A Phase 1, Dosage-Escalation Study of the Safety and Immunogenicity of a Novel Rabies Vaccine ChAd155-RG vs. the Comparator RABAVERT Vaccine in Healthy Adult Subjects

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STATEMENT OF COMPLIANCE

This trial will be carried out in accordance with Good Clinical Practice (GCP) and as required by:

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 (Institutional Review Boards), 21 CFR Part 11 (Electronic Records and Electronic Signatures), 21 CFR Part 312 (Investigational New Drug Application), and 21 CFR 812 (Investigational Device Exemptions)

International Conference on Harmonisation (ICH) E6 GCP; 62 Federal Register 25691 (1997); and future revisions

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 (NIAID) 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 this protocol, including all statements regarding confidentiality, and according to local legal and regulatory requirements and applicable US federal regulations and ICH E6 GCP guidelines.

I agree to conduct this trial in compliance with GCP and applicable regulatory requirements.

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

Site Principal Investigator Signature:

Signed: _____ Date: _____

Name

Title

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LIST OF ABBREVIATIONS

Ad	Adenovirus
AE	Adverse Event/Adverse Experience
ALT	Alanine Transaminase
aPTT	Activated Partial Thromboplastin Time
ASC	Antibody Secreting Cell
AST	Aspartate Transaminase
BMI	Body Mass Index
BUN	Blood Urea Nitrogen
CFR	Code of Federal Regulations
CFSE	Carboxyfluorescein Diacetate Succinimidyl Ester
ChAd	Chimpanzee Adenovirus
CMS	Clinical Materials Services
COI	Conflict of Interest
COVID-19	Coronavirus Disease 2019
CPM	Clinical Project Manager
CROMS	Clinical Research Operations and Management Support
CSR	Clinical Study Report
DHHS	Department of Health and Human Services
DMID	Division of Microbiology and Infectious Diseases, NIAID, NIH, DHHS
eCRF	Electronic Case Report Form
ELISA	Enzyme-linked Immuno-sorbent Assay
ELISPOT	Enzyme-linked Immunospot
FDA	Food and Drug Administration

FWA	Federal Wide Assurance
GCP	Good Clinical Practice
GMT	Geometric Mean Titer
HCV	Hepatitis C Virus
HIPAA	Health Insurance Portability and Accountability Act
IB	Investigator's Brochure
ICF	Informed Consent Form
ICH	International Conference on Harmonisation
ICMJE	International Committee of Medical Journal Editors
ICS	Intracellular Cytokine Staining
IDES	Internet Data Entry System
IEC	Independent or Institutional Ethics Committee
IM	Intramuscular
IND	Investigational New Drug Application
IRB	Institutional Review Board
ISM	Independent Safety Monitor
ITT	Intention-to-Treat
IU	International Unit
IV	Intravenous
MOP	Manual of Procedures
N	Number (typically refers to subjects)
NDA	New Drug Application
NHP	Non-Human Primate
NIAID	National Institute of Allergy and Infectious Diseases, NIH, DHHS
NIH	National Institutes of Health
OCRR	Office of Clinical Research Resources

OHRP	Office for Human Research Protections
OHSR	Office for Human Subjects Research
PEP	Post-Exposure Prophylaxis
PHI	Protected Health Information
PI	Principal Investigator
PrEP	Pre-Exposure Prophylaxis
PT	Prothrombin Time
QA	Quality Assurance
QC	Quality Control
RFFIT	Rapid Fluorescence Focus Inhibition Test
RG	Rabies Protein G
RIG	Rabies Immunoglobulin
RSV	Respiratory Syncytial Virus
SAE	Serious Adverse Event/Serious Adverse Experience
SAP	Statistical Analysis Plan
SARS-CoV-2	Severe Acute Respiratory Syndrome Coronavirus 2
SDCC	Statistical and Data Coordinating Center
SMC	Safety Monitoring Committee
SOC	System Organ Class
SOP	Standard Operating Procedure
ULN	Upper Limit of Normal
US	United States
VNA	Virus neutralizing antibody
VP	Viral Particle
VTEU	Vaccine and Treatment Evaluation Unit
WBC	White Blood Cell

WHO	World Health Organization

PROTOCOL SUMMARY

Title:	A Phase 1, Dosage-Escalation Study of the Safety and Immunogenicity of a Novel Rabies Vaccine ChAd155-RG vs. the Comparator RABAVERT Vaccine in Healthy Adult Subjects
Phase:	1
Population:	This trial will enroll 50 rabies- and adenovirus vaccine-naive healthy male and non-pregnant female subjects aged 18 to 49 years, and will be conducted in the US.
Number of Sites:	1 (Emory University VTEU)
Study Duration:	This trial is expected to take approximately 48 months to complete, from initiation through availability of a final report on the primary and secondary outcomes of safety and rabies virus neutralizing antibody (VNA) responses.
Estimated Time to Complete Enrollment:	Enrollment is expected to take up to 18 months from study initiation.
Subject Participation Duration:	The duration of each subject's participation is approximately 13 months, from recruitment through the last study visit.
Description of Agent or Intervention:	The investigational product is ChAd155-RG Vaccine, administered in one or two doses, and the commercially available, active comparator is RABAVERT Vaccine, administered in three doses, by the intramuscular (IM) route. All subjects will receive one or more doses of an experimental or licensed vaccine.
Objectives	Primary • Assessment of the safety, tolerability, and reactogenicity of one dose of ChAd155-RG at $5x10^{10}$vp per dose, or one or two doses of ChAd155-RG at $1x10^{11}$vp per dose.

- Comparison of the safety, tolerability, and reactogenicity of one or two doses of ChAd155-RG, with three doses of RABAVERT.

Secondary

- Assessment of serum rabies VNA levels by a standard, WHO-approved, RFFIT, as assessed by immune response kinetics (through approximately 12 months after first dose of vaccine), seroconversion rates, and peak GMT in each treatment arm.

Exploratory (if applicable)

- Quantification of circulating T-cell responses against RG by ex vivo IFN-gamma ELISpot.
- Characterization of circulating CD4 and CD8 T-cell responses against RG at peak time point by multiparameter ICS.
- Comparison of durability of immune response to ChAd155-RG and RABAVERT at approximately 3, 6, and 12 months after first dose of vaccine by each treatment arm tested.
- Measurement of RG-specific memory B cells by ELISpot.
    - Measurement of an antibody response to the vaccine vector (ChAd155).

Outcome Measures:

Primary

- Frequency and severity of solicited injection site and systemic reactogenicity events from the time of each vaccination through Day 7 after each vaccination, in each treatment arm and overall.
- Frequency and severity of SAEs considered study vaccine-related and reported at any time after the first vaccination through the end of the study, in each treatment arm and overall.

- Frequency and severity of study vaccine-related lab AEs through Day 22 from the time of the first vaccination , in each treatment arm and overall.

- Frequency and severity of unsolicited study vaccine-related AEs from the time of the first vaccination through Day 28 after the last vaccination, in each treatment arm and overall.

- Number of subjects with new onset of a chronic medical condition at any time after the first vaccination, in each treatment arm and overall.

- Frequency and severity of any SAEs at any time after the first vaccination through the end of the study, in each treatment arm and overall.

Secondary

- Proportion of subjects seroconverting to rabies virus at each antibody time point (seroconversion is defined as VNA concentration ≥0.5 IU/mL), in each treatment arm and overall.

- GMT (as measured by rabies VNA using a standard, WHO-approved, RFFIT) at each antibody time point within each treatment arm.

- Peak GMT (as measured by rabies VNA using a standard, WHO-approved, RFFIT; peak GMT is defined as highest GMT measured across all post-vaccination antibody time points) within each treatment arm.

Exploratory

- Frequencies of circulating T cell responses against RG by ex vivo IFN-ELISpot.

- Characteristics of circulating CD4 and CD8 T cell responses against RG by multiparameter ICS.

- Magnitude and durability of RG-specific memory B cells by ELISpot.
    - GMT to ChAd155 vector at one month after first vaccination

Description of Study Design:

This is a single-center, observer-blinded, Phase 1, dosage-escalation trial to evaluate the safety, tolerability, reactogenicity, and immunogenicity of ChAd155-RG compared with RABAVERT in rabies virus-naïve healthy male and non-pregnant female adult subjects. There are 4 dose groups (Table 1).

Subjects who have never received a licensed or investigational rabies virus vaccine, or an Adenovirus (Ad)-based investigational vaccine, and who have never been exposed to a rabid animal will be eligible for enrollment.

Since this is a dosage-escalation study, sentinel subjects will be used at each dosage level. The first four subjects will be randomized to Group A or D and will receive either a single dose of ChAd155-RG at the lower dosage ($5x10^{10}$vp, N=3) (followed by placebo on Days 8, 15, and 22) or RABAVERT (N=1) (on Days 1, 8, and 22, and placebo on Day 15) (see Table 1). These subjects will be monitored for safety, tolerability, and reactogenicity through day 7 after first study vaccination, and if no pre-defined halting rule is met (Section 9), then four additional sentinel subjects will be randomized to Group B or D and will receive either a single dose of ChAd155-RG at the higher dosage ($1x10^{11}$vp, N=3) (followed by placebo on Days 8, 15, and 22) or RABAVERT (N=1) (on Days 1, 8, and 22, and placebo on Day 15) (see Table 1). These subjects will be monitored for safety, tolerability, and reactogenicity through day 7 after first study vaccination. The Safety Monitoring Committee (SMC) will then review the available safety, reactogenicity, AE, and lab data of all the sentinel subjects, and will decide if the remaining non-sentinel subjects should be enrolled.

If the SMC recommends proceeding with the remainder of trial enrollment, eligible individuals will be enrolled and randomized to one of the four dose groups (Groups A-D). To maintain

blinding of study personnel conducting surveillance and assessment of AEs, subjects in these groups will receive 4 sequential injections, 1 ml per injection, in alternating arms, on the same schedule:

- Subjects randomized to Group A will receive ChAd155-RG at the lower dosage ($5x10^{10}$vp) on Day 1, then placebo injections on Days 8, 15, and 22
- Subjects randomized to Group B will receive ChAd155-RG at the higher dosage ($1x10^{11}$vp) on Day 1, then placebo injections on Days 8, 15, and 22
- Subjects randomized to Group C will receive ChAd155-RG at the higher dosage ($1x10^{11}$vp) on Days 1 and 15, and placebo injections on Days 8 and 22
- Subjects randomized to Group D will receive RABAVERT at the standard dose (1 mL) on Days 1, 8, and 22, and a placebo injection on Day 15

Vaccine preparation and administration will be unblinded. The investigational vaccine doses will be prepared just prior to administration to ensure that the volume of each injection is identical between Groups A-D. Follow-up will be double-blinded, and the SMC will review the available safety, reactogenicity, AE, and lab data collected from all subjects for 7 days after the last vaccination and as needed.

Subjects will be followed using a memory aid, face-to-face scheduled clinic visits, and safety lab monitoring as outlined in Section 7 and Appendix A. Following enrollment and vaccination, safety lab blood samples will be collected on Days 2, 8, 16, and 22. Blood samples for immunologic assays will be collected on Days 1, 2, 8, 15, 16, 22, 29, 91, 181, and 381. The duration of each subject's participation will be approximately 13 months, from recruitment to collection of data for safety and immunogenicity outcomes.

This trial is expected to take approximately 30 months to complete, from initiation through availability of a final report on the primary outcomes of safety and the secondary outcomes of VNA levels to RG. Exploratory outcomes are discussed in an

addendum. A planned interim safety, reactogenicity, and immunogenicity analysis is detailed in Section 11.3.

Table 1: Treatment Arms

Study Group	N	Vaccine	Dosage of Vaccine	Number of Vaccine Doses	Schedule of Vaccine and Placebo Injections	Route	Description of Study Group
A	14 including 3 sentinels[#]	ChAd155-RG	$5x10^{10}$vp*	1	Day 1 vaccine; Days 8, 15, and 22 placebo	IM	Lower dosage, 1 dose
B	14 including 3 sentinels	ChAd155-RG	$1x10^{11}$vp	1	Day 1 vaccine; Days 8, 15, and 22 placebo	IM	Higher dosage, 1 dose
C	10	ChAd155-RG	$1x10^{11}$vp	2	Days 1 and 15 vaccine; Days 8 and 22 placebo	IM	Higher dosage, 2 doses
D	12 including 2 sentinels	RABA-VERT	1 mL	3	Days 1, 8, and 22 vaccine; Day 15 placebo	IM	Active comparator (licensed vaccine)

*Abbreviations: vp, viral particles; IM, intramuscular

[#]Groups A, B and D each include sentinel subjects as indicated. Note: Two of the sentinels will receive a three-dose series of RABAVERT. To maintain blinding, the other sentinels will receive placebo injections on Days 8, 15, and 22 as indicated in the table.

1. KEY ROLES

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2. BACKGROUND INFORMATION AND SCIENTIFIC RATIONALE

2.1. Background Information

Rabies is an acute, progressive viral encephalitis caused by neurotropic single-stranded RNA viruses (genus Lyssavirus, family Rhabdoviridae). This zoonotic infection is acquired after exposure to the saliva of infected domestic or wild mammals and is almost invariably fatal. Rabies is estimated to cause more than 50,000 deaths annually, mostly in Asia and Africa, but the true disease burden is likely higher due to underreporting and lack of confirmatory testing in many endemic regions.[1, 2]

Highly effective, inactivated viral vaccines against rabies have been available for use as pre-exposure prophylaxis (PrEP) or post-exposure prophylaxis (PEP) for decades. In the US, two vaccine formulations are currently available, a purified chick embryo cell vaccine (PCECV; RabAvert, GSK Vaccines), and a human diploid cell vaccine (HDCV; Imodex, Sanofi Pasteur).[3, 4] It is recommended that both vaccines be administered in a three-dose series (Days 0, 7, and 21 or 28)[3] for PrEP, and a four-dose series (Days 0, 3, 7, and 14; a fifth dose on Day 28 is recommended for immunocompromised individuals) for PEP, with or without rabies immunoglobulin (RIG) depending on the severity of the exposure.[4] Virtually all recipients of a recommended vaccine series mount a seroprotective titer of rabies VNA, defined as a post-vaccination VNA concentratin ≥0.5 IU/mL, or complete virus neutralization at a 1:5 serum dilution, by a WHO-approved RFFIT.

Despite the existence of effective vaccines, rabies remains endemic throughout the world. This is largely because RIG and/or rabies vaccines are not readily available, especially in countries with a high animal rabies burden, and because the recommended schedule of vaccine administration is cumbersome and cost-prohibitive. Even in the US, compliance with the recommended PEP vaccine series is suboptimal; for example, in one prospective study of rabies PEP, over one-third of individuals failed to complete the PEP vaccine series at 1 month.[5] Thus, novel vaccines against rabies that are safe, tolerable, economical, and as immunogenic as current formulations, and that can be administered for PrEP in one dose, and for PEP in one or two doses, are urgently needed.

Modified live viral-vectored rabies vaccines – given as a single dose – have been used successfully in animals for decades; thus, a similar approach could be applied against human rabies. For example, RG-canarypox virus and RG-vaccinia virus vaccines are currently in use in the US for vaccination of domestic cats and wildlife, respectively.[6] In humans, replication-defective chimpanzee adenovirus (ChAd) vectors are being increasingly recognized as attractive vaccine vector candidates.[7] Given the lower probability of pre-existing immunity to the ChAd vector in humans, ChAd-vectored vaccines have the potential to elicit high-quality, durable

immune responses.[8] In fact, safe and immunogenic ChAd-vectored human vaccines have already been described for Ebola,[9-12] malaria,[13] hepatitis C virus (HCV),[14] and respiratory syncytial virus (RSV).[15]

Ad vaccine carriers have also been shown to be effective for rabies, but only in animals. For example, in Canada, an oral rabies vaccine (ORV) consisting of replication-competent recombinant human adenovirus type 5 (Ad5) expressing RG within an E3 deletion (referred to as AdRG1.3,[16] or ONRAB®) is licensed for use as vaccine bait in animal rabies control efforts,[17] and has also been evaluated in multiple US field trials.[18-20] More recently, Xiang *et al*. described a novel rabies vaccine consisting of replication-defective recombinant ChAd vector serotype SAd-V24 (referred to as Ad68C) expressing RG using a non-human primate (NHP) model.[21] Notably, after only a single IM injection in this model, this vaccine elicited sustained levels of seroprotective VNAs through 21 months post-vaccination, and provided 100% protection from lethal rabies virus challenge at 22 months post vaccination.[21]

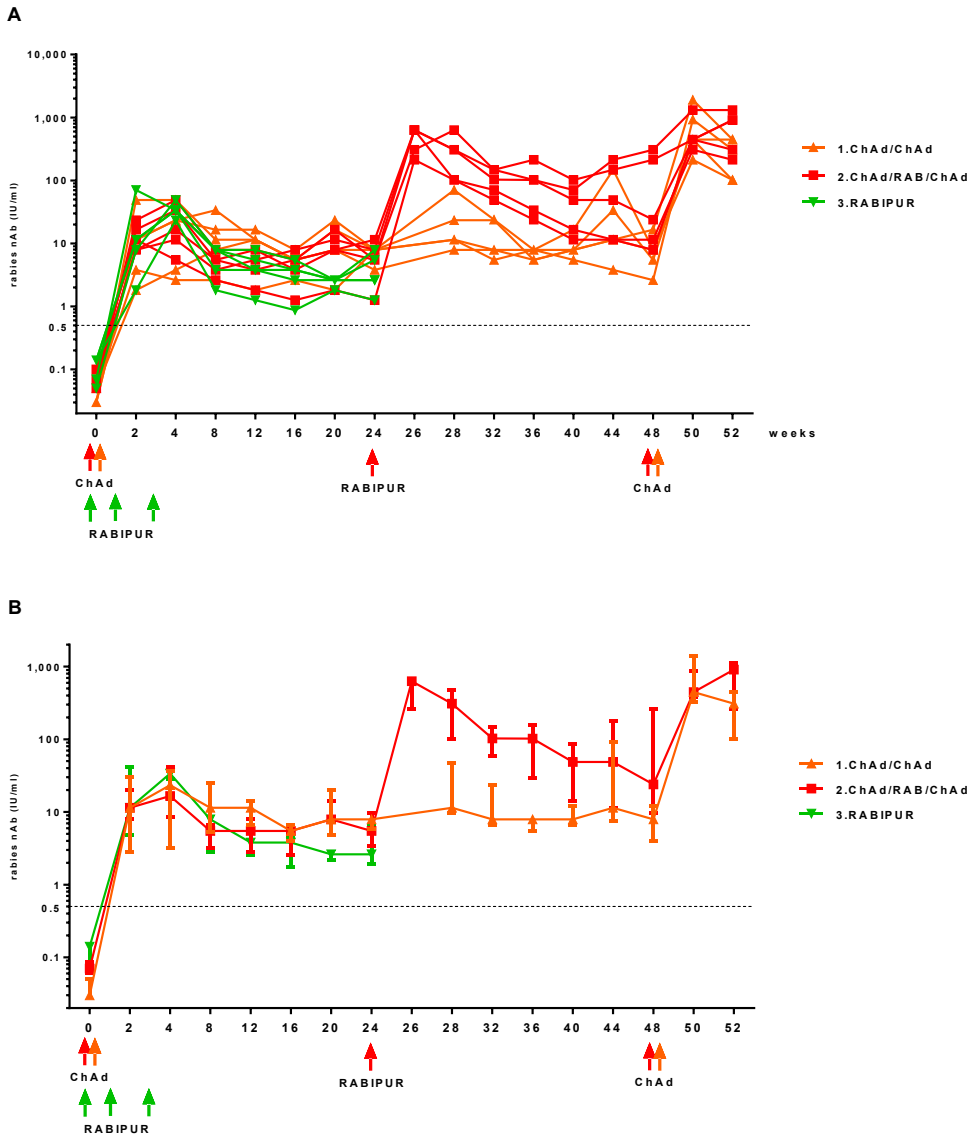
ReiThera has recently developed a novel rabies vaccine aimed at extending these findings into humans. The vaccine, ChAd155-RG, consists of a replication-defective group C ChAd 155 (ChAd155) expressing RG under the control of the CMV promoter. Group C Ad vectors (including human Ad5 and ChAd3) have previously been shown to be safe, tolerable, and highly immunogenic in humans.[9-12, 14, 15] A recombinant RSV-ChAd155 vaccine has also been tested successfully in a Phase 1 clinical trial.[22] To increase the cross-protective breadth of ChAd155-RG, the RG sequence is a medoid, a natural viral strain with the highest average percent of amino acid identity among all RG sequences annotated in NCBI's database. The selected RG protein (NCBI strain AGN94271) shares an average 94% percent identity to the RG proteins of current vaccines.

In preclinical studies in mice, ChAd155-RG elicited seroprotective VNA concentrations (≥0.5 IU/mL) over a wide dosage range and conferred protection against a lethal challenge with a rabies virus strain (unpublished data). Immunogenicity studies were subsequently conducted in NHP models with the aim of assessing up to 1 year the kinetics, breadth, and longevity of VNA titers induced by a single IM immunization with ChAd155-RG compared with three doses (Days 0, 7, and 21) of a comparator cell culture-derived vaccine (CCV), RABIPUR (sold in the US as RABAVERT). In these experiments, three groups of five cynomolgus monkeys (*Macaca fascicularis*) received rabies vaccine: Groups 1 and 2 received a single dose of $5x10^{10}$vp ChAd155-RG at Day 0, whereas Group 3 received three doses of RABIPUR at Days 0, 7, and 21. For both vaccines, half of the intended human dosage was used. Group 2 monkeys subsequently received one dose of RABIPUR 24 weeks after the first vaccination to analyze the capacity of VNA titers induced by the vectored vaccine to be boosted by a CCV vaccine. Near the end of the study at week 48 after the first vaccination, all animals in Groups 1 and 2 received a final $5x10^{10}$vp booster dose of ChAd155-RG as a recall vaccination.

The results (Figure 1) showed that a single IM immunization with ChAd155-RG elicited VNA titers above the protective threshold in all 10 animals in Groups 1 and 2 by just two weeks after vaccination. Titers peaked between week 2 and 4 post-vaccination and were of comparable magnitude to those induced by three vaccinations with RABIPUR. VNA kinetics and levels remained similar between

ChAd155-RG and RABIPUR vaccinated animals over 6 months of follow up. VNA concentrations were stable and above 0.5 IU/mL up to week 48 (~1 year) after a single ChAd155-RG administration in Group 1 animals.

Figure 1: Rabies VNA titer kinetics in NHP



Rabies vaccine immunogenicity study in NHPs. Groups of 5 cynomolgus monkeys were vaccinated as highlighted by arrows on the x-axis with half of the human dose of ChAd155-RG vector (Groups 1 and 2) and RABIPUR, injected in the muscle. Rabies VNA titers were measured with a Fluorescent Antibody

Virus Neutralization (FAVN) assay (at IDEXX; Germany) and followed starting at 2 weeks post vaccination and then monthly. A: Data for individual animals, B: Group median +/- IQR

The RABIPUR boost in Group 2 animals at week 24 was highly efficient in raising VNA titers even above the peak level achieved with the ChAd155-RG prime, suggesting compatibility of the two rabies vaccines. The boost with ChAd155-RG at week 48 was also highly effective: VNA titers increased well above peak post prime in Group 1, similar to what was seen in Group 2 upon RABIPUR boost, highlighting the possibility and potential efficacy of re-administering the candidate ChAd155-RG vaccine.

Together, these preclinical data demonstrate the potential for ChAd155-RG to be a promising novel rabies vaccine candidate in humans, and thus warrant further evaluation in a Phase 1 clinical trial.

2.2 Rationale

This trial will assess the safety, tolerability, and reactogenicity of ChAd155-RG in comparison with RABAVERT. This trial will also determine whether the immunogenicity data generated in NHP studies can be replicated in a clinical setting, using a standard WHO-approved RFFIT to measure VNA responses, and evaluate the durability of the immune response to ChAd155-RG in comparison with RABAVERT up to 1 year after vaccination.

The ChAd155-RG dosage and regimen selected for this trial are based on past experience with ChAd-vectored vaccines, non-clinical data from ChAd155-RG immunizations in NHPs, as well as a desire to evaluate this novel vaccine as rabies PrEP (one or two doses) in comparison with commercially available RABAVERT as PrEP (recommended three doses).

Finally, exploratory endpoint assays will be performed to broadly characterize the short- and long-term immune responses to ChAd155-RG compared with RABAVERT.

2.3 Potential Risks and Benefits

2.3.1. Potential Risks

This is a Phase 1 dosage-escalation study of the investigational ChAd155-RG Vaccine. The potential risks of this trial are those related to ChAd155-RG, the comparator RABAVERT Vaccine, having blood drawn, IM injection, other risks, and breach of confidentiality.

RABAVERT

RABAVERT was licensed by the FDA for use as rabies PrEP or PEP in 1997. A comprehensive description of the risks of AEs are described in the package insert.[23] Notable potential risks related to RABAVERT administration are the potential occurrences of severe rare adverse neurological complications that have been previously reported in temporal association with vaccine administration. These include meningitis, encephalitis, neuroparalysis, transient

paralysis, Guillain-Barre syndrome, myelitis, retrobulbar neuritis, multiple sclerosis, vertigo, and visual disturbances.[23, 24] An analysis of post-marketing surveillance data from 1997 to 2005 from the US Vaccine Adverse Event Reporting System (VAERS) reported 336 AEs for 1.1 million RABAVERT doses administered (0.03% of all doses). Of these, 13 were neurological events (4% of all the AEs), but there was no pattern in the type or time of onset of symptoms to indicate a plausible association with the vaccine.[25] RABAVERT is reactogenic in healthy people, causing local reactions (pain, induration, erythema), lymphadenopathy, fever, myalgias, arthralgias, dizziness, nausea, headache, and rash. The following adverse reactions have been identified during post approval use of RabAvert: presyncope, syncope, palpitations, hot flush, extensive limb swelling and angioedema. Once reconstituted, RABAVERT contains processed bovine gelatin and trace amounts of chicken protein, neomycin, chlortetracycline, and amphotericin B; therefore, individuals with a history of allergy or hypersensitivity to these ingredients should be monitored for the possibility of allergic reactions following vaccination.

ChAd Vector

Administration of ChAd vectors can be associated with local reactions (pain, tenderness, redness, and swelling) and systemic symptoms including fatigue, myalgias, and arthralgias, or flu-like symptoms such as fever, chills, and headache.

In recent clinical trials of Ebola virus vaccine candidates using ChAd3 as the vaccine vector (a group C Ad, like ChAd155), there were no vaccine-related SAEs. Most AEs were mild (Grades 1-3) and self-limited, resolving within 48 hours after vaccination. The most common systemic AEs were fatigue and headache. All abnormal lab values recorded during these studies were Grade 1, with the most common abnormalities being transient lymphopenia, anemia, or thrombocytopenia that developed on Day 1 after vaccination, and which had resolved by Day 7 after vaccination.[9-12] Similar findings were reported for a novel ChAd3-vectored HCV vaccine,[14] as well as multiple malaria vaccine candidates that used ChAd63 (a group E adenovirus) as the vaccine carrier.[13] A small number of participants in each of the ChAd3 Ebola vaccine studies (<5 in each study) developed asymptomatic prolongation of the activated partial thromboplastin time (aPTT) which resolved by the end of the follow-up period.[9-12] In one of the trials, further investigation of the prolonged aPTT revealed induction of an antiphospholipid antibody.[12]

The ChAd155 vector being used in this trial has been previously tested in humans in a Phase 1 trial of a novel RSV vaccine candidate.[22] The vaccine dosages used in that study (low-dosage, $5x10^9$vp; high-dosage, $5x10^{10}$vp) were half of those proposed in this trial. Per the investigator's brochure for ChAd155-RG, in that previous study, there were no vaccine-related SAEs, and the most common AEs were local injection site pain (100% of vaccine recipients vs. 5.3% of placebo recipients), fatigue (50% of high-dosage ($5x10^{10}$vp) vaccine recipients, only 1 subject or 3% with Grade 3 fatigue) and headache (34.5% of high-dosage vaccine recipients).

In recent studies of COVID-19 vaccines that use an adenovirus vector, there were a small number of individuals who experienced rare but serious thrombotic events, including cerebral

venous sinus thrombosis, associated with thrombocytopenia. These events occurred after the Oxford-AstraZeneca COVID-19 vaccine, which uses a chimpanzee group E adenovirus vector (ChAdOx1), and the Johnson and Johnson/Janssen Biotech Inc's COVID-19 vaccine, which uses a human group D adenovirus vector (Ad26). The exact mechanism of this vaccine-induced thrombotic thrombocytopenia is not fully understood, and may or may not be related to the adenovirus vectors used in these vaccines. There have been no thrombotic events identified in published studies of vaccines based on group C adenoviruses (similar to ChAd155), though the number of subjects in these studies was significantly smaller than the number of people who have received an adenovirus-vector based COVID-19 vaccine.

Allergic Reaction

Acute and potentially life-threatening allergic reactions are also possible. Up to 1 in 4 million vaccinated individuals may develop a serious allergic reaction, which can manifest as hives, angioedema, bronchospasm, tachycardia, or hypotension, and can usually be stopped by the administration of emergency medications by the study personnel. As with any vaccine, there is a very small chance of a fatal reaction, although researchers do not expect this to occur.

Blood Draw and IM Injection

Blood draws and IM injections may cause transient discomfort; fainting, which can managed by having the subject lie down; bruising, which can be prevented or lessened by applying pressure to the blood draw site for several minutes; nerve injury, which is very unlikely; and infection, which can be made extremely unlikely by using sterile technique.

Respiratory Virus Testing

Subjects may experience reactogenicity phenomena following study vaccination that overlap with symptoms of community-acquired respiratory viruses, including SARS-CoV-2. To accurately assess relatedness of systemic symptoms to study product, subjects who meet symptom criteria (detailed in the MOP) may undergo viral testing during the study. Sampling procedures (including, but not limited to, nasopharyngeal swab, nasal swab, or saliva sampling) may cause transient discomfort.

Pregnancy

It is unknown if these vaccines pose any risks to an unborn child. As such, women of childbearing potential (women who have not reached menopause >1 year) or who have not been surgically sterilized (tubal ligation, bilateral oophorectomy, or hysterectomy) must agree to use effective contraception for ≥28 days before the first vaccination through ≥60 days after the last vaccination.

Breach of Confidentiality

Subjects will be asked to provide personal health information (PHI). All attempts will be made to keep this PHI confidential within the limits of the law. However, there is a chance that

unauthorized persons will see a subject's PHI. All records will be kept in a locked file cabinet or maintained in a locked room at the site. Electronic files will be password protected. Only people who are involved in the conduct, oversight, monitoring, or auditing of this trial will be allowed access to this PHI. Any publications from this trial will not use information that will identify subjects by name. Organizations that may inspect and/or copy research records maintained at the site for quality assurance (QA) and data analysis include groups such as NIAID (or its designee) and FDA.

A description of this trial will be available on http://www.ClinicalTrials.gov, as required by US Law. This web site will not include information that can identify subjects. At most, this web site will include a summary of the results.

There may be other unknown risks, discomforts, or side effects.

2.3.2 Known Potential Benefits

As the efficacy of ChAd155-RG in protecting against human rabies has not been proven, there are no known potential benefits to the subjects receiving ChAd155-RG in this trial. Subjects who receive RABAVERT will be immunized against rabies. Society benefits if a safe, tolerable, equally immunogenic vaccine for rabies which can be given in fewer doses than the currently available rabies virus vaccines, is ultimately developed as a result of this and other vaccine trials.

3. OBJECTIVES AND OUTCOME MEASURES

3.1. Study Objectives

3.1.1. Primary

- Assessment of the safety, tolerability, and reactogenicity of one dose of ChAd155-RG at $5x10^{10}$vp per dose, or one or two doses of ChAd155-RG at $1x10^{11}$vp per dose.
- Comparison of the safety, tolerability, and reactogenicity of one or two doses of ChAd155-RG, with three doses of RABAVERT.

3.1.2. Secondary

- Assessment of serum rabies VNA levels by a standard, WHO-approved, RFFIT, as assessed by immune response kinetics (through approximately 12 months after first dose of vaccine), seroconversion rates, and peak GMT in each treatment arm.

3.1.3. Exploratory (if applicable)

- Quantification of circulating T-cell responses against RG by ex vivo IFN-gamma ELISpot.
- Characterization of circulating CD4 and CD8 T-cell responses against RG at peak time point by multiparameter ICS.
- Comparison of durability of immune response to ChAd155-RG and RABAVERT at approximately 3, 6, and 12 months after first dose of vaccine by each treatment arm tested.
- Measurement of RG-specific memory B cells by ELISpot.
- Measurement of an antibody response to the vaccine vector (ChAd155).

3.2. Study Outcome Measures

3.2.1. Primary

- Frequency and severity of solicited injection site and systemic reactogenicity events from the time of each vaccination through Day 7 after each vaccination, in each treatment arm and overall.
- Frequency and severity of SAEs considered study vaccine-related and reported at any time after the first vaccination through the end of the study, in each treatment arm and overall.

- Frequency and severity of study vaccine-related lab AEs through Day 22 from the time of the first vaccination , in each treatment arm and overall.
- Frequency and severity of unsolicited study vaccine-related AEs from the time of the first vaccination through Day 28 after the last vaccination, in each treatment arm and overall.
- Number of subjects with new onset of a chronic medical condition at any time after the first vaccination, in each treatment arm and overall.
- Frequency and severity of any SAEs at any time after the first vaccination through the end of the study, in each treatment arm and overall.

3.2.2. Secondary

- Proportion of subjects seroconverting to rabies virus at each antibody time point (seroconversion is defined as VNA concentration ≥0.5 IU/mL), in each treatment arm and overall.
- GMT (as measured by rabies VNA using a standard, WHO-approved, RFFIT) at each antibody time point within each treatment arm.
- Peak GMT (as measured by rabies VNA using a standard, WHO-approved, RFFIT; peak GMT is defined as highest GMT measured across all post-vaccination antibody time points) within each treatment arm.

3.2.3. Exploratory

- Frequencies of circulating T cell responses against RG by ex vivo IFN-ELISpot.
- Characteristics of circulating CD4 and CD8 T cell responses against RG by multiparameter ICS.
- Magnitude and durability of RG-specific memory B cells by ELISpot.
- GMT to ChAd155 vector at one month after first vaccination

4. STUDY DESIGN

This is a single-center, observer-blinded, Phase 1, dosage-escalation trial to evaluate the safety, tolerability, reactogenicity, and immunogenicity of ChAd155-RG compared with RABAVERT in rabies virus-naïve healthy male and non-pregnant female adult subjects. There are 4 dose groups (see Table 1).

Subjects who have never received a licensed or investigational rabies virus vaccine, or an Ad-based investigational vaccine, and who have never been exposed to a rabid animal will be eligible for enrollment.

Since this is a dosage-escalation study, sentinel subjects will be used at each dosage level. The first four subjects will be randomized to Group A or D and will receive either a single dose of ChAd155-RG at the lower dosage ($5x10^{10}$vp, N=3) (followed by placebo on Days 8, 15, and 22) or RABAVERT (N=1) (on Days 1, 8, and 22, and placebo on Day 15) (see Table 1). These subjects will be monitored for safety, tolerability, and reactogenicity through day 7 after first study vaccination, and if no pre-defined halting rule is met (Section 9), then four additional sentinel subjects will be randomized to Group B or D and will receive either a single dose of ChAd155-RG at the higher dosage ($1x10^{11}$vp, N=3) (followed by placebo on Days 8, 15, and 22) or RABAVERT (N=1) (on Days 1, 8, and 22, and placebo on Day 15) (see Table 1). These subjects will be monitored for safety, tolerability, and reactogenicity through day 7 after first study vaccination. The SMC will then review the available safety, reactogenicity, AE, and lab data of all the sentinel subjects, and will decide if the remaining non-sentinel subjects should be enrolled.

If the SMC recommends proceeding with the remainder of trial enrollment, eligible individuals will be enrolled and randomized to one of the four dose groups (Groups A-D). To maintain blinding of study personnel conducting surveillance and assessment of AEs, subjects in these groups will receive 4 sequential injections, 1 ml per injection, in alternating arms, on the same schedule:

- Subjects randomized to Group A will receive ChAd155-RG at the lower dosage ($5x10^{10}$vp) on Day 1, then placebo injections on Days 8, 15, and 22
- Subjects randomized to Group B will receive ChAd155-RG at the higher dosage ($1x10^{11}$vp) on Day 1, then placebo injections on Days 8, 15, and 22
- Subjects randomized to Group C will receive ChAd155-RG at the higher dosage ($1x10^{11}$vp) on Days 1 and 15, and placebo injections on Days 8 and 22
- Subjects randomized to Group D will receive RABAVERT at the standard dose (1 mL) on Days 1, 8, and 22, and a placebo injection on Day 15

Vaccine preparation and administration will be unblinded. The investigational vaccine doses will be prepared just prior to administration to ensure that the volume of each injection is identical

between Groups A-D. Follow-up will be double-blinded, and the SMC will review the available safety, reactogenicity, AE, and lab data collected from all subjects for 15 days after the last vaccination and as needed.

Subjects will be followed using a memory aid, face-to-face scheduled clinic visits, and safety lab monitoring as outlined in Section 7 and Appendix A. Following enrollment and vaccination, safety lab blood samples will be collected on Days 2, 8, 16, and 22. Blood samples for immunologic assays will be collected on Days 1, 2, 8, 15, 16, 22, 29, 91, 181, and 381. The duration of each subject's participation will be approximately 13 months, from recruitment to collection of data for safety and immunogenicity outcomes.

This trial is expected to take approximately 30 months to complete, from initiation through availability of a final report on the primary outcomes of safety and the secondary outcomes of VNA levels to RG. Exploratory outcomes are discussed in an addendum. A planned interim safety, reactogenicity, and immunogenicity analysis is detailed in Section 11.3.

4.1. Sub-studies (if applicable)

No sub-studies are planned.

5. STUDY ENROLLMENT AND WITHDRAWAL

Subject inclusion and exclusion criteria must be confirmed by a study clinician licensed to make medical diagnoses. No exemptions are granted on subject inclusion/exclusion criteria in DMID-sponsored studies. Questions about eligibility will be directed toward the DMID Medical Officer.

5.1. Subject Inclusion Criteria

Subjects must meet all the following inclusion criteria to be eligible for this trial, and for subsequent doses of vaccine/placebo:

1. Must be a male or female aged 18-49 years old (inclusive) at the time of first vaccination.
2. Must be able to provide written informed consent.
3. Must have a body mass index (BMI) ≥18.5 and <35.0 kg/m^2
4. Must be in good health based on physical examination, vital signs[a], medical history, safety labs[b], and the investigator's clinical judgment.

[a]*Vital signs must be within the normal ranges in Appendix C. If a subject has elevated systolic or diastolic blood pressure, subject may rest for 10 minutes in a quiet room and the blood pressure may be retaken.*

[b]*Safety lab normal ranges will be those used by the reference clinical lab. Protocol-specific criteria for individual subjects are listed in criteria #5.*

5. Must have acceptable* lab values within 28 days before enrollment.

**Acceptable values include:*

-Hemoglobin: women >11.6 g/dL, men >13.1 g/dL

-White blood cells: >3,700 but <10,900 cells/mm^3

-Absolute neutrophil count: ≥1,500 cells/mm^3

-Absolute lymphocyte count: ≥850 cells/mm^3

-Platelets: >139,000 but <401,000 per mm^3

-Urine dipstick (clean urine sample): protein <1+, glucose negative

-Alanine transaminase and aspartate transaminase (ALT, AST) <1.1 x institutional upper limit of normal (ULN)

-Total bilirubin <1.1x institutional ULN

-Blood urea nitrogen (BUN) <1 x institutional ULN

-Serum creatinine <1x institutional ULN

-If lab screening tests are out of range, repeating them is permitted once, provided there is an alternative explanation for the out-of-range value.

6. Women of childbearing potential* must have a negative serum pregnancy test at screening and negative urine pregnancy tests within 24 hours before each vaccination.

   **Women of non-childbearing potential, defined as postmenopausal (any age with amenorrhea for ≥12 months without other known or suspected cause for amenorrhea), or surgically sterile [hysterectomy, bilateral tubal ligation, bilateral oophorectomy, or successful Essure® placement (permanent, non-surgical, non-hormonal sterilization)] with documented confirmation test ≥3 months after the procedure), are not required to use contraceptive methods.*

7. Women of childbearing potential must use an acceptable method of contraception* from 28 days before the first vaccination until ≥60 days after the last vaccination.

   **Acceptable methods of contraception include: prescription oral contraceptives, contraceptive injections, intrauterine device (IUD), implants, vaginal ring, double-barrier method, contraceptive patch, male partner who had a vasectomy at least 6 months prior to study enrollment, abstinence (defined as refraining from heterosexual intercourse during participation in this trial [from 28 days before the first vaccination until ≥60 days after the last vaccination]).*

8. Female subjects must agree to not donate eggs (ova, oocytes) from the start of screening until ≥60 days after the last vaccination.

9. Male subjects who have not had a vasectomy* and are sexually active with a woman of childbearing potential must agree to use an acceptable method of contraception**.

   **Men who have had a vasectomy must have had the procedure performed at least 6 months prior to study enrollment*

   **Acceptable methods of contraception must be used from the first vaccination until ≥60 days after the last vaccination, and include: abstinence (defined as refraining from heterosexual intercourse with a female partner of childbearing potential during participation in this trial [from 28 days before the first vaccination until ≥60 days after the last vaccination]; a double-barrier method, such as condom with spermicidal foam/gel/film/cream/suppository and partner with occlusive cap (diaphragm, cervical/vault caps); if the female partner is using an acceptable method of contraception (see Inclusion Criterion #7), a single-barrier method for the male subject is acceptable.*

10. Male subjects must agree to not donate sperm from the start of screening until ≥60 days after the last vaccination.
11. Must be available and willing to participate for the duration of this trial.
12. Must have a means to be contacted by telephone.

5.2. Subject Exclusion Criteria

Subjects meeting any of the following exclusion criteria at baseline will be excluded from study participation.

1. Was ever vaccinated with a licensed or investigational rabies vaccine* or was diagnosed with rabies exposure, infection, or disease.

   **Includes RABAVERT and Imovax. Subject's verbal history will suffice.*

2. Has a higher risk than the average US resident with regard to exposure to rabies, per the Rabavert package insert and rabies vaccination recommendations from the CDC*

   **People at high risk of exposure to rabies, such as veterinarians, animal handlers, rabies laboratory workers, spelunkers, and rabies biologics production workers.*
   **People whose activities bring them into frequent contact with rabies virus or with possibly rabid animals.*
   **International travelers who are likely to come in contact with animals in parts of the world where rabies is common.*

3. Was ever vaccinated with a licensed or investigational Ad vector or Ad vaccine.
4. Is currently taking chloroquine or hydroxychloroquine[26]
5. Was diagnosed with laboratory-confirmed COVID-19 (PCR or antigen-based test) in the preceding 28 days.
6. Positive serology for HIV antibody, HCV antibody, or Hepatitis B surface antigen (HBsAg).
7. Has known allergy or history of anaphylaxis or other serious adverse reaction to a vaccine or vaccine products*.

   **Including egg products, aminoglycosides, gelatin, sorbitol, tris (hydroxymethyl)-amino methane (THAM), or any of the constituents of the study vaccines.*

8. Has severe allergy or anaphylaxis to latex.
9. Has an acute illness or temperature ≥38.0ºC on Day 1*.

**Subjects with fever or acute illness on the day of vaccination may be re-assessed and enrolled if healthy or only minor residual symptoms remain within 3 days.*

10. Female subjects who are pregnant or breastfeeding, or planning to become pregnant while enrolled in this trial and at least 60 days after last vaccination.
11. Has history of autoimmune disease, or clinically significant cardiac, pulmonary, hepatic, rheumatologic, or renal disease by history, physical examination, and/or lab studies.
12. Has history of malignancy other than squamous cell or basal cell skin cancer, unless there has been surgical excision that is considered to have achieved cure*.

    **Subjects with a history of skin cancer must not be vaccinated at the previous tumor site.*
13. Has known or suspected congenital or acquired immunodeficiency, or recent history or current use of immunosuppressive therapy*.

    **Anti-cancer chemotherapy or radiation therapy within the preceding 6 months, or long-term (≥2 weeks within the previous 3 months) systemic corticosteroid therapy (at a dosage of ≥0.5 mg/kg/day). Intranasal or topical prednisone (or equivalent) are allowed.*
14. Is post-organ and/or stem cell transplant, whether or not on chronic immunosuppressive therapy.
15. Had major surgery (per the investigator's judgment) within 4 weeks before study entry or planned major surgery during this trial.
16. Has history of diabetes mellitus type 1 or type 2, including cases controlled with diet alone.

    **<u>Note</u>: history of isolated gestational diabetes is not an exclusion criterion.*
17. Has history of thyroidectomy, or thyroid disease requiring medication in the last 12 months.
18. Has history of hypertension, even if medically controlled.

    **<u>Note</u>: Vital signs must be normal by protocol toxicity grading scale. In the event of an abnormal heart rate or blood pressure due to physiological variation or activity, the subject may rest for 10 minutes in a quiet room, and then blood pressure and/or heart rate re-measured. Repeated vital signs may be used to determine eligibility.*
19. Received live attenuated vaccines from 30 days before first vaccination until 30 days after final vaccination.*

    *Not including licensed or authorized COVID-19 vaccines

20. Received killed or inactivated vaccines from 14 days before first vaccination until 30 days after final vaccination.*

    *Not including licensed or authorized COVID-19 vaccines

21. Received experimental therapeutic agents within 3 months before first vaccination or plans to receive any experimental therapeutic agents during this trial.*

    **That that in the opinion of the investigator would interfere with safety or immunogenicity assessments.*

22. Is currently participating or plans to participate in another clinical study which would involve receipt of the following:*

    ** An investigational product, blood drawing, or an invasive medical procedure that would require administration of anesthetics, intravenous (IV) dyes, or removal of tissue during this trial and, in the opinion of the investigator, would interfere with safety or immunogenicity assessments.*

    -Includes endoscopy, bronchoscopy, and administration of IV contrast.

23. Received blood products or immunoglobulin in the 3 months before study entry or planned use during this trial.

24. Donated a unit of blood or blood products within 8 weeks before Day 1 or plans to donate blood or blood products during this trial.

25. Has major psychiatric illness in the past 12 months that in the opinion of the investigator would preclude participation.

26. Has current alcohol use or current or past abuse of recreational or narcotic drugs by history as judged by the investigator to potentially interfere with study adherence.

27. Has a history of chronic urticaria.

28. Has tattoos, scars, or other marks on both deltoid areas which would, in the opinion of the investigator, interfere with assessment of the vaccination site.

29. Is a site employee* or staff who are paid entirely or partially by/through the OCRR contract for this trial, or staff who are supervised by the Principal Investigator (PI) or sub-investigators.

    **Including the PI, sub-investigators listed on Form FDA 1572 or Investigator of Record Form.*

30. In the opinion of the investigator cannot communicate reliably, is unlikely to adhere to the requirements of this trial, or has any condition which would limit the ability to complete this trial.

31. Has history of Guillain-Barre syndrome, meningitis, encephalitis, neuroparalysis, transient paralysis, myelitis, retrobulbar neuritis, multiple sclerosis, vertigo, or visual disturbances.

32. Has a history of arterial or venous thrombosis, or thrombocytopenia that required medical attention.

5.3. Treatment Assignment Procedures

5.3.1. Randomization Procedures

The list of randomized vaccine assignments will be prepared by statisticians at The Emmes Corporation and included in the enrollment module of The Emmes Corporation's Internet Data Entry System (IDES). IDES will assign each subject a vaccine code from the list after demographic and eligibility data have been entered into it. A designated individual at the site will be provided with a vaccine key, which links the vaccine code to the vaccine assignment, which will be kept in a secure place.

Subjects in sentinel group 1 will be randomized 3:1 to either dose Group A or D. If no halting criteria are met through Day 8, then subjects in sentinel group 2 will be randomized 3:1 to either dose Group B or D. The SMC will review safety data from both sentinel groups through Day 15 for sentinel group 1 (Day 8 for sentinel group 2), and if they approve, then the remaining non-sentinel subjects will be enrolled and randomized to the 4 dose groups (Groups A-D).

Instructions for use of the enrollment module are included in the IDES User's Guide. Manual back-up randomization procedures are provided in the Manual of Procedures (MOP) for use in case the site temporarily loses access to the Internet or the online enrollment system is unavailable.

Study product will be administered by an unblinded administrator. The subjects, the study personnel who perform study assessments after administration, data entry personnel at the site, and laboratory personnel performing immunologic assays will be blinded to vaccine assignment.

The SMC may receive data in aggregate and presented by dose group, but without the dose group (or dose level) identified. The SMC may be unblinded to individual vaccine assignments, as needed, to adequately assess safety issues. Refer to the MOP for unblinding procedures.

5.3.2. Masking Procedures

Investigators and study personnel performing any study-related assessments following vaccination will be blinded to study vaccine. Syringes will be labeled with an overlay/blinding tape containing the subject ID, and the treatment number from the treatment key, and expiration time for the syringe, and provided to the unblinded vaccine administrator. Lab personnel performing assays will be blinded to all subjects.

The randomization scheme will be generated by the Statistical and Data Coordinating Center (SDCC) and provided to unblinded study personnel (i.e., pharmacists preparing vaccines, clinical staff administering vaccines) at the site.

The unblinded vaccine administrator will be credentialed to administer vaccines, but will not be involved in study-related assessments, subject contact, or data collection following vaccination.

5.3.3. Reasons for Withdrawal and Discontinuation of Study Product Administration

Subjects may voluntarily withdraw their consent for study participation at any time without penalty or loss of benefits to which they are otherwise entitled. A study subject will be discontinued from participation in the trial if any of the following reasons occur before dosing:

- Request by the subject to terminate participation
- Lost to follow-up
- As deemed necessary by the site principal investigator or appropriate sub-investigator for noncompliance or other reasons.
- New information becomes available that makes further participation unsafe.

An investigator may also withdraw a subject from receiving subsequent doses of vaccine for any reason. Follow-up safety and immunogenicity evaluations will be conducted, if the subject agrees. The reasons may include, but are not limited to, the following:

- Subject no longer meets eligibility criteria
- Subject meets individual halting criteria (listed in Section 9.5)
- Subject becomes noncompliant
- Subject develops a medical disease or condition, 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 trial, or interfere with the evaluation of responses
- Female subject becomes pregnant
- Male subject if his female partner becomes pregnant
- Determined by a physician’s discretion to require additional therapy not indicated in the protocol to ensure the subject’s health and well-being

If a subject withdraws or is withdrawn before completing this trial, the reason for this decision will be recorded in the electronic case report form (eCRF). The investigator will be explicit regarding study follow-up (e.g., safety follow-up) that might be carried out despite the fact the

subject will not receive further vaccine. If the subject consents, every attempt will be made to follow all AEs through resolution. The procedures that collect safety data for the purposes of research will be inclusive in the original ICF or the investigator may seek subsequent informed consent using an IRB/IEC-approved ICF with the revised procedures.

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

5.3.4. Handling of Withdrawals and Discontinuation of Administration

The primary reason for withdrawal from this trial will be recorded on the Study Status form. If a subject discontinues product administration, the primary reason will be recorded on the Discontinuation of Treatment form. Subjects will be asked to complete the Early Termination Visit (procedures listed in Section 7.6).

Although subjects have the right to withdraw from this trial at any time or may be withdrawn by the site PI or appropriate sub-investigator at any time, those subjects who received only one dose of vaccine will be asked to remain in this trial for follow-up safety and immunogenicity assessments. Safety assessments may be done by phone call, rather than in person, if this is the only means available of obtaining safety information. Subjects who withdraw from this trial will be asked to provide blood samples for immunogenicity testing and exploratory endpoints. See the MOP for alternate follow-up requirements. There will be no exceptions to the planned dose schedule.

Every attempt will be made to follow all AEs, including solicited injection site and systemic reactions, unsolicited non-serious AEs, and SAEs, ongoing at the time of early withdrawal through resolution as per applicable collection times defined for specific types of AEs.

If subjects fail to appear for a safety follow-up assessment, extensive effort (i.e., three documented contact attempts via phone calls, e-mails, etc., made on separate occasions and followed by a certified letter) will be made to locate or recall them, or at least to determine their health status. These efforts will be documented in the subject's records.

Subjects may be replaced following written approval from the sponsor (detailed in next sub-section). Subjects who withdraw, or are withdrawn or terminated from this trial, or are lost to follow-up after signing the ICF and randomization but before receipt of the first dose of vaccine will be replaced.

5.3.5. Subject Replacement

Subjects who withdraw, or are withdrawn or terminated from this trial, or are lost to follow-up after signing the ICF and randomization, but before receipt of any doses of vaccine will be replaced. Subjects who do not receive protocol-specified subsequent doses of vaccine may be replaced following written approval of the sponsor (Section 5.3). Subjects who are discontinued or withdraw from this trial will be replaced irrespective of the dose group to which they were

assigned, and replacement subjects will be assigned according to usual enrollment/randomization procedures.

5.3.6. Termination of Study

If this trial is prematurely terminated by the sponsor, any regulatory authority, or the investigator for any reason, the investigator will promptly inform the 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/IEC.

6. STUDY INTERVENTION/INVESTIGATIONAL PRODUCT

6.1. Study Product Description

ChAd155-RG Vaccine

The ChAd155-RG Vaccine consists of a replication-defective group C ChAd, ChAd155, expressing RG under the control of the CMV promoter. The RG sequence cloned into the ChAd155 vector is a medoid, a natural viral strain with the highest average percent of amino acid identity among all RG sequences annotated in the NCBI database. The selected RG (NCBI strain AGN94271) shares an average 94% percent identity to the RGs in current vaccines.

RABAVERT Vaccine

The RABAVERT Vaccine is an inactivated, purified chick embryo cell vaccine (PCECV). It consists of lyophilized rabies virus (strain Flury LEP) that has been propagated in chicken fibroblasts, inactivated with β-propiolactone, and concentrated and purified by centrifugation**Placebo**

The placebo is 0.9% Sodium Chloride, USP injection.

6.1.1. Acquisition

ChAd155-RG and RABAVERT will be provided by GSK Vaccines under agreement with DHHS, and will be supplied through DMID CMS to the site before the start of this trial upon request and with prior approval from DMID. Should the site PI require additional vaccine during this trial, further instructions are provided in the MOP.

Placebo

Normal saline will be used as the placebo, and will be provided by DMID Clinical Materials Services (CMS, Fisher BioServices

6.1.2 Formulation, Packaging, and Labeling

ChAd155-RG Vaccine
The purified ChAd155-RG bulk drug substance is processed as follows to obtain drug product: Purified ChAd155-RG drug substance is diluted in buffer A 195 (Tris base 10mM, NaCl 75 mM, L-Histidine 10mM, $MgCl_2$ 1 mM, EDTA 0.1 mM, Polysorbate 80 0.02% (w/v), sucrose 5% (w/v), ethanol 0.5% (v/v), HCl for adjustment to pH 7.4). It then undergoes sterile filtration and

is then transferred into final containers. The ChAd155-RG drug product is a liquid formulation contained in vials. It is presented as a sterile suspension in a 3-mL clear glass, stoppered vial, with a 1-mL extractable volume (nominal single dose).

The vaccine is suppled as a single 1mL-dose and is formulated without preservative.

The study product will be labelled according to manufacturer specifications or regulatory specifications and include the statement "Caution: New Drug-Limited by Federal Law to Investigational Use."

RABAVERT Vaccine[23]

RABAVERT is a freeze-dried vaccine obtained by growing the fixed-virus strain Flury LEP in primary cultures of chicken fibroblasts. The virus is inactivated with β-propiolactone, processed by zonal centrifugation, and then lyophilized after adding a stabilizer solution consisting of buffered polygeline and potassium glutamate. One dose of reconstituted vaccine contains ≤12 mg polygeline (processed bovine gelatin), ≤0.3 mg human serum albumin, 1 mg potassium glutamate, and 0.3 mg sodium EDTA. Small quantities of bovine serum (originating from the US, Australia, and New Zealand) are used in the cell culture process; ovalbumin content is ≤3 ng/dose (1 mL), based on ELISA. Antibiotics (neomycin, chlortetracycline, amphotericin B) added during cell and virus propagation are largely removed during subsequent steps in the manufacturing process; thus, in the final vaccine, neomycin is present at ≤10 µg, chlortetracycline at ≤200 ng, and amphotericin B at ≤20 ng per dose. Rabavert is supplied in a package that contains a vial of the freeez-dried vaccine, a syringe containing 1 mL of sterile diluent (sterile water for injection), a sterile needle for reconstitution, and a sterile needle suitable for IM injection.

The study product will be labelled according to manufacturer specifications or regulatory specifications and include the statement "Caution: New Drug-Limited by Federal Law to Investigational Use."

Placebo

Placebo will be supplied as 0.9% Sodium Chloride Injection, USP which is a colorless, sterile, nonpyrogenic, isotonic solution of sodium chloride and water for injection (WFI). Each mL contains supplied chloride 9 mg. It contains no bacteriostatic, antimicrobial agent, or added buffer and is supplied only in single-dose containers. The placebo, 0.9% Sodium Chloride, contains no preservatives. The solution may contain hydrochloric acid and/or sodium hydroxide for pH adjustment (pH 5.3 [4.5 to 7.0]).

6.1.3 Product Storage and Stability

ChAd155-RG Vaccine

The vaccine product requires storage at ≤ -60˚C

The stability of ChAd155-RG final container lots will be followed for up to 60 months at the recommended storage condition of ≤ -60°C.

RABAVERT Vaccine[23]

RABAVERT contains no preservative, and should be stored protected from light at 2°C to 8°C (36°F to 46°F). After reconstitution with the supplied sterile diluent (water for injection), the vaccine should be used immediately (as defined in the MOP). The reconstituted vaccine is a clear-to-slightly opalescent, colorless-to-slightly pink suspension. The vaccine may not be used after the expiration date given on package and container.

Placebo

0.9% Sodium Chloride, USP injection must be stored at 20ºC to 25ºC (68°F to 77ºF) [See USP Controlled Room Temperature; excursions between 15°C and 30°C (59°F and 86°F) are permitted]. See protocol-specific MOP for further instructions..

Additional information

All vaccines will be stored in the site research pharmacy. The temperature of the storage unit will be continuously monitored and recorded during this trial per the site's SOPs, and documentation will be maintained. If the temperature fluctuates outside of the required range, the affected study product(s) will be quarantined at the correct storage temperature and labeled "Do Not Use" (until further notice). The pharmacist will alert the site PI and study coordinator if the temperature fluctuates outside of the required range. If the temperature fluctuates outside of the required range, including accidental deep-freezing or disruption of the cold chain, the affected vaccine(s) will not be administered. The site PI or responsible person should immediately contact the DMID Product Support Team at DMIDProductSupportTeam@niaid.nih.gov and DMID Clinical Project Manager (CPM) for further instructions before any additional study vaccines are administered. Based on the information collected, DMID and/or the manufacturer will determine whether the affected vaccine(s) can be used. If it cannot be used, the site will receive specific instructions on how to return the affected vaccine(s) to DMID CMS or destroy it on site. Additional instructions for quarantine are provided in the MOP.

Stability studies to support study vaccine storage conditions have been conducted. The sponsor will continue to monitor the stability of the vaccines and will alert the site if a lot is nearing the end of its anticipated shelf life.

6.2. Dosage, Preparation and Administration of Study Intervention/Investigational Product

ChAd155-RG Investigational Vaccine
The ChAd155-RG candidate vaccine is formulated without preservative and is presented as a sterile suspension in a 3-mL clear glass, stoppered vial, at a concentration of $1.3x10^{11}$vp/mL.

The dosages of ChAd155-RG to be used in this study are: $5x10^{10}$vp (low dose) and $1x10^{11}$vp (high dose). These dosages will be prepared at site's pharmacy. See the MOP for detailed information on the preparation, labeling, storage, and administration of the ChAd155-RG investigational vaccine. Vaccine preparation will be performed by the site pharmacist on the same day of vaccine administration.

On vaccination days, vaccines will be allowed to thaw to room temperature and administered within 1 hour. The vaccine is administered IM over the deltoid region of the arm. Qualified medical personnel will wear gloves. During administration of the vaccine, medication and resuscitation equipment will be immediately available for the management of anaphylaxis.

In order to minimize dissemination of the recombinant vectored vaccine virus into the environment, the inoculation site will be covered with a dressing after immunization. This should absorb any virus that may leak out through the needle track. The dressing will be removed from the injection site after 30 minutes and will be disposed as Genetically Modified Organisms (GMO) waste by autoclaving, in accordance with the relevant SOP and current standard US practice.

RABAVERT Vaccine[23]

The RABAVERT Vaccine will be prepared as outlined in the package insert. Additional information: See the MOP for detailed information on the preparation, labeling, storage, and administration of study vaccine for each group. Vaccine preparation will be performed by the site pharmacist on the same day of vaccine administration.

Visually inspect the ChAd155-RG Vaccine, RABAVERT Vaccine, and admixture vials, upon receipt and before use. If the vaccine(s) appear(s) to be damaged, contaminated, or discolored, contain visible particulate matter, or if there are any concerns regarding its integrity, do not use the affected vaccine(s). Instead, quarantine it at the correct storage temperature and label it "Do Not Use" (until further notice). The site PI or responsible person should immediately contact the DMID Product Support Team at DMIDProductSupportTeam@niaid.nih.gov and DMID CPM for further instructions before any additional vaccines are administered. Based on the information collected, DMID and/or the manufacturer will determine whether the affected vaccine(s) or admixture can be used. If it cannot be used, the site will receive specific final disposition instructions from DMID (e.g. how to return the affected vaccine(s) to DMID CMS or destroy it on site). If the vaccine(s) or admixture is unusable, the site pharmacist will prepare another dose

of admixture vial. Replacement vials may be requested by contacting DMID. Additional instructions for quarantine and DMID contact information are provided in the MOP.

Study vaccine administration will be performed by an unblinded vaccine administrator who is credentialed to administer vaccines, but will not participate in dose preparation, and may not be involved in study-related assessments, subject contact, or data collection after vaccine administration. On the assigned dosage days, each dose of vaccine will be administered to subjects via a single IM injection (depending on group assignment and/or randomization) in the preferred arm. The dose administration schedule is as follows:

- Subjects randomized to Group A will receive ChAd155-RG at the lower dosage ($5x10^{10}$vp) on Day 1, then placebo injections on Days 8, 15, and 22
- Subjects randomized to Group B will receive ChAd155-RG at the higher dosage ($1x10^{11}$vp) on Day 1, then placebo injections on Days 8, 15, and 22
- Subjects randomized to Group C will receive ChAd155-RG at the higher dosage ($1x10^{11}$vp) on Days 1 and 15, and placebo injections on Days 8 and 22
- Subjects randomized to Group D will receive RABAVERT at the standard dose (1mL) on Days 1, 8, and 22, and a placebo injection on Day 15

Placebo

Gently invert the normal saline vial 5 to 7 times. Using aseptic technique, puncture the septum top of the saline vial with a 1-inch, 23- or 25-gauge disposable, sterile needle attached to a 1-mL disposable, sterile syringe. Withdraw 1.0 mL from the saline vial. The prepared saline dose will be allowed to store at room temperature and administered within 1 hour. The dose will be administered IM over the deltoid region of the preferred arm.

6.3. Accountability Procedures for the Study Investigational Product(s)

Study vaccines will be stored and shipped from DMID CMS to the site. Once received, vaccines will be stored in and dispensed by the Emory Hope Clinic Investigational Pharmacy. Unused vaccines will be returned to DMID CMS.

The FDA requires accounting for the disposition of all investigational products. After receipt of the study vaccine, the site principal investigator is responsible for ensuring that a current record of product disposition is maintained and product is dispensed only at an official study site by authorized personnel as required by applicable regulations and guidelines. Records of product

disposition, as required by federal law, consist of the date received, date administered, quantity administered, and the subject number to whom the study product was administered.

As this is a blinded study, the site principal investigator will delegate this responsibility to the unblinded site pharmacist. The unblinded site Pharmacist will be responsible for maintaining accurate records of the shipment and dispensing of the investigational product. The pharmacy records will be available for inspection by the DMID monitoring contractors, and is subject to inspection by a regulatory agency (e.g., FDA) at any time. An assigned Study Monitor will review the pharmacy records.

Unused reconstituted vaccines will be stored at [2ºC-8ºC for RABAVERT, ≤ -60 ºC for ChAd155-RG] in the Investigational Pharmacy until trial accountability is completed. At trial termination, all unused vaccines will be disposed in accordance with the MOP following their complete accountability and monitoring.

6.4. Concomitant Medications/Treatments

Administration of any medications, therapies, or vaccines will be documented on the appropriate eCRF. Concomitant medications will include all current medications and non-study vaccinations taken within 30 days before signing the ICF through approximately 28 days after the last study vaccination, and for new-onset chronic medical conditions through approximately 12 months after the last vaccination for each subject. Subjects who do not receive all vaccinations will have concomitant medications collected through approximately 28 days after the last vaccination, or early termination, whichever occurs first. Prescription and over-the-counter drugs will be included as well as herbals, vitamins, and supplements.

Use of new medication should prompt evaluation for the presence of a new diagnosis of chronic medical disease or condition.

Medications that might interfere with the evaluation of the study vaccines should not be used during the trial unless absolutely necessary. Medications in this category include the prohibited medications per the subject exclusion criteria (see Section 5.2, #3 and #11). In addition, the site PI or appropriate sub-investigator may identify other medications that should not be used due to a risk to subject safety or assessment of reactogenicity and immunogenicity. Use of medications as prophylaxis before study vaccination is prohibited.

To the sponsor's knowledge, there are no known drug-vaccine interactions with the study vaccines and subjects are not being asked to discontinue current medications not listed in the exclusion criteria. In the event medical conditions dictate use of medications, subjects are encouraged to obtain adequate care, comply with the course of therapy as prescribed by their physician, and inform the Investigator as soon as practicable. Details of all medications taken during this trial (date, brand or generic name) will be recorded.

7. STUDY SCHEDULE

7.1. Recruitment

Subjects will be recruited through: posting of IRB-approved flyers on the Emory University campus; use of social media, list serves (such as CDC, ID, and Vaccine Dinner Club), and clinical trial recruitment websites; a HIPAA-compliant clinical trials database to identify subjects of previous trials at the Hope Clinic who have agreed to future contact; presentations by Hope Clinic faculty at various University and community venues; and volunteer word-of-mouth.

If subjects are agreeable and interested, they will be screened initially over the phone for general eligibility criteria and their PHI will be saved in a locked cabinet in a secure office of the study coordinator/recruiter. When appropriate, an appointment at the Hope Clinic is then scheduled for an in-person clinic visit. Research staff will then obtain written consent per the standard informed consent process before conducting protocol-specific screening activities.

7.2. Screening

7.2.1. Visit 00A, Day -28 to -1, 1st Screen, Clinic Visit

- Potential subjects will be screened for eligibility -28 to -1 days before administration of the first study vaccination. If a subject has a positive symptom screen prior to the visit (per the MOP), subsequent procedures will be based on whether viral testing was pursued by the subject and the test result if applicable (as outlined in the MOP).

- The following activities will be performed at the 1st Screen:
- Begin the informed consent process. Provide subjects with a description of this trial (purpose and study procedures) and ask them to read the ICF. Provide an opportunity to the potential subject to ask questions. If she/he wishes to proceed with participation, the ICF should be signed before performing any screening procedures.
- Review eligibility criteria.
- Interview subjects to collect medical history, and recent (i.e., within past 30 days) travel, blood donation (8 weeks prior to Day 1), vaccination history, and pre-study medication use. Interview of subjects is sufficient for obtaining medical history. Solicitation of medical records from the subject's primary care provider is not required.
- Review subjects' concomitant medications taken within 90 days before signing the ICF.
- Obtain vital signs, including oral temperature, pulse, and blood pressure to assure eligibility.

- Measure height and weight and calculate BMI.
- Perform a full physical examination, including the following organs and organ systems: general appearance, head/eyes/ears/nose/throat (HEENT), neck, lungs, heart, abdomen, extremities, musculoskeletal, lymph nodes, skin, and nervous system. All physical exams will be done by a clinician licensed to make medical diagnoses and listed on Form FDA 1572 as site PI or sub-investigator.
- Collect venous blood for WBCs, hemoglobin, platelets, absolute neutrophil count, absolute lymphocyte count, ALT, AST, total bilirubin, BUN, creatinine, HIV-1/2 antibody, HBsAg, and HCV antibody.
- Collect serum for pregnancy test from all female subjects of childbearing potential.
- Collect urine for dipstick glucose and protein.
- The overall eligibility of the subject to participate in this trial will be assessed once all screening test values and results of any other required evaluations are available. Subjects who qualify for inclusion will be contacted and scheduled for enrollment and **first** vaccination within 28 days.

7.2.2. Visit 00B, Day -28 to -1, 2nd Screen, Clinic Visit – This visit may occur to repeat a screening assessment whose initial result is thought to be temporary OR for subjects previously identified as eligible but who are out of window for enrollment

- Subjects may return for a 2nd Screen for eligibility -28 to -1 days before administration of the first study vaccination. If a subject has a positive symptom screen prior to the visit (per the MOP), subsequent procedures will be based on whether viral testing was pursued by the subject and the test result if applicable (as outlined in the MOP).
- Review eligibility criteria, including results of available clinical screening lab evaluations.
- Review medical history and any updates obtained by interview of subjects since the screening visit (Visit 00A) to assure continued eligibility.
- Review all concomitant medications recorded on the appropriate eCRF.
- Obtain vital signs including oral temperature, blood pressure, and pulse to assure eligibility.
- Measure height and weight and calculate BMI.
- Perform a targeted physical examination, if needed.

- Collect blood and urine for safety labs, if needed.

7.3. Enrollment/Baseline

7.3.1. Visit 01, Day 1, Enrollment and First Vaccination

- If a subject has a positive symptom screen prior to the visit (per the MOP), subsequent procedures will be based on whether viral testing was pursued by the subject and the test result, if applicable (as outlined in the MOP).
- Reconfirm subject's willingness to participate before performing any study procedures, including the first vaccination.
- Review eligibility criteria, including results of all clinical screening lab evaluations, with subjects before the first vaccination to assure continued eligibility. **If the initial safety labs are >28 days old, these tests will need to be repeated and results obtained before first vaccination (see Section 7.2).*
- Review medical history and any updates obtained by interview of subjects since the last screening visit (Visit 00A or, if performed, Visit 00B) to assure continued eligibility.
- Review all concomitant medications and recent (within 28 days) vaccinations with subjects before the first vaccination for accuracy and completeness. Any new medications taken since the last screening visit (Visit 00A or, if performed, Visit00B) will be recorded on the appropriate eCRF and assessed for continued eligibility.
- Assess all AE/SAEs and new onset chronic medical conditions and record them on the appropriate eCRF.
- Obtain vital signs including oral temperature, blood pressure, and pulse before the first vaccination. Perform a targeted physical examination as needed before the first vaccination, if indicated based on review of complete medical history and updates obtained by interview of subjects since the last screening visit (Visit 00A or, if performed, Visit 00B).
- Perform a urine pregnancy test within 24 hours before first vaccination on all female subjects of childbearing potential. Results must be negative and known before enrollment and first vaccination.
- Collect blood samples for baseline immunogenicity (RFFIT assay), memory B cell (MBC), and T cell assays (IFN-gamma ELISPOT) before vaccination.
- Collect blood samples for developer assays before vaccination.

- Collect blood samples for PBMCs for future use, if the subject has provided consent.
- Subjects will be enrolled in Advantage EDC^{SM} and randomly assigned before the first vaccination.
- Perform pre-administration reactogenicity assessments before the first vaccination to establish baseline. Subjects will then receive a single dose of vaccine via IM injection into the deltoid muscle of the preferred arm. The site of injection (right or left arm) and time of administration will be recorded on the appropriate eCRF.
- Observe subjects in the clinic for ≥30 minutes after vaccination to monitor for any acute reactions.
- Evaluate the vaccination site and assess for reactogenicity and AE/SAEs for ≥30 minutes after vaccination and before discharge from clinic and record on the appropriate CRF.
- Subjects will enter their reactogenicity information on a memory aid. Subjects will be provided a thermometer, ruler and memory aid and will record daily oral temperature, solicited injection site and systemic reactions, any unsolicited AEs, and concomitant medications. Subjects will be asked to bring their memory aid with them at their next study visit.

7.4. Follow-up

7.4.1. Visit 02, Day 2 (window: +2 days)

- If a subject has a positive symptom screen prior to the visit (per the MOP), viral testing may be performed during this study visit with a research-based assay
- Obtain interim medical history, including an assessment for new medical conditions by interview of subjects and note any changes since the previous clinic visit.
- Record all concomitant medications on the appropriate eCRF.
- Obtain vital signs including oral temperature, blood pressure, and pulse.
- A targeted physical examination will be performed, if needed, by a study clinician licensed to make medical diagnoses and listed on Form FDA 1572 as the site PI or sub-investigator, if indicated based on review of complete medical history.
- Examine vaccination site for local reactions and determine whether information recorded by subjects on the memory aid conforms to examination.
- Collect blood for safety labs (chemistry, hematology) and developer assays.

- Assess all AE/SAEs and new onset chronic medical conditions and record them on the appropriate eCRF.

7.4.2. Visit 03, Day 8 (window: +2 days), Second Vaccination

- If a subject has a positive symptom screen prior to the visit (per the MOP), viral testing may be performed during this study visit with a research-based assay
- Obtain interim medical history, including an assessment for new medical conditions by interview of subjects and note any changes since the previous clinic visit or phone call.
- Review eligibility criteria with subjects, and review results of all safety lab evaluations, before vaccination to assure continued eligibility for vaccination.
- Record all concomitant medications on the appropriate eCRF.
- Perform a targeted physical examination, if indicated based on review of complete medical history.
- Obtain vital signs including oral temperature, blood pressure, and pulse.
- Examine vaccination site for local reactions and determine information recorded by subjects on the memory aid conforms to examination.
- Collect blood for safety labs (chemistry, hematology), immunogenicity (RFFIT assay), and developer assays before vaccination.
- Perform a urine pregnancy test within 24 hours before second vaccination on all female subjects of childbearing potential. Results must be negative and known before second vaccination.
- Perform pre-administration reactogenicity assessments before the second vaccination to establish baseline. Subjects will then receive a single dose of vaccine via IM injection into the deltoid muscle of the arm opposite to that of the first injection. The site of injection (right or left arm) and time of administration will be recorded on the appropriate eCRF.
- Observe subjects in the clinic for ≥30 minutes after vaccination to monitor for any acute reactions.
- Evaluate the vaccination site and assess for reactogenicity and AE/SAEs for ≥30 minutes after vaccination and before discharge from clinic and record on the appropriate CRF.

- Subjects will enter their reactogenicity information on a memory aid. Subjects will be provided a thermometer, ruler and memory aid and will record daily oral temperature, solicited injection site and systemic reactions, any unsolicited AEs, and concomitant medications. Subjects will be asked to bring their memory aid with them at their next study visit.
- Assess all AE/SAEs and new onset chronic medical conditions and record them on the appropriate eCRF.

7.4.3. Visit 04, Day 15 (window: +2 days), Third Vaccination

- If a subject has a positive symptom screen prior to the visit (per the MOP), viral testing may be performed during this study visit with a research-based assay
- Obtain interim medical history, including an assessment for new medical conditions by interview of subjects and note any changes since the previous clinic visit or phone call.
- Review eligibility criteria with subjects, and review results of all safety lab evaluations, before vaccination to assure continued eligibility for vaccination.
- Record all concomitant medications on the appropriate eCRF.
- Perform a targeted physical examination, if indicated based on review of complete medical history.
- Obtain vital signs including oral temperature, blood pressure, and pulse.
- Examine vaccination site for local reactions and determine whether information recorded by subjects on the memory aid conforms to examination.
- Collect blood for immunogenicity (RFFIT assay), T cell (IFN-gamma ELISPOT and ICS), and developer assays before vaccination. Blood for safety labs may be collected at the discretion of the investigator as specified in the MOP.
- Collect blood samples for PBMCs for future use, if the subject has provided consent.
- Perform a urine pregnancy test within 24 hours before third vaccination on all female subjects of childbearing potential. Results must be negative and known before third vaccination.
- Perform pre-administration reactogenicity assessments before the third vaccination to establish baseline. Subjects will then receive a single dose of vaccine via IM injection into the deltoid muscle of the arm opposite to that of the second injection. The site of injection (right or left arm) and time of administration will be recorded on the appropriate eCRF.

- Observe subjects in the clinic for ≥30 minutes after vaccination to monitor for any acute reactions.
- Evaluate the vaccination site and assess for reactogenicity and AE/SAEs for ≥30 minutes after vaccination and before discharge from clinic and record on the appropriate CRF.
- Subjects will enter their reactogenicity information on a memory aid. Subjects will be provided a thermometer, ruler and memory aid and will record daily oral temperature, solicited injection site and systemic reactions, any unsolicited AEs, and concomitant medications. Subjects will be asked to bring their memory aid with them at their next study visit.
- Assess all AE/SAEs and new onset chronic medical conditions and record them on the appropriate eCRF.

7.4.4. Visit 05, Day 16 (window: +2 days)

- If a subject has a positive symptom screen prior to the visit (per the MOP), viral testing may be performed during this study visit with a research-based assay
- Obtain interim medical history, including an assessment for new medical conditions by interview of subjects and note any changes since the previous clinic visit or phone call.
- Record all concomitant medications on the appropriate eCRF.
- Obtain vital signs including oral temperature, blood pressure, and pulse.
- A targeted physical examination will be performed, if needed, by a study clinician licensed to make medical diagnoses and listed on Form FDA 1572 as the site PI or sub-investigator, if indicated based on review of complete medical history.
- Examine vaccination site for local reactions and determine whether information recorded by subjects on the memory aid conforms to examination.
- Collect blood for safety labs (chemistry and hematology) and developer assays.
- Assess all AE/SAEs and new onset chronic medical conditions and record them on the appropriate eCRF.

7.4.5. Visit 06, Day 22 (window: +2 days), Fourth Vaccination

- If a subject has a positive symptom screen prior to the visit (per the MOP), viral testing may be performed during this study visit with a research-based assay

- Review medical history to assure continued eligibility.
- Review eligibility criteria with subjects, and review results of all safety lab evaluations, before vaccination to assure continued eligibility for vaccination.
- Review all concomitant medications for accuracy and completeness. Any new medications or vaccinations will be recorded and assessed for continued eligibility.
- Obtain vital signs including oral temperature, blood pressure, and pulse.
- Perform a targeted physical examination, if indicated based on review of complete medical history.
- Examine vaccination site for local reactions before vaccination and determine whether information recorded by subjects on the memory aid conforms to examination.
- Collect blood for safety labs (chemistry and hematology), immunogenicity (RFFIT assay), and developer assays before vaccination.
- Perform a urine pregnancy test within 24 hours before fourth vaccination on all female subjects of childbearing potential. Results must be negative and known before fourth vaccination.
- Perform pre-administration reactogenicity assessments before the fourth vaccination to establish baseline. Subjects will then receive a single dose of vaccine via IM injection into the deltoid muscle of the arm opposite to that of the third injection. The site of injection (right or left arm) and time of administration will be recorded on the appropriate eCRF.
- Observe subjects in the clinic for ≥30 minutes after vaccination to monitor for any acute reactions.
- Evaluate the vaccination site and assess for reactogenicity and AE/SAEs for ≥30 minutes after vaccination and before discharge from clinic and record on the appropriate CRF.
- Subjects will enter their reactogenicity information on a memory aid. Subjects will be provided a thermometer, ruler and memory aid and will record daily oral temperature, solicited injection site and systemic reactions, any unsolicited AEs, and concomitant medications. Subjects will be asked to bring their memory aid with them at their next study visit.
- Assess all AE/SAEs and new onset chronic medical conditions, then record them on the appropriate eCRF.

7.4.6. Visit 07, Day 29 (window: +14 days)

- If a subject has a positive symptom screen prior to the visit (per the MOP), viral testing may be performed during this study visit with a research-based assay
- Obtain interim medical history, including an assessment for new medical conditions by interview of subjects and note any changes since the previous clinic visit.
- Record all concomitant medications on the appropriate eCRF.
- Obtain vital signs including oral temperature, blood pressure, and pulse.
- Perform a targeted physical examination, if indicated based on review of complete medical history.
- Examine vaccination site for local reactions and determine whether information recorded by subjects on the memory aid conforms to examination.
- Collect blood for immunogenicity (RFFIT assay), T cell (IFN-gamma ELISPOT and ICS), and developer assays.
- Collect blood samples for PBMCs for future use, if the subject has provided consent.
- Assess all AE/SAEs and new onset chronic medical conditions and record them on the appropriate eCRF.

7.4.7. Phone Call, Visit 08, Day 50 (window +7 days)

- Obtain interim medical history, including an assessment for new medical conditions by telephone interview of subjects and note any changes since the previous clinic visit.
- Record all concomitant medications taken through 28 days after the last study vaccination on the appropriate eCRF.
- Assess all AEs that occurred through 28 days after the last study vaccination and record them on the appropriate eCRF.
- Assess all SAEs and new onset chronic medical conditions and record them on the appropriate eCRF.

7.4.8. Visit 09, Day 91 (window ±14 days)

- Obtain interim medical history, including an assessment for new medical conditions by interview of subjects and note any changes since the previous clinic visit.
- Obtain vital signs including oral temperature, blood pressure, and pulse.

- Perform a targeted physical examination, if indicated based on review of complete medical history.
- Collect blood for immunogenicity (RFFIT assay), MBC, and T cell assays (IFN-gamma ELISPOT).
- Collect blood samples for PBMCs for future use, if the subject has provided consent.
- Assess all SAEs and new onset chronic medical conditions and associated concomitant medications and record them on the appropriate eCRF.

7.4.9. Visit 10, Day 181 (window ±14 days)

- Obtain interim medical history, including an assessment for new medical conditions by interview of subjects and note any changes since the previous clinic visit.
- Obtain vital signs including oral temperature, blood pressure, and pulse.
- Perform a targeted physical examination, if indicated based on review of complete medical history.
- Collect blood for immunogenicity (RFFIT assay) and MBC assays.
- Collect blood samples for PBMCs for future use, if the subject has provided consent.
- Assess all SAEs and new onset chronic medical conditions and associated concomitant medications and record them on the appropriate eCRF.

7.5. Final Study Visit

7.5.1. Visit 11, Day 381 (window ±14 days)

- Obtain interim medical history, including an assessment for new medical conditions by interview of subjects and note any changes since the previous clinic visit.
- Obtain vital signs including oral temperature, blood pressure, and pulse.
- Perform a targeted physical examination, if indicated based on review of complete medical history.
- Collect blood for immunogenicity (RFFIT assay), MBC, and T cell assays (IFN-gamma ELISPOT).
- Collect blood samples for PBMCs for future use, if the subject has provided consent.

- Assess all SAEs and new onset chronic medical conditions and associated concomitant medications and record them on the appropriate eCRF.

7.6. Early Termination Visit

The following activities will be performed at the early termination visit for subjects who withdraw, or are withdrawn or terminated from this trial:

- Obtain interim medical history, including an assessment for new medical conditions by interview of subjects and note any changes since the previous clinic visit or phone call.
- Record all concomitant medications on the appropriate eCRF (if before 28 days after the last vaccination).
- Obtain vital signs including oral temperature, blood pressure, and pulse.
- Perform a targeted physical examination, if indicated based on review of complete medical history.
- Examine vaccination site for local reactions if visit occurs ≤28 days after last injection.
- Review memory aid if visit occurs ≤15 days after last injection.
- Obtain labs (safety and immunogenicity) appropriate to that visit if visit occurs within window of a regular study visit.
- Perform a urine pregnancy test on all female subjects of childbearing potential (if indicated)
- Assess patient for AE/SAEs and new onset chronic medical conditions and associated concomitant medications and record on the appropriate eCRF (AEs only through Day 28).

7.7. Unscheduled Visit

Unscheduled visits may occur at any time during this trial. Labs may be drawn at PI discretion. Any of the following activities may be performed:

- If an unscheduled visit occurs ≤14 days after any study vaccination, the subject will undergo a symptom screen prior to the visit, and if the screen is positive (per the MOP), viral testing may be performed during this visit with a research-based assay
- Obtain interim medical history, including an assessment for new medical conditions by interview of subjects and note any changes since the previous clinic visit or phone call.

- Record all concomitant medications on the appropriate eCRF (if before 28 days after the last vaccination).
- Obtain vital signs including oral temperature, blood pressure, and pulse.
- A targeted physical examination will be performed, if needed, by a study clinician licensed to make medical diagnoses and listed on Form FDA 1572 as the site PI or sub-investigator, if indicated based on review of complete medical history.
- Examine vaccination site for local reactions if visit occurs ≤28 days after any vaccination, and if so, determine whether information recorded by subjects on the memory aid conforms to examination.
- Obtain blood for safety if needed.
- Perform a urine pregnancy test on all female subjects of childbearing potential (if indicated)
- Assess all AE/SAEs and new onset chronic medical conditions and associated concomitant medications and record them on the appropriate eCRF.

8. STUDY PROCEDURES/EVALUATIONS

8.1. Clinical Evaluations

Complete medical history will be obtained by interviewing the subjects at the first study visit. Subjects will be queried regarding a history of significant medical disorders of the head, eyes, ears, nose, throat, mouth, cardiovascular system, lungs, gastrointestinal tract, liver, pancreas, kidney, urologic system, nervous system, blood, lymph nodes, 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. At subsequent follow-up visits, an interim medical history will be obtained by interview of the subjects noting any changes since the previous clinic visit or phone call. The interim medical history will include an assessment for new medical conditions and symptoms.

Concomitant medications will be collected as described in Section 6.4.

At the first screening visit (Visit 00A), a physical examination (excluding pelvic and rectal examinations) will be performed by a study clinician licensed to make medical diagnoses and listed on Form FDA 1572 as the site PI or sub-investigator. For all subsequent visits, a targeted physical examination may be performed by a study clinician licensed to make medical diagnoses and listed on Form FDA 1572 as the site PI or sub-investigator, if indicated based on subject's interim medical history.

Vital signs (oral temperature, pulse, and blood pressure) will be collected through Visit 11. Vital signs assessed on the screening visit will be considered as baseline. Subjects must not eat or drink anything hot or cold, or smoke within 10 minutes before taking oral temperature. In the event of an abnormal heart rate or blood pressure due to physiological variation or activity, the subject may rest for 10 minutes in a quiet room, and then blood pressure and/or heart rate may be re-measured. The repeated measurement may be used to determine eligibility per the judgement of the investigator.

Height and weight will be collected on the screening visit to determine BMI.

Pre-administration reactogenicity assessments will be performed before each vaccination. A subject with mild (Grade 1) pre-administration reactogenicity which is transient, resolving, or clinically insignificant may be enrolled at the investigator's discretion.

Subjects will be observed in the clinic for ≥30 minutes after each vaccination. The vaccination site will be examined, post-administration reactogenicity assessments will be performed, and any AE/SAEs will be assessed and recorded on the appropriate eCRF before discharge from the clinic.

All subjects will complete a subject memory aid on the day of each vaccination through Day 7 following each vaccination. Subject memory aids will be reviewed with the subjects for AEs

(solicited injection site and systemic reactions, and unsolicited AEs). In addition, after each vaccination, whenever the subject is seen in the clinic or a phone call occurs (i.e., at all opportunities), solicited and unsolicited reactogenicity and AEs will be reviewed.

Reactogenicity assessments will include an assessment of solicited AEs occurring from the time of each vaccination through Day 7 following each vaccination, which include injection site reactions (pruritus, erythema, ecchymosis, induration/swelling, pain, and tenderness) and systemic reactions (fever, chills/shivering/sweating, fatigue, malaise, myalgia and arthralgia (exclusive of the injection site), headache, and nausea).

8.2. Laboratory Evaluations

8.2.1. Clinical Laboratory Evaluations

Serum pregnancy tests will be performed by the clinical laboratory at the first screening visit (Visit 00A). Urine pregnancy tests will be performed by the site laboratory within 24 hours before each vaccination (Day 1 (Visit 01), Day 8 (Visit 03), Day 15 (Visit 04) and Day 22 (Visit 06)) on all female subjects of childbearing potential. Results must be negative and known before randomization on Day 1 (Visit 01) and before receipt of any vaccination.

To be eligible for participation in this trial and receipt of the first vaccine, the subject's clinical screening lab evaluations must be confirmed to meet the subject inclusion criteria (see Section 5.1). WBCs, hemoglobin, platelets, absolute neutrophil count, absolute lymphocyte count, ALT, AST, total bilirubin BUN, creatinine, and urine dipstick for glucose and protein will be measured as part of screening.

Subjects will be screened for HIV-1/2 antibody, hepatitis B surface antigen (HBsAg), and hepatitis C virus (HCV) antibody. If a positive result occurs, the subject will be referred for appropriate follow-up and results reported as required by state law. These screening tests must be negative for the subject to be eligible to participate.

Clinical safety lab parameters evaluated after receipt of vaccine (on Days 2, 8, 16, and Day 22) will include WBCs, hemoglobin, platelets, absolute neutrophil count, absolute lymphocyte count, ALT, AST, total bilirubin, BUN, and creatinine.

8.2.2. Special Assays

Immunogenicity Assays

8.2.2.1. Rabies virus neutralization assay

The RFFIT is a serum neutralization assay performed on chamber slides. The RFFIT has a 24-hour incubation period for virus-serum-cell and includes a 5-fold serial dilution of the serum. The RFFIT reading is virus positive counts per microscopic field (20 fields per well). In the Kansas State University (KSU) rabies laboratory, the RFFIT is fully validated per FDA and ICH

guidelines for human testing. The laboratory is CLIA certified. The lab's RFFIT is approved by most rabies-free areas including the EU (https://ec.europa.eu/food/animals/pet-movement/approved-labs_en) and is recognized by the USDA for rabies serology testing for pet export.

RFFIT assays will be performed on Days 1, 8, 15, 22, 29, 91, 181, and 381 (Visits: 1, 3, 4, 6, 7, 9, 10, and 11).

8.2.2.2. Cell-mediated immunity assays

IFN-gamma ELISPOT assays will be performed on Days 1, 15, 29, 91, and 381 (Visits: 1, 4, 7, 9, and 11).

ICS will be run at peak of T cell response, which is expected on Day 15 for subjects assigned to Groups A and B and on Day 29 for subjects assigned to Group C.

MBC ELISPOT will be done at Days 1, 91, 181 and 381 (Visits 1, 9, 10 and 11).

8.2.2.3. Developer assays

Blood will be collected for the vaccine developer to conduct assays that may include early vaccine responses, multi-plex protein assays, and anti-vector antibody assays. Blood (PAXgene tubes) for early vaccine responses will be collected on Days 1, 2, 8, 15, 16, 22, and 29. Blood for multi-plex protein assays will be collected on Days 1, 2, 8, 15, 16, 22 and 29. These assays are to support the design of future studies with this vaccine. Data from these assays will not be collected as part of this study. Blood for anti-vector antibody assays will be collected on Days 1 and 29. Anti-vector antibody data will be collected as a exploratory endpoint.

8.2.2.4. Virus/SARS-CoV-2 testing

Subjects that have a positive symptom screen prior to Visits 02, 03, 04, 05, 06, and 07 (per the MOP), may be tested for SARS-CoV-2 using a research-based assay. Samples may be collected during or after visits on Days 2, 8, 15, 16, 22, and 29. If a positive result occurs, the subject will be referred for appropriate follow-up and results reported as required by state law.

8.2.3. Specimen Preparation, Handling, and Shipping

PBMC samples for T cell assays will be collected on Days 1, 15, 29, 91, 181, and 381. For samples scheduled to be collected during vaccination visits, samples will be collected before vaccination. Serum for antibody assays will be collected on Days 1, 8, 15, 22, 29, 91, 181, and 381. Plasma from the PBMC separations will be saved as described in the MOP.

8.2.3.1. Instructions for Specimen Preparation, Handling, and Storage

Instructions for specimen preparation, handling, and storage are included in the MOP.

8.2.3.2. Specimen Shipment

Specimen shipment will occur at intervals during this trial following all applicable International Air Transport Association (IATA) requirements and according to the specifics for storage temperature and documentation as detailed in the MOP.

9. ASSESSMENT OF SAFETY

9.1. Specification of Safety Parameters

Safety will be assessed by the frequency and/or severity of:

- Solicited injection site and systemic reactogenicity events from the time of each vaccination through Day 7 after each vaccination, overall and in each dose group.
- Serious SAEs considered study vaccine-related and reported at any time after the first vaccination through the end of this trial, overall and in each dose group.
- Vaccine-related lab AEs through study Day 22, overall and in each dose group.
- Unsolicited vaccine-related AEs from the time of the first vaccination through Day 28 after the last vaccination, overall and in each dose group.
- New onset chronic medical conditions at any time after the first vaccination and through the end of this trial, overall and in each dose group.
- Local, systemic, or lab toxicities after any vaccination, overall and in each dose group.
- Local reactogenicity through Day 7 after any vaccination, overall and in each dose group.
- Any SAEs, overall and in each dose group.

9.2. Methods and Timing for Assessing, Recording, and Analyzing Safety Parameters

9.2.1. Adverse Events

ICH E6 defines an AE as any untoward medical occurrence in a patient or clinical investigation subject administered a study product regardless of its causal relationship to the study product administration. FDA defines an AE as any untoward medical occurrence associated with the use of a study product in humans, whether or not considered study product-related.

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

AEs, including solicited local (injection site) and systemic (subjective and quantitative) reactions, will be captured on the appropriate data collection form and eCRF. Information to be collected for unsolicited non-serious AEs includes event description, date of onset, assessment of severity, relationship to study product, 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. AEs occurring during this trial and reporting period will

be documented appropriately regardless of relationship to study product. AEs 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 should be recorded as an AE.

All AEs will be graded for severity and relationship to study product (see definitions). AEs 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.

Severity of Event:

AEs will be assessed by the investigator using a protocol-defined grading system (toxicity table included as an appendix). For events not included in the protocol-defined grading system, the following guidelines will be used to quantify severity:

Mild (Grade 1): Events require minimal or no treatment; do not interfere with the subject's daily activities.

Moderate (Grade 2): Events result in a low level of inconvenience or concern with therapeutic measures; may cause some interference with functioning and daily activities.

Severe (Grade 3): Events interrupt the subject's daily activities and may require systemic drug therapy or other treatment; are usually incapacitating.

Changes in the severity of an AE should be documented to allow an assessment of the duration of the event at each level of intensity to be performed.

Relationship to Study Products:

The assessment of the relationship of an AE to administration of a study product is made only by those with the training and authority to make a diagnosis and listed on Form FDA 1572 as an investigator based on all available information at the time of the completion of the eCRF. Whether the AE is related or not is not a factor in determining what is or is not reported in this trial. If there is any doubt as to whether a clinical observation is an AE, the event should be reported. Solicited injection site AEs reported during the reporting period will be considered related.

In a clinical trial, the study product must always be suspect. To help assess relatedness, the following guidelines are used.

Related – There is a reasonable possibility that the study product caused the AE; that is, there is evidence to suggest a causal relationship between the study product and the AE.

Not Related – There is not a reasonable possibility that the administration of the study product caused the AE.

9.2.2. Reactogenicity

Reactogenicity events are AEs that are common and known to occur following administration of vaccines. The Toxicity Table (Appendix C) will be used to grade solicited local (injection site) and systemic (subjective and quantitative) reactions.

9.2.3. Serious Adverse Events

Serious Adverse Event (SAE):

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

Death,

- a life-threatening AE
- inpatient hospitalization or prolongation of existing hospitalization,
- a persistent or significant incapacity or substantial disruption of the ability to conduct normal life functions, or
- a congenital anomaly/birth defect.

Important medical events that may not result in death, be life-threatening, or require hospitalizations may be considered serious when, based upon appropriate medical judgment they may jeopardize the patient or subject and may require medical or surgical intervention to prevent one of the outcomes listed in this definition. Examples of such medical events include allergic bronchospasm requiring intensive treatment in an emergency room or at home, blood dyscrasias or convulsions that do not result in inpatient hospitalization, or the development of drug dependency or drug abuse.

An AE is considered "life-threatening" if, in the view of either the site PI or sponsor, its occurrence places the patient or subject at immediate risk of death. It does not include an AE, had it occurred in a more severe form, might have caused death.

SAEs will be:

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

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

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

The site PI or appropriate sub-investigator is responsible for recording all AE/SAEs that are observed or reported during this trial, regardless of the relationship to the vaccine. AE/SAEs or abnormal clinical findings will be collected, assessed, documented, reported, and followed appropriately.

9.3. Reporting Procedures

9.3.1. Serious Adverse Events

SAEs 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 will be submitted immediately (within 24 hours of site awareness) on an SAE form to the DMID Pharmacovigilance Group, at the following address:

DMID Pharmacovigilance Group

Clinical Research Operations and Management Support (CROMS)

6500 Rock Spring Dr., Suite 650

Bethesda, MD 20817, USA

SAE Hot Line: 1-800-537-9979 (US) or 1-301-897-1709 (outside US)

SAE FAX Number: 1-800-275-7619 (US) or 1-301-897-1710 (outside US)

SAE Email Address: PVG@dmidcroms.com

In addition to the SAE form, select SAE data fields will also be entered into Advantage EDC^{SM}. Please see the MOP for details regarding this procedure.

Other supporting documentation of the event may be requested by the DMID Pharmacovigilance Group and should be provided as soon as possible.

The site will send a copy of the SAE report(s) to the ISM (as deemed necessary) when they are provided to the DMID Pharmacovigilance Group. The DMID Medical Monitor and DMID CPM will be notified of the SAE by the DMID Pharmacovigilance Group. The DMID Medical

Monitor 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 this trial, if the site PI or appropriate sub-investigator becomes aware of an SAE that is suspected to be related to the vaccine, the site PI or appropriate sub-investigator will report the event to the DMID Pharmacovigilance Group.

9.3.2. Regulatory Reporting for Studies Conducted Under DMID-Sponsored IND

Following notification from the investigator, DMID, the Investigational New Drug (IND) sponsor, will report any suspected adverse reaction that is both serious and unexpected. DMID will report an AE as a suspected adverse reaction only if there is evidence to suggest a causal relationship between the vaccine and the AE. DMID will notify FDA and all participating investigators (i.e., all investigators to whom the sponsor is providing vaccine under its IND(s) or under any PI's IND(s)) in an IND safety report of potential serious risks from clinical trials or any other source, as soon as possible, but in no case later than 15 calendar days after the sponsor determines that the information qualifies for reporting as specified in 21 CFR Part 312.32. DMID will also notify FDA of any unexpected fatal or life-threatening suspected adverse reaction as soon as possible, but in no case later than 7 calendar days after the sponsor's initial receipt of the information. DMID will also notify FDA if the study is halted for a safety concern as soon as possible, but in no case later than 48 hours after the sponsor's initial receipt of the information. Relevant follow up information to an IND safety report will be submitted as soon as the information is available. Upon request from FDA, DMID will submit to FDA any additional data or information that the agency deems necessary, as soon as possible, but in no case later than 15 calendar days after receiving the request.

All serious events designated as "not related" to vaccine(s), will be reported to the FDA at least annually in a summary format.

9.3.3. Reporting of Pregnancy

Pregnancies occurring in subjects will be reported via Advantage eClinical^{SM} on the Pregnancy Report form. No further study vaccinations will be administered to pregnant subjects, but with the subject's permission all study-related blood samples will be obtained and the subject will continue in follow-up for safety events. Efforts will be made to follow all pregnancies reported during this trial to pregnancy outcome pending the subject's permission. The suggested follow up schedule is detailed in the MOP.

9.4. Type and Duration of Follow-up of Subjects after Adverse Events

Solicited injection site and systemic reactogenicity events will be documented and reported from Day 1 through Day 8.

Clinical safety labs will be collected at baseline and at Days 2, 8, 16 and 22. Solicited clinical safety lab results will be documented in Advantage eClinical in the Local Laboratory Results eCRF for these days. Abnormal clinical safety laboratory results for solicited laboratory parameters ARE NOT reported as unsolicited AEs on the Adverse Event eCRF.

Unsolicited AEs will be collected and assessed through 28 days after the final vaccination. Unsolicited AEs are followed through resolution even if this extends beyond the reporting period (approximately 28 days after the last vaccination).

SAEs and new-onset chronic medical conditions will be collected, assessed, and followed from the time of the first vaccination through approximately 12 months after the last vaccination or until resolution or stabilization.

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

Follow-up procedures, evaluations, and outcomes will be recorded on the appropriate eCRF.

9.5. Halting Rules

This trial may be halted due to ISM, SRC, SMC, or PI review or at the discretion of DMID.

Halting Rules for Sentinel Subjects (safety reviews through Day 8, which occurs 7 days after first study vaccination)

If any of the following halting rules are met by the first group of sentinel subjects, this trial will not proceed with dose escalation to the second group of sentinel subjects without a DMID-approved SMC recommendation to proceed.

- One or more subjects experiences a study vaccine-related Grade 3 solicited systemic AE.
- One or more subjects experiences a study vaccine-related Grade 3 unsolicited AE.
- One or more subjects experiences a study vaccine-related Grade 2 or greater laboratory AE.
- One or more subjects experiences a study vaccine-related Grade 3 solicited local injection site AE.
- One or more subjects experiences injection site ulceration, abscess, or necrosis judged to be related to study vaccine.
- One or more subjects experiences laryngospasm, bronchospasm, anaphylaxis judged to be related to study vaccine within 1 day of vaccination.

- One or more subjects experiences an allergic reaction (e.g., generalized urticaria) judged to be related to study vaccine within 3 days of vaccination.
- Any death occurs following vaccination.
- Any subject experiences a non-fatal SAE judged to be related to study vaccine.

Halting Rules for Discontinuation of an Individual Subject from Additional Vaccination

The following assessments should be performed before an additional vaccination to determine if the subject remains eligible. If any of the following criteria are met, a subject will not be given subsequent vaccinations:

- Allergic reaction (e.g., generalized urticaria) within 3 days after vaccination
- Anaphylaxis, laryngospasm, or bronchospasm within 1 day after vaccination
- An SAE judged to be related to study vaccine
- A Grade 3 lab AE that is considered to be related to any of the study vaccines
- A Grade 3 unsolicited AE that is considered to be related to any of the study vaccines
- A Grade 3 solicited injection site AE that does not resolve or decrease to a lower grade (the measurement of ecchymosis, erythema or induration by itself will not be used as halting criteria) within 2 days.
- A Grade 3 solicited systemic AE (except fatigue, malaise and headache) considered to be related to any of the study vaccines
- A Grade 3 solicited systemic AE of fatigue, malaise or headache considered to be related to the study vaccines, that does not resolve or decrease to a lower grade within 2 days.
- Female subject becomes pregnant
- Subject develops a new illness or condition that meets the Exclusion Criteria
- Subject develops a new medical condition or medication change for which continued participation, in the opinion of the investigator, would pose a risk to the subject, would interfere with the subject's ability to complete this trial, or would be likely to confound interpretation of the results
- Subject is no longer willing and/or able to adhere to study restrictions outlined in the Inclusion and Exclusion Criteria

- Subject has a fever (defined as an oral temperature ≥38.0 °C) or an acute illness at the time of boost vaccination administration. The next study vaccination can be deferred until the fever or illness resolves or only minor residual symptoms remain, that in the opinion of the investigator, will not interfere with the ability to assess safety parameters as required by the protocol, provided that the boost is given within the protocol-defined study window. Permission to administer vaccination outside the protocol-defined study window must be obtained from DMID Medical Officer
- Subject has a Grade 2 or greater solicited injection site or systemic AE at the time of boost vaccination administration. The next study vaccination can be deferred until the AE resolves or decreases to a lower grade, such that in the opinion of the investigator, it will not interfere with the ability to assess safety parameters as required by the protocol, provided that the boost is given within the protocol-defined study window

Note: In case the boost vaccination is postponed, the timing of the safety/immunogenicity visits post-boost will be planned relative to actual vaccination day.

Halting Rules for Discontinuing Further Enrollments and Vaccinations Across this Trial

If one or more of the following halting rules is met, further enrollment and vaccinations will be withheld until the SMC reviews the safety data:

- If any subject experiences anaphylaxis, laryngospasm, bronchospasm within 24 hours after study vaccination
- If two or more subjects experience an allergic reaction (e.g. generalized urticaria) within 3 days after study vaccination
- If any subject experiences injection site ulceration, abscess, or necrosis judged to be related to study vaccine
- If any subject experiences a study vaccine-related SAE
- If any subject experiences any life-threatening or fatal SAE without an obvious, alternative explanation (such as trauma)
- If any subject experiences thrombosis (arterial or venous) through study Day 50
- If two or more subjects experience the same study vaccine-related Grade 3 laboratory AE.
- If two or more subjects experience the same study vaccine-related Grade 3 unsolicited AE

- If two or more subjects experience the same study vaccine-related Grade 3 solicited local injection site AE (except measured ecchymosis, erythema and induration) that does not resolve or decrease to a lower grade within 2 days
- If two or more subjects experience the same study vaccine-related Grade 3 solicited systemic AE (except fatigue, malaiase and headache) that does not resolve or decrease to a lower grade within 1 day
- If two or more subjects experience the same study vaccine-related Grade 3 solicited systemic AE of fatigue, malaise or headache that does not resolve or decrease to a lower grade within 2 days

9.6. Safety Oversight

9.6.1. Safety Review Committee (SRC)

The SRC will be composed of:

- PI, or designee
- Independent Safety Monitor (ISM)
- DMID Medical Monitor (MM) or designee;

The SRC will be responsible for reviewing halting criteria notifications in real time. If a halting rule is met, the SRC will meet for safety review and assessment to confirm the event. If any of the study halting criteria are confirmed, the SMC (section 9.6.2) will meet to evaluate the data and recommend appropriateness of further enrollments and/or vaccinations. The SRC will remain blinded as to treatment assignment.

9.6.2. Safety Monitoring Committee (SMC)

This clinical study will utilize an SMC, which is an independent group of experts that advises DMID. The primary responsibility of the SMC is to monitor subject safety. The SMC is external to DMID and comprises at least three voting members. The SMC will consist of members with appropriate Phase 1 study expertise to help interpret data from this trial. Its activities will be delineated in a SMC charter that will describe the membership, responsibilities, and scope and frequency of data reviews. The SMC will operate on a conflict-free basis independently of the study team. The DMID or SMC may convene *ad hoc* SMC meetings according to protocol criteria or if concerns arise during this trial.

The SMC will review the safety data at the following milestones:

- Organizational Meeting – conducted prior to the enrollment of study participants

- Scheduled Data Review Meetings – a preplanned, scheduled data review meeting – a scheduled meeting will take place to review cumulative data up to day 8 after dosing of the sentinel subject and recommend whether to proceed to enrollment of non-sentinel subjects

- Ad hoc meetings – an unplanned meeting convened for a specific safety concern – the committee will convene to review events that meet halting rules for sentinel and non-sentinel subjects

- Final Meeting – to review cumulative safety data (6 months following database lock)

All reviews by the SMC will be performed using blinded data initially (open session) and if necessary for safety of subject unblinded data will be reviewed in closed session. Vaccination data, including dose interruptions, modifications, and the associated reason(s), will be reported to the SMC.

9.6.3. Independent Safety Monitor (ISM)

An Independent Safety Monitor (ISM) will be assigned for the study site. An ISM is a physician with relevant expertise whose primary responsibility is to provide to DMID an independent safety assessment in a timely fashion. Participation is for the duration of the study and is a voluntary position that does not receive payment. The ISM must meet the requirements of the NIAID conflict of interest policy.

The ISM:

- Is in close proximity to the study site and has the authority and ability to readily access study participant records in real time.
- May be a member of the participating institution's staff but preferably be from a different organizational group within the institution.
- Should not be in a direct supervisory relationship with the investigator.
- Should have no direct involvement in the conduct of the study.

The ISM will:

- Sign a Conflict of Interest (COI) certification at the time they are asked to participate and provide updates to this information as needed.
- Receive reports of SAEs from the site PI and will be notified by email when DMID is notified of the SAE.
- Evaluate the SAE and report their clinical assessment to DMID, through DMID-CROMS SOCS in a timely manner and email the report to DMID-CROMS SOCS.
- Communicate with the investigator at the participating site as needed.

- Review additional safety related events at the request of DMID.
- Provide additional information to DMID and/or the SMC by teleconference as requested.

10. CLINICAL MONITORING

10.1. Site Monitoring Plan

Site monitoring is conducted to ensure that the human subjects' protections, study and laboratory procedures, and data collection processes are of high quality and meet sponsor requirements, ICH/GCP guidelines, and applicable regulations, and that this trial is conducted in accordance with the protocol, MOP, and applicable sponsor SOPs. The sponsor, DMID, 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 DMID, and may be made more frequently as directed by DMID. 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 site, study personnel, and all study documentation according to the DMID-approved site monitoring plan. Study monitors will meet with the site PI to discuss any problems and actions to be taken, and will document site visit findings and discussions.

11. STATISTICAL CONSIDERATIONS

11.1. Study Hypotheses

There are two primary objectives for this trial: to assess the safety, tolerability, and reactogenicity of one dose of ChAd155-RG at $5x10^{10}$vp per dose, or one or two doses of ChAd155-RG at $1x10^{11}$vp per dose; and to compare the safety, tolerability, and reactogenicity of one or two doses of ChAd155-RG with three doses of RABAVERT. A secondary goal is to assess serum rabies VNA levels by a standard, WHO-approved, RFFIT, as assessed by immune response kinetics (through 12 months after the first dose of vaccination), seroconversion rates, and peak GMT. Also of interest are innate T cell and B cell responses, as well as ELISA binding antibodies. This trial, like other Phase I trials, is exploratory rather than confirmatory, and was not designed to test a formal null hypothesis.

11.2. Sample Size Considerations

This trial will enroll 50 subjects into four treatment arms: Group A (N=14) will receive one dose of ChAd155-RG at the lower dosage (5×10^{10}vp); Group B (N=14) will receive one dose of ChAd155-RG at the higher dosage (1×10^{11}vp); Group C (N=10) will receive two doses of ChAd155-RG at the higher dosage ($1x10^{11}$vp); and Group D (N=12) will receive three doses of RABAVERT. Randomization and safety, tolerability, and reactogenicity monitoring will be conducted as described in Section 5.3.1and Section 9.6.1 respectively.

11.2.1 Adverse Events

The sample size of 10-14 subjects in each treatment arm is small given the early stage (Phase I) of the product's development; thus, the precision of estimates for AEs is limited. Rare AEs associated with dose level or number of doses are not demonstrable in a trial of this size; however, the probabilities of observing one or more AEs within a treatment arm given various true event rates are presented in Table 2. The minimum detectable event rates for various levels of power are displayed in Table 3.

Table 2: Probability of Observing an Adverse Event for Various Event Rates and Sample Sizes

N	"True" Event Rate	Probability of Observation (%)	N	"True" Event Rate	Probability of Observation (%)
8	0.1 %	0.8	12	0.1%	1.2
	0.5 %	3.9		0.5%	5.8
	1.0 %	7.7		1.0%	11.4
	2.0 %	14.9		2.0%	21.5
	3.0 %	21.6		3.0%	30.6
	4.0 %	27.9		4.0%	38.7
	5.0 %	33.7		5.0%	46.0
	10.0 %	57.0		10.0%	71.8
	20.0 %	83.2		20.0%	93.1
10	0.1%	1.0	14	0.1%	1.4
	0.5%	4.9		0.5%	6.8
	1.0%	9.6		1.0%	13.1
	2.0%	18.3		2.0%	24.6
	3.0%	26.3		3.0%	34.7
	4.0%	33.5		4.0%	43.5
	5.0%	40.1		5.0%	51.2
	10.0%	65.1		10.0%	77.1
	20.0%	89.3		20.0%	95.6

Table 3: Minimum Detectable Event Rates for Various Levels of Power and Sample Size

N	Desired Power Level	Detectable Event Rate (%)	N	Desired Power Level	Detectable Event Rate (%)
8	0.80	18.2	12	0.80	12.6
	0.90	25.0		0.90	17.5
	0.95	31.2		0.95	22.1
	0.99	43.8		0.99	31.9
10	0.80	14.9	14	0.80	10.9
	0.90	20.6		0.90	15.2
	0.95	25.9		0.95	19.3
	0.99	36.9		0.99	28.0

11.2.2 Immunogenicity

This trial was not designed to formally test any hypotheses associated with the immunogenicity data. The 95% confidence intervals for multiple observed seroconversion rates at various sample sizes were calculated assuming observed seroconversion rates of 90%, 95%, and 100%, as shown in Table 4.

Table 4: 95% Confidence Intervals for Seroconversion Rate for Various Rates and Sample Sizes

N	Seroconversion Rate	95% CI	N	Seroconversion Rate	95% CI
8	90%	(50 %, 100 %)	12	90%	(59 %, 100 %)
	95%	(56 %, 100 %)		95%	(66 %, 100 %)
	100%	(63 %, 100 %)		100%	(74 %, 100 %)
10	90%	(55 %, 100 %)	14	90%	(62 %, 99 %)
	95%	(62 %, 100 %)		95%	(69 %, 100 %)
	100%	(69 %, 100 %)		100%	(77 %, 100 %)

11.3. Planned Interim Analyses

For purposes of planning subsequent research and to allow internal and external presentations of data (e.g., national and/or international conferences and early publication of data), there will be one planned interim safety and immunogenicity analysis. Results of the interim analysis will not be used to make any decisions concerning the conduct of this trial.

Once at least half the subjects complete the Day 91 visit the clinical database will be cleaned, monitored, and a data freeze created. Unblinded analyses of safety, reactogenicity, and available immunogenicity data are planned. The interim report will be prepared by the SDCC after the interim clinical database is frozen and all VNA data through Day 91 for subjects for whom data are available at the time of database freeze are received.

Investigators will remain blinded to individual subject treatment assignment, and will be provided only data summaries aggregated by dose-group or blinded by-subject data.

For the purpose of advancing from the two groups of sentinel subjects to enrollment of the remainder of subjects, the SMC will review safety data. The halting rules described elsewhere in this protocol (Section 9.5) will be utilized to pause this trial if certain criteria are met.

11.3.1. Interim Safety Review

Safety data for the first set of four sentinel subjects will not be reviewed by the SMC before enrollment of the second set of four sentinel subjects if no pre-defined halting rule is met. Safety data for all 8 sentinels will be reviewed by the SMC prior to enrollment of the remaining 42 subjects. These sentinel reviews will not involve hypothesis testing and will not be considered in estimating the precision of any estimates made at the conclusion of this trial.

The interim analysis will include all safety and immunogenicity data through Day 91 for subjects for whom data are available at the time of database freeze. The interim safety analyses will be specified in the Statistical Analysis Plan and may include tables summarizing demographics, study status, and protocol adherence information. Tables and figures will present unsolicited adverse events by MedDRA System Organ Class (SOC) and Preferred Team (PT), severity, and relationship to study product. Solicited adverse events will be summarized by symptom, severity, and study day. Laboratory results will be summarized by parameter, severity, and study day. Blinded listings of early terminations, protocol deviations, and clinical data may be included. Since this early analysis of the data is not intended to impact the conduct of this trial, it has no impact on Type I error and adjustments are not planned.

11.3.2. Interim Immunogenicity Review

The interim analysis will include all available immunogenicity (VNA-RFFIT) data through Day 91 for the subjects for whom data are available at the time of database freeze, respectively addressing secondary objective. The interim immunogenicity analyses will include summaries of geometric mean titers (GMTs) and their associated 95% confidence intervals (CIs) as well as peak GMT by treatment arm at study timepoints outlined in Appendix A. Titer results will also be displayed in reverse cumulative distribution (RCD) curves by visit and treatment arm. No hypothesis tests or modeling will be included in the interim immunogenicity analysis. Since this early analysis of the data is not intended to impact the conduct of this trial, it has no impact on Type I error and adjustments are not planned.

11.4. Final Analysis Plan

The primary Clinical Study Report (CSR) will summarize all safety and protocol-specified humoral immunogenicity results, including all primary and secondary endpoint data, collected through the final study visit for the last subject. Prior to the interim database freeze, a formal Statistical Analysis Plan will be developed that specifies all planned analyses of primary and secondary endpoints, and indicates which analyses will be included in the interim analyses and primary CSR . In the following an abbreviated version of the planned primary and secondary analysis is provided. Additional exploratory humoral and cellular immunogenicity assessments may be summarized in one or more addenda to the main CSR.

11.4.1 Analysis Populations

Intent to Treat Population

The modified intent-to-treat (mITT) population includes all subjects who received at least one dose of study vaccine and contributed both pre- and at least one post-study vaccination venous blood samples for immunogenicity testing for which valid results were reported. For analyses using the mITT population, subjects will be grouped based on randomized treatment arm.

Per Protocol Population

The per protocol (PP) population includes all subjects in the mITT subset with the following exclusions:

- Data from all available visits for subjects found to be ineligible at baseline, according to the inclusion and exclusion criteria at the time of the subject's enrollment.
- Data from subjects who have a baseline rabies VNA concentration ≥ 0.5 IU/mL (for the immunogenicity analyses)

- Data from all visits subsequent to the following, such as:
    - Study withdrawal or treatment discontinuation,
    - Any study vaccination dose received out of window,
    - Receipt of non-study licensed live vaccine within 30 days before or after each study vaccination,
    - Receipt of non-study licensed inactivated vaccine within 14 days before or after each study vaccination,
    - Receipt of immunosuppressive therapy (e.g., corticosteroids) within 30 days before or after each study vaccination.
    - Receipt of chloroquine or hydroxychloroquine
- Data from any visit that occurs out of window, as defined in Appendix A "Window for each study visit".

Safety Population

The Safety Population will consist of all subjects who have received at least one dose of vaccine and for whom any data on safety are available. Subjects will be classified according to the vaccine and dosage received. The primary safety analysis will be performed using this population.

11.4.2 Baseline Characteristics

Baseline characteristics overall and within each treatment arm will be summarized. For both continuous and categorical variables, appropriate summary statistics will be applied. For continuous variables, descriptive statistics will include the number of non-missing values, mean, standard deviation, median, minimum, and maximum. For categorical variables, descriptive statistics will include counts and percentages per category.

11.4.3 Safety Analysis Plan

Safety evaluations will be based on the incidence, severity and type of AEs. Clinically significant physical examination findings and vital signs will be documented on an AE case report form. Safety variables will be tabulated and presented for all subjects in the safety population, overall and for each treatment arm.

AEs will be coded by the Medical Dictionary for Regulatory Activities (MedDRA) for preferred term and SOC. The number and percentage of subjects experiencing each specific AE, along with 95% confidence intervals, will be tabulated by severity and by relatedness to vaccine. The number of AEs and SAEs will be reported by a detailed listing showing the type, MedDRA

coding, relevant dates (administration and AE), severity, relatedness, and outcome for each event.

Solicited local injection site and systemic reactogenicity will be summarized by severity for each day post vaccination (Days 1-7 post each vaccination) and as the maximum severity over all 7 days. Additionally, solicited AEs will be analyzed by taking the most severe response over the follow-up period, dichotomizing into a binary variable (none versus mild, moderate, or severe) and using exact confidence intervals to summarize the proportion of subjects reporting each symptom, any injection site symptom, and any systemic symptom. Solicited AEs will be summarized separately for each vaccination, and over all vaccinations by treatment arm.

Laboratory toxicities will be analyzed by taking the most severe response over the follow-up period, dichotomizing into a binary variable (none versus mild, moderate, or severe) and using exact confidence intervals to summarize the event and toxicity rates. Tabular and graphical summaries of events will be presented for each laboratory parameter, by type, severity (none, mild, moderate, severe), and time point post-administration.

Laboratory and vital sign data will also be presented as change from baseline. Variable transformations will be applied as appropriate. Shift tables may be produced for select laboratory parameters.

11.4.4 Immunology Analysis Plan

Seroconversion is defined as achieving a serum antibody level post-vaccination that is greater than 0.5 IU/mL, the pre-defined protective titer in the KSU RFFIT assay.

Seroconversion rates and GMT for rabies VNA titers will be calculated for each antibody timepoint by treatment arm, and will be summarized graphically.

Seroconversion rates and GMT will be presented with their corresponding 95% confidence interval estimates at each time point and overall peak GMT, and the pair-wise differences between seroconversion rates by treatment arm will be summarized by study day along with 95% confidence intervals. In an exploratory analysis, the relationship between the proportion of seroconverters and vaccine type, dosage, and number of doses may be examined using mixed-effects logistic regression models for longitudinal data.

All GMT calculations will use log10 transformed data, and the anti-log of the resulting point estimates will be reported for means and 95% confidence intervals. The relationship between the log-transformed titer values, vaccine type, and dosage, and dose number will be modeled using longitudinal regression methods in an exploratory analysis. The distribution of antibody titers will also be graphically summarized using the reverse cumulative frequency distribution of titers by study day, and treatment arm.

Cellular immunology assays and anti-vector antibody responses are exploratory endpoints and will be analyzed as described in the Statistical Analysis Plan (SAP).

11.4.5 Missing Values and Outliers

Missing safety and/or immunogenicity data will not be imputed. No search for outliers will be performed. However, the logarithmic transform will be used as appropriate to improve the distributional properties of the data and reduce the impact of potential outliers.

12. SOURCE DOCUMENTS AND ACCESS TO SOURCE DATA/DOCUMENTS

The site will maintain appropriate medical and research records in compliance with ICH E6, Section 4.9 and regulatory and institutional requirements for the protection of subject confidentiality. The site will permit authorized representatives of DMID, 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 aids 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 pharmacies, laboratories, and medico-technical departments involved in this trial.

13. QUALITY CONTROL AND QUALITY ASSURANCE

Following a written DMID-approved site quality management plan, the site and its subcontractors are responsible for conducting routine QA and quality control (QC) activities to internally monitor study progress and protocol compliance. The site PI will provide direct access to the study sites, source data/data collection forms, and reports for monitoring and auditing by the sponsor, and inspection by local and regulatory authorities. The site PI will ensure all study personnel are appropriately trained and applicable documentations are maintained at the site.

The SDCC will implement QC procedures beginning with the data entry system, and generate data QC checks that will be run on the database. Any missing data or data anomalies will be communicated to the site for clarification and resolution.

DMID-designated clinical monitors will verify that this trial is conducted, and data are generated, documented (recorded), and reported in compliance with the protocol, ICH/GCP guidelines, and applicable regulatory requirements. Clinical monitoring reports will be submitted to DMID.

14. ETHICS/PROTECTION OF HUMAN SUBJECTS

14.1. Ethical Standard

The site PI will ensure that this trial is conducted in full conformity with principles of the Belmont Report: Ethical Principles and Guidelines for the Protection of Human Subjects of Research (National Commission for the Protection of Human Subjects of Biomedical and Behavioral Research [April 18, 1979]) and codified in 45 CFR 46, 21 CFR 50, and 21 CFR 56, as applicable. The site PI will also ensure conformity with ICH E6 GCP, and applicable federal regulations, guidance, and guidelines for GCP and Clinical Trials with humans.

14.2. Institutional Review Board

The site PI will obtain IRB approval for this protocol to be conducted at the site, and send supporting documentation to DMID before initiating recruitment of subjects. The investigator will submit applicable information to the IRB/IEC 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, 21 CFR 50, and other federal, state, and local regulations. The IRB/IEC will be registered with the Office of Human Research Protection (OHRP) as applicable to the research. DMID will receive the documentation that verifies IRB/IEC-approval for this protocol, associated ICFs, and upon request any recruitment material and handouts or surveys intended for the subjects, before the recruitment and enrollment of subjects.

Any amendments to the protocol or consent materials will be approved by the IRB/IEC before they are implemented. IRB/IEC 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. The investigator will notify the IRB/IEC of deviations from the protocol and reportable SAEs, as applicable to the IRB/IEC policy.

Each institution engaged in this research will hold a current Federalwide Assurance (FWA) issued by OHRP for federally-funded research.

14.3. Informed Consent Process

Informed consent is a process that is initiated before an individual agrees to participate in a trial and continues throughout the individual's trial participation. Before any study procedures are performed, informed consent will be obtained and documented. Subjects will receive a concise and focused presentation of key information about this trial, verbally and with a written ICF. The explanation will be organized, and presented in lay terminology and language that facilitates understanding of why one might or might not want to participate.

An investigator or designee will describe the protocol to potential subjects face-to-face. The key information about the purpose of this trial, the procedures and experimental aspects of this trial, risks and discomforts, any expected benefits to the subject, and alternative treatment will be presented first to the subject.

Subjects will also receive an explanation that this trial involves research, and a detailed summary of the proposed study procedures and study interventions/products. This will include aspects of this trial that are experimental, the probability for random assignment to dose groups, any expected benefits, all possible risks (including a statement that the particular vaccine or procedure may involve risks to the subject or to the embryo or fetus, if the subject is or may become pregnant, that are currently unforeseeable), the expected duration of the subject's participation in this trial, alternative procedures that may be available, and the potential benefits and risks of these available alternative procedures.

Subjects will be informed that they will be notified in a timely manner if information becomes available that may be relevant to their willingness to continue participation in this trial. Subjects will receive an explanation as to whether any compensation and any medical treatments are available if injury occurs, and, if so, what they consist of, or where further information may be obtained. Subjects will be informed of the anticipated financial expenses, if any, to them for participating in this trial, as well as any anticipated prorated payments, if any, to them for participating in this trial. They will be informed of whom to contact (e.g., the investigator) for answers to any questions relating to the research project.

Information will also include the foreseeable circumstances and/or reasons under which the subject's participation in this trial may be terminated. The subjects will be informed that participation is voluntary and that they are free to withdraw from this trial for any reason at any time without penalty or loss of benefits to which subjects are otherwise entitled.

The extent of the confidentiality of the subjects' records will be defined, and subjects will be informed that applicable data protection legislation will be followed. Subjects will be informed that the monitor(s), auditors(s), IRB, NIAID, and regulatory authority(ies) will be granted direct access to the subject's original medical records for verification of clinical trial procedures and/or data without violating the confidentiality of the subject, to the extent permitted by applicable laws and regulations, and that, by signing a written ICF, the subject is authorizing such access.

Subjects will be informed that records identifying the subject will be kept confidential, and, to the extent permitted by applicable laws and/or regulations, will not be made publicly available and, if the results of this trial are published, the subject's identity will remain confidential. Subjects will be informed whether confidential information collected from this research and/or specimens will be used for additional research, even if identifiers are removed.

Subjects will be allowed sufficient time to consider participation in this trial, and to have the opportunity to discuss it with their family, friends, or legally authorized representative, or think about it before agreeing to participate.

Subjects will be asked to read and review the IRB-approved ICF. Subjects must sign the ICF before starting any study procedures being done specifically for this trial.

Once signed, a copy of the ICF will be given to subjects for their records. Subjects may withdraw consent at any time during this trial. The rights and welfare of subjects will be protected by emphasizing to them that the quality of their medical care will not be adversely affected if they decline to participate in this trial.

Study personnel may employ recruitment efforts before obtaining study consent if a patient-specific screening consent is on record or if the IRB has agreed that chart review is allowed without a fully executed screening consent. In cases where there is not a patient-specific screening consent on record, site clinical staff may pre-screen via chart review and refer potential subjects to the research staff. Research staff would obtain written consent per the standard informed consent process before conducting protocol-specific screening activities.

New information will be communicated by the site PI to subjects who consent to participate in this trial in accordance with IRB requirements. The ICF will be updated and subjects will be re-consented per IRB requirements, if necessary. Subjects will be given a copy of all ICFs that they sign.

14.4. Exclusion of Women, Minorities, and Children (Special Populations)

This trial will include all healthy adults who meet the inclusion/exclusion criteria, regardless of religion, sex, or ethnic background. Only subjects, aged 18 to 49 years inclusive, will be included. Children will be excluded from this first-in-humans trial for safety reasons. Special populations, e.g., non-English speakers, illiterate or non-writing individuals, and vulnerable populations, for which no benefit of trial participation has been identified, will not be enrolled in this trial.

14.5. Subject Confidentiality

Subject confidentiality is strictly held in trust by the participating investigators, their staff, and the sponsor(s) and their agents. This confidentiality includes documentation, investigation data, subject clinical information, and all other information generated during participation in this trial. No information concerning this trial or the data generated from it will be released to any unauthorized third party without prior written approval of DMID and the subject. Subject confidentiality will be maintained when study results are published or discussed in conferences. Study monitors 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 trial. The 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 non-clinical specimens, evaluation forms, reports, and other records that leave the site will be identified only by a coded number.

Specimens provided to research labs for the assays and to GlaxoSmithKline Vaccines for testing with developer assays are barcoded with no subject identifiers. A copy of the final dataset from this study will be provided to the pharmaceutical partner, GlaxoSmithKline Vaccines, which has a Federal Wide Assurance with the Office of Human Research Protections (OHRP, HHS) to maintain the confidentiality of individual patients. The final dataset contains subject level data linked to a coded subject identifier and not subject ID number.

14.6. Study Discontinuation

If this trial is prematurely terminated by the sponsor, any regulatory authority, or the investigator for any reason, the investigator will promptly inform 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/IEC. If any subject's confidential information will continue to be collected for this trial, the IRB/IEC will approve an ICF with the study procedures, any risks and discomforts, and applicable elements, and the investigator or designee will re-consent the subjects as approved by the IRB/IEC.

14.7. Costs, Subject Compensation, and Research Related Injuries

There is no cost to subjects for the research tests, procedures, and vaccines 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 trial. Compensation will be in accordance with the local IRB's policies and procedures, and subject to IRB approval.

If it is determined by the site PI that an injury occurred to a subject as a direct result of the tests or treatments that are done for this trial, 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 trial. Immediate medical treatment may be provided by the site. No financial compensation will be provided to the subject by the NIAID, NIH to the subject. However, the manufacturers of the vaccines will cover research-related injury.

14.8. Future Use of Stored Specimens and Data

At enrollment, subjects will be asked for permission to obtain blood samples for PBMC collection specifically for future use to perform the immunologic assays in line with the

exploratory objectives. Any remaining blood samples may also be used in future research studies, such as examining additional immunological assessments or testing for antibodies against other viruses or bacteria. These residual samples will be stored indefinitely at a central clinical storage facility and may be shared with investigators at the site or other institutions.

It is anticipated that residual serum and PBMCs will be available specifically for other future research, including but not limited to non-traditional immune assay development, assessing innate immune factors and the ability of vaccine-induced antibodies to cross-react with other rabies viruses. These future use clinical samples will be stored indefinitely at a central clinical storage facility.

Residual clinical samples will be available upon the completion of this trial; however, future use clinical samples may be requested from DMID and shipped from DMID CMS at any time.

The samples will not be sold or used directly for production of any commercial product. No human genetic tests will be performed on the samples. Each sample will be encoded (labeled) only with a barcode and a unique tracking number to protect subject confidentiality.

There are no benefits to subjects in the collection, storage and subsequent use of their specimens for future research. Reports about future research done with subjects' samples will not be kept in their health records.

Subjects may be given the option to decide if they want their samples to be used for future research or have their samples destroyed at the end of this trial. The subject's decision can be changed at any time by notifying the study doctors or nurses in writing. However, if the subject originally consents to future use and subsequently changes his/her decision, any data from a previously collected sample may still be used for this research.

15. DATA HANDLING AND RECORD KEEPING

The investigator is responsible to ensure the accuracy, completeness, legibility, and timeliness of the data reported.

Data collection forms will be derived from the eCRF and provided by the SDCC to record and maintain data for each subject enrolled in this trial.

All source documents should be completed in a neat, legible manner to ensure accurate interpretation of data. Black or blue permanent ink is required to ensure clarity of reproduced copies. When making changes or corrections, cross out the original entry with a single line, and initial and date the change. DO NOT ERASE, OVERWRITE, OR USE CORRECTION FLUID OR TAPE ON THE ORIGINAL.

Copies of the eCRF will be provided for use as source data collection forms and maintained for recording data for each subject enrolled in this trial. Data reported in the eCRF derived from source data collection forms should be consistent or the discrepancies should be explained.

The sponsor and/or its designee will provide guidance to the site PI and other study personnel on making corrections to the data collection forms and eCRF.

15.1. Data Management Responsibilities

All data collection forms and laboratory reports must be reviewed by the clinical team and data entry personnel, who will ensure that they are accurate and complete. AEs must be recorded on the appropriate data collection form, assessed for severity and relationship, and reviewed by the site principal investigator or appropriate sub-investigator.

Data collection is the responsibility of the study personnel at the participating clinical study site under the supervision of the site PI. During this trial, the site PI will maintain complete and accurate documentation for this trial.

The SDCC for this trial will be responsible for data management, quality review, analysis, and reporting of the study data.

15.2. Data Capture Methods

Clinical (including, but not limited to, AE/SAEs, concomitant medications, medical history, physical assessments, and clinical lab values) and reactogenicity 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 the SDCC. The data system includes password protection and internal quality checks, such as automatic range checks, to identify data that appear inconsistent, incomplete, or inaccurate.

15.3. Types of Data

Data for this trial will include clinical, safety, and outcome measures (e.g., clinical lab values, reactogenicity, and immunogenicity data).

15.4. Timing/Reports

The SDCC will produce one interim statistical report, and one final clinical study report (CSR). A final report will be prepared following the availability of all the clinical, safety, reactogenicity, and immunogenicity data. Timing and contents of the interim report is described in Section 11.3 of this protocol. Additional interim statistical reports may be generated as deemed necessary and appropriate by DMID. Safety and immunogenicity summary reports may be generated for the SMC.

After full analysis and final reporting is complete, and upon request and DMID approval, the SDCC will provide the VTEU site and subcontract site with a summary of results by treatment group and/or subject treatment assignments. In this regard, the VTEU site and subcontract site requesting such information to share with study subjects must do so in compliance with their respective IRB guidelines.

15.5. Study Records Retention

Study documents will be retained for at least 2 years after the last approval of a marketing application in an ICH region and until there are no pending or contemplated marketing applications in an ICH region or at least 2 years have elapsed since the formal discontinuation of clinical development of the vaccine. These documents will be retained for a longer period, however, if required by local regulations. 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.

15.6. Protocol Deviations

A protocol deviation is any noncompliance with the protocol, GCP, or MOP requirements. The noncompliance may be either on the part of the subject, the site PI, or the site personnel. As a result of deviations, corrective actions will be developed by the site and implemented promptly.

These practices are consistent with ICH E6:

4.5 Compliance with Protocol, Sections 4.5.1, 4.5.2, and 4.5.3

5.1 Quality Assurance and Quality Control, Section 5.1.1

5.20 Noncompliance, Sections 5.20.1, and 5.20.2.

It is the responsibility of the site PI and personnel to use continuous vigilance to identify and report deviations within 5 working days of identification of the protocol deviation, or within 5 working days of the scheduled protocol-required activity. All deviations will be promptly reported to DMID per the SDCC protocol deviation reporting procedures.

All protocol deviations, as defined above, will be addressed in study subject data collection forms. A completed copy of the DMID Protocol Deviation Form will be maintained in the Regulatory File, as well as in the subject's chart. Protocol deviations will be sent to the local IRB/IEC per their guidelines. The site PI and personnel are responsible for knowing and adhering to their IRB requirements.

16. PUBLICATION POLICY

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 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 (OER) 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 supported by the NIH, generally, 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 SAP will be posted on ClincialTrials.gov.

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

DMID does not plan to request certification of delayed posting.

Refer to:

Public Law 110-85, Section 801, Clinical Trial Databases

42 CFR Part 11

NIH NOT-OD-16-149

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18. SUPPLEMENTS/APPENDICES

APPENDIX A. SCHEDULE OF STUDY PROCEDURES AND EVALUATIONS

(see next page)

Study Visit		1st Screen (D-28 to -1)	2nd Screen (D -28 to -1)	V1	V2	V3	V4	V5	V6	V7	V8	V9	V10	V11	Early termination	Unscheduled
Study Day post Vaccination #1 (D1)				Enrollment Dose 1 D1	D2 +2d	D8 +2d	D15 +2d	D16 +2d	D22 +2d	D29+14d	D50+ 7d	D91 ± 14d	D181± 14d	D381± 14d		
Study Day post Vaccination #2						Dose 2 D1	D8 +2d	D9 +2d	D15 +2d	D22+14d	D43+ 7d	D84 ± 14d	D174± 14d	D374± 14d		
Study Day post Vaccination #3							Dose 3 D1	D2 +2d	D8 +2d	D15+14d	D36+ 7d	D77 ± 14d	D167± 14d	D367± 14d		
Study Day post Vaccination #4									Dose 4 D1	D8+14d	D29+ 7d	D70 ± 14d	D160± 14d	D360± 14d		
Signed ICF		X														
Assessment of Eligibility Criteria		X	X	X		X	X		X							
Symptom Screen		X	X	X	X	X	X	X	X	X	X	X	X	X		
Review of Medical History		X	X	X	X	X	X	X	X	X	X	X	X	X	X	(X)
Review of Concomitant Medications		X	X	X	X	X	X	X	X	X	X	X[a]	X[a]	X[a]	X	(X)
Vaccinations (X) Placebo (P)	A			X		P	P		P							
	B			X		P	P		P							
	C			X		P	X		P							
	D			X		X	P		X							
Physical Exam	Complete	X														
	Symptom-Directed		(X)	(X)	(X)	(X)	(X)	(X)	(X)	(X)		(X)	(X)	(X)	(X)	(X)
	Vital Signs	X[b]	X[b]	X	X	X	X	X	X	X		X	X	X	X	(X)

Study Visit		1st Screen (D-28 to -1)	2nd Screen (D -28 to -1)	V1	V2	V3	V4	V5	V6	V7	V8	V9	V10	V11	Early termination	Unscheduled
Study Day post Vaccination #1 (D1)				Enrollment Dose 1 D1	D2 +2d	D8 +2d	D15 +2d	D16 +2d	D22 +2d	D29+14d	D50+ 7d	D91 ± 14d	D181± 14d	D381± 14d		
Study Day post Vaccination #2						Dose 2 D1	D8 +2d	D9 +2d	D15 +2d	D22+14d	D43+ 7d	D84 ± 14d	D174± 14d	D374± 14d		
Study Day post Vaccination #3							Dose 3 D1	D2 +2d	D8 +2d	D15+14d	D36+ 7d	D77 ± 14d	D167± 14d	D367± 14d		
Study Day post Vaccination #4									Dose 4 D1	D8+14d	D29+ 7d	D70 ± 14d	D160± 14d	D360± 14d		
Assessment of Adverse Events and New Onset Chronic Medical Conditions				X	X	X	X	X	X	X	X	X[c]	X[c]	X[c]	X	X
Examine Vaccination Site for Local Reactions (before vaccination on injection visits) and Review Memory Aid					X	X	X	X	X	X					X[d]	X[e]
Pre-vaccine Administration Reactogenicity Assessment				X		X	X		X							
Post-vaccination Observation (for 30 minutes) to Assess Acute Reactions				X		X	X		X							
Evaluation of Vaccination Site to Assess for Reactogenicity and AE/SAEs after 30 minutes				X		X	X		X							
Subject provided with Memory Aid to Enter Reactogenicity Information				X		X	X		X							
Pregnancy test	Serum	X														
	Urine			X		X	X		X							

Study Visit		1st Screen (D-28 to -1)	2nd Screen (D -28 to -1)	V1	V2	V3	V4	V5	V6	V7	V8	V9	V10	V11	Early termination	Unscheduled
Study Day post Vaccination #1 (D1)				Enrollment Dose 1 D1	D2 +2d	D8 +2d	D15 +2d	D16 +2d	D22 +2d	D29+14d	D50+ 7d	D91 ± 14d	D181± 14d	D381± 14d		
Study Day post Vaccination #2						Dose 2 D1	D8 +2d	D9 +2d	D15 +2d	D22+14d	D43+ 7d	D84 ± 14d	D174± 14d	D374± 14d		
Study Day post Vaccination #3							Dose 3 D1	D2 +2d	D8 +2d	D15+14d	D36+ 7d	D77 ± 14d	D167± 14d	D367± 14d		
Study Day post Vaccination #4									Dose 4 D1	D8+14d	D29+ 7d	D70 ± 14d	D160± 14d	D360± 14d		
Blood draw		X	(X)	X	X	X	X	X	X	X		X	X	X	X	(X)
Clinical Laboratory	Chemistry	X[f]	(X)		X	X		X	X						X	(X)
	Hematology	X	(X)		X	X		X	X						X	(X)
	Urine dipstick	X	(X)													
Research Assay	SARS-CoV-2 test				(X)	(X)	(X)	(X)	(X)	(X)						
Immunology (all samples on vaccination days will be collected before the vaccine dose is given)	Rabies nAb (RFFIT)			X		X	X		X	X		X	X	X	X	
	T cell (IFNγ ELISpot) ($5x10^6$ $10x10^6$ PBMCs)			X			X			X		X		X		
	T cell (ICS) ($10x10^6$ PBMCs)						X[g]			X[g]						
	MBC ELISpot ($5x10^6$ $10x10^6$ PBMCs)			X								X	X	X		
	PBMCs for future use			X			X			X		X	X	X		

Study Visit		1st Screen (D-28 to -1)	2nd Screen (D -28 to -1)	V1	V2	V3	V4	V5	V6	V7	V8	V9	V10	V11	Early termination	Unscheduled
Study Day post Vaccination #1 (D1)				Enrollment Dose 1 D1	D2 +2d	D8 +2d	D15 +2d	D16 +2d	D22 +2d	D29+14d	D50+ 7d	D91 ± 14d	D181± 14d	D381± 14d		
Study Day post Vaccination #2						Dose 2 D1	D8 +2d	D9 +2d	D15 +2d	D22+14d	D43+ 7d	D84 ± 14d	D174± 14d	D374± 14d		
Study Day post Vaccination #3							Dose 3 D1	D2 +2d	D8 +2d	D15+14d	D36+ 7d	D77 ± 14d	D167± 14d	D367± 14d		
Study Day post Vaccination #4									Dose 4 D1	D8+14d	D29+ 7d	D70 ± 14d	D160± 14d	D360± 14d		
	Blood for developer assays[h]			X	X	X	X	X	X	X						

[a]Conccomitant medications associated with new onset chronic medical condition(s)

[b]Height and weight will also be measured at the 1st and 2nd screening visits to calculate BMI

[c]Only SAEs

[d]Vaccination site will be examined for local reactions if early termination visit occurs ≤28 days after last injection; memory aid will be reviewed if early termination visit occurs ≤15 days after last injection

[e]Vaccination site will be examined for local reactions if unscheduled visit occurs ≤28 days after any vaccination, and if so, determine whether information recorded by subject on the memory aid conforms to examination

[f]Clinical labs drawn at 1st screening visit will also include HIV-1/2 antibody, HBsAg, and HCV antibody

[g]Assay will be done only for those with positive IFNγ reponse by T cell ELISpot assay

[h]Detailed in Appendix B

Abbreviations: X = study procedure for all Groups, (X) = optional procedures to be determined by study staff, RFFIT = rapid fluorescent focus inhibition test

APPENDIX B. TABLE OF BLOOD VOLUME COLLECTION (ML)

Study Visit		1st Screen (D-42 to -1)	2nd Screen (D -42 to -1)	V1	V2	V3	V4	V5	V6	V7	V8	V9	V10	V11	Early termination	Unscheduled
Study Day post Vaccination #1 (D1)				D1	D2	D8	D15	D16	D22	D29	D50	D91	D181	D381		
Blood draw		X		X	X	X	X	X	X	X		X	X	X	X	(X)
Clinical Laboratory	Chemistry (SST 8mL)	8			8	8		8	8						X	
	Hematology (EDTA 4mL)	4			4	4		4	4						X	
	Screening serology (SST 8mL)	8														
	Serum Preg (SST 3.5mL)	3.5														
	Urine dipstick	X														
Immunology (all samples on vaccination days will be collected before the vaccine dose is given)	Rabies nAb (RFFIT) (SST, 8mL)			16		16	16		16	16		16	16	16	X	
	Anti-vector Ab (SST, 4mL)			4						4						
	T cell (IFNγ ELISpot) (8mL CPT tubes)			8			8			8		8		8		
	T cell (ICS) (8mL CPT tubes)						16*			16*						
	MBC ELISpot (8mL CPT tubes)			8								8	8	8		
	PBMCs for future use			16			16			16		16	16	16		
	Blood for PAXgene (early vaccine response) (future use)			5	5	5	5	5	5	5						

Study Visit		1st Screen (D-42 to -1)	2nd Screen (D -42 to -1)	V1	V2	V3	V4	V5	V6	V7	V8	V9	V10	V11	Early termination	Unscheduled
Study Day post Vaccination #1 (D1)				D1	D2	D8	D15	D16	D22	D29	D50	D91	D181	D381		
	Blood for multiplex protein assay (early vaccine response) (future use) (SST, 8mL)			8	8	8	8	8	8	8						
	Blood volume per visit	23.5		65	25	41	69	25	41	73		48	40	48		
	Total blood volume (cumulative)	**23.5**		**88.5**	**113.5**	**154.5**	**223.5**	**248.5**	**289.5**	**362.5**		**410.5**	**450.5**	**498.5**		

*Assay will be done only for those with positive IFNγ reponse by T cell ELISpot assay

APPENDIX C. TOXICITY TABLE

Local (Injection Site) Reactogenicity Grading

Local (Injection Site) Reaction	Mild (Grade 1)	Moderate (Grade 2)	Severe (Grade 3)
Pain – experienced without touching the injection site (spontaneous discomfort)	Subject is aware of pain, but it does not interfere with daily activity, **and** it does not require pain medication or it requires use of a non-narcotic pain reliever ≤24hours	Subject is aware of pain; there is interference with daily activity **or** it requires repeated use of a non-narcotic pain reliever for >24 hours	Subject is aware of pain, **and** it prevents daily activity or requires any use of a narcotic pain reliever
Tenderness – hurts only when injection site is touched or the arm is moved	The area immediately surrounding the injection site hurts only when touched or with arm motion, **and** it does **not** interfere with daily activity	The area immediately surrounding the injection site hurts when touched or with arm motion, **and** it interferes with daily activity	The area immediately surrounding the injection site hurts when touched or with arm motion, **and** it prevents daily activity
Pruritus (Itching)	Does not interfere with daily activity	Interferes with daily activity	Prevents daily activity

Local (Injection Site) Reaction	Mild (Grade 1)	Moderate (Grade 2)	Severe (Grade 3)
Ecchymosis (Bruising)*	Does not interfere with daily activity	Interferes with daily activity	Prevents daily activity
Erythema (Redness)*	Does not interfere with daily activity	Interferes with daily activity	Prevents daily activity
Induration (Hardness)/Swelling*	Does not interfere with daily activity	Interferes with daily activity	Prevents daily activity

* Size will also be measured in mm but will not be used as a halting criterion.

Ecchymosis (bruising), erythema (redness), and induration (hardness)/swelling as analyzed by measurement will be graded as follows:

Local (Injection Site) Reactogenicity Measurements

Local (Injection Site) Reaction	Mild (Grade 1)	Moderate (Grade 2)	Severe (Grade 3)
Ecchymosis (Bruising)*	25 mm – 50 mm	51 mm – 100 mm	>100 mm
Erythema (Redness)*	25 mm – 50 mm	51 mm – 100 mm	>100 mm
Induration (Hardness)/Swelling*	25 mm – 50 mm	51 mm – 100 mm	>100 mm

* Will not be used as halting criteria.

Subjective Systemic Reactogenicity Grading

Systemic (Subjective)	Mild (Grade 1)	Moderate (Grade 2)	Severe (Grade 3)
Feverishness (Chills/Shivering/Sweating)	No interference with daily activity	Some interference with daily activity	Significant interference, prevents daily activity
Fatigue (Tiredness)	No interference with daily activity	Some interference with daily activity	Significant interference, prevents daily activity
Malaise (General Unwell Feeling)	No interference with daily activity	Some interference with daily activity	Significant interference, prevents daily activity
Myalgia (Body Aches/Muscular Pain)*	No interference with daily activity	Some interference with daily activity	Significant interference, prevents daily activity
Arthralgia (Joint Pain)*	No interference with daily activity	Some interference with daily activity	Significant interference, prevents daily activity
Headache	No interference with daily activity	Some interference with daily activity	Significant interference, prevents daily activity
Nausea	No interference with daily activity	Some interference with daily activity	Significant interference, prevents daily activity

* Not at injection site.

Oral temperature[#] will be graded as follows:

Quantitative Systemic Reactogenicity Grading

Systemic (Quantitative)	Mild (Grade 1)	Moderate (Grade 2)	Severe (Grade 3)
Fever* - oral[†]	38.0°C – 38.4°C 100.4°F – 101.1°F	38.5°C – 38.9°C 101.2°F – 102.0°F	>38.9°C >102.0°F

[#] Oral temperature assessed on Day 1 prior to the first study vaccination will be considered as baseline.

* A fever can be considered not related to the vaccine if an alternative etiology can be documented.

† Subjects must not eat or drink anything hot or cold, or smoke within 10 minutes prior to taking oral temperature.

Additional Adverse Event Severity Grading

Pulse and blood pressure[#] will be graded as follows:

Physiologic Parameter	Mild (Grade 1)	Moderate (Grade 2)	Severe (Grade 3)
Bradycardia - beats per minute	50 – 54	45 – 49	<45
Tachycardia - beats per minute	101 – 115	116 – 130	>130

Hypotension (systolic) mmHg	85 – 89	80 – 84	<80
Hypotension (diastolic) mmHg	50 – 54	45 – 49	<45
Hypertension (systolic) mmHg	141 – 150	151 – 155	>155
Hypertension (diastolic) mmHg	91 – 95	96 – 100	>100

[#] Pulse and blood pressure assessed on Day 1 prior to the first study vaccination will be considered as baseline.

Clinical safety laboratory results* will be graded as follows:

Clinical Safety Laboratory Adverse Event*

Hematology	Normal Range	Mild (Grade 1)	Moderate (Grade 2)	Severe (Grade 3)
WBC $10^3/\mu L$ (Decrease)	3.8 – 10.8	2.5 – 3.7	1.5 – 2.4	<1.5
WBC $10^3/\mu L$ (Increase)	3.8 – 10.8	10.9 – 15.0	15.1 – 20.0	>20.0
Hgb g/dL (Decrease) (Female)	11.7 – 15.5	11.0 – 11.6	9.5 – 10.9	<9.5
Hgb g/dL (Decrease) (Male)	13.2 – 17.1	12.0 – 13.1	10.0 – 11.9	<10.0
Platelets $10^3/\mu L$ (Decrease)	140 – 400	125 – 139	100 – 124	<100
Platelets $10^3/\mu L$ (Increase)	140 – 400	401 – 550	551– 750	>750
Absolute Lymphocyte Count $10^3/\mu L$	0.85 – 3.9	0.65 – 0.84	0.5 – 0.64	<0.5
Absolute Neutrophil Count $10^3/\mu L$	1.5 – 7.8	1.2 – 1.49	1.0 – 1.19	<1.0

Chemistry	Normal Range	Mild (Grade 1)	Moderate (Grade 2)	Severe (Grade 3)
AST IU/L (increase) (Male)	10 – 40	1.1 – 2.5 x ULN	> 2.5 – 5.0 x ULN	> 5.0 – 10 x ULN
AST IU/L (increase) (Female)	10 – 30	1.1 – 2.5 x ULN	> 2.5 – 5.0 x ULN	> 5.0 – 10 x ULN
ALT IU/L (increase) (Male)	9 – 46	1.1 – 2.5 x ULN	> 2.5 – 5.0 x ULN	> 5.0 – 10 x ULN
ALT IU/L (increase) (Female)	6 – 29	1.1 – 2.5 x ULN	> 2.5 – 5.0 x ULN	> 5.0 – 10 x ULN

Total Bilirubin mg/dL (Increase) – when accompanied by any increase in ALT	0.2 – 1.2	1.1 – 1.25 x ULN	> 1.25 – 1.5 x ULN	> 1.5 – 1.75 x ULN
Total Bilirubin mg/dL (Increase) – when ALT is normal	0.2 – 1.2	1.1 – 1.5 x ULN	> 1.5 – 2.0 x ULN	> 2.0 – 3.0 x ULN
Blood urea nitrogen (BUN) mg/dL (increase)	7 – 25	26 – 29	30 – 34	>34
Creatinine mg/dL (Increase) (Male)	0.60 – 1.35	1.36 – 1.70	1.71 – 2.00	>2.00
Creatinine mg/dL (Increase) (Female)	0.50 – 1.10	1.11 – 1.70	1.71 – 2.00	>2.00

*Clinical laboratory evaluations assessed at the screening visit will be considered as baseline