collection processes are of high quality and meet Sponsor, ICH/GCP guidelines and applicable regulations, and that this trial is conducted in accordance with the protocol, protocol-specific MOP and applicable Sponsor SOPs. Experienced clinical monitors of the Sponsor or its designee will conduct site-monitoring visits as detailed in the clinical monitoring plan.

Site visits will be made at standard intervals as defined by the Sponsor and may be made more frequently as directed by the Sponsor. Monitoring visits will include, but are not limited to, review of regulatory files, accountability records, CRFs, ICFs, medical and laboratory reports, and protocol and GCP compliance. Clinical monitors will have access to each participating site, study personnel, and all study documentation according to the site monitoring plan. Clinical monitors will meet with the site PI to discuss any problems and actions to be taken and will document site visit findings and discussions. As data are entered into a CRF, data will be monitored to ensure accuracy as well as adherence to data entry instructions provided in the CRF guidelines.

11.7. Quality Control and Quality Assurance

A quality management plan will be put in place for this study. The site is responsible for conducting routine quality assurance and quality control activities to internally monitor study progress and protocol compliance. The PI will provide direct access to all study-related sites, source data/data collection forms, and reports for the purpose of monitoring and auditing by the Sponsor, and inspection by local and regulatory authorities. The PI will ensure all study personnel are appropriately trained and applicable documentations are maintained on site.

11.8. Study Termination and Closure Procedures

11.8.1. Study Termination

This study may be temporarily suspended or prematurely terminated if there is sufficient reasonable cause. Written notification, documenting the reason for study suspension or termination, will be provided to the site. If the study is terminated or suspended, the PI (or designee) will inform study participants and the EC. The Sponsor will notify appropriate regulatory authorities. The Sponsor will provide the reason(s) for the termination or suspension. Study participants will be contacted, as applicable, and be informed of changes to study visit schedule.

If suspended, the study may resume once issues that caused suspension of the study are resolved.

11.8.2. Termination Procedures

If the study is prematurely terminated, then the site must return all appropriate study data, resolve all data queries, complete final drug accountability, return any study drug remaining on site, and ship all biological samples (including PK and PBMCs) to the laboratory designated by the Sponsor. The PI (or designee) must notify the EC of study termination.

11.9. Information Disclosure

11.9.1. Confidentiality

Subject confidentiality and privacy is strictly held in trust by the PI, site staff, and the Sponsor(s) and their agents. The study protocol, documentation, data, and all other information generated will be held in strict confidence. No information concerning the study or the data will be released to any unauthorized third party without prior written approval of the Sponsor.

The study monitor or other authorized representatives of the Sponsor or regulatory agencies may inspect all documents and records required to be maintained by the PI, including but not limited to, medical records (office, clinic, or hospital) and pharmacy records for the participants in this study. The clinical study site will permit access to such records.

The study participant's contact information will be securely stored at the clinical site for internal use during the study. At the end of the study, all records will continue to be kept in a secure location for as long a period as dictated by the Sponsor, site, or regulatory requirements.

Study participant research data will be transmitted to and stored securely by the Sponsor's designated data center. This will not include the participant's contact or identifying information. Individual participants and their research data will be identified by a unique study identification number. The study data entry and study management systems used by clinical sites will be secured and password protected. At the end of the study, all study databases will be de-identified and archived at a secure location.

11.9.2. Clinicaltrials.gov

This clinical study will be registered on clinicaltrials.gov as required.

11.9.3. Publication Policy

All information generated from this study is the proprietary property of the Sponsor. It is the intent of the Sponsor to publish the results of the study in their entirety as deemed appropriate.

12. REFERENCES

Agostini ML, Pruijssers AJ, Chappell JD, Gribble J, Lu X, Andres EL, et al. Small-Molecule Antiviral β-d-N4-Hydroxycytidine Inhibits a Proofreading-Intact Coronavirus with a High Genetic Barrier to Resistance. J Virol. 2019 Dec 15;93(24).

Flavell RA, Sabo DL, Bandle EF, Weissmann C. Site-directed mutagenesis: generation of an extracistronic mutation in bacteriophage Q beta RNA. J Mol Biol. 1974 Oct 25;89(2):255–72.

Hernandez-Santiago BI, Beltran T, Stuyver L, Chu CK, Schinazi RF. Metabolism of the anti-hepatitis C virus nucleoside beta-D-N4-hydroxycytidine in different liver cells. Antimicrob Agents Chemother. 2004 Dec;48(12):4636–42.

Mullins JI, Heath L, Hughes JP, Li D, Essigmann JM, Harris KS, et al. Mutations based on viral decay acceleration in the HIV-1 genomes of a clinical population treated with the mutagenic nucleoside KP1461. Retrovirology. 2011 Oct 3;8(Suppl 2):P84.

Painter GR, Bowen RA, Bluemling GR, DeBergh J, Edpuganti V, Gruddanti PR, et al. The prophylactic and therapeutic activity of a broadly active ribonucleoside analog in a murine model of intranasal venezuelan equine encephalitis virus infection. Antiviral Res. 2019;171:104597.

Sticher ZM, Lu G, Mitchell DG, Marlow J, Moellering L, Bluemling GR, et al. Analysis of the Potential for N4-Hydroxycytidine To Inhibit Mitochondrial Replication and Function. Antimicrob Agents Chemother. 2020 27;64(2).

Stuyver LJ, Whitaker T, McBrayer TR, Hernandez-Santiago BI, Lostia S, Tharnish PM, et al. Ribonucleoside analogue that blocks replication of bovine viral diarrhea and hepatitis C viruses in culture. Antimicrob Agents Chemother. 2003 Jan;47(1):244–54.

Toots M, Yoon J-J, Cox RM, Hart M, Sticher ZM, Makhsous N, et al. Characterization of orally efficacious influenza drug with high resistance barrier in ferrets and human airway epithelia. Sci Transl Med. 2019 Oct 23;11(515):eaax5866.

Urakova N, Kuznetsova V, Crossman DK, Sokratian A, Guthrie DB, Kolykhalov AA, et al. β-d-N4-Hydroxycytidine Is a Potent Anti-alphavirus Compound That Induces a High Level of Mutations in the Viral Genome. J Virol. 2018 01;92(3).

Yoon J-J, Toots M, Lee S, Lee M-E, Ludeke B, Luczo JM, et al. Orally Efficacious Broad-Spectrum Ribonucleoside Analog Inhibitor of Influenza and Respiratory Syncytial Viruses. Antimicrob Agents Chemother. 2018;62(8).

13. APPENDICES

Appendix 1: Time and Events Schedule

Parts 1 and 2

Visit	Screening (Days -28 to -2)	Day -1	Day 1	Day 2 (24 hr)	Day 3 (48 hr)	Day 4 (72 hr)	Day 9	Day 15 / EOS[1] (non-FE cohorts)
Food-Effect Cohort		**Day 14**	**Day 15**	**Day 16 (24 hr)**	**Day 17 (48 hr)**	**Day 18 (72 hr)**	**Day 23**	**Day 30 / EOS**
ICF; Demography	x							
I/E; Medical history	x	x						
Physical examination[2]	x	x		x		x	x	x
Qualifying laboratory analyses[3]	x	x						
Drug screening and pregnancy test[4]	x	x					x	x
Height, weight (BMI)	x							
Clinic confinement[5]		x	x	x	x	x		
Non-residential visit	x						x	x
Clinical chemistry and urinalysis[6]	x	x		x		x	x	x
Hematology[7]	x	x		x	x	x	x	x
PBMC collection (FE cohort ONLY)			x[8]	x[8]				
ECG	x		x[9]			x		x
Vital signs	x	x	x[10]	x	x	x	x	x
Administer study drug			x					
Plasma PK sample collection			x[11]	x[12]	x[12]	x[12]		
Urine PK sample collection			x[13]	x[14]	x[14]			
Adverse events	x[15]	x[15]	x[15]	x	x	x	x	x
Prior and/or Concomitant medications	x	x	x	x	x	x	x	x

Abbreviations: BMI (body mass index); EOS (End of Study); FE (food-effect); ICF (Informed Consent Form); I/E (Inclusion/Exclusion Criteria); ECG (electrocardiogram); PBMC (peripheral blood mononuclear cell); PK (pharmacokinetic).

[1] On Day 15, conduct the EOS visit for all subjects except those in the FE cohort.

[2] A complete PE will be performed at Screening and/or Check-in, and EOS visits; on all other days, a limited PE will be conducted targeting any change from the previous status or any body system deemed pertinent to that subject by the PI (or designee).

[3] Laboratory analyses to qualify for study entry include: total white cell count or absolute lymphocyte count (values must be in the normal range), hemoglobin or platelet levels (values must not be below the lower limit of normal), and ALT/SGPT, alkaline phosphatase, AST/SGOT (values must not be above the upper limit of normal), plus virology (HIV/HCV/HBV) (all must be negative), and FSH (if required).

[4] Drug screening (urine drug screen, UDS), including a cotinine test, should be conducted at Screening, Day -1, Days 9 and 23, and at EOS. Pregnancy tests should be conducted in serum at Screening and in urine at all other times (i.e., Days -1, 9, and 23, and EOS). A positive urine pregnancy test will be confirmed with a serum pregnancy test. An alcohol breath test should be conducted at Screening and Day -1.

[5] Participants should be admitted to the clinic following review of final I/E criteria on Day -1 and discharged from the clinic following completion of study specific assessments on Day 4. Subjects in the FE cohort should be readmitted to the clinic on Study Day 14.

[6] Safety laboratory assessments include clinical chemistry and urinalysis. Baseline samples will be collected after clinic admission on Day -1.

[7] Hematology values include, but are not limited to, RBC, WBC with differential and platelets. PT/PTT and INR assessed only at

Screening.

[8] PBMCs will be collected 2- and 12-hr postdose on Day 1/15 and 24 hrs postdose (on Day 2/16). Depending on ongoing review of the data, timepoints may be modified or removed, and up to 2 additional samples may be collected.

[9] The baseline ECG should be conducted prior to dosing on Day 1/15.

[10] Vital signs include pulse rate, respiratory rate, blood pressure, and body temperature; on Day 1/15, VS should be taken predose, and at 2, 4, 8 and 12 hr postdose.

[11] PK samples should be collected predose and at 0.5, 1, 1.5, 2, 2.5, 3, 4, 6, 9, 12, and 15 hr postdose on Day 1/15. Depending on ongoing review of the PK data, timepoints may be modified or removed, and up to 3 additional samples (in each period for the food-effect) may be added.

[12] PK samples should be collected at 24, 36, 48, and 72 hr postdose on Days 2/16, 3/17, 4/18 respectively. Depending on ongoing review of the PK data, timepoints may be modified or removed, and up to 3 additional samples (in each period for the food-effect) may be added.

[13] Urine samples for PK analysis should be collected predose (spot collection), and from 0 to 4, 4 to 8, 8 to 12, and 12 to 24 hr postdose.

[14] Urine samples for PK analysis should be collected from 24 to 36 and 36 to 48 hr postdose.

[15] Adverse events occurring prior to study drug administration should be recorded as medical history unless the event is directly related to a study specified procedure.

Appendix 2: DMID Toxicity Grading Scale

Clinical Adverse Events			
VITAL SIGNS	**Mild (Grade 1)**	**Moderate Grade 2)**	**Severe (Grade 3)**
Fever (°C) Oral temperature; no recent hot or cold beverages or smoking.	38.0 – 38.4	38.5 – 38.9	>39.0
Tachycardia - beats per minute	101 – 115	116 – 130	> 130 or ventricular dysrhythmias
Bradycardia - beats per minute	50 – 54 or 45-50 bpm if baseline <60 bpm	45 – 49 or 40-44 if baseline <60 bpm	< 45 or <40 bpm if baseline <60 bpm
Hypertension (systolic) - mm Hg [Assuming supine position, 10 min at rest conditions, not sleeping subjects, measurements on the same arm and several concordant results.]	141-150	151-160	> 160
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**
Arrythmia		asymptomatic, transient signs, no Rx required	recurrent/persistent; symptomatic Rx required
Hemorrhage, Blood Loss	Estimated blood loss ≤ 100 mL	Estimated blood loss > 100 mL, no transfusion required	Transfusion required
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 60-70% (of peak flow)	no normalization with bronchodilator; FEV1 <60% 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 gms/24 hours	4 - 5 loose or watery stools or 400 - 800 gms/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	Any use of narcotic pain reliever or prevents daily

		interferes with activity	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
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
SYSTEMIC	**Mild (Grade 1)**	**Moderate (Grade 2)**	**Severe (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
All Other conditions	**Mild (Grade 1)**	**Moderate (Grade 2)**	**Severe (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

*In addition to grading the measured local reaction at the greatest single diameter, the measurement should be recorded as a continuous variable.

**Induration/Swelling should be evaluated and graded using the functional scale as well as the actual measurement

Laboratory Adverse Events			
Blood, Serum, or Plasma *	**Mild (Grade 1)**	**Moderate (Grade 2)**	**Severe (Grade 3)**
Sodium – Hyponatremia mEq/L	132 – <LLN	130 – 131	<130
Sodium – Hypernatremia mEq/L	>ULN – 148	149 – 150	>150
Potassium – Hyperkalemia mEq/L	>ULN – 5.2	5.3 – 5.4	>5.4
Potassium – Hypokalemia mEq/L	<LLN-3.1	<3.1 – 3.0	<3.0
Glucose – Hypoglycemia mg/dL	65 – 67	55 – 64	<55
Glucose – Hyperglycemia Fasting – mg/dL	>ULN - 120	121 – 130	>130
Glucose – Hyperglycemia Random – mg/dL	140 – 159	160 – 200	>200
Blood Urea Nitrogen mg/dL	23-26	27 – 31	> 31
Creatinine – mg/dL	>ULN – 1.7	1.8 – 2.0	>2.0
Calcium – hypocalcemia mg/dL	8.0 – <LLN	7.5 – 7.9	<7.5
Calcium – hypercalcemia mg/dL	>ULN – 11.0	11.1 – 11.5	>11.5
Magnesium – hypomagnesemia mg/dL	1.3 – 1.5	1.1 – 1.2	<1.1
Phosphorus – hypophosphatemia mg/dL	2.3 – 2.5	2.0 – 2.2	<2.0
CPK – mg/dL	400-1000	1001-1500	>1500
Albumin – Hypoalbuminemia g/dL	2.8 – 3.0	2.5 – 2.7	< 2.5
Total Protein – Hypoproteinemia g/dL	5.2 – 5.4	4.8 – 5.1	< 4.8
Alkaline phosphate – U/L	132-240	241– 360	>360
AST U/L	44 – 105	106-175	>175
ALT U/L	44 – 105	106-175	>175
Bilirubin (serum total) mg/dL	1.3 - 2.0	2.1 - 2.5	> 2.5
Bilirubin – when ALT ≥105 (Hy's law)	1.3 -1.5	1.6 – 2.0	> 2.0
Amylase- U/L	200-270	271-360	>360
Lipase- U/L	176-270	271-360	>360
Hemoglobin (Female) - g/dL	11.0 – 11.5	9.5 – 10.9	< 9.5
Hemoglobin (Male) - g/dL	12.0 – 12.5	10.0 – 11.9	<10.0
WBC Increase - cell/mm3	11,001 – 15,000	15,001 – 20,000	> 20,000
WBC Decrease - cell/mm3	2,500 – 3,500	1,500 – 2,499	< 1500
Lymphocytes Decrease - cell/mm3	750 – 1,000	500 – 749	< 500
Neutrophils Decrease - cell/mm3	1,500 – 2,000	1,000 – 1,499	< 1000
Eosinophils - cell/mm3	500-750	751-1500	> 1500
Platelets Decreased - cell/mm3	120,000 – 130,000	100,000 – 119,999	<100,000
PT – seconds (prothrombin time)	> ULN-14.4	14.5 – 15.7	> 15.7
PTT – seconds (partial thromboplastin time)	>ULN-42.1	42.2– 50.0	> 50.0
Fibrinogen increase - mg/dL	>ULN – 500	501 – 600	> 600
Fibrinogen decrease - mg/dL	<LLN – 140	125 – 139	<125
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	5-10	11-50	> 50 and/or gross

power field (rbc/hpf)			blood

* The laboratory values provided in the tables serve as guidelines and are dependent upon institutional normal parameters.
* Institutional normal reference ranges should be provided to demonstrate that they are appropriate.

Appendix 3: Contraception Guidance

Male subjects (even with a history of vasectomy) with partners of childbearing potential must use a male barrier method of contraception (i.e., male condom with spermicide) in addition to a second method of acceptable contraception from Check-in until 90 days after the EOS visit. Acceptable methods of contraception for female partners include:

- hormonal injection
- combined oral contraceptive pill or progestin/progestogen-only pill
- combined hormonal patch
- combined hormonal vaginal ring
- surgical method (bilateral tubal ligation or Essure® [hysteroscopic bilateral tubal occlusion])
- hormonal implant
- hormonal or non-hormonal intrauterine device
- OTC sponge with spermicide
- cervical cap with spermicide
- diaphragm with spermicide.

An acceptable second method of contraception for male subjects is vasectomy that has been performed at least 90 days prior to the screening visit, with verbal confirmation of surgical success.

For male subjects (even with a history of vasectomy), sexual intercourse with female partners who are pregnant or breastfeeding should be avoided unless condoms are used from the time of the first dose until 90 days after the EOS visit. Male subjects are required to refrain from donation of sperm from Check-in until 90 days after the EOS visit.

Sexual Abstinence and Same-sex Relationships

Subjects who practice true abstinence, because of the subject's lifestyle choice (i.e., the subject should not become abstinent just for the purpose of study participation), are exempt from contraceptive requirements. Periodic abstinence (eg, calendar, ovulation, symptothermal, postovulation methods) and withdrawal are not acceptable methods of contraception. If a subject who is abstinent at the time of signing the ICF becomes sexually active they must agree to use contraception as described previously.

For subjects who are exclusively in same-sex relationships, contraceptive requirements do not apply. If a subject who is in a same-sex relationship at the time of signing the ICF becomes engaged in a heterosexual relationship, they must agree to use contraception as described previously.

Title of study: COVID-19 Generic Procedures.

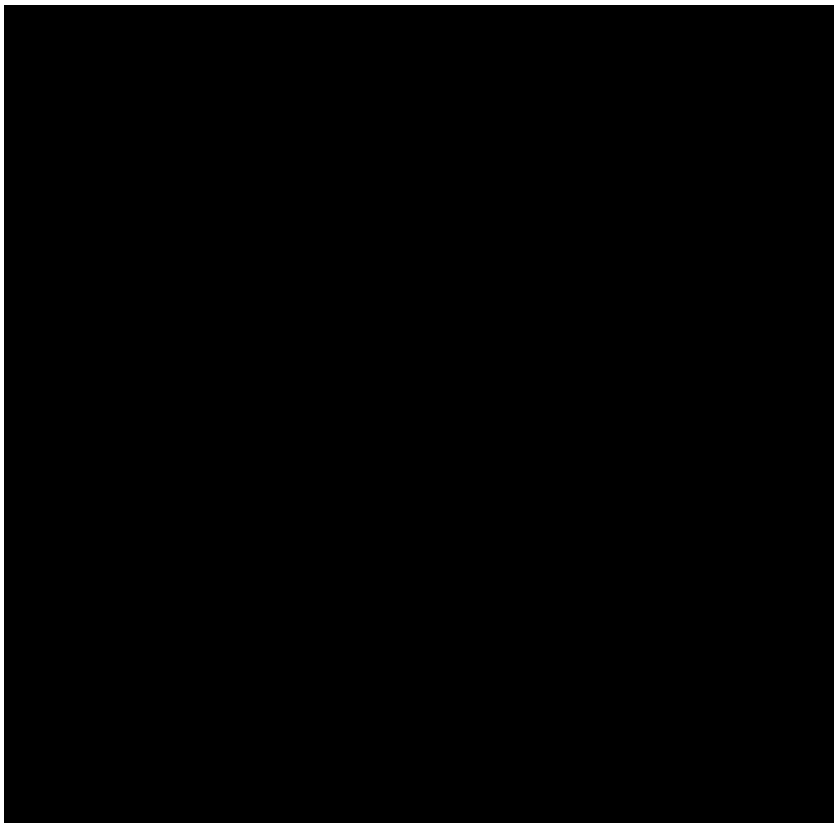
NCT Number: NCT04392219

Document: Protocol, Version 1.3

Date: 17 June 2020

Protocol

COVID-19 Generic Procedures



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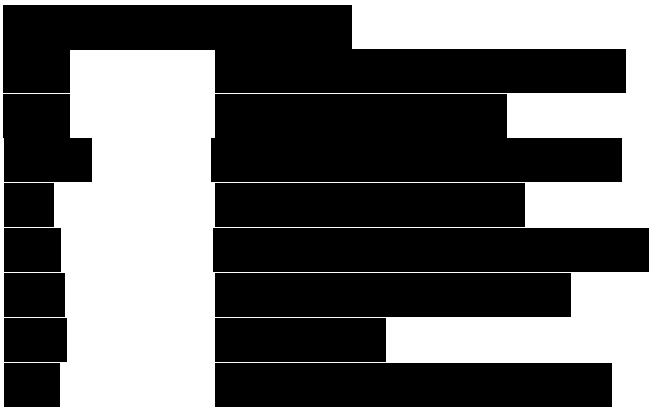

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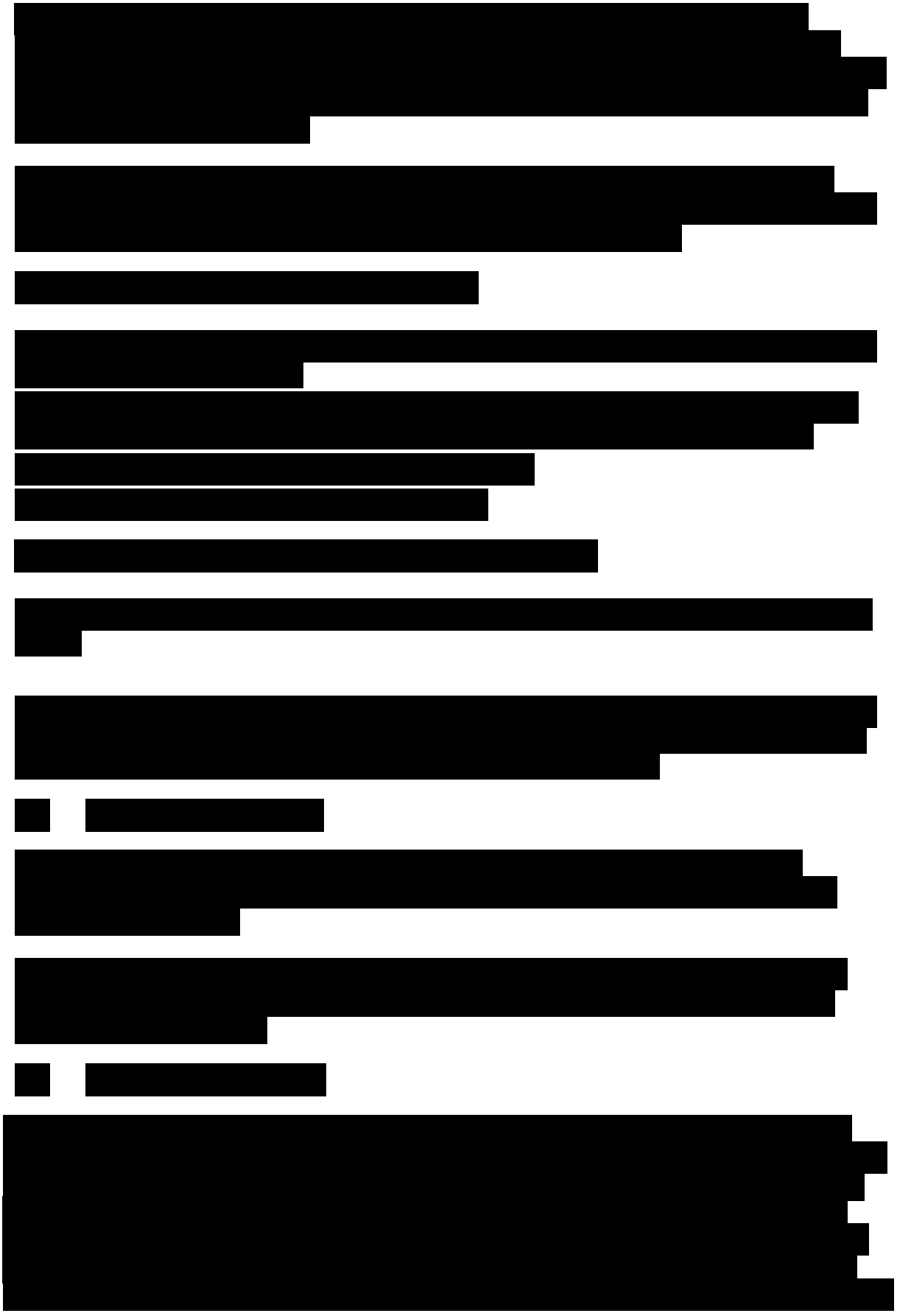

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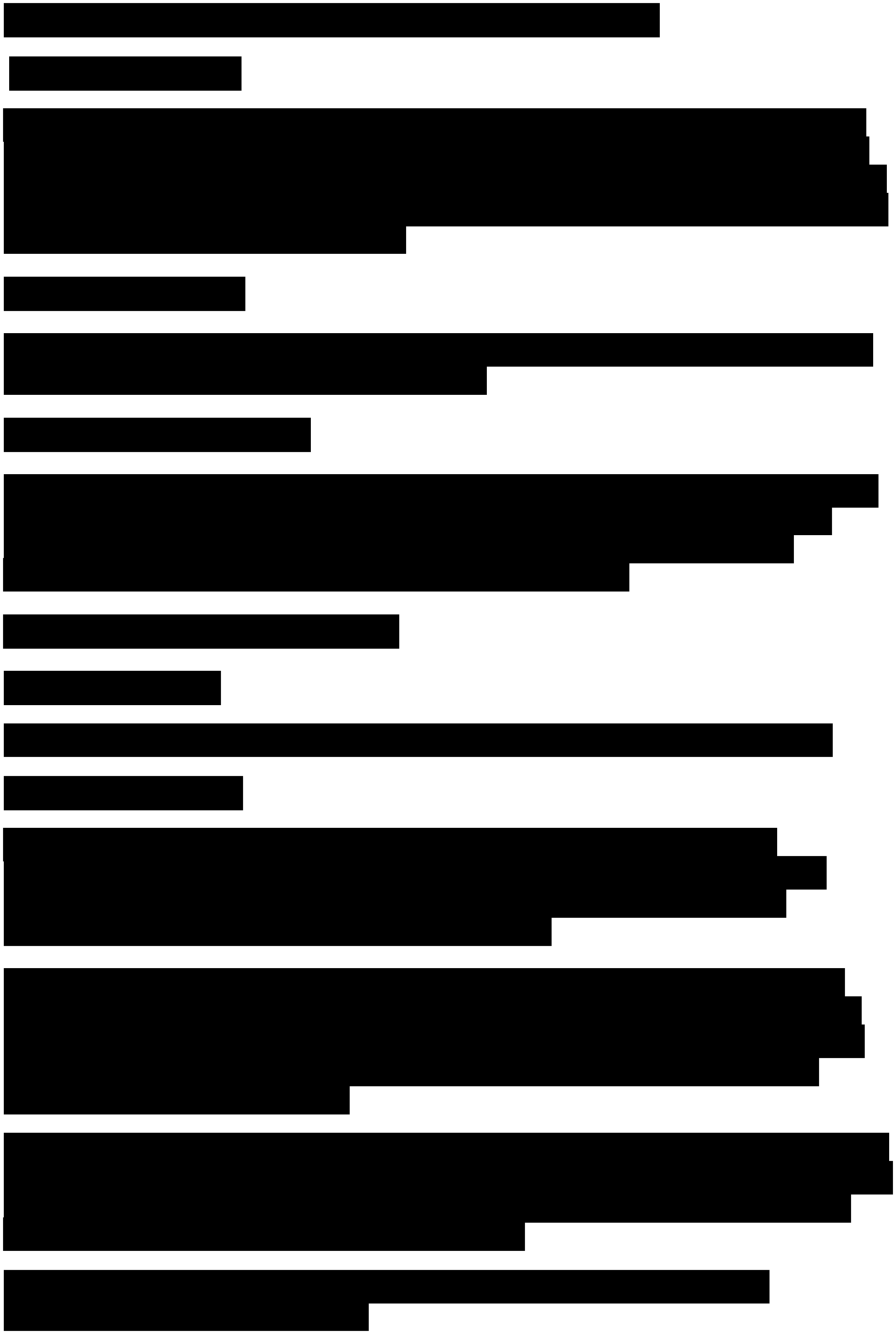

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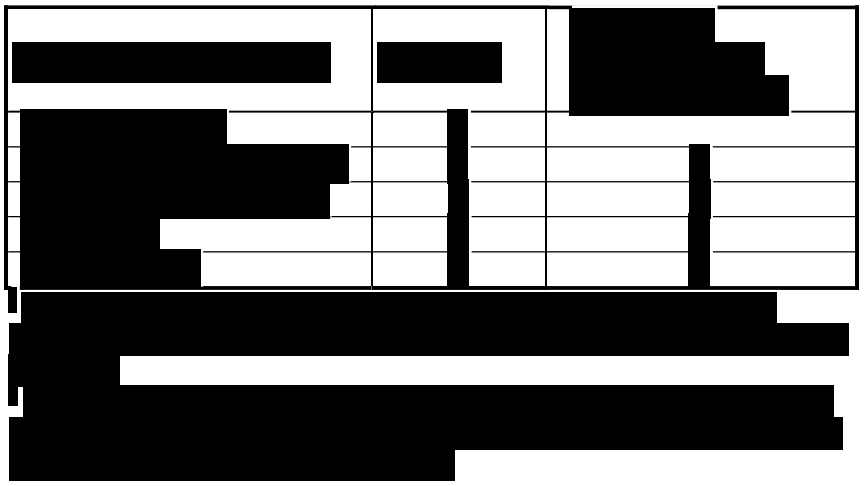

Title of study: A Randomized, Double-blind, Placebo-controlled, First-in-human Study Designed to Evaluate the Safety, Tolerability, and Pharmacokinetics of EIDD-2801 Following Oral Administration to Healthy Volunteers

NCT Number: NCT04392219

Document: Protocol, Version 2.1

Date: 06 April 2020

A Randomized, Double-Blind, Placebo-Controlled, First-in-Human Study Designed to Evaluate the Safety, Tolerability, and Pharmacokinetics of EIDD-2801 Following Oral Administration to Healthy Volunteers

(EIDD-2801-1001-UK)

Protocol Version	2.1 (United Kingdom)
Version Date	06 April 2020

Sponsor Team:

Ridgeback Biotherapeutics	3162 Commodore Plaza, Suite 3E Miami, FL 33133-5815 United States
Medical Officer	[redacted]
EudraCT Number	2020-001407-17

SUMMARY OF CHANGES

Changes from Version 1.1 (United Kingdom) to Version 2.1 (United Kingdom).

The primary reason for this amendment was to include Part 3, a multiple ascending dose study of EIDD-2801 in healthy subjects. Additionally, the following updates were made:

- Results of an *in vivo* micronucleus assay were added.
- The guidelines used to assess the severity of adverse events not presented in the DMID Toxicity Grading Scale were updated, specifically those that were life-threatening or led to death.
- The timepoints for collection of peripheral blood mononuclear cells in Part 2 were modified.
- Minor clarifications were made and typographical errors were corrected.

STATEMENT OF COMPLIANCE

The study will be carried out in accordance with Good Clinical Practice (GCP) as required by the following:

- Consensus ethical principles derived from international guidelines including the Declaration of Helsinki and Council for International Organizations of Medical Sciences International Ethical Guidelines.
- Applicable International Conference on Harmonisation (ICH) GCP Guidelines.
- United States Code of Federal Regulations (CFR) applicable to clinical studies (45 CFR Part 46; 21 CFR Part 50, 21 CFR Part 56, and 21 CFR Part 312).
- Applicable laws and regulations.

SIGNATURE PAGE

The signature below constitutes the approval of this protocol and the attachments and provides the necessary assurances that this study will be conducted according to all stipulations of the protocol, including all statements regarding confidentiality, and according to local legal and regulatory requirements and applicable regulations and ICH guidelines.

Principal Investigator: [redacted]

Signed: _____ Date: 06 APR 2020

Name

Title EXECUTIVE MEDICAL DIRECTOR

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ABBREVIATIONS

AE	adverse event
AGP	alpha1-acid glycoprotein
ALT/SGPT	alanine aminotransferase/serum glutamic pyruvic transaminase
AST/SGOT	aspartate aminotransferase/serum glutamic oxaloacetic transaminase
AUC	area under the curve
BID	twice-daily
BMI	body mass index
CDC	Centers for Disease Control and Prevention
CEM	a cell line of lymphoblastic cells derived from a child with leukemia
CFR	Code of Federal Regulations
CHKV	Chikungunya virus
CI	confidence interval
CoV	coronavirus
CRF	case report form
CTP	cytosine triphosphate cytidine triphosphate
CYP	cytochrome P450
DMID	Division of Microbiology and Infectious Diseases
DMSO	dimethylsulfoxide
DPP4	dipeptidyl peptidase 4
DRF	dose range finding
DSS	Drug Safety Services
EBOV	Ebola virus
EC	Ethics Committee
EC_{50}	Half-maximal effective concentration
ECG	electrocardiogram
EEEV	Eastern equine encephalitis virus
EIDD	Emory Institute for Drug Development
EOS	End of Study
FDA	Food and Drug Administration
FE	food-effect
FIH	first-in- human
FSH	follicle stimulating hormone
GCP	Good Clinical Practice
GI	gastrointestinal
GLP	Good Laboratory Practice

HAE	human airway epithelium
HBV	hepatitis b virus
hBTEC	human bronchial/tracheal epithelial cells
HCV	hepatitis c virus
HED	human equivalent dose
HEENT	head, eye, ear, nose and throat
HIV	human immunodeficiency virus
HPMC	hydroxypropyl methylcellulose
IAV	influenza a virus
IB	Investigator's Brochure
IBV	influenza b virus
IC_{50}	half maximal inhibitory concentration
ICF	informed consent form
ICH	International Council for Harmonisation
MAD	multiple ascending dose
MDCK	Madin-Darby Canine Kidney Cells
MedDRA	Medical Dictionary for Regulatory Activities
MERS	Middle East respiratory syndrome
%MN-PCE	percentage of micronuclei-polychromatic erythrocytes
MRSD	maximum recommended starting dose
MTD	maximum tolerated dose
NOAEL	no-observed-adverse-effect-level
NSAID	nonsteroidal anti-inflammatory drug
OTC	over-the-counter
PBMC	peripheral blood mononuclear cells
PBO	placebo
PCE:TE	polychromatic erythrocytes to total erythrocytes
PE	physical examination
PI	principal investigator
[redacted]	[redacted]
PK	pharmacokinetic
QTc(F)	QT interval corrected for heart rate (using Fridericia's formula)
RNA	ribonucleic acid
RSV	respiratory syncytial virus
SAD	single ascending dose
SAE	serious adverse event

SAP	statistical analysis plan
SARS	severe acute respiratory syndrome
SI	selectivity index
SOP	standard operating procedure
UTP	uridine 5'-triphosphate
VEEV	Venezuelan equine encephalitis virus
ZIKV	Zika virus

PROTOCOL SYNOPSIS

Sponsor: Ridgeback Biotherapeutics	
Title: A Randomized, Double-Blind, Placebo-Controlled, First-in-Human Study Designed to Evaluate the Safety, Tolerability, and Pharmacokinetics of EIDD-2801 Following Oral Administration to Healthy Volunteers (EIDD-2801-1001-UK)	
Short Title: EIDD-2801-1001-UK	**Development Phase:** Phase 1
Description of Study Drugs, Dose and Mode of Administration: EIDD-2801 and matching placebo (PBO) will be supplied [redacted] **Part 1:** A single oral dose of EIDD-2801 or PBO will be administered to subjects enrolled in Part 1 (P1; single ascending dose [SAD]) cohorts. The starting dose in the first SAD cohort will be 50 mg. The doses will be administered in an escalating manner. Dose levels will not increase by more than 3-fold for predicted nonpharmacologically active dose levels and by 2-fold for predicted pharmacologically active dose levels. [redacted] **Part 2:** Subjects in Part 2 (P2)/food-effect (FE) will receive 2 single doses of EIDD-2801 with a 14-day (minimum) washout period between doses. The dose for the P2 cohort is planned to be 100 mg but may be adjusted based on safety, tolerability, and/or pharmacokinetic (PK) data from P1. In any case, the dose assessed in P2 will have been given previously to subjects in a P1 cohort successfully (i.e., no halting rules were met following dosing). **Part 3:** Multiple doses of EIDD-2801 or PBO will be administered to subjects enrolled in Part 3 (P3; multiple ascending dose [MAD]) cohorts. Subjects will receive twice-daily (BID) doses for 6 days, with a single dose administered on the morning of Day 7. The total daily dose will not exceed a dose shown to be safe and well-tolerated in P1. Doses in P3 may be administered in the fed state, following review of the PK data from P2. Additionally, depending on ongoing review of the safety, tolerability, and PK data, the dosing frequency and number of days of dosing may be changed; however, the dosing frequency will be no less than once-daily or no greater than three-times daily, and the number of days of dosing will be no fewer than 5 days and no greater than 10 days.	
Treatment Duration: **Part 1:** Subjects enrolled into P1/SAD cohorts will receive a single dose of study drug (EIDD-2801 or matching PBO). **Part 2:** Subjects enrolled into the P2/FE cohort will receive 2 single doses of EIDD-2801, with a washout period between doses. **Part 3:** Subjects in P3/MAD cohorts will receive study drug (EIDD-2801 or matching PBO) for 6 days BID followed by a single dose of EIDD-2801 or PBO on Day 7. Depending on ongoing review of the safety, tolerability, and PK data, the number of days of dosing may be changed; however, will be no fewer than 5 days and no greater than 10 days.	

Subject Duration:

Part 1: The maximum possible study duration for participants enrolled in P1/SAD cohorts will be approximately 43 days.

Part 2: The maximum possible study duration for participants enrolled in the P2/FE cohort will be approximately 58 days.

Part 3: The maximum possible study duration for participants enrolled in P3/MAD will be approximately 49 days.

Objectives and Endpoints:

Part 1: SAD Cohorts

Primary:

- Objective: To determine the safety and tolerability of single ascending doses of EIDD-2801.
  - Endpoints:
    - Results of safety evaluations including safety laboratory assessments, physical examination (PE), electrocardiograms (ECGs), vital signs, and adverse events (AEs).

Secondary:

- Objective: To define PK of EIDD-2801 and EIDD-1931 in plasma and urine following single doses administered to healthy volunteers.
  - Endpoints:
    - Plasma PK parameters, including C_{max}, t_{max}, $t_{1/2}$, CL/F, λz, Vz/F, $AUC_{0\text{-}inf}$ and AUC_{last} as appropriate
    - Urinary excretion of EIDD-2801 and EIDD-1931 following single-dose administration.

Part 2: Single-Dose Food-Effect

Primary:

- Objective: To assess the effect of food on the PK of EIDD-2801 and EIDD-1931 following a single oral dose.
  - Endpoints:
    - Plasma PK parameters, including C_{max}, t_{max}, $t_{1/2}$, CL/F, λz, Vz/F, $AUC_{0\text{-}inf}$ and AUC_{last} as appropriate
    - Urinary excretion of EIDD-2801 and EIDD-1931 following single-dose administration.

Secondary:

- Objective: To determine the safety and tolerability of single doses of EIDD-2801.
  - Endpoints:
    - Results of safety evaluations, including safety laboratory assessments, PEs, ECG, vital signs, and AEs.

Part 3: MAD Cohorts

Primary:

- Objective: To determine the safety and tolerability of multiple ascending doses of EIDD-2801.

- Endpoints:
  - Results of safety evaluations, including safety laboratory assessments, PE, ECGs, vital signs, and AEs.

Secondary:

- Objective: To define PK of EIDD-2801 and EIDD-1931 in plasma and urine following multiple doses administered to healthy volunteers.
  - Endpoints:
    - Plasma PK parameters, including C_{trough}, C_{max}, t_{max}, $t_{1/2}$, CL/F, λz, Vz/F, $AUC_{0\text{-}inf}$ (Day 1 dose only), AUC_{τ}, $RA_{AUC\tau}$, and RA_{Cmax} as appropriate
    - Urinary excretion of EIDD-2801 and EIDD-1931 following multiple-dose administration.

Exploratory:

- Objective: To collect data to assess the relationship between EIDD-2801/EIDD-1931 concentrations and QT interval corrected for heart rate (QTc).

Population:

Part 1: P1/SAD cohorts will include 8 subjects each, 2 randomized to receive PBO and 6 randomized to receive EIDD-2801. Initially, 4 SAD cohorts are planned for P1 with the option to add an additional 3 cohorts based on study results.

Part 2: Ten subjects will be enrolled into the P2/FE cohort; all subjects enrolled in the P2 cohort will receive EIDD-2801. One P2 cohort is planned for the study. If PK results obtained are equivocal, additional subjects may be enrolled into the P2 cohort at the previously tested dose or an additional P2 cohort may be added at a different dose.

Part 3: P3/MAD cohorts will include 8 subjects, 2 randomized to receive PBO and 6 randomized to receive EIDD-2801. Initially, 4 MAD cohorts are planned for P3 with the option to add an additional 3 cohorts based on study results. However, the number of cohorts may be reduced if the study objectives are met. Dose levels in P3 may be repeated for collection of peripheral blood mononuclear cells (PBMCs).

The site is strongly encouraged to ensure that women are represented in each cohort. Inclusion and exclusion criteria for study participation are as follows:

Inclusion Criteria: subjects who meet all of the criteria at Screening will be eligible for study participation

1. Male or female between the ages of 18 and 60, inclusive.
2. Female subjects must be of nonchildbearing potential. Nonchildbearing potential is defined as women who:
   - are physiologically incapable of becoming pregnant including those who are surgically sterile, via hysterectomy, bilateral salpingectomy, and/or bilateral oophorectomy by reported medical history and/or medical records. Surgical sterilization to have occurred a minimum of 6 weeks, or at the Principal Investigator's (PI's; or designee's) discretion, prior to Screening.
   - are postmenopausal (at least 45 years of age with amenorrhea for 12 months without an alternative medical reason with confirmatory follicle stimulating

hormone (FSH) levels of ≥ 40 mIU/mL. The amenorrhea should not be induced by a medical condition such as anorexia nervosa, hypothyroid disease or polycystic ovarian disease, or by extreme exercise. It should not be due to concomitant medications that may have induced the amenorrhea such as oral contraceptives, hormones, gonadotropin-releasing hormones, anti-estrogens, or selective estrogen receptor modulators).

3. Male participants must agree to the use of effective contraception for study duration, i.e., from Check-in until 90 days following the last dose of study drug.
4. Is in generally good health as determined by medical history, PE (at Screening and/or Check-in), vital signs, and other screening procedures.
5. Has a body mass index (BMI) of 18 to 30 kg/m^2.
6. Is willing and able to comply with all study procedures including providing informed consent.

Exclusion Criteria: subjects who meet any of the following criteria at Screening (unless otherwise stated) will not be eligible for participation

1. Females who are pregnant, planning to become pregnant, or breastfeeding.
2. Has a clinically relevant acute or chronic medical condition or disease of the cardiovascular, gastrointestinal (GI), hepatic, renal, endocrine, pulmonary, neurologic, or dermatologic systems as determined by the PI (or designee).
3. Has any current or historical disease or disorder of the hematological system or significant liver disease or family history of bleeding/platelet disorders.
4. Has a history of cancer (other than basal cell or squamous cell cancer of the skin), rheumatologic disease or blood dyscrasias.
5. Has a history of GI surgery or other condition that may interfere with absorption of study drug, in the option of the PI (or designee).
6. Has a history of febrile illness within the 14 days prior to the first dose of study drug.
7. Has a positive alcohol or drug screen at Screening or the Day -1 visit or has a history of alcohol or drug abuse within the past 5 years.
8. Currently uses tobacco, nicotine or tobacco products, e-cigarettes or has stopped using tobacco products within the past 3 months and/or has a positive cotinine test at Screening or Check-in.
9. Has any abnormal laboratory value of Grade 2 or higher, which is considered to be clinically significant by the PI (or designee).
10. Has a total white cell count or absolute lymphocyte count outside the normal range, or hemoglobin or platelet levels below the lower limit of normal, at Screening of Day -1.
11. Has values above the upper limit of normal for the following laboratory analytes: alanine aminotransferase (ALT/SGPT), alkaline phosphatase (serum), aspartate aminotransferase (AST/SGOT), at Screening or Day -1.
12. Positive test result for human immunodeficiency virus (HIV), hepatitis B virus (HBV), or hepatitis C virus (HCV).
13. Has an autoimmune disease, is immunosuppressed or is in any way immunocompromised.

14. Has any of the following:
    - QT interval corrected for heart rate using Fridericia's formula (QTcF) >450 ms confirmed by repeat measurement
    - QRS duration >110 ms confirmed by repeat measurement
    - PR interval >220 ms confirmed by repeat measurement
    - findings which would make QTc measurements difficult or QTc data uninterpretable
    - history of additional risk factors for torsades de pointes (e.g., heart failure, hypokalemia, family history of long QT syndrome).
15. Except as noted, has used prescription drugs (other than hormone replacement therapy) within 14 days prior to the first dose of study drug unless, in the opinion of the PI (or designee), the drug will not interfere with study assessments. The following prescription drugs will be allowed:
    - Statins: Subjects who have been on a stable dose of statins for a minimum of 6 months and who have normal creatine kinase at Screening.
    - Antihypertensives: Subjects who have been on a stable dose of antihypertensives for a minimum of 6 months and have controlled blood pressure and no active heart disease.
16. Has received an experimental agent (vaccine, drug, biologic, device, blood product or medication) within 3 months prior to the first dose of study drug and agrees not to receive another experimental agent during the duration of this trial.
17. Has donated blood within 30 days, plasma within 2 weeks, or platelets within 6 weeks prior to Screening or refuses to agree to not donate blood until 3 months after the End of Study (EOS) visit.
18. Uses over-the-counter (OTC) medications or nutritional supplements (including, e.g., gingko biloba, aspirin at any dose, and nonsteroidal anti-inflammatory drugs [NSAIDs]) on a routine/scheduled basis, and/or is unwilling to abstain from such use for the duration of the study.
19. Is unwilling to abstain from quinine containing products including quinine water, tonic water, bitter lemon, jello shots, any quinine preparations or Cinchona tree bark preparations (e.g., herbal medications containing Cinchona tree bark).
20. Has any condition that would, in the opinion of the PI (or designee), put the subject at increased risk for participation in a clinical trial or confinement in a clinical research unit.

Retesting for Inclusion/Exclusion Criteria: In the case where a parameter is outside of the acceptable limits for enrollment, measurement of the parameter can be repeated one time if both the following are met:

- In the opinion of the PI (or designee), the parameter value does not represent a chronic condition that otherwise will preclude the volunteer from enrolling in the study; and
- In the opinion of the PI (or designee), the abnormality is not likely to recur.

General Investigational Plan: For all potential subjects, volunteers who express interest in the study will report to the clinic for informed consent. The study will be explained to the subject

and the Ethics Committee (EC)-approved informed consent form (ICF) will be presented. The subject will be given the chance to review the document and ask any questions he/she may have. If, after reviewing the consent, the subject would like to participate in the study, then he/she will sign the ICF and begin screening for study entry; those satisfying all criteria will be enrolled into the study and admitted to the clinic on Day -1. Retesting will be allowed as described above.

Part 1: For P1/SAD, the first cohort will be divided into 2 groups. The first group, a sentinel group, will include 2 subjects.

On Day 1, one subject in the sentinel group will be administered PBO and one will be administered EIDD-2801. Analysis of the safety results for the sentinel group will be reviewed 24 hours postdose. If no halting criteria have been met, the remainder of the subjects in Cohort 1 (the second group) will be dosed. There will be no sentinel groups for the remaining cohorts.

Following dosing on Day 1, subjects will remain in the clinic through completion of study assessments on Day 4, returning to the clinic on Day 9 for study assessments, and Day 15 for the EOS visit.

Doses will be administered in an escalating manner following satisfactory review by the Sponsor and PI of the blinded safety and tolerability data (up to 72 hours post final dose). Dose-escalation will only occur if data from a minimum of 6 subjects have been reviewed from the previous lower dose group, such that data from a minimum of 4 subjects who have received EIDD-2801 will be used to make the dose-escalation decision. Dose levels will not increase by more than 3-fold for predicted nonpharmacologically active dose levels and by 2-fold for predicted pharmacologically active dose levels. The highest dose to be studied under this protocol will not exceed 200 mg.

Part 2: For P2/FE, one cohort assessing the effect of food on EIDD-2801 and EIDD-1931 PK parameters will be enrolled; it is planned that the P2 cohort will be at a dose of 100 mg EIDD-2801, although higher or lower doses may be selected based on safety and available PK data from P1.

In addition to assessing the effect of food on dosing, PBMCs will be collected from subjects following each dose; the PBMCs will be stored for analysis of EIDD-2061 levels at some point in the future.

The 10 subjects enrolled in the FE cohort will all receive EIDD-2801; subjects will be randomized to a treatment sequence (i.e., to receive drug in the fed then fasted state [Sequence 1] versus fasted then fed state [Sequence 2]). Half of the subjects in the cohort will be randomized to Sequence 1 and the other half will be randomized to Sequence 2.

Following dosing on Day 1, subjects will remain in the clinic through completion of study assessments on Day 4, returning to the clinic on Day 9 for study assessments, and Day 14 to begin the second dosing sequence on Day 15. Subjects will then be discharged from the clinic on Day 18, returning to the clinic on Day 23 for study assessments, and Day 30 for the EOS visit. There will be a 14-day (minimum) washout period between doses.

Part 3: For P3/MAD, subjects will receive BID doses on Day 1 through Day 6, inclusive, and will receive the final dose of study drug on the morning of Day 7. Subjects will remain domiciled at the site during the dosing period and until Day 10, returning to the clinic on Day 15 for completion of study assessments, and Day 21 for the EOS visit.

Doses in P3 may be administered in the fed state, following review of the PK data from P2. Additionally, depending on ongoing review of the safety, tolerability, and PK data, the dosing

frequency and number of days of dosing may be changed; however, the dosing frequency will be no less than once-daily or no greater than three-times daily, and the number of days of dosing will be no fewer than 5 days and no greater than 10 days.

Part 3 may run in parallel with P1, providing that the total daily dose to be administered does not exceed a dose already shown to be safe and well-tolerated in P1.

Peripheral blood mononuclear cells may be collected from subjects in P3, depending upon ongoing review of the data. The collection of these samples may be omitted from some cohorts, depending on ongoing review of the data. The PBMCs will be stored for analysis of EIDD-2061 levels at some point in the future. In addition, continuous 12-lead ECG monitoring will be performed to collect data to assess the relationship between study drug concentrations and QTc interval. All data will be archived without extraction or analysis and will not be reported in the scope of this study.

Additional parts may be added to this study, including a SAD part in elderly healthy subjects and a study of the safety, tolerability, and PK in Japanese subjects. These additional parts are not described in this protocol but may be added via a protocol amendment.

Safety Monitoring and Potential Unblinding: Safety for this study will be continually monitored by the PI and Sponsor.

Blinding: All study personnel will remain blinded to treatment assignment (i.e., EIDD-2801 or PBO) in P1 and P3, except for personnel at the bioanalytical laboratory, and the unblinded pharmacy staff and pharmacokineticist. If unblinding is required to manage subject safety or to support dose-escalation decisions, the decision to unblind lies solely with the PI. If possible and providing that it does not interfere with or delay any decision in the best interest of the subject, the PI will discuss the intended code-break with the Sponsor.

Dose-Escalation Halting Rules:

For Group 1 in P1/SAD, sentinel dosing will occur whereby 2 subjects (1 active and 1 PBO) will be dosed on 1 day and, providing no safety concerns arise, the remaining 6 subjects will be dosed after at least 24 hours.

The study will be halted if one or more subjects experience a serious adverse event (SAE) that is considered to be related to EIDD-2801 or 2 or more subjects in the same group experience severe AEs that are considered to be related to EIDD-2801.

Dose-escalation will stop if clinically relevant signs or symptoms of similar nature occur in 2 or more subjects in a group that, in the opinion of the PI, warrant stopping of dose-escalation.

Data Monitoring, Safety Reporting and Unblinding: Data will be monitored throughout the course of the study by experienced clinical monitors according to the clinical monitoring plan. As data will be entered into an electronic case report form (CRF), data will be monitored to ensure accuracy as well as adherence to data entry instructions provided in the CRF guidelines. Procedures for reporting any SAE will be detailed in the protocol and forms and instructions provided to the site.

Statistical Considerations: A complete description of all statistical analyses and methods will be presented in the statistical analysis plan (SAP). The SAP will be reviewed and approved by the Sponsor and will be finalized prior to database lock. Plans for PK analyses will be included in the SAP.

Determination of Sample Size: The sample sizes for the P1 and P3 cohorts are typical for a Phase 1 first in human (FIH) study. The sample size for the P2 cohort is in accordance with Food and Drug Administration (FDA) guidelines for sample size in FE studies.

Study Populations:

Safety Population: All subjects who receive at least one dose of study drug.

Pharmacokinetic Population: All subjects receiving study drug who comply sufficiently with protocol requirements (i.e., no major protocol deviations) and for whom a sufficient number of analyzable PK samples have been obtained to permit the determination of the PK profile for EIDD-2801/EIDD-1931 and the statistical analysis of the results. Subjects who experience an AE of vomiting before 2× the median t_{max} of the group may be excluded from the PK population.

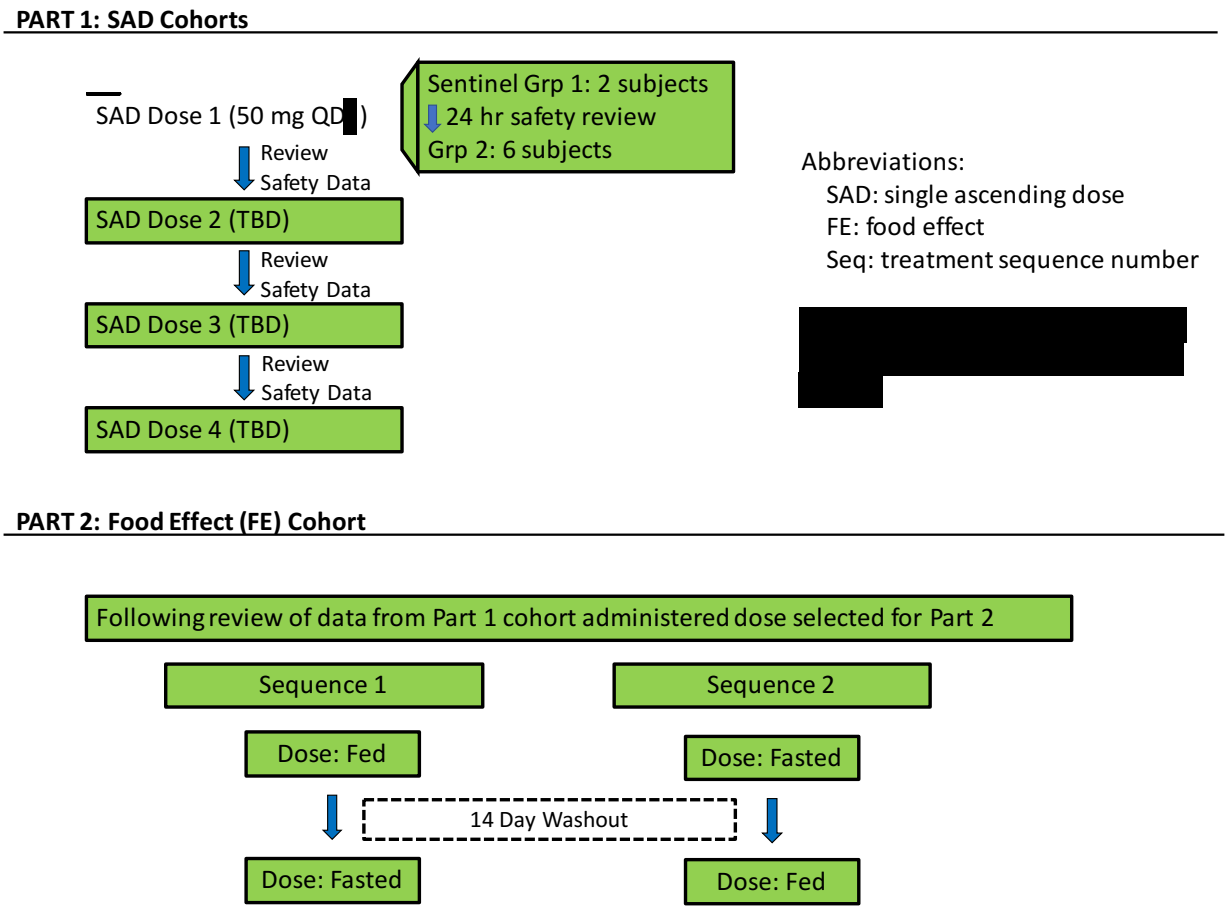
Safety Analyses: Statistical methods for the safety analyses will be descriptive in nature. Safety data, including AEs, clinical laboratory data, vital signs, ECG parameters, and PEs. All appropriate AEs will be graded using the Division of Microbiology and Infectious Diseases (DMID) toxicity scale (March 2014). Change from baseline will be included in summary tables for laboratory parameters. All laboratory data will be included in the data listings and all test values outside the normal range will be flagged.

Pharmacokinetic Analyses: Plasma PK parameters for each dose level will be calculated from the concentrations of EIDD-2801 and EIDD-1931 measured in predose and postdose plasma samples. For each dose level, descriptive statistics will be presented. Figures will be created to display mean and individual subject EIDD-2801 and EIDD-1931 concentration versus time. Urine PK parameters will be calculated whenever possible for each subject based on the urine concentrations of EIDD-2801 and EIDD-1931. The following PK parameters will be calculated:

Plasma PK Parameter	Description
AUC_{last}	The area under the plasma concentration-time curve, from time 0 to the last measurable non-zero concentration, as calculated by the linear up/log down trapezoidal method (P1 and P2 only).
$AUC_{0\text{-}inf}$	The area under the plasma concentration-time curve from time 0 extrapolated to infinity. $AUC_{0\text{-}inf}$ is calculated as the sum of AUC_{last} plus the ratio of the last measurable plasma concentration to the elimination rate constant (λz) (P1, P2, and Day 1 dose of P3).
C_{max}	Maximum observed concentration.
t_{max}	Time to reach C_{max}. If the maximum value occurs at more than one time point, t_{max} is defined as the first time point with this value.
λz	Apparent terminal elimination rate constant; represents the fraction of medication eliminated per unit time.
$t_{1/2}$	Apparent terminal elimination half-life of medication, calculated as $0.693/\lambda z$.
Vz/F	Apparent volume of distribution (EIDD-2801 only).
CL/F	Apparent oral drug clearance (EIDD-2801 only).
C_{trough}	Trough concentration (P3 only).
AUC_{τ}	The area under the plasma concentration-time curve during a dosing interval (P3 only).
$RA_{AUC\tau}$	Observed accumulation ratio based on AUC_{τ} (P3 only).
RA_{Cmax}	Observed accumulation ratio based on C_{max} (P3 only).

Urine PK Parameter	Description
A_e	Amount excreted in urine over the sampling interval.
F_e	Fraction of drug excreted in the urine over sampling interval.
CL_R	Renal clearance, $CL_R = A_e/AUC$ where both A_e and AUC are determined over co-incident time ranges after dosing.
In P1, dose-proportionality for PK parameters of EIDD-2801 and EIDD-1931 ($AUC_{0\text{-}inf}$, AUC_{last}, and C_{max}) will be assessed by the power model. The slope and the associated 90% confidence interval (CI) based on the power model will be reported. In P3 on Day 7, dose-proportionality for PK parameters of EIDD-2801 and EIDD-1931 (AUC_τ and C_{max}) will be assessed by the power model. The slope and associated 90% CI based on the power model will be reported. To assess the effect of food on the PK of EIDD-2801 and EIDD-1931, natural log-transformed PK parameters from P2 ($AUC_{0\text{-}inf}$, AUC_{last}, and C_{max}) will be analyzed using a mixed-effects model with fixed effect terms for treatment (with or without food), period, and sequence and with subject within sequence as a random effect. Point estimates and their associated 90% CIs for the natural log-transformed PK parameters for the difference between EIDD-2801 administered under fed relative to fasted conditions will be constructed. The point estimates and interval estimates will be back transformed to give estimates of the ratio and 90% CI for the geometric least squares mean ratio for fed/fasted to assess the effect of food.	
Interim Analyses: No formal interim analyses are planned for this study. Data from P1 cohorts may be locked, unblinded, and analyzed prior to conducting P2 and P3.	

Figure 1: Study Schema



1. INTRODUCTION

This study is a FIH study designed to assess the safety, tolerability and PK of EIDD-2801 in healthy human volunteers. EIDD-2801 is a ribonucleoside analog with broad-spectrum activity against many RNA viruses. It is currently being developed by Ridgeback Biotherapeutics as a treatment of infections caused by highly pathogenic coronaviruses (CoV), including COVID-19. In addition, EIDD-2801 is being developed in parallel as a treatment of uncomplicated influenza caused by all subtypes of circulating and emerging (drifted and shifted) influenza A virus (IAV) and influenza B virus (IBV), including seasonal, epidemic and pandemic strains.

1.1. Background

EIDD-2801 is the 5'-isopropyl ester prodrug of the broadly active, direct-acting antiviral ribonucleoside analog EIDD-1931. After oral delivery, the prodrug (EIDD-2801) is rapidly hydrolyzed by circulating esterases to produce high circulating (plasma) levels of EIDD-1931. In cell culture systems, EIDD-1931 has been shown to inhibit replication of multiple viral pathogens from multiple RNA virus families including pathogenic CoV (e.g., Middle East respiratory syndrome [MERS], severe acute respiratory syndrome [SARS]-CoV and SARS-CoV-2), influenza viruses (seasonal, pandemic, and avian subtypes), respiratory syncytial virus (RSV), alphaviruses (e.g., Eastern equine encephalitis virus [EEEV], Venezuelan equine encephalitis virus [VEEV], and Chikungunya virus [CHKV]), Filoviruses (e.g., Ebola virus [EBOV]), and Zika virus (ZIKV). In addition, EIDD-2801 is active against orthopoxviruses (tested against vaccinia virus) probably because orthopoxviruses encode their own unique RNA polymerase. [redacted] The primary mechanism of action of EIDD-2801 is inhibition of viral RNA replication by incorporation of the EIDD-1931 monophosphate metabolite into the viral RNA genome resulting in induction of viral error catastrophe.

1.2. Rationale for Development

EIDD-2801 is being developed for the treatment of infections caused by RNA viruses, specifically for COVID-19 and other CoV infections, influenza, and VEEV. During conduct of the FIH study, the Sponsor intends to define a dose that may be active in treating SARS-CoV-2 in patient studies. The multiple-dose data will also be informative with respect to the influenza development program.

EIDD-2801 has a unique dual mechanism of action against RNA viruses, including COVID-19 and other CoV infections. The compound acts as a competitive, alternative substrate for the virally encoded RNA-dependent RNA-polymerase that upon incorporation into nascent chain RNA induces increased mutational frequency in the viral genome. Incorporation quickly results in the production of non-viable virus. Additionally, the active metabolite, EIDD-1931-5'-triphosphate (EIDD-2061), may act directly as a chain terminator and arrest replication by exerting a next nucleoside effect. It is anticipated that the high barrier to resistance observed during *in vitro* passaging studies will translate to slow, if any, emergence of viral resistance. Resilience to viral escape is a distinguishing feature of EIDD-2801.

Currently, there is no approved antiviral therapeutic for the treatment of SARS-CoV-2. An antiviral drug is urgently needed. [redacted]

1.3. Nonclinical Overview

1.3.1. Mechanism of Action

The mechanism of antiviral activity of EIDD-2801 is "lethal mutagenesis"; a concept that is predicated on increasing the viral mutation rate beyond a biologically-tolerable threshold, resulting in impairment of viral fitness and leading to viral extinction.

The specifics of the mechanism are as follows. EIDD-2801 is rapidly taken up by cells and the 5'-isopropylester cleaved to liberate EIDD-1931, which is in turn phosphorylated to EIDD-2061 by host kinases (Hernandez-Santiago et al., 2004; Painter et al., 2019). The 5'-triphosphate, EIDD-2061, acts as a competitive alternative substrate for virally encoded RNA-directed RNA polymerases and EIDD-2061 is incorporated into nascent viral RNA. Owing to the ability of the N^4-hydroxycytosine base of EIDD-1931 to tautomerize, EIDD-2061 can pair with either guanosine or adenosine, and consequently can substitute for either CTP or UTP, respectively (Flavell et al., 1974). This results in an accumulation of mutations that increases with each cycle of viral replication. The process whereby the mutation rate is increased by exposure to a drug is referred to as Viral Decay Acceleration (Mullins et al., 2011) and results in viral ablation.

Significant work has gone into validating this mechanism of action for EIDD-2801/1931, and it has been shown for MERS-CoV, VEEV, and IAV that viruses grown in the presence of EIDD-1931 have significantly increased levels of transition mutations (Agostini et al., 2019; Toots et al., 2019; Urakova et al., 2018). Multi-log decreases in virus yields were observed after treatment with EIDD-1931. Additionally, it was demonstrated for VEEV that the infectivity of virions formed in the presence of EIDD-1931 decreases from ~20% to <0.2%, and that the infectious virions are significantly Impaired in their replication ability (Urakova et al., 2018). As a consequence of this mechanism of action, the generation of drug-resistant escape mutants is practically impossible. This same effect was demonstrated for CoV (Agostini et al., 2019) and influenza virus (Toots et al., 2019). Furthermore, given the unique mechanism of action, EIDD-2801 is expected to be active against viruses resistant to other antiviral agents which have a different mechanism of action. The only data generated to date regarding the activity of EIDD-1931 against viruses resistant to other nucleoside analogs found that EIDD-1931 was active against CoV resistant to remdesivir in cell culture assays (T. Sheahan et al, preprint available at https://www.biorxiv.org/content/10.1101/2020.03.19.997890v1).

As an alternative or additional mechanism of action, it has been theorized that incorporation of EIDD-2061 into viral genomic RNA can change the thermodynamics of RNA secondary

structure and thus decrease the efficiency of the promoter regions involved in RNA genome replication (Stuyver et al., 2003).

1.3.2. *In Vitro* Pharmacology

1.3.2.1. Antiviral Activity in Tissue Culture and in Human Airway Epithelium

The ribonucleoside analog EIDD-1931 is the parent of the prodrug EIDD-2801. EIDD-1931 shows specific antiviral activity in different tissue culture cells and in the differentiated organoid model of human airway epithelium (HAE) with a selectivity index (SI) ranging from 21 to >100 for all influenza viral isolates tested. It is active against IAV (pandemic and seasonal) and IBV strains, as well as against highly pathogenic H5N1 and H7N9 strains (Table 1).

Table 1: EIDD-1931 Antiviral Activity Against Influenza A and B Viruses in Tissue Culture and Primary Human Bronchial/Tracheal Epithelial Cells

Virus	Strain	Cell line	EC_{50}* (µM)	CC_{50} (µM)	SI	Reference
IAV H1N1	Ca/07/2009	MDCK	1.24	68	55	NIAID Antiviral Testing Program
IAV H1N1	WSN/33	MDCK	1.1	299.8	275	Yoon et al., 2018
IAV H1N2	WSN/33	primary hBTEC	5.4	-	-	Yoon et al., 2018
IAV H2N3	Perth/16/2009	MDCK	0.88	52	59	NIAID Antiviral Testing Program
IAV H2N3	Ohio/sw-10-132/2010	MDCK	3.2	299.8	94	Yoon et al., 2018
IAV H5N1	Duck/MN/1525/81	MDCK	1.28	27	21	NIAID Antiviral Testing Program
IAV H5N1	Vietnam/1203/2004	MDCK	0.14	299.8	2100	Yoon et al., 2018
IAV H7N9	Anhui/1/2013	MDCK	0.13	299.8	2300	Yoon et al., 2018
IBV	Florida/4/2006	MDCK	<0.4	76	>190	NIAID Antiviral Testing Program
IBV	Brisbane/60/08	MDCK	0.006	299.8	50000	Yoon et al., 2018
IAV H1N1	Ca/07/2009	HAE-3D*	0.08	50	625	Toots et al., 2019
IAV H1N1	WSN/33	HAE-3D*	0.08	50	625	Toots et al., 2019
IBV	Brisbane/60/08	HAE-3D*	0.06	50	833	Toots et al., 2019

* Human Airway Epithelium organoid model.

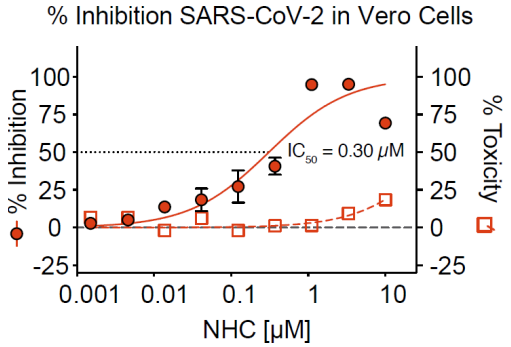
EIDD-1931 also showed specific antiviral activity against human SARS-CoV-1, MERS-CoV (Table 2) and SARS-CoV-2 (Figure 2), against togaviruses VEEV, EEEV and CHKV (Table 3) [redacted]

Table 2: EIDD-1931 Antiviral Activity Against SARS and MERS Coronaviruses in Tissue Culture and Primary Human Bronchial/Tracheal Epithelial Cells

Virus	Strain	Cell line	EC_{50} (µM)	CC_{50} (µM)	SI	Reference
SARS-CoV-1	Urbani	Vero76	<0.4	144	>360	NIAID Antiviral Testing Program
SARS-CoV-1	SARS-CoV-GFP(†)	HAE-3D(*)	<1	>100	>100	Tech. Report 25.038
MERS-CoV	GenBank Ac.No JX869059**	DBT-9	0.56	>200	>357	Agostini et al., 2019
MERS-CoV	Human ß-CoV C, Novel 2912	Vero E6	<0.8	20	>25	NIAID Antiviral Testing Program

* Human Airway Epithelium organoid model; ** cDNA Derived clone

Figure 2: Inhibition of SARS-CoV-2 by EIDD-1931



EIDD-1931 (NHC) antiviral activity (closed circles) and cytotoxicity (open squares) in Vero Cells infected with SARS-CoV-2. Vero cells were infected in duplicate with SARS-CoV-2 clinical isolate virus at a multiplicity of infection (MOI) of 0.05 in the presence of a dose response of EIDD-2801 for 48 hours after which replication was measured through quantitation of cell viability by Cell-Titer-Glo assay. Cytotoxicity was measured in similarly treated but uninfected cultures. Reproduced from Sheahan et al 2020.

Table 3: EIDD-1931 Antiviral Activity Against Togaviruses in Tissue Culture

Virus	Strain	Cell line	EC_{50} (µM)	CC_{50} (µM)	SI	Reference
VEEV	TC-83	Vero	0.43	>200	>930	Urakova et al., 2018
VEEV	TC-83	Vero76	1.92	32	17	NIAID Antiviral Testing Program
EEEV	FL93-939	Vero76	1.08	84	78	NIAID Antiviral Testing Program
CHKV	S27 (VR-64)	Vero76	1.8	96	53	NIAID Antiviral Testing Program

[redacted]

1.3.2.2. Cytotoxicity of EIDD-1931 in Tissue Culture Utilizing Cells from Different Organs and Species

EIDD-1931 was tested for cytotoxicity in human hepatic origin Huh7 and HepG2 cells, in human lymphoid CEM, human pancreatic BxPC-3, human prostate cancer PC-3, human muscle A204, human lung A549, human epithelial hEp-2, rat heart muscle H9c2, monkey kidney Vero, and canine kidney MDCK cell lines (Table 5). The compound exhibits low cytotoxicity in the majority of cells tested (half-maximal effective concentration [EC_{50}] values are in the range of 40 to >100 µM) except in lymphoid origin CEM cells where the compound shows a 7.5 µM EC_{50} value (Sticher et al., 2020; Urakova et al., 2018; Yoon et al., 2018).

Table 5: Cytotoxicity (CC50) of EIDD-1931 in Mammalian Cell Lines

Cell Line	CEM	HepG2	PC-3	A204	A549	BxPC-3	Huh-7	H9c-2	Vero	hEp-2	MDCK
CC50 (µM)	7.5	42.3	267.1	84	46	48	165.5	81	53	272.4	299.8

Sources: Sticher et al., 2020, Yoon et al., 2018

EIDD-2801 typically showed 2-4× lower activity and cytotoxicity than EIDD-1931 due to slightly slower uptake and anabolism in tissue culture.

1.3.2.3. Assessment of Mitochondrial Toxicity

Since EIDD-1931 is a nucleoside analog, additional investigations were performed to analyze whether observed cytotoxicity of EIDD-1931 is caused by mitochondrial toxicity. It was demonstrated that the prolonged treatment (14 days) with the compound does not result in selective killing of mitochondria or in mitochondrial dysfunction in CEM and HepG2 cells (Sticher et al., 2020).

1.3.3. *In Vivo* Pharmacology

The prodrug EIDD-2801 or its parent EIDD-1931 have been tested in animal models of RNA viral infection. An overview of results from the animal studies in indications to be pursued are described below. Additional detail is provided in the Investigator's Brochure (IB).

1.3.3.1. Coronavirus: SARS-CoV and MERS-CoV

In mouse models of SARS and MERS infection and disease, coronaviral disease was assessed by changes in body weight, measured daily, and lung hemorrhage, assessed in the large left lobe of the lung following sacrifice on Day 5 (SARS-CoV) or Day 6 (MERS-CoV). To assess production of infectious virions, virus was isolated from the lower left lobe of the lung following sacrifice on Day 5 (SARS-CoV) or Day 6 (MERS-CoV) and quantified using a plaque assay.

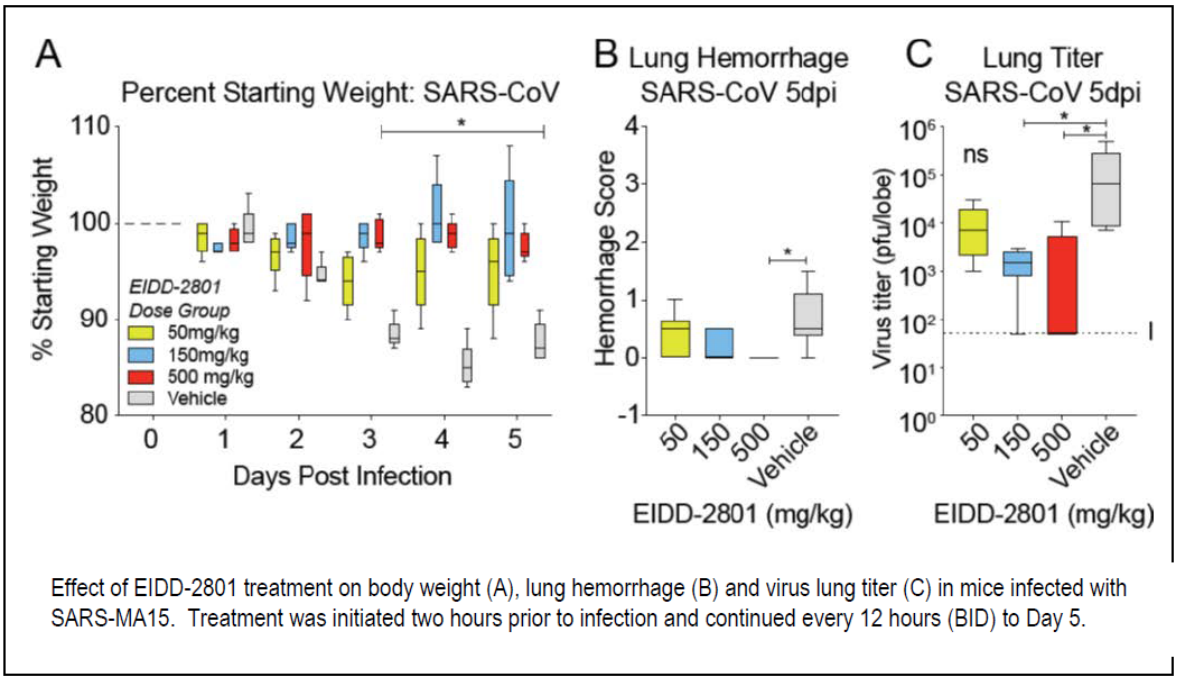
The results demonstrated that, in mice infected with either SARS- or MERS-CoV, both prophylactic and therapeutic treatment with EIDD-2801 resulted in a reduction in virus replication, improvements in pulmonary function, and improvements in maintaining body weight

(i.e., reduced body weight loss). While EIDD-2801 doses of 50, 150 and 500 mg/kg BID were assessed in the CoV mouse experiments, 500 mg/kg BID yielded the most consistent therapeutic effect.

A prophylactic, dose-escalation study was conducted in C57BL/6 mice infected with mouse-adapted SARS-CoV (SARS-MA15). Prophylactic oral treatment with EIDD-2801 was initiated 2 hours before intranasal infection and continued every 12 hours thereafter through the end of the study (Day 5; Figure 3).

In mice treated BID with EIDD-2801, body weight loss observed with vehicle treatment was diminished in the 50 mg/kg treatment group, beginning on Day 3 post-infection. No body weight loss was seen in the 150 and 500 mg/kg treatment groups (Figure 3, Panel A). Lung hemorrhage was also significantly reduced on Day 5 post-infection, following treatment with 500 mg/kg EIDD-2801 (Figure 3, Panel B). When compared to vehicle control, a dose-dependent reduction in SARS-CoV lung titers at Day 5 was seen across all 3 treatment groups (Figure 3, Panel C) with significant differences among the vehicle, 150 mg/kg and 500 mg/kg groups. Thus, prophylactic treatment with EIDD-2801 resulted in a robust antiviral effect that was able to prevent SARS-CoV replication and disease.

Figure 3: EIDD-2801 Prophylactic Treatment of SARS-CoV Infected Mice

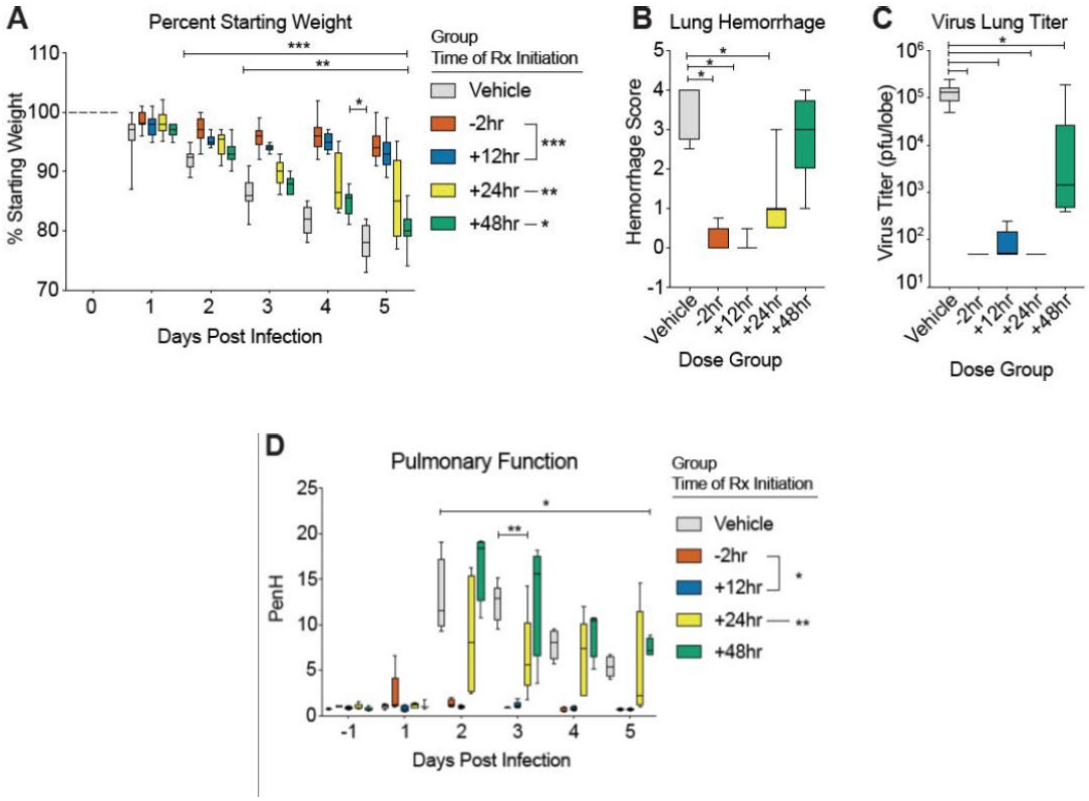


Effect of EIDD-2801 treatment on body weight (A), lung hemorrhage (B) and virus lung titer (C) in mice infected with SARS-MA15. Treatment was initiated two hours prior to infection and continued every 12 hours (BID) to Day 5.

The antiviral activity of EIDD-2801 against SARS-CoV was compared when treatment was initiated at -2 hours (pre-infection) and 12, 24, or 48 hours post-infection. After initiation of treatment, all groups were dosed every 12 hours for the duration of the study (Figure 4). For SARS-challenged mice, initiating treatment at 12 hours post-infection significantly prevented body weight loss beginning on Day 2, a result similar to that seen when dosing prophylactically

(i.e., beginning at 2 hours pre-infection). Initiation of treatment with EIDD-2801 at 24 hours post-infection also significantly reduced body weight loss on Days 3 through 5 post-infection. When EIDD-2801 treatment was initiated at 48 hours post-infection, body weight loss was only statistically different from vehicle on Day 4 post-infection (Figure 4, Panel A). Significant reductions in lung hemorrhage were seen when EIDD-2801 treatment was initiated before (-2 hours) and up to 24 hours after infection; a result that mirrored body weight loss data (Figure 4, Panel B). All mice treated with EIDD-2801 had significantly reduced viral loads in the lungs, even in the group where treatment was initiated 48-hour post-infection (Figure 4, Panel C). Pulmonary function, measured via whole body plethysmography, was assessed using the PenH metric which is a surrogate marker for bronchoconstriction or pulmonary obstruction. The administration of EIDD-2801 prior to infection (-2 hours) and at 12 hour post-infection completely abrogated the loss of pulmonary function observed in vehicle-treated animals (Figure 4, Panel D). Improved pulmonary function was also seen in the group where treatment was initiated 24 hours after virus challenge.

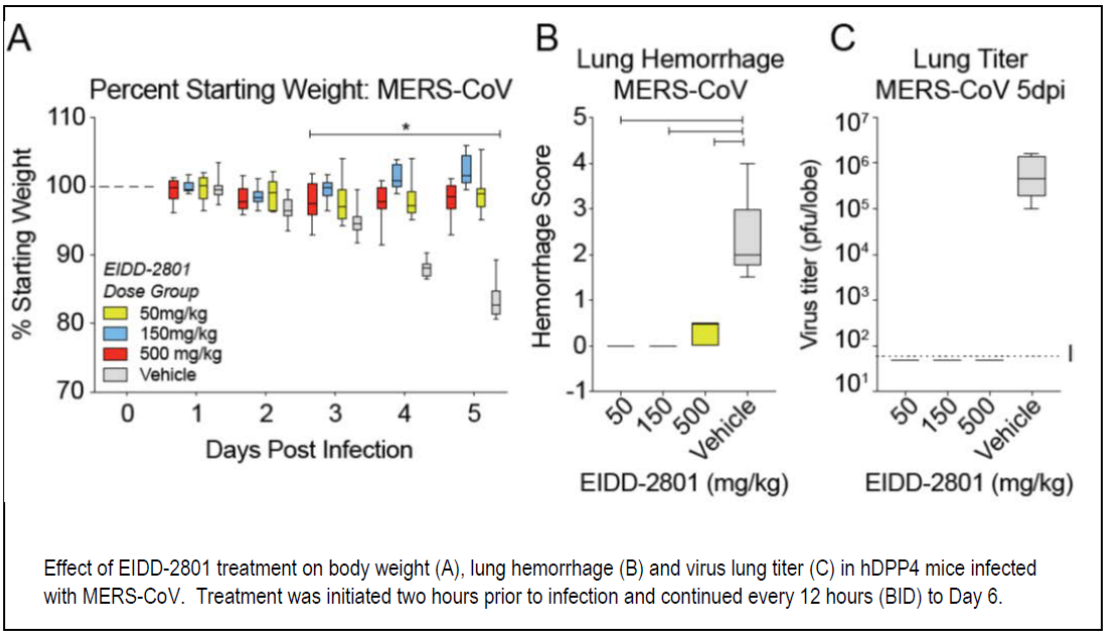
Figure 4: EIDD-2801 Treatment of SARS-CoV Infected Mice



EIDD-2801 was also tested to determine if it is active *in vivo* against MERS-CoV as described by Sheahan et. al (draft manuscript). The murine ortholog of the MERS-CoV receptor, dipeptidyl peptidase 4 (DPP4), does not support viral binding and entry. Thus, all *in vivo* studies described below were performed in genetically modified hDPP4 mice permissive for MERS infection. Prophylactic treatment starting at 2 hours before viral challenge with either 50, 150, or

500 mg/kg EIDD-2801 prevented body weight loss on Days 2 through 6 post-infection (Figure 5, Panel A), prevented lung hemorrhage measured on Day 6 (Figure 5, Panel B), and reduced virus lung titer on Day 6 to the limit of detection (Figure 5, Panel C).

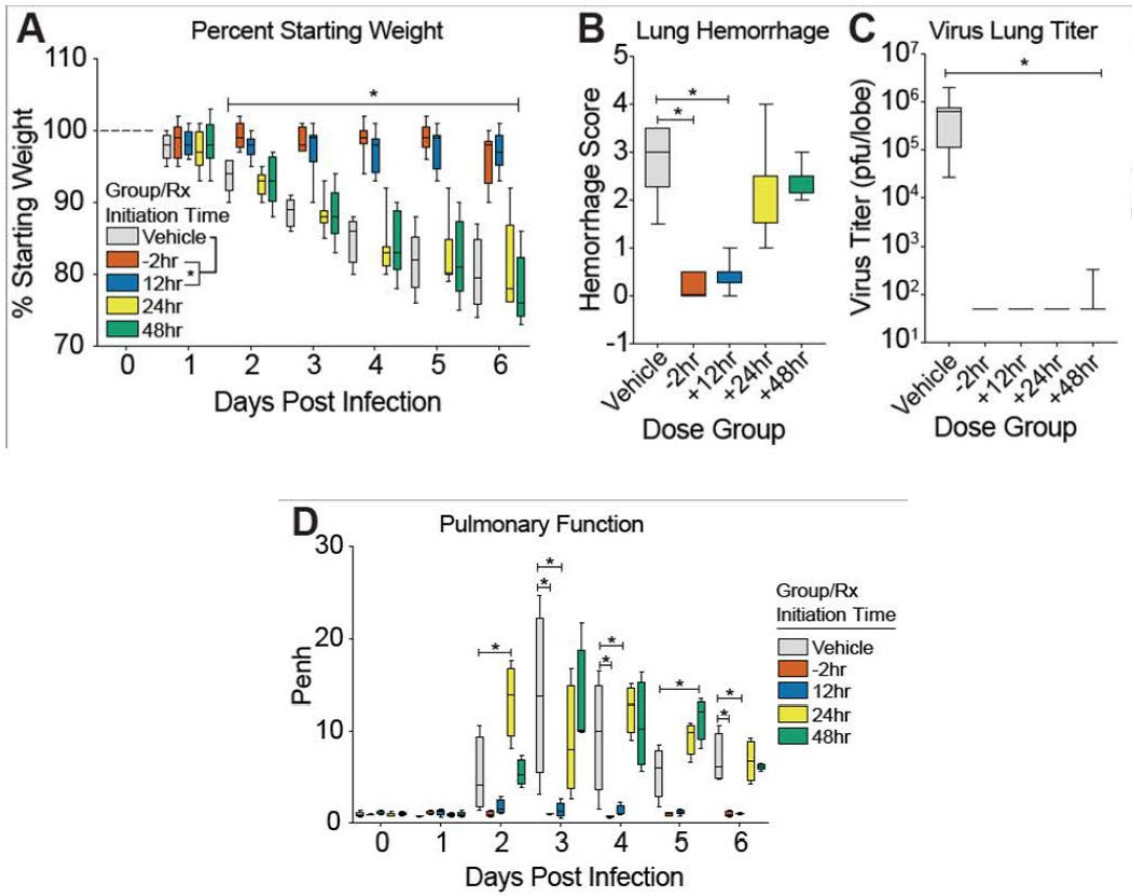
Figure 5: EIDD-2801 Prophylactic Treatment of MERS-CoV Infected Mice



Effect of EIDD-2801 treatment on body weight (A), lung hemorrhage (B) and virus lung titer (C) in hDPP4 mice infected with MERS-CoV. Treatment was initiated two hours prior to infection and continued every 12 hours (BID) to Day 6.

The effect of EIDD-2801 treatment on MERS-CoV infected mice is shown in Figure 6. When EIDD-2801 treatment was initiated 12 hours post-infection, there was no loss in body weight from Days 2 through 6 post-infection (Figure 6, Panel A) and no evidence of lung hemorrhage on Day 6 post-infection (Figure 6, Panel B). However, protection was not observed in groups where treatment was initiated either 24- or 48-hours post-infection. Conversely, virus lung titer on Day 6 post-infection was significantly reduced to the limit of detection in all treatment groups, regardless of the time treatment began (Figure 6, Panel C). To gauge the effect of the timing of EIDD-2801 treatment initiation on physiologic measures of lung disease, pulmonary function was assessed by whole body plethysmography. As expected, a loss of pulmonary function, as determined by measuring the PenH metric, was observed in vehicle-treated animals infected with MERS-CoV beginning on Day 2 post-infection (Figure 6, Panel D). Mirroring the body weight loss data, normal pulmonary function was observed in groups where treatment was initiated prior to or at 12 hours post-infection (Figure 6, Panel D).

Figure 6: EIDD-2801 Treatment of MERS-CoV Infected Mice



1.3.3.2. Influenza Virus

EIDD-2801 was tested in a ferret model of influenza virus infection and disease. Ferrets recapitulate hallmarks of human influenza infection, providing a clinically relevant animal model to investigate therapeutic intervention. Therapeutic oral dosing of influenza virus-infected ferrets reduced shed levels of pandemic and seasonal IAV by multiple orders of magnitude, and alleviated fever and inflammation while improving airway epithelium histopathology. Post-exposure prophylactic dosing was sterilizing (Toots et al., 2019).

Ferrets infected with pandemic IAV and treated with EIDD-2801 at a dose of 7 mg/kg demonstrated significantly less viral shedding and inflammatory cellular infiltrates in nasal lavages, but mounted a normal humoral antiviral response (Toots et al., 2019).

When examining the effect of delayed dosing, Toots et. al. (2019) demonstrated that treatment with 20 mg/kg of EIDD-2801 initiated 24 hours after challenge with IAV (H1N1) resulted in a rapid decrease in both shed virus and fever relative to control, with fever returning to baseline at 24 hours. When oseltamivir (20 mg/kg) was dosed prophylactically as a positive control, a modest suppression of fever was observed; however, there was no reduction in shed virus titer.

1.3.3.3. Venezuelan Equine Encephalitis Virus

Treatment with EIDD-1931 was evaluated in a mouse model of lethal VEEV infections. To be truly effective as a therapeutic agent for VEEV infection, a drug must penetrate the blood brain barrier and arrest virus replication in the brain. High plasma levels of EIDD-1931 are rapidly achieved in mice after oral dosing. Once in the plasma, EIDD-1931 is efficiently distributed into organs important in the pathology of VEEV infection, including the brain, where it is rapidly converted to its active 5'-triphosphate (EIDD-2061). EIDD-1931 showed a good safety profile in mice after 7 days of dosing with up to 1,000 mg/kg/day. In mouse model studies of VEEV infection, EIDD-1931 was 90-100% effective in protecting mice against lethal intranasal infection when therapeutic treatment was started as late as 24 hours post-infection, and partial protection was achieved when treatment was delayed for 48 hours post-infection (Painter et al., 2019).

[redacted]

1.4. Safety and Secondary Pharmacology

The standard battery of safety pharmacology studies including studies assessing the cardiovascular, respiratory and central nervous systems have been conducted. The studies are discussed in the IB; results indicated that there were no adverse pharmacologic effects of EIDD-2801 on the cardiovascular, respiratory or central nervous systems.

1.5. Nonclinical Pharmacokinetics and Metabolism

1.5.1. Overview

The uptake, metabolism and protein binding of EIDD-2801 and EIDD-1931 have been studied in plasma, microsomes, and non-hepatic cells from several species as outlined below. The PK and tissue distribution of prodrug EIDD-2801 and its active parent EIDD-1931 have been studied extensively in rats, dogs and ferrets. Key results from these studies are presented below; additional detail can be found in the IB.

1.5.2. Absorption

EIDD-1931 is parent of the prodrug EIDD-2801. The appearance of EIDD-1931 is dependent on the absorption of EIDD-2801 and the rate of its conversion to EIDD-1931.

EIDD-2801 PK studies have been completed in dog, rat, mouse, ferret, and monkey. EIDD-2801 was efficiently absorbed and rapidly converted to EIDD-1931 in each species. The t_{max} for EIDD-2801 (not observed in rodents) occurred at 0.5-1 hours, while the t_{max} for EIDD-1931 occurred at 1-2 hours.

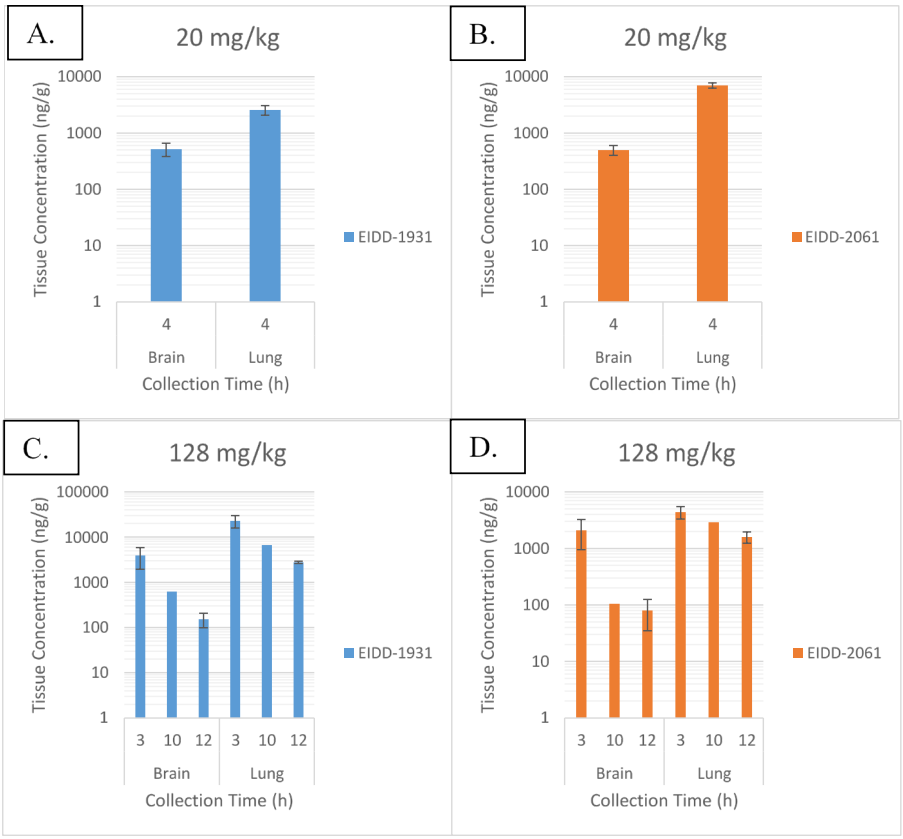
1.5.3. Distribution

[redacted]

1.5.3.2. Tissue Distribution Studies

EIDD-2801 is rapidly absorbed in the gut and converted to EIDD-1931 reaching C_{max} in 1-3 hours in mice, rats, ferrets, dogs and monkeys. EIDD-1931 is then widely distributed to tissues including lungs and brain, where it is rapidly taken up into cells and converted to EIDD-2061. Figure 7 shows the concentration of EIDD-1931 and EIDD-2061 in ferret brain and lung following single doses of 20 (Panels A and B) and 128 (Panels C and D) mg/kg.

Figure 7: Tissue Distribution of EIDD-1931 and EIDD-2061 in Ferret Brain and Lung



1.5.4. Metabolism

1.5.4.1. Metabolic Stability of EIDD-1931 and EIDD-2801

EIDD-2801 was designed to be converted to EIDD-1931 by esterases in plasma or in cells. Stability has been assessed in plasma and liver microsomes from mouse, rat, dog, monkey and humans. The stability of EIDD-2801 in mouse, rat and monkey plasma is relatively short (≤0.4 hours) while the stability is longer in human and dog plasma (1-3 hours). EIDD-2801 stability in mouse, rat, dog and monkey liver microsomes is very short, ranging from 0.02 to 0.08 hours while the stability in human liver microsomes is 1.2 hours (Table 6).

Table 6: Metabolic Stability of EIDD-2801 in Plasma and Liver Microsomes

	Plasma stability	LM stability
Species	t1/2 (h)	t1/2 (h)
Mouse	0.017	0.033
Rat	0.033	0.017
Dog	3.2	0.083
Monkey	0.40	0.017
Human	1.05	1.22

EIDD-2801 is stable in simulated gastric and intestinal fluids (Table 7).

Table 7: Metabolic Stability of EIDD-2801 in Simulated Gastric and Intestinal Fluids and in Buffered Saline with Fetal Bovine Serum

Matrix	t1/2 (hr)
Simulated Gastric Fluid	>24
Simulated Intestinal Fluid	>24
Phosphate Buffered Saline plus 10% Fetal Bovine Serum*	>24

EIDD-1931 was found to be stable when incubated with all tested plasmas, whole blood, liver microsomes and liver S9 extracts and intestinal microsomes (Table 8).

Table 8: Metabolic Stability of EIDD-1931 in Plasma, Whole Blood and Liver and Intestinal Microsomes

Medium	Plasma	Whole Blood	Liver Microsomes	Liver S9 Stability	Intestinal Microsomes
Species	t1/2 (h)	t1/2 (h)	t1/2 (h)	t1/2 (h)	t1/2 (h)
Mouse		>24	>24		>24
Rat	17		>24	>24	
Dog	>24			>24	
Monkey	6.5	>24	>24	>24	>24
Human	10	7	>24	20	>24

1.5.4.2. Uptake and Anabolism of EIDD-1931 in Tissue Culture and Primary Cells

EIDD-1931 is efficiently taken up by tissue culture cells and converted to its pharmacologically active metabolite EIDD-2061 (EIDD-1931-5'-triphosphate). Intracellular EIDD-2061 accumulates dose-dependently, with C_{max} levels ~200-2000 pmol/10^6 cells (at 10-20 µM dose) in different cell lines. It reaches high levels relatively quickly, typically within 1-3 hours, though the t_{max} values vary widely between 1 and 24 hours depending on the cell line and dose concentration tested. Detailed data on the uptake and anabolism of EIDD-2801 is presented in the IB.

EIDD-2801 is also taken up by tissue culture cells and is converted to EIDD-1931 and then to EIDD-2061, but the process is slightly delayed compared to dosing with EIDD-1931. EIDD-1931 is also taken up and metabolized to EIDD-2061 by primary cells. EIDD-2061 is accumulated in all primary cells tested except in mouse primary hepatocytes where EIDD-1931 is apparently extensively metabolized to cytidine and uridine which, in turn, quickly metabolize into CTP and UTP. The quick metabolism of EIDD-1931 consequently results in low levels of EIDD-2061 in mouse hepatocytes. The intracellular stability ($t_{1/2}$) of EIDD-2061 is 4-5 hours in human astrocytes and hBTEC and is significantly shorter (0.2-1.1 hours) in primary hepatocytes.

1.5.5. Excretion

Currently, there is no data on excretion of EIDD-2801 or EIDD-1931. Excretion will be measured in urine during this study.

1.5.6. Pharmacokinetic Drug Interactions

1.5.6.1. Cytochrome P450 (CYP) Inhibition

The purpose of this non-GLP *in vitro* study was to determine the time-dependent inhibitory potential of EIDD-2801 and EIDD-1931 on human cytochromes P450 (CYP) 1A2, 2B6, 2C8, 2C9, 2C19, 2D6, and 3A4 (midazolam and testosterone substrates) enzyme activity, using pooled human liver microsomes in an half-maximal inhibitory concentration (IC_{50}) shift assay.

Neither EIDD-2801 nor EIDD-1931 demonstrated inhibition greater than 31.4% for any of the CYP isozymes tested nor could the data for each assay condition be curve fit to determine time-dependent inhibition by these compounds. Full dose-response curves were not achieved at concentrations ranging from 0.00545 to 50.0 µM indicating EIDD-2801 and EIDD-1931 have no CYP inhibition potential at concentrations ranging from 0.00545 to 50.0 µM. Assay performance was acceptable based on the results for the positive control inhibitors.

1.5.6.2. CYP Induction

An assay was performed to determine the induction potential of EIDD-2801 on human CYP isoenzyme (1A2, 2B6, and 3A4) activity using 3 single-donor lots of inducible, cryopreserved human hepatocytes. Both enzyme activity and mRNA results demonstrated that EIDD-2801 did not show induction for any of the CYP isozymes.

[redacted]

[redacted] [redacted]

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[redacted]	[redacted]	[redacted]	[redacted]	[redacted]
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1.7. Potential Risks and Benefits

1.7.1. Potential Benefits

As this is a FIH study in healthy volunteers, there is no direct benefit to subjects enrolled in the study. However, given the current pandemic situation and *in vitro* antiviral activity of EIDD-2801 against SARS-CoV-2, and the activity against several other viruses of public health concern, it is possible that participants may benefit from future availability of the drug.

1.7.2. Potential Risks

EIDD-2801 has never been administered to humans; therefore, the risks from EIDD-2801 to subjects participating in this trial are unknown. Although toxicology studies have been done, unexpected AEs may occur. [redacted] Subjects will also be monitored for other end-organ effects through a range of safety assessments.

2. OBJECTIVES

2.1. Study Objectives

2.1.1. Primary Objectives

The primary objective of Part 1 of the study is to determine the safety and tolerability of single ascending doses of EIDD-2801.

The primary objective of Part 2 of the study is to assess the effect of food on the PK on EIDD-2801 and EIDD-1931 following a single oral dose.

The primary objective of Part 3 of the study is to determine the safety and tolerability of multiple ascending doses of EIDD-2801.

2.1.2. Secondary Objectives

The secondary objectives of Part 1 and Part 3 of the study is to define the PK of EIDD-2801 and EIDD-1931 in plasma and urine following single and multiple doses administered to healthy volunteers.

The secondary objective of Part 2 of the study is to determine the safety and tolerability of single doses of EIDD-2801.

2.1.3. Exploratory Objectives

The exploratory objective of Part 3 is to collect data to assess the relationship between EIDD-2801/EIDD-1931 concentrations and QTc

2.2. Study Outcome Measures

2.2.1. Primary Outcome Measures

The primary outcome measures for Parts 1 and 3 of are results of safety evaluations including safety laboratory assessments, PEs, ECGs, vital signs, and AEs.

The primary outcome measures for Part 2 of the study are plasma PK parameters including C_{max}, t_{max}, $t_{1/2}$, CL/F, λz, Vz/F, $AUC_{0\text{-}inf}$ and AUC_{last} as appropriate.

2.2.2. Secondary Outcome Measures

The secondary outcome measures are as follows:

- Single-dose plasma PK parameters, including C_{max}, t_{max}, $t_{1/2}$, CL/F, λz, Vz/F, $AUC_{0\text{-}inf}$ and AUC_{last} as appropriate (Part 1)

- Multiple-dose plasma PK parameters, including C_{trough}, C_{max}, t_{max}, $t_{1/2}$, CL/F, λz,

V_z/F, AUC_{τ}, $AUC_{0\text{-inf}}$ (Day 1 dose only), $RA_{AUC\tau}$ and RA_{Cmax}, as appropriate (Part 3)

- Urinary excretion of EIDD-2801 and EIDD-1931 following single- and multiple-dose administration (Parts 1 and 3).

- Results of safety evaluations including safety laboratory assessments, PEs, ECGs, vital signs, and AEs (Part 2).

3. STUDY DESIGN

3.1. Overview

EIDD-2801-1001-UK is a Phase 1, randomized, double-blind, placebo-controlled, FIH, SAD, and MAD study of the safety, tolerability and PK of EIDD-2801 and EIDD-1931 following oral administration of single and multiple doses of EIDD-2801 to healthy volunteers. In addition, for a minimum of one cohort, the effect of food on the single-dose EIDD-2801 and EIDD-1931 PK parameters will be assessed in subjects taking open-label EIDD-2801. The overall objective of the study is to identify a starting dose for future safety and therapeutic intervention trials.

The study is composed of 3 parts; P1 is the SAD study, P2 is the FE cohort study, and P3 is the MAD study.

3.1.1. Part 1 (Single Ascending Dose)

A single oral dose of EIDD-2801 or PBO will be administered to subjects. Subjects will be randomized in a 3:1 ratio to receive either EIDD-2801 or PBO.

Following dosing on Day 1, subjects will remain in the clinic through completion of study assessments on Day 4, returning to the clinic on Day 9 for study assessments, and Day 15 for the EOS visit.

The first cohort will be divided into 2 groups. The first group, a sentinel group, will include 2 subjects. On Day 1, one subject in the sentinel group will be administered PBO and one will be administered EIDD-2801. Analysis of the safety results for the sentinel group will be reviewed 24 hours postdose. If no halting criteria (Section 9) have been met, the remainder of the subjects in Cohort 1 (the second group) will be dosed. There will be no sentinel groups for the remaining cohorts.

After completion of each dosing cohort, safety and tolerability data will be reviewed to determine if any of the halting rules have been met. If not, then the subsequent cohort may be dosed. As PK data become available, these data may be used for dose-escalation decisions.

The proposed dose-escalation scheme is shown in Figure 1, however, planned dose escalations will be determined based on ongoing review of the safety, tolerability, and available PK data. The starting dose in the first SAD cohort will be 50 mg. Dose levels will not increase by more than 3-fold for predicted nonpharmacologically active dose levels and by 2-fold for predicted pharmacologically active dose levels. [redacted]

Four cohorts are initially planned for P1; however, up to an additional 3 cohorts may be enrolled.

3.1.2. Part 2 (Food-Effect)

Two single oral doses of EIDD-2801 will be administered to subjects, in an open-label manner. The dose for the P2/FE cohort is planned to be 100 mg but may be adjusted based on safety, tolerability, and/or PK data from P1. The dose assessed in the P2 cohort will have been given previously to subjects in a P1 cohort successfully (i.e., no halting rules were met following dosing).

Subjects will be randomized to a treatment sequence; i.e., to receive drug in the fed then fasted state (Sequence 1) versus fasted then fed state (Sequence 2; Figure 1). Half of the subjects in the cohort will be randomized to Sequence 1 and the other half will be randomized to Sequence 2.

Following dosing on Day 1, subjects will remain in the clinic through completion of study assessments on Day 4, returning to the clinic on Day 9 for study assessments, and Day 14 to begin the second dosing sequence on Day 15. Subjects will then be discharged from the clinic on Day 18, returning to the clinic on Day 23 for study assessments, and Day 30 for the EOS visit. There will be a 14-day (minimum) washout period between doses.

One cohort of 10 subjects is planned for P2. However, if PK results obtained are equivocal, additional subjects may be enrolled into the P2/FE cohort at the previously tested dose or an additional P2/FE cohort may be added at a different dose.

3.1.3. Part 3 (Multiple Ascending Dose)

Subjects in P3 will be randomized in a 3:1 ratio to receive either EIDD-2801 or PBO. Twice-daily dosing will be administered to subjects on Day 1 through Day 6, inclusive, and a final dose will be administered on the morning of Day 7. Depending on ongoing review of the safety, tolerability, and PK data, the dosing frequency and number of days of dosing may be changed; however, the dosing frequency will be no less than once-daily or no greater than three-times daily, and the number of days of dosing will be no fewer than 5 days and no greater than 10 days.

This protocol has been written assuming 7 days of dosing; however, should the number of days of dosing be modified, the timepoints for study assessments may similarly be changed; representative examples of Time and Events Schedules are presented for 5 and 7 days of dosing.

The total daily dose will not exceed a dose shown to be safe and well-tolerated in P1. Doses in P3 may be administered in the fed state, following review of the PK data obtained from P2. Part 3 may run in parallel with P1, providing that the total daily dose to be administered does not exceed a dose already shown to be safe and well-tolerated in P1.

Subjects will remain domiciled at the site during the dosing period and until Day 10, returning to the clinic on Day 15 for completion of study assessments, and Day 21 for the EOS visit.

Four cohorts are initially planned for P3; however, up to an additional 3 cohorts may be enrolled, or the number of cohorts may be reduced if the study objectives are met. Dose levels in P3 may be repeated for collection of PBMCs.

Additional parts may be added to this study, including a SAD part in elderly healthy subjects and a study of the safety, tolerability, and PK in Japanese subjects. These additional parts are not described in this protocol, but may be added via a protocol amendment.

3.2. Rationale and Justification

3.2.1. Justification of Design

The FIH study is a typical dose-escalation study designed to provide the maximum amount of data in the minimum number of subjects. The cohort size in P1 and P3 is planned to be 8 subjects (6 active:2 PBO). This number of subjects allows for a sufficient PK analysis, considered to be important because dose extrapolation from efficacious animal models to humans will be based on exposure. The duration of participation for each subject following dosing well exceeds 5 drug half-lives (up to 9.1 hours in dogs; 5 hours in ferrets) [redacted]. The FE cohort is considered to be important to maximize exposure based on fed vs. fasted condition and obtaining this information early in Phase 1 will minimize study drug dose in all future studies. This design follows the FDA guidance document on assessing FE in clinical studies.

3.2.2. Justification of Starting Dose

[redacted] The FDA guidance document *("Estimating the Maximum Safe Starting Dose in Initial Clinical Trials for Therapeutics in Adult Healthy Volunteers")* provides guidance for when a safety factor smaller than 10 may be used to calculate the starting dose:

- A smaller safety factor might also be used when toxicities produced by the therapeutic are easily monitored, reversible, predictable, and exhibit a moderate-to-shallow dose-response relationship with toxicities that are consistent across the tested species (both qualitatively and with respect to appropriately scaled dose and exposure).

- A safety factor smaller than 10 could be justified when the NOAEL was determined based on toxicity studies of longer duration compared to the proposed clinical schedule in healthy volunteers. In this case, a greater margin of safety should be built into the NOAEL, as it was associated with a longer duration of exposure than that proposed in the clinical setting. This assumes that toxicities are cumulative, are not associated with acute peaks in therapeutic concentration (e.g., hypotension), and did not occur early in the repeat dose study.

EIDD-2801 satisfies both of these criteria as follows:

[redacted]

Importantly, given the activity of EIDD-2801 versus SARS-CoV-2 (cause of COVID-19), the Sponsor thinks it is most prudent to start with a dose that is predicted to be a safe starting dose and is also as close as possible to a potential therapeutic dose that would allow the Sponsor to move into COVID-19 patients as safely and expeditiously as possible. Based on modeling to animal data, a dose of 100 mg BID is projected to be an active dose in humans.

3.2.3. Justification of Study Population

Healthy volunteers are considered to be the appropriate population for conduct of the FIH study. Healthy volunteers without confounding medical conditions that may obscure the interpretation of AEs or affect absorption, distribution, metabolism and excretion of study drug will provide the most valuable data regarding the tolerability, safety, and plasma exposures observed and expected following single doses and multiple doses up to 10 days. EIDD-2801 is intended for eventual study in patients with potential CoV-2 infection as defined by the Centers for Disease Control and Prevention (CDC) in whom a range of AEs are expected based on the disease under study. Understanding the safety and PK profile in a normal population will better inform the use of EIDD-2801 in disease settings where complications are frequent, and AEs will need to be interpreted in context. While EIDD-2801 is being developed for the treatment of highly pathogenic CoV, there are plans underway to develop protocols for the treatment of influenza. Without safety data from healthy individuals, it will be more difficult to extrapolate the interpretation of AEs from one disease state to another.

4. STUDY POPULATION

This study will enroll healthy volunteers; 8 subjects will be enrolled into each SAD and MAD cohort in P1 and P3, and 10 subjects will be enrolled into the FE cohort in P2.

The site is strongly encouraged to ensure that women are represented in each cohort.

4.1. Subject Inclusion Criteria

Subjects who meet all of the criteria at Screening will be eligible for study participation

1. Male or female between the ages of 18 and 60, inclusive.

2. Female subjects must be of nonchildbearing potential. Nonchildbearing potential is defined as women who:

    - are physiologically incapable of becoming pregnant including those who are surgically sterile, via hysterectomy, bilateral salpingectomy, and/or bilateral oophorectomy by reported medical history and/or medical records. Surgical sterilization to have occurred a minimum of 6 weeks, or at the PI's (or designee's) discretion, prior to Screening.

    - are post-menopausal (at least 45 years of age with amenorrhea for 12 months without an alternative medical reason with confirmatory FSH levels of ≥40 mIU/mL. The amenorrhea should not be induced by a medical condition such as anorexia nervosa, hypothyroid disease or polycystic ovarian disease, or by extreme exercise. It should not be due to concomitant medications that may have induced the amenorrhea such as oral contraceptives, hormones, gonadotropin-releasing hormones, anti-estrogens, or selective estrogen receptor modulators).

3. Male participants must agree to the use of effective contraception for study duration, i.e., from Check-in until 90 days after the EOS visit.

4. Is in generally good health as determined by medical history, PEs (at Screening and/or Check-in), vital signs, and other screening procedures.

5. Has a BMI of 18 to 30 kg/m^2.

6. Is willing and able to comply with all study procedures including providing informed consent.

4.2. Subject Exclusion Criteria

Subjects who meet any of the following criteria at Screening (unless otherwise stated) will not be eligible for participation:

1. Females who are pregnant, planning to become pregnant, or breastfeeding.
2. Has a clinically relevant acute or chronic medical condition or disease of the cardiovascular, GI, hepatic, renal, endocrine, pulmonary, neurologic, or dermatologic systems as determined by the PI (or designee).
3. Has any current or historical disease or disorder of the hematological system or significant liver disease or family history of bleeding/platelet disorders.
4. Has a history of cancer (other than basal cell or squamous cell cancer of the skin), rheumatologic disease or blood dyscrasias.
5. Has a history of GI surgery or other condition that may interfere with absorption of study drug, in the option of the PI (or designee).
6. Has a history of febrile illness within the 14 days prior to the first dose of study drug.
7. Has a positive alcohol or drug screen at Screening or the Day -1 visit or has a history of alcohol or drug abuse within the past 5 years.
8. Currently uses tobacco, nicotine or tobacco products or e-cigarettes or stopped using tobacco products within the past 3 months and/or has a positive cotinine test at Screening or Check-in.
9. Has any abnormal laboratory value of Grade 2 or higher, which is considered to be clinically significant by the PI (or designee).
10. Has a total white cell count or absolute lymphocyte count outside the normal range, or hemoglobin or platelet levels below the lower limit of normal, at Screening or Day -1.
11. Has values above the upper limit of normal for the following laboratory analytes: ALT/SGPT, alkaline phosphatase (serum), AST/SGOT, at Screening or Day -1.
12. Positive test result for HIV, HBV, or HCV.
13. Has an autoimmune disease, is immunosuppressed or is in any way immunocompromised.
14. Has any of the following:
    - QTcF >450 ms confirmed by repeat measurement

- QRS duration >110 ms confirmed by repeat measurement
- PR interval >220 ms confirmed by repeat measurements
- findings which would make QTc measurements difficult or QTc data uninterpretable
- history of additional risk factors for torsades de pointes (e.g., heart failure, hypokalemia, family history of long QT syndrome).

15. Except as noted, has used prescription drugs (other than hormone replacement therapy) within 14 days prior to the first dose of study drug unless, in the opinion of the PI (or designee), the drug will not interfere with study assessments. The following prescription drugs will be allowed:
    - Statins: Subjects who have been on a stable dose of statins for a minimum of 6 months and who have normal creatine kinase at Screening.
    - Antihypertensives: Subjects who have been on a stable dose of antihypertensives for a minimum of 6 months and have controlled blood pressure and no active heart disease.
16. Has received an experimental agent (vaccine, drug, biologic, device, blood product or medication) within 3 months prior to the first dose of study drug and agrees not to receive another experimental agent during the duration of this trial.
17. Has donated blood within 30 days, plasma within 2 weeks, or platelets within 6 weeks prior to Screening or refuses to agree to not donate blood until 3 months after the EOS visit.
18. Uses OTC medications or nutritional supplements (including, e.g., gingko biloba, aspirin at any dose, and NSAIDs) on a routine/scheduled basis and/or is unwilling to abstain from such use for the duration of the study.
19. Is unwilling to abstain from quinine containing products including quinine water, tonic water, bitter lemon, jello shots, any quinine preparations or Cinchona tree bark preparations (e.g., herbal medications containing Cinchona tree bark).
20. Has any condition that would, in the opinion of the PI (or designee), put the subject at increased risk for participation in a clinical trial or confinement in a clinical research unit.

5. STUDY MEDICATION, RANDOMIZATION AND DOSE ADMINISTRATION

5.1. Study Drug Description

EIDD-2801 and matching PBO will be supplied [redacted].

[redacted]

[redacted]

[redacted]

5.1.1. Acquisition, Formulation, Packaging and Labeling

[redacted]

All study drug will be labeled according to the regulatory requirements for investigational product.

5.1.2. Product Storage and Stability

Study drug should be stored at controlled room temperature defined as [redacted]. If excursions occur which are outside of this range, the pharmacy staff should contact Sponsor to determine the course of action. Additional stability data may be available which would allow continued use of the study drug, or study drug may need to be replaced.

5.2. Randomization

Unmasked study drug (EIDD-2801 and matching PBO) will be supplied to the study site pharmacy. The pharmacy staff will be unmasked with regards to treatment assignment. A randomization list will be provided to the pharmacy staff who will use that list to dispense masked study drug for administration to each study participant. In P1 and P3, 6 subjects will

receive EIDD-2801 while 2 subjects will be randomized to PBO. In P2, all 10 subjects in the FE cohort will receive EIDD-2801. The pharmacy staff will maintain the security of the randomization list ensuring that no study personnel outside of the pharmacy have access to identify treatment assignment. In the case that it becomes necessary to know a subject's treatment assignment, unmasking procedures will be followed as discussed in Section 8.4.

5.3. Dosage, Preparation and Administration of Study Drug

Detailed instructions for extemporaneous compounding (as necessary), dispensing and administering study drug can be found in the pharmacy manual.

[redacted]

5.4. Drug Accountability

The site pharmacy must maintain records of receipt and disposition of all study drug supplied to the site by the Sponsor. The records must be maintained according to site standard operating procedures (SOPs) and should include at a minimum, receipt date, lot or batch number, amount and formulation received, [redacted] date, time and amount dispensed, subject identifier for each aliquot of study drug dispensed. In addition, the clinic staff must ensure that the time of dosing is identified for each subject.

Monitors must verify drug accountability/dispensing records during the monitoring visit.

Unused study drug must be disposed of according to the procedures described in the pharmacy manual.

5.5. Concomitant Medication/Treatments

[redacted] In this FIH study, limited types of concomitant medications are permitted. Adjustment of routine medications taken by subjects should be avoided during study participation except when subject safety could be affected by lack of

adjustment. There are no restrictions on treatment medications prescribed by the PI (or designee) to be used for AEs that occur during study participation. For additional restrictions, see Section 6.1.2.

6. STUDY CONDUCT AND VISIT SCHEDULE

All study assessments will be conducted according to the Time and Events Schedule.

6.1. Study Conduct

6.1.1. Study Windows and Rounding Principles

The time schedule described in the protocol for each scheduled activity should be followed as closely as possible. However, if it is not possible and if it is not otherwise specifically contraindicated per protocol, then the time windows detailed in Table 10 are allowed without incurring a protocol deviation.

Table 10: Allowable Time Windows for Study Assessments/Visits

Protocol Specified Time	Allowable Window
PK Samples and Associated Assessments	
Predose	within 2 hours prior to dosing
<2 hours	± 5 minutes
≥2 hours to 24 hours	± 15 minutes (± 2 hours for urinalysis)
>24 hours to 48 hours	± 30 minutes
>48 hours	± 1 hour (PK) and ± 2 hours (safety)
Study Visits - Parts 1 and 2	
Day −1 to Day 4 (Part 1)	must occur on scheduled day
Day −1 to Day 4 and Day 14 to Day 18 (Part 2)	must occur on scheduled day
Day 9 (Part 1) and Days 9 and 23 (Part 2)	± 1 day
End-of-Study (EOS) Visit	± 2 days
Study Visits - Part 3	
Day −1 to Day 10	must occur on scheduled day
Day 15	± 1 day
Day 21/EOS Visit	± 2 days

6.1.2. Restrictions

Prior to arriving at the clinic for the Day -1 visit, subjects must abstain from consumption of alcoholic beverages for a minimum of 72 hours prior to Check-in. Subjects must continue to abstain from consumption of alcoholic beverages throughout clinic confinement. Subjects enrolled in P2 must continue to refrain from consuming alcoholic beverages from discharge on Day 4 through Check-in on Day 14, and then through the second clinic confinement to Day 18. After discharge on Day 4 (P1), Day 18 (P2), or Day 10 (P3), subjects must minimize consumption of alcoholic beverages (i.e., limit of up to one serving per day) until the EOS procedures have been completed.

All subjects must refrain from the following:

- consuming quinine containing products from 72 hours prior to Check-in through to completion of the EOS procedures.
- using nutraceuticals and nutritional/vitamin supplements (e.g., gingko biloba, multivitamins) from 72 hours prior to Check-in through to completion of the EOS procedures. However, vitamin supplements required by a physician are exempt from this restriction.
- taking OTC analgesics including aspirin (any dose) and NSAIDS from 72 hours before Check-in until completion of EOS study procedures unless prescribed by the PI (or designee).
- use of tobacco, nicotine or tobacco products, or e-cigarettes from 3 months prior to Screening until the EOS visit.
- strenuous exercise from 7 days before Check-in until the EOS visit. Subjects will otherwise maintain their normal level of physical activity during this time (i.e., will not begin a new exercise program nor participate in any unusually strenuous physical exertion).
- Female subjects must not donate eggs/ovum from the time of Check-in until 3 months after the EOS visit.

All subjects in P1 should be dosed in the fasted state. Subjects should fast overnight for a minimum of 10 hours prior to dosing in the morning on Day 1. Following dosing, subjects may have water after 2 hours and food beginning 4 hours postdose. For subjects to be dosed in the fed state, subjects should have a high-fat breakfast as defined in the FDA guidance. Subjects must complete the meal within 30 minutes of starting the meal and should be dosed after 30 minutes of starting the meal. Doses in P3 may be administered in the fed state, following review of the PK data from P2.

6.2. Screening

6.2.1. Screening Visit

Subjects who meet preliminary pre-screening criteria (as defined by the site) and are interested in participating in the study will arrive at the study site for administration of informed consent according to site standard operating procedures (SOPs). After the subject has signed and dated the ICF, screening procedures can begin. Assessments and procedures should be conducted as shown in the Time and Events Schedule Screening may be conducted as early as 28 days prior to dosing.

6.2.2. Retesting Procedures

In the case where a parameter is outside of the acceptable limits for enrollment, measurement of the parameter can be repeated one time if both the following are met:

- In the opinion of the PI (or designee), the parameter value does not represent a chronic condition that otherwise will preclude the volunteer from enrolling in the study; and
- In the opinion of the PI (or designee), the abnormality is not likely to recur.

6.2.3. Study Visits

Subjects who satisfy entry criteria will return to the clinic for the Day -1 visit. Following review of I/E criteria, subjects who still qualify will be checked into the clinic and enrolled into the study. Following enrollment, clinical chemistry, hematology and urine samples will be collected to determine baseline values. Based on site standard practices, alternate subjects will also be enrolled into the study in case one of the selected subjects cannot be dosed. If all subjects can be dosed on the morning of Day 1, the alternates will be released and may be enrolled into subsequent cohorts. All assessments and procedures will be performed according to the Time and Events Schedule.

6.2.4. Part 1 (Single Ascending Dose)

Subjects will remain in the clinic through completion of study procedures on Day 4, returning to the clinic for procedures on Day 9.

6.2.5. Part 2 (Food-Effect)

Subjects will remain in the clinic through completion of study procedures on Day 4, returning to the clinic on Day 9 for study assessments, and Day 14 to begin the second dosing sequence on Day 15. Subjects will then be discharged from the clinic on Day 18, returning to the clinic on Day 23 for study assessments.

6.2.6. Part 3 (Multiple Ascending Dose)

Subjects will remain in the clinic through dosing and completion of study procedures on Day 10, returning to the clinic on Day 15 for study assessments. Depending on ongoing review of the safety, tolerability, and PK data, the number of days of dosing may be changed; however, the number of days of dosing will be no fewer than 5 days and no greater than 10 days.

6.3. Safety Follow-up and End-of-Study Visit

Subjects in P1 will return to the clinic for the EOS visit on Day 15. Subjects in the FE cohort (P2) will return for the EOS visit on Day 30. Subjects in P3 will return for the EOS visit on Day 21. Subjects with drug-related AEs at the EOS visit will be followed as discussed in Section 8.2.1.

6.4. Subject Withdrawal and Replacement

As this is a small study with a limited number of subjects per cohort, it is critical that all subjects complete the study including postdose study assessments. Site personnel should emphasize this to study subjects at the time of informed consent so that subjects will understand this fact before agreeing to participate in the study.

Subjects may voluntarily withdraw their consent for study participation at any time without penalty or loss of benefits to which they are otherwise entitled. An PI (or designee) may also withdraw a subject from receiving study drug or participation in the study for any reason. Subjects who withdraw or are withdrawn from the study should undergo withdrawal procedures as discussed below. These procedures would include follow-up safety evaluations.

6.4.1. Reasons for Withdrawal

If a subject withdraws or is withdrawn from the study, the primary consideration must be the health and welfare of the subject. The reasons for withdrawal might include but are not limited to the following:

- Subject no longer meets eligibility criteria including subject withdraws consent from study participation (with or without a reason)
- Subject becomes noncompliant
- Medical disease or condition, or new clinical finding(s) for which continued participation, in the opinion of the PI (or designee) might compromise the safety of the subject, interfere with the subject's successful completion of this study, or interfere with the evaluation of responses
- Subject Lost-to-Follow-up
- Subject becomes pregnant, if applicable
- Determined by a physician's discretion to require additional therapy not indicated in the protocol to ensure subject's health and well-being (or treatment failure, if applicable)

The PI should be explicit regarding study follow-up (e.g. safety follow-up) that might be carried out. If the subject consents, every attempt will be made to follow all AEs through resolution, return to baseline, or until stabilized with sequelae for a maximum of 30 days following discontinuation. The procedures that collect safety data for the purposes of research must be inclusive in the original ICF or the PI may seek subsequent informed consent using an EC-approved ICF with the revised procedures.

The PI will inform the subject that already collected data will be retained and analyzed even if the subject withdraws from this study.

6.4.2. Handling of Withdrawals

Subjects who withdraw from the study prior to receiving study drug (i.e., on Day -1 or before dosing on Day 1) will be discharged from the clinic and followed only if AEs are present which occurred due to participation in the study (e.g., AE resulting from a study procedure). In this case, the subject should be followed until the AE resolves or the PI determines that the AE has stabilized.

Subjects who withdraw from the study after receiving study drug should have EOS assessments at the time of withdrawal or as quickly thereafter as possible.

Subjects who do not return for follow-up procedures on Days 9/23 or 15/30 (P1 and P2), or Days 15 or 21 (P3) will be contacted by the site at least 3 times using the subject's preferred method of communication (as determined at Check-in). If the site is unable to contact the person, then a certified letter will be sent. If the subject still cannot be reached or refuses to come back to the clinic after all attempts, the subject will be considered Lost-to-Follow-up and withdrawn from the study.

6.4.3. Documentation of Withdrawals

If a subject withdraws or is withdrawn prior to completion of the study, the reason for this decision must be recorded in the CRF. If the subject is Lost-to-Follow-up, the site should document the attempts to contact the subject in the source documents. If the subject has an ongoing AE at the time of withdrawal, then the AE should be followed as detailed in Section 8.2.1.

6.4.4. Subject Replacement

If a subject withdraws from the study prior to receiving study drug, the subject will be replaced. In this case, designated alternate subjects, if available, will be first in line to replace the withdrawn subject.

If a subject withdraws from the study after receiving study drug, then the decision to replace the subject will be made by the PI (or designee) in consultation with the Sponsor. Factors to consider will be the timing postdose of withdrawal and the number of safety and PK assessments completed prior to withdrawal. Subjects who are withdrawn because of an AE related to the study drug will not be replaced.

6.5. Unscheduled Visit(s)

If a subject experiences an AE after discharge from the clinic but prior to the EOS visit, the subject should be instructed to call the site. Based on the issue, the PI (or designee) may request that the subject return to the site for an unscheduled visit. In this case, procedures/assessments should be conducted as deemed appropriate for the situation by the PI (or designee). The visit should be recorded in the unscheduled visit page of the CRF.

7. STUDY PROCEDURES/EVALUATIONS

7.1. Demography and Medical History

Demographics including age, gender, race, ethnicity, and medical history will be recorded for each subject. All significant medical history should be recorded. In general, significant medical history should include all ongoing events and all events occurring within the last 6 months. Clinically relevant or clinically significant events occurring greater than 6 months ago should be recorded. All surgeries occurring in adulthood should be recorded. If surgeries occurred more than 2 years ago, then only the year needs to be recorded on the CRF.

7.2. Clinical Evaluations

7.2.1. Physical Examinations

The PE will be performed by the PI or a designee that is licensed to perform a PE per local requirements. The initial PE performed at Screening and the final PE conducted at the EOS visit will include examination of all pertinent body systems as defined by the site standard PE body systems (general appearance, HEENT, lymphatic, cardiovascular, respiratory, GI, musculoskeletal, neurological, dermatological).

Subsequent PEs will be performed as shown in the Time and Events Schedule and will be targeted to any new signs or symptoms, any change from the previous status or any body system deemed pertinent to that subject by the PI (or designee). Clinically significant abnormalities should be recorded in the CRF; those occurring prior to dosing will be included in medical history unless the abnormality was the direct result of study participation.

7.2.2. Vital Sign Measurements and ECGs

Vital signs include pulse rate, respiratory rate, blood pressure, and body temperature. Vital signs should be measured after the subject has been supine for a minimum of 5 minutes. Site standard ranges will be used for determining any out-of-range values.

Height and weight should be measured, and BMI calculated at Screening as indicated in the Time and Events Schedule.

Resting 12-lead ECGs should be recorded at the visits indicated in the Time and Events Schedule after the subject has been supine for a minimum of 5 minutes. The PI (or designee) will evaluate the ECG tracings to determine if there are out-of-range values; if out-or-range values are detected, the PI (or designee) will determine if they are clinically significant. Site standard ranges will be used to determine if any parameters are considered out-of-range. At the discretion of the PI (or designee), the ECG may be repeated if erroneous readings are suspected.

7.2.2.1. Continuous 12-lead ECG Monitoring (Part 3 Only)

Continuous 12-lead ECG monitoring using a digital recorder will take place at the times indicated in the Time and Events Schedule, in P3 only.

All continuous 12-lead ECG data collected will be archived without extraction or analysis and will not be reported in the scope of this study.

Subjects will be supine for at least 10 minutes before the extraction timepoint and for 5 minutes from the start of each extraction timepoint (each extraction will last for 5 minutes). Environmental distractions (e.g., television, radio, conversation) should be avoided during the pre-ECG resting period and during ECG recording.

When coinciding, PK sampling should always be performed after the ECG extraction time window. If a separate ECG machine is being used for safety assessments, that machine should be in place prior to the extraction window to permit safety ECGs to be recorded irrespective of the extraction window. If the machine is not in place prior to the extraction window, safety ECGs must be recorded after the extraction window. If an integral system is used, safety ECGs may be recorded irrespective of the extraction window.

7.2.3. Adverse Events and Concomitant Medications

Adverse Events: The PI is responsible for identifying and documenting events meeting the definition of an AE or SAE (Section 8.1). Once each day while the subject is in the clinic and once during each out-patient visit, the PI (or designee) should inquire about the occurrence of AEs. The following are examples of open-ended questions that may be used to obtain this information: *"How are you feeling?"; "Have you had any medical problems recently?"; "Have you taken any new medicines since your last visit/assessment?"*

All AEs and SAEs must be documented in the source documents and recorded in the CRF.

Concomitant Medications: All medications (prescription or over-the-counter), nutritional supplements, and nutraceuticals taken by the subject from 30 days prior to dosing through the EOS visit must be recorded in the CRF. Medication information should include indication, dose, frequency, and route of administration. Any medication taken for an AE/SAE should be documented as such. Refer to Section 5.5 for additional information.

7.3. Laboratory Evaluations

The laboratory will perform standard routine testing, and processing of all blood samples. For the entire study, the amount of blood collected from any one subject will not exceed 500 mL.

7.3.1. Routine Laboratory Panels

Blood and urine samples will be collected at the times indicated in the Time and Events Schedule. The analytes shown in the table below (Table 11) will be assessed.

If a method to determine COVID-19 status becomes readily available, subjects may be tested at Screening and Check-in to confirm they are not positive for COVID-19 prior to dosing.

Table 11: Laboratory Evaluations

CHEMISTRY PANEL Alanine Aminotransferase (ALT/SGPT) Albumin, Serum Albumin/Globulin (A/G) Ratio (calculation) Alkaline Phosphatase, Serum Amylase Aspartate Aminotransferase (AST/SGOT) Bilirubin, Total and Direct BUN BUN/Creatinine Ratio (calculation) Calcium, Serum Creatinine, Serum Creatinine Kinase (CK) Gamma Glutamyl Transferase (GGT) Lactate Dehydrogenase (LDH) Uric Acid Electrolyte Panel (Na+, K+, Cl-, Bicarb.) Phosphorus Globulin, Total Glucose, Serum Lipase Protein, Total, Serum	**HEMATOLOGY PANEL** Hemoglobin Hematocrit White Blood Cell Count with differential (absolute and percentage) Red Blood Count Prothrombin Time (PT)/Partial Prothrombin Time (PTT) and International Normalized Ratio (INR) Mean cell hemoglobin (MCH), mean cell hemoglobin concentration (MCHC), mean cell volume (MCV), red cell distribution width (RDW; may be a Grade 1 abnormality) Platelets
	ADDITIONAL ASSESSMENTS Virology: Human Immunodeficiency Virus (HIV) serology, Hepatitis B Virus (HBV; Surface Antigen [HBsAg]), Hepatitis C Virus (HCV) Follicle-Stimulating Hormone (FSH; as applicable)
	PREGNANCY TEST Serum Pregnancy Test Urine Dipstick (optional blood follow-up)
	DRUG SCREENING Serum/urinalysis (per site SOP) Cotinine Urine Dipstick Alcohol Breathalyzer
	ROUTINE URINALYSIS Bilirubin Color and appearance Glucose Ketones Leukocytes Microscopic (including red blood cells [RBCs] and white blood cells [WBCs]) Nitrite Occult blood pH Protein Specific Gravity Urobilinogen

7.3.2. Pharmacokinetic Sampling

Samples for PK analysis will be collected as shown in the Time and Events Schedule. All samples will be analyzed to define the PK parameters for prodrug EIDD-2801 and the active parent, EIDD-1931. In order to preserve EIDD-2801, special handling procedures will be put in place. These procedures will be documented in the laboratory manual for the study.

Depending on ongoing review of the PK data, timepoints may be modified or removed, and up to 3 additional samples (in each period for the food-effect) may be added.

7.3.3. Peripheral Blood Mononuclear Cell Collection

Peripheral blood mononuclear cells will be collected in P2 and P3. Blood will be collected into specialized tubes and processed according to procedures described in the laboratory manual.

Depending on ongoing review of the data, timepoints may be modified or removed, and up to 2 additional samples may be added.

7.3.4. Urine Collection

Urine will be collected over the time periods noted in the Time and Events Schedule. Samples will be collected for routine urinalysis and PK analysis according to site standard practices and as described in the laboratory manual.

8. SAFETY MONITORING, MANAGEMENT AND REPORTING

8.1. Definitions

8.1.1. Adverse Events

An AE is 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 an abnormal laboratory finding), symptom, or disease temporally associated with the use of a medicinal (investigational) product, whether or not related to the medicinal (investigational) product.

Abnormal clinical laboratory findings (e.g. clinical chemistry, hematology) or other abnormal assessments that are judged by the PI (or designee) as clinically significant will be recorded as AEs or SAEs if they meet the definitions of an AE or an SAE as defined in this Section 8.1.2. Disease specific signs and symptoms which were ongoing prior to study entry will not be considered AEs unless they worsen (e.g. increase in frequency or severity) unexpectedly during the course of the trial.

8.1.2. Serious Adverse Events

An AE or suspected adverse reaction is considered "serious" if, in the view of either the PI (or designee) or Sponsor, it results in any of the following outcomes:

- Death,
- A life-threatening AE: An AE or suspected adverse reaction is considered "life-threatening" if, in the view of either the PI or Sponsor, its occurrence places the patient or subject at immediate risk of death. It does not include an AE or suspected adverse reaction 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 hospitalizations may be considered serious when, based upon appropriate medical judgment they may jeopardize the patient or 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.

Suspected adverse reaction means any AE for which there is a reasonable possibility that the drug caused the AE. "Reasonable possibility" means there is evidence to suggest a causal relationship between the drug and the adverse event. Suspected adverse reaction implies a lesser degree of certainty about causality than adverse reaction, which means any adverse event caused by a drug.

An AE or suspected adverse reaction is considered "unexpected" if

- It is not listed in the IB or is not listed at the specificity or severity that has been observed; or, if an IB is not required or available,
- Is not consistent with the risk information described in the general investigational plan or elsewhere in the current application as amended. For example, under this definition, hepatic necrosis would be unexpected (by virtue of greater severity) if the IB referred only to elevated hepatic enzymes or hepatitis. Similarly, cerebral thromboembolism and cerebral vasculitis would be unexpected (by virtue of greater specificity) if the IB listed only cerebral vascular accidents.
- "Unexpected" as used in this definition, also refers to AE or suspected adverse reactions that are mentioned in the IB as occurring with a class of drugs or as anticipated from the pharmacological properties of the drug, but are not specifically mentioned as occurring with the particular drug under investigation.

As of the date of this protocol, there are no expected events listed in the current version of the IB; therefore, all AEs will be considered unexpected until such a time that the reference safety information in the IB is updated with any identified, expected events.

8.2. Documenting Adverse Events

8.2.1. Timeframe for Collection and Follow-up of AEs/SAEs

All AEs/SAEs will be collected from the time of the first study drug administration until the subject has completed the EOS visit and been discontinued from the study. This includes subjects who discontinue early. Events considered related to study drug will be followed as noted:

- AEs that are related to study drug and continue beyond the normal collection period (i.e., are ongoing at the time a subject exits the study) will be followed until resolution, return to baseline or until stabilized with sequelae for a maximum of 30 days following discontinuation. After 30 days, the AE will be closed, and the outcome noted (see Table 12).
- SAEs that are related to study drug and continue beyond the normal collection period

(i.e., are ongoing at the time a subject exits the study) will be followed until resolution, return to baseline or until stabilized with sequelae.

- Serious AEs that are reported to the site within 30 days after the subject has been discontinued from the study (i.e., completed the EOS visit) will be recorded. Those that are considered related to study drug will be followed as noted in the bullet above.

Note that all events which occurred prior to dosing with study drug should be recorded as medical history unless the event is directly related to study procedures.

8.2.2. Recording of Adverse Events/Serious Adverse Events

AEs/SAEs must be recorded in the CRF as indicated in the CRF completion instructions. Information to be collected includes event description, time of onset, clinician's assessment of severity (Section 8.2.3), relationship to study drug (Section 8.2.4), outcome (Section 8.2.5), and time of resolution/stabilization of the event. All AEs occurring while on study must be documented appropriately regardless of relationship and will be followed to adequate resolution as described above (Section 8.2.1). All SAEs will be recorded as noted above; SAEs reported to the site within 30 days following the EOS visit will also be recorded. All SAEs must be entered onto the SAE form and reported as discussed below (Section 8.3.1).

If an AE changes in severity, the highest severity will be recorded in the CRF. AEs characterized as intermittent require documentation of onset and duration of each episode.

8.2.3. Assessing Severity of Adverse Events

All AEs/SAEs will be assessed by the PI or those with the training and authority to make a medical judgment. AEs/SAEs will be graded according to the DMID Toxicity Grading Scale. For any AEs not specifically listed in the tables, the following guidelines should be used to grade severity:

- **Mild** (Grade 1); asymptomatic or mild symptoms; clinical or diagnostic observations only; intervention not indicated.
- **Moderate** (Grade 2); minimal, local or noninvasive intervention indicated; limiting age-appropriate instrumental activities of daily living.
- **Severe** (Grade 3); medically significant but not immediately life-threatening; hospitalization or prolongation of hospitalization indicated; disabling; limiting self-care activities of daily living.
    - **Life-threatening**; life-threatening consequences; urgent intervention indicated.
    - **Death**; death related to AE.

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

8.2.4. Relationship to Study Drug

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

- **Definite –** The AE is clearly related to the study drug**.**
- **Probable –** The AE is likely related to the study drug.
- **Possible –** The AE may be related to the study drug.
- **Unlikely –** The AE is doubtfully related to the study drug.
- **Unrelated –** The AE is clearly NOT related to the study drug.

8.2.5. Classifying Adverse Event Outcome

All AEs/SAEs in the study must be assigned an outcome by site staff. The outcome will be included on the AE CRF. Possible outcomes are shown below:

Table 12: Adverse Event Outcomes

Outcome	Description
Recovered / Resolved	AE resolved with no residual signs or symptoms; an event is considered resolved if it returns to baseline (pretreatment) values.
Recovered / Resolved with sequelae	AE stabilized but residual signs or symptoms remain; this includes stabilization of an event/condition with the expectation that it will remain chronic.
Not Recovered / Not Resolved	AE remains ongoing AND no or only minimal improvement has occurred.
Ongoing	AE has not yet resolved, but continues to improve/resolve and complete resolution is expected over time.
Fatal	Outcome of the AE is death.
Unknown	AE outcome is not known; usually because the subject has been Lost-to-Follow-up.

8.3. Reporting Procedures

8.3.1. Serious Adverse Events

The PI or clinical site personnel should notify Covance Drug Safety Services (DSS) of all SAEs, regardless of relationship to the investigational drug, within 24 hours of clinical site personnel becoming aware of the event. The PI (or designee) will provide the initial notification by sending a completed "SAE Notification Form," which must include the PI's (or designee's) assessment of the relationship of the event to investigational drug and must be signed by the PI. Follow-up information, or new information regarding an ongoing SAE, must be provided promptly to Covance DSS.

The responsibilities of Covance DSS include the following:

- Prepare an AE reporting plan prior to the start of the study. Where this plan differs from the applicable site standard operating procedure on SAE reporting, the AE reporting plan will always take precedence.

- Receive and review SAE report forms from the site and inform the Sponsor of the SAE within 1 working day of the initial notification to DSS. Drug Safety Services will delete any information from the SAE report forms that may identify the subject.

- Write case narratives and enter the case into Covance's safety database as defined in the AE reporting plan.

- Produce appropriate reports of all Suspected Unexpected Serious Adverse Reactions and forward to the EC, Medicines and Healthcare Products Regulatory Agency, PI's (or designee's), and the Sponsor within the timeframes stipulated in the Clinical Trials Directive Guideline (ENTR/CT 3).

The responsibility for reporting SAEs will be transferred to the Sponsor 28 days after the end of the study.

8.3.2. Pregnancy

Subjects in this study must be of non-childbearing potential. However, should a pregnancy occur any time from informed consent to the EOS visit, subjects must immediately report the event to the clinical site which in turn must immediately report the pregnancy to the Sponsor or their designee. The subject will be followed until the end of the pregnancy. A separate ICF will be used for consenting for follow-up pregnancy activities. Pregnancy will not be considered an AE unless deemed likely related to study drug. Pregnancy will not be considered an SAE unless there is an associated SAE. A spontaneous abortion (miscarriage) or abnormal outcome (including congenital anomalies) will be reported as an SAE.

8.4. Unmasking Treatment Assignment

The following controls will be employed to maintain the double-blind status of the study:

- Placebo will be identical in appearance to the EIDD-2801.
- The PI and other members of staff involved with the study will remain blinded to the treatment randomization code in P1 and P3.
- Interim bioanalytical data will be provided in a blinded manner.

To maintain the blind, the PI will be provided with a sealed randomization code for each subject in P1 and P3, containing details of their treatment. These individual sealed envelopes will be kept in a limited access area that is accessible 24 hours a day. In order to manage subject safety or to support dose-escalation decisions (in the event of possibly treatment-related SAEs or severe AEs), the decision to unblind resides solely with the PI. Whenever possible and providing it does not interfere with or delay any decision in the best interest of the subject, the PI will discuss the intended code-break with the Sponsor. If it becomes necessary to break the code during the study, the date, time, and reason will be recorded in the subject's source data and on the individual envelope and will be witnessed by a second person.

The pharmacy and bioanalytical lab will have access to the treatment randomization and will be unblinded. Pharmacokinetic personnel may be unblinded to perform interim PK analysis and to ensure that PK data are provided in a blinded manner for dose-escalation decisions.

9. DOSE-ESCALATION AND HALTING RULES

9.1. Guidelines for Dose-Escalation

Doses will be administered in an escalating manner following satisfactory review by the Sponsor and PI of the blinded safety and tolerability data up to 72 hours post final dose. [redacted] As PK data become available, these data will be reviewed and may be used to assist in dose selection.

Doses may be reduced and may be lower than the starting dose. There will be a minimum of 4 days between dose escalations to allow sufficient time for an adequate safety review.

Dose-escalation in P1 and P3 will only occur if data from a minimum of 6 subjects have been reviewed from the previous lower dose group, such that data from a minimum of 4 subjects who have received EIDD-2801 will be used to make the dose-escalation decision.

The justification for this is as follows:

- The study treatment is of a known pharmacological class for which the on-target effects in humans are well characterized. Based upon nonclinical data, no clinically important off target effects are expected within the proposed dose range.

- A minimum of 4 subjects receiving the active drug is considered sufficient to characterize the safety profile of EIDD-2801.

Between each dose-escalation, the PI will review all available data in a blinded manner to ensure it is safe to proceed with the planned dose-escalation. The results from all available safety assessments will be sent to the Sponsor prior to the start of each successive group/treatment period. Any clinically significant results will be discussed with the Sponsor before dose-escalation continues. Interim PK data may also be reviewed in terms of dose-escalation and to confirm that the study design remains appropriate. In the event of a disagreement between Sponsor and PI on the dose-escalation decision, the most conservative decision will be upheld.

Dose levels will not increase by more than 3-fold for predicted nonpharmacologically active dose levels and by 2-fold for predicted pharmacologically active dose levels. [redacted]

Dose levels in P3 may be repeated for collection of PBMCs.

9.2. Dose-Escalation Halting Rules

For Group 1 in P1, sentinel dosing will occur whereby 2 subjects (1 active and 1 PBO) will be dosed on 1 day and, providing no safety concerns arise, the remaining 6 subjects will be dosed after at least 24 hours.

The study will be halted (i.e., no further dosing will occur) if one or more subjects experience an SAE that is considered to be related to EIDD-2801 or 2 or more subjects in the same group experience severe AEs that are considered to be related to EIDD-2801. If, following an internal safety review, the Sponsor deems it appropriate to restart the study, this can be done following approval of a substantial protocol amendment.

Dose-escalation will stop if clinically relevant signs or symptoms of similar nature occur in 2 or more subjects in a group that, in the opinion of the PI, warrant stopping of dose-escalation.

10. STATISTICAL CONSIDERATIONS

10.1. General Considerations

All summaries will be provided by study part and treatment. Continuous variables will be summarized using descriptive statistics including number of observations (n), mean, standard deviation, minimum (min), median (med), and maximum (max). Categorical variables will be summarized using frequency counts and percentages. Note that PBO data from each dose level in Parts 1 and 3 will be combined into one PBO group.

No missing data imputation will be performed.

Subject listings will be provided for all the data collected during the study period.

Specific information about the statistical analysis will be provided in a SAP that will be reviewed and approved by the Sponsor and will be finalized before final database lock. If there is a discrepancy between the methods described in the protocol and final approved SAP, the SAP will take precedence.

10.2. Sample Size Considerations

No formal sample size calculation was conducted. The sample size of 8 per cohort (6 active: 2 PBO) for the SAD and MAD cohorts is considered adequate for a Phase 1 FIH study. The sample size of 10 subjects (all administered EIDD-2801) is in accordance with FDA guidelines for sample size in FE studies.

10.3. Analysis Populations

Safety Population: All subjects who receive at least one dose of study drug.

Pharmacokinetic Population: All subjects receiving study drug who comply sufficiently with protocol requirements (i.e., no major protocol violations) and for whom a sufficient number of analyzable PK samples have been obtained to permit the determination of the PK profile for EIDD-2801/EIDD-1931 and the statistical analysis of the results. Subjects who experience an AE of vomiting before 2× the median t_{max} of the group may be excluded from the PK population.

10.4. Analysis of Safety Data

All safety analyses will be performed on the Safety Population as defined in Section 10.3. Safety will be assessed on the basis of AEs, clinical laboratory data, vital signs, ECG parameters, and PEs.

10.4.1. Extent of Exposure

Dosing data will be listed by study part.

10.4.2. Adverse Events

Adverse events will be coded using Medical Dictionary for Regulatory Activities (MedDRA) preferred term and system organ classification. Any events reported after the initiation of study treatment and through the EOS are defined as treatment-emergent. The occurrence of treatment-emergent AEs will be summarized using MedDRA preferred terms, system organ classifications, and severity. Separate summaries of treatment-emergent SAEs and AEs considered related to study treatment and AEs leading to study treatment discontinuation will be generated. All AEs will be listed for individual subjects showing both verbatim and preferred terms.

10.4.3. Clinical Laboratory Results

Laboratory abnormalities will be graded according to the DMID Toxicity Grading Scale. Any graded abnormality that occurs following the initiation of study treatment and represents at least a one-grade increase from the baseline assessment is defined as treatment-emergent. The number and percentage of subjects experiencing treatment-emergent graded toxicities will be summarized. Raw values and mean changes from baseline in clinical laboratory measures will be summarized.

Listings of the clinical laboratory test results will be provided. Abnormal laboratory values will be flagged in the listings.

10.4.4. Other Parameters

Individual data for ECG parameters and vital sign measurements will be listed by subject and time point and summarized for each treatment. Individual data for PE will be listed by subject and time point.

Concomitant medications will be coded using the World Health Organization-Drug Dictionary (WHO-DD) and summarized.

10.5. Analysis of Pharmacokinetic Data

Pharmacokinetic analysis as defined in the SAP will be conducted using the PK population defined in Section 10.3. In the event of discrepancies between analyses described in the SAP and this clinical study protocol, the SAP will supersede the protocol.

- All samples will be analyzed and all concentrations listed.
- Descriptive statistics will be performed for all time points available, with the exclusion of subjects who had any significant protocol deviation.
- Pharmacokinetic parameters will be derived where possible for all subjects. Data from subjects with incomplete profiles (missed blood draws, lost samples, samples unable to be quantified) may be used if PK parameters can be estimated using the remaining data points.

- Descriptive statistics will be performed on all parameters available, and any missing parameters will be flagged.

Plasma concentration data for EIDD-2801 and EIDD-1931 will be listed for individual subjects and summarized by study part and treatment. Individual and mean plasma concentration versus time plots for EIDD-2801 and EIDD-1931 will be provided. Urine concentration data for EIDD-2801 and EIDD-1931 will be listed.

Plasma PK parameters of EIDD-2801 and EIDD-1931 for each subject will be estimated over the sampling interval using noncompartmental analysis and summarized by study part and treatment using descriptive statistics. Actual blood sampling times will be used for plasma PK analysis.

Urine PK parameters of EIDD-2801 and EIDD-1931 will also be analyzed and summarized when possible. The PK parameters that will be estimated are listed in the table below.

Table 13: Pharmacokinetic Parameters

Plasma PK Parameter	Description
AUC_{last}	The area under the plasma concentration-time curve, from time 0 to the last measurable non-zero concentration, as calculated by the linear up/log down trapezoidal method (P1 and P2 only).
$AUC_{0\text{-}inf}$	The area under the plasma concentration-time curve from time 0 extrapolated to infinity. $AUC_{0\text{-}inf}$ is calculated as the sum of AUC_{last} plus the ratio of the last measurable plasma concentration to the elimination rate constant (λz) (P1, P2, and Day 1 dose of P3).
C_{max}	Maximum observed concentration.
t_{max}	Time to reach C_{max}. If the maximum value occurs at more than one time point, t_{max} is defined as the first time point with this value.
λz	Apparent terminal elimination rate constant; represents the fraction of medication eliminated per unit time.
t½	Apparent terminal elimination half-life of medication, calculated as 0.693/λz.
Vz/F	Apparent volume of distribution (EIDD-2801 only).
CL/F	Apparent oral drug clearance (EIDD-2801 only).
C_{trough}	Trough concentration (P3 only).
AUC_{τ}	The area under the plasma concentration-time curve during a dosing interval (P3 only).
$RA_{AUC\tau}$	Observed accumulation ratio based on AUC_{τ} (P3 only).
RA_{Cmax}	Observed accumulation ratio based on C_{max} (P3 only).
Urine PK Parameter	**Description**
A_e	Amount excreted in urine over the sampling interval.
F_e	Fraction of drug excreted in the urine over sampling interval.
CL_R	Renal clearance, $CL_R = A_e/AUC$ where both A_e and AUC are determined over co-incident time ranges after dosing.

Additional PK parameters may be analyzed as appropriate.

10.5.1. Statistical Analysis of Pharmacokinetic Data

10.5.1.1. Dose Proportionality

In P1, dose-proportionality for PK parameters of EIDD-2801 and EIDD-1931 ($AUC_{0\text{-}inf}$, AUC_{last}, and C_{max}) will be assessed by the power model. The slope and the associated 90% CI based on the power model will be reported.

In Part 3 on Day 7 dose-proportionality for PK parameters of EIDD-2801 and EIDD-1931 (AUC_{τ}, and C_{max}) will be assessed by the power model. The slope and the associated 90% CI based on the power model will be reported.

10.5.1.2. Food-Effect

To assess the effect of food on the PK of EIDD-2801 and EIDD-1931, natural log transformed PK parameters from P2 ($AUC_{0\text{-}inf}$, AUC_{last}, and C_{max}) will be analyzed using a mixed-effects model with fixed-effect terms for treatment (with or without food), period, and sequence and with subject within sequence as a random effect. Point estimates and their associated 90% CIs for the natural log-transformed PK parameters for the difference between EIDD-2801 administered under fed relative to fasted conditions will be constructed. The point estimates and interval estimates will be back transformed to give estimates of the ratio and 90% CI for the geometric least squares mean ratio for fed/fasted to assess the effect of food.

10.6. Peripheral Blood Mononuclear Cells

Peripheral blood mononuclear cellss will be collected from subjects following each dose in Part 2. Depending upon ongoing review of the data, PBMCs may be collected from subjects in P3; the collection of these samples may be omitted from some cohorts in P3.

The PBMCs will be stored for analysis of EIDD-2061 levels at some point in the future and reported separately.

10.7. Interim Analyses

There are no formal interim analyses planned for this study. However, interim analyses may be implemented at the discretion of the Sponsor or health authority request. In addition, data from P1 cohorts may be locked, unblinded, and analyzed prior to conducting P2 and P3.

11. STUDY ADMINISTRATION

11.1. Regulatory and Ethical Considerations

11.2. Ethical Standard

This study will be conducted in accordance with the protocol and with the following:

- Consensus ethical principles derived from international guidelines including the Declaration of Helsinki and Council for International Organizations of Medical Sciences International Ethical Guidelines.
- Applicable ICH GCP Guidelines.
- United States CRFs applicable to clinical studies (45 CFR Part 46; 21 CFR Part 50, 21 CFR Part 56, and 21 CFR Part 312).
- Applicable laws and regulations.

11.2.1. Ethics Committee Approval

The PI (or designee) must ensure that all required study-specific documents and/or information are submitted to the EC for review and approval as appropriate including but not limited to:

- the protocol and any future protocol amendments
- ICF and any other documents (electronic or paper) given to the subject
- IB

Any substantial protocol amendments, likely to affect the safety of the subjects or the conduct of the study, will require EC and regulatory authority (as locally required) approval before implementation of changes made to the study design, except for changes necessary to eliminate an immediate hazard to study subjects or any nonsubstantial changes, as defined by regulatory requirements.

11.2.2. Informed Consent

Informed consent is a process that is initiated prior to the individual's agreeing to participate in the study and continuing throughout the individual's study participation. Extensive discussion of risks and possible benefits of this therapy will be provided to the subjects and, if applicable, their families. Consent forms describing in detail the study interventions/products, study procedures, and risks are given to the subject and written documentation of informed consent is required prior to starting any screening procedures or intervention/administering study product. Consent forms will be EC-approved and the subject will be asked to read and review the document.

Upon reviewing the document, the PI (or designee) will explain the research study to the subject and answer any questions that may arise. The subjects will sign the ICF prior to any procedures being done specifically for the study. The subjects should have the opportunity to discuss the study with their surrogates or think about it prior to agreeing to participate. The subjects may withdraw consent at any time throughout the course of the trial. A copy of the ICF will be given to the subjects for their records. The rights and welfare of the subjects will be protected by emphasizing to them that the quality of their medical care will not be adversely affected if they decline to participate in this study.

11.3. Financing and Insurance

Financing and insurance will be addressed in a separate agreement.

11.4. Source Documentation and Access

The site will maintain appropriate medical and research records in compliance with ICH E6, Section 4.9 and regulatory and institutional requirements for the protection of confidentiality of subjects. The site will permit authorized representatives of the Sponsor, its designees, and appropriate regulatory agencies to examine (and when required by applicable law, to copy) clinical records for the purposes of quality assurance reviews, audits, and evaluation of the study safety and progress. These representatives will be permitted access to all source data and source documents, which include, but are not limited to, clinical and office charts, laboratory notes, memoranda, subjects' memory aid or evaluation checklists, pharmacy dispensing records, recorded data from automated instruments, copies or transcriptions certified after verification as being accurate and complete, photographic negatives, microfilm or magnetic media, x-rays, and subject files and records kept at the pharmacy, at the laboratory, and medico-technical departments involved in the clinical trial.

11.5. Data Collection and Record Keeping

11.5.1. Data Collection

Data collection and data entry are the responsibility of the clinical trial staff at the site. The PI is responsible for ensuring the accuracy, completeness, legibility, and timeliness of the data reported. All source documents should be completed in a neat, legible manner to ensure accurate interpretation of data.

A CRF must be completed for every subject who signs the ICF and has at least one protocol-specified assessment conducted. The CRF must be completed and processed according to the CRF guidelines and the SOPs of the site. All data should be entered into the CRF, where possible, within 3 days after each visit for any one subject. After the subject has completed the study, the PI must review and sign the signature page of the CRF indicating that he has reviewed the completed CRF and pertinent clinical data for that subject and that, to the best of his knowledge, all data recorded in the CRF accurately reflects the subject's clinical performance in the study

11.5.2. Study Records Retention

Study documents should be retained for a minimum of 5 years after the end of the study. These documents should be retained for a longer period; however, if required by local regulations. No records will be destroyed without the written consent of the Sponsor, if applicable. It is the responsibility of the Sponsor to inform the PI when these documents no longer need to be retained.

11.5.3. Protocol Deviations

A protocol deviation is any noncompliance with the protocol or study procedures detailed in the laboratory or pharmacy manuals. The noncompliance may have been the result of action by the PI, site staff, or subject. All deviations should be handled in accordance with site SOPs.

It is the responsibility of the site to use continuous vigilance to identify and report deviations within 5 working days of identification of the protocol deviation, or within 5 working days of the scheduled protocol-required activity.

11.6. Clinical Monitoring

Site monitoring is conducted to ensure that the human subjects' protections, study and laboratory procedures, study intervention administration, and data collection processes are of high quality and meet Sponsor, ICH/GCP guidelines and applicable regulations, and that this trial is conducted in accordance with the protocol, protocol-specific MOP and applicable Sponsor SOPs. Experienced clinical monitors of the Sponsor or its designee will conduct site-monitoring visits as detailed in the clinical monitoring plan.

Site visits will be made at standard intervals as defined by the Sponsor and may be made more frequently as directed by the Sponsor. Monitoring visits will include, but are not limited to, review of regulatory files, accountability records, CRFs, ICFs, medical and laboratory reports, and protocol and GCP compliance. Clinical monitors will have access to each participating site, study personnel, and all study documentation according to the site monitoring plan. Clinical monitors will meet with the site PI to discuss any problems and actions to be taken and will document site visit findings and discussions. As data are entered into a CRF, data will be monitored to ensure accuracy as well as adherence to data entry instructions provided in the CRF guidelines.

11.7. Quality Control and Quality Assurance

A quality management plan will be put in place for this study. The site is responsible for conducting routine quality assurance and quality control activities to internally monitor study progress and protocol compliance. The PI will provide direct access to all study-related sites, source data/data collection forms, and reports for the purpose of monitoring and auditing by the Sponsor, and inspection by local and regulatory authorities. The PI will ensure all study personnel are appropriately trained and applicable documentations are maintained on site.

11.8. Study Termination and Closure Procedures

11.8.1. Study Termination

This study may be temporarily suspended or prematurely terminated if there is sufficient reasonable cause. Written notification, documenting the reason for study suspension or termination, will be provided to the site. If the study is terminated or suspended, the PI (or designee) will inform study participants and the EC. The Sponsor will notify appropriate regulatory authorities. The Sponsor will provide the reason(s) for the termination or suspension. Study participants will be contacted, as applicable, and be informed of changes to study visit schedule.

If suspended, the study may resume once issues that caused suspension of the study are resolved.

11.8.2. Termination Procedures

If the study is prematurely terminated, then the site must return all appropriate study data, resolve all data queries, complete final drug accountability, return any study drug remaining on site, and ship all biological samples (including PK and PBMCs) to the laboratory designated by the Sponsor. The PI (or designee) must notify the EC of study termination.

11.9. Information Disclosure

11.9.1. Confidentiality

Subject confidentiality and privacy is strictly held in trust by the PI, site staff, and the Sponsor(s) and their agents. The study protocol, documentation, data, and all other information generated will be held in strict confidence. No information concerning the study or the data will be released to any unauthorized third party without prior written approval of the Sponsor.

The study monitor or other authorized representatives of the Sponsor or regulatory agencies may inspect all documents and records required to be maintained by the PI, including but not limited to, medical records (office, clinic, or hospital) and pharmacy records for the participants in this study. The clinical study site will permit access to such records.

The study participant's contact information will be securely stored at the clinical site for internal use during the study. At the end of the study, all records will continue to be kept in a secure location for as long a period as dictated by the Sponsor, site, or regulatory requirements.

Study participant research data will be transmitted to and stored securely by the Sponsor's designated data center. This will not include the participant's contact or identifying information. Individual participants and their research data will be identified by a unique study identification number. The study data entry and study management systems used by clinical sites will be secured and password-protected. At the end of the study, all study databases will be de-identified and archived at a secure location.

11.9.2. Clinicaltrials.gov

This clinical study will be registered on clinicaltrials.gov as required.

11.9.3. Publication Policy

All information generated from this study is the proprietary property of the Sponsor. It is the intent of the Sponsor to publish the results of the study in their entirety as deemed appropriate.

12. REFERENCES

Agostini ML, Pruijssers AJ, Chappell JD, Gribble J, Lu X, Andres EL, et al. Small-Molecule Antiviral β-d-N4-Hydroxycytidine Inhibits a Proofreading-Intact Coronavirus with a High Genetic Barrier to Resistance. J Virol. 2019 Dec 15;93(24).

Flavell RA, Sabo DL, Bandle EF, Weissmann C. Site-directed mutagenesis: generation of an extracistronic mutation in bacteriophage Q beta RNA. J Mol Biol. 1974 Oct 25;89(2):255–72.

Hernandez-Santiago BI, Beltran T, Stuyver L, Chu CK, Schinazi RF. Metabolism of the anti-hepatitis C virus nucleoside beta-D-N4-hydroxycytidine in different liver cells. Antimicrob Agents Chemother. 2004 Dec;48(12):4636–42.

Mullins JI, Heath L, Hughes JP, Li D, Essigmann JM, Harris KS, et al. Mutations based on viral decay acceleration in the HIV-1 genomes of a clinical population treated with the mutagenic nucleoside KP1461. Retrovirology. 2011 Oct 3;8(Suppl 2):P84.

Painter GR, Bowen RA, Bluemling GR, DeBergh J, Edpuganti V, Gruddanti PR, et al. The prophylactic and therapeutic activity of a broadly active ribonucleoside analog in a murine model of intranasal venezuelan equine encephalitis virus infection. Antiviral Res. 2019;171:104597.

Sticher ZM, Lu G, Mitchell DG, Marlow J, Moellering L, Bluemling GR, et al. Analysis of the Potential for N4-Hydroxycytidine To Inhibit Mitochondrial Replication and Function. Antimicrob Agents Chemother. 2020 27;64(2).

Stuyver LJ, Whitaker T, McBrayer TR, Hernandez-Santiago BI, Lostia S, Tharnish PM, et al. Ribonucleoside analogue that blocks replication of bovine viral diarrhea and hepatitis C viruses in culture. Antimicrob Agents Chemother. 2003 Jan;47(1):244–54.

Toots M, Yoon J-J, Cox RM, Hart M, Sticher ZM, Makhsous N, et al. Characterization of orally efficacious influenza drug with high resistance barrier in ferrets and human airway epithelia. Sci Transl Med. 2019 Oct 23;11(515):eaax5866.

Urakova N, Kuznetsova V, Crossman DK, Sokratian A, Guthrie DB, Kolykhalov AA, et al. β-d-N4-Hydroxycytidine Is a Potent Anti-alphavirus Compound That Induces a High Level of Mutations in the Viral Genome. J Virol. 2018 01;92(3).

Yoon J-J, Toots M, Lee S, Lee M-E, Ludeke B, Luczo JM, et al. Orally Efficacious Broad-Spectrum Ribonucleoside Analog Inhibitor of Influenza and Respiratory Syncytial Viruses. Antimicrob Agents Chemother. 2018;62(8).

13. APPENDICES

Appendix 1: Time and Events Schedule

Parts 1 and 2

Visit	Screening (Days -28 to -2)	Day -1	Day 1	Day 2 (24 hr)	Day 3 (48 hr)	Day 4 (72 hr)	Day 9	Day 15 / EOS[1] (non-FE cohorts)
Food-Effect Cohort		**Day 14**	**Day 15**	**Day 16 (24 hr)**	**Day 17 (48 hr)**	**Day 18 (72 hr)**	**Day 23**	**Day 30 / EOS**
ICF; Demography	x							
I/E; Medical history	x	x						
Physical examination[2]	x	x		x		x	x	x
Qualifying laboratory analyses[3]	x	x						
Drug screening and pregnancy test[4]	x	x					x	x
Height, weight (BMI)	x							
Clinic confinement[5]		x	x	x	x	x		
Non-residential visit	x						x	x
Clinical chemistry and urinalysis[6]	x	x		x		x	x	x
Hematology[7]	x	x		x	x	x	x	x
PBMC collection (FE cohort ONLY)			x[8]	x[8]				
ECG	x		x[9]			x		x
Vital signs	x	x	x[10]	x	x	x	x	x
Administer study drug			x					
Plasma PK sample collection			x[11]	x[12]	x[12]	x[12]		
Urine PK sample collection			x[13]	x[14]	x[14]			
Adverse events	x[15]	x[15]	x[15]	x	x	x	x	x
Prior and/or Concomitant medications	x	x	x	x	x	x	x	x

Abbreviations: BMI (body mass index); EOS (End of Study); FE (food-effect); ICF (Informed Consent Form); I/E (Inclusion/Exclusion Criteria); ECG (electrocardiogram); PBMC (peripheral blood mononuclear cell); PK (pharmacokinetic).

[1] On Day 15, conduct the EOS visit for all subjects except those in the FE cohort.

[2] A complete PE will be performed at Screening and/or Check-in, and EOS visits; on all other days, a limited PE will be conducted targeting any change from the previous status or any body system deemed pertinent to that subject by the PI (or designee).

[3] Laboratory analyses to qualify for study entry include: total white cell count or absolute lymphocyte count (values must be in the normal range), hemoglobin or platelet levels (values must not be below the lower limit of normal), and ALT/SGPT, alkaline phosphatase, AST/SGOT (values must not be above the upper limit of normal), plus virology (HIV/HCV/HBV) (all must be negative), and FSH (if required).

[4] Drug screening (urine drug screen, UDS), including a cotinine test, should be conducted at Screening, Day -1, Days 9 and 23, and at EOS. Pregnancy tests should be conducted in serum at Screening and in urine at all other times (i.e., Days -1, 9, and 23, and EOS). A positive urine pregnancy test will be confirmed with a serum pregnancy test. An alcohol breath test should be conducted at Screening and Day -1.

[5] Participants should be admitted to the clinic following review of final I/E criteria on Day -1 and discharged from the clinic following completion of study specific assessments on Day 4. Subjects in the FE cohort should be readmitted to the clinic on Study Day 14.

[6] Safety laboratory assessments include clinical chemistry and urinalysis. Baseline samples will be collected after clinic admission on Day -1.

[7] Hematology values include, but are not limited to, RBC, WBC with differential and platelets. PT/PTT and INR assessed only at

Screening.

[8] PBMCs will be collected 2 hr postdose on Day 1/15, and predose and 8 hrs postdose (on Day 16). Depending on ongoing review of the data, timepoints may be modified or removed, and up to 2 additional samples may be collected.

[9] The baseline ECG should be conducted prior to dosing on Day 1/15.

[10] Vital signs include pulse rate, respiratory rate, blood pressure, and body temperature; on Day 1/15, VS should be taken predose, and at 2, 4, 8 and 12 hr postdose.

[11] PK samples should be collected predose and at 0.5, 1, 1.5, 2, 2.5, 3, 4, 6, 9, 12, and 15 hr postdose on Day 1/15. Depending on ongoing review of the PK data, timepoints may be modified or removed, and up to 3 additional samples (in each period for the food-effect) may be added.

[12] PK samples should be collected at 24, 36, 48, and 72 hr postdose on Days 2/16, 3/17, 4/18 respectively. Depending on ongoing review of the PK data, timepoints may be modified or removed, and up to 3 additional samples (in each period for the food-effect) may be added.

[13] Urine samples for PK analysis should be collected predose (spot collection), and from 0 to 4, 4 to 8, 8 to 12, and 12 to 24 hr postdose.

[14] Urine samples for PK analysis should be collected from 24 to 36 and 36 to 48 hr postdose.

[15] Adverse events occurring prior to study drug administration should be recorded as medical history unless the event is directly related to a study specified procedure.

Part 3 (7 Days of Dosing)

Visit	Screening (Days -28 to -2)	Day -1	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7	Day 8	Day 9	Day 10	Day 15	Day 21 / EOS
ICF; Demography	x													
I/E; Medical history	x	x												
Physical examination[1]	x	x		x						x			x	x
Qualifying laboratory analyses[2]	x	x												
Drug screening and pregnancy test[3]	x	x											x	x
Height, weight (BMI)	x													
Clinic confinement[4]		x	x	x	x	x	x	x	x	x	x	x		
Non-residential visit	x												x	x
Clinical chemistry and urinalysis[5]	x	x			x			x				x		x
Hematology[6]	x	x			x			x				x	x	x
PBMC collection[7]			x				x							
ECG[8]	x		x						x			x		x
Continuous 12-lead ECG			x[9]						x[10]	x[10]				x
Vital signs[11]	x	x	x	x	x	x	x	x	x	x	x	x	x	x
Administer study drug[12]			x	x	x	x	x	x	x					
Plasma PK sample collection[13]			x			x	x	x	x	x	x	x	x	
Urine PK sample collection[14]			x						x	x	x	x		
Adverse events	x[15]	x[15]	x[15]	x	x	x	x	x	x	x	x	x	x	x
Prior and/or Concomitant medications	x	x	x	x	x	x	x	x	x	x	x	x	x	x

Abbreviations: BID (twice daily); BMI (body mass index); EOS (End of Study); ICF (Informed Consent Form); I/E (Inclusion/Exclusion Criteria); ECG (electrocardiogram); PK (pharmacokinetic).

[1] A complete PE will be performed at Screening and/or Check-in, and EOS visits; on all other days, a limited PE will be conducted targeting any change from the previous status or any body system deemed pertinent to that subject by the PI (or designee).

[2] Laboratory analyses to qualify for study entry include: total white cell count or absolute lymphocyte count (values must be in the normal range), hemoglobin or platelet levels (values must not be below the lower limit of normal), and ALT/SGPT, alkaline phosphatase, AST/SGOT (values must not be above the upper limit of normal), plus virology

(HIV/HCV/HBV) (all must be negative), and FSH (if required).

[3] Drug screening (urine drug screen, UDS), including cotinine, should be conducted at Screening, Day -1, Day 15, and at EOS. Pregnancy tests should be conducted in serum at Screening and in urine at all other times (i.e., Days -1 and 15, and EOS). A positive urine pregnancy test will be confirmed with a serum pregnancy test. An alcohol breath test should be conducted at Screening and Day -1.

[4] Participants should be admitted to the clinic following review of final I/E criteria on Day -1 and discharged from the clinic following completion of study specific assessments on Day 10.

[5] Safety laboratory assessments include clinical chemistry and urinalysis. Baseline samples will be collected after clinic admission on Day -1. On Days 3 and 6, the assessment will be pre-am dose. On Day 10, the assessment will be 72 hrs post-am dose on Day 7.

[6] Hematology values include, but are not limited to, RBC, WBC with differential and platelets. PT/PTT and INR assessed only at Screening.

[7] PBMCs will be collected 2 hrs postdose, relative to the first daily dose on Day 1, and pre-am dose and 2 and 8 hrs postdose relative to the first daily dose on Day 5. Depending on ongoing review of the data, timepoints may be modified or removed, and up to 2 additional samples may be collected. Additionally, PMBC collection may be omitted from some cohorts.

[8] The baseline ECG should be conducted prior to first dosing on Day 1. On Day 1, ECG should be taken predose and 2 hours post-am dose. On Day 7, ECG should be taken predose, 2 hrs, and 72 hrs (Day 10) postdose.

[9] On Day 1, monitor for 12-lead ECG recording will be worn from 2 hrs predose to 12 hrs postdose. Extraction timepoints will be 60, 45, and 30 minutes predose and at 0.5, 1, 1.5, 2, 2.5, 3, 4, 6, 9, and 12 hrs postdose.

[10] On Day 7, monitor for 12-lead ECG recording will be worn from dosing (0 hr) to 25 hrs (Day 8) postdose. Extraction timepoints will be at 0.5, 1, 1.5, 2, 2.5, 3, 4, 6, 9, 12, and 24 hrs postdose.

[11] Vital signs include pulse rate, respiratory rate, blood pressure, and body temperature. On Day 1, VS should be taken pre-am dose, and at 2, 4, and 8 hrs post-am dose. On Day 2 through 6, VS should be taken pre-am dose. On Day 7, VS should be taken predose, and 2, 4, 8, 24 (Day 8), 48 (Day 9), and 72 (Day 10) hrs postdose.

[12] Study drug should be administered BID, 12 hrs apart on Days 1 through 6. On Day 7, study drug should be administered once in the morning only (0 hr). Depending on ongoing review of the safety, tolerability, and PK data, the dosing frequency and the number of days of dosing may be changed; however, the dosing frequency will be no less than once-daily or no greater than three-times daily, and the number of days of dosing will be no fewer than 5 days and no greater than 10 days.

[13] On Day 1, PK samples should be collected pre-am dose and at 0.5, 1, 1.5, 2, 2.5, 3, 4, 6, 9, and 12 hrs post-am dose. On Days 4 through 6, PK samples should be collected pre-am dose. On Day 7, PK samples should be collected predose and at 0.5, 1, 1.5, 2, 2.5, 3, 4, 6, 9, 12, 15, 24 (Day 8), 48 (Day 9), 72 (Day 10), and 192 (Day 15) hrs postdose. Depending on ongoing review of the PK data, timepoints may be modified or removed, and up to 3 additional samples may be added.

[14] On Day 1, urine samples for PK analysis should be collected predose (spot collection), and from 0 to 4, 4 to 8, and 8 to 12 hrs postdose. On Day 7, urine samples for PK analysis should be collected predose (spot collection), and from 0 to 4, 4 to 8, 8 to 12, 12 to 24 (Day 8), 24 to 48 (Day 9), and 48 to 72 hrs (Day 10). Sampling timepoints may be modified, removed, or additional timepoints added depending upon ongoing review of the data. The 12-hour urine collection should occur prior to the second daily dose administration.

[15] Adverse events occurring prior to study drug administration should be recorded as medical history unless the event is directly related to a study specified procedure.

Part 3 (5 Days of Dosing)

Visit	Screening (Days -28 to -2)	Day -1	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7	Day 8	Day 13	Day 19 /EOS
ICF; Demography	x											
I/E; Medical history	x	x										
Physical examination[1]	x	x		x				x			x	x
Qualifying laboratory analyses[2]	x	x										
Drug screening and pregnancy test[3]	x	x									x	x
Height, weight (BMI)	x											
Clinic confinement[4]		x	x	x	x	x	x	x	x	x		
Non-residential visit	x										x	x
Clinical chemistry and urinalysis[5]	x	x			x		x			x		x
Hematology[6]	x	x			x		x			x	x	x
PBMC collection[7]			x				x					
ECG[8]	x		x				x			x		x
Continuous 12-lead ECG			x[9]				x[10]	x[10]				x
Vital signs[11]	x	x	x	x	x	x	x	x	x	x	x	x
Administer study drug[12]			x	x	x	x	x					
Plasma PK sample collection[13]			x			x	x	x	x	x	x	
Urine PK sample collection[14]			x				x	x	x	x		
Adverse events	x[15]	x[15]	x[15]	x	x	x	x	x	x	x	x	x
Prior and/or Concomitant medications	x	x	x	x	x	x	x	x	x	x	x	x

Abbreviations: BID (twice daily); BMI (body mass index); EOS (End of Study); ICF (Informed Consent Form); I/E (Inclusion/Exclusion Criteria); ECG (electrocardiogram); PK (pharmacokinetic).

[1] A complete PE will be performed at Screening and/or Check-in, and EOS visits; on all other days, a limited PE will be conducted targeting any change from the previous status or any body system deemed pertinent to that subject by the PI (or designee).

[2] Laboratory analyses to qualify for study entry include: total white cell count or absolute lymphocyte count (values must be in the normal range), hemoglobin or platelet levels (values must not be below the lower limit of normal), and ALT/SGPT, alkaline phosphatase, AST/SGOT (values must not be above the upper limit of normal), plus virology (HIV/HCV/HBV) (all must be negative), and FSH (if required).

[3] Drug screening (urine drug screen, UDS), including cotinine, should be conducted at Screening, Day -1, Day 13, and at EOS. Pregnancy tests should be conducted in serum at Screening and in urine at all other times (i.e., Days -1 and 13, and EOS). A positive urine pregnancy test will be confirmed with a serum pregnancy test. An alcohol breath test should be conducted at Screening and Day -1.

[4] Participants should be admitted to the clinic following review of final I/E criteria on Day -1 and discharged from the clinic following completion of study specific assessments on Day 8.

[5] Safety laboratory assessments include clinical chemistry and urinalysis. Baseline samples will be collected after clinic admission on Day -1. On Days 3 and 5, the assessment will be pre-am dose. On Day 8, the assessment will be 72 hrs post-am dose on Day 5.

[6] Hematology values include, but are not limited to, RBC, WBC with differential and platelets. PT/PTT and INR assessed only at

Screening.

[7] PBMCs will be collected 2 hrs postdose, relative to the first daily dose on Day 1, and pre-am dose and 2 and 8 hrs postdose relative to the first daily dose on Day 5. Depending on ongoing review of the data, timepoints may be modified or removed, and up to 2 additional samples may be collected. Additionally, PMBC collection may be omitted from some cohorts.

[8] The baseline ECG should be conducted prior to first dosing on Day 1. On Day 1, ECG should be taken predose and 2 hours post-am dose. On Day 5, ECG should be taken predose, 2 hrs, and 72 hrs (Day 8) postdose.

[9] On Day 1, monitor for 12-lead ECG recording will be worn from 2 hrs predose to 12 hrs postdose. Extraction timepoints will be 60, 45, and 30 minutes predose and at 0.5, 1, 1.5, 2, 2.5, 3, 4, 6, 9, and 12 hrs postdose.

[10] On Day 5, monitor for 12-lead ECG recording will be worn from dosing (0 hr) to 25 hrs (Day 6) postdose. Extraction timepoints will be at 0.5, 1, 1.5, 2, 2.5, 3, 4, 6, 9, 12, and 24 hrs postdose.

[11] Vital signs include pulse rate, respiratory rate, blood pressure, and body temperature. On Day 1, VS should be taken pre-am dose, and at 2, 4, and 8 hrs post-am dose. On Day 2 through 4, VS should be taken pre-am dose. On Day 5, VS should be taken predose, and 2, 4, 8, 24 (Day 6), 48 (Day 7), and 72 (Day 8) hrs postdose.

[12] Study drug should be administered BID, 12 hrs apart on Days 1 through 4. On Day 5, study drug should be administered once in the morning only (0 hr). Depending on ongoing review of the safety, tolerability, and PK data, the dosing frequency and the number of days of dosing may be changed; however, the dosing frequency will be no less than once-daily or no greater than three-times daily, and the number of days of dosing will be no fewer than 5 days and no greater than 10 days.

[13] On Day 1, PK samples should be collected pre-am dose and at 0.5, 1, 1.5, 2, 2.5, 3, 4, 6, 9, and 12 hrs post-am dose. On Days 2 through 4, PK samples should be collected pre-am dose. On Day 5, PK samples should be collected predose and at 0.5, 1, 1.5, 2, 2.5, 3, 4, 6, 9, 12, 15, 24 (Day 6), 48 (Day 7), 72 (Day 8), and 192 (Day 13) hrs postdose. Depending on ongoing review of the PK data, timepoints may be modified or removed, and up to 3 additional samples may be added.

[14] On Day 1, urine samples for PK analysis should be collected predose (spot collection), and from 0 to 4, 4 to 8, and 8 to 12 hrs postdose. On Day 5, urine samples for PK analysis should be collected predose (spot collection), and from 0 to 4, 4 to 8, 8 to 12, 12 to 24 (Day 6), 24 to 48 (Day 7), and 48 to 72 hrs (Day 8). Sampling timepoints may be modified, removed, or additional timepoints added depending upon ongoing review of the data. The 12-hour urine collection should occur prior to the second daily dose administration.

[15] Adverse events occurring prior to study drug administration should be recorded as medical history unless the event is directly related to a study specified procedure.

Appendix 2: DMID Toxicity Grading Scale

Clinical Adverse Events			
VITAL SIGNS	**Mild (Grade 1)**	**Moderate Grade 2)**	**Severe (Grade 3)**
Fever (°C) Oral temperature; no recent hot or cold beverages or smoking.	38.0 – 38.4	38.5 – 38.9	>39.0
Tachycardia - beats per minute	101 – 115	116 – 130	> 130 or ventricular dysrhythmias
Bradycardia - beats per minute	50 - 54 or 45 - 50 bpm if baseline <60 bpm	45 - 49 or 40 - 44 if baseline <60 bpm	< 45 or <40 bpm if baseline <60 bpm
Hypertension (systolic) - mm Hg [Assuming supine position, 10 min at rest conditions, not sleeping subjects, measurements on the same arm and several concordant results.]	141 - 150	151 - 160	> 160
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**
Arrythmia		asymptomatic, transient signs, no Rx required	recurrent/persistent; symptomatic Rx required
Hemorrhage, Blood Loss	Estimated blood loss ≤ 100 mL	Estimated blood loss > 100 mL, no transfusion required	Transfusion required
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 60 - 70% (of peak flow)	no normalization with bronchodilator; FEV1 <60% 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 gms/24 hours	4 - 5 loose or watery stools or 400 - 800 gms/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
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
SYSTEMIC	**Mild (Grade 1)**	**Moderate (Grade 2)**	**Severe (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
All Other conditions	**Mild (Grade 1)**	**Moderate (Grade 2)**	**Severe (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

*In addition to grading the measured local reaction at the greatest single diameter, the measurement should be recorded as a continuous variable.

**Induration/Swelling should be evaluated and graded using the functional scale as well as the actual measurement

Laboratory Adverse Events			
Blood, Serum, or Plasma *	**Mild (Grade 1)**	**Moderate (Grade 2)**	**Severe (Grade 3)**
Sodium – Hyponatremia mEq/L	132 – <LLN	130 – 131	<130
Sodium – Hypernatremia mEq/L	>ULN – 148	149 – 150	>150
Potassium – Hyperkalemia mEq/L	>ULN – 5.2	5.3 – 5.4	>5.4
Potassium – Hypokalemia mEq/L	<LLN-3.1	<3.1 – 3.0	<3.0
Glucose – Hypoglycemia mg/dL	65 – 67	55 – 64	<55
Glucose – Hyperglycemia Fasting – mg/dL	>ULN - 120	121 – 130	>130
Glucose – Hyperglycemia Random – mg/dL	140 – 159	160 – 200	>200
Blood Urea Nitrogen mg/dL	23-26	27 – 31	> 31
Creatinine – mg/dL	>ULN – 1.7	1.8 – 2.0	>2.0
Calcium – hypocalcemia mg/dL	8.0 – <LLN	7.5 – 7.9	<7.5
Calcium – hypercalcemia mg/dL	>ULN – 11.0	11.1 – 11.5	>11.5
Magnesium – hypomagnesemia mg/dL	1.3 – 1.5	1.1 – 1.2	<1.1
Phosphorus – hypophosphatemia mg/dL	2.3 – 2.5	2.0 – 2.2	<2.0
CPK – mg/dL	400-1000	1001-1500	>1500
Albumin – Hypoalbuminemia g/dL	2.8 – 3.0	2.5 – 2.7	< 2.5
Total Protein – Hypoproteinemia g/dL	5.2 – 5.4	4.8 – 5.1	< 4.8
Alkaline phosphate – U/L	132-240	241– 360	>360
AST U/L	44 – 105	106-175	>175
ALT U/L	44 – 105	106-175	>175
Bilirubin (serum total) mg/dL	1.3 - 2.0	2.1 - 2.5	> 2.5
Bilirubin – when ALT ≥105 (Hy's law)	1.3 -1.5	1.6 – 2.0	> 2.0
Amylase- U/L	200-270	271-360	>360
Lipase- U/L	176-270	271-360	>360
Hemoglobin (Female) - g/dL	11.0 – 11.5	9.5 – 10.9	< 9.5
Hemoglobin (Male) - g/dL	12.0 – 12.5	10.0 – 11.9	<10.0
WBC Increase - cell/mm3	11,001 – 15,000	15,001 – 20,000	> 20,000
WBC Decrease - cell/mm3	2,500 – 3,500	1,500 – 2,499	< 1500
Lymphocytes Decrease - cell/mm3	750 – 1,000	500 – 749	< 500
Neutrophils Decrease - cell/mm3	1,500 – 2,000	1,000 – 1,499	< 1000
Eosinophils - cell/mm3	500-750	751-1500	> 1500
Platelets Decreased - cell/mm3	120,000 – 130,000	100,000 – 119,999	<100,000
PT – seconds (prothrombin time)	> ULN-14.4	14.5 – 15.7	> 15.7
PTT – seconds (partial thromboplastin time)	>ULN-42.1	42.2– 50.0	> 50.0
Fibrinogen increase - mg/dL	>ULN – 500	501 – 600	> 600
Fibrinogen decrease - mg/dL	<LLN – 140	125 – 139	<125
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	11-50	> 50 and/or gross blood

* The laboratory values provided in the tables serve as guidelines and are dependent upon institutional normal parameters.

* Institutional normal reference ranges should be provided to demonstrate that they are appropriate.

Appendix 3: Contraception Guidance

Male subjects (even with a history of vasectomy) with partners of childbearing potential must use a male barrier method of contraception (i.e., male condom with spermicide) in addition to a second method of acceptable contraception from Check-in until 90 days after the EOS visit. Acceptable methods of contraception for female partners include:

- hormonal injection
- combined oral contraceptive pill or progestin/progestogen-only pill
- combined hormonal patch
- combined hormonal vaginal ring
- surgical method (bilateral tubal ligation or Essure® [hysteroscopic bilateral tubal occlusion])
- hormonal implant
- hormonal or non-hormonal intrauterine device
- OTC sponge with spermicide
- cervical cap with spermicide
- diaphragm with spermicide.

An acceptable second method of contraception for male subjects is vasectomy that has been performed at least 90 days prior to the screening visit, with verbal confirmation of surgical success.

For male subjects (even with a history of vasectomy), sexual intercourse with female partners who are pregnant or breastfeeding should be avoided unless condoms are used from the time of the first dose until 90 days after the EOS visit. Male subjects are required to refrain from donation of sperm from Check-in until 90 days after the EOS visit.

Sexual Abstinence and Same-sex Relationships

Subjects who practice true abstinence, because of the subject's lifestyle choice (i.e., the subject should not become abstinent just for the purpose of study participation), are exempt from contraceptive requirements. Periodic abstinence (eg, calendar, ovulation, symptothermal, postovulation methods) and withdrawal are not acceptable methods of contraception. If a subject who is abstinent at the time of signing the ICF becomes sexually active they must agree to use contraception as described previously.

For subjects who are exclusively in same-sex relationships, contraceptive requirements do not apply. If a subject who is in a same-sex relationship at the time of signing the ICF becomes engaged in a heterosexual relationship, they must agree to use contraception as described previously.

Title of study: A Randomized, Double-blind, Placebo-controlled, First-in-human Study Designed to Evaluate the Safety, Tolerability, and Pharmacokinetics of EIDD-2801 Following Oral Administration to Healthy Volunteers

NCT Number: NCT04392219

Document: Protocol, Version 3.1

Date: 13 April 2020

A Randomized, Double-Blind, Placebo-Controlled, First-in-Human Study Designed to Evaluate the Safety, Tolerability, and Pharmacokinetics of EIDD-2801 Following Oral Administration to Healthy Volunteers

(EIDD-2801-1001-UK)

Protocol Version	3.1 (United Kingdom)
Version Date	13 April 2020

Sponsor Team:

Ridgeback Biotherapeutics	3162 Commodore Plaza, Suite 3E Miami, FL 33133-5815 United States
Medical Officer	[redacted]
EudraCT Number	2020-001407-17

SUMMARY OF CHANGES

Changes from Version 1.1 (United Kingdom) to Version 2.1 (United Kingdom).

The primary reason for this amendment was to include Part 3, a multiple ascending dose study of EIDD-2801 in healthy subjects. Additionally, the following updates were made:

- Results of an *in vivo* micronucleus assay were added.
- The guidelines used to assess the severity of adverse events not presented in the DMID Toxicity Grading Scale were updated, specifically those that were life-threatening or led to death.
- The timepoints for collection of peripheral blood mononuclear cells in Part 2 were modified.
- Minor clarifications were made and typographical errors were corrected.

Changes from Version 2.1 (United Kingdom) to Version 3.1 (United Kingdom).

- Clarified that this study will be conducted at 1 site in the United Kingdom and up to 3 sites in the United States, and that integrated data across all sites will be used for dose-escalation decisions.
- The number of cohorts initially planned for Part 1 (single ascending dose) has been reduced from 4 to 3.
- The number of days of dosing in Part 3 (multiple ascending dose) was decreased from 7 days to 5.5 days. However, the number of days of dosing may be further reduced depending on ongoing review of the safety, tolerability, and pharmacokinetic data.
- Added a rationale for the appropriateness of a 4-day interval between dose escalations.
- Added a dose-escalation stopping criteria for reductions in platelets and lymphocytes.
- Clarified that assessments of human immunodeficiency virus, hepatitis B virus, and hepatitis C virus will only be conducted at Screening.
- Minor clarification and typographical corrections.

STATEMENT OF COMPLIANCE

The study will be carried out in accordance with Good Clinical Practice (GCP) as required by the following:

- Consensus ethical principles derived from international guidelines including the Declaration of Helsinki and Council for International Organizations of Medical Sciences International Ethical Guidelines.
- Applicable International Conference on Harmonisation (ICH) GCP Guidelines.
- United States Code of Federal Regulations (CFR) applicable to clinical studies (45 CFR Part 46; 21 CFR Part 50, 21 CFR Part 56, and 21 CFR Part 312).
- Applicable laws and regulations.

SIGNATURE PAGE

The signature below constitutes the approval of this protocol and the attachments and provides the necessary assurances that this study will be conducted according to all stipulations of the protocol, including all statements regarding confidentiality, and according to local legal and regulatory requirements and applicable regulations and ICH guidelines.

Principal Investigator:

Signed: [redacted] Date: 14 APR 2020

Executive Medical Director
Covance Clinical Research Unit Limited

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ABBREVIATIONS

AE	adverse event
AGP	alpha1-acid glycoprotein
ALT/SGPT	alanine aminotransferase/serum glutamic pyruvic transaminase
AST/SGOT	aspartate aminotransferase/serum glutamic oxaloacetic transaminase
AUC	area under the curve
BID	twice-daily
BMI	body mass index
CDC	Centers for Disease Control and Prevention
CEM	a cell line of lymphoblastic cells derived from a child with leukemia
CFR	Code of Federal Regulations
CHKV	Chikungunya virus
CI	confidence interval
CoV	coronavirus
CRF	case report form
CTP	cytosine triphosphate cytidine triphosphate
CYP	cytochrome P450
DMID	Division of Microbiology and Infectious Diseases
DMSO	dimethylsulfoxide
DPP4	dipeptidyl peptidase 4
DRF	dose range finding
DSS	Drug Safety Services
EBOV	Ebola virus
EC	Ethics Committee
EC_{50}	Half-maximal effective concentration
ECG	electrocardiogram
EEEV	Eastern equine encephalitis virus
EIDD	Emory Institute for Drug Development
EOS	End of Study
FDA	Food and Drug Administration
FE	food-effect
FIH	first-in- human
FSH	follicle stimulating hormone
GCP	Good Clinical Practice
GI	gastrointestinal
GLP	Good Laboratory Practice

HAE	human airway epithelium
HBV	hepatitis b virus
hBTEC	human bronchial/tracheal epithelial cells
HCV	hepatitis c virus
HED	human equivalent dose
HEENT	head, eye, ear, nose and throat
HIV	human immunodeficiency virus
HPMC	hydroxypropyl methylcellulose
IAV	influenza a virus
IB	Investigator's Brochure
IBV	influenza b virus
IC_{50}	half maximal inhibitory concentration
ICF	informed consent form
ICH	International Council for Harmonisation
MAD	multiple ascending dose
MDCK	Madin-Darby Canine Kidney Cells
MedDRA	Medical Dictionary for Regulatory Activities
MERS	Middle East respiratory syndrome
%MN-PCE	percentage of micronuclei-polychromatic erythrocytes
MRSD	maximum recommended starting dose
MTD	maximum tolerated dose
NOAEL	no-observed-adverse-effect-level
NSAID	nonsteroidal anti-inflammatory drug
OTC	over-the-counter
PBMC	peripheral blood mononuclear cells
PBO	placebo
PCE:TE	polychromatic erythrocytes to total erythrocytes
PE	physical examination
PI	principal investigator
[redacted]	[redacted]
PK	pharmacokinetic
QTc(F)	QT interval corrected for heart rate (using Fridericia's formula)
RNA	ribonucleic acid
RSV	respiratory syncytial virus
SAD	single ascending dose
SAE	serious adverse event

SAP	statistical analysis plan
SARS	severe acute respiratory syndrome
SI	selectivity index
SOP	standard operating procedure
UTP	uridine 5'-triphosphate
VEEV	Venezuelan equine encephalitis virus
ZIKV	Zika virus

PROTOCOL SYNOPSIS

Sponsor: Ridgeback Biotherapeutics	
Title: A Randomized, Double-Blind, Placebo-Controlled, First-in-Human Study Designed to Evaluate the Safety, Tolerability, and Pharmacokinetics of EIDD-2801 Following Oral Administration to Healthy Volunteers (EIDD-2801-1001-UK)	
Short Title: EIDD-2801-1001-UK	**Development Phase:** Phase 1
Study Sites: This study will be conducted at 1 site in the United Kingdom and up to 3 sites in the United States.	
Description of Study Drugs, Dose and Mode of Administration: EIDD-2801 and matching placebo (PBO) will be supplied [redacted] **Part 1:** A single oral dose of EIDD-2801 or PBO will be administered to subjects enrolled in Part 1 (P1; single ascending dose [SAD]) cohorts. The starting dose in the first SAD cohort will be 50 mg. The doses will be administered in an escalating manner. Dose levels will not increase by more than 3-fold for predicted nonpharmacologically active dose levels and by 2-fold for predicted pharmacologically active dose levels. [redacted] **Part 2:** Subjects in Part 2 (P2)/food-effect (FE) will receive 2 single doses of EIDD-2801 with a 14-day (minimum) washout period between doses. The dose for the P2 cohort is planned to be 100 mg but may be adjusted based on safety, tolerability, and/or pharmacokinetic (PK) data from P1. In any case, the dose assessed in P2 will have been given previously to subjects in a P1 cohort successfully (i.e., no halting rules were met following dosing). **Part 3:** Multiple doses of EIDD-2801 or PBO will be administered to subjects enrolled in Part 3 (P3; multiple ascending dose [MAD]) cohorts. Subjects will receive twice-daily (BID) doses for 5 days, with a single dose administered on the morning of Day 6 for collection of steady-state PK samples during waking hours. The total daily dose will not exceed a dose shown to be safe and well-tolerated in P1. Doses in P3 may be administered in the fed state, following review of the PK data from P2. Additionally, depending on ongoing review of the safety, tolerability, and PK data, the number of days of dosing may be reduced and the dosing frequency may be changed; the dosing frequency will be no less than once-daily or no greater than three-times daily.	
Treatment Duration: **Part 1:** Subjects enrolled into P1/SAD cohorts will receive a single dose of study drug (EIDD-2801 or matching PBO). **Part 2:** Subjects enrolled into the P2/FE cohort will receive 2 single doses of EIDD-2801, with a washout period between doses. **Part 3:** Subjects in P3/MAD cohorts will receive study drug (EIDD-2801 or matching PBO) for 5 days BID followed by a single dose of EIDD-2801 or PBO on Day 6. Depending on ongoing review of the safety, tolerability, and PK data, the number of days of dosing may be reduced.	

Subject Duration:

Part 1: The maximum possible study duration for participants enrolled in P1/SAD cohorts will be approximately 43 days.

Part 2: The maximum possible study duration for participants enrolled in the P2/FE cohort will be approximately 58 days.

Part 3: The maximum possible study duration for participants enrolled in P3/MAD will be approximately 48 days.

Objectives and Endpoints:

Part 1: SAD Cohorts

Primary:

- Objective: To determine the safety and tolerability of single ascending doses of EIDD-2801.
  - Endpoints:
    - Results of safety evaluations including safety laboratory assessments, physical examination (PE), electrocardiograms (ECGs), vital signs, and adverse events (AEs).

Secondary:

- Objective: To define PK of EIDD-2801 and EIDD-1931 in plasma and urine following single doses administered to healthy volunteers.
  - Endpoints:
    - Plasma PK parameters, including C_{max}, t_{max}, $t_{1/2}$, CL/F, λz, Vz/F, $AUC_{0\text{-}inf}$ and AUC_{last} as appropriate
    - Urinary excretion of EIDD-2801 and EIDD-1931 following single-dose administration.

Part 2: Single-Dose Food-Effect

Primary:

- Objective: To assess the effect of food on the PK of EIDD-2801 and EIDD-1931 following a single oral dose.
  - Endpoints:
    - Plasma PK parameters, including C_{max}, t_{max}, $t_{1/2}$, CL/F, λz, Vz/F, $AUC_{0\text{-}inf}$ and AUC_{last} as appropriate
    - Urinary excretion of EIDD-2801 and EIDD-1931 following single-dose administration.

Secondary:

- Objective: To determine the safety and tolerability of single doses of EIDD-2801.
  - Endpoints:
    - Results of safety evaluations, including safety laboratory assessments, PEs, ECG, vital signs, and AEs.

Part 3: MAD Cohorts

Primary:

- Objective: To determine the safety and tolerability of multiple ascending doses of EIDD-2801.

- Endpoints:
  - Results of safety evaluations, including safety laboratory assessments, PE, ECGs, vital signs, and AEs.

Secondary:

- Objective: To define PK of EIDD-2801 and EIDD-1931 in plasma and urine following multiple doses administered to healthy volunteers.
  - Endpoints:
    - Plasma PK parameters, including C_{trough}, C_{max}, t_{max}, $t_{1/2}$, CL/F, λz, Vz/F, $AUC_{0\text{-}inf}$ (Day 1 dose only), AUC_{τ}, $RA_{AUC\tau}$, and RA_{Cmax} as appropriate
    - Urinary excretion of EIDD-2801 and EIDD-1931 following multiple-dose administration.

Exploratory:

- Objective: To collect data to assess the relationship between EIDD-2801/EIDD-1931 concentrations and QT interval corrected for heart rate (QTc).

Population:

Part 1: P1/SAD cohorts will include 8 subjects each, 2 randomized to receive PBO and 6 randomized to receive EIDD-2801. Initially, 3 SAD cohorts are planned for P1 with the option to add an additional 3 cohorts based on study results.

Part 2: Ten subjects will be enrolled into the P2/FE cohort; all subjects enrolled in the P2 cohort will receive EIDD-2801. One P2 cohort is planned for the study. If PK results obtained are equivocal, additional subjects may be enrolled into the P2 cohort at the previously tested dose or an additional P2 cohort may be added at a different dose.

Part 3: P3/MAD cohorts will include 8 subjects, 2 randomized to receive PBO and 6 randomized to receive EIDD-2801. Initially, 3 MAD cohorts are planned for P3 with the option to add an additional 3 cohorts based on study results. However, the number of cohorts may be reduced if the study objectives are met. Dose levels in P3 may be repeated for collection of peripheral blood mononuclear cells (PBMCs), providing that the dose level did not meet the dose-escalation halting criteria.

The site is strongly encouraged to ensure that women are represented in each cohort. Inclusion and exclusion criteria for study participation are as follows:

Inclusion Criteria: subjects who meet all of the criteria at Screening will be eligible for study participation

1. Male or female between the ages of 18 and 60, inclusive.
2. Female subjects must be of nonchildbearing potential. Nonchildbearing potential is defined as women who:
   - are physiologically incapable of becoming pregnant including those who are surgically sterile, via hysterectomy, bilateral salpingectomy, and/or bilateral oophorectomy by reported medical history and/or medical records. Surgical sterilization to have occurred a minimum of 6 weeks, or at the Principal Investigator's (PI's; or designee's) discretion, prior to Screening.
   - are postmenopausal (at least 45 years of age with amenorrhea for 12 months

without an alternative medical reason with confirmatory follicle stimulating hormone (FSH) levels of ≥ 40 mIU/mL. The amenorrhea should not be induced by a medical condition such as anorexia nervosa, hypothyroid disease or polycystic ovarian disease, or by extreme exercise. It should not be due to concomitant medications that may have induced the amenorrhea such as oral contraceptives, hormones, gonadotropin-releasing hormones, anti-estrogens, or selective estrogen receptor modulators).

3. Male participants must agree to the use of effective contraception for study duration, i.e., from Check-in until 90 days following the last dose of study drug.
4. Is in generally good health as determined by medical history, PE (at Screening and/or Check-in), vital signs, and other screening procedures.
5. Has a body mass index (BMI) of 18 to 30 kg/m^2.
6. Is willing and able to comply with all study procedures including providing informed consent.

Exclusion Criteria: subjects who meet any of the following criteria at Screening (unless otherwise stated) will not be eligible for participation

1. Females who are pregnant, planning to become pregnant, or breastfeeding.
2. Has a clinically relevant acute or chronic medical condition or disease of the cardiovascular, gastrointestinal (GI), hepatic, renal, endocrine, pulmonary, neurologic, or dermatologic systems as determined by the PI (or designee).
3. Has any current or historical disease or disorder of the hematological system or significant liver disease or family history of bleeding/platelet disorders.
4. Has a history of cancer (other than basal cell or squamous cell cancer of the skin), rheumatologic disease or blood dyscrasias.
5. Has a history of GI surgery or other condition that may interfere with absorption of study drug, in the option of the PI (or designee).
6. Has a history of febrile illness within the 14 days prior to the first dose of study drug.
7. Has a positive alcohol or drug screen at Screening or the Day -1 visit or has a history of alcohol or drug abuse within the past 5 years.
8. Currently uses tobacco, nicotine or tobacco products, e-cigarettes or has stopped using tobacco products within the past 3 months and/or has a positive cotinine test at Screening or Check-in.
9. Has any abnormal laboratory value of Grade 2 or higher, which is considered to be clinically significant by the PI (or designee).
10. Has a total white cell count or absolute lymphocyte count outside the normal range, or hemoglobin or platelet levels below the lower limit of normal, at Screening of Day -1.
11. Has values above the upper limit of normal for the following laboratory analytes: alanine aminotransferase (ALT/SGPT), alkaline phosphatase (serum), aspartate aminotransferase (AST/SGOT), at Screening or Day -1.
12. Positive test result for human immunodeficiency virus (HIV), hepatitis B virus (HBV), or hepatitis C virus (HCV).
13. Has an autoimmune disease, is immunosuppressed or is in any way

immunocompromised.

14. Has any of the following:
    - QT interval corrected for heart rate using Fridericia's formula (QTcF) >450 ms confirmed by repeat measurement
    - QRS duration >110 ms confirmed by repeat measurement
    - PR interval >220 ms confirmed by repeat measurement
    - findings which would make QTc measurements difficult or QTc data uninterpretable
    - history of additional risk factors for torsades de pointes (e.g., heart failure, hypokalemia, family history of long QT syndrome).
15. Except as noted, has used prescription drugs (other than hormone replacement therapy) within 14 days prior to the first dose of study drug unless, in the opinion of the PI (or designee), the drug will not interfere with study assessments. The following prescription drugs will be allowed:
    - Statins: Subjects who have been on a stable dose of statins for a minimum of 6 months and who have normal creatine kinase at Screening.
    - Antihypertensives: Subjects who have been on a stable dose of antihypertensives for a minimum of 6 months and have controlled blood pressure and no active heart disease.
16. Has received an experimental agent (vaccine, drug, biologic, device, blood product or medication) within 3 months prior to the first dose of study drug and agrees not to receive another experimental agent during the duration of this trial.
17. Has donated blood within 30 days, plasma within 2 weeks, or platelets within 6 weeks prior to Screening or refuses to agree to not donate blood until 3 months after the End of Study (EOS) visit.
18. Uses over-the-counter (OTC) medications or nutritional supplements (including, e.g., gingko biloba, aspirin at any dose, and nonsteroidal anti-inflammatory drugs [NSAIDs]) on a routine/scheduled basis, and/or is unwilling to abstain from such use for the duration of the study.
19. Is unwilling to abstain from quinine containing products including quinine water, tonic water, bitter lemon, jello shots, any quinine preparations or Cinchona tree bark preparations (e.g., herbal medications containing Cinchona tree bark).
20. Has any condition that would, in the opinion of the PI (or designee), put the subject at increased risk for participation in a clinical trial or confinement in a clinical research unit.

Retesting for Inclusion/Exclusion Criteria: In the case where a parameter is outside of the acceptable limits for enrollment, measurement of the parameter can be repeated one time if both the following are met:

- In the opinion of the PI (or designee), the parameter value does not represent a chronic condition that otherwise will preclude the volunteer from enrolling in the study; and
- In the opinion of the PI (or designee), the abnormality is not likely to recur.

General Investigational Plan: For all potential subjects, volunteers who express interest in the study will report to the clinic for informed consent. The study will be explained to the subject and the Ethics Committee (EC)-approved informed consent form (ICF) will be presented. The subject will be given the chance to review the document and ask any questions he/she may have. If, after reviewing the consent, the subject would like to participate in the study, then he/she will sign the ICF and begin screening for study entry; those satisfying all criteria will be enrolled into the study and admitted to the clinic on Day -1. Retesting will be allowed as described above.

Part 1: For P1/SAD, the first cohort will be divided into 2 groups. The first group, a sentinel group, will include 2 subjects.

On Day 1, one subject in the sentinel group will be administered PBO and one will be administered EIDD-2801. Analysis of the safety results for the sentinel group will be reviewed 24 hours postdose. If no halting criteria have been met, the remainder of the subjects in Cohort 1 (the second group) will be dosed. There will be no sentinel groups for the remaining cohorts.

Following dosing on Day 1, subjects will remain in the clinic through completion of study assessments on Day 4, returning to the clinic on Day 9 for study assessments, and Day 15 for the EOS visit.

Doses will be administered in an escalating manner following satisfactory review by the Sponsor and PI of the blinded safety and tolerability data (up to 72 hours post final dose). Integrated data from all applicable sites will be used to make dose-escalation decisions. Dose-escalation will only occur if data from a minimum of 6 subjects (across all sites) have been reviewed from the previous lower dose group, such that data from a minimum of 4 subjects (across all sites) who have received EIDD-2801 will be used to make the dose-escalation decision. Dose levels will not increase by more than 3-fold for predicted nonpharmacologically active dose levels and by 2-fold for predicted pharmacologically active dose levels. The highest dose to be studied under this protocol will not exceed 200 mg.

Part 2: For P2/FE, one cohort assessing the effect of food on EIDD-2801 and EIDD-1931 PK parameters will be enrolled; it is planned that the P2 cohort will be at a dose of 100 mg EIDD-2801, although higher or lower doses may be selected based on safety and available PK data from P1.

In addition to assessing the effect of food on dosing, PBMCs will be collected from subjects following each dose; the PBMCs will be stored for analysis of EIDD-2061 levels at some point in the future.

The 10 subjects enrolled in the FE cohort will all receive EIDD-2801; subjects will be randomized to a treatment sequence (i.e., to receive drug in the fed then fasted state [Sequence 1] versus fasted then fed state [Sequence 2]). Half of the subjects in the cohort will be randomized to Sequence 1 and the other half will be randomized to Sequence 2.

Following dosing on Day 1, subjects will remain in the clinic through completion of study assessments on Day 4, returning to the clinic on Day 9 for study assessments, and Day 14 to begin the second dosing sequence on Day 15. Subjects will then be discharged from the clinic on Day 18, returning to the clinic on Day 23 for study assessments, and Day 30 for the EOS visit. There will be a 14-day (minimum) washout period between doses.

Part 3: For P3/MAD, subjects will receive BID doses on Day 1 through Day 5, inclusive, and will receive the final dose of study drug on the morning of Day 6 for collection of steady-state PK samples during waking hours. Subjects will remain domiciled at the site during the dosing

period and until Day 9, returning to the clinic on Day 14 for completion of study assessments, and Day 20 for the EOS visit.

Doses in P3 may be administered in the fed state, following review of the PK data from P2. Additionally, depending on ongoing review of the safety, tolerability, and PK data, the number of days of dosing may be reduced and the dosing frequency changed; the dosing frequency will be no less than once-daily or no greater than three-times daily. If the number of days of dosing are reduced, the timepoints for subsequent assessments and the period for which subjects are domiciled at the clinic will similarly be changed (subjects will be domiciled for 3 days post final dose).

Part 3 may run in parallel with P1, providing that the total daily dose to be administered does not exceed a dose already shown to be safe and well-tolerated in P1.

Peripheral blood mononuclear cells may be collected from subjects in P3, depending upon ongoing review of the data. The collection of these samples may be omitted from some cohorts, depending on ongoing review of the data. The PBMCs will be stored for analysis of EIDD-2061 levels at some point in the future. In addition, continuous 12-lead ECG monitoring will be performed to collect data to assess the relationship between study drug concentrations and QTc interval. All data will be archived without extraction or analysis and will not be reported in the scope of this study.

Additional parts may be added to this study, including a SAD part in elderly healthy subjects and a study of the safety, tolerability, and PK in Japanese subjects. These additional parts are not described in this protocol but may be added via a protocol amendment.

Safety Monitoring and Potential Unblinding: Safety for this study will be continually monitored by the PI and Sponsor.

Blinding: All study personnel will remain blinded to treatment assignment (i.e., EIDD-2801 or PBO) in P1 and P3, except for personnel at the bioanalytical laboratory, and the unblinded pharmacy staff and pharmacokineticist. If unblinding is required to manage subject safety or to support dose-escalation decisions, the decision to unblind lies solely with the PI. If possible and providing that it does not interfere with or delay any decision in the best interest of the subject, the PI will discuss the intended code-break with the Sponsor.

Dose-Escalation Halting Rules:

For Group 1 in P1/SAD, sentinel dosing will occur whereby 2 subjects (1 active and 1 PBO) will be dosed on 1 day and, providing no safety concerns arise, the remaining 6 subjects will be dosed after at least 24 hours.

The study will be halted if one or more subjects experience a serious adverse event (SAE) that is considered to be related to EIDD-2801 or 2 or more subjects in the same group experience severe AEs that are considered to be related to EIDD-2801.

Additionally, if 2 or more subjects experience a reduction of platelets greater than 50% over baseline levels or a reduction in lymphocytes (absolute count) to Grade 3/severe levels without an alternate cause, the PI and Sponsor will review the data. If, after review of unblinded data, it is determined that these events are probably related to study drug, no further dose escalations will occur.

Dose-escalation will stop if clinically relevant signs or symptoms of similar nature occur in 2 or

more subjects in a group that, in the opinion of the PI, warrant stopping of dose-escalation.

Data Monitoring, Safety Reporting and Unblinding: Data will be monitored throughout the course of the study by experienced clinical monitors according to the clinical monitoring plan. As data will be entered into an electronic case report form (CRF), data will be monitored to ensure accuracy as well as adherence to data entry instructions provided in the CRF guidelines. Procedures for reporting any SAE will be detailed in the protocol and forms and instructions provided to the site.

Statistical Considerations: A complete description of all statistical analyses and methods will be presented in the statistical analysis plan (SAP). The SAP will be reviewed and approved by the Sponsor and will be finalized prior to database lock. Plans for PK analyses will be included in the SAP.

Determination of Sample Size: The sample sizes for the P1 and P3 cohorts are typical for a Phase 1 first in human (FIH) study. The sample size for the P2 cohort is in accordance with Food and Drug Administration (FDA) guidelines for sample size in FE studies.

Study Populations:

Safety Population: All subjects who receive at least one dose of study drug.

Pharmacokinetic Population: All subjects receiving study drug who comply sufficiently with protocol requirements (i.e., no major protocol deviations) and for whom a sufficient number of analyzable PK samples have been obtained to permit the determination of the PK profile for EIDD-2801/EIDD-1931 and the statistical analysis of the results. Subjects who experience an AE of vomiting before 2× the median t_{max} of the group may be excluded from the PK population.

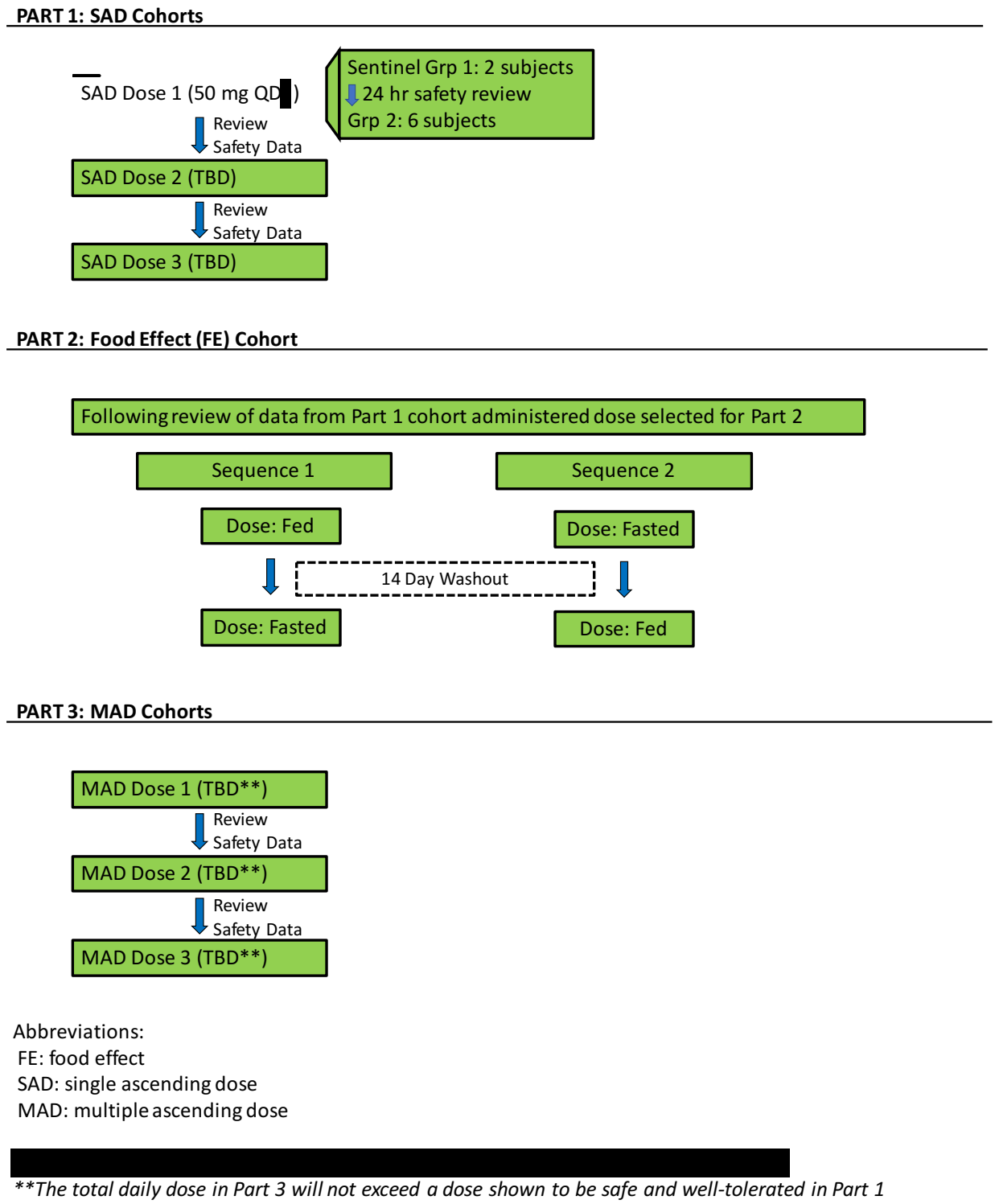
Safety Analyses: Statistical methods for the safety analyses will be descriptive in nature. Safety data, including AEs, clinical laboratory data, vital signs, ECG parameters, and PEs. All appropriate AEs will be graded using the Division of Microbiology and Infectious Diseases (DMID) toxicity scale (March 2014). Change from baseline will be included in summary tables for laboratory parameters. All laboratory data will be included in the data listings and all test values outside the normal range will be flagged.

Pharmacokinetic Analyses: Plasma PK parameters for each dose level will be calculated from the concentrations of EIDD-2801 and EIDD-1931 measured in predose and postdose plasma samples. For each dose level, descriptive statistics will be presented. Figures will be created to display mean and individual subject EIDD-2801 and EIDD-1931 concentration versus time. Urine PK parameters will be calculated whenever possible for each subject based on the urine concentrations of EIDD-2801 and EIDD-1931. The following PK parameters will be calculated:

Plasma PK Parameter	Description
AUC_{last}	The area under the plasma concentration-time curve, from time 0 to the last measurable non-zero concentration, as calculated by the linear up/log down trapezoidal method (P1 and P2 only).
$AUC_{0\text{-}inf}$	The area under the plasma concentration-time curve from time 0 extrapolated to infinity. $AUC_{0\text{-}inf}$ is calculated as the sum of AUC_{last} plus the ratio of the last measurable plasma concentration to the elimination rate constant (λz) (P1, P2, and Day 1 dose of P3).
C_{max}	Maximum observed concentration.
t_{max}	Time to reach C_{max}. If the maximum value occurs at more than one time point, t_{max} is defined as the first time point with this value.

λz	Apparent terminal elimination rate constant; represents the fraction of medication eliminated per unit time.
$t_{1/2}$	Apparent terminal elimination half-life of medication, calculated as $0.693/\lambda z$.
Vz/F	Apparent volume of distribution (EIDD-2801 only).
CL/F	Apparent oral drug clearance (EIDD-2801 only).
C_{trough}	Trough concentration (P3 only).
AUC_{τ}	The area under the plasma concentration-time curve during a dosing interval (P3 only).
$RA_{AUC\tau}$	Observed accumulation ratio based on AUC_{τ} (P3 only).
RA_{Cmax}	Observed accumulation ratio based on C_{max} (P3 only).
Urine PK Parameter	**Description**
A_e	Amount excreted in urine over the sampling interval.
F_e	Fraction of drug excreted in the urine over sampling interval.
CL_R	Renal clearance, $CL_R = A_e/AUC$ where both A_e and AUC are determined over co-incident time ranges after dosing.
In P1, dose-proportionality for PK parameters of EIDD-2801 and EIDD-1931 ($AUC_{0\text{-}inf}$, AUC_{last}, and C_{max}) will be assessed by the power model. The slope and the associated 90% confidence interval (CI) based on the power model will be reported. In P3 on Day 6, dose-proportionality for PK parameters of EIDD-2801 and EIDD-1931 (AUC_{τ} and C_{max}) will be assessed by the power model. The slope and associated 90% CI based on the power model will be reported. To assess the effect of food on the PK of EIDD-2801 and EIDD-1931, natural log-transformed PK parameters from P2 ($AUC_{0\text{-}inf}$, AUC_{last}, and C_{max}) will be analyzed using a mixed-effects model with fixed effect terms for treatment (with or without food), period, and sequence and with subject within sequence as a random effect. Point estimates and their associated 90% CIs for the natural log-transformed PK parameters for the difference between EIDD-2801 administered under fed relative to fasted conditions will be constructed. The point estimates and interval estimates will be back transformed to give estimates of the ratio and 90% CI for the geometric least squares mean ratio for fed/fasted to assess the effect of food.	
Interim Analyses: No formal interim analyses are planned for this study. Data from P1 cohorts may be locked, unblinded, and analyzed prior to conducting P2 and P3.	

Figure 1: Study Schema



Abbreviations:
FE: food effect
SAD: single ascending dose
MAD: multiple ascending dose

[redacted]

***The total daily dose in Part 3 will not exceed a dose shown to be safe and well-tolerated in Part 1*

1. INTRODUCTION

This study is being conducted at sites across the United Kingdom and the United States. Country-specific protocol amendments have been prepared; with the exception of regional regulatory requirements, the study is consistent across countries and data will be reported in a single clinical study report.

This study is a FIH study designed to assess the safety, tolerability and PK of EIDD-2801 in healthy human volunteers. EIDD-2801 is a ribonucleoside analog with broad-spectrum activity against many RNA viruses. It is currently being developed by Ridgeback Biotherapeutics as a treatment of infections caused by highly pathogenic coronaviruses (CoV), including COVID-19. In addition, EIDD-2801 is being developed in parallel as a treatment of uncomplicated influenza caused by all subtypes of circulating and emerging (drifted and shifted) influenza A virus (IAV) and influenza B virus (IBV), including seasonal, epidemic and pandemic strains.

1.1. Background

EIDD-2801 is the 5'-isopropyl ester prodrug of the broadly active, direct-acting antiviral ribonucleoside analog EIDD-1931. After oral delivery, the prodrug (EIDD-2801) is rapidly hydrolyzed by circulating esterases to produce high circulating (plasma) levels of EIDD-1931. In cell culture systems, EIDD-1931 has been shown to inhibit replication of multiple viral pathogens from multiple RNA virus families including pathogenic CoV (e.g., Middle East respiratory syndrome [MERS], severe acute respiratory syndrome [SARS]-CoV and SARS-CoV-2), influenza viruses (seasonal, pandemic, and avian subtypes), respiratory syncytial virus (RSV), alphaviruses (e.g., Eastern equine encephalitis virus [EEEV], Venezuelan equine encephalitis virus [VEEV], and Chikungunya virus [CHKV]), Filoviruses (e.g., Ebola virus [EBOV]), and Zika virus (ZIKV). In addition, EIDD-2801 is active against orthopoxviruses (tested against vaccinia virus) probably because orthopoxviruses encode their own unique RNA polymerase. [redacted] The primary mechanism of action of EIDD-2801 is inhibition of viral RNA replication by incorporation of the EIDD-1931 monophosphate metabolite into the viral RNA genome resulting in induction of viral error catastrophe.

1.2. Rationale for Development

EIDD-2801 is being developed for the treatment of infections caused by RNA viruses, specifically for COVID-19 and other CoV infections, influenza, and VEEV. During conduct of the FIH study, the Sponsor intends to define a dose that may be active in treating COVID-19 in patient studies.

EIDD-2801 has a unique dual mechanism of action against RNA viruses, including SARS-CoV-2 and other CoV infections. The compound acts as a competitive, alternative substrate for the virally encoded RNA-dependent RNA-polymerase that upon incorporation into nascent chain RNA induces increased mutational frequency in the viral genome. Incorporation quickly results in the production of non-viable virus. Additionally, the active metabolite,

EIDD-1931-5'-triphosphate (EIDD-2061), may act directly as a chain terminator and arrest replication by exerting a next nucleoside effect. It is anticipated that the high barrier to resistance observed during *in vitro* passaging studies will translate to slow, if any, emergence of viral resistance. Resilience to viral escape is a distinguishing feature of EIDD-2801.

Currently, there is no approved antiviral therapeutic for the treatment of COVID-19. An antiviral drug is urgently needed. [redacted]

1.3. Nonclinical Overview

1.3.1. Mechanism of Action

The mechanism of antiviral activity of EIDD-2801 is "lethal mutagenesis"; a concept that is predicated on increasing the viral mutation rate beyond a biologically-tolerable threshold, resulting in impairment of viral fitness and leading to viral extinction.

The specifics of the mechanism are as follows. EIDD-2801 is rapidly taken up by cells and the 5'-isopropylester cleaved to liberate EIDD-1931, which is in turn phosphorylated to EIDD-2061 by host kinases (Hernandez-Santiago et al., 2004; Painter et al., 2019). The 5'-triphosphate, EIDD-2061, acts as a competitive alternative substrate for virally encoded RNA-directed RNA polymerases and EIDD-2061 is incorporated into nascent viral RNA. Owing to the ability of the N^4-hydroxycytosine base of EIDD-1931 to tautomerize, EIDD-2061 can pair with either guanosine or adenosine, and consequently can substitute for either CTP or UTP, respectively (Flavell et al., 1974). This results in an accumulation of mutations that increases with each cycle of viral replication. The process whereby the mutation rate is increased by exposure to a drug is referred to as Viral Decay Acceleration (Mullins et al., 2011) and results in viral ablation.

Significant work has gone into validating this mechanism of action for EIDD-2801/1931, and it has been shown for MERS-CoV, VEEV, and IAV that viruses grown in the presence of EIDD-1931 have significantly increased levels of transition mutations (Agostini et al., 2019; Toots et al., 2019; Urakova et al., 2018). Multi-log decreases in virus yields were observed after treatment with EIDD-1931. Additionally, it was demonstrated for VEEV that the infectivity of virions formed in the presence of EIDD-1931 decreases from ~20% to <0.2%, and that the infectious virions are significantly Impaired in their replication ability (Urakova et al., 2018). As a consequence of this mechanism of action, the generation of drug-resistant escape mutants is practically impossible. This same effect was demonstrated for CoV (Agostini et al., 2019) and influenza virus (Toots et al., 2019). Furthermore, given the unique mechanism of action, EIDD-2801 is expected to be active against viruses resistant to other antiviral agents which have a different mechanism of action. The only data generated to date regarding the activity of EIDD-1931 against viruses resistant to other nucleoside analogs found that EIDD-1931 was

active against CoV resistant to remdesivir in cell culture assays (T. Sheahan et al, preprint available at https://www.biorxiv.org/content/10.1101/2020.03.19.997890v1).

As an alternative or additional mechanism of action, it has been theorized that incorporation of EIDD-2061 into viral genomic RNA can change the thermodynamics of RNA secondary structure and thus decrease the efficiency of the promoter regions involved in RNA genome replication (Stuyver et al., 2003).

1.3.2. *In Vitro* Pharmacology

1.3.2.1. Antiviral Activity in Tissue Culture and in Human Airway Epithelium

The ribonucleoside analog EIDD-1931 is the parent of the prodrug EIDD-2801. EIDD-1931 shows specific antiviral activity in different tissue culture cells and in the differentiated organoid model of human airway epithelium (HAE) with a selectivity index (SI) ranging from 21 to >100 for all influenza viral isolates tested. It is active against IAV (pandemic and seasonal) and IBV strains, as well as against highly pathogenic H5N1 and H7N9 strains (Table 1).

Table 1: EIDD-1931 Antiviral Activity Against Influenza A and B Viruses in Tissue Culture and Primary Human Bronchial/Tracheal Epithelial Cells

Virus	Strain	Cell line	EC_{50}* (µM)	CC_{50} (µM)	SI	Reference
IAV H1N1	Ca/07/2009	MDCK	1.24	68	55	NIAID Antiviral Testing Program
IAV H1N1	WSN/33	MDCK	1.1	299.8	275	Yoon et al., 2018
IAV H1N2	WSN/33	primary hBTEC	5.4	-	-	Yoon et al., 2018
IAV H2N3	Perth/16/2009	MDCK	0.88	52	59	NIAID Antiviral Testing Program
IAV H2N3	Ohio/sw-10-132/2010	MDCK	3.2	299.8	94	Yoon et al., 2018
IAV H5N1	Duck/MN/1525/81	MDCK	1.28	27	21	NIAID Antiviral Testing Program
IAV H5N1	Vietnam/1203/2004	MDCK	0.14	299.8	2100	Yoon et al., 2018
IAV H7N9	Anhui/1/2013	MDCK	0.13	299.8	2300	Yoon et al., 2018
IBV	Florida/4/2006	MDCK	<0.4	76	>190	NIAID Antiviral Testing Program
IBV	Brisbane/60/08	MDCK	0.006	299.8	50000	Yoon et al., 2018
IAV H1N1	Ca/07/2009	HAE-3D*	0.08	50	625	Toots et al., 2019
IAV H1N1	WSN/33	HAE-3D*	0.08	50	625	Toots et al., 2019
IBV	Brisbane/60/08	HAE-3D*	0.06	50	833	Toots et al., 2019

* Human Airway Epithelium organoid model.

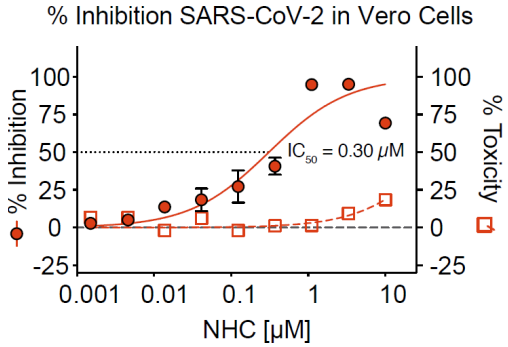
EIDD-1931 also showed specific antiviral activity against human SARS-CoV, MERS-CoV (Table 2) and SARS-CoV-2 (Figure 2), against togaviruses VEEV, EEEV and CHKV (Table 3) [redacted]

Table 2: EIDD-1931 Antiviral Activity Against SARS and MERS Coronaviruses in Tissue Culture and Primary Human Bronchial/Tracheal Epithelial Cells

Virus	Strain	Cell line	EC_{50} (µM)	CC_{50} (µM)	SI	Reference
SARS-CoV-1	Urbani	Vero76	<0.4	144	>360	NIAID Antiviral Testing Program
SARS-CoV-1	SARS-CoV-GFP(†)	HAE-3D(*)	<1	>100	>100	Tech. Report 25.038
MERS-CoV	GenBank Ac.No JX869059**	DBT-9	0.56	>200	>357	Agostini et al., 2019
MERS-CoV	Human ß-CoV C, Novel 2912	Vero E6	<0.8	20	>25	NIAID Antiviral Testing Program

* Human Airway Epithelium organoid model; ** cDNA Derived clone

Figure 2: Inhibition of SARS-CoV-2 by EIDD-1931



EIDD-1931 (NHC) antiviral activity (closed circles) and cytotoxicity (open squares) in Vero Cells infected with SARS-CoV-2. Vero cells were infected in duplicate with SARS-CoV-2 clinical isolate virus at a multiplicity of infection (MOI) of 0.05 in the presence of a dose response of EIDD-2801 for 48 hours after which replication was measured through quantitation of cell viability by Cell-Titer-Glo assay. Cytotoxicity was measured in similarly treated but uninfected cultures. Reproduced from Sheahan et al 2020.

Table 3: EIDD-1931 Antiviral Activity Against Togaviruses in Tissue Culture

Virus	Strain	Cell line	EC_{50} (µM)	CC_{50} (µM)	SI	Reference
VEEV	TC-83	Vero	0.43	>200	>930	Urakova et al., 2018
VEEV	TC-83	Vero76	1.92	32	17	NIAID Antiviral Testing Program
EEEV	FL93-939	Vero76	1.08	84	78	NIAID Antiviral Testing Program
CHKV	S27 (VR-64)	Vero76	1.8	96	53	NIAID Antiviral Testing Program

[redacted]

1.3.2.2. Cytotoxicity of EIDD-1931 in Tissue Culture Utilizing Cells from Different Organs and Species

EIDD-1931 was tested for cytotoxicity in human hepatic origin Huh7 and HepG2 cells, in human lymphoid CEM, human pancreatic BxPC-3, human prostate cancer PC-3, human muscle A204, human lung A549, human epithelial hEp-2, rat heart muscle H9c2, monkey kidney Vero, and canine kidney MDCK cell lines (Table 5). The compound exhibits low cytotoxicity in the majority of cells tested (half-maximal effective concentration [EC_{50}] values are in the range of 40 to >100 µM) except in lymphoid origin CEM cells where the compound shows a 7.5 µM EC_{50} value (Sticher et al., 2020; Urakova et al., 2018; Yoon et al., 2018).

Table 5: Cytotoxicity (CC50) of EIDD-1931 in Mammalian Cell Lines

Cell Line	CEM	HepG2	PC-3	A204	A549	BxPC-3	Huh-7	H9c-2	Vero	hEp-2	MDCK
CC50 (µM)	7.5	42.3	267.1	84	46	48	165.5	81	53	272.4	299.8

Sources: Sticher et al., 2020, Yoon et al., 2018

EIDD-2801 typically showed 2-4× lower activity and cytotoxicity than EIDD-1931 due to slightly slower uptake and anabolism in tissue culture.

1.3.2.3. Assessment of Mitochondrial Toxicity

Since EIDD-1931 is a nucleoside analog, additional investigations were performed to analyze whether observed cytotoxicity of EIDD-1931 is caused by mitochondrial toxicity. It was demonstrated that the prolonged treatment (14 days) with the compound does not result in selective killing of mitochondria or in mitochondrial dysfunction in CEM and HepG2 cells (Sticher et al., 2020).

1.3.3. *In Vivo* Pharmacology

The prodrug EIDD-2801 or its parent EIDD-1931 have been tested in animal models of RNA viral infection. An overview of results from the animal studies in indications to be pursued are described below. Additional detail is provided in the Investigator's Brochure (IB).

1.3.3.1. Coronavirus: SARS-CoV and MERS-CoV

In mouse models of SARS and MERS infection and disease, coronaviral disease was assessed by changes in body weight, measured daily, and lung hemorrhage, assessed in the large left lobe of the lung following sacrifice on Day 5 (SARS-CoV) or Day 6 (MERS-CoV). To assess production of infectious virions, virus was isolated from the lower left lobe of the lung following sacrifice on Day 5 (SARS-CoV) or Day 6 (MERS-CoV) and quantified using a plaque assay.

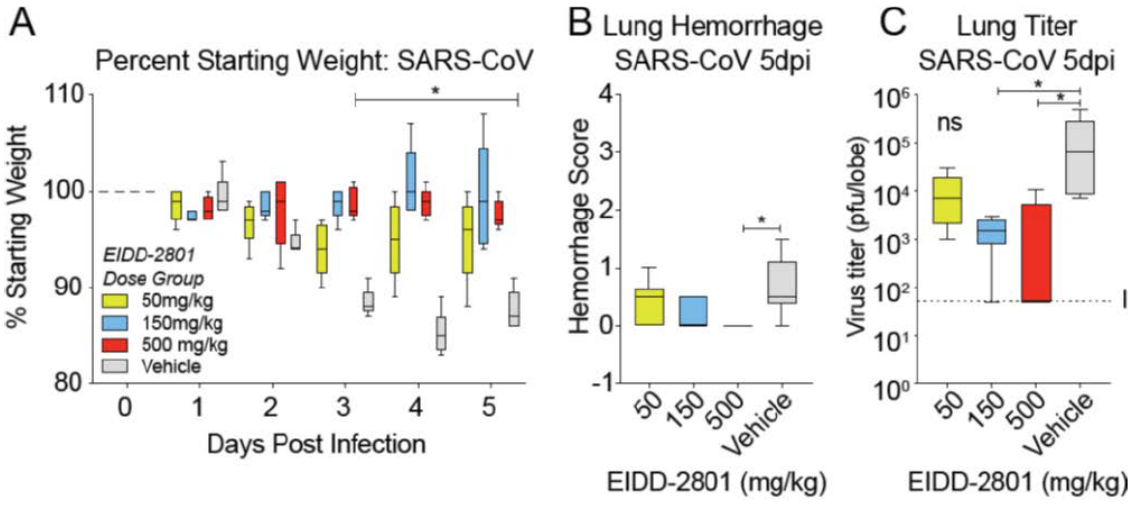
The results demonstrated that, in mice infected with either SARS- or MERS-CoV, both prophylactic and therapeutic treatment with EIDD-2801 resulted in a reduction in virus replication, improvements in pulmonary function, and improvements in maintaining body weight

(i.e., reduced body weight loss). While EIDD-2801 doses of 50, 150 and 500 mg/kg BID were assessed in the CoV mouse experiments, 500 mg/kg BID yielded the most consistent therapeutic effect.

A prophylactic, dose-escalation study was conducted in C57BL/6 mice infected with mouse-adapted SARS-CoV (SARS-MA15). Prophylactic oral treatment with EIDD-2801 was initiated 2 hours before intranasal infection and continued every 12 hours thereafter through the end of the study (Day 5; Figure 3).

In mice treated BID with EIDD-2801, body weight loss observed with vehicle treatment was diminished in the 50 mg/kg treatment group, beginning on Day 3 post-infection. No body weight loss was seen in the 150 and 500 mg/kg treatment groups (Figure 3, Panel A). Lung hemorrhage was also significantly reduced on Day 5 post-infection, following treatment with 500 mg/kg EIDD-2801 (Figure 3, Panel B). When compared to vehicle control, a dose-dependent reduction in SARS-CoV lung titers at Day 5 was seen across all 3 treatment groups (Figure 3, Panel C) with significant differences among the vehicle, 150 mg/kg and 500 mg/kg groups. Thus, prophylactic treatment with EIDD-2801 resulted in a robust antiviral effect that was able to prevent SARS-CoV replication and disease.

Figure 3: EIDD-2801 Prophylactic Treatment of SARS-CoV Infected Mice

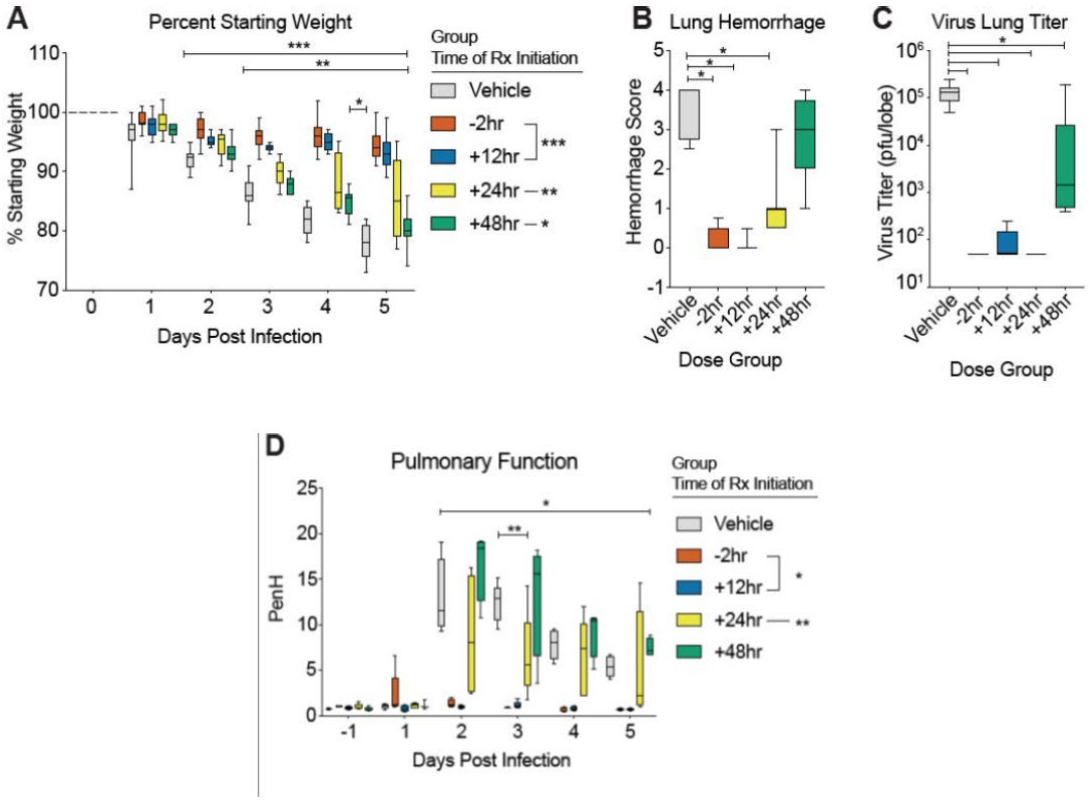


Effect of EIDD-2801 treatment on body weight (A), lung hemorrhage (B) and virus lung titer (C) in mice infected with SARS-MA15. Treatment was initiated two hours prior to infection and continued every 12 hours (BID) to Day 5.

The antiviral activity of EIDD-2801 against SARS-CoV was compared when treatment was initiated at -2 hours (pre-infection) and 12, 24, or 48 hours post-infection. After initiation of treatment, all groups were dosed every 12 hours for the duration of the study (Figure 4). For SARS-challenged mice, initiating treatment at 12 hours post-infection significantly prevented body weight loss beginning on Day 2, a result similar to that seen when dosing prophylactically

(i.e., beginning at 2 hours pre-infection). Initiation of treatment with EIDD-2801 at 24 hours post-infection also significantly reduced body weight loss on Days 3 through 5 post-infection. When EIDD-2801 treatment was initiated at 48 hours post-infection, body weight loss was only statistically different from vehicle on Day 4 post-infection (Figure 4, Panel A). Significant reductions in lung hemorrhage were seen when EIDD-2801 treatment was initiated before (-2 hours) and up to 24 hours after infection; a result that mirrored body weight loss data (Figure 4, Panel B). All mice treated with EIDD-2801 had significantly reduced viral loads in the lungs, even in the group where treatment was initiated 48-hour post-infection (Figure 4, Panel C). Pulmonary function, measured via whole body plethysmography, was assessed using the PenH metric which is a surrogate marker for bronchoconstriction or pulmonary obstruction. The administration of EIDD-2801 prior to infection (-2 hours) and at 12 hour post-infection completely abrogated the loss of pulmonary function observed in vehicle-treated animals (Figure 4, Panel D). Improved pulmonary function was also seen in the group where treatment was initiated 24 hours after virus challenge.

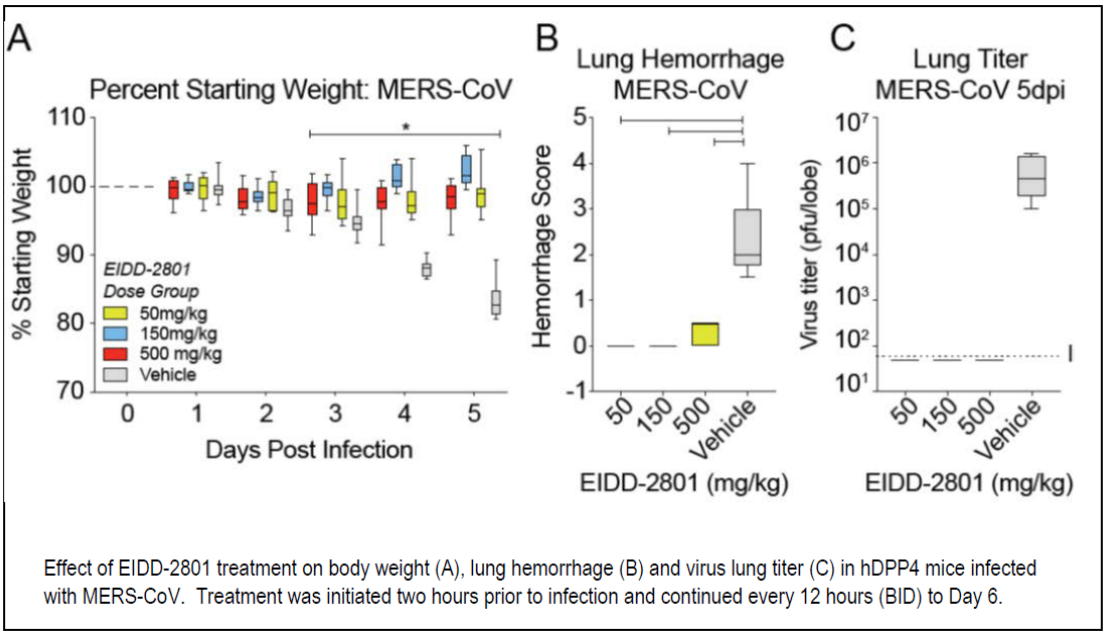
Figure 4: EIDD-2801 Treatment of SARS-CoV Infected Mice



EIDD-2801 was also tested to determine if it is active *in vivo* against MERS-CoV as described by Sheahan et. al (draft manuscript). The murine ortholog of the MERS-CoV receptor, dipeptidyl peptidase 4 (DPP4), does not support viral binding and entry. Thus, all *in vivo* studies described below were performed in genetically modified hDPP4 mice permissive for MERS infection. Prophylactic treatment starting at 2 hours before viral challenge with either 50, 150, or

500 mg/kg EIDD-2801 prevented body weight loss on Days 2 through 6 post-infection (Figure 5, Panel A), prevented lung hemorrhage measured on Day 6 (Figure 5, Panel B), and reduced virus lung titer on Day 6 to the limit of detection (Figure 5, Panel C).

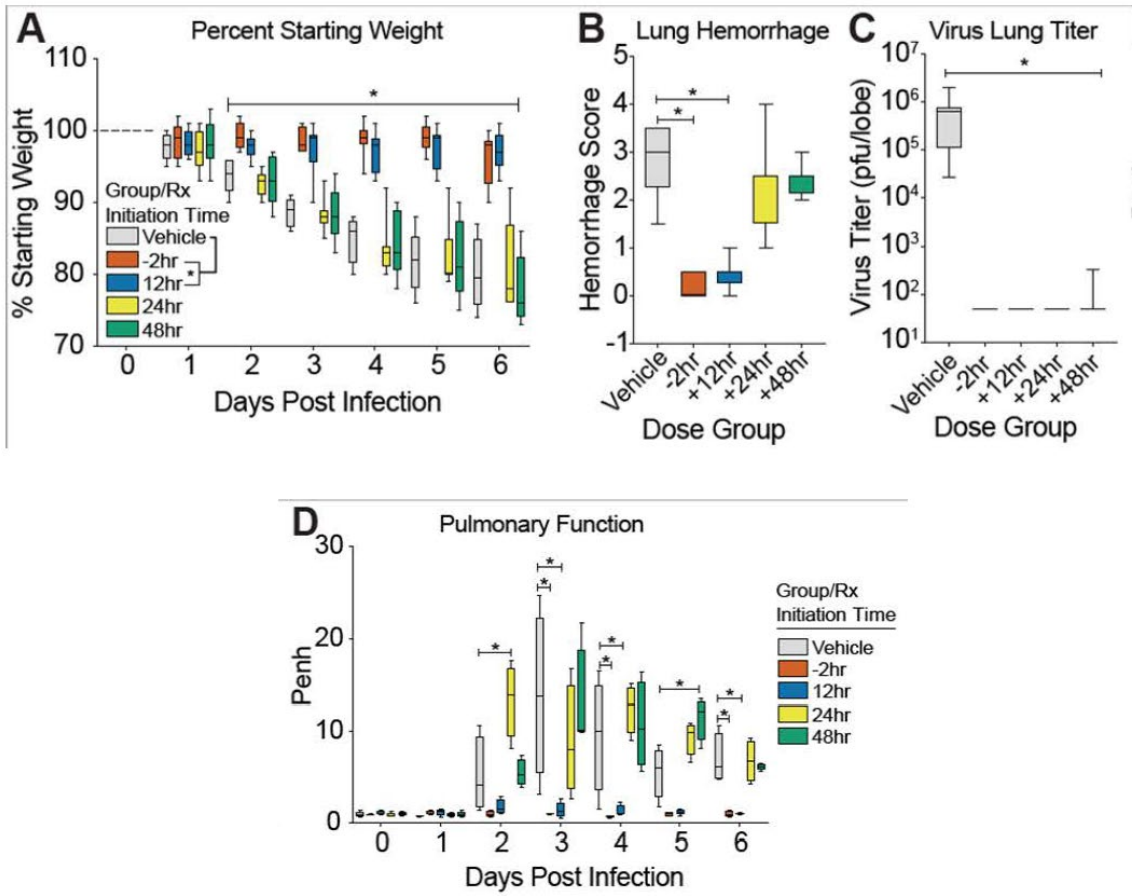
Figure 5: EIDD-2801 Prophylactic Treatment of MERS-CoV Infected Mice



Effect of EIDD-2801 treatment on body weight (A), lung hemorrhage (B) and virus lung titer (C) in hDPP4 mice infected with MERS-CoV. Treatment was initiated two hours prior to infection and continued every 12 hours (BID) to Day 6.

The effect of EIDD-2801 treatment on MERS-CoV infected mice is shown in Figure 6. When EIDD-2801 treatment was initiated 12 hours post-infection, there was no loss in body weight from Days 2 through 6 post-infection (Figure 6, Panel A) and no evidence of lung hemorrhage on Day 6 post-infection (Figure 6, Panel B). However, protection was not observed in groups where treatment was initiated either 24- or 48-hours post-infection. Conversely, virus lung titer on Day 6 post-infection was significantly reduced to the limit of detection in all treatment groups, regardless of the time treatment began (Figure 6, Panel C). To gauge the effect of the timing of EIDD-2801 treatment initiation on physiologic measures of lung disease, pulmonary function was assessed by whole body plethysmography. As expected, a loss of pulmonary function, as determined by measuring the PenH metric, was observed in vehicle-treated animals infected with MERS-CoV beginning on Day 2 post-infection (Figure 6, Panel D). Mirroring the body weight loss data, normal pulmonary function was observed in groups where treatment was initiated prior to or at 12 hours post-infection (Figure 6, Panel D).

Figure 6: EIDD-2801 Treatment of MERS-CoV Infected Mice



1.3.3.2. Influenza Virus

EIDD-2801 was tested in a ferret model of influenza virus infection and disease. Ferrets recapitulate hallmarks of human influenza infection, providing a clinically relevant animal model to investigate therapeutic intervention. Therapeutic oral dosing of influenza virus-infected ferrets reduced shed levels of pandemic and seasonal IAV by multiple orders of magnitude, and alleviated fever and inflammation while improving airway epithelium histopathology. Post-exposure prophylactic dosing was sterilizing (Toots et al., 2019).

Ferrets infected with pandemic IAV and treated with EIDD-2801 at a dose of 7 mg/kg demonstrated significantly less viral shedding and inflammatory cellular infiltrates in nasal lavages, but mounted a normal humoral antiviral response (Toots et al., 2019).

When examining the effect of delayed dosing, Toots et. al. (2019) demonstrated that treatment with 20 mg/kg of EIDD-2801 initiated 24 hours after challenge with IAV (H1N1) resulted in a rapid decrease in both shed virus and fever relative to control, with fever returning to baseline at 24 hours. When oseltamivir (20 mg/kg) was dosed prophylactically as a positive control, a modest suppression of fever was observed; however, there was no reduction in shed virus titer.

1.3.3.3. Venezuelan Equine Encephalitis Virus

Treatment with EIDD-1931 was evaluated in a mouse model of lethal VEEV infections. To be truly effective as a therapeutic agent for VEEV infection, a drug must penetrate the blood brain barrier and arrest virus replication in the brain. High plasma levels of EIDD-1931 are rapidly achieved in mice after oral dosing. Once in the plasma, EIDD-1931 is efficiently distributed into organs important in the pathology of VEEV infection, including the brain, where it is rapidly converted to its active 5'-triphosphate (EIDD-2061). EIDD-1931 showed a good safety profile in mice after 7 days of dosing with up to 1,000 mg/kg/day. In mouse model studies of VEEV infection, EIDD-1931 was 90-100% effective in protecting mice against lethal intranasal infection when therapeutic treatment was started as late as 24 hours post-infection, and partial protection was achieved when treatment was delayed for 48 hours post-infection (Painter et al., 2019).

[redacted]

1.4. Safety and Secondary Pharmacology

The standard battery of safety pharmacology studies including studies assessing the cardiovascular, respiratory and central nervous systems have been conducted. The studies are discussed in the IB; results indicated that there were no adverse pharmacologic effects of EIDD-2801 on the cardiovascular, respiratory or central nervous systems.

1.5. Nonclinical Pharmacokinetics and Metabolism

1.5.1. Overview

The uptake, metabolism and protein binding of EIDD-2801 and EIDD-1931 have been studied in plasma, microsomes, and non-hepatic cells from several species as outlined below. The PK and tissue distribution of prodrug EIDD-2801 and its active parent EIDD-1931 have been studied extensively in rats, dogs and ferrets. Key results from these studies are presented below; additional detail can be found in the IB.

1.5.2. Absorption

EIDD-1931 is parent of the prodrug EIDD-2801. The appearance of EIDD-1931 is dependent on the absorption of EIDD-2801 and the rate of its conversion to EIDD-1931.

EIDD-2801 PK studies have been completed in dog, rat, mouse, ferret, and monkey. EIDD-2801 was efficiently absorbed and rapidly converted to EIDD-1931 in each species. The t_{max} for EIDD-2801 (not observed in rodents) occurred at 0.5-1 hours, while the t_{max} for EIDD-1931 occurred at 1-2 hours.

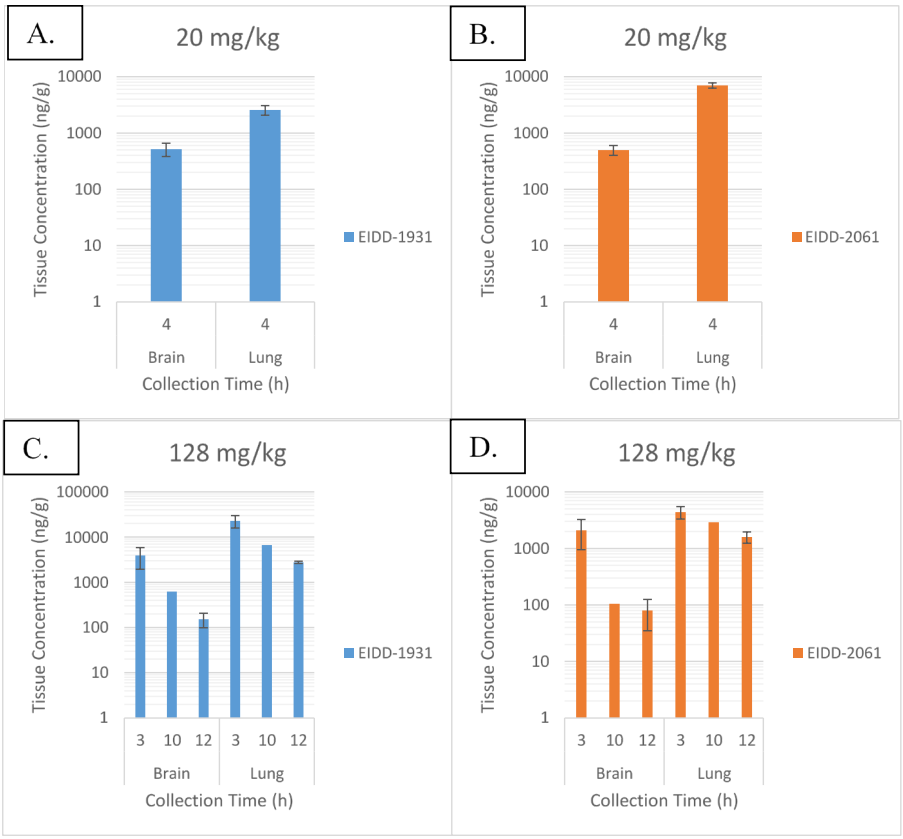
1.5.3. Distribution

[redacted]

1.5.3.2. Tissue Distribution Studies

EIDD-2801 is rapidly absorbed in the gut and converted to EIDD-1931 reaching C_{max} in 1-3 hours in mice, rats, ferrets, dogs and monkeys. EIDD-1931 is then widely distributed to tissues including lungs and brain, where it is rapidly taken up into cells and converted to EIDD-2061. Figure 7 shows the concentration of EIDD-1931 and EIDD-2061 in ferret brain and lung following single doses of 20 (Panels A and B) and 128 (Panels C and D) mg/kg.

Figure 7: Tissue Distribution of EIDD-1931 and EIDD-2061 in Ferret Brain and Lung



1.5.4. Metabolism

1.5.4.1. Metabolic Stability of EIDD-1931 and EIDD-2801

EIDD-2801 was designed to be converted to EIDD-1931 by esterases in plasma or in cells. Stability has been assessed in plasma and liver microsomes from mouse, rat, dog, monkey and humans. The stability of EIDD-2801 in mouse, rat and monkey plasma is relatively short (≤0.4 hours) while the stability is longer in human and dog plasma (1-3 hours). EIDD-2801 stability in mouse, rat, dog and monkey liver microsomes is very short, ranging from 0.02 to 0.08 hours while the stability in human liver microsomes is 1.2 hours (Table 6).

Table 6: Metabolic Stability of EIDD-2801 in Plasma and Liver Microsomes

	Plasma stability	LM stability
Species	t1/2 (h)	t1/2 (h)
Mouse	0.017	0.033
Rat	0.033	0.017
Dog	3.2	0.083
Monkey	0.40	0.017
Human	1.05	1.22

EIDD-2801 is stable in simulated gastric and intestinal fluids (Table 7).

Table 7: Metabolic Stability of EIDD-2801 in Simulated Gastric and Intestinal Fluids and in Buffered Saline with Fetal Bovine Serum

Matrix	t1/2 (hr)
Simulated Gastric Fluid	>24
Simulated Intestinal Fluid	>24
Phosphate Buffered Saline plus 10% Fetal Bovine Serum*	>24

EIDD-1931 was found to be stable when incubated with all tested plasmas, whole blood, liver microsomes and liver S9 extracts and intestinal microsomes (Table 8).

Table 8: Metabolic Stability of EIDD-1931 in Plasma, Whole Blood and Liver and Intestinal Microsomes

Medium	Plasma	Whole Blood	Liver Microsomes	Liver S9 Stability	Intestinal Microsomes
Species	t1/2 (h)	t1/2 (h)	t1/2 (h)	t1/2 (h)	t1/2 (h)
Mouse		>24	>24		>24
Rat	17		>24	>24	
Dog	>24			>24	
Monkey	6.5	>24	>24	>24	>24
Human	10	7	>24	20	>24

1.5.4.2. Uptake and Anabolism of EIDD-1931 in Tissue Culture and Primary Cells

EIDD-1931 is efficiently taken up by tissue culture cells and converted to its pharmacologically active metabolite EIDD-2061 (EIDD-1931-5'-triphosphate). Intracellular EIDD-2061 accumulates dose-dependently, with C_{max} levels ~200-2000 pmol/10^6 cells (at 10-20 µM dose) in different cell lines. It reaches high levels relatively quickly, typically within 1-3 hours, though the t_{max} values vary widely between 1 and 24 hours depending on the cell line and dose concentration tested. Detailed data on the uptake and anabolism of EIDD-2801 is presented in the IB.

EIDD-2801 is also taken up by tissue culture cells and is converted to EIDD-1931 and then to EIDD-2061, but the process is slightly delayed compared to dosing with EIDD-1931. EIDD-1931 is also taken up and metabolized to EIDD-2061 by primary cells. EIDD-2061 is accumulated in all primary cells tested except in mouse primary hepatocytes where EIDD-1931 is apparently extensively metabolized to cytidine and uridine which, in turn, quickly metabolize into CTP and UTP. The quick metabolism of EIDD-1931 consequently results in low levels of EIDD-2061 in mouse hepatocytes. The intracellular stability ($t_{1/2}$) of EIDD-2061 is 4-5 hours in human astrocytes and hBTEC and is significantly shorter (0.2-1.1 hours) in primary hepatocytes.

1.5.5. Excretion

Currently, there is no data on excretion of EIDD-2801 or EIDD-1931. Excretion will be measured in urine during this study.

1.5.6. Pharmacokinetic Drug Interactions

1.5.6.1. Cytochrome P450 (CYP) Inhibition

The purpose of this non-GLP *in vitro* study was to determine the time-dependent inhibitory potential of EIDD-2801 and EIDD-1931 on human cytochromes P450 (CYP) 1A2, 2B6, 2C8, 2C9, 2C19, 2D6, and 3A4 (midazolam and testosterone substrates) enzyme activity, using pooled human liver microsomes in an half-maximal inhibitory concentration (IC_{50}) shift assay.

Neither EIDD-2801 nor EIDD-1931 demonstrated inhibition greater than 31.4% for any of the CYP isozymes tested nor could the data for each assay condition be curve fit to determine time-dependent inhibition by these compounds. Full dose-response curves were not achieved at concentrations ranging from 0.00545 to 50.0 µM indicating EIDD-2801 and EIDD-1931 have no CYP inhibition potential at concentrations ranging from 0.00545 to 50.0 µM. Assay performance was acceptable based on the results for the positive control inhibitors.

1.5.6.2. CYP Induction

An assay was performed to determine the induction potential of EIDD-2801 on human CYP isoenzyme (1A2, 2B6, and 3A4) activity using 3 single-donor lots of inducible, cryopreserved human hepatocytes. Both enzyme activity and mRNA results demonstrated that EIDD-2801 did not show induction for any of the CYP isozymes.

[redacted]

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			[redacted]	[redacted]
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1.7. Potential Risks and Benefits

1.7.1. Potential Benefits

As this is a FIH study in healthy volunteers, there is no direct benefit to subjects enrolled in the study. However, given the current pandemic situation and *in vitro* antiviral activity of EIDD-2801 against SARS-CoV-2, and the activity against several other viruses of public health concern, it is possible that participants may benefit from future availability of the drug.

1.7.2. Potential Risks

EIDD-2801 has never been administered to humans; therefore, the risks from EIDD-2801 to subjects participating in this trial are unknown. Although toxicology studies have been done, unexpected AEs may occur. [redacted]. Subjects will also be monitored for other end-organ effects through a range of safety assessments.

2. OBJECTIVES

2.1. Study Objectives

2.1.1. Primary Objectives

The primary objective of Part 1 of the study is to determine the safety and tolerability of single ascending doses of EIDD-2801.

The primary objective of Part 2 of the study is to assess the effect of food on the PK on EIDD-2801 and EIDD-1931 following a single oral dose.

The primary objective of Part 3 of the study is to determine the safety and tolerability of multiple ascending doses of EIDD-2801.

2.1.2. Secondary Objectives

The secondary objectives of Part 1 and Part 3 of the study is to define the PK of EIDD-2801 and EIDD-1931 in plasma and urine following single and multiple doses administered to healthy volunteers.

The secondary objective of Part 2 of the study is to determine the safety and tolerability of single doses of EIDD-2801.

2.1.3. Exploratory Objectives

The exploratory objective of Part 3 is to collect data to assess the relationship between EIDD-2801/EIDD-1931 concentrations and QTc

2.2. Study Outcome Measures

2.2.1. Primary Outcome Measures

The primary outcome measures for Parts 1 and 3 of are results of safety evaluations including safety laboratory assessments, PEs, ECGs, vital signs, and AEs.

The primary outcome measures for Part 2 of the study are plasma PK parameters including C_{max}, t_{max}, $t_{1/2}$, CL/F, λz, Vz/F, $AUC_{0\text{-}inf}$ and AUC_{last} as appropriate.

2.2.2. Secondary Outcome Measures

The secondary outcome measures are as follows:

- Single-dose plasma PK parameters, including C_{max}, t_{max}, $t_{1/2}$, CL/F, λz, Vz/F, $AUC_{0\text{-}inf}$ and AUC_{last} as appropriate (Part 1)

- Multiple-dose plasma PK parameters, including C_{trough}, C_{max}, t_{max}, $t_{1/2}$, CL/F, λz,

Vz/F, AUC_{τ}, $AUC_{0\text{-}inf}$ (Day 1 dose only), $RA_{AUC\tau}$ and RA_{Cmax}, as appropriate (Part 3)

- Urinary excretion of EIDD-2801 and EIDD-1931 following single- and multiple-dose administration (Parts 1 and 3).

- Results of safety evaluations including safety laboratory assessments, PEs, ECGs, vital signs, and AEs (Part 2).

3. STUDY DESIGN

3.1. Overview

EIDD-2801-1001-UK is a Phase 1, randomized, double-blind, placebo-controlled, FIH, SAD, and MAD study of the safety, tolerability and PK of EIDD-2801 and EIDD-1931 following oral administration of single and multiple doses of EIDD-2801 to healthy volunteers. In addition, for a minimum of one cohort, the effect of food on the single-dose EIDD-2801 and EIDD-1931 PK parameters will be assessed in subjects taking open-label EIDD-2801. The overall objective of the study is to identify a starting dose for future safety and therapeutic intervention trials.

The study is composed of 3 parts; P1 is the SAD study, P2 is the FE cohort study, and P3 is the MAD study.

This study will be conducted at 1 site in the United Kingdom and up to 3 sites in the United States.

3.1.1. Part 1 (Single Ascending Dose)

A single oral dose of EIDD-2801 or PBO will be administered to subjects. Subjects will be randomized in a 3:1 ratio to receive either EIDD-2801 or PBO.

Following dosing on Day 1, subjects will remain in the clinic through completion of study assessments on Day 4, returning to the clinic on Day 9 for study assessments, and Day 15 for the EOS visit.

The first cohort will be divided into 2 groups. The first group, a sentinel group, will include 2 subjects. On Day 1, one subject in the sentinel group will be administered PBO and one will be administered EIDD-2801. Analysis of the safety results for the sentinel group will be reviewed 24 hours postdose. If no halting criteria (Section 9) have been met, the remainder of the subjects in Cohort 1 (the second group) will be dosed. There will be no sentinel groups for the remaining cohorts.

After completion of each dosing cohort, safety and tolerability data will be reviewed to determine if any of the halting rules have been met. If not, then the subsequent cohort may be dosed following review of the 72-hour safety data. As PK data become available, these data may be used for dose-escalation decisions.

The proposed dose-escalation scheme is shown in Figure 1, however, planned dose escalations will be determined based on ongoing review of the safety, tolerability, and available PK data. The starting dose in the first SAD cohort will be 50 mg. Dose levels will not increase by more than 3-fold for predicted nonpharmacologically active dose levels and by 2-fold for predicted pharmacologically active dose levels. [redacted]

Three cohorts are initially planned for P1; however, up to an additional 3 cohorts may be enrolled.

3.1.2. Part 2 (Food-Effect)

Two single oral doses of EIDD-2801 will be administered to subjects, in an open-label manner. The dose for the P2/FE cohort is planned to be 100 mg but may be adjusted based on safety, tolerability, and/or PK data from P1. The dose assessed in the P2 cohort will have been given previously to subjects in a P1 cohort successfully (i.e., no halting rules were met following dosing).

Subjects will be randomized to a treatment sequence; i.e., to receive drug in the fed then fasted state (Sequence 1) versus fasted then fed state (Sequence 2; Figure 1). Half of the subjects in the cohort will be randomized to Sequence 1 and the other half will be randomized to Sequence 2.

Following dosing on Day 1, subjects will remain in the clinic through completion of study assessments on Day 4, returning to the clinic on Day 9 for study assessments, and Day 14 to begin the second dosing sequence on Day 15. Subjects will then be discharged from the clinic on Day 18, returning to the clinic on Day 23 for study assessments, and Day 30 for the EOS visit. There will be a 14-day (minimum) washout period between doses.

One cohort of 10 subjects is planned for P2. However, if PK results obtained are equivocal, additional subjects may be enrolled into the P2/FE cohort at the previously tested dose or an additional P2/FE cohort may be added at a different dose.

3.1.3. Part 3 (Multiple Ascending Dose)

Subjects in P3 will be randomized in a 3:1 ratio to receive either EIDD-2801 or PBO. Twice-daily dosing will be administered to subjects on Day 1 through Day 5, inclusive, and a final dose will be administered on the morning of Day 6 for collection of steady-state PK samples during waking hours. Depending on ongoing review of the safety, tolerability, and PK data, the number of days of dosing may be reduced and the dosing frequency may be changed; the dosing frequency will be no less than once-daily or no greater than three-times daily.

The proposed dose-escalation scheme is shown in Figure 1. The total daily dose will not exceed a dose shown to be safe and well-tolerated in P1. Doses in P3 may be administered in the fed state, following review of the PK data obtained from P2. Part 3 may run in parallel with P1, providing that the total daily dose to be administered does not exceed a dose already shown to be safe and well-tolerated in P1.

Subjects will remain domiciled at the site during the dosing period and until Day 9, returning to the clinic on Day 14 for completion of study assessments, and Day 20 for the EOS visit. If the number of days of dosing are reduced, the timepoints for subsequent assessments and the period for which subjects are domiciled at the clinic will similarly be changed (subjects will be domiciled for 3 days post final dose).

Three cohorts are initially planned for P3; however, up to an additional 3 cohorts may be enrolled, or the number of cohorts may be reduced if the study objectives are met. Dose levels in P3 may be repeated for collection of PBMCs, providing that the dose level did not meet the dose-escalation halting criteria.

Additional parts may be added to this study, including a SAD part in elderly healthy subjects and a study of the safety, tolerability, and PK in Japanese subjects. These additional parts are not described in this protocol, but may be added via a protocol amendment.

3.2. Rationale and Justification

3.2.1. Justification of Design

The FIH study is a typical dose-escalation study designed to provide the maximum amount of data in the minimum number of subjects. The cohort size in P1 and P3 is planned to be 8 subjects (6 active:2 PBO). This number of subjects allows for a sufficient PK analysis, considered to be important because dose extrapolation from efficacious animal models to humans will be based on exposure. The duration of participation for each subject following dosing well exceeds 5 drug half-lives (up to 9.1 hours in dogs; 5 hours in ferrets) [redacted] The FE cohort is considered to be important to maximize exposure based on fed vs. fasted condition and obtaining this information early in Phase 1 will minimize study drug dose in all future studies. This design follows the FDA guidance document on assessing FE in clinical studies.

3.2.2. Justification of Starting Dose

[redacted] The FDA guidance document *("Estimating the Maximum Safe Starting Dose in Initial Clinical Trials for Therapeutics in Adult Healthy Volunteers")* provides guidance for when a safety factor smaller than 10 may be used to calculate the starting dose:

- A smaller safety factor might also be used when toxicities produced by the therapeutic are easily monitored, reversible, predictable, and exhibit a moderate-to-shallow dose-response relationship with toxicities that are consistent across the tested species (both qualitatively and with respect to appropriately scaled dose and exposure).

- A safety factor smaller than 10 could be justified when the NOAEL was determined based on toxicity studies of longer duration compared to the proposed clinical schedule in healthy volunteers. In this case, a greater margin of safety should be built

into the NOAEL, as it was associated with a longer duration of exposure than that proposed in the clinical setting. This assumes that toxicities are cumulative, are not associated with acute peaks in therapeutic concentration (e.g., hypotension), and did not occur early in the repeat dose study.

EIDD-2801 satisfies both of these criteria as follows:

Importantly, given the activity of EIDD-2801 versus SARS-CoV-2 (cause of COVID-19), the Sponsor thinks it is most prudent to start with a dose that is predicted to be a safe starting dose and is also as close as possible to a potential therapeutic dose that would allow the Sponsor to move into COVID-19 patients as safely and expeditiously as possible. Based on modeling to animal data, a dose of 100 mg BID is projected to be an active dose in humans.

3.2.3. Justification of Study Population

Healthy volunteers are considered to be the appropriate population for conduct of the FIH study. Healthy volunteers without confounding medical conditions that may obscure the interpretation of AEs or affect absorption, distribution, metabolism and excretion of study drug will provide the most valuable data regarding the tolerability, safety, and plasma exposures observed and expected following single doses and multiple doses up to 10 days. EIDD-2801 is intended for eventual study in patients with potential CoV-2 infection as defined by the Centers for Disease Control and Prevention (CDC) in whom a range of AEs are expected based on the disease under study. Understanding the safety and PK profile in a normal population will better inform the use of EIDD-2801 in disease settings where complications are frequent, and AEs will need to be interpreted in context.

4. STUDY POPULATION

This study will enroll healthy volunteers; 8 subjects will be enrolled into each SAD and MAD cohort in P1 and P3, and 10 subjects will be enrolled into the FE cohort in P2.

The site is strongly encouraged to ensure that women are represented in each cohort.

4.1. Subject Inclusion Criteria

Subjects who meet all of the criteria at Screening will be eligible for study participation

1. Male or female between the ages of 18 and 60, inclusive.

2. Female subjects must be of nonchildbearing potential. Nonchildbearing potential is defined as women who:

    - are physiologically incapable of becoming pregnant including those who are surgically sterile, via hysterectomy, bilateral salpingectomy, and/or bilateral oophorectomy by reported medical history and/or medical records. Surgical sterilization to have occurred a minimum of 6 weeks, or at the PI's (or designee's) discretion, prior to Screening.

    - are post-menopausal (at least 45 years of age with amenorrhea for 12 months without an alternative medical reason with confirmatory FSH levels of ≥40 mIU/mL. The amenorrhea should not be induced by a medical condition such as anorexia nervosa, hypothyroid disease or polycystic ovarian disease, or by extreme exercise. It should not be due to concomitant medications that may have induced the amenorrhea such as oral contraceptives, hormones, gonadotropin-releasing hormones, anti-estrogens, or selective estrogen receptor modulators).

3. Male participants must agree to the use of effective contraception for study duration, i.e., from Check-in until 90 days after the EOS visit.

4. Is in generally good health as determined by medical history, PEs (at Screening and/or Check-in), vital signs, and other screening procedures.

5. Has a BMI of 18 to 30 kg/m^2.

6. Is willing and able to comply with all study procedures including providing informed consent.

4.2. Subject Exclusion Criteria

Subjects who meet any of the following criteria at Screening (unless otherwise stated) will not be eligible for participation:

1. Females who are pregnant, planning to become pregnant, or breastfeeding.
2. Has a clinically relevant acute or chronic medical condition or disease of the cardiovascular, GI, hepatic, renal, endocrine, pulmonary, neurologic, or dermatologic systems as determined by the PI (or designee).
3. Has any current or historical disease or disorder of the hematological system or significant liver disease or family history of bleeding/platelet disorders.
4. Has a history of cancer (other than basal cell or squamous cell cancer of the skin), rheumatologic disease or blood dyscrasias.
5. Has a history of GI surgery or other condition that may interfere with absorption of study drug, in the option of the PI (or designee).
6. Has a history of febrile illness within the 14 days prior to the first dose of study drug.
7. Has a positive alcohol or drug screen at Screening or the Day -1 visit or has a history of alcohol or drug abuse within the past 5 years.
8. Currently uses tobacco, nicotine or tobacco products or e-cigarettes or stopped using tobacco products within the past 3 months and/or has a positive cotinine test at Screening or Check-in.
9. Has any abnormal laboratory value of Grade 2 or higher, which is considered to be clinically significant by the PI (or designee).
10. Has a total white cell count or absolute lymphocyte count outside the normal range, or hemoglobin or platelet levels below the lower limit of normal, at Screening or Day -1.
11. Has values above the upper limit of normal for the following laboratory analytes: ALT/SGPT, alkaline phosphatase (serum), AST/SGOT, at Screening or Day -1.
12. Positive test result for HIV, HBV, or HCV.
13. Has an autoimmune disease, is immunosuppressed or is in any way immunocompromised.
14. Has any of the following:
    - QTcF >450 ms confirmed by repeat measurement

- QRS duration >110 ms confirmed by repeat measurement
- PR interval >220 ms confirmed by repeat measurements
- findings which would make QTc measurements difficult or QTc data uninterpretable
- history of additional risk factors for torsades de pointes (e.g., heart failure, hypokalemia, family history of long QT syndrome).

15. Except as noted, has used prescription drugs (other than hormone replacement therapy) within 14 days prior to the first dose of study drug unless, in the opinion of the PI (or designee), the drug will not interfere with study assessments. The following prescription drugs will be allowed:
    - Statins: Subjects who have been on a stable dose of statins for a minimum of 6 months and who have normal creatine kinase at Screening.
    - Antihypertensives: Subjects who have been on a stable dose of antihypertensives for a minimum of 6 months and have controlled blood pressure and no active heart disease.
16. Has received an experimental agent (vaccine, drug, biologic, device, blood product or medication) within 3 months prior to the first dose of study drug and agrees not to receive another experimental agent during the duration of this trial.
17. Has donated blood within 30 days, plasma within 2 weeks, or platelets within 6 weeks prior to Screening or refuses to agree to not donate blood until 3 months after the EOS visit.
18. Uses OTC medications or nutritional supplements (including, e.g., gingko biloba, aspirin at any dose, and NSAIDs) on a routine/scheduled basis and/or is unwilling to abstain from such use for the duration of the study.
19. Is unwilling to abstain from quinine containing products including quinine water, tonic water, bitter lemon, jello shots, any quinine preparations or Cinchona tree bark preparations (e.g., herbal medications containing Cinchona tree bark).
20. Has any condition that would, in the opinion of the PI (or designee), put the subject at increased risk for participation in a clinical trial or confinement in a clinical research unit.

5. STUDY MEDICATION, RANDOMIZATION AND DOSE ADMINISTRATION

5.1. Study Drug Description

EIDD-2801 and matching PBO will be supplied [redacted]

[redacted]

[redacted]

[redacted]

5.1.1. Acquisition, Formulation, Packaging and Labeling

[redacted]

All study drug will be labeled according to the regulatory requirements for investigational product.

5.1.2. Product Storage and Stability

Study drug should be stored at controlled room temperature defined as [redacted]s. If excursions occur which are outside of this range, the pharmacy staff should contact Sponsor to determine the course of action. Additional stability data may be available which would allow continued use of the study drug, or study drug may need to be replaced.

5.2. Randomization

Unmasked study drug (EIDD-2801 and matching PBO) will be supplied to the study site pharmacy. The pharmacy staff will be unmasked with regards to treatment assignment. A randomization list will be provided to the pharmacy staff who will use that list to dispense masked study drug for administration to each study participant. In P1 and P3, 6 subjects will

receive EIDD-2801 while 2 subjects will be randomized to PBO. In P2, all 10 subjects in the FE cohort will receive EIDD-2801. The pharmacy staff will maintain the security of the randomization list ensuring that no study personnel outside of the pharmacy have access to identify treatment assignment. In the case that it becomes necessary to know a subject's treatment assignment, unmasking procedures will be followed as discussed in Section 8.4.

5.3. Dosage, Preparation and Administration of Study Drug

Detailed instructions for extemporaneous compounding (as necessary), dispensing and administering study drug can be found in the pharmacy manual.

[redacted]

5.4. Drug Accountability

The site pharmacy must maintain records of receipt and disposition of all study drug supplied to the site by the Sponsor. The records must be maintained according to site standard operating procedures (SOPs) and should include at a minimum, receipt date, lot or batch number, amount and formulation received, [redacted], date, time and amount dispensed, subject identifier for each aliquot of study drug dispensed. In addition, the clinic staff must ensure that the time of dosing is identified for each subject.

Monitors must verify drug accountability/dispensing records during the monitoring visit.

Unused study drug must be disposed of according to the procedures described in the pharmacy manual.

5.5. Concomitant Medication/Treatments

[redacted] In this FIH study, limited types of concomitant medications are permitted. Adjustment of routine medications taken by subjects should be avoided during study participation except when subject safety could be affected by lack of

adjustment. There are no restrictions on treatment medications prescribed by the PI (or designee) to be used for AEs that occur during study participation. For additional restrictions, see Section 6.1.2.

6. STUDY CONDUCT AND VISIT SCHEDULE

All study assessments will be conducted according to the Time and Events Schedule.

6.1. Study Conduct

6.1.1. Study Windows and Rounding Principles

The time schedule described in the protocol for each scheduled activity should be followed as closely as possible. However, if it is not possible and if it is not otherwise specifically contraindicated per protocol, then the time windows detailed in Table 10 are allowed without incurring a protocol deviation.

Table 10: Allowable Time Windows for Study Assessments/Visits

Protocol Specified Time	Allowable Window
PK Samples and Associated Assessments	
Predose	within 2 hours prior to dosing
<2 hours	± 5 minutes
≥2 hours to 24 hours	± 15 minutes (± 2 hours for urinalysis)
>24 hours to 48 hours	± 30 minutes
>48 hours	± 1 hour (PK) and ± 2 hours (safety)
Study Visits - Parts 1 and 2	
Day −1 to Day 4 (Part 1)	must occur on scheduled day
Day −1 to Day 4 and Day 14 to Day 18 (Part 2)	must occur on scheduled day
Day 9 (Part 1) and Days 9 and 23 (Part 2)	± 1 day
End-of-Study (EOS) Visit	± 2 days
Study Visits - Part 3	
Day −1 to Day 9	must occur on scheduled day
Day 14	± 1 day
Day 20/EOS Visit	± 2 days

6.1.2. Restrictions

Prior to arriving at the clinic for the Day -1 visit, subjects must abstain from consumption of alcoholic beverages for a minimum of 72 hours prior to Check-in. Subjects must continue to abstain from consumption of alcoholic beverages throughout clinic confinement. Subjects enrolled in P2 must continue to refrain from consuming alcoholic beverages from discharge on Day 4 through Check-in on Day 14, and then through the second clinic confinement to Day 18. After discharge on Day 4 (P1), Day 18 (P2), or Day 9 (P3), subjects must minimize consumption of alcoholic beverages (i.e., limit of up to one serving per day) until the EOS procedures have been completed.

All subjects must refrain from the following:

- consuming quinine containing products from 72 hours prior to Check-in through to completion of the EOS procedures.
- using nutraceuticals and nutritional/vitamin supplements (e.g., gingko biloba, multivitamins) from 72 hours prior to Check-in through to completion of the EOS procedures. However, vitamin supplements required by a physician are exempt from this restriction.
- taking OTC analgesics including aspirin (any dose) and NSAIDS from 72 hours before Check-in until completion of EOS study procedures unless prescribed by the PI (or designee).
- use of tobacco, nicotine or tobacco products, or e-cigarettes from 3 months prior to Screening until the EOS visit.
- strenuous exercise from 7 days before Check-in until the EOS visit. Subjects will otherwise maintain their normal level of physical activity during this time (i.e., will not begin a new exercise program nor participate in any unusually strenuous physical exertion).
- Female subjects must not donate eggs/ovum from the time of Check-in until 3 months after the EOS visit.

All subjects in P1 should be dosed in the fasted state. Subjects should fast overnight for a minimum of 10 hours prior to dosing in the morning on Day 1. Following dosing, subjects may have water after 2 hours and food beginning 4 hours postdose. For subjects to be dosed in the fed state, subjects should have a high-fat breakfast as defined in the FDA guidance. Subjects must complete the meal within 30 minutes of starting the meal and should be dosed after 30 minutes of starting the meal. Doses in P3 may be administered in the fed state, following review of the PK data from P2.

6.2. Screening

6.2.1. Screening Visit

Subjects who meet preliminary pre-screening criteria (as defined by the site) and are interested in participating in the study will arrive at the study site for administration of informed consent according to site standard operating procedures (SOPs). After the subject has signed and dated the ICF, screening procedures can begin. Assessments and procedures should be conducted as shown in the Time and Events Schedule Screening may be conducted as early as 28 days prior to dosing.

6.2.2. Retesting Procedures

In the case where a parameter is outside of the acceptable limits for enrollment, measurement of the parameter can be repeated one time if both the following are met:

- In the opinion of the PI (or designee), the parameter value does not represent a chronic condition that otherwise will preclude the volunteer from enrolling in the study; and
- In the opinion of the PI (or designee), the abnormality is not likely to recur.

6.2.3. Study Visits

Subjects who satisfy entry criteria will return to the clinic for the Day -1 visit. Following review of I/E criteria, subjects who still qualify will be checked into the clinic and enrolled into the study. Following enrollment, clinical chemistry, hematology and urine samples will be collected to determine baseline values. Based on site standard practices, alternate subjects will also be enrolled into the study in case one of the selected subjects cannot be dosed. If all subjects can be dosed on the morning of Day 1, the alternates will be released and may be enrolled into subsequent cohorts. All assessments and procedures will be performed according to the Time and Events Schedule.

6.2.4. Part 1 (Single Ascending Dose)

Subjects will remain in the clinic through completion of study procedures on Day 4, returning to the clinic for procedures on Day 9.

6.2.5. Part 2 (Food-Effect)

Subjects will remain in the clinic through completion of study procedures on Day 4, returning to the clinic on Day 9 for study assessments, and Day 14 to begin the second dosing sequence on Day 15. Subjects will then be discharged from the clinic on Day 18, returning to the clinic on Day 23 for study assessments.

6.2.6. Part 3 (Multiple Ascending Dose)

Subjects will remain in the clinic through dosing and completion of study procedures on Day 9, returning to the clinic on Day 14 for study assessments. Depending on ongoing review of the safety, tolerability, and PK data, the number of days of dosing may be reduced. If the number of days of dosing are reduced, the timepoints for subsequent assessments and the period for which subjects are domiciled at the clinic will similarly be changed (subjects will be domiciled for 3 days post final dose).

6.3. Safety Follow-up and End-of-Study Visit

Subjects in P1 will return to the clinic for the EOS visit on Day 15. Subjects in the FE cohort (P2) will return for the EOS visit on Day 30. Subjects in P3 will return for the EOS visit on

Day 20. Subjects with drug-related AEs at the EOS visit will be followed as discussed in Section 8.2.1.

6.4. Subject Withdrawal and Replacement

As this is a small study with a limited number of subjects per cohort, it is critical that all subjects complete the study including postdose study assessments. Site personnel should emphasize this to study subjects at the time of informed consent so that subjects will understand this fact before agreeing to participate in the study.

Subjects may voluntarily withdraw their consent for study participation at any time without penalty or loss of benefits to which they are otherwise entitled. An PI (or designee) may also withdraw a subject from receiving study drug or participation in the study for any reason. Subjects who withdraw or are withdrawn from the study should undergo withdrawal procedures as discussed below. These procedures would include follow-up safety evaluations.

6.4.1. Reasons for Withdrawal

If a subject withdraws or is withdrawn from the study, the primary consideration must be the health and welfare of the subject. The reasons for withdrawal might include but are not limited to the following:

- Subject no longer meets eligibility criteria including subject withdraws consent from study participation (with or without a reason)
- Subject becomes noncompliant
- Medical disease or condition, or new clinical finding(s) for which continued participation, in the opinion of the PI (or designee) might compromise the safety of the subject, interfere with the subject's successful completion of this study, or interfere with the evaluation of responses
- Subject Lost-to-Follow-up
- Subject becomes pregnant, if applicable
- Determined by a physician's discretion to require additional therapy not indicated in the protocol to ensure subject's health and well-being (or treatment failure, if applicable)

The PI should be explicit regarding study follow-up (e.g. safety follow-up) that might be carried out. If the subject consents, every attempt will be made to follow all AEs through resolution, return to baseline, or until stabilized with sequelae for a maximum of 30 days following discontinuation. The procedures that collect safety data for the purposes of research must be inclusive in the original ICF or the PI may seek subsequent informed consent using an EC-approved ICF with the revised procedures.

The PI will inform the subject that already collected data will be retained and analyzed even if the subject withdraws from this study.

6.4.2. Handling of Withdrawals

Subjects who withdraw from the study prior to receiving study drug (i.e., on Day -1 or before dosing on Day 1) will be discharged from the clinic and followed only if AEs are present which occurred due to participation in the study (e.g., AE resulting from a study procedure). In this case, the subject should be followed until the AE resolves or the PI determines that the AE has stabilized.

Subjects who withdraw from the study after receiving study drug should have EOS assessments at the time of withdrawal or as quickly thereafter as possible.

Subjects who do not return for follow-up procedures on Days 9/23 or 15/30 (P1 and P2), or Days 14 or 20 (P3) will be contacted by the site at least 3 times using the subject's preferred method of communication (as determined at Check-in). If the site is unable to contact the person, then a certified letter will be sent. If the subject still cannot be reached or refuses to come back to the clinic after all attempts, the subject will be considered Lost-to-Follow-up and withdrawn from the study.

6.4.3. Documentation of Withdrawals

If a subject withdraws or is withdrawn prior to completion of the study, the reason for this decision must be recorded in the CRF. If the subject is Lost-to-Follow-up, the site should document the attempts to contact the subject in the source documents. If the subject has an ongoing AE at the time of withdrawal, then the AE should be followed as detailed in Section 8.2.1.

6.4.4. Subject Replacement

If a subject withdraws from the study prior to receiving study drug, the subject will be replaced. In this case, designated alternate subjects, if available, will be first in line to replace the withdrawn subject.

If a subject withdraws from the study after receiving study drug, then the decision to replace the subject will be made by the PI (or designee) in consultation with the Sponsor. Factors to consider will be the timing postdose of withdrawal and the number of safety and PK assessments completed prior to withdrawal. Subjects who are withdrawn because of an AE related to the study drug will not be replaced.

6.5. Unscheduled Visit(s)

If a subject experiences an AE after discharge from the clinic but prior to the EOS visit, the subject should be instructed to call the site. Based on the issue, the PI (or designee) may request that the subject return to the site for an unscheduled visit. In this case, procedures/assessments

should be conducted as deemed appropriate for the situation by the PI (or designee). The visit should be recorded in the unscheduled visit page of the CRF.

7. STUDY PROCEDURES/EVALUATIONS

7.1. Demography and Medical History

Demographics including age, gender, race, ethnicity, and medical history will be recorded for each subject. All significant medical history should be recorded. In general, significant medical history should include all ongoing events and all events occurring within the last 6 months. Clinically relevant or clinically significant events occurring greater than 6 months ago should be recorded. All surgeries occurring in adulthood should be recorded. If surgeries occurred more than 2 years ago, then only the year needs to be recorded on the CRF.

7.2. Clinical Evaluations

7.2.1. Physical Examinations

The PE will be performed by the PI or a designee that is licensed to perform a PE per local requirements. The initial PE performed at Screening and the final PE conducted at the EOS visit will include examination of all pertinent body systems as defined by the site standard PE body systems (general appearance, HEENT, lymphatic, cardiovascular, respiratory, GI, musculoskeletal, neurological, dermatological).

Subsequent PEs will be performed as shown in the Time and Events Schedule and will be targeted to any new signs or symptoms, any change from the previous status or any body system deemed pertinent to that subject by the PI (or designee). Clinically significant abnormalities should be recorded in the CRF; those occurring prior to dosing will be included in medical history unless the abnormality was the direct result of study participation.

7.2.2. Vital Sign Measurements and ECGs

Vital signs include pulse rate, respiratory rate, blood pressure, and body temperature. Vital signs should be measured after the subject has been supine for a minimum of 5 minutes. Site standard ranges will be used for determining any out-of-range values.

Height and weight should be measured, and BMI calculated at Screening as indicated in the Time and Events Schedule.

Resting 12-lead ECGs should be recorded at the visits indicated in the Time and Events Schedule after the subject has been supine for a minimum of 5 minutes. The PI (or designee) will evaluate the ECG tracings to determine if there are out-of-range values; if out-or-range values are detected, the PI (or designee) will determine if they are clinically significant. Site standard ranges will be used to determine if any parameters are considered out-of-range. At the discretion of the PI (or designee), the ECG may be repeated if erroneous readings are suspected.

7.2.2.1. Continuous 12-lead ECG Monitoring (Part 3 Only)

Continuous 12-lead ECG monitoring using a digital recorder will take place at the times indicated in the Time and Events Schedule, in P3 only.

All continuous 12-lead ECG data collected will be archived without extraction or analysis and will not be reported in the scope of this study.

Subjects will be supine for at least 10 minutes before the extraction timepoint and for 5 minutes from the start of each extraction timepoint (each extraction will last for 5 minutes). Environmental distractions (e.g., television, radio, conversation) should be avoided during the pre-ECG resting period and during ECG recording.

When coinciding, PK sampling should always be performed after the ECG extraction time window. If a separate ECG machine is being used for safety assessments, that machine should be in place prior to the extraction window to permit safety ECGs to be recorded irrespective of the extraction window. If the machine is not in place prior to the extraction window, safety ECGs must be recorded after the extraction window. If an integral system is used, safety ECGs may be recorded irrespective of the extraction window.

7.2.3. Adverse Events and Concomitant Medications

Adverse Events: The PI is responsible for identifying and documenting events meeting the definition of an AE or SAE (Section 8.1). Once each day while the subject is in the clinic and once during each out-patient visit, the PI (or designee) should inquire about the occurrence of AEs. The following are examples of open-ended questions that may be used to obtain this information: *"How are you feeling?"; "Have you had any medical problems recently?"; "Have you taken any new medicines since your last visit/assessment?"*

All AEs and SAEs must be documented in the source documents and recorded in the CRF.

Concomitant Medications: All medications (prescription or over-the-counter), nutritional supplements, and nutraceuticals taken by the subject from 30 days prior to dosing through the EOS visit must be recorded in the CRF. Medication information should include indication, dose, frequency, and route of administration. Any medication taken for an AE/SAE should be documented as such. Refer to Section 5.5 for additional information.

7.3. Laboratory Evaluations

The laboratory will perform standard routine testing, and processing of all blood samples. For the entire study, the amount of blood collected from any one subject will not exceed 500 mL.

7.3.1. Routine Laboratory Panels

Blood and urine samples will be collected at the times indicated in the Time and Events Schedule. The analytes shown in the table below (Table 11) will be assessed.

If a method to determine COVID-19 status becomes readily available, subjects may be tested at Screening and Check-in to confirm they are not positive for COVID-19 prior to dosing.

Table 11: Laboratory Evaluations

CHEMISTRY PANEL Alanine Aminotransferase (ALT/SGPT) Albumin, Serum Albumin/Globulin (A/G) Ratio (calculation) Alkaline Phosphatase, Serum Amylase Aspartate Aminotransferase (AST/SGOT) Bilirubin, Total and Direct BUN BUN/Creatinine Ratio (calculation) Calcium, Serum Creatinine, Serum Creatinine Kinase (CK) Gamma Glutamyl Transferase (GGT) Lactate Dehydrogenase (LDH) Uric Acid Electrolyte Panel (Na+, K+, Cl-, Bicarb.) Phosphorus Globulin, Total Glucose, Serum Lipase Protein, Total, Serum	**HEMATOLOGY PANEL** Hemoglobin Hematocrit White Blood Cell Count with differential (absolute and percentage) Red Blood Count Prothrombin Time (PT)/Partial Prothrombin Time (PTT) and International Normalized Ratio (INR) Mean cell hemoglobin (MCH), mean cell hemoglobin concentration (MCHC), mean cell volume (MCV), red cell distribution width (RDW; may be a Grade 1 abnormality) Platelets
	ADDITIONAL ASSESSMENTS Virology: Human Immunodeficiency Virus (HIV) serology, Hepatitis B Virus (HBV; Surface Antigen [HBsAg]), Hepatitis C Virus (HCV) Follicle-Stimulating Hormone (FSH; as applicable)
	PREGNANCY TEST Serum Pregnancy Test Urine Dipstick (optional blood follow-up)
	DRUG SCREENING Serum/urinalysis (per site SOP) Cotinine Urine Dipstick Alcohol Breathalyzer
	ROUTINE URINALYSIS Bilirubin Color and appearance Glucose Ketones Leukocytes Microscopic (including red blood cells [RBCs] and white blood cells [WBCs]) Nitrite Occult blood pH Protein Specific Gravity Urobilinogen

7.3.2. Pharmacokinetic Sampling

Samples for PK analysis will be collected as shown in the Time and Events Schedule. All samples will be analyzed to define the PK parameters for prodrug EIDD-2801 and the active parent, EIDD-1931. In order to preserve EIDD-2801, special handling procedures will be put in place. These procedures will be documented in the laboratory manual for the study.

Depending on ongoing review of the PK data, timepoints may be modified or removed, and up to 3 additional samples (in each period for the food-effect) may be added.

7.3.3. Peripheral Blood Mononuclear Cell Collection

Peripheral blood mononuclear cells will be collected in P2 and P3. Blood will be collected into specialized tubes and processed according to procedures described in the laboratory manual.

Depending on ongoing review of the data, timepoints may be modified or removed, and up to 2 additional samples may be added.

7.3.4. Urine Collection

Urine will be collected over the time periods noted in the Time and Events Schedule. Samples will be collected for routine urinalysis and PK analysis according to site standard practices and as described in the laboratory manual.

8. SAFETY MONITORING, MANAGEMENT AND REPORTING

8.1. Definitions

8.1.1. Adverse Events

An AE is 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 an abnormal laboratory finding), symptom, or disease temporally associated with the use of a medicinal (investigational) product, whether or not related to the medicinal (investigational) product.

Abnormal clinical laboratory findings (e.g. clinical chemistry, hematology) or other abnormal assessments that are judged by the PI (or designee) as clinically significant will be recorded as AEs or SAEs if they meet the definitions of an AE or an SAE as defined in this Section 8.1.2. Disease specific signs and symptoms which were ongoing prior to study entry will not be considered AEs unless they worsen (e.g. increase in frequency or severity) unexpectedly during the course of the trial.

8.1.2. Serious Adverse Events

An AE or suspected adverse reaction is considered "serious" if, in the view of either the PI (or designee) or Sponsor, it results in any of the following outcomes:

- Death,
- A life-threatening AE: An AE or suspected adverse reaction is considered "life-threatening" if, in the view of either the PI or Sponsor, its occurrence places the patient or subject at immediate risk of death. It does not include an AE or suspected adverse reaction 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 hospitalizations may be considered serious when, based upon appropriate medical judgment they may jeopardize the patient or 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.

Suspected adverse reaction means any AE for which there is a reasonable possibility that the drug caused the AE. "Reasonable possibility" means there is evidence to suggest a causal relationship between the drug and the adverse event. Suspected adverse reaction implies a lesser degree of certainty about causality than adverse reaction, which means any adverse event caused by a drug.

An AE or suspected adverse reaction is considered "unexpected" if

- It is not listed in the IB or is not listed at the specificity or severity that has been observed; or, if an IB is not required or available,
- Is not consistent with the risk information described in the general investigational plan or elsewhere in the current application as amended. For example, under this definition, hepatic necrosis would be unexpected (by virtue of greater severity) if the IB referred only to elevated hepatic enzymes or hepatitis. Similarly, cerebral thromboembolism and cerebral vasculitis would be unexpected (by virtue of greater specificity) if the IB listed only cerebral vascular accidents.
- "Unexpected" as used in this definition, also refers to AE or suspected adverse reactions that are mentioned in the IB as occurring with a class of drugs or as anticipated from the pharmacological properties of the drug, but are not specifically mentioned as occurring with the particular drug under investigation.

As of the date of this protocol, there are no expected events listed in the current version of the IB; therefore, all AEs will be considered unexpected until such a time that the reference safety information in the IB is updated with any identified, expected events.

8.2. Documenting Adverse Events

8.2.1. Timeframe for Collection and Follow-up of AEs/SAEs

All AEs/SAEs will be collected from the time of the first study drug administration until the subject has completed the EOS visit and been discontinued from the study. This includes subjects who discontinue early. Events considered related to study drug will be followed as noted:

- AEs that are related to study drug and continue beyond the normal collection period (i.e., are ongoing at the time a subject exits the study) will be followed until resolution, return to baseline or until stabilized with sequelae for a maximum of 30 days following discontinuation. After 30 days, the AE will be closed, and the outcome noted (see Table 12).
- SAEs that are related to study drug and continue beyond the normal collection period

(i.e., are ongoing at the time a subject exits the study) will be followed until resolution, return to baseline or until stabilized with sequelae.

- Serious AEs that are reported to the site within 30 days after the subject has been discontinued from the study (i.e., completed the EOS visit) will be recorded. Those that are considered related to study drug will be followed as noted in the bullet above.

Note that all events which occurred prior to dosing with study drug should be recorded as medical history unless the event is directly related to study procedures.

8.2.2. Recording of Adverse Events/Serious Adverse Events

AEs/SAEs must be recorded in the CRF as indicated in the CRF completion instructions. Information to be collected includes event description, time of onset, clinician's assessment of severity (Section 8.2.3), relationship to study drug (Section 8.2.4), outcome (Section 8.2.5), and time of resolution/stabilization of the event. All AEs occurring while on study must be documented appropriately regardless of relationship and will be followed to adequate resolution as described above (Section 8.2.1). All SAEs will be recorded as noted above; SAEs reported to the site within 30 days following the EOS visit will also be recorded. All SAEs must be entered onto the SAE form and reported as discussed below (Section 8.3.1).

If an AE changes in severity, the highest severity will be recorded in the CRF. AEs characterized as intermittent require documentation of onset and duration of each episode.

8.2.3. Assessing Severity of Adverse Events

All AEs/SAEs will be assessed by the PI or those with the training and authority to make a medical judgment. AEs/SAEs will be graded according to the DMID Toxicity Grading Scale. For any AEs not specifically listed in the tables, the following guidelines should be used to grade severity:

- **Mild** (Grade 1); asymptomatic or mild symptoms; clinical or diagnostic observations only; intervention not indicated.
- **Moderate** (Grade 2); minimal, local or noninvasive intervention indicated; limiting age-appropriate instrumental activities of daily living.
- **Severe** (Grade 3); medically significant but not immediately life-threatening; hospitalization or prolongation of hospitalization indicated; disabling; limiting self-care activities of daily living.
    - **Life-threatening**; life-threatening consequences; urgent intervention indicated.
    - **Death**; death related to AE.

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

8.2.4. Relationship to Study Drug

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

- **Definite –** The AE is clearly related to the study drug**.**
- **Probable –** The AE is likely related to the study drug.
- **Possible –** The AE may be related to the study drug.
- **Unlikely –** The AE is doubtfully related to the study drug.
- **Unrelated –** The AE is clearly NOT related to the study drug.

8.2.5. Classifying Adverse Event Outcome

All AEs/SAEs in the study must be assigned an outcome by site staff. The outcome will be included on the AE CRF. Possible outcomes are shown below:

Table 12: Adverse Event Outcomes

Outcome	Description
Recovered / Resolved	AE resolved with no residual signs or symptoms; an event is considered resolved if it returns to baseline (pretreatment) values.
Recovered / Resolved with sequelae	AE stabilized but residual signs or symptoms remain; this includes stabilization of an event/condition with the expectation that it will remain chronic.
Not Recovered / Not Resolved	AE remains ongoing AND no or only minimal improvement has occurred.
Ongoing	AE has not yet resolved, but continues to improve/resolve and complete resolution is expected over time.
Fatal	Outcome of the AE is death.
Unknown	AE outcome is not known; usually because the subject has been Lost-to-Follow-up.

8.3. Reporting Procedures

8.3.1. Serious Adverse Events

The PI or clinical site personnel should notify Covance Drug Safety Services (DSS) of all SAEs, regardless of relationship to the investigational drug, within 24 hours of clinical site personnel becoming aware of the event. The PI (or designee) will provide the initial notification by sending a completed "SAE Notification Form," which must include the PI's (or designee's) assessment of the relationship of the event to investigational drug and must be signed by the PI. Follow-up information, or new information regarding an ongoing SAE, must be provided promptly to Covance DSS.

The responsibilities of Covance DSS include the following:

- Prepare an AE reporting plan prior to the start of the study. Where this plan differs from the applicable site standard operating procedure on SAE reporting, the AE reporting plan will always take precedence.

- Receive and review SAE report forms from the site and inform the Sponsor of the SAE within 1 working day of the initial notification to DSS. Drug Safety Services will delete any information from the SAE report forms that may identify the subject.

- Write case narratives and enter the case into Covance's safety database as defined in the AE reporting plan.

- Produce appropriate reports of all Suspected Unexpected Serious Adverse Reactions and forward to the EC, Medicines and Healthcare Products Regulatory Agency, PI's (or designee's), and the Sponsor within the timeframes stipulated in the Clinical Trials Directive Guideline (ENTR/CT 3).

The responsibility for reporting SAEs will be transferred to the Sponsor 28 days after the end of the study.

8.3.2. Pregnancy

Subjects in this study must be of non-childbearing potential. However, should a pregnancy occur any time from informed consent to the EOS visit, subjects must immediately report the event to the clinical site which in turn must immediately report the pregnancy to the Sponsor or their designee. The subject will be followed until the end of the pregnancy. A separate ICF will be used for consenting for follow-up pregnancy activities. Pregnancy will not be considered an AE unless deemed likely related to study drug. Pregnancy will not be considered an SAE unless there is an associated SAE. A spontaneous abortion (miscarriage) or abnormal outcome (including congenital anomalies) will be reported as an SAE.

8.4. Unmasking Treatment Assignment

The following controls will be employed to maintain the double-blind status of the study:

- Placebo will be identical in appearance to the EIDD-2801.
- The PI and other members of staff involved with the study will remain blinded to the treatment randomization code in P1 and P3.
- Interim bioanalytical data will be provided in a blinded manner.

To maintain the blind, the PI will be provided with a sealed randomization code for each subject in P1 and P3, containing details of their treatment. These individual sealed envelopes will be kept in a limited access area that is accessible 24 hours a day. In order to manage subject safety or to support dose-escalation decisions (in the event of possibly treatment-related SAEs or severe AEs), the decision to unblind resides solely with the PI. Whenever possible and providing it does not interfere with or delay any decision in the best interest of the subject, the PI will discuss the intended code-break with the Sponsor. If it becomes necessary to break the code during the study, the date, time, and reason will be recorded in the subject's source data and on the individual envelope and will be witnessed by a second person.

The pharmacy and bioanalytical lab will have access to the treatment randomization and will be unblinded. Pharmacokinetic personnel may be unblinded to perform interim PK analysis and to ensure that PK data are provided in a blinded manner for dose-escalation decisions.

9. DOSE-ESCALATION AND HALTING RULES

9.1. Guidelines for Dose-Escalation

Doses will be administered in an escalating manner following satisfactory review by the Sponsor and PI of the blinded safety and tolerability data up to 72 hours post final dose. [redacted] As PK data become available, these data will be reviewed and may be used to assist in dose selection.

Doses may be reduced and may be lower than the starting dose. There will be a minimum of 4 days between dose escalations to allow sufficient time for an adequate safety review. [redacted]

Integrated data from all applicable sites will be used to make dose-escalation decisions.

Dose-escalation in P1 and P3 will only occur if data from a minimum of 6 subjects (across all sites) have been reviewed from the previous lower dose group, such that data from a minimum of 4 subjects (across all sites) who have received EIDD-2801 will be used to make the dose-escalation decision.

The justification for this is as follows:

- The study treatment is of a known pharmacological class for which the on-target effects in humans are well characterized. Based upon nonclinical data, no clinically important off target effects are expected within the proposed dose range.

- A minimum of 4 subjects receiving the active drug is considered sufficient to characterize the safety profile of EIDD-2801.

Between each dose-escalation, the PI will review all available data in a blinded manner to ensure it is safe to proceed with the planned dose-escalation. The results from all available safety assessments will be sent to the Sponsor prior to the start of each successive group/treatment period. Any clinically significant results will be discussed with the Sponsor before dose-escalation continues. Interim PK data may also be reviewed in terms of dose-escalation and to confirm that the study design remains appropriate. In the event of a disagreement between Sponsor and PI on the dose-escalation decision, the most conservative decision will be upheld.

Dose levels will not increase by more than 3-fold for predicted nonpharmacologically active dose levels and by 2-fold for predicted pharmacologically active dose levels. [redacted]

[redacted]

Dose levels in P3 may be repeated for collection of PBMCs, providing that the dose level did not meet the dose-escalation halting criteria.

9.2. Dose-Escalation Halting Rules

For Group 1 in P1, sentinel dosing will occur whereby 2 subjects (1 active and 1 PBO) will be dosed on 1 day and, providing no safety concerns arise, the remaining 6 subjects will be dosed after at least 24 hours.

The study will be halted (i.e., no further dosing will occur) if one or more subjects experience an SAE that is considered to be related to EIDD-2801 or 2 or more subjects in the same group experience severe AEs that are considered to be related to EIDD-2801. If, following an internal safety review, the Sponsor deems it appropriate to restart the study, this can be done following approval of a substantial protocol amendment.

Additionally, if 2 or more subjects experience a reduction of platelets greater than 50% over baseline levels or a reduction in lymphocytes (absolute count) to Grade 3/severe levels without an alternate cause, the PI and Sponsor will review the data. If, after review of unblinded data, it is determined that these events are probably related to study drug, no further dose escalations will occur. In this case, additional cohorts may be enrolled at a lower dose.

Dose-escalation will stop if clinically relevant signs or symptoms of similar nature occur in 2 or more subjects in a group that, in the opinion of the PI, warrant stopping of dose-escalation.

10. STATISTICAL CONSIDERATIONS

10.1. General Considerations

All summaries will be provided by study part and treatment. Continuous variables will be summarized using descriptive statistics including number of observations (n), mean, standard deviation, minimum (min), median (med), and maximum (max). Categorical variables will be summarized using frequency counts and percentages. Note that PBO data from each dose level in Parts 1 and 3 will be combined into one PBO group.

No missing data imputation will be performed.

Subject listings will be provided for all the data collected during the study period.

Specific information about the statistical analysis will be provided in a SAP that will be reviewed and approved by the Sponsor and will be finalized before final database lock. If there is a discrepancy between the methods described in the protocol and final approved SAP, the SAP will take precedence.

10.2. Sample Size Considerations

No formal sample size calculation was conducted. The sample size of 8 per cohort (6 active: 2 PBO) for the SAD and MAD cohorts is considered adequate for a Phase 1 FIH study. The sample size of 10 subjects (all administered EIDD-2801) is in accordance with FDA guidelines for sample size in FE studies.

10.3. Analysis Populations

Safety Population: All subjects who receive at least one dose of study drug.

Pharmacokinetic Population: All subjects receiving study drug who comply sufficiently with protocol requirements (i.e., no major protocol violations) and for whom a sufficient number of analyzable PK samples have been obtained to permit the determination of the PK profile for EIDD-2801/EIDD-1931 and the statistical analysis of the results. Subjects who experience an AE of vomiting before 2× the median t_{max} of the group may be excluded from the PK population.

10.4. Analysis of Safety Data

All safety analyses will be performed on the Safety Population as defined in Section 10.3. Safety will be assessed on the basis of AEs, clinical laboratory data, vital signs, ECG parameters, and PEs.

10.4.1. Extent of Exposure

Dosing data will be listed by study part.

10.4.2. Adverse Events

Adverse events will be coded using Medical Dictionary for Regulatory Activities (MedDRA) preferred term and system organ classification. Any events reported after the initiation of study treatment and through the EOS are defined as treatment-emergent. The occurrence of treatment-emergent AEs will be summarized using MedDRA preferred terms, system organ classifications, and severity. Separate summaries of treatment-emergent SAEs and AEs considered related to study treatment and AEs leading to study treatment discontinuation will be generated. All AEs will be listed for individual subjects showing both verbatim and preferred terms.

10.4.3. Clinical Laboratory Results

Laboratory abnormalities will be graded according to the DMID Toxicity Grading Scale. Any graded abnormality that occurs following the initiation of study treatment and represents at least a one-grade increase from the baseline assessment is defined as treatment-emergent. The number and percentage of subjects experiencing treatment-emergent graded toxicities will be summarized. Raw values and mean changes from baseline in clinical laboratory measures will be summarized.

Listings of the clinical laboratory test results will be provided. Abnormal laboratory values will be flagged in the listings.

10.4.4. Other Parameters

Individual data for ECG parameters and vital sign measurements will be listed by subject and time point and summarized for each treatment. Individual data for PE will be listed by subject and time point.

Concomitant medications will be coded using the World Health Organization-Drug Dictionary (WHO-DD) and summarized.

10.5. Analysis of Pharmacokinetic Data

Pharmacokinetic analysis as defined in the SAP will be conducted using the PK population defined in Section 10.3. In the event of discrepancies between analyses described in the SAP and this clinical study protocol, the SAP will supersede the protocol.

- All samples will be analyzed and all concentrations listed.
- Descriptive statistics will be performed for all time points available, with the exclusion of subjects who had any significant protocol deviation.
- Pharmacokinetic parameters will be derived where possible for all subjects. Data from subjects with incomplete profiles (missed blood draws, lost samples, samples unable to be quantified) may be used if PK parameters can be estimated using the remaining data points.

- Descriptive statistics will be performed on all parameters available, and any missing parameters will be flagged.

Plasma concentration data for EIDD-2801 and EIDD-1931 will be listed for individual subjects and summarized by study part and treatment. Individual and mean plasma concentration versus time plots for EIDD-2801 and EIDD-1931 will be provided. Urine concentration data for EIDD-2801 and EIDD-1931 will be listed.

Plasma PK parameters of EIDD-2801 and EIDD-1931 for each subject will be estimated over the sampling interval using noncompartmental analysis and summarized by study part and treatment using descriptive statistics. Actual blood sampling times will be used for plasma PK analysis.

Urine PK parameters of EIDD-2801 and EIDD-1931 will also be analyzed and summarized when possible. The PK parameters that will be estimated are listed in the table below.

Table 13: Pharmacokinetic Parameters

Plasma PK Parameter	Description
AUC_{last}	The area under the plasma concentration-time curve, from time 0 to the last measurable non-zero concentration, as calculated by the linear up/log down trapezoidal method (P1 and P2 only).
$AUC_{0\text{-}inf}$	The area under the plasma concentration-time curve from time 0 extrapolated to infinity. $AUC_{0\text{-}inf}$ is calculated as the sum of AUC_{last} plus the ratio of the last measurable plasma concentration to the elimination rate constant (λz) (P1, P2, and Day 1 dose of P3).
C_{max}	Maximum observed concentration.
t_{max}	Time to reach C_{max}. If the maximum value occurs at more than one time point, t_{max} is defined as the first time point with this value.
λz	Apparent terminal elimination rate constant; represents the fraction of medication eliminated per unit time.
t½	Apparent terminal elimination half-life of medication, calculated as 0.693/λz.
Vz/F	Apparent volume of distribution (EIDD-2801 only).
CL/F	Apparent oral drug clearance (EIDD-2801 only).
C_{trough}	Trough concentration (P3 only).
AUC_{τ}	The area under the plasma concentration-time curve during a dosing interval (P3 only).
$RA_{AUC\tau}$	Observed accumulation ratio based on AUC_{τ} (P3 only).
RA_{Cmax}	Observed accumulation ratio based on C_{max} (P3 only).
Urine PK Parameter	**Description**
A_e	Amount excreted in urine over the sampling interval.
F_e	Fraction of drug excreted in the urine over sampling interval.
CL_R	Renal clearance, $CL_R = A_e/AUC$ where both A_e and AUC are determined over co-incident time ranges after dosing.

Additional PK parameters may be analyzed as appropriate.

10.5.1. Statistical Analysis of Pharmacokinetic Data

10.5.1.1. Dose Proportionality

In P1, dose-proportionality for PK parameters of EIDD-2801 and EIDD-1931 ($AUC_{0\text{-}inf}$, AUC_{last}, and C_{max}) will be assessed by the power model. The slope and the associated 90% CI based on the power model will be reported.

In Part 3 on Day 6 dose-proportionality for PK parameters of EIDD-2801 and EIDD-1931 (AUC_{τ}, and C_{max}) will be assessed by the power model. The slope and the associated 90% CI based on the power model will be reported.

10.5.1.2. Food-Effect

To assess the effect of food on the PK of EIDD-2801 and EIDD-1931, natural log transformed PK parameters from P2 ($AUC_{0\text{-}inf}$, AUC_{last}, and C_{max}) will be analyzed using a mixed-effects model with fixed-effect terms for treatment (with or without food), period, and sequence and with subject within sequence as a random effect. Point estimates and their associated 90% CIs for the natural log-transformed PK parameters for the difference between EIDD-2801 administered under fed relative to fasted conditions will be constructed. The point estimates and interval estimates will be back transformed to give estimates of the ratio and 90% CI for the geometric least squares mean ratio for fed/fasted to assess the effect of food.

10.6. Peripheral Blood Mononuclear Cells

Peripheral blood mononuclear cellss will be collected from subjects following each dose in P2. Depending upon ongoing review of the data, PBMCs may be collected from subjects in P3; the collection of these samples may be omitted from some cohorts in P3.

The PBMCs will be stored for analysis of EIDD-2061 levels at some point in the future and reported separately.

10.7. Interim Analyses

There are no formal interim analyses planned for this study. However, interim analyses may be implemented at the discretion of the Sponsor or health authority request. In addition, data from P1 cohorts may be locked, unblinded, and analyzed prior to conducting P2 and P3.

11. STUDY ADMINISTRATION

11.1. Regulatory and Ethical Considerations

11.2. Ethical Standard

This study will be conducted in accordance with the protocol and with the following:

- Consensus ethical principles derived from international guidelines including the Declaration of Helsinki and Council for International Organizations of Medical Sciences International Ethical Guidelines.
- Applicable ICH GCP Guidelines.
- United States CRFs applicable to clinical studies (45 CFR Part 46; 21 CFR Part 50, 21 CFR Part 56, and 21 CFR Part 312).
- Applicable laws and regulations.

11.2.1. Ethics Committee Approval

The PI (or designee) must ensure that all required study-specific documents and/or information are submitted to the EC for review and approval as appropriate including but not limited to:

- the protocol and any future protocol amendments
- ICF and any other documents (electronic or paper) given to the subject
- IB

Any substantial protocol amendments, likely to affect the safety of the subjects or the conduct of the study, will require EC and regulatory authority (as locally required) approval before implementation of changes made to the study design, except for changes necessary to eliminate an immediate hazard to study subjects or any nonsubstantial changes, as defined by regulatory requirements.

11.2.2. Informed Consent

Informed consent is a process that is initiated prior to the individual's agreeing to participate in the study and continuing throughout the individual's study participation. Extensive discussion of risks and possible benefits of this therapy will be provided to the subjects and, if applicable, their families. Consent forms describing in detail the study interventions/products, study procedures, and risks are given to the subject and written documentation of informed consent is required prior to starting any screening procedures or intervention/administering study product. Consent forms will be EC-approved and the subject will be asked to read and review the document.

Upon reviewing the document, the PI (or designee) will explain the research study to the subject and answer any questions that may arise. The subjects will sign the ICF prior to any procedures being done specifically for the study. The subjects should have the opportunity to discuss the study with their surrogates or think about it prior to agreeing to participate. The subjects may withdraw consent at any time throughout the course of the trial. A copy of the ICF will be given to the subjects for their records. The rights and welfare of the subjects will be protected by emphasizing to them that the quality of their medical care will not be adversely affected if they decline to participate in this study.

11.3. Financing and Insurance

Financing and insurance will be addressed in a separate agreement.

11.4. Source Documentation and Access

The site will maintain appropriate medical and research records in compliance with ICH E6, Section 4.9 and regulatory and institutional requirements for the protection of confidentiality of subjects. The site will permit authorized representatives of the Sponsor, its designees, and appropriate regulatory agencies to examine (and when required by applicable law, to copy) clinical records for the purposes of quality assurance reviews, audits, and evaluation of the study safety and progress. These representatives will be permitted access to all source data and source documents, which include, but are not limited to, clinical and office charts, laboratory notes, memoranda, subjects' memory aid or evaluation checklists, pharmacy dispensing records, recorded data from automated instruments, copies or transcriptions certified after verification as being accurate and complete, photographic negatives, microfilm or magnetic media, x-rays, and subject files and records kept at the pharmacy, at the laboratory, and medico-technical departments involved in the clinical trial.

11.5. Data Collection and Record Keeping

11.5.1. Data Collection

Data collection and data entry are the responsibility of the clinical trial staff at the site. The PI is responsible for ensuring the accuracy, completeness, legibility, and timeliness of the data reported. All source documents should be completed in a neat, legible manner to ensure accurate interpretation of data.

A CRF must be completed for every subject who signs the ICF and has at least one protocol-specified assessment conducted. The CRF must be completed and processed according to the CRF guidelines and the SOPs of the site. All data should be entered into the CRF, where possible, within 3 days after each visit for any one subject. After the subject has completed the study, the PI must review and sign the signature page of the CRF indicating that he has reviewed the completed CRF and pertinent clinical data for that subject and that, to the best of his knowledge, all data recorded in the CRF accurately reflects the subject's clinical performance in the study

11.5.2. Study Records Retention

Study documents should be retained for a minimum of 5 years after the end of the study. These documents should be retained for a longer period; however, if required by local regulations. No records will be destroyed without the written consent of the Sponsor, if applicable. It is the responsibility of the Sponsor to inform the PI when these documents no longer need to be retained.

11.5.3. Protocol Deviations

A protocol deviation is any noncompliance with the protocol or study procedures detailed in the laboratory or pharmacy manuals. The noncompliance may have been the result of action by the PI, site staff, or subject. All deviations should be handled in accordance with site SOPs.

It is the responsibility of the site to use continuous vigilance to identify and report deviations within 5 working days of identification of the protocol deviation, or within 5 working days of the scheduled protocol-required activity.

11.6. Clinical Monitoring

Site monitoring is conducted to ensure that the human subjects' protections, study and laboratory procedures, study intervention administration, and data collection processes are of high quality and meet Sponsor, ICH/GCP guidelines and applicable regulations, and that this trial is conducted in accordance with the protocol, protocol-specific MOP and applicable Sponsor SOPs. Experienced clinical monitors of the Sponsor or its designee will conduct site-monitoring visits as detailed in the clinical monitoring plan.

Site visits will be made at standard intervals as defined by the Sponsor and may be made more frequently as directed by the Sponsor. Monitoring visits will include, but are not limited to, review of regulatory files, accountability records, CRFs, ICFs, medical and laboratory reports, and protocol and GCP compliance. Clinical monitors will have access to each participating site, study personnel, and all study documentation according to the site monitoring plan. Clinical monitors will meet with the site PI to discuss any problems and actions to be taken and will document site visit findings and discussions. As data are entered into a CRF, data will be monitored to ensure accuracy as well as adherence to data entry instructions provided in the CRF guidelines.

11.7. Quality Control and Quality Assurance

A quality management plan will be put in place for this study. The site is responsible for conducting routine quality assurance and quality control activities to internally monitor study progress and protocol compliance. The PI will provide direct access to all study-related sites, source data/data collection forms, and reports for the purpose of monitoring and auditing by the Sponsor, and inspection by local and regulatory authorities. The PI will ensure all study personnel are appropriately trained and applicable documentations are maintained on site.

11.8. Study Termination and Closure Procedures

11.8.1. Study Termination

This study may be temporarily suspended or prematurely terminated if there is sufficient reasonable cause. Written notification, documenting the reason for study suspension or termination, will be provided to the site. If the study is terminated or suspended, the PI (or designee) will inform study participants and the EC. The Sponsor will notify appropriate regulatory authorities. The Sponsor will provide the reason(s) for the termination or suspension. Study participants will be contacted, as applicable, and be informed of changes to study visit schedule.

If suspended, the study may resume once issues that caused suspension of the study are resolved.

11.8.2. Termination Procedures

If the study is prematurely terminated, then the site must return all appropriate study data, resolve all data queries, complete final drug accountability, return any study drug remaining on site, and ship all biological samples (including PK and PBMCs) to the laboratory designated by the Sponsor. The PI (or designee) must notify the EC of study termination.

11.9. Information Disclosure

11.9.1. Confidentiality

Subject confidentiality and privacy is strictly held in trust by the PI, site staff, and the Sponsor(s) and their agents. The study protocol, documentation, data, and all other information generated will be held in strict confidence. No information concerning the study or the data will be released to any unauthorized third party without prior written approval of the Sponsor.

The study monitor or other authorized representatives of the Sponsor or regulatory agencies may inspect all documents and records required to be maintained by the PI, including but not limited to, medical records (office, clinic, or hospital) and pharmacy records for the participants in this study. The clinical study site will permit access to such records.

The study participant's contact information will be securely stored at the clinical site for internal use during the study. At the end of the study, all records will continue to be kept in a secure location for as long a period as dictated by the Sponsor, site, or regulatory requirements.

Study participant research data will be transmitted to and stored securely by the Sponsor's designated data center. This will not include the participant's contact or identifying information. Individual participants and their research data will be identified by a unique study identification number. The study data entry and study management systems used by clinical sites will be secured and password-protected. At the end of the study, all study databases will be de-identified and archived at a secure location.

11.9.2. Clinicaltrials.gov

This clinical study will be registered on clinicaltrials.gov as required.

11.9.3. Publication Policy

All information generated from this study is the proprietary property of the Sponsor. It is the intent of the Sponsor to publish the results of the study in their entirety as deemed appropriate.

12. REFERENCES

Agostini ML, Pruijssers AJ, Chappell JD, Gribble J, Lu X, Andres EL, et al. Small-Molecule Antiviral β-d-N4-Hydroxycytidine Inhibits a Proofreading-Intact Coronavirus with a High Genetic Barrier to Resistance. J Virol. 2019 Dec 15;93(24).

Flavell RA, Sabo DL, Bandle EF, Weissmann C. Site-directed mutagenesis: generation of an extracistronic mutation in bacteriophage Q beta RNA. J Mol Biol. 1974 Oct 25;89(2):255–72.

Hernandez-Santiago BI, Beltran T, Stuyver L, Chu CK, Schinazi RF. Metabolism of the anti-hepatitis C virus nucleoside beta-D-N4-hydroxycytidine in different liver cells. Antimicrob Agents Chemother. 2004 Dec;48(12):4636–42.

Mullins JI, Heath L, Hughes JP, Li D, Essigmann JM, Harris KS, et al. Mutations based on viral decay acceleration in the HIV-1 genomes of a clinical population treated with the mutagenic nucleoside KP1461. Retrovirology. 2011 Oct 3;8(Suppl 2):P84.

Painter GR, Bowen RA, Bluemling GR, DeBergh J, Edpuganti V, Gruddanti PR, et al. The prophylactic and therapeutic activity of a broadly active ribonucleoside analog in a murine model of intranasal venezuelan equine encephalitis virus infection. Antiviral Res. 2019;171:104597.

Sticher ZM, Lu G, Mitchell DG, Marlow J, Moellering L, Bluemling GR, et al. Analysis of the Potential for N4-Hydroxycytidine To Inhibit Mitochondrial Replication and Function. Antimicrob Agents Chemother. 2020 27;64(2).

Stuyver LJ, Whitaker T, McBrayer TR, Hernandez-Santiago BI, Lostia S, Tharnish PM, et al. Ribonucleoside analogue that blocks replication of bovine viral diarrhea and hepatitis C viruses in culture. Antimicrob Agents Chemother. 2003 Jan;47(1):244–54.

Toots M, Yoon J-J, Cox RM, Hart M, Sticher ZM, Makhsous N, et al. Characterization of orally efficacious influenza drug with high resistance barrier in ferrets and human airway epithelia. Sci Transl Med. 2019 Oct 23;11(515):eaax5866.

Urakova N, Kuznetsova V, Crossman DK, Sokratian A, Guthrie DB, Kolykhalov AA, et al. β-d-N4-Hydroxycytidine Is a Potent Anti-alphavirus Compound That Induces a High Level of Mutations in the Viral Genome. J Virol. 2018 01;92(3).

Yoon J-J, Toots M, Lee S, Lee M-E, Ludeke B, Luczo JM, et al. Orally Efficacious Broad-Spectrum Ribonucleoside Analog Inhibitor of Influenza and Respiratory Syncytial Viruses. Antimicrob Agents Chemother. 2018;62(8).

13. APPENDICES

Appendix 1: Time and Events Schedule

Parts 1 and 2

Visit	Screening (Days -28 to -2)	Day -1	Day 1	Day 2 (24 hr)	Day 3 (48 hr)	Day 4 (72 hr)	Day 9	Day 15 / EOS[1] (non-FE cohorts)
Food-Effect Cohort		**Day 14**	**Day 15**	**Day 16 (24 hr)**	**Day 17 (48 hr)**	**Day 18 (72 hr)**	**Day 23**	**Day 30 / EOS**
ICF; Demography	x							
I/E; Medical history	x	x						
Physical examination[2]	x	x		x		x	x	x
Qualifying laboratory analyses[3]	x	x						
Drug screening and pregnancy test[4]	x	x					x	x
Height, weight (BMI)	x							
Clinic confinement[5]		x	x	x	x	x		
Non-residential visit	x						x	x
Clinical chemistry and urinalysis[6]	x	x		x		x	x	x
Hematology[7]	x	x		x	x	x	x	x
PBMC collection (FE cohort ONLY)			x[8]	x[8]				
ECG	x		x[9]			x		x
Vital signs	x	x	x[10]	x	x	x	x	x
Administer study drug			x					
Plasma PK sample collection			x[11]	x[12]	x[12]	x[12]		
Urine PK sample collection			x[13]	x[14]	x[14]			
Adverse events	x[15]	x[15]	x[15]	x	x	x	x	x
Prior and/or Concomitant medications	x	x	x	x	x	x	x	x

Abbreviations: BMI (body mass index); EOS (End of Study); FE (food-effect); ICF (Informed Consent Form); I/E (Inclusion/Exclusion Criteria); ECG (electrocardiogram); PBMC (peripheral blood mononuclear cell); PK (pharmacokinetic).

[1] On Day 15, conduct the EOS visit for all subjects except those in the FE cohort.

[2] A complete PE will be performed at Screening and/or Check-in, and EOS visits; on all other days, a limited PE will be conducted targeting any change from the previous status or any body system deemed pertinent to that subject by the PI (or designee).

[3] Laboratory analyses to qualify for study entry include: total white cell count or absolute lymphocyte count (values must be in the normal range), hemoglobin or platelet levels (values must not be below the lower limit of normal), and ALT/SGPT, alkaline phosphatase, AST/SGOT (values must not be above the upper limit of normal), plus virology (HIV/HCV/HBV; tested at Screening only; all must be negative), and FSH (if required).

[4] Drug screening (urine drug screen, UDS), including a cotinine test, should be conducted at Screening, Day -1, Days 9 and 23, and at EOS. Pregnancy tests should be conducted in serum at Screening and in urine at all other times (i.e., Days -1, 9, and 23, and EOS). A positive urine pregnancy test will be confirmed with a serum pregnancy test. An alcohol breath test should be conducted at Screening and Day -1.

[5] Participants should be admitted to the clinic following review of final I/E criteria on Day -1 and discharged from the clinic following completion of study specific assessments on Day 4. Subjects in the FE cohort should be readmitted to the clinic on Study Day 14.

[6] Safety laboratory assessments include clinical chemistry and urinalysis. Baseline samples will be collected after clinic admission on Day -1.

[7] Hematology values include, but are not limited to, RBC, WBC with differential and platelets. PT/PTT and INR assessed only at

Screening.

[8] PBMCs will be collected 2 hr postdose on Day 1/15, and predose and 8 hrs postdose (on Day 16). Depending on ongoing review of the data, timepoints may be modified or removed, and up to 2 additional samples may be collected.

[9] The baseline ECG should be conducted prior to dosing on Day 1/15.

[10] Vital signs include pulse rate, respiratory rate, blood pressure, and body temperature; on Day 1/15, VS should be taken predose, and at 2, 4, 8 and 12 hr postdose.

[11] PK samples should be collected predose and at 0.5, 1, 1.5, 2, 2.5, 3, 4, 6, 9, 12, and 15 hr postdose on Day 1/15. Depending on ongoing review of the PK data, timepoints may be modified or removed, and up to 3 additional samples (in each period for the food-effect) may be added.

[12] PK samples should be collected at 24, 36, 48, and 72 hr postdose on Days 2/16, 3/17, 4/18 respectively. Depending on ongoing review of the PK data, timepoints may be modified or removed, and up to 3 additional samples (in each period for the food-effect) may be added.

[13] Urine samples for PK analysis should be collected predose (spot collection), and from 0 to 4, 4 to 8, 8 to 12, and 12 to 24 hr postdose.

[14] Urine samples for PK analysis should be collected from 24 to 36 and 36 to 48 hr postdose.

[15] Adverse events occurring prior to study drug administration should be recorded as medical history unless the event is directly related to a study specified procedure.

Part 3

Visit	Screening (Days -28 to -2)	Day -1	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7	Day 8	Day 9	Day 14	Day 20 /EOS
ICF; Demography	x												
I/E; Medical history	x	x											
Physical examination[1]	x	x		x					x			x	x
Qualifying laboratory analyses[2]	x	x											
Drug screening and pregnancy test[3]	x	x										x	x
Height, weight (BMI)	x												
Clinic confinement[4]		x	x	x	x	x	x	x	x	x	x		
Non-residential visit	x											x	x
Clinical chemistry and urinalysis[5]	x	x			x			x			x		x
Hematology[6]	x	x			x			x			x	x	x
PBMC collection[7]			x					x					
ECG[8]	x		x					x			x		x
Continuous 12-lead ECG			x[9]					x[10]		x[10]			x
Vital signs[11]	x	x	x	x	x	x	x	x	x	x	x	x	x
Administer study drug[12]			x	x	x	x	x	x					
Plasma PK sample collection[13]			x			x		x	x	x	x	x	
Urine PK sample collection[14]			x					x	x	x	x		
Adverse events	x[15]	x[15]	x[15]	x	x	x	x	x	x	x	x	x	x
Prior and/or Concomitant medications	x	x	x	x	x	x	x	x	x	x	x	x	x

Abbreviations: BID (twice daily); BMI (body mass index); EOS (End of Study); ICF (Informed Consent Form); I/E (Inclusion/Exclusion Criteria); ECG (electrocardiogram); PK (pharmacokinetic).

[1] A complete PE will be performed at Screening and/or Check-in, and EOS visits; on all other days, a limited PE will be conducted targeting any change from the previous status or any body system deemed pertinent to that subject by the PI (or designee).

[2] Laboratory analyses to qualify for study entry include: total white cell count or absolute lymphocyte count (values must be in the normal range), hemoglobin or platelet levels (values must not be below the lower limit of normal), and ALT/SGPT, alkaline phosphatase, AST/SGOT (values must not be above the upper limit of normal), plus virology (HIV/HCV/HBV; tested at Screening only; all must be negative), and FSH (if required).

[3] Drug screening (urine drug screen, UDS), including cotinine, should be conducted at Screening, Day -1, Day 14, and at EOS. Pregnancy tests should be conducted in serum at all timepoints. An alcohol breath test should be conducted at Screening and Day -1.

[4] Participants should be admitted to the clinic following review of final I/E criteria on Day -1 and discharged from the clinic following completion of study specific assessments on Day 9.

[5] Safety laboratory assessments include clinical chemistry and urinalysis. Baseline samples will be collected after clinic admission on Day -1. On Days 3 and 6, the assessment will be pre-am dose. On Day 9, the assessment will be 72 hrs post-am dose on Day 6.

[6] Hematology values include, but are not limited to, RBC, WBC with differential and platelets. PT/PTT and INR assessed only at Screening.

[7] PBMCs will be collected 2 hrs postdose, relative to the first daily dose on Day 1, and pre-am dose and 2 and 8 hrs postdose relative to the first daily dose on Day 6. Depending on ongoing review of the data, timepoints may be modified or removed, and up to 2 additional samples may be collected. Additionally, PMBC collection may be omitted from some cohorts.

[8] The baseline ECG should be conducted prior to first dosing on Day 1. On Day 1, ECG should be taken predose and 2 hours post-am dose. On Day 6, ECG should be taken predose, 2 hrs, and 72 hrs (Day 9) postdose.

[9] On Day 1, monitor for 12-lead ECG recording will be worn from 2 hrs predose to 12 hrs postdose. Extraction timepoints will be 60, 45, and 30 minutes predose and at 0.5, 1, 1.5, 2, 2.5, 3, 4, 6, 9, and 12 hrs postdose.

[10] On Day 6, monitor for 12-lead ECG recording will be worn from dosing (0 hr) to 25 hrs (Day 6) postdose. Extraction timepoints will be at 0.5, 1, 1.5, 2, 2.5, 3, 4, 6, 9, 12, and 24 hrs postdose.

[11] Vital signs include pulse rate, respiratory rate, blood pressure, and body temperature. On Day 1, VS should be taken pre-am dose, and at 2, 4, and 8 hrs post-am dose. On Day 2 through 5, VS should be taken pre-am dose. On Day 6, VS should be taken predose, and 2, 4, 8, 24 (Day 7), 48 (Day 8), and 72 (Day 9) hrs postdose.

[12] Study drug should be administered BID, 12 hrs apart on Days 1 through 5. On Day 6, study drug should be administered once in the morning only (0 hr). Depending on ongoing review of the safety, tolerability, and PK data, the number of days of dosing may be reduced and the dosing frequency changed; the dosing frequency will be no less than once-daily or no greater than three-times daily. If the number of days of dosing are reduced, the timepoints for subsequent assessments and the period for which subjects are domiciled at the clinic will similarly be changed (subjects will be domiciled for 3 days post final dose).

[13] On Day 1, PK samples should be collected pre-am dose and at 0.5, 1, 1.5, 2, 2.5, 3, 4, 6, 9, and 12 hrs post-am dose. On Day 4, the PK sample should be collected pre-am dose. On Day 6, PK samples should be collected predose and at 0.5, 1, 1.5, 2, 2.5, 3, 4, 6, 9, 12, 15, 24 (Day 7), 48 (Day 8), 72 (Day 9), and 192 (Day 14) hrs postdose. Depending on ongoing review of the PK data, timepoints may be modified or removed, and up to 3 additional samples may be added.

[14] On Day 1, urine samples for PK analysis should be collected predose (spot collection), and from 0 to 4, 4 to 8, and 8 to 12 hrs postdose. On Day 6, urine samples for PK analysis should be collected predose (spot collection), and from 0 to 4, 4 to 8, 8 to 12, 12 to 24 (Day 7), 24 to 48 (Day 8), and 48 to 72 hrs (Day 9). Sampling timepoints may be modified, removed, or additional timepoints added depending upon ongoing review of the data. The 12-hour urine collection should occur prior to the second daily dose administration.

[15] Adverse events occurring prior to study drug administration should be recorded as medical history unless the event is directly related to a study specified procedure.

Appendix 2: DMID Toxicity Grading Scale

Clinical Adverse Events			
VITAL SIGNS	**Mild (Grade 1)**	**Moderate Grade 2)**	**Severe (Grade 3)**
Fever (°C) Oral temperature; no recent hot or cold beverages or smoking.	38.0 – 38.4	38.5 – 38.9	>39.0
Tachycardia - beats per minute	101 – 115	116 – 130	> 130 or ventricular dysrhythmias
Bradycardia - beats per minute	50 - 54 or 45 - 50 bpm if baseline <60 bpm	45 - 49 or 40 - 44 if baseline <60 bpm	< 45 or <40 bpm if baseline <60 bpm
Hypertension (systolic) - mm Hg [Assuming supine position, 10 min at rest conditions, not sleeping subjects, measurements on the same arm and several concordant results.]	141 - 150	151 - 160	> 160
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**
Arrythmia		asymptomatic, transient signs, no Rx required	recurrent/persistent; symptomatic Rx required
Hemorrhage, Blood Loss	Estimated blood loss ≤ 100 mL	Estimated blood loss > 100 mL, no transfusion required	Transfusion required
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 60 - 70% (of peak flow)	no normalization with bronchodilator; FEV1 <60% 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 gms/24 hours	4 - 5 loose or watery stools or 400 - 800 gms/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
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
SYSTEMIC	**Mild (Grade 1)**	**Moderate (Grade 2)**	**Severe (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
All Other conditions	**Mild (Grade 1)**	**Moderate (Grade 2)**	**Severe (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

*In addition to grading the measured local reaction at the greatest single diameter, the measurement should be recorded as a continuous variable.

**Induration/Swelling should be evaluated and graded using the functional scale as well as the actual measurement

Laboratory Adverse Events			
Blood, Serum, or Plasma *	**Mild (Grade 1)**	**Moderate (Grade 2)**	**Severe (Grade 3)**
Sodium – Hyponatremia mEq/L	132 – <LLN	130 – 131	<130
Sodium – Hypernatremia mEq/L	>ULN – 148	149 – 150	>150
Potassium – Hyperkalemia mEq/L	>ULN – 5.2	5.3 – 5.4	>5.4
Potassium – Hypokalemia mEq/L	<LLN-3.1	<3.1 – 3.0	<3.0
Glucose – Hypoglycemia mg/dL	65 – 67	55 – 64	<55
Glucose – Hyperglycemia Fasting – mg/dL	>ULN - 120	121 – 130	>130
Glucose – Hyperglycemia Random – mg/dL	140 – 159	160 – 200	>200
Blood Urea Nitrogen mg/dL	23-26	27 – 31	> 31
Creatinine – mg/dL	>ULN – 1.7	1.8 – 2.0	>2.0
Calcium – hypocalcemia mg/dL	8.0 – <LLN	7.5 – 7.9	<7.5
Calcium – hypercalcemia mg/dL	>ULN – 11.0	11.1 – 11.5	>11.5
Magnesium – hypomagnesemia mg/dL	1.3 – 1.5	1.1 – 1.2	<1.1
Phosphorus – hypophosphatemia mg/dL	2.3 – 2.5	2.0 – 2.2	<2.0
CPK – mg/dL	400-1000	1001-1500	>1500
Albumin – Hypoalbuminemia g/dL	2.8 – 3.0	2.5 – 2.7	< 2.5
Total Protein – Hypoproteinemia g/dL	5.2 – 5.4	4.8 – 5.1	< 4.8
Alkaline phosphate – U/L	132-240	241– 360	>360
AST U/L	44 – 105	106-175	>175
ALT U/L	44 – 105	106-175	>175
Bilirubin (serum total) mg/dL	1.3 - 2.0	2.1 - 2.5	> 2.5
Bilirubin – when ALT ≥105 (Hy's law)	1.3 -1.5	1.6 – 2.0	> 2.0
Amylase- U/L	200-270	271-360	>360
Lipase- U/L	176-270	271-360	>360
Hemoglobin (Female) - g/dL	11.0 – 11.5	9.5 – 10.9	< 9.5
Hemoglobin (Male) - g/dL	12.0 – 12.5	10.0 – 11.9	<10.0
WBC Increase - cell/mm3	11,001 – 15,000	15,001 – 20,000	> 20,000
WBC Decrease - cell/mm3	2,500 – 3,500	1,500 – 2,499	< 1500
Lymphocytes Decrease - cell/mm3	750 – 1,000	500 – 749	< 500
Neutrophils Decrease - cell/mm3	1,500 – 2,000	1,000 – 1,499	< 1000
Eosinophils - cell/mm3	500-750	751-1500	> 1500
Platelets Decreased - cell/mm3	120,000 – 130,000	100,000 – 119,999	<100,000
PT – seconds (prothrombin time)	> ULN-14.4	14.5 – 15.7	> 15.7
PTT – seconds (partial thromboplastin time)	>ULN-42.1	42.2– 50.0	> 50.0
Fibrinogen increase - mg/dL	>ULN – 500	501 – 600	> 600
Fibrinogen decrease - mg/dL	<LLN – 140	125 – 139	<125
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	11-50	> 50 and/or gross blood

* The laboratory values provided in the tables serve as guidelines and are dependent upon institutional normal parameters.

* Institutional normal reference ranges should be provided to demonstrate that they are appropriate.

Appendix 3: Contraception Guidance

Male subjects (regardless of fertility status) with partners of childbearing potential must use a male barrier method of contraception (i.e., male condom with spermicide) in addition to a second method of acceptable contraception from Check-in until 90 days after the EOS visit. Acceptable methods of contraception for female partners include:

- hormonal injection
- combined oral contraceptive pill or progestin/progestogen-only pill
- combined hormonal patch
- combined hormonal vaginal ring
- surgical method (bilateral tubal ligation or Essure® [hysteroscopic bilateral tubal occlusion])
- hormonal implant
- hormonal or non-hormonal intrauterine device
- OTC sponge with spermicide
- cervical cap with spermicide
- diaphragm with spermicide.

An acceptable second method of contraception for male subjects is vasectomy that has been performed at least 90 days prior to the screening visit, with verbal confirmation of surgical success.

For male subjects (even with a history of vasectomy), sexual intercourse with female partners who are pregnant or breastfeeding should be avoided unless condoms are used from the time of the first dose until 90 days after the EOS visit. Male subjects are required to refrain from donation of sperm from Check-in until 90 days after the EOS visit.

Sexual Abstinence and Same-sex Relationships

Subjects who practice true abstinence, because of the subject's lifestyle choice (i.e., the subject should not become abstinent just for the purpose of study participation), are exempt from contraceptive requirements. Periodic abstinence (eg, calendar, ovulation, symptothermal, postovulation methods) and withdrawal are not acceptable methods of contraception. If a subject who is abstinent at the time of signing the ICF becomes sexually active they must agree to use contraception as described previously.

For subjects who are exclusively in same-sex relationships, contraceptive requirements do not apply. If a subject who is in a same-sex relationship at the time of signing the ICF becomes engaged in a heterosexual relationship, they must agree to use contraception as described previously.

Title of study: A Randomized, Double-blind, Placebo-controlled, First-in-human Study Designed to Evaluate the Safety, Tolerability, and Pharmacokinetics of EIDD-2801 Following Oral Administration to Healthy Volunteers

NCT Number: NCT04392219

Document: Protocol, Version 4.1

Date: 28 April 2020

A Randomized, Double-Blind, Placebo-Controlled, First-in-Human Study Designed to Evaluate the Safety, Tolerability, and Pharmacokinetics of EIDD-2801 Following Oral Administration to Healthy Volunteers

(EIDD-2801-1001-UK)

Protocol Version	4.1 (United Kingdom)
Version Date	28 April 2020

Sponsor Team:

Ridgeback Biotherapeutics	3162 Commodore Plaza, Suite 3E Miami, FL 33133-5815 United States
Medical Officer	[redacted]
EudraCT Number	2020-001407-17

SUMMARY OF CHANGES

Changes from Version 1.1 (United Kingdom) to Version 2.1 (United Kingdom).

The primary reason for this amendment was to include Part 3, a multiple ascending dose study of EIDD-2801 in healthy subjects. Additionally, the following updates were made:

- Results of an *in vivo* micronucleus assay were added.
- The guidelines used to assess the severity of adverse events not presented in the DMID Toxicity Grading Scale were updated, specifically those that were life-threatening or led to death.
- The timepoints for collection of peripheral blood mononuclear cells in Part 2 were modified.
- Minor clarifications were made and typographical errors were corrected.

Changes from Version 2.1 (United Kingdom) to Version 3.1 (United Kingdom).

- Clarified that this study will be conducted at 1 site in the United Kingdom and up to 3 sites in the United States, and that integrated data across all sites will be used for dose-escalation decisions.
- The number of cohorts initially planned for Part 1 (single ascending dose) has been reduced from 4 to 3.
- The number of days of dosing in Part 3 (multiple ascending dose) was decreased from 7 days to 5.5 days. However, the number of days of dosing may be further reduced depending on ongoing review of the safety, tolerability, and pharmacokinetic data.
- Added a rationale for the appropriateness of a 4-day interval between dose escalations.
- Added a dose-escalation stopping criteria for reductions in platelets and lymphocytes.
- Clarified that assessments of human immunodeficiency virus, hepatitis B virus, and hepatitis C virus will only be conducted at Screening.
- Minor clarification and typographical corrections.

Changes from Version 3.1 (United Kingdom) to Version 4.1 (United Kingdom).

- The **total daily** dose to be administered in this protocol will not exceed 800 mg. A rationale for the increase in the maximum dose to be studied is provided in Section 9.2.
- The number of additional cohorts in Parts 1 and 3 has been increased to 4.

- Pregnancy tests in Part 3 will be conducted in serum at Screening and in urine at all other timepoints. A positive urine pregnancy test will be confirmed with a serum pregnancy test.

- Clarified that pregnancy, urine drug of abuse, cotinine, and alcohol breath tests will additionally be performed at Check-in to second treatment period in Part 2.

- An additional pharmacokinetic blood sampling timepoint was added in Part 1, and two additional pharmacokinetic blood sampling timepoints were added to each of Parts 2 and 3.

- Timepoints for continuous 12-lead monitoring in Part 3 were updated.

- Timepoints for collection of peripheral blood mononuclear cells in Part 2 were updated.

- The storage conditions for the study drug were updated.

- Minor clarification and typographical corrections.

STATEMENT OF COMPLIANCE

The study will be carried out in accordance with Good Clinical Practice (GCP) as required by the following:

- Consensus ethical principles derived from international guidelines including the Declaration of Helsinki and Council for International Organizations of Medical Sciences International Ethical Guidelines.
- Applicable International Conference on Harmonisation (ICH) GCP Guidelines.
- United States Code of Federal Regulations (CFR) applicable to clinical studies (45 CFR Part 46; 21 CFR Part 50, 21 CFR Part 56, and 21 CFR Part 312).
- Applicable laws and regulations.

SIGNATURE PAGE

The signature below constitutes the approval of this protocol and the attachments and provides the necessary assurances that this study will be conducted according to all stipulations of the protocol, including all statements regarding confidentiality, and according to local legal and regulatory requirements and applicable regulations and ICH guidelines.

Principal Investigator:

Signed: [redacted] Date: 29 APR 2020

Executive Medical Director
Covance Clinical Research Unit Limited

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ABBREVIATIONS

AE	adverse event
AGP	alpha1-acid glycoprotein
ALT/SGPT	alanine aminotransferase/serum glutamic pyruvic transaminase
AST/SGOT	aspartate aminotransferase/serum glutamic oxaloacetic transaminase
AUC	area under the curve
BID	twice-daily
BMI	body mass index
CDC	Centers for Disease Control and Prevention
CEM	a cell line of lymphoblastic cells derived from a child with leukemia
CFR	Code of Federal Regulations
CHKV	Chikungunya virus
CI	confidence interval
CoV	coronavirus
CRF	case report form
CTP	cytosine triphosphate cytidine triphosphate
CYP	cytochrome P450
DMID	Division of Microbiology and Infectious Diseases
DMSO	dimethylsulfoxide
DPP4	dipeptidyl peptidase 4
DRF	dose range finding
DSS	Drug Safety Services
EBOV	Ebola virus
EC	Ethics Committee
EC_{50}	Half-maximal effective concentration
ECG	electrocardiogram
EEEV	Eastern equine encephalitis virus
EIDD	Emory Institute for Drug Development
EOS	End of Study
FDA	Food and Drug Administration
FE	food-effect
FIH	first-in- human
FSH	follicle stimulating hormone
GCP	Good Clinical Practice
GI	gastrointestinal
GLP	Good Laboratory Practice

HAE	human airway epithelium
HBV	hepatitis b virus
hBTEC	human bronchial/tracheal epithelial cells
HCV	hepatitis c virus
HED	human equivalent dose
HEENT	head, eye, ear, nose and throat
HIV	human immunodeficiency virus
HPMC	hydroxypropyl methylcellulose
IAV	influenza a virus
IB	Investigator's Brochure
IBV	influenza b virus
IC_{50}	half maximal inhibitory concentration
ICF	informed consent form
ICH	International Council for Harmonisation
MAD	multiple ascending dose
MDCK	Madin-Darby Canine Kidney Cells
MedDRA	Medical Dictionary for Regulatory Activities
MERS	Middle East respiratory syndrome
%MN-PCE	percentage of micronuclei-polychromatic erythrocytes
MRSD	maximum recommended starting dose
MTD	maximum tolerated dose
NOAEL	no-observed-adverse-effect-level
NSAID	nonsteroidal anti-inflammatory drug
OTC	over-the-counter
PBMC	peripheral blood mononuclear cells
PBO	placebo
PCE:TE	polychromatic erythrocytes to total erythrocytes
PE	physical examination
PI	principal investigator
[redacted]	[redacted]
PK	pharmacokinetic
QTc(F)	QT interval corrected for heart rate (using Fridericia's formula)
RNA	ribonucleic acid
RSV	respiratory syncytial virus
SAD	single ascending dose
SAE	serious adverse event

SAP	statistical analysis plan
SARS	severe acute respiratory syndrome
SI	selectivity index
SOP	standard operating procedure
UTP	uridine 5'-triphosphate
VEEV	Venezuelan equine encephalitis virus
ZIKV	Zika virus

PROTOCOL SYNOPSIS

Sponsor: Ridgeback Biotherapeutics	
Title: A Randomized, Double-Blind, Placebo-Controlled, First-in-Human Study Designed to Evaluate the Safety, Tolerability, and Pharmacokinetics of EIDD-2801 Following Oral Administration to Healthy Volunteers (EIDD-2801-1001-UK)	
Short Title: EIDD-2801-1001-UK	**Development Phase:** Phase 1
Study Sites: This study will be conducted at 1 site in the United Kingdom and up to 3 sites in the United States.	
Description of Study Drugs, Dose and Mode of Administration: EIDD-2801 and matching placebo (PBO) will be supplied [redacted] **Part 1:** A single oral dose of EIDD-2801 or PBO will be administered to subjects enrolled in Part 1 (P1; single ascending dose [SAD]) cohorts. The starting dose in the first SAD cohort will be 50 mg. The doses will be administered in an escalating manner. Dose levels will not increase by more than 3-fold for predicted nonpharmacologically active dose levels and by 2-fold for predicted pharmacologically active dose levels. [redacted] **Part 2:** Subjects in Part 2 (P2)/food-effect (FE) will receive 2 single doses of EIDD-2801 with a 14-day (minimum) washout period between doses. The dose for the P2 cohort is planned to be 100 mg but may be adjusted based on safety, tolerability, and/or pharmacokinetic (PK) data from P1. In any case, the dose assessed in P2 will have been given previously to subjects in a P1 cohort successfully (i.e., no halting rules were met following dosing). **Part 3:** Multiple doses of EIDD-2801 or PBO will be administered to subjects enrolled in Part 3 (P3; multiple ascending dose [MAD]) cohorts. Subjects will receive twice-daily (BID) doses for 5 days, with a single dose administered on the morning of Day 6 for collection of steady-state PK samples during waking hours. The total daily dose will not exceed a dose shown to be safe and well-tolerated in P1. Doses in P3 may be administered in the fed state, following review of the PK data from P2. Additionally, depending on ongoing review of the safety, tolerability, and PK data, the number of days of dosing may be reduced and the dosing frequency may be changed; the dosing frequency will be no less than once-daily or no greater than three-times daily.	
Treatment Duration: **Part 1:** Subjects enrolled into P1/SAD cohorts will receive a single dose of study drug (EIDD-2801 or matching PBO). **Part 2:** Subjects enrolled into the P2/FE cohort will receive 2 single doses of EIDD-2801, with a washout period between doses. **Part 3:** Subjects in P3/MAD cohorts will receive study drug (EIDD-2801 or matching PBO) for 5 days BID followed by a single dose of EIDD-2801 or PBO on Day 6. Depending on ongoing review of the safety, tolerability, and PK data, the number of days of dosing may be reduced.	

Subject Duration:

Part 1: The maximum possible study duration for participants enrolled in P1/SAD cohorts will be approximately 43 days.

Part 2: The maximum possible study duration for participants enrolled in the P2/FE cohort will be approximately 58 days.

Part 3: The maximum possible study duration for participants enrolled in P3/MAD will be approximately 48 days.

Objectives and Endpoints:

Part 1: SAD Cohorts

Primary:

- Objective: To determine the safety and tolerability of single ascending doses of EIDD-2801.
  - Endpoints:
    - Results of safety evaluations including safety laboratory assessments, physical examination (PE), electrocardiograms (ECGs), vital signs, and adverse events (AEs).

Secondary:

- Objective: To define PK of EIDD-2801 and EIDD-1931 in plasma and urine following single doses administered to healthy volunteers.
  - Endpoints:
    - Plasma PK parameters, including C_{max}, t_{max}, $t_{1/2}$, CL/F, λz, Vz/F, $AUC_{0\text{-}inf}$ and AUC_{last} as appropriate
    - Urinary excretion of EIDD-2801 and EIDD-1931 following single-dose administration.

Part 2: Single-Dose Food-Effect

Primary:

- Objective: To assess the effect of food on the PK of EIDD-2801 and EIDD-1931 following a single oral dose.
  - Endpoints:
    - Plasma PK parameters, including C_{max}, t_{max}, $t_{1/2}$, CL/F, λz, Vz/F, $AUC_{0\text{-}inf}$ and AUC_{last} as appropriate
    - Urinary excretion of EIDD-2801 and EIDD-1931 following single-dose administration.

Secondary:

- Objective: To determine the safety and tolerability of single doses of EIDD-2801.
  - Endpoints:
    - Results of safety evaluations, including safety laboratory assessments, PEs, ECG, vital signs, and AEs.

Part 3: MAD Cohorts

Primary:

- Objective: To determine the safety and tolerability of multiple ascending doses of EIDD-2801.

- Endpoints:
  - Results of safety evaluations, including safety laboratory assessments, PE, ECGs, vital signs, and AEs.

Secondary:

- Objective: To define PK of EIDD-2801 and EIDD-1931 in plasma and urine following multiple doses administered to healthy volunteers.
  - Endpoints:
    - Plasma PK parameters, including C_{trough}, C_{max}, t_{max}, $t_{1/2}$, CL/F, λz, Vz/F, $AUC_{0\text{-}inf}$ (Day 1 dose only), AUC_{τ}, $RA_{AUC\tau}$, and RA_{Cmax} as appropriate
    - Urinary excretion of EIDD-2801 and EIDD-1931 following multiple-dose administration.

Exploratory:

- Objective: To collect data to assess the relationship between EIDD-2801/EIDD-1931 concentrations and QT interval corrected for heart rate (QTc).

Population:

Part 1: P1/SAD cohorts will include 8 subjects each, 2 randomized to receive PBO and 6 randomized to receive EIDD-2801. Initially, 3 SAD cohorts are planned for P1 with the option to add an additional 4 cohorts based on study results.

Part 2: Ten subjects will be enrolled into the P2/FE cohort; all subjects enrolled in the P2 cohort will receive EIDD-2801. One P2 cohort is planned for the study. If PK results obtained are equivocal, additional subjects may be enrolled into the P2 cohort at the previously tested dose or an additional P2 cohort may be added at a different dose.

Part 3: P3/MAD cohorts will include 8 subjects, 2 randomized to receive PBO and 6 randomized to receive EIDD-2801. Initially, 3 MAD cohorts are planned for P3 with the option to add an additional 4 cohorts based on study results. However, the number of cohorts may be reduced if the study objectives are met. Dose levels in P3 may be repeated for collection of peripheral blood mononuclear cells (PBMCs), providing that the dose level did not meet the dose-escalation halting criteria.

The site is strongly encouraged to ensure that women are represented in each cohort. Inclusion and exclusion criteria for study participation are as follows:

Inclusion Criteria: subjects who meet all of the criteria at Screening will be eligible for study participation

1. Male or female between the ages of 18 and 60, inclusive.
2. Female subjects must be of nonchildbearing potential. Nonchildbearing potential is defined as women who:
   - are physiologically incapable of becoming pregnant including those who are surgically sterile, via hysterectomy, bilateral salpingectomy, and/or bilateral oophorectomy by reported medical history and/or medical records. Surgical sterilization to have occurred a minimum of 6 weeks, or at the Principal Investigator's (PI's; or designee's) discretion, prior to Screening.
   - are postmenopausal (at least 45 years of age with amenorrhea for 12 months

without an alternative medical reason with confirmatory follicle stimulating hormone (FSH) levels of ≥ 40 mIU/mL. The amenorrhea should not be induced by a medical condition such as anorexia nervosa, hypothyroid disease or polycystic ovarian disease, or by extreme exercise. It should not be due to concomitant medications that may have induced the amenorrhea such as oral contraceptives, hormones, gonadotropin-releasing hormones, anti-estrogens, or selective estrogen receptor modulators).

3. Male participants must agree to the use of effective contraception for study duration, i.e., from Check-in until 90 days following the last dose of study drug.
4. Is in generally good health as determined by medical history, PE (at Screening and/or Check-in), vital signs, and other screening procedures.
5. Has a body mass index (BMI) of 18 to 30 kg/m^2.
6. Is willing and able to comply with all study procedures including providing informed consent.

Exclusion Criteria: subjects who meet any of the following criteria at Screening (unless otherwise stated) will not be eligible for participation

1. Females who are pregnant, planning to become pregnant, or breastfeeding.
2. Has a clinically relevant acute or chronic medical condition or disease of the cardiovascular, gastrointestinal (GI), hepatic, renal, endocrine, pulmonary, neurologic, or dermatologic systems as determined by the PI (or designee).
3. Has any current or historical disease or disorder of the hematological system or significant liver disease or family history of bleeding/platelet disorders.
4. Has a history of cancer (other than basal cell or squamous cell cancer of the skin), rheumatologic disease or blood dyscrasias.
5. Has a history of GI surgery or other condition that may interfere with absorption of study drug, in the option of the PI (or designee).
6. Has a history of febrile illness within the 14 days prior to the first dose of study drug.
7. Has a positive alcohol or drug screen at Screening or the Day -1 visit or has a history of alcohol or drug abuse within the past 5 years.
8. Currently uses tobacco, nicotine or tobacco products, e-cigarettes or has stopped using tobacco products within the past 3 months and/or has a positive cotinine test at Screening or Check-in.
9. Has any abnormal laboratory value of Grade 2 or higher, which is considered to be clinically significant by the PI (or designee).
10. Has a total white cell count or absolute lymphocyte count outside the normal range, or hemoglobin or platelet levels below the lower limit of normal, at Screening of Day -1.
11. Has values above the upper limit of normal for the following laboratory analytes: alanine aminotransferase (ALT/SGPT), alkaline phosphatase (serum), aspartate aminotransferase (AST/SGOT), at Screening or Day -1.
12. Positive test result for human immunodeficiency virus (HIV), hepatitis B virus (HBV), or hepatitis C virus (HCV).
13. Has an autoimmune disease, is immunosuppressed or is in any way

immunocompromised.

14. Has any of the following:
    - QT interval corrected for heart rate using Fridericia's formula (QTcF) >450 ms confirmed by repeat measurement
    - QRS duration >110 ms confirmed by repeat measurement
    - PR interval >220 ms confirmed by repeat measurement
    - findings which would make QTc measurements difficult or QTc data uninterpretable
    - history of additional risk factors for torsades de pointes (e.g., heart failure, hypokalemia, family history of long QT syndrome).
15. Except as noted, has used prescription drugs (other than hormone replacement therapy) within 14 days prior to the first dose of study drug unless, in the opinion of the PI (or designee), the drug will not interfere with study assessments. The following prescription drugs will be allowed:
    - Statins: Subjects who have been on a stable dose of statins for a minimum of 6 months and who have normal creatine kinase at Screening.
    - Antihypertensives: Subjects who have been on a stable dose of antihypertensives for a minimum of 6 months and have controlled blood pressure and no active heart disease.
16. Has received an experimental agent (vaccine, drug, biologic, device, blood product or medication) within 3 months prior to the first dose of study drug and agrees not to receive another experimental agent during the duration of this trial.
17. Has donated blood within 30 days, plasma within 2 weeks, or platelets within 6 weeks prior to Screening or refuses to agree to not donate blood until 3 months after the End of Study (EOS) visit.
18. Uses over-the-counter (OTC) medications or nutritional supplements (including, e.g., gingko biloba, aspirin at any dose, and nonsteroidal anti-inflammatory drugs [NSAIDs]) on a routine/scheduled basis, and/or is unwilling to abstain from such use for the duration of the study.
19. Is unwilling to abstain from quinine containing products including quinine water, tonic water, bitter lemon, jello shots, any quinine preparations or Cinchona tree bark preparations (e.g., herbal medications containing Cinchona tree bark).
20. Has any condition that would, in the opinion of the PI (or designee), put the subject at increased risk for participation in a clinical trial or confinement in a clinical research unit.

Retesting for Inclusion/Exclusion Criteria: In the case where a parameter is outside of the acceptable limits for enrollment, measurement of the parameter can be repeated one time if both the following are met:

- In the opinion of the PI (or designee), the parameter value does not represent a chronic condition that otherwise will preclude the volunteer from enrolling in the study; and
- In the opinion of the PI (or designee), the abnormality is not likely to recur.

General Investigational Plan: For all potential subjects, volunteers who express interest in the study will report to the clinic for informed consent. The study will be explained to the subject and the Ethics Committee (EC)-approved informed consent form (ICF) will be presented. The subject will be given the chance to review the document and ask any questions he/she may have. If, after reviewing the consent, the subject would like to participate in the study, then he/she will sign the ICF and begin screening for study entry; those satisfying all criteria will be enrolled into the study and admitted to the clinic on Day -1. Retesting will be allowed as described above.

Part 1: For P1/SAD, the first cohort will be divided into 2 groups. The first group, a sentinel group, will include 2 subjects.

On Day 1, one subject in the sentinel group will be administered PBO and one will be administered EIDD-2801. Analysis of the safety results for the sentinel group will be reviewed 24 hours postdose. If no halting criteria have been met, the remainder of the subjects in Cohort 1 (the second group) will be dosed. There will be no sentinel groups for the remaining cohorts.

Following dosing on Day 1, subjects will remain in the clinic through completion of study assessments on Day 4, returning to the clinic on Day 9 for study assessments, and Day 15 for the EOS visit.

Doses will be administered in an escalating manner following satisfactory review by the Sponsor and PI of the blinded safety and tolerability data (up to 72 hours post final dose). Integrated data from all applicable sites will be used to make dose-escalation decisions. Dose-escalation will only occur if data from a minimum of 6 subjects (across all sites) have been reviewed from the previous lower dose group, such that data from a minimum of 4 subjects (across all sites) who have received EIDD-2801 will be used to make the dose-escalation decision. Dose levels will not increase by more than 3-fold for predicted nonpharmacologically active dose levels and by 2-fold for predicted pharmacologically active dose levels. The highest total daily dose to be studied under this protocol will not exceed 800 mg.

Part 2: For P2/FE, one cohort assessing the effect of food on EIDD-2801 and EIDD-1931 PK parameters will be enrolled; it is planned that the P2 cohort will be at a dose of 100 mg EIDD-2801, although higher or lower doses may be selected based on safety and available PK data from P1.

In addition to assessing the effect of food on dosing, PBMCs will be collected from subjects following each dose; the PBMCs will be stored for analysis of EIDD-2061 levels at some point in the future.

The 10 subjects enrolled in the FE cohort will all receive EIDD-2801; subjects will be randomized to a treatment sequence (i.e., to receive drug in the fed then fasted state [Sequence 1] versus fasted then fed state [Sequence 2]). Half of the subjects in the cohort will be randomized to Sequence 1 and the other half will be randomized to Sequence 2.

Following dosing on Day 1, subjects will remain in the clinic through completion of study assessments on Day 4, returning to the clinic on Day 9 for study assessments, and Day 14 to begin the second dosing sequence on Day 15. Subjects will then be discharged from the clinic on Day 18, returning to the clinic on Day 23 for study assessments, and Day 30 for the EOS visit. There will be a 14-day (minimum) washout period between doses.

Part 3: For P3/MAD, subjects will receive BID doses on Day 1 through Day 5, inclusive, and will receive the final dose of study drug on the morning of Day 6 for collection of steady-state PK samples during waking hours. Subjects will remain domiciled at the site during the dosing

period and until Day 9, returning to the clinic on Day 14 for completion of study assessments, and Day 20 for the EOS visit.

Doses in P3 may be administered in the fed state, following review of the PK data from P2. Additionally, depending on ongoing review of the safety, tolerability, and PK data, the number of days of dosing may be reduced and the dosing frequency changed; the dosing frequency will be no less than once-daily or no greater than three-times daily. If the number of days of dosing are reduced, the timepoints for subsequent assessments and the period for which subjects are domiciled at the clinic will similarly be changed (subjects will be domiciled for 3 days post final dose).

Part 3 may run in parallel with P1, providing that the total daily dose to be administered does not exceed a dose already shown to be safe and well-tolerated in P1.

Peripheral blood mononuclear cells may be collected from subjects in P3, depending upon ongoing review of the data. The collection of these samples may be omitted from some cohorts, depending on ongoing review of the data. The PBMCs will be stored for analysis of EIDD-2061 levels at some point in the future. In addition, continuous 12-lead ECG monitoring will be performed to collect data to assess the relationship between study drug concentrations and QTc interval. All data will be archived without extraction or analysis and will not be reported in the scope of this study.

Additional parts may be added to this study, including a SAD part in elderly healthy subjects and a study of the safety, tolerability, and PK in Japanese subjects. These additional parts are not described in this protocol but may be added via a protocol amendment.

Safety Monitoring and Potential Unblinding: Safety for this study will be continually monitored by the PI and Sponsor.

Blinding: All study personnel will remain blinded to treatment assignment (i.e., EIDD-2801 or PBO) in P1 and P3, except for personnel at the bioanalytical laboratory, and the unblinded pharmacy staff and pharmacokineticist. If unblinding is required to manage subject safety or to support dose-escalation decisions, the decision to unblind lies solely with the PI. If possible and providing that it does not interfere with or delay any decision in the best interest of the subject, the PI will discuss the intended code-break with the Sponsor.

Dose-Escalation Halting Rules:

For Group 1 in P1/SAD, sentinel dosing will occur whereby 2 subjects (1 active and 1 PBO) will be dosed on 1 day and, providing no safety concerns arise, the remaining 6 subjects will be dosed after at least 24 hours.

The study will be halted if one or more subjects experience a serious adverse event (SAE) that is considered to be related to EIDD-2801 or 2 or more subjects in the same group experience severe AEs that are considered to be related to EIDD-2801.

Additionally, if 2 or more subjects experience a reduction of platelets greater than 50% over baseline levels or a reduction in lymphocytes (absolute count) to Grade 3/severe levels without an alternate cause, the PI and Sponsor will review the data. If, after review of unblinded data, it is determined that these events are probably related to study drug, no further dose escalations will occur.

Dose-escalation will stop if clinically relevant signs or symptoms of similar nature occur in 2 or

more subjects in a group that, in the opinion of the PI, warrant stopping of dose-escalation.

Data Monitoring, Safety Reporting and Unblinding: Data will be monitored throughout the course of the study by experienced clinical monitors according to the clinical monitoring plan. As data will be entered into an electronic case report form (CRF), data will be monitored to ensure accuracy as well as adherence to data entry instructions provided in the CRF guidelines. Procedures for reporting any SAE will be detailed in the protocol and forms and instructions provided to the site.

Statistical Considerations: A complete description of all statistical analyses and methods will be presented in the statistical analysis plan (SAP). The SAP will be reviewed and approved by the Sponsor and will be finalized prior to database lock. Plans for PK analyses will be included in the SAP.

Determination of Sample Size: The sample sizes for the P1 and P3 cohorts are typical for a Phase 1 first in human (FIH) study. The sample size for the P2 cohort is in accordance with Food and Drug Administration (FDA) guidelines for sample size in FE studies.

Study Populations:

Safety Population: All subjects who receive at least one dose of study drug.

Pharmacokinetic Population: All subjects receiving study drug who comply sufficiently with protocol requirements (i.e., no major protocol deviations) and for whom a sufficient number of analyzable PK samples have been obtained to permit the determination of the PK profile for EIDD-2801/EIDD-1931 and the statistical analysis of the results. Subjects who experience an AE of vomiting before 2× the median t_{max} of the group may be excluded from the PK population.

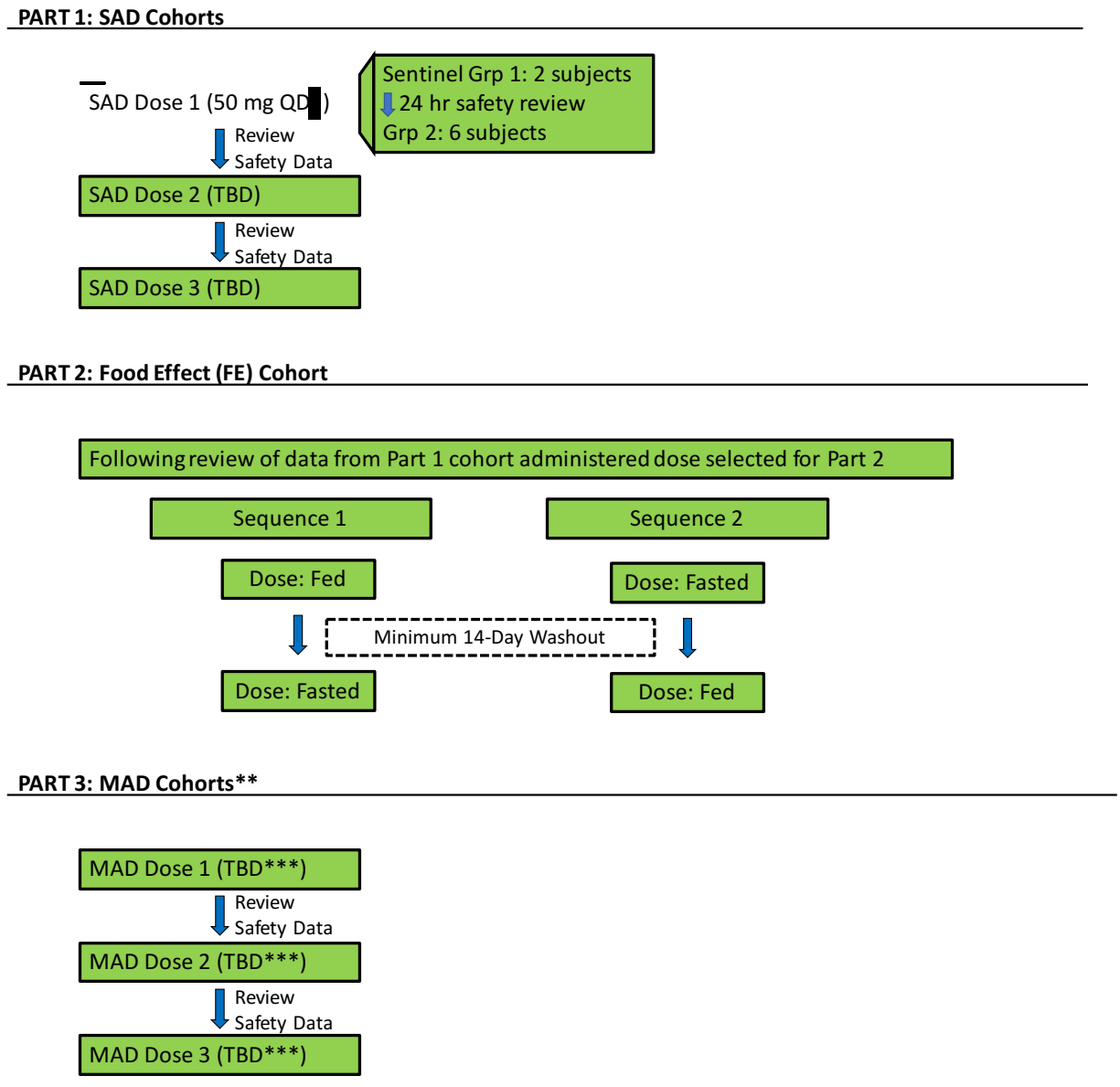
Safety Analyses: Statistical methods for the safety analyses will be descriptive in nature. Safety data, including AEs, clinical laboratory data, vital signs, ECG parameters, and PEs. All appropriate AEs will be graded using the Division of Microbiology and Infectious Diseases (DMID) toxicity scale (March 2014). Change from baseline will be included in summary tables for laboratory parameters. All laboratory data will be included in the data listings and all test values outside the normal range will be flagged.

Pharmacokinetic Analyses: Plasma PK parameters for each dose level will be calculated from the concentrations of EIDD-2801 and EIDD-1931 measured in predose and postdose plasma samples. For each dose level, descriptive statistics will be presented. Figures will be created to display mean and individual subject EIDD-2801 and EIDD-1931 concentration versus time. Urine PK parameters will be calculated whenever possible for each subject based on the urine concentrations of EIDD-2801 and EIDD-1931. The following PK parameters will be calculated:

Plasma PK Parameter	Description
AUC_{last}	The area under the plasma concentration-time curve, from time 0 to the last measurable non-zero concentration, as calculated by the linear up/log down trapezoidal method (P1 and P2 only).
$AUC_{0\text{-}inf}$	The area under the plasma concentration-time curve from time 0 extrapolated to infinity. $AUC_{0\text{-}inf}$ is calculated as the sum of AUC_{last} plus the ratio of the last measurable plasma concentration to the elimination rate constant (λz) (P1, P2, and Day 1 dose of P3).
C_{max}	Maximum observed concentration.
t_{max}	Time to reach C_{max}. If the maximum value occurs at more than one time point, t_{max} is defined as the first time point with this value.

λz	Apparent terminal elimination rate constant; represents the fraction of medication eliminated per unit time.
$t_{1/2}$	Apparent terminal elimination half-life of medication, calculated as $0.693/\lambda z$.
Vz/F	Apparent volume of distribution (EIDD-2801 only).
CL/F	Apparent oral drug clearance (EIDD-2801 only).
C_{trough}	Trough concentration (P3 only).
AUC_{τ}	The area under the plasma concentration-time curve during a dosing interval (P3 only).
$RA_{AUC\tau}$	Observed accumulation ratio based on AUC_{τ} (P3 only).
RA_{Cmax}	Observed accumulation ratio based on C_{max} (P3 only).
Urine PK Parameter	**Description**
A_e	Amount excreted in urine over the sampling interval.
F_e	Fraction of drug excreted in the urine over sampling interval.
CL_R	Renal clearance, $CL_R = A_e/AUC$ where both A_e and AUC are determined over co-incident time ranges after dosing.
In P1, dose-proportionality for PK parameters of EIDD-2801 and EIDD-1931 ($AUC_{0\text{-}inf}$, AUC_{last}, and C_{max}) will be assessed by the power model. The slope and the associated 90% confidence interval (CI) based on the power model will be reported. In P3 on Day 6, dose-proportionality for PK parameters of EIDD-2801 and EIDD-1931 (AUC_{τ} and C_{max}) will be assessed by the power model. The slope and associated 90% CI based on the power model will be reported. To assess the effect of food on the PK of EIDD-2801 and EIDD-1931, natural log-transformed PK parameters from P2 ($AUC_{0\text{-}inf}$, AUC_{last}, and C_{max}) will be analyzed using a mixed-effects model with fixed effect terms for treatment (with or without food), period, and sequence and with subject within sequence as a random effect. Point estimates and their associated 90% CIs for the natural log-transformed PK parameters for the difference between EIDD-2801 administered under fed relative to fasted conditions will be constructed. The point estimates and interval estimates will be back transformed to give estimates of the ratio and 90% CI for the geometric least squares mean ratio for fed/fasted to assess the effect of food.	
Interim Analyses: No formal interim analyses are planned for this study. Data from P1 cohorts may be locked, unblinded, and analyzed prior to conducting P2 and P3.	

Figure 1: Study Schema



Abbreviations:
FE: food effect
SAD: single ascending dose
MAD: multiple ascending dose

[redacted]

*** Part 3 may run in parallel with Part 1, providing that the total daily dose to be administered does not exceed a dose already shown to be safe and well tolerated in Part 1*

****The total daily dose in Part 3 will not exceed a dose shown to be safe and well-tolerated in Part 1*

1. INTRODUCTION

This study is being conducted at sites across the United Kingdom and the United States. Country-specific protocol amendments have been prepared; with the exception of regional regulatory requirements, the study is consistent across countries and data will be reported in a single clinical study report.

This study is a FIH study designed to assess the safety, tolerability and PK of EIDD-2801 in healthy human volunteers. EIDD-2801 is a ribonucleoside analog with broad-spectrum activity against many RNA viruses. It is currently being developed by Ridgeback Biotherapeutics as a treatment of infections caused by highly pathogenic coronaviruses (CoV), including COVID-19. In addition, EIDD-2801 is being developed in parallel as a treatment of uncomplicated influenza caused by all subtypes of circulating and emerging (drifted and shifted) influenza A virus (IAV) and influenza B virus (IBV), including seasonal, epidemic and pandemic strains.

1.1. Background

EIDD-2801 is the 5'-isopropyl ester prodrug of the broadly active, direct-acting antiviral ribonucleoside analog EIDD-1931. After oral delivery, the prodrug (EIDD-2801) is rapidly hydrolyzed by circulating esterases to produce high circulating (plasma) levels of EIDD-1931. In cell culture systems, EIDD-1931 has been shown to inhibit replication of multiple viral pathogens from multiple RNA virus families including pathogenic CoV (e.g., Middle East respiratory syndrome [MERS], severe acute respiratory syndrome [SARS]-CoV and SARS-CoV-2), influenza viruses (seasonal, pandemic, and avian subtypes), respiratory syncytial virus (RSV), alphaviruses (e.g., Eastern equine encephalitis virus [EEEV], Venezuelan equine encephalitis virus [VEEV], and Chikungunya virus [CHKV]), Filoviruses (e.g., Ebola virus [EBOV]), and Zika virus (ZIKV). In addition, EIDD-2801 is active against orthopoxviruses (tested against vaccinia virus) probably because orthopoxviruses encode their own unique RNA polymerase. [redacted] The primary mechanism of action of EIDD-2801 is inhibition of viral RNA replication by incorporation of the EIDD-1931 monophosphate metabolite into the viral RNA genome resulting in induction of viral error catastrophe.

1.2. Rationale for Development

EIDD-2801 is being developed for the treatment of infections caused by RNA viruses, specifically for COVID-19 and other CoV infections, influenza, and VEEV. During conduct of the FIH study, the Sponsor intends to define a dose that may be active in treating COVID-19 in patient studies.

EIDD-2801 has a unique dual mechanism of action against RNA viruses, including SARS-CoV-2 and other CoV infections. The compound acts as a competitive, alternative substrate for the virally encoded RNA-dependent RNA-polymerase that upon incorporation into nascent chain RNA induces increased mutational frequency in the viral genome. Incorporation quickly results in the production of non-viable virus. Additionally, the active metabolite,

EIDD-1931-5'-triphosphate (EIDD-2061), may act directly as a chain terminator and arrest replication by exerting a next nucleoside effect. It is anticipated that the high barrier to resistance observed during *in vitro* passaging studies will translate to slow, if any, emergence of viral resistance. Resilience to viral escape is a distinguishing feature of EIDD-2801.

Currently, there is no approved antiviral therapeutic for the treatment of COVID-19. An antiviral drug is urgently needed. [redacted]

1.3. Nonclinical Overview

1.3.1. Mechanism of Action

The mechanism of antiviral activity of EIDD-2801 is "lethal mutagenesis"; a concept that is predicated on increasing the viral mutation rate beyond a biologically-tolerable threshold, resulting in impairment of viral fitness and leading to viral extinction.

The specifics of the mechanism are as follows. EIDD-2801 is rapidly taken up by cells and the 5'-isopropylester cleaved to liberate EIDD-1931, which is in turn phosphorylated to EIDD-2061 by host kinases (Hernandez-Santiago et al., 2004; Painter et al., 2019). The 5'-triphosphate, EIDD-2061, acts as a competitive alternative substrate for virally encoded RNA-directed RNA polymerases and EIDD-2061 is incorporated into nascent viral RNA. Owing to the ability of the N^4-hydroxycytosine base of EIDD-1931 to tautomerize, EIDD-2061 can pair with either guanosine or adenosine, and consequently can substitute for either CTP or UTP, respectively (Flavell et al., 1974). This results in an accumulation of mutations that increases with each cycle of viral replication. The process whereby the mutation rate is increased by exposure to a drug is referred to as Viral Decay Acceleration (Mullins et al., 2011) and results in viral ablation.

Significant work has gone into validating this mechanism of action for EIDD-2801/1931, and it has been shown for MERS-CoV, VEEV, and IAV that viruses grown in the presence of EIDD-1931 have significantly increased levels of transition mutations (Agostini et al., 2019; Toots et al., 2019; Urakova et al., 2018). Multi-log decreases in virus yields were observed after treatment with EIDD-1931. Additionally, it was demonstrated for VEEV that the infectivity of virions formed in the presence of EIDD-1931 decreases from ~20% to <0.2%, and that the infectious virions are significantly Impaired in their replication ability (Urakova et al., 2018). As a consequence of this mechanism of action, the generation of drug-resistant escape mutants is practically impossible. This same effect was demonstrated for CoV (Agostini et al., 2019) and influenza virus (Toots et al., 2019). Furthermore, given the unique mechanism of action, EIDD-2801 is expected to be active against viruses resistant to other antiviral agents which have a different mechanism of action. The only data generated to date regarding the activity of EIDD-1931 against viruses resistant to other nucleoside analogs found that EIDD-1931 was

active against CoV resistant to remdesivir in cell culture assays (T. Sheahan et al, preprint available at https://www.biorxiv.org/content/10.1101/2020.03.19.997890v1).

As an alternative or additional mechanism of action, it has been theorized that incorporation of EIDD-2061 into viral genomic RNA can change the thermodynamics of RNA secondary structure and thus decrease the efficiency of the promoter regions involved in RNA genome replication (Stuyver et al., 2003).

1.3.2. *In Vitro* Pharmacology

1.3.2.1. Antiviral Activity in Tissue Culture and in Human Airway Epithelium

The ribonucleoside analog EIDD-1931 is the parent of the prodrug EIDD-2801. EIDD-1931 shows specific antiviral activity in different tissue culture cells and in the differentiated organoid model of human airway epithelium (HAE) with a selectivity index (SI) ranging from 21 to >100 for all influenza viral isolates tested. It is active against IAV (pandemic and seasonal) and IBV strains, as well as against highly pathogenic H5N1 and H7N9 strains (Table 1).

Table 1: EIDD-1931 Antiviral Activity Against Influenza A and B Viruses in Tissue Culture and Primary Human Bronchial/Tracheal Epithelial Cells

Virus	Strain	Cell line	EC_{50}* (µM)	CC_{50} (µM)	SI	Reference
IAV H1N1	Ca/07/2009	MDCK	1.24	68	55	NIAID Antiviral Testing Program
IAV H1N1	WSN/33	MDCK	1.1	299.8	275	Yoon et al., 2018
IAV H1N2	WSN/33	primary hBTEC	5.4	-	-	Yoon et al., 2018
IAV H2N3	Perth/16/2009	MDCK	0.88	52	59	NIAID Antiviral Testing Program
IAV H2N3	Ohio/sw-10-132/2010	MDCK	3.2	299.8	94	Yoon et al., 2018
IAV H5N1	Duck/MN/1525/81	MDCK	1.28	27	21	NIAID Antiviral Testing Program
IAV H5N1	Vietnam/1203/2004	MDCK	0.14	299.8	2100	Yoon et al., 2018
IAV H7N9	Anhui/1/2013	MDCK	0.13	299.8	2300	Yoon et al., 2018
IBV	Florida/4/2006	MDCK	<0.4	76	>190	NIAID Antiviral Testing Program
IBV	Brisbane/60/08	MDCK	0.006	299.8	50000	Yoon et al., 2018
IAV H1N1	Ca/07/2009	HAE-3D*	0.08	50	625	Toots et al., 2019
IAV H1N1	WSN/33	HAE-3D*	0.08	50	625	Toots et al., 2019
IBV	Brisbane/60/08	HAE-3D*	0.06	50	833	Toots et al., 2019

* Human Airway Epithelium organoid model.

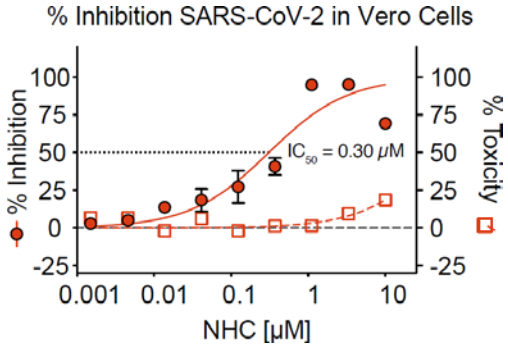
EIDD-1931 also showed specific antiviral activity against human SARS-CoV, MERS-CoV (Table 2) and SARS-CoV-2 (Figure 2), against togaviruses VEEV, EEEV and CHKV (Table 3) [redacted]

Table 2: EIDD-1931 Antiviral Activity Against SARS and MERS Coronaviruses in Tissue Culture and Primary Human Bronchial/Tracheal Epithelial Cells

Virus	Strain	Cell line	EC_{50} (µM)	CC_{50} (µM)	SI	Reference
SARS-CoV-1	Urbani	Vero76	<0.4	144	>360	NIAID Antiviral Testing Program
SARS-CoV-1	SARS-CoV-GFP(†)	HAE-3D(*)	<1	>100	>100	Tech. Report 25.038
MERS-CoV	GenBank Ac.No JX869059**	DBT-9	0.56	>200	>357	Agostini et al., 2019
MERS-CoV	Human ß-CoV C, Novel 2912	Vero E6	<0.8	20	>25	NIAID Antiviral Testing Program

* Human Airway Epithelium organoid model; ** cDNA Derived clone

Figure 2: Inhibition of SARS-CoV-2 by EIDD-1931



EIDD-1931 (NHC) antiviral activity (closed circles) and cytotoxicity (open squares) in Vero Cells infected with SARS-CoV-2. Vero cells were infected in duplicate with SARS-CoV-2 clinical isolate virus at a multiplicity of infection (MOI) of 0.05 in the presence of a dose response of EIDD-2801 for 48 hours after which replication was measured through quantitation of cell viability by Cell-Titer-Glo assay. Cytotoxicity was measured in similarly treated but uninfected cultures. Reproduced from Sheahan et al 2020.

Table 3: EIDD-1931 Antiviral Activity Against Togaviruses in Tissue Culture

Virus	Strain	Cell line	EC_{50} (µM)	CC_{50} (µM)	SI	Reference
VEEV	TC-83	Vero	0.43	>200	>930	Urakova et al., 2018
VEEV	TC-83	Vero76	1.92	32	17	NIAID Antiviral Testing Program
EEEV	FL93-939	Vero76	1.08	84	78	NIAID Antiviral Testing Program
CHKV	S27 (VR-64)	Vero76	1.8	96	53	NIAID Antiviral Testing Program

[redacted]

1.3.2.2. Cytotoxicity of EIDD-1931 in Tissue Culture Utilizing Cells from Different Organs and Species

EIDD-1931 was tested for cytotoxicity in human hepatic origin Huh7 and HepG2 cells, in human lymphoid CEM, human pancreatic BxPC-3, human prostate cancer PC-3, human muscle A204, human lung A549, human epithelial hEp-2, rat heart muscle H9c2, monkey kidney Vero, and canine kidney MDCK cell lines (Table 5). The compound exhibits low cytotoxicity in the majority of cells tested (half-maximal effective concentration [EC_{50}] values are in the range of 40 to >100 µM) except in lymphoid origin CEM cells where the compound shows a 7.5 µM EC_{50} value (Sticher et al., 2020; Urakova et al., 2018; Yoon et al., 2018).

Table 5: Cytotoxicity (CC50) of EIDD-1931 in Mammalian Cell Lines

Cell Line	CEM	HepG2	PC-3	A204	A549	BxPC-3	Huh-7	H9c-2	Vero	hEp-2	MDCK
CC50 (µM)	7.5	42.3	267.1	84	46	48	165.5	81	53	272.4	299.8

Sources: Sticher et al., 2020, Yoon et al., 2018

EIDD-2801 typically showed 2-4× lower activity and cytotoxicity than EIDD-1931 due to slightly slower uptake and anabolism in tissue culture.

1.3.2.3. Assessment of Mitochondrial Toxicity

Since EIDD-1931 is a nucleoside analog, additional investigations were performed to analyze whether observed cytotoxicity of EIDD-1931 is caused by mitochondrial toxicity. It was demonstrated that the prolonged treatment (14 days) with the compound does not result in selective killing of mitochondria or in mitochondrial dysfunction in CEM and HepG2 cells (Sticher et al., 2020).

1.3.3. *In Vivo* Pharmacology

The prodrug EIDD-2801 or its parent EIDD-1931 have been tested in animal models of RNA viral infection. An overview of results from the animal studies in indications to be pursued are described below. Additional detail is provided in the Investigator's Brochure (IB).

1.3.3.1. Coronavirus: SARS-CoV and MERS-CoV

In mouse models of SARS and MERS infection and disease, coronaviral disease was assessed by changes in body weight, measured daily, and lung hemorrhage, assessed in the large left lobe of the lung following sacrifice on Day 5 (SARS-CoV) or Day 6 (MERS-CoV). To assess production of infectious virions, virus was isolated from the lower left lobe of the lung following sacrifice on Day 5 (SARS-CoV) or Day 6 (MERS-CoV) and quantified using a plaque assay.

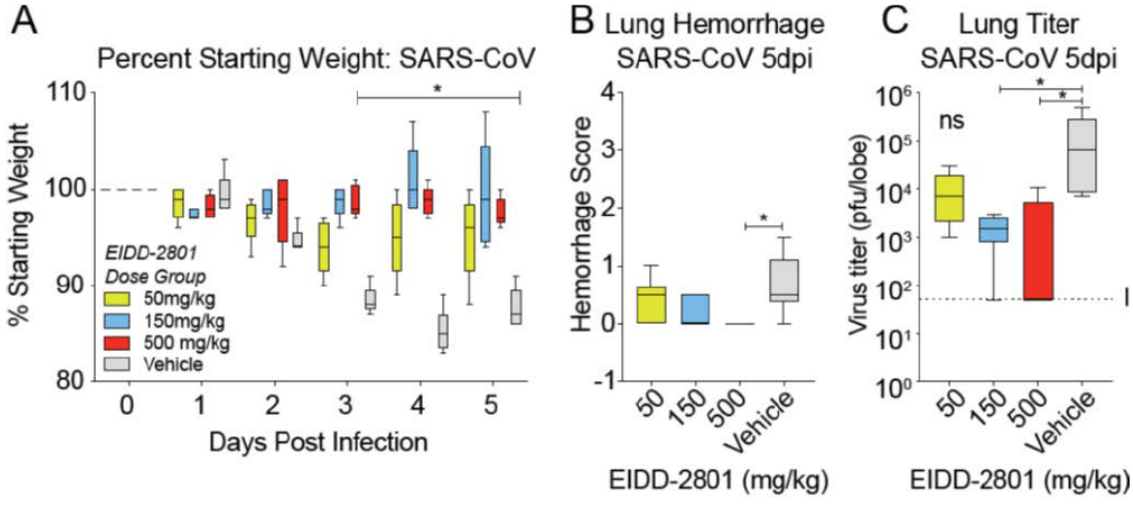
The results demonstrated that, in mice infected with either SARS- or MERS-CoV, both prophylactic and therapeutic treatment with EIDD-2801 resulted in a reduction in virus replication, improvements in pulmonary function, and improvements in maintaining body weight

(i.e., reduced body weight loss). While EIDD-2801 doses of 50, 150 and 500 mg/kg BID were assessed in the CoV mouse experiments, 500 mg/kg BID yielded the most consistent therapeutic effect.

A prophylactic, dose-escalation study was conducted in C57BL/6 mice infected with mouse-adapted SARS-CoV (SARS-MA15). Prophylactic oral treatment with EIDD-2801 was initiated 2 hours before intranasal infection and continued every 12 hours thereafter through the end of the study (Day 5; Figure 3).

In mice treated BID with EIDD-2801, body weight loss observed with vehicle treatment was diminished in the 50 mg/kg treatment group, beginning on Day 3 post-infection. No body weight loss was seen in the 150 and 500 mg/kg treatment groups (Figure 3, Panel A). Lung hemorrhage was also significantly reduced on Day 5 post-infection, following treatment with 500 mg/kg EIDD-2801 (Figure 3, Panel B). When compared to vehicle control, a dose-dependent reduction in SARS-CoV lung titers at Day 5 was seen across all 3 treatment groups (Figure 3, Panel C) with significant differences among the vehicle, 150 mg/kg and 500 mg/kg groups. Thus, prophylactic treatment with EIDD-2801 resulted in a robust antiviral effect that was able to prevent SARS-CoV replication and disease.

Figure 3: EIDD-2801 Prophylactic Treatment of SARS-CoV Infected Mice

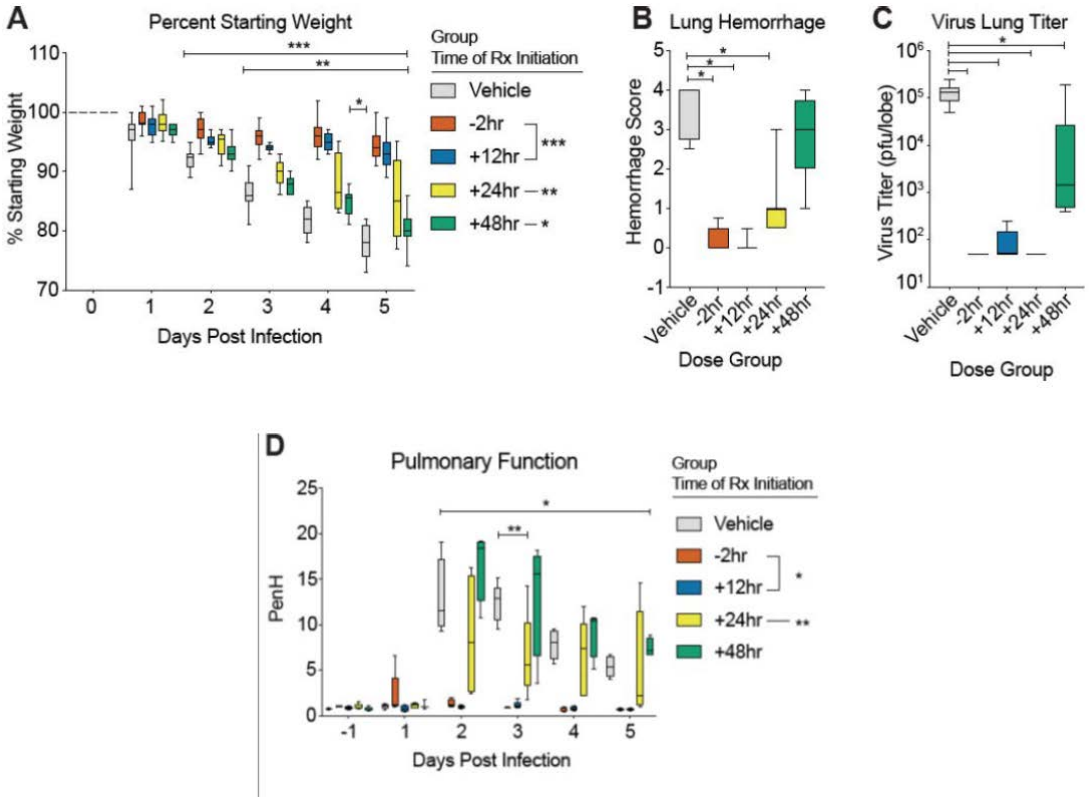


Effect of EIDD-2801 treatment on body weight (A), lung hemorrhage (B) and virus lung titer (C) in mice infected with SARS-MA15. Treatment was initiated two hours prior to infection and continued every 12 hours (BID) to Day 5.

The antiviral activity of EIDD-2801 against SARS-CoV was compared when treatment was initiated at -2 hours (pre-infection) and 12, 24, or 48 hours post-infection. After initiation of treatment, all groups were dosed every 12 hours for the duration of the study (Figure 4). For SARS-challenged mice, initiating treatment at 12 hours post-infection significantly prevented body weight loss beginning on Day 2, a result similar to that seen when dosing prophylactically

(i.e., beginning at 2 hours pre-infection). Initiation of treatment with EIDD-2801 at 24 hours post-infection also significantly reduced body weight loss on Days 3 through 5 post-infection. When EIDD-2801 treatment was initiated at 48 hours post-infection, body weight loss was only statistically different from vehicle on Day 4 post-infection (Figure 4, Panel A). Significant reductions in lung hemorrhage were seen when EIDD-2801 treatment was initiated before (-2 hours) and up to 24 hours after infection; a result that mirrored body weight loss data (Figure 4, Panel B). All mice treated with EIDD-2801 had significantly reduced viral loads in the lungs, even in the group where treatment was initiated 48-hour post-infection (Figure 4, Panel C). Pulmonary function, measured via whole body plethysmography, was assessed using the PenH metric which is a surrogate marker for bronchoconstriction or pulmonary obstruction. The administration of EIDD-2801 prior to infection (-2 hours) and at 12 hour post-infection completely abrogated the loss of pulmonary function observed in vehicle-treated animals (Figure 4, Panel D). Improved pulmonary function was also seen in the group where treatment was initiated 24 hours after virus challenge.

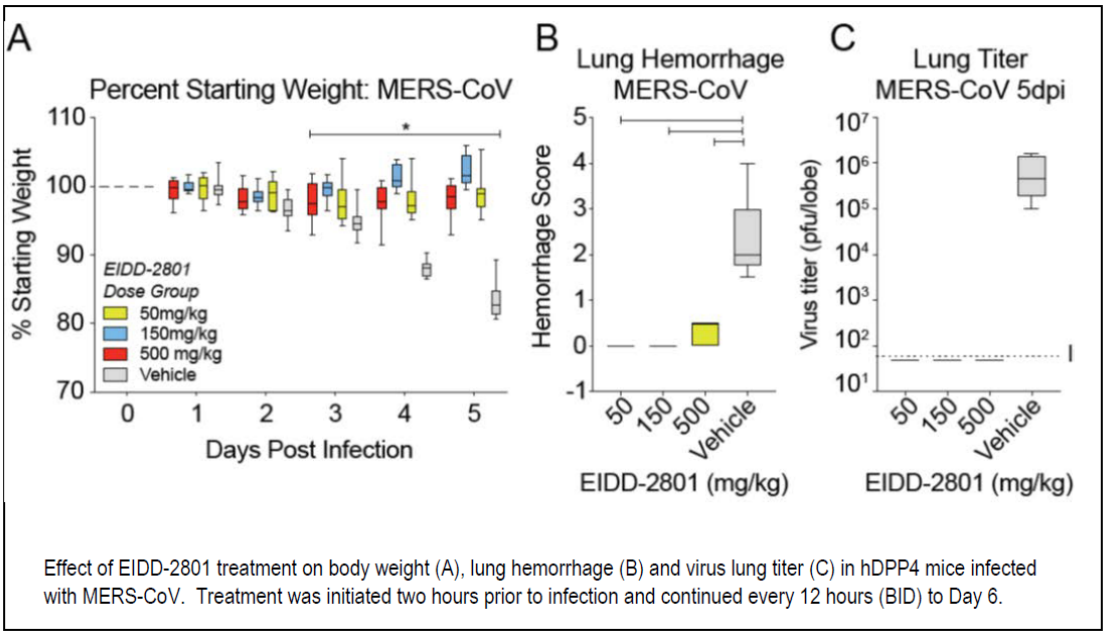
Figure 4: EIDD-2801 Treatment of SARS-CoV Infected Mice



EIDD-2801 was also tested to determine if it is active *in vivo* against MERS-CoV as described by Sheahan et. al (draft manuscript). The murine ortholog of the MERS-CoV receptor, dipeptidyl peptidase 4 (DPP4), does not support viral binding and entry. Thus, all *in vivo* studies described below were performed in genetically modified hDPP4 mice permissive for MERS infection. Prophylactic treatment starting at 2 hours before viral challenge with either 50, 150, or

500 mg/kg EIDD-2801 prevented body weight loss on Days 2 through 6 post-infection (Figure 5, Panel A), prevented lung hemorrhage measured on Day 6 (Figure 5, Panel B), and reduced virus lung titer on Day 6 to the limit of detection (Figure 5, Panel C).

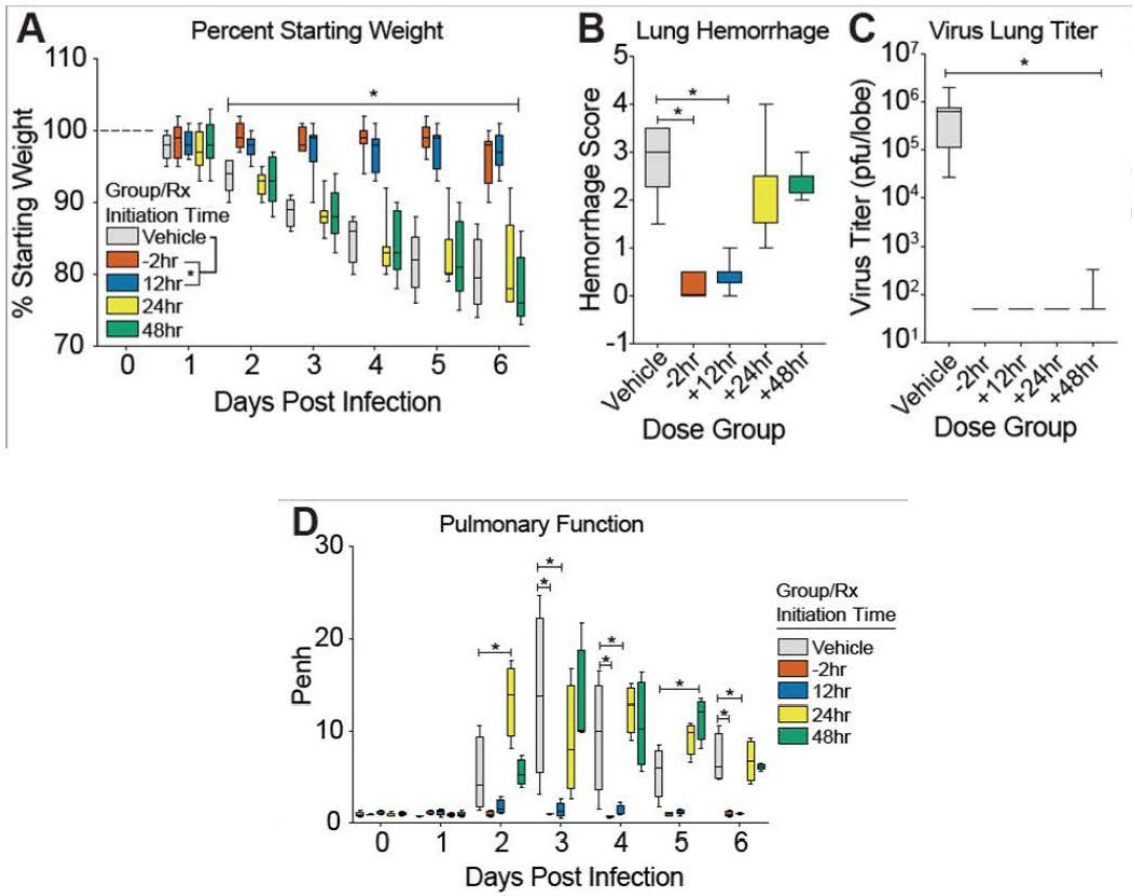
Figure 5: EIDD-2801 Prophylactic Treatment of MERS-CoV Infected Mice



Effect of EIDD-2801 treatment on body weight (A), lung hemorrhage (B) and virus lung titer (C) in hDPP4 mice infected with MERS-CoV. Treatment was initiated two hours prior to infection and continued every 12 hours (BID) to Day 6.

The effect of EIDD-2801 treatment on MERS-CoV infected mice is shown in Figure 6. When EIDD-2801 treatment was initiated 12 hours post-infection, there was no loss in body weight from Days 2 through 6 post-infection (Figure 6, Panel A) and no evidence of lung hemorrhage on Day 6 post-infection (Figure 6, Panel B). However, protection was not observed in groups where treatment was initiated either 24- or 48-hours post-infection. Conversely, virus lung titer on Day 6 post-infection was significantly reduced to the limit of detection in all treatment groups, regardless of the time treatment began (Figure 6, Panel C). To gauge the effect of the timing of EIDD-2801 treatment initiation on physiologic measures of lung disease, pulmonary function was assessed by whole body plethysmography. As expected, a loss of pulmonary function, as determined by measuring the PenH metric, was observed in vehicle-treated animals infected with MERS-CoV beginning on Day 2 post-infection (Figure 6, Panel D). Mirroring the body weight loss data, normal pulmonary function was observed in groups where treatment was initiated prior to or at 12 hours post-infection (Figure 6, Panel D).

Figure 6: EIDD-2801 Treatment of MERS-CoV Infected Mice



1.3.3.2. Influenza Virus

EIDD-2801 was tested in a ferret model of influenza virus infection and disease. Ferrets recapitulate hallmarks of human influenza infection, providing a clinically relevant animal model to investigate therapeutic intervention. Therapeutic oral dosing of influenza virus-infected ferrets reduced shed levels of pandemic and seasonal IAV by multiple orders of magnitude, and alleviated fever and inflammation while improving airway epithelium histopathology. Post-exposure prophylactic dosing was sterilizing (Toots et al., 2019).

Ferrets infected with pandemic IAV and treated with EIDD-2801 at a dose of 7 mg/kg demonstrated significantly less viral shedding and inflammatory cellular infiltrates in nasal lavages, but mounted a normal humoral antiviral response (Toots et al., 2019).

When examining the effect of delayed dosing, Toots et. al. (2019) demonstrated that treatment with 20 mg/kg of EIDD-2801 initiated 24 hours after challenge with IAV (H1N1) resulted in a rapid decrease in both shed virus and fever relative to control, with fever returning to baseline at 24 hours. When oseltamivir (20 mg/kg) was dosed prophylactically as a positive control, a modest suppression of fever was observed; however, there was no reduction in shed virus titer.

1.3.3.3. Venezuelan Equine Encephalitis Virus

Treatment with EIDD-1931 was evaluated in a mouse model of lethal VEEV infections. To be truly effective as a therapeutic agent for VEEV infection, a drug must penetrate the blood brain barrier and arrest virus replication in the brain. High plasma levels of EIDD-1931 are rapidly achieved in mice after oral dosing. Once in the plasma, EIDD-1931 is efficiently distributed into organs important in the pathology of VEEV infection, including the brain, where it is rapidly converted to its active 5'-triphosphate (EIDD-2061). EIDD-1931 showed a good safety profile in mice after 7 days of dosing with up to 1,000 mg/kg/day. In mouse model studies of VEEV infection, EIDD-1931 was 90-100% effective in protecting mice against lethal intranasal infection when therapeutic treatment was started as late as 24 hours post-infection, and partial protection was achieved when treatment was delayed for 48 hours post-infection (Painter et al., 2019).

[redacted]

1.4. Safety and Secondary Pharmacology

The standard battery of safety pharmacology studies including studies assessing the cardiovascular, respiratory and central nervous systems have been conducted. The studies are discussed in the IB; results indicated that there were no adverse pharmacologic effects of EIDD-2801 on the cardiovascular, respiratory or central nervous systems.

1.5. Nonclinical Pharmacokinetics and Metabolism

1.5.1. Overview

The uptake, metabolism and protein binding of EIDD-2801 and EIDD-1931 have been studied in plasma, microsomes, and non-hepatic cells from several species as outlined below. The PK and tissue distribution of prodrug EIDD-2801 and its active parent EIDD-1931 have been studied extensively in rats, dogs and ferrets. Key results from these studies are presented below; additional detail can be found in the IB.

1.5.2. Absorption

EIDD-1931 is parent of the prodrug EIDD-2801. The appearance of EIDD-1931 is dependent on the absorption of EIDD-2801 and the rate of its conversion to EIDD-1931.

EIDD-2801 PK studies have been completed in dog, rat, mouse, ferret, and monkey. EIDD-2801 was efficiently absorbed and rapidly converted to EIDD-1931 in each species. The t_{max} for EIDD-2801 (not observed in rodents) occurred at 0.5-1 hours, while the t_{max} for EIDD-1931 occurred at 1-2 hours.

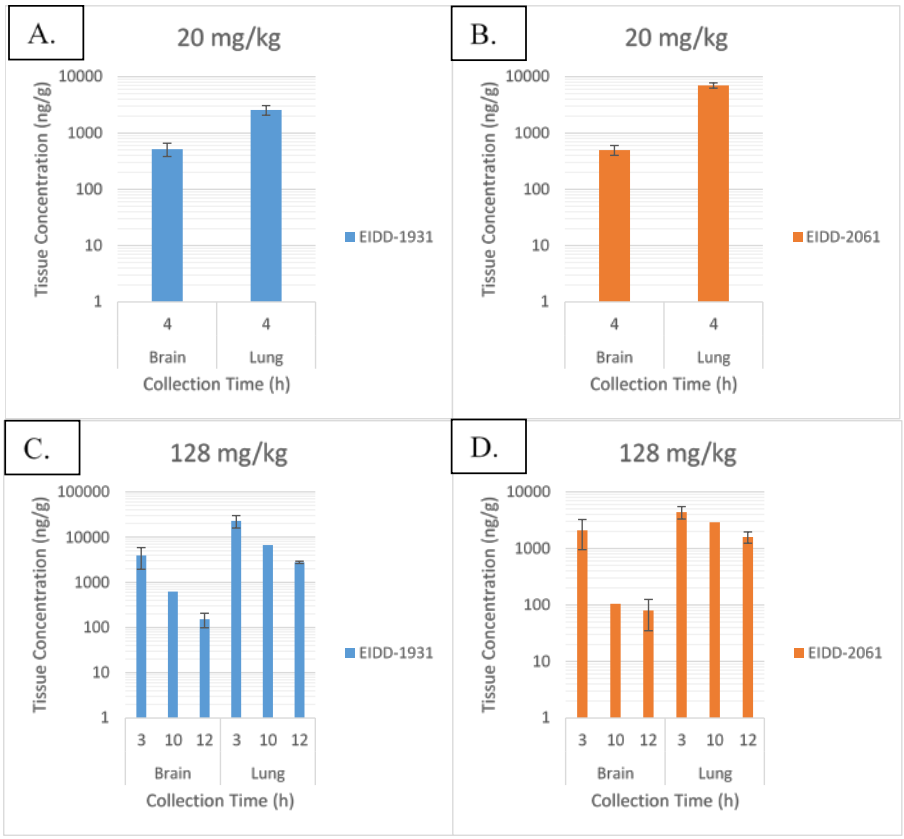
1.5.3. Distribution

[redacted]

1.5.3.2. Tissue Distribution Studies

EIDD-2801 is rapidly absorbed in the gut and converted to EIDD-1931 reaching C_{max} in 1-3 hours in mice, rats, ferrets, dogs and monkeys. EIDD-1931 is then widely distributed to tissues including lungs and brain, where it is rapidly taken up into cells and converted to EIDD-2061. Figure 7 shows the concentration of EIDD-1931 and EIDD-2061 in ferret brain and lung following single doses of 20 (Panels A and B) and 128 (Panels C and D) mg/kg.

Figure 7: Tissue Distribution of EIDD-1931 and EIDD-2061 in Ferret Brain and Lung



1.5.4. Metabolism

1.5.4.1. Metabolic Stability of EIDD-1931 and EIDD-2801

EIDD-2801 was designed to be converted to EIDD-1931 by esterases in plasma or in cells. Stability has been assessed in plasma and liver microsomes from mouse, rat, dog, monkey and humans. The stability of EIDD-2801 in mouse, rat and monkey plasma is relatively short (≤0.4 hours) while the stability is longer in human and dog plasma (1-3 hours). EIDD-2801 stability in mouse, rat, dog and monkey liver microsomes is very short, ranging from 0.02 to 0.08 hours while the stability in human liver microsomes is 1.2 hours (Table 6).

Table 6: Metabolic Stability of EIDD-2801 in Plasma and Liver Microsomes

	Plasma stability	LM stability
Species	t1/2 (h)	t1/2 (h)
Mouse	0.017	0.033
Rat	0.033	0.017
Dog	3.2	0.083
Monkey	0.40	0.017
Human	1.05	1.22

EIDD-2801 is stable in simulated gastric and intestinal fluids (Table 7).

Table 7: Metabolic Stability of EIDD-2801 in Simulated Gastric and Intestinal Fluids and in Buffered Saline with Fetal Bovine Serum

Matrix	t1/2 (hr)
Simulated Gastric Fluid	>24
Simulated Intestinal Fluid	>24
Phosphate Buffered Saline plus 10% Fetal Bovine Serum*	>24

EIDD-1931 was found to be stable when incubated with all tested plasmas, whole blood, liver microsomes and liver S9 extracts and intestinal microsomes (Table 8).

Table 8: Metabolic Stability of EIDD-1931 in Plasma, Whole Blood and Liver and Intestinal Microsomes

Medium	Plasma	Whole Blood	Liver Microsomes	Liver S9 Stability	Intestinal Microsomes
Species	t1/2 (h)	t1/2 (h)	t1/2 (h)	t1/2 (h)	t1/2 (h)
Mouse		>24	>24		>24
Rat	17		>24	>24	
Dog	>24			>24	
Monkey	6.5	>24	>24	>24	>24
Human	10	7	>24	20	>24

1.5.4.2. Uptake and Anabolism of EIDD-1931 in Tissue Culture and Primary Cells

EIDD-1931 is efficiently taken up by tissue culture cells and converted to its pharmacologically active metabolite EIDD-2061 (EIDD-1931-5'-triphosphate). Intracellular EIDD-2061 accumulates dose-dependently, with C_{max} levels ~200-2000 pmol/10^6 cells (at 10-20 µM dose) in different cell lines. It reaches high levels relatively quickly, typically within 1-3 hours, though the t_{max} values vary widely between 1 and 24 hours depending on the cell line and dose concentration tested. Detailed data on the uptake and anabolism of EIDD-2801 is presented in the IB.

EIDD-2801 is also taken up by tissue culture cells and is converted to EIDD-1931 and then to EIDD-2061, but the process is slightly delayed compared to dosing with EIDD-1931. EIDD-1931 is also taken up and metabolized to EIDD-2061 by primary cells. EIDD-2061 is accumulated in all primary cells tested except in mouse primary hepatocytes where EIDD-1931 is apparently extensively metabolized to cytidine and uridine which, in turn, quickly metabolize into CTP and UTP. The quick metabolism of EIDD-1931 consequently results in low levels of EIDD-2061 in mouse hepatocytes. The intracellular stability ($t_{1/2}$) of EIDD-2061 is 4-5 hours in human astrocytes and hBTEC and is significantly shorter (0.2-1.1 hours) in primary hepatocytes.

1.5.5. Excretion

Currently, there is no data on excretion of EIDD-2801 or EIDD-1931. Excretion will be measured in urine during this study.

1.5.6. Pharmacokinetic Drug Interactions

1.5.6.1. Cytochrome P450 (CYP) Inhibition

The purpose of this non-GLP *in vitro* study was to determine the time-dependent inhibitory potential of EIDD-2801 and EIDD-1931 on human cytochromes P450 (CYP) 1A2, 2B6, 2C8, 2C9, 2C19, 2D6, and 3A4 (midazolam and testosterone substrates) enzyme activity, using pooled human liver microsomes in an half-maximal inhibitory concentration (IC_{50}) shift assay.

Neither EIDD-2801 nor EIDD-1931 demonstrated inhibition greater than 31.4% for any of the CYP isozymes tested nor could the data for each assay condition be curve fit to determine time-dependent inhibition by these compounds. Full dose-response curves were not achieved at concentrations ranging from 0.00545 to 50.0 µM indicating EIDD-2801 and EIDD-1931 have no CYP inhibition potential at concentrations ranging from 0.00545 to 50.0 µM. Assay performance was acceptable based on the results for the positive control inhibitors.

1.5.6.2. CYP Induction

An assay was performed to determine the induction potential of EIDD-2801 on human CYP isoenzyme (1A2, 2B6, and 3A4) activity using 3 single-donor lots of inducible, cryopreserved human hepatocytes. Both enzyme activity and mRNA results demonstrated that EIDD-2801 did not show induction for any of the CYP isozymes.

[redacted]

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[redacted]	[redacted]	[redacted]	[redacted]	[redacted]
[redacted]	[redacted]	[redacted]	[redacted]	[redacted]
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1.7. Potential Risks and Benefits

1.7.1. Potential Benefits

As this is a FIH study in healthy volunteers, there is no direct benefit to subjects enrolled in the study. However, given the current pandemic situation and *in vitro* antiviral activity of EIDD-2801 against SARS-CoV-2, and the activity against several other viruses of public health concern, it is possible that participants may benefit from future availability of the drug.

1.7.2. Potential Risks

EIDD-2801 has never been administered to humans; therefore, the risks from EIDD-2801 to subjects participating in this trial are unknown. Although toxicology studies have been done, unexpected AEs may occur. [redacted]. Subjects will also be monitored for other end-organ effects through a range of safety assessments.

2. OBJECTIVES

2.1. Study Objectives

2.1.1. Primary Objectives

The primary objective of Part 1 of the study is to determine the safety and tolerability of single ascending doses of EIDD-2801.

The primary objective of Part 2 of the study is to assess the effect of food on the PK on EIDD-2801 and EIDD-1931 following a single oral dose.

The primary objective of Part 3 of the study is to determine the safety and tolerability of multiple ascending doses of EIDD-2801.

2.1.2. Secondary Objectives

The secondary objectives of Part 1 and Part 3 of the study is to define the PK of EIDD-2801 and EIDD-1931 in plasma and urine following single and multiple doses administered to healthy volunteers.

The secondary objective of Part 2 of the study is to determine the safety and tolerability of single doses of EIDD-2801.

2.1.3. Exploratory Objectives

The exploratory objective of Part 3 is to collect data to assess the relationship between EIDD-2801/EIDD-1931 concentrations and QTc.

2.2. Study Outcome Measures

2.2.1. Primary Outcome Measures

The primary outcome measures for Parts 1 and 3 of are results of safety evaluations including safety laboratory assessments, PEs, ECGs, vital signs, and AEs.

The primary outcome measures for Part 2 of the study are plasma PK parameters including C_{max}, t_{max}, $t_{1/2}$, CL/F, λz, Vz/F, $AUC_{0\text{-}inf}$ and AUC_{last} as appropriate.

2.2.2. Secondary Outcome Measures

The secondary outcome measures are as follows:

- Single-dose plasma PK parameters, including C_{max}, t_{max}, $t_{1/2}$, CL/F, λz, Vz/F, $AUC_{0\text{-}inf}$ and AUC_{last} as appropriate (Part 1)

- Multiple-dose plasma PK parameters, including C_{trough}, C_{max}, t_{max}, $t_{1/2}$, CL/F, λz,

Vz/F, AUC_{τ}, $AUC_{0\text{-inf}}$ (Day 1 dose only), $RA_{AUC\tau}$ and RA_{Cmax}, as appropriate (Part 3)

- Urinary excretion of EIDD-2801 and EIDD-1931 following single- and multiple-dose administration (Parts 1 and 3).

- Results of safety evaluations including safety laboratory assessments, PEs, ECGs, vital signs, and AEs (Part 2).

3. STUDY DESIGN

3.1. Overview

EIDD-2801-1001-UK is a Phase 1, randomized, double-blind, placebo-controlled, FIH, SAD, and MAD study of the safety, tolerability and PK of EIDD-2801 and EIDD-1931 following oral administration of single and multiple doses of EIDD-2801 to healthy volunteers. In addition, for a minimum of one cohort, the effect of food on the single-dose EIDD-2801 and EIDD-1931 PK parameters will be assessed in subjects taking open-label EIDD-2801. The overall objective of the study is to identify a starting dose for future safety and therapeutic intervention trials.

The study is composed of 3 parts; P1 is the SAD study, P2 is the FE cohort study, and P3 is the MAD study.

This study will be conducted at 1 site in the United Kingdom and up to 3 sites in the United States.

3.1.1. Part 1 (Single Ascending Dose)

A single oral dose of EIDD-2801 or PBO will be administered to subjects. Subjects will be randomized in a 3:1 ratio to receive either EIDD-2801 or PBO.

Following dosing on Day 1, subjects will remain in the clinic through completion of study assessments on Day 4, returning to the clinic on Day 9 for study assessments, and Day 15 for the EOS visit.

The first cohort will be divided into 2 groups. The first group, a sentinel group, will include 2 subjects. On Day 1, one subject in the sentinel group will be administered PBO and one will be administered EIDD-2801. Analysis of the safety results for the sentinel group will be reviewed 24 hours postdose. If no halting criteria (Section 9) have been met, the remainder of the subjects in Cohort 1 (the second group) will be dosed. There will be no sentinel groups for the remaining cohorts.

After completion of each dosing cohort, safety and tolerability data will be reviewed to determine if any of the halting rules have been met. If not, then the subsequent cohort may be dosed following review of the 72-hour safety data. As PK data become available, these data may be used for dose-escalation decisions.

The proposed dose-escalation scheme is shown in Figure 1, however, planned dose escalations will be determined based on ongoing review of the safety, tolerability, and available PK data. The starting dose in the first SAD cohort will be 50 mg. Dose levels will not increase by more than 3-fold for predicted nonpharmacologically active dose levels and by 2-fold for predicted pharmacologically active dose levels. The highest total daily dose to be studied under this protocol will not exceed 800 mg (Section 9.2).

Three cohorts are initially planned for P1; however, up to an additional 4 cohorts may be enrolled.

3.1.2. Part 2 (Food-Effect)

Two single oral doses of EIDD-2801 will be administered to subjects, in an open-label manner. The dose for the P2/FE cohort is planned to be 100 mg but may be adjusted based on safety, tolerability, and/or PK data from P1. The dose assessed in the P2 cohort will have been given previously to subjects in a P1 cohort successfully (i.e., no halting rules were met following dosing).

Subjects will be randomized to a treatment sequence; i.e., to receive drug in the fed then fasted state (Sequence 1) versus fasted then fed state (Sequence 2; Figure 1). Half of the subjects in the cohort will be randomized to Sequence 1 and the other half will be randomized to Sequence 2.

Following dosing on Day 1, subjects will remain in the clinic through completion of study assessments on Day 4, returning to the clinic on Day 9 for study assessments, and Day 14 to begin the second dosing sequence on Day 15. Subjects will then be discharged from the clinic on Day 18, returning to the clinic on Day 23 for study assessments, and Day 30 for the EOS visit. There will be a 14-day (minimum) washout period between doses.

One cohort of 10 subjects is planned for P2. However, if PK results obtained are equivocal, additional subjects may be enrolled into the P2/FE cohort at the previously tested dose or an additional P2/FE cohort may be added at a different dose.

3.1.3. Part 3 (Multiple Ascending Dose)

Subjects in P3 will be randomized in a 3:1 ratio to receive either EIDD-2801 or PBO. Twice-daily dosing will be administered to subjects on Day 1 through Day 5, inclusive, and a final dose will be administered on the morning of Day 6 for collection of steady-state PK samples during waking hours. Depending on ongoing review of the safety, tolerability, and PK data, the number of days of dosing may be reduced and the dosing frequency may be changed; the dosing frequency will be no less than once-daily or no greater than three-times daily.

The proposed dose-escalation scheme is shown in Figure 1. The total daily dose will not exceed a dose shown to be safe and well-tolerated in P1. Doses in P3 may be administered in the fed state, following review of the PK data obtained from P2. Part 3 may run in parallel with P1, providing that the total daily dose to be administered does not exceed a dose already shown to be safe and well-tolerated in P1.

Subjects will remain domiciled at the site during the dosing period and until Day 9, returning to the clinic on Day 14 for completion of study assessments, and Day 20 for the EOS visit. If the number of days of dosing are reduced, the timepoints for subsequent assessments and the period for which subjects are domiciled at the clinic will similarly be changed (subjects will be domiciled for 3 days post final dose).

Three cohorts are initially planned for P3; however, up to an additional 4 cohorts may be enrolled, or the number of cohorts may be reduced if the study objectives are met. Dose levels in P3 may be repeated for collection of PBMCs, providing that the dose level did not meet the dose-escalation halting criteria.

Additional parts may be added to this study, including a SAD part in elderly healthy subjects and a study of the safety, tolerability, and PK in Japanese subjects. These additional parts are not described in this protocol, but may be added via a protocol amendment.

3.2. Rationale and Justification

3.2.1. Justification of Design

The FIH study is a typical dose-escalation study designed to provide the maximum amount of data in the minimum number of subjects. The cohort size in P1 and P3 is planned to be 8 subjects (6 active:2 PBO). This number of subjects allows for a sufficient PK analysis, considered to be important because dose extrapolation from efficacious animal models to humans will be based on exposure. The duration of participation for each subject following dosing well exceeds 5 drug half-lives (up to 9.1 hours in dogs; 5 hours in ferrets) [redacted] The FE cohort is considered to be important to maximize exposure based on fed vs. fasted condition and obtaining this information early in Phase 1 will minimize study drug dose in all future studies. This design follows the FDA guidance document on assessing FE in clinical studies.

3.2.2. Justification of Starting Dose

[redacted] The FDA guidance document *("Estimating the Maximum Safe Starting Dose in Initial Clinical Trials for Therapeutics in Adult Healthy Volunteers")* provides guidance for when a safety factor smaller than 10 may be used to calculate the starting dose:

- A smaller safety factor might also be used when toxicities produced by the therapeutic are easily monitored, reversible, predictable, and exhibit a moderate-to-shallow dose-response relationship with toxicities that are consistent across the tested species (both qualitatively and with respect to appropriately scaled dose and exposure).

- A safety factor smaller than 10 could be justified when the NOAEL was determined based on toxicity studies of longer duration compared to the proposed clinical schedule in healthy volunteers. In this case, a greater margin of safety should be built

into the NOAEL, as it was associated with a longer duration of exposure than that proposed in the clinical setting. This assumes that toxicities are cumulative, are not associated with acute peaks in therapeutic concentration (e.g., hypotension), and did not occur early in the repeat dose study.

EIDD-2801 satisfies both of these criteria as follows:

[redacted]

Importantly, given the activity of EIDD-2801 versus SARS-CoV-2 (cause of COVID-19), the Sponsor thinks it is most prudent to start with a dose that is predicted to be a safe starting dose and is also as close as possible to a potential therapeutic dose that would allow the Sponsor to move into COVID-19 patients as safely and expeditiously as possible. Based on modeling to animal data, a dose of 100 mg BID is projected to be an active dose in humans.

3.2.3. Justification of Study Population

Healthy volunteers are considered to be the appropriate population for conduct of the FIH study. Healthy volunteers without confounding medical conditions that may obscure the interpretation of AEs or affect absorption, distribution, metabolism and excretion of study drug will provide the most valuable data regarding the tolerability, safety, and plasma exposures observed and expected following single doses and multiple doses up to 10 days. EIDD-2801 is intended for eventual study in patients with potential CoV-2 infection as defined by the Centers for Disease Control and Prevention (CDC) in whom a range of AEs are expected based on the disease under study. Understanding the safety and PK profile in a normal population will better inform the use of EIDD-2801 in disease settings where complications are frequent, and AEs will need to be interpreted in context.

4. STUDY POPULATION

This study will enroll healthy volunteers; 8 subjects will be enrolled into each SAD and MAD cohort in P1 and P3, and 10 subjects will be enrolled into the FE cohort in P2.

The site is strongly encouraged to ensure that women are represented in each cohort.

4.1. Subject Inclusion Criteria

Subjects who meet all of the criteria at Screening will be eligible for study participation

1. Male or female between the ages of 18 and 60, inclusive.

2. Female subjects must be of nonchildbearing potential. Nonchildbearing potential is defined as women who:

    - are physiologically incapable of becoming pregnant including those who are surgically sterile, via hysterectomy, bilateral salpingectomy, and/or bilateral oophorectomy by reported medical history and/or medical records. Surgical sterilization to have occurred a minimum of 6 weeks, or at the PI's (or designee's) discretion, prior to Screening.

    - are post-menopausal (at least 45 years of age with amenorrhea for 12 months without an alternative medical reason with confirmatory FSH levels of ≥40 mIU/mL. The amenorrhea should not be induced by a medical condition such as anorexia nervosa, hypothyroid disease or polycystic ovarian disease, or by extreme exercise. It should not be due to concomitant medications that may have induced the amenorrhea such as oral contraceptives, hormones, gonadotropin-releasing hormones, anti-estrogens, or selective estrogen receptor modulators).

3. Male participants must agree to the use of effective contraception for study duration, i.e., from Check-in until 90 days after the EOS visit.

4. Is in generally good health as determined by medical history, PEs (at Screening and/or Check-in), vital signs, and other screening procedures.

5. Has a BMI of 18 to 30 kg/m^2.

6. Is willing and able to comply with all study procedures including providing informed consent.

4.2. Subject Exclusion Criteria

Subjects who meet any of the following criteria at Screening (unless otherwise stated) will not be eligible for participation:

1. Females who are pregnant, planning to become pregnant, or breastfeeding.
2. Has a clinically relevant acute or chronic medical condition or disease of the cardiovascular, GI, hepatic, renal, endocrine, pulmonary, neurologic, or dermatologic systems as determined by the PI (or designee).
3. Has any current or historical disease or disorder of the hematological system or significant liver disease or family history of bleeding/platelet disorders.
4. Has a history of cancer (other than basal cell or squamous cell cancer of the skin), rheumatologic disease or blood dyscrasias.
5. Has a history of GI surgery or other condition that may interfere with absorption of study drug, in the option of the PI (or designee).
6. Has a history of febrile illness within the 14 days prior to the first dose of study drug.
7. Has a positive alcohol or drug screen at Screening or the Day -1 visit or has a history of alcohol or drug abuse within the past 5 years.
8. Currently uses tobacco, nicotine or tobacco products or e-cigarettes or stopped using tobacco products within the past 3 months and/or has a positive cotinine test at Screening or Check-in.
9. Has any abnormal laboratory value of Grade 2 or higher, which is considered to be clinically significant by the PI (or designee).
10. Has a total white cell count or absolute lymphocyte count outside the normal range, or hemoglobin or platelet levels below the lower limit of normal, at Screening or Day -1.
11. Has values above the upper limit of normal for the following laboratory analytes: ALT/SGPT, alkaline phosphatase (serum), AST/SGOT, at Screening or Day -1.
12. Positive test result for HIV, HBV, or HCV.
13. Has an autoimmune disease, is immunosuppressed or is in any way immunocompromised.
14. Has any of the following:
    - QTcF >450 ms confirmed by repeat measurement

- QRS duration >110 ms confirmed by repeat measurement
- PR interval >220 ms confirmed by repeat measurements
- findings which would make QTc measurements difficult or QTc data uninterpretable
- history of additional risk factors for torsades de pointes (e.g., heart failure, hypokalemia, family history of long QT syndrome).

15. Except as noted, has used prescription drugs (other than hormone replacement therapy) within 14 days prior to the first dose of study drug unless, in the opinion of the PI (or designee), the drug will not interfere with study assessments. The following prescription drugs will be allowed:
    - Statins: Subjects who have been on a stable dose of statins for a minimum of 6 months and who have normal creatine kinase at Screening.
    - Antihypertensives: Subjects who have been on a stable dose of antihypertensives for a minimum of 6 months and have controlled blood pressure and no active heart disease.
16. Has received an experimental agent (vaccine, drug, biologic, device, blood product or medication) within 3 months prior to the first dose of study drug and agrees not to receive another experimental agent during the duration of this trial.
17. Has donated blood within 30 days, plasma within 2 weeks, or platelets within 6 weeks prior to Screening or refuses to agree to not donate blood until 3 months after the EOS visit.
18. Uses OTC medications or nutritional supplements (including, e.g., gingko biloba, aspirin at any dose, and NSAIDs) on a routine/scheduled basis and/or is unwilling to abstain from such use for the duration of the study.
19. Is unwilling to abstain from quinine containing products including quinine water, tonic water, bitter lemon, jello shots, any quinine preparations or Cinchona tree bark preparations (e.g., herbal medications containing Cinchona tree bark).
20. Has any condition that would, in the opinion of the PI (or designee), put the subject at increased risk for participation in a clinical trial or confinement in a clinical research unit.

5. STUDY MEDICATION, RANDOMIZATION AND DOSE ADMINISTRATION

5.1. Study Drug Description

EIDD-2801 and matching PBO will be supplied [redacted]

[redacted]

[redacted]

[redacted]

5.1.1. Acquisition, Formulation, Packaging and Labeling

[redacted]

All study drug will be labeled according to the regulatory requirements for investigational product.

5.1.2. Product Storage and Stability

Study drug should be stored at controlled room temperature defined as [redacted]. If excursions occur which are outside of this range, the pharmacy staff should contact Sponsor to determine the course of action. Additional stability data may be available which would allow continued use of the study drug, or study drug may need to be replaced.

5.2. Randomization

Unmasked study drug (EIDD-2801 and matching PBO) will be supplied to the study site pharmacy. The pharmacy staff will be unmasked with regards to treatment assignment. A randomization list will be provided to the pharmacy staff who will use that list to dispense masked study drug for administration to each study participant. In P1 and P3, 6 subjects will

receive EIDD-2801 while 2 subjects will be randomized to PBO. In P2, all 10 subjects in the FE cohort will receive EIDD-2801. The pharmacy staff will maintain the security of the randomization list ensuring that no study personnel outside of the pharmacy have access to identify treatment assignment. In the case that it becomes necessary to know a subject's treatment assignment, unmasking procedures will be followed as discussed in Section 8.4.

5.3. Dosage, Preparation and Administration of Study Drug

Detailed instructions for extemporaneous compounding (as necessary), dispensing and administering study drug can be found in the pharmacy manual.

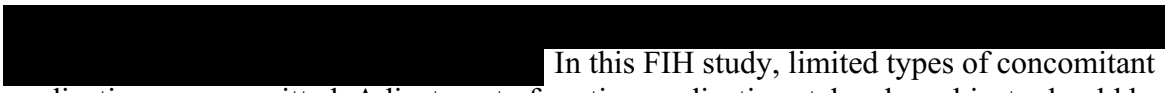
5.4. Drug Accountability

The site pharmacy must maintain records of receipt and disposition of all study drug supplied to the site by the Sponsor. The records must be maintained according to site standard operating procedures (SOPs) and should include at a minimum, receipt date, lot or batch number, amount and formulation received, , date, time and amount dispensed, subject identifier for each aliquot of study drug dispensed. In addition, the clinic staff must ensure that the time of dosing is identified for each subject.

Monitors must verify drug accountability/dispensing records during the monitoring visit.

Unused study drug must be disposed of according to the procedures described in the pharmacy manual.

5.5. Concomitant Medication/Treatments



In this FIH study, limited types of concomitant medications are permitted. Adjustment of routine medications taken by subjects should be avoided during study participation except when subject safety could be affected by lack of

adjustment. There are no restrictions on treatment medications prescribed by the PI (or designee) to be used for AEs that occur during study participation. For additional restrictions, see Section 6.1.2.

6. STUDY CONDUCT AND VISIT SCHEDULE

All study assessments will be conducted according to the Time and Events Schedule.

6.1. Study Conduct

6.1.1. Study Windows and Rounding Principles

The time schedule described in the protocol for each scheduled activity should be followed as closely as possible. However, if it is not possible and if it is not otherwise specifically contraindicated per protocol, then the time windows detailed in Table 10 are allowed without incurring a protocol deviation.

Table 10: Allowable Time Windows for Study Assessments/Visits

Protocol Specified Time	Allowable Window
PK Samples and Associated Assessments	
Predose	within 2 hours prior to dosing
<2 hours	± 5 minutes
≥2 hours to 24 hours	± 15 minutes (± 2 hours for urinalysis)
>24 hours to 48 hours	± 30 minutes
>48 hours	± 1 hour (PK) and ± 2 hours (safety)
Study Visits - Parts 1 and 2	
Day −1 to Day 4 (Part 1)	must occur on scheduled day
Day −1 to Day 4 and Day 14 to Day 18 (Part 2)	must occur on scheduled day
Day 9 (Part 1) and Days 9 and 23 (Part 2)	± 1 day
End-of-Study (EOS) Visit	± 2 days
Study Visits - Part 3	
Day −1 to Day 9	must occur on scheduled day
Day 14	± 1 day
Day 20/EOS Visit	± 2 days

6.1.2. Restrictions

Prior to arriving at the clinic for the Day -1 visit, subjects must abstain from consumption of alcoholic beverages for a minimum of 72 hours prior to Check-in. Subjects must continue to abstain from consumption of alcoholic beverages throughout clinic confinement. Subjects enrolled in P2 must continue to refrain from consuming alcoholic beverages from discharge on Day 4 through Check-in on Day 14, and then through the second clinic confinement to Day 18. After discharge on Day 4 (P1), Day 18 (P2), or Day 9 (P3), subjects must minimize consumption of alcoholic beverages (i.e., limit of up to one serving per day) until the EOS procedures have been completed.

All subjects must refrain from the following:

- consuming quinine containing products from 72 hours prior to Check-in through to completion of the EOS procedures.
- using nutraceuticals and nutritional/vitamin supplements (e.g., gingko biloba, multivitamins) from 72 hours prior to Check-in through to completion of the EOS procedures. However, vitamin supplements required by a physician are exempt from this restriction.
- taking OTC analgesics including aspirin (any dose) and NSAIDS from 72 hours before Check-in until completion of EOS study procedures unless prescribed by the PI (or designee).
- use of tobacco, nicotine or tobacco products, or e-cigarettes from 3 months prior to Screening until the EOS visit.
- strenuous exercise from 7 days before Check-in until the EOS visit. Subjects will otherwise maintain their normal level of physical activity during this time (i.e., will not begin a new exercise program nor participate in any unusually strenuous physical exertion).
- Female subjects must not donate eggs/ovum from the time of Check-in until 3 months after the EOS visit.

All subjects in P1 should be dosed in the fasted state. Subjects should fast overnight for a minimum of 10 hours prior to dosing in the morning on Day 1. Following dosing, subjects may have water after 2 hours and food beginning 4 hours postdose. For subjects to be dosed in the fed state, subjects should have a high-fat breakfast as defined in the FDA guidance. Subjects must complete the meal within 30 minutes of starting the meal and should be dosed after 30 minutes of starting the meal. Doses in P3 may be administered in the fed state, following review of the PK data from P2.

6.2. Screening

6.2.1. Screening Visit

Subjects who meet preliminary pre-screening criteria (as defined by the site) and are interested in participating in the study will arrive at the study site for administration of informed consent according to site standard operating procedures (SOPs). After the subject has signed and dated the ICF, screening procedures can begin. Assessments and procedures should be conducted as shown in the Time and Events Schedule Screening may be conducted as early as 28 days prior to dosing.

6.2.2. Retesting Procedures

In the case where a parameter is outside of the acceptable limits for enrollment, measurement of the parameter can be repeated one time if both the following are met:

- In the opinion of the PI (or designee), the parameter value does not represent a chronic condition that otherwise will preclude the volunteer from enrolling in the study; and
- In the opinion of the PI (or designee), the abnormality is not likely to recur.

6.2.3. Study Visits

Subjects who satisfy entry criteria will return to the clinic for the Day -1 visit. Following review of I/E criteria, subjects who still qualify will be checked into the clinic and enrolled into the study. Following enrollment, clinical chemistry, hematology and urine samples will be collected to determine baseline values. Based on site standard practices, alternate subjects will also be enrolled into the study in case one of the selected subjects cannot be dosed. If all subjects can be dosed on the morning of Day 1, the alternates will be released and may be enrolled into subsequent cohorts. All assessments and procedures will be performed according to the Time and Events Schedule.

6.2.4. Part 1 (Single Ascending Dose)

Subjects will remain in the clinic through completion of study procedures on Day 4, returning to the clinic for procedures on Day 9.

6.2.5. Part 2 (Food-Effect)

Subjects will remain in the clinic through completion of study procedures on Day 4, returning to the clinic on Day 9 for study assessments, and Day 14 to begin the second dosing sequence on Day 15. Subjects will then be discharged from the clinic on Day 18, returning to the clinic on Day 23 for study assessments.

6.2.6. Part 3 (Multiple Ascending Dose)

Subjects will remain in the clinic through dosing and completion of study procedures on Day 9, returning to the clinic on Day 14 for study assessments. Depending on ongoing review of the safety, tolerability, and PK data, the number of days of dosing may be reduced. If the number of days of dosing are reduced, the timepoints for subsequent assessments and the period for which subjects are domiciled at the clinic will similarly be changed (subjects will be domiciled for 3 days post final dose).

6.3. Safety Follow-up and End-of-Study Visit

Subjects in P1 will return to the clinic for the EOS visit on Day 15. Subjects in the FE cohort (P2) will return for the EOS visit on Day 30. Subjects in P3 will return for the EOS visit on

Day 20. Subjects with drug-related AEs at the EOS visit will be followed as discussed in Section 8.2.1.

6.4. Subject Withdrawal and Replacement

As this is a small study with a limited number of subjects per cohort, it is critical that all subjects complete the study including postdose study assessments. Site personnel should emphasize this to study subjects at the time of informed consent so that subjects will understand this fact before agreeing to participate in the study.

Subjects may voluntarily withdraw their consent for study participation at any time without penalty or loss of benefits to which they are otherwise entitled. An PI (or designee) may also withdraw a subject from receiving study drug or participation in the study for any reason. Subjects who withdraw or are withdrawn from the study should undergo withdrawal procedures as discussed below. These procedures would include follow-up safety evaluations.

6.4.1. Reasons for Withdrawal

If a subject withdraws or is withdrawn from the study, the primary consideration must be the health and welfare of the subject. The reasons for withdrawal might include but are not limited to the following:

- Subject no longer meets eligibility criteria including subject withdraws consent from study participation (with or without a reason)
- Subject becomes noncompliant
- Medical disease or condition, or new clinical finding(s) for which continued participation, in the opinion of the PI (or designee) might compromise the safety of the subject, interfere with the subject's successful completion of this study, or interfere with the evaluation of responses
- Subject Lost-to-Follow-up
- Subject becomes pregnant, if applicable
- Determined by a physician's discretion to require additional therapy not indicated in the protocol to ensure subject's health and well-being (or treatment failure, if applicable)

The PI should be explicit regarding study follow-up (e.g. safety follow-up) that might be carried out. If the subject consents, every attempt will be made to follow all AEs through resolution, return to baseline, or until stabilized with sequelae for a maximum of 30 days following discontinuation. The procedures that collect safety data for the purposes of research must be inclusive in the original ICF or the PI may seek subsequent informed consent using an EC-approved ICF with the revised procedures.

The PI will inform the subject that already collected data will be retained and analyzed even if the subject withdraws from this study.

6.4.2. Handling of Withdrawals

Subjects who withdraw from the study prior to receiving study drug (i.e., on Day -1 or before dosing on Day 1) will be discharged from the clinic and followed only if AEs are present which occurred due to participation in the study (e.g., AE resulting from a study procedure). In this case, the subject should be followed until the AE resolves or the PI determines that the AE has stabilized.

Subjects who withdraw from the study after receiving study drug should have EOS assessments at the time of withdrawal or as quickly thereafter as possible.

Subjects who do not return for follow-up procedures on Days 9/23 or 15/30 (P1 and P2), or Days 14 or 20 (P3) will be contacted by the site at least 3 times using the subject's preferred method of communication (as determined at Check-in). If the site is unable to contact the person, then a certified letter will be sent. If the subject still cannot be reached or refuses to come back to the clinic after all attempts, the subject will be considered Lost-to-Follow-up and withdrawn from the study.

6.4.3. Documentation of Withdrawals

If a subject withdraws or is withdrawn prior to completion of the study, the reason for this decision must be recorded in the CRF. If the subject is Lost-to-Follow-up, the site should document the attempts to contact the subject in the source documents. If the subject has an ongoing AE at the time of withdrawal, then the AE should be followed as detailed in Section 8.2.1.

6.4.4. Subject Replacement

If a subject withdraws from the study prior to receiving study drug, the subject will be replaced. In this case, designated alternate subjects, if available, will be first in line to replace the withdrawn subject.

If a subject withdraws from the study after receiving study drug, then the decision to replace the subject will be made by the PI (or designee) in consultation with the Sponsor. Factors to consider will be the timing postdose of withdrawal and the number of safety and PK assessments completed prior to withdrawal. Subjects who are withdrawn because of an AE related to the study drug will not be replaced.

6.5. Unscheduled Visit(s)

If a subject experiences an AE after discharge from the clinic but prior to the EOS visit, the subject should be instructed to call the site. Based on the issue, the PI (or designee) may request that the subject return to the site for an unscheduled visit. In this case, procedures/assessments

should be conducted as deemed appropriate for the situation by the PI (or designee). The visit should be recorded in the unscheduled visit page of the CRF.

7. STUDY PROCEDURES/EVALUATIONS

7.1. Demography and Medical History

Demographics including age, gender, race, ethnicity, and medical history will be recorded for each subject. All significant medical history should be recorded. In general, significant medical history should include all ongoing events and all events occurring within the last 6 months. Clinically relevant or clinically significant events occurring greater than 6 months ago should be recorded. All surgeries occurring in adulthood should be recorded. If surgeries occurred more than 2 years ago, then only the year needs to be recorded on the CRF.

7.2. Clinical Evaluations

7.2.1. Physical Examinations

The PE will be performed by the PI or a designee that is licensed to perform a PE per local requirements. The initial PE performed at Screening and the final PE conducted at the EOS visit will include examination of all pertinent body systems as defined by the site standard PE body systems (general appearance, HEENT, lymphatic, cardiovascular, respiratory, GI, musculoskeletal, neurological, dermatological).

Subsequent PEs will be performed as shown in the Time and Events Schedule and will be targeted to any new signs or symptoms, any change from the previous status or any body system deemed pertinent to that subject by the PI (or designee). Clinically significant abnormalities should be recorded in the CRF; those occurring prior to dosing will be included in medical history unless the abnormality was the direct result of study participation.

7.2.2. Vital Sign Measurements and ECGs

Vital signs include pulse rate, respiratory rate, blood pressure, and body temperature. Vital signs should be measured after the subject has been supine for a minimum of 5 minutes. Site standard ranges will be used for determining any out-of-range values.

Height and weight should be measured, and BMI calculated at Screening as indicated in the Time and Events Schedule.

Resting 12-lead ECGs should be recorded at the visits indicated in the Time and Events Schedule after the subject has been supine for a minimum of 5 minutes. The PI (or designee) will evaluate the ECG tracings to determine if there are out-of-range values; if out-or-range values are detected, the PI (or designee) will determine if they are clinically significant. Site standard ranges will be used to determine if any parameters are considered out-of-range. At the discretion of the PI (or designee), the ECG may be repeated if erroneous readings are suspected.

7.2.2.1. Continuous 12-lead ECG Monitoring (Part 3 Only)

Continuous 12-lead ECG monitoring using a digital recorder will take place at the times indicated in the Time and Events Schedule, in P3 only.

All continuous 12-lead ECG data collected will be archived without extraction or analysis and will not be reported in the scope of this study.

Subjects will be supine for at least 10 minutes before the extraction timepoint and for 5 minutes from the start of each extraction timepoint (each extraction will last for 5 minutes). Environmental distractions (e.g., television, radio, conversation) should be avoided during the pre-ECG resting period and during ECG recording.

When coinciding, PK sampling should always be performed after the ECG extraction time window. If a separate ECG machine is being used for safety assessments, that machine should be in place prior to the extraction window to permit safety ECGs to be recorded irrespective of the extraction window. If the machine is not in place prior to the extraction window, safety ECGs must be recorded after the extraction window. If an integral system is used, safety ECGs may be recorded irrespective of the extraction window.

7.2.3. Adverse Events and Concomitant Medications

Adverse Events: The PI is responsible for identifying and documenting events meeting the definition of an AE or SAE (Section 8.1). Once each day while the subject is in the clinic and once during each out-patient visit, the PI (or designee) should inquire about the occurrence of AEs. The following are examples of open-ended questions that may be used to obtain this information: *"How are you feeling?"; "Have you had any medical problems recently?"; "Have you taken any new medicines since your last visit/assessment?"*

All AEs and SAEs must be documented in the source documents and recorded in the CRF.

Concomitant Medications: All medications (prescription or over-the-counter), nutritional supplements, and nutraceuticals taken by the subject from 30 days prior to dosing through the EOS visit must be recorded in the CRF. Medication information should include indication, dose, frequency, and route of administration. Any medication taken for an AE/SAE should be documented as such. Refer to Section 5.5 for additional information.

7.3. Laboratory Evaluations

The laboratory will perform standard routine testing, and processing of all blood samples. For the entire study, the amount of blood collected from any one subject will not exceed 500 mL.

7.3.1. Routine Laboratory Panels

Blood and urine samples will be collected at the times indicated in the Time and Events Schedule. The analytes shown in the table below (Table 11) will be assessed.

If a method to determine COVID-19 status becomes readily available, subjects may be tested at Screening and Check-in to confirm they are not positive for COVID-19 prior to dosing.

Table 11: Laboratory Evaluations

CHEMISTRY PANEL Alanine Aminotransferase (ALT/SGPT) Albumin, Serum Albumin/Globulin (A/G) Ratio (calculation) Alkaline Phosphatase, Serum Amylase Aspartate Aminotransferase (AST/SGOT) Bilirubin, Total and Direct BUN BUN/Creatinine Ratio (calculation) Calcium, Serum Creatinine, Serum Creatinine Kinase (CK) Gamma Glutamyl Transferase (GGT) Lactate Dehydrogenase (LDH) Uric Acid Electrolyte Panel (Na+, K+, Cl-, Bicarb.) Phosphorus Globulin, Total Glucose, Serum Lipase Protein, Total, Serum	**HEMATOLOGY PANEL** Hemoglobin Hematocrit White Blood Cell Count with differential (absolute and percentage) Red Blood Count Prothrombin Time (PT)/Partial Prothrombin Time (PTT) and International Normalized Ratio (INR) Mean cell hemoglobin (MCH), mean cell hemoglobin concentration (MCHC), mean cell volume (MCV), red cell distribution width (RDW; may be a Grade 1 abnormality) Platelets
	ADDITIONAL ASSESSMENTS Virology: Human Immunodeficiency Virus (HIV) serology, Hepatitis B Virus (HBV; Surface Antigen [HBsAg]), Hepatitis C Virus (HCV) Follicle-Stimulating Hormone (FSH; as applicable)
	PREGNANCY TEST Serum Pregnancy Test Urine Dipstick (optional blood follow-up)
	DRUG SCREENING Serum/urinalysis (per site SOP) Cotinine Urine Dipstick Alcohol Breathalyzer
	ROUTINE URINALYSIS Bilirubin Color and appearance Glucose Ketones Leukocytes Microscopic (including red blood cells [RBCs] and white blood cells [WBCs]) Nitrite Occult blood pH Protein Specific Gravity Urobilinogen

7.3.2. Pharmacokinetic Sampling

Samples for PK analysis will be collected as shown in the Time and Events Schedule. All samples will be analyzed to define the PK parameters for prodrug EIDD-2801 and the active parent, EIDD-1931. In order to preserve EIDD-2801, special handling procedures will be put in place. These procedures will be documented in the laboratory manual for the study.

Depending on ongoing review of the PK data, timepoints may be modified or removed, and up to 3 additional samples (in each period for the food-effect) may be added.

7.3.3. Peripheral Blood Mononuclear Cell Collection

Peripheral blood mononuclear cells will be collected in P2 and P3. Blood will be collected into specialized tubes and processed according to procedures described in the laboratory manual.

Depending on ongoing review of the data, timepoints may be modified or removed, and up to 2 additional samples may be added.

7.3.4. Urine Collection

Urine will be collected over the time periods noted in the Time and Events Schedule. Samples will be collected for routine urinalysis and PK analysis according to site standard practices and as described in the laboratory manual.

8. SAFETY MONITORING, MANAGEMENT AND REPORTING

8.1. Definitions

8.1.1. Adverse Events

An AE is 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 an abnormal laboratory finding), symptom, or disease temporally associated with the use of a medicinal (investigational) product, whether or not related to the medicinal (investigational) product.

Abnormal clinical laboratory findings (e.g. clinical chemistry, hematology) or other abnormal assessments that are judged by the PI (or designee) as clinically significant will be recorded as AEs or SAEs if they meet the definitions of an AE or an SAE as defined in this Section 8.1.2. Disease specific signs and symptoms which were ongoing prior to study entry will not be considered AEs unless they worsen (e.g. increase in frequency or severity) unexpectedly during the course of the trial.

8.1.2. Serious Adverse Events

An AE or suspected adverse reaction is considered "serious" if, in the view of either the PI (or designee) or Sponsor, it results in any of the following outcomes:

- Death,
- A life-threatening AE: An AE or suspected adverse reaction is considered "life-threatening" if, in the view of either the PI or Sponsor, its occurrence places the patient or subject at immediate risk of death. It does not include an AE or suspected adverse reaction 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 hospitalizations may be considered serious when, based upon appropriate medical judgment they may jeopardize the patient or 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.

Suspected adverse reaction means any AE for which there is a reasonable possibility that the drug caused the AE. "Reasonable possibility" means there is evidence to suggest a causal relationship between the drug and the adverse event. Suspected adverse reaction implies a lesser degree of certainty about causality than adverse reaction, which means any adverse event caused by a drug.

An AE or suspected adverse reaction is considered "unexpected" if

- It is not listed in the IB or is not listed at the specificity or severity that has been observed; or, if an IB is not required or available,
- Is not consistent with the risk information described in the general investigational plan or elsewhere in the current application as amended. For example, under this definition, hepatic necrosis would be unexpected (by virtue of greater severity) if the IB referred only to elevated hepatic enzymes or hepatitis. Similarly, cerebral thromboembolism and cerebral vasculitis would be unexpected (by virtue of greater specificity) if the IB listed only cerebral vascular accidents.
- "Unexpected" as used in this definition, also refers to AE or suspected adverse reactions that are mentioned in the IB as occurring with a class of drugs or as anticipated from the pharmacological properties of the drug, but are not specifically mentioned as occurring with the particular drug under investigation.

As of the date of this protocol, there are no expected events listed in the current version of the IB; therefore, all AEs will be considered unexpected until such a time that the reference safety information in the IB is updated with any identified, expected events.

8.2. Documenting Adverse Events

8.2.1. Timeframe for Collection and Follow-up of AEs/SAEs

All AEs/SAEs will be collected from the time of the first study drug administration until the subject has completed the EOS visit and been discontinued from the study. This includes subjects who discontinue early. Events considered related to study drug will be followed as noted:

- AEs that are related to study drug and continue beyond the normal collection period (i.e., are ongoing at the time a subject exits the study) will be followed until resolution, return to baseline or until stabilized with sequelae for a maximum of 30 days following discontinuation. After 30 days, the AE will be closed, and the outcome noted (see Table 12).
- SAEs that are related to study drug and continue beyond the normal collection period

(i.e., are ongoing at the time a subject exits the study) will be followed until resolution, return to baseline or until stabilized with sequelae.

- Serious AEs that are reported to the site within 30 days after the subject has been discontinued from the study (i.e., completed the EOS visit) will be recorded. Those that are considered related to study drug will be followed as noted in the bullet above.

Note that all events which occurred prior to dosing with study drug should be recorded as medical history unless the event is directly related to study procedures.

8.2.2. Recording of Adverse Events/Serious Adverse Events

AEs/SAEs must be recorded in the CRF as indicated in the CRF completion instructions. Information to be collected includes event description, time of onset, clinician's assessment of severity (Section 8.2.3), relationship to study drug (Section 8.2.4), outcome (Section 8.2.5), and time of resolution/stabilization of the event. All AEs occurring while on study must be documented appropriately regardless of relationship and will be followed to adequate resolution as described above (Section 8.2.1). All SAEs will be recorded as noted above; SAEs reported to the site within 30 days following the EOS visit will also be recorded. All SAEs must be entered onto the SAE form and reported as discussed below (Section 8.3.1).

If an AE changes in severity, the highest severity will be recorded in the CRF. AEs characterized as intermittent require documentation of onset and duration of each episode.

8.2.3. Assessing Severity of Adverse Events

All AEs/SAEs will be assessed by the PI or those with the training and authority to make a medical judgment. AEs/SAEs will be graded according to the DMID Toxicity Grading Scale. For any AEs not specifically listed in the tables, the following guidelines should be used to grade severity:

- **Mild** (Grade 1); asymptomatic or mild symptoms; clinical or diagnostic observations only; intervention not indicated.
- **Moderate** (Grade 2); minimal, local or noninvasive intervention indicated; limiting age-appropriate instrumental activities of daily living.
- **Severe** (Grade 3); medically significant but not immediately life-threatening; hospitalization or prolongation of hospitalization indicated; disabling; limiting self-care activities of daily living.
    - **Life-threatening**; life-threatening consequences; urgent intervention indicated.
    - **Death**; death related to AE.

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

8.2.4. Relationship to Study Drug

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

- **Definite –** The AE is clearly related to the study drug**.**
- **Probable –** The AE is likely related to the study drug.
- **Possible –** The AE may be related to the study drug.
- **Unlikely –** The AE is doubtfully related to the study drug.
- **Unrelated –** The AE is clearly NOT related to the study drug.

8.2.5. Classifying Adverse Event Outcome

All AEs/SAEs in the study must be assigned an outcome by site staff. The outcome will be included on the AE CRF. Possible outcomes are shown below:

Table 12: Adverse Event Outcomes

Outcome	Description
Recovered / Resolved	AE resolved with no residual signs or symptoms; an event is considered resolved if it returns to baseline (pretreatment) values.
Recovered / Resolved with sequelae	AE stabilized but residual signs or symptoms remain; this includes stabilization of an event/condition with the expectation that it will remain chronic.
Not Recovered / Not Resolved	AE remains ongoing AND no or only minimal improvement has occurred.
Ongoing	AE has not yet resolved, but continues to improve/resolve and complete resolution is expected over time.
Fatal	Outcome of the AE is death.
Unknown	AE outcome is not known; usually because the subject has been Lost-to-Follow-up.

8.3. Reporting Procedures

8.3.1. Serious Adverse Events

The PI or clinical site personnel should notify Covance Drug Safety Services (DSS) of all SAEs, regardless of relationship to the investigational drug, within 24 hours of clinical site personnel becoming aware of the event. The PI (or designee) will provide the initial notification by sending a completed "SAE Notification Form," which must include the PI's (or designee's) assessment of the relationship of the event to investigational drug and must be signed by the PI. Follow-up information, or new information regarding an ongoing SAE, must be provided promptly to Covance DSS.

The responsibilities of Covance DSS include the following:

- Prepare an AE reporting plan prior to the start of the study. Where this plan differs from the applicable site standard operating procedure on SAE reporting, the AE reporting plan will always take precedence.

- Receive and review SAE report forms from the site and inform the Sponsor of the SAE within 1 working day of the initial notification to DSS. Drug Safety Services will delete any information from the SAE report forms that may identify the subject.

- Write case narratives and enter the case into Covance's safety database as defined in the AE reporting plan.

- Produce appropriate reports of all Suspected Unexpected Serious Adverse Reactions and forward to the EC, Medicines and Healthcare Products Regulatory Agency, PI's (or designee's), and the Sponsor within the timeframes stipulated in the Clinical Trials Directive Guideline (ENTR/CT 3).

The responsibility for reporting SAEs will be transferred to the Sponsor 28 days after the end of the study.

8.3.2. Pregnancy

Subjects in this study must be of non-childbearing potential. However, should a pregnancy occur any time from informed consent to the EOS visit, subjects must immediately report the event to the clinical site which in turn must immediately report the pregnancy to the Sponsor or their designee. The subject will be followed until the end of the pregnancy. A separate ICF will be used for consenting for follow-up pregnancy activities. Pregnancy will not be considered an AE unless deemed likely related to study drug. Pregnancy will not be considered an SAE unless there is an associated SAE. A spontaneous abortion (miscarriage) or abnormal outcome (including congenital anomalies) will be reported as an SAE.

8.4. Unmasking Treatment Assignment

The following controls will be employed to maintain the double-blind status of the study:

- Placebo will be identical in appearance to the EIDD-2801.
- The PI and other members of staff involved with the study will remain blinded to the treatment randomization code in P1 and P3.
- Interim bioanalytical data will be provided in a blinded manner.

To maintain the blind, the PI will be provided with a sealed randomization code for each subject in P1 and P3, containing details of their treatment. These individual sealed envelopes will be kept in a limited access area that is accessible 24 hours a day. In order to manage subject safety or to support dose-escalation decisions (in the event of possibly treatment-related SAEs or severe AEs), the decision to unblind resides solely with the PI. Whenever possible and providing it does not interfere with or delay any decision in the best interest of the subject, the PI will discuss the intended code-break with the Sponsor. If it becomes necessary to break the code during the study, the date, time, and reason will be recorded in the subject's source data and on the individual envelope and will be witnessed by a second person.

The pharmacy and bioanalytical lab will have access to the treatment randomization and will be unblinded. Pharmacokinetic personnel may be unblinded to perform interim PK analysis and to ensure that PK data are provided in a blinded manner for dose-escalation decisions.

9. DOSE-ESCALATION AND HALTING RULES

9.1. Guidelines for Dose-Escalation

Doses will be administered in an escalating manner following satisfactory review by the Sponsor and PI of the blinded safety and tolerability data up to 72 hours post final dose. [redacted] As PK data become available, these data will be reviewed and may be used to assist in dose selection.

Doses may be reduced and may be lower than the starting dose. There will be a minimum of 4 days between dose escalations to allow sufficient time for an adequate safety review. [redacted]

Integrated data from all applicable sites will be used to make dose-escalation decisions.

Dose-escalation in P1 and P3 will only occur if data from a minimum of 6 subjects (across all sites) have been reviewed from the previous lower dose group, such that data from a minimum of 4 subjects (across all sites) who have received EIDD-2801 will be used to make the dose-escalation decision.

The justification for this is as follows:

- The study treatment is of a known pharmacological class for which the on-target effects in humans are well characterized. Based upon nonclinical data, no clinically important off target effects are expected within the proposed dose range.

- A minimum of 4 subjects receiving the active drug is considered sufficient to characterize the safety profile of EIDD-2801.

Between each dose-escalation, the PI will review all available data in a blinded manner to ensure it is safe to proceed with the planned dose-escalation. The results from all available safety assessments will be sent to the Sponsor prior to the start of each successive group/treatment period. Any clinically significant results will be discussed with the Sponsor before dose-escalation continues. Interim PK data may also be reviewed in terms of dose-escalation and to confirm that the study design remains appropriate. In the event of a disagreement between Sponsor and PI on the dose-escalation decision, the most conservative decision will be upheld.

Dose levels will not increase by more than 3-fold for predicted nonpharmacologically active dose levels and by 2-fold for predicted pharmacologically active dose levels. The highest total daily dose to be studied under this protocol will not exceed 800 mg (Section 9.2).

Dose levels in P3 may be repeated for collection of PBMCs, providing that the dose level did not meet the dose-escalation halting criteria.

9.2. Rationale for Highest Dose

9.2.1. Clinical Safety

At the time of this amendment, 24 healthy subjects were administered a single dose of 50, 100, or 200 mg EIDD-2801 or placebo in P1. There were 6 males and 2 females at each dose level, and the age range was 22 to 60 years. No clinically significant abnormalities or changes from baseline were reported for vital signs, ECGs, or PEs.

Thirteen treatment-emergent AEs were reported across the 3 dose levels; 11 of these were mild and 2 were moderate in severity. One AE was considered to be probably related, 6 were possibly related, and 6 were unlikely related to EIDD-2801.

The most frequently reported AEs were headache (4 reports), aching legs, and nausea (2 reports each).

- Two of the reports of headache (1 at 100mg and 1 at 200mg) were considered possibly related to EIDD-2801 because they started within 2 hours of dosing; 1 was mild in severity (200 mg) and did not require treatment, and 1 was moderate in severity (100 mg), which resolved after a single dose of 1 g acetaminophen. Both headaches resolved within 24 hours.

- The two AEs of aching legs were considered unlikely related to EIDD-2801.

- At 100 mg, 1 subject reported nausea that was considered possibly related to EIDD-2801 because it started approximately 4 hours after dosing; this event was moderate in severity and resolved approximately 40 minutes after onset following a single episode of vomiting. At 200 mg, 1 subject reported nausea that was considered unlikely related to EIDD-2801 because it started > 36 hours after dosing.

- Other AEs of interest included 1 report of each of loose stools and abdominal pain; both were mild in severity and reported by the same subject at the 200 mg dose level. The loose stools were considered probably related to EIDD-2801; they started approximately 13.5 hours after dosing and resolved 3 days after dosing. The abdominal pain started approximately 24 hours after dosing and resolved after 4.5 hours.

Laboratory safety data have been reviewed through Day 9 at the 50 and 100 mg dose levels and through Day 4 at the 200 mg dose level. There were no clinically significant abnormalities or changes from baseline. Specifically, there were no reductions in platelets or absolute lymphocyte counts.

9.2.2. Pharmacokinetics

Pharmacokinetic data have been reviewed post dosing for the 50, 100, and 200 mg dose levels. No concentrations of EIDD-2801 (the ester prodrug) were quantifiable in any of the samples at either dose level, thus PK parameters were not calculable for EIDD-2801. EIDD-1931 (the active drug) appeared rapidly in plasma, with a median t_{max} of 1 hour at all dose levels. EIDD-1931 concentrations increased in a dose-proportional manner between 50 and 200 mg. EIDD-1931 concentrations declined in a monophasic manner and remained quantifiable until 6 hours postdose for the 50 and 100 mg doses, and until between 6 and 9 hours for the 200 mg dose. The geometric mean terminal elimination half-life was approximately 1.0 hour at all dose levels. At 400 mg, geometric mean systemic exposure is predicted to achieve an approximate area under the concentration versus time curve from time zero to 24 hours postdose (AUC_{0-24}) of 3593 ng•h/mL and C_{max} of 1889 ng/mL [redacted]

9.2.3. Rationale to Exceed the Maximum Single Dose Limit of 200 mg

[redacted]

Based on the clinical data collected to date, it is considered acceptable to exceed 200 mg because EIDD-2801 has been well tolerated and the PK behaved in a predictable manner.

[redacted]

Ongoing modelling suggests that the therapeutic dose level required to treat highly pathogenic CoV may be 200 mg BID. In order to escalate to this dose level in P3, a dose level of 400 mg must be shown to be safe and well tolerated in P1. Additionally, in order to allow for unpredicted differences in PK at higher dose levels compared with that seen at the dose levels already evaluated, dose levels of 600 and 800 mg are planned for P1; this will allow escalation to a maximum of 400 mg BID in P3 if required to achieve therapeutic exposures.

[redacted]

9.3. Dose-Escalation Halting Rules

For Group 1 in P1, sentinel dosing will occur whereby 2 subjects (1 active and 1 PBO) will be dosed on 1 day and, providing no safety concerns arise, the remaining 6 subjects will be dosed after at least 24 hours.

The study will be halted (i.e., no further dosing will occur) if one or more subjects experience an SAE that is considered to be related to EIDD-2801 or 2 or more subjects in the same group experience severe AEs that are considered to be related to EIDD-2801. If, following an internal safety review, the Sponsor deems it appropriate to restart the study, this can be done following approval of a substantial protocol amendment.

Additionally, if 2 or more subjects experience a reduction of platelets greater than 50% over baseline levels or a reduction in lymphocytes (absolute count) to Grade 3/severe levels without an alternate cause, the PI and Sponsor will review the data. If, after review of unblinded data, it is determined that these events are probably related to study drug, no further dose escalations will occur. In this case, additional cohorts may be enrolled at a lower dose.

Dose-escalation will stop if clinically relevant signs or symptoms of similar nature occur in 2 or more subjects in a group that, in the opinion of the PI, warrant stopping of dose-escalation.

10. STATISTICAL CONSIDERATIONS

10.1. General Considerations

All summaries will be provided by study part and treatment. Continuous variables will be summarized using descriptive statistics including number of observations (n), mean, standard deviation, minimum (min), median (med), and maximum (max). Categorical variables will be summarized using frequency counts and percentages. Note that PBO data from each dose level in Parts 1 and 3 will be combined into one PBO group.

No missing data imputation will be performed.

Subject listings will be provided for all the data collected during the study period.

Specific information about the statistical analysis will be provided in a SAP that will be reviewed and approved by the Sponsor and will be finalized before final database lock. If there is a discrepancy between the methods described in the protocol and final approved SAP, the SAP will take precedence.

10.2. Sample Size Considerations

No formal sample size calculation was conducted. The sample size of 8 per cohort (6 active: 2 PBO) for the SAD and MAD cohorts is considered adequate for a Phase 1 FIH study. The sample size of 10 subjects (all administered EIDD-2801) is in accordance with FDA guidelines for sample size in FE studies.

10.3. Analysis Populations

Safety Population: All subjects who receive at least one dose of study drug.

Pharmacokinetic Population: All subjects receiving study drug who comply sufficiently with protocol requirements (i.e., no major protocol violations) and for whom a sufficient number of analyzable PK samples have been obtained to permit the determination of the PK profile for EIDD-2801/EIDD-1931 and the statistical analysis of the results. Subjects who experience an AE of vomiting before 2× the median t_{max} of the group may be excluded from the PK population.

10.4. Analysis of Safety Data

All safety analyses will be performed on the Safety Population as defined in Section 10.3. Safety will be assessed on the basis of AEs, clinical laboratory data, vital signs, ECG parameters, and PEs.

10.4.1. Extent of Exposure

Dosing data will be listed by study part.

10.4.2. Adverse Events

Adverse events will be coded using Medical Dictionary for Regulatory Activities (MedDRA) preferred term and system organ classification. Any events reported after the initiation of study treatment and through the EOS are defined as treatment-emergent. The occurrence of treatment-emergent AEs will be summarized using MedDRA preferred terms, system organ classifications, and severity. Separate summaries of treatment-emergent SAEs and AEs considered related to study treatment and AEs leading to study treatment discontinuation will be generated. All AEs will be listed for individual subjects showing both verbatim and preferred terms.

10.4.3. Clinical Laboratory Results

Laboratory abnormalities will be graded according to the DMID Toxicity Grading Scale. Any graded abnormality that occurs following the initiation of study treatment and represents at least a one-grade increase from the baseline assessment is defined as treatment-emergent. The number and percentage of subjects experiencing treatment-emergent graded toxicities will be summarized. Raw values and mean changes from baseline in clinical laboratory measures will be summarized.

Listings of the clinical laboratory test results will be provided. Abnormal laboratory values will be flagged in the listings.

10.4.4. Other Parameters

Individual data for ECG parameters and vital sign measurements will be listed by subject and time point and summarized for each treatment. Individual data for PE will be listed by subject and time point.

Concomitant medications will be coded using the World Health Organization-Drug Dictionary (WHO-DD) and summarized.

10.5. Analysis of Pharmacokinetic Data

Pharmacokinetic analysis as defined in the SAP will be conducted using the PK population defined in Section 10.3. In the event of discrepancies between analyses described in the SAP and this clinical study protocol, the SAP will supersede the protocol.

- All samples will be analyzed and all concentrations listed.
- Descriptive statistics will be performed for all time points available, with the exclusion of subjects who had any significant protocol deviation.
- Pharmacokinetic parameters will be derived where possible for all subjects. Data from subjects with incomplete profiles (missed blood draws, lost samples, samples unable to be quantified) may be used if PK parameters can be estimated using the remaining data points.

- Descriptive statistics will be performed on all parameters available, and any missing parameters will be flagged.

Plasma concentration data for EIDD-2801 and EIDD-1931 will be listed for individual subjects and summarized by study part and treatment. Individual and mean plasma concentration versus time plots for EIDD-2801 and EIDD-1931 will be provided. Urine concentration data for EIDD-2801 and EIDD-1931 will be listed.

Plasma PK parameters of EIDD-2801 and EIDD-1931 for each subject will be estimated over the sampling interval using noncompartmental analysis and summarized by study part and treatment using descriptive statistics. Actual blood sampling times will be used for plasma PK analysis.

Urine PK parameters of EIDD-2801 and EIDD-1931 will also be analyzed and summarized when possible. The PK parameters that will be estimated are listed in the table below.

Table 13: Pharmacokinetic Parameters

Plasma PK Parameter	Description
AUC_{last}	The area under the plasma concentration-time curve, from time 0 to the last measurable non-zero concentration, as calculated by the linear up/log down trapezoidal method (P1 and P2 only).
$AUC_{0\text{-}inf}$	The area under the plasma concentration-time curve from time 0 extrapolated to infinity. $AUC_{0\text{-}inf}$ is calculated as the sum of AUC_{last} plus the ratio of the last measurable plasma concentration to the elimination rate constant (λz) (P1, P2, and Day 1 dose of P3).
C_{max}	Maximum observed concentration.
t_{max}	Time to reach C_{max}. If the maximum value occurs at more than one time point, t_{max} is defined as the first time point with this value.
λz	Apparent terminal elimination rate constant; represents the fraction of medication eliminated per unit time.
t½	Apparent terminal elimination half-life of medication, calculated as 0.693/λz.
Vz/F	Apparent volume of distribution (EIDD-2801 only).
CL/F	Apparent oral drug clearance (EIDD-2801 only).
C_{trough}	Trough concentration (P3 only).
AUC_{τ}	The area under the plasma concentration-time curve during a dosing interval (P3 only).
$RA_{AUC\tau}$	Observed accumulation ratio based on AUC_{τ} (P3 only).
RA_{Cmax}	Observed accumulation ratio based on C_{max} (P3 only).
Urine PK Parameter	**Description**
A_e	Amount excreted in urine over the sampling interval.
F_e	Fraction of drug excreted in the urine over sampling interval.
CL_R	Renal clearance, $CL_R = A_e/AUC$ where both A_e and AUC are determined over co-incident time ranges after dosing.

Additional PK parameters may be analyzed as appropriate.

10.5.1. Statistical Analysis of Pharmacokinetic Data

10.5.1.1. Dose Proportionality

In P1, dose-proportionality for PK parameters of EIDD-2801 and EIDD-1931 ($AUC_{0\text{-}inf}$, AUC_{last}, and C_{max}) will be assessed by the power model. The slope and the associated 90% CI based on the power model will be reported.

In Part 3 on Day 6 dose-proportionality for PK parameters of EIDD-2801 and EIDD-1931 (AUC_{τ}, and C_{max}) will be assessed by the power model. The slope and the associated 90% CI based on the power model will be reported.

10.5.1.2. Food-Effect

To assess the effect of food on the PK of EIDD-2801 and EIDD-1931, natural log transformed PK parameters from P2 ($AUC_{0\text{-}inf}$, AUC_{last}, and C_{max}) will be analyzed using a mixed-effects model with fixed-effect terms for treatment (with or without food), period, and sequence and with subject within sequence as a random effect. Point estimates and their associated 90% CIs for the natural log-transformed PK parameters for the difference between EIDD-2801 administered under fed relative to fasted conditions will be constructed. The point estimates and interval estimates will be back transformed to give estimates of the ratio and 90% CI for the geometric least squares mean ratio for fed/fasted to assess the effect of food.

10.6. Peripheral Blood Mononuclear Cells

Peripheral blood mononuclear cells will be collected from subjects following each dose in P2. Depending upon ongoing review of the data, PBMCs may be collected from subjects in P3; the collection of these samples may be omitted from some cohorts in P3.

The PBMCs will be stored for analysis of EIDD-2061 levels at some point in the future and reported separately.

10.7. Interim Analyses

There are no formal interim analyses planned for this study. However, interim analyses may be implemented at the discretion of the Sponsor or health authority request. In addition, data from P1 cohorts may be locked, unblinded, and analyzed prior to conducting P2 and P3.

11. STUDY ADMINISTRATION

11.1. Regulatory and Ethical Considerations

11.2. Ethical Standard

This study will be conducted in accordance with the protocol and with the following:

- Consensus ethical principles derived from international guidelines including the Declaration of Helsinki and Council for International Organizations of Medical Sciences International Ethical Guidelines.
- Applicable ICH GCP Guidelines.
- United States CRFs applicable to clinical studies (45 CFR Part 46; 21 CFR Part 50, 21 CFR Part 56, and 21 CFR Part 312).
- Applicable laws and regulations.

11.2.1. Ethics Committee Approval

The PI (or designee) must ensure that all required study-specific documents and/or information are submitted to the EC for review and approval as appropriate including but not limited to:

- the protocol and any future protocol amendments
- ICF and any other documents (electronic or paper) given to the subject
- IB

Any substantial protocol amendments, likely to affect the safety of the subjects or the conduct of the study, will require EC and regulatory authority (as locally required) approval before implementation of changes made to the study design, except for changes necessary to eliminate an immediate hazard to study subjects or any nonsubstantial changes, as defined by regulatory requirements.

11.2.2. Informed Consent

Informed consent is a process that is initiated prior to the individual's agreeing to participate in the study and continuing throughout the individual's study participation. Extensive discussion of risks and possible benefits of this therapy will be provided to the subjects and, if applicable, their families. Consent forms describing in detail the study interventions/products, study procedures, and risks are given to the subject and written documentation of informed consent is required prior to starting any screening procedures or intervention/administering study product. Consent forms will be EC-approved and the subject will be asked to read and review the document.

Upon reviewing the document, the PI (or designee) will explain the research study to the subject and answer any questions that may arise. The subjects will sign the ICF prior to any procedures being done specifically for the study. The subjects should have the opportunity to discuss the study with their surrogates or think about it prior to agreeing to participate. The subjects may withdraw consent at any time throughout the course of the trial. A copy of the ICF will be given to the subjects for their records. The rights and welfare of the subjects will be protected by emphasizing to them that the quality of their medical care will not be adversely affected if they decline to participate in this study.

11.3. Financing and Insurance

Financing and insurance will be addressed in a separate agreement.

11.4. Source Documentation and Access

The site will maintain appropriate medical and research records in compliance with ICH E6, Section 4.9 and regulatory and institutional requirements for the protection of confidentiality of subjects. The site will permit authorized representatives of the Sponsor, its designees, and appropriate regulatory agencies to examine (and when required by applicable law, to copy) clinical records for the purposes of quality assurance reviews, audits, and evaluation of the study safety and progress. These representatives will be permitted access to all source data and source documents, which include, but are not limited to, clinical and office charts, laboratory notes, memoranda, subjects' memory aid or evaluation checklists, pharmacy dispensing records, recorded data from automated instruments, copies or transcriptions certified after verification as being accurate and complete, photographic negatives, microfilm or magnetic media, x-rays, and subject files and records kept at the pharmacy, at the laboratory, and medico-technical departments involved in the clinical trial.

11.5. Data Collection and Record Keeping

11.5.1. Data Collection

Data collection and data entry are the responsibility of the clinical trial staff at the site. The PI is responsible for ensuring the accuracy, completeness, legibility, and timeliness of the data reported. All source documents should be completed in a neat, legible manner to ensure accurate interpretation of data.

A CRF must be completed for every subject who signs the ICF and has at least one protocol-specified assessment conducted. The CRF must be completed and processed according to the CRF guidelines and the SOPs of the site. All data should be entered into the CRF, where possible, within 3 days after each visit for any one subject. After the subject has completed the study, the PI must review and sign the signature page of the CRF indicating that he has reviewed the completed CRF and pertinent clinical data for that subject and that, to the best of his knowledge, all data recorded in the CRF accurately reflects the subject's clinical performance in the study

11.5.2. Study Records Retention

Study documents should be retained for a minimum of 5 years after the end of the study. These documents should be retained for a longer period; however, if required by local regulations. No records will be destroyed without the written consent of the Sponsor, if applicable. It is the responsibility of the Sponsor to inform the PI when these documents no longer need to be retained.

11.5.3. Protocol Deviations

A protocol deviation is any noncompliance with the protocol or study procedures detailed in the laboratory or pharmacy manuals. The noncompliance may have been the result of action by the PI, site staff, or subject. All deviations should be handled in accordance with site SOPs.

It is the responsibility of the site to use continuous vigilance to identify and report deviations within 5 working days of identification of the protocol deviation, or within 5 working days of the scheduled protocol-required activity.

11.6. Clinical Monitoring

Site monitoring is conducted to ensure that the human subjects' protections, study and laboratory procedures, study intervention administration, and data collection processes are of high quality and meet Sponsor, ICH/GCP guidelines and applicable regulations, and that this trial is conducted in accordance with the protocol, protocol-specific MOP and applicable Sponsor SOPs. Experienced clinical monitors of the Sponsor or its designee will conduct site-monitoring visits as detailed in the clinical monitoring plan.

Site visits will be made at standard intervals as defined by the Sponsor and may be made more frequently as directed by the Sponsor. Monitoring visits will include, but are not limited to, review of regulatory files, accountability records, CRFs, ICFs, medical and laboratory reports, and protocol and GCP compliance. Clinical monitors will have access to each participating site, study personnel, and all study documentation according to the site monitoring plan. Clinical monitors will meet with the site PI to discuss any problems and actions to be taken and will document site visit findings and discussions. As data are entered into a CRF, data will be monitored to ensure accuracy as well as adherence to data entry instructions provided in the CRF guidelines.

11.7. Quality Control and Quality Assurance

A quality management plan will be put in place for this study. The site is responsible for conducting routine quality assurance and quality control activities to internally monitor study progress and protocol compliance. The PI will provide direct access to all study-related sites, source data/data collection forms, and reports for the purpose of monitoring and auditing by the Sponsor, and inspection by local and regulatory authorities. The PI will ensure all study personnel are appropriately trained and applicable documentations are maintained on site.

11.8. Study Termination and Closure Procedures

11.8.1. Study Termination

This study may be temporarily suspended or prematurely terminated if there is sufficient reasonable cause. Written notification, documenting the reason for study suspension or termination, will be provided to the site. If the study is terminated or suspended, the PI (or designee) will inform study participants and the EC. The Sponsor will notify appropriate regulatory authorities. The Sponsor will provide the reason(s) for the termination or suspension. Study participants will be contacted, as applicable, and be informed of changes to study visit schedule.

If suspended, the study may resume once issues that caused suspension of the study are resolved.

11.8.2. Termination Procedures

If the study is prematurely terminated, then the site must return all appropriate study data, resolve all data queries, complete final drug accountability, return any study drug remaining on site, and ship all biological samples (including PK and PBMCs) to the laboratory designated by the Sponsor. The PI (or designee) must notify the EC of study termination.

11.9. Information Disclosure

11.9.1. Confidentiality

Subject confidentiality and privacy is strictly held in trust by the PI, site staff, and the Sponsor(s) and their agents. The study protocol, documentation, data, and all other information generated will be held in strict confidence. No information concerning the study or the data will be released to any unauthorized third party without prior written approval of the Sponsor.

The study monitor or other authorized representatives of the Sponsor or regulatory agencies may inspect all documents and records required to be maintained by the PI, including but not limited to, medical records (office, clinic, or hospital) and pharmacy records for the participants in this study. The clinical study site will permit access to such records.

The study participant's contact information will be securely stored at the clinical site for internal use during the study. At the end of the study, all records will continue to be kept in a secure location for as long a period as dictated by the Sponsor, site, or regulatory requirements.

Study participant research data will be transmitted to and stored securely by the Sponsor's designated data center. This will not include the participant's contact or identifying information. Individual participants and their research data will be identified by a unique study identification number. The study data entry and study management systems used by clinical sites will be secured and password-protected. At the end of the study, all study databases will be de-identified and archived at a secure location.

11.9.2. Clinicaltrials.gov

This clinical study will be registered on clinicaltrials.gov as required.

11.9.3. Publication Policy

All information generated from this study is the proprietary property of the Sponsor. It is the intent of the Sponsor to publish the results of the study in their entirety as deemed appropriate.

12. REFERENCES

Agostini ML, Pruijssers AJ, Chappell JD, Gribble J, Lu X, Andres EL, et al. Small-Molecule Antiviral β-d-N4-Hydroxycytidine Inhibits a Proofreading-Intact Coronavirus with a High Genetic Barrier to Resistance. J Virol. 2019 Dec 15;93(24).

Flavell RA, Sabo DL, Bandle EF, Weissmann C. Site-directed mutagenesis: generation of an extracistronic mutation in bacteriophage Q beta RNA. J Mol Biol. 1974 Oct 25;89(2):255–72.

Hernandez-Santiago BI, Beltran T, Stuyver L, Chu CK, Schinazi RF. Metabolism of the anti-hepatitis C virus nucleoside beta-D-N4-hydroxycytidine in different liver cells. Antimicrob Agents Chemother. 2004 Dec;48(12):4636–42.

Mullins JI, Heath L, Hughes JP, Li D, Essigmann JM, Harris KS, et al. Mutations based on viral decay acceleration in the HIV-1 genomes of a clinical population treated with the mutagenic nucleoside KP1461. Retrovirology. 2011 Oct 3;8(Suppl 2):P84.

Painter GR, Bowen RA, Bluemling GR, DeBergh J, Edpuganti V, Gruddanti PR, et al. The prophylactic and therapeutic activity of a broadly active ribonucleoside analog in a murine model of intranasal venezuelan equine encephalitis virus infection. Antiviral Res. 2019;171:104597.

Sticher ZM, Lu G, Mitchell DG, Marlow J, Moellering L, Bluemling GR, et al. Analysis of the Potential for N4-Hydroxycytidine To Inhibit Mitochondrial Replication and Function. Antimicrob Agents Chemother. 2020 27;64(2).

Stuyver LJ, Whitaker T, McBrayer TR, Hernandez-Santiago BI, Lostia S, Tharnish PM, et al. Ribonucleoside analogue that blocks replication of bovine viral diarrhea and hepatitis C viruses in culture. Antimicrob Agents Chemother. 2003 Jan;47(1):244–54.

Toots M, Yoon J-J, Cox RM, Hart M, Sticher ZM, Makhsous N, et al. Characterization of orally efficacious influenza drug with high resistance barrier in ferrets and human airway epithelia. Sci Transl Med. 2019 Oct 23;11(515):eaax5866.

Urakova N, Kuznetsova V, Crossman DK, Sokratian A, Guthrie DB, Kolykhalov AA, et al. β-d-N4-Hydroxycytidine Is a Potent Anti-alphavirus Compound That Induces a High Level of Mutations in the Viral Genome. J Virol. 2018 01;92(3).

Yoon J-J, Toots M, Lee S, Lee M-E, Ludeke B, Luczo JM, et al. Orally Efficacious Broad-Spectrum Ribonucleoside Analog Inhibitor of Influenza and Respiratory Syncytial Viruses. Antimicrob Agents Chemother. 2018;62(8).

13. APPENDICES

Appendix 1: Time and Events Schedule

Parts 1 and 2

Visit	Screening (Days -28 to -2)	Day -1	Day 1	Day 2 (24 hr)	Day 3 (48 hr)	Day 4 (72 hr)	Day 9	Day 15 / EOS[1] (non-FE cohorts)
Food-Effect Cohort		**Day 14**	**Day 15**	**Day 16 (24 hr)**	**Day 17 (48 hr)**	**Day 18 (72 hr)**	**Day 23**	**Day 30 / EOS**
ICF; Demography	x							
I/E; Medical history	x	x						
Physical examination[2]	x	x		x		x	x	x
Qualifying laboratory analyses[3]	x	x						
Drug screening and pregnancy test[4]	x	x					x	x
Height, weight (BMI)	x							
Clinic confinement[5]		x	x	x	x	x		
Non-residential visit	x						x	x
Clinical chemistry and urinalysis[6]	x	x		x		x	x	x
Hematology[7]	x	x		x	x	x	x	x
PBMC collection (FE cohort ONLY)			x[8]	x[8]				
ECG	x		x[9]			x		x
Vital signs	x	x	x[10]	x	x	x	x	x
Administer study drug			x					
Plasma PK sample collection			x[11]	x[12]	x[12]	x[12]		
Urine PK sample collection			x[13]	x[14]	x[14]			
Adverse events	x[15]	x[15]	x[15]	x	x	x	x	x
Prior and/or Concomitant medications	x	x	x	x	x	x	x	x

Abbreviations: BMI (body mass index); EOS (End of Study); FE (food-effect); ICF (Informed Consent Form); I/E (Inclusion/Exclusion Criteria); ECG (electrocardiogram); PBMC (peripheral blood mononuclear cell); PK (pharmacokinetic).

[1] On Day 15, conduct the EOS visit for all subjects except those in the FE cohort.

[2] A complete PE will be performed at Screening and/or Check-in, and EOS visits; on all other days, a limited PE will be conducted targeting any change from the previous status or any body system deemed pertinent to that subject by the PI (or designee).

[3] Laboratory analyses to qualify for study entry include: total white cell count or absolute lymphocyte count (values must be in the normal range), hemoglobin or platelet levels (values must not be below the lower limit of normal), and ALT/SGPT, alkaline phosphatase, AST/SGOT (values must not be above the upper limit of normal), plus virology (HIV/HCV/HBV; tested at Screening only; all must be negative), and FSH (if required).

[4] Drug screening (urine drug screen, UDS), including a cotinine test, should be conducted at Screening, Day -1, Days 9, 14, and 23, and at EOS. Pregnancy tests should be conducted in serum at Screening and in urine at all other times (i.e., Days -1, 9, 14, and 23, and at EOS). A positive urine pregnancy test will be confirmed with a serum pregnancy test. An alcohol breath test should be conducted at Screening and Days -1 and 14.

[5] Participants should be admitted to the clinic following review of final I/E criteria on Day -1 and discharged from the clinic following completion of study specific assessments on Day 4. Subjects in the FE cohort should be readmitted to the clinic on Study Day 14.

[6] Safety laboratory assessments include clinical chemistry and urinalysis. Baseline samples will be collected after clinic admission on Day -1.

[7] Hematology values include, but are not limited to, RBC, WBC with differential and platelets. PT/PTT and INR assessed only at

Screening.

[8] PBMCs will be collected predose, and 2, and 8 hrs postdose on Days 1 and15. Depending on ongoing review of the data, timepoints may be modified or removed, and up to 2 additional samples may be collected.

[9] The baseline ECG should be conducted prior to dosing on Day 1/15.

[10] Vital signs include pulse rate, respiratory rate, blood pressure, and body temperature; on Day 1/15, VS should be taken predose, and at 2, 4, 8 and 12 hr postdose.

[11] PK samples should be collected predose and at 0.25, 0.5, 1, 1.5, 2, 2.5, 3, 4, 6, 9, 12, and 15 hr postdose on Day 1/15. Depending on ongoing review of the PK data, timepoints may be modified or removed, and up to 3 additional samples (in each period for the food-effect) may be added.

[12] PK samples should be collected at 24, 36, 48, and 72 hr postdose on Days 2/16, 3/17, 4/18 respectively. Depending on ongoing review of the PK data, timepoints may be modified or removed, and up to 3 additional samples (in each period for the food-effect) may be added.

[13] Urine samples for PK analysis should be collected predose (spot collection), and from 0 to 4, 4 to 8, 8 to 12, and 12 to 24 hr postdose.

[14] Urine samples for PK analysis should be collected from 24 to 36 and 36 to 48 hr postdose.

[15] Adverse events occurring prior to study drug administration should be recorded as medical history unless the event is directly related to a study specified procedure.

Part 3

Visit	Screening (Days -28 to -2)	Day -1	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7	Day 8	Day 9	Day 14	Day 20 /EOS
ICF; Demography	x												
I/E; Medical history	x	x											
Physical examination[1]	x	x		x					x			x	x
Qualifying laboratory analyses[2]	x	x											
Drug screening and pregnancy test[3]	x	x										x	x
Height, weight (BMI)	x												
Clinic confinement[4]		x	x	x	x	x	x	x	x	x	x		
Non-residential visit	x											x	x
Clinical chemistry and urinalysis[5]	x	x			x			x			x		x
Hematology[6]	x	x			x			x			x	x	x
PBMC collection[7]			x					x					
ECG[8]	x		x					x			x		x
Continuous 12-lead ECG			x[9]					x[10]	x[10]				
Vital signs[11]	x	x	x	x	x	x	x	x	x	x	x	x	x
Administer study drug[12]			x	x	x	x	x	x					
Plasma PK sample collection[13]			x			x		x	x	x	x	x	
Urine PK sample collection[14]			x					x	x	x	x		
Adverse events	x[15]	x[15]	x[15]	x	x	x	x	x	x	x	x	x	x
Prior and/or Concomitant medications	x	x	x	x	x	x	x	x	x	x	x	x	x

Abbreviations: BID (twice daily); BMI (body mass index); EOS (End of Study); ICF (Informed Consent Form); I/E (Inclusion/Exclusion Criteria); ECG (electrocardiogram); PK (pharmacokinetic).

[1] A complete PE will be performed at Screening and/or Check-in, and EOS visits; on all other days, a limited PE will be conducted targeting any change from the previous status or any body system deemed pertinent to that subject by the PI (or designee).

[2] Laboratory analyses to qualify for study entry include: total white cell count or absolute lymphocyte count (values must be in the normal range), hemoglobin or platelet levels (values must not be below the lower limit of normal), and ALT/SGPT, alkaline phosphatase, AST/SGOT (values must not be above the upper limit of normal), plus virology (HIV/HCV/HBV; tested at Screening only; all must be negative), and FSH (if required).

[3] Drug screening (urine drug screen, UDS), including cotinine, should be conducted at Screening, Day -1, Day 14, and at EOS. Pregnancy tests should be conducted in serum at Screening and in urine at all other timepoints. A positive urine pregnancy test will be confirmed with a serum pregnancy test. An alcohol breath test should be conducted at Screening and Day -1.

[4] Participants should be admitted to the clinic following review of final I/E criteria on Day -1 and discharged from the clinic following completion of study specific assessments on Day 9.

[5] Safety laboratory assessments include clinical chemistry and urinalysis. Baseline samples will be collected after clinic admission on Day -1. On Days 3 and 6, the assessment will be pre-am dose. On Day 9, the assessment will be 72 hrs post-am dose on Day 6.

[6] Hematology values include, but are not limited to, RBC, WBC with differential and platelets. PT/PTT and INR assessed only at Screening.

[7] PBMCs will be collected 2 hrs postdose, relative to the first daily dose on Day 1, and pre-am dose and 2 and 8 hrs postdose relative to the first daily dose on Day 6. Depending on ongoing review of the data, timepoints may be modified or removed, and up to 2 additional samples may be collected. Additionally, PMBC collection may be omitted from some cohorts.

[8] The baseline ECG should be conducted prior to first dosing on Day 1. On Day 1, ECG should be taken predose and 2 hours post-am dose. On Day 6, ECG should be taken predose, 2 hrs, and 72 hrs (Day 9) postdose.

[9] On Day 1, monitor for 12-lead ECG recording will be worn from 2 hrs prior to the first of the daily doses to 12 hrs following the first of the daily doses. Extraction timepoints will be 60, 45, and 30 minutes prior to the first of the daily doses and at 0.25, 0.5, 1, 1.5, 2, 2.5, 3, 4, 6, 9, and 12 hrs following the first of the daily doses.

[10] On Day 6, monitor for 12-lead ECG recording will be worn from 2 hours prior to dosing (0 hr) to 25 hrs (Day 7) postdose. Extraction timepoints will be at 0.25, 0.5, 1, 1.5, 2, 2.5, 3, 4, 6, 9, 12, and 24 hrs postdose.

[11] Vital signs include pulse rate, respiratory rate, blood pressure, and body temperature. On Day 1, VS should be taken pre-am dose, and at 2, 4, and 8 hrs post-am dose. On Day 2 through 5, VS should be taken pre-am dose. On Day 6, VS should be taken predose, and 2, 4, 8, 24 (Day 7), 48 (Day 8), and 72 (Day 9) hrs postdose.

[12] Study drug should be administered BID, 12 hrs apart on Days 1 through 5. On Day 6, study drug should be administered once in the morning only (0 hr). Depending on ongoing review of the safety, tolerability, and PK data, the number of days of dosing may be reduced and the dosing frequency changed; the dosing frequency will be no less than once-daily or no greater than three-times daily. If the number of days of dosing are reduced, the timepoints for subsequent assessments and the period for which subjects are domiciled at the clinic will similarly be changed (subjects will be domiciled for 3 days post final dose).

[13] On Day 1, PK samples should be collected pre-am dose and at 0.25, 0.5, 1, 1.5, 2, 2.5, 3, 4, 6, 9, and 12 hrs post-am dose. On Day 4, the PK sample should be collected pre-am dose. On Day 6, PK samples should be collected predose and at 0.25, 0.5, 1, 1.5, 2, 2.5, 3, 4, 6, 9, 12, 15, 24 (Day 7), 48 (Day 8), 72 (Day 9), and 192 (Day 14) hrs postdose. Depending on ongoing review of the PK data, timepoints may be modified or removed, and up to 3 additional samples may be added.

[14] On Day 1, urine samples for PK analysis should be collected predose (spot collection), and from 0 to 4, 4 to 8, and 8 to 12 hrs postdose. On Day 6, urine samples for PK analysis should be collected predose (spot collection), and from 0 to 4, 4 to 8, 8 to 12, 12 to 24 (Day 7), 24 to 48 (Day 8), and 48 to 72 hrs (Day 9). Sampling timepoints may be modified, removed, or additional timepoints added depending upon ongoing review of the data. The 12-hour urine collection should occur prior to the second daily dose administration.

[15] Adverse events occurring prior to study drug administration should be recorded as medical history unless the event is directly related to a study specified procedure.

Appendix 2: DMID Toxicity Grading Scale

Clinical Adverse Events			
VITAL SIGNS	**Mild (Grade 1)**	**Moderate Grade 2)**	**Severe (Grade 3)**
Fever (°C) Oral temperature; no recent hot or cold beverages or smoking.	38.0 – 38.4	38.5 – 38.9	>39.0
Tachycardia - beats per minute	101 – 115	116 – 130	> 130 or ventricular dysrhythmias
Bradycardia - beats per minute	50 - 54 or 45 - 50 bpm if baseline <60 bpm	45 - 49 or 40 - 44 if baseline <60 bpm	< 45 or <40 bpm if baseline <60 bpm
Hypertension (systolic) - mm Hg [Assuming supine position, 10 min at rest conditions, not sleeping subjects, measurements on the same arm and several concordant results.]	141 - 150	151 - 160	> 160
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**
Arrythmia		asymptomatic, transient signs, no Rx required	recurrent/persistent; symptomatic Rx required
Hemorrhage, Blood Loss	Estimated blood loss ≤100 mL	Estimated blood loss > 100 mL, no transfusion required	Transfusion required
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 60 - 70% (of peak flow)	no normalization with bronchodilator; FEV1 <60% 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 gms/24 hours	4 - 5 loose or watery stools or 400 - 800 gms/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
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
SYSTEMIC	**Mild (Grade 1)**	**Moderate (Grade 2)**	**Severe (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
All Other conditions	**Mild (Grade 1)**	**Moderate (Grade 2)**	**Severe (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

*In addition to grading the measured local reaction at the greatest single diameter, the measurement should be recorded as a continuous variable.

**Induration/Swelling should be evaluated and graded using the functional scale as well as the actual measurement

Laboratory Adverse Events			
Blood, Serum, or Plasma *	**Mild (Grade 1)**	**Moderate (Grade 2)**	**Severe (Grade 3)**
Sodium – Hyponatremia mEq/L	132 – <LLN	130 – 131	<130
Sodium – Hypernatremia mEq/L	>ULN – 148	149 – 150	>150
Potassium – Hyperkalemia mEq/L	>ULN – 5.2	5.3 – 5.4	>5.4
Potassium – Hypokalemia mEq/L	<LLN-3.1	<3.1 – 3.0	<3.0
Glucose – Hypoglycemia mg/dL	65 – 67	55 – 64	<55
Glucose – Hyperglycemia Fasting – mg/dL	>ULN - 120	121 – 130	>130
Glucose – Hyperglycemia Random – mg/dL	140 – 159	160 – 200	>200
Blood Urea Nitrogen mg/dL	23-26	27 – 31	> 31
Creatinine – mg/dL	>ULN – 1.7	1.8 – 2.0	>2.0
Calcium – hypocalcemia mg/dL	8.0 – <LLN	7.5 – 7.9	<7.5
Calcium – hypercalcemia mg/dL	>ULN – 11.0	11.1 – 11.5	>11.5
Magnesium – hypomagnesemia mg/dL	1.3 – 1.5	1.1 – 1.2	<1.1
Phosphorus – hypophosphatemia mg/dL	2.3 – 2.5	2.0 – 2.2	<2.0
CPK – mg/dL	400-1000	1001-1500	>1500
Albumin – Hypoalbuminemia g/dL	2.8 – 3.0	2.5 – 2.7	< 2.5
Total Protein – Hypoproteinemia g/dL	5.2 – 5.4	4.8 – 5.1	< 4.8
Alkaline phosphate – U/L	132-240	241– 360	>360
AST U/L	44 – 105	106-175	>175
ALT U/L	44 – 105	106-175	>175
Bilirubin (serum total) mg/dL	1.3 - 2.0	2.1 - 2.5	> 2.5
Bilirubin – when ALT ≥105 (Hy's law)	1.3 -1.5	1.6 – 2.0	> 2.0
Amylase- U/L	200-270	271-360	>360
Lipase- U/L	176-270	271-360	>360
Hemoglobin (Female) - g/dL	11.0 – 11.5	9.5 – 10.9	< 9.5
Hemoglobin (Male) - g/dL	12.0 – 12.5	10.0 – 11.9	<10.0
WBC Increase - cell/mm3	11,001 – 15,000	15,001 – 20,000	> 20,000
WBC Decrease - cell/mm3	2,500 – 3,500	1,500 – 2,499	< 1500
Lymphocytes Decrease - cell/mm3	750 – 1,000	500 – 749	< 500
Neutrophils Decrease - cell/mm3	1,500 – 2,000	1,000 – 1,499	< 1000
Eosinophils - cell/mm3	500-750	751-1500	> 1500
Platelets Decreased - cell/mm3	120,000 – 130,000	100,000 – 119,999	<100,000
PT – seconds (prothrombin time)	> ULN-14.4	14.5 – 15.7	> 15.7
PTT – seconds (partial thromboplastin time)	>ULN-42.1	42.2– 50.0	> 50.0
Fibrinogen increase - mg/dL	>ULN – 500	501 – 600	> 600
Fibrinogen decrease - mg/dL	<LLN – 140	125 – 139	<125
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	11-50	> 50 and/or gross blood

* The laboratory values provided in the tables serve as guidelines and are dependent upon institutional normal parameters.

* Institutional normal reference ranges should be provided to demonstrate that they are appropriate.

Appendix 3: Contraception Guidance

Male subjects (regardless of fertility status) with partners of childbearing potential must use a male barrier method of contraception (i.e., male condom with spermicide) in addition to a second method of acceptable contraception from Check-in until 90 days after the EOS visit. Acceptable methods of contraception for female partners include:

- hormonal injection
- combined oral contraceptive pill or progestin/progestogen-only pill
- combined hormonal patch
- combined hormonal vaginal ring
- surgical method (bilateral tubal ligation or Essure® [hysteroscopic bilateral tubal occlusion])
- hormonal implant
- hormonal or non-hormonal intrauterine device
- OTC sponge with spermicide
- cervical cap with spermicide
- diaphragm with spermicide.

An acceptable second method of contraception for male subjects is vasectomy that has been performed at least 90 days prior to the screening visit, with verbal confirmation of surgical success.

For male subjects (even with a history of vasectomy), sexual intercourse with female partners who are pregnant or breastfeeding should be avoided unless condoms are used from the time of the first dose until 90 days after the EOS visit. Male subjects are required to refrain from donation of sperm from Check-in until 90 days after the EOS visit.

Sexual Abstinence and Same-sex Relationships

Subjects who practice true abstinence, because of the subject's lifestyle choice (i.e., the subject should not become abstinent just for the purpose of study participation), are exempt from contraceptive requirements. Periodic abstinence (eg, calendar, ovulation, symptothermal, postovulation methods) and withdrawal are not acceptable methods of contraception. If a subject who is abstinent at the time of signing the ICF becomes sexually active they must agree to use contraception as described previously.

For subjects who are exclusively in same-sex relationships, contraceptive requirements do not apply. If a subject who is in a same-sex relationship at the time of signing the ICF becomes engaged in a heterosexual relationship, they must agree to use contraception as described previously.

Title of study: A Randomized, Double-blind, Placebo-controlled, First-in-human Study Designed to Evaluate the Safety, Tolerability, and Pharmacokinetics of EIDD-2801 Following Oral Administration to Healthy Volunteers

NCT Number: NCT04392219

Document: Protocol, Version 5.1

Date: 11 June 2020

A Randomized, Double-Blind, Placebo-Controlled, First-in-Human Study Designed to Evaluate the Safety, Tolerability, and Pharmacokinetics of EIDD-2801 Following Oral Administration to Healthy Volunteers

(EIDD-2801-1001-UK)

Protocol Version	5.1 (United Kingdom)
Version Date	11 June 2020

Sponsor Team:

Ridgeback Biotherapeutics	3162 Commodore Plaza, Suite 3E Miami, FL 33133-5815 United States
Medical Officer	[redacted]
EudraCT Number	2020-001407-17

SUMMARY OF CHANGES

Changes from Version 1.1 (United Kingdom) to Version 2.1 (United Kingdom).

The primary reason for this amendment was to include Part 3, a multiple ascending dose study of EIDD-2801 in healthy subjects. Additionally, the following updates were made:

- Results of an *in vivo* micronucleus assay were added.
- The guidelines used to assess the severity of adverse events not presented in the DMID Toxicity Grading Scale were updated, specifically those that were life-threatening or led to death.
- The timepoints for collection of peripheral blood mononuclear cells in Part 2 were modified.
- Minor clarifications were made and typographical errors were corrected.

Changes from Version 2.1 (United Kingdom) to Version 3.1 (United Kingdom).

- Clarified that this study will be conducted at 1 site in the United Kingdom and up to 3 sites in the United States, and that integrated data across all sites will be used for dose-escalation decisions.
- The number of cohorts initially planned for Part 1 (single ascending dose) has been reduced from 4 to 3.
- The number of days of dosing in Part 3 (multiple ascending dose) was decreased from 7 days to 5.5 days. However, the number of days of dosing may be further reduced depending on ongoing review of the safety, tolerability, and pharmacokinetic data.
- Added a rationale for the appropriateness of a 4-day interval between dose escalations.
- Added a dose-escalation stopping criteria for reductions in platelets and lymphocytes.
- Clarified that assessments of human immunodeficiency virus, hepatitis B virus, and hepatitis C virus will only be conducted at Screening.
- Minor clarification and typographical corrections.

Changes from Version 3.1 (United Kingdom) to Version 4.1 (United Kingdom).

- The **total daily** dose to be administered in this protocol will not exceed 800 mg. A rationale for the increase in the maximum dose to be studied is provided in Section 9.2.
- The number of additional cohorts in Parts 1 and 3 has been increased to 4.

- Pregnancy tests in Part 3 will be conducted in serum at Screening and in urine at all other timepoints. A positive urine pregnancy test will be confirmed with a serum pregnancy test.

- Clarified that pregnancy, urine drug of abuse, cotinine, and alcohol breath tests will additionally be performed at Check-in to second treatment period in Part 2.

- An additional pharmacokinetic blood sampling timepoint was added in Part 1, and two additional pharmacokinetic blood sampling timepoints were added to each of Parts 2 and 3.

- Timepoints for continuous 12-lead monitoring in Part 3 were updated.

- Timepoints for collection of peripheral blood mononuclear cells in Part 2 were updated.

- The storage conditions for the study drug were updated.

- Minor clarification and typographical corrections.

Changes from Version 4.1 (United Kingdom) to Version 5.1 (United Kingdom).

- The **total daily** dose to be administered in this protocol will not exceed 1600 mg. A rationale for the increase in the maximum dose to be studied is provided in Section 9.2.

- The number of additional cohorts in Part 1 was increased to 7.

- The number of additional cohorts in Part 3 was increased to 6.

- Continuous 12-lead electrocardiogram monitoring was added in Part 1.

STATEMENT OF COMPLIANCE

The study will be carried out in accordance with Good Clinical Practice (GCP) as required by the following:

- Consensus ethical principles derived from international guidelines including the Declaration of Helsinki and Council for International Organizations of Medical Sciences International Ethical Guidelines.
- Applicable International Conference on Harmonisation (ICH) GCP Guidelines.
- United States Code of Federal Regulations (CFR) applicable to clinical studies (45 CFR Part 46; 21 CFR Part 50, 21 CFR Part 56, and 21 CFR Part 312).
- Applicable laws and regulations.

SIGNATURE PAGE

The signature below constitutes the approval of this protocol and the attachments and provides the necessary assurances that this study will be conducted according to all stipulations of the protocol, including all statements regarding confidentiality, and according to local legal and regulatory requirements and applicable regulations and ICH guidelines.

Principal Investigator:

Signed: [redacted] Date: 11 JUN 2020.

Executive Medical Director
Covance Clinical Research Unit Limited

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ABBREVIATIONS

AE	adverse event
AGP	alpha1-acid glycoprotein
ALT/SGPT	alanine aminotransferase/serum glutamic pyruvic transaminase
AST/SGOT	aspartate aminotransferase/serum glutamic oxaloacetic transaminase
AUC	area under the curve
BID	twice-daily
BMI	body mass index
CDC	Centers for Disease Control and Prevention
CEM	a cell line of lymphoblastic cells derived from a child with leukemia
CFR	Code of Federal Regulations
CHKV	Chikungunya virus
CI	confidence interval
CoV	coronavirus
CRF	case report form
CTP	cytosine triphosphate cytidine triphosphate
CYP	cytochrome P450
DMID	Division of Microbiology and Infectious Diseases
DMSO	dimethylsulfoxide
DPP4	dipeptidyl peptidase 4
DRF	dose range finding
DSS	Drug Safety Services
EBOV	Ebola virus
EC	Ethics Committee
EC_{50}	Half-maximal effective concentration
ECG	electrocardiogram
EEEV	Eastern equine encephalitis virus
EIDD	Emory Institute for Drug Development
EOS	End of Study
FDA	Food and Drug Administration
FE	food-effect
FIH	first-in- human
FSH	follicle stimulating hormone
GCP	Good Clinical Practice
GI	gastrointestinal
GLP	Good Laboratory Practice

HAE	human airway epithelium
HBV	hepatitis b virus
hBTEC	human bronchial/tracheal epithelial cells
HCV	hepatitis c virus
HED	human equivalent dose
HEENT	head, eye, ear, nose and throat
HIV	human immunodeficiency virus
HPMC	hydroxypropyl methylcellulose
IAV	influenza a virus
IB	Investigator's Brochure
IBV	influenza b virus
IC_{50}	half maximal inhibitory concentration
ICF	informed consent form
ICH	International Council for Harmonisation
MAD	multiple ascending dose
MDCK	Madin-Darby Canine Kidney Cells
MedDRA	Medical Dictionary for Regulatory Activities
MERS	Middle East respiratory syndrome
%MN-PCE	percentage of micronuclei-polychromatic erythrocytes
MRSD	maximum recommended starting dose
MTD	maximum tolerated dose
NOAEL	no-observed-adverse-effect-level
NSAID	nonsteroidal anti-inflammatory drug
OTC	over-the-counter
PBMC	peripheral blood mononuclear cells
PBO	placebo
PCE:TE	polychromatic erythrocytes to total erythrocytes
PE	physical examination
PI	principal investigator
[redacted]	[redacted]
PK	pharmacokinetic
QTc(F)	QT interval corrected for heart rate (using Fridericia's formula)
RNA	ribonucleic acid
RSV	respiratory syncytial virus
SAD	single ascending dose
SAE	serious adverse event

SAP	statistical analysis plan
SARS	severe acute respiratory syndrome
SI	selectivity index
SOP	standard operating procedure
UTP	uridine 5'-triphosphate
VEEV	Venezuelan equine encephalitis virus
ZIKV	Zika virus

PROTOCOL SYNOPSIS

Sponsor: Ridgeback Biotherapeutics	
Title: A Randomized, Double-Blind, Placebo-Controlled, First-in-Human Study Designed to Evaluate the Safety, Tolerability, and Pharmacokinetics of EIDD-2801 Following Oral Administration to Healthy Volunteers (EIDD-2801-1001-UK)	
Short Title: EIDD-2801-1001-UK	**Development Phase:** Phase 1
Study Sites: This study will be conducted at 1 site in the United Kingdom and up to 3 sites in the United States.	
Description of Study Drugs, Dose and Mode of Administration: EIDD-2801 and matching placebo (PBO) will be supplied [redacted] **Part 1:** A single oral dose of EIDD-2801 or PBO will be administered to subjects enrolled in Part 1 (P1; single ascending dose [SAD]) cohorts. The starting dose in the first SAD cohort will be 50 mg. The doses will be administered in an escalating manner. Dose levels will not increase by more than 3-fold for predicted nonpharmacologically active dose levels and by 2-fold for predicted pharmacologically active dose levels. [redacted] **Part 2:** Subjects in Part 2 (P2)/food-effect (FE) will receive 2 single doses of EIDD-2801 with a 14-day (minimum) washout period between doses. The dose for the P2 cohort is planned to be 100 mg but may be adjusted based on safety, tolerability, and/or pharmacokinetic (PK) data from P1. In any case, the dose assessed in P2 will have been given previously to subjects in a P1 cohort successfully (i.e., no halting rules were met following dosing). **Part 3:** Multiple doses of EIDD-2801 or PBO will be administered to subjects enrolled in Part 3 (P3; multiple ascending dose [MAD]) cohorts. Subjects will receive twice-daily (BID) doses for 5 days, with a single dose administered on the morning of Day 6 for collection of steady-state PK samples during waking hours. The total daily dose will not exceed a dose shown to be safe and well-tolerated in P1. Doses in P3 may be administered in the fed state, following review of the PK data from P2. Additionally, depending on ongoing review of the safety, tolerability, and PK data, the number of days of dosing may be reduced and the dosing frequency may be changed; the dosing frequency will be no less than once-daily or no greater than three-times daily.	
Treatment Duration: **Part 1:** Subjects enrolled into P1/SAD cohorts will receive a single dose of study drug (EIDD-2801 or matching PBO). **Part 2:** Subjects enrolled into the P2/FE cohort will receive 2 single doses of EIDD-2801, with a washout period between doses. **Part 3:** Subjects in P3/MAD cohorts will receive study drug (EIDD-2801 or matching PBO) for 5 days BID followed by a single dose of EIDD-2801 or PBO on Day 6. Depending on ongoing review of the safety, tolerability, and PK data, the number of days of dosing may be reduced.	

Subject Duration:

Part 1: The maximum possible study duration for participants enrolled in P1/SAD cohorts will be approximately 43 days.

Part 2: The maximum possible study duration for participants enrolled in the P2/FE cohort will be approximately 58 days.

Part 3: The maximum possible study duration for participants enrolled in P3/MAD will be approximately 48 days.

Objectives and Endpoints:

Part 1: SAD Cohorts

Primary:

- Objective: To determine the safety and tolerability of single ascending doses of EIDD-2801.
  - Endpoints:
    - Results of safety evaluations including safety laboratory assessments, physical examination (PE), electrocardiograms (ECGs), vital signs, and adverse events (AEs).

Secondary:

- Objective: To define PK of EIDD-2801 and EIDD-1931 in plasma and urine following single doses administered to healthy volunteers.
  - Endpoints:
    - Plasma PK parameters, including C_{max}, t_{max}, $t_{1/2}$, CL/F, λz, Vz/F, $AUC_{0\text{-}inf}$ and AUC_{last} as appropriate
    - Urinary excretion of EIDD-2801 and EIDD-1931 following single-dose administration.

Exploratory:

- Objective: To collect data to assess the relationship between EIDD-2801/EIDD-1931 concentrations and QT interval corrected for heart rate (QTc).

Part 2: Single-Dose Food-Effect

Primary:

- Objective: To assess the effect of food on the PK of EIDD-2801 and EIDD-1931 following a single oral dose.
  - Endpoints:
    - Plasma PK parameters, including C_{max}, t_{max}, $t_{1/2}$, CL/F, λz, Vz/F, $AUC_{0\text{-}inf}$ and AUC_{last} as appropriate
    - Urinary excretion of EIDD-2801 and EIDD-1931 following single-dose administration.

Secondary:

- Objective: To determine the safety and tolerability of single doses of EIDD-2801.
  - Endpoints:
    - Results of safety evaluations, including safety laboratory assessments, PEs, ECG, vital signs, and AEs.

Part 3: MAD Cohorts

Primary:

- Objective: To determine the safety and tolerability of multiple ascending doses of EIDD-2801.
    - Endpoints:
        - Results of safety evaluations, including safety laboratory assessments, PE, ECGs, vital signs, and AEs.

Secondary:

- Objective: To define PK of EIDD-2801 and EIDD-1931 in plasma and urine following multiple doses administered to healthy volunteers.
    - Endpoints:
        - Plasma PK parameters, including C_{trough}, C_{max}, t_{max}, $t_{1/2}$, CL/F, λz, Vz/F, $AUC_{0\text{-}inf}$ (Day 1 dose only), AUC_{τ}, $RA_{AUC\tau}$, and RA_{Cmax} as appropriate
        - Urinary excretion of EIDD-2801 and EIDD-1931 following multiple-dose administration.

Exploratory:

- Objective: To collect data to assess the relationship between EIDD-2801/EIDD-1931 concentrations and QTc.

Population:

Part 1: P1/SAD cohorts will include 8 subjects each, 2 randomized to receive PBO and 6 randomized to receive EIDD-2801. Initially, 3 SAD cohorts are planned for P1 with the option to add an additional 7 cohorts based on study results.

Part 2: Ten subjects will be enrolled into the P2/FE cohort; all subjects enrolled in the P2 cohort will receive EIDD-2801. One P2 cohort is planned for the study. If PK results obtained are equivocal, additional subjects may be enrolled into the P2 cohort at the previously tested dose or an additional P2 cohort may be added at a different dose.

Part 3: P3/MAD cohorts will include 8 subjects, 2 randomized to receive PBO and 6 randomized to receive EIDD-2801. Initially, 3 MAD cohorts are planned for P3 with the option to add an additional 6 cohorts based on study results. However, the number of cohorts may be reduced if the study objectives are met. Dose levels in P3 may be repeated for collection of peripheral blood mononuclear cells (PBMCs), providing that the dose level did not meet the dose-escalation halting criteria.

The site is strongly encouraged to ensure that women are represented in each cohort. Inclusion and exclusion criteria for study participation are as follows:

Inclusion Criteria: subjects who meet all of the criteria at Screening will be eligible for study participation

1. Male or female between the ages of 18 and 60, inclusive.
2. Female subjects must be of nonchildbearing potential. Nonchildbearing potential is defined as women who:
    - are physiologically incapable of becoming pregnant including those who are surgically sterile, via hysterectomy, bilateral salpingectomy, and/or bilateral oophorectomy by reported medical history and/or medical records. Surgical

sterilization to have occurred a minimum of 6 weeks, or at the Principal Investigator's (PI's; or designee's) discretion, prior to Screening.

- are postmenopausal (at least 45 years of age with amenorrhea for 12 months without an alternative medical reason with confirmatory follicle stimulating hormone (FSH) levels of ≥ 40 mIU/mL. The amenorrhea should not be induced by a medical condition such as anorexia nervosa, hypothyroid disease or polycystic ovarian disease, or by extreme exercise. It should not be due to concomitant medications that may have induced the amenorrhea such as oral contraceptives, hormones, gonadotropin-releasing hormones, anti-estrogens, or selective estrogen receptor modulators).

3. Male participants must agree to the use of effective contraception for study duration, i.e., from Check-in until 90 days following the last dose of study drug.
4. Is in generally good health as determined by medical history, PE (at Screening and/or Check-in), vital signs, and other screening procedures.
5. Has a body mass index (BMI) of 18 to 30 kg/m^2.
6. Is willing and able to comply with all study procedures including providing informed consent.

Exclusion Criteria: subjects who meet any of the following criteria at Screening (unless otherwise stated) will not be eligible for participation

1. Females who are pregnant, planning to become pregnant, or breastfeeding.
2. Has a clinically relevant acute or chronic medical condition or disease of the cardiovascular, gastrointestinal (GI), hepatic, renal, endocrine, pulmonary, neurologic, or dermatologic systems as determined by the PI (or designee).
3. Has any current or historical disease or disorder of the hematological system or significant liver disease or family history of bleeding/platelet disorders.
4. Has a history of cancer (other than basal cell or squamous cell cancer of the skin), rheumatologic disease or blood dyscrasias.
5. Has a history of GI surgery or other condition that may interfere with absorption of study drug, in the option of the PI (or designee).
6. Has a history of febrile illness within the 14 days prior to the first dose of study drug.
7. Has a positive alcohol or drug screen at Screening or the Day -1 visit or has a history of alcohol or drug abuse within the past 5 years.
8. Currently uses tobacco, nicotine or tobacco products, e-cigarettes or has stopped using tobacco products within the past 3 months and/or has a positive cotinine test at Screening or Check-in.
9. Has any abnormal laboratory value of Grade 2 or higher, which is considered to be clinically significant by the PI (or designee).
10. Has a total white cell count or absolute lymphocyte count outside the normal range, or hemoglobin or platelet levels below the lower limit of normal, at Screening of Day -1.
11. Has values above the upper limit of normal for the following laboratory analytes: alanine aminotransferase (ALT/SGPT), alkaline phosphatase (serum), aspartate aminotransferase (AST/SGOT), at Screening or Day -1.
12. Positive test result for human immunodeficiency virus (HIV), hepatitis B virus (HBV),

or hepatitis C virus (HCV).

13. Has an autoimmune disease, is immunosuppressed or is in any way immunocompromised.
14. Has any of the following:
    - QT interval corrected for heart rate using Fridericia's formula (QTcF) >450 ms confirmed by repeat measurement
    - QRS duration >110 ms confirmed by repeat measurement
    - PR interval >220 ms confirmed by repeat measurement
    - findings which would make QTc measurements difficult or QTc data uninterpretable
    - history of additional risk factors for torsades de pointes (e.g., heart failure, hypokalemia, family history of long QT syndrome).
15. Except as noted, has used prescription drugs (other than hormone replacement therapy) within 14 days prior to the first dose of study drug unless, in the opinion of the PI (or designee), the drug will not interfere with study assessments. The following prescription drugs will be allowed:
    - Statins: Subjects who have been on a stable dose of statins for a minimum of 6 months and who have normal creatine kinase at Screening.
    - Antihypertensives: Subjects who have been on a stable dose of antihypertensives for a minimum of 6 months and have controlled blood pressure and no active heart disease.
16. Has received an experimental agent (vaccine, drug, biologic, device, blood product or medication) within 3 months prior to the first dose of study drug and agrees not to receive another experimental agent during the duration of this trial.
17. Has donated blood within 30 days, plasma within 2 weeks, or platelets within 6 weeks prior to Screening or refuses to agree to not donate blood until 3 months after the End of Study (EOS) visit.
18. Uses over-the-counter (OTC) medications or nutritional supplements (including, e.g., gingko biloba, aspirin at any dose, and nonsteroidal anti-inflammatory drugs [NSAIDs]) on a routine/scheduled basis, and/or is unwilling to abstain from such use for the duration of the study.
19. Is unwilling to abstain from quinine containing products including quinine water, tonic water, bitter lemon, jello shots, any quinine preparations or Cinchona tree bark preparations (e.g., herbal medications containing Cinchona tree bark).
20. Has any condition that would, in the opinion of the PI (or designee), put the subject at increased risk for participation in a clinical trial or confinement in a clinical research unit.

Retesting for Inclusion/Exclusion Criteria: In the case where a parameter is outside of the acceptable limits for enrollment, measurement of the parameter can be repeated one time if both the following are met:

- In the opinion of the PI (or designee), the parameter value does not represent a chronic condition that otherwise will preclude the volunteer from enrolling in the study; and

- In the opinion of the PI (or designee), the abnormality is not likely to recur.

General Investigational Plan: For all potential subjects, volunteers who express interest in the study will report to the clinic for informed consent. The study will be explained to the subject and the Ethics Committee (EC)-approved informed consent form (ICF) will be presented. The subject will be given the chance to review the document and ask any questions he/she may have. If, after reviewing the consent, the subject would like to participate in the study, then he/she will sign the ICF and begin screening for study entry; those satisfying all criteria will be enrolled into the study and admitted to the clinic on Day -1. Retesting will be allowed as described above.

Part 1: For P1/SAD, the first cohort will be divided into 2 groups. The first group, a sentinel group, will include 2 subjects.

On Day 1, one subject in the sentinel group will be administered PBO and one will be administered EIDD-2801. Analysis of the safety results for the sentinel group will be reviewed 24 hours postdose. If no halting criteria have been met, the remainder of the subjects in Cohort 1 (the second group) will be dosed. There will be no sentinel groups for the remaining cohorts.

Following dosing on Day 1, subjects will remain in the clinic through completion of study assessments on Day 4, returning to the clinic on Day 9 for study assessments, and Day 15 for the EOS visit.

Doses will be administered in an escalating manner following satisfactory review by the Sponsor and PI of the blinded safety and tolerability data (up to 72 hours post final dose). Integrated data from all applicable sites will be used to make dose-escalation decisions. Dose-escalation will only occur if data from a minimum of 6 subjects (across all sites) have been reviewed from the previous lower dose group, such that data from a minimum of 4 subjects (across all sites) who have received EIDD-2801 will be used to make the dose-escalation decision. Dose levels will not increase by more than 3-fold for predicted nonpharmacologically active dose levels and by 2-fold for predicted pharmacologically active dose levels. The highest total daily dose to be studied under this protocol will not exceed 1600 mg.

Continuous 12-lead ECG monitoring will be performed to collect data to assess the relationship between study drug concentrations and QTc interval. All data will be archived without extraction or analysis and will not be reported in the scope of this study.

Part 2: For P2/FE, one cohort assessing the effect of food on EIDD-2801 and EIDD-1931 PK parameters will be enrolled; it is planned that the P2 cohort will be at a dose of 100 mg EIDD-2801, although higher or lower doses may be selected based on safety and available PK data from P1.

In addition to assessing the effect of food on dosing, PBMCs will be collected from subjects following each dose; the PBMCs will be stored for analysis of EIDD-2061 levels at some point in the future.

The 10 subjects enrolled in the FE cohort will all receive EIDD-2801; subjects will be randomized to a treatment sequence (i.e., to receive drug in the fed then fasted state [Sequence 1] versus fasted then fed state [Sequence 2]). Half of the subjects in the cohort will be randomized to Sequence 1 and the other half will be randomized to Sequence 2.

Following dosing on Day 1, subjects will remain in the clinic through completion of study assessments on Day 4, returning to the clinic on Day 9 for study assessments, and Day 14 to begin the second dosing sequence on Day 15. Subjects will then be discharged from the clinic on Day 18, returning to the clinic on Day 23 for study assessments, and Day 30 for the EOS visit.

There will be a 14-day (minimum) washout period between doses.

Part 3: For P3/MAD, subjects will receive BID doses on Day 1 through Day 5, inclusive, and will receive the final dose of study drug on the morning of Day 6 for collection of steady-state PK samples during waking hours. Subjects will remain domiciled at the site during the dosing period and until Day 9, returning to the clinic on Day 14 for completion of study assessments, and Day 20 for the EOS visit.

Doses in P3 may be administered in the fed state, following review of the PK data from P2. Additionally, depending on ongoing review of the safety, tolerability, and PK data, the number of days of dosing may be reduced and the dosing frequency changed; the dosing frequency will be no less than once-daily or no greater than three-times daily. If the number of days of dosing are reduced, the timepoints for subsequent assessments and the period for which subjects are domiciled at the clinic will similarly be changed (subjects will be domiciled for 3 days post final dose).

Part 3 may run in parallel with P1, providing that the total daily dose to be administered does not exceed a dose already shown to be safe and well-tolerated in P1.

Peripheral blood mononuclear cells may be collected from subjects in P3, depending upon ongoing review of the data. The collection of these samples may be omitted from some cohorts, depending on ongoing review of the data. The PBMCs will be stored for analysis of EIDD-2061 levels at some point in the future. In addition, continuous 12-lead ECG monitoring will be performed to collect data to assess the relationship between study drug concentrations and QTc interval. All data will be archived without extraction or analysis and will not be reported in the scope of this study.

Additional parts may be added to this study, including a SAD part in elderly healthy subjects and a study of the safety, tolerability, and PK in Japanese subjects. These additional parts are not described in this protocol but may be added via a protocol amendment.

Safety Monitoring and Potential Unblinding: Safety for this study will be continually monitored by the PI and Sponsor.

Blinding: All study personnel will remain blinded to treatment assignment (i.e., EIDD-2801 or PBO) in P1 and P3, except for personnel at the bioanalytical laboratory, and the unblinded pharmacy staff and pharmacokineticist. If unblinding is required to manage subject safety or to support dose-escalation decisions, the decision to unblind lies solely with the PI. If possible and providing that it does not interfere with or delay any decision in the best interest of the subject, the PI will discuss the intended code-break with the Sponsor.

Dose-Escalation Halting Rules:

For Group 1 in P1/SAD, sentinel dosing will occur whereby 2 subjects (1 active and 1 PBO) will be dosed on 1 day and, providing no safety concerns arise, the remaining 6 subjects will be dosed after at least 24 hours.

The study will be halted if one or more subjects experience a serious adverse event (SAE) that is considered to be related to EIDD-2801 or 2 or more subjects in the same group experience severe AEs that are considered to be related to EIDD-2801.

Additionally, if 2 or more subjects experience a reduction of platelets greater than 50% over baseline levels or a reduction in lymphocytes (absolute count) to Grade 3/severe levels without

an alternate cause, the PI and Sponsor will review the data. If, after review of unblinded data, it is determined that these events are probably related to study drug, no further dose escalations will occur.

Dose-escalation will stop if clinically relevant signs or symptoms of similar nature occur in 2 or more subjects in a group that, in the opinion of the PI, warrant stopping of dose-escalation.

Data Monitoring, Safety Reporting and Unblinding: Data will be monitored throughout the course of the study by experienced clinical monitors according to the clinical monitoring plan. As data will be entered into an electronic case report form (CRF), data will be monitored to ensure accuracy as well as adherence to data entry instructions provided in the CRF guidelines. Procedures for reporting any SAE will be detailed in the protocol and forms and instructions provided to the site.

Statistical Considerations: A complete description of all statistical analyses and methods will be presented in the statistical analysis plan (SAP). The SAP will be reviewed and approved by the Sponsor and will be finalized prior to database lock. Plans for PK analyses will be included in the SAP.

Determination of Sample Size: The sample sizes for the P1 and P3 cohorts are typical for a Phase 1 first in human (FIH) study. The sample size for the P2 cohort is in accordance with Food and Drug Administration (FDA) guidelines for sample size in FE studies.

Study Populations:

Safety Population: All subjects who receive at least one dose of study drug.

Pharmacokinetic Population: All subjects receiving study drug who comply sufficiently with protocol requirements (i.e., no major protocol deviations) and for whom a sufficient number of analyzable PK samples have been obtained to permit the determination of the PK profile for EIDD-2801/EIDD-1931 and the statistical analysis of the results. Subjects who experience an AE of vomiting before 2× the median t_{max} of the group may be excluded from the PK population.

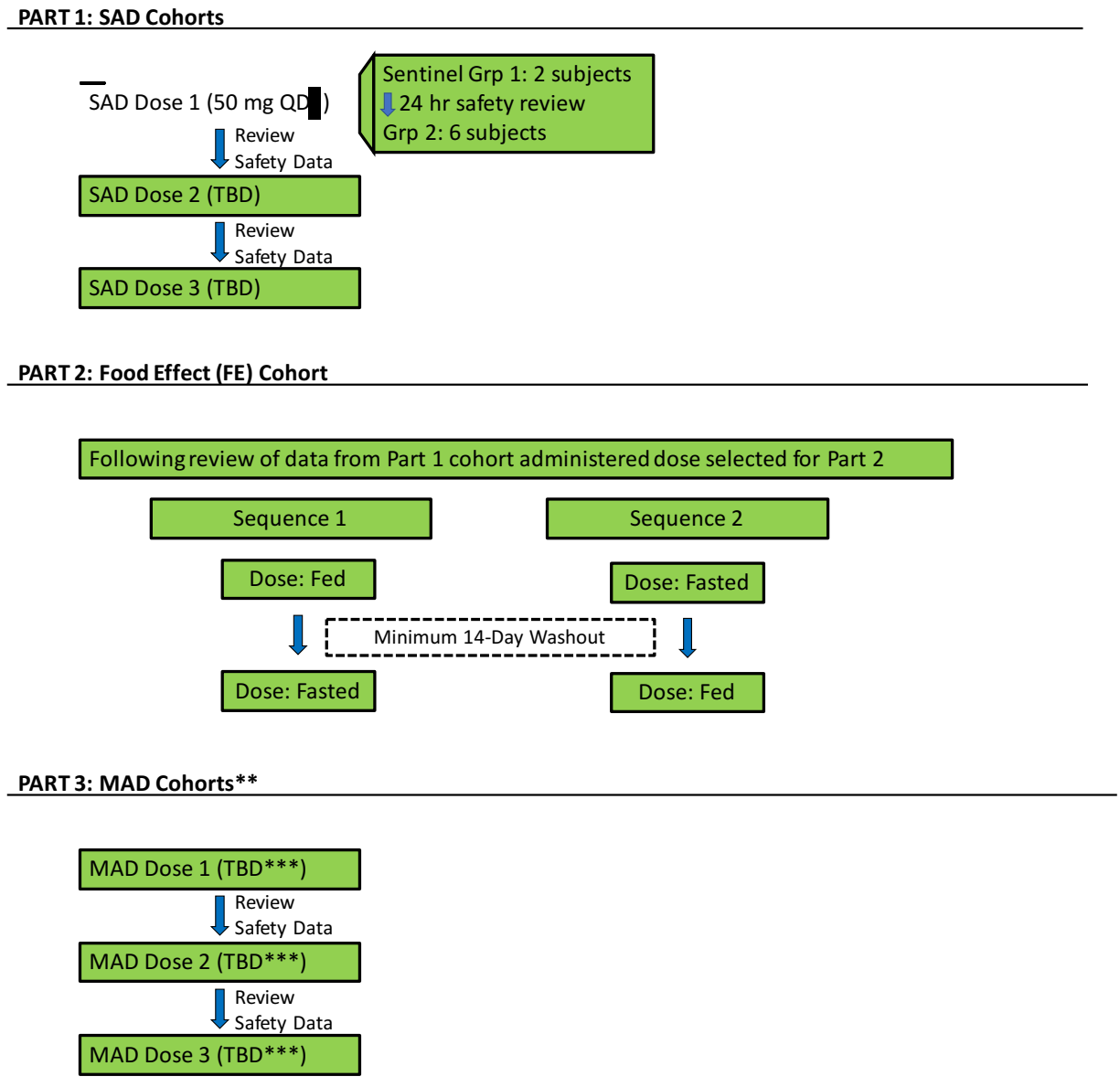
Safety Analyses: Statistical methods for the safety analyses will be descriptive in nature. Safety data, including AEs, clinical laboratory data, vital signs, ECG parameters, and PEs. All appropriate AEs will be graded using the Division of Microbiology and Infectious Diseases (DMID) toxicity scale (March 2014). Change from baseline will be included in summary tables for laboratory parameters. All laboratory data will be included in the data listings and all test values outside the normal range will be flagged.

Pharmacokinetic Analyses: Plasma PK parameters for each dose level will be calculated from the concentrations of EIDD-2801 and EIDD-1931 measured in predose and postdose plasma samples. For each dose level, descriptive statistics will be presented. Figures will be created to display mean and individual subject EIDD-2801 and EIDD-1931 concentration versus time. Urine PK parameters will be calculated whenever possible for each subject based on the urine concentrations of EIDD-2801 and EIDD-1931. The following PK parameters will be calculated:

Plasma PK Parameter	Description
AUC_{last}	The area under the plasma concentration-time curve, from time 0 to the last measurable non-zero concentration, as calculated by the linear up/log down trapezoidal method (P1 and P2 only).

$AUC_{0\text{-}inf}$	The area under the plasma concentration-time curve from time 0 extrapolated to infinity. $AUC_{0\text{-}inf}$ is calculated as the sum of AUC_{last} plus the ratio of the last measurable plasma concentration to the elimination rate constant (λz) (P1, P2, and Day 1 dose of P3).
C_{max}	Maximum observed concentration.
t_{max}	Time to reach C_{max}. If the maximum value occurs at more than one time point, t_{max} is defined as the first time point with this value.
λz	Apparent terminal elimination rate constant; represents the fraction of medication eliminated per unit time.
$t_{1/2}$	Apparent terminal elimination half-life of medication, calculated as $0.693/\lambda z$.
Vz/F	Apparent volume of distribution (EIDD-2801 only).
CL/F	Apparent oral drug clearance (EIDD-2801 only).
C_{trough}	Trough concentration (P3 only).
AUC_{τ}	The area under the plasma concentration-time curve during a dosing interval (P3 only).
$RA_{AUC\tau}$	Observed accumulation ratio based on AUC_{τ} (P3 only).
RA_{Cmax}	Observed accumulation ratio based on C_{max} (P3 only).
Urine PK Parameter	**Description**
A_e	Amount excreted in urine over the sampling interval.
F_e	Fraction of drug excreted in the urine over sampling interval.
CL_R	Renal clearance, $CL_R = A_e/AUC$ where both A_e and AUC are determined over co-incident time ranges after dosing.
In P1, dose-proportionality for PK parameters of EIDD-2801 and EIDD-1931 ($AUC_{0\text{-}inf}$, AUC_{last}, and C_{max}) will be assessed by the power model. The slope and the associated 90% confidence interval (CI) based on the power model will be reported. In P3 on Day 6, dose-proportionality for PK parameters of EIDD-2801 and EIDD-1931 (AUC_{τ} and C_{max}) will be assessed by the power model. The slope and associated 90% CI based on the power model will be reported. To assess the effect of food on the PK of EIDD-2801 and EIDD-1931, natural log-transformed PK parameters from P2 ($AUC_{0\text{-}inf}$, AUC_{last}, and C_{max}) will be analyzed using a mixed-effects model with fixed effect terms for treatment (with or without food), period, and sequence and with subject within sequence as a random effect. Point estimates and their associated 90% CIs for the natural log-transformed PK parameters for the difference between EIDD-2801 administered under fed relative to fasted conditions will be constructed. The point estimates and interval estimates will be back transformed to give estimates of the ratio and 90% CI for the geometric least squares mean ratio for fed/fasted to assess the effect of food.	
Interim Analyses: No formal interim analyses are planned for this study. Data from P1 cohorts may be locked, unblinded, and analyzed prior to conducting P2 and P3.	

Figure 1: Study Schema



Abbreviations:
FE: food effect
SAD: single ascending dose
MAD: multiple ascending dose

[redacted]

*** Part 3 may run in parallel with Part 1, providing that the total daily dose to be administered does not exceed a dose already shown to be safe and well tolerated in Part 1*

****The total daily dose in Part 3 will not exceed a dose shown to be safe and well-tolerated in Part 1*

1. INTRODUCTION

This study is being conducted at sites across the United Kingdom and the United States. Country-specific protocol amendments have been prepared; with the exception of regional regulatory requirements, the study is consistent across countries and data will be reported in a single clinical study report.

This study is a FIH study designed to assess the safety, tolerability and PK of EIDD-2801 in healthy human volunteers. EIDD-2801 is a ribonucleoside analog with broad-spectrum activity against many RNA viruses. It is currently being developed by Ridgeback Biotherapeutics as a treatment of infections caused by highly pathogenic coronaviruses (CoV), including COVID-19. In addition, EIDD-2801 is being developed in parallel as a treatment of uncomplicated influenza caused by all subtypes of circulating and emerging (drifted and shifted) influenza A virus (IAV) and influenza B virus (IBV), including seasonal, epidemic and pandemic strains.

1.1. Background

EIDD-2801 is the 5'-isopropyl ester prodrug of the broadly active, direct-acting antiviral ribonucleoside analog EIDD-1931. After oral delivery, the prodrug (EIDD-2801) is rapidly hydrolyzed by circulating esterases to produce high circulating (plasma) levels of EIDD-1931. In cell culture systems, EIDD-1931 has been shown to inhibit replication of multiple viral pathogens from multiple RNA virus families including pathogenic CoV (e.g., Middle East respiratory syndrome [MERS], severe acute respiratory syndrome [SARS]-CoV and SARS-CoV-2), influenza viruses (seasonal, pandemic, and avian subtypes), respiratory syncytial virus (RSV), alphaviruses (e.g., Eastern equine encephalitis virus [EEEV], Venezuelan equine encephalitis virus [VEEV], and Chikungunya virus [CHKV]), Filoviruses (e.g., Ebola virus [EBOV]), and Zika virus (ZIKV). In addition, EIDD-2801 is active against orthopoxviruses (tested against vaccinia virus) probably because orthopoxviruses encode their own unique RNA polymerase. [redacted] The primary mechanism of action of EIDD-2801 is inhibition of viral RNA replication by incorporation of the EIDD-1931 monophosphate metabolite into the viral RNA genome resulting in induction of viral error catastrophe.

1.2. Rationale for Development

EIDD-2801 is being developed for the treatment of infections caused by RNA viruses, specifically for COVID-19 and other CoV infections, influenza, and VEEV. During conduct of the FIH study, the Sponsor intends to define a dose that may be active in treating COVID-19 in patient studies.

EIDD-2801 has a unique dual mechanism of action against RNA viruses, including SARS-CoV-2 and other CoV infections. The compound acts as a competitive, alternative substrate for the virally encoded RNA-dependent RNA-polymerase that upon incorporation into nascent chain RNA induces increased mutational frequency in the viral genome. Incorporation quickly results in the production of non-viable virus. Additionally, the active metabolite,

EIDD-1931-5'-triphosphate (EIDD-2061), may act directly as a chain terminator and arrest replication by exerting a next nucleoside effect. It is anticipated that the high barrier to resistance observed during *in vitro* passaging studies will translate to slow, if any, emergence of viral resistance. Resilience to viral escape is a distinguishing feature of EIDD-2801.

Currently, there is no approved antiviral therapeutic for the treatment of COVID-19. An antiviral drug is urgently needed. [redacted]

1.3. Nonclinical Overview

1.3.1. Mechanism of Action

The mechanism of antiviral activity of EIDD-2801 is "lethal mutagenesis"; a concept that is predicated on increasing the viral mutation rate beyond a biologically-tolerable threshold, resulting in impairment of viral fitness and leading to viral extinction.

The specifics of the mechanism are as follows. EIDD-2801 is rapidly taken up by cells and the 5'-isopropylester cleaved to liberate EIDD-1931, which is in turn phosphorylated to EIDD-2061 by host kinases (Hernandez-Santiago et al., 2004; Painter et al., 2019). The 5'-triphosphate, EIDD-2061, acts as a competitive alternative substrate for virally encoded RNA-directed RNA polymerases and EIDD-2061 is incorporated into nascent viral RNA. Owing to the ability of the N^4-hydroxycytosine base of EIDD-1931 to tautomerize, EIDD-2061 can pair with either guanosine or adenosine, and consequently can substitute for either CTP or UTP, respectively (Flavell et al., 1974). This results in an accumulation of mutations that increases with each cycle of viral replication. The process whereby the mutation rate is increased by exposure to a drug is referred to as Viral Decay Acceleration (Mullins et al., 2011) and results in viral ablation.

Significant work has gone into validating this mechanism of action for EIDD-2801/1931, and it has been shown for MERS-CoV, VEEV, and IAV that viruses grown in the presence of EIDD-1931 have significantly increased levels of transition mutations (Agostini et al., 2019; Toots et al., 2019; Urakova et al., 2018). Multi-log decreases in virus yields were observed after treatment with EIDD-1931. Additionally, it was demonstrated for VEEV that the infectivity of virions formed in the presence of EIDD-1931 decreases from ~20% to <0.2%, and that the infectious virions are significantly Impaired in their replication ability (Urakova et al., 2018). As a consequence of this mechanism of action, the generation of drug-resistant escape mutants is practically impossible. This same effect was demonstrated for CoV (Agostini et al., 2019) and influenza virus (Toots et al., 2019). Furthermore, given the unique mechanism of action, EIDD-2801 is expected to be active against viruses resistant to other antiviral agents which have a different mechanism of action. The only data generated to date regarding the activity of EIDD-1931 against viruses resistant to other nucleoside analogs found that EIDD-1931 was

active against CoV resistant to remdesivir in cell culture assays (T. Sheahan et al, preprint available at https://www.biorxiv.org/content/10.1101/2020.03.19.997890v1).

As an alternative or additional mechanism of action, it has been theorized that incorporation of EIDD-2061 into viral genomic RNA can change the thermodynamics of RNA secondary structure and thus decrease the efficiency of the promoter regions involved in RNA genome replication (Stuyver et al., 2003).

1.3.2. *In Vitro* Pharmacology

1.3.2.1. Antiviral Activity in Tissue Culture and in Human Airway Epithelium

The ribonucleoside analog EIDD-1931 is the parent of the prodrug EIDD-2801. EIDD-1931 shows specific antiviral activity in different tissue culture cells and in the differentiated organoid model of human airway epithelium (HAE) with a selectivity index (SI) ranging from 21 to >100 for all influenza viral isolates tested. It is active against IAV (pandemic and seasonal) and IBV strains, as well as against highly pathogenic H5N1 and H7N9 strains (Table 1).

Table 1: EIDD-1931 Antiviral Activity Against Influenza A and B Viruses in Tissue Culture and Primary Human Bronchial/Tracheal Epithelial Cells

Virus	Strain	Cell line	EC_{50}* (µM)	CC_{50} (µM)	SI	Reference
IAV H1N1	Ca/07/2009	MDCK	1.24	68	55	NIAID Antiviral Testing Program
IAV H1N1	WSN/33	MDCK	1.1	299.8	275	Yoon et al., 2018
IAV H1N2	WSN/33	primary hBTEC	5.4	-	-	Yoon et al., 2018
IAV H2N3	Perth/16/2009	MDCK	0.88	52	59	NIAID Antiviral Testing Program
IAV H2N3	Ohio/sw-10-132/2010	MDCK	3.2	299.8	94	Yoon et al., 2018
IAV H5N1	Duck/MN/1525/81	MDCK	1.28	27	21	NIAID Antiviral Testing Program
IAV H5N1	Vietnam/1203/2004	MDCK	0.14	299.8	2100	Yoon et al., 2018
IAV H7N9	Anhui/1/2013	MDCK	0.13	299.8	2300	Yoon et al., 2018
IBV	Florida/4/2006	MDCK	<0.4	76	>190	NIAID Antiviral Testing Program
IBV	Brisbane/60/08	MDCK	0.006	299.8	50000	Yoon et al., 2018
IAV H1N1	Ca/07/2009	HAE-3D*	0.08	50	625	Toots et al., 2019
IAV H1N1	WSN/33	HAE-3D*	0.08	50	625	Toots et al., 2019
IBV	Brisbane/60/08	HAE-3D*	0.06	50	833	Toots et al., 2019

* Human Airway Epithelium organoid model.

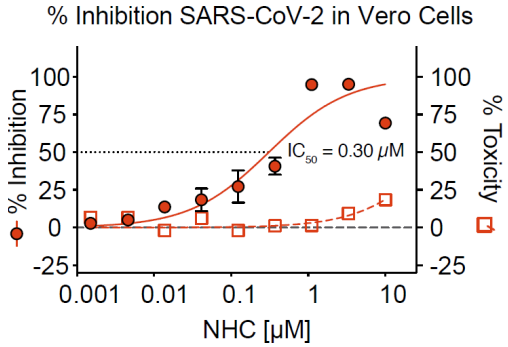
EIDD-1931 also showed specific antiviral activity against human SARS-CoV, MERS-CoV (Table 2) and SARS-CoV-2 (Figure 2), against togaviruses VEEV, EEEV and CHKV (Table 3) [redacted]

Table 2: EIDD-1931 Antiviral Activity Against SARS and MERS Coronaviruses in Tissue Culture and Primary Human Bronchial/Tracheal Epithelial Cells

Virus	Strain	Cell line	EC_{50} (µM)	CC_{50} (µM)	SI	Reference
SARS-CoV-1	Urbani	Vero76	<0.4	144	>360	NIAID Antiviral Testing Program
SARS-CoV-1	SARS-CoV-GFP(†)	HAE-3D(*)	<1	>100	>100	Tech. Report 25.038
MERS-CoV	GenBank Ac.No JX869059**	DBT-9	0.56	>200	>357	Agostini et al., 2019
MERS-CoV	Human ß-CoV C, Novel 2912	Vero E6	<0.8	20	>25	NIAID Antiviral Testing Program

* Human Airway Epithelium organoid model; ** cDNA Derived clone

Figure 2: Inhibition of SARS-CoV-2 by EIDD-1931



EIDD-1931 (NHC) antiviral activity (closed circles) and cytotoxicity (open squares) in Vero Cells infected with SARS-CoV-2. Vero cells were infected in duplicate with SARS-CoV-2 clinical isolate virus at a multiplicity of infection (MOI) of 0.05 in the presence of a dose response of EIDD-2801 for 48 hours after which replication was measured through quantitation of cell viability by Cell-Titer-Glo assay. Cytotoxicity was measured in similarly treated but uninfected cultures. Reproduced from Sheahan et al 2020.

Table 3: EIDD-1931 Antiviral Activity Against Togaviruses in Tissue Culture

Virus	Strain	Cell line	EC_{50} (µM)	CC_{50} (µM)	SI	Reference
VEEV	TC-83	Vero	0.43	>200	>930	Urakova et al., 2018
VEEV	TC-83	Vero76	1.92	32	17	NIAID Antiviral Testing Program
EEEV	FL93-939	Vero76	1.08	84	78	NIAID Antiviral Testing Program
CHKV	S27 (VR-64)	Vero76	1.8	96	53	NIAID Antiviral Testing Program

[redacted]

1.3.2.2. Cytotoxicity of EIDD-1931 in Tissue Culture Utilizing Cells from Different Organs and Species

EIDD-1931 was tested for cytotoxicity in human hepatic origin Huh7 and HepG2 cells, in human lymphoid CEM, human pancreatic BxPC-3, human prostate cancer PC-3, human muscle A204, human lung A549, human epithelial hEp-2, rat heart muscle H9c2, monkey kidney Vero, and canine kidney MDCK cell lines (Table 5). The compound exhibits low cytotoxicity in the majority of cells tested (half-maximal effective concentration [EC_{50}] values are in the range of 40 to >100 µM) except in lymphoid origin CEM cells where the compound shows a 7.5 µM EC_{50} value (Sticher et al., 2020; Urakova et al., 2018; Yoon et al., 2018).

Table 5: Cytotoxicity (CC50) of EIDD-1931 in Mammalian Cell Lines

Cell Line	CEM	HepG2	PC-3	A204	A549	BxPC-3	Huh-7	H9c-2	Vero	hEp-2	MDCK
CC50 (µM)	7.5	42.3	267.1	84	46	48	165.5	81	53	272.4	299.8

Sources: Sticher et al., 2020, Yoon et al., 2018

EIDD-2801 typically showed 2-4× lower activity and cytotoxicity than EIDD-1931 due to slightly slower uptake and anabolism in tissue culture.

1.3.2.3. Assessment of Mitochondrial Toxicity

Since EIDD-1931 is a nucleoside analog, additional investigations were performed to analyze whether observed cytotoxicity of EIDD-1931 is caused by mitochondrial toxicity. It was demonstrated that the prolonged treatment (14 days) with the compound does not result in selective killing of mitochondria or in mitochondrial dysfunction in CEM and HepG2 cells (Sticher et al., 2020).

1.3.3. *In Vivo* Pharmacology

The prodrug EIDD-2801 or its parent EIDD-1931 have been tested in animal models of RNA viral infection. An overview of results from the animal studies in indications to be pursued are described below. Additional detail is provided in the Investigator's Brochure (IB).

1.3.3.1. Coronavirus: SARS-CoV and MERS-CoV

In mouse models of SARS and MERS infection and disease, coronaviral disease was assessed by changes in body weight, measured daily, and lung hemorrhage, assessed in the large left lobe of the lung following sacrifice on Day 5 (SARS-CoV) or Day 6 (MERS-CoV). To assess production of infectious virions, virus was isolated from the lower left lobe of the lung following sacrifice on Day 5 (SARS-CoV) or Day 6 (MERS-CoV) and quantified using a plaque assay.

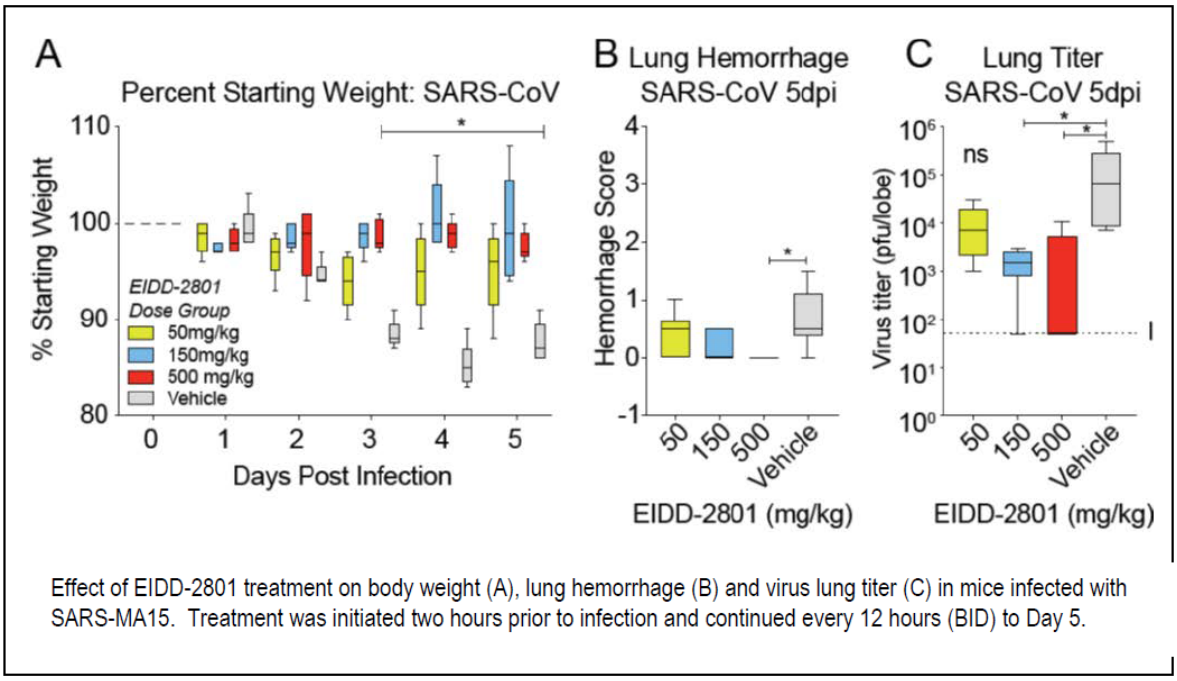
The results demonstrated that, in mice infected with either SARS- or MERS-CoV, both prophylactic and therapeutic treatment with EIDD-2801 resulted in a reduction in virus replication, improvements in pulmonary function, and improvements in maintaining body weight

(i.e., reduced body weight loss). While EIDD-2801 doses of 50, 150 and 500 mg/kg BID were assessed in the CoV mouse experiments, 500 mg/kg BID yielded the most consistent therapeutic effect.

A prophylactic, dose-escalation study was conducted in C57BL/6 mice infected with mouse-adapted SARS-CoV (SARS-MA15). Prophylactic oral treatment with EIDD-2801 was initiated 2 hours before intranasal infection and continued every 12 hours thereafter through the end of the study (Day 5; Figure 3).

In mice treated BID with EIDD-2801, body weight loss observed with vehicle treatment was diminished in the 50 mg/kg treatment group, beginning on Day 3 post-infection. No body weight loss was seen in the 150 and 500 mg/kg treatment groups (Figure 3, Panel A). Lung hemorrhage was also significantly reduced on Day 5 post-infection, following treatment with 500 mg/kg EIDD-2801 (Figure 3, Panel B). When compared to vehicle control, a dose-dependent reduction in SARS-CoV lung titers at Day 5 was seen across all 3 treatment groups (Figure 3, Panel C) with significant differences among the vehicle, 150 mg/kg and 500 mg/kg groups. Thus, prophylactic treatment with EIDD-2801 resulted in a robust antiviral effect that was able to prevent SARS-CoV replication and disease.

Figure 3: EIDD-2801 Prophylactic Treatment of SARS-CoV Infected Mice

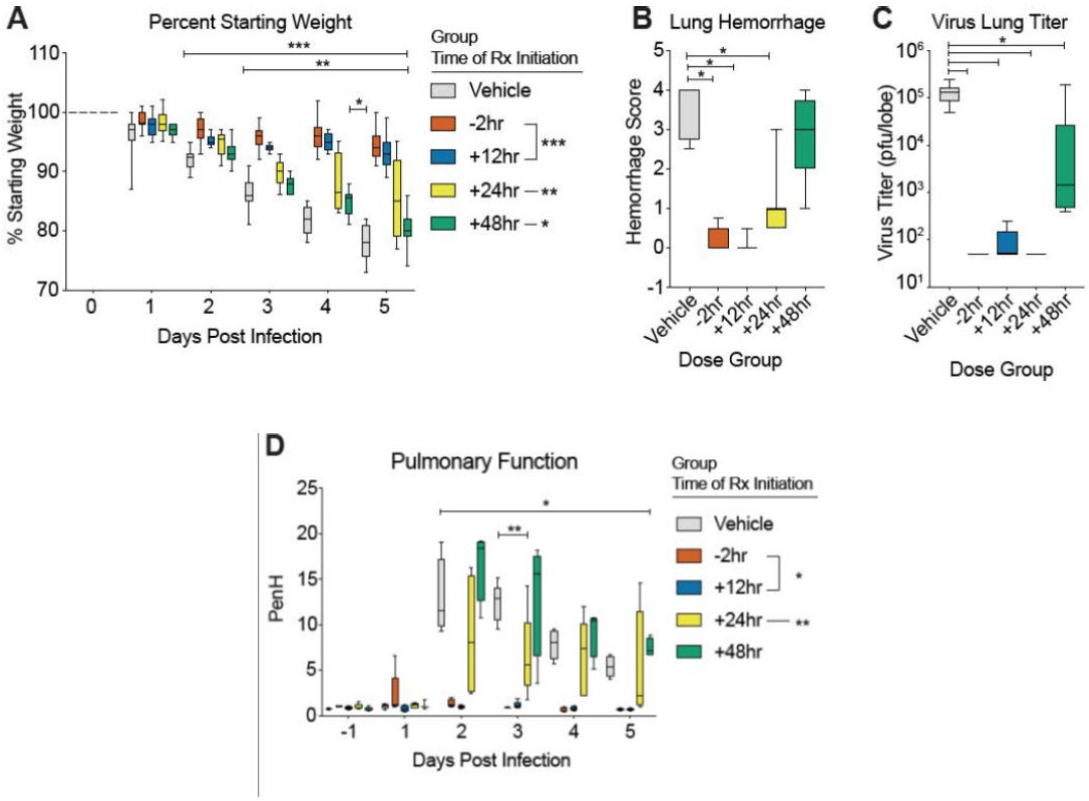


Effect of EIDD-2801 treatment on body weight (A), lung hemorrhage (B) and virus lung titer (C) in mice infected with SARS-MA15. Treatment was initiated two hours prior to infection and continued every 12 hours (BID) to Day 5.

The antiviral activity of EIDD-2801 against SARS-CoV was compared when treatment was initiated at -2 hours (pre-infection) and 12, 24, or 48 hours post-infection. After initiation of treatment, all groups were dosed every 12 hours for the duration of the study (Figure 4). For SARS-challenged mice, initiating treatment at 12 hours post-infection significantly prevented body weight loss beginning on Day 2, a result similar to that seen when dosing prophylactically

(i.e., beginning at 2 hours pre-infection). Initiation of treatment with EIDD-2801 at 24 hours post-infection also significantly reduced body weight loss on Days 3 through 5 post-infection. When EIDD-2801 treatment was initiated at 48 hours post-infection, body weight loss was only statistically different from vehicle on Day 4 post-infection (Figure 4, Panel A). Significant reductions in lung hemorrhage were seen when EIDD-2801 treatment was initiated before (-2 hours) and up to 24 hours after infection; a result that mirrored body weight loss data (Figure 4, Panel B). All mice treated with EIDD-2801 had significantly reduced viral loads in the lungs, even in the group where treatment was initiated 48-hour post-infection (Figure 4, Panel C). Pulmonary function, measured via whole body plethysmography, was assessed using the PenH metric which is a surrogate marker for bronchoconstriction or pulmonary obstruction. The administration of EIDD-2801 prior to infection (-2 hours) and at 12 hour post-infection completely abrogated the loss of pulmonary function observed in vehicle-treated animals (Figure 4, Panel D). Improved pulmonary function was also seen in the group where treatment was initiated 24 hours after virus challenge.

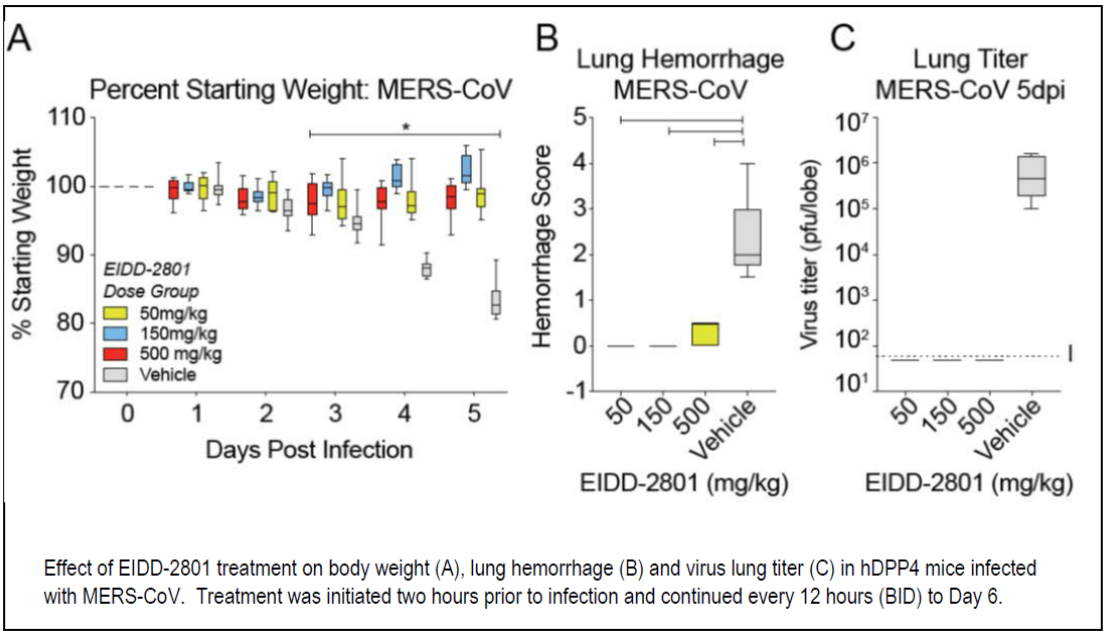
Figure 4: EIDD-2801 Treatment of SARS-CoV Infected Mice



EIDD-2801 was also tested to determine if it is active *in vivo* against MERS-CoV as described by Sheahan et. al (draft manuscript). The murine ortholog of the MERS-CoV receptor, dipeptidyl peptidase 4 (DPP4), does not support viral binding and entry. Thus, all *in vivo* studies described below were performed in genetically modified hDPP4 mice permissive for MERS infection. Prophylactic treatment starting at 2 hours before viral challenge with either 50, 150, or

500 mg/kg EIDD-2801 prevented body weight loss on Days 2 through 6 post-infection (Figure 5, Panel A), prevented lung hemorrhage measured on Day 6 (Figure 5, Panel B), and reduced virus lung titer on Day 6 to the limit of detection (Figure 5, Panel C).

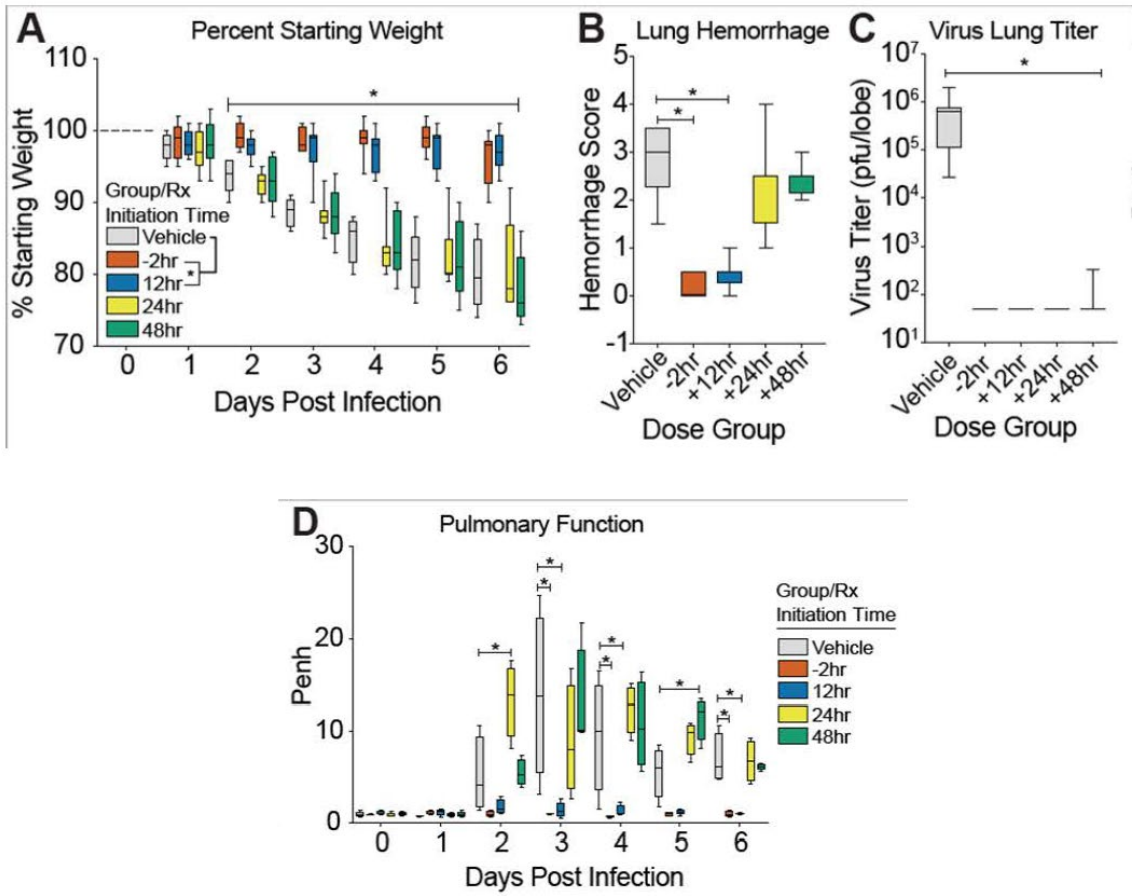
Figure 5: EIDD-2801 Prophylactic Treatment of MERS-CoV Infected Mice



Effect of EIDD-2801 treatment on body weight (A), lung hemorrhage (B) and virus lung titer (C) in hDPP4 mice infected with MERS-CoV. Treatment was initiated two hours prior to infection and continued every 12 hours (BID) to Day 6.

The effect of EIDD-2801 treatment on MERS-CoV infected mice is shown in Figure 6. When EIDD-2801 treatment was initiated 12 hours post-infection, there was no loss in body weight from Days 2 through 6 post-infection (Figure 6, Panel A) and no evidence of lung hemorrhage on Day 6 post-infection (Figure 6, Panel B). However, protection was not observed in groups where treatment was initiated either 24- or 48-hours post-infection. Conversely, virus lung titer on Day 6 post-infection was significantly reduced to the limit of detection in all treatment groups, regardless of the time treatment began (Figure 6, Panel C). To gauge the effect of the timing of EIDD-2801 treatment initiation on physiologic measures of lung disease, pulmonary function was assessed by whole body plethysmography. As expected, a loss of pulmonary function, as determined by measuring the PenH metric, was observed in vehicle-treated animals infected with MERS-CoV beginning on Day 2 post-infection (Figure 6, Panel D). Mirroring the body weight loss data, normal pulmonary function was observed in groups where treatment was initiated prior to or at 12 hours post-infection (Figure 6, Panel D).

Figure 6: EIDD-2801 Treatment of MERS-CoV Infected Mice



1.3.3.2. Influenza Virus

EIDD-2801 was tested in a ferret model of influenza virus infection and disease. Ferrets recapitulate hallmarks of human influenza infection, providing a clinically relevant animal model to investigate therapeutic intervention. Therapeutic oral dosing of influenza virus-infected ferrets reduced shed levels of pandemic and seasonal IAV by multiple orders of magnitude, and alleviated fever and inflammation while improving airway epithelium histopathology. Post-exposure prophylactic dosing was sterilizing (Toots et al., 2019).

Ferrets infected with pandemic IAV and treated with EIDD-2801 at a dose of 7 mg/kg demonstrated significantly less viral shedding and inflammatory cellular infiltrates in nasal lavages, but mounted a normal humoral antiviral response (Toots et al., 2019).

When examining the effect of delayed dosing, Toots et. al. (2019) demonstrated that treatment with 20 mg/kg of EIDD-2801 initiated 24 hours after challenge with IAV (H1N1) resulted in a rapid decrease in both shed virus and fever relative to control, with fever returning to baseline at 24 hours. When oseltamivir (20 mg/kg) was dosed prophylactically as a positive control, a modest suppression of fever was observed; however, there was no reduction in shed virus titer.

1.3.3.3. Venezuelan Equine Encephalitis Virus

Treatment with EIDD-1931 was evaluated in a mouse model of lethal VEEV infections. To be truly effective as a therapeutic agent for VEEV infection, a drug must penetrate the blood brain barrier and arrest virus replication in the brain. High plasma levels of EIDD-1931 are rapidly achieved in mice after oral dosing. Once in the plasma, EIDD-1931 is efficiently distributed into organs important in the pathology of VEEV infection, including the brain, where it is rapidly converted to its active 5'-triphosphate (EIDD-2061). EIDD-1931 showed a good safety profile in mice after 7 days of dosing with up to 1,000 mg/kg/day. In mouse model studies of VEEV infection, EIDD-1931 was 90-100% effective in protecting mice against lethal intranasal infection when therapeutic treatment was started as late as 24 hours post-infection, and partial protection was achieved when treatment was delayed for 48 hours post-infection (Painter et al., 2019).

[redacted]

1.4. Safety and Secondary Pharmacology

The standard battery of safety pharmacology studies including studies assessing the cardiovascular, respiratory and central nervous systems have been conducted. The studies are discussed in the IB; results indicated that there were no adverse pharmacologic effects of EIDD-2801 on the cardiovascular, respiratory or central nervous systems.

1.5. Nonclinical Pharmacokinetics and Metabolism

1.5.1. Overview

The uptake, metabolism and protein binding of EIDD-2801 and EIDD-1931 have been studied in plasma, microsomes, and non-hepatic cells from several species as outlined below. The PK and tissue distribution of prodrug EIDD-2801 and its active parent EIDD-1931 have been studied extensively in rats, dogs and ferrets. Key results from these studies are presented below; additional detail can be found in the IB.

1.5.2. Absorption

EIDD-1931 is parent of the prodrug EIDD-2801. The appearance of EIDD-1931 is dependent on the absorption of EIDD-2801 and the rate of its conversion to EIDD-1931.

EIDD-2801 PK studies have been completed in dog, rat, mouse, ferret, and monkey. EIDD-2801 was efficiently absorbed and rapidly converted to EIDD-1931 in each species. The t_{max} for EIDD-2801 (not observed in rodents) occurred at 0.5-1 hours, while the t_{max} for EIDD-1931 occurred at 1-2 hours.

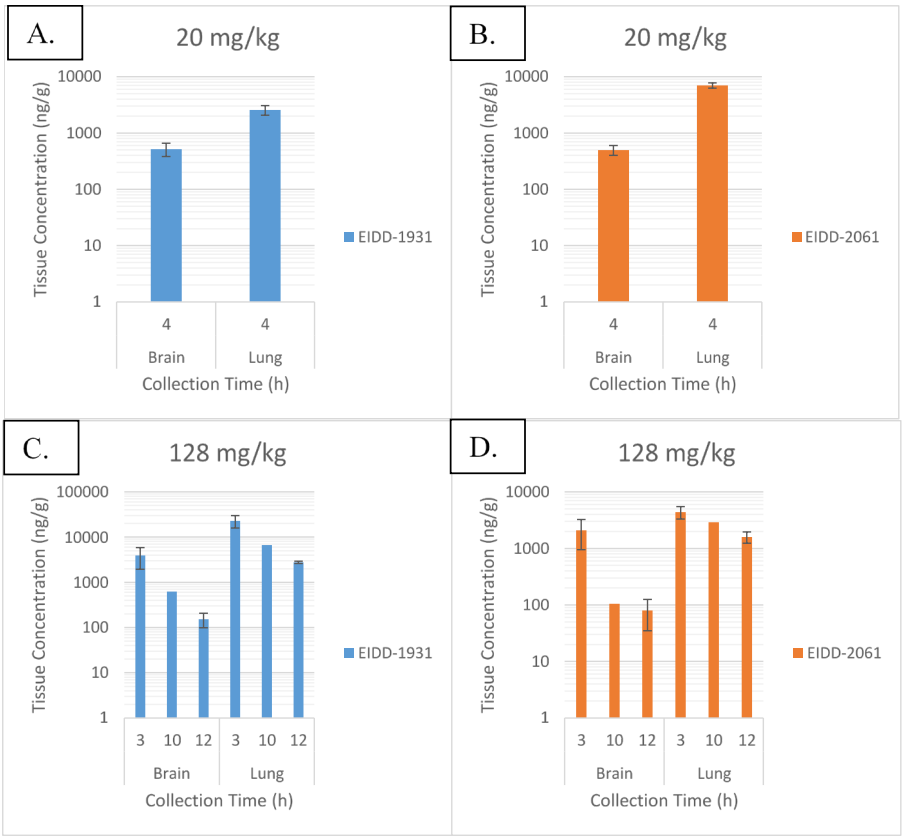
1.5.3. Distribution

[redacted]

1.5.3.2. Tissue Distribution Studies

EIDD-2801 is rapidly absorbed in the gut and converted to EIDD-1931 reaching C_{max} in 1-3 hours in mice, rats, ferrets, dogs and monkeys. EIDD-1931 is then widely distributed to tissues including lungs and brain, where it is rapidly taken up into cells and converted to EIDD-2061. Figure 7 shows the concentration of EIDD-1931 and EIDD-2061 in ferret brain and lung following single doses of 20 (Panels A and B) and 128 (Panels C and D) mg/kg.

Figure 7: Tissue Distribution of EIDD-1931 and EIDD-2061 in Ferret Brain and Lung



1.5.4. Metabolism

1.5.4.1. Metabolic Stability of EIDD-1931 and EIDD-2801

EIDD-2801 was designed to be converted to EIDD-1931 by esterases in plasma or in cells. Stability has been assessed in plasma and liver microsomes from mouse, rat, dog, monkey and humans. The stability of EIDD-2801 in mouse, rat and monkey plasma is relatively short (≤0.4 hours) while the stability is longer in human and dog plasma (1-3 hours). EIDD-2801 stability in mouse, rat, dog and monkey liver microsomes is very short, ranging from 0.02 to 0.08 hours while the stability in human liver microsomes is 1.2 hours (Table 6).

Table 6: Metabolic Stability of EIDD-2801 in Plasma and Liver Microsomes

	Plasma stability	LM stability
Species	t1/2 (h)	t1/2 (h)
Mouse	0.017	0.033
Rat	0.033	0.017
Dog	3.2	0.083
Monkey	0.40	0.017
Human	1.05	1.22

EIDD-2801 is stable in simulated gastric and intestinal fluids (Table 7).

Table 7: Metabolic Stability of EIDD-2801 in Simulated Gastric and Intestinal Fluids and in Buffered Saline with Fetal Bovine Serum

Matrix	t1/2 (hr)
Simulated Gastric Fluid	>24
Simulated Intestinal Fluid	>24
Phosphate Buffered Saline plus 10% Fetal Bovine Serum*	>24

EIDD-1931 was found to be stable when incubated with all tested plasmas, whole blood, liver microsomes and liver S9 extracts and intestinal microsomes (Table 8).

Table 8: Metabolic Stability of EIDD-1931 in Plasma, Whole Blood and Liver and Intestinal Microsomes

Medium	Plasma	Whole Blood	Liver Microsomes	Liver S9 Stability	Intestinal Microsomes
Species	t1/2 (h)	t1/2 (h)	t1/2 (h)	t1/2 (h)	t1/2 (h)
Mouse		>24	>24		>24
Rat	17		>24	>24	
Dog	>24			>24	
Monkey	6.5	>24	>24	>24	>24
Human	10	7	>24	20	>24

1.5.4.2. Uptake and Anabolism of EIDD-1931 in Tissue Culture and Primary Cells

EIDD-1931 is efficiently taken up by tissue culture cells and converted to its pharmacologically active metabolite EIDD-2061 (EIDD-1931-5'-triphosphate). Intracellular EIDD-2061 accumulates dose-dependently, with C_{max} levels ~200-2000 pmol/10^6 cells (at 10-20 µM dose) in different cell lines. It reaches high levels relatively quickly, typically within 1-3 hours, though the t_{max} values vary widely between 1 and 24 hours depending on the cell line and dose concentration tested. Detailed data on the uptake and anabolism of EIDD-2801 is presented in the IB.

EIDD-2801 is also taken up by tissue culture cells and is converted to EIDD-1931 and then to EIDD-2061, but the process is slightly delayed compared to dosing with EIDD-1931. EIDD-1931 is also taken up and metabolized to EIDD-2061 by primary cells. EIDD-2061 is accumulated in all primary cells tested except in mouse primary hepatocytes where EIDD-1931 is apparently extensively metabolized to cytidine and uridine which, in turn, quickly metabolize into CTP and UTP. The quick metabolism of EIDD-1931 consequently results in low levels of EIDD-2061 in mouse hepatocytes. The intracellular stability ($t_{1/2}$) of EIDD-2061 is 4-5 hours in human astrocytes and hBTEC and is significantly shorter (0.2-1.1 hours) in primary hepatocytes.

1.5.5. Excretion

Currently, there is no data on excretion of EIDD-2801 or EIDD-1931. Excretion will be measured in urine during this study.

1.5.6. Pharmacokinetic Drug Interactions

1.5.6.1. Cytochrome P450 (CYP) Inhibition

The purpose of this non-GLP *in vitro* study was to determine the time-dependent inhibitory potential of EIDD-2801 and EIDD-1931 on human cytochromes P450 (CYP) 1A2, 2B6, 2C8, 2C9, 2C19, 2D6, and 3A4 (midazolam and testosterone substrates) enzyme activity, using pooled human liver microsomes in an half-maximal inhibitory concentration (IC_{50}) shift assay.

Neither EIDD-2801 nor EIDD-1931 demonstrated inhibition greater than 31.4% for any of the CYP isozymes tested nor could the data for each assay condition be curve fit to determine time-dependent inhibition by these compounds. Full dose-response curves were not achieved at concentrations ranging from 0.00545 to 50.0 µM indicating EIDD-2801 and EIDD-1931 have no CYP inhibition potential at concentrations ranging from 0.00545 to 50.0 µM. Assay performance was acceptable based on the results for the positive control inhibitors.

1.5.6.2. CYP Induction

An assay was performed to determine the induction potential of EIDD-2801 on human CYP isoenzyme (1A2, 2B6, and 3A4) activity using 3 single-donor lots of inducible, cryopreserved human hepatocytes. Both enzyme activity and mRNA results demonstrated that EIDD-2801 did not show induction for any of the CYP isozymes.

[redacted] [redacted]

[redacted]	[redacted]	[redacted]	[redacted]	
			[redacted]	[redacted]
	[redacted]	[redacted]	[redacted]	[redacted]
	[redacted]	[redacted]	[redacted]	[redacted]
[redacted]	[redacted]	[redacted]	[redacted]	[redacted]
	[redacted]	[redacted]	[redacted]	[redacted]
	[redacted]	[redacted]	[redacted]	[redacted]
	[redacted]	[redacted]	[redacted]	[redacted]
	[redacted]	[redacted]	[redacted]	[redacted]
[redacted]	[redacted]	[redacted]	[redacted]	[redacted]
	[redacted]	[redacted]	[redacted]	[redacted]
	[redacted]	[redacted]	[redacted]	[redacted]

[redacted]

[redacted] [redacted]

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[redacted]

[redacted]

[redacted]

[redacted]

1.7. Potential Risks and Benefits

1.7.1. Potential Benefits

As this is a FIH study in healthy volunteers, there is no direct benefit to subjects enrolled in the study. However, given the current pandemic situation and *in vitro* antiviral activity of EIDD-2801 against SARS-CoV-2, and the activity against several other viruses of public health concern, it is possible that participants may benefit from future availability of the drug.

1.7.2. Potential Risks

EIDD-2801 has never been administered to humans; therefore, the risks from EIDD-2801 to subjects participating in this trial are unknown. Although toxicology studies have been done, unexpected AEs may occur. [redacted]. Subjects will also be monitored for other end-organ effects through a range of safety assessments.

2. OBJECTIVES

2.1. Study Objectives

2.1.1. Primary Objectives

The primary objective of Part 1 of the study is to determine the safety and tolerability of single ascending doses of EIDD-2801.

The primary objective of Part 2 of the study is to assess the effect of food on the PK on EIDD-2801 and EIDD-1931 following a single oral dose.

The primary objective of Part 3 of the study is to determine the safety and tolerability of multiple ascending doses of EIDD-2801.

2.1.2. Secondary Objectives

The secondary objectives of Part 1 and Part 3 of the study is to define the PK of EIDD-2801 and EIDD-1931 in plasma and urine following single and multiple doses administered to healthy volunteers.

The secondary objective of Part 2 of the study is to determine the safety and tolerability of single doses of EIDD-2801.

2.1.3. Exploratory Objectives

The exploratory objectives of Parts 1 and 3 are to collect data to assess the relationship between EIDD-2801/EIDD-1931 concentrations and QTc.

2.2. Study Outcome Measures

2.2.1. Primary Outcome Measures

The primary outcome measures for Parts 1 and 3 of are results of safety evaluations including safety laboratory assessments, PEs, ECGs, vital signs, and AEs.

The primary outcome measures for Part 2 of the study are plasma PK parameters including C_{max}, t_{max}, $t_{1/2}$, CL/F, λz, Vz/F, $AUC_{0\text{-}inf}$ and AUC_{last} as appropriate.

2.2.2. Secondary Outcome Measures

The secondary outcome measures are as follows:

- Single-dose plasma PK parameters, including C_{max}, t_{max}, $t_{1/2}$, CL/F, λz, Vz/F, $AUC_{0\text{-}inf}$ and AUC_{last} as appropriate (Part 1)

- Multiple-dose plasma PK parameters, including C_{trough}, C_{max}, t_{max}, $t_{1/2}$, CL/F, λz,

Vz/F, AUC_{τ}, $AUC_{0\text{-inf}}$ (Day 1 dose only), $RA_{AUC\tau}$ and RA_{Cmax}, as appropriate (Part 3)

- Urinary excretion of EIDD-2801 and EIDD-1931 following single- and multiple-dose administration (Parts 1 and 3).

- Results of safety evaluations including safety laboratory assessments, PEs, ECGs, vital signs, and AEs (Part 2).

3. STUDY DESIGN

3.1. Overview

EIDD-2801-1001-UK is a Phase 1, randomized, double-blind, placebo-controlled, FIH, SAD, and MAD study of the safety, tolerability and PK of EIDD-2801 and EIDD-1931 following oral administration of single and multiple doses of EIDD-2801 to healthy volunteers. In addition, for a minimum of one cohort, the effect of food on the single-dose EIDD-2801 and EIDD-1931 PK parameters will be assessed in subjects taking open-label EIDD-2801. The overall objective of the study is to identify a starting dose for future safety and therapeutic intervention trials.

The study is composed of 3 parts; P1 is the SAD study, P2 is the FE cohort study, and P3 is the MAD study.

This study will be conducted at 1 site in the United Kingdom and up to 3 sites in the United States.

3.1.1. Part 1 (Single Ascending Dose)

A single oral dose of EIDD-2801 or PBO will be administered to subjects. Subjects will be randomized in a 3:1 ratio to receive either EIDD-2801 or PBO.

Following dosing on Day 1, subjects will remain in the clinic through completion of study assessments on Day 4, returning to the clinic on Day 9 for study assessments, and Day 15 for the EOS visit.

The first cohort will be divided into 2 groups. The first group, a sentinel group, will include 2 subjects. On Day 1, one subject in the sentinel group will be administered PBO and one will be administered EIDD-2801. Analysis of the safety results for the sentinel group will be reviewed 24 hours postdose. If no halting criteria (Section 9) have been met, the remainder of the subjects in Cohort 1 (the second group) will be dosed. There will be no sentinel groups for the remaining cohorts.

After completion of each dosing cohort, safety and tolerability data will be reviewed to determine if any of the halting rules have been met. If not, then the subsequent cohort may be dosed following review of the 72-hour safety data. As PK data become available, these data may be used for dose-escalation decisions.

The proposed dose-escalation scheme is shown in Figure 1, however, planned dose escalations will be determined based on ongoing review of the safety, tolerability, and available PK data. The starting dose in the first SAD cohort will be 50 mg. Dose levels will not increase by more than 3-fold for predicted nonpharmacologically active dose levels and by 2-fold for predicted pharmacologically active dose levels. The highest total daily dose to be studied under this protocol will not exceed 1600 mg (Section 9.2).

Three cohorts are initially planned for P1; however, up to an additional 7 cohorts may be enrolled.

3.1.2. Part 2 (Food-Effect)

Two single oral doses of EIDD-2801 will be administered to subjects, in an open-label manner. The dose for the P2/FE cohort is planned to be 100 mg but may be adjusted based on safety, tolerability, and/or PK data from P1. The dose assessed in the P2 cohort will have been given previously to subjects in a P1 cohort successfully (i.e., no halting rules were met following dosing).

Subjects will be randomized to a treatment sequence; i.e., to receive drug in the fed then fasted state (Sequence 1) versus fasted then fed state (Sequence 2; Figure 1). Half of the subjects in the cohort will be randomized to Sequence 1 and the other half will be randomized to Sequence 2.

Following dosing on Day 1, subjects will remain in the clinic through completion of study assessments on Day 4, returning to the clinic on Day 9 for study assessments, and Day 14 to begin the second dosing sequence on Day 15. Subjects will then be discharged from the clinic on Day 18, returning to the clinic on Day 23 for study assessments, and Day 30 for the EOS visit. There will be a 14-day (minimum) washout period between doses.

One cohort of 10 subjects is planned for P2. However, if PK results obtained are equivocal, additional subjects may be enrolled into the P2/FE cohort at the previously tested dose or an additional P2/FE cohort may be added at a different dose.

3.1.3. Part 3 (Multiple Ascending Dose)

Subjects in P3 will be randomized in a 3:1 ratio to receive either EIDD-2801 or PBO. Twice-daily dosing will be administered to subjects on Day 1 through Day 5, inclusive, and a final dose will be administered on the morning of Day 6 for collection of steady-state PK samples during waking hours. Depending on ongoing review of the safety, tolerability, and PK data, the number of days of dosing may be reduced and the dosing frequency may be changed; the dosing frequency will be no less than once-daily or no greater than three-times daily.

The proposed dose-escalation scheme is shown in Figure 1. The total daily dose will not exceed a dose shown to be safe and well-tolerated in P1. Doses in P3 may be administered in the fed state, following review of the PK data obtained from P2. Part 3 may run in parallel with P1, providing that the total daily dose to be administered does not exceed a dose already shown to be safe and well-tolerated in P1.

Subjects will remain domiciled at the site during the dosing period and until Day 9, returning to the clinic on Day 14 for completion of study assessments, and Day 20 for the EOS visit. If the number of days of dosing are reduced, the timepoints for subsequent assessments and the period for which subjects are domiciled at the clinic will similarly be changed (subjects will be domiciled for 3 days post final dose).

Three cohorts are initially planned for P3; however, up to an additional 6 cohorts may be enrolled, or the number of cohorts may be reduced if the study objectives are met. Dose levels in P3 may be repeated for collection of PBMCs, providing that the dose level did not meet the dose-escalation halting criteria.

Additional parts may be added to this study, including a SAD part in elderly healthy subjects and a study of the safety, tolerability, and PK in Japanese subjects. These additional parts are not described in this protocol, but may be added via a protocol amendment.

3.2. Rationale and Justification

3.2.1. Justification of Design

The FIH study is a typical dose-escalation study designed to provide the maximum amount of data in the minimum number of subjects. The cohort size in P1 and P3 is planned to be 8 subjects (6 active:2 PBO). This number of subjects allows for a sufficient PK analysis, considered to be important because dose extrapolation from efficacious animal models to humans will be based on exposure. The duration of participation for each subject following dosing well exceeds 5 drug half-lives (up to 9.1 hours in dogs; 5 hours in ferrets) [redacted] The FE cohort is considered to be important to maximize exposure based on fed vs. fasted condition and obtaining this information early in Phase 1 will minimize study drug dose in all future studies. This design follows the FDA guidance document on assessing FE in clinical studies.

3.2.2. Justification of Starting Dose

[redacted] The FDA guidance document *("Estimating the Maximum Safe Starting Dose in Initial Clinical Trials for Therapeutics in Adult Healthy Volunteers")* provides guidance for when a safety factor smaller than 10 may be used to calculate the starting dose:

- A smaller safety factor might also be used when toxicities produced by the therapeutic are easily monitored, reversible, predictable, and exhibit a moderate-to-shallow dose-response relationship with toxicities that are consistent across the tested species (both qualitatively and with respect to appropriately scaled dose and exposure).

- A safety factor smaller than 10 could be justified when the NOAEL was determined based on toxicity studies of longer duration compared to the proposed clinical schedule in healthy volunteers. In this case, a greater margin of safety should be built

into the NOAEL, as it was associated with a longer duration of exposure than that proposed in the clinical setting. This assumes that toxicities are cumulative, are not associated with acute peaks in therapeutic concentration (e.g., hypotension), and did not occur early in the repeat dose study.

EIDD-2801 satisfies both of these criteria as follows:

[redacted]

Importantly, given the activity of EIDD-2801 versus SARS-CoV-2 (cause of COVID-19), the Sponsor thinks it is most prudent to start with a dose that is predicted to be a safe starting dose and is also as close as possible to a potential therapeutic dose that would allow the Sponsor to move into COVID-19 patients as safely and expeditiously as possible. Based on modeling to animal data, a dose of 100 mg BID is projected to be an active dose in humans.

3.2.3. Justification of Study Population

Healthy volunteers are considered to be the appropriate population for conduct of the FIH study. Healthy volunteers without confounding medical conditions that may obscure the interpretation of AEs or affect absorption, distribution, metabolism and excretion of study drug will provide the most valuable data regarding the tolerability, safety, and plasma exposures observed and expected following single doses and multiple doses up to 10 days. EIDD-2801 is intended for eventual study in patients with potential CoV-2 infection as defined by the Centers for Disease Control and Prevention (CDC) in whom a range of AEs are expected based on the disease under study. Understanding the safety and PK profile in a normal population will better inform the use of EIDD-2801 in disease settings where complications are frequent, and AEs will need to be interpreted in context.

4. STUDY POPULATION

This study will enroll healthy volunteers; 8 subjects will be enrolled into each SAD and MAD cohort in P1 and P3, and 10 subjects will be enrolled into the FE cohort in P2.

The site is strongly encouraged to ensure that women are represented in each cohort.

4.1. Subject Inclusion Criteria

Subjects who meet all of the criteria at Screening will be eligible for study participation

1. Male or female between the ages of 18 and 60, inclusive.

2. Female subjects must be of nonchildbearing potential. Nonchildbearing potential is defined as women who:

    - are physiologically incapable of becoming pregnant including those who are surgically sterile, via hysterectomy, bilateral salpingectomy, and/or bilateral oophorectomy by reported medical history and/or medical records. Surgical sterilization to have occurred a minimum of 6 weeks, or at the PI's (or designee's) discretion, prior to Screening.

    - are post-menopausal (at least 45 years of age with amenorrhea for 12 months without an alternative medical reason with confirmatory FSH levels of ≥40 mIU/mL. The amenorrhea should not be induced by a medical condition such as anorexia nervosa, hypothyroid disease or polycystic ovarian disease, or by extreme exercise. It should not be due to concomitant medications that may have induced the amenorrhea such as oral contraceptives, hormones, gonadotropin-releasing hormones, anti-estrogens, or selective estrogen receptor modulators).

3. Male participants must agree to the use of effective contraception for study duration, i.e., from Check-in until 90 days after the EOS visit.

4. Is in generally good health as determined by medical history, PEs (at Screening and/or Check-in), vital signs, and other screening procedures.

5. Has a BMI of 18 to 30 kg/m^2.

6. Is willing and able to comply with all study procedures including providing informed consent.

4.2. Subject Exclusion Criteria

Subjects who meet any of the following criteria at Screening (unless otherwise stated) will not be eligible for participation:

1. Females who are pregnant, planning to become pregnant, or breastfeeding.

2. Has a clinically relevant acute or chronic medical condition or disease of the cardiovascular, GI, hepatic, renal, endocrine, pulmonary, neurologic, or dermatologic systems as determined by the PI (or designee).

3. Has any current or historical disease or disorder of the hematological system or significant liver disease or family history of bleeding/platelet disorders.

4. Has a history of cancer (other than basal cell or squamous cell cancer of the skin), rheumatologic disease or blood dyscrasias.

5. Has a history of GI surgery or other condition that may interfere with absorption of study drug, in the option of the PI (or designee).

6. Has a history of febrile illness within the 14 days prior to the first dose of study drug.

7. Has a positive alcohol or drug screen at Screening or the Day -1 visit or has a history of alcohol or drug abuse within the past 5 years.

8. Currently uses tobacco, nicotine or tobacco products or e-cigarettes or stopped using tobacco products within the past 3 months and/or has a positive cotinine test at Screening or Check-in.

9. Has any abnormal laboratory value of Grade 2 or higher, which is considered to be clinically significant by the PI (or designee).

10. Has a total white cell count or absolute lymphocyte count outside the normal range, or hemoglobin or platelet levels below the lower limit of normal, at Screening or Day -1.

11. Has values above the upper limit of normal for the following laboratory analytes: ALT/SGPT, alkaline phosphatase (serum), AST/SGOT, at Screening or Day -1.

12. Positive test result for HIV, HBV, or HCV.

13. Has an autoimmune disease, is immunosuppressed or is in any way immunocompromised.

14. Has any of the following:

    - QTcF >450 ms confirmed by repeat measurement

- QRS duration >110 ms confirmed by repeat measurement
- PR interval >220 ms confirmed by repeat measurements
- findings which would make QTc measurements difficult or QTc data uninterpretable
- history of additional risk factors for torsades de pointes (e.g., heart failure, hypokalemia, family history of long QT syndrome).

15. Except as noted, has used prescription drugs (other than hormone replacement therapy) within 14 days prior to the first dose of study drug unless, in the opinion of the PI (or designee), the drug will not interfere with study assessments. The following prescription drugs will be allowed:
    - Statins: Subjects who have been on a stable dose of statins for a minimum of 6 months and who have normal creatine kinase at Screening.
    - Antihypertensives: Subjects who have been on a stable dose of antihypertensives for a minimum of 6 months and have controlled blood pressure and no active heart disease.
16. Has received an experimental agent (vaccine, drug, biologic, device, blood product or medication) within 3 months prior to the first dose of study drug and agrees not to receive another experimental agent during the duration of this trial.
17. Has donated blood within 30 days, plasma within 2 weeks, or platelets within 6 weeks prior to Screening or refuses to agree to not donate blood until 3 months after the EOS visit.
18. Uses OTC medications or nutritional supplements (including, e.g., gingko biloba, aspirin at any dose, and NSAIDs) on a routine/scheduled basis and/or is unwilling to abstain from such use for the duration of the study.
19. Is unwilling to abstain from quinine containing products including quinine water, tonic water, bitter lemon, jello shots, any quinine preparations or Cinchona tree bark preparations (e.g., herbal medications containing Cinchona tree bark).
20. Has any condition that would, in the opinion of the PI (or designee), put the subject at increased risk for participation in a clinical trial or confinement in a clinical research unit.

5. STUDY MEDICATION, RANDOMIZATION AND DOSE ADMINISTRATION

5.1. Study Drug Description

EIDD-2801 and matching PBO will be supplied [redacted]

[redacted]

[redacted]

[redacted]

5.1.1. Acquisition, Formulation, Packaging and Labeling

[redacted]

All study drug will be labeled according to the regulatory requirements for investigational product.

5.1.2. Product Storage and Stability

Study drug should be stored at controlled room temperature defined as [redacted] If excursions occur which are outside of this range, the pharmacy staff should contact Sponsor to determine the course of action. Additional stability data may be available which would allow continued use of the study drug, or study drug may need to be replaced.

5.2. Randomization

Unmasked study drug (EIDD-2801 and matching PBO) will be supplied to the study site pharmacy. The pharmacy staff will be unmasked with regards to treatment assignment. A randomization list will be provided to the pharmacy staff who will use that list to dispense masked study drug for administration to each study participant. In P1 and P3, 6 subjects will

receive EIDD-2801 while 2 subjects will be randomized to PBO. In P2, all 10 subjects in the FE cohort will receive EIDD-2801. The pharmacy staff will maintain the security of the randomization list ensuring that no study personnel outside of the pharmacy have access to identify treatment assignment. In the case that it becomes necessary to know a subject's treatment assignment, unmasking procedures will be followed as discussed in Section 8.4.

5.3. Dosage, Preparation and Administration of Study Drug

Detailed instructions for extemporaneous compounding (as necessary), dispensing and administering study drug can be found in the pharmacy manual.

[redacted]

5.4. Drug Accountability

The site pharmacy must maintain records of receipt and disposition of all study drug supplied to the site by the Sponsor. The records must be maintained according to site standard operating procedures (SOPs) and should include at a minimum, receipt date, lot or batch number, amount and formulation received, [redacted] date, time and amount dispensed, subject identifier for each aliquot of study drug dispensed. In addition, the clinic staff must ensure that the time of dosing is identified for each subject.

Monitors must verify drug accountability/dispensing records during the monitoring visit.

Unused study drug must be disposed of according to the procedures described in the pharmacy manual.

5.5. Concomitant Medication/Treatments

[redacted] In this FIH study, limited types of concomitant medications are permitted. Adjustment of routine medications taken by subjects should be avoided during study participation except when subject safety could be affected by lack of

adjustment. There are no restrictions on treatment medications prescribed by the PI (or designee) to be used for AEs that occur during study participation. For additional restrictions, see Section 6.1.2.

6. STUDY CONDUCT AND VISIT SCHEDULE

All study assessments will be conducted according to the Time and Events Schedule.

6.1. Study Conduct

6.1.1. Study Windows and Rounding Principles

The time schedule described in the protocol for each scheduled activity should be followed as closely as possible. However, if it is not possible and if it is not otherwise specifically contraindicated per protocol, then the time windows detailed in Table 10 are allowed without incurring a protocol deviation.

Table 10: Allowable Time Windows for Study Assessments/Visits

Protocol Specified Time	Allowable Window
PK Samples and Associated Assessments	
Predose	within 2 hours prior to dosing
<2 hours	± 5 minutes
≥2 hours to 24 hours	± 15 minutes (± 2 hours for urinalysis)
>24 hours to 48 hours	± 30 minutes
>48 hours	± 1 hour (PK) and ± 2 hours (safety)
Study Visits - Parts 1 and 2	
Day −1 to Day 4 (Part 1)	must occur on scheduled day
Day −1 to Day 4 and Day 14 to Day 18 (Part 2)	must occur on scheduled day
Day 9 (Part 1) and Days 9 and 23 (Part 2)	± 1 day
End-of-Study (EOS) Visit	± 2 days
Study Visits - Part 3	
Day −1 to Day 9	must occur on scheduled day
Day 14	± 1 day
Day 20/EOS Visit	± 2 days

6.1.2. Restrictions

Prior to arriving at the clinic for the Day -1 visit, subjects must abstain from consumption of alcoholic beverages for a minimum of 72 hours prior to Check-in. Subjects must continue to abstain from consumption of alcoholic beverages throughout clinic confinement. Subjects enrolled in P2 must continue to refrain from consuming alcoholic beverages from discharge on Day 4 through Check-in on Day 14, and then through the second clinic confinement to Day 18. After discharge on Day 4 (P1), Day 18 (P2), or Day 9 (P3), subjects must minimize consumption of alcoholic beverages (i.e., limit of up to one serving per day) until the EOS procedures have been completed.

All subjects must refrain from the following:

- consuming quinine containing products from 72 hours prior to Check-in through to completion of the EOS procedures.
- using nutraceuticals and nutritional/vitamin supplements (e.g., gingko biloba, multivitamins) from 72 hours prior to Check-in through to completion of the EOS procedures. However, vitamin supplements required by a physician are exempt from this restriction.
- taking OTC analgesics including aspirin (any dose) and NSAIDS from 72 hours before Check-in until completion of EOS study procedures unless prescribed by the PI (or designee).
- use of tobacco, nicotine or tobacco products, or e-cigarettes from 3 months prior to Screening until the EOS visit.
- strenuous exercise from 7 days before Check-in until the EOS visit. Subjects will otherwise maintain their normal level of physical activity during this time (i.e., will not begin a new exercise program nor participate in any unusually strenuous physical exertion).
- Female subjects must not donate eggs/ovum from the time of Check-in until 3 months after the EOS visit.

All subjects in P1 should be dosed in the fasted state. Subjects should fast overnight for a minimum of 10 hours prior to dosing in the morning on Day 1. Following dosing, subjects may have water after 2 hours and food beginning 4 hours postdose. For subjects to be dosed in the fed state, subjects should have a high-fat breakfast as defined in the FDA guidance. Subjects must complete the meal within 30 minutes of starting the meal and should be dosed after 30 minutes of starting the meal. Doses in P3 may be administered in the fed state, following review of the PK data from P2.

6.2. Screening

6.2.1. Screening Visit

Subjects who meet preliminary pre-screening criteria (as defined by the site) and are interested in participating in the study will arrive at the study site for administration of informed consent according to site standard operating procedures (SOPs). After the subject has signed and dated the ICF, screening procedures can begin. Assessments and procedures should be conducted as shown in the Time and Events Schedule Screening may be conducted as early as 28 days prior to dosing.

6.2.2. Retesting Procedures

In the case where a parameter is outside of the acceptable limits for enrollment, measurement of the parameter can be repeated one time if both the following are met:

- In the opinion of the PI (or designee), the parameter value does not represent a chronic condition that otherwise will preclude the volunteer from enrolling in the study; and
- In the opinion of the PI (or designee), the abnormality is not likely to recur.

6.2.3. Study Visits

Subjects who satisfy entry criteria will return to the clinic for the Day -1 visit. Following review of I/E criteria, subjects who still qualify will be checked into the clinic and enrolled into the study. Following enrollment, clinical chemistry, hematology and urine samples will be collected to determine baseline values. Based on site standard practices, alternate subjects will also be enrolled into the study in case one of the selected subjects cannot be dosed. If all subjects can be dosed on the morning of Day 1, the alternates will be released and may be enrolled into subsequent cohorts. All assessments and procedures will be performed according to the Time and Events Schedule.

6.2.4. Part 1 (Single Ascending Dose)

Subjects will remain in the clinic through completion of study procedures on Day 4, returning to the clinic for procedures on Day 9.

6.2.5. Part 2 (Food-Effect)

Subjects will remain in the clinic through completion of study procedures on Day 4, returning to the clinic on Day 9 for study assessments, and Day 14 to begin the second dosing sequence on Day 15. Subjects will then be discharged from the clinic on Day 18, returning to the clinic on Day 23 for study assessments.

6.2.6. Part 3 (Multiple Ascending Dose)

Subjects will remain in the clinic through dosing and completion of study procedures on Day 9, returning to the clinic on Day 14 for study assessments. Depending on ongoing review of the safety, tolerability, and PK data, the number of days of dosing may be reduced. If the number of days of dosing are reduced, the timepoints for subsequent assessments and the period for which subjects are domiciled at the clinic will similarly be changed (subjects will be domiciled for 3 days post final dose).

6.3. Safety Follow-up and End-of-Study Visit

Subjects in P1 will return to the clinic for the EOS visit on Day 15. Subjects in the FE cohort (P2) will return for the EOS visit on Day 30. Subjects in P3 will return for the EOS visit on

Day 20. Subjects with drug-related AEs at the EOS visit will be followed as discussed in Section 8.2.1.

6.4. Subject Withdrawal and Replacement

As this is a small study with a limited number of subjects per cohort, it is critical that all subjects complete the study including postdose study assessments. Site personnel should emphasize this to study subjects at the time of informed consent so that subjects will understand this fact before agreeing to participate in the study.

Subjects may voluntarily withdraw their consent for study participation at any time without penalty or loss of benefits to which they are otherwise entitled. An PI (or designee) may also withdraw a subject from receiving study drug or participation in the study for any reason. Subjects who withdraw or are withdrawn from the study should undergo withdrawal procedures as discussed below. These procedures would include follow-up safety evaluations.

6.4.1. Reasons for Withdrawal

If a subject withdraws or is withdrawn from the study, the primary consideration must be the health and welfare of the subject. The reasons for withdrawal might include but are not limited to the following:

- Subject no longer meets eligibility criteria including subject withdraws consent from study participation (with or without a reason)
- Subject becomes noncompliant
- Medical disease or condition, or new clinical finding(s) for which continued participation, in the opinion of the PI (or designee) might compromise the safety of the subject, interfere with the subject's successful completion of this study, or interfere with the evaluation of responses
- Subject Lost-to-Follow-up
- Subject becomes pregnant, if applicable
- Determined by a physician's discretion to require additional therapy not indicated in the protocol to ensure subject's health and well-being (or treatment failure, if applicable)

The PI should be explicit regarding study follow-up (e.g. safety follow-up) that might be carried out. If the subject consents, every attempt will be made to follow all AEs through resolution, return to baseline, or until stabilized with sequelae for a maximum of 30 days following discontinuation. The procedures that collect safety data for the purposes of research must be inclusive in the original ICF or the PI may seek subsequent informed consent using an EC-approved ICF with the revised procedures.

The PI will inform the subject that already collected data will be retained and analyzed even if the subject withdraws from this study.

6.4.2. Handling of Withdrawals

Subjects who withdraw from the study prior to receiving study drug (i.e., on Day -1 or before dosing on Day 1) will be discharged from the clinic and followed only if AEs are present which occurred due to participation in the study (e.g., AE resulting from a study procedure). In this case, the subject should be followed until the AE resolves or the PI determines that the AE has stabilized.

Subjects who withdraw from the study after receiving study drug should have EOS assessments at the time of withdrawal or as quickly thereafter as possible.

Subjects who do not return for follow-up procedures on Days 9/23 or 15/30 (P1 and P2), or Days 14 or 20 (P3) will be contacted by the site at least 3 times using the subject's preferred method of communication (as determined at Check-in). If the site is unable to contact the person, then a certified letter will be sent. If the subject still cannot be reached or refuses to come back to the clinic after all attempts, the subject will be considered Lost-to-Follow-up and withdrawn from the study.

6.4.3. Documentation of Withdrawals

If a subject withdraws or is withdrawn prior to completion of the study, the reason for this decision must be recorded in the CRF. If the subject is Lost-to-Follow-up, the site should document the attempts to contact the subject in the source documents. If the subject has an ongoing AE at the time of withdrawal, then the AE should be followed as detailed in Section 8.2.1.

6.4.4. Subject Replacement

If a subject withdraws from the study prior to receiving study drug, the subject will be replaced. In this case, designated alternate subjects, if available, will be first in line to replace the withdrawn subject.

If a subject withdraws from the study after receiving study drug, then the decision to replace the subject will be made by the PI (or designee) in consultation with the Sponsor. Factors to consider will be the timing postdose of withdrawal and the number of safety and PK assessments completed prior to withdrawal. Subjects who are withdrawn because of an AE related to the study drug will not be replaced.

6.5. Unscheduled Visit(s)

If a subject experiences an AE after discharge from the clinic but prior to the EOS visit, the subject should be instructed to call the site. Based on the issue, the PI (or designee) may request that the subject return to the site for an unscheduled visit. In this case, procedures/assessments

should be conducted as deemed appropriate for the situation by the PI (or designee). The visit should be recorded in the unscheduled visit page of the CRF.

7. STUDY PROCEDURES/EVALUATIONS

7.1. Demography and Medical History

Demographics including age, gender, race, ethnicity, and medical history will be recorded for each subject. All significant medical history should be recorded. In general, significant medical history should include all ongoing events and all events occurring within the last 6 months. Clinically relevant or clinically significant events occurring greater than 6 months ago should be recorded. All surgeries occurring in adulthood should be recorded. If surgeries occurred more than 2 years ago, then only the year needs to be recorded on the CRF.

7.2. Clinical Evaluations

7.2.1. Physical Examinations

The PE will be performed by the PI or a designee that is licensed to perform a PE per local requirements. The initial PE performed at Screening and the final PE conducted at the EOS visit will include examination of all pertinent body systems as defined by the site standard PE body systems (general appearance, HEENT, lymphatic, cardiovascular, respiratory, GI, musculoskeletal, neurological, dermatological).

Subsequent PEs will be performed as shown in the Time and Events Schedule and will be targeted to any new signs or symptoms, any change from the previous status or any body system deemed pertinent to that subject by the PI (or designee). Clinically significant abnormalities should be recorded in the CRF; those occurring prior to dosing will be included in medical history unless the abnormality was the direct result of study participation.

7.2.2. Vital Sign Measurements and ECGs

Vital signs include pulse rate, respiratory rate, blood pressure, and body temperature. Vital signs should be measured after the subject has been supine for a minimum of 5 minutes. Site standard ranges will be used for determining any out-of-range values.

Height and weight should be measured, and BMI calculated at Screening as indicated in the Time and Events Schedule.

Resting 12-lead ECGs should be recorded at the visits indicated in the Time and Events Schedule after the subject has been supine for a minimum of 5 minutes. The PI (or designee) will evaluate the ECG tracings to determine if there are out-of-range values; if out-or-range values are detected, the PI (or designee) will determine if they are clinically significant. Site standard ranges will be used to determine if any parameters are considered out-of-range. At the discretion of the PI (or designee), the ECG may be repeated if erroneous readings are suspected.

7.2.2.1. Continuous 12-lead ECG Monitoring (Parts 1 and 3 Only)

Continuous 12-lead ECG monitoring using a digital recorder will take place at the times indicated in the Time and Events Schedule, in P1 and P3 only.

All continuous 12-lead ECG data collected will be archived without extraction or analysis and will not be reported in the scope of this study.

Subjects will be supine for at least 10 minutes before the extraction timepoint and for 5 minutes from the start of each extraction timepoint (each extraction will last for 5 minutes). Environmental distractions (e.g., television, radio, conversation) should be avoided during the pre-ECG resting period and during ECG recording.

When coinciding, PK sampling should always be performed after the ECG extraction time window. If a separate ECG machine is being used for safety assessments, that machine should be in place prior to the extraction window to permit safety ECGs to be recorded irrespective of the extraction window. If the machine is not in place prior to the extraction window, safety ECGs must be recorded after the extraction window. If an integral system is used, safety ECGs may be recorded irrespective of the extraction window.

7.2.3. Adverse Events and Concomitant Medications

Adverse Events: The PI is responsible for identifying and documenting events meeting the definition of an AE or SAE (Section 8.1). Once each day while the subject is in the clinic and once during each out-patient visit, the PI (or designee) should inquire about the occurrence of AEs. The following are examples of open-ended questions that may be used to obtain this information: *"How are you feeling?"; "Have you had any medical problems recently?"; "Have you taken any new medicines since your last visit/assessment?"*

All AEs and SAEs must be documented in the source documents and recorded in the CRF.

Concomitant Medications: All medications (prescription or over-the-counter), nutritional supplements, and nutraceuticals taken by the subject from 30 days prior to dosing through the EOS visit must be recorded in the CRF. Medication information should include indication, dose, frequency, and route of administration. Any medication taken for an AE/SAE should be documented as such. Refer to Section 5.5 for additional information.

7.3. Laboratory Evaluations

The laboratory will perform standard routine testing, and processing of all blood samples. For the entire study, the amount of blood collected from any one subject will not exceed 500 mL.

7.3.1. Routine Laboratory Panels

Blood and urine samples will be collected at the times indicated in the Time and Events Schedule. The analytes shown in the table below (Table 11) will be assessed.

If a method to determine COVID-19 status becomes readily available, subjects may be tested at Screening and Check-in to confirm they are not positive for COVID-19 prior to dosing.

Table 11: Laboratory Evaluations

CHEMISTRY PANEL Alanine Aminotransferase (ALT/SGPT) Albumin, Serum Albumin/Globulin (A/G) Ratio (calculation) Alkaline Phosphatase, Serum Amylase Aspartate Aminotransferase (AST/SGOT) Bilirubin, Total and Direct BUN BUN/Creatinine Ratio (calculation) Calcium, Serum Creatinine, Serum Creatinine Kinase (CK) Gamma Glutamyl Transferase (GGT) Lactate Dehydrogenase (LDH) Uric Acid Electrolyte Panel (Na+, K+, Cl-, Bicarb.) Phosphorus Globulin, Total Glucose, Serum Lipase Protein, Total, Serum	**HEMATOLOGY PANEL** Hemoglobin Hematocrit White Blood Cell Count with differential (absolute and percentage) Red Blood Count Prothrombin Time (PT)/Partial Prothrombin Time (PTT) and International Normalized Ratio (INR) Mean cell hemoglobin (MCH), mean cell hemoglobin concentration (MCHC), mean cell volume (MCV), red cell distribution width (RDW; may be a Grade 1 abnormality) Platelets
	ADDITIONAL ASSESSMENTS Virology: Human Immunodeficiency Virus (HIV) serology, Hepatitis B Virus (HBV; Surface Antigen [HBsAg]), Hepatitis C Virus (HCV) Follicle-Stimulating Hormone (FSH; as applicable)
	PREGNANCY TEST Serum Pregnancy Test Urine Dipstick (optional blood follow-up)
	DRUG SCREENING Serum/urinalysis (per site SOP) Cotinine Urine Dipstick Alcohol Breathalyzer
	ROUTINE URINALYSIS Bilirubin Color and appearance Glucose Ketones Leukocytes Microscopic (including red blood cells [RBCs] and white blood cells [WBCs]) Nitrite Occult blood pH Protein Specific Gravity Urobilinogen

7.3.2. Pharmacokinetic Sampling

Samples for PK analysis will be collected as shown in the Time and Events Schedule. All samples will be analyzed to define the PK parameters for prodrug EIDD-2801 and the active parent, EIDD-1931. In order to preserve EIDD-2801, special handling procedures will be put in place. These procedures will be documented in the laboratory manual for the study.

Depending on ongoing review of the PK data, timepoints may be modified or removed, and up to 3 additional samples (in each period for the food-effect) may be added.

7.3.3. Peripheral Blood Mononuclear Cell Collection

Peripheral blood mononuclear cells will be collected in P2 and P3. Blood will be collected into specialized tubes and processed according to procedures described in the laboratory manual.

Depending on ongoing review of the data, timepoints may be modified or removed, and up to 2 additional samples may be added.

7.3.4. Urine Collection

Urine will be collected over the time periods noted in the Time and Events Schedule. Samples will be collected for routine urinalysis and PK analysis according to site standard practices and as described in the laboratory manual.

8. SAFETY MONITORING, MANAGEMENT AND REPORTING

8.1. Definitions

8.1.1. Adverse Events

An AE is 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 an abnormal laboratory finding), symptom, or disease temporally associated with the use of a medicinal (investigational) product, whether or not related to the medicinal (investigational) product.

Abnormal clinical laboratory findings (e.g. clinical chemistry, hematology) or other abnormal assessments that are judged by the PI (or designee) as clinically significant will be recorded as AEs or SAEs if they meet the definitions of an AE or an SAE as defined in this Section 8.1.2. Disease specific signs and symptoms which were ongoing prior to study entry will not be considered AEs unless they worsen (e.g. increase in frequency or severity) unexpectedly during the course of the trial.

8.1.2. Serious Adverse Events

An AE or suspected adverse reaction is considered "serious" if, in the view of either the PI (or designee) or Sponsor, it results in any of the following outcomes:

- Death,
- A life-threatening AE: An AE or suspected adverse reaction is considered "life-threatening" if, in the view of either the PI or Sponsor, its occurrence places the patient or subject at immediate risk of death. It does not include an AE or suspected adverse reaction 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 hospitalizations may be considered serious when, based upon appropriate medical judgment they may jeopardize the patient or 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.

Suspected adverse reaction means any AE for which there is a reasonable possibility that the drug caused the AE. "Reasonable possibility" means there is evidence to suggest a causal relationship between the drug and the adverse event. Suspected adverse reaction implies a lesser degree of certainty about causality than adverse reaction, which means any adverse event caused by a drug.

An AE or suspected adverse reaction is considered "unexpected" if

- It is not listed in the IB or is not listed at the specificity or severity that has been observed; or, if an IB is not required or available,
- Is not consistent with the risk information described in the general investigational plan or elsewhere in the current application as amended. For example, under this definition, hepatic necrosis would be unexpected (by virtue of greater severity) if the IB referred only to elevated hepatic enzymes or hepatitis. Similarly, cerebral thromboembolism and cerebral vasculitis would be unexpected (by virtue of greater specificity) if the IB listed only cerebral vascular accidents.
- "Unexpected" as used in this definition, also refers to AE or suspected adverse reactions that are mentioned in the IB as occurring with a class of drugs or as anticipated from the pharmacological properties of the drug, but are not specifically mentioned as occurring with the particular drug under investigation.

As of the date of this protocol, there are no expected events listed in the current version of the IB; therefore, all AEs will be considered unexpected until such a time that the reference safety information in the IB is updated with any identified, expected events.

8.2. Documenting Adverse Events

8.2.1. Timeframe for Collection and Follow-up of AEs/SAEs

All AEs/SAEs will be collected from the time of the first study drug administration until the subject has completed the EOS visit and been discontinued from the study. This includes subjects who discontinue early. Events considered related to study drug will be followed as noted:

- AEs that are related to study drug and continue beyond the normal collection period (i.e., are ongoing at the time a subject exits the study) will be followed until resolution, return to baseline or until stabilized with sequelae for a maximum of 30 days following discontinuation. After 30 days, the AE will be closed, and the outcome noted (see Table 12).
- SAEs that are related to study drug and continue beyond the normal collection period

(i.e., are ongoing at the time a subject exits the study) will be followed until resolution, return to baseline or until stabilized with sequelae.

- Serious AEs that are reported to the site within 30 days after the subject has been discontinued from the study (i.e., completed the EOS visit) will be recorded. Those that are considered related to study drug will be followed as noted in the bullet above.

Note that all events which occurred prior to dosing with study drug should be recorded as medical history unless the event is directly related to study procedures.

8.2.2. Recording of Adverse Events/Serious Adverse Events

AEs/SAEs must be recorded in the CRF as indicated in the CRF completion instructions. Information to be collected includes event description, time of onset, clinician's assessment of severity (Section 8.2.3), relationship to study drug (Section 8.2.4), outcome (Section 8.2.5), and time of resolution/stabilization of the event. All AEs occurring while on study must be documented appropriately regardless of relationship and will be followed to adequate resolution as described above (Section 8.2.1). All SAEs will be recorded as noted above; SAEs reported to the site within 30 days following the EOS visit will also be recorded. All SAEs must be entered onto the SAE form and reported as discussed below (Section 8.3.1).

If an AE changes in severity, the highest severity will be recorded in the CRF. AEs characterized as intermittent require documentation of onset and duration of each episode.

8.2.3. Assessing Severity of Adverse Events

All AEs/SAEs will be assessed by the PI or those with the training and authority to make a medical judgment. AEs/SAEs will be graded according to the DMID Toxicity Grading Scale. For any AEs not specifically listed in the tables, the following guidelines should be used to grade severity:

- **Mild** (Grade 1); asymptomatic or mild symptoms; clinical or diagnostic observations only; intervention not indicated.
- **Moderate** (Grade 2); minimal, local or noninvasive intervention indicated; limiting age-appropriate instrumental activities of daily living.
- **Severe** (Grade 3); medically significant but not immediately life-threatening; hospitalization or prolongation of hospitalization indicated; disabling; limiting self-care activities of daily living.
    - **Life-threatening**; life-threatening consequences; urgent intervention indicated.
    - **Death**; death related to AE.

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

8.2.4. Relationship to Study Drug

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

- **Definite –** The AE is clearly related to the study drug**.**
- **Probable –** The AE is likely related to the study drug.
- **Possible –** The AE may be related to the study drug.
- **Unlikely –** The AE is doubtfully related to the study drug.
- **Unrelated –** The AE is clearly NOT related to the study drug.

8.2.5. Classifying Adverse Event Outcome

All AEs/SAEs in the study must be assigned an outcome by site staff. The outcome will be included on the AE CRF. Possible outcomes are shown below:

Table 12: Adverse Event Outcomes

Outcome	Description
Recovered / Resolved	AE resolved with no residual signs or symptoms; an event is considered resolved if it returns to baseline (pretreatment) values.
Recovered / Resolved with sequelae	AE stabilized but residual signs or symptoms remain; this includes stabilization of an event/condition with the expectation that it will remain chronic.
Not Recovered / Not Resolved	AE remains ongoing AND no or only minimal improvement has occurred.
Ongoing	AE has not yet resolved, but continues to improve/resolve and complete resolution is expected over time.
Fatal	Outcome of the AE is death.
Unknown	AE outcome is not known; usually because the subject has been Lost-to-Follow-up.

8.3. Reporting Procedures

8.3.1. Serious Adverse Events

The PI or clinical site personnel should notify Covance Drug Safety Services (DSS) of all SAEs, regardless of relationship to the investigational drug, within 24 hours of clinical site personnel becoming aware of the event. The PI (or designee) will provide the initial notification by sending a completed "SAE Notification Form," which must include the PI's (or designee's) assessment of the relationship of the event to investigational drug and must be signed by the PI. Follow-up information, or new information regarding an ongoing SAE, must be provided promptly to Covance DSS.

The responsibilities of Covance DSS include the following:

- Prepare an AE reporting plan prior to the start of the study. Where this plan differs from the applicable site standard operating procedure on SAE reporting, the AE reporting plan will always take precedence.

- Receive and review SAE report forms from the site and inform the Sponsor of the SAE within 1 working day of the initial notification to DSS. Drug Safety Services will delete any information from the SAE report forms that may identify the subject.

- Write case narratives and enter the case into Covance's safety database as defined in the AE reporting plan.

- Produce appropriate reports of all Suspected Unexpected Serious Adverse Reactions and forward to the EC, Medicines and Healthcare Products Regulatory Agency, PI's (or designee's), and the Sponsor within the timeframes stipulated in the Clinical Trials Directive Guideline (ENTR/CT 3).

The responsibility for reporting SAEs will be transferred to the Sponsor 28 days after the end of the study.

8.3.2. Pregnancy

Subjects in this study must be of non-childbearing potential. However, should a pregnancy occur any time from informed consent to the EOS visit, subjects must immediately report the event to the clinical site which in turn must immediately report the pregnancy to the Sponsor or their designee. The subject will be followed until the end of the pregnancy. A separate ICF will be used for consenting for follow-up pregnancy activities. Pregnancy will not be considered an AE unless deemed likely related to study drug. Pregnancy will not be considered an SAE unless there is an associated SAE. A spontaneous abortion (miscarriage) or abnormal outcome (including congenital anomalies) will be reported as an SAE.

8.4. Unmasking Treatment Assignment

The following controls will be employed to maintain the double-blind status of the study:

- Placebo will be identical in appearance to the EIDD-2801.
- The PI and other members of staff involved with the study will remain blinded to the treatment randomization code in P1 and P3.
- Interim bioanalytical data will be provided in a blinded manner.

To maintain the blind, the PI will be provided with a sealed randomization code for each subject in P1 and P3, containing details of their treatment. These individual sealed envelopes will be kept in a limited access area that is accessible 24 hours a day. In order to manage subject safety or to support dose-escalation decisions (in the event of possibly treatment-related SAEs or severe AEs), the decision to unblind resides solely with the PI. Whenever possible and providing it does not interfere with or delay any decision in the best interest of the subject, the PI will discuss the intended code-break with the Sponsor. If it becomes necessary to break the code during the study, the date, time, and reason will be recorded in the subject's source data and on the individual envelope and will be witnessed by a second person.

The pharmacy and bioanalytical lab will have access to the treatment randomization and will be unblinded. Pharmacokinetic personnel may be unblinded to perform interim PK analysis and to ensure that PK data are provided in a blinded manner for dose-escalation decisions.

9. DOSE-ESCALATION AND HALTING RULES

9.1. Guidelines for Dose-Escalation

Doses will be administered in an escalating manner following satisfactory review by the Sponsor and PI of the blinded safety and tolerability data up to 72 hours post final dose. [redacted] As PK data become available, these data will be reviewed and may be used to assist in dose selection.

Doses may be reduced and may be lower than the starting dose. There will be a minimum of 4 days between dose escalations to allow sufficient time for an adequate safety review. [redacted]

Integrated data from all applicable sites will be used to make dose-escalation decisions.

Dose-escalation in P1 and P3 will only occur if data from a minimum of 6 subjects (across all sites) have been reviewed from the previous lower dose group, such that data from a minimum of 4 subjects (across all sites) who have received EIDD-2801 will be used to make the dose-escalation decision.

The justification for this is as follows:

- The study treatment is of a known pharmacological class for which the on-target effects in humans are well characterized. Based upon nonclinical data, no clinically important off target effects are expected within the proposed dose range.

- A minimum of 4 subjects receiving the active drug is considered sufficient to characterize the safety profile of EIDD-2801.

Between each dose-escalation, the PI will review all available data in a blinded manner to ensure it is safe to proceed with the planned dose-escalation. The results from all available safety assessments will be sent to the Sponsor prior to the start of each successive group/treatment period. Any clinically significant results will be discussed with the Sponsor before dose-escalation continues. Interim PK data may also be reviewed in terms of dose-escalation and to confirm that the study design remains appropriate. In the event of a disagreement between Sponsor and PI on the dose-escalation decision, the most conservative decision will be upheld.

Dose levels will not increase by more than 3-fold for predicted nonpharmacologically active dose levels and by 2-fold for predicted pharmacologically active dose levels. The highest total daily dose to be studied under this protocol will not exceed 1600 mg (Section 9.2).

Dose levels in P3 may be repeated for collection of PBMCs, providing that the dose level did not meet the dose-escalation halting criteria.

9.2. Rationale for Highest Dose

9.2.1. Clinical Safety

9.2.1.1. Single Ascending Doses

At the time of Protocol Version 5.1, 48 healthy subjects were administered a single dose of 50, 100, 200, 400, 600, and 800 mg EIDD-2801 or placebo in P1. There were 6 males and 2 females at each dose level, with the exception of the 800 mg dose level, which included 8 males. The age range across all dose levels was 21 to 60 years. No clinically significant abnormalities or changes from baseline were reported for vital signs, ECGs, laboratory safety assessments or PEs.

Thirty-three treatment-emergent AEs were reported across the 6 dose levels; 32 of these were mild and 1 were moderate in severity. Two AEs were considered to be probably related, 9 were possibly related, 15 were unlikely related, and 6 were unrelated to EIDD-2801.

The most frequently reported AEs were headache (13 reports), nausea (4 reports), and backache (3 reports).

- Four reports of headache (1 at 100 mg, 1 at 200mg, and 2 at 600 mg) were considered possibly related to EIDD-2801 and 1 (at 600 mg) was considered to be probably related; all were mild in severity.
    - One headache (at 600 mg) that was probably related and 2 headaches (1 at 100 mg and 1 at 200mg) that were possibly related started within 2 hours of dosing and resolved without treatment within 24 hours, and 2 headaches (both at 600 mg) that were possibly related started later, but had resolved without treatment within 30 hours.
    - One headache (at 100 mg) resolved after a single dose of 1 g acetaminophen.
- At 100 mg, 1 subject reported nausea that was considered possibly related to EIDD-2801 because it started approximately 4 hours after dosing; this event was moderate in severity and resolved approximately 40 minutes after onset following a single episode of vomiting. At 200 mg, 1 subject reported nausea that was considered possibly related to EIDD-2801. At 600 mg, 1 subject reported 2 episodes of nausea that were considered unlikely related to EIDD-2801 because they started at 28 hours and nearly 36 hours postdose.
- Three reports of backache were considered unrelated to EIDD-2801.
- Other AEs of interest included 1 report of each of loose stools and abdominal pain; both were mild in severity and reported by the same subject at 200 mg. The loose stools were

considered probably related to EIDD-2801; they started approximately 13.5 hours after dosing and resolved 3 days after dosing. The abdominal pain started approximately 24 hours after dosing, resolved after 4.5 hours, and was considered to be probably related to EIDD-2801.

Laboratory safety data have been reviewed through Day 14 at all dose levels. There were no clinically significant abnormalities or changes from baseline. There was 1 decrease in platelet counts to below the lower limit of normal: 1 subject (at 600 mg) had a value of $147 \times 10^3/\mu L$ (normal range: 150 to $357 \times 10^3/\mu L$) on Day 9; however, this was not considered clinically significant because the subject had values of $150 \times 10^3/\mu L$ and $178 \times 10^3/\mu L$ on Days -1 and 14, respectively.

9.2.1.2. Multiple Ascending Doses

At the time of Protocol Version 5.1, 32 healthy subjects were administered multiple doses of 50, 100, 200, and 300 mg EIDD-2801 or placebo BID in P3. There were 27 males and 5 females across all dose levels, with ages ranging from 20 to 60 years. No clinically significant abnormalities or changes from baseline were reported for vital signs, ECGs, laboratory safety assessments, or PEs.

Seventeen treatment-emergent AEs were reported across the 4 dose levels; 15 of these were mild and 2 were moderate in severity. One AE was considered to be probably related, 4 were possibly related, 6 were unlikely related, and 6 were unrelated to EIDD-2801.

The most frequently reported AEs were lower back pain (4 reports), headache (2 reports), and loose stools (2 reports).

- Four reports of lower back pain were mild in severity and were considered unlikely related or unrelated to EIDD-2801.
- Two reports of headache (both at 300 mg BID) were mild in severity.
    - One was considered unlikely related as it started at predose on the final day of dosing.
    - One was considered possibly related because it started 8 hours and 40 minutes after the first dose on Day 1 and lasted for 4 hours.
- Two reports of loose stools were mild in severity and considered possibly related to EIDD-2801.
    - One (100 mg BID) started approximately 48 hours after the final dose on Day 6 and resolved after 5 hours and 30 minutes.
    - One (200 mg BID) started 5 hours and 30 minutes after the first dose on Day 1 and resolved after approximately 30 hours.

One report of gurgling stomach was considered probably related to EIDD-2801 (200 mg BID). This occurred in the subject who reported loose stools, started 5 hours after the first dose on Day 1, and lasted for 20 minutes.

The only other AE considered possibly related to EIDD-2801 was a report of dizziness while standing (200 mg BID), which started 1 hour postdose on Day 1 and lasted for 30 minutes. This event was not associated with any changes in vital signs or ECGs.

Laboratory safety data have been reviewed through Day 20 at all dose levels. There were no clinically significant abnormalities or changes from baseline. There was 1 decrease in platelet counts to below the lower limit of normal: 1 subject (300 mg BID) had a value of $130 \times 10^3/\mu L$ on Day 9 (normal range: 150 to $357 \times 10^3/\mu L$; Screening value was $171 \times 10^3/\mu L$); however, this was not considered clinically significant because the subject had a value of $149 \times 10^3/\mu L$ on Day 14.

9.2.2. Pharmacokinetics

9.2.2.1. Single Ascending Doses

Pharmacokinetic data [redacted] have been reviewed up to 72 hours postdose for doses up to 800 mg. Concentrations of EIDD-2801 (the ester prodrug) were quantifiable in the 0.25 hours postdose samples only for the majority of subjects following administration of 600 and 800 mg doses of EIDD-2801, thus PK parameters, with the exception of C_{max} and t_{max}, were not calculable for EIDD-2801. EIDD-1931 (the active drug) appeared rapidly in plasma, with a median t_{max} of 1 hour at all dose levels. EIDD-1931 exposure increased in a dose-proportional manner between 50 and 800 mg. EIDD-1931 concentrations declined in a monophasic manner and remained quantifiable until 4 to 6 hours postdose up to 100 mg, until 9 hours at 200 mg, and between 9 and 12 hours at 400 mg and above. The geometric mean apparent terminal elimination half-life was approximately 1 hour (0.945 to 1.06 hours across dose groups) at doses up to 600 mg, and increased to 1.29 hours at 800 mg.

Geometric mean systemic exposure is predicted to achieve an approximate area under the concentration versus time curve from time zero to 24 hours postdose (AUC_{0-24}) of 14,000 ng•h/mL and C_{max} of 5480 ng/mL at 1200 mg and is predicted to achieve an approximate AUC_{0-24} of 18,700 ng•h/mL and C_{max} of 7310 ng/mL at 1600 mg [redacted]

9.2.2.2. Multiple Ascending Doses

Pharmacokinetic data [redacted] have been reviewed up to 72 hours postdose for doses up to 300 mg BID. A single concentration of EIDD-2801 (the ester prodrug) was quantifiable in a single sample at each of the 100 (Day 1, 0.5 hours) and 200 mg (Day 6, 0.5 hours) BID dose levels; however, no concentrations of EIDD-2801 were quantifiable in any of the samples at 300 mg BID, thus PK parameters were not calculable for EIDD-2801 at any dose level. EIDD-1931 (the active drug) appeared rapidly in plasma, with a median t_{max} on Days 1 and 6 of

1 hour at 50 mg BID, 1.25 hours at 100 mg BID, and 1.5 hours at 200 and 300 mg BID. EIDD-1931 exposure increased in a dose-proportional manner between 50 and 300 mg BID on Day 6. EIDD-1931 concentrations declined in a monophasic manner and remained quantifiable until 4 and 9 hours postdose up to 100 mg BID on Days 1 and 6, increasing to between 9 and 12 hours at 200 mg BID and 300 mg BID on Day 1 and Day 6, with 1 subject maintaining quantifiable concentrations until 24 hours postdose at 300 mg BID on Day 6. The geometric mean terminal elimination half-life was approximately 0.9 to 1.0 hours on Day 1 for all dose levels. On Day 6, the geometric mean terminal elimination half-life increased from 0.968 hours at 50 mg BID to 1.71 hours at 300 mg BID. There was no significant accumulation at any dose level. Exposure on Day 1 were generally similar to those at the corresponding dose in Part 1 (comparable up to 200 mg), i.e., there was no clear difference in PK between the [redacted] formulations. This assessment is made with the caveat that the data have not been formally, statistically assessed and the results are from different subjects (not crossover).

Geometric mean systemic exposure is predicted to achieve an approximate AUC_{τ} of 9060 ng•h/mL and C_{max} of 2900 ng/mL at 800 mg BID.

9.2.3. Rationale to Exceed the Maximum Single Dose Limit of 800 mg

[redacted]

Based on the clinical data collected to date, it is considered acceptable to exceed 800 mg because EIDD-2801 has been well tolerated at the single and multiple dose levels tested to date, and the PK has behaved in a predictable manner.

[redacted]

Because of the uncertainty of the virologic course of COVID-19, it is prudent to establish a safety margin above the predicted effective dose to allow for maximum viral suppression, especially where treatment is started later in disease where a rapid reduction of viral replication is important. Therefore, a potential treatment of 400 and/or 800 mg BID will be investigated. In order to escalate to this dose level in P3, a dose level of 1600 mg must be shown to be safe and well tolerated in P1. Additionally, in order to allow for unpredicted differences in PK at higher

dose levels compared with that seen at the dose levels already evaluated, dose levels of 1200 and 1600 mg are planned for P1; this will allow escalation to a maximum of 800 mg BID in P3 if required to achieve therapeutic exposures.

[redacted]

9.3. Dose-Escalation Halting Rules

For Group 1 in P1, sentinel dosing will occur whereby 2 subjects (1 active and 1 PBO) will be dosed on 1 day and, providing no safety concerns arise, the remaining 6 subjects will be dosed after at least 24 hours.

The study will be halted (i.e., no further dosing will occur) if one or more subjects experience an SAE that is considered to be related to EIDD-2801 or 2 or more subjects in the same group experience severe AEs that are considered to be related to EIDD-2801. If, following an internal safety review, the Sponsor deems it appropriate to restart the study, this can be done following approval of a substantial protocol amendment.

Additionally, if 2 or more subjects experience a reduction of platelets greater than 50% over baseline levels or a reduction in lymphocytes (absolute count) to Grade 3/severe levels without an alternate cause, the PI and Sponsor will review the data. If, after review of unblinded data, it is determined that these events are probably related to study drug, no further dose escalations will occur. In this case, additional cohorts may be enrolled at a lower dose.

Dose-escalation will stop if clinically relevant signs or symptoms of similar nature occur in 2 or more subjects in a group that, in the opinion of the PI, warrant stopping of dose-escalation.

10. STATISTICAL CONSIDERATIONS

10.1. General Considerations

All summaries will be provided by study part and treatment. Continuous variables will be summarized using descriptive statistics including number of observations (n), mean, standard deviation, minimum (min), median (med), and maximum (max). Categorical variables will be summarized using frequency counts and percentages. Note that PBO data from each dose level in Parts 1 and 3 will be combined into one PBO group.

No missing data imputation will be performed.

Subject listings will be provided for all the data collected during the study period.

Specific information about the statistical analysis will be provided in a SAP that will be reviewed and approved by the Sponsor and will be finalized before final database lock. If there is a discrepancy between the methods described in the protocol and final approved SAP, the SAP will take precedence.

10.2. Sample Size Considerations

No formal sample size calculation was conducted. The sample size of 8 per cohort (6 active: 2 PBO) for the SAD and MAD cohorts is considered adequate for a Phase 1 FIH study. The sample size of 10 subjects (all administered EIDD-2801) is in accordance with FDA guidelines for sample size in FE studies.

10.3. Analysis Populations

Safety Population: All subjects who receive at least one dose of study drug.

Pharmacokinetic Population: All subjects receiving study drug who comply sufficiently with protocol requirements (i.e., no major protocol violations) and for whom a sufficient number of analyzable PK samples have been obtained to permit the determination of the PK profile for EIDD-2801/EIDD-1931 and the statistical analysis of the results. Subjects who experience an AE of vomiting before 2× the median t_{max} of the group may be excluded from the PK population.

10.4. Analysis of Safety Data

All safety analyses will be performed on the Safety Population as defined in Section 10.3. Safety will be assessed on the basis of AEs, clinical laboratory data, vital signs, ECG parameters, and PEs.

10.4.1. Extent of Exposure

Dosing data will be listed by study part.

10.4.2. Adverse Events

Adverse events will be coded using Medical Dictionary for Regulatory Activities (MedDRA) preferred term and system organ classification. Any events reported after the initiation of study treatment and through the EOS are defined as treatment-emergent. The occurrence of treatment-emergent AEs will be summarized using MedDRA preferred terms, system organ classifications, and severity. Separate summaries of treatment-emergent SAEs and AEs considered related to study treatment and AEs leading to study treatment discontinuation will be generated. All AEs will be listed for individual subjects showing both verbatim and preferred terms.

10.4.3. Clinical Laboratory Results

Laboratory abnormalities will be graded according to the DMID Toxicity Grading Scale. Any graded abnormality that occurs following the initiation of study treatment and represents at least a one-grade increase from the baseline assessment is defined as treatment-emergent. The number and percentage of subjects experiencing treatment-emergent graded toxicities will be summarized. Raw values and mean changes from baseline in clinical laboratory measures will be summarized.

Listings of the clinical laboratory test results will be provided. Abnormal laboratory values will be flagged in the listings.

10.4.4. Other Parameters

Individual data for ECG parameters and vital sign measurements will be listed by subject and time point and summarized for each treatment. Individual data for PE will be listed by subject and time point.

Concomitant medications will be coded using the World Health Organization-Drug Dictionary (WHO-DD) and summarized.

10.5. Analysis of Pharmacokinetic Data

Pharmacokinetic analysis as defined in the SAP will be conducted using the PK population defined in Section 10.3. In the event of discrepancies between analyses described in the SAP and this clinical study protocol, the SAP will supersede the protocol.

- All samples will be analyzed and all concentrations listed.
- Descriptive statistics will be performed for all time points available, with the exclusion of subjects who had any significant protocol deviation.
- Pharmacokinetic parameters will be derived where possible for all subjects. Data from subjects with incomplete profiles (missed blood draws, lost samples, samples unable to be quantified) may be used if PK parameters can be estimated using the remaining data points.

- Descriptive statistics will be performed on all parameters available, and any missing parameters will be flagged.

Plasma concentration data for EIDD-2801 and EIDD-1931 will be listed for individual subjects and summarized by study part and treatment. Individual and mean plasma concentration versus time plots for EIDD-2801 and EIDD-1931 will be provided. Urine concentration data for EIDD-2801 and EIDD-1931 will be listed.

Plasma PK parameters of EIDD-2801 and EIDD-1931 for each subject will be estimated over the sampling interval using noncompartmental analysis and summarized by study part and treatment using descriptive statistics. Actual blood sampling times will be used for plasma PK analysis.

Urine PK parameters of EIDD-2801 and EIDD-1931 will also be analyzed and summarized when possible. The PK parameters that will be estimated are listed in the table below.

Table 13: Pharmacokinetic Parameters

Plasma PK Parameter	Description
AUC_{last}	The area under the plasma concentration-time curve, from time 0 to the last measurable non-zero concentration, as calculated by the linear up/log down trapezoidal method (P1 and P2 only).
$AUC_{0\text{-}inf}$	The area under the plasma concentration-time curve from time 0 extrapolated to infinity. $AUC_{0\text{-}inf}$ is calculated as the sum of AUC_{last} plus the ratio of the last measurable plasma concentration to the elimination rate constant (λz) (P1, P2, and Day 1 dose of P3).
C_{max}	Maximum observed concentration.
t_{max}	Time to reach C_{max}. If the maximum value occurs at more than one time point, t_{max} is defined as the first time point with this value.
λz	Apparent terminal elimination rate constant; represents the fraction of medication eliminated per unit time.
t½	Apparent terminal elimination half-life of medication, calculated as 0.693/λz.
Vz/F	Apparent volume of distribution (EIDD-2801 only).
CL/F	Apparent oral drug clearance (EIDD-2801 only).
C_{trough}	Trough concentration (P3 only).
AUC_{τ}	The area under the plasma concentration-time curve during a dosing interval (P3 only).
$RA_{AUC\tau}$	Observed accumulation ratio based on AUC_{τ} (P3 only).
RA_{Cmax}	Observed accumulation ratio based on C_{max} (P3 only).
Urine PK Parameter	**Description**
A_e	Amount excreted in urine over the sampling interval.
F_e	Fraction of drug excreted in the urine over sampling interval.
CL_R	Renal clearance, $CL_R = A_e/AUC$ where both A_e and AUC are determined over co-incident time ranges after dosing.

Additional PK parameters may be analyzed as appropriate.

10.5.1. Statistical Analysis of Pharmacokinetic Data

10.5.1.1. Dose Proportionality

In P1, dose-proportionality for PK parameters of EIDD-2801 and EIDD-1931 ($AUC_{0\text{-}inf}$, AUC_{last}, and C_{max}) will be assessed by the power model. The slope and the associated 90% CI based on the power model will be reported.

In Part 3 on Day 6 dose-proportionality for PK parameters of EIDD-2801 and EIDD-1931 (AUC_{τ}, and C_{max}) will be assessed by the power model. The slope and the associated 90% CI based on the power model will be reported.

10.5.1.2. Food-Effect

To assess the effect of food on the PK of EIDD-2801 and EIDD-1931, natural log transformed PK parameters from P2 ($AUC_{0\text{-}inf}$, AUC_{last}, and C_{max}) will be analyzed using a mixed-effects model with fixed-effect terms for treatment (with or without food), period, and sequence and with subject within sequence as a random effect. Point estimates and their associated 90% CIs for the natural log-transformed PK parameters for the difference between EIDD-2801 administered under fed relative to fasted conditions will be constructed. The point estimates and interval estimates will be back transformed to give estimates of the ratio and 90% CI for the geometric least squares mean ratio for fed/fasted to assess the effect of food.

10.6. Peripheral Blood Mononuclear Cells

Peripheral blood mononuclear cells will be collected from subjects following each dose in P2. Depending upon ongoing review of the data, PBMCs may be collected from subjects in P3; the collection of these samples may be omitted from some cohorts in P3.

The PBMCs will be stored for analysis of EIDD-2061 levels at some point in the future and reported separately.

10.7. Interim Analyses

There are no formal interim analyses planned for this study. However, interim analyses may be implemented at the discretion of the Sponsor or health authority request. In addition, data from P1 cohorts may be locked, unblinded, and analyzed prior to conducting P2 and P3.

11. STUDY ADMINISTRATION

11.1. Regulatory and Ethical Considerations

11.2. Ethical Standard

This study will be conducted in accordance with the protocol and with the following:

- Consensus ethical principles derived from international guidelines including the Declaration of Helsinki and Council for International Organizations of Medical Sciences International Ethical Guidelines.
- Applicable ICH GCP Guidelines.
- United States CRFs applicable to clinical studies (45 CFR Part 46; 21 CFR Part 50, 21 CFR Part 56, and 21 CFR Part 312).
- Applicable laws and regulations.

11.2.1. Ethics Committee Approval

The PI (or designee) must ensure that all required study-specific documents and/or information are submitted to the EC for review and approval as appropriate including but not limited to:

- the protocol and any future protocol amendments
- ICF and any other documents (electronic or paper) given to the subject
- IB

Any substantial protocol amendments, likely to affect the safety of the subjects or the conduct of the study, will require EC and regulatory authority (as locally required) approval before implementation of changes made to the study design, except for changes necessary to eliminate an immediate hazard to study subjects or any nonsubstantial changes, as defined by regulatory requirements.

11.2.2. Informed Consent

Informed consent is a process that is initiated prior to the individual's agreeing to participate in the study and continuing throughout the individual's study participation. Extensive discussion of risks and possible benefits of this therapy will be provided to the subjects and, if applicable, their families. Consent forms describing in detail the study interventions/products, study procedures, and risks are given to the subject and written documentation of informed consent is required prior to starting any screening procedures or intervention/administering study product. Consent forms will be EC-approved and the subject will be asked to read and review the document.

Upon reviewing the document, the PI (or designee) will explain the research study to the subject and answer any questions that may arise. The subjects will sign the ICF prior to any procedures being done specifically for the study. The subjects should have the opportunity to discuss the study with their surrogates or think about it prior to agreeing to participate. The subjects may withdraw consent at any time throughout the course of the trial. A copy of the ICF will be given to the subjects for their records. The rights and welfare of the subjects will be protected by emphasizing to them that the quality of their medical care will not be adversely affected if they decline to participate in this study.

11.3. Financing and Insurance

Financing and insurance will be addressed in a separate agreement.

11.4. Source Documentation and Access

The site will maintain appropriate medical and research records in compliance with ICH E6, Section 4.9 and regulatory and institutional requirements for the protection of confidentiality of subjects. The site will permit authorized representatives of the Sponsor, its designees, and appropriate regulatory agencies to examine (and when required by applicable law, to copy) clinical records for the purposes of quality assurance reviews, audits, and evaluation of the study safety and progress. These representatives will be permitted access to all source data and source documents, which include, but are not limited to, clinical and office charts, laboratory notes, memoranda, subjects' memory aid or evaluation checklists, pharmacy dispensing records, recorded data from automated instruments, copies or transcriptions certified after verification as being accurate and complete, photographic negatives, microfilm or magnetic media, x-rays, and subject files and records kept at the pharmacy, at the laboratory, and medico-technical departments involved in the clinical trial.

11.5. Data Collection and Record Keeping

11.5.1. Data Collection

Data collection and data entry are the responsibility of the clinical trial staff at the site. The PI is responsible for ensuring the accuracy, completeness, legibility, and timeliness of the data reported. All source documents should be completed in a neat, legible manner to ensure accurate interpretation of data.

A CRF must be completed for every subject who signs the ICF and has at least one protocol-specified assessment conducted. The CRF must be completed and processed according to the CRF guidelines and the SOPs of the site. All data should be entered into the CRF, where possible, within 3 days after each visit for any one subject. After the subject has completed the study, the PI must review and sign the signature page of the CRF indicating that he has reviewed the completed CRF and pertinent clinical data for that subject and that, to the best of his knowledge, all data recorded in the CRF accurately reflects the subject's clinical performance in the study

11.5.2. Study Records Retention

Study documents should be retained for a minimum of 5 years after the end of the study. These documents should be retained for a longer period; however, if required by local regulations. No records will be destroyed without the written consent of the Sponsor, if applicable. It is the responsibility of the Sponsor to inform the PI when these documents no longer need to be retained.

11.5.3. Protocol Deviations

A protocol deviation is any noncompliance with the protocol or study procedures detailed in the laboratory or pharmacy manuals. The noncompliance may have been the result of action by the PI, site staff, or subject. All deviations should be handled in accordance with site SOPs.

It is the responsibility of the site to use continuous vigilance to identify and report deviations within 5 working days of identification of the protocol deviation, or within 5 working days of the scheduled protocol-required activity.

11.6. Clinical Monitoring

Site monitoring is conducted to ensure that the human subjects' protections, study and laboratory procedures, study intervention administration, and data collection processes are of high quality and meet Sponsor, ICH/GCP guidelines and applicable regulations, and that this trial is conducted in accordance with the protocol, protocol-specific MOP and applicable Sponsor SOPs. Experienced clinical monitors of the Sponsor or its designee will conduct site-monitoring visits as detailed in the clinical monitoring plan.

Site visits will be made at standard intervals as defined by the Sponsor and may be made more frequently as directed by the Sponsor. Monitoring visits will include, but are not limited to, review of regulatory files, accountability records, CRFs, ICFs, medical and laboratory reports, and protocol and GCP compliance. Clinical monitors will have access to each participating site, study personnel, and all study documentation according to the site monitoring plan. Clinical monitors will meet with the site PI to discuss any problems and actions to be taken and will document site visit findings and discussions. As data are entered into a CRF, data will be monitored to ensure accuracy as well as adherence to data entry instructions provided in the CRF guidelines.

11.7. Quality Control and Quality Assurance

A quality management plan will be put in place for this study. The site is responsible for conducting routine quality assurance and quality control activities to internally monitor study progress and protocol compliance. The PI will provide direct access to all study-related sites, source data/data collection forms, and reports for the purpose of monitoring and auditing by the Sponsor, and inspection by local and regulatory authorities. The PI will ensure all study personnel are appropriately trained and applicable documentations are maintained on site.

11.8. Study Termination and Closure Procedures

11.8.1. Study Termination

This study may be temporarily suspended or prematurely terminated if there is sufficient reasonable cause. Written notification, documenting the reason for study suspension or termination, will be provided to the site. If the study is terminated or suspended, the PI (or designee) will inform study participants and the EC. The Sponsor will notify appropriate regulatory authorities. The Sponsor will provide the reason(s) for the termination or suspension. Study participants will be contacted, as applicable, and be informed of changes to study visit schedule.

If suspended, the study may resume once issues that caused suspension of the study are resolved.

11.8.2. Termination Procedures

If the study is prematurely terminated, then the site must return all appropriate study data, resolve all data queries, complete final drug accountability, return any study drug remaining on site, and ship all biological samples (including PK and PBMCs) to the laboratory designated by the Sponsor. The PI (or designee) must notify the EC of study termination.

11.9. Information Disclosure

11.9.1. Confidentiality

Subject confidentiality and privacy is strictly held in trust by the PI, site staff, and the Sponsor(s) and their agents. The study protocol, documentation, data, and all other information generated will be held in strict confidence. No information concerning the study or the data will be released to any unauthorized third party without prior written approval of the Sponsor.

The study monitor or other authorized representatives of the Sponsor or regulatory agencies may inspect all documents and records required to be maintained by the PI, including but not limited to, medical records (office, clinic, or hospital) and pharmacy records for the participants in this study. The clinical study site will permit access to such records.

The study participant's contact information will be securely stored at the clinical site for internal use during the study. At the end of the study, all records will continue to be kept in a secure location for as long a period as dictated by the Sponsor, site, or regulatory requirements.

Study participant research data will be transmitted to and stored securely by the Sponsor's designated data center. This will not include the participant's contact or identifying information. Individual participants and their research data will be identified by a unique study identification number. The study data entry and study management systems used by clinical sites will be secured and password-protected. At the end of the study, all study databases will be de-identified and archived at a secure location.

11.9.2. Clinicaltrials.gov

This clinical study will be registered on clinicaltrials.gov as required.

11.9.3. Publication Policy

All information generated from this study is the proprietary property of the Sponsor. It is the intent of the Sponsor to publish the results of the study in their entirety as deemed appropriate.

12. REFERENCES

Agostini ML, Pruijssers AJ, Chappell JD, Gribble J, Lu X, Andres EL, et al. Small-Molecule Antiviral β-d-N4-Hydroxycytidine Inhibits a Proofreading-Intact Coronavirus with a High Genetic Barrier to Resistance. J Virol. 2019 Dec 15;93(24).

Flavell RA, Sabo DL, Bandle EF, Weissmann C. Site-directed mutagenesis: generation of an extracistronic mutation in bacteriophage Q beta RNA. J Mol Biol. 1974 Oct 25;89(2):255–72.

Hernandez-Santiago BI, Beltran T, Stuyver L, Chu CK, Schinazi RF. Metabolism of the anti-hepatitis C virus nucleoside beta-D-N4-hydroxycytidine in different liver cells. Antimicrob Agents Chemother. 2004 Dec;48(12):4636–42.

Mullins JI, Heath L, Hughes JP, Li D, Essigmann JM, Harris KS, et al. Mutations based on viral decay acceleration in the HIV-1 genomes of a clinical population treated with the mutagenic nucleoside KP1461. Retrovirology. 2011 Oct 3;8(Suppl 2):P84.

Painter GR, Bowen RA, Bluemling GR, DeBergh J, Edpuganti V, Gruddanti PR, et al. The prophylactic and therapeutic activity of a broadly active ribonucleoside analog in a murine model of intranasal venezuelan equine encephalitis virus infection. Antiviral Res. 2019;171:104597.

Sticher ZM, Lu G, Mitchell DG, Marlow J, Moellering L, Bluemling GR, et al. Analysis of the Potential for N4-Hydroxycytidine To Inhibit Mitochondrial Replication and Function. Antimicrob Agents Chemother. 2020 27;64(2).

Stuyver LJ, Whitaker T, McBrayer TR, Hernandez-Santiago BI, Lostia S, Tharnish PM, et al. Ribonucleoside analogue that blocks replication of bovine viral diarrhea and hepatitis C viruses in culture. Antimicrob Agents Chemother. 2003 Jan;47(1):244–54.

Toots M, Yoon J-J, Cox RM, Hart M, Sticher ZM, Makhsous N, et al. Characterization of orally efficacious influenza drug with high resistance barrier in ferrets and human airway epithelia. Sci Transl Med. 2019 Oct 23;11(515):eaax5866.

Urakova N, Kuznetsova V, Crossman DK, Sokratian A, Guthrie DB, Kolykhalov AA, et al. β-d-N4-Hydroxycytidine Is a Potent Anti-alphavirus Compound That Induces a High Level of Mutations in the Viral Genome. J Virol. 2018 01;92(3).

Yoon J-J, Toots M, Lee S, Lee M-E, Ludeke B, Luczo JM, et al. Orally Efficacious Broad-Spectrum Ribonucleoside Analog Inhibitor of Influenza and Respiratory Syncytial Viruses. Antimicrob Agents Chemother. 2018;62(8).

13. APPENDICES

Appendix 1: Time and Events Schedule

Parts 1 and 2

Visit	Screening (Days -28 to -2)	Day -1	Day 1	Day 2 (24 hr)	Day 3 (48 hr)	Day 4 (72 hr)	Day 9	Day 15 / EOS[1] (non-FE cohorts)
Food-Effect Cohort		**Day 14**	**Day 15**	**Day 16 (24 hr)**	**Day 17 (48 hr)**	**Day 18 (72 hr)**	**Day 23**	**Day 30 / EOS**
ICF; Demography	x							
I/E; Medical history	x	x						
Physical examination[2]	x	x		x		x	x	x
Qualifying laboratory analyses[3]	x	x						
Drug screening and pregnancy test[4]	x	x					x	x
Height, weight (BMI)	x							
Clinic confinement[5]		x	x	x	x	x		
Non-residential visit	x						x	x
Clinical chemistry and urinalysis[6]	x	x		x		x	x	x
Hematology[7]	x	x		x	x	x	x	x
PBMC collection (FE cohort ONLY)			x[8]	x[8]				
ECG	x		x[9]			x		x
Continuous 12-lead ECG (Part 1 only)			x[16]	x[16]				
Vital signs	x	x	x[10]	x	x	x	x	x
Administer study drug			x					
Plasma PK sample collection			x[11]	x[12]	x[12]	x[12]		
Urine PK sample collection			x[13]	x[14]	x[14]			
Adverse events	x[15]	x[15]	x[15]	x	x	x	x	x
Prior and/or Concomitant medications	x	x	x	x	x	x	x	x

Abbreviations: BMI (body mass index); EOS (End of Study); FE (food-effect); ICF (Informed Consent Form); I/E (Inclusion/Exclusion Criteria); ECG (electrocardiogram); PBMC (peripheral blood mononuclear cell); PK (pharmacokinetic).

[1] On Day 15, conduct the EOS visit for all subjects except those in the FE cohort.

[2] A complete PE will be performed at Screening and/or Check-in, and EOS visits; on all other days, a limited PE will be conducted targeting any change from the previous status or any body system deemed pertinent to that subject by the PI (or designee).

[3] Laboratory analyses to qualify for study entry include: total white cell count or absolute lymphocyte count (values must be in the normal range), hemoglobin or platelet levels (values must not be below the lower limit of normal), and ALT/SGPT, alkaline phosphatase, AST/SGOT (values must not be above the upper limit of normal), plus virology (HIV/HCV/HBV; tested at Screening only; all must be negative), and FSH (if required).

[4] Drug screening (urine drug screen, UDS), including a cotinine test, should be conducted at Screening, Day -1, Days 9, 14, and 23, and at EOS. Pregnancy tests should be conducted in serum at Screening and in urine at all other times (i.e., Days -1, 9, 14, and 23, and at EOS). A positive urine pregnancy test will be confirmed with a serum pregnancy test. An alcohol breath test should be conducted at Screening and Days -1 and 14.

[5] Participants should be admitted to the clinic following review of final I/E criteria on Day -1 and discharged from the clinic following completion of study specific assessments on Day 4. Subjects in the FE cohort should be readmitted to the clinic on Study Day 14.

[6] Safety laboratory assessments include clinical chemistry and urinalysis. Baseline samples will be collected after clinic admission on Day -1.

[7] Hematology values include, but are not limited to, RBC, WBC with differential and platelets. PT/PTT and INR assessed only at Screening.

[8] PBMCs will be collected predose, and 2, and 8 hrs postdose on Days 1 and 15. Depending on ongoing review of the data, timepoints may be modified or removed, and up to 2 additional samples may be collected.

[9] The baseline ECG should be conducted prior to dosing on Day 1/15.

[10] Vital signs include pulse rate, respiratory rate, blood pressure, and body temperature; on Day 1/15, VS should be taken predose, and at 2, 4, 8 and 12 hr postdose.

[11] PK samples should be collected predose and at 0.25, 0.5, 1, 1.5, 2, 2.5, 3, 4, 6, 9, 12, and 15 hr postdose on Day 1/15. Depending on ongoing review of the PK data, timepoints may be modified or removed, and up to 3 additional samples (in each period for the food-effect) may be added.

[12] PK samples should be collected at 24, 36, 48, and 72 hr postdose on Days 2/16, 3/17, 4/18 respectively. Depending on ongoing review of the PK data, timepoints may be modified or removed, and up to 3 additional samples (in each period for the food-effect) may be added.

[13] Urine samples for PK analysis should be collected predose (spot collection), and from 0 to 4, 4 to 8, 8 to 12, and 12 to 24 hr postdose.

[14] Urine samples for PK analysis should be collected from 24 to 36 and 36 to 48 hr postdose.

[15] Adverse events occurring prior to study drug administration should be recorded as medical history unless the event is directly related to a study specified procedure.

[16] Monitor for 12-lead ECG recording will be worn from approximately 2 hours prior to dosing (0 hr) to 25 hrs (Day 2) postdose. Extraction timepoints will be at 60, 45, and 30 minutes prior to dosing and 0.25, 0.5, 1, 1.5, 2, 2.5, 3, 4, 6, 9, 12, 15 and 24 hrs postdose.

Part 3

Visit	Screening (Days -28 to -2)	Day -1	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7	Day 8	Day 9	Day 14	Day 20 /EOS
ICF; Demography	x												
I/E; Medical history	x	x											
Physical examination[1]	x	x		x					x			x	x
Qualifying laboratory analyses[2]	x	x											
Drug screening and pregnancy test[3]	x	x										x	x
Height, weight (BMI)	x												
Clinic confinement[4]		x	x	x	x	x	x	x	x	x	x		
Non-residential visit	x											x	x
Clinical chemistry and urinalysis[5]	x	x			x			x			x		x
Hematology[6]	x	x			x			x			x	x	x
PBMC collection[7]			x					x					
ECG[8]	x		x					x			x		x
Continuous 12-lead ECG			x[9]					x[10]	x[10]				
Vital signs[11]	x	x	x	x	x	x	x	x	x	x	x	x	x
Administer study drug[12]			x	x	x	x	x	x					
Plasma PK sample collection[13]			x			x		x	x	x	x	x	
Urine PK sample collection[14]			x					x	x	x	x		
Adverse events	x[15]	x[15]	x[15]	x	x	x	x	x	x	x	x	x	x
Prior and/or Concomitant medications	x	x	x	x	x	x	x	x	x	x	x	x	x

Abbreviations: BID (twice daily); BMI (body mass index); EOS (End of Study); ICF (Informed Consent Form); I/E (Inclusion/Exclusion Criteria); ECG (electrocardiogram); PK (pharmacokinetic).

[1] A complete PE will be performed at Screening and/or Check-in, and EOS visits; on all other days, a limited PE will be conducted targeting any change from the previous status or any body system deemed pertinent to that subject by the PI (or designee).

[2] Laboratory analyses to qualify for study entry include: total white cell count or absolute lymphocyte count (values must be in the normal range), hemoglobin or platelet levels (values must not be below the lower limit of normal), and ALT/SGPT, alkaline phosphatase, AST/SGOT (values must not be above the upper limit of normal), plus virology (HIV/HCV/HBV; tested at Screening only; all must be negative), and FSH (if required).

[3] Drug screening (urine drug screen, UDS), including cotinine, should be conducted at Screening, Day -1, Day 14, and at EOS. Pregnancy tests should be conducted in serum at Screening and in urine at all other timepoints. A positive urine pregnancy test will be confirmed with a serum pregnancy test. An alcohol breath test should be conducted at Screening and Day -1.

[4] Participants should be admitted to the clinic following review of final I/E criteria on Day -1 and discharged from the clinic following completion of study specific assessments on Day 9.

[5] Safety laboratory assessments include clinical chemistry and urinalysis. Baseline samples will be collected after clinic admission on Day -1. On Days 3 and 6, the assessment will be pre-am dose. On Day 9, the assessment will be 72 hrs post-am dose on Day 6.

[6] Hematology values include, but are not limited to, RBC, WBC with differential and platelets. PT/PTT and INR assessed only at Screening.

[7] PBMCs will be collected 2 hrs postdose, relative to the first daily dose on Day 1, and pre-am dose and 2 and 8 hrs postdose relative to the first daily dose on Day 6. Depending on ongoing review of the data, timepoints may be modified or removed, and up to 2 additional samples may be collected. Additionally, PMBC collection may be omitted from some cohorts.

[8] The baseline ECG should be conducted prior to first dosing on Day 1. On Day 1, ECG should be taken predose and 2 hours post-am dose. On Day 6, ECG should be taken predose, 2 hrs, and 72 hrs (Day 9) postdose.

[9] On Day 1, monitor for 12-lead ECG recording will be worn from 2 hrs prior to the first of the daily doses to 12 hrs following the first of the daily doses. Extraction timepoints will be 60, 45, and 30 minutes prior to the first of the daily doses and at 0.25, 0.5, 1, 1.5, 2, 2.5, 3, 4, 6, 9, and 12 hrs following the first of the daily doses.

[10] On Day 6, monitor for 12-lead ECG recording will be worn from 2 hours prior to dosing (0 hr) to 25 hrs (Day 7) postdose. Extraction timepoints will be at 0.25, 0.5, 1, 1.5, 2, 2.5, 3, 4, 6, 9, 12, and 24 hrs postdose.

[11] Vital signs include pulse rate, respiratory rate, blood pressure, and body temperature. On Day 1, VS should be taken pre-am dose, and at 2, 4, and 8 hrs post-am dose. On Day 2 through 5, VS should be taken pre-am dose. On Day 6, VS should be taken predose, and 2, 4, 8, 24 (Day 7), 48 (Day 8), and 72 (Day 9) hrs postdose.

[12] Study drug should be administered BID, 12 hrs apart on Days 1 through 5. On Day 6, study drug should be administered once in the morning only (0 hr). Depending on ongoing review of the safety, tolerability, and PK data, the number of days of dosing may be reduced and the dosing frequency changed; the dosing frequency will be no less than once-daily or no greater than three-times daily. If the number of days of dosing are reduced, the timepoints for subsequent assessments and the period for which subjects are domiciled at the clinic will similarly be changed (subjects will be domiciled for 3 days post final dose).

[13] On Day 1, PK samples should be collected pre-am dose and at 0.25, 0.5, 1, 1.5, 2, 2.5, 3, 4, 6, 9, and 12 hrs post-am dose. On Day 4, the PK sample should be collected pre-am dose. On Day 6, PK samples should be collected predose and at 0.25, 0.5, 1, 1.5, 2, 2.5, 3, 4, 6, 9, 12, 15, 24 (Day 7), 48 (Day 8), 72 (Day 9), and 192 (Day 14) hrs postdose. Depending on ongoing review of the PK data, timepoints may be modified or removed, and up to 3 additional samples may be added.

[14] On Day 1, urine samples for PK analysis should be collected predose (spot collection), and from 0 to 4, 4 to 8, and 8 to 12 hrs postdose. On Day 6, urine samples for PK analysis should be collected predose (spot collection), and from 0 to 4, 4 to 8, 8 to 12, 12 to 24 (Day 7), 24 to 48 (Day 8), and 48 to 72 hrs (Day 9). Sampling timepoints may be modified, removed, or additional timepoints added depending upon ongoing review of the data. The 12-hour urine collection should occur prior to the second daily dose administration.

[15] Adverse events occurring prior to study drug administration should be recorded as medical history unless the event is directly related to a study specified procedure.

Appendix 2: DMID Toxicity Grading Scale

Clinical Adverse Events			
VITAL SIGNS	**Mild (Grade 1)**	**Moderate Grade 2)**	**Severe (Grade 3)**
Fever (°C) Oral temperature; no recent hot or cold beverages or smoking.	38.0 – 38.4	38.5 – 38.9	>39.0
Tachycardia - beats per minute	101 – 115	116 – 130	> 130 or ventricular dysrhythmias
Bradycardia - beats per minute	50 - 54 or 45 - 50 bpm if baseline <60 bpm	45 - 49 or 40 - 44 if baseline <60 bpm	< 45 or <40 bpm if baseline <60 bpm
Hypertension (systolic) - mm Hg [Assuming supine position, 10 min at rest conditions, not sleeping subjects, measurements on the same arm and several concordant results.]	141 - 150	151 - 160	> 160
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**
Arrythmia		asymptomatic, transient signs, no Rx required	recurrent/persistent; symptomatic Rx required
Hemorrhage, Blood Loss	Estimated blood loss ≤100 mL	Estimated blood loss > 100 mL, no transfusion required	Transfusion required
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 60 - 70% (of peak flow)	no normalization with bronchodilator; FEV1 <60% 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 gms/24 hours	4 - 5 loose or watery stools or 400 - 800 gms/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
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
SYSTEMIC	**Mild (Grade 1)**	**Moderate (Grade 2)**	**Severe (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
All Other conditions	**Mild (Grade 1)**	**Moderate (Grade 2)**	**Severe (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

*In addition to grading the measured local reaction at the greatest single diameter, the measurement should be recorded as a continuous variable.

**Induration/Swelling should be evaluated and graded using the functional scale as well as the actual measurement

Laboratory Adverse Events			
Blood, Serum, or Plasma *	**Mild (Grade 1)**	**Moderate (Grade 2)**	**Severe (Grade 3)**
Sodium – Hyponatremia mEq/L	132 – <LLN	130 – 131	<130
Sodium – Hypernatremia mEq/L	>ULN – 148	149 – 150	>150
Potassium – Hyperkalemia mEq/L	>ULN – 5.2	5.3 – 5.4	>5.4
Potassium – Hypokalemia mEq/L	<LLN-3.1	<3.1 – 3.0	<3.0
Glucose – Hypoglycemia mg/dL	65 – 67	55 – 64	<55
Glucose – Hyperglycemia Fasting – mg/dL	>ULN - 120	121 – 130	>130
Glucose – Hyperglycemia Random – mg/dL	140 – 159	160 – 200	>200
Blood Urea Nitrogen mg/dL	23-26	27 – 31	> 31
Creatinine – mg/dL	>ULN – 1.7	1.8 – 2.0	>2.0
Calcium – hypocalcemia mg/dL	8.0 – <LLN	7.5 – 7.9	<7.5
Calcium – hypercalcemia mg/dL	>ULN – 11.0	11.1 – 11.5	>11.5
Magnesium – hypomagnesemia mg/dL	1.3 – 1.5	1.1 – 1.2	<1.1
Phosphorus – hypophosphatemia mg/dL	2.3 – 2.5	2.0 – 2.2	<2.0
CPK – mg/dL	400-1000	1001-1500	>1500
Albumin – Hypoalbuminemia g/dL	2.8 – 3.0	2.5 – 2.7	< 2.5
Total Protein – Hypoproteinemia g/dL	5.2 – 5.4	4.8 – 5.1	< 4.8
Alkaline phosphate – U/L	132-240	241– 360	>360
AST U/L	44 – 105	106-175	>175
ALT U/L	44 – 105	106-175	>175
Bilirubin (serum total) mg/dL	1.3 - 2.0	2.1 - 2.5	> 2.5
Bilirubin – when ALT ≥105 (Hy's law)	1.3 -1.5	1.6 – 2.0	> 2.0
Amylase- U/L	200-270	271-360	>360
Lipase- U/L	176-270	271-360	>360
Hemoglobin (Female) - g/dL	11.0 – 11.5	9.5 – 10.9	< 9.5
Hemoglobin (Male) - g/dL	12.0 – 12.5	10.0 – 11.9	<10.0
WBC Increase - cell/mm3	11,001 – 15,000	15,001 – 20,000	> 20,000
WBC Decrease - cell/mm3	2,500 – 3,500	1,500 – 2,499	< 1500
Lymphocytes Decrease - cell/mm3	750 – 1,000	500 – 749	< 500
Neutrophils Decrease - cell/mm3	1,500 – 2,000	1,000 – 1,499	< 1000
Eosinophils - cell/mm3	500-750	751-1500	> 1500
Platelets Decreased - cell/mm3	120,000 – 130,000	100,000 – 119,999	<100,000
PT – seconds (prothrombin time)	> ULN-14.4	14.5 – 15.7	> 15.7
PTT – seconds (partial thromboplastin time)	>ULN-42.1	42.2– 50.0	> 50.0
Fibrinogen increase - mg/dL	>ULN – 500	501 – 600	> 600
Fibrinogen decrease - mg/dL	<LLN – 140	125 – 139	<125
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	11-50	> 50 and/or gross blood

* The laboratory values provided in the tables serve as guidelines and are dependent upon institutional normal parameters.

* Institutional normal reference ranges should be provided to demonstrate that they are appropriate.

Appendix 3: Contraception Guidance

Male subjects (regardless of fertility status) with partners of childbearing potential must use a male barrier method of contraception (i.e., male condom with spermicide) in addition to a second method of acceptable contraception from Check-in until 90 days after the EOS visit. Acceptable methods of contraception for female partners include:

- hormonal injection
- combined oral contraceptive pill or progestin/progestogen-only pill
- combined hormonal patch
- combined hormonal vaginal ring
- surgical method (bilateral tubal ligation or Essure® [hysteroscopic bilateral tubal occlusion])
- hormonal implant
- hormonal or non-hormonal intrauterine device
- OTC sponge with spermicide
- cervical cap with spermicide
- diaphragm with spermicide.

An acceptable second method of contraception for male subjects is vasectomy that has been performed at least 90 days prior to the screening visit, with verbal confirmation of surgical success.

For male subjects (even with a history of vasectomy), sexual intercourse with female partners who are pregnant or breastfeeding should be avoided unless condoms are used from the time of the first dose until 90 days after the EOS visit. Male subjects are required to refrain from donation of sperm from Check-in until 90 days after the EOS visit.

Sexual Abstinence and Same-sex Relationships

Subjects who practice true abstinence, because of the subject's lifestyle choice (i.e., the subject should not become abstinent just for the purpose of study participation), are exempt from contraceptive requirements. Periodic abstinence (eg, calendar, ovulation, symptothermal, postovulation methods) and withdrawal are not acceptable methods of contraception. If a subject who is abstinent at the time of signing the ICF becomes sexually active they must agree to use contraception as described previously.

For subjects who are exclusively in same-sex relationships, contraceptive requirements do not apply. If a subject who is in a same-sex relationship at the time of signing the ICF becomes engaged in a heterosexual relationship, they must agree to use contraception as described previously.