A PHASE 1A, DOUBLE-BLIND, PLACEBO-CONTROLLED, SINGLE ASCENDING DOSE STUDY TO DETERMINE THE SAFETY AND PHARMACOKINETICS OF AV-1 IN HEALTHY MALE AND FEMALE ADULT SUBJECTS

Sponsor Protocol Number:	AV1-PPD-0005
DMID Protocol Number:	18-0016
NIAID Contract Number:	HHSN272201600028C
IND Sponsor:	AbViro LLC [redacted]
Principal Investigator:	[redacted]
Sponsor Representative:	[redacted]
Version Number:	5.0
Protocol Date:	06 November 2019
Protocol Amendment 1 Date:	25 March 2020
Protocol Amendment 2 Date:	07 May 2020
Protocol Amendment 3 Date:	25 February 2021
Protocol Amendment 4 Date:	23 April 2021

STATEMENT OF ASSURANCE

Each institution will hold a current Federal Wide Assurance (FWA) issued by the Office of Human Research Protections (OHRP) for federally funded human subjects research. Each FWA will designate at least one institutional review board (IRB) registered with OHRP, for which the research will be reviewed and approved by the IRB and will be subject to continuing review (45 CFR 46.103[b]). The IRB designated under an FWA may include an institution's IRB, an independent IRB, or an IRB of another institution after establishing a written agreement with that other institution.

STATEMENT OF COMPLIANCE

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

- United States (US) Code of Federal Regulations (CFR) 45 CFR Part 46: Protection of Human Subjects
- Food and Drug Administration (FDA) Regulations, as applicable: 21 CFR Part 50 (Protection of Human Subjects), 21 CFR Part 54 (Financial Disclosure by Clinical Investigators), 21 CFR Part 56 (IRBs), 21 CFR Part 11, and 21 CFR Part 312 (Investigational New Drug Application)
- International Council for Harmonisation (ICH) E6 (R2) GCP: Integrated Addendum to ICH E6 (R1) Guidance for Industry, published in the Federal Register (83 Federal Register 8882 [2018])
- Belmont Report: Ethical Principles and Guidelines for the Protection of Human Subjects of Research, Report of the National Commission for the Protection of Human Subjects of Biomedical and Behavioral Research
- National Institutes of Health (NIH) Office of Extramural Research, Research Involving Human Subjects, as applicable
- National Institute of Allergy and Infectious Diseases Clinical Terms of Award, as applicable
- Applicable federal, state, and local regulations and guidance

SIGNATURE PAGE

The signature below provides the necessary assurance 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 US federal regulations and current ICH E6 GCP guidelines.

I agree to conduct the study in compliance with GCP and applicable regulatory requirements.

I agree to conduct the study in accordance with the current protocol and will not make changes to the protocol without obtaining the Sponsor's approval and IRB approval, except when necessary to protect the safety, rights, or welfare of subjects.

Investigator Signature:

Sign/Date: [redacted]

[redacted]
Principal Investigator

Sponsor Signature:

Sign/Date: [redacted]

[redacted]
Sponsor Representative

PROTOCOL AMENDMENT SUMMARY OF CHANGES

Date of Amendment 1: 25 March 2020 (Version 2.0)

The reasons for this protocol amendment are summarized below:

- DMID Medical Monitor name, address, phone number, and email address were updated in Section 1.
- Modified the screening period from 28 days to 35 days in the protocol summary, Section 3.1, Section 5.1.2, and schedule of events (Section 17.1 Appendix A).
- A pharmacokinetic blood draw was added on Day 43 (±3) and Day 57 (±3) throughout the protocol.
- To clarify the duration of nonclinical studies in mice from the survival perspective, the following text "for the duration of the study (10 days)" was added in Section 2.2.
- Text was added to clarify subject retesting of laboratory parameters that are outside the acceptable range in Section 5.1.
- Clarified inclusion criterion 3.b.II "A pre-existing medical condition that is not exclusionary but has worsened in terms of clinical outcome within 3 months of enrollment" in Section 5.1.1.
- Removed "will" and "unless otherwise specified" and replaced with "may" in Section 5.2.2.
- Respiratory rate was added as part of vital sign measurements in Section 7.1.1 and footnote "j" in the schedule of events (Section 17.1 Appendix A).
- Added "alkaline phosphatase, aspartate aminotransferase" to Section 7.2.1 to match the safety laboratory tests that will be performed.
- Text was added to clarify when dosing escalation should be halted in Section 8.6.1.
- Clarified when the infusion should be halted and adjusted language to allow reductions in infusion rate for symptom management in Section 8.6.2.

- Clarified that the physical examination performed at Check-in (Day –1) will be a focused examination in Section 7.1.4 and footnote "i" in the schedule of events (Section 17.1 Appendix A).

- Updated "immunogenicity" to "anti-AV-1 antibody" in Section 3.1, Section 7.2.3, and the schedule of events (Section 17.1 Appendix A).

- Removed "Water is permitted as desired except for the period of 1 hour prior to the start of the IP infusion until 1 hour after the end of the infusion." in footnote "s" in the schedule of events (Section 17.1 Appendix A). There is no restriction for water prior to and after dosing.

- Removed reference range from the title and removed the reference ranges column for laboratory values in Table 17-3 Laboratory Toxicity Grading.

- Administrative changes were made throughout the protocol.

Details of the changes within the protocol can be found in the tracked-changes document, which accompanies the amended protocol.

Date of Amendment 2: 07 May 2020 (Version 3.0)

The reasons for this protocol amendment are summarized below:

- The approximate study duration was updated from 16 to 22 weeks in the protocol summary and Section 3.1.

- The number of follow-up visits was updated from 7 to 8 in the protocol summary and Sections 2.3.1 and 3.1.

- A Day 120 study visit was added to include safety activities as well as pharmacokinetic and anti-AV-1 antibody sampling throughout the protocol.

- The schedule of events table (Section 17.1 Appendix A) was updated to add the Day 120 visit, to reflect that Day 120 is the end of study (EOS) visit, as well as procedures to be performed in the event of early termination from the study. The abbreviation EOS and definition was added to the schedule of events table abbreviation list.

- Final follow-up visit in footnote "c" in the schedule of events (Section 17.1 Appendix A) was updated from "Day 85 (Week 12)" to "Day 120 (approximately Week 17)".

- AV-1 product stability and planned stability study duration was updated in Section 4.1.2.

- Typographical error corrected in Section 5.1 to change “hepatitis B surface antibody” to “hepatitis B surface antigen”.

- For clarity, “Anti-drug antibody” was changed to “Anti-AV-1 antibody” in the schedule of events table (Section 17.1 Appendix A).

- A urine drug screen test was added to Day 85 and the text was updated in Section 7.2.1 and the schedule of events table (Section 17.1 Appendix A).

Details of the changes within the protocol can be found in the tracked-changes document, which accompanies the amended protocol.

Date of Amendment 3: 25 February 2021 (Version 4.0)

The reasons for this protocol amendment are summarized below:

- The Principal Investigator for this study changed. The Principal Investigator’s name was updated on the title page and the signature page. The Principal Investigator’s name, telephone number, and email address were updated in the key roles in Section 1.

- The lead statistician for this study changed. The lead statistician’s name and email address were updated in the key roles in Section 1.

Date of Amendment 4: 23 April 2021 (Version 5.0)

The reasons for this protocol amendment are summarized below:

- The Principal Investigator for this study changed. The Principal Investigator’s name was updated on the title page and the signature page. The Principal Investigator’s name, telephone number, and email address were updated in the key roles in Section 1.

- Updated exclusion criterion 23 to include language prohibiting receipt of any COVID-19 vaccine within 14 days prior to or after Day 1 in Section 5.1.2.

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LIST OF ABBREVIATIONS

λ_z	apparent terminal elimination rate constant
$\%AUC_{exp}$	percentage of the area extrapolated for calculation of area under the serum concentration-time curve from time 0 extrapolated to infinity
AE	adverse event
$AUC_{0-\infty}$	area under the serum concentration-time curve from time 0 extrapolated to infinity
AUC_{0-48}	area under the serum concentration-time curve from time 0 to 48 hours postdose
AUC	area under the serum concentration-time curve
$AUC_{0-tlast}$	area under the serum concentration-time curve from time 0 to the last quantifiable concentration
BLQ	below the limit of quantification
bpm	beats per minute
CFR	Code of Federal Regulations
CL	total serum clearance
C_{max}	maximum observed serum concentration
C_t	last observed serum drug concentration
CV	coefficient of variation
DENV	dengue virus
DHHS	Department of Health and Human Services
DMID	Division of Microbiology and Infectious Diseases
ECL	electrochemiluminescence
eCRF	electronic case report form
ECG	electrocardiogram
ELISA	enzyme-linked immunosorbent assay
ET	early termination
FDA	Food and Drug Administration
FWA	Federal Wide Assurance
GCP	Good Clinical Practice
GLP	Good Laboratory Practice
ICF	informed consent form
ICH	International Council for Harmonisation
IFN	interferon
IP	investigational product
IRB	Institutional Review Board
IV	intravenous
mAb	monoclonal antibody
NaCl	sodium chloride
NIH	National Institutes of Health
NOAEL	no observed adverse effect level

OHRP	Office for Human Research Protections
PK	pharmacokinetic(s)
QTcF	QTc interval corrected by Fridericia's formula
SAE	serious adverse event
SD	standard deviation
SMC	safety monitoring committee
SRC	safety review committee
$t_{1/2}$	apparent terminal half-life
T_{max}	time to reach maximum observed serum concentration
US	United States
V_z	volume of distribution during the terminal phase

PROTOCOL SUMMARY

Title:	A Phase 1a, Double-Blind, Placebo-Controlled, Single Ascending Dose Study to Determine the Safety and Pharmacokinetics of AV-1 in Healthy Male and Female Adult Subjects
Design of the Study:	Double-blind, randomized, placebo-controlled, sequential-group, single ascending dose study in healthy adult subjects. Five different doses will be tested in sequential cohorts of 8 subjects each (randomly assigned 6:2 to receive a single intravenous dose of AV-1 or placebo)
Study Phase:	1a
Study Population:	40 healthy male and female subjects between 18 and 55 years of age
Number of Sites:	One site as follows: PPD Phase 1 Clinic [redacted]
Description of the Investigational Product:	AV-1 is a fully human anti-flavivirus IgG1κ monoclonal antibody to be administered intravenously in fixed single doses of 30, 90, 250, 500, and 1000 mg.
Study Objectives:	Primary: • To determine the safety of a single intravenous dose of AV-1 in healthy adult subjects. Secondary: • To characterize the pharmacokinetics of AV-1 following a single intravenous dose. • To evaluate if subjects develop anti-AV-1 antibodies following a single intravenous dose of AV-1.

Duration of Individual Subject Participation:	Subject participation will include an up-to-35-day screening period, admission to the clinic for a 5-day period, and 8 follow-up visits through Day 120 (±5).
Estimated Time to Last Subject/Last Study Day:	Approximately 22 weeks

1 KEY ROLES

Principal Investigator:
[redacted]
PPD Phase 1 Clinic
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Email: [redacted]

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2 BACKGROUND AND SCIENTIFIC RATIONALE

2.1 Background

AV-1 is a human monoclonal antibody (mAb) being investigated as a potential therapy for dengue, a mosquito-borne viral disease with extensive global public health impact. Current treatment options for dengue are limited to supportive care. Thus, a safe and effective therapeutic treatment would provide substantial public health benefits.

Dengue virus (DENV) is a flavivirus transmitted by mosquitos of the *Aedes* genus and a major cause of disease in tropical and subtropical areas (Henchal and Putnak 1990). Over 2 billion people are thought to be at risk of DENV infection globally (Brady *et al* 2012) with an estimated 390 million infections annually (95% credible interval 284 to 528 million), resulting in 96 million clinical cases (95% credible interval 67 to 136 million) and 500,000 severe cases (Bhatt *et al* 2013; Guzman *et al* 2010).

There are 4 known DENV serotypes (DENV-1, -2, -3, and -4). Exposure to any one serotype confers lifelong immunity to that serotype, however; subsequent exposure to a different serotype is associated with increased risk of severe disease. Data suggests that antibody-dependent enhancement due to subneutralizing antibodies may contribute to severe disease resulting from exposure to a second serotype (Schmidt 2010; Dejnirattisai *et al* 2010).

While the majority of DENV infections are asymptomatic or result in a mild flu-like illness lasting 4 to 10 days, the disease known as dengue can be severe and life-threatening. Signs and symptoms of dengue disease include fever (≥38.5°C), headache, retro-orbital or ocular pain, muscle and joint pain, macular or petechial rash, bruising, vomiting, abdominal pain, and enlarged liver. Clinical laboratory features typically include thrombocytopenia, leukopenia, and elevated aminotransferases (Simmons *et al* 2012). The condition of some patients rapidly deteriorates after a 3- to 7-day febrile phase around the time of defervescence. During this critical period, which lasts 48 to 72 hours, some patients develop a systemic vascular leakage syndrome with hemoconcentration, hypoproteinemia, pleural effusions, and ascites. Patients with clinically significant vascular leakage may develop persistent vomiting, severe abdominal pain, tender hepatomegaly, serosal effusions, mucosal bleeding, and lethargy or restlessness. These severe dengue cases may develop hemorrhage, hypovolemic shock, respiratory distress, and organ failure, requiring prompt supportive care measures and judicious intravascular resuscitation. Other complications include liver failure, myocarditis, and encephalopathy but these are less frequent (Simmons *et al* 2012).

AV-1 specifically recognizes a conformational epitope in the fusion loop of Domain II of the flavivirus envelope protein. The highly conserved epitope gives AV-1 the potential for broad-spectrum antiviral activity against flaviviruses, including DENV-1, -2, -3, and -4, and limits the emergence of viral resistance. It is thought that AV-1 elicits its antiviral effects by reduction in viral load through inhibition of virus membrane fusion with the host cell, although

mechanism of action studies to confirm this have not yet been performed. AV-1 has leucine to alanine substitutions at amino acid positions 234 and 235 in its Fc component (LALA substitutions), rendering it unable to bind to Fcγ receptors and complement.

AV-1 is manufactured from a stable Chinese hamster ovary master cell bank. The drug product is aseptically filled into vials with closure, resulting in a sterile injectable buffered solution suitable for clinical use. AV-1 will be administered in the Phase 1a clinical study as a single intravenous (IV) dose prepared in United States Pharmacopeia grade sterile 0.9% sodium chloride (NaCl) in water for injection (0.9% NaCl).

The target population for AV-1 is anticipated to include those with suspected or confirmed dengue.

2.2 Scientific Rationale

2.2.1 Purpose of Study

AV-1 has not previously been tested in humans. The primary objective of this study is to determine the safety of AV-1 in healthy male and female adult subjects when administered as a single IV infusion. Additionally, the pharmacokinetics (PK) and the incidence of anti-AV-1 antibody responses will be studied. This first-in-human clinical safety study will provide important information to help guide further development of AV-1 as a potential countermeasure against dengue.

2.2.2 Rationale for Study

There are currently no approved treatments for dengue. AV-1, a fully human mAb, is being developed to address this unmet medical need. The nonclinical studies supporting the potential of AV-1 are briefly summarized. A more detailed summary of the nonclinical studies is available in the investigator's brochure (AbViro, LLC 2019).

AV-1 safety studies were conducted in compliance with Title 21 Code of Federal Regulations (CFR) Part 58 Good Laboratory Practice (GLP) for Nonclinical Laboratory Studies, including a human tissue cross-reactivity study and a 2-week IV dose toxicity and toxicokinetic study in Sprague-Dawley rats with a 1-week interim necropsy and an 8-week recovery. The human tissue cross-reactivity study showed AV-1 does not bind to normal human tissue. No AV-1-related deaths or adverse effects from AV-1 administered as either a single dose or multiple doses were observed in rats up to the highest tested dose of 300 mg/kg/day, which was the determined no-observed-adverse-effect level (NOAEL) dose.

Nonclinical results suggest AV-1 has potential for treatment of dengue. In vitro studies showed AV-1 neutralizes all 4 DENV serotypes (DENV-1, -2, -3, -4), including 2 strains from each serotype prepared from recent clinical cases (2008 to 2016) and support the conclusion that DENV does not develop resistance to AV-1. *In vivo*, AV-1 protected 90 to 100% of AG129 mice (129/Sv, lacking receptors for interferon [IFN]-α, IFN-β, and IFN-γ, while keeping the adaptive

immune system intact) from death for the duration of the study (10 days) when administered at a dose as low as 7.5 mg/kg prior to or 1 day after lethal challenge with DENV. These doses, given prophylactically, also resulted in reduction of virus levels in serum and tissues and reduced tumor necrosis factor alpha levels in serum. AV-1 protected 80% of mice from death for the duration of the study (10 days) when administered 2 days following the virus challenge at a dose of 37.5 mg/kg. Results from both in vitro and in vivo studies indicate AV-1 avoid antibody-dependent enhancement of DENV as a result of the LALA substitutions.

The initial starting dose of 30 mg considers the standard maximum starting dose calculations as described in the FDA Guidance for Industry Estimating the Maximum Safe Starting Dose in Initial Clinical Trials for Therapeutics in Adult Healthy Volunteers (DHHS 2005), which indicates a minimum 10 times safety margin for selection of an initial clinical dose. The proposed initial starting dose provides a >90-fold safety margin when compared to the human equivalent dose NOAEL identified from the nonclinical safety study. The highest planned dose of 1000 mg was selected as it provides an approximately 3-fold safety margin compared with the NOAEL.

Doses will be increased approximately 3-fold for the initial lower doses (30, 90, and 250 mg) and then increased 2-fold at the higher dose levels (500 and 1000 mg). No drug-related toxicity is anticipated based on nonclinical safety data; however, the more conservative dose increases at higher doses will provide greater opportunity to assess and identify safety risks.

The nonclinical data supports AV-1 safety and potential efficacy and justifies the planned Phase 1a clinical study to assess AV-1 safety in a healthy adult population. The first step will be to conduct a Phase 1a safety trial. The study will be a double-blind, randomized, placebo-controlled, sequential-group, single-ascending dose study in healthy adult subjects. Five different doses will be tested in sequential cohorts of 8 subjects each, of which, 6 subjects will receive AV-1 and 2 subjects will receive placebo per cohort.

2.2.3 Study Population

Because this is an exploratory Phase 1a study to assess the safety and PK of a single IV dose of AV-1, and to determine if subjects develop anti-AV-1 antibodies, the most relevant population is healthy subjects as recommended by the FDA (ICH 1997). A combination of factors have been used to select the study population, including consideration of the patient population for AV-1, the estimated blood sample volume, and the stage of development.

Both males and females aged 18 to 55 years will be enrolled in this study to represent the potential patient population who may receive AV-1 in future clinical studies.

2.3 Potential Risks and Benefits

2.3.1 Potential Risks

No clinical trials in humans have been conducted. Nonclinical GLP-compliant safety studies have indicated no safety concerns. However, as this is a first-in-human for AV-1, unexpected adverse events (AEs) are possible in humans.

Potential theoretical risks associated with the clinical evaluation of AV-1 in humans include immune reactions to AV-1 or its excipients or unintended adverse effects related to the specificity of the antibody. Potential AEs may occur immediately following infusion or be delayed and may range from mild to severe.

Consideration to these potential risks was included in the design of both the AV-1 product and the first-in-human clinical study. Subjects will be screened to ensure enrollment of only healthy adult subjects without significant underlying medical issues or history of hypersensitivity.

AV-1 is a fully human mAb that reduces the risk of immunogenicity-related complications. Additionally, subjects will receive only a single dose of AV-1, further limiting risk of immunogenicity more likely to result from repeated exposures. A single dose will also limit exposure and allow reversibility of any treatment-related adverse effects. The initial fixed starting dose of 30 mg allows for a >90-fold safety margin based on nonclinical data based on the FDA guidance (DHHS 2005). Sentinel dosing will be employed for each dose level to further limit the impact of unforeseen drug-related adverse safety outcomes.

Risk of acute reactions will be managed through close monitoring of subjects during and following administration of the investigational product (IP) infusion. Subjects will be admitted and remain in the clinical site beginning the day prior to the IP infusion (Day –1) through discharge on Day 5 to allow for regular monitoring for hypersensitivity and both laboratory or systemic adverse effects of IP dosing. Eight planned outpatient visits will follow subjects for approximately 120 (±5) days after dosing to afford an appropriate amount of time to observe delayed AV-1-related AEs prior to subject completion of the study. Review of cumulative safety data will be performed prior to dose escalations.

2.3.2 Potential Benefits

There are no benefits to the subjects from taking part in this study. This study is necessary to characterize the initial safety of AV-1 prior to additional clinical studies to further evaluate the safety and efficacy of AV-1 for treatment of dengue, an unmet medical need.

2.3.3 Risk-Benefit Ratio

AV-1 is a fully human mAb that lacks human tissue targets and has an unremarkable nonclinical safety profile. The nonclinical profile of AV-1 indicated no safety signals and supports evaluation in humans starting at an initial dose of 30 mg. The clinical study design includes monitoring for potential risks of AV-1. No therapy is currently approved for treatment of DENV

disease, leaving an unmet medical need. Therefore, the overall risk-benefit balance for evaluating the safety, PK, and incidence of anti-AV-1 antibody responses to AV-1 in healthy subjects is considered to be acceptable.

3 STUDY DESIGN, OBJECTIVES, AND ENDPOINTS

3.1 Study Design Description

This is a Phase 1a, first-in-human, single-center, double-blind, randomized, placebo-controlled, sequential-group, single ascending dose study in healthy adult subjects. Five sequential cohorts of 8 subjects each (Cohorts 1 to 5) are planned for this study. The starting dose of AV-1 will be 30 mg and the planned doses for subsequent cohorts are 90, 250, 500, and 1000 mg. All references to the IP within the content of the protocol apply to AV-1 and placebo.

The study will consist of a screening period, check-in, a treatment period, and 8 follow-up visits for each cohort.

Potential subjects will be screened to assess their eligibility to enter the study within 35 days prior to IP administration. Study site staff will obtain written consent per the standard informed consent process outlined in Section 9.2 before conducting protocol-specific screening activities. Eligible subjects will check in to the clinical site on Day –1 (the day before dosing). After reverification of eligibility, subjects will be randomly assigned before dosing on Day 1 to AV-1 or placebo in a 6:2 ratio. In each cohort, subjects will fast overnight (nothing to eat or drink except water) for at least 10 hours before the start of the IP infusion. Subjects will remain fasted for 4 hours after dosing with the IP. Subjects will be randomly assigned to receive AV-1 or placebo as follows:

- Cohort 1: AV-1 30 mg (n = 6) or placebo (n = 2) administered as an IV dose
- Cohort 2: AV-1 90 mg (n = 6) or placebo (n = 2) administered as an IV dose
- Cohort 3: AV-1 250 mg (n = 6) or placebo (n = 2) administered as an IV dose
- Cohort 4: AV-1 500 mg (n = 6) or placebo (n = 2) administered as an IV dose
- Cohort 5: AV-1 1000 mg (n = 6) or placebo (n = 2) administered as an IV dose

Sentinel dosing will be used in each cohort as discussed in Section 8.2.3. In each cohort, the Investigator will review safety data for the sentinel subjects and notify the Sponsor of intent to proceed with dosing the rest of the cohort with a minimum of 72 hours between dosing of the sentinel subjects and dosing the rest of the cohort. Safety data through Day 8 will be reviewed in a blinded fashion by the safety review committee (SRC) (Section 8.7.1) for each dose cohort before escalating to the next dose cohort (Section 8.2.2). Based on the review of safety and PK data, the Sponsor and the Investigator may choose to repeat a dose level, administer a dose less than the previous dose, escalate to a dose lower than the next planned dose, or prolong the duration of the infusion.

On Day 5, subjects will be discharged after all protocol-specified assessments have been completed. Subjects will return for 8 follow-up visits on Days 8 (+2), 15 (±2), 22 (±2), 29 (±2), 43 (±3), 57 (±3), 85 (±5), and 120 (±5). Subjects will be asked to attend an early termination

(ET) visit if they withdraw or are withdrawn from the study prior to the final follow-up visit on Day 120 (±5).

Safety, PK, and anti-AV-1 antibody endpoints will be evaluated in the study. Clinical laboratory evaluations, physical examination findings, vital sign measurements, safety 12-lead electrocardiograms (ECGs), incidence and severity of AEs, and incidence of serious AEs (SAEs) will be monitored to assess safety (Section 7 and Section 8).

Blood samples for PK analysis of AV-1 will be collected predose (within 15 minutes); at 0.5, 1 (end of infusion), 1.25, 1.5, 2, 3, 4, 6, 8, 12, 24, and 48 hours after the start of infusion; Day 5 prior to discharge; and at the follow-up visits on Days 8 (+2), 15 (±2), 22 (±2), 29 (±2), 43 (±3), 57 (±3), 85 (±5), and 120 (±5) (or ET) (Section 7.2.2).

Blood samples for anti-AV-1 antibody analysis will be collected on Day –1 and on Days 29 (±2) 85 (±5), and 120 (±5) (or ET) to evaluate for the presence of pre-existing or treatment-emergent anti-AV-1 antibodies.

A blood sample will be collected at baseline for hypersensitivity testing and additional samples will be collected if a suspected hypersensitivity reaction or anaphylactic response occurs (Section 7.2.4).

Subjects will be confined to the clinical unit from Day –1 until discharge on Day 5. The planned duration for each cohort (screening to final follow-up visit) is approximately 22 weeks.

3.2 Study Objectives

3.2.1 Primary

The primary objective of this study is to determine the safety of a single IV dose of AV-1 in healthy adult subjects.

3.2.2 Secondary

The secondary objectives of the study are the following:

- To characterize the PK of AV-1 following a single IV dose.
- To evaluate if subjects develop anti-AV-1 antibodies following a single IV dose of AV-1.

3.3 Study Endpoints

3.3.1 Primary Endpoint

The primary safety endpoints are clinical laboratory evaluations (hematology, chemistry, urinalysis), physical examination findings, vital sign measurements, safety 12-lead ECG parameters, incidence and severity of AEs, and incidence of SAEs.

3.3.2 Secondary Endpoints

The secondary endpoints of the study are the following:

- Single dose PK including area under the serum concentration-time curve (AUC) from time 0 to infinity ($AUC_{0-\infty}$), AUC from time 0 to 48 hours postdose (AUC_{0-48}), AUC from time 0 to the time of the last quantifiable concentration ($AUC_{0-tlast}$), maximum observed serum concentration (C_{max}), time of the C_{max} (T_{max}), apparent serum terminal elimination half-life ($t_{1/2}$), total serum clearance (CL), and volume of distribution during the terminal phase (V_z) calculated from serum AV-1 levels measured using an electrochemiluminescence (ECL) enzyme-linked immunosorbent assay (ELISA).
- Incidence of anti-AV-1 antibodies as measured by the proportion of subjects with detectable anti-AV-1 antibody signal in serum, including treatment-emergent anti-AV-1 antibodies, using an ECL ELISA.

4 INVESTIGATIONAL PRODUCT

4.1 Investigational Product Description

AV-1

AV-1 is a fully human IgG1$_K$ mAb that targets the conserved fusion loop structure of the virus envelope protein. AV-1's Fc portion has been engineered with leucine to alanine substitution of amino acids at positions 234 and 235 to avoid dengue antibody-dependent enhancement. Clinical doses will be prepared at the study site pharmacy in sterile 0.9% NaCl. AV-1 is being developed for treatment of dengue.

Placebo

The placebo is commercially available sterile 0.9% NaCl.

4.1.1 Formulation, Packaging, and Labeling

The AV-1 drug substance is formulated in 20 mM citrate (pH 5.6), 150 mM NaCl, and 0.025% polysorbate 80 to a nominal concentration of 25 mg/mL of the AV-1 antibody.

AV-1 will be supplied as a sterile, clear, colorless to pale yellow solution filled in a primary glass container with coated stopper and aluminum seal. The lower end of the fill volume range is controlled to ensure at least 4 mL recovery (100 mg total) from each vial during use.

The AV-1 drug product will be labeled according to manufacturer or regulatory specifications and include the statement "Caution: New Drug – Limited by Federal Law to Investigational Use."

4.1.2 Product Storage and Stability

AV-1

AV-1 must be stored in a secure area at 2°C to 8°C (36°F to 46°F). The study site pharmacy has continuous temperature monitoring with alarm notification. Vials should not be shaken.

Current stability data shows AV-1 is stable when stored at 2°C to 8°C (36°F to 46°F) for at least 18 months. Stability testing is ongoing and planned for at least 24 months. Infusion set compatibility and stability testing was performed using the same materials intended for the conduct of the clinical study.

Placebo

Placebo solution will be stored at room temperature within the study site pharmacy.

4.2 Acquisition

The Sponsor will provide the Investigator and clinical unit with adequate quantities of AV-1 and placebo. AV-1 will be stored and shipped from the AbViro-specified distribution center to the clinical site. Once received, AV-1 will be stored in the study site pharmacy and dispensed by the study site pharmacy to the study site staff. Unused product may be destroyed.

4.3 Preparation and Administration of Investigational Product

The IP will be aseptically prepared as per the pharmacy manual by delegated unblinded pharmacy personnel in INTRAVIA bags with pre-primed infusion set with a blunt tip cannula prior to IV infusion.

AV-1 will be diluted to the appropriate dose in sterile 0.9% NaCl. Diluted AV-1 should be clear and colorless.

The IP will be administered on the morning of Day 1 as a single IV infusion at planned nominal doses of 30, 90, 250, 500, and 1000 mg delivering the full product volume in 60 minutes. The exact dose will be determined following a safety data review after each cohort.

Complete instructions for the preparation of the IP are described in the pharmacy manual.

4.4 Accountability Procedures for the Investigational Product

The FDA requires accountability of the IPs. The pharmacist is responsible for ensuring that a current record of IP accountability is maintained, and that the IP is dispensed only at an official clinical site by authorized personnel as required by applicable regulations and guidelines. Records of IP accountability will consist of the date received, date administered, quantity administered, and the subject number to whom the drug was administered.

The pharmacist will be responsible for maintaining accurate records of the shipment and dispensing of the IP. The pharmacy records must be available for inspection by the Division of Microbiology and Infectious Diseases (DMID) monitoring contractors and is subject to inspection by a regulatory agency (eg, the FDA) at any time. An assigned blinded study monitor will review the pharmacy records.

Unused AV-1 vials will be stored upright at 2ºC to 8ºC in the study site pharmacy until clinical trial accountability is completed. At study termination, all unused AV-1 will be disposed of in accordance with the pharmacy manual.

5 SELECTION OF SUBJECTS AND STUDY ENROLLMENT AND WITHDRAWAL

5.1 Eligibility Criteria

Eligible subjects unable to participate in the study due to a scheduling conflict or if the group they are screening for has completed enrollment may be rescreened for participation in a later group.

A subject who has failed screening may also be rescreened for participation in a later group at the discretion of the Investigator and with Sponsor agreement based on the following criteria:

1. In the opinion of the Investigator, the parameter value does not represent a chronic condition that otherwise will preclude the volunteer from enrolling in the study; and

2. In the opinion of the Investigator, the abnormality is not likely to recur.

Tests that do not allow for a rescreen include the following:

- Positive human immunodeficiency virus, hepatitis B surface antigen, or hepatitis C virus antibody
- Positive flavivirus screen
- Positive urine drug screen

In these cases, a new screening number must be assigned for each subject who is rescreened, and a new informed consent form (ICF) must be signed.

5.1.1 Inclusion Criteria

Each volunteer must meet all the following criteria to be enrolled in this study:

1. Is a male or nonpregnant, nonlactating female, of any race, between 18 and 55 years of age, inclusive, at Screening.

2. Has a body mass index between 18.5 and 29.9 kg/m^2, inclusive, at Screening.

3. Is in good health at Screening and reaffirmed on Day –1.

    a. Good health is defined by the absence of a medical condition described in the exclusion criteria and based on screening medical history, physical examination, vital signs, and 12-lead ECG.

    b. If the subject has another current, ongoing chronic medical condition, the condition cannot meet any of the following criteria:

I. First diagnosed within 3 months of enrollment; or

II. A pre-existing medical condition that is not exclusionary but has worsened in terms of clinical outcome within 3 months of enrollment; or

III. Involves need for medication that may pose a risk to the subject's safety or impede assessment of AEs or anti-AV-1 antibody response if they participate in the study.

4. Female subjects must have a negative serum beta-human chorionic gonadotropin pregnancy test at Screening and Check-in on Day –1 and, on admission to the unit, must not be lactating.

5. Female subjects must fulfill one of the following criteria as confirmed at Screening:

   a. *Postmenopausal women must have had ≥12 months of spontaneous amenorrhea with follicle-stimulating hormone concentration consistently ≥40 mIU/mL and must have a negative pregnancy test result at Screening and Check-in on Day –1.*

   b. *Surgically sterile women, defined as those who have had a hysterectomy, bilateral ovariectomy (oophorectomy), or bilateral tubal ligation. Women who are surgically sterile must provide documentation of the procedure by an operative report or relevant medical records, and must have a negative pregnancy test result at Screening and Check-in.*

   c. *Be willing to remain abstinent (not engage in sexual intercourse) from Check-in on Day -1 until the final follow-up visit on Day 120 (±5).*

   d. *Be willing to use one of the following acceptable methods of birth control until the final follow-up visit on Day 120 (±5): intrauterine device with spermicide; a female condom with spermicide; contraceptive sponge with spermicide; an intravaginal system (eg, NuvaRing®); a diaphragm with spermicide; a cervical cap with spermicide; or oral, implantable, transdermal, or injectable hormonal contraceptives beginning 3 months prior to dosing.*

6. Male subjects who are biologically capable of fathering children must agree and commit to use of an adequate form of double-barrier contraception and refrain from sperm donation from Check-in on Day –1 until the final follow-up visit on Day 120 (±5). A male subject is considered capable of fathering children even if his sexual partner is sterile or using contraceptives.

7. Has not traveled outside the US within 60 days prior to Day –1 and agrees to not travel outside the US through the final follow-up visit on Day 120 (±5).

8. Agrees to abide by the study restrictions and is willing to sign an ICF.

5.1.2 Exclusion Criteria

Volunteers meeting any of the following criteria will be excluded from the study:

1. Has any significant medical condition that, in the opinion of the Investigator or Sponsor, would interfere with the subject's ability to participate in the study or increase the risk of participating for that subject based on the investigator's brochure and the safety profile of AV-1.

    a. *Significant medical condition includes, but is not limited to, kidney disease with creatinine clearance <90 mL/min/1.73 cm^2 (CKD-EPI method), known active liver disease (including steatosis), ischemic heart disease, cardiac conduction disorder, chronic intestinal disease, hypertension (including treated), arrythmia requiring treatment, diabetes requiring insulin, neuropathy, myopathy, and malignancy (not including squamous cell skin cancer, basal cell skin cancer, or cervical low-grade squamous intraepithelial lesions).*

    b. *Congenital nonhemolytic hyperbilirubinemia syndromes (eg, Gilbert's syndrome) are not acceptable.*

2. Has one or more symptoms of a urinary tract infection (eg, dysuria, frequent, urgency, or suprapubic pain) at Screening or Check-in on Day –1.

3. Has a 12-lead ECG with:

    a. *Any clinically important abnormalities in rhythm, conduction, or morphology of the resting ECG and any clinically important abnormalities in the 12-lead ECG that, in the Investigator's opinion, may interfere with the interpretation of QTc interval changes, including abnormal ST-T-wave morphology.*

    b. *QT interval corrected by Fridericia's formula (QTcF) of >450 msec in men or >460 msec in women.*

    c. *PR >220 msec.*

    d. *Second or third degree atrioventricular block or atrioventricular dissociation.*

    e. *Complete left or right bundle branch block.*

    f. *Ventricular or atrial premature contractions in couplets or higher in grouping.*

4. Has abnormal laboratory values for any of the hematology, serum chemistry, coagulation, or urinalysis tests outlined in Section 7.2.1 at Screening or Check-in on Day –1.

5. Is positive for hepatitis B surface antigen, hepatitis C virus antibody, or human immunodeficiency virus antibody types 1 and 2 within 35 days of enrollment.

6. Has any psychiatric condition or history of psychiatric condition that, in the opinion of the Investigator or Sponsor, would interfere with the subject's ability to participate in the study or increase the risk of the participation for that subject.

7. Is unwilling to abstain from alcohol, caffeine-, or other xanthine-containing foods or beverages, tobacco or nicotine-containing products, and all bergamottin-containing fruits and fruit juices (eg, Seville-oranges, grapefruit or grapefruit juice, pomelos, pomegranate or pomegranate juice, cranberry or cranberry juice) 72 hours prior to Day –1 and through discharge on Day 5.

8. Is unwilling to abstain from strenuous exercise 7 days before Day –1 through Day 15.

9. Has a history of alcoholism or drug/chemical abuse within 6 months prior to Day –1.

10. Has excessive alcohol consumption (regular alcohol intake >21 units per week for male subjects and >14 units of alcohol per week for female subjects) (1 unit is equal to approximately ½ pint [200 mL] of beer, 1 small glass [100 mL] of wine, or 1 measure [25 mL] of spirits).

11. Has a positive urine drug screen at Screening or Check-in on Day –1.

12. Has a positive cotinine urine test at Check-in on Day –1.

13. Has any confirmed or suspected immunosuppressive or immunodeficient condition, including, but not limited to, human immunodeficiency virus infection, or use of anti-cancer chemotherapy or radiation therapy (cytotoxic) in the 3 years prior to Screening.

14. Provides verbal history of vaccination with a licensed or investigational flavivirus vaccine for any of the following diseases: Zika virus, DENV, yellow fever virus, Japanese encephalitis virus, West Nile virus, St. Louis encephalitis virus, or tick-borne encephalitis virus or reportedly diagnosed with a flavivirus infection or disease.

15. Plans to receive a licensed flavivirus vaccine or participate in a flavivirus vaccine trial during the study.

16. Is positive for DENV or West Nile virus by immunoglobulin M, or immunoglobulin G, or Zika by immunoglobulin M serology testing within 35 days of enrollment.

17. Has a history of significant hypersensitivity, intolerance, or allergy to any drug compound, food, or other substance, unless approved by the Investigator (or designee).

18. Has had major surgery within 3 months prior to Screening or plans to have a major surgery during the study through the final follow-up on Day 120 (±5).

19. Has been previously treated for a medical condition with a licensed monoclonal or polyclonal antibody any time in the past.

20. Has been dosed in a clinical study involving administration of an investigational monoclonal or polyclonal antibody in the 18 months (550 days) prior to IP administration on Day 1.

21. Has received an investigational drug within 28 days of IP administration on Day 1.

22. Has used any prohibited medication within 30 days prior to Day –1 or plans to use prohibited medication during the study.

    a. *Prohibited medications include immunosuppressive drugs, immune modulators (except acetaminophen), oral corticosteroids, inhaled or intranasal steroids (<800 µg/day beclomethasone is acceptable), and anti-neoplastic agents. Topical steroids are acceptable.*

23. Has received or plans to receive any live vaccination, experimental or otherwise, within 28 days prior to or after Day 1; and receipt or planned receipt of an inactivated vaccination (or any COVID-19 vaccine), experimental or otherwise, within 14 days prior to or after Day 1.

24. Has received blood products within 60 days prior to Day –1.

25. Has donated or lost more than 450 mL of blood or plasma within 56 days of IP infusion on Day 1. The subject must also agree to refrain from donating blood or plasma during the study.

26. Has poor peripheral venous access.

27. Has previously completed or withdrawn from this study.

28. Is a current study site staff paid entirely or partially by the contractor for this trial, or staff who are supervised by the Investigator or subinvestigators.

29. In the opinion of the Investigator (or designee), the subject is not suitable for entry into the study.

5.2 Withdrawal from the Study, Discontinuation of Investigational Product, or Study Termination

5.2.1 Withdrawal from the Study or Discontinuation of the Investigational Product

Subjects are free to withdraw from the study at any time upon request. Subject participation in the study may be stopped at any time at the discretion of the Investigator or at the request of the Sponsor.

The Sponsor will be notified in advance whenever possible if subjects are stopped at the discretion of the Investigator. When a subject withdraws from the study or is stopped, the reason(s) are to be recorded by the Investigator on the relevant page of the electronic case report form (eCRF). Whenever possible, any subject who withdraws or is stopped from the study prematurely will undergo all ET assessments. Any subject who fails to return for final assessments will be contacted by the clinical site in an attempt to have them comply with the protocol. The status of subjects who fail to complete final assessments will be documented in the eCRF.

The reasons for stopping subject participation by the Investigator or Sponsor include, but are not limited to the following:

- The subject meets individual halting criteria (Section 8.6.2).
- Subject noncompliance based on any of the following criteria after IP administration and before completion of all follow-up visits through Day 120 (±5):
    - Positive urine drug screen
    - Has major surgery
    - Received another IP
    - Uses prohibited medication
    - Receives blood products or donates blood or plasma
- An AE or new clinical finding(s) for which continued participation, in the opinion of the Investigator, might compromise the safety of the subject, interfere with the subject's successful completion of this study, or interfere with the evaluation of responses.
- The subject is lost to follow-up.
- Subjects becoming pregnant prior to or following IP administration prior to the final follow-up visit on Day 120 (±5). Subject becoming pregnant after IP administration will not be included in data analysis but will be followed for safety.

If the subject consents, every attempt will be made to follow all AEs through resolution including those subjects that elect to withdraw. The procedures that collect safety data for the

purposes of research must be inclusive in the original ICF or the Investigator may seek subsequent informed consent using an IRB-approved consent form with the revised procedures.

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

5.2.2 Subject Replacement

Any subject who withdraws before completing the study but after receiving IP may be replaced at the discretion of the Investigator and in consultation with the Sponsor. Any replacement subject will be assigned to receive the same treatment as the subject he or she is replacing.

5.2.3 Study Termination

Although the Sponsor has every intention of completing the study, they reserve the right to discontinue it at any time for clinical or administrative reasons.

If the study is prematurely terminated by the Sponsor, any regulatory authority, or the Investigator for any reason, the Investigator will promptly inform the study subjects and assure appropriate therapy or follow-up for the subjects, as necessary. The Investigator will provide a detailed written explanation of the termination to the IRB.

The end of the study is defined as the date on which the last subject completes the last visit (including the ET visit and any additional long-term follow-up). Any additional long-term follow-up that is required for monitoring of the resolution of an AE or finding may be appended to the clinical study report.

6 STUDY PROCEDURES

Before performing any study procedures, all potential subjects will sign an ICF as outlined in Section 9.2.

A complete schedule for all study procedures is detailed in the schedule of events (Section 17.1).

The IP will be administered in the clinical unit under direct observation of clinic personnel and will be recorded in the eCRF. Clinic personnel will confirm that the subject has received the entire dose of the IP. Complete information regarding any partial or interrupted infusions will be documented. The date and time (start and stop) of IP dosing will be recorded on the appropriate page in the eCRF.

A study physician will be present at the time of IP administration and the subject will be under observation during the 60-minute infusion. Vital signs will be monitored, and an ECG will be recorded prior to, during, and after the infusion as outlined in the schedule of events (Section 17.1). The study site staff will observe the subjects closely for signs and symptoms of infusion-related reactions, including anaphylaxis and anaphylactoid type reactions. Should an adverse reaction of this type occur, the subject will be treated using the standard protocol at the clinical site. Infusion-related AEs and suspected hypersensitivity reactions will be documented, including the clinical presentation and severity (based on the grading scales in Section 17.2) of the event.

For the specified time points, PK sample collection will take precedence over all other study sampling procedures and will therefore occur at the nominal time as described in Section 7.2.2. Multiple non-PK study procedures scheduled at the same time point should be performed as close as possible to the nominal time point (within 60 minutes predose and within 30 minutes for postdose time points). When procedures overlap or occur at the same time point, all blood draws should follow ECGs or vital signs, and PK sampling should be timed to occur last and as close to the scheduled time window as possible. The total amount of blood collected from each subject over the duration of the study, including any extra assessments that may be required, will not exceed 500 mL. Refer to the laboratory manual for specific details on blood volumes used for laboratory testing.

7 DESCRIPTION OF CLINICAL AND LABORATORY EVALUATIONS

The timing and frequency of all safety assessments are listed in the schedule of events (Section 17.1).

7.1 Clinical Evaluations

Safety endpoints will include monitoring and recording of AEs and SAEs, clinical laboratory test results (hematology, chemistry, and urinalysis), vital sign measurements, 12-lead ECG results, and physical examination findings.

For all safety assessments, the Investigator will determine whether results are clinically significant, which is defined as any variation in a result that has medical relevance and may result in an alteration in medical care (eg, active observation, diagnostic measures, or therapeutic measures). If clinical significance is noted, the result and reason for significance will be documented on the AE page in the subject's eCRF and the Investigator will monitor the subject until the result has reached the reference range or the result at Screening, or until the Investigator determines that follow-up is no longer medically necessary.

7.1.1 Vital Signs

Vital sign measurements will include systolic and diastolic blood pressure, oral body temperature, heart rate, and respiratory rate. The subject will have rested comfortably in a seated or supine position for at least 5 minutes before all measurements are taken. For time points when ECGs and vital signs occur at the same time point, it is acceptable to perform the vital sign measurements approximately 2 minutes after the ECG as long as the subject remains resting in the supine position. Additional vital signs may be measured for safety of the subjects at the discretion of the Investigator. All measurements will be recorded in the subject's eCRF.

7.1.2 Twelve-lead ECG

Twelve-lead ECG recordings will be obtained after the subject has rested comfortably in a supine position, for at least 10 minutes. The Investigator should review and sign the ECG for any immediate issues. Electrocardiogram assessments will include comments on whether the tracings are normal or abnormal, rhythm, presence of arrhythmia or conduction defects, morphology, any evidence of myocardial infarction, or ST-segment, T-wave, and U-wave abnormalities. In addition, measurements of the following intervals will be measured and reported: RR interval, PR interval, QRS width, and QTcF.

7.1.3 Medical History

Medical history will be obtained by direct interview and include a history of significant medical disorders of the head, eyes, ears, nose, throat, cardiovascular system, lungs, gastrointestinal tract,

liver, kidney, urologic, nervous system, blood, lymph glands, endocrine system, musculoskeletal system, skin, and genital/reproductive tract. A history of any allergies, cancer, immunodeficiency, psychiatric illness, substance abuse, and autoimmune disease will be solicited. The medical history will include current and past medical diagnoses, hospitalizations, and major surgical procedures.

At each visit, subjects will be asked about any changes in health, new or worsening symptoms, or medications taken since their last visit. Any new adverse health changes will be recorded as an AE.

7.1.4 Physical Examination

A full physical examination will be performed at Screening (skin, head, ears, eyes, nose, throat, neck, thyroid, lungs, heart, cardiovascular, abdomen, lymph nodes, and musculoskeletal system/extremities). Focused examination (lungs, cardiovascular, abdomen, and skin) will be performed at Check-in (Day –1) and on Days 5, 8, and 15. Targeted, symptom-directed physical examinations will be performed at all other follow-up visits.

7.1.5 Assessment of Concomitant Medications/Treatments other than the Investigational Product

Restrictions for prior and concomitant medications and therapies are provided in Section 5.1.1 and Section 5.1.2. Prior and concomitant medications and therapies will be coded using the latest version of the World Health Organization Drug Dictionary.

7.1.5.1 Prior Medications

Information regarding prior medications taken by the subject within the 30 days before signing the ICF will be recorded in the subject's eCRF.

7.1.5.2 Concomitant Medications

The Investigator is responsible for ensuring that details regarding the medication are adequately recorded in the subject's source documents and in the eCRF. Medication information will be recorded at the Screening visit for the prior 30 days and at Day –1. At each follow-up study visit, new concomitant medication and changes to existing medications will be recorded. Any concomitant medication deemed necessary for the welfare of the subject during the study may be given at the discretion of the Investigator. If a concomitant medication is taken, except for those specified in the protocol, a joint decision will be made by the Investigator and the Sponsor to continue or discontinue the subject based on the time the medication was administered, its pharmacology and PK, and whether the use of the medication will compromise the safety of the subject or the interpretation of the data.

7.2 Laboratory Evaluations

7.2.1 Clinical Laboratory Evaluations

The following clinical laboratory assessments will be performed at the time points indicated in the schedule of events (Section 17.1):

Hematology	Absolute neutrophil count, hemoglobin, platelet count, and white blood cell count
Serum Chemistry	Alanine aminotransferase, albumin[a], alkaline phosphatase, aspartate aminotransferase, bicarbonate[b], blood urea nitrogen, calcium[a], chloride[b], creatinine[c], glucose (fasted), potassium, sodium, and total bilirubin
Coagulation[a]	Prothrombin time and activated partial thromboplastin time
Urinalysis	Blood, glucose[a], and protein
Serology (IgG)[a]	Hepatitis B surface antigen, hepatitis C virus antibody, and human immunodeficiency virus antibody types 1 and 2
Flavivirus Screening (IgM and IgG)[a]	DENV antibody, Zika virus antibody (IgM only), and West Nile virus antibody
Urine Drug Screen[d]	Amphetamines (includes methamphetamines and ecstasy/methylenedioxymethamphetamine), barbiturates, benzodiazepines, cannabinoids, cocaine metabolites, cotinine, ethyl alcohol, methadone, methamphetamines, opiates (including heroin, codeine, and oxycodone/oxymorphone)
Pregnancy Test (female subjects only)[e]	Follicle-stimulating hormone, serum pregnancy test (human chorionic gonadotropin)

Abbreviations: IgG, immunoglobulin G; IgM, immunoglobulin M.

Note: Results of alanine aminotransferase, aspartate aminotransferase, alkaline phosphatase, blood urea nitrogen, creatinine, and total bilirubin that are below the reference ranges are acceptable and will not exclude participation.

[a] Clinical laboratory assessments will be performed at Screening and Check-in on Day –1 only, except serology and flavivirus screening will occur only at Screening.

[b] Bicarbonate and chloride will only be used for calculation of anion gap ([sodium + potassium] - [chloride + bicarbonate)]. Volunteers with an anion gap greater than the upper limit of normal at Screening and Check-in on Day –1 will be excluded from participation in the study.

[c] Volunteers with an estimated creatinine clearance (calculated using CKD-EPI method) <90 mL/min/1.73 cm^2 at Screening will be excluded from participation in the study.

[d] Urine drug screening will be performed at Screening, Check-in on Day –1, on Day 22 (±2), on Day 43 (±3), and on Day 85 (±5) except cotinine screening will occur only on Day –1.

[e] For females in menses, screening urinalysis may be postponed or repeated, but a result should be available prior to Day 1.

Results of the absolute neutrophil count, hemoglobin, platelet count, white blood cell count, alanine aminotransferase, albumin, anion gap, aspartate aminotransferase, blood urea nitrogen, calcium, creatinine, creatinine clearance, glucose (fasting), potassium, sodium, prothrombin time and activated partial thromboplastin time, and urinalysis for blood, glucose, and protein must be normal at Screening and Check-in on Day –1 for eligibility. A repeat is allowed for out-of-range values, when not considered clinically significant by the Investigator, for the purpose of determining eligibility. The Investigator may choose to run addition clinical laboratory assessments at Screening or Check-in to assess volunteer eligibility to enroll. Results of these tests that fall outside of the reference ranges will be evaluated for inclusion into the study at the discretion of the Investigator.

Laboratory assessments for monitoring safety and determining dose escalation include absolute neutrophil count, hemoglobin, platelet count, white blood cell count, alanine aminotransferase, alkaline phosphatase, aspartate aminotransferase, anion gap, blood urea nitrogen, creatinine, glucose (fasted), potassium, sodium, and total bilirubin. Instruments will be programmed to only generate results for hematology (including % relative neutrophils used to calculate absolute neutrophil count) and chemistry tests (including chloride and bicarbonate used to calculate anion gap) used to monitor safety and dose escalation after enrollment. Results of % relative neutrophils, chloride and bicarbonate will not individually be reviewed for evaluation of dose escalation or study halting criteria.

Instruments will be programmed to only generate results for hematology and chemistry tests used to monitor safety and determining dose escalation include absolute neutrophil count, hemoglobin, platelet count, white blood cell count, alanine aminotransferase, anion gap, blood urea nitrogen, creatinine, glucose (fasted), potassium, sodium, and total bilirubin.

The clinical laboratory that performs the tests will provide the reference ranges for all clinical laboratory parameters. Clinical laboratory tests that are thought to be due to a laboratory error may be repeated once at the discretion of the Investigator for assessment of inclusion and exclusion criteria or evaluation of clinical laboratory abnormalities.

7.2.2 Pharmacokinetic Sampling

Blood samples for PK analysis of concentrations of AV-1 in serum will be collected predose (within 15 minutes) and at 0.5, 1 (end of infusion), 1.25, 1.5, 2, 3, 4, 6, 8, 12, 24 and 48 hours after the start of the infusion, on Day 5 prior to discharge, and at the follow-up visits on Days 8 (+2), 15 (±2), 22 (±2), 29 (±2), 43 (±3), 57 (±3), 85 (±5), and 120 (±5) (or ET). A validated ECL ELISA method will be used to quantify AV-1 in serum samples for PK analyses as outlined in Section 10.4.3.

7.2.3 Anti-AV-1 Antibody Sampling

Blood samples for anti-AV-1 antibody analysis will be collected at Check-in (Day –1) and on Days 29 (±2), 85 (±5), and 120 (±5) (or ET). A validated ECL ELISA method will be used for detection and confirmation of pre-existing and treatment-emergent anti-AV-1 antibodies in

serum samples based on the MesoScale Discovery platform that utilizes labeled AV-1 for detection of anti-AV-1 antibodies and treatment-emergent antibodies.

7.2.4 Hypersensitivity Testing

Subjects will be monitored closely for signs and symptoms of hypersensitivity during and following infusion of the IP. A blood sample will be collected for all subjects on Day –1 and tested only if a suspected hypersensitivity reaction occurs during or following IP infusion on Day 1 at the discretion of the Investigator in consultation with the Sponsor. The subject will be treated using the standard site procedures if a subject develops a suspected hypersensitivity reaction. At the discretion of the Investigator, an additional blood sample will be drawn as soon as possible after the event occurs (Day 1) and again the following day (Day 2). Hypersensitivity testing of the Day –1, Day 1, and Day 2 samples will only occur in the event of a suspected hypersensitivity reaction at the discretion of the Investigator and in consultation with the Sponsor. Refer to the manual of procedures for further details.

7.2.5 Laboratory Specimen Preparation, Handling, and Storage

Details for the collection, processing, and storage of samples for assessment of PK and anti-AV-1 antibody will be provided to the clinical unit separately and outlined in the laboratory manual.

7.2.6 Laboratory Specimen Shipping

Details for the shipping of samples for the assessment of PK and anti-AV-1 antibody will be provided to the clinical unit separately and outlined in the laboratory manual.

8 ASSESSMENT OF SAFETY

8.1 Assessing and Recording Safety Parameters

Subjects will be monitored throughout the duration of the study to assess safety of a single IV infusion of AV-1 as per the schedule of events (Section 17.1).

Safety assessments will include:

- Incidence and severity of AEs
- Incidence of SAEs
- Physical examinations
- Clinical safety laboratory tests
- Vital sign measurements
- 12-lead ECGs

8.1.1 Adverse Events

An AE is defined by ICH E6(R2) as any untoward medical occurrence in a patient or clinical investigation subject administered a pharmaceutical product regardless of its causal relationship to the study treatment. The FDA defines an AE as any untoward medical occurrence associated with the use of a drug in humans, whether or not considered drug related.

An AE can therefore be any unfavorable and unintended sign (including an abnormal laboratory finding), symptom, or disease temporally associated with the use of medicinal (investigational) product. The occurrence of an AE may come to the attention of study personnel during study visits and interviews of a study recipient presenting for medical care, or upon review by a study monitor.

All AEs, including solicited local (infusion site) and systemic (subjective and quantitative) reactions, will be captured on the appropriate data collection form and in the eCRF. Information to be collected for AEs includes event description, date of onset, assessment of severity, relationship to the IP and alternate etiology (assessed only by those with the training and authority to make a diagnosis and listed on Form FDA 1572 as an Investigator), date of resolution, seriousness, and outcome. Adverse events occurring during the study collection and reporting period will be documented appropriately regardless of relationship. Adverse events will be followed through resolution.

Any medical condition that is present at the time that the subject is screened will be considered as baseline and not reported as an AE. However, if the severity of any pre-existing medical condition increases, it will be recorded as an AE.

8.1.1.1 Adverse Events Grading

All AEs (laboratory and clinical symptoms) will be graded for severity and assessed for relationship to the IP (see definitions below). Adverse events characterized as intermittent require documentation of onset and duration of each episode. The start and stop date of each reported AE will be recorded on the appropriate data collection form and eCRF. Changes in the severity of an AE will be documented to allow an assessment of the duration of the event at each level of intensity.

Severity of Event: Adverse events will be assessed by the Investigator using a protocol-defined grading system (toxicity table included in Section 17.2). For events not included in the protocol-defined grading system, the following guidelines will be used to quantify severity:

- Mild (Grade 1): Events that are transient and may require only minimal or no treatment or therapeutic intervention and do not interfere with the subject's usual activities of daily living.
- Moderate (Grade 2): Events that are alleviated with additional specific therapeutic intervention. The event interferes with usual activities of daily living, causing discomfort but poses no significant or permanent risk of harm to the subject.
- Severe (Grade 3): Events interrupt usual activities of daily living, or significantly affects clinical status, or may require intensive therapeutic intervention. Severe events are usually incapacitating.

Relationship to Investigational Product: The assessment of the AE's relationship to the IP will be done by the licensed study physician indicated on Form FDA 1572 and the assessment will be part of the documentation process. Whether the AE is related or not, is not a factor in determining what is or is not reported in this study. If there is any doubt as to whether a clinical observation is an AE, the event should be reported.

In a clinical trial, the IP must always be suspect. The relationship to the IP will be assessed for AEs using the terms related or not related:

- Related – There is a reasonable possibility that the IP caused the AE. Reasonable possibility means that there is evidence to suggest a causal relationship between the IP and the AE.
- Not Related – There is not a reasonable possibility that the administration of the IP caused the event.

8.1.2 Serious Adverse Events

An AE or suspected adverse reaction is considered an SAE if, in the view of either the Investigator or Sponsor, it results in any of the following outcomes:

- Death
- A life-threatening AE[1]

- 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 subject and may require medical or surgical intervention to prevent one of the outcomes listed in this definition. Examples of such medical events include, but are not limited to, 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.

[1]*An AE or suspected adverse reaction is considered "life-threatening" if, in the view of either the Investigator or Sponsor, its occurrence places the 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.*

Serious AEs will be:

- Assessed for severity and relationship to the IP and alternate etiology (if not related to the IP) by a licensed study physician listed on Form FDA 1572 or by the Investigator or subinvestigator.
- Recorded on the appropriate SAE data collection form and in the eCRF.
- Followed through resolution by a licensed study physician (a physician listed on Form FDA 1572 as the Investigator or subinvestigator).
- Reviewed and evaluated by the SRC, the safety monitoring committee (SMC) (if related), and the IRB.

8.2 Specification of Safety Parameters

Safety will be assessed by the frequency and severity of:

- Occurrence of SAEs from the administration of AV-1 to final follow-up visit on Day 120 (±5).
- Occurrence of unsolicited AEs from the administration of AV-1 to the final follow-up visit on Day 120 (±5).
- Occurrence of solicited AEs from the administration of AV-1 to Day 8.

- Occurrence of changes from baseline in physical examination, vital sign measurements, and clinical safety laboratory values from the administration of AV-1 to the final follow-up visit on Day 120 (±5).
- Occurrence of changes from baseline in ECG parameters in the 48 hours after starting the AV-1 infusion and at Days 8 (+2), 43 (±3), 85 (±5), and 120 (±5).

8.2.1 Solicited and Unsolicited Events

Solicited events are AEs that are common and known to occur following administration of the IP. Solicited events are defined in Table 17-1.

Unsolicited events are any other AEs that occur following administration of the IP.

8.2.2 Dose Escalation Criteria

This study is designed such that dose escalation is allowed only after a blinded safety data review of each cohort by the SRC has occurred as outlined in Section 8.7.1. Study assessments (eg, AEs, clinical laboratory test results, vital sign measurements, and 12-lead ECG results) will be used to evaluate safety of a given dose level. Dose escalation will be dependent on review of the safety data. Clinically significant results will be discussed with the Sponsor before dose escalation continues. Data from a minimum of 6 subjects dosed per cohort must be reviewed before each dose escalation.

Based on the review of safety data and recommendations by the SRC, SMC, or both, the Sponsor and the Investigator may choose to repeat a dose level, administer a dose less than the previous dose, escalate to a dose lower than the next planned dose, or prolong the duration of the infusion. Before implementing any change, the IRB will be notified and provided with the rationale for approval.

8.2.3 Sentinel Subjects

As a safety precaution in this first-in-human study, each cohort is divided into 2 groups with each group being dosed at least 72 hours apart. On the first day of dosing in all cohorts, the first group in each cohort will be comprised of 2 sentinel subjects. The randomization schedule will be constructed such that one of the sentinel subjects dosed on the first day will be randomly assigned to receive AV-1 and one of the sentinel subjects will be randomly assigned to receive placebo. After Investigator review of the safety data from the 48-hour postdose period for the sentinel subjects, which includes review of any AEs, abnormalities in ECGs, safety laboratory assessments, and vital signs, the remainder of the cohort (5 subjects randomly assigned to AV-1; one subject randomly assigned to placebo) will be dosed at least 72 hours after the sentinel subjects. Continuation from the sentinel subjects to dose the remaining 6 subjects will proceed if no study halting criteria are met as outlined in Section 8.6.3.

8.3 Reporting Procedures

8.3.1 Reporting Serious Adverse Events

Serious AEs will be followed until resolution even if this extends beyond the study-reporting period. Resolution of an AE is defined as the return to pretreatment status or stabilization of the condition with the expectation that it will remain chronic.

Any AE that meets a protocol-defined serious criterion must be submitted immediately (within 24 hours of site awareness) on an SAE form to:

PPD PVG Safety:
Telephone (24 hour): [redacted]
Fax: [redacted]
Email: [redacted]

PPD PVG Safety will notify and submit to the DMID Pharmacovigilance Group within 1 business day to:

DMID Pharmacovigilance Group
Clinical Research Operations and Management Support
[redacted]
SAE Hot Line: [redacted]
SAE FAX Number: [redacted]
SAE Email Address: [redacted]

In addition to the SAE form, select SAE data fields must also be entered into the Electronic Data Capture system.

Other supporting documentation of the event may be requested by the Sponsor Representative or DMID Pharmacovigilance Group and should be provided as specified in the safety management plan.

The Sponsor's Representative, Sponsor's Medical Monitor, and DMID Clinical Project Manager will also be notified of the SAE by the PPD Pharmacovigilance Group within 1 business day and will review and assess the SAE for regulatory reporting and potential impact on study subject safety and protocol conduct.

At any time after completion of the study, if the Investigator or subinvestigator becomes aware of an SAE that is suspected to be related to the IP, the Investigator or subinvestigator will report the event to the Sponsor Representative and DMID Pharmacovigilance Group.

8.3.2 Regulatory Reporting

Following notification from the Investigator, PPD will report events that are both serious and unexpected that are related to the IP to the FDA on behalf of AbViro, LLC (the Sponsor) and

within the required timelines as specified in 21 CFR Part 312.32: fatal and life-threatening events within 7 calendar days (by telephone or fax). All written reports will be sent to the FDA within 15 calendar days. All SAEs designated as "not related" to the IP, will be reported to the FDA at least annually in a summary format.

8.3.3 Reporting of Pregnancy

Subjects will be instructed that if they or their partner becomes pregnant during the study, this should be reported to the Investigator. The Investigator should also be notified of a pregnancy occurring during the study but confirmed after completion of the study. In the event that a subject or a subject's partner is subsequently found to be pregnant after the subject is enrolled in the study and has received the IP, the pregnancy will be followed through its resolution, and the status of the mother and child will be reported to the Sponsor after delivery.

A pregnancy notification form and follow-up will be completed.

8.4 Type and Duration of Follow-up of Subjects after Adverse Events

Adverse events will be assessed and followed from initial recognition of the AE through resolution even if the duration of follow-up goes beyond the final follow-up visit.

Serious AEs will be followed through resolution even if duration of follow-up goes beyond the final follow-up visit.

Resolution of an AE is defined as the return to pretreatment status or stabilization of the condition with the expectation that it will remain chronic.

8.5 Procedures to be Followed in the Event of Abnormal Laboratory Test Values or Abnormal Clinical Findings

Results of laboratory values collected during the conduct of the study will be reviewed by the Investigator or designee within 24 hours of receipt at the clinical site. As this study will be conducted in healthy subjects, assessment of laboratory abnormalities will be performed in accordance with the grading scale provided in Section 17.2, which is adapted from the FDA guideline, "Toxicity Grading Scale for Healthy Adult and Adolescent Volunteers Enrolled in Preventive Vaccine Clinical Trials" (DHHS 2007). The adaptation is applied to account for local laboratory normal values and for compliance with the Clinical Data Interchange Standards Consortium. Any findings meeting Grade 3 criteria must be repeated immediately to rule out spurious values. All abnormal laboratory test values and findings on physical examination will be included in the study data and summarized in the safety analyses as described in Section 10.4.5. Any treatment-emergent AEs that are appropriately graded according to the toxicity scale in Section 17.2 will be captured as AEs in the eCRF.

Abnormal laboratory values occurring during the study will be followed until repeat test results return to normal, stabilize, or a cause is identified. If the laboratory abnormality is identified at the final scheduled visit, the subject will be requested to return for additional testing.

8.6 Halting Rules

8.6.1 Dose Escalation Halting Criteria

If either of the following criteria is met, escalation to the next planned dose cohort will not proceed until all available study data have been reviewed by the SMC.

- Two or more subjects in the same cohort experience a Grade 2 related AE (laboratory or systemic) that is attributed to the study drug and coded under the same high-level term per the Medical Dictionary for Regulatory Activities coding, throughout the duration of the study.
- Two or more subjects in the same cohort experience a Grade 3 AE (laboratory or systemic) that is coded under the same high-level term per the Medical Dictionary for Regulatory Activities coding, regardless of causality assessment by the Investigator, throughout the duration of the study.

If dose escalation is halted, the study may resume if deemed acceptable by the SMC following review of all available study data.

8.6.2 Halting Criteria for Subject Infusions

The Investigator and study site staff must carefully observe the subjects for any AE during the infusion.

The infusion will be halted for an individual subject and will not resume if any of the following manifestations occur during the 1-hour infusion:

- Occurrence during dosing that, in the opinion of the Investigator, results in the inability to have confidence that the intended dose of IP was delivered.
- Infusion-related reaction of Grade 2 or higher.

  In case of a suspected infusion-related reaction, at the discretion of the Investigator, the infusion rate may be slowed from a rate of 250 mL/hour to a rate of 125 mL/hour for symptom management. The infusion may also be stopped. The subject will be observed to determine the severity of the reaction for a period of up to 15 minutes. Depending on the assessment of the subject, supportive care can be administered, and the infusion may permanently be stopped. If the Investigator determines that there is no infusion reaction, no ongoing supportive care is required after dosing is stopped, or if the symptoms of the clinical reaction have been improved after 15 minutes, the infusion may be restarted. The decision to

continue the infusion is based on the assessment by the Investigator. The infusion will be halted if there is a reoccurrence after a pause in dosing.

- Symptomatic low blood pressure of <90 mm Hg systolic or >30% decrease from baseline in systolic or diastolic blood pressure at predose.
- Confirmed tachycardia with an increase in resting heat rate to ≥130 beats per minute (bpm), or development of ventricular dysrhythmia, or bradycardia (<45 bpm, or <40 bpm in subjects with baseline of <60 bpm) associated with complaints of dizziness, nausea, or feeling faint.
- Syncope.
- Confusion.
- Suspected Grade 3 AE or SAE.

8.6.3 Study Halting Criteria

Study dosing must be stopped, and a review of available safety data will be conducted by the SMC if any of the following occur:

- Death of a dosed subject following infusion and prior to the subject's last follow-up visit that was not the result of trauma or accident, regardless of relatedness to the IP.
- Occurrence of a life-threatening allergic/hypersensitivity reaction (eg, anaphylaxis) in any subject, manifested by bronchospasm with or without urticaria, or angioedema requiring hemodynamic support with pressor medications or mechanical ventilation.
- One subject with an IP-related SAE of any grade.
- Two or more subjects with a Grade 3 AE in the same system organ class (systemic toxicity, clinical laboratory tests, or vital sign measurements) considered related to the IP, except scenarios where there are acceptable physiological explanations for a Grade 3 abnormality (eg, Grade 3 hematuria in a menstruating female).
- One or more subjects experience a confirmed Grade 3 QTcF (≥500 msec or increase ≥60 msec), or 2 or more subjects cumulatively have experienced a Grade 2 change in QTcF (≥480 msec).

8.7 Safety Oversight

8.7.1 Safety Review Committee

The SRC is composed of the DMID Medical Monitor, Sponsor Medical Monitor, and the Investigator at the clinical site. Objective dose escalation criteria and safety evaluations will be utilized. The Investigator will evaluate safety data from the sentinel group and notify the Sponsor Representative of intent to proceed if no study halting criteria (Section 8.6.3) are met. The SRC will evaluate the safety data through Day 8 for each cohort to determine whether dose escalation

can occur. These reviews do not necessitate formal meetings. The SRC will inform the Sponsor Representative that the study can proceed to begin dosing the next cohort if none of the events described in the study halting criteria (Section 8.6.3) or dose escalation criteria (Section 8.2.2) are met.

Should any of the study halting criteria or dose escalation halting criteria be met, the SMC will meet to evaluate the data and recommend appropriateness of further dosing. The decision to advance to the next dosing cohort will be documented and provided to the SRC and Sponsor Representative.

8.7.2 Safety Monitoring Committee

The SMC is an independent group of experts (at least 3 members) that monitor subject safety and advises the Sponsor. The SMC members will be separate and independent of study personnel participating in this study and should not have scientific, financial, or other conflicts of interest related to the study. The SMC will consist of members with appropriate expertise to contribute to the interpretation of the data from this trial. A quorum will consist of a simple majority.

The SMC will review all available safety data during all scheduled ad hoc meetings:

- When study halting criteria or dose escalation criteria are met.
- At the request of the Sponsor to review a potential safety concern identified by the Investigator, Sponsor Medical Monitor, or DMID Medical Monitor.
- When an SAE occurs.

Procedures for the SMC reviews and meetings will be defined in the SMC charter. The SMC will review applicable aggregate data including, but not limited to, enrollment, demographics, dosing, laboratory, and safety data at scheduled time points during the study as defined in the charter. The SMC may request to receive data by AV-1 versus placebo in a closed session. The SMC may also request that the blind be broken for individual subjects, as needed, to assess safety issues. The SMC will make a recommendation to the Sponsor Representative in writing to continue, modify, or terminate the study as an outcome of each review or meeting.

9 HUMAN SUBJECTS PROTECTION

9.1 Institutional Review Board

The Investigator will obtain IRB approval for this protocol to be conducted at the clinical site and send supporting documentation to the Sponsor Representative before initiating recruitment of subjects. The Investigator will submit applicable information to the IRB on which it relies for the review, to conduct the review in accordance with 45 CFR 46, ICH E6 GCP, and as applicable, 21 CFR 56 and 21 CFR 50 (Protection of Human Subjects), other federal, state, and local regulations. The IRB must be registered with OHRP as applicable to the research. The DMID Clinical Project Manager must receive the documentation that verifies IRB approval for this protocol, associated informed consent documents, and upon request any recruitment material and handouts or surveys intended for the subjects, prior to the recruitment and enrollment of subjects.

Any amendments to the protocol or consent materials will be approved by the IRB before they are implemented. Institutional review board review and approval will occur at least annually throughout the enrollment and follow-up of subjects and may cease if annual review is no longer required by applicable regulations and the IRB. The Investigator will notify the IRB of deviations from the protocol and reportable SAEs, as applicable to the IRB policy.

Each institution engaged in this research will hold a current FWA issued by the OHRP for federally funded research.

9.2 Informed Consent Process

Written informed consent in compliance with US Title 21 CFR Part 50 shall be obtained from each subject before he or she enters the study or before performing any unusual or nonroutine procedure that involves risk to the subject. If any institution-specific modifications to study-related procedures are proposed or made by the clinical site, the consent should be reviewed by the Sponsor before IRB submission. Once reviewed, the Investigator will submit the ICF to the IRB for review and approval before the start of the study. If the ICF is revised during the course of the study, all active participating subjects must sign the revised form.

Before recruitment and enrollment, each prospective subject will be given a full explanation of the study and will be allowed to read the approved ICF. Once the Investigator is assured that the subject understands the implications of participating in the study, the subject will be asked to give his or her consent to participate in the study by signing the ICF. A copy of the ICF will be provided to the subject.

9.3 Subject Confidentiality

Subject confidentiality is strictly held in trust by the participating Investigator, their study site staff, and the Sponsor and their agents. This confidentiality includes documentation, investigational data, subject's clinical information, and all other information generated during participation in the study. No information concerning the study, or the data generated from the study will be released to any unauthorized third party without prior written approval of the Sponsor and the subject. Subject confidentiality will be maintained when study results are published or discussed in conferences. The study monitor, or other authorized representatives of the Sponsor or governmental regulatory agencies may inspect all documents and records required to be maintained by the Investigator, including but not limited to, medical records (office, clinic, or hospital) and pharmacy records for the subjects in this study. The clinical site will permit access to such records.

All records will be kept locked and all computer entry and networking programs will be carried out with coded numbers only and with password-protected systems. All specimens, evaluation forms, reports, and other records that leave the clinical site will be identified only by a coded number.

9.4 Certificate of Confidentiality

To protect privacy, we have received a Certificate of Confidentiality. With this Certificate, the researchers cannot be forced to release information that may identify the research subject, even by a court subpoena, in any federal, state, or local civil, criminal, administrative, legislative, or other proceedings. The researchers will use the Certificate to resist any demands for information that would identify the subject, except as explained below.

The Certificate cannot be used to resist a demand for information from personnel of the US Government that is used for auditing or evaluation of federally funded projects, like this study, or for information that must be released in order to meet the requirements of the FDA.

A Certificate of Confidentiality does not prevent the subject from voluntarily releasing information about themselves or their involvement in this research. If any person or agency obtains a written consent to receive research information, then the researchers may not use the Certificate to withhold that information.

The Certificate of Confidentiality does not prevent the researchers from reporting without the subject's consent, information that would identify the subject as a participant in the research project regarding matters that must be legally reported including: child and elder abuse, sexual abuse, or wanting to harm themselves or others.

The release of individual private information or specimens for other research will only occur if consent was obtained from the individual to whom the information, document, or biospecimen

pertains, or for the purposes of other research that is in compliance with applicable federal regulations governing the protection of human subjects in research.

9.5 Costs, Subject Compensation, and Research-Related Injuries

There is no cost to subjects for the research tests, procedures, and IP while taking part in this trial. Procedures and treatment for clinical care may be billed to the subject, subject's insurance, or third party. Subjects may be compensated for their participation in this study. Compensation will be in accordance with the local IRB's policies and procedures, and subject to IRB approval.

If it is determined by the Investigator that an injury occurred to a subject as a direct result of the tests or treatments that are done for this study, then referrals to appropriate health care facilities will be provided to the subject. Study personnel will try to reduce, control, and treat any complications from this study. Immediate medical treatment may be provided by the participating clinical site. No financial compensation will be provided to the subject by the National Institute of Allergy and Infectious Diseases, the National Institutes of Health (NIH), or by the participating clinical site for any injury suffered due to participation in this study.

10 STATISTICAL CONSIDERATIONS

10.1 Study Hypotheses

No hypothesis will be formally tested.

10.2 Sample Size Considerations

This study will enroll a total of 40 subjects in 5 cohorts of 8 subjects each and up to 8 subjects for replacements as necessary. Each cohort will consist of 6 AV-1 treated and 2 placebo treated subjects.

10.3 Treatment Assignment Procedures

The PPD randomization statistician will generate the randomization schedule. The PPD randomization statistician will not directly be involved in study conduct, data management, or data analysis. The first group will be comprised of 2 eligible subjects (sentinel subjects), with 1 subject randomly assigned to receive AV-1 and 1 subject randomly assigned to receive placebo. The second group will be comprised of 6 eligible subjects (remaining cohort), with 5 subjects randomly assigned to receive AV-1 and 1 subject randomly assigned to receive placebo with an overall ratio of 6:2 in each cohort. Continuation to dose the remaining 6 subjects in the second group will be at the discretion of the Investigator following consultation with the Sponsor Representative.

10.3.1 Randomization Procedures

This is a double-blind study. Neither the subjects, the Investigator, nor the Sponsor will be aware of the treatment assignment. Subjects will be randomly assigned to receive AV-1 or placebo in a 6:2 randomization ratio. Access to the randomization code will be strictly controlled according to the standard operating procedures of PPD.

10.3.2 Blinding

The randomization list, as well as individual, subject-specific blind breaking documents, will be generated by the PPD randomization statistician and transferred to the unblinded study pharmacist prior to the start of the study. Pharmacy staff will store the subject-specific blind breaking documents in a tamper-evident container for afterhours access per standard clinic procedures. Study site staff participating in the administration of the IP and assessment of the subjects will not be aware of the contents of the IV bag. It is not anticipated that AV-1 and placebo will be distinguishable; however, masking overlays will be employed to maintain blinding and ensure the study site staff and the subject cannot determine whether AV-1 or placebo is being infused.

To maintain the study blind, AV-1 and placebo doses will be labeled with subject number, expiration date, expiration time, and volume. If the dose is not administered before the expiration time, a new dose should be requested from the unblinded pharmacy, if needed. The prepared dose, AV-1 or placebo, will be administered by appropriately trained neutral study site dosing staff as a single IV infusion over approximately 60 minutes by a controlled infusion device.

10.3.3 Breaking the Blind

Blind breaking envelopes will be prepared. A subject or subjects may be unblinded in the event of a dose-limiting toxicity, SAE, or other event, or if there is a medical emergency where the identity of the drug must be known to properly treat a subject. A cohort may be unblinded to determine if dose escalation to the next dose level will terminate. If a subject becomes seriously ill or pregnant during the study, the blind will be broken only if knowledge of the administered study drug will affect that subject's treatment options. In the event of a medical emergency requiring identification of the IP administered to an individual subject, the Investigator will make every attempt to contact the Sponsor Medical Monitor to explain the need for opening the code within 24 hours of opening the code. The Investigator will be responsible for documenting the time, date, reason for the code break, and the names of the personnel involved.

10.4 Final Analysis Plan

Prior to clinical database lock for this study, a statistical analysis plan will be finalized. The statistical analysis plan will detail the analyses to be conducted, the statistical methods that will be employed, and the formats and content of the final tables, listings, and figures to be produced.

10.4.1 Populations for Analysis

The analysis populations are as follows:

- The safety population will include all subjects who receive the full dose of the IP.
- The PK population will include subjects who receive the full dose of AV-1 and have at minimum all samples from predose through 1.5 hours from the start of infusion to have sufficient concentration data to support accurate estimation of at least 1 PK parameter.

10.4.2 Statistical Analyses

Details of all statistical analyses will be described in a separate statistical analysis plan. All data collected will be presented in data listings. Data from subjects excluded from an analysis population will be presented in the data listings, but not included in the calculation of summary statistics.

For categorical variables, frequencies and percentages will be presented. Continuous variables will be summarized using descriptive statistics (number of subjects, mean, standard deviation [SD], minimum, median, and maximum).

All subjects receiving placebo across cohorts will be pooled into 1 treatment group for summary purposes.

Baseline demographic and background variables will be summarized overall for all subjects. The number of subjects who enroll in the study and the number and percentage of subjects who complete the study will be presented. Frequency and percentage of subjects who withdraw or discontinue from the study, and the reason for withdrawal or discontinuation, will also be summarized.

10.4.3 Pharmacokinetic Analyses

Serum concentration data will be listed and summarized by time point for each dose level using descriptive statistics (number of subjects, mean, SD, coefficient of variation [CV], minimum, median, and maximum). Serum concentration versus time profiles for each subject will be presented graphically. The mean serum concentrations versus scheduled time profiles will be presented graphically by dose.

The PK parameters of AV-1 will be analyzed based on the actual sampling times. All parameters will be calculated using the latest version of Phoenix® WinNonlin® (Certara USA Inc., Princeton, New Jersey) or SAS® (SAS Institute Inc., Cary, North Carolina). Pharmacokinetic parameters will be summarized by time point for each dose level using descriptive statistics (number of subjects, arithmetic mean, SD, CV, minimum, median, and maximum). Geometric means and geometric CV will be reported for $AUC_{0\text{-}tlast}$, $AUC_{0\text{-}\infty}$, and C_{max}. The following serum PK parameters will be calculated as data permit:

C_{max}	Maximum observed serum concentration
T_{max}	Time to reach maximum observed serum concentration
$AUC_{0\text{-}48}$	Area under the serum concentration-time curve from time 0 to 48 hours postdose, calculated using the linear trapezoidal rule method
$AUC_{0\text{-}tlast}$	Area under the serum concentration-time curve from time 0 to the last quantifiable concentration, calculated using the linear trapezoidal method
$AUC_{0\text{-}\infty}$	Area under the serum concentration-time curve from time 0 extrapolated to infinity, calculated as $[AUC_{0\text{-}tlast} + (C_t/\lambda_z)]$ where C_t is the last observed serum drug concentration. $\%AUC_{exp}$ must be <0.20 to retain $AUC_{0\text{-}\infty}$ in the summary and statistical tables
$t_{1/2}$	Apparent terminal half-life, calculated as $\ln(2)/\lambda_z$
CL	Total serum clearance, calculated as $Dose/AUC_{0\text{-}\infty}$
V_z	Volume of distribution during the terminal phase, calculated as $Dose/AUC_{0\text{-}\infty} * \lambda_z$

In addition to the above PK parameters, which will be listed and summarized, the following parameters will also be listed to document the selection of data points used to estimate $t_{1/2}$ using noncompartmental procedures:

$\%AUC_{exp}$	Percentage of the area extrapolated for calculation of $AUC_{0-\infty}$
λ_z	Apparent terminal elimination rate constant
Number of points	Number of data points used to estimate λ_z; a minimum of 3 data points must be used, and C_{max} must not be included
λ_z lower (h)	Lower bound used for the estimation of λ_z
λ_z upper (h)	Upper bound used for the estimation of λ_z
Rsq	r^2, the coefficient of determination (goodness of fit statistic); visual inspection of the terminal slope will be performed. In general, λ_z may only be retained if $r^2 \geq 0.80$

Where data are available, AV-1 dose proportionality will be examined across the dose groups. The PK parameters will be analyzed for dose proportionality using a power model approach or analysis of variance model as appropriate. Additional analysis may be conducted if deemed appropriate.

Dose proportionality will be tested using the power regression model for $AUC_{0-tlast}$, $AUC_{0-\infty}$, and C_{max}, defined as:

$$\ln[\text{PK parameter}] = \beta_0 + \beta_1 \ln[\text{dose}]$$

where the PK parameter is an AUC or C_{max}. The null hypothesis being tested is that the AUC and C_{max} values are dose proportional, or slope (β_1) = 1.

10.4.4 Anti-AV-1 Antibody Analyses

Serum samples for measurement of anti-AV-1 antibody levels collected on Days –1, 29, 85, and 120 (±5) (or ET) will be analyzed by an ECL ELISA method. The proportion of subjects with detectable anti-AV-1 antibody responses prior to AV-1 dosing and treatment-emergent anti-AV-1 antibodies will be reported.

10.4.5 Safety Analyses

Adverse events will be coded by preferred term and system organ class using the latest version of the Medical Dictionary for Regulatory Activities and summarized by treatment and overall. All AE data will be presented in a data listing. Treatment-emergent AEs will be summarized by treatment and overall, as well as by severity and relationship to the IP. Serious AEs and AEs leading to discontinuation of the IP will also be presented in the data listings and summarized by treatment and overall.

Actual values and changes from baseline for clinical laboratory test results, vital sign measurements, and 12-lead ECG results will be summarized by treatment at each time point using descriptive statistics (number of subjects, mean, SD, minimum, median, and maximum). Shift tables will be generated for clinical laboratory test results. Physical examination findings will be presented in a data listing.

10.5 Handling of Missing Data

Concentrations that are below the limit of quantification (BLQ) will be treated as zero for descriptive statistics. Mean BLQ concentrations will be presented as BLQ, and the SD and CV will be reported as not applicable. Missing concentrations will be excluded from the calculations.

For the PK analysis, BLQ values will be treated as zero with the exception that a BLQ value between 2 quantifiable concentrations will be set as missing. Missing concentrations will be treated as missing from the PK parameter calculations. If consecutive BLQ concentrations are followed by quantifiable concentrations in the terminal phase, those concentrations after BLQ concentrations will be treated as missing.

10.6 Interim Analyses

No formal interim analyses will be performed in this study. A blinded safety data review will be performed for each cohort before dose escalation is allowed (Section 8.2.2).

11 SOURCE DOCUMENTS AND ACCESS TO SOURCE DATA/DOCUMENTS

The clinical 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 clinical 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, hospital records, 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, microfiches, photographic negatives, microfilm or magnetic media, x-rays, and subject files and records kept at the clinical site pharmacy, at the laboratories, and medico-technical departments involved in the clinical trial.

12 QUALITY CONTROL AND QUALITY ASSURANCE

This study will be conducted using the quality processes described in applicable procedural documents. The quality management approach to be implemented will be documented and will comply with current ICH guidance on quality and risk management. All aspects of the study will be monitored for compliance with applicable government regulatory requirements, current GCP, the protocol, and standard operating procedures. The monitor will maintain current personal knowledge of the study through observation, review of study records and source documentation, and discussion of the conduct of the study with the Investigator and study site staff. Electronic CRFs and electronic data capture will be utilized. The electronic data capture system is validated and compliant with US Title 21 CFR Part 11. Each person involved with the study will have an individual identification code and password that allows for record traceability. There may be an internal quality review audit of the data and additional reviews by the clinical monitor.

13 DATA HANDLING AND RECORD KEEPING

13.1 Data Management Responsibilities

All source documents and laboratory reports must be reviewed by the clinical team and data entry staff who will ensure that they are accurate and complete. Adverse events must be graded, assessed for severity and causality, and reviewed by the Investigator or designee.

Data collection is the responsibility of the study site staff at the clinical site under the supervision of the Investigator. During the study, the Investigator must maintain complete and accurate documentation of the study.

PPD, the contract research organization, will serve as the statistical and data coordinating center for this study and will be responsible for data management, quality review, analysis, and study data reports. The Investigator is responsible to ensure the accuracy, completeness, legibility, and timeliness of the data reported.

13.2 Data Coordinating Center/Biostatistician Responsibilities

Data collection is the responsibility of the study site staff at the participating clinical site under the supervision of the Investigator. During the study, the Investigator must maintain complete and accurate documentation for the study.

The statistical and data coordination center for this study will be responsible for data management, quality review, analysis, and reporting of the study data.

13.3 Data Capture Methods

Clinical (including AE/SAEs, concomitant medications, medical history, and physical assessments) and clinical laboratory values will be collected on data collection forms by study personnel then entered into eCRFs via a 21 CFR Part 11-compliant internet data entry system provided by PPD. The data system includes password protection and internal quality checks, such as automatic range checks, to identify data that appear inconsistent, incomplete, or inaccurate.

13.4 Types of Data

Data for this trial will include clinical, safety, and outcome measures.

13.5 Study Records Retention

Study records and reports must be maintained for a minimum of 2 years after a marketing application is approved for AV-1 for the indication for which it is being investigated; or, if no

application is to be filed or if the application is not approved for AV-1, until 2 years after the investigation is discontinued and the FDA has been notified. These documents will be retained for a longer period, however, if required by local regulations. Informed consent forms will be maintained as long as the sample/specimen exists.

No records will be destroyed without the written consent of the Sponsor. It is the responsibility of the Sponsor to inform the Investigator when these documents no longer need to be retained.

13.6 Protocol Deviations

The Investigator or designee must document and explain in the subject's source documentation any deviation from the approved protocol. The Investigator may implement a deviation from, or a change to, the protocol to eliminate an immediate hazard to study subjects without prior IRB approval. As soon as possible after such an occurrence, the implemented deviation or change, the reasons for it, and any proposed protocol amendments should be submitted to the IRB for review and approval, to the Sponsor Representative for agreement, and to the regulatory authorities, if required.

A protocol deviation is any change, divergence, or departure from the study design or procedures defined in the protocol. An important deviation (sometimes referred to as a major or significant deviation) is a subset of protocol deviations that leads to a subject being discontinued from the study, or significantly affects the subject's rights, safety, or well-being or the completeness, accuracy, and reliability of the study data. An important deviation can include nonadherence to inclusion or exclusion criteria or nonadherence to FDA regulations or ICH E6(R2) guidelines.

Protocol deviations will be documented by the clinical monitor throughout the course of monitoring visits. The Investigator will be notified in writing by the monitor of deviations. The IRB should be notified of all protocol deviations, if appropriate, in a timely manner.

14 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 and applicable standard operating procedures. PPD, 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, eCRFs, ICFs, medical and laboratory reports, and protocol and GCP compliance. Site monitors will have access to the clinical site, study site staff, and all study documentation according to the Sponsor-approved site monitoring plan. Study monitors will meet with the Investigator to discuss any problems and actions to be taken and will document clinical site visit findings and discussions.

15 PUBLICATION POLICY

Following completion of the study, the Sponsor is expected to publish the results of this research in a scientific journal. All Investigators funded by the NIH must submit or have submitted for them to the National Library of Medicine's PubMed Central (http://www.ncbi.nlm.nih.gov/pmc/) an electronic version of their final, peer-reviewed manuscripts upon acceptance for publication, to be made publicly available no later than 12 months after the official date of publication. The NIH Public Access Policy ensures the public has access to the published results of NIH funded research. It requires the Investigators to submit final peer-reviewed journal manuscripts that arise from NIH funds to the digital archive PubMed Central upon acceptance for publication. Further, the policy stipulates that these papers must be accessible to the public on PubMed Central no later than 12 months after publication.

Refer to:

- NIH Public Access Policy, http://publicaccess.nih.gov/
- NIH Office of Extramural Research Grants and Funding, http://grants.nih.gov/grants/oer.htm

As of January 2018, all clinical trials supported by the NIH must be registered on ClinicalTrials.gov, no later than 21 days after the enrollment of the first subject. Results of all clinical trials need to be submitted no later than 12 months following the primary completion date. A delay of up to 2 years is available for trials that meet certain criteria and have applied for certification of delayed posting.

As part of the result posting a copy of this protocol (and its amendments) and a copy of the statistical analysis plan will be posted on ClincialTrials.gov.

For this trial the responsible party is AbViro which will register the trial and post results.

The responsible party does not plan to request certification of delayed posting.

Refer to:

- Public Law 110-85, Section 801, Clinical Trial Databases
- 42CFR11
- NIH NOT-OD-16-149

16 LITERATURE REFERENCES

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17 APPENDICES

17.1 Appendix A. Schedule of Events

Phase	Screening	Check-in	Treatment Period											EOS/ ET
Procedure[a] / Day	–35 to –2	–1	1	2	3	5	8 (+2)	15 (±2)	22 (±2)	29 (±2)	43 (±3)	57 (±3)	85 (±5)	120 (±5)
Admission to clinic		X												
Discharge from clinic[b]						X								
Outpatient visit							X	X	X	X	X	X	X	X[c]
Informed consent	X													
Demographics	X													
Serology[d]	X													
Flavivirus serology test[e]	X													
Serum FSH[f]	X													
Inclusion/exclusion criteria	X	X												
Medical history[g]	X	X				X	X	X	X	X	X	X	X	X
Height, weight, and BMI[h]	X	X											X	X
Physical examination[i]	X	X				X	X	X	X	X	X	X	X	X
Vital sign measurements[j]	X	X	X	X	X	X	X	X	X		X		X	X
12-lead ECG[k]	X		X	X			X				X		X	X
Clinical laboratory testing[l]	X	X	X		X		X	X	X		X		X	X
Urine drug[m]	X	X							X		X		X	
Urine cotinine testing		X												
Pregnancy test[n]	X	X												
Randomization[o]			X											
AV-1 or placebo administration[p]			X											
PK blood sample collection[q]			X	X	X	X	X	X	X	X	X	X	X	X
Anti-AV-1 antibody blood sample collection		X								X			X	X
Hypersensitivity panel collection[r]		X	X	X										
Fasting period[s]		X	X											
Non-fasting period[t]				X	X	X								

Phase	Screening	Check-in	Treatment Period											EOS/ ET
Procedure[a] **Day**	**–35 to –2**	**–1**	**1**	**2**	**3**	**5**	**8 (+2)**	**15 (±2)**	**22 (±2)**	**29 (±2)**	**43 (±3)**	**57 (±3)**	**85 (±5)**	**120 (±5)**
Solicited AEs, Unsolicited AEs, and SAEs[u]			◄ X ►											
Prior/concomitant medications		◄ X ►												

Abbreviations: AEs, adverse events; BMI, body mass index; ECG, electrocardiogram; EOS, end of study; ET, early termination; FSH, follicle-stimulating hormone; IP, investigational product; PK, pharmacokinetic; QTcF, QT interval corrected for heart rate using Fridericia's formula; SAE, serious adverse event.

Notes:

[a] When procedures overlap or occur at the same time point, they should be performed in the following order: ECG, vital signs, then PK sampling which should be timed to occur last and as close to the scheduled time window as possible.

[b] Discharge from the clinical site on Day 5 after all study related activities are completed.

[c] The final follow-up visit will occur on Day 120 (approximately Week 17).

[d] Serology testing will include hepatitis B surface antigen, hepatitis C virus antibody, and human immunodeficiency virus antibody types 1 and 2.

[e] Flavivirus testing will include dengue, Zika, and West Nile viruses.

[f] Females only. A serum FSH test may be performed at Screening to confirm postmenopausal status along with at least 12 months of amenorrhea.

[g] Interim medical history will be done after initial thorough medical and medication history taken at Screening. Any new disorders after dosing on Day 1 will be recorded as an AE.

[h] Height and weight will be measured, and BMI will be calculated at Screening only. Only weight will be measured at Check-in (Day –1) and on Days 85 and 120.

[i] A full physical examination will be performed at Screening (skin, head, ears, eyes, nose, throat, neck, thyroid, lungs, heart, cardiovascular, abdomen, lymph nodes, and musculoskeletal system/extremities). Focused examination (lungs, cardiovascular, abdomen, skin) will be performed at Check-in (Day –1) and on Days 5, 8, and 15. Targeted, symptom-directed physical examination will be performed at all other time points.

[j] Vital signs will be measured at Screening and Check-in; within 60 minutes prior to start of the IP infusion; at 0.5, 1 (±10 minutes), 2, 4, 8, 24, and 48 hours after the start of the IP infusion; and on Days 5, 8, 15, 22, 43, 85, and 120. Vital signs will be measured after the subject has been in the seated or supine position for at least 5 minutes. Vital signs will include systolic and diastolic blood pressure, oral body temperature, heart rate, and respiratory rate.

[k] Single 12-lead ECG recordings will be made at Screening, within 60 minutes prior to the start of the IP infusion; at 1 (±10 minutes), 2, 4, 8, and 24 hours after the start of the IP infusion; and on Days 8, 43, 85, and 120. Electrocardiograms will be recorded after the subject has been in the supine position for at least 10 minutes. A single repeat measurement is permitted at Screening for eligibility determination. Measurements of the following intervals will be reported: RR interval, PR interval, QRS width, QT interval, and QTcF. Assessments should include comments on whether the tracings are normal or abnormal; rhythm; presence of arrhythmia or conduction defects; morphology; any evidence of myocardial infarction; or ST-segment, T-wave, and U-wave abnormalities.

[l] Clinical laboratory testing will occur at Screening, Check-in, predose (within 60 minutes) and 48 hours after the start of the IP infusion on Day 1; and on Days 8, 15, 22, 43, 85, and 120. A complete list of assessments is provided in Section 7.2. Blood and urine samples will be collected under fasted conditions and prepared per the clinic's standard procedures.

[m] Urine drug screen will occur at Screening and Check-in and on Days 22, 43, and 85 per the clinic's standard procedures and per Section 7.2.

(n) Women of childbearing potential only. A serum pregnancy test will be performed at Screening and Check-in, and tests must be negative prior to enrollment and dosing, respectively.
(o) After verification of eligibility, subjects will be randomly assigned before dosing on Day 1.
(p) The start of the IP infusion will be called "0" hour and is denoted with grey shading. All doses of the IP will be administered intravenously over a period of 60 minutes. Subjects will maintain an upright (ie, seated or standing) position for at least 4 hours after the end of infusion.
(q) Blood samples for PK analysis of AV-1 in serum will be collected predose (within 15 minutes); at 0.5, 1 (end of infusion), 1.25, 1.5, 2, 3, 4, 6, 8, 12, 24, and 48 hours after the start of infusion, Day 5 prior to discharge, and at the follow-up visits on Days 8, 15, 22, 29, 43, 57, 85, and 120 (or ET).
(r) A blood sample will be drawn on Day –1 and tested only if a suspected hypersensitivity reaction occurs during or following infusion on Day 1 at the discretion of the Investigator in consultation with the Sponsor. At the discretion of the Investigator, an additional blood sample will be drawn as soon as possible after the event occurs (Day 1) and again the following day (Day 2).
(s) During fasting periods, subjects should have nothing to eat or drink except water from 10 hours prior to start of the IP infusion until 4 hours after the end of infusion.
(t) During non-fasting periods when subjects are at the clinical site, subjects should receive standardized meals per the clinic's standard procedures that are scheduled at the same time in each treatment period during the study.
(u) Solicited AEs will be assessed from the start of IP infusion until Day 8 and unsolicited AEs will be assessed from the start of IP infusion through the final follow-up visit on Day 120. All AEs should be followed until they are resolved, stable, or judged by the Investigator to be not clinically significant.

17.2 Appendix B. Toxicity Tables

Table 17-1 Toxicity Grading Scales for Clinical Adverse Events and Reactions

Cardiovascular	**Mild (Grade 1)**	**Moderate (Grade 2)**	**Severe (Grade 3)**
Arrhythmia	N/A	Asymptomatic; transient signs; no medical intervention required	Recurrent or persistent symptoms; medical intervention required
Hemorrhage, Blood Loss*	Estimated blood loss is <100 mL	Estimated blood loss is ≥100 mL; no transfusion required	Transfusion required
QTc (Fridericia's correction)	Asymptomatic, QT interval 450 to 479 msec (male) and 460 to 479 msec (female) OR increase in interval <30 msec above baseline	Asymptomatic, QTc interval 480 to 499 msec OR increase in interval 30 to 50 msec above baseline	Asymptomatic, QTc interval ≥500 msec OR increase in interval ≥60 msec above baseline
PR Interval (prolonged)	PR interval 0.21 to 0.25 sec	PR interval >0.25 sec	Type II 2nd degree AV block OR ventricular pause >3 sec
Respiratory	**Mild (Grade 1)**	**Moderate (Grade 2)**	**Severe (Grade 3)**
Cough	Transient; no treatment	Persistent	Interferes with daily activities
Bronchospasm, Acute	Transient; no treatment; forced expiratory volume 71 to 80% of predicted peak flow	Requires medical intervention; normalizes with bronchodilator; forced expiratory volume 60 to 70% of predicted peak flow	No normalization with bronchodilator; forced expiratory volume <60% of predicted 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	**Mild (Grade 1)**	**Moderate (Grade 2)**	**Severe (Grade 3)**
Nausea	No interference with activity	Some interference with activity	Prevents daily activity
Vomiting	No interference with activity OR 1 to 2 episodes/24 hours	Some interference with activity OR >2 episodes/24 hours	Prevents daily activity OR requires IV hydration OR requires medical intervention
Diarrhea	2 to 3 loose or watery stools OR <400 grams/24 hours	4 to 5 loose or watery stools OR 400 to 800 grams/24 hours	6 or more loose or watery stools OR >800 grams/24 hours OR requires IV hydration OR requires medical intervention
Local Reactions	**Mild (Grade 1)**	**Moderate (Grade 2)**	**Severe (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 or mild discomfort at rest	Significant discomfort at rest
Erythema/Redness**	2.5 to 5 cm	5.1 to 10 cm	>10 cm

Induration/Swelling***	2.5 to 5 cm AND does not interfere with activity	5.1 to 10 cm OR interferes with activity	>10 cm OR prevents daily activity
Systemic Reactions	**Mild (Grade 1)**	**Moderate (Grade 2)**	**Severe (Grade 3)**
Allergic Reaction	Pruritus without rash	Localized urticaria OR requires oral therapy	Generalized urticaria; angioedema OR anaphylaxis OR requires epinephrine
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 OR requires triptans
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
Systemic Illness	**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

*Does not include blood loss from study.
**The local reaction should be measured at the greatest single diameter and recorded as a continuous variable in addition to grading.
***Induration/swelling should be evaluated and graded using the functional scale as well as the actual measurement.

Table 17-2 Vital Signs Toxicity Grading

Vital Signs*	Mild (Grade 1)	Moderate (Grade 2)	Severe (Grade 3)
Fever (°C) ** (°F) **	38.0 to 38.4 100.4 to 101.1	38.5 to 38.9 101.2 to 102.0	39.0 to 40 102.1 to 104
Tachycardia - beats per minute	101 to 115	116 to 130	>130
Bradycardia - beats per minute***	50 to 54	45 to 49	<45
Hypertension (systolic) - mm Hg	141 to 150	151 to 155	>155
Hypertension (diastolic) - mm Hg	91 to 95	96 to 100	>100
Hypotension (systolic) – mm Hg	85 to 89	80 to 84	<80
Respiratory rate – breaths per minute	17 to 20	21 to 25	>25

*Subject should be at rest for all vital sign measurements.
**Oral temperature; no recent hot or cold beverages or smoking.
***When resting heart rate is between 60 to 100 beats per minute. Use clinical judgement when characterizing bradycardia among some healthy subject populations, for example, conditioned athletes.

Table 17-3 Laboratory Toxicity Grading

Laboratory Tests	Mild (Grade 1)	Moderate (Grade 2)	Severe (Grade 3)
Liver Function Tests – ALT increase by factor	1.1 – 2.5 × ULN	2.6 – 5.0 × ULN	>5.0 × ULN
Liver Function Tests – AST increase by factor	1.1 – 2.5 × ULN	2.6 – 5.0 × ULN	>5.0 × ULN
Albumin – Hypoalbuminemia g/dL	2.8 – 3.1	2.5 – 2.7	<2.5
Alkaline Phosphatase – ALP increase by factor	1.1 – 2.0 × ULN	>2.0 – 3.0 × ULN	>3.0 × ULN
Anion Gap* – mEq/L	N/A	N/A	N/A
Bilirubin (total) – mg/dL	1.1 – <1.6 × ULN	1.6 – < 2.6 × ULN	≥ 2.6 × ULN
Blood Urea Nitrogen mg/dL	27 – 30	31 – 35	> 35
Calcium – Hypocalcemia mg/dL	8.0 – 8.4	7.5 – 7.9	7.0 – 7.4
Calcium – Hypercalcemia mg/dL	10.5 – 11.0	11.1 – 11.5	11.6 – 12.0
Creatinine – mg/dL	1.2 – 1.3 × ULN	>1.3 – 1.8 × ULN	>1.8 × ULN
Glucose (fasted) – Hypoglycemia mg/dL	50 – 54	40 – 49	30 – 39
Glucose (fasted) – Hyperglycemia Fasting – mg/dL	117 – 127	128 – 142	>143
Potassium – Hypokalemia mEq/L	3.3 – 3.4	3.1 – 3.2	2.9 – 3.0
Potassium – Hyperkalemia mEq/L	5.1 – 5.2	5.3 – 5.4	5.5 – 5.6
Sodium – Hyponatremia mEq/L	129 – 131	127 – 128	122 – 126
Sodium – Hypernatremia mEq/L	146 – 147	148 – 149	150 – 152
Hematology			
WBC increase - cell/mm^3	9,800 – 14,000	14,001 – 19,000	19,001 – 24,000
WBC decrease - cell/mm^3	2,000 – 3,099	1,000 – 1,999	500 – 999
Absolute Neutrophil Count - cell/mm^3	1,000 – 1.499	500 – 999	0 – 499
Hemoglobin Decrease (female) - gm/dL	9.7 – 10.7	8.2 – 9.6	6.7 – 8.1
Hemoglobin Decrease (male) - gm/dL	11.7 – 12.7	9.76 – 11.6	7.6 – 9.6
Platelets Decrease - cell/mm^3	125,000 – 139,000	100,000 – 124,000	<100,000
Coagulation			
Prothrombin Time – increase by factor	>1.0 – 1.2 × ULN	1.2 – 1.4 × ULN	>1.4 × ULN
Activated Partial thromboplastin time – increase by factor	>1.0 – 1.2 × ULN	1.2 – 1.4 × ULN	>1.4 × ULN

Laboratory Tests	Mild (Grade 1)	Moderate (Grade 2)	Severe (Grade 3)
Urine			
Blood**	1+	2+	>2+
Glucose	1+	2+	>2+
Protein	1+	2+	>2+

Abbreviations: ALP, alkaline phosphatase; ALT, alanine transferase; AST, aspartate aminotransferase; ULN, upper limit of the normal range, WBC, white blood cell.

*Only values above the ULN and below the LLN are considered abnormal.

**For females: In case of menstruation, screening urinalysis must be postponed, but a result should be available prior to Day 1. Hematuria is acceptable for females in menses for the purposes of evaluating study halting criteria and dose escalation.

Source: DHHS 2007.

Table 17-4 Infusion-related Reactions and Toxicity Grading

Mild (Grade 1)	Moderate (Grade 2)	Severe (Grade 3)
Mild transient reaction; infusion interruption not indicated; intervention not indicated.	Therapy or infusion interruption indicated but responds promptly to symptomatic treatment (eg, antihistamines, nonsteroidal anti-inflammatory drugs, narcotics [opioids], intravenous fluids); prophylactic medication indicated for less than or equal to 24 hours.	Prolonged (eg, not rapidly responsive to symptomatic medication and/or brief interruption of infusion); recurrence of symptoms following initial improvement; hospitalization indicated for other clinical sequelae.

Source: Adapted from the National Cancer Institute Common Terminology Criteria for Adverse Events (Version 5.0) for infusion-related reactions.