SCHARP
Statistical Center for HIV/AIDS Research and Prevention

STATISTICAL ANALYSIS PLAN FOR HVTN SAFETY

Protocol HVTN 139 (v1.0)

A phase I clinical trial to evaluate the safety and immunogenicity of HIV-1 vaccines based on chimpanzee serotypes of adenovirus expressing Clade C gp140 and a CH505TF gp120 protein boost in healthy, HIV- uninfected adult participants

Date finalized for signature: 25 July 2024

Document will become effective on date of last signature.

SAP version: 3.0

Statistical Analysis Plan for Safety

Protocol: HVTN 139 (v1.0)

Document will become effective on date of last signature.

Author:

Legal Name	Pei-Chun Yu
Job Title	Statistical Research Associate III
Signature & Date	See eTMF for signature manifest

Approval:

Legal Name	Youyi Fong
Job Title	Professor
Signature & Date	See eTMF for signature manifest

SAP Modification History

The version history of, and modifications to, this statistical analysis plan are described below.

SAP Version	Modification
1.0	Initial
2.0	Add MAAE in participant listing Refresh TOC, and formatting Revise typo in section 4 participants bullet
3.0	Update using current Safety SAP template. Updating TLFs section 11.

Table of Contents

1 LIST OF ABBREVIATIONS AND ACRONYMS

AE	Adverse Experience
EAE	Expedited Adverse Experience
FSR	Final Study Report
RSC	Regulatory Support Center
SAE	Serious Adverse Event
SAP	Statistical Analysis Plan
SMB	Safety Monitoring Board

2 OVERVIEW

The following describes the Statistical Analysis Plan (SAP) for the analysis of safety data from HVTN **139** for Safety Monitoring Board (SMB) reports and the Final Study Report (FSR) for Safety.

3 PROTOCOL SUMMARY

Title

A phase 1 clinical trial to evaluate the safety and immunogenicity of HIV-1 vaccines based on chimpanzee serotypes of adenovirus expressing clade C gp140 and a CH505TF gp120 protein boost in healthy, HIV- uninfected adult participants

Primary objective

To evaluate the safety and tolerability of AdC6-HIVgp140 and AdC7-HIVgp140 at doses of 1 x 10^{10} virus particles (vp) and 5 x 10^{10} vp, alone and in combination with CH505TF gp120 adjuvanted with GLA-SE in HIV- uninfected adults.

Study products and routes of administration

- **AdC6-HIVgp140**: a chimpanzee-derived replication-defective adenovirus (Ad) vector expressing codon optimized gp140 of clade C isolate Du422 administered at a dose of 1 x 10^{10} vp or 5 x 10^{10} vp. The vaccine dose will be divided equally into two separate 1mL intramuscular (IM) injections, administered into the deltoid of the non-dominant arm unless medically contraindicated.

- **AdC7-HIVgp140**: a chimpanzee-derived replication-defective Ad vector expressing codon optimized gp140 of clade C isolate Du172 administered at a dose of 1 x 10^{10} vp or 5 x 10^{10} vp. The vaccine dose will be divided equally into two separate 1mL IM injections administered into the deltoid of the non-dominant arm unless medically contraindicated.

- **CH505TF gp120**: CH505 transmitted/founder gp120 mixed with GLA-SE [the immunological adjuvant Glucopyranosyl Lipid A (GLA) in an oil-in-water stable emulsion (SE)], administered at a dose of 400 mcg with 10 mcg GLA-SE as a 1 mL injection in the thigh unless medically contraindicated.

- Placebo control for AdC6 -HIVgp140 and AdC7-HIVgp140 vaccine: Sodium Chloride for Injection, 0.9% , administered as two separate 1 mL IM injections into the deltoid of the non-dominant arm unless medically contraindicated.

- Placebo control for CH505TF gp120: Sodium Chloride for Injection, 0.9% administered as a 1 mL IM injection in the thigh unless medically contraindicated.

Table 1-1 Schema

Study arm	N	AdC dose (vp)	M0	M3	M6
Part A: Low Dose					
Group 1	5	1x 10^{10}	AdC6-HIVgp140	-	-
Group 2	5	1x 10^{10}	AdC7-HIVgp140	-	-
Group 3	2	0	Placebo	-	-
Part B: High Dose					
Group 4	10	5 x 10^{10}	AdC6-HIVgp140	AdC7-HIVgp140	400 mcg CH505TF gp120/GLA-SE
Group 5	10	5 x 10^{10}	AdC7-HIVgp140	AdC6-HIVgp140	400 mcg CH505TF gp120/GLA-SE
Group 6	2	0	Placebo	Placebo	Placebo
Total	34 (30 vaccinees / 4 placebos)				

Notes:

Study is blinded as to Group assignment (Part assignment is not blinded). Enrollment will be stepwise starting with Part A. To ensure the safety of participants, a series of pre-planned enrollment pauses will occur.

Safety Review #1: Low-dose Initial Vaccination. Enrollment for Part A will be restricted to a maximum of 1 participant per day across all participating HVTN Clinical Research Sites (CRSs) until a total of 5 participants have been enrolled. The HVTN Protocol Safety Review Team (PSRT) will review available safety and reactogenicity data reported for each of these 5 participants, up to and including the data reported for the first 72 hours postvaccination for the 5th enrolled participant, and determine whether it is safe to proceed with full enrollment in Part A.

Safety Review #2: Low-dose (Part A) Safe-to-Proceed. The HVTN PSRT will review cumulative safety data, including at a minimum the 2-weeks following the vaccination visit, available on all 12 participants in Part A to determine whether dose-escalation may occur from Part A to Part B.

Safety Review #3: Full Enrollment at Targeted Dose. The HVTN PSRT will also review cumulative safety data available on the first 12 participants enrolled in Part B, including the 2-week post Month 0 vaccination visit, to determine whether it is safe to proceed with full enrollment.

Participants

34 healthy, HIV– uninfected volunteers aged 18 through 50 years, inclusive; 30 vaccinees, 4 placebo recipients

Design

Multicenter, randomized, controlled, double-blind trial

Duration per participant

Part A participants: 6 months of scheduled clinic visits (main study) followed by AESI (Adverse Events of Special Interest) health contacts at month 12, and then annual health contacts at month 24 and 36.

Part B participants: 12 months of scheduled clinic visits (main study) followed by an AESI health contact at month 18, and then annual health contacts at month 24 and 36.

Estimated total study duration

48 months, includes enrollment, planned safety holds, AESI and annual health contacts.

Clinical trial sponsor

DAIDS, NIAID, NIH, DHHS (Bethesda, Maryland, USA)

Study product providers

- AdC6-HIVgp140: provided by DAIDS, NIAID, NIH, DHHS (Bethesda, Maryland, USA)
- AdC7-HIVgp140: provided by DAIDS, NIAID, NIH, DHHS (Bethesda, Maryland, USA)
- CH505TF gp120: DAIDS, NIAID, NIH, DHHS (Bethesda, Maryland, USA)
- GLA-SE adjuvant: DAIDS, NIAID, NIH, DHHS (Bethesda, Maryland, USA)

HVTN Leadership Operations Center (LOC) operations

HVTN Vaccine Leadership Group/Core Operations Center, Fred Hutchinson Cancer Research Center (Fred Hutch) (Seattle, Washington, USA)

HVTN Statistical and Data Management Center (SDMC)

Statistical Center for HIV/AIDS Research and Prevention (SCHARP), Fred Hutch (Seattle, Washington, USA)

HVTN Laboratory Center (LC)

HIV diagnostic laboratories

HIV Sero-Molecular Laboratory-National Institute for Communicable Diseases (HSML-NICD) (Johannesburg, South Africa)

University of Washington Virology Specialty Laboratory (UW-VSL) (Seattle, Washington, USA)

Endpoint assay laboratories

- Cape Town HVTN Immunology Laboratory (CHIL, Cape Town, Africa);
- South African Immunology Laboratory-National Institute for Communicable Diseases (SAIL-NICD, Johannesburg, South Africa)
- Duke University Medical Center (Durham, North Carolina, USA)

- Fred Hutch/University of Washington (Seattle, Washington, USA)
- The Ertl Lab at the Wistar Institute Vaccine and Immunotherapy Center (Philadelphia, Pennsylvania, USA)

Study sites

HVTN Clinical Research Sites (HVTN CRSs) in South Africa:

- Cape Town – Emavundleni
- Durban - eThekwini
- Durban - Isipingo
- Klerksdorp
- Soshanguve
- Soweto - Bara

Safety monitoring

HVTN 139 PSRT; HVTN Safety Monitoring Board (SMB)

4 SAFETY OBJECTIVES AND ENDPOINTS

Primary objective 1:

- To evaluate the safety and tolerability of AdC6-HIVgp140 and AdC7-HIVgp140 at doses from 1 x 10^{10} vp and 5 x 10^{10} vp, followed by boosting with CH505TF gp120 adjuvanted with GLA-SE in HIV- uninfected adults.

Primary endpoint 1:

- Local and systemic reactogenicity signs and symptoms, laboratory measures of safety, and adverse and serious adverse events, assessed for six months following the final vaccine administration. Adverse events of special interest (AESI), and medically attended adverse events (MAAE) for one year following the final vaccination.

5 COHORT DEFINITION

Recruitment will target enrolling 34 healthy, HIV-uninfected adult participants aged 18 through 50 years. Part A and B are single- and multiple-administration groups, respectively. Enrollment to the groups in the two parts will have a series of pre-planned enrollment pauses as indicated in Section 1.

To ensure balance of both sexes assigned at birth, the trial will enroll at least approximately 40% of each sex assigned at birth across all treatment groups. The study team will work closely with the sites to monitor enrollment progress to ensure appropriate balance.

Since enrollment is concurrent with receiving the first study vaccination, all participants will provide some safety data.

6 POTENTIAL CONFOUNDERS

Characterization of the safety of the vaccine is susceptible to confounding by adverse events not related to the vaccine that by chance occur more often in one arm of the trial than another. Therefore analyses involving adverse events will incorporate the reported relationship to product as assessed by HVTN staff.

7 RANDOMIZATION

A participant's randomization assignment will be computer generated and provided to the HVTN CRS pharmacist through a Web-based randomization system. At each institution, the pharmacist with primary responsibility for dispensing study products is charged with maintaining security of the treatment assignments (except in emergency situations as specified in the HVTN MOP).

8 BLINDING

Participants and site staff (except for site pharmacists) will be blinded as to participant treatment arm assignments (eg, vaccine or control). Study product assignments are accessible to those HVTN CRS pharmacists, DAIDS protocol pharmacists and contract monitors, and SDMC staff who are required to know this information in order to ensure proper trial conduct. Any discussion of study product assignment between pharmacy staff and any other HVTN CRS staff is prohibited. The HVTN SMB members also are unblinded to treatment assignment in order to conduct review of trial safety.

When a participant leaves the trial prior to study completion, the participant will be told he or she must wait until all participants are unblinded to learn his or her treatment assignment.

In some cases, the CRS, PSRT, or study sponsor may believe unblinding of the site PI and participant would be appropriate to facilitate the clinical management of an AE or SAE. The HVTN Unblinding MOP specifies procedures for emergency unblinding, and for early unblinding for medical reasons.

9 SAMPLE SIZE

9.1 Accrual and sample size calculations

Recruitment will target enrolling 34 healthy, HIV-uninfected adult participants aged 18 through 50 years. Part A and B are single- and multiple-administration groups, respectively. Enrollment to the groups in the two parts will have a series of pre-planned enrollment pauses as indicated in Section 1.

To ensure balance of both sexes assigned at birth, the trial will enroll at least approximately 40% of each sex assigned at birth across all treatment groups. The study team will work closely with the sites to monitor enrollment progress to ensure appropriate balance.

Since enrollment is concurrent with receiving the first study vaccination, all participants will provide some safety data. However, for immunogenicity analyses, it is possible that data may be missing for various reasons, such as participants terminating from the study early, problems in shipping specimens, low cell viability of processed peripheral blood mononuclear cells (PBMCs), or high assay background. Immunogenicity data from 17 phase 1 and 2 phase 2a HVTN vaccine trials, which began enrolling after June 2005 (data as of September 2014), indicate that 10% is a reasonable estimate for the rate of missing data. For this reason, the sample size calculations in Section 9.2 account for 10% enrolled participants having missing data for the primary immunogenicity endpoint.

9.2 Sample size calculations for safety

The goal of the safety evaluation for this study is to identify safety concerns associated with product administration. The ability of the study to detect serious adverse events (SAEs) (see Section 10, Table 6-1) can be expressed by the true event rate above which at least 1 SAE would likely be observed and the true event rate below which no events would likely be observed. Specifically, for each vaccine arm of the study in Part A (n =5), there is at least an 83% chance of observing at least 1 event if the true rate of such an event is 30% or more; and there is at least a 95% chance of observing no events if the true rate is 1% or less. As a reference, in HVTN vaccine trials from April 2008 through March 2018, about 1.6% of participants who received placebos experienced an SAE.

Binomial probabilities of observing 0, 1 or more, and 2 or more events among arms of size 5 and 10 are presented in Table 6-1 for a range of possible true adverse event rates. Ten participants does not account for historical potential dropout because every enrolled subject provides some safety data. These calculations provide a more complete picture of the sensitivity of this study design to identify potential safety problems with the vaccine.

Table 9-1 Probability of observing 0 events, 1 or more events, and 2 or more events, among arms of size 5 or 10, for different true event rates

True event rate (%)	Pr(0/5)	Pr(1+/5)	Pr(2+/5)	Pr(0/10)	Pr(1+/10)	Pr(2+/10)
1	0.95	0.05	<0.01	0.90	0.10	<0.01
3.5	0.84	0.16	0.01	0.70	0.30	0.05
5	0.77	0.23	0.02	0.60	0.40	0.09
10	0.59	0.41	0.08	0.35	0.65	0.26
20	0.33	0.67	0.26	0.11	0.89	0.62
30	0.17	0.83	0.47	0.03	0.97	0.85
40	0.08	0.92	0.66	0.01	0.99	0.95

An alternative way of describing the statistical properties of the study design is in terms of the 95% confidence interval for the true rate of an adverse event based on the observed data. Table 6-2 shows the 2-sided 95% confidence intervals for the probability of an event based on a particular observed rate. Calculations are done using the score test method (1, 2). If none of the 10 participants receiving a vaccine regimen experience a safety event, the 95% 2-sided upper confidence bound for the true rate of such events in the total vaccinated population is 27.8%.

Table 9-1 Two-sided 95% confidence intervals for the probability of observing a safety event based on observing a particular rate of safety endpoints for arms of sizes 5 and 10

Observed event rate	95% Confidence interval (%)
0/5	0.0 – 43.4
1/5	1.0 – 62.4
2/5	11.8 – 76.9
0/10	0.0 – 27.8
1/10	0.5 – 40.4
2/10	5.7 – 51.0

9.2.1 Sample size calculations for immunogenicity

The main goals of this trial regarding immunogenicity outcomes involve a preliminary estimation of response rates based on data from the BAMA assays among vaccinees. No adjustment for multiple comparisons will be made for the use of multiple assays. The precision with which the true response rate can be estimated from the observed data depends on the true underlying response rate and the sample size. Two-sided 95% confidence intervals for the response rate based on observing a particular rate of responses in the vaccinees is shown in Table 9-2. Calculations are done using the score test method (1). The n = 9 assumes 10% missing immunogenicity data.

Table 9-2 Two-sided 95% confidence intervals for the true response rate based on observing a particular rate of responses in the vaccinees (n = 9, assuming 10% missing data)

No. of responses	Observed response rate (%)	95% Confidence interval
5	56	[26.7, 81.1]
6	67	[35.4, 87.9]
7	78	[45.3, 93.7]
8	89	[56.5, 99.4]
9	100	[70.1, 100.0]

10 STATISTICAL ANALYSIS

This section describes the final study analyses, unblinded as to treatment arm assignment. All data from enrolled participants will be analyzed according to the initial randomization assignment regardless of how many vaccinations they received. Analyses are modified intent-to-treat in that individuals who are randomized but not enrolled do not contribute data and hence are excluded. Because of blinding and the brief length of time between randomization and enrollment—typically no more than 4 working days—very few such individuals are expected.

Analyses for primary endpoints will be performed using SAS and R. All other descriptive and inferential statistical analyses will be performed using SAS, StatXact, or R statistical software.

No formal multiple comparison adjustments will be employed for multiple safety endpoints, multiple primary immunogenicity endpoints, or secondary endpoints. However, multiplicity adjustments will be made for certain immunogenicity assays, as discussed below, when the assay endpoint is viewed as a collection of hypotheses (eg, testing multiple peptide pools to determine a positive response).

10.1 Analysis variables

The analysis variables consist of baseline participant characteristics, safety, and immunogenicity for primary and secondary objective analyses.

10.2 Baseline comparability

Treatment arms will be compared for baseline participant characteristics using descriptive statistics.

10.3 Safety/tolerability analysis

Since enrollment is concurrent with receiving the first vaccination, all participants will have received at least 1 vaccination and therefore will provide some safety data.

10.4 Reactogenicity

The number and percentage of participants experiencing each type of reactogenicity sign or symptom will be tabulated by severity and treatment arm and the percentages displayed graphically by arm. For a given sign or symptom, each participant's reactogenicity will be counted once under the maximum severity for all injection visits. In addition, to the individual types of events, the maximum severity of local pain or tenderness, induration or erythema, and of systemic symptoms will be calculated. Kruskal-Wallis tests will be used to test for differences in severity between arms.

10.5 AEs and SAEs

AEs will be summarized using MedDRA System Organ Class and preferred terms. Tables will show by treatment arm the number and percentage of participants experiencing an AE within a System Organ Class or within preferred term category by severity or by relationship to study product. For the calculations in these tables, a participant with multiple AEs within a category will be counted once under the maximum severity or the strongest recorded causal relationship to study product. Formal statistical testing comparing arms is not planned since interpretation of differences must rely heavily upon clinical judgment.

A listing of SAEs reported to the DAIDS Regulatory Support Center (RSC) Safety Office will provide details of the events including severity, relationship to study

product, time between onset and last vaccination, and number of vaccinations received.

10.6 Local laboratory values

Box plots of local laboratory values will be generated for baseline values and for values measured during the course of the study by treatment arm and visit. Each box plot will show the first quartile, the median, and the third quartile. Outliers (values outside the box plot) will also be plotted. If appropriate, horizontal lines representing boundaries for abnormal values will be plotted.

For each local laboratory measure, summary statistics will be presented by treatment arm and timepoint, as well as changes from baseline for post-enrollment values. In addition, the number (percentage) of participants with local laboratory values recorded as meeting Grade 1 AE criteria or above as specified in the Division of AIDS Table for Grading the Severity of Adult and Pediatric Adverse Events will be tabulated by treatment arm for each postvaccination timepoint. Reportable clinical laboratory abnormalities without an associated clinical diagnosis will also be included in the tabulation of AEs described above.

10.7 Reasons for vaccination discontinuation and early study termination

The number and percentage of participants who discontinue vaccination and who terminate the study early will be tabulated by reason and treatment arm.

11 SAFETY TABLES, PARTICIPANT LISTINGS, AND FIGURES

11.1 List of Tables

SMB reports and Safety FSRs include the following tables.

- Enrollment Report
- Demographics and Vaccination Frequencies
- Overall Protocol Status
- Maximum Local and Systemic Reactogenicity Summaries
- Adverse Experiences by Body System and Severity – By Decreasing Frequency
- Adverse Experiences by Preferred Term and Severity – By Decreasing Frequency – Includes Severe, Life-threatening or Fatal Events Only
- Adverse Experiences by Preferred Term and Severity – By Decreasing Frequency – Includes Events of All Severities
- Adverse Experiences by Preferred Term and Relationship to Study Product – By Decreasing Frequency – Includes Events of Any Relationship
- Expedited Adverse Experiences (EAEs) / Serious Adverse Events (SAEs) Reported to the Regulatory Support Center (RSC) listing.

- HIV Acquisition Results from Lab and Reported by Site listing.
- Study Product Administration Errors listing
- Pregnancy listing

Additional tables included in the FSR for Safety:

- Adverse Experiences of Special Interest (AESIs) listing
- Medically Attended Adverse Events (MAAEs) listing
- End of Study HIV Diagnostic Testing Results (Vaccine-Induced Seropositivity/Reactivity)
- Social Impacts
- Local Lab Value Summary Statistics
- Local Laboratory Values Meeting Grade 1 AE Criteria or Above

11.2 List of Participant Listings

These participant listings are included in the SMB reports:

- Discontinuation Status
- Pregnancies
- Severe or Life-Threatening Local and Systemic Reactogenicities
- Moderate Erythema and Induration
- Expedited Adverse Experiences (EAEs)
- Adverse Experiences of Special Interest (AESIs)
- Medically Attended Adverse Events (MAAEs)
- Study Product Administration Errors
- Severe, Life-Threatening, or Fatal Adverse Experiences
- Adverse Experiences with Relationship to Study Product
- HIV Acquisition Results from Lab and Reported by Site

11.3 List of Figures

These graphs are included in the SMB reports and FSR for Safety:

- Maximum Local Reactogenicities
- Maximum Systemic Reactogenicities
- Boxplots for laboratory measures Alkaline Phosphatase, ALT, Creatinine, WBC, Hemoglobin, Platelets, Lymphocyte Count

12 REFERENCES

1. Agresti A, Coull BA. Approximate is better than "exact" for interval estimation of binomial proportions. Am Stat 1998;52:119-26.
2. Liu W. On sample size determination of Dunnett's procedure for comparing several treatments with a control. Journal of Statistical Planning and Inference 1997;62:255-261.