PROTOCOL

HVTN 705/VAC89220HPX2008

A multicenter, randomized, double-blind, placebo-controlled phase 2b efficacy study of a heterologous prime/boost vaccine regimen of Ad26.Mos4.HIV and aluminum phosphate-adjuvanted Clade C gp140 in preventing HIV-1 infection in women in sub-Saharan Africa

DAIDS DOCUMENT ID 34567

BB IND 016263 HELD BY JANSSEN VACCINES & PREVENTION B.V.

CLINICAL TRIAL SPONSORED BY

JANSSEN VACCINES & PREVENTION B.V.

STUDY PRODUCTS PROVIDED BY

JANSSEN VACCINES & PREVENTION B.V.
Leiden, The Netherlands

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Contents

1 Ethical considerations

Multiple candidate HIV vaccines will need to be studied simultaneously in different populations around the world before a successful HIV preventive vaccine is found. It is critical that universally accepted ethical guidelines are followed at all sites involved in the conduct of these clinical trials. Janssen Vaccines is the Sponsor and a co-funder of the trial, together with NIH/NIAID/DAIDS and the Bill & Melinda Gates Foundation, and therefore is responsible for the oversight of the trial. The HIV Vaccine Trials Network (HVTN) will be primarily responsible for operational execution, as well as involved in other scientific and trial-related activities. Janssen fully endorses the way in which the HVTN addresses ethical concerns as listed below. The HVTN has addressed ethical concerns in the following ways:

- All trials are designed and conducted to enhance the knowledge base necessary to find a preventive vaccine, using methods that are scientifically rigorous and valid, and in accordance with Good Clinical Practice (GCP) guidelines and local in-country adaptations of GCP. Good Clinical Practice is an international ethical and scientific quality standard for designing, conducting, recording, and reporting studies that involve the participation of human subjects. Compliance with this standard and adherence to local adaptations of GCP provides public assurance that the rights, safety, and well-being of study participants are protected, consistent with the principles that originated in the Declaration of Helsinki, and that the study data are credible.
- HVTN and Janssen scientists and operational staff incorporate the philosophies underlying major codes (UNAIDS, Helsinki, CIOMS) [1-3], declarations, and other guidance documents relevant to human subjects research into the design and conduct of HIV vaccine clinical trials.
- HVTN and Janssen scientists and operational staff are committed to substantive community input—into the planning, conduct, and follow-up of its research—to help ensure that locally appropriate cultural and linguistic needs of study populations are met. Community Advisory Boards (CAB) are required by DAIDS and supported at all HVTN research sites to ensure community input, in accordance with Good Participatory Practices (GPP) and all local and national guidelines [4].
- HVTN clinical trial staff counsel study participants routinely on how to reduce HIV risk. Participants who become HIV infected during the trial are provided referral into care and counseling on notifying their partners and about treatment for HIV infection according to local guidelines. Staff members will also counsel them about reducing their risk of transmitting HIV to others.
- The HVTN requires that all international HVTN sites lacking national plans for providing antiretroviral therapy (ART) develop plans for the care and treatment of participants who acquire HIV infection during a trial. Each plan is developed in consultation with representatives of host countries, communities from which potential trial participants will be drawn, sponsors, and the HVTN. If a program for ART provision is not available at a site and ART is needed, a privately established fund will be used to pay for access to treatment to the fullest extent possible.
- The HVTN and Janssen are committed to ensuring that all trial participants receive access to the highest standard of prevention, which may include, but is not limited to, risk reduction counseling, provision of male and female condoms, diagnostic testing and access to treatment for sexually transmitted infections (STIs), and appropriate

referrals to pre- and postexposure prophylaxis (PrEP and PEP) according to national and/or local guidelines.

- The HVTN provides training so that all participating sites similarly ensure fair participant selection, protect the privacy of research participants, and obtain meaningful informed consent. During the study, participants will have their wellbeing monitored, and to the fullest extent possible, their privacy protected. Participants may withdraw from the study at any time.
- Prior to implementation, HVTN trials are rigorously reviewed by scientists who are not involved in the conduct of the trials under consideration.
- All trials are reviewed by local and national regulatory bodies and are conducted in compliance with all applicable national and local regulations.
- The HVTN and Janssen design its research to minimize risk and maximize benefit to both study participants and their local communities. For example, the protocols provide enhancement of participants' knowledge of HIV and HIV prevention, as well as counseling, guidance, and assistance with any social impacts that may result from research participation. HVTN protocols also include careful medical review of each research participant's health conditions and reactions to study products while in the study.
- HVTN research aims to benefit local communities by directly addressing the health and HIV prevention needs of those communities and by strengthening the capacity of the communities through training, support, shared knowledge, and equipment. Researchers involved in HVTN trials are able to conduct other critical research in their local research settings.
- The HVTN and Janssen value the role of in-country Institutional Review Boards (IRBs), Ethics Committees (ECs), and other Regulatory Entities (REs) as custodians responsible for ensuring the ethical conduct of research in each setting.

2 IRB/EC review considerations

US Food and Drug Administration (FDA) and other US federal regulations require IRBs/ECs/REs to ensure that certain requirements are satisfied on initial and continuing review of research (Title 45, Code of Federal Regulations (CFR), Part 46.111(a) 1-7; 21 CFR 56.111(a) 1-7). The following section highlights how this protocol addresses each of these research requirements. Each HVTN Investigator welcomes IRB/EC/RE questions or concerns regarding these research requirements.

This trial is being conducted in sub-Saharan Africa, with funding from the US NIH among others. Due to this, the trial is subject to both US and local regulations and guidelines on the protection of human research subjects and ethical research conduct. These research regulations and guidelines are based on ethical principles of respect for persons, beneficence and nonmaleficence, and justice. Where there is a conflict in regulations or guidelines, the regulation or guideline providing the maximum protection of human research subjects will be followed.

In compliance with international and local (as appropriate) Good Clinical Practice guidelines, each research location has a locally-based Principal Investigator (PI) who is qualified to conduct (and supervise the conduct of) the research. The investigators take responsibility for the conduct of the study and the control of the study products, including obtaining all appropriate regulatory and ethical reviews of the research.

2.1 Minimized risks to participants

45 CFR 46.111 (a) 1 and 21 CFR 56.111 (a) 1: Risks to subjects are minimized.

This protocol minimizes risks to participants by (a) correctly and promptly informing participants about risks so that they can join in partnership with the researcher in recognizing and reporting harms; (b) respecting local/national blood draw limits; (c) performing direct observation of participants postvaccination and collecting information regarding side effects for several days postvaccination; (d) having staff properly trained in administering study procedures that may cause physical harm or psychological distress, such as blood draws, vaccinations, HIV testing and counseling and HIV risk reduction counseling; (e) providing HIV risk reduction counseling and checking on contraception use (for women); and (f) providing safety monitoring.

2.2 Reasonable risk/benefit balance

45 CFR 46.111(a) 2 and 21 CFR 56.111(a) 2: Risks to subjects are reasonable in relation to anticipated benefits, if any, to subjects, and the importance of the knowledge that may reasonably be expected to result.

In all public health research, the risk-benefit ratio may be difficult to assess because the benefits to a healthy participant are not as apparent as they would be in treatment protocols, where a study participant may be ill and may have exhausted all conventional treatment options. However, this protocol is designed to minimize the risks to participants while maximizing the potential value of the knowledge it is designed to generate.

2.3 Equitable participant selection

45 CFR 46.111 (a) 3 and 21 CFR 56.111 (a) 3: Subject selection is equitable

This protocol has specific inclusion and exclusion criteria for investigators to follow in admitting participants into the protocol. Participants are selected because of these criteria and not because of positions of vulnerability or privilege. Investigators are required to maintain screening and enrollment logs to document volunteers who screened into and out of the protocol and for what reasons.

2.4 Appropriate informed consent

45 CFR 46.111 (a) 4 & 5 and 21 CFR 56.111 (a) 4 & 5: Informed consent is sought from each prospective subject or the subject's legally authorized representative as required by 45 CFR 46.116 and 21 CFR Part 50; informed consent is appropriately documented as required by 45 CFR 46.117 and 21 CFR 50.27

The protocol specifies that informed consent must be obtained before any study procedures are initiated and assessed throughout the trial (see Section 9.1). Each site is provided training in informed consent by the HVTN as part of its entering the HVTN. The HVTN requires a signed consent document for documentation, in addition to chart notes or a consent checklist. During a pandemic (such as COVID-19), alternative methods to written consent may be allowed (eg, verbal consent) as further detailed in the HVTN/VAC89220HPX2008 SSP. Prior to implementation, local IRB/ECs should be consulted to verify acceptability of alternative methods.

2.5 Adequate safety monitoring

45 CFR 46.111 (a) 6 and 21 CFR 56.111 (a) 6: There is adequate provision for monitoring the data collected to ensure the safety of subjects.

This protocol has extensive safety monitoring in place (see Section 12). Safety is monitored daily by HVTN Core, Janssen Study Responsible Physicians, and routinely by the HVTN 705/HPX2008 Protocol Safety Review Team (PSRT). In addition, an independent Data and Safety Monitoring Board (DSMB) periodically reviews study data, including unblinded study data if/when needed.

2.6 Protect privacy/confidentiality

45 CFR 46.111 (a) 7 and 21 CFR 56.111 (a) 7: There are adequate provisions to protect the privacy of subjects and maintain the confidentiality of data.

Privacy refers to an individual's right to be free from unauthorized or unreasonable intrusion into his/her private life and the right to control access to individually identifiable information about him/her. The term "privacy" concerns research participants or potential research participants as individuals whereas the term "confidentiality" is used to refer to the treatment of information about those individuals. This protocol respects the privacy of participants by informing them about who will have access to their personal information and study data (see Appendix A). The privacy of participants is protected by

assigning unique identifiers in place of the participant's name on study data and specimens. In addition, each staff member at each study site in this protocol signs an Agreement on Confidentiality and Use of Data and Specimens with the HVTN and each study site participating in the protocol is required to have a standard operating procedure on how the staff members will protect the confidentiality of study participants.

3 Overview

Title

A multicenter, randomized, double-blind, placebo-controlled phase 2b efficacy study of a heterologous prime/boost vaccine regimen of Ad26.Mos4.HIV and aluminum phosphate-adjuvanted Clade C gp140 in preventing HIV-1 infection in women in sub-Saharan Africa

Primary objectives

Primary objective 1:

To evaluate the preventive vaccine efficacy (VE) of a heterologous prime/boost regimen utilizing Ad26.Mos4.HIV and aluminum-phosphate adjuvanted Clade C gp140 for the prevention of HIV infection in HIV-seronegative women residing in sub-Saharan Africa from confirmed HIV-1 infections diagnosed between the Month 7 and Month 24 visits

Primary objective 2:

To evaluate the safety and tolerability of a heterologous prime/boost regimen utilizing Ad26.Mos4.HIV and aluminum-phosphate adjuvanted Clade C gp140 for the prevention of HIV infection in HIV-seronegative women residing in sub-Saharan Africa

Study products and routes of administration

- **Ad26.Mos4.HIV**: a tetravalent adenovirus 26 vaccine containing a premixed ratio (1:1:1:1) of Ad26.Mos1.Gag-Pol, Ad26.Mos2.Gag-Pol, Ad26.Mos1.Env, and Ad26.Mos2S.Env. A dose of 5 x 10^{10} virus particles (vp) will be administered by intramuscular (IM) injection into the deltoid.
- **Clade C gp140**: a trimeric recombinant HIV-1 Env gp140 of clade C. A dose of 250 mcg mixed with aluminum phosphate adjuvant (0.425 mg) will be administered by IM injection into the deltoid.
- **Placebo control for Ad26.Mos4.HIV**: Sodium Chloride for Injection USP, 0.9% administered by IM injection into the deltoid.
- **Placebo control for Clade C gp140**: Sodium Chloride for Injection USP, 0.9% administered by IM injection into the deltoid.

Table 3-1 Schema

Group	N	Month 0	Month 3	Month 6	Month 12
1	1300	Ad26.Mos4.HIV	Ad26.Mos4.HIV	Ad26.Mos4.HIV + Clade C gp140 (250 mcg + adjuvant)[a]	Ad26.Mos4.HIV + Clade C gp140 (250 mcg + adjuvant)[a]
2	1300	Placebo	Placebo	Placebo + Placebo	Placebo + Placebo

[a] 250 mcg refers to total protein content (Clade C gp140). Sterile aluminum phosphate suspension will be used as adjuvant. Aluminum content will be 0.425 mg/0 5 mL dose.

Participants

2600 healthy, HIV-1–uninfected women aged 18 to 35 years; 1300 vaccinees, 1300 placebo control recipients

Design

Multinational randomized, placebo-controlled, double-blind trial

Duration per participant

Minimum 24, maximum 57 months per participant (including 6 months follow-up should a participant be diagnosed with HIV infection at their last scheduled study visit). All HIV-1–uninfected study participants will be followed for at least 24 months following enrollment unless an interim monitoring boundary is reached. Stage 1 is the calendar time since trial start until the last enrolled participant reaches the Month 24 visit. Stage 2 is the calendar time since the last enrolled participant reached the Month 24 visit until the last enrolled participant reaches the Month 36 visit. If the vaccine efficacy analysis in Stage 1 supports continuation of the study through Stage 2, all HIV-1–uninfected study participants will be followed for at least 36 months of scheduled clinic visits. Participants who completed their Month 36 visit will be followed further with scheduled visits every 3 months, until the end of the study (around July 2022, when the last Month 36 visit is completed). Participants who become HIV-1–infected during the study will be followed for approximately 6 months after confirmation of diagnosis.

Estimated total study duration

About 57 months (includes enrollment and follow-up).

Regulatory sponsor

Janssen Vaccines & Prevention B.V. (Leiden, The Netherlands)

Study product providers

- Ad26.Mos4.HIV: Janssen Vaccines & Prevention B.V. (Leiden, The Netherlands)
- Clade C gp140 and aluminum phosphate: Janssen Vaccines & Prevention B.V. (Leiden, The Netherlands)

HVTN Core operations

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

Janssen Operations

Janssen Pharmaceutica NV, Global Clinical Development Operations (Beerse, Belgium)

Statistical and data management center (SDMC)

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

HIV diagnostic laboratory

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

- South Africa Immunology Laboratory and National Institute for Communicable Diseases (SAIL-NICD) (Johannesburg, South Africa)
- HIV Sero-Molecular Laboratory, National Institute for Communicable Diseases (HSML-NICD) (Johannesburg, South Africa)
- Cape Town HVTN Immunology Laboratory (CHIL) (Cape Town, South Africa)
- Duke University Medical Center (Durham, North Carolina, USA)
- FHCRC/University of Washington (Seattle, Washington, USA)
- Janssen Vaccines & Prevention B.V. (Leiden, The Netherlands)
- University of Cape Town (UCT) (Cape Town, South Africa)
- University of Colorado Denver (UC Denver) (Aurora, Colorado, USA)

Study sites

HVTN Clinical Research Sites (HVTN CRSs), in Southern Africa where clade C predominates, to be specified in the Site Announcement Memo

Safety monitoring

HVTN 705/HPX2008 PSRT; NIAID Data Safety Monitoring Board (DSMB)

3.1 Protocol team

Protocol leadership

Chair
PPD
Chris Hani Baragwanath Hospital, Soweto; PPD, South African Medical Research Council

Cochairs
PPD
Aurum Institute

PPD

PPD
San Francisco Department of Public Health

PPD

PPD
Janssen Research & Development
Janssen Vaccines & Prevention, B.V.

PPD

PPD

HVTN Protocol Team leader and Core medical monitor
PPD
HVTN Core, FHCRC

PPD

Statisticians
PPD
SCHARP, FHCRC

PPD

PPD
SCHARP, FHCRC

PPD

PPD
Janssen Vaccines & Prevention, B PPD

Medical officers
PPD
DAIDS, NIAID

PPD

PPD
DAIDS, NIAID

PPD

Laboratory leads
PPD
HVTN Laboratory Program

PPD

PPD
HVTN Laboratory

PPD

PPD
Janssen Vaccines & Prevention, B.V.

PPD

Janssen Protocol Team Leader and Study Responsible Physician
PPD
Janssen Research & Development
Janssen Vaccines & Prevention, B.V.

PPD

Other contributors to the protocol

Janssen Medical Safety Officer	PPD Janssen Vaccines & Prevention, B.V.
Laboratory center representatives	PPD PPD HVTN Laboratory Program, FHCRC
Regulatory affairs	PPD PPD HVTN Core, FHCRC PPD Janssen Vaccines & Prevention, B.V.
Community Advisory Board (CAB) members	PPD Seke South PPD CAPRISA
HVTN Regional medical liaison Southern Africa	PPD HVTN Core, FHCRC
Statistical research associates	PPD SCHARP, FHCRC PPD Janssen Vaccines & Prevention, B.V
IPCAVD representative	PPD Harvard Medical School; Ragon Institute
Senior Clinical Data managers	PPD SCHARP, FHCRC PPD Janssen Vaccines & Prevention, B.V.
Clinical trials manager	PPD HVTN Core, FHCRC PPD Janssen Vaccines & Prevention, B.V.
Clinical safety specialist	PPD HVTN Core, FHCRC
Clinic coordinators	PPD Aurum Institute PPD Perinatal HIV Research Unit
Protocol development managers	PPD HVTN Core, FHCRC
Laboratory Representative	PPD HVTN, FHCRC PPD Janssen Vaccines & Prevention, B.V.
Community engagement unit representative	PPD HVTN Core, FHCRC
Community educators/recruiters	PPD Seke South PPD CAPRISA
Technical editor	PPD HVTN Core, FHCRC

4 Background

4.1 Rationale for trial concept

A safe and effective HIV vaccine is presently the elusive cornerstone of HIV prevention despite advances in other preventive measures [5]. Optimally, researchers believe that both a robust CD4+ and CD8+ T-cell response and a potent humoral response with multiple effector functions should be induced by a vaccine. A successful global prophylactic HIV vaccine will likely need to protect against the diverse strains and clades of HIV-1 as present in different geographic regions. Current strategies are targeting clade specific vaccine approaches, aimed at specific populations. We aim to develop a single global vaccine which offers important scientific and logistic advantages over multiple clade-specific vaccines. Optimizing the magnitude and breadth of epitope coverage is thought to be key to development of a successful antibody and T-cell based preventive HIV vaccine.

Strategies to accomplish this include the use of vaccines containing immunogens from a number of prevalent clades and/or using mosaic sequences, ie, proteins assembled from natural sequences of the different clades [6]. Mosaic antigens could potentially increase the breadth of humoral and cellular immune responses for improved immunologic coverage of diverse clades. This approach has been demonstrated in nonhuman primates showing that mosaic antigens elicit increased cellular immune breadth and depth, and augmented antibody responses, compared with naturally occurring sequences and consensus or conserved antigens [7-9]. This will be the first time to evaluate the efficacy of mosaic HIV-1 antigens and their impact on protection.

There is a paucity of vaccine regimens that have advanced into late-stage clinical development. Only 4 vaccine strategies have proceeded to efficacy studies, and only 1 has demonstrated modest efficacy [10-15]. The RV144 trial in Thailand demonstrated that a heterologous prime-boost regimen using a canarypox vector ALVAC (vCP1521) expressing env, gag, and pol genes with a boost of a bivalent gp120 env glycoprotein, AIDSVAX B/E could provide protection against acquisition. Recently, study HVTN 702 or Uhambo, a Phase 2b/3 evaluating the efficacy of an investigational heterologous vaccine regimen (ie, ALVAC-HIV [vCP2438] + Bivalent Subtype C gp120/MF59) based on the regimen evaluated in the RV144 clinical study but adapted to HIV-1 Clade C which is most common in southern Africa, was stopped early because the pre-established criteria for non-efficacy had been met [56, 57].

Janssen Vaccines and its partners are currently evaluating a novel strategy that involves the use of adenovirus serotype 26 (Ad26) vectors as a prime, and a combination of Ad26 vectors and adjuvanted recombinant glycoproteins as a boost. Although the clinical studies with Ad5 failed to demonstrate protection against HIV infection, the rationale to advance clinical development of Ad26 vector-based vaccines for HIV-1 is based on data [16] showing that (1) Ad26 is biologically substantially different than Ad5; (2) Ad26-based vaccines afford superior protective efficacy compared with Ad5-based vaccines against stringent SIVmac251 challenges in rhesus monkeys; and (3) Ad26 did not increase the number or activation status of total or vector-specific CD4+ T lymphocytes at mucosal surfaces in humans in a randomized, double-blind, placebo-controlled clinical study (IPCAVD 003) [44].

The following candidate HIV vaccine components are being evaluated in clinical studies: Ad26.Mos.HIV (a trivalent Ad26), Modified Vaccinia Ankara (MVA)-Mosaic and Clade C glycoprotein 140 (Clade C gp140). Several prime boost vaccine regimens, either heterologous or homologous, are currently being studied (the APPROACH study, HIV-V-A004) in Southern and East Africa, Thailand, and the US. In addition, Janssen Vaccines has optimized the design of the Ad26 component by adding a fourth vector, encoding a second mosaic Env antigen (Ad26.Mos2S.Env), to the trimeric Ad26.Mos.HIV (generating the tetravalent Ad26.Mos4.HIV) that demonstrated increased breadth of immune response in preclinical studies. This tetravalent Ad26.Mos4.HIV is currently also being evaluated in a clinical study.

The vaccine components chosen for this efficacy study are Ad26.Mos4.HIV and aluminum phosphate-adjuvanted Clade C gp140 (Table 4-1). A heterologous prime-boost regimen will be administered, consisting of an Ad26 mosaic component, followed by the co-administration of the Ad26 mosaic with a Clade C gp140, adjuvanted with aluminum phosphate. The immune response is expected to be active cross clades as animal studies have shown that there is cross clade immunogenicity after administration of Clade C gp140 as a boost in animals primed with the Ad26 mosaic component. Data from this study will guide future development of the vaccine. The study will enroll female participants from populations at high risk of acquiring HIV infection in southern African settings with overall moderate to high HIV incidence, predominantly with clade C. Women in these settings have among the highest HIV infection rates globally, making them one of the populations in greatest need of effective prevention interventions. Should this trial demonstrate efficacy of this regimen, it is likely that additional efficacy studies will include men in these same settings. Other vaccine trials have adopted this staged strategy based on gender (eg, HPV).

In an international adenovirus seroprevalence study [45], Ad5 seroprevalence was 87.9-89.5% in several cohorts of adults in South Africa (N=1,551). The majority of individuals also exhibited moderate Ad5 nAb titers >200 (61.1-63.5%), and a substantial fraction had high Ad5 nAb titers >1000 (25.1-27.9%). In contrast, Ad26 seroprevalence was lower at 43.1-53.2%. Fewer individuals demonstrated moderate Ad26 nAb titers >200 (5.4-7.6%), and only rare individuals had high Ad26 nAb titers >1000 (0.0-0.9%). In the IPCAVD004 clinical trial (N=217) [41], we observed that the baseline Ad26 nAb titers observed in South and East Africa did not impact the humoral or cellular immune responses elicited by the prototype Ad26.ENVA.01 vaccine. Therefore, participants will be enrolled regardless of baseline Ad26 positivity.

Table 4-1 Vaccine Components

Vaccine	JNJ Number	Dose/IM Injection
Ad26.Mos4.HIV:	NA	$5x10^{10}$ vp (1:1:1:1)
Ad26.Mos1.Gag-Pol*	JNJ-55471494-AAA	-
Ad26.Mos2.Gag-Pol*	JNJ-55471520-AAA	-
Ad26.Mos1.Env*	JNJ-55471468-AAA	-
Ad26.Mos2S.Env	JNJ-64219324-AAA	-
Clade C gp140/aluminum phosphate	JNJ-55471585-AAA	250 mcg Clade C gp140, mixed with aluminum phosphate adjuvant (0.425 mg aluminum)
Placebo	NA	0.9% saline

gp, glycoprotein; IM, intramuscular; NA, not applicable; vp, virus particles
* These are the components of the trivalent Ad26.Mos.HIV

4.2 Ad26.Mos4.HIV vaccine

Ad26.Mos4.HIV is a tetravalent recombinant, replication-incompetent Adenovirus 26-based vaccine. Replication-incompetent Ad26.Mos4.HIV vectors are derived from wild-type human adenovirus 26. Wild-type human adenoviruses are common pathogens and endemic to all countries. Adenoviral infections in humans are usually mild and self-limiting. Ad26.Mos4.HIV vectors do not replicate in unmodified human cells and do not produce any toxins. Therefore, no adverse effects would be expected for healthy individuals who inadvertently come into contact with it.

The Ad26 vectors have been made replication-incompetent by deletion of the E1 region of the adenovirus type 26 genome, which is required for replication. A large portion of the E3 region, which promotes persistence within the host cell, has also been removed to create sufficient space in the viral genome for insertion of foreign antigens. For productive infection and replication during manufacturing the E1 defect is supplemented by engineered E1 (from Ad5) complementing cell lines (HEK293, PER.C6®). In unmodified human cells the Ad26.Mos4.HIV vectors cannot replicate. The absence of E1 and presence of the transgene expression cassette is confirmed by identity PCR and sequencing at several stages of vector production. Additionally, replication-incompetence is confirmed by a functional test for replication competent Adenovirus (RCA). The absence of any DNA sequence overlap between the Ad26 vector and the PER.C6® cell line prevents the formation of RCA.

Ad26.Mos4.HIV contains the following 4 active pharmaceutical ingredients (APIs) in a 1:1:1:1virus particle (vp) ratio:

- Ad26.Mos1.Env: recombinant, replication incompetent adenovirus serotype 26 expressing a mosaic 1 HIV-1 Env protein, manufactured in PER.C6 Cells.
- Ad26.Mos2S.Env: recombinant, replication incompetent adenovirus serotype 26 expressing a mosaic 2S HIV-1 Env protein, manufactured in PER.C6 Cells.
- Ad26.Mos1.Gag-Pol: recombinant, replication incompetent adenovirus serotype 26 expressing mosaic 1 HIV-1 Gag and Pol proteins, manufactured in PER.C6 Cells.
- Ad26.Mos2.Gag-Pol: recombinant, replication incompetent adenovirus serotype 26 expressing mosaic 2 HIV-1 Gag and Pol proteins, manufactured in PER.C6 Cells.

4.2.1 Construct

Ad26.Mos4.HIV is comprised of 4 replication-incompetent, Early region 1/Early region 3 (E1/E3)-deleted Adenovirus 26 vectors engineered to express mosaic Gag-Pol and Env sequences. This set of mosaic proteins was assembled *in silico* from fragments derived from natural sequences via a computational optimization method [6]. The dataset used for generating the fragments was restricted to include only full length proteins and only 1 sequence per patient. Ad26.Mos4.HIV consists of Ad26.Mos1.Gag-Pol, Ad26.Mos2.Gag-Pol, Ad26.Mos1.Env, and Ad26.Mos2S.Env. A schematic representation of these 4 vectors of Ad26.Mos4.HIV is shown in Figure 4-1.

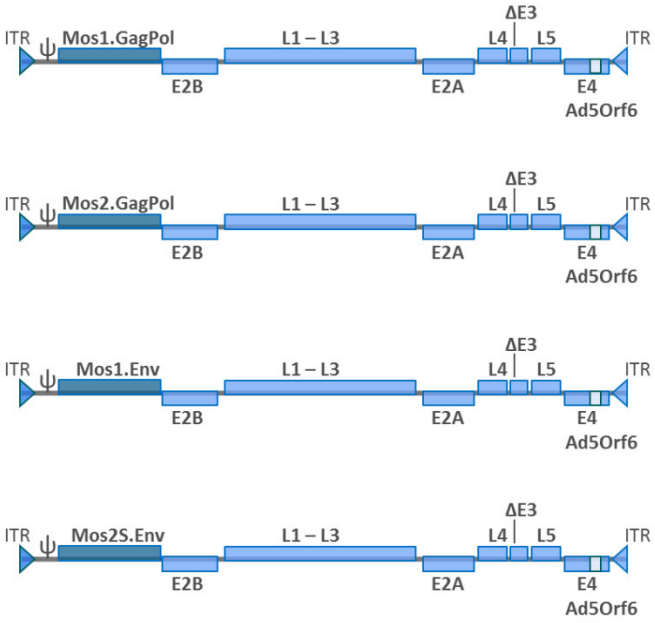

Figure 4-1 Schematic representation of the 4 vectors comprising Ad26.Mos4.HIV

4.2.2 Formulation characteristics

Ad26. Mos4.HIV will be provided in single dose vials for IM injection at a concentration of 1×10^{11} vp/mL. The dose to be administered is 5×10^{10} vp (per 0.5 mL dose). The quantitative composition of Ad26.Mos4.HIV Drug Product (DP) is shown in Table 4-2.

Table 4-2 Targeted composition and concentration of Ad26.Mos4.HIV DP, pH 6.2

Ingredient	Grade	Concentration	Function
Ad26.Mos1.Gag-Pol	NA	2.5×10^{10} vp/mL	Active
Ad26.Mos2.Gag-Pol	NA	2.5×10^{10} vp/mL	Active
Ad26.Mos1.Env	NA	2.5×10^{10} vp/mL	Active
Ad26.Mos2S.Env	NA	2.5×10^{10} vp/mL	Active
NaCl	Ph. Eur. / USP	75 mM	Tonicity agent
Citric acid monohydrate	Ph. Eur. / USP	15 mM	Buffer
PS-80	Ph. Eur.	0.03% w/w	Stabilizer
HBCD	Ph. Eur. / USP	5 % w/w	Stabilizer
Ethanol	Ph. Eur. / USP	0.4% w/w	Stabilizer
NaOH	Ph. Eur. / USP	q.s.	pH modifier
WFI	Ph. Eur.	q.s.	Solvent

PS-80, Polysorbate 80; HBCD, 2-hydroxypropyl-β-cyclodextrin; WFI, Water for Injection; USP, United States Pharmacopeia; q.s., sufficient quantity

4.2.3 Manufacturing

Ad26.Mos4.HIV active ingredients are produced in PER.C6 cells by Janssen Vaccines & Prevention B.V., Leiden, The Netherlands. The PER.C6 cells have been qualified in line with current US Guidance for Industry, International Conference on Harmonization (ICH) guidelines and the European Pharmacopeia. Formulation and filling of the Ad26.Mos4.HIV DP is performed at a qualified current Good Manufacturing Practice

(cGMP) fill and finish contract manufacturer. Manufacturing and release are performed according to cGMP and the facilities involved are in possession of the relevant licenses.

4.3 Clade C gp140

4.3.1 Construct

Clade C gp140 drug substance (DS) = trimeric, recombinant HIV-1 Env gp140 of clade C, produced on a PER.C6 cell line. Aluminum phosphate suspension is used as adjuvant (JNJ-55471585-AAA: Clade C gp140 + aluminum phosphate adjuvant). A schematic representation of the foldon (Fd)-stabilized trimeric gp140 is shown in Figure 4-2.

The clade C antigen is derived from a clade C isolate CZA97012 [17].

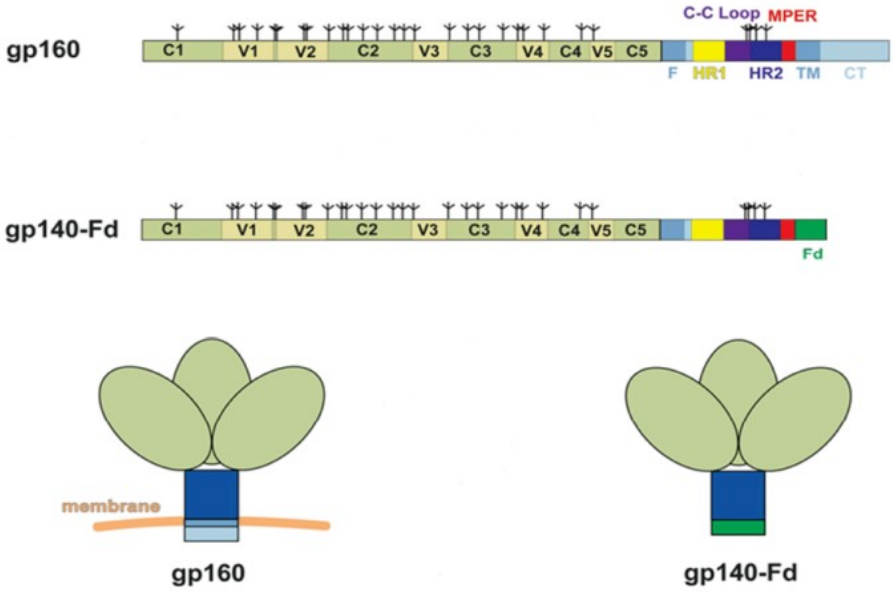

Figure 4-2 Schematic of the Fd-stabilized HIV-1 trimeric gp140. gp140-Fd represents the uncleaved ectodomain of gp160 with a T4-fibritin foldon trimerization tag. Segments of gp120 and gp41 are designated as follows: C1-C5, conserved regions 1-5; V1-V5, variable regions 1-5; F, fusion peptide; HR1, heptad repeat 1; C-C loop, the immunodominant loop with a conserved disulfide bond; HR2, heptad repeat 2; MPER, membrane proximal external region; TM., transmembrane; CT, cytoplasmic tail; Fd, foldon trimerization motif.

4.3.2 Formulation characteristics

Clade C gp140 and aluminum phosphate adjuvant will either be co-formulated or supplied in separate vials.

4.3.2.1 Co-formulation of Clade C gp140 and adjuvant

The phase 2 DP is being developed as a co-formulated sterile liquid, adjuvanted protein with 250 mcg Clade C gp140 dosage strength, with 425 mcg aluminum phosphate

adjuvant at a fill volume of 0.7mL (extractable volume ≥ 0.5 mL) in a single vial. The target composition and concentration of the co-formulation is shown in Table 4-3.

Table 4-3 Target composition and concentration of the co- formulation of Clade C gp140 and Adjuvant

Component	Grade	Clade C gp140, 250mcg Aluminum Phosphate, 425 mcg	Function
Clade C gp140	N/A	0.5 mg/mL	Active
Aluminum phosphate Adjuvant	N/A	0.85 mg Al/mL	Adjuvant
L-Histidine	USP/EP/JP	1.15 mg/mL	Buffer
L-Histidine monohydrochloride monohydrate	EP/JP	0.54 mg/mL	Buffer
Polysorbate 20	NF/EP/JP	0.02% w/v	Surfactant
Sorbitol	NF/EP/JP	5% w/v	Stabilizer/Tonicifier
Volume of WFI	EP	q.s.	Solvent

N/A, not applicable; EP, European Pharmacopoeia; NF, National Formulary; USP, United States Pharmacopeia; JP, Japan Pharmacopoeia; WFI, water for injection; q.s., sufficient quantity

4.3.2.2 Clade C gp140 and separate aluminum phosphate adjuvant drug product

Clade C gp140 and aluminum phosphate adjuvant drug product will be supplied in separate vials, to be mixed at the site pharmacy prior to administration, with a resulting 250 mcg Clade C gp140 dosage strength and a 425 mcg aluminum phosphate adjuvant dosage strength, with a nominal fill volume of 0.5 mL. The target composition and concentration of Clade C gp140 drug product (HEPES buffer) and Clade C gp140 drug product (L-Histidine buffer) is shown in Table 4-4 and Table 4-5, respectively. The target composition and concentration of the aluminum phosphate adjuvant drug product is shown in Table 4-6.

Table 4-4 Target composition and concentration of Clade C gp140 Drug Product (HEPES Buffer)

Component	Grade	Clade C gp140 250mcg	Function
Clade C gp140	N/A	1.0 mg/mL	Active
NaCl	USP/EP	90 mM	Tonicity agent
HEPES	Exipient grade	20 mM	Buffer
Polysorbate 80	EP	0.02% w/v	Surfactant
Sucrose	NF/EP	4% w/v	Stabilizer
Volume of WFI	USP/EP	q.s.	Solvent

N/A, not applicable; EP, European Pharmacopoeia; NF, National Formulary; USP, United States Pharmacopeia; WFI, water for injection; q.s., sufficient quantity

Table 4-5 Targeted composition and concentration of Clade C gp140 Drug Product (L-Histidine buffer)

Component	Grade	Clade C gp140 250 mcg	Function
Clade C gp140	N/A	1.0 mg/mL	Active
L-Histidine	USP/EP/JP	1.15 mg/mL	Buffer
L-Histidine monohydrochloride monohydrate	EP/JP	0.54 mg/mL	Buffer
Sorbitol	NF/EP/JP	50 mg/mL	Stabilizer/ Tonicifier
Polysorbate 20	NF/EP/JP	0.02% w/v	Surfactant
Volume of WFI	USP/EP	q.s.	Solvent

N/A, not applicable; EP, European Pharmacopoeia; NF, National Formulary; USP, United States Pharmacopeia; JP, Japan Pharmacopeia; WFI, water for injection; q.s., sufficient quantity

Table 4-6 Target composition and concentration of Aluminum Phosphate Adjuvant

Component	Grade	Aluminum phosphate, 425 mcg	Function
Aluminum phosphate adjuvant	N/A	1.7 mg Al/mL	Adjuvant
NaCl	USP/EP	90 mM	Tonicity agent
HEPES	Exipient grade	20 mM	Buffer
Polysorbate 80	EP	0.02% w/v	Surfactant
Sucrose	NF/EP	4% w/v	Stabilizer
Volume of WFI	USP/EP	q.s.	Solvent

N/A, not applicable; EP, European Pharmacopoeia; NF, National Formulary; USP, United States Pharmacopeia; WFI, water for injection; q.s., sufficient quantity

4.3.3 Manufacturing

4.3.3.1 Co-formulation of Clade C gp140 and adjuvant

Clade C gp140 DS is produced in PER.C6 cells by Janssen Biotech, Inc. at the contract manufacturing organization Cook Pharmica LLC, Bloomington, IN, USA (or manufactured at another qualified manufacturing site). The Clade C gp140 co-formulated DP is manufactured at Janssen Vaccine Corporation, South Korea (or manufactured at another qualified manufacturing site). All manufacturing is performed according to cGMP requirements and the facilities are in possession of the relevant licenses.

4.3.3.2 Clade C gp140 and separate aluminum phosphate adjuvant drug product

Clade C gp140 DS (HEPES buffer) is produced at Patheon Biologics (formerly Gallus Biopharmaceuticals, LLC) Princeton, NJ, USA (or manufactured at another qualified manufacturing site). For Clade C gp140 the DP manufacturing site is Patheon Biologics (formerly Gallus Biopharmaceuticals, LLC) Princeton, NJ, USA (or manufactured at another qualified manufacturing site).

Clade C gp140 DS (L-Histidine buffer) is produced at Cook Pharmica LLC, Bloomington, US. For Clade C gp140 (L-Histidine buffer) the DP manufacturing site is

Patheon Ferentino, Ferentino, Italy (or manufactured at another qualified manufacturing site).

For aluminum phosphate adjuvant the DP manufacturing site is IDT Biologika Corporation (formerly Aeras) Rockville, MD, USA (or manufactured at another qualified manufacturing site).

4.3.4 Aluminum phosphate adjuvant

4.3.4.1 Co-formulation of Clade C gp140 and adjuvant

For the co-formulated product, the aluminum phosphate adjuvant material is sourced from Pfizer Ireland Pharmaceuticals, Ireland. The DS will be thawed, pooled and mixed. The adjuvant will be mixed with sterile filtered formulation buffer. The drug substance is filtered through a bioburden reduction filter and a 0.22 µm sterilizing filter prior to mixing with adjuvant. The mixed adjuvant and Clade C gp140 protein will be aseptically filled into Type I glass vials. The glass vials are fitted with a grey butyl rubber FluroTec® Film Coated stopper.

4.3.4.2 Clade C gp140 and separate aluminum phosphate adjuvant drug product

For the Clade C gp140 with separate aluminum phosphate adjuvant, the aluminum phosphate adjuvant material is sourced from BRENNTAG BIOSECTOR A/S, Denmark (or manufactured at another qualified manufacturing site). Formulation buffer is mixed aseptically with aluminum phosphate adjuvant. Vials are filled, stoppered, and capped at the manufacturing site IDT Biologika Corporation (formerly Aeras) Rockville, MD, USA (or manufactured at another qualified manufacturing site).

4.4 Trial design

4.4.1 Dose (amount and number)

Ad26.Mos4.HIV will be administered at a dose of 5 x 10^{10} vp by IM injection into the deltoid. This dose has been chosen based on 3 previous phase 1 studies of the related Ad26.ENVA.01 vaccine (see Section 4.9). In these studies Ad26.ENVA.01, at doses over the range of 10^{9} to 10^{11} vp, induced Env-specific humoral and cell-mediated responses when given up to 3 times to more than 200 healthy participants in the United States and Africa. The dose of 5 x 10^{10} vp was found to provide the optimal balance of immunogenicity and reactogenicity. The same dose has been used in the NHP 13-19/15-06 study (Section 4.8.4) and is also being used in the ongoing clinical Phase 1/2a studies HIV-V-A004 (Section 4.9.2.2) and HPX2004 (Section 4.9.2.4).

Based on the final results of HIV-V-A003 and the results of the primary analysis (Week 28) of HIV-V-A004, a dose of 250 mcg Clade C gp140, adjuvanted with aluminum phosphate, has been selected for use in the current study (see Section 4.9).

4.4.2 Schedule

The choice of schedule is based on observations from a variety of pre-clinical and clinical studies using different adenovirus vectors with a variety of inserts, as well as immunogenicity generated in RV144 (which demonstrated the probable benefit of a late

boost) (see Sections 4.8 and 4.9). Given that RV144 demonstrated modest efficacy, we have built on the vector prime, protein boost concept, choosing a similar schedule but adding a late boost in an attempt to compensate for the waning effect of RV144 after the first year.

This schedule was used in NHP study 13-19/15-06, which demonstrated encouraging immunogenicity and protection against challenge (see Section 4.8.4).

4.4.3 Prime-boost regimen

The prime-boost strategy consists of Ad26.Mos4.HIV primes with Ad26.Mos4.HIV plus Clade C gp140 boosts.

In rhesus macaque SIV challenge models, heterologous boost immunizations with a SIV model vaccine potently expanded cellular and humoral immune parameters that correlated with protection against viral challenge [18]. These results were confirmed for the clinically applied mosaic/recombinant protein HIV vaccine concept in rhesus macaque SHIV challenge models [19,20].

The concept of priming with 1 HIV vaccine component followed by boosting with another heterologous HIV vaccine component emerged in the 1990s as a strategy to enhance vaccine immunogenicity and, potentially, vaccine efficacy. While Env glycoproteins elicit strong humoral responses, they have not been associated with potent cellular immunity or efficacy when administered alone as in the first efficacy trials of AIDSVAX [11,14]. On the other hand, although viral vector constructs, including Ad26, often elicit both antibody and cell-mediated responses, it was observed that the magnitude of antibody responses could be increased significantly by subsequent administration of Env glycoproteins. In addition, prime-boost vaccine regimens were explored as means of increasing the breadth of HIV strains to which vaccination might induce responses. Response breadth and the capacity to induce both potent cytotoxic T lymphocyte and potent antibody responses is a combination believed to be important in conferring protective immunity against HIV infection [21,22]. These desirable immune response characteristics were often observed in early phase clinical trials using a variety of prime-boost vaccine regimens [22-32].

The use of a prime-boost regimen is currently being evaluated in the clinical studies HIV-V-A004 (Section 4.9.2.2) and HPX2004 (Section 4.9.2.4).

4.4.4 Choice of control

The placebo control for Ad26.Mos4.HIV and Clade C gp140 is sodium chloride for injection, 0.9%.

4.4.5 HIV-1 incidence estimates from previous clinical trials among at-risk study populations in Southern Africa

The high rates of HIV acquisition amongst young women in Southern Africa highlight the need for biomedical interventions that are female controlled. Recent longitudinal studies evaluating various interventions indicate HIV incidence rates of between 4-11% and are summarized below:

Vaccine efficacy studies

- The HVTN 503 (Phambili) HIV vaccine study (2007), in which HIV-1 incidence among placebo recipients, 18-35 years of age was estimated at 3.7 per 100 person-years (5.86 for females and 1.93 for males) [33].

Topical and Systemic Pre-Exposure Prophylaxis Studies

- The FACTS 001 vaginal microbicide study (2011-2014), in which HIV-1 incidence was estimated at 4.0 per 100 person-years in women age 18-30 [34].
- The VOICE study of vaginal microbicide and systemic prophylactic antiretroviral drugs (2009-2013), in which overall HIV-1 incidence was 5.7% and incidence in the 3 placebo arms of the trial varied from 4.2 to 6.8 per 100 person-years, while the annual incidence in single women in South Africa aged 25 or younger was up to 10% [35].
- The MTN-020-ASPIRE dapivirine vaginal ring study (2012-2015), in which HIV-1 incidence was estimated at 4.5 per 100 person-years in female placebo recipients aged 18-45 in Malawi, South Africa, Uganda, and Zimbabwe [36].
- The RING study (IPM-027), a vaginal ring study (2012-2015), in which HIV-1 incidence was estimated at 6.1 per 100 person-years in female placebo recipients aged 18-45 [37].
- The Fem-PrEP, a study of PrEP effectiveness in HIV-negative women in Kenya, South Africa, and Tanzania (2009-2012), in which HIV-1 incidence was estimated at 5.0 per 100 person-years overall and ranged from 3.4–6 per 100 person-years in South African female placebo recipients aged 18-35 [38].

Based on a synthesis of the above references the estimated annual incidence rate of HIV-1 in Sub-Saharan women is 5.0% (total HIV acquisition events divided by total person years at risk). Under an assumed 90% PrEP efficacy during PrEP use, a conservative estimate of 4.2% annual HIV incidence can tolerate up to 15 out of 100 person years at risk during PrEP use.

A conservative estimate of the background HIV-1 incidence is 4.2 per 100 person-years, and this level of incidence is expected for the duration of our study. Self-reported risk has been shown to decline over time in HIV prevention studies, attributed to risk reduction counseling and the provision of other interventions provided to study participants as part of a package of prevention. While incidence of HIV infection may be expected to decrease over time in trial populations due to the provision of HIV prevention packages, as well as the heterogeneity in risk for HIV-1, the increasing access to pre-exposure prophylaxis at a country level may impact HIV incidence more profoundly.

4.5 Combination prevention of HIV acquisition

HVTN 705 embraces the highest South African standards of prevention for all participants. Participants will be provided with a comprehensive HIV prevention package. This package includes evidence-based behavioral risk reduction counseling [39], advocacy and referral of partner for medical male circumcision as appropriate, free condoms and lubricant (where available), regular testing as well as treatment or appropriate referral for sexually transmitted infections (STIs), counseling and referral for postexposure prophylaxis (PEP) when indicated, and, where appropriate, access to oral drugs for pre-exposure prophylaxis (PrEP). Participants will be informed as new prevention modalities are proven effective and become available. These activities can be

expected to reduce HIV acquisition below historical levels. The contribution of these efforts will be monitored. Further details on these efforts are provided in the HVTN 705 Study Specific Procedures (SSP) and the HVTN 705 website.

4.6 Plans for future product development and testing

Upon confirmation of the safety and immunogenicity of Ad26.Mos4.HIV in HVTN 117/HPX2004 (see Section4.9.2.4), HVTN 705/VAC89220HPX2008 will be initiated in southern Africa utilizing a regimen of Ad26.Mos4.HIV and Clade C gp140.

Upon confirmation of the safety and immunogenicity of the combination of Clade C gp140 and Mosaic gp140, with Ad26.Mos4.HIV in HVTN 118/HPX2003 (see Section 4.9.2.5), a phase 3 global efficacy trial will be performed to determine if this regimen can sufficiently prevent infection globally with HIV-1 of multiple clades.

4.7 Nonclinical safety studies

Table 4-7 Summary of nonclinical safety studies

Study number	Product	Type of study	Animal	N	Route	Schedule
TOX10872	Control Clade C gp140 Clade C gp140 + Aluminum Phosphate	Double dose Toxicity	NZW[1] Rabbits	30m/30f	IM	day 0, 21
TOX10873	Control Ad26.Mos.HIV MVA Mosaic Clade C gp140 + Aluminum Phosphate	4 Cycle Toxicity	NZW Rabbits	60m/60f	IM	day 0, 21, 42, 63

NZW, New Zealand white; IM, intramuscular; m, male; f, female

4.7.1 Double dose IM toxicity study of Clade C gp140 in New Zealand White rabbits (TOX10872)

This study assessed potential toxicity and local tolerance of Clade C gp140 and Clade C gp140/aluminum phosphate administered IM to male and female NZW rabbits (see Table 4-7). Animals were injected with 250 mcg of Clade C gp140 +/- 425 mcg of aluminum phosphate (the proposed clinical dose) 3 weeks apart and any adverse effects were monitored during a 21 day recovery period.

The vaccine was well tolerated when administered 2 times with an interval of 3 weeks between injections. An immune response was triggered, as shown by the IgG levels measured in both groups. Increases in C-reactive protein (CRP) were seen in both groups (but more consistently in the adjuvant group) and had resolved by the end of the recovery period. These CRP increases are typical for a mild immune and/or inflammatory response related to vaccine administration. Mild reactions were seen at the injection sites, which largely recovered 3 weeks after the last dose administration. For additional information please see the Investigator's Brochure.

4.7.2 Four-cycle IM toxicity study with prime-boost combinations of Ad26.Mos.HIV, MVA-Mosaic, and Clade C gp140 in New Zealand White rabbits (TOX10873)

This study assessed potential toxicity and local tolerance of prime-boost combinations of Ad26.Mos.HIV, MVA-Mosaic, and Clade C gp140/aluminum phosphate administered once every 3 weeks for up to 4 injections (see Table 4-7). Ad26.Mos.HIV was administered at a dose of 5×10^{10} vp (the proposed clinical dose) and Clade C gp140/aluminum phosphate was administered at a dose of 250 mcg/425 mcg (the proposed clinical dose).

The different dose regimens were well tolerated when administered once every 3 weeks for up to 9 weeks (ie, 4 injections). All tested vaccine regimens were immunogenic and induced Clade C gp140-specific serum IgG levels. The observed increases in CRP, fibrinogen, globulin and body temperature and the transient decrease in food consumption are considered to reflect a normal, non-adverse response to the vaccine administration. Non-adverse test article-related findings were seen in iliac lymph nodes, in the spleen, and at the injection sites. Injection-site lesions showed ongoing recovery at the 3 weeks recovery interval while the findings in lymph nodes and spleen (ie, immunogenic response) were still present after recovery.

For additional information please see the Investigator's Brochure.

4.7.3 Nonclinical assessment of Ad26.Mos4.HIV and formulation buffer changes

No repeated dose GLP toxicity study has been performed with Ad26.Mos4.HIV (ie. containing the Ad26.Mos2S.HIV vector). There is significant nonclinical experience with Ad26 vectored vaccines using various HIV and non-HIV gene-inserts, showing that these vaccines are safe and well tolerated. At least 7 GLP toxicity studies have been performed in rabbits testing the nonclinical safety of various prime-boost regimens with Ad26 vectored HIV and non-HIV vaccines at full human doses of up to $1.2x10^{11}$ vp. All effects observed in these studies were considered to be reflective of a physiological inflammatory/immune response to the vaccines administered and seem to be independent of the specific gene-insert used in the Ad26 vector. This indicates that differences in the expressed inserts do not pose a significant safety concern, when using the same vector backbone and vector (vp) dose.

Ad26.Mos4.HIV (including the new Ad26.Mos2S.Env vector) vaccine was tested in a non-GLP immunogenicity study in rabbits (See Section 4.8.5). Two full human doses ($5x10^{10}$ vp per dose) were administered via IM injection to the animals with a 6-week interval. The animals were observed daily for signs of toxicity and body weights were measured monthly. No test article-related adverse effects were observed.

The Ad26.Mos2S.Env construct is produced using the same production platform as was used for the Ad26.Mos1.Env, Ad26.Mos1.Gag-Pol and Ad26.Mos2.Gag-Pol constructs that were part of Ad26.Mos.HIV and which was toxicologically assessed previously (TOX10873).

Ad26.Mos4.HIV will be formulated in a different formulation buffer as used for Ad26.Mos.HIV that was toxicologically assessed in TOX10873. This change in the formulation buffer does not raise a safety concern, as the excipients in the post-change formulation buffer are either used in the pre-change formulation buffer or are accepted excipients for parenteral use. In addition, the new formulation buffer (without vaccine)

has been tested in a supportive single dose intramuscular local tolerability and tissue reaction study in rabbits (1 mL injection volume), not showing any adverse local effects (TOX11521).

Based on these considerations it was not deemed necessary to perform an additional GLP toxicity study including Ad26.Mos4.HIV. This has been agreed with the US FDA in a pre-IND meeting on September 24, 2015, as well as in a Type C meeting on January 21, 2016.

The co-formulated Clade C gp140 + aluminum phosphate will be using a different formulation buffer than the buffer used for the Clade C gp140 + aluminum phosphate mix that will be prepared at the pharmacy and which was used in the HIV-V-A003 and HIV-V-A004 clinical trials (See Section 4.9), as well as in the available GLP toxicity studies TOX10872 and TOX10873. For the co-formulated product, this initial (sucrose-based) buffer will be replaced by a buffer composed of histidine, sorbitol, and PS-20. This buffer change is not considered to impact on the safety profile of the co-formulated Clade C gp140 DP, as the excipients are used in licensed vaccines (with histidine also being a component of the current Ad26.Mos4.HIV formulation buffer) and are listed as approved excipients for parenteral use. Based on this rationale it was not deemed necessary to perform an additional GLP toxicity study with the post-change Clade C gp140 DP. This approach has been agreed with the US FDA through a Type C meeting briefing document in a Type C meeting on January 21, 2016. A rabbit immunogenicity study has been performed to evaluate potential influence of the formulation change on immunogenicity (Section 4.8.6). In this study, visual inspection of the injection sites was performed to assess possible local (skin) effects. No safety signals were observed in this study.

4.8 Nonclinical immunogenicity and efficacy studies

4.8.1 Nonclinical models

The nonclinical data presented here is primarily based on data from 2 rhesus macaque challenge models, using simian immunodeficiency virus (SIV) or simian-human immunodeficiency virus (SHIV) [18-20].

In the SIV model, NHP were immunized with vaccines carrying SIV antigens (analogues of HIV) and then challenged with neutralization-resistant SIVmac251. This model was used to evaluate potential viral vectors and inserts capable of inducing cellular and humoral immune responses with broad specificity and high magnitude. In the SHIV model, the mosaic HIV-1 vectors were used to immunize NHP prior to challenge with neutralization-resistant SHIV SF162P3. SHIV is an engineered virus comprising the genetic backbone of SIVmac239 with the envelope of HIV-1 and accessory genes. The clade B-derived challenge virus SF162P3 is fully heterologous in relation to all used vaccine antigens (Clade C gp140, Mos1 and Mos2/Mos2S).To mimic natural exposure to HIV-1, multiple low doses of SIV or SHIV were delivered intra-rectally, and neutralization-resistant challenge strains were used.

In addition, a rabbit immunogenicity model was used. This species is able to generate antibodies with long CDR H3 loops, which is a function-related characteristic of most neutralizing antibodies isolated from HIV-1 infected humans.

4.8.2 Immunogenicity and protective efficacy of vectors encoding mosaic antigens in rhesus macaques

A proof-of-concept study in rhesus macaques demonstrated that adenoviral heterologous Ad26/Ad35 vaccine regimens encoding bivalent HIV-1 Mosaic Env/Gag/Pol antigens afforded partial protection against acquisition of infection following repetitive, heterologous, intrarectal challenges with the difficult-to-neutralize SHIV-SF162P3 (tier 2; Clade B; HIV-1 Env and SIV Gag/Pol) [19]. This corresponded to a per exposure risk reduction of 87%, although absolute protection against the full challenge series was low.

Protection against acquisition of infection correlated with (1) binding antibody titers against the homologous Mosaic 1 Env antigen, (2) vaccine-elicited neutralizing antibodies against SF162 (tier 1; Clade B), a neutralization sensitive virus that is related to the challenge virus SHIV-SF162P3, and (3) functional antibody-dependent cellular phagocytosis responses (ADCP) which are usually dependent on neutralizing and non-neutralizing binding antibodies. Taken together, the identified correlates of protection suggest that the coordinated activity of multiple antibody functions may contribute to protective efficacy.

4.8.3 Immunogenicity and protective efficacy of Ad26 vectors followed by a boost with Clade C gp140 in rhesus macaques

Clade C gp140 protein was selected as protein boost component of the proposed investigational vaccine regimen in order to maximize the humoral immune response, to increase the chances of eliciting HIV-1 specific functional antibodies [17,40].

A study was conducted in rhesus macaques to assess the effect of boosting with Env gp140 protein after priming with Ad26 [20]. In the SIV model, boosting with adjuvanted SIVmac32H gp140 protein afforded 50% protection from infection in animals that were primed with an Ad26 vector expressing the SIVsmE543 Env/Gag/Pol antigens. This protection was significantly higher than in animals that received prime and boost vaccinations with adenoviral vectors (17% protection). This suggests that recombinant Env gp140 is an important component of an efficacious vaccine regimen. Antibody Env binding capacity and Env specific antibody functionality significantly correlated with protection against SIV challenge. Env gp140 boosting primarily expanded Env-specific IFN-γ CD4+ T-lymphocyte responses in the Ad26/gp140 group [20].

In addition to the protective advantage of a boost immunization with gp140, an adenovirus-based prime immunization was demonstrated to be an important factor in mediating protection from infection. Prime immunization with adenoviral vectors Ad26 and Ad5 coding for HIV-1 antigens followed by an HIV-1 Clade C gp140 protein boost (with AS01B as adjuvant), was able to partly protect rhesus macaques against stringent heterologous SHIV-SF162P3 challenge while repeated immunization with the similarly adjuvanted Clade C gp140 alone mediated a lower level of protection. Forty percent (8 of 20) of Ad/Env-vaccinated animals were completely protected against the series of 6 challenges. Binding antibody titers and ADCP responses correlated with protection against acquisition of infection [20].

Taken together, these data indicate that Ad26 priming followed by Env gp140 protein boosting is the most advantageous regimen of those examined and warrants further testing as an efficacious prototype HIV-1 vaccine.

4.8.4 Immunogenicity and protective efficacy of Ad26.Mos.HIV and Clade C gp140 in rhesus macaques

An NHP study (Study 13-19/15-06) was performed to test, among other compounds, the combination of Ad26.Mos.HIV priming and Clade C gp140 boost, using comparable regimens, starting materials and manufacturing processes as those being used in clinical studies. Briefly, 6 cohorts of 12 rhesus macaques were primed in Month 0 and Month 3 with trivalent Ad26.Mos.HIV, followed by heterologous boosts with vector-based vaccine components or Clade C gp140 with aluminum phosphate adjuvant or a combination thereof in Month 6 and Month 12.

Immunogenicity data from this study (Figure 4-3, Panels A-C) showed that group geometric mean binding antibody titers were highest in all groups boosted with Clade C gp140 protein. Boosting with Clade C gp140 also increased the Env-specific cellular response, measured by ELISpot. HIV-1 pseudovirus neutralization data overall followed the trend of humoral responses; however, substantial neutralization was only observed for easy-to-neutralize Tier 1 Env pseudotyped viruses (data not shown).

Viral load data were determined after each challenge of a series of 6 IR weekly challenges with SHIV-SF162P3 (Figure 4-3, Panel D). The 3 most protective regimens contained Clade C gp140 as a boost vaccine component. The regimen boosted with a combination of Ad26.Mos.HIV and Clade C gp140 led to the highest level of protection in this study (8 out of 12 monkeys protected after the full course of challenges). Statistical analysis of the data showed that this group had a 94% per exposure risk reduction, which was associated with a 66% complete protection after the full series of 6 challenges (Table 4-8).

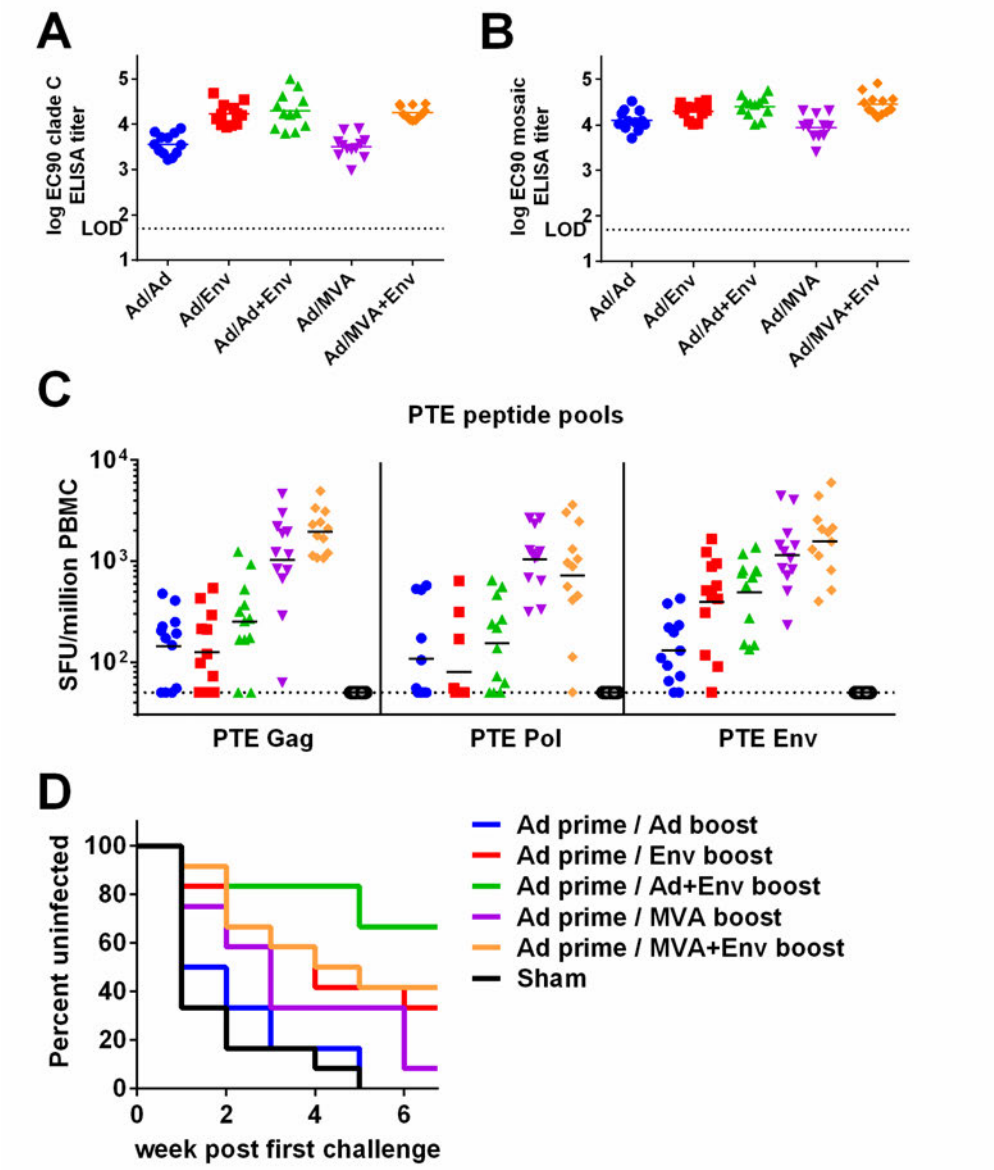

Figure 4-3 Immunogenicity and protective efficacy of various Ad26.Mos.HIV/Clade C gp140 prime-boost regimens in rhesus macaques. Results of NHP Study 13-19/15-06. Clade C gp140 (A) and Mosaic gp140 (B) Binding Antibody Responses Generated in Rhesus Macaques in Week 56 Post First Immunization (4 Weeks Post Final Immunization), (C) ELISpot Responses Against PTE (Potential T-cell Epitopes From Worldwide Circulating HIV-1 Strains) Peptides in Week 54 Post First Immunization. All Animals Were Primed in Month 0 and Month 3 With Trivalent Ad26.Mos.HIV, Followed by Heterologous Boosts in Month 6 and 12 as Indicated in the Graphs and in Table 2. (D) Number of Challenges Required for Acquisition of Infection Displayed as Kaplan-Meier Plots. Ad - Ad26.Mos.HIV; Env – Clade C gp140; MVA – MVA-Mosaic.

Table 4-8 Statistical analysis of NHP study 13-19/15-06

Regimen	Hazard Ratio (95% CI)	Per-exposure risk reduction	p-value versus sham	Complete protection
Ad **Ad**	0.647 (0.205-2.039)	35%	1.000	0%
Ad **Env**	0.161 (0.050-0.516)	84%	0.011	33%
Ad **Ad + Env**	0.055 (0.014-0.215)	94%	0.001	66%
Ad **MVA**	0.288 (0.094-0.879)	71%	0.144	8%
Ad26 **MVA + Env**	0.130 (0.040-0.430)	87%	0.004	42%
Sham	1 (reference)	N/A	N/A	0%

[1] Cox proportional hazard model

p-values were adjusted with a 5-fold Bonferroni correction for multiple comparisons

Ad, Ad26.Mos.HIV; Env, Clade C gp140; MVA, MVA-Mosaic

4.8.5 Immunogenicity of Ad26.Mos4.HIV with Clade C gp140 in rabbits

The immunogenicity and antigenicity of Ad26.Mos4.HIV compared to Ad26.Mos.HIV was tested in rabbits (study 0095-14). Six rabbits per group were immunized with 2 prime immunizations with Ad26.Mos.HIV or Ad26.Mos4.HIV, each at doses of $5x10^9$ or $5x10^{10}$ vp, and 2 aluminum phosphate-adjuvanted Clade C gp140 protein boost immunizations.

The neutralization capacity of immune sera collected after the second, third and fourth immunization was determined against easy to neutralize Tier 1 pseudoviruses (Figure 4-4). For the clade C pseudovirus MW965, the Ad26.Mos4.HIV vaccine (Groups 3+4, Panels A+B, Figure 4-4) elicited significantly increased neutralization capacity in comparison to the Ad26.Mos.HIV combination (Groups 1+2, Panels A+B, Figure 4-4). For the clade B pseudovirus SF162, the neutralization capacity elicited by Ad26.Mos4.HIV did not significantly differ from the neutralization capacity elicited by Ad26.Mos.HIV (data not shown). This demonstrates that the proposed combination of Ad26.Mos4.HIV and Clade C gp140 shows improved clade C pseudovirus recognition in the absence of negative effects on clade B pseudovirus recognition.

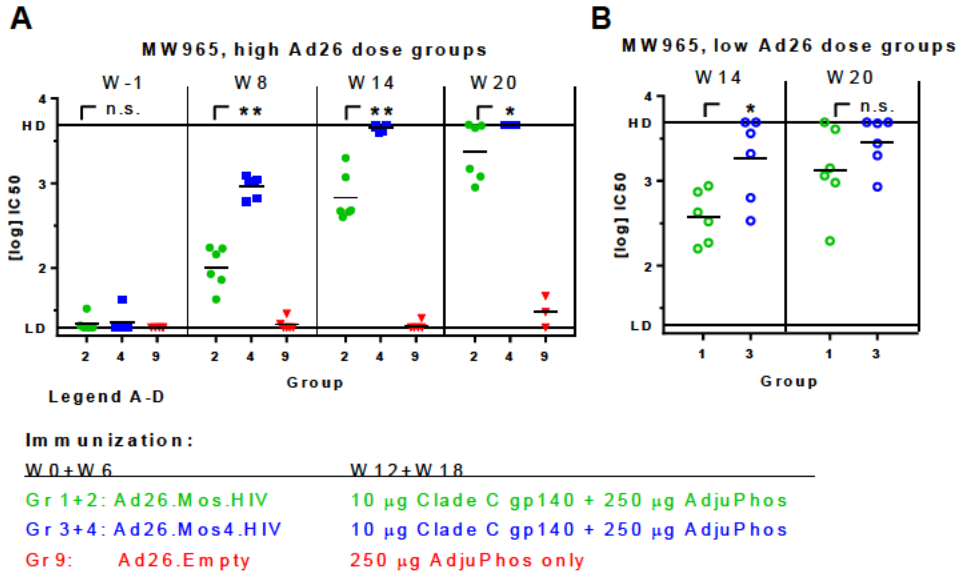

Figure 4-4 Virus neutralization titers against HIV-1 ENV pseudotyped virus particles (EVP's) in rabbits in the TZM-bl cell-based neutralization assay. Log10-transformed IC50 values were determined for the HIV-1 Env pseudotyped virus particles (EVP's) MW965 (Tier 1A clade C, A-B), at week -1, 8, 14, and 20 for the high adeno dose groups and at week 14 and 20 for the low adeno dose groups. Each dot represents the log10-transformed IC50 value of an individual rabbit, with group geometric means indicated as horizontal lines. HD: Highest dilution tested (upper solid line) LD: Lowest dilution tested (lower solid line). A one-way non-parametric comparison with control using Dunn's method for joint ranking was performed for each timepoint, EVP and dose group. Additional groups (# 5-8) assessing the immunogenicity of other components were included in the study but are not displayed here. Statistical analyses were performed including these groups and were thus corrected for the resulting multiple comparisons. * P<0.05; **P<0.01; n.s. = not significant.

4.8.6 Formulation changes

To improve stability of the vaccine, Ad26.Mos4.HIV will be formulated in a different buffer as used in immunogenicity studies 13-19/15-06 (NHP) and 0095-14 (rabbits). A potential influence of the formulation change on immunogenicity has been assessed in a mouse immunogenicity study using buffer-exchanged materials. No difference between the old and new formulation was observed. To exclude any effect of the new formulation on immunogenicity of Ad26.Mos4.HIV produced with the final process, an additional mouse immunogenicity study will be conducted comparing clinical trial materials produced in the old versus new formulation. Briefly, cohorts of mice will be immunized with 3 different dose levels of each formulation (typically $1x10^9$, $5x10^9$ or $1x10^{10}$ vp per animal and dose). Vaccine-induced humoral and cellular responses will be determined by ELISA and ELISpot, respectively. Data will be analyzed using a non-inferiority testing method comparing pre-change and post-change formulations across dose levels based on antigen-specific serum antibody binding data determined by ELISA.

The combination of Clade C gp140 and aluminum phosphate adjuvant will be administered either through a co-formulation or through pharmacy mixing of separate vials of Clade C gp140 and the adjuvant. A potential influence of the formulation change on immunogenicity has been evaluated in the rabbit immunogenicity study 0028-17. Briefly, rabbits were primed with a low dose of Ad26.Mos4.HIV to mimic the clinically applied prime immunizations and then boosted with 3 different dose levels of each Clade C gp140 formulation using the antigen-adjuvant ratio that was selected in clinical study

HIV-V-A004. Readout parameters included antigen-specific antibody binding (ELISA or comparable assay) and viral neutralization assays. Data were analyzed using a non-inferiority testing method comparing pre-change and postchange formulations across dose levels based on antigen-specific serum antibody binding data determined by ELISA. This study demonstrated non-inferiority of the co-formulation compared with the pharmacy mixture of Clade C gp140 and aluminum phosphate adjuvant.

4.9 Clinical studies

4.9.1 Clinical studies with related Adenovirus 26 vaccine, Ad26.ENVA.01

In Ad26.ENVA.01 a gene insert was used that coded for a clade A env protein, while Ad26.Mos4.HIV is a tetravalent vaccine containing 4 Ad26 vectors encoding mosaic antigens. These mosaic antigens were shown in pre-clinical NHP studies to increase the breadth of induced immune responses in comparison to antigens derived from circulation viruses or consensus virus sequences [7]. Ad26.Mos4.HIV consists of 2 Env antigen-encoding vectors (Ad26.Mos1.Env, Ad26.Mos2S.Env) and 2 Gag-Pol antigen-encoding vectors (Ad26.Mos1.GagPol, Ad26.Mos2.GagPol). Thus, each antigen is encoded by 2 complementary mosaic sequences that were designed with the aim of providing maximal coverage of globally circulating HIV-1 strains [6].

In 3 phase 1 studies (IPCAVD 001, IPCAVD 003, and IPCAVD 004 [41-44]), Ad26.ENVA.01, at IM doses over the range 10^9 to 10^{11} vp, was found to induce Env-specific humoral and cell-mediated responses when given on up to 3 occasions to more than 200 healthy participants. Ad26.ENVA.01 was generally well tolerated in these studies. An IM dose of $5x10^{10}$ vp was found to provide the optimal balance of immunogenicity and reactogenicity. Therefore, this Ad26.Mos4.HIV vaccine will be administered in a 1:1:1:1 ratio totaling $5x10^{10}$ vp for evaluation in the current HVTN705/VAC89220HPX2008 study.

In IPCAVD 003, 24 HIV-1 negative participants were randomized 3:1 to receive a single vaccination with Ad26.ENVA.01 or placebo. Eight of the participants were Ad26 seropositive at screening. The T-cell responses by IFNγ ELISpot assays were slightly lower in the baseline Ad26 seropositive participants; ICS and enzyme-linked immunosorbent assay responses proved comparable between participants who were Ad26 seropositive and Ad26 seronegative at baseline, both in peripheral blood and in colorectal mucosa. In addition, systemic and mucosal responses persisted for at least 1 year in the majority of participants after a single IM vaccine dose. These data suggest that the impact of baseline Ad26 nAbs, at the titers observed in that study, on the immunogenicity of this Ad26 vaccine is modest. Additionally, there were no consistent increases in Ad26-specific CD4+ T lymphocyte responses at mucosal surfaces following vaccination in either Ad26-seronegative or Ad26 seropositive participants.

In the IPCAVD 004 clinical trial (N=217) [41], the safety and immunogenicity of IM doses of Ad26.ENVA.01 and Ad35.ENV ($5x10^{10}$vp), administered in heterologous and homologous prime-boost regimens at 3- versus 6-month intervals, were evaluated. We observed that the baseline Ad26 nAb titers observed in South and East Africa did not impact the humoral or cellular immune responses elicited by the prototype Ad26.ENVA.01 vaccine. See Investigator's Brochure for additional information.

4.9.2 Clinical studies with vaccine components

Study	N	Product	Dose	Route
Completed Study				
HIV-V-A003	50	Clade C gp140 Clade C gp140/AP*	50/ 250 mcg 50/ 250 mcg	IM
Ongoing Studies				
HIV-V-A004	400	Ad26.Mos.HIV MVA-Mosaic Clade C gp140/AP	5×10^{10} vp 1×10^{8} pfu 50/250 mcg	IM
HPX1002	36	Ad26.Mos.HIV Clade C gp140	5×10^{10} vp 250 mcg	IM
HPX2004/HVTN 117	198	Ad26.Mos.HIV Ad26.Mos4.HIV Clade C gp140/AP	5×10^{10} vp 5×10^{10} vp 250 mcg	IM
Planned study				
HPX2003/HVTN 118	150	Ad26.Mos4.HIV Clade C gp140/AP Clade C gp140/AP + Mosaic gp140/AP	5×10^{10} vp 250 mcg 125 mcg each	IM

*AP, aluminum phosphate adjuvant, 425mcg

Three first-in-human (FIH) studies in healthy HIV-uninfected participants are currently ongoing or being analyzed. In these studies safety/tolerability and immunogenicity of Clade C gp140 (study HIV-V-A003), Ad26.Mos.HIV, MVA-Mosaic and Clade C gp140 (study HIV-V-A004, FIH for Ad26.Mos.HIV) and Ad26.Mos.HIV, Ad26.Mos4.HIV and Clade C gp140 (study VAC89220HPX2004/HVTN117, FIH for Ad26.Mos.4.HIV) are being evaluated. One additional study is ongoing, VAC89220HPX1002, which is a randomized, parallel-group, placebo-controlled, double-blind phase 1 study to evaluate the safety/tolerability and immunogenicity of different vaccine schedules with Ad26.Mos.HIV and Clade C gp140.

An additional FIH study in healthy HIV-uninfected adults is currently planned, in which safety/tolerability and immunogenicity of Ad26.Mos4.HIV, Clade C gp140 and Mosaic gp140 (study VAC89220HPX2003/HVTN118, FIH for Mosaic gp140) will be evaluated.

4.9.2.1 HIV-V-A003

HIV-V-A003 is a single-center, randomized, placebo-controlled, double-blind, FIH phase 1 study to evaluate safety/tolerability, and immunogenicity of 2 dose levels (50 and 250 mcg) of Clade C gp140, with or without aluminum phosphate adjuvant, in healthy HIV-uninfected adult participants. The schema is shown in Table 4-9. The clinical phase of the study was completed on 28 April 2016 (Last Subject Last Visit). The Clinical Study Report is currently in preparation.

All treatments were well tolerated. All adverse events were grade 1 or grade 2 in severity. The most frequent solicited systemic AEs were headache, nausea, and fatigue. Injection site pain was the main observed solicited reaction. It was reported by 3 participants in the low dose (LD) - no adjuvant (50 mcg Clade C gp140) group, by 2 participants in each of the other 3 active vaccine groups, and by 1 subject in the placebo group. The most frequent unsolicited AEs were upper respiratory tract infection (reported by 6 participants in the active vaccine groups, all events considered not related to treatment by the investigator), and back pain, cough, and hypertension (reported by 2 participants each in the active vaccine groups), compared with no participants in the placebo groups.

There were no SAEs, and none of the participants discontinued due to AEs.

Vaccine-induced binding antibody responses were investigated using ELISA performed at the Beth Israel and Deaconess Medical Center, Boston, MA, USA. Antibodies were mainly detected at 4 weeks after the boost injection (when tested at baseline, Day 29, and Day 57 [4 weeks after the 2nd injection]). Although the limited number of participants precludes a formal comparison, the highest response was observed in the high dose (HD) with adjuvant (250 mcg Clade C gp140/AP) group. See the Investigator's Brochure for more details.

Table 4-9 HIV-V-A003 Schema

Group	N	Dose	Month 0	Month 1 (day 29)
1	10	50 mcg	Clade C gp140	Clade C gp140
2	10	50 mcg	Clade C gp140/AP*	Clade C gp140/AP
3	5	0	Placebo for 50 mcg	Placebo for 50 mcg
4	10	250 mcg	Clade C gp140	Clade C gp140
5	10	250 mcg	Clade C gp140/AP	Clade C gp140/AP
6	5	0	Placebo for 250 mcg	Placebo for 250 mcg

* AP, aluminum phosphate adjuvant, 425 mcg

4.9.2.2 HIV-V-A004

HIV-V-A004 is a multi-center, randomized, parallel-group, placebo-controlled, double-blind phase 1/2a study to evaluate safety/tolerability, and immunogenicity of various prime/boost regimens containing Ad26.Mos.HIV, MVA-Mosaic, and/or Clade C gp140 (with aluminum phosphate adjuvant) components in approximately 400 healthy HIV-uninfected adult participants in the US, Uganda, Rwanda, South Africa, and Thailand. The schema is depicted in Table 4-10.

Table 4-10 HIV-V-A004 Schema

Group	N	Prime Month 0, Month 3	Boost Month 6, Month 12
1	50	Ad26.Mos.HIV	Ad26.Mos.HIV + 250 mcg Clade C gp140/AP*
2	50	Ad26.Mos.HIV	Ad26.Mos.HIV + 50 mcg Clade C gp140/AP
3	50	Ad26.Mos.HIV	Ad26.Mos.HIV + Placebo
4	50	Ad26.Mos.HIV	MVA-Mosaic + 250 mcg Clade C gp140/AP
5	50	Ad26.Mos.HIV	MVA-Mosaic +50 mcg Clade C gp140/AP
6	50	Ad26.Mos.HIV	MVA-Mosaic + Placebo
7	50	Ad26.Mos.HIV	250 mcg Clade C gp140 +Placebo
8	50	Placebo	Placebo + Placebo

*AP, aluminum phosphate adjuvant, 425 mcg

The study is currently ongoing and is still blinded for subjects and sites. At the time of the primary analysis (Week 28), results of which are described below, all subjects had received their 3rd vaccination or discontinued earlier. For more details, see the Investigator's Brochure.

The results from the primary analysis (Week 28) showed that all vaccine regimens were found to be well tolerated.

Solicited events, both local and systemic, were overall mild to moderate in severity. Injection site pain, headache and fatigue were the most frequent reported solicited events overall.

Most solicited systemic AE were reported after the first dose. The frequency of grade 3 related solicited systemic AEs was low, with headache and fatigue most frequently reported. Only one serious adverse event was assessed as related to the study products by the investigator, ie, severe allergic reaction/hypersensitivity:

> A 50 years-old male subject reported a severe allergic reaction/hypersensitivity having started 12 hours after first vaccine administration. Symptoms and signs mentioned by the subject included facial swelling, blurred vision, difficulty swallowing, and generalized rash of the face, extremities, and chest tightness. The subject was brought to the emergency room per ambulance. Clinical observations by the emergency room physician noted an alert and oriented person, with symptoms of, but with no clinical signs of allergic reaction (no rash, no lip/tongue/oropharyngeal/uvular swelling, normal findings of cardiovascular and respiratory system). Diphenhydramine was administered; no hospitalization or administration of corticosteroids occurred. The reaction was resolved within 1 day of onset. Concurrent medical conditions included PPD (not disclosed by the subject to the investigator) with hallucinatory decompensation and chest pain 1 week after the severe allergic reaction. The subject was withdrawn from the study due to non-compliance (history of PPD and non-reported PPD).
>
> Three on-study HIV infections occurred. For all 3 subjects, factors known to increase the risk for HIV infection were present.
>
> No deaths were reported.

All active vaccine regimens were immunogenic as most groups showed 100% of subjects having a detectable antibody response post the third vaccination. The ELISA utilizing the same ELISA antigen as used for protein boost immunization showed a clear increase in antibodies upon the Clade C gp140 boost (irrespective of vector boost) (Figure 4-5): for HIV Env Clade C C97ZA.012 the geometric mean ratio of the Ad26/Ad26+gp140HD group versus the Ad26/Ad26 group was 5.5 (with a 95% CI between 3.5 and 8.6). In addition, the ELISA showed a clear increase in antibodies upon boost with a high dose of the Clade C gp140 in the boost compared to the low dose, and inclusion of a vector in the boost with the Clade C gp140 in comparison to Clade C gp140 alone. Up to 100% cross clade antibody responses to clade A, B and C were detected in most vaccine groups. The Ad26/Ad26+gp140HD group showed highest humoral responses overall. Clear IgG3, ADCP and neutralizing antibody responses were detected. IgG subclass responses to the vaccine protein were of the IgG1 and IgG3 subtype, with little to no IgG2 and IgG4 detected in most vaccine groups.

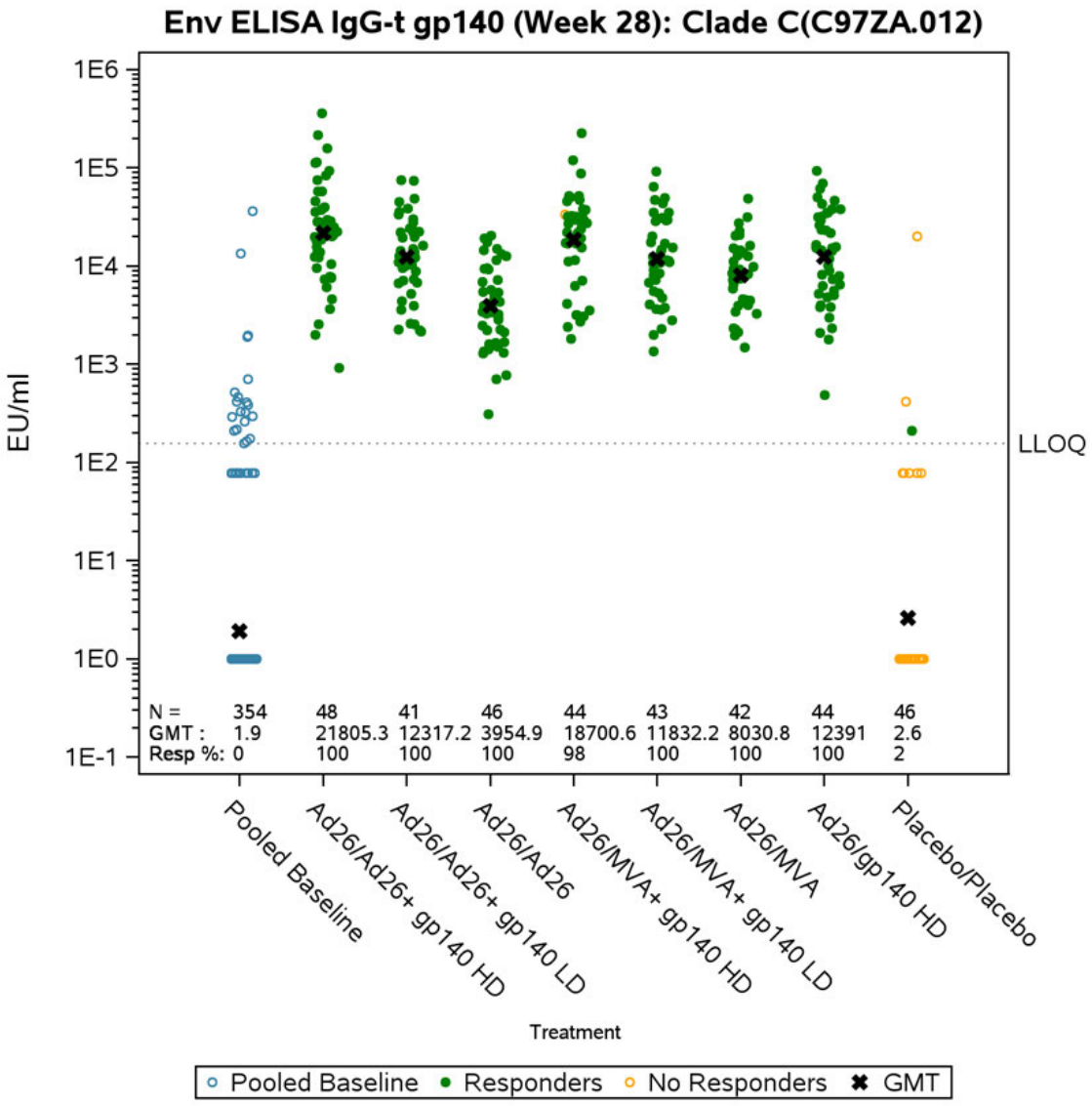

Figure 4-5 IgG gp140 ENV ELISA Clade C (C97ZA.012) (HIV-V-A004, Week 28 Analysis)

High numbers of responders were identified by ELISPOT (Figure 4-6), with some of the highest numbers of responders in the groups boosted with Ad26+gp140HD as well as MVA in the boost, with the highest median responses in the groups with MVA in the boost. Responses for CD4 and CD8 T cells producing IFNγ and/or IL2 were detected by Intracellular Cytokine Staining (ICS). A clear contribution of both the vector and the protein to boosting of both humoral and cellular immune responses was observed.

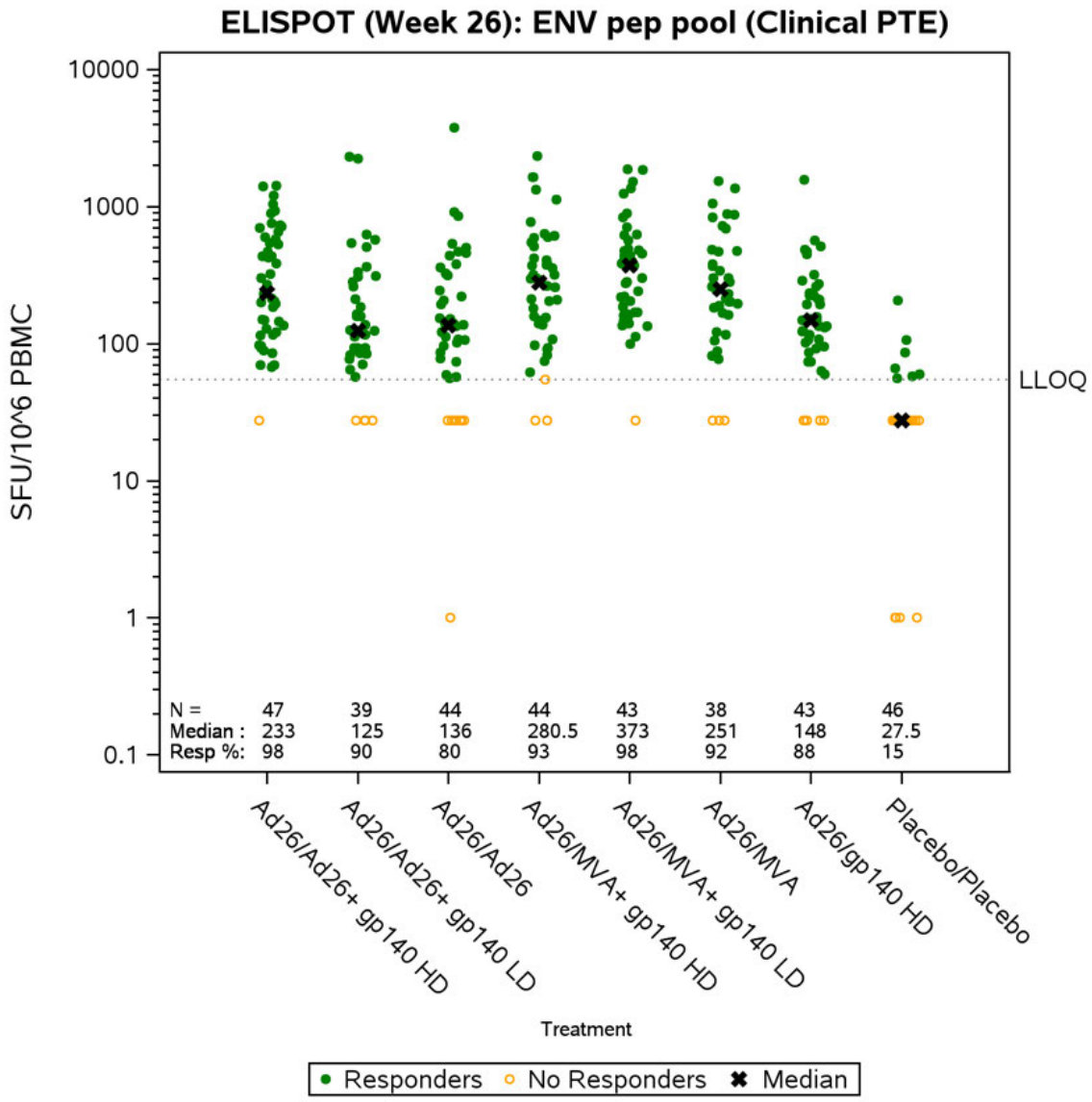

Figure 4-6 IFNγ ELISPOT ENV PTE peptide pool (HIV-V-A004, Week 28 Analysis)

4.9.2.3 HPX1002

VAC89220HPX1002 is a single-center, randomized, parallel-group, placebo-controlled, double-blind phase 1 clinical study in healthy HIV-uninfected adults to evaluate the safety, tolerability, and immunogenicity of different regimen durations (24 or 48 weeks) and different number of dose administrations with Ad26.Mos.HIV (3 or 4 dose administrations) and Clade C gp140 (2 or 3 dose administrations). The study is fully enrolled. Participants will be followed up until Week 72.

Table 4-11 HPX1002 study design

Group	Subgroup	N	Week 0	Week 8	Week 12	Week 24	Week 48
1	A	10	Ad26.Mos.HIV		Ad26.Mos.HIV	Ad26.Mos.HIV + Clade C gp140 (250 mcg + adjuvant)[a]	Ad26.Mos.HIV + Clade C gp140 (250 mcg + adjuvant)[a]
	B	2	Placebo		Placebo	Placebo + Placebo	Placebo + Placebo
2	A	10	Ad26.Mos.HIV + Clade C gp140 (250 mcg + adjuvant)[a]		Ad26.Mos.HIV + Clade C gp140 (250 mcg + adjuvant)[a]	Ad26.Mos.HIV + Clade C gp140 (250 mcg + adjuvant)[a]	
	B	2	Placebo + Placebo		Placebo + Placebo	Placebo + Placebo	
3	A	10	Ad26.Mos.HIV	Ad26.Mos.HIV + Clade C gp140 (250 mcg + adjuvant)[a]		Ad26.Mos.HIV + Clade C gp140 (250 mcg + adjuvant)[a]	
	B	2	Placebo	Placebo + Placebo		Placebo + Placebo	

[a] Sterile aluminum phosphate suspension is used as adjuvant. Aluminum content is 0.425 mg/0.5 mL dose.

4.9.2.4 HPX2004 (HVTN117)

VAC89220HPX2004 (HVTN117) is a randomized, parallel-group, placebo-controlled, double-blind phase 1/2a study to evaluate the safety/tolerability and immunogenicity of priming with trivalent Ad26.Mos.HIV and boosting with trivalent Ad26.Mos.HIV plus Clade C gp140 (with aluminum phosphate adjuvant), or priming with tetravalent Ad26.Mos4.HIV and boosting with Ad26.Mos4.HIV plus Clade C gp140 (with aluminum phosphate adjuvant). Approximately 198 participants will be enrolled in the study. Better Clade C responses are to be expected with the tetravalent Ad26.Mos4.HIV compared to the trivalent Ad26.Mos.HIV. A Data Review Committee (DRC) will review blinded safety data (4 weeks of follow-up) after 15% of participants have received their first injection (Ad26.Mos.HIV/Ad26.Mos4.HIV/placebo). Administration of the first dose of Ad26.Mos4.HIV (first injection) in VAC89220HPX2003 (see below), will be allowed only if no significant safety concerns are identified. Interim safety results on Ad26.Mos4.HIV are expected to be available before dosing of this vaccine component in the current HVTN705/VAC89220HPX2008 study.

Table 4-12 HPX2004 (HVTN117) schema

Group	N	Prime Month 0, Month 3	Boost Month 6, Month 12
1A	55	Ad26.Mos.HIV	Ad26.Mos.HIV + 250 mcg Clade C gp140/AP*
1B	11	Placebo	Placebo + Placebo
2A	110	Ad26.Mos4.HIV	Ad26.Mos4.HIV + 250 mcg Clade C gp140/AP*
2B	22	Placebo	Placebo + Placebo
Total 198 (165 vaccine/33 placebo)			

*Aluminum Phosphate

4.9.2.5 HPX2003 (HVTN 118)

VAC89220HPX2003 (HVTN 118) will be a randomized, parallel-group, placebo-controlled, double-blind phase 1/2a study in healthy HIV-uninfected adults to evaluate safety/tolerability and immunogenicity of different regimens of tetravalent Ad26.Mos4.HIV together with either Clade C gp140 or a combination of Mosaic and Clade C gp140 (with aluminum phosphate adjuvant). Approximately 150 participants will be enrolled in the study.

Table 4-13 HPX2003 (HVTN118) schema

Group	N	Prime Month 0, Month 3	Boost Month 6, Month 12
1	25	Ad26.Mos4.HIV	Ad26.Mos4.HIV + 250 mcg Clade C gp140/AP*
2	100	Ad26.Mos4.HIV	Ad26.Mos4.HIV + 125 mcg Clade C gp140/AP + 125 mcg Mosaic gp140/AP
3	25	Placebo	Placebo + Placebo
Total 150 (125 vaccine/ 25 placebo)			

*Aluminum Phosphate

4.10 Potential risks of study products and administration

4.10.1 Risks related to vaccines

Subjects may exhibit general signs and symptoms associated with administration of a vaccine, or vaccination with placebo, including fever, chills, rash, aches and pains, nausea, headache, dizziness and fatigue. These side effects will be monitored, but are generally short-term and do not require treatment.

Vasovagal reactions, lightheadedness or dizziness, mostly related to injections procedures, have also been observed, and therefore the vaccine should be administered while subjects are sitting or lying down and precautions must be taken to avoid falls potentially due to those events. All subjects will be observed in the clinic for 25-60 minutes after each vaccination.

Subjects may have an allergic reaction to the vaccination. An allergic reaction may cause a rash, hives or even difficulty breathing. Severe reactions are rare. Medications must be available in the clinic to treat serious allergic reactions. The effect of this vaccine on a

fetus or nursing baby is unknown so female subjects of child bearing potential will be required to agree to use birth control for sexual intercourse beginning prior to the first vaccination and through 3 months after the last vaccination. Women who are pregnant or nursing will be excluded from the study.

Risks related to VISP are discussed in Section 9.7.2.

4.10.2 Risks from blood draws

Blood drawing may cause pain, bruising, and, rarely, infection at the site where the blood is taken. Large volume blood draws can cause transient anemia.

4.10.3 Risks from HLA testing

Tests results can be used to provide information about how susceptible subjects are to certain diseases. Used inappropriately, this information could be discriminatory (for example, by insurance companies). Human Leukocyte Antigen typing can also be used to determine paternity. However, the blood samples donated will not be used for this purpose; they will be used only to provide study investigators information about the immune system. The results will be coded to protect subject identity.

4.10.4 Unknown risks

There may be other serious risks that are not known. Subjects may believe that this vaccine provides protection against acquiring HIV infection, and therefore practice riskier behavior. They will receive extensive counseling throughout the study to address this potential problem. It is not known if the study vaccines increase or decrease the chance of becoming HIV infected when exposed, or if upon becoming HIV infected, the person's disease course progresses faster or slower to AIDS. In previous HIV-efficacy studies utilizing Ad5, an increase in HIV-1 infections was observed in male vaccine recipients as compared with placebo recipients. Adenovirus serotype 26 is biologically substantially different than Ad5 and Ad26-based vaccines afford superior protective efficacy compared with Ad5-based vaccines against SIVMAC251 challenges in rhesus monkeys [16]. Further, Ad26 did not increase the number or activation status of total or vector-specific CD4+ T-lymphocytes at mucosal surfaces in humans following vaccination in a randomized, double-blind, placebo-controlled clinical study [44].

5 Objectives and endpoints

5.1 Primary objectives and endpoints

Primary objective 1:

To evaluate the preventive vaccine efficacy (VE) of a heterologous prime/boost regimen utilizing Ad26.Mos4.HIV and aluminum-phosphate adjuvanted Clade C gp140 for the prevention of HIV infection in HIV-seronegative women residing in sub-Saharan Africa from confirmed HIV-1 infections diagnosed between the Month 7 and Month 24 visits

Primary endpoint 1:

Vaccine efficacy as derived from confirmed HIV-1 infections diagnosed between the Month 7 and Month 24 visits

Primary objective 2:

To evaluate the safety and tolerability of a heterologous prime/boost regimen utilizing Ad26.Mos4.HIV and aluminum-phosphate adjuvanted Clade C gp140 for the prevention of HIV infection in HIV-seronegative women residing in sub-Saharan Africa

Primary endpoint 2:

Local and systemic reactogenicity signs and symptoms for 3 days after each vaccination, adverse events for 30 days after each vaccination, and serious adverse events, AESIs, and adverse events leading to early participant withdrawal or early discontinuation of study product(s) administration for the entire duration of the study.

5.2 Secondary objectives and endpoints

Secondary objective 1:

To evaluate vaccine efficacy from enrollment through 24 months

Secondary endpoint 1:

HIV-1 infection diagnosed after enrollment through 24 months post enrollment

Secondary objective 2:

To evaluate vaccine efficacy from enrollment through the end of the study if Stage 2 occurs

Secondary endpoint 2:

HIV-1 infection diagnosed after enrollment through the end of the study

Secondary objective 3:

To evaluate vaccine efficacy from month 12 through month 24

Secondary endpoint 3:

HIV-1 infection diagnosed after month 12 through 24 months post enrollment

Secondary objective 4:

To evaluate vaccine efficacy from month 12 through the end of the study if Stage 2 occurs

Secondary endpoint 4:

HIV-1 infection diagnosed after month 12 through the end of the study post enrollment

Secondary objective 5:

To evaluate the immunogenicity of the vaccine regimen

Secondary endpoint 5:

Immune responses at the study visits following the third and fourth vaccinations from assays based on the HVTN Laboratory Assay Algorithm such as vaccine-specific binding antibodies and T-cell responses.

Secondary objective 6:

To evaluate immunogenicity and immune response biomarkers among vaccine recipients after the third vaccination as correlates of risk of subsequent HIV acquisition and correlates of vaccine efficacy, if deemed applicable.

Secondary endpoint 6:

Immune responses from assays based on the HVTN Laboratory Assay Algorithm (available at https://atlas.scharp.org/) and/or more assays down-selected from a larger pool of pilot studies, in HIV-1–infected vaccine cases and HIV-1–uninfected vaccine controls

Secondary objective 7:

To evaluate VE adjusting for various demographic and other baseline characteristics

Secondary endpoint 7:

HIV-1 infection diagnosed after the third vaccination by demographic and other baseline characteristics

Secondary objective 8:

If significant positive evidence of vaccine efficacy from month 7 through 24 months is seen, to assess if and how vaccine efficacy depends on genotypic characteristics of HIV such as signature mutations

Secondary endpoint 8:

HIV-1 infection diagnosed after month 7 through Month 24 and genotypic characteristics of viral sequences from HIV-1–infected participants at HIV-1 diagnosis, such as signature site mutations

Secondary objective 9:

To evaluate and compare genomic sequences of viral isolates from HIV-1–infected vaccine and placebo recipients, and use sieve analysis methods to assess whether VE differs by genotypic or phenotypic characteristics of exposing HIVs and whether there is evidence of vaccine-induced immune pressure on the viral sequences

Secondary endpoint 9:

Viral sequences from HIV-1–infected participants at the earliest available postinfection timepoint and possible subsequent visits

5.3 Exploratory objectives

Exploratory objective 1:

To evaluate vaccine effects ("vaccine activity") on virologic and immunologic outcomes (eg, HIV-1 viral load (VL) and postdiagnosis CD4+ T-cell count) among HIV-1–infected participants for 6 months post diagnosis accounting for ARV use

Exploratory objective 2:

To explore the association between the vaginal microbiome as well as genital inflammation, and HIV infection risk

Exploratory objective 3

To evaluate early and innate immune responses (eg, whole blood transcriptomics, serum cytokines) one day after the third vaccination (ie, the first protein boost) as correlates of risk of subsequent HIV acquisition

Exploratory objective 4:

To evaluate local and systemic reactogenicity signs and symptoms that arise from day 4 to day 7 at a subset of clinical research sites

Exploratory objective 5:

To further evaluate the immunogenicity of the vaccine regimen, additional immunogenicity assays may be performed, and assays may be performed on samples from other timepoints, based on the HVTN Laboratory Assay Algorithm

Exploratory objective 6:

To assess use of biomedical interventions and biological and behavioral factors in the study cohort and how they modify vaccine efficacy

Exploratory objective 7:

To evaluate the role of host genetic factors in the immune response to the vaccine regimen and in vaccine effects on study endpoints

Exploratory objective 8:

To perform comparative analyses of correlates of risk identified in HVTN 705 and those identified in other HIV vaccine efficacy studies

Exploratory objective 9:

To conduct analyses related to furthering the understanding of HIV, immunology, vaccines, and clinical trial conduct

6 Statistical considerations

6.1 Study populations

The following study populations or analysis sets are used for addressing the study objectives. This terminology is used throughout this protocol and the statistical analysis plan (SAP).

1. Full Analysis Set (FAS): all randomized participants who receive at least 1 vaccine administration

2. Modified Intent-to-Treat (MITT) Population: participants in the FAS who are HIV-1 uninfected on the date of first vaccination.

3. Per-Protocol (PP) Population: participants in the FAS who are HIV-1 uninfected 4 weeks after the 3rd vaccination visit, who received all planned vaccinations at the first 3 vaccination visits within the respective visit windows and have no other major protocol deviations that were judged to possibly impact the efficacy of the vaccine.

4. Full Immunization Set (FIS): participants in the FAS who are HIV-1 uninfected 4 weeks after the 4th vaccination visit and who receive all planned vaccinations within the respective visit windows.

5. At risk Immunogenicity Cohort (IC-at risk): participants in the FAS who are selected for measurement of immune response endpoints at the primary immunogenicity timepoints and who are HIV-1 uninfected 4 weeks after the 3rd vaccination visit, who have no other major protocol deviations that were judged to possibly impact the efficacy of the vaccine.

6. Per Protocol Immunogenicity Cohort (IC-PP): Participants in the IC-at risk who received all planned vaccinations at the first 3 vaccination visits within the respective visit windows.

The MITT population and the FAS are very similar but not identical to a full Intention-to-Treat Cohort (ie, all randomized participants); the FAS differs by excluding randomized volunteers who do not enroll (ie, don't receive any vaccinations); and the MITT population is the subset of the FAS that also excludes randomized participants discovered later to be HIV-1 positive by day 0. Because of blinding and the brief length of time between randomization and enrollment—typically no more than 4 working days—we expect almost all randomized volunteers to be in the FAS. Given that eligibility for the study requires recent evidence of being HIV-1 uninfected (within 45 days prior to enrollment), we expect almost all enrolled participants to also be in the MITT Cohort.

The analyses of safety will be performed on the FAS. The primary analysis of vaccine efficacy will be based on the PP population. Secondary analyses of vaccine efficacy will be based on the MITT population and the FIS. Analyses of vaccine immunogenicity and immune correlates of risk will be based on IC-at risk, IC-PP and the FIS (for those with immunogenicity outcomes).

Since this is a proof-of-concept trial, all efficacy analyses will be done according to the as treated principle (ie, actually received treatments), except for analyses using the MITT population.

In addition, 4 cohorts of participants who are diagnosed with HIV-1 infection during the trial are analyzed for addressing various study objectives. Terminology for these cohorts is defined in Table 6-1, which will be used throughout the protocol and SAP.

Table 6-1 Cohorts of HIV-1–infected study participants

Cohort Name	Definition of cohort
MITT infected by 24 Months cohort	Participants in the MITT population who are diagnosed with HIV-1 infection during the follow-up period after enrollment through the Month 24 visit.
MITT infected by end of study cohort	Participants in the MITT population who are diagnosed with HIV-1 infection during the follow-up period after enrollment through the end of the study.
Per-Protocol infected by 24 Months cohort	Participants in the PP population who are diagnosed with HIV-1 infection during the follow-up period on or after the Month 7 visit through the Month 24 visit.
Per-Protocol infected by end of study cohort	Participants in the PP population who are diagnosed with HIV-1 infection during the follow-up period on or after the Month 7 visit through the end of the study.

6.2 Objectives

The primary, secondary, and exploratory objectives are defined in Section 5. Primary and secondary endpoints are described in Sections 5.1 and 5.2.

Since this is a proof-of-concept study, primary analyses of the HIV-1 infection endpoint and of vaccine activity endpoints will be based on the PP population (Section 6.1). Secondary analyses of the vaccine efficacy objectives will be based on the MITT population, the PP population and the FIS.

Safety analyses will be based on the FAS. Analyses of vaccine immunogenicity and immune correlates of risk will be based on IC-at risk, IC-PP and the FIS (for those with immunogenicity outcomes).

6.3 Primary analysis of vaccine efficacy

We define the vaccine efficacy parameter, VE(7-24), for the primary analysis as 1 minus the cumulative incidence ratio (vaccine/placebo) of the HIV-1 endpoint between Month 7 and Month 24 after enrollment in the PP population.

The cumulative incidence parameters used to define VE(7-24) will be estimated using the transformed Nelson-Aalen estimator for the cumulative hazard function at Month 24 in the PP population.

The target parameter for the primary analysis of VE is VE(7-24). A two-sided 95% log-cumulative-hazard based Wald confidence interval will be reported for VE(7-24), without adjustment for interim monitoring.

The primary analysis also reports a test of the null hypothesis

H0: VE(7-24) = 0% against the alternative hypothesis H1: VE(7-24) ≠ 0%

using a 2-sided α=0.05 level Wald test of the equality of log cumulative hazard functions at Month 24 in the PP population for the vaccine group and the placebo group. The p-value of this test will also be reported, where this p-value will not be adjusted for interim monitoring.

6.3.1 Analysis to determine whether or not the trial will advance into Stage 2

If the lower bound of the 2-sided 95% confidence interval for VE(7-24) is > 0% (equivalently, the 1-sided p-value for testing H0: VE(7-24) ≤0% vs. H1: VE(7-24)>0% is below 0.025), trial participants will continue blinded follow-up through the end of the study (timepoint of final analysis). On the other hand, if the lower bound of the 95% confidence interval for VE(7-24) is ≤ 0%, Stage 2 will not occur and the trial participants will be unblinded at the end of Stage 1 (timepoint of primary/final analysis). Pending availability and outcome of primary analysis results, participants will continue their normal scheduled visits beyond Month 24.

6.4 Accrual and sample size

6.4.1 Sample size calculation for proof of concept endpoints (vaccine efficacy of a prime-boost regimen to prevent HIV-1 infection)

A targeted total of 2,600 HIV-uninfected adult women will be recruited. Trial participants will be enrolled over a 14-month period approximately and randomized to a placebo or vaccine regimen (1:1 randomization). Participants will receive vaccinations at Months 0, 3, 6, and 12 and be followed for HIV infection for a period of at least 2 years (stage 1) after enrollment until the primary analysis at the end of stage 1 is performed (when the last subject reaches the month 24 visit). If H0: VE(7-24) ≤ 0% is rejected at the end of Stage 1, then the trial participants will continue blinded follow-up for at least 36 months (stage 2). If stage 2 does not occur, then all trial participants will be unblinded at the end of stage 1.

The trial is designed so that a 0.025-level Wald test has approximately 90% power in the PP population to reject the null hypothesis:

H0: VE(7-24) ≤ 0% versus the alternative hypothesis H1: VE(7-24) > 0% if the level of VE(7-24) is 50%.

The sample size calculations are based on the power of a 1-sided 0.025-level Wald test for comparing cumulative incidences of HIV-1 infection by the Month 24 visit between randomized groups, in the presence of the sequential monitoring described below. Power is computed by simulating a large number of efficacy trials under assumptions described below using the R package seqDesign [46,47] ,

The following assumptions are made for the sample size calculations:

- 10% annual dropout incidence in each of the study groups
- halved VE in the first 7 months after enrollment
- 14-month uniform accrual with halved accrual during the first 3 months
- visits approximately every 3months for HIV-1 diagnostic tests
- 4.2% annual HIV-1 incidence that is constant over time in the placebo group
- Sequential monitoring of vaccine efficacy for the following outcomes:
    - Potential harm [Conclude that VE(0-24)< 0% based on the one-sided exact binomial test of the proportion of infections assigned to the vaccine group]
    - Non-efficacy [Conclude that VE(7-24) < 40% based on a 2-sided 95% nominal confidence interval for VE(7-24) lying completely below 40%, and VE(0-24) < 40% based on a 2-sided 95% nominal confidence interval for VE(0-24) lying completely below 40%]
    - High efficacy [Conclude that VE(0-36) >70% based on a 2-sided 95% nominal confidence interval for VE(0-36) lying completely above 70%]
- 5% of subjects with missed vaccinations, i.e. subjects who would be discarded from the PP population

Based on the references in Section 4.4.5, the estimated annual incidence rate of HIV-1 in Sub-Saharan women is 5.0% (total HIV acquisition events divided by total person years at risk). For our power calculations a more conservative incidence of 4.2% was chosen to provide some margin for reduced incidence due to PrEP use during the study. Table 6-2 provides the power estimates to reject the null hypothesis for the assumed incidence of 4.2%, as well as for situations with lower (3.1%) or higher (5.5%) than expected incidence. With the current sample size this trial would still have approximately 80% power to detect VE(7-24) of 50% or more if the incidence in the control group were to drop as low as 3.1%. As can be seen from the table (VE=0%) the type I error is controlled in the presence of sequential monitoring.

Table 6-2 Estimated power to detect different levels of VE(7-24) in the presence of sequential monitoring for VE, based on a randomized sample size of N = 1300/group§:

	Power to reject H0: VE(7-24) ≤ 0%with a 1-sided 0.025-level test		
True VE(7-24)	**Annual placebo incidence 3.1%**	**Annual placebo incidence 4.2%**	**Annual placebo incidence 5.5%**
0%	2.11	2.06	1.70
10%	6.69	5.72	7.79
20%	15.66	17.63	22.39
30%	28.94	41.86	48.38
40%	57.19	66.03	75.61
50%	79.70	87.56	93.26
60%	93.56	98.00	98.19
70%	98.90	99.60	99.60
80%	99.70	99.70	99.90
90%	100.00	100.00	100.00

§ Halved VE in first 7 months
N=1300:1300 placebo:vaccine group
14-month enrollment period with a uniform enrollment rate, halved in the first 3 months
10% annual dropout

Under the incidence assumptions, the study sample size also provides sufficient and slightly higher power for the secondary MITT analyses assuming VE is halved during the first 7 months after enrollment, as shown in Figure 6-1 below (dark blue bars).

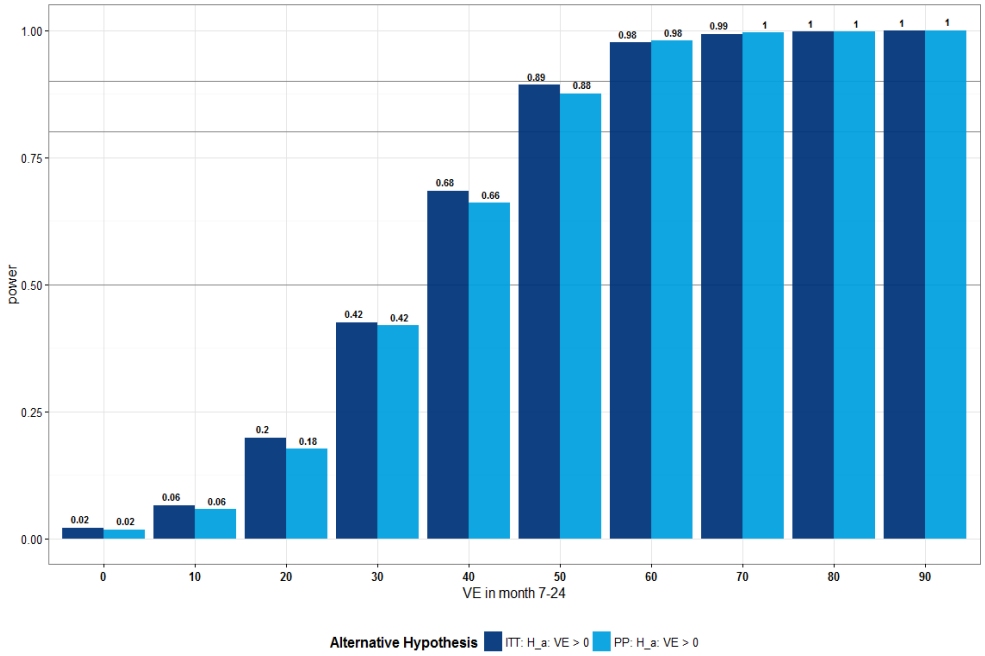

Figure 6-1 Estimated power to detect different levels of VE(0-24) and VE(7-24) based on a sample size of N = 1300/group and an HIV-1 incidence of 4.2%

6.4.2 Sample size calculations for primary safety endpoint

A total of 2600 participants in the trial provides enough power for assessing safety of the vaccine regimen. The probability of observing at least 1 adverse event occurring at a rate

of 1/100 is >99.9% with 1300 participants. The probability of observing at least 1 adverse event occurring at a rate of 1/1,000 is 73% with 1300 participants.

No (S)AE observed in the vaccine group (N = 1300) would provide us with 95% confidence that the true incidence is no more than 0.29%.

If no events will be observed for a specific adverse event, for example an SAE, then the Bayesian posterior probability that the adverse event rate is below 1/1,000 equals 89.3% for 1300 participants when using Jeffrey's prior, and the posterior probability that the adverse event rate is below 1/100 equals 99.99%. If no AE is observed in the vaccine group (N = 1300), the upper limit of the equal-tailed 95% credible interval will be 0.19%.

As shown in Figure 6-2, there is 80% power to detect a small event rate difference between the vaccine and placebo groups using a 2-sided 0.05-level Fisher's exact test. For example, if the true safety event rate in the placebo group is 2%, there is 80% power to detect an event rate difference of 2% or higher in the vaccine group.

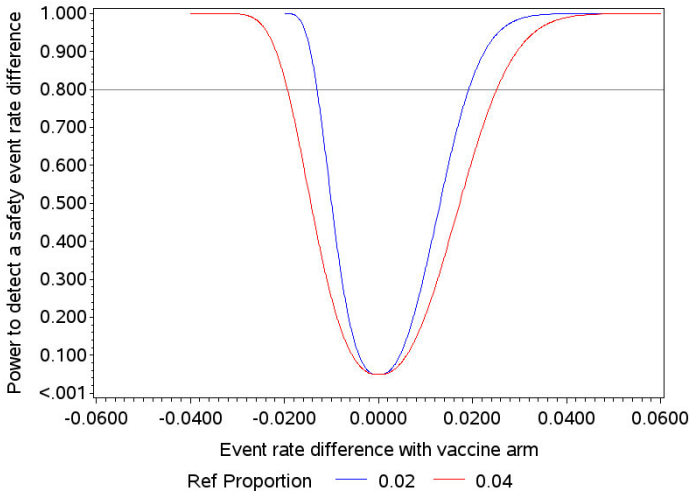

Figure 6-2 Power to detect safety event rate differences between the vaccine (N = 1300) and placebo (N = 1300) arms for assumed true event rates of 2% (blue) and 4% (red) in the placebo arm

6.5 Trial monitoring

The trial will be formally monitored. This may lead to a modification or termination of the trial.

Table 6-3 lists the specification of parameters under which the power calculations were done. The details of the actual monitoring plan will be discussed in the Statistical Analysis Plan (SAP). Table 6-4 provides the different outcome probabilities when applying these sequential monitoring rules to the study, assuming an incidence of 4.2%. More details on the individual monitoring rules follow in the subsequent sections.

Table 6-3 Sequential monitoring specifications used for power calculations

Monitoring Type	Hypotheses	Statistical Method	Monitoring Plan	Timing of Analyses
Potential Harm	H0: VE(0-24) ≥ 0% vs. H1: VE(0-24) < 0%	Exact 1-sided binomial test of the fraction of infections assigned to receive the vaccine.	Constant p-value cut-off controlling the FWER at α=5%	After every MITT infection starting by the 10th infection until the first non-efficacy analysis
Non- Efficacy	H0: VE(0-24) ≥ 40% H0: VE(7-24) ≥ 40% vs. H1: VE(0-24) <40% H1: VE(7-24)<40%	For both VE(0-24) and VE(7-24): LCLs of 95% CI<0% and UCLs of 95%CI<40%	Unadjusted 95% confidence intervals around VE(0-24) and VE(7-24)	6-monthly starting once all participants reach Month 13, and 60 MITT infections are observed, then through the end of Stage 1
High Efficacy	H0: VE(0-36) ≤ 70% vs. H1: VE(0-36) > 70%	95%CI for VE(0-36) lies completely above 70%	Unadjusted 95% confidence interval around VE(0-36)	Harmonized with non-efficacy monitoring starting when 150 participants reach 36 months of follow-up. If Stage 2 occurs, one additional analysis half-way through Stage 2

Table 6-4 Outcome probabilities under sequential monitoring of vaccine efficacy

True VE(7-24)	Potential Harm	Non-efficacy (interim)	High Efficacy	UC Power (PP) (end Stage 1)	UC Power (MITT) (end Stage 1)
0	4.57 %	81.18 %	0.0%	2.18 %	2.18 %
10	2.88 %	65.97 %	0.0%	7.19 %	7.41 %
20	1.70 %	41.08 %	0.0%	20.17 %	21.79 %
30	0.90 %	19.32 %	0.0%	43.33 %	45.80 %
40	0.64 %	5.55 %	0.0%	69.90 %	73.10 %
50	0.31 %	1.26 %	0.0%	90.86 %	91.58 %
60	0.21 %	0.14 %	0.02 %	98.58 %	98.70 %
70	0.14 %	0.00 %	0.13 %	99.92 %	99.83 %
80	0.04 %	0.00 %	1.20 %	99.99 %	99.95 %
90	0.03 %	0.00 %	8.26 %	100.0%	99.97 %

In the simulations, the start of the non-efficacy and high-efficacy monitoring was set to assure that the trial was continued to be monitored for harm until the M13 time point under the trial pre-trial assumptions:
VE halved in first 7 months
N=1300:1300 placebo:vaccine group
4.2% annual incidence in placebo group
10% annual dropout
5% missed vaccinations

6.5.1 Monitoring for potential harm

The unblinded statistician will continuously monitor the trial for early evidence of a higher infection rate of MITT HIV-1 infection in the vaccine group compared to the placebo group to ensure participant safety. The monitoring will start from the 10th infection onward

(pooled over vaccine and placebo) and after each additional infection it is decided whether the stopping boundary has been reached [VE(0-24)<0%]. It will continue until the first analysis for non-efficacy is performed. The potential harm monitoring uses the one-sided exact binomial test resulting in a boundary that is determined via controlling the overall type I error rate across tests at 5%. Figure 6-3 shows for example that when 60 infections are counted, with 40 infections in the active vaccine arm, the boundary has been reached. In general, the null hypothesis will be rejected when the probability that the observed infections are from the vaccine arm is significantly larger than 50% (blue line in Figure 6-3). If the prespecified stopping boundary is reached, then the unblinded statisticians will immediately inform the DSMB. In addition, the DSMB chair will be updated on the accruing unblinded HIV-1 infection data after each confirmed MITT infection. This monitoring guideline is chosen to allow stopping for prudence as early as possible, maximizing participant safety.

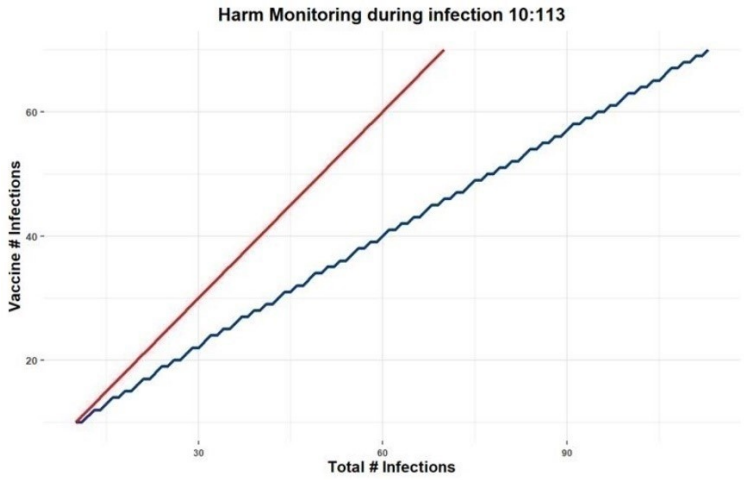

Figure 6-3 Potential harm stopping boundary for the total number of HIV-1 infections in the active vaccine regimen versus placebo. Blue line is the potential harm stopping boundary, red line displays the case that all of the infections are in the vaccine arm.

6.5.2 Monitoring for non-efficacy

Non-efficacy will be followed by means of a double monitoring rule. These analyses will start when all participants have reached the Month 13 timepoint, and 60 MITT infections have occurred, then approximately every 6 months until the end of Stage 1. The boundary is hit when the VE 95% CI upper limit is <40% for both VE(0-24) and VE(7-24). Figure 6-4 shows the cumulative probability of hitting a non-efficacy bound in time. If the non-efficacy boundary is hit prior to reaching the end of Stage 1, the final analysis will be performed at the time the boundary is hit (prior to reaching the end of Stage 1) and the results will be reported. For participants that are already beyond 24 months of follow-up, no further follow-up will be required. For the other participants, a decision would be made, based on input from the OG, on whether to still follow them up until their month 24 visit. Stage 2 will not be started in this case.

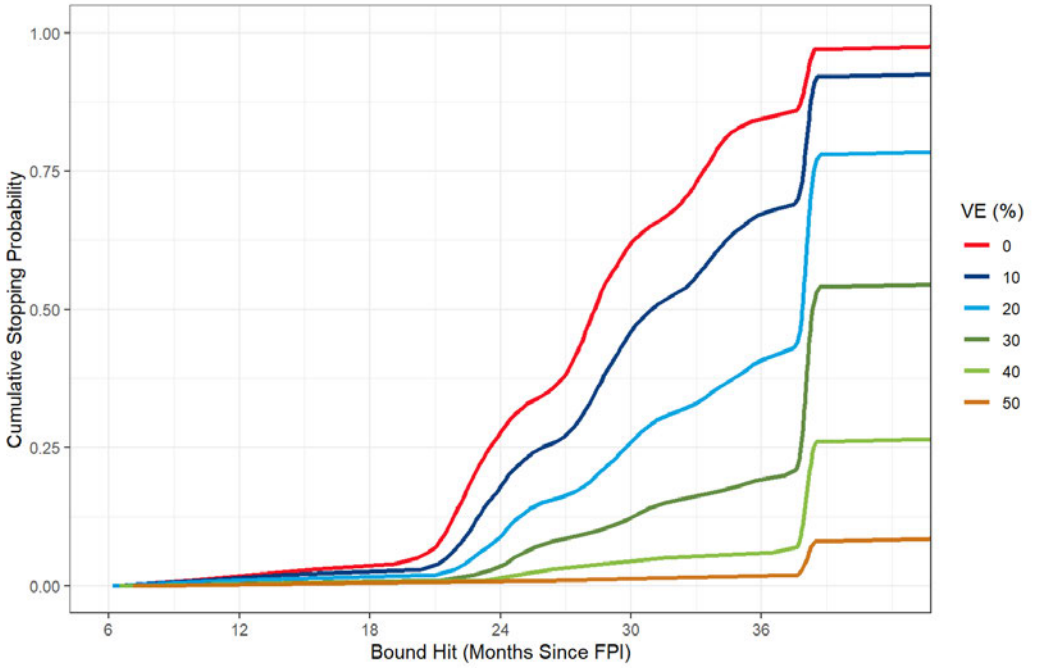

Figure 6-4 Cumulative probability of hitting a potential harm or non-efficacy bound in time

Additionally, the DSMB will monitor the trial for non-efficacy with the aim to detect early on scenarios where the null hypothesis would not be rejected if the study were to continue to its end. At each DSMB meeting, conditional power calculations will be provided. These calculations will help in interpreting the projected treatment-arm-pooled infection totals. The conditional power is defined as the power for rejecting H0: VE(0-24) ≤ 0%, setting VE(0-24) = 50%. The conditional power will be calculated given the actual enrollment, dropout, and treatment-arm-pooled HIV-1 incidence data from trial start through the time of the interim analysis and compared to a benchmark of 50% power.

6.5.3 Monitoring for high efficacy

The DSMB will also monitor for high vaccine efficacy (95% CI lower limit for VE [0-36] >70%). Interim analyses for high efficacy will begin following the first participant's 36 month visit (end of Stage 2), and will be performed together with those for non-efficacy monitoring. Stopping for high efficacy would entail study unblinding and offering placebo recipients vaccination. The criterion for continuing from Stage 1 to Stage 2 is given in Section 6.3.

Figure 6-5 shows the trial design. Whereas the timing of interim analyses is event-driven, the total number of Stage 1 infections is not fixed. The end of Stage 1 is when the last enrolled participant reaches the Month 24 visit. Ensuring all participants are followed to Month 24 maximizes power and precision for assessing VE(7-24).

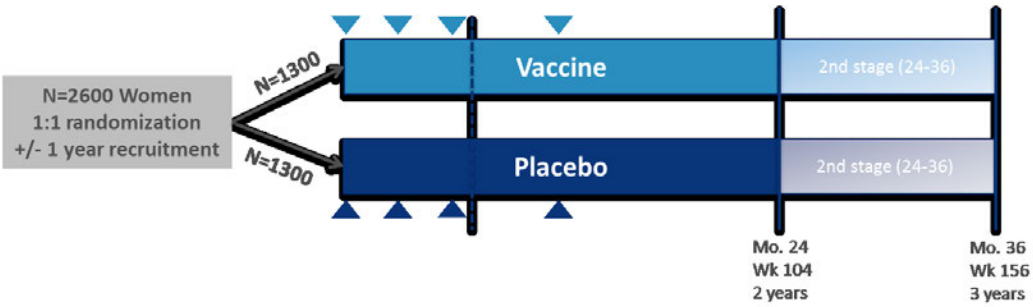

Figure 6-5 Study design and stages. Triangles represent vaccination timepoints. Stage 1 is the calendar time since trial start until the last enrolled participant reaches the Month 24 visit. Stage 2 is the calendar time since the last enrolled participant reached the Month 24 visit until the last enrolled participant reaches the Month 36 visit. Participants who completed their Month 36 visit will be followed further with scheduled clinical visits until the end of the study (ie, when the last Month 36 visit is completed).

Early stopping of the study while Stage 1 is still ongoing will be recommended if the potential harm or non-efficacy boundary is met at a pre-specified analysis time, in which case the final analysis will be performed and Stage 2 will not occur. If none of the stopping boundaries are reached by the end of Stage 1, the primary endpoint of vaccine efficacy from Month 7 through Month 24 [VE(7-24)] is evaluated at the end of Stage 1. If the lower bound of the 2-sided 95% confidence interval for VE(7-24) is > 0% at the end of Stage 1 (equivalently, the 1-sided p-value for testing H0: VE(7-24) ≤ 0% is below 0.025), trial participants will continue blinded follow-up through Stage 2. On the other hand, if at the Stage 1 efficacy analysis the lower bound of the 95% confidence interval for VE(7-24) ≤ 0%, Stage 2 will not occur even if none of the stopping boundaries were reached by the end of Stage 1.

6.5.4 Monitoring for futility to assess VE

The DSMB monitors the trial for futility to assess VE defined as overly slow progress toward the Target Number of Infections (defined below) or insufficient conditional power for the primary analysis given the observed data. This may occur if enrollment rates are too low and/or the incidence of HIV-1 infections is too low. The DSMB will monitor for futility to assess VE every 6 months starting no later than 12 months after the first participant is enrolled. All relevant data that are available regarding enrollment and dropout will be provided to the DSMB. Timings can be adapted at the discretion of the DSMB in case signs of futility to assess VE are seen. According to the design, enrollment is expected to take place within 14 months.

We define the Target Number of Infections to be the number of primary HIV-1 endpoints after Month 7 and by the Month 24 visit such that the trial has approximately 70% power to reject H0: VE ≤ 0% if VE = 50% (using a 1-sided 0.025-level cumulative hazard-based Wald test).

The DSMB will be provided with the distribution of infections together with the expected distribution of infections under the design assumptions (incidence, VE and dropout rates; table below). The conditional power for rejecting VE(7-24) ≤ 0% using the actual data, given a true vaccine efficacy of 50% in the future data, will also be provided to the DSMB at each interim analysis. More details on the data provided to the DSMB will be described in the DSMB charter.

Table 6-5 Probability distribution of all Stage 1 infections

VE*	Percentiles of distribution of number of Infections (Post Month 7)														
	1%	2.5%	5%	10%	20%	30%	40%	50%	60%	70%	80%	90%	95%	97.5%	99%
0%	105	110	112	116	120	123	126	128	131	134	138	143	148	151	154
50%	76	80	83	85	89	92	94	97	99	102	105	110	113	117	120
	Percentiles of distribution of number of Infections (MITT)														
0%	157	164	169	173	179	183	186	190	193	196	201	206	212	216	221
50%	124	128	131	136	141	144	147	150	153	156	160	166	171	174	179

*Halved in first 7 months
N=1300:1300 placebo:vaccine group
4.2% annual incidence in placebo group
10% annual dropout

6.5.5 Monitoring for expanding enrollment

At the time of each DSMB meeting for which there are enough HIV infections to conduct an analysis, the HVTN 705/HPX2008 Oversight Group (OG) will receive the results of the operational futility analysis (which are blinded—pooled across treatment arms). The OG will also be provided additional information at the time of a special DSMB meeting scheduled to take place approximately 2 months before the completion of enrollment—based on ongoing projections of time until full enrollment by study statisticians. For this meeting, study statisticians will prepare a report that includes the calculations typically in an operational futility analysis, along with additional guidance as to whether the HIV incidence – pooled over treatment arms—is "too low" or "acceptable" to sufficiently power the study. (The details of this guidance will be described in the study SAP.) The report will be shared both with the DSMB and with the study OG. The OG will then have the opportunity to decide whether, if incidence is lower than anticipated but not so low that operational futility is declared, to expand enrollment beyond the originally planned 2600, in order to maintain the ability of the study to meet the study's primary objectives. The meeting will be timed before but near to the end of enrollment, to allow a potential decision to expand enrollment to occur before the enrollment apparatus is scaled down, while also allowing the decision to be made on the maximal amount of primary endpoint infections. The OG will also be presented information on the degrees of enrollment expansion that would be required in order to achieve conditional power and treatment-arm-pooled infection totals at various levels.

6.5.6 Roles of study statisticians

HVTN SDMC statisticians will be "blinded" or "unblinded". Janssen statisticians will remain blinded until the primary analysis at Month 24. During protocol development and after primary follow-up is completed, there will be no distinction between the roles; both types of statisticians will be responsible for designing and analyzing the study. During the primary follow-up period, however, only the unblinded statisticians will see interim data broken down by treatment arm. Their role will be to conduct the interim monitoring and to produce and present reports on accruing data to the study DSMB. During the primary follow-up period, blinded statisticians will see only the interim data pooled across treatment arms. This way, blinded statisticians can assist protocol leadership in making decisions about modifications to the protocol without being influenced by interim efficacy results.

6.6 Assessment of PrEP use

The use of oral FTC/TDF as PrEP (either off-study or provided in the study) may impact study outcomes (e.g., by lowering HIV-1 incidence with a loss of study power). Dried blood spot (DBS) samples will be collected and stored for assessment of quantitative concentrations of intracellular tenofovir diphosphate (TFV-DP) (see Section 11.9). DBS samples will be collected and stored at pre-specified DBS sample collection days, which may vary by study site. The calendar-based selection of sample collection days will ensure representative sampling of participants across the entire visit schedule at all sites throughout the study. A detailed DBS sampling plan, specified in the statistical analysis plan, will be developed to prospectively monitor FTC/TDF use during the trial to inform the DSMB and to assess the potential impact of FTC/TDF use on endpoint accrual during the study. Prevalence of oral PrEP use will be reported both as the estimated percentage of person-years at-risk during highly adherent PrEP use (above the concentration of TFV-DP designated as consistent with daily dosing) and the estimated percentage of person-years at-risk during any detectable PrEP use (concentration of TFV-DP above the lower limit of detection for the DBS assay). PrEP use measures will be reported by arm to the DSMB in the closed session; in addition both the OG and the protocol team leadership will see pooled estimates of FTC/TDF use.

6.7 Randomization of treatment assignments

The randomization sequence will be obtained by computer-generated random numbers through an Interactive Web Response System (IWRS) under provision by the sponsor and provided to each HVTN CRS. The randomization will be stratified by HVTN CRS and done in blocks to ensure balance across arms. At each institution, the pharmacist with primary responsibility for dispensing study products is charged with maintaining security of the treatment assignments. Participants and site staff will be blinded as to the treatment group assignment throughout both Stage 1 and Stage 2, if it occurs.

6.8 Blinding

The study participants, study-site personnel (except for those with primary responsibility for study vaccine preparation and dispensing), and investigators will be blinded to study vaccine allocation throughout the study (both Stage 1 and Stage 2, if it occurs).

Study product assignments are accessible to those HVTN CRS 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 DSMB members also are unblinded to treatment assignment in order to conduct review of trial safety and efficacy.

When a participant leaves the trial prior to study completion, the participant will be told she must wait until all participants have completed follow-up to learn her treatment assignment.

Emergency individual unblinding decisions will be made by the site investigator when the timing is critical for medical management. Otherwise, the HVTN 705/HPX2008 PSRT should be consulted before emergency unblinding occurs.

6.9 Statistical analysis

Statistical analysis will be done by close collaboration between HVTN SDMC and Janssen Vaccines. A general description of the statistical methods to be used to analyze the efficacy, safety and immunogenicity data is outlined below. Specific details will be provided in the SAP.

This section describes the final study analysis with unblinded treatment groups. The primary efficacy analysis will be performed when all participants have reached the Month 24 visit or discontinued earlier. The primary analyses of safety will be performed on the FAS. The primary analysis of vaccine efficacy will be based on the PP population. Secondary analyses of vaccine efficacy will be based on the MITT population and the FIS. Analyses of vaccine immunogenicity and immune correlates of risk will be based on the IC-at risk, IC-PP and the FIS. In general vaccination assignment will follow the as treated principle, except for analyses using the MITT population.

Analyses for primary endpoints will be performed using SAS and R.

No multiplicity corrections will be used unless stated otherwise.

6.9.1 Analysis variables

The analysis variables consist of baseline participant characteristics, safety, efficacy, and immunogenicity.

6.9.2 Baseline comparability

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

6.9.3 Safety/tolerability analysis

All safety analyses will be tabulated by treatment group (active vaccine, placebo) and based on the FAS.

6.9.3.1 Reactogenicity

The number and percentage of participants experiencing each type of reactogenicity sign or symptom will be tabulated by severity and treatment group 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 each injection visit. 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.

6.9.3.2 AEs and SAEs

AEs will be summarized using MedDRA System Organ Class and preferred terms. Tables will show by treatment group the number and percentage of participants experiencing an AE within a System Organ Class or within preferred term category by severity and 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 and by causal relationship to study product. Formal statistical testing comparing

arms is not planned since interpretation of differences must rely heavily upon clinical judgment. Parallel analyses will include all AEs and AEs leading to participant withdrawal or early discontinuation of study product(s).

A listing of SAEs reported to the Janssen Global Medical Safety Group will provide details of the events including severity, relationship to study product, time between onset and last vaccination, and number of vaccinations received.

6.9.3.3 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 treatment arm and including the reason for discontinuation.

6.9.4 Immunogenicity analysis

Data from quantitative assays will be summarized (tabulated/graphically presented per timepoint available) by treatment group: N, geometric means and corresponding 95% CIs, percentage positive responses/responders (if available).

Data from qualitative (ie, positive or negative) assays will be summarized by tabulating the frequency of positive responses for each assay by group at each timepoint that an assessment is performed.

6.9.5 Vaccine efficacy analyses

6.9.5.1 Primary vaccine efficacy analysis: HIV-1 infection

The primary analysis will be done in the PP population where participants becoming HIV infected or dropping out before the visit after the third vaccination or not having received all of the first 3 vaccinations within the specified time window or having a major specified protocol deviation, will be excluded from the analysis. The date of HIV-1 diagnosis will be the draw date of the first sample that leads to a positive test result by the diagnostic algorithm described in Section 10.3. Dropouts will be censored at the time of their last HIV-1 negative test. To evaluate the primary vaccine efficacy endpoint, the ratio of cumulative incidences of HIV-1 infection between Months 7 and 24 (vaccine vs. placebo), estimated using the transformed Nelson-Aalen cumulative hazard function estimator and tested via a Wald test. Cox proportional hazards model will also be used for estimating VE(7-24), measured by 1 minus the hazard ratio (vaccine vs. placebo) and for score testing whether the VE(7-24) differs from 0%.

As a sensitivity analysis to the primary analysis of vaccine efficacy in the PP population, targeted minimum loss-based estimation (TMLE) is used to estimate of this vaccine efficacy parameter. In particular, TMLE is used to estimate the cumulative incidences of HIV-1 infection over time for each of the vaccine and placebo groups, through to the final timepoint Month 24. These estimates will then be contrasted to estimate the primary VE parameter VE(7-24), which adjusts for covariates. This analysis has the ability to correct for bias due to measured participant covariates that predict both per-protocol status and HIV-1 infection. Iterative mean-based TMLE is used for this analysis as described in Benkeser et al. (2016, PhD dissertation). As part of the implementation of the TMLE the Super Learner is used to generate initial estimates of the conditional censoring distribution and the iterated conditional means [48]. The Super Learner library includes both parametric and nonparametric learning algorithms that are specified in the

SAP; the SAP also specifies the input variables considered by the different learning algorithms. Each learning algorithm considers adjustment for baseline demographic covariates and the baseline behavioral risk score built via supervised learning.

In addition, to assess potential time-effects of vaccine efficacy, the Kaplan-Meier method will be used to plot the estimated cumulative incidence rates over time for the vaccine and placebo groups. This method will be used to estimate cumulative vaccine efficacy over time, defined as [(1–ratio(vaccine/placebo) of cumulative incidence by time t)*100%], with the method of Parzen, Wei and Ying applied to estimate pointwise and simultaneous 95% Cis [49].

6.9.5.2 Secondary vaccine efficacy analyses: HIV-1 acquisition

Vaccine efficacy for MITT and FIS

The analyses above will be repeated for the MITT population and the FIS.

Durability of vaccine efficacy

Vaccine efficacy over all available follow-up times will be assessed using the method of Parzen, Wei, and Ying described above, as well as with nonparametric hazard ratio estimation [50] and with Cox proportional hazards modeling with time-dependent covariates. If the analysis suggests time-invariant vaccine efficacy, then the Cox model will be used to estimate proportional-hazards VE and to test for VE different from zero accounting for all available follow-up time. The SAP will provide details of these analyses.

Vaccine efficacy in the absence of PrEP

The primary analysis will be repeated, where only MITT or PP infections in participants who were not using prophylactic ARVs at the time of HIV-1 diagnosis or first evidence of infection will be included in the analysis. Plasma drug levels will be used to determine eligibility for this analysis. A participant is eligible if the plasma drug levels are undetectable at the diagnosis visit and at the visit with earliest evidence of HIV-1 infection (if different from the diagnosis visit). Since the ARVs are only detectable in plasma through roughly 14 days [51] and some participants may have become infected before the 14-day window, with this approach we are not assured that all those included in the analysis were not using prophylactic ARVs at the time of infection. Therefore, an additional analysis will address this issue by also excluding participants from the HIV-1 acquisition analysis if they self-reported drug use in the last 30 days at either the diagnosis visit or the last visit prior to diagnosis. This additional analysis will evaluate uninfected participants without accounting for data on their plasma drug levels.

Vaccine efficacy by subgroups

The primary analysis may be repeated for different subgroups. Wald tests for interaction in the context of Cox proportional hazards models will be used to test for evidence of differential vaccine efficacy by subgroup.

6.9.5.3 Secondary vaccine efficacy and activity analyses of viral sequences

Vaccine efficacy and genotypic characteristics

A variety of methods may be used to assess genotypic characteristics of HIV as potential effect modifiers of VE(0-24) [52]. The TMLE for estimating cumulative incidences in the presence of competing risks will also be applied as described in Benkeser et al. (2016, PhD dissertation). Like the TMLE efficacy analysis, this analysis incorporates Super Learner to increase precision. Details can be found in the SAP.

Sieve analysis

Acquisition sieve analysis methods including genome scanning tests described in Gilbert et al. and their extensions will be used to evaluate the relationship between VE(7-24) and VE(0-24), and the genotypic differences between the incoming exposing/founder sequences and the HIV insert sequence(s) represented in the vaccine construct [53].

7 Selection and withdrawal of participants

Participants will be healthy, HIV-uninfected (seronegative) women who comprehend the purpose of the study and have provided informed consent. Volunteers will be recruited and screened; those determined to be eligible, based on the inclusion and exclusion criteria, will be enrolled in the study. Final eligibility determination will depend on results of medical history, hemoglobin, physical examinations, and answers to self-administered and/or interview questions.

Investigators should always use good clinical judgment in considering a volunteer's overall fitness for trial participation. Some volunteers may not be appropriate for enrollment even if they meet all inclusion/exclusion criteria. Medical, psychiatric, occupational, limited ability to communicate, or other conditions may make evaluation of safety and/or immunogenicity difficult, and some volunteers may be poor candidates for retention.

Determination of eligibility, taking into account all inclusion and exclusion criteria, must be made within 45 days prior to enrollment unless otherwise noted in Sections 7.1 and 7.2.

The HVTN 705/HPX2008 PSRT will be consulted for questions related to selection and withdrawal of participants that are not specifically addressed below or are ambiguous. All attempts to achieve consensus will be made. In the event that consensus cannot be reached, the participant will not be enrolled.

7.1 Inclusion criteria

General and Demographic Criteria

1. **Persons born female (assigned female sex at birth)**

2. **Age** of 18 to 35 years

3. **Sexually active**, defined as having had sexual intercourse with a male partner at least twice in the past 30 days prior to screening, and is considered by the site staff to be at risk for HIV infection.

4. **Access to a participating HVTN CRS** and willingness to be followed for the planned duration of the study

5. Ability and willingness to provide **informed consent**

6. **Assessment of understanding**: volunteer demonstrates understanding of this study prior to first vaccination with verbal demonstration of understanding of all questions.

7. **Is not enrolled in, and agrees not to enroll in, another study** of an investigational research agent until the participant is unblinded or their study participation ends, whichever occurs last

8. **Good general health** as shown by medical history, hemoglobin, and physical exam. STIs are not exclusionary, unless deemed to impact general health by the investigator.

HIV-Related Criteria:

9. Willingness to receive **HIV test results**

10. **Willingness to discuss HIV infection risks** and willing to receive HIV risk reduction counseling and appropriate referrals to minimize HIV acquisition, as applicable.

Laboratory Inclusion Values

Virology

11. **Negative HIV-1 and -2 blood test**: Sites may use locally available assays that have been approved by HVTN Laboratory Operations.

Hemoglobin

12. **Hemoglobin** ≥ 10.5 g/dL

Reproductive Status

13. Negative beta human chorionic gonadotropin (β-HCG) pregnancy test performed prior to vaccination on the day of initial vaccination. Persons who are NOT of reproductive potential due to having undergone total hysterectomy or bilateral oophorectomy (verified by medical records), are not required to undergo pregnancy testing.

14. All volunteers must:

- Agree to consistently use effective contraception (Appendix B) for sexual activity that could lead to pregnancy from at least 21 days prior to enrollment through 3 months after the last vaccination. Effective contraception is defined as using 1 of the following methods of birth control.
    - Intrauterine device (IUD),
    - Hormonal contraception (in accordance with applicable national contraception guidelines), or
    - Any other contraceptive method approved by the HVTN 705/HPX2008 PSRT;
- Or not be of reproductive potential, such as having been diagnosed with premature menopause (with no menses for 1 year) or having undergone hysterectomy, bilateral oophorectomy, or tubal ligation.

15. **Volunteers must also agree not to seek pregnancy through alternative methods**, such as artificial insemination or *in vitro* fertilization until 3 months after the last vaccination

7.2 Exclusion criteria

General

1. **Blood products** received within 90 days before first vaccination

2. **Investigational research agents** received within 30 days before first vaccination

3. **Intent to participate in another study** of an investigational research agent or any other study that requires non-HVTN HIV antibody testing or blood draws during the planned duration of the HVTN 705/VAC89220HPX2008 study

4. **Pregnant or breastfeeding** (breastfeeding includes wet nursing)

Vaccines and other Injections

5. **HIV vaccine(s)** received in a prior HIV vaccine trial. For volunteers who have received control/placebo in an HIV vaccine trial, the HVTN 705/HPX2008 PSRT will determine eligibility on a case-by-case basis.

6. **Non-HIV experimental vaccine(s) received within the last yea**r in a prior vaccine trial. Exceptions may be made for vaccines that have subsequently undergone licensure. For volunteers who have received control/placebo in an experimental vaccine trial, the HVTN 705/HPX2008 PSRT will determine eligibility on a case-by-case basis. For volunteers who have received an experimental vaccine(s) greater than 1 year ago, eligibility for enrollment will be determined by the HVTN 705/HPX2008 PSRT on a case-by-case basis.

7. **Live attenuated vaccines** received within 30 days before first vaccination or scheduled within 14 days after injection (eg, measles, mumps, and rubella [MMR]; oral polio vaccine [OPV]; varicella; yellow fever)

8. **Any vaccines that are not live attenuated vaccines** and were received within 14 days prior to first vaccination (eg, tetanus, pneumococcal, Hepatitis A or B)

Immune System

9. **Immunosuppressive medications** received within 6 months before first vaccination. (Not exclusionary: [1] corticosteroid nasal spray; [2] inhaled corticosteroids; [3] topical corticosteroids for mild, uncomplicated dermatitis; or [4] a single course of oral/parenteral prednisone or equivalent at doses < 60 mg /day and length of therapy < 11 days with completion at least 30 days prior to enrollment.)

10. **Serious adverse reactions to vaccines** including history of anaphylaxis and related symptoms such as hives, respiratory difficulty, angioedema, and/or abdominal pain. (Not excluded from participation: a volunteer who had a non-anaphylactic adverse reaction to pertussis vaccine as a child.)

11. **Immunoglobulin** received within 60 days before first vaccination

12. **Immunodeficiency**

Clinically significant medical conditions

13. **Clinically significant medical condition**, physical examination findings, clinically significant abnormal laboratory results, or past medical history with clinically significant

implications for current health. A clinically significant condition or process includes but is not limited to:

- A process that would affect the immune response,
- A process that would require medication that affects the immune response,
- Any contraindication to repeated injections or blood draws,
- A condition that requires active medical intervention or monitoring to avert grave danger to the volunteer's health or well-being during the study period,
- A condition or process for which signs or symptoms could be confused with reactions to vaccine, or
- Any condition specifically listed among the exclusion criteria below.

14. **Any medical, occupational, or other condition** that, in the judgment of the investigator, would interfere with, or serve as a contraindication to, protocol adherence, assessment of safety or reactogenicity, or a volunteer's ability to give informed consent

15. **Psychiatric condition or substance abuse issue that precludes compliance with the protocol, in the opinion of the investigator**. Specifically excluded are persons with psychoses within the past 3 years, ongoing risk for suicide, or history of suicide attempt or gesture within the past 3 years.

16. **Active tuberculosis (TB) disease**

17. **Uncontrolled BP elevation**: systolic blood pressure (SBP) ≥ 160 mm Hg or diastolic blood pressure (DBP) ≥ 100 mm Hg

18. **Bleeding disorder** (diagnosed by a doctor) contraindicating IM injection and/or blood draws, based on investigator's judgment

19. **Malignancy** (Not excluded from participation: Volunteer who has had malignancy excised surgically and who, in the investigator's estimation, has a reasonable assurance of sustained cure or who is unlikely to experience recurrence of malignancy during the period of the study)

20. History of hereditary **angioedema**, acquired angioedema, or idiopathic angioedema

21. **Employee** of the investigator or study site, with direct involvement in the proposed study or other studies under the direction of that investigator or study site

7.3 Participant departure from vaccination schedule or withdrawal

This section concerns an individual participant's departure from the vaccination schedule. Pause rules for the trial as a whole are described in Section 12.3.

7.3.1 Delaying vaccinations for a participant

Under certain circumstances, a participant's scheduled vaccination will be delayed. The factors to be considered in such a decision include but are not limited to the following:

- Within 45 days prior to any study injection
  - Receipt of blood products or immunoglobulin
- Within 30 days prior to any study injection
  - Receipt of live attenuated vaccines
  - Receipt of allergy treatment with antigen injections
- Within 14 days prior to any study injection
  - Receipt of any vaccines that are not live attenuated vaccines (eg, Hepatitis)
- Pre-vaccination abnormal vital signs or clinical symptoms that may mask assessment of vaccine reaction.
- Pregnancy: For participants, who become pregnant, no study vaccinations will be given; except for participants who may have been pregnant during the study but are no longer pregnant as shown by 2 negative urine pregnancy tests taken from 2 different urine samples; in this circumstance, the HVTN 705/HPX2008 PSRT should be consulted to determine if the participant may resume vaccinations.

In order to avoid vaccination delays and missed vaccinations, participants who plan to receive licensed vaccines should be counseled to schedule receipt of these substances, when possible, outside the intervals indicated above. The effects of these substances on safety and immunogenicity assessments and their interactions with study vaccines are unknown. Therefore, if circumstances allow, these substances should also be avoided in the 4 weeks following study vaccination.

7.3.2 Participant departure from vaccination schedule

Every effort should be made to follow the vaccination schedule per the protocol. Vaccinations should generally not be administered outside the visit window period specified in the HVTN 705/VAC89220HPX2008 Study Specific Procedures (SSP). If a participant cannot be vaccinated within the allowable window, the PSRT can determine on a case-by-case basis if the participant can still be vaccinated.

If a participant misses a vaccination, the participant should be asked to continue study visits. The participant should resume the vaccination schedule with the next vaccination unless there are circumstances that require further delay or permanent discontinuation of vaccination (see Sections 7.3.1 and 7.3.3).

7.3.3 Discontinuing vaccination for a participant

Under certain circumstances, an individual participant's vaccinations will be temporarily or permanently discontinued. Specific events that will result in stopping a participant's vaccination schedule include:

- Co-enrollment in a study with an investigational research agent (rare exceptions allowing for the continuation of vaccinations may be granted with the unanimous consent of the HVTN 705/HPX2008 PSRT).
- Clinically significant condition (ie, a condition that affects the immune system or for which continued vaccinations and/or blood draws may pose additional risk), including but not limited to the following:

- Pregnancy: Vaccinations will be stopped while a participant is pregnant. If the participant is no longer pregnant, vaccinations may resume (Sections 7.3.1 and 7.3.2);
- Any related SAE;
- Any grade 4 local or systemic reactogenicity symptom, lab abnormality, or AE that is subsequently considered to be related to vaccination;
- Any grade 3 lab abnormality or other clinical AE (exception: fever or vomiting and subjective local and systemic symptoms) that is subsequently considered to be related to vaccination after confirmation by the PSRT. For grade 3 injection site erythema and/or induration, upon review, the PSRT may allow continuation of vaccinations; or
- Clinically significant type 1 hypersensitivity reaction associated with study vaccination. Consultation with the HVTN 705/HPX2008 PSRT is required prior to subsequent vaccinations following any type 1 hypersensitivity reaction associated with study vaccination; or

- Investigator determination in consultation with PSRT and sponsor (eg, for repeated nonadherence to study staff instructions; recognition of non-disclosed preexisting condition of clinical significance).

Such participants should be counseled on the importance of continuing with the study and strongly encouraged to participate in follow-up visits and protocol-related procedures per the protocol for the remainder of the trial, unless medically contraindicated.

In addition, vaccinations will be stopped for participants diagnosed with HIV infection.

7.3.4 Participant termination from the study

Under certain circumstances, an individual participant may be terminated from participation in this study. Specific events that will result in early termination include:

- Participant refuses further participation,
- Participant relocates and remote follow-up or transfer to another HVTN CRS is not possible,
- HVTN CRS determines that the participant is lost to follow-up,
- Investigator decides, in consultation with the PSRT, to terminate participation (eg, if participant exhibits inappropriate behavior toward clinic staff).
- Any condition where termination from the study is required by applicable regulations.

8 Study product preparation and administration

CRS pharmacists should consult the Site Investigational Product Procedures Manual, provided by the Sponsor for further instructions on study product preparations. The protocol schema is shown in Table 3-1. See the Investigator's Brochures for further information about study products.

All study products must be stored at 2°C - 8°C. Refer to the Site Investigational Product Procedures Manual for additional guidance on study drug preparation, handling, and storage.

8.1 Vaccine regimen

The schedule of vaccination is shown in Table 3-1.The vaccine is given on Months 0, 3, 6, and 12 and additional information is given below.

Group 1

Treatment 1 (T1):

Ad26.Mos4.HIV 5 x 10^{10} vp to be administered as 0.5 mL IM into the LEFT deltoid (unless medically contraindicated) on Months 0, 3, 6, and 12.

AND

Clade C gp140 (250 mcg) mixed with Aluminum phosphate adjuvant to be administered as 0.5 mL IM into the RIGHT deltoid (unless medically contraindicated) on Months 6 and 12.

Group 2

Placebo 2 (P2):

Placebo for Ad26.Mos4.HIV to be administered as 0.5 mL IM into the LEFT deltoid (unless medically contraindicated) on Months 0, 3, 6, and 12.

AND

Placebo for Clade C gp140 / Aluminum phosphate adjuvant to be administered as 0.5 mL IM into the RIGHT deltoid (unless medically contraindicated) on Months 6 and 12.

8.2 Study product formulation

8.2.1 Ad26.Mos4.HIV [Labeled as Ad26.Mos4.HIV]

The Ad26.Mos4.HIV vaccine is formulated as a tetravalent vaccine 1x10^{11} vp/mL , and is supplied as a colorless to slightly yellowish/brownish solution for IM injection. The

vaccine will be provided in single-use vial containing a volume to deliver 0.5 mL of $5x10^{10}$ Ad26 Mos4.HIV.

The study product is described in further detail in the Investigator's Brochure (IB).

8.2.2 Placebo for Ad26.Mos4.HIV

Sodium chloride for injection USP, 0.9% will be used as the placebo for Ad26.Mos4.HIV.

8.2.3 Clade C gp140 (250 mcg) + Aluminum Phosphate adjuvant

Clade C gp140 and aluminum phosphate adjuvant will either be co-formulated or supplied in separate vials:

The Clade C gp140 (250mcg) and aluminum phosphate adjuvant are co-formulated as a white to off-white suspension in single-use vials with a deliverable volume of 0.5 mL of 0.5 mg/mL Clade C gp140 and 0.85 mg/mL aluminum phosphate adjuvant. The study product is described in detail in the Investigator's Brochure (IB).

OR

Clade C gp140 is formulated as a colorless to slightly yellowish/brownish solution and will be practically free from particles. Clade C gp140 will be supplied at a nominal strength of 1 mg/mL. The aluminum phosphate adjuvant is formulated as a white to off-white suspension with a nominal aluminum content of 1.7 mg/mL. It will be mixed with Clade C gp140 at the site pharmacy prior to administration. The study products are described in detail in the IB.

8.2.4 Placebo for Clade C gp140 /Aluminum Phosphate adjuvant

Sodium chloride for injection, 0.9% will be used as the placebo for Clade C gp140/Aluminum Phosphate adjuvant.

8.3 Preparation of study products

CRS pharmacists should consult the Site Investigational Product Procedures Manual, provided by the Sponsor for further instructions on study product preparations.

8.3.1 Procedures to preserve blinding

The pharmacist will prepare all doses for administration and dispense to the clinic. In order to preserve blinding, the pharmacist will place an overlay on ALL the syringes.

8.4 Administration

All injections are to be given IM by blinded study staff in the deltoid indicated.

CRS pharmacists should consult the Site Investigational Product Procedures Manual, provided by the Sponsor for further instructions on study product administration.

8.5 Acquisition of study products

Ad26.Mos4.HIV, the co-formulated Clade C gp140 with aluminum phosphate adjuvant, the individual Clade C gp140 product, the individual aluminum phosphate adjuvant, and Placebo will be provided by Janssen Vaccines & Prevention B.V. (Leiden, The Netherlands).

Once an HVTN CRS has completed all procedures for study start, study products can be obtained by the pharmacist from the sponsor.

8.6 Pharmacy records

The HVTN CRS pharmacist is required to maintain complete records of all study products. The pharmacist of record is responsible for maintaining randomization codes and randomization confirmation notices for each participant in a secure manner.

8.7 Product quality complaint handling

A product quality complaint (PQC) is defined as any suspicion of a product defect related to manufacturing, labeling, or packaging, ie, any dissatisfaction relative to the identity, quality, durability, or reliability of a product, including its labeling or package integrity. A PQC may have an impact on the safety and efficacy of the product. Timely, accurate, and complete reporting and analysis of PQC information from studies are crucial for the protection of participants, for the investigators, and for the sponsor, and are mandated by regulatory agencies worldwide. Janssen has established procedures in conformity with regulatory requirements worldwide to ensure appropriate reporting of PQC information; all studies conducted by the sponsor or its affiliates will have to be conducted in accordance with those procedures.

8.7.1 Procedures

All initial PQCs must be reported to Janssen by the HVTN CRS staff within 3 business days after being made aware of the event. If the defect is combined with an SAE, the CRS staff must report the PQC to Janssen according to the SAE reporting timelines. A sample of the suspected product should be maintained for further investigation if requested by the sponsor.

8.7.2 Contacting Sponsor Regarding Product Quality

The names (and corresponding contact information) of the individuals who should be contacted regarding product quality issues can be found on the protocol home page on the HVTN Members' site (https://members.hvtn.org/protocols/hvtn705).

8.8 Final disposition of study products

All unused study products must be returned to the sponsor or destroyed after the study is completed or terminated according to the instructions provided in the Site Investigational Product Procedures Manual.

9 Clinical procedures

The schedule of clinical procedures is shown in Appendix G and Appendix H.

All study data will be recorded on paper source documents prior to being entered into the study database (see the HVTN 705/VAC89220HPX2008 SSP).

During the COVID-19 pandemic, procedures are in place so that study visits may be conducted remotely, such as via phone, text message, email, or other electronic means, in lieu of, or in combination with, in-person visits (see HVTN 705/VAC89220HPX2008 SSP for additional details).

9.1 Informed consent

Informed consent is the process of working with participants so that they fully understand what will and may happen to them while participating in a research study. The HVTN informed consent form documents that a participant (1) has been informed about the potential risks, benefits, and alternatives to participation, and (2) is willing to participate in an HVTN study. Informed consent encompasses all written or verbal study information HVTN CRS staff provide to the participant, before and during the trial. HVTN CRS staff will obtain informed consent of participants according to HVTN policies and procedures.

The informed consent process continues throughout the study. Key study concepts should be reviewed periodically with the participant and the review should be documented. At each study visit, HVTN CRS staff should consider reviewing the procedures and requirements for that visit and for the remaining visits. Additionally, if any new information is learned that might affect the participants' decisions to stay in the trial, this information will be shared with trial participants. If necessary, participants will be asked to provide revised informed consent.

An HVTN CRS may employ recruitment efforts prior to the participant consenting. For example, some HVTN CRSs use a telephone script to prescreen people before they come to the clinic for a full screening visit. Participants must sign a screening or protocol-specific consent before any procedures are performed to determine eligibility. HVTN CRSs must submit recruitment and prescreening materials to their IRB/EC and any applicable regulatory entity (RE) for human subjects protection review and approval.

Note: An RE is "Any group other than the local IRB/EC responsible for reviewing and/or approving a clinical research protocol and site-specific ICFs [informed consent forms] prior to implementation at a site." CRSs are responsible for knowing the requirements of their applicable REs.

9.1.1 Screening consent form

Some HVTN CRSs have approval from their IRB/EC and any applicable RE to use a general screening consent form that allows screening for an unspecified HIV vaccine trial. In this way, HVTN CRS staff can continually screen potential participants and, when needed, proceed quickly to obtain protocol-specific enrollment consent. Sites conducting general screening or prescreening approved by their IRB/EC and any applicable RE may use the results from this screening to determine eligibility for this

protocol, provided the tests are conducted within the time periods specified in the eligibility criteria.

9.1.2 Protocol-specific consent forms

The protocol-specific consent forms describe the study products to be used and all aspects of protocol participation, including screening and enrollment procedures. A sample protocol-specific consent form for the main study is located in Appendix A. A separate sample consent form for other uses of specimens is located in Appendix C.

Each HVTN CRS is responsible for developing a protocol-specific consent form(s) for local use, based on the sample protocol-specific consent forms in Appendix A and Appendix C. The consent form(s) must be developed in accordance with requirements of the following:

- CRS's IRB/EC and any applicable REs,
- CRS's institution, and
- Elements of informed consent as described in Title 45, CFR Part 46 and Title 21 CFR, Part 50, and in the International Conference on Harmonisation (ICH) E6, Good Clinical Practice: Consolidated Guidance 4.8.

Study sites are strongly encouraged to have their local CABs review their site-specific consent forms. This review should include, but should not be limited to, issues of cultural competence, local language considerations, and the level of understandability.

The sample informed consent form includes instructions throughout the document for developing specific content.

Sites should follow the instructions in the Protocol-specific Official Memo distributed along with this protocol regarding when they may begin using their site-specific protocol consent forms.

9.1.3 Assessment of Understanding

Study staff are responsible for ensuring that participants fully understand the study before enrolling them. This process involves reviewing the informed consent form with the participant, allowing time for the participant to reflect on the procedures and issues presented, and answering all questions completely.

An Assessment of Understanding is used to document the participant's understanding of key concepts in this HIV vaccine trial and that in a previous trial there was an association of increased HIV acquisition with receipt of that study vaccine. The participant must complete the Assessment of Understanding before enrollment. Staff may provide assistance in reading and understanding the questions and responses, if necessary. Participants must verbalize understanding of all questions answered incorrectly. This process and the participant's understanding of the key concepts should be recorded in source documentation at the site.

IRB/EC and any applicable RE may require that a participant has signed either a screening or protocol-specific consent document prior to administering the Assessment of

Understanding. The consent process (including the use of the Assessment of Understanding) should be explained thoroughly to the IRB/EC and any applicable RE, whose recommendations should be followed.

9.2 Pre-enrollment procedures

Screening may occur over the course of several contacts/visits, up to and including before vaccination on day 0. All inclusion and exclusion criteria must be assessed within 45 days before enrollment, unless otherwise specified in the eligibility criteria (or below in this section).

After the appropriate informed consent has been obtained and before enrollment, the following procedures are performed:

- Medical history, documented in the case history record;
- Assessment of Understanding (see Section 9.1.3);
- Assessment of risk for HIV infection
- Complete physical examination, including height, weight, vital signs, and clinical assessments of head, ears, eyes, nose, and throat; neck; lymph nodes; heart; chest; abdomen; extremities; neurological function; and skin;
- Assessment of concomitant medications the volunteer is taking, including prescription and nonprescription drugs, vitamins, topical products, alternative/complementary medicines (eg, herbal and health food supplements), recreational drugs, vaccinations, and allergy shots;
- Laboratory tests as defined in the inclusion and exclusion criteria, including:
    - Screening HIV testing at local lab,
    - Pregnancy test,
    - Hemoglobin;
- Obtaining of volunteer demographics in compliance with the NIH Policy on Reporting Race and Ethnicity Data: Subjects in Clinical Research, Aug. 8, 2001 (available at http://grants.nih.gov/grants/guide/notice-files/NOT-OD-01-053.html);
- Counseling on HIV testing and risk reduction, as described in Section 9.7; and
- Discussion of pregnancy prevention. A pregnant or breastfeeding person may not be enrolled in this trial. Specific criteria and assessment of contraception and pregnancy status are described in study inclusion criteria.

9.2.1 Use of screening results from another HVTN study

If a participant screens for an HVTN study at the same HVTN CRS but then does not join that study, screening results from that effort may be applied to the screening for this protocol, as long as the screening was done under participant consent, the participant has signed a consent form to begin screening for this study, and the tests were conducted within the time periods specified in the eligibility criteria (see Sections 7.1 and 7.2).

9.3 Enrollment and vaccination visits

Enrollment is simultaneous with first vaccination. The time interval between randomization and enrollment should not exceed 4 working days. The HVTN CRS registers the participant by scheduling the day 0 visit (enrollment) via the Web-based randomization system, and requests the randomization assignment. Circumstances may require a participant's enrollment visit to be changed. This may exceed the 4-day randomization time limit.

At all vaccination visits, the following procedures are performed before vaccination:

- Abbreviated physical examination, including weight, vital signs, and a symptom-directed evaluation by history and/or appropriate physical exam based on participant self-reported symptoms or complaints;
- Assessment of baseline reactogenicity parameters;
- Assessment of concomitant medications (as described in Section 9.2);
- Assessment of any new or unresolved AEs/ intercurrent illnesses;
- Confirm that participants received HIV test results from previous visit. If not provide test results and post-test counseling as appropriate;
- Pregnancy test. Persons who are NOT of reproductive potential due to having undergone total hysterectomy or bilateral oophorectomy (verified by medical records), are not required to undergo pregnancy testing; and
- Specimen collection as applicable.

Following completion of all procedures in the preceding list, and if results indicate that vaccination may proceed, vaccination is prepared and administered (see Sections 8.3 and 8.4).

Administration of all injections during a vaccination visit must be accomplished within 1 calendar day.

Immediately following vaccination, the participant remains in the clinic for observation. An initial reactogenicity assessment is made at a target of 30 minutes after injection, with an acceptable range of 25-60 minutes. Before leaving the clinic, the participant is given a postvaccination memory tool and is instructed on how to complete it. The site will make arrangements to obtain a report of reactogenicity events from the participant after the reactogenicity period (as described in Section 9.9).

The following procedures will be performed at all vaccination visits. These procedures may be performed prior to or following vaccination:

- Risk reduction counseling (as described in Section 9.7);
- Pregnancy prevention assessment (as described in Section 9.2 and 9.8);
- Assessment of new or unresolved social impacts (site staff will ask participant about the status of any unresolved social impacts and if she has experienced any new social impacts as a result of the trial participation).

Additional procedures will be performed at scheduled visits as specified in Appendix G:

- Gonorrhea and Chlamydia trachomatis (GC/CT) testing (before vaccination);
- Trichomonas vaginalis testing (before vaccination);
- Syphilis testing (before vaccination);
- Microbiome/mucosal collections: vaginal swab and cervical cup (before vaccination);
- Behavioral risk assessment questionnaire;
- Outside testing and belief questionnaire; and
- HIV infection assessment.

9.4 Follow-up visits

The following procedures are performed at all scheduled follow-up visits:

- Risk reduction counseling (as described in Section 9.7);
- Assessment of new or unresolved social impacts (site staff will ask participant about the status of any unresolved social impacts and if s/he has experienced any new social impacts as a result of the trial participation);
- Specimen collection as applicable;
- Assessment of new or continuing concomitant medications (as described in Section 9.2); and
- Assessment of new or unresolved AEs/intercurrent illnesses.

Additional procedures will be performed at scheduled follow-up visits as specified in Appendix E and Appendix G:

- Pregnancy prevention assessment (as described in Section 9.2 and 9.8);
- Microbiome/mucosal collections: vaginal swab and cervical cup (before vaccination);
- GC/CT testing;
- Trichomonas vaginalis testing;
- Syphilis testing;
- Abbreviated physical examination, including weight, vital signs, and a symptom-directed evaluation by history and/or appropriate physical exam based on participant self-reported symptoms or complaints;
- Complete physical examination, including height, weight, vital signs, and clinical assessments of head, ears, eyes, nose, and throat; neck; lymph nodes; heart; chest; abdomen; extremities; neurological function; and skin;
- HIV infection assessment including pre-test counseling. A subsequent follow-up contact is conducted to provide post-test counseling and to report results to participant;
- Confirm that participants received HIV test results from previous visit. If not, provide test results and post-test counseling as appropriate;
- Behavioral risk assessment questionnaire;

- Outside Testing and Belief Questionnaire; and
- Urine pregnancy test. Persons who are NOT of reproductive potential due to having undergone total hysterectomy or bilateral oophorectomy (verified by medical records), are not required to undergo pregnancy testing.

9.5 Procedures for HIV-infected participants

The following procedures are performed at follow-up visits for HIV-infected participants, as specified in Appendix F and Appendix H:

- Counseling on HIV-1 testing/diagnosis;
- Referral to medical professionals for care;
- Abbreviated physical examination including weight, vital signs, and a symptom-directed evaluation by history and/or appropriate physical exam based on participant self-reported symptoms or complaints;
- Complete physical examination, including clinical assessments of head, ears, eyes, nose, and throat; neck; lymph nodes; heart; chest; abdomen; extremities; neurological function; and skin;
- Assessment of HIV/AIDS-related conditions and non-HIV disease progression-related events;
- Assessment of new or continuing concomitant medications (as described in 9.2);
- Assessment of new or continuing antiretroviral therapies and reason for initiation or change in therapy;
- Assessment of new or unresolved AEs/intercurrent illnesses;
- Counseling to reduce the risk of HIV transmission;
- Administration of behavioral risk assessment questionnaire;
- Assessment of new or unresolved social impacts (site staff will ask participant about the status of any unresolved social impacts and about any new social impacts experienced as a result of the trial participation);
- Specimen collection (Appendix F).

9.6 Mucosal sampling

Vaginal swabs and cups will be collected from study participants at the visits indicated in Appendix E and Appendix G. These samples will be used for vaginal microbiome analysis and assessment of markers of inflammation including but not limited to cytokines, chemokines and other soluble mediators.

9.7 HIV counseling and testing

HIV counseling will be performed in compliance with the CDC's guidelines or other local guidelines for HIV counseling and referral. HIV testing will be performed in accordance with the current HVTN HIV testing algorithm following enrollment.

Participants will be counseled routinely during the trial on the avoidance of HIV infection and on the potential negative social impacts of testing antibody positive due to the vaccine. They will also be counseled on the risks of HIV antibody testing outside of the HVTN CRSs and will be discouraged from doing so during study participation and/or during any period of vaccine-induced positive serology.

Study staff will take particular care to inform study participants of the likelihood of routine HIV testing being offered or performed outside the study CRS at emergency rooms, clinics, and medical offices. CRS staff should inform participants of their right to opt out of HIV testing outside the study site. CRS staff should inform study participants if regulations permit medical providers to perform HIV testing without first informing patients. If this is the case, then CRS staff should advise study participants that they may decline testing preemptively. CRS staff should also inform participants if positive results must be reported to local public health authorities. CRS staff should also inform participants of the need to maintain study blinding by getting HIV testing only at the study CRS. CRS staff should provide participants with CRS contact information and should encourage participants to ask medical providers to contact the CRS. The CRS can verify that the participant is in an HIV vaccine clinical trial and should only be tested at the study CRS.

Potential participants identified as being HIV infected during screening are not enrolled. All participants who are identified as being HIV infected during the study will be followed for approximately 6 months as detailed in Appendix F and Appendix H.

9.7.1 Distinguishing intercurrent HIV infection from vaccine-induced positive serology

The study product may elicit an antibody response to HIV proteins. Therefore, vaccine-induced positive serology may occur in this study. Several precautionary measures will be taken to distinguish intercurrent HIV infection from vaccine-induced positive serology. These precautionary measures include:

- Participants will have physical examinations at visits specified in Appendix G. Signs or symptoms of an acute HIV infection syndrome, an intercurrent illness consistent with HIV-1 infection, or probable HIV exposure (ie, unprotected sex with a known HIV positive partner), would prompt a diagnostic workup per the HVTN algorithm for Recent Exposure/Acute Infection Testing to determine HIV infection.
- HIV testing will be performed at multiple timepoints throughout the study (see Appendix E and Appendix G). The Laboratory Program (or approved diagnostic laboratory) will follow the HVTN HIV testing algorithm (as described in the HVTN Laboratory Manual of Operations), which is able to distinguish vaccine-induced antibody responses from actual HIV infections.
- All participants can receive HIV-1 diagnostic testing from the site following their last scheduled visit until they are told that they did not receive an HIV vaccine
- All participants who received vaccine product and who have vaccine-induced positive or indeterminate HIV-1 serology (as measured by the standard anti-HIV

antibody screening tests) at or after the study is unblinded will be offered poststudy HIV-1 diagnostic testing (per the HVTN poststudy HIV-1 testing algorithm) periodically and free of charge as medically/socially indicated (approximately every 6 months) unless or until HIV antibody testing is no longer the standard test in clinical settings.

9.7.2 VISP registry

Experimental HIV vaccines may induce antibody production to HIV antigens, producing reactive results on commercially available HIV test kits. This is called "vaccine-induced seropositivity" (VISP). In order to provide poststudy HIV testing to distinguish between VISP and HIV infection, and to mitigate potential social harms resulting from VISP in HIV vaccine recipients who are not infected with HIV, the HVTN has created a VISP registry. Following study unblinding, the registry will allow trained staff to verify that an individual has received an HIV vaccine, and therefore has the potential for VISP. Information in the VISP registry will not be used for research. Rather, the registry exists to support provision of poststudy testing and counseling services to HIV vaccine recipients. The registry contains the names of all study participants, unless they request that their names be removed.

9.8 Pregnancy prevention assessment

Contraception status is assessed and documented at scheduled clinic visits as specified in Appendix G for a participant who is sexually active in a way that could cause that participant to become pregnant. Prior to enrollment and throughout the study, staff will ask participants to verbally confirm their use of adequate contraceptive methods. A participant who is sexually active in a way that could cause that participant to become pregnant should be reminded at scheduled clinic visits, specified in Appendix G, of the importance of using contraception and should be referred to specific counseling, information, and advice as needed. (Specific contraception requirements are listed in Section 7.1). This reminder should be documented in the participant's study record.

Self-reported infertility—including having reached menopause (no menses for 1 year) or having undergone hysterectomy, bilateral oophorectomy, or tubal ligation—must be documented in the participant's study record.

9.9 Assessments of reactogenicity

For all participants, baseline assessments are performed before and reactogenicity assessments are performed after each vaccination. All reactogenicity symptoms are followed until resolution and graded according to the Division of AIDS Table for Grading the Severity of Adult and Pediatric Adverse Events, Corrected Version 2.1, July 2017, except as noted in Section 12.2.2.

The reactogenicity assessment period is 3 full days following each vaccination. At some sites the reactogenicity assessment period may be 7 full days following each vaccination. The maximum severity reached for each symptom during the assessment period is reported on CRFs. Participants will be given a postvaccination memory tool to assist with recall of symptoms. The site staff and the participant will make multiple efforts in good faith to be in contact after the last day of the reactogenicity period, or sooner if indicated,

to review reactogenicity data. Clinic staff will follow new or unresolved reactogenicity symptoms present at the last day of the reactogenicity assessment period to resolution. Participants are instructed to contact the clinic for events that arise during the period between vaccination and the next scheduled visit. In general, a participant who self-reports any postvaccination reaction greater than mild is seen by a clinician within 48 hours after onset, unless the reaction is improving and/or has resolved completely.

Reactogenicity assessments include assessments of systemic and local symptoms and vaccine-related lesions. Events not listed on a CRF, or with an onset after the reactogenicity assessment period, or those meeting SAE/AEs criteria requiring expedited reporting, are recorded on an AE log form.

9.9.1 Assessment of systemic and local symptoms

Systemic symptoms include increased body temperature, malaise and/or fatigue, myalgia, headache, chills, arthralgia, nausea, and vomiting. Local symptoms include pain and/or tenderness proximal to the injection site.

Body temperature is measured by oral or infrared thermometry and reported in degrees Celsius. A measurement is taken once daily during the assessment period and should be repeated if participant is feeling feverish.

9.9.2 Assessment of injection site

Typical injection site reactions are erythema/redness and induration/swelling. The maximum horizontal and maximum vertical measurements for all injection site reactions are recorded.

All injection site reactions are monitored until resolution. Areas greater than 25 cm^2 are followed daily; otherwise, the frequency of follow-up is based on clinician judgment.

9.10 Visit windows and missed visits

Visit windows are defined in HVTN 705/VAC89220HPX2008 Study Specific Procedures. For a visit not performed, a Missed Visit form is completed. If the missed visit is one that required safety assessments or local safety labs, HVTN CRS staff should attempt to bring the participant in for an interim visit as soon as possible.

Procedures performed at an interim visit are usually toxicity/safety assessments (including local safety labs) and HIV testing. With the exception of HIV testing, these procedures are performed only if they were required at the missed visit or if clinically indicated. HIV testing may be performed as deemed appropriate by the study staff. Blood samples for immunogenicity assays are not typically collected at interim visits.

If a missed visit required vaccination, please refer to Section 7.3.2 and Section 7.3.3 for resolution.

9.11 Early termination visit

In the event of early participant termination, site staff should consider if the following assessments are appropriate: a final physical examination, AE and concomitant medication reporting, pregnancy testing, social impact assessment, risk reduction counseling, and HIV test.

9.12 Pregnancy

All initial reports of pregnancy must be reported to Janssen Vaccines by the study-site personnel within 3 business days of their knowledge of the event using the appropriate form(s). If a participant becomes pregnant during the course of the study, no more injections of study product will be given during the pregnancy, but remaining visits and study procedures should be completed unless medically contraindicated or applicable regulations require termination from the study. For participants who are no longer pregnant, see Section 7.3.1. If the participant terminates from the study prior to the pregnancy outcome, the site should make every effort to keep in touch with the participant in order to ascertain the pregnancy outcome. All pregnancies and pregnancy outcomes should be recorded and reported (see the HVTN 705/VAC89220HPX2008 Study Specific Procedures).

10 HIV-1 infection assessment and clinical management

10.1 HIV-1 symptom assessment

At scheduled visits indicated in Appendix G and at unscheduled visits due to illness or suspected exposure, if necessary, information will be collected about any signs or symptoms suggestive of acute HIV-1 infection. At visits following recent high-risk exposure, participants will be queried about any signs/symptoms suggestive of acute HIV-1 infection. Presence of signs/symptoms suggestive of acute HIV-1 infection, an intercurrent illness consistent with acute retroviral syndrome, or probable recent HIV exposure (eg, unprotected sex with a known HIV-positive partner) would prompt a diagnostic work-up per the protocol-specific algorithm to determine HIV infection, except that in this instance the algorithm is modified to conduct serology and nucleic acid testing simultaneously.

10.2 HIV-1 screening test

As part of screening, participants will be screened for HIV-1/2 infection by blood tests approved by the US FDA or locally available assays that have been approved by HVTN Laboratory Operations. Potential participants identified as being HIV infected during screening will be referred for medical treatment and management of the HIV infection. These individuals may also be referred to appropriate ongoing clinical trials or observational studies.

10.3 HIV-1 testing postvaccination

Surveillance for HIV-1 takes place at vaccination visits and follow-up visits specified in Appendix E and Appendix G.

In-study HIV testing will be performed according to the HVTN HIV diagnostic testing algorithms. Routinely, specimens are initially assayed with an HVTN Lab Program–approved HIV 1/2 enzyme immunoassay (EIA) or chemiluminescent microparticle immunoassay (CMIA). If the EIA/CMIA is reactive, nucleic acid polymerase chain reaction (PCR) test to detect HIV-1 RNA will be performed as indicated in the algorithm. The algorithm is repeated on a second specimen to confirm a diagnosis of HIV-1 infection. The second specimen for confirmatory testing may be collected at an interim visit (designated as visit #.X, where # is the visit at which the first reactive test was obtained and X designates the interim visit; specified in Appendix F and Appendix H). Samples to be stored for future immunogenicity or virology studies will also be collected at this time (Appendix F).

A ‘case’ will be defined as a participant with confirmed detectable HIV-1 nucleic acid PCR on 2 different specimen collection dates. The nucleic acid test will most commonly be the HIV-1 RNA PCR viral load test. Confirmation of HIV-1 infection will be determined as dictated by the HVTN HIV testing algorithm (available on the HVTN 705 protocol-specific website). Before issuing an HIV-1 infection report for a participant diagnosed with HIV-1 infection prior to study unblinding, all testing results will be reviewed by a blinded, independent Endpoint Adjudicator(s) and/or designee(s) (Section 10.4).

If a participant is confirmed to have become HIV-1 infected prior to study unblinding, plasma HIV-1 viral RNA will be measured on archived samples collected according to Appendix E. In addition to plasma HIV-1 viral RNA testing, participants may also have measurements of immunogenicity assessments and a clinical assessment performed at all postinfection visits (see Appendix F).

The HVTN Laboratory Program is responsible for all in-study diagnostic HIV testing.

10.4 Endpoint adjudication

The diagnostic criteria for HIV-1 infection outside the setting of a vaccine trial are well accepted. However, definitive diagnosis of HIV-1 infection in the context of having received an HIV vaccine that is even partially effective may be more difficult. Specifically, if the immune responses elicited by vaccination are capable of completely suppressing viral replication, or if vaccination alters the normal serological response upon exposure to HIV-1, standard diagnostic tests may be more difficult to assess. Therefore, the HVTN will have an endpoint adjudication process to assess all serological and virological testing, in a blinded manner, on each participant in the trial who, prior to study unblinding, tests positive per the HVTN 705 HIV-1 diagnostic testing algorithm. The assessment of the Endpoint Adjudicator(s) or designee(s) will be reported to the SDMC and to the HIV diagnostics laboratory.

The Endpoint Adjudicator(s) and/or designee(s) must notify the SDMC within 1 working day of any confirmed HIV-1 infection. The HIV diagnostics lab will inform the clinic of the outcome of the HIV testing algorithm (ie, HIV infected, HIV uninfected, or redraw required).

The Endpoint Adjudicator(s) and/or designee(s) will be an expert in the fields of infectious diseases or laboratory medicine independent of the clinical investigators participating in this trial.

10.5 HIV-1 infection during the study

It is critical to the success of the study that HIV-1–infected participants are properly identified and all data postdiagnosis are carefully recorded. Information obtained from these cases of HIV-1 infection will form the basis of the primary endpoint assessment.

Participants who develop HIV-1 infection following receipt of study product may remain in the study for follow-up but will receive no further injections. All participants who become HIV-1 infected following enrollment will be monitored as indicated in Appendix F and Appendix H. Archived samples from earlier visits may also be tested to determine the earliest date of HIV-1 infection.

Once the last HIV-1 uninfected participant completes the Month 36 visit, the study is considered completed. Further monitoring as indicated in Appendix F and Appendix H will not be required for participants who received confirmation of HIV-1 infection status after the study is considered complete. Redraw Visits (Visit #.X) to confirm HIV infection status may still be performed after the study is complete.

10.6 Medical care for participants who become HIV-1 infected

It is anticipated that some study participants, whether they are randomized to receive vaccine or placebo, will become HIV-1 infected during the course of the trial. It is critical that these HIV-1–infected participants receive appropriate medical care.

The investigators associated with this trial will refer participants who develop HIV infection while participating in this trial to medical professionals for care.

11 Laboratory

11.1 HVTN CRS laboratory procedures

The HVTN 705/HPX2008 Site Lab Instructions and SSP provide further guidelines for operational issues concerning the clinical and processing laboratories. These documents include guidelines for general specimen collection, special considerations for phlebotomy, specimen labeling, whole blood processing, HIV screening/diagnostic testing, and general screening and safety testing.

Tube types for blood collection are specified in Appendix E and Appendix F. For tests performed locally, the local lab may assign appropriate tube types.

In specific situations, the blood collection tubes may be redirected to another laboratory or may require study-specific processing techniques. In these cases, laboratory special instructions will be posted on the protocol-specific section of the HVTN website.

11.2 Total blood volume

Required blood volumes per visit are shown in Appendix E and Appendix F. Not shown is any additional blood volume needed if additional testing were required, or if a serum pregnancy test needs to be performed. The additional blood volume would likely be minimal. The total blood volume drawn for each participant will not exceed 500 mL in any 56-day (8-week) period.

11.3 Immunogenicity assays

Endpoint assays for humoral and cellular responses will be run primarily in the context of case control studies. Pilot studies with a selected subset of samples may be conducted to inform the choice of assays for the case control studies. The primary immunogenicity timepoints in this study are 4 weeks after the third vaccination visit and 4 weeks after the fourth vaccination visit. Assays for humoral and cellular responses may be performed on participants at other timepoints; the schedules are shown in Appendix E and Appendix F.

11.4 Endpoint assays: humoral

11.4.1 HIV-1 multiplex antibody assay

Total binding IgG and IgA antibodies to HIV-1 Env proteins contained in the vaccine regimen will be assessed on serum samples from study participants taken at the primary immunogenicity timepoints and baseline. In addition, binding to cross-clade Env proteins and IgG isotypes will be assessed. Specimens from other timepoints as well as other HIV antigens may also be assayed based on the results of the initial assay.

11.4.2 Env-specific binding antibody assay

Total IgG Env-specific binding antibodies against HIV Env clade A (92UG037), B (1990A), C (ConC) and vaccine proteins gp140 Clade C and Mosaic (C97ZA and Mos1) will be assessed by ELISA in serum samples from study participants.

11.4.3 Neutralizing antibody (nAb) assay

HIV-1–specific nAb assays will be performed on serum samples from study participants taken at the primary immunogenicity timepoints. Specimens from the baseline and other timepoints may also be analyzed at the discretion of the HVTN Laboratory Program, which may be contingent on the results of the primary immunogenicity timepoints.

11.4.4 Antibody-dependent cellular phagocytosis (ADCP) assay

Antibody-dependent cellular phagocytosis assesses the ability of antibodies to induce the phagocytosis of antigen-coated beads by monocytes. Antigen-coated beads are incubated with increasing concentrations of purified antibodies and the monocytic cell line THP-1. The extent of phagocytosis is measured via flow cytometry, and the data is reported as a phagocytic score, which takes into account the proportion of effector cells that phagocytosed a bead and the degree of phagocytosis.

11.4.5 Antivector antibody studies

Baseline Ad26 neutralizing antibodies (nAb) will be assessed from cryopreserved specimens in a subset of participants.

11.5 Endpoint assays: cellular

11.5.1 Flow cytometry

Flow cytometry will be used to examine vaccine-specific CD4+ and CD8+ T-cell responses following stimulation of PBMCs with synthetic HIV peptides that span the proteins encoded by the vaccine. ICS parameters will include cytokines such as IFN-γ, interleukin (IL)-2, and tumor necrosis factor (TNF)-α, and may include other cytokines (such as cytokines relevant to Th2 and Th17 responses) to identify T cells of specific functionality. Markers of cytotoxic potential (eg, Granzyme B) and of Tfh functionality (eg, CXCR5 and PD-1) may also be included. Data will be reported as percentages of CD4+ or CD8+ T cells responding to a specific peptide pool. Additional cell surface markers, cytokines, or functional markers may also be analyzed.

11.5.2 IFN-γ ELISpot

Ex vivo HIV-specific T-cells will be assessed by IFN-γ ELISpot. To this end, PBMCs will be stimulated with synthetic peptide pools that span the proteins encoded by the vaccine constructs. Data will be reported as the number of spot-forming cells (SFC) per 10^6 cells recognizing specific peptide pools.

11.6 Innate immunity assays

Innate immune and anamnestic responses may be assessed at baseline, on the day of third vaccination and one day later.

11.6.1 Soluble factors in serum

Multiplex cytokine assays and/or enzyme-linked immunosorbent assays (ELISA) may be used to measure soluble cytokines, chemokines, and other immunomodulatory factors in the serum. Analytes may include IFN-γ, IL-2, IL-4, IL-10, IL-12, IL-13 TNF- α, IL-1 β, MIP-1α, MIP-1β and IP-10. Other analytes may also be included.

11.6.2 RNA gene expression

Whole blood will be cryopreserved in an RNA protection reagent. RNA from whole blood or from sorted PBMC may be isolated and used for RNAseq and/or real-time PCR. Signatures of gene expression changes for different cell types will be compared pre- and post-vaccination. Data will be reported as fold change over baseline pre-vaccine expression.

11.7 Mucosal studies

11.7.1 Vaginal microbiome analysis

Vaginal swab specimens are processed to enable nucleic acid sequencing. 16S rRNA sequences will then be determined by high throughput sequencing methods.

11.7.2 Inflammatory marker analysis

Assays may be performed using cervical secretion specimens to examine cytokines, chemokines, and other immunomodulatory factors using a multiplex bead array (Luminex) or Meso-Scale Discovery (MSD) platform.

11.8 Viral sequencing

Viral sequencing may be conducted on the earliest available plasma specimens with positive HIV-1 RNA PCR tests from study participants who are diagnosed with HIV-1 infection. Viral sequencing may be conducted on subsequently collected samples to assess viral evolution.

11.9 ARV detection

ARV testing will be performed using a test that measures the amount of tenofovir-diphosphate (a metabolite of tenofovir) in red blood cells. This test utilizes a dried blood spot as the sample source and analyzes for tenofovir diphosphate by liquid chromatography and tandem mass-spectroscopy, as previously described [54,55]. Stored serum and plasma (see Appendix F) will allow additional assessment of PrEP use if warranted.

11.10 Genotyping

Molecular human leukocyte antigen (HLA) typing may be performed on enrolled participants using cryopreserved PBMC collected at baseline, initially on specimens from participants who demonstrate vaccine-induced T-cell responses at postvaccination timepoints. Other participants (including control recipients) may be HLA-typed to support future studies of immunological interest at the discretion of the HVTN Laboratory Program. Other markers, such as genes associated with immune responses or HIV-1 disease progression may also be assessed.

11.11 Lab assay algorithm

The Lab Assay Algorithm lists assays to characterize cellular, humoral, and innate immune responses as well as host genetics that may be conducted to determine endpoints in HVTN vaccine trials. The type of assay(s) employed will be dependent on the response obtained by the primary immunogenicity assays at relevant timepoints. Please note that the Lab Assay Algorithm will be updated periodically to include new assays.

11.12 Exploratory studies

Samples may be used for other testing and research related to furthering the understanding of HIV immunology or vaccines. In addition, cryopreserved samples may be used to perform additional assays to support standardization and validation of existing or newly developed methods.

11.13 Other use of stored specimens

The HVTN stores specimens from all study participants indefinitely, unless a participant requests that specimens be destroyed or if required by IRB/EC or RE.

Other use of specimens is defined as studies not described in the protocol.

This research may relate to HIV, vaccines, the immune system, and other diseases. This could include limited genetic testing and, potentially, genome-wide association studies. This research is done only to the extent authorized in each study site's informed consent form, or as otherwise authorized under applicable law. Other testing on specimens will occur only after review and approval by the HVTN and Janssen Vaccines, the IRB/EC of the researcher requesting the specimens, and the CRS's IRBs/ECs if required.

The protocol sample informed consent form is written so that the participant either explicitly allows or does not allow their samples to be used in other research when they sign the form. Participants who initially agree to other use of their samples may rescind their approval once they enter the study; such participants will remain in this study and their samples will only be used for the studies described in this protocol. If a participant decides against allowing other research using his or her samples, or at any time rescinds prior approval for such other use, the study site investigator or designee must notify HVTN Regulatory Affairs in writing. In either case, HVTN Regulatory Affairs directs the HVTN Lab Program not to use samples from these participants for such other uses.

CRSs must notify HVTN Regulatory Affairs if institutional or local governmental requirements pose a conflict with or impose restrictions on other use of specimens.

11.14 Biohazard containment

As the transmission of HIV and other blood-borne pathogens can occur through contact with contaminated needles, blood, and blood products, appropriate precautions will be employed by all personnel in the drawing of blood and shipping and handling of all specimens for this study, as currently recommended by the CDC and the NIH or other applicable agencies.

All dangerous goods materials, including Biological Substances, Category A or Category B, must be transported according to instructions detailed in the International Air Transport Association Dangerous Goods Regulations.

12 Safety monitoring and safety review

12.1 Safety monitoring and oversight

12.1.1 HVTN 705/HPX2008 PSRT

The HVTN 705/HPX2008 PSRT is composed of the following members:

- Sponsor study responsible physician,
- Sponsor Global Medical Safety physician,
- DAIDS medical officer representative,
- Protocol chair and cochairs,
- Protocol Team leader/Core medical monitor,
- Clinical safety specialist (CSS), and
- Regional medical liaison (RML).

The clinician members of HVTN 705/HPX2008 PSRT are responsible for decisions related to participant safety, as outlined in the PSRT charter.

The Protocol Team clinic coordinator, clinical data manager, vaccine developer representative, clinical trial manager, and others may also be included in HVTN 705/HPX2008 PSRT meetings.

12.1.2 NIAID DSMB

The NIAID DSMB assesses the effects of the study vaccine during the trial, provides other monitoring as described in Section 12.4.3, and may give advice to the HVTN 705/VAC89220HPX2008 Protocol Team leadership, the OG, and PSRT. More details on the role of and the data provided to the DSMB will be described in a DSMB charter and SAP.

12.1.3 SDMC roles and responsibilities in safety monitoring

The roles and responsibilities of the SDMC in relation to safety monitoring include:

- Maintaining a central database management system in coordination with Janssen;
- Providing reports of clinical data to appropriate groups such as the HVTN 705/HPX2008 PSRT, Janssen Global Medical Safety, and NIAID DSMB (see Section 12.4.3).

12.1.4 HVTN Core roles and responsibilities in safety monitoring

- Daily monitoring of clinical data for events that meet the safety pause and HVTN 705/HPX2008 PSRT AE review criteria (see Section 12.3);
- Notifying HVTN CRSs and other groups when safety pauses are instituted and lifted (see Section 12.3);

- Querying HVTN CRSs for additional information regarding reported clinical data; and
- Providing support to the HVTN 705/HPX2008 PSRT.

12.1.5 Sponsor roles and responsibilities in safety monitoring

- Report SAEs/SUSARs to FDA or other Regulatory Authorities according to local requirements
- Notifying the FDA, the SAHPRA, and other health authorities as applicable when safety pauses are instituted and lifted (see Section 12.3);
- Providing support to the HVTN 705/HPX2008 PSRT.

12.2 Safety reporting

12.2.1 Submission of safety forms to SDMC

Sites must enter all safety data (eg, reactogenicity, AE, concomitant medications) before the end of the next business day after receiving the information, excluding federal or bank holidays. The forms should not be held in anticipation of additional information at a later date. If additional information is received at a later date, the forms should be updated and resubmitted before the end of the next business day after receiving the new information. For the case of a longer site holiday closure, site staff must submit the data by the end of the 5th day (local time) after receiving the information even if this day is a holiday.

For example: If the site becomes aware of an AE on Thursday (Day 0), the site must submit the data by the end of the next business day, on Friday. If there is a longer site holiday closure, then this AE must be reported no later than the end of the fifth day, Monday (Day 4). If Monday is a holiday as well, all safety forms still need to be submitted by the end of Monday (Day 4).

12.2.2 AE reporting

Sites are expected to notify the CSS/RML of any serious safety concern requiring their attention (see Table 12-1). Telephone numbers and email addresses are found on the protocol home page on the HVTN Members' site (https://members.hvtn.org/protocols/hvtn705). Concerns requiring immediate attention should be communicated by calling the clinical safety phone.

In the case of email notification, the CSS/RML will reply during working hours (US Pacific Time or South Africa Time) to confirm that the email has been received and reviewed. If email service is not available, the HVTN CRS should notify the CSS/RML of the event by telephone, then submit CRFs.

In addition, site investigators are required to submit AE information in accordance with IRB/EC and any applicable RE requirements.

12.2.2.1 Reactogenicity event

Reactogenicity events are predefined local (at the injection site) and systemic events occurring in a predefined postvaccination period for which the subject is specifically questioned (see Section 9.9).

12.2.2.2 Adverse event

An AE is any untoward medical occurrence in a clinical investigation participant administered a study product and which does not necessarily have a causal relationship with this treatment. An AE can therefore be any unfavorable and unintended sign (including an abnormal laboratory finding), symptom, or disease temporally associated with the use of an investigational study product/procedure(s), whether or not related to the investigational study product/procedure(s).

All AEs are graded according to the Division of AIDS Table for Grading the Severity of Adult and Pediatric Adverse Events, Corrected Version 2.1, July 2017, available on the RSC website at http://rsc.tech-res.com/clinical-research-sites/safety-reporting/daids-grading-tables, except:

- Unintentional weight loss is required to be reported as an AE only if it is considered to be potentially deleterious to the participant's health (see HVTN 705 Study Specific Procedures);
- Injection Site Erythema or Redness and Injection Site Induration or Swelling will not consider interference with usual social and functional activities such that:
  - Grade 1 is: 2.5 to < 5 cm in diameter OR 6.25 to < 25 cm^2 surface area;
  - Grade 2 is: ≥ 5 to < 10 cm in diameter OR ≥ 25 to < 100 cm^2 surface area;
  - Grade 3 is: ≥ 10 cm in diameter OR ≥ 100 cm^2 surface area OR Ulceration OR Secondary infection OR Phlebitis OR Sterile abscess OR Drainage;
  - Grade 4 is: Potentially life-threatening consequences (eg, abscess, exfoliative dermatitis, necrosis involving dermis or deeper tissue).

Adverse Events (AE) will be collected over a period of 30 days after each vaccination visit. AEs with the onset date outside the timeframe defined above (>30 days after previous study vaccination), which are ongoing on the day of the subsequent vaccination, should be recorded on the AE log CRF. All collected AEs are reported to the SDMC on the appropriate CRF. Clinic staff should evaluate every AE to determine if (1) the AE meets SAE reporting criteria (Section 12.2.3), (2) if the AE meets the criteria for a safety pause/prompt AE review (Section 12.3), and (3) if the AE is a potential immune-mediated disease that may be listed as an AE of special interest (AESI). A sample list of AESI is provided in Appendix I.

Certain AEs will be collected and reported throughout the entire study:

- SAEs,
- AESIs,
- STIs,

- AEs leading to early participant withdrawal or early discontinuation of study product(s) administration.

12.2.2.3 Serious adverse event

A serious adverse event based on ICH and EU Guidelines on Pharmacovigilance for Medicinal Products for Human Use (as well as other pertinent national guidelines) is any untoward medical occurrence that at any dose:

- Results in death
- Is life-threatening (The subject was at risk of death at the time of the event. It does not refer to an event that hypothetically might have caused death if it were more severe.)
- Requires inpatient hospitalization or prolongation of existing hospitalization
- Results in persistent or significant disability/incapacity
- Is a congenital anomaly/birth defect
- Suspected transmission of any infectious agent via a medicinal product
- Is Medically Important*

*Medical and scientific judgment should be exercised in deciding whether expedited reporting is also appropriate in other situations, such as important medical events that may not be immediately life threatening or result in death or hospitalization but may jeopardize the subject or may require intervention to prevent one of the other outcomes listed in the definition above. These should usually be considered serious.

For the duration of the study, all events that meet the definition of a serious adverse event will be reported from the site to Janssen Vaccines as serious adverse events, regardless of whether they are protocol-specific assessments. Janssen Vaccines will evaluate any safety information that is spontaneously reported by an investigator beyond the time frame specified in the protocol.

All related SAEs that are ongoing at study completion should be followed by the investigator until resolution or until clinically stable.

12.2.3 SAE reporting to Janssen Vaccines

All SAEs, as defined in Section 12.2.2 must be reported to Janssen Vaccines by the study-site personnel within 3 business days of their knowledge of the event. Sites will submit SAE reports by documenting the information on the SAE Form and e-mailing or faxing it to the Janssen Local Safety Officer (LSO). The email address and fax number can be found on the protocol home page on the HVTN Members' site (https://members.hvtn.org/protocols/hvtn705).

The SAE reporting period for this study comprises the entire study period for each individual participant (from study enrollment until study completion or discontinuation from the study).

The study products for which expedited reporting are required are:

- Ad26.Mos4.HIV

- Placebo for Ad26.Mos4.HIV
- Clade C gp140 with alum
- Placebo for Clade C gp140 with alum

If the PSRT believes unblinding of the site PI to treatment assignment will assist with the clinical management of the SAE, the PSRT may consult the independent NIAID DSMB for a recommendation. In the event the PSRT and/or the NIAID DSMB determines that unblinding is indicated, the unblinded statistician, DSMB Chair, or designee will inform the site physician of the participant's treatment assignment in such a manner as to maintain the study blind of the PSRT and study team. For emergency medical management of a participant, unblinding decisions will be made by the site investigator, when the timing is critical for medical management. Otherwise, the HVTN 705/HPX2008 PSRT should be consulted before emergency unblinding occurs.

12.2.4 Expedited reporting of AEs to pertinent national regulatory authorities

The study sponsor or designee(s) prepares and files expedited reports to appropriate regulatory authorities within the timelines required.

Site IoRs/designees will submit AE information and any other relevant safety information to their ECs/IRBs and any other relevant authority in accordance with applicable local requirements.

12.3 Safety pause and prompt PSRT AE review

When a trial is placed on safety pause, all enrollments and vaccinations with the product related to the event that triggered the pause will be held until further notice. The AEs that will lead to a safety pause or prompt HVTN 705/HPX2008 PSRT AE review are summarized in Table 12-1. Vaccinations may be suspended for safety concerns other than those described in the table, or before pause rules are met, if, in the judgment of the HVTN 705/HPX2008 PSRT, participant safety may be threatened. Criteria for an individual participant's departure from the schedule of vaccinations are listed in Section 7.3.

Table 12-1 AE notification and safety pause/AE review rules

Event and relationship to study products	Severity	HVTN CRS action[a]	Sponsor action
SAE, related	Grade 5 or Grade 4 or Grade 3	Phone immediately, email/fax and submit applicable forms immediately	Immediate pause and HVTN 705/HPX2008 PSRT notification
SAE, related	Grade 2 or Grade 1	Email/fax and submit applicable forms immediately	Prompt HVTN 705/HPX2008 PSRT AE review to consider pause
SAE, not related	Grade 5	Phone immediately, email/fax and submit applicable forms immediately	Immediate HVTN 705/HPX2008 PSRT notification
AE[b], related	Grade 4 or 3	Email/fax and submit applicable forms immediately	Prompt HVTN 705/HPX2008 PSRT AE review to consider pause

[a] Phone numbers, fax numbers and email addresses are found on the Protocol home page on the HVTN Members' site (https://members hvtn org/protocols/hvtn705)

[b] Does not include subjective reactogenicity symptoms (injection site pain, tenderness, fatigue/malaise, myalgia, arthralgia, chills, headache, nausea) Objective reactogenicity symptoms, grade 3, need to be ongoing for 3 days

For all safety pauses, HVTN Core notifies the HVTN 705/HPX2008 PSRT, Janssen Global Regulatory Affairs, HVTN Regulatory Affairs, and participating HVTN CRSs. When an immediate safety pause is triggered, HVTN Core notifies the DSMB.

Once a trial is paused, the HVTN 705/HPX2008 PSRT reviews safety data and decides whether the pause can be lifted or permanent discontinuation of vaccination is appropriate, consulting the DSMB if necessary. HVTN Core notifies the participating HVTN CRSs, Janssen Global Regulatory Affairs, and HVTN Regulatory Affairs of the decision regarding resumption or discontinuation of study vaccinations. Based on the HVTN 705/HPX2008 PSRT assessment, Janssen Vaccines notifies the FDA, the SAHPRA, and other pertinent national and regional authorities as appropriate.

If an immediate HVTN 705/HPX2008 PSRT notification or prompt HVTN 705/HPX2008 PSRT AE review is triggered, HVTN Core notifies the HVTN 705/HPX2008 PSRT as soon as possible during working hours (US Pacific Time)—or, if the information was received during off hours, by the morning of the next work day. If a prompt HVTN 705/HPX2008 PSRT AE review cannot be completed within 72 hours of notification (excluding weekends), an automatic safety pause occurs.

The HVTN requires that each CRS submit to its IRB/EC and any applicable RE protocol-related safety information (such as IND safety reports, notification of vaccine holds due to the pause rules, and notification of other unplanned safety pauses). CRSs must also follow all applicable RE reporting requirements.

In addition, all other AEs are reviewed routinely by the HVTN 705/HPX2008 PSRT (see Section 12.4.2).

12.4 Review of cumulative safety data

Routine safety review occurs at the start of enrollment and then throughout the study.

Reviews proceed from a standardized set of protocol-specific safety data reports. These reports are produced by the SDMC and include queries to the HVTN CRSs. Events are tracked by internal reports until resolution.

12.4.1 Daily review

Blinded daily safety reviews are routinely conducted by HVTN Core for events that meet immediate HVTN 705 PSRT notification criteria, as described in Section 12.3.

12.4.2 Twice-monthly review

During the injection phase of the trial, the HVTN 705/HPX2008 PSRT reviews clinical safety reports on a twice-monthly basis and conducts calls to review the data as appropriate. After the 4-week-post-final-vaccination visits are completed, less frequent reporting and safety reviews may be conducted at the discretion of the HVTN 705/HPX2008 PSRT. HVTN Core reviews reports of all reported AEs. Events identified during the review that are considered questionable, inconsistent, or unexplained are referred to the HVTN CRS clinic coordinator for verification.

12.4.3 DSMB review of cumulative safety data

The DSMB will periodically review accumulating unblinded safety data by group. Prior to each meeting, the SDMC will provide the DSMB with data as described in Section 6.5. Reports will be cumulative, generated from an up-to-date data file.

Based upon the reports, the DSMB will determine whether to recommend that the study should be continued, modified, or stopped, including for safety reasons.

12.5 Study termination

This study may be terminated early upon recommendation by the DSMB and/or the OG.

13 Protocol conduct

This protocol and all actions and activities connected with it will be conducted in compliance with the principles of GCP (ICHe6), and according to DAIDS, Janssen Vaccines, and HVTN policies and procedures as specified in the *HVTN Manual of Operations,* Janssen Vaccines Standard Operation Procedures, and DAIDS Clinical Research Policies and Standard Procedures Documents including procedures for the following:

- Protocol activation and implementation;
- Site initiation;
- Informed consent, screening, and enrollment;
- Study participant reimbursement;
- Clinical and safety assessments;
- Safety monitoring and reporting;
- Data collection, documentation, transfer, and storage;
- Participant confidentiality;
- Study follow-up and close-out;
- Unblinding of staff and participants;
- Quality control;
- Protocol monitoring and compliance;
- Advocacy and assistance to participants regarding negative social impacts associated with the vaccine trial;
- Risk reduction counseling;
- Specimen collection, processing, and analysis;
- Exploratory and ancillary studies and sub-studies, and
- Destruction of specimens.

Any policies or procedures that vary from DAIDS, Janssen Vaccines, and HVTN standards or require additional instructions (eg, instructions for randomization specific to this study) will be described in the HVTN 705/VAC89220HPX2008 *Study Specific Procedures.*

13.1 Protocol governance

13.1.1 Protocol team

The Protocol Team will be responsible for administrative oversight of the study, provides the overall operational direction for the trial, and is responsible for the conduct of the trial according to the highest scientific and ethical standards, as well as approving revisions and amendments to the protocol. The Protocol Team will remain blinded to the treatment group assignment of individual participants during the course of the study.

13.1.2 PSRT

The PSRT will review all clinical and laboratory safety data during the course of the study as described in Section 12.4.

13.1.3 Oversight group (OG)

The OG provides the overall scientific direction for the trial, and will receive and decide on any recommendations made by the DSMB, including stopping the study.

13.1.4 NIAID DSMB

The NIAID DSMB assesses the effects of the study vaccine during the trial and may give advice to Janssen Vaccines and to the Oversight Group. The DSMB membership for this trial will include representatives from Africa. With the exception of an unblinded statistician, the members of the committee are independent of Janssen Vaccines, DAIDS, the HVTN, and clinical investigators participating in this trial. They will not have any other involvement in the study, and will not have any relation to study participants. More details will be provided in the DSMB charter. The DSMB will monitor the trial for evidence of beneficial or adverse effects of the study vaccine using the guidelines proposed by the protocol. The DSMB may recommend any steps to ensure the safety of study participants and the integrity of the trial. Furthermore, it may recommend that the trial be terminated or that specific groups be withdrawn from the study, if any subgroup manifests serious or widespread side effects. To guarantee the unrestricted performance of its task, the DSMB may receive the individual study morbidity and mortality data from the unblinded statistician.

The DSMB will be informed immediately by an independent statistician if the pre-specified stopping boundary is met, indicating that the vaccine causes harm by increasing the rate of HIV acquisition (Section 6.5.1). In addition, the DSMB will monitor the study for futility to detect vaccine activity (Section 6.5.2), for high vaccine efficacy (Section 6.5.3), and for operational futility, defined as an unacceptably low rate of HIV-1 infections and by other measures of under-performance (Section 6.5.4). Semiannual DSMB meetings will be held for monitoring operational futility.

Should ongoing clinical trials of HIV prevention modalities produce results having implications for recruitment or endpoint enrollment in HVTN 705/VAC89220HPX2008, those results will be reported to the DSMB, whose recommendations will inform consultations with applicable experts and stakeholders regarding possible changes in trial design and conduct.

13.2 Social impacts

Participants in this study risk experiencing discrimination or other personal problems, resulting from the study participation itself or from the development of VISP. The HVTN CRS is obliged to provide advocacy for and assistance to participants regarding these negative social impacts associated with the vaccine trial. If HVTN CRS staff have questions regarding ways to assist a participant dealing with a social impact, a designated NIAID or HVTN Core representative can be contacted.

Social harms are tabulated by the SDMC and are subjected to descriptive analysis. The goal is to reduce their incidence and enhance the ability of study staff to mitigate them when possible.

Summary tables of social impact events will be generated weekly, and made available for review by the protocol chairs, protocol team leader, and Janssen Vaccines and NIAID representatives.

13.3 Compliance with NIH guidelines for research involving products containing recombinant DNA

Because this study is evaluating products containing recombinant or synthetic DNA, it must comply with regulations set forth in the *NIH Guidelines for Research Involving Recombinant or Synthetic Nucleic Acid Molecules(April 2016).* Information about the study must be submitted to site Institutional Biosafety Committees (IBCs). Investigators at each site are responsible for obtaining IBC approval per NIH Guidelines *section IV-B7-a-(1).* IBC review and approval must be documented by the investigator before participants can be enrolled at the site. If this protocol is amended, investigators should follow the requirements of their IBC.

13.4 Emergency communication with study participants

As in all clinical research, this study may generate a need to reach participants quickly to avoid imminent harm, or to report study findings that may otherwise concern their health or welfare.

When such communication is needed, the CRS will request that its IRB/EC and any applicable RE expedite review of the message. If this review cannot be completed in a timeframe consistent with the urgency of the required communication, the site should contact the participant first, and then notify the IRB/EC and any applicable RE of the matter as soon as possible.

14 Version history

The Protocol Team may modify the original version of the protocol. Modifications are made to HVTN protocols via clarification memos, letters of amendment, or full protocol amendments.

The version history of, and modifications to, Protocol HVTN 705/VAC89220HPX2008 are described below.

Protocol history and modifications

Date: June 12, 2020

Protocol version: Version 4.0
Protocol modification: Full Protocol Amendment 3

Item 1 Revised in Section 3, *Overview*, Section 5.2, *Secondary objectives and endpoints*, Table 6-1, *Cohorts of HIV-1-infected study participants*, Figure 6.5, *Study design and stages*, Section 10.5, *HIV infection during the study*, and Appendices A, D, E, G: redefining end of study, adding additional visits every 3 months until last Month 36 visit is complete

Item 2 Revised in Section 6.5, *Trial monitoring*, Section 6.5.1, *Monitoring for potential harm*, Section 6.5.2, *Monitoring for non-efficacy*: timing of assessment of non-efficacy to the point when all participants have reached Month 13.

Item 3 Added to Section 2.4, *Appropriate informed consent*, Section 7, *Selection and withdrawal of participants*, Section 9, *Clinical procedures,* and Appendix A, *Sample informed consent form*: information regarding remote study visits and consenting during the COVID-19 pandemic

Item 4 Added to Section 3, *Overview*, Section 6.3.1, *Analysis to determine whether or not the trial will advance into Stage 2*, and Figure 6.5, *Study design and stages*: text clarifying definition of Stage 1 and 2

Item 5 Updated in Section 3, *Overview*: protocol team members

Item 6 Added to Section 4.1, *Rationale for trial concept,* and Section 17, *Literature cited*: recent information regarding the HIV vaccine efficacy study HVTN 702

Item 7 Updated in Appendix A, *Sample informed consent form* and Appendix C, *Sample consent form for use of samples and information in other studies*: SAHPRA contact information

Item 8 Updated with changes described in Letter of Amendment 1 dated May 6, 2019

Item 9 Updated with changes described in Clarification memo 1 dated March 08, 2019

Item 10 Version history updated in Section 14

Date: May 06, 2019

Protocol version: Version 3.0
Protocol modification: Letter of Amendment 1

Item 1 Revised in Section 7.3.1, *Delaying vaccinations for a participant*, Section 7.3.2, *Participant departure from vaccination schedule,* and Section 7.3.3*, Discontinuing vaccination for a participant* :guidance regarding vaccination out of window

Date: March 08, 2019

Protocol version: Version 3.0
Protocol modification: Clarification memo 1

Item1 Revised in Section 9.10, *Visit windows and missed visits*: Missed Visit form completion

Date: July 26, 2018

Protocol version: Version 3.0
Protocol modification: Full Protocol Amendment 2

Item 1 Added in Section 4.3, *Clade C gp140*: Clade C gp140 (L-Histidine buffer) study product to be mixed with aluminum phosphate adjuvant by the pharmacist

Item 2 Corrected in Section 9.4, *Follow-up visits*, Section 9.8, *Pregnancy prevention assessment*, and Appendix G, *Schedule 1:Procedures at HVTN CRS for HIV-uninfected participants:* pregnancy prevention assessment timepoints

Item 3 Added in Appendix G, *Schedule 1:Procedures at HVTN CRS for HIV-uninfected participants*: behavioral risk assessment timepoints at month 7 and month 36

Item 4 Corrected in Appendix F, *Laboratory procedures for HIV-infected participants*, footnote 5: visits in Schedule 1 that may affect redraw guidance.

Item 5 Corrected in Appendix E, *Schedule 1:Laboratory procedures for HIV-uninfected participants*, footnote 14 and Appendix G, *Procedures at HVTN CRS for HIV-uninfected participants*, footnote 8: hemoglobin assessment and potential blood draw adjustment visits

Item 6 Added in Section 3, *Overview*, and Appendices E and F, *Laboratory Procedures*: UW-VSL as a backup laboratory for HIV diagnostics

Item 7 Updated in Section 11.1, *HVTN CRS laboratory procedures*: source for guidelines concerning the clinical and processing laboratories

Item 8 Updated in Section 12.1.5, S*ponsor roles and responsibilities in safety monitoring*, Section 12.3, *Safety pause and prompt PSRT AE review*, Appendix A, *Sample Informed Consent*, Appendix C, *Sample consent form for use of samples and information in other studies*: MCC and contact information updated to SAHPRA

Item 9 Clarified in Appendix A, *Sample Informed Consent*, item 26: insurance company and funds refer to the clinic's insurance company and funds

Item 10 Added in Appendix A, *Sample Informed Consent* and Appendix C, *Sample consent form for use of samples and information in other studies*: broad regulatory access to participant study records

Item 11 Clarified in Section 9.9, *Assessments of reactogenicity*: contact between staff and participant

Item 12 Added in Appendix G, *Procedures at HVTN CRS for HIV-uninfected participants*: HIV infection assessment at screening

Item 13 Corrected in Section 4.5, *Combination prevention of HIV acquisition*: misplaced reference

Item 14 Clarified in Section 7.3.3, *Discontinuing vaccination for a participant*: vaccinations may continue upon review by the PSRT for grade 3 erythema and/or induration

Item 15 Corrected in Section 9.1.1, *Screening consent form*: when screening can take place

Item 16 Clarified in Section 12.2.1, *Submission of safety forms to SDMC*: timing of submission of safety forms and federal or bank holidays

Item 17 Revised in Section 7.2, *Exclusion criteria* items 7 and 8, and Section 7.3.1, Delaying vaccination for a participant: timing of influenza vaccination made consistent with other live attenuated viruses

Item 18 Updated in Section 3.1, *Protocol Team*: affiliations and new members

Date: April 13, 2018

Protocol version: Version 2.0
Protocol modification: Full Protocol Amendment 1

Item 1 Added: Clade C gp140 and separate aluminum phosphate adjuvant study product formulation

Item 2 Updated in Section 3.1, Protocol team: email address and phone number for PPD PPD

Item 3 Updated: items from Clarification memo 1 dated September 25, 2017

Date: September 25, 2017

Protocol version: Version 1.0
Protocol modification: Clarification memo 1

Item 1 Updated in Section 9.9, *Assessments of reactogenicity*, Section 12.2.2 *AE reporting*, and Section 15, *Document references*: AE Grading table version

Item 2 Clarified in Appendix D, *Table of procedures (for sample informed consent) HIV-infected participants*, Appendix F, *Schedule 2 Laboratory procedures for HIV-infected participants*, and Appendix H, *Schedule 2 Procedures at HVTN CRS for HIV-infected participant*s: pregnancy testing for HIV-infected participants at months 3 and 6

Item 3 Updated in Section 3.1, *Protocol team*: email address for PPD

Item 4 Updated in Section 3.1, *Protocol team*: new team member PPD

Date: February 17, 2017

Protocol version: 1.0
Protocol modification:
Original protocol

15 Document references (other than literature citations)

Other documents referred to in this protocol, and containing information relevant to the conduct of this study, include:

- Assessment of Understanding. Accessible through the HVTN protocol-specific website.
- Current CDC Guidelines. Revised Recommendations for HIV Testing of Adults, Adolescents, and Pregnant Women in Health-Care Settings. Available at http://www.cdc.gov/mmwr/PDF/rr/rr5514.pdf
- Division of AIDS (DAIDS) Clinical Research Policies and Standard Procedures Documents. Available at https://www.niaid.nih.gov/research/daids-clinical-research-policies-standard-procedures
- Division of AIDS Table for Grading the Severity of Adult and Pediatric Adverse Events. Corrected Version 2.1, July 2017. Available at http://rsc.tech-res.com/clinical-research-sites/safety-reporting/daids-grading-tables
- HVTN Certificate of Confidentiality. Accessible through the HVTN website.
- HVTN 705/VAC89220HPX2008 Special Instructions. Accessible through the HVTN protocol-specific website.
- HVTN 705/VAC89220HPX2008 Study Specific Procedures. Accessible through the HVTN protocol-specific website.
- HVTN 705/VAC89220HPX2008 Site Lab Instructions. Accessible through the HVTN protocol-specific website.
- HVTN Laboratory Manual of Operations. Accessible through the HVTN website.
- HVTN Manual of Operations. Accessible through the HVTN website.
- Dangerous Goods Regulations (updated annually), International Air Transport Association. Available for purchase at http://www.iata.org/publications/dgr/Pages/index.aspx
- Lab assay algorithm
- HVTN algorithm for diagnosis of HIV infections. Part of the HVTN Laboratory Manual of Operations (see above).
- International Conference on Harmonisation (ICH) E6 (R1), Guideline for Good Clinical Practice: Section 4.8, Informed consent of trial subjects. Available at http://www.ich.org/products/guidelines/efficacy/article/efficacy-guidelines.html
- Participants' Bill of Rights and Responsibilities. Accessible through the HVTN website.
- NIH *Guidelines for Research Involving Recombinant or Synthetic Nucleic Acid Molecules, April 2016*. Available at http://osp.od.nih.gov/sites/default/files/resources/NIH_Guidelines.pdf
- NIH Policy on Reporting Race and Ethnicity Data: Subjects in Clinical Research. Available at http://grants1.nih.gov/grants/guide/notice-files/NOT-OD-01-053.html.
- Janssen Site Investigational Product Procedures Manual.

- Requirements for Source Documentation in DAIDS Funded and/or Sponsored Clinical Trials. Available at https://www.niaid.nih.gov/sites/default/files/daids-sourcedocpolicy.pdf
- Title 21, Code of Federal Regulations, Part 50. Available at http://www.ecfr.gov/cgi-bin/text-idx?SID=7e6fd19033c54083955c83c75675f72e&mc=true&node=pt21.1.50&rgn=div5
- Title 45, Code of Federal Regulations, Part 46. Available at http://www.hhs.gov/ohrp/humansubjects/guidance/45cfr46.html

See Section 17 for literature cited in the background and statistics sections of this protocol.

16 Acronyms and abbreviations

Ad	adenovirus
AE	adverse event
ART	antiretroviral therapy
CAB	Community Advisory Board
CBC	complete blood count
CDC	US Centers for Disease Control and Prevention
CFR	Code of Federal Regulations
CIOMS	Council for International Organizations of Medical Sciences
CRF	case report form
CRPMC	NIAID Clinical Research Products Management Center
CRS*	clinical research site
DAERS	DAIDS Adverse Experience Reporting System
DAIDS	Division of AIDS (US NIH)
DHHS	US Department of Health and Human Services
DSMB	NIAID Data and Safety Monitoring Board
EC	Ethics Committee
EIA	enzyme immunoassay
ELISA	enzyme-linked immunosorbent assay
ELISpot	enzyme-linked immunospot
FDA	US Food and Drug Administration
FHCRC	Fred Hutchinson Cancer Research Center
GCP	Good Clinical Practice
GMP	Good Manufacturing Practice
HLA	human leukocyte antigen
HVTN	HIV Vaccine Trials Network
IB	Investigator's Brochure
IBC	Institutional Biosafety Committee
ICH	International Conference on Harmonisation
ICS	intracellular cytokine staining
IFN-γ	interferon gamma
IND	Investigational New Drug
IRB	Institutional Review Board
IUD	intrauterine device
MMR	measles, mumps, and rubella
nAb	neutralizing antibody
NHP	nonhuman primate
NIAID	National Institute of Allergy and Infectious Diseases (US NIH)
NICD	National Institute for Communicable Diseases (Johannesburg, South Africa)
NIH	US National Institutes of Health
OG	Oversight Group
OPV	oral polio vaccine

PAB	DAIDS Pharmaceutical Affairs Branch
PBMC	peripheral blood mononuclear cell
PCR	polymerase chain reaction
PI	Principal Investigator
PSRT	Protocol Safety Review Team
RAB	DAIDS Regulatory Affairs Branch
RE	regulatory entity
RSC	DAIDS Regulatory Support Center
SAE	serious adverse event
SCHARP	Statistical Center for HIV/AIDS Research and Prevention
SDMC	statistical and data management center
SFC	spot-forming cell
SHIV	simian-human immunodeficiency virus
SIV	simian immunodeficiency virus
TB	tuberculosis
VISP	Vaccine induced seropositivity

* CRSs were formerly referred to as HIV Vaccine Trial Units (HVTUs). Conversion to use of the term CRS is in process, and some HVTN documents may still refer to HVTUs.

17 Literature cited

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Appendix A Sample informed consent form

Title: A multicenter, randomized, double-blind, placebo-controlled phase 2b efficacy study of a heterologous prime/boost vaccine regimen of Ad26.Mos4.HIV and aluminum phosphate-adjuvanted Clade C gp140 in preventing HIV-1 infection in women in sub-Saharan Africa

Protocol number: HVTN 705/VAC89220HPX2008

Site: [Insert site name]

Thank you for your interest in our research study. Please read this consent form or ask someone to read it to you. If you decide to join the study, we will ask you to sign or make your mark on this form. We will offer you a copy to keep. We will ask you questions to see if we have explained everything clearly. You can also ask us questions about the study.

Research is not the same as treatment or medical care. The purpose of a research study is to answer scientific questions.

About the study

The HIV Vaccine Trials Network (HVTN), Janssen Vaccines and Prevention B.V., and [Insert site name] are doing a study to test HIV vaccines. HIV is the virus that causes AIDS.

About 2600 women will take part in this study at multiple sites in several sub-Saharan African countries. The researcher in charge of this study at this clinic is [Insert name of site PI]. The US National Institutes of Health (NIH), the Bill & Melinda Gates Foundation, and Janssen Vaccines & Prevention, B.V. are paying for the study.

1. **We are doing this study to answer several questions.**
   - Do the study vaccines lower people's chances of getting infected with HIV?
   - Are the study vaccines safe to give to people?
   - Are people able to take the study vaccines without becoming too uncomfortable?
   - How do people's immune systems respond to the study vaccines? (Your immune system protects you from disease.)
   - Do the study vaccines lower how much HIV is in people's blood if they get HIV?

2. **The study vaccines cannot give you HIV.**

   It is impossible for the study vaccines to give you HIV. Also, they cannot cause you to give HIV to someone else. The study vaccines are not made from actual HIV. Below, we will tell you about how the vaccines in this study are made.

3. **We do not know if the study vaccines will decrease, increase, or not change your risk of becoming infected with HIV if you are exposed to the virus.**

   Sites: Any change to the language in this section requires approval from HVTN Regulatory Affairs.

Several studies have tested whether HIV vaccines can reduce the risk of getting HIV from another person. In some studies, people who got the vaccine seemed to have the *same* risk of getting HIV as people who did not get the vaccine. In one study, people who got the vaccine seemed to have a *lower* risk of getting HIV than people who did not get the vaccine. In studies with a different vaccine, some people who got the vaccine had a *higher* risk of getting HIV than people who did not get the vaccine.

The studies that found a higher risk of HIV infection were testing a vaccine made from a weakened common cold virus called adenovirus type 5 (Ad5).

In one study, the group of men at higher risk had some things in common. In addition to getting the vaccine, they either:

- had antibodies to Ad5 (your body makes antibodies to fight infection),
- or they were uncircumcised (still had the foreskin on their penises),
- or both.

Men who were circumcised and did not have these antibodies did not have higher risk when they got the vaccine. In the group with higher risk after vaccination, the risk seemed to lessen after about a year and a half.

Only a few women in that study got HIV, so we can't tell if the vaccine affected their risk.

In another study, men who got the vaccine had higher risk whether or not they were circumcised or had antibodies to Ad5. Many women became HIV infected during the study but we can't tell if the vaccine affected their risk.

This study differs from those studies in several ways. It is testing a vaccine made from adenovirus type 26 (Ad26), not Ad5. We can tell you about other differences.

We do not know whether the vaccines in this study will affect your risk of getting HIV from another person. The risk could be higher, lower, or unchanged. It's very important to avoid exposure to HIV during and after the study. We will tell you how to avoid HIV.

4. These study vaccines are experimental.

The study vaccines are called Ad26.Mos4.HIV and Clade C gp140. From here on, we will call them Ad26 and Protein, or the study vaccines.

The study vaccines are experimental HIV vaccines. That means we do not know whether the vaccines will be safe to use in people, or whether they will work to prevent HIV infection. These vaccines are used only in research studies.

The vaccines were developed by Janssen Vaccines & Prevention B.V. The Ad26 vaccine is made from a virus called Adenovirus type 26. Adenovirus type 26 is a virus that can cause the common cold. However, the adenovirus used in this study vaccine has been weakened so it cannot cause colds. This study vaccine has been designed to tell the body to make proteins similar to proteins found in HIV.

The Protein vaccine includes a man-made protein called gp140 that is similar to the protein found on the outside of HIV. The gp140 protein vaccine is mixed with an adjuvant called aluminum phosphate, or alum for short. An adjuvant is something added to a vaccine to help the immune system respond better. Alum is an approved adjuvant that is used in many common vaccines such as those for Hepatitis A and B, diphtheria, and tetanus.

General risks of vaccines:

All vaccines can cause fever, chills, rash, aches and pains, nausea, headache, dizziness, and feeling tired. Vaccines can also cause pain, redness, swelling, or itching where you got the injection. Most people can still do their planned activities after getting a vaccine. Rarely, people experience side effects that limit their normal activities or make them go to the doctor.

Rarely, a vaccine can cause an allergic reaction, including a rash, hives, or difficulty breathing. Allergic reactions can be life-threatening. You should tell us if you have ever had a bad reaction to any injection or vaccine.

Very rarely, a vaccine causes an autoimmune disease in a person, or makes an autoimmune disease worse. An autoimmune disease happens when your immune system attacks your own body, instead of attacking an infection.

Risks of the study vaccines:

This section lists the side effects we know about. There may be others that we don't yet know about, even serious ones. We will tell you if we learn about any new side effects.

The Ad26 vaccine:

The Ad26 vaccine is being tested in 2 other studies (HVTN 117/HPX2004 and HVTN 118/HPX2003) that started before this study. About 235 people in the United States, Rwanda, and Kenya are participating in those studies.

Two similar vaccines have been tested in other studies. One of the similar vaccines has been given to over 200 people. That vaccine did not make people too uncomfortable or cause them serious health problems. The other similar vaccine has been given to about 380 people. In those studies, that vaccine did not make people too uncomfortable. One person did have a serious allergic reaction and chest pain. The symptoms went away in about 1 day. Studies with a small number of people do not tell us everything about the safety of a study vaccine.

The gp140 Protein vaccine:

The Protein vaccine is being tested in 5 other studies (HVTN 117/HPX2004, HVTN 118/HPX2003, HPX1002, HIV-V-A003, and HIV-V-A004) that started before this study. As of January 2017, about 300 people received the gp140 Protein vaccine. So far, it has not made them too uncomfortable or caused them serious health problems. However, studies with a small number of people do not tell us everything about the safety of a study vaccine.

A similar Protein vaccine (gp120 Protein) has been given to thousands of people in other studies over 10 years. In these studies, the gp120 Protein vaccine did not cause serious health problems.

Aluminum phosphate adjuvant:

As we told you, gp140 Protein vaccine is mixed with an adjuvant called aluminum phosphate. Too much aluminum in the body can cause problems with nerves and bones, mostly for people who already have problems with their kidneys. The aluminum in vaccines is much less than what people eat or take in medicines such as antacids.

5. It is completely up to you whether or not to join the study.

Take your time in deciding. If it helps, talk to people you trust, such as your doctor, friends or family. If you decide not to join this study, or if you leave it after you have joined, your other care at this clinic and the benefits or rights you would normally have will not be affected.

If you join this study, you may not be allowed to join other HIV vaccine or HIV prevention studies now or in the future. You cannot be in this study while you are in another study where you get a study product. Also during the study, you should not donate blood or tissue. We will tell you more about this below.

If you choose not to join this study, you may be able to join another study.

Site: Remove item 6 if you use a separate screening consent that covers these procedures.

6. If you want to join the study, we will screen you to see if you are eligible.

Screening involves a physical exam, HIV test and health history. A physical exam may include, but is not limited to:

- Checking your weight, temperature and blood pressure
- Looking in your mouth and throat
- Listening to your heart and lungs
- Feeling your abdomen (stomach and liver)

We will also test your blood for hemoglobin. This test tells us about the health of your blood. We will also do a pregnancy test. We will ask you about medications you are taking. We will ask you about behaviors that might put you at risk for getting HIV.

We will review the screening results with you. The screening results may show you are not eligible to join the study, even if you want to.

Sites: adapt the following section so it is applicable to the care available at your site.

7. If we find that you have a health problem during screening or during the study, we will tell you about the care that we can give here for free.

For the care that we cannot give, we will explain how we will help you get care elsewhere. For health problems that are unrelated to the study, we will not pay for care.

8. **If you could become pregnant, you must agree to use birth control to join this study.**

   Site: If you want to include Appendix B, Approved birth control methods (for sample informed consent form), in this consent form, paste it below and delete paragraph below.

   You should not become pregnant during the study because we do not know how the study vaccines could affect the developing baby. You must agree to use effective birth control from 21 days before your first injection until 3 months after your last study injection. We will talk to you about effective birth control methods. They are listed on a handout that we will give to you.

Being in the study

If you meet the study requirements and want to join, here is what will happen:

9. **You will come to the clinic for scheduled visits about [#] times over [Insert period of time].**

   Site: Insert number of visits and range of visit lengths. (There is site-specific variation in screening protocols and in the number of possible follow-up visits between protocol-mandated visits.)

   Visits can last from [#] to [#] hours.

   You may have to come for more visits if you have a lab or health issue.

   We may contact you after the study ends (for example, to tell you about the study results).

   In response to the COVID-19 pandemic, we want to make it easier for you to stay in the study. If lockdowns or stay-at-home orders make travel to the clinic difficult, we may offer to arrange transport for you to the clinic and back home. Or we can do some parts of these visits over the phone or through electronic communication such as text messages and email. We will discuss with you what works best.

10. **We will give you [Site: Insert compensation] for each study visit you complete.**

    This amount is to cover the costs of [Site: Insert text]

    Site: Insert any costs to participants (eg, birth control costs for female participants who could become pregnant).

    You do not have to pay anything to be in this study.

11. **We will give you either the study vaccines or a placebo.**

    Not everyone in this study will get the study vaccines. Some people will get a placebo, a substance that does not contain vaccine. We will compare the results from people who got the placebo with results from people who got the study vaccines. In this study, the placebo is sterile salt water.

You have a 1-in-2 chance of getting the study vaccines. *Site: Modify the randomization metaphor in the next sentence as appropriate to your local culture.* Whether you get the study vaccines or the placebo is completely random, like flipping a coin.

We have no say in whether you get the study vaccines or the placebo. We will not know which one you are getting, and neither will you. Only the pharmacist at this clinic will have this information while the study is going on.

Unless the study is stopped early, you will have to wait until everyone completes their final study visits to find out whether you got the study vaccines or the placebo. This could be several years. But, if you have a serious medical problem and need to know what you got before the end of the study, we can find out and tell you.

If the study shows that the vaccines are working, the researchers will talk to you about how this may affect your participation.

12. We will give you the study products on a schedule.

You will be in 1 of 2 groups, vaccine or placebo. You will get a total of 6 injections into the upper arms during the study. You will get 1 injection of either the Ad26 vaccine or placebo at your first injection visit. Three (3) months later, you will get the same injection. Six (6) and 12 months after your first injection, you will get 2 injections at each visit. You will get either the Ad26 vaccine or placebo in your left arm, and the Protein vaccine or placebo in your right arm.

Site: You may insert the picture version of the injection schedule (Appendix J) in place of (or in addition to) the text version or give it as a separate document to volunteers if you believe it will be helpful to them. You are not required to do either.

Injection Schedule

Group	First injection	3 months after first injection	6 months after first injection	12 months after first injection
1	Ad26	Ad26	Ad26 + Protein/Alum	Ad26 + Protein/Alum
2	Placebo	Placebo	Placebo	Placebo

Site: If your site has been selected for the 7-day reactogenicity assessment then replace 3 more days in the next paragraph with 7 more days.

You will have to wait in the clinic for about a half hour after each injection to see if there are any problems. Then for that night and for 3 more days, you will need to keep track of how you are feeling and if you have any symptoms. To help you do this we will give you a memory tool and show you how to use it. We will call you about a week after your injection visit to see how you are doing. Contact the clinic staff if you have any issues or concerns after getting an injection. If you have a problem, we will continue to check on you until it goes away.

13. In addition to giving you the study products, we will:

- Do regular HIV testing, as well as counseling on your results and on how to avoid getting HIV;
- Do physical exams;
- Do pregnancy tests;
- Ask questions about your health, including medications you may be taking;
- Ask questions about any personal problems or benefits you may have from being in the study; and
- Take blood and urine samples.

When we take blood, the amount will depend on the lab tests we need to do. It will be some amount between 10 mL and 350 mL (2 teaspoons to 1½ cup). Your body will make new blood to replace the blood we take out.

Site: You may want to add a sentence to the end of the previous paragraph contextualizing the blood volumes described (eg, "To compare, people who donate blood in the US can give a total of about 500 mL in an 8-week period."). Modify the example for cultural relevance and alter blood volumes as necessary.

Site: Insert Appendix D, "Table of procedures (for informed consent form), HIV-uninfected participants" in this section or distribute it as a separate sheet if it is helpful to your study participants. You are not required to do either.

We will be looking for side effects. We will review the results of these procedures and tests with you at your next visit, or sooner if necessary. If any of the results are important to your health, we will tell you.

14. You are being asked to have additional visits in this study.

The purpose of the additional visits is to collect more information about the safety of this vaccine and how your immune system responds to the vaccine. The number of additional visits will depend on when you started the study. For example, if you started in November 2017 then you may have 6 or 7 extra visits; if you started in May 2019 you may have no or 1 extra visits. We will complete the study for all people at the same time about July 2022.

We will not give you any more study products at these extra visits. We will:

- Do regular HIV testing, as well as counseling on your results and on how to avoid getting HIV;
- Ask questions about your health, including medications you may be taking;
- Do physical exams if required;
- Ask questions about any personal problems or benefits you may have from being in the study; and
- Take blood and urine samples.

Site: Insert Appendix D, "HIV-uninfected participants: Additional visits" in this section or distribute it as a separate sheet if it is helpful to your study participants. You are not required to do either.

15. We will also collect vaginal fluid and swabs.

We want to see if the bacteria that live in your vagina affect your risk of getting HIV.

At 5 visits, we will ask you to collect vaginal fluid using a disposable menstrual cup. We will explain how you can insert and remove the cup, or we can do it for you. We will explain how long you will wear the cup. At these same visits, we will also ask you to collect a vaginal swab. We will ask you to place the swab into your vagina. We will teach you how to do this. If you do not want to swab yourself, we can do it for you.

At these same 5 visits, plus up to 5 more additional visits, we will also test you for gonorrhea, chlamydia, syphilis, and Trichomonas vaginalis. These are all common sexually transmitted infections. In order to do some of these tests, we will ask you to collect another vaginal swab at those visits. We will give you your test results. If we find that you have an infection and you need care, we will tell you about the care we can give you here. We will also tell you about care we can help you get elsewhere.

Sites: Do not make changes to the following section without obtaining approval from HVTN Regulatory Affairs at vtn.core.reg@hvtn.org.

16. We will counsel you on avoiding HIV infection.

We will ask you personal questions about your HIV risk factors such as sexual behavior, alcohol, and drug use. *Site: Modify this next sentence based on what you provide:* We will provide you with condoms and lubricant. We will talk with you about ways of lowering your risk of getting HIV. We will help you develop a risk reduction plan. Our counseling may include:

- What you think causes risky behavior for you.
- Ways to avoid getting HIV.
- If you become infected with HIV, we will talk with you about ways to avoid giving the virus to someone else.

These may include not having sex, using condoms, or other behavior changes, such as cutting down on alcohol. We will talk about new methods of HIV prevention as these become available, and will give you information on how to access them.

These methods may include the use of anti-HIV drugs taken every day to prevent HIV. This is called pre-exposure prophylaxis or PrEP. A combination of two anti-HIV drugs (TDF/FTC) has been licensed in South Africa for use to prevent HIV infection in people at high risk, and we think that PrEP will become more available in other southern African countries. Studies have shown this drug combination may reduce the risk of getting HIV when it is taken every day. The drug combination is not always effective and HIV infections may happen, especially if the medicine is not taken every day. Where PrEP is licensed, this drug combination will be available in the private health sector by prescription from a doctor. In southern Africa, the national departments of health are working with international donors to establish "demonstration projects" that may provide

this drug combination to people free of charge. Where available, we can refer you to the nearest of these programs if you are interested. In this study, we will look at how many participants use PrEP. We will also try to find out if using PrEP has any effect on how the study products work.

17. We will test your samples for this study.

We will send your samples (without your name) to labs approved by the HVTN for this study, which are located in the United States, South Africa, and Europe. Researchers at these labs will test your samples to see how your immune system responds to the study products. In rare cases, some of your samples may be sent to labs approved by the HVTN in other countries for research related to this study.

Researchers may also do genetic testing related to this study on your samples. Your genes are passed to you from your birth parents. They affect how you look and how your body works. The differences in people's genes can help explain why some people get a disease while others do not. These types of genetic tests involve some of your genes, not all of your genes (your genome). The researchers will study the genes related to the immune system and HIV and those that affect how people get HIV.

The researchers may look at all of the genes of the bacteria found in your vaginal samples. If you become HIV infected, the researchers may look at all of the genes of the virus found in your samples. In both cases, the researchers will use this information to learn more about HIV and the study products. The researchers may put this information about the virus and/or bacteria into a protected database so that other researchers can access it. They would not be able to link the information to you.

In some cases, researchers may take cells from your samples and grow more of them over time, so that they can continue to contribute to this study.

In some participants, we may test for antibodies against Ad26. This can tell us if you have been previously exposed to this virus. We want to see if this has an effect on how the vaccine works.

Tests done on your samples are for research purposes, not to check your health. The labs will not give the results to you or this clinic because their tests are not approved for use in making health care decisions. These labs are only approved to do research tests.

When your samples are no longer needed for this study, the HVTN will continue to store them.

Site: Delete next section if using separate consent for use of samples and information in other studies

18. When samples are no longer needed for this study, the HVTN wants to keep them for use in other studies by HVTN or other researchers. We will call these "extra samples."

This section gives you information so you can decide if you want your extra samples and study-related information used in other studies. You will mark your decision at the end of the form. If you have any questions, please ask.

Do I have to agree? No. You are free to say yes or no, or to change your mind after you sign this form. At your request, we will destroy all extra samples that we have. Your decision will not affect your being in this study or have any negative consequences here.

Where are the samples stored? Extra samples are stored in a secure central place called a repository. *[Site: insert specific information if your regulatory authority requires it.]* Your samples will be stored in the HVTN repository in South Africa.

How long will the samples be stored? There is no limit on how long your extra samples will be stored. *[Site: insert limits if your regulatory authority imposes them.]*

Will I be paid for the use of my samples? No. Also, a researcher may make a new scientific discovery or product based on the use of your samples. If this happens, there is no plan to share any money with you. The researcher is not likely to ever know who you are.

Will I benefit from allowing my samples to be used in other studies? Probably not. Results from these other studies are not given to you, this clinic, or your doctor. They are not part of your medical record. The studies are only being done for research purposes.

Will the HVTN sell my samples and information? No, but the HVTN may share your samples with other researchers. Once we share your samples and information, we will not be able to get them back.

How do other researchers get my samples and information? When a researcher wants to use your samples and/or information, their research plan must be approved by the HVTN. Also, the researcher's institutional review board (IRB) or ethics committee (EC) will review their plan. *[Site: If review by your institution's IRB/EC/RE is also required, insert a sentence stating this.]* IRBs/ECs protect the rights and well-being of people in research. If the research plan is approved, the HVTN will send your samples to the researcher's location.

What information is shared with other researchers? The samples and limited information will be labeled with a code number. Your name will not be part of the information. However, some information that we share may be personal, such as your race, ethnicity, sex, health information from the study, and HIV status. We may share information about the study product you received and how your body responded to the study product.

What kind of studies might be done with my extra samples and information? The studies will be related to HIV, vaccines, the immune system and other diseases.

Researchers may also do genetic testing on your samples.

Researchers may look at all of the genes of the bacteria found in your vaginal samples. If you become HIV infected, the researchers may look at all of the genes of the virus found in your samples. In both cases, the researchers will use this information to learn more about HIV and the study products.

In some cases, researchers may take cells from your samples and grow more of them over time, so that they can continue to contribute to this study.

If you agree, your samples could also be used for genome wide studies. In these studies, researchers will look at all of your genes (your genome). The researchers compare the genomes of many people, looking for common patterns of genes that could help them understand diseases. The researchers may put the information from the genome-wide studies into a protected database so that other researchers can access it. Usually, no one would be able to look at your genome and link it to you as a person. However, if another database exists that also has information on your genome and your name, someone might be able to compare the databases and identify you. If others found out, it could lead to discrimination or other problems. The risk of this is very small.

Who will have access to my information in studies using my extra samples?

People who may see your information are:

- Researchers who use your stored samples and limited information for other research
- Government agencies that fund or monitor the research using your samples or information
- The researcher's Institutional Review Board or Ethics Committee
- Any regulatory agency that reviews clinical trials
- The people who work with the researcher

All of these people will do their best to protect your information. The results of any new studies that use your extra samples or information may be published. No publication will use your name or identify you personally.

19. We will do our best to protect your private information.

Your study records and samples will be kept in a secure location. We will label all of your samples and most of your records with a code number, not your name or other personal information. However, it is possible to identify you, if necessary. We will not share your name with the lab that does the tests on your samples, or with anyone else who does not need to know your name.

Sites: Any change to the following boxed text requires approval from HVTN Regulatory Affairs.

We do need to share your name with the HVTN in case you need proof in the future that you participated in an HIV vaccine study. The HVTN will keep your name in a secure file with these items:

- The name of your study
- Your age or date of birth
- Your study ID number
- What study product(s) you received

There are no HIV test results kept in this file. The HVTN will not share any information that could identify you without your agreement. The HVTN will remove your name from the file if you do not want it there.

Clinic staff will have access to your study records. Your records may also be reviewed by groups who watch over this study to see that we are protecting your rights, keeping you safe, and following the study plan. These groups include:

- The US National Institutes of Health and its study monitors,
- The US Food and Drug Administration,
- Any regulatory agency that reviews clinical trials,
- [Insert name of local IBC],
- [Insert name of local IRB/EC] ,
- [Insert name of local and/or national regulatory authority as appropriate],
- Janssen Vaccines & Prevention B.V., people who work for them, and their chosen South African representatives,
- The HVTN and people who work for them,
- The NIAID Data Safety and Monitoring Board, and
- The US Office for Human Research Protections.

All reviewers will take steps to keep your records private.

We cannot guarantee absolute privacy. At this clinic, we have to report the following information:

Site: Include any public health or legal reporting requirements. Bulleted examples should include all appropriate cases (reportable communicable disease, risk of harm to self or others, etc.).

- [Item 1]
- [Item 2]
- [Item 3]

The results of this study may be published. No publication will use your name or identify you personally.

We may share information from the study with other researchers and those groups paying for this study. We will not share your name or information that can identify you.

Sites: The text below may not be deleted or changed, per FDA requirement. It's OK to remove the box around it.

A description of this clinical trial will be available on http://www.ClinicalTrials.gov, as required by U.S. Law. This website will not include information that can identify you. At most, the website will include a summary of the results. You can search this website at any time.

20. We may stop your injections or take you out of the study at any time. We may do this even if you want to stay in the study and even if you were scheduled for more injections.

This may happen if:

- you do not follow instructions,
- we think that staying in the study might harm you,
- you enroll in a different research study where you get another study product,
- people who got the study vaccines are getting infected with HIV more than people who got the placebo, or
- the study is stopped for any reason.

If we stop your injections, we may ask you to stay in the study to complete other study procedures.

21. We will stop your injections if you become pregnant before your last injection.

We will encourage you to stay in the study if you choose. We will discuss your study options with you.

If you leave the study while you are still pregnant, we will contact you after your due date to ask some questions about your pregnancy and delivery.

22. If you get infected with HIV during the study, we will help you get care and support.

We will counsel you about your HIV infection and about telling your partner(s). We will tell you where you can get support and medical care. *Site: Modify the following sentence as appropriate.* We will not provide or pay for any of your HIV care directly.

We will also ask you to come to the clinic for about 3 visits over the following 6 months, but we will not give you any more injections. Even though your study injections will have stopped, learning about your health is an essential part of this study. At these visits, we will:

- Ask questions about your health;
- Do physical exams based on how you tell us you are feeling;
- Ask questions about your risk of infecting others with HIV, including sexual behavior and drug use;
- Counsel you on how to avoid infecting others with HIV;
- Do pregnancy tests;
- Collect blood samples (about 230 mL; about a cup at each visit); and
- Ask questions about any personal problems or benefits you may have from participating in the study.

Site: Insert Appendix D, "Table of procedures (for informed consent form), HIV-infected participants" in this section or distribute it as a separate sheet if it is helpful to your study participants. You are not required to do either.

Other Risks

23. There are other risks to being in this study.

This section describes the other risks and restrictions we know about. There may also be unknown risks, even serious ones. If you have a problem because you are in this study, we will help you. We will tell you if we learn anything new that may affect your willingness to stay in the study.

Risks of routine medical procedures:

In this study, we will do some routine medical procedures. These are taking blood and giving injections. These procedures can cause bruising, pain, fainting, soreness, redness, swelling, itching, a sore, bleeding, and (rarely) muscle damage or infection where you got the injection. Taking blood can cause a low blood cell count (anemia), making you feel tired.

Risks of menstrual cup:

You may feel some discomfort when inserting or removing the menstrual cup.

Risks of vaginal swabs:

You may have some discomfort when you swab your vagina.

Personal problems/discrimination/testing HIV antibody positive:

About 1 or 2 in 10 people who join HVTN studies report personal problems or discrimination because of joining an HIV vaccine study. Family or friends may worry, get upset or angry, or assume that you are infected with HIV or at high risk and treat you unfairly as a result. Rarely, a person has lost a job because the study took too much time away from work, or because their employer thought they had HIV.

The body makes antibodies to fight or prevent infection. Most vaccines cause the body to make antibodies as a way of preventing infection. Your body may make antibodies to HIV because you received HIV study vaccines. The study vaccines are likely to cause you to test positive on some types of HIV antibody tests, even if you are not infected with HIV. This is called vaccine-induced seropositivity (VISP). VISP means that after you get the study vaccines, a routine HIV test done outside this clinic is likely to say you have HIV, even if you don't. For this reason, you should plan to get HIV tests only at this clinic during the study. Our tests can tell the difference between true HIV infection and a positive result that is caused by the study vaccines.

If you have a positive test result caused by the study vaccines at any time, it does not mean that there is a problem with your health. But, we can arrange free HIV testing for as long as you need it. If this happens, we do not know how long you will test HIV positive due to the study vaccines. If you receive a positive HIV test result and we determine it is because you have HIV, we will refer you for follow-up care.

It is unlikely, but you could test negative at the end of the study and positive some time later, even though you don't have HIV. This could happen if different HIV tests come into use. We will give you a phone number to call for more information.

Site: Modify the following paragraph if applicable. If someone believes you are infected with HIV even if you are not, you could face discrimination and other problems. For example, in some countries, you could be denied medical or dental care, employment, insurance, a visa, or entry into the military. If you do have a positive HIV antibody test caused by the study vaccines, you will not be able to donate blood or organs. Your family and friends may treat you differently. We will give you a brochure that tells you more about testing HIV positive because of an HIV vaccine, and how you can avoid some of these problems.

If you become pregnant during or after the study and have VISP, we don't know if the antibodies could be passed to your baby. We know that this happens with other vaccines, like tetanus vaccine. These antibodies from the mother are not a danger to the baby, and they go away over time. For most babies antibodies from the mother last for about six months.

You should always tell the delivery staff if you have VISP. However, you may still be tested for HIV using the antibody test when you deliver your baby. If your test is positive and the delivery staff believes you have an HIV infection, your baby may be started on antiretroviral treatment when it is not needed. If this happens, we can arrange for you and the baby to have a test that can tell the difference between true HIV infection and a VISP result. If you or the baby continue to have VISP, we can arrange this testing for free for as long as it is needed.

Embarrassment/anxiety:

You may feel embarrassed when we ask about your HIV risks, such as having sex and using drugs. Also, waiting for your HIV test results or other health test results could make you feel anxious. You could feel worried if your test results show that you are infected with HIV. If you feel embarrassed or anxious, please tell us and we will try to help you.

Risks of disclosure of your personal information:

We will take several steps to protect your personal information. Although the risk is very low, it is possible that your personal information could be given to someone who should not have it. If that happened, you could face discrimination, stress, and embarrassment. We can tell you more about how we will protect your personal information if you would like it.

Risks of genetic testing:

The genetic testing could show you may be at risk for certain diseases. If others found out, it could lead to discrimination or other problems. However, it is almost impossible for you or others to know your test results from the genetic testing. The results are not part of your study records and are not given to you.

Unknown risks:

We do not know if the study vaccines will increase, decrease, or not change your risk of becoming infected with HIV if exposed. If you get infected with HIV, we do not know how the study vaccines might affect your HIV infection or how long it takes to develop AIDS.

We do not know if getting these study vaccines will affect how you respond to any future approved HIV vaccine. It could be that a future HIV vaccine may not work as well for you because you got the study vaccines. Currently, no HIV vaccine has been approved for use.

We do not know how the study vaccines will affect a pregnant participant or a developing baby.

Benefits

24. The study may not benefit you.

We do not know whether getting the study vaccines might benefit you in any way. However, being in the study might still help you in some ways. The counseling that you get as part of the study may help you avoid getting HIV. The lab tests and physical exams that you get while in this study might detect health problems you don't yet know about.

This study may help in the search for a vaccine to prevent HIV. However, if the study vaccines later become approved and sold, there are no plans to share any money with you.

Your rights and responsibilities

25. If you join the study, you have rights and responsibilities.

You have many rights that we will respect. You also have responsibilities. We list these in the Participant's Bill of Rights and Responsibilities. We will give you a copy of it.

Leaving the study

26. Tell us if you decide to leave the study.

You are free to leave the study at any time and for any reason. Your care at this clinic and your legal rights will not be affected, but it is important for you to let us know.

We will ask you to come back to the clinic one last time for a physical exam, and we may ask to take some blood and urine samples. We will also ask about any personal problems or benefits you have experienced from being in the study. We believe these steps are important to protecting your health, but it is up to you whether to complete them.

Injuries

Sites: Do not make changes to the following section without obtaining approval from HVTN Regulatory Affairs at vtn.core.reg@hvtn.org.

27. If you get sick or injured during the study, contact us immediately.

Your health is important to us. *(Sites: adjust the following 2 sentences if applicable to the care available at your site)* We will tell you about the care that we can give here. For the care that we cannot provide, we will explain how we will help you get care elsewhere.

If you become sick or injured in this study, there is a process to decide if it is related to the study products and/or procedures. If it is, we call it a study-related injury. There are funds to pay for treatment of study-related injuries if certain conditions are met. The funds may come from different groups, as described below.

(Sites: adjust the language in this paragraph (or remove altogether) so it is applicable to your site. Note: insurance is purchased for all southern African countries; however, the ABPI guidelines apply to South Africa and Mozambique only.) In this study, our clinic has insurance to cover your medical treatment in the case of a study-related injury, following the Association of the British Pharmaceutical Industry guidelines for payment of study-related injury. We can give you a copy of these guidelines. In rare cases, the clinic's insurance funds may not be enough. In case the clinic's insurance company does not fully cover those medical costs, the sponsor of the study (Janssen Vaccines) can pay the remaining medical costs that are not covered by the clinic's insurance company, provided the injury is objectively determined to be caused directly by the vaccination in the trial with its candidate vaccine.

For study-related injuries that cannot be funded as described above, the HVTN has limited funds to pay medical costs that it determines are reasonable. *(Sites: insert locale-appropriate medical insurance language in the following sentence)* If the injury is not study related, then you and your health insurance will be responsible for treatment costs.

Some injuries are not physical. For example, you might be harmed emotionally by being in an HIV vaccine study. Or you might lose wages because you cannot go to work. However, there are no funds to pay for these kinds of injuries, even if they are study related.

You may disagree with the decision about whether your injury is study related. If you wish, the HVTN will ask independent experts to review the decision. You always have the right to use the court system if you are not satisfied.

Questions

28. If you have questions or problems at any time during your participation in this study, use the following important contacts.

If you have questions about this study, contact
[name and telephone number of the investigator or other study staff].

If you have any symptoms that you think may be related to this study, contact
[name and telephone number of the investigator or other study staff].

If you have questions about your rights as a research participant, or problems or concerns about how you are being treated in this study, contact
[name/title/phone of person on IRB or other appropriate organization].

If you want to leave this study, contact
[name and telephone number of the investigator or other study staff].

Remainder of section for South African sites only.

If you have questions about this trial you should first discuss them with your doctor or the ethics committee (contact details as provided on this form). After you have consulted your doctor or the ethics committee and if they have not provided you with answers to your satisfaction, you should write to the South African Healthcare Products Regulatory Authority (SAHPRA) at:

PPD

South African Health Products Regulatory Authority
Department of Health
Private Bag X828
PRETORIA
0001

Tel: PPD

e-mail: PPD

Your permissions and signature

Site: Delete this section if using a separate consent for use of samples and information in other studies

29. In Section 17 of this form, we told you about possible other uses of your extra samples and limited information, outside this study. Please choose only one of the options below and write your initials or make your mark in the box next to it. Whatever you choose, the HVTN keeps track of your decision about how your samples and information can be used.

☐ I allow my extra samples combined with limited information to be used for other studies related to HIV, vaccines, the immune system, and other diseases. This may include genetic testing and keeping my cells growing over time.

OR

☐ I agree to the option above *and* also to allow my extra samples combined with limited information to be used in genome wide studies.

OR

☐ I do not allow my extra samples to be used in any other studies. This includes not allowing genetic testing, growing more of my cells, or genome wide studies.

30. If you agree to join this study, you will need to sign or make your mark below. Before you sign or make your mark on this consent form, make sure of the following:

- You have read this consent form, or someone has read it to you.
- You feel that you understand what the study is about and what will happen to you if you join. You understand what the possible risks and benefits are.
- You have had your questions answered and know that you can ask more.
- You agree to join this study.

You will not be giving up any of your rights by signing this consent form.

Participant's name (print)	Participant's signature or mark	Date	Time

Clinic staff conducting consent discussion (print)	Clinic staff signature	Date	Time

For participants who are unable to read or write, a witness should complete the signature block below:

Witness's name (print)	Witness's signature	Date	Time

*Witness is impartial and was present for the consent process.

Appendix B Approved birth control methods (for sample informed consent form)

You should not become pregnant during the study because we do not know how the study vaccines could affect the developing baby.

You must agree to use effective birth control from 21 days before your first injection until 3 months after your last study injection.

Effective birth control means using any of the following methods every time you have sex:

- Birth control drugs that prevent pregnancy – given by pills, shots, patches, vaginal rings, or inserts under the skin; or
- Intrauterine device (IUD); or
- Any other contraceptive method approved by the researchers.

You do not have to use birth control if:

- You have been diagnosed with early menopause, with no menstrual periods for one year; or
- You have had a hysterectomy (your uterus removed); or
- You have had your ovaries removed; or
- You have a tubal ligation (your "tubes tied") or confirmed successful placement of a product that blocks the fallopian tubes.

Remember: You need to use male or female condoms to protect yourself from HIV infection.

If you join the study, we will test you for pregnancy at some visits, including before each study injection.

Appendix C Sample consent form for use of samples and information in other studies

Title: A multicenter, randomized, double-blind, placebo-controlled phase 2b efficacy study of a heterologous prime/boost vaccine regimen of Ad26.Mos4.HIV and aluminum phosphate-adjuvanted Clade C gp140 in preventing HIV-1 infection in women in sub-Saharan Africa

Protocol number: HVTN 705/VAC89220HPX2008

Site: [Insert site name]

When samples are no longer needed for this study, the HVTN wants to keep them for use in other studies. We will call these "extra samples."

This form gives you information so you can decide if you want your extra samples and study-related information used in other studies. You will mark your decision at the end of the form. If you have any questions, please ask.

1. Do I have to agree?

No. You are free to say yes or no, or to change your mind after you sign this form. At your request, we will destroy all extra samples that we have. Your decision will not affect your being in this study or have any negative consequences here.

2. Where are the samples stored?

Extra samples are stored in a secure central place called a repository. *[Site: insert specific information if your regulatory authority requires it.]* Your samples will be stored in the HVTN repository in South Africa.

3. How long will the samples be stored?

There is no limit on how long your extra samples will be stored. *[Site: insert limits if your regulatory authority imposes them.]*

4. Will I be paid for the use of my samples?

No. Also, a researcher may make a new scientific discovery or product based on the use of your samples. If this happens, there is no plan to share any money with you. The researcher is not likely to ever know who you are.

5. Will I benefit from allowing my samples to be used in other studies?

Probably not. Results from these other studies are not given to you, this clinic, or your doctor. They are not part of your medical record. The studies are only being done for research purposes.

6. Will the HVTN sell my samples and information?

No, but the HVTN may share your samples with other researchers. Once we share your samples and information, we will not be able to get them back.

7. How do other researchers get my samples and information?

When a researcher wants to use your samples and/or information, their research plan must be approved by the HVTN. Also, the researcher's institutional review board (IRB) or ethics committee (EC) will review their plan. *[Site: If review by your institution's IRB/EC/RE is also required, insert a sentence stating this.]* IRBs/ECs protect the rights and well-being of people in research. If the research plan is approved, the HVTN will send your samples to the researcher's location.

8. What information is shared with other researchers?

The samples and limited information will be labeled with a code number. Your name will not be part of the information. However, some information that we share may be personal, such as your race, ethnicity, gender, health information from the study, and HIV status. We may share information about the study product you received and how your body responded to the study product.

9. What kind of studies might be done with my extra samples and information?

The studies will be related to HIV, vaccines, the immune system and other diseases.

Researchers may also do genetic testing related to this study on your samples.

Researchers may look at all of the genes of the bacteria found in your vaginal samples. If you become HIV infected, the researchers may look at all of the genes of the virus found in your samples. In both cases, the researchers will use this information to learn more about HIV and the study products.

In some cases, researchers may take cells from your samples and grow more of them over time, so that they can continue to contribute to this study.

If you agree, your samples could also be used for genome wide studies. In these studies, researchers will look at all of your genes (your genome). The researchers compare the genomes of many people, looking for common patterns of genes that could help them understand diseases. The researchers may put the information from the genome-wide studies into a protected database so that other researchers can access it. Usually, no one would be able to look at your genome and link it to you as a person. However, if another database exists that also has information on your genome and your name, someone might be able to compare the databases and identify you. If others found out, it could lead to discrimination or other problems. The risk of this is very small.

10. What are the risks of genetic testing?

The genetic testing could show you may be at risk for certain diseases. If others found out, it could lead to discrimination or other problems. However, it is almost impossible for you or others to know your test results from the genetic testing. The results are not part of your study records and are not given to you.

11. Who will have access to my information in studies using my extra samples?

People who may see your information are:

- Researchers who use your stored samples and limited information for other research

- Government agencies that fund or monitor the research using your samples or information
- The researcher's Institutional Review Board or Ethics Committee
- Any regulatory agency that reviews clinical trials
- The people who work with the researcher

All of these people will do their best to protect your information. The results of any new studies that use your extra samples or information may be published. No publication will use your name or identify you personally.

Questions

12. If you have questions or problems about allowing your samples and information to be used in other studies, use the following important contacts.

If you have questions about the use of your samples or information or if you want to change your mind about their use, contact
[name and telephone number of the investigator or other study staff].

If you think you may have been harmed because of studies using your samples or information, contact [name and telephone number of the investigator or other study staff].

If you have questions about your rights as a research participant, contact
[name/title/phone of person on IRB or other appropriate organization].

Remainder of section for South African sites only.

If you have questions about this trial you should first discuss them with your doctor or the ethics committee (contact details as provided on this form). After you have consulted your doctor or the ethics committee and if they have not provided you with answers to your satisfaction, you should write to the South African Healthcare Products Regulatory Authority (SAHPRA) at:

PPD
South African Health Products Regulatory Authority
Department of Health
Private Bag XPP
PRETORIA
0001

Tel: PPD

e-mail: PPD

Please choose only one of the options below and write your initials or make your mark in the box next to it. Whatever you choose, the HVTN keeps track of your choice about how your samples and information can be used.

☐ I allow my extra samples combined with limited information to be used for other studies related to HIV, vaccines, the immune system, and other diseases. This may include genetic testing and keeping my cells growing over time.

OR

☐ I agree to the option above and also to allow my extra samples combined with limited information to be used in genome wide studies.

OR

☐ I do not allow my extra samples to be used in any other studies. This includes not allowing genetic testing, growing more of my cells, or genome wide studies.

Participant's name (print)	Participant's signature or mark	Date	Time

Clinic staff conducting consent discussion (print)	Clinic staff signature	Date	Time

For participants who are unable to read or write, a witness should complete the signature block below:

Witness's name (print)	Witness's signature	Date	Time

*Witness is impartial and was present for the consent process.

Appendix D Table of procedures (for sample informed consent form)

HIV-uninfected participants

Procedure	Screening visit(s)	First injection visit	Time after 1st injection visits (in months)														
			3	6	6#	7	9	12	13	15	18	21	24	27	30	33	36
Injection		√	√	√				√									
Medical history	√																
Complete physical	√																√
Brief physical		√	√	√	√	√	√	√	√	√	√	√	√	√	√	√	
Blood drawn	√	√	√	√	√	√	√	√	√	√	√	√	√	√	√	√	√
Pregnancy test*	√	√	√	√				√								√	
Vaginal swab(s)		√		√				√			√		√		√		√
Cervical fluid		√		√				√			√		√				
STI testing		√		√				√			√		√		√		√
HIV testing & pretest counseling	√	√	√	√		√	√	√		√	√	√	√	√	√	√	√
Risk reduction counseling	√	√	√	√		√	√	√	√	√	√	√	√	√	√	√	√
Interview/ questionnaire	√	√	√	√		√	√	√	√	√	√	√	√	√	√	√	√

Not shown in this table is a time after all study participants have completed their last scheduled visit when you can find out what products you received.

One day after vaccination visit.

*Persons who had a complete hysterectomy (removal of the uterus verified by medical records) or removal of both ovaries (verified by medical records), are not required to have a pregnancy test.

HIV-uninfected participants: Additional visits

The number of extended follow-up visits for any individual will depend in part on the time of enrollment. For example, if you enrolled around November 2017 then you may have 6 -7 extra visits; if you enrolled around May 2019 you may have 0 or 1 extra visits. We will end the study for all participants at the same time around July 2022.

Procedure	Time after 1st injection visits (in months)						
	39	42	45	48	51	54	57
Brief physical	√	√	√	√	√	√	√
Blood drawn	√	√	√	√	√	√	√
STI testing		√		√		√	
HIV testing & pretest counseling	√	√	√	√	√	√	√
Risk reduction counseling	√	√	√	√	√	√	√
Interview/questionnaire	√	√	√	√	√	√	√

Not shown in this table is a time after all study participants have completed their last scheduled visit when you can find out what products you received.

Appendix D (continued) Table of procedures (for sample informed consent form)

HIV-infected participants

	Time after 1st positive HIV test (in months)		
Procedure	About 2 weeks	3	6
Medical history (as needed)	√	√	√
Brief physical (as needed)	√	√	
Complete physical			√
HIV testing & pretest counseling	√		
Risk reduction counseling	√	√	√
Questions/questionnaire	√	√	√
Blood drawn	√	√	√
Vaginal swab	√		
Pregnancy test*		√	√

Not shown in this table is a time when you can find out what products you received.

*Persons who had a complete hysterectomy (removal of the uterus verified by medical records) or removal of both ovaries (verified by medical records), are not required to have a pregnancy test.

**Once the last HIV uninfected participant completes their Month 36 visit, the study is completed. Redraw visits to confirm HIV infection status may still be performed after the study is complete.

Appendix E Schedule1: Laboratory procedures for HIV-uninfected participants

					Visit	1	2	3	4[1]	5[1]	6[1]	7	8[1]	9[1]	10	11	12	13	14	15	16	17	
					Day		D0	D84	D168	D169	D182-196	D273	D364	D378-394	D455	D546	D637	D728	D819	D910	D1001	D1092	
					Week	Screening visit[3]	W0	W12	W24		W26-28	W39	W52	W54-56	W65	W78	W91	W104	W117	W130	W143	W156	
					Month		M0	M3	M6		M6.5-7	M9	M12	M12.5-13	M15	M18	M21	M24	M27	M30	M33	M36	
							Ad26.Mos4.HIV or Placebo	**Ad26.Mos4.HIV or Placebo**	**Ad26.Mos4.HIV + gp140/Alum or Placebo**				**Ad26.Mos4.HIV + gp140/Alum or Placebo**										
Procedure	**Ship to[1,2]**	**Assay location[2]**	**Tube Type**	**Tube size (vol. capacity)**																			**Total**
BLOOD COLLECTION																							
Screening/Diagnostic																							
Screening HIV test	Local lab	Local lab	SST	5mL		5	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	**5**
HIV diagnostics[8]	HSML-NICD / UW-VSL	HSML-NICD / UW-VSL	EDTA	10mL		—	10	10	10	—	10	10	10	—	10	10	10	10	10	10	10	20[8]	**150**
Safety labs																							
Hemoglobin	Local lab	Local lab	EDTA	5mL		5	—	—	5	—	—	—	5	—	—	—	—	—	—	—	—	—	**15**
STI Serology																							
Syphilis[15]	Local lab	Local lab	SST	5mL		—	5[15]	—	5[15]	—	—	—	5[15]	—	—	5[15]	—	5[15]	—	5[15]	—	5[15]	**35**
ARV detection by serum or plasma[9, 10]	BARC	TBD	SST	8.5mL		—	y	—	y	—	—	—	y	—	—	y	—	y	—	y	—	y	**0**
ARV detection by dried blood spots[11]	BARC	HVTN Labs	EDTA	2mL		—	See footnote 11																**0**
Immunogenicity & virologic assays[5]																							
Host genetics[6]	BARC	HVTN Labs	Na Hep	10mL		—	20	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	**20**
Cellular assays																							
ICS	BARC	HVTN Labs	Na Hep	10mL		—	50	—	—	—	***50***	—	50	***50***	—	50	—	50	—	50	—	50	**400**
IFN-g ELISpot	BARC	HVTN Labs	Na Hep	10mL		—	30	—	—	—	***30***	—	30	***30***	—	30	—	30	—	30	—	30	**240**
Humoral assays																							
Binding Ab	BARC	HVTN Labs	SST	8.5mL		—	8.5	—	—	—	***8.5***	—	8.5	***8.5***	—	8.5	—	8.5	—	8.5	—	8.5	**68**
Neutralizing Ab	BARC	HVTN Labs	SST	8.5mL		—	8.5	—	—	—	***8.5***	—	8.5	***8.5***	—	8.5	—	8.5	—	8.5	—	8.5	**68**
Env-specific ELISA	BARC	Janssen	SST	8.5mL		—	8.5	—	—	—	***8.5***	—	8.5	***8.5***	—	8.5	—	8.5	—	8.5	—	8.5	**68**
Ad26 neutralizing Ab	BARC	Janssen	SST	8.5mL		—	y	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	**0**
Antibody-dependent cellular phagocytos	BARC	HVTN Labs	SST	8.5mL		—	y	—	—	—	***y***	—	y	***y***	—	y	—	y	—	y	—	y	**0**
Innate Immunity																							
RNA gene expression	BARC	HVTN Labs	Tempus	3mL		—	3	—	3	3	—	—	—	—	—	—	—	—	—	—	—	—	**9**
Serum cytokines	BARC	HVTN Labs	SST	8.5mL		—	8.5	—	8.5	8.5	—	—	—	—	—	—	—	—	—	—	—	—	**26**
Transcriptional profiling	BARC	HVTN Labs	Na Hep	10mL		—	40	—	40	40	—	—	—	—	—	—	—	—	—	—	—	—	**120**
Viral isolation/sequencing[9]	BARC	TBD	EDTA	10mL		—	10	10	10	—	10	10	10	20[13]	10	10	10	10	10	10	10	10	**160**
STORAGE																							
PBMC	BARC		Na Hep	10mL		—	100	—	—	—	100	—	50	100	—	50	—	50	—	50	—	50	**550**
Serum	BARC		SST	8.5mL		—	42.5	—	—	—	42.5	—	17	42.5	—	17	—	17	—	17	—	17	**213**
Visit total						**10**	**347**	**22**	**84**	**54**	**270**	**22**	**205**	**270**	**22**	**200**	**22**	**200**	**22**	**200**	**22**	**210**	**2178**
56 Day total[12]						**10**	**357**	**22**	**84**	**137**	**407**	**22**	**205**	**475**	**22**	**200**	**22**	**200**	**22**	**200**	**22**	**210**	
URINE COLLECTION																							
Pregnancy test[7]	Local lab	Local lab				X	X	X	X	—	—	—	X	—	—	—	—	—	—	—	—	X	—	
EITHER URINE OR CERVICAL/VAGINAL SWAB COLLECTION																							
Urine - Chlamydia/Gonorrhea[16]	Local lab	Local lab				—	X[17]	—	X[17]	—	—	—	X[17]	—	—	X[17]	—	X[17]	—	X[17]	—	X[17]	
Cervical/vaginal swab - Chlamydia/Gonorrhea[16]	Local lab	Local lab				—	X[17]	—	X[17]	—	—	—	X[17]	—	—	X[17]	—	X[17]	—	X[17]	—	X[17]	
CERVICAL/VAGINAL SWAB COLLECTION																							
Trichomonas vaginalis[18]	Local lab	Local lab				—	X[18]	—	X[18]	—	—	—	X[18]	—	—	X[18]	—	X[18]	—	X[18]	—	X[18]	
MUCOSAL COLLECTIONS																							
Vaginal swab	BARC	TBD				—	X	—	X	—	—	—	X	—	—	X	—	X	—	—	—	—	
Cervical secretions	BARC	HVTN Labs				—	X	—	X	—	—	—	X	—	—	X	—	X	—	—	—	—	

[1]BARC = Bio Analytical Research Corporation South Africa (Pty) Ltd (Johannesburg, South Africa); HSML-NICD = HIV Sero-Molecular Laboratory-National Institute for Communicable Diseases (Johannesburg, South Africa); UW-VSL = University of Washington Virology Specialty Laboratory (Seattle, Washington, USA)

[2]HVTN Laboratories include: Cape Town HVTN Immunology Laboratory (CHIL, Cape Town, South Africa); South African Immunology, Laboratory–National Institute for Communicable Diseases (SAIL-NICD, Johannesburg, South Africa); Fred Hutchinson Cancer Research Center (FHCRC, Seattle, Washington, USA); Duke University Medical Center (Duke, Durham, North Carolina, USA); University of Cape Town (Cape Town, South Africa), University of Colorado Denver (Aurora, Colorado, USA) Non-HVTN laboratory: Janssen = Janssen Prevention and Vaccines (Leiden, The Netherlands) TBD = Laboratories to be determined by the HVTN laboratory Program

[3]Screening may occur over the course of several contacts/visits up to and including day 0 prior to vaccination

[4]Local labs may assign appropriate alternative tube types for locally performed tests

[5]Immunogenicity assays will be performed at M0 (for binding antibody assay), M7 and M13 Based on the number of responders observed at these timepoints, lab assays may be performed on participants for humoral and cellular responses at other timepoints

[6]Genotyping may be performed on enrolled participants using cryopreserved PBMC collected at baseline, initially in participants who demonstrate vaccine-induced T-cell responses at postvaccination timepoints

[7]Pregnancy test must be performed on the day of vaccination with negative results received prior to vaccination Persons who have undergone total hysterectomy or bilateral oophorectomy (verified by medical records) are not required to undergo pregnancy testing

[8]At an early termination visit for a withdrawn or terminated participant (see Section 9 11), blood should be drawn for HIV diagnostic testing, as shown for visit 17 above
[9]EDTA blood collected for plasma may also be used for ARV detection, if necessary
[10]Testing plan for ARV detection to be determined
[11]EDTA blood for dried blood spots will be collected on regularly scheduled calendar days throughout the study Refer to the Specimen Collection SSP for more information
[12]The total 56-day blood collection includes 2mL EDTA blood collection for dried blood spots It is highly improbably that the 2mL EDTA blood collection will occur at every possible timepoint, but it is reflected in the blood draw totals for every visit to account for this extreme possibility
[13]EDTA blood collected for viral isolation/sequencing may also be used for HIV diagnostics, if necessary
[14]Based on hemoglobin levels at visits 4 and 8, blood draws at visits 5, 6 and 9 may be reduced Refer to the Specimen Collection SSP for more information
[15]Syphilis testing will be done by serology and will be performed at visits 2, 4, 8, 11, 13, 15 and 17; in addition, testing may occur at any other visit if clinically indicated
[16] Chlamydia/gonorrhea testing will be done with either urine or cervical/vaginal swabs Either urine or swab is acceptable, and clinics will determine which specimen will be collected and tested
[17]Chlamydia/gonorrhea testing will be performed at visits 2, 4, 8, 11, 13, 15 and 17; in addition, testing may occur at any other visit if clinically indicated
[18]Cervical/vaginal swab will be collected for Trichomonas testing at visits 2, 4, 8, 11, 13, 15 and 17; in addition, testing may occur at any other visit if clinically indicated
y = SST blood collected for humoral assays and serum storage will also cover specimen needs for ARV detection, Ad26 neutralizing antibody, and antibody-dependent cellular phagocytosis; no separate blood draw is needed

Appendix E (continued) Schedule1: Laboratory procedures for HIV-uninfected participants, extension phase

				Visit	**18**	**19**	**20**	**21**	**22**	**23**	**24**	
				Month	M39	M42	M45	M48	M51	M54	M57	
Procedure	**Ship to**[1,2]	**Assay location**[2]	**Tube Type**[3]	**Tube size (vol. capacity)**[3]								**Total**
BLOOD COLLECTION												
Diagnostic												
HIV diagnostics[8]	HSML-NICD / UW-VSL	HSML-NICD / UW-VSL	EDTA	10mL	10	10	10	10	10	10	10	**90**
Safety labs												
Hemoglobin	Local lab	Local lab	EDTA	5mL	—	—	—	—	—	—	—	**0**
STI Serology												
Syphilis[8]	Local lab	Local lab	SST	5mL	—	5	—	5	—	5	—	**35**
ARV detection by serum or plasma	BARC	TBD	SST	8.5mL		y		y	—	y	—	**0**
ARV detection by dried blood spots[5]	BARC	HVTN Labs	EDTA	2mL	See footnote 5							**0**
Immunogenicity & virologic assays												
Cellular assays												
ICS	BARC	HVTN Labs	Na Hep	10mL	—	50	—	50	—	50	—	**150**
IFN-g ELISpot	BARC	HVTN Labs	Na Hep	10mL	—	30	—	30	—	30	—	**90**
Humoral assays												
Binding Ab	BARC	HVTN Labs	SST	8.5mL	—	8.5	—	8.5	—	8.5	—	**26**
Neutralizing Ab	BARC	HVTN Labs	SST	8.5mL	—	8.5	—	8.5	—	8.5	—	**26**
Env-specific ELISA	BARC	Janssen	SST	8.5mL	—	8.5	—	8.5	—	8.5	—	**26**
Antibody-dependent cellular phagocytos	BARC	HVTN Labs	SST	8.5mL	—	y	—	y	—	y	—	**0**
Viral isolation/sequencing[7]	BARC	TBD	EDTA	10mL	10	10	10	10	10	10	10	**90**
STORAGE												
PBMC	BARC		Na Hep	10mL	—	50	—	50	—	50	—	**150**
Serum	BARC		SST	8.5mL	—	17	—	17	—	17	—	**51**
Visit total					**22**	**200**	**22**	**200**	**22**	**200**	**22**	**687**
56-Day total[6]					**22**	**200**	**22**	**200**	**22**	**200**	**22**	
EITHER URINE OR CERVICAL/VAGINAL SWAB COLLECTION												
Urine - Chlamydia/Gonorrhea[9]	Local lab	Local lab			—	X	—	X	—	X	—	
Cervical/vaginal swab - Chlamydia/Gonorrhea[9]	Local lab	Local lab			—	X	—	X	—	X	—	
CERVICAL/VAGINAL SWAB COLLECTION												
Trichomonas vaginalis[10]	Local lab	Local lab			—	X	—	X	—	X	—	

1BARC = Bio Analytical Research Corporation South Africa (Pty) Ltd (Johannesburg, South Africa); HSML-NICD = HIV Sero-Molecular Laboratory-National Institute for Communicable Diseases (Johannesburg, South Africa); UW-VSL = University of Washington Virology Specialty Laboratory (Seattle, Washington, USA)
2 HVTN Laboratories include: Cape Town HVTN Immunology Laboratory (CHIL, Cape Town, South Africa); South African Immunology Laboratory–National Institute for Communicable Diseases (SAIL-NICD, Johannesburg, South Africa); Fred Hutchinson Cancer Research Center (Seattle, Washington, USA); Duke University Medical Center (Durham, North Carolina, USA); University of Cape Town (Cape Town, South Africa); University of Colorado Denver (Aurora, Colorado, USA) Non-HVTN laboratory: Janssen = Janssen Prevention and Vaccines (Leiden, The Netherlands) TBD = Laboratories to be determined by HVTN Laboratory Program
3 Local labs may assign appropriate alternative tube types for locally performed tests
4 EDTA blood collected for plasma may also be used for ARV detection, if necessary
5EDTA blood for dried blood spots will be collected on regularly scheduled calendar days throughout the study Refer to the Specimen Collection SSP for more information
6The total 56-day blood collection includes 2mL EDTA blood collection for dried blood spots It is highly improbable that the 2mL EDTA blood collection will occur at every possible timepoint, but it is reflected in the blood draw totals for every visit to account for this extreme possibility
7EDTA blood collected for viral isolation/sequencing may also be used for HIV diagnostics, if necessary
8Syphilis testing will be done by serology at visits 19, 21, and 23; in addition, testing may occur at any visit if clinically indicated
9Chlamydia/gonorrhea testing will be performed at visits 19, 21 and 23; in addition, testing may occur at any visit if clinically indicated Chlamydia/gonorrhea testing will be done with either urine or cervical/vaginal swab Either urine or swab is acceptable, and clinics will determine which specimen will be collected and tested
10Cervical/vaginal swab will be collected for Trichomonas testing at visits 19, 21, and 23; in addition, testing may occur at any visit if clinically indicated
y = SST blood collected for humoral assays and serum storage will also cover specimen needs for ARV detection, and antibody-dependent cellular phagocytosis assays; no separate blood draw is needed

Appendix F Schedule 2: Laboratory procedures for HIV-infected participants

Procedure	Ship to[1,2]	Assay location[2]	Tube Type[3]	Tube size (vol. capacity)[3]	Visit: #.X[4,5]		Visit: 31	32	Total
							Weeks after diagnosis[8]: W12	W24	
							Month: M3	M6	
BLOOD COLLECTION									
Screening/Diagnostic									
HIV diagnostics	HSML-NICD / UW-VSL	HSML-NICD / UW-VSL	EDTA	10mL	20[6]		—	—	**20**
HIV PCR viral load	HSML-NICD / UW-VSL	HSML-NICD / UW-VSL	EDTA	10mL	—		10	10	**20**
CD4+T cell count	Local lab	Local lab	EDTA	5mL	5		5	5	**15**
Immunogenicity & virologic assays									
Cellular assays									
ICS	BARC	HVTN Labs	Na Hep	10mL	50		50	50	**150**
IFN-g ELISpot	BARC	HVTN Labs	Na Hep	10mL	20		20	20	**60**
Humoral assays									
Binding Ab	BARC	HVTN Labs	SST	8 5mL	8 5		8 5	8 5	**25.5**
Neutralizing Ab	BARC	HVTN Labs	SST	8 5mL	8 5		8 5	8 5	**25.5**
Env-specific ELISA	BARC	Janssen	SST	8 5mL	y		y	y	**0**
Antibody-dependent cellular phagocytosis	BARC	HVTN Labs	SST	8 5mL	y		y	y	**0**
Viral isolation/sequencing	BARC	TBD	EDTA	10mL	10		10	10	**30**
STORAGE									
PBMC	BARC		Na Hep	10mL	50		50	50	**150**
Plasma	BARC		EDTA	10mL	10		10	10	**30**
Serum	BARC		SST	8 5mL	17		17	17	**51**
Visit total					**199**		**189**	**189**	**577**
56-Day total					**199**		**189**	**189**	
URINE COLLECTION									
Pregnancy Test[9]	Local lab	Local lab			—		X	X	
MUCOSAL COLLECTION									
Vaginal swab	BARC	TBD			X[7]		—	—	

[1]BARC = Bio Analytical Research Corporation, South Africa (Pth) Ltd (Johannesburg, South Africa); HSML-NICD = HIV Sero-Molecular Laboratory-National Institute for Communicable Diseases (Johannesburg, South Africa); UW-VSL = University of Washington Virology Specialty Laboratory (Seattle, Washington, USA)

[2]HVTN Laboratories include: Cape Town HVTN Immunology Laboratory (CHIL, Cape Town, South Africa); South African Immunology, Laboratory–National Institute for Communicable Diseases (SAIL-NICD, Johannesburg, South Africa); Fred Hutchinson Cancer Research Center (FHCRC, Seattle, Washington, USA); Duke University Medical Center (Duke, Durham, North Carolina, USA) Non-HVTN laboratory: Janssen = Janssen Prevention and Vaccines (Leiden, The Netherlands) TBD = Laboratories to be determined by the HVTN laboratory Program

[3]Local labs may assign appropriate alternative tube types for locally performed tests

[4]Visit # X indicates a Redraw visit where # is a Schedule 1 visit number Confirmatory draw for HIV diagnostics will be collected at the Redraw visit The Redraw visit should occur as soon as possible after the clinic receives a Redraw Request from the HIV diagnostics laboratory Multiple subsequent Redraw visits may be necessary; only the EDTA blood specimen for HIV diagnostics, viral isolation/sequencing, and plasma storage will be collected at the subsequent Redraw visits

[5]If the first Redraw visit is preceded by visit 2, 6, 8, or 9, in Schedule 1, the clinic should contact HVTN Lab Ops immediately prior to scheduling the participant for the Redraw visit HVTN Lab Ops will provide guidance on what blood specimens to collect in order to not exceed the 56-day blood volume limit

[6]One tube will remain as a whole blood specimen and be shipped ambient to the HIV diagnostics laboratory; it should not be processed for plasma by the site-associated laboratory The other tube will be processed for plasma as usual

[7]Vaginal swab will only be collected at the first Redraw visit

[8]Date of diagnosis = date of initial specimen draw that led to first Redraw Request

[9]Persons who have undergone total hysterectomy or bilateral oophorectomy (verified by medical records), are not required to undergo pregnancy testing

y = SST blood collected for humoral assays and serum storage will also cover specimen needs for Env-specific binding Ab and antibody-dependent cellular phagocytosis; no separate blood draw is needed

Appendix G Schedule 1: Procedures at HVTN CRS for HIV-uninfected participants

Visit:	01[1]	02[2]	03	04	05[11]	06	07	08	09	10	11	12	13	14	15	16	17	Post
Day:		D0	D84	D168	D169	D196	D273	D364	D394	D455	D546	D637	D728	D819	D910	D1001	D1092	
Month:		M0	M3	M6	M6	M7	M09	M12	M13	M15	M18	M21	M24	M27	M30	M33	M36	
Procedure	Scr.	VAC1	VAC2	VAC3				VAC4										
Study procedures[3]																		
Signed screening consent (if used)	X	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—
Assessment of understanding	X	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—
Signed protocol consent	X	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—
Medical history	X	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—
HIV risk assessment	X	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—
Complete physical exam	X	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	X	—
Abbreviated physical exam	—	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	—	—
Risk reduction counseling	X	X	X	X	—	X	X	X	X	X	X	X	X	X	X	X	X	—
Pregnancy prevention assessment[4]	X	X	X	X	—	X	X	X	X	X	—	—	—	—	—	—	—	—
Behavioral risk assessment questionnaire	—	X	—	—	—	X	—	X	—	—	—	—	X	—	—	—	X	—
Confirm eligibility, obtain demographics, randomize	X	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—
Social impact assessment	—	X	X	X	—	X	X	X	X	X	X	X	X	X	X	X	X	—
Outside testing and belief questionnaire	—	—	—	X	—	—	—	—	—	—	—	—	X	—	—	—	—	—
Concomitant medications	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	—
Intercurrent illness/adverse experience	—	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	—
HIV infection assessment[5]	X	X	X	X	—	X	X	X	—	X	X	X	X	X	X	X	X	—
Confirm HIV test results provided to participant	—	X	X	X	—	X	X	X	X	—	X	X	X	X	X	X	X	X
Local lab assessment																		
Screening HIV test	X	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—
Pregnancy[6]	X	X	X	X	—	—	—	X	—	—	—	—	—	—	—	X	—	—
GC/CT[7]	—	X	—	X	—	—	—	X	—	—	X	—	X	—	X	—	X	—
Syphilis[7]	—	X	—	X	—	—	—	X	—	—	X	—	X	—	X	—	X	—
Trichomonas vaginalis[7]	—	X	—	X	—	—	—	X	—	—	X	—	X	—	X	—	X	—
Cervicovaginal secretion collection[7]	—	X	—	X	—	—	—	X	—	—	X	—	X	—	—	—	—	—
Vaginal swabs[7]	—	X	—	X	—	—	—	X	—	—	X	—	X	—	—	—	—	—
Hemoglobin (Hb)[8]	X	—	—	X	—	—	—	X	—	—	—	—	—	—	—	—	—	—
Vaccination procedures																		
Vaccination[9]	—	X	X	X	—	—	—	X	—	—	—	—	—	—	—	—	—	—
Reactogenicity assessments[10]	—	X	X	X		—	—	X	—	—	—	—	—	—	—	—	—	—
Poststudy																		
Unblind participant	—	—	—	—		—	—	—	—	—	—	—	—	—	—	—	—	X

[1] Screening may occur over the course of several contacts/visits up to and including day 0 prior to vaccination

[2] Specimens collected at the enrollment visit may be obtained within the 14 days prior to vaccination, except for a pregnancy test which must be performed on the day of vaccination with negative results received prior to vaccination

[3] For specimen collection requirements, see Appendix E

[4] Pregnancy prevention assessment is required only for participants who are capable of becoming pregnant

[5] Includes pre-test counseling A subsequent follow-up contact is conducted to provide post-test counseling and to report results to participant

[6] Pregnancy test must be performed on the day of vaccination with negative results received prior to vaccination Pregnancy test to determine initial eligibility may be performed at screening, but must also be done on day 0 prior to first vaccination Persons who have undergone total hysterectomy or bilateral oophorectomy (verified by medical records), are not required to undergo pregnancy testing

[7] Must be performed prior to vaccination

[8] Based on hemoglobin levels at visits 4 and 8, blood draws at visits 5, 6, and 9 may be reduced, respectively Refer to the Specimen Collection SSP for more information

[9] Specimen collection required at vaccination visits must be performed prior to vaccination; however, it is not necessary to have results prior to vaccination, except for results of a pregnancy test, if indicated

[10] Reactogenicity assessments are performed as described in Section 9 9

[11]May be performed offsite

Schedule 1: Procedures at HVTN CRS for HIV-uninfected participants during extension phase

Extension phase visit:	**18**	**19**	**20**	**21**	**22**	**23**	**24**	**Post**
Month:	M39	M42	M45	M48	M51	M54	M57	
Study procedures[1]								
Abbreviated physical exam	X	X	X	X	X	X	X	—
Risk reduction counseling	X	X	X	X	X	X	X	—
Behavioral risk assessment questionnaire	—	—	—	X	—	—	—	—
Social impact assessment	X	X	X	X	X	X	X	—
Concomitant medications	X	X	X	X	X	X	X	—
Intercurrent illness/adverse experience	X	X	X	X	X	X	X	—
HIV infection assessment[2]	X	X	X	X	X	X	X	—
Confirm HIV test results provided to participant	X	X	X	X	X	X	X	X
Local lab assessment								
GC/CT	—	X	—	X	—	X	—	—
Syphilis	—	X	—	X	—	X	—	—
Trichomonas vaginalis	—	X	—	X	—	X	—	—
Poststudy								
Unblind participant	—	—	—	—	—	—	—	X

[1] For specimen collection requirements, see Appendix E, Table for Extension phase.

[2] Includes pre-test counseling. A subsequent follow-up contact is conducted to provide post-test counseling and to report results to participant.

Appendix H Schedule 2: Procedures at HVTN CRS for HIV-infected participants

Visit:	#.X[1]	Visit:	31	32
		Weeks after diagnosis:	W12	W24
		Month:	M3	M6
Study procedures[2]				
Counseling on HIV-1 testing/diagnosis	X		—	—
Abbreviated physical exam	X		X	—
Complete physical exam[3]	—		—	X
ART assessment	X		X	X
Concomitant medications	—		X	X
Intercurrent illness/adverse experience	—		X	X
Transmission risk reduction counseling	X		X	X
Behavioral risk assessment questionnaire	—		—	X
Social impact assessment	X		X	X
Vaginal swab[4]	X		—	—
Pregnancy test[5]	—		X	X

[1] Visit #.X = interim visit for the purpose of drawing samples for confirmatory HIV testing

[2] For specimen collection requirements, see Appendix F.

[3] Includes assessment of HIV/AIDS-related conditions.

[4] Vaginal swab collected at first Redraw visit only.

[5] Persons who have undergone total hysterectomy or bilateral oophorectomy (verified by medical records), are not required to undergo pregnancy testing.

Appendix I Adverse events of special interest

AEs of special interest (AESI) for this protocol include but are not limited to potential immune-mediated diseases; representative examples of AESI are listed below. Updates to AESI will be provided as an appendix to the *HVTN 705/HPX2008 Study Specific Procedures.*

Neuroinflammatory disorders	Musculoskeletal disorders	Skin disorders
• Cranial nerve disorders, including paralyses/paresis (eg Bell's palsy) • Optic neuritis • Multiple sclerosis • Transverse myelitis • Guillain-Barré syndrome, including Miller Fisher syndrome and other variants • Acute disseminated encephalomyelitis, including site specific variants: eg non-infectious encephalitis, encephalomyelitis, myelitis, myeloradiculoneuritis • Myasthenia gravis, including Lambert-Eaton myasthenic syndrome • Immune-mediated peripheral neuropathies and plexopathies, (including chronic inflammatory demyelinating polyneuropathy, multifocal motor neuropathy and polyneuropathies associated with monoclonal gammopathy). • Narcolepsy	• Systemic lupus erythematosus and associated conditions • Systemic scleroderma (Systemic sclerosis), including diffuse systemic form and CREST syndrome • Idiopathic inflammatory myopathies, including dermatomyositis • Polymyositis • Antisynthetase syndrome • Rheumatoid arthritis, and associated conditions including juvenile chronic arthritis and Still's disease • Polymyalgia rheumatica • Spondyloarthritis, including ankylosing spondylitis, reactive arthritis (Reiter's Syndrome) and undifferentiated spondyloarthritis • Psoriatic arthropathy • Relapsing polychondritis • Mixed connective tissue disorder	• Psoriasis • Vitiligo • Erythema nodosum • Autoimmune bullous skin diseases (including pemphigus, pemphigoid and dermatitis herpetiformis) • Alopecia areata • Lichen planus • Sweet's syndrome • Localized Scleroderma (Morphea) • Cutaneous lupus erythematosus **Metabolic disorders** • Addison's disease • Autoimmune thyroiditis (including Hashimoto thyroiditis) • Diabetes mellitus type I • Grave's or Basedow's disease
Vasculitides	**Blood disorders**	**Others**
• Large vessels vasculitis including: giant cell arteritis such as Takayasu's arteritis and temporal arteritis. • Medium sized and/or small vessels vasculitis including: polyarteritis nodosa, Kawasaki's disease, microscopic polyangiitis, Wegener's granulomatosis, Churg-Strauss syndrome (allergic granulomatous angiitis), Buerger's disease (thromboangiitis obliterans), necrotizing vasculitis and anti-neutrophil cytoplasmic antibody (ANCA) positive vasculitis (type unspecified), Henoch-Schonlein purpura, Behcet's syndrome, leukocytoclastic vasculitis.	• Autoimmune hemolytic anemia • Autoimmune thrombocytopenia • Antiphospholipid syndrome • Pernicious anemia • Autoimmune aplastic anemia • Autoimmune neutropenia • Autoimmune pancytopenia **Gastrointestinal disorders** • Celiac disease • Crohn's disease • Ulcerative colitis • Ulcerative proctitis **Liver disorders** • Autoimmune cholangitis • Autoimmune hepatitis • Primary biliary cirrhosis • Primary sclerosing cholangitis	• Autoimmune glomerulonephritis (including IgA nephropathy, glomerulonephritis rapidly progressive, membranous glomerulonephritis, membranoproliferative glomerulonephritis, and mesangioproliferative glomerulonephritis) • Ocular autoimmune diseases (including autoimmune uveitis and autoimmune retinopathy) • Autoimmune myocarditis/cardiomyopathy • Sarcoidosis • Stevens-Johnson syndrome • Sjögren's syndrome • Idiopathic pulmonary fibrosis • Goodpasture syndrome • Raynaud's phenomenon

Appendix J Injection Schedule for sample informed consent