CHAVI 001

ACUTE HIV-1 INFECTION PROSPECTIVE COHORT STUDY

A Study of the Center for HIV/AIDS Vaccine Immunology (CHAVI)

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Protocol Chair:

Myron S. Cohen

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PROTOCOL TEAM ROSTER

Sarah Chiduo, A.M.O., M.A.
Kilimanjaro Christian Medical Centre
Department of Community Health
P.O. Box 3010
Moshi
Tanzania
sarahchiduo@kilinet.co.tz
T: 255-744-434355
F: 255-272-754381

Gavin Churchyard, MBBCh, FCP (SA), M.Med., Ph.D.
Aurum Institute for Health Research
6th Floor, 47 Main Street
Ferreirasdorp, Johannesburg, 2001, South Africa
Postal Address: PO Box 61587, Marshalltown, 2107, South Africa
gchurch@mjvn.co.za
T: 27-11-638-2253
F: 27-11-638-2179

Myron S. Cohen, M.D.
J. Herbert Bate Professor
Medicine, Microbiology and Immunology and Public Health Chief, Division of Infectious Diseases Director, UNC Center for Infectious Diseases
Bioinformatics Building
130 Mason Farm Road
Chapel Hill, NC 27599-7030
T: 919-966-2536
F: 919-966-6714
mscohen@med.unc.edu

Joseph Eron, M.D.
Professor of Medicine
University of North Carolina
Division of Infectious Disease
Department of Medicine
547 Burnett Womack
Chapel Hill, NC 27599-7030
jeron@med.unc.edu
T: 919-966-2536
F: 919-966-6714

Guido Ferrari, Ph.D.
Research Associate Professor
Duke University
Medicine
115 SORF Building, Research Dr Box 2926
Durham, North Carolina 27710
gflmp@duke.edu
T: 919-684-2862
F: 919-684-4288

Susan A. Fiscus, Ph.D.
Professor
University of North Carolina School of Medicine
Department of Microbiology & Immunology
709 Mary Ellen Jones Building
Box # CB#7290
Chapel Hill NC 27599-7290
fiscussa@med.unc.edu
T: 919-966-6872
F: 919-966-9873

Shannon Galvin, M.D.
Assistant Professor of Medicine
Division of Infectious Diseases
University of North Carolina School of Medicine
130 Mason Farm Road CB# 7030
Chapel Hill, NC 27599 USA
shannon_galvin@med.unc.edu
T: 919- 843-0025
F: 919- 966-6714

Peter Gilbert, Ph.D.
Faculty Statistician
Statistical Center for HIV/AIDS Research & Prevention (SCHARP) Fred Hutchinson Cancer Research Center
1100 Fairview Ave. N., LE-400
Seattle, WA 98109-1024
pgilbert@scharp.org
T: 206-667-7299
F: 206-667-4812

Clive Gray, Ph.D.
Chief Specialist Scientist and Head of HIV Immunology
National Institute for Communicable Diseases
Private Bag X4
Sandringham 2131
Johannesburg
South Africa
cliveg@nicd.ac.za
T: 27 11 386 6372
F: 27 11 386 6310

Barton F. Haynes, M.D.
Director, Duke Human Vaccine Institute
Research Drive
224 CARL Bldg.
Box 3258
Duke University Medical Center
Durham, NC 27710
hayne002@mc.duke.edu
T: 919-684-5279
F: 919-684-5230

Charles Hicks, M.D.
Associate Professor of Medicine
Room 34225 - Duke South Red Zone
Duke University Medical Center
Durham, NC 27710
charles.hicks@duke.edu
T: 919-684-4474
F: 919-681-7494

Irving Hoffman, P.A., M.P.H.
Center for Infectious Diseases
University of North Carolina
School of Medicine
1700 Airport Road
CB # 3368
Chapel Hill, NC 27599 USA
Hoffmani@med.unc.edu
T: 919-966-6324
F: 919-966-8536

Pontiano Kaleebu, MD PhD
MRC/UVRI Uganda Research Unit on AIDS
Medical Research Council
Uganda Virus Research Institute
P. O Box 49
Entebbe Uganda
pontiano.kaleebu@mrcuganda.org
T: 256- 41-320042/320272
F: 256-41 321137

Saidi H. Kapiga, M.D., M.P.H., Sc.D.
Harvard School of Public Health
Department of Population and International Health
665 Huntington Avenue, Bldg 1-1105
Boston, MA 02115
USA
skapiga@hsph.harvard.edu
T: 617-432-1237
F: 617-566-0365

Salim Abdool Karim, MBChB, Ph.D.
CAPRISA
University of KwaZulu-Natal
King George V Avenue
Durban 4041, South Africa
karims1@ukzn.ac.za
T: 27-31-2602381
F: 27-31-2602384

Johnstone Kumwenda, MBChB, MSc, MRCP
College of Medicine-Johns Hopkins Project
P.O. Box 1131 Chipatala Avenue
Blantrye, Malawi
jonnykumwenda@yahoo.com
T: 265-884-1875
F: 265-167-0132

Marybeth McCauley, M.P.H.
Senior Clinical Research Manager
Family Health International
2101 Wilson Boulevard, Suite 700
Arlington, VA 22201
mmccauley@fhi.org
T: 703-516-9779
F: 703-516-0295

Newton Kumwenda, M.P.H., Ph.D.
Field Director
Johns Hopkins Project
P.O. Box 1131
Blantyre, MALAWI
jhopkins@sdnp.org.mw
T: 011-265-631-527
F: 011-265-631-527

Peter Leone, M.D.
Division of Infectious Diseases
University of North Carolina, Chapel Hill CB# 7030 BioInformatics
130 Mason Farm Road
University of North Carolina
Chapel Hill, NC
27599-7030
peter_leone@med.unc.edu
T : 919-966-2536
F: 919-966-6714

Francis Martinson, M.D.
Project Director
UNC Project Private Bag A-104
Lilongwe, MALAWI
fmartinson@malawi.net
T: 011-265-755-056
F: 011-265-755-954

Helen Rees, MBBCHIR MA (CANTAB) MRCGP
Reproductive Health and HIV Research Unit
Department of Obstetrics and Gynaecology
University of the Witwatersrand
Chris Hani Baragwanath Hospital, Soweto
P.O.Bertsham, 2013
Johannesburg, South Africa
h.rees@rhrujhb.co.za
T: 27 11 989 – 9208
F: 27 11 989- 9271

Noel E. Sam, M.D., M.Med.
Kilimanjaro Christian Medical Centre
Department of Clinical Laboratories
P.O. Box 3010
Moshi
Tanzania
noelsam@kcmc.ac.tz
T: 255-744-372211
F: 255-272-754777

Paul Sato, M.D., M.P.H.
Medical Officer
Division of AIDS/NIAID/NIH
6700B Rockledge Drive, Rm 5248
Bethesda, MD 20892-7628 USA
psato@niaid.nih.gov
T: 301-896-4068
F: 301-402-3684

Steve G. Self, Ph.D.
Executive Director
Statistical Center for HIV/AIDS Research & Prevention (SCHARP) Fred Hutchinson Cancer Research Center
1100 Fairview Ave. N., LE-400
Seattle, WA 98109-1024
sgs@scharp.org
T: 206-667-4944
F: 206-667-4812

Jennifer Serwanga, BVM PhD
MRC/UVRI Uganda Research Unit on AIDS
Medical Research Council
Uganda Virus Research Institute
P. O Box 49
Entebbe Uganda
Jennifer.serwanga@mrcuganda.org
T: 256- 41-320042/320272
F: 256-41 321137

Stuart Shapiro, M.D., Ph.D.
DAIDS Program Officer
Division of AIDS/NIAID/NIH 6700B
Rockledge Drive, Rm 5146
Bethesda, MD 20892-7628 USA
sshapiro@niaid.nih.gov
T: 301-402-0122
F: 301-402-3684

Wendy Stevens. M.D.
Professor and Head
Department of Molecular Medicine and
Haematology University of
Witwatersrand Johannesburg South Africa
wendy.stevens@nhls.ac.za, or
wendy@dlatech.co.za
T: 27-11-489-8505
F: 27-11-484-5812

Taha Taha, M.D.
Johns Hopkins Bloomberg School Public Health
615 North Wolfe Street, Room E-6011
Baltimore, MD 21205 USA
ttaha@jhsph.edu
T: 410-614-5255
F: 410-955-1836

ABBREVIATIONS AND ACRONYMS

AIDS	Acquired Immunodeficiency Syndrome
CDC	US Centers for Disease Control and Prevention
CHAVI	Center for HIV/AIDS Vaccine Immunology
CRF	case report form
CTL	cytotoxic T-lymphocytes
DAIDS	Division of AIDS
DC	dendritic cells
EC	Ethical Committee
EIA	enzyme immunoassay
GCP	Good Clinical Practice
GEE	generalized estimating equations
HIV	Human Immunodeficiency Virus
HLA	human lymphocyte antigen
IATA	International Air Transport Association
ICF	informed consent form
IRB	Institutional Review Board
m	myeloid
ml	milliliter
MPER	membrane proximal external region
NAT	nucleic acid test
NC	North Carolina
NIAID	(United States) National Institute of Allergy and Infectious Diseases
NIH	(United States) National Institutes of Health
NK	natural killer
p	plasmacytoid
PBMC	peripheral blood mononuclear cell
PCR	polymerase chain reaction
RNA	ribonucleic acid
SIV	simian immunodeficiency virus
SLG	(CHAVI) Scientific Leadership Group
SNP	single-nucleotide polymorphism
SOP	standard operating procedure
SSP	Study-Specific Procedures (Manual)
STARHS	Serologic Testing Algorithm for Recent HIV Seroconversions
STD	sexually transmitted disease
STI	sexually transmitted infection
TLR	toll like receptors
UNC	University of North Carolina
US	United States
USA	United States of America
VCT	voluntary HIV counseling and testing
WB	Western blot

ACUTE HIV-1 INFECTION PROSPECTIVE COHORT STUDY

SCHEMA

Purpose: To collect biological specimens to study the HIV-1 virus, the host response, the genetic factors that determine HIV transmission and viral set point.

Design: Multi-center, prospective, observational, cohort study

Study Population and Size: Up to approximately 1300 participants who are 18 years or older will be enrolled into this study. Eligible participants will be placed into one of the following four study groups:

Group 1: Proven acute HIV-1 infection: approximately 600 total

Group 1A: Sexual partners of members of Group 1: approximately 600 total

Group 2: Established HIV-1 infection: approximately 50 total

Group 3: HIV-1 negative: approximately 50 total

Study Duration: Approximately 84 months, with accrual requiring approximately 60 months, and with each participant followed for up to approximately 24 months.

Primary Objectives:

- To determine the prevalence of acute HIV infection at the participating sites.
- To determine the characteristics of the transmitted virus in acute HIV-1 infection.
- To determine the host acquired and innate immunes responses that contribute to virus control and/or protection against infection with HIV-1.
- To determine the genetic factors that contributes to early virus control and/or protection from infection with HIV-1.
- To examine the clinical, laboratory, and behavioral characteristics of individuals at diverse sites with acute HIV infection and their sexual partners, and appropriate controls.

SCHEMA (continued)

Study Sites: The study will be conducted at clinical sites located in the following cities and countries:

Blantyre and Lilongwe, Malawi

Durban, Klerksdorp and Johannesburg, South Africa

Moshi and Arusha, Tanzania

Kampala, Uganda

Chapel Hill and Durham, North Carolina, USA

Other sites may be added as necessary.

1.0 BACKGROUND AND RATIONALE

Now fully twenty five years after the beginning of the AIDS pandemic, enormous gaps in knowledge limit both prevention and treatment. Understanding HIV transmission has been difficult in the absence of identifying large numbers of transmission events and collecting specimens in the first days and weeks of infection.

In order to fill some of the critical gaps in knowledge, the US National Institutes of Health (NIH) has awarded a large grant to a group of investigators to implement The Center for HIV/AIDS Vaccine Immunology (CHAVI), as a US Government contribution to the Global HIV-1 Enterprise. CHAVI investigators are comprised of clinical and laboratory investigators located throughout the world. The clinical site investigators will lead the effort in implementing prospective observational studies, and the laboratory investigators will study the specimens obtained from the observational studies using state-of-the art technology through carefully designed, focused, and coordinated laboratory studies to address Enterprise-identified gaps in our knowledge targeted at enabling the production of a successful HIV-1 vaccine. To do this CHAVI will work on the discovery of new information about acute HIV-1 infection, the correlates of protective immunity to HIV-1, and the development of novel methods of inducing protective immunity at mucosal sites.

1.1 Acute HIV Infection

During acute HIV infection, the individual has not yet mounted an immune response detectable by tests traditionally used to screen for HIV infection. While seroconversion has been shown to occur as late as 6 months post exposure, detectable levels of viremia and p24 antigenemia develop over the first three to four weeks of infection (Busch & Satten. 1997; Lindback, Thorstensson, Karlsson, et al. 2000). Several studies suggest that individuals with acute HIV infection can be identified in STD clinics and perhaps other high risk settings. Bollinger et al. enrolled 6495 consecutive attendees to two sexually transmitted infection (STI) clinics in Pune, India (Bollinger, Brookmeyer, Mehendale, et al. 1997). Of 5033 HIV negative samples, 3874 were tested for p24 antigen and 58 (1.5%) were found to be positive. These positive p24 antigen results were later confirmed by HIV seroconversion. The majority of these acute infections (51, 88%) were identified in men. Two hundred and ninety controls were matched by age and gender and compared for clinical signs of acute HIV infection. The most common symptoms associated with p24 antigenemia were fever, joint pain, inguinal lymphadenopathy, and night sweats with at least one of these symptoms being found in 47% of those reporting with acute HIV infection.

Based on these observations researchers at the University of North Carolina (UNC) (Chapel Hill, North Carolina [NC]) have developed rapid and simple strategies that facilitate the large-scale identification of acute HIV infections (Pilcher, McPherson, Leone, et al. 2002). This system was initiated in a statewide public testing system for identification of both acute and established HIV infections in collaboration with the NC Department of Health and the NC State Laboratory of Public Health. In the first 12 months of this enhanced testing, 109,250 patients at risk of HIV were tested at 110 sites

located in antenatal clinics, STD clinics, family planning clinics, jail sites, and in non-traditional venues statewide. Of 606 new infections identified, 583 were antibody positive but an additional 23 cases (4% of all HIV infections identified in the state system) were antibody negative acute infections confirmed at follow-up in year one (Pilcher, 2005). These subjects could only be identified by HIV RNA testing. 107 of the antibody positives were also identified as "recent" by the Serologic Testing Algorithm for Recent HIV Seroconversions (STARHS) algorithm, which uses detection of low-titer antibodies to define recent infection. Two-thirds of the acute patients (16 of 23) were VCT clients that attended STD clinics. All clients identified by this mechanism are referred directly for evaluation at UNC and Duke Acute HIV Program sites as part of NC's program for notification and counseling (called the Screening and Tracing Active Transmission, or STAT Program). Partner notification and testing of the first 23 antibody negative acute clients by the STAT rapid public health response team resulted in identification of 18 HIV+ partners (5 new infections, 11 likely transmitters, and 2 acute-to-acute transmissions) and counseling of 48 high-risk HIV- sexual partners. The North Carolina Program will serve as the model and flagship for the CHAVI acute HIV infection Program.

Similar methods have been implemented in Africa in clinics that will form the second critical network for the goals of the CHAVI. In a study of 1,361 men visiting an STD clinic in Lilongwe, Malawi (Pilcher, Price, Hoffman, et al. 2004), a 2.1% prevalence of acute HIV infection was detected. Among men presenting with a new genital ulcer and adenopathy, 11% had acute HIV infection. Despite this diagnosis, only 46% of these men had signs or symptoms consistent with acute HIV infection, either because they were discovered so early in infection (before symptoms developed), or because symptoms are truly rare in the general population. The median blood viral burden in these subjects was more than 1,000,000 copies/ml. Similar results were found in a more recent study of 1441 men and women in the same clinic (Fiscus, Pilcher, Miller, et al. 2005).

In anticipation of formation of the CHAVI Acute HIV infection Program, a preliminary study has been conducted at the Hillbrow Esselen Street STD Clinic in Johannesberg, South Africa (Stevens, Akkers, Myers, Motloung, Pilcher, & Venter, 2005.). 1418 consecutive clinic attendees (47% female) were recruited April-October 2004. As in the Malawi program, HIV antibody testing, specimen pooling, and HIV RNA determinations were performed locally. Of those tested, 572 individuals were HIV antibody positive (40.3%) and 8 individuals (1% of antibody negatives) were HIV antibody negative and HIV RNA positive, indicative of acute HIV infection.

1.2 Acute HIV Infection and Vaccine Development

In recent years a remarkable effort has been made to understand HIV transmission and the evolving immune response. However, this work and the interpretation of results have suffered from lack of sufficient study subjects, insufficient number of specimens for study, and limited numbers of HIV transmission pairs. A key strength of CHAVI will be the coordinated recruitment of subjects and their sexual partners, as well as specimen collection, to generate novel, more reliable, and reproducible results. To understand HIV transmission, viral isolates harvested from index cases and their sexual partners at the

earliest point after transmission possible are needed. In addition, semen and female mucosal secretions are critical to determine whether blood specimens are a suitable surrogate for mucosal events, and to determine the importance of compartmentalization on the HIV transmission event(s).

These specimens will be used to study the virus, the host response, and genetic factors that determine HIV transmission and viral set point.

1.3 The Virus and the Host Response

Numerous studies have examined the complexity of the virus population during acute or early infection. Some investigators have reported a homogeneous virus population (Pang, Shlesinger, Daar, Moudgil, Ho, & Chen. 1992; Wolfs, Zwart, Bakker, & Goudsmit. 1992; Mulder-Kampinga, Kuiken, Dekker, Scherpbier, Boer, & Goudsmit. 1993; Zhu, Mo, Wang, et al. 1993; Delwart, Sheppard, Walker, Goudsmit, & Mullins. 1994; Furuta, Bergstrom, Norkrans, & Horal. 1994; Kliks, Contag, Corliss, et al. 2000; Shankarappa, Margolick, Gange, et al. 1999) and others a more heterogeneous population (Lamers, Sleasman, She, et al. 1993; Scarlatti, Leitner, Halapi, et al. 1993; Wolinsky, Korber, Neumann, et al. 1996; Zhu, Wang, Carr, et al. 1996; Delwart, Pan, Sheppard, et al. 1997; Kampinga, Simonon, Van de Perre, Karita, Msellati, & Goudsmit. 1997; Liu, Schacker, Musey, et al. 1997; Sutthent, Foongladda, Chearskul, et al. 1998; Dickover, Garratty, Plaeger, & Bryson. 2001; Learn, Muthui, Brodie, et al. 2002; Machado, Delwart, Diaz, et al. 2002; Renjifo, Chung, Gilbert, et al. 2003; Verhofstede, Demecheleer, De Cabooter, et al. 2003). The basis for discrepancies in these reports is not known, but has generally been ascribed to substantial differences in study populations, timing and type(s) of specimen collected, and differences in laboratory methodology.

Poss and colleagues have reported that the sex of the infected person is a determinant of the complexity of the transmitted virus with women frequently having multiple variants and men usually having a homogeneous population (Poss, Martin, Kreiss, et al. 1995; Long, Martin, Kreiss, et al. 2000). This gender difference can in part be explained by different regions of the viral genome that were analyzed, the different modes of transmission among populations, the different definitions of primary infection that were used, and the different criteria by which viral populations were categorized as homogeneous versus heterogeneous.

Control of viral replication and phenotypic evolution of the virus must to some extent reflect interaction with the individual host. The effect of cytotoxic T-lymphocyte (CTL) selection in resolving the initial viremia can be seen in the appearance of early CTL escape mutants. O'Connor and Watkins have use whole genome sequencing of SIVmac239-infected macaques to detect CTL escape mutants as early as 4-8 weeks after infection (O'Connor, Allen, Vogel, et al. 2002). The early evolution of sequences at these sites must represent strong selective pressure due to the CTL response. Conversely, the absence of evolution at other putative sites must be equated with little or no selective pressure.

Several studies have documented the presence of a persistent CTL response in infected people, albeit with CD8+ cells with lower avidity. Large scale screening of such responses and/or viral polymorphisms in high incidence areas are starting to provide a picture of the most frequent targets (Moore, John, James, Christiansen, Witt, & Mallal. 2002; Novitsky, Cao, Rybak, et al. 2002; Yusim, Kesmir, Gaschen, et al. 2002; Frahm, Korber, Adams, et al. 2004; Leslie, Pfafferott, Chetty, et al. 2004; Masemola, Mashishi, Khoury, et al. 2004). The early CTL response may differ from the response measured later (Cao, McNevin, Holte, Fink, Corey, & McElrath. 2003), and these early epitopes may play an important role in slowing disease progression (O'Connor, Mothe, Weinfurter, et al. 2003). Knowledge of the human lymphocyte antigen (HLA) background also permits an assessment of sequence changes that are likely to be observed after transmission (Peyerl, Barouch, Yeh, et al. 2003; Friedrich, Dodds, Yant, et al. 2004; Leslie, Pfafferott, Chetty, et al. 2004), and the temporal appearance of mutations can suggest either different levels of selective pressure or the addition of compensatory mutations to restore fitness (Geels, Cornelissen, Schuitemaker, et al. 2003; Jamieson, Yang, Hultin, et al. 2003; Peyerl, Barouch, Yeh, et al. 2003; Friedrich, Frye, Yant, et al. 2004). Swanstrom et al. (personal communication) found only a single coding substitution when approximately one fourth of the viral proteome (including most of gag) was queried in comparing a week two time point with a four month time point. Thus the selective pressure may be exerted at a few number of sites, although in the Cao study (Cao, McNevin, Holte, Fink, Corey, and McElrath. 2003) gag was not a strong early target of CTL.

Characteristically, the humoral response results in the appearance of neutralizing antibodies. The first HIV-1 specific immune response is formation of non-neutralizing antibodies detected by ELISA 2-4 weeks after infection (Fiebig, Wright, Rawal, et al. 2003), which serves as the basis for the diagnosis. Neutralizing antibodies are detected 8 weeks or more after infection (Rybarczyk, Montefiori, Johnson, West, Johnston, & Swanstrom. 2004); (Wei, Decker, Wang, et al. 2003; Richman, Wrin, Little, & Petropoulos. 2003). Neutralizing antibodies seem to place strong immune pressure on the virus since escape variants appear relatively early and are followed by an explosion of viral diversity (Rybarczyk, Montefiori, Johnson, West, Johnston, & Swanstrom. 2004). Neutralizing antibodies are directed at the glycosylated surface envelope proteins (Rybarczyk, Montefiori, Johnson, West, Johnston, & Swanstrom. 2004; Wei, Decker, Wang, et al. 2003) and sequence change(s) in the highly variable surface loops (with addition or movement of carbohydrate side chains) is the primary mechanism of HIV escape.

There is a maturation of this response over the first year (Cole, Murphey-Corb, Narayan, Joag, Shaw, & Montelaro. 1998; Cole, Paliotti, Murphey-Corb, & Montelaro. 2000). However, the earliest effects of this response can be measured in the activity of neutralizing antibodies and in sequence changes in the env gene sequence. Richman and colleagues (Richman, Wrin, Little, & Petropoulos. 2003) have employed assays using full length env genes pseudotyping reporter viruses to demonstrate an early response to the autologous virus followed by successive waves of virus escape. Shaw and colleagues (Wei, Decker, Wang, et al. 2003) have used similar assays to show that the movement of carbohydrate side chains on the surface of gp120 can lead to neutralization escape. The

SIV-macaque model system and V1/V2 heteroduplex tracking assay have been used to determine the timing of the diversification of the env gene (Rybarczyk, Montefiori, Johnson, West, Johnston, & Swanstrom. 2004). Rybarcyzk et al. found a correlation between the timing of env diversification and the appearance of neutralizing antibodies, and found a lack of env diversification in those monkeys that failed to mount an immune response. The timing of the diversification differed by route of infection, with earlier env changes seen after IV infection and later with an IR/mucosal challenge (even though there was no difference in the timing or peak of viremia). No changes in the env gene population between week 2 and weeks 8-12 were observed, indicating that efforts to identify primary infection within the first 2 months should yield information about the complexity of the inoculum prior to the effects of antibody selection. However, the potential for bias in samples tested longitudinally for seroconversion is clear, and any delay in subject identification will yield artifactually heterogeneous populations whose complexity is unrelated to the complexity of the transmitted inoculum. The initial sequence changes found with the diversification of env by Rybarcyzk were largely in V1 and not V2, and did not involve changes in glycosylation sites. Frost and Richman (Frost, Liu, Pond, et al. 2005) did not see changes in glycosylation in env in two subjects. There is a clear need for a much larger sample size to determine both the range of escape pathways and the frequency with which the different mechanisms are used.

Several other models for early sequence evolution have been proposed. An early report indicated that the gag gene was more diverse than the env gene (Zhu, Mo, Wang, et al. 1993), suggesting an early bottleneck in env after infection, something not seen in monkeys (Rybarczyk, Montefiori, Johnson, West, Johnston, & Swanstrom. 2004). This suggestion was supported by an observation that early diversity in env was followed by a period of homogenization (Learn, Muthui, Brodie, et al. 2002), again quite different from what was observed in the SIV model where the env gene population was extremely stable until the appearance of neutralizing antibody.

Recently an even more provocative model for HIV transmission has been proposed (Derdeyn, Decker, Bibollet-Ruche, et al. 2004). Derdeyn et al. presented evidence from eight subjects suggesting that neutralization sensitive viruses with short variable loops are selectively transmitted during heterosexual transmission of subtype C HIV-1 (male and female). These results however have not been reproduced by other investigators to date.

1.4 The Antibody Immune Response in Greater Detail

Why are broadly reactive neutralizing antibodies not made in acute or early infection, why are they rarely made in chronic disease, and why are they not made in response to vaccination with HIV-1 envelope?

The majority of studies focused on these questions have targeted the viral envelope and not the host immune response. Autologous, strain-specific neutralizing antibodies are routinely made early in primary infection. They generally target exposed variable loop epitopes including those present on V1, V2, V3, and possibly V4, and virus escape from neutralization is rapid (Wei, Decker, Wang, et al. 2003; Richman, Wrin, Little, & Petropoulos. 2003). Antibody responses to CD4+ or co-receptor binding surfaces have

been documented, but except for the CD4bs mAb IgG1b12, such antibodies generally have weak neutralizing potency (Burton, Desrosiers, Doms, et al. 2004).

The defined epitopes on HIV-1 envelope to which rare broadly reactive neutralizing antibodies bind are thus the CD4+ binding site (CD4BS) (mAb IgG1b12)(Zwick, Parren, Saphire, et al. 2003)and the membrane proximal external region (MPER) epitopes defined by human mAbs 2F5 and 4E10 (Stiegler & Katinger. 2003; Armbruster, Stiegler, Vcelar, et al. 2004; Zwick, Jensen, Church, et al. 2005) and the glycan epitope defined by mAb 2G12 (Scanlan, Pantophlet, Wormald, et al. 2003). These mAbs are all unusual: two are IgG3 (2F5 and 4E10), one has a unique Ig dimer structure (2G12), and one has a very hydrophobic CDR3 (2F5). Moreover, all four have unusually long CDR3 regions (Burton, Desrosiers, Doms, et al. 2004; Kunert, Wolbank, Stiegler, Weik, & Katinger. 2004; Zwick, Komori, Stanfield, et al. 2004), and three of the four mAbs (2F5, 4E10 and IgG1b12) have recently been found to be autoreactive (Haynes, Fleming, St Clair, et al. 2005).

Recent data demonstrate that mAbs 2F5, 4E10 and IgG1b12 are indeed polyspecific, autoreactive Abs that bind with high affinity to multiple human autoantigens (Haynes, Fleming, St Clair, et al. 2005). These data thus suggest a new explanation and paradigm for understanding the ineffective host neutralizing antibody response to HIV-1 in both acute infection and in normal subjects following vaccination with HIV-1 envelope. Thus, HIV-1 may have evolved to escape antibody responses by having conserved neutralizing epitopes as mimics of autoantibody epitopes.

These data suggest the hypothesis that acute HIV infection and current HIV-1 vaccines do not routinely induce robust anti-envelope neutralizing antibodies because antibodies targeting conserved epitopes are derived from autoreactive B cell clones that are normally deleted or made tolerant upon antigenic stimulation by HIV-1 env. Thus, by carefully defining the ontogeny and regulation of anti-MPER and other broadly neutralizing antibody-producing B cells, cloning and sequencing acute HIV infection antibodies and their ancestral immunoglobulin genes, and making human monoclonal antibodies against the repertoire of neutralizing determinants on the transmitted envelope, future studies hold promise for designing strategies for eliciting durable systemic mucosal neutralizing antibodies against a broad spectrum of HIV primary isolates.

In addition, George Shaw and colleagues at the University of Alabama have recently demonstrated broad neutralizing antibodies in the sera of subjects that react with epitopes other than the 2F5 and 4E10 epitopes of gp41 (unpublished observations). Moreover, Gorny et al. have demonstrated that human monoclonal antibodies from non-B clade infected subjects have broader neutralizing capacity than anti-V3 monoclonal antibodies derived from clade B infected patients (unpublished observations). Taken together, these data demonstrate that a comprehensive survey of the types and specifics of both polyreactive and monospecific antibodies produced in acute HIV infection is required for analysis of antibody types systemically and at mucosal sites.

1.5 Cell Mediated Immunity in Greater Detail

Retrovirus specific cytotoxic CD8+ T lymphocytes (CTL) detected early after infection are strongly associated with the control of viremia (Borrow, Lewicki, Hahn, Shaw, & Oldstone. 1994; Koup, Safrit, Cao, et al. 1994; Schmitz, Kuroda, Santra, et al. 1999). The initial response(s) may be directed against a few epitopes, and subsequently broaden during prolonged antigen stimulation (Yu, Addo, Rosenberg, et al. 2002; Goulder, Altfeld, Rosenberg, et al. 2001), as epitopes targeted during early infection often differ from those recognized later in infection. The importance of the CTL response in modulating HIV-1 viremia can be inferred by the association of specific HLA types with low HIV RNA levels and slowed disease progression (Malhotra, Holte, Dutta, et al. 2001; Gao, Nelson, Karacki, et al. 2001; Kaslow, Carrington, Apple, et al. 1996), the demonstration that specific HLA restricted CTL are associated with better control of SIV and HIV replication (Carrington, Nelson, Martin, et al. 1999; Carrington & O'Brien. 2003), and the selection for viral escape mutants (Borrow, Lewicki, Wei, et al. 1997). Some specific CD8+T lymphocyte responses in the SIV model lead rapidly to generation of escape variants, while others appear to apply little selective pressure (Borrow, Lewicki, Wei, et al. 1997; Allen, O'Connor, Jing, et al. 2000). HIV specific CD4+ responses may also be an essential component of the initial adaptive immune response (Rosenberg, Billingsley, Caliendo, et al. 1997; Pitcher, Quittner, Peterson, et al. 1999).

Empirical data from SIV-infected primates treated with CD8+ antibodies to deplete CD8+ T cells (Jin, Bauer, Tuttleton, et al. 1999; Schmitz, Kuroda, Santra, et al. 1999; Matano, Shibata, Siemon, Connors, Lane, & Martin. 1998) and from HIV-1 infected humans evaluated for virus load, CTL frequencies, and viral escape kinetics (Bhardwaj. 2001; Jones, Wei, Flower, et al. 2004), all suggest that the viral load setpoint is influenced by CD8+ T cell immune pressures. But the practical significance of this immune pressure for vaccine development remains unclear. T-cells are a heterogeneous cell population exhibiting multiple phenotypes that reflect different states of maturation, activation, and function. The relative representation of these subpopulations changes during the primary antiviral CD8+ T cell response to HIV-1 infection (Appay, Dunbar, Callan, et al. 2002; Appay, Papagno, Spina, et al. 2002). It has been postulated that the breadth and magnitude of the virus-specific CD8+ T cell response might correlate with HIV-1 virus containment (Ogg, Jin, Bonhoeffer, et al. 1998), but three published studies have indicated otherwise (Betts, Ambrozak, Douek, et al. 2001; Cao, McNevin, Holte, Fink, Corey, & McElrath. 2003; Addo, Yu, Rathod, et al. 2003). Thus, other aspects of CD8+ T-cell activity, including but not limited to the breadth and magnitude of response, coupled with other facets of the cellular and humoral responses are likely to contribute to virus containment in complex ways.

CD8+ T-cells are dependent on CD4+ T-cell help. T helper cells are impaired early in HIV-1 infection (Pitcher, Quittner, Peterson, et al. 1999; Rosenberg, Altfeld, Poon, et al. 2000). In this phase, much damage is done to gut-associated CD4+ T-cells (Brenchley, Schacker, Ruff, et al. 2004; Mehandru, Poles, Tenner-Racz, et al. 2004) while the loss of CD4+ T-cells is slower in the blood (Picker, Hagen, Lum, et al. 2004). The damage is due in part to preferential infection of HIV-1-specific CD4+ T cells, which may be mediated by specific contact with dendritic cells (DC) infected or associated with HIV-1,

as the virus attaches to, and infects DC (Pope, Betjes, Romani, et al. 1994). HIV-1 can infect tissue plasmacytoid (p) DC by binding to CD4+ and mannose binding proteins; following internalization, production of alpha interferon is triggered and CCR7 is expressed, directing pDC to lymph nodes (Fong, Mengozzi, Abbey, Herndier, & Engleman. 2002; Fong, Mengozzi, Abbey, Herndier, & Engleman. 2002; Fonteneau, Larsson, Beignon, et al. 2004) where HIV-1 attaches to myeloid (m) DC by binding to DC-SIGN, which in turn targets virus to HIV-1 specific CD4+ T-cells (Geijtenbeek, Kwon, Torensma, et al. 2000; Kwon, Gregorio, Bitton, Hendrickson, and Littman, 2002). However, at the same time in acute and chronic HIV-1 infection, mDC and pDC are lost (Donaghy, Pozniak, Gazzard, et al. 2001; Pacanowski, Kahi, Baillet, et al. 2001). A characterization of CD4+ T-cell and DC responses during very early HIV-1 infection is therefore needed to understand how cellular immunity contributes to early virus containment. Plasmacytoid DCs may also be activated in acute HIV-1 infection, directly by HIV-1 or through toll like receptors (TLR), to generate alpha interferon and IL-15, which likely activates natural killer (NK) cells (Bhardwaj. 2001; Pulendran, Palucka, & Banchereau. 2001; Fonteneau, Larsson, Beignon, et al. 2004; Alter, Malenfant, Delabre, et al. 2004).

NK cells are activated during acute viral infections by innate cytokine production and contact with DCs or other innate cell subsets. Their effector functions are triggered by contact with virally-infected cells, where expression of ligands for inhibitory NK receptors (*e.g.,* HLA locus proteins) is reduced and ligands for activating KIR, CD94/NKG2C, and Nkp46 receptors may be increased (Pessino, Sivori, Bottino, et al. 1998). The importance of NK cells in HIV-1 infection is suggested by the epistatic association of HLA-Bw4 and KIR3DS1 with slow progression to AIDS. NK cells may directly kill HIV-1-infected cells, and, by the release of cytokines or the interaction with DCs, may also favor Th1 immune responses (Pulendran, Palucka, & Banchereau. 2001). The degree to which NK cells contribute to the early control of HIV-1 infection remains unknown and needs to be determined. Likewise, the role of T/NK cells (a subset of T cells expressing an invariant Vα chain and limited repertoire of Vβ chains) that recognises αGalCer or other ligands presented by CD1d, and like NK cells, mediates both effector and immunoregulatory functions during the early stages of viral infections (Kronenberg. 2005) and requires investigation.

1.6 Host Genetic Selection

Surprisingly little is known about the basis of inter-individual variation in HIV-1 susceptibility, progression to AIDS, or response to vaccine and ART treatment. A growing body of evidence, however, makes clear that both the host and virus genetic make-up play important roles.

The first AIDS related genetic variant – the CCR5δ32 deletion – was identified nearly 10 years ago (Dean, Carrington, Winkler, et al. 1996). Most individuals homozygous for this variant are completely protected from HIV-1 infection. However, only a handful of other variants have been consistently shown to be associated with the risk of HIV-1 transmission. In fact, ignoring results from the HLA-B alleles (Gao, Nelson, Karacki, et al. 2001; Kiepiela, Leslie, Honeyborne, et al. 2004), the only other genes repeatedly

implicated in HIV-1 transmission, baseline measures, and/or the progression to AIDS have been those that encode the CC and CXC family of chemokines and receptors (Smith, Dean, Carrington, et al. 1997; Winkler, Modi, Smith, et al. 1998; Martin, Dean, Smith, et al. 1998; Gonzalez, Rovin, Sen, et al. 2002; Duggal, An, Beaty, et al. 2003; Modi, Goedert, Strathdee, et al. 2003; Gonzalez, Kulkarni, Bolivar, et al. 2005). It is clear that these variants can explain only a fraction of the variation in virus handling that is due to host genetic variation.

In addition to the absence of large cohorts of patients monitored during the acute infection phase, genetic association studies of HIV disease to date have had two major shortcomings. First, the number of genes examined in all genetic association studies has been very small (O'Brien & Nelson. 2004). Second, the representation of genetic variation in the genes under study has not been comprehensive, a point that is exemplified by studies showing specific genetic associations observed in one ethnic group, but not another, in spite of the presence of that polymorphism in both groups. Striking advances in knowledge of polymorphism in the human genome (*e.g.* www.hapmap.org) and methodologies for the selection of validated sets of tagging single-nucleotide polymorphisms (SNPs) (Ahmadi, Weale, Xue, et al. 2005) allow significantly more comprehensive genetic association studies to be carried out than have previously been possible. Host genetic studies in the HIV field are only now beginning to use the most contemporary standards for genetic analyses, such as formal checks for stratification.

Across all participating sites except for those located in North Carolina, individuals will be asked during the screening process if their screening blood sample may be used for additional experimental testing, including genetic testing. For those who agree, minimal demographic information will be collected, including age, HIV serostatus, country of origin, and ethnic background or tribe, if applicable. This data will be used to perform genome-wide association analyses on disease predisposition and protective genes for HIV. (Screening for the study in North Carolina is done through the state system and not performed by the clinical site; therefore, they are unable to participate in this analysis).

In addition, a subset of individuals in Malawi who have agreed to allow their HIV screening sample to be used for genetic testing will also be asked if they would be willing to answer a short, one-time only risk behavior questionnaire. The additional information gathered from these individuals will provide more specific data related to genetic factors associated with HIV acquisition and transmission. This specific data set will include individuals who upon initial screening for the study are found to be HIV positive or HIV negative. It is anticipated that approximately 1000 individuals of each HIV serostatus group will be necessary to perform this analyses. This work will provide a unique description of how host genetic variation influences HIV infection in individuals from Malawi. As it is anticipated that thousands of individuals will be screened for this study over the five year enrollment period, this subset of individuals will represent a very small proportion of the total screened. Other clinical sites also may contribute to this data set if deemed necessary.

The scientific basis for the information collected via the risk behavior questionnaire is that there is clear evidence that individuals vary dramatically in many aspects of HIV disease including both susceptibility to infection and control of viral replication. Not all people exposed to HIV become infected, and those who do progress to an AIDS-defining pathology do this at different rates. Infected individuals have heterogeneity in the strength of their immune responses as well as differences in how they respond to antiretroviral treatment. Much work has been done to characterize the virulence factors of the pathogen (HIV), while the knowledge of the relevant host genetic factors is currently limited. The main reason for this is that a minimal number of genes have been screened when using a candidate gene approach. This approach has identified a limited number of gene variants that have a major impact on the susceptibility to infection (e.g. CCR5δ32 and MIP1αP). For this reason there is an urgent need to study large cohorts of individuals using a genome-wide approach. The aim is to identify gene variants that are involved with susceptibility and resistance to HIV infection.

1.7 Specific Aims

The specimens collected during the course of this study will be used to investigate the following specific aims:

Aims directed at the transmitted virus

- To determine the genetic, biologic, antigenic, and structural characteristics of sexually transmitted virus and mechanisms underlying virus transmission.
- To determine the source (cell free and/or cell associated) of sexually acquired HIV-1.
- To determine the cellular and molecular events in mucosal epithelial cell induction of virion production from virus infected cells.
- To determine whether acutely transmitted virus differs from chronic HIV-1 in co-receptor usage, relative CD4+ dependence and neutralization susceptibility.
- To identify sequence and structural signatures associated with acute HIV-1 infection.
- To model the transmitted HIV-1 trimer.
- To crystallize the transmitted HIV-1 trimer.
- To design new immunogens from structural analysis of the transmitted HIV-1 env.

Aims Directed at the Cellular Responses in Acute HIV Infection

- To examine cytotoxic CD8 T-cells, CD4+ T-cells, regulatory T-cells and DC, NK, T/NK cells in subjects with acute HIV infection.

- To characterize early anti-HIV-1 CD8 T-cell responses and its relation to the changing viral phenotype and viral set point.

- To determine the influence of CD4+ cells on CD8 T-cell response.

Aims Directed at the Antibody Response

- To characterize the ontogeny of anti-HIV systemic and mucosal antibody responses early in HIV infection through 24 months of infection.

- To make humans monoclonal antibodies reflective of neutralizing antibodies that evolve during HIV infection.

- To characterize systemic and mucosal neutralizing antibodies that arise early and late in HIV-1 infection.

- To characterize systemic and mucosal B cells that generate neutralizing antibodies.

- To characterize polyspecificity in the mediation and regulation of neutralizing antibody response systemically and at mucosal sites.

Aims Directed at Host Genetics

- To perform genome-wide association analyses using chips specific for Africa.

- To examine the genotype haplotype tagging SNPs comparing cases and control subjects in order to correlate genotype with susceptibility to infection and virus control.

- To study the HLA types of HIV positive and negative individuals.

The remainder of this protocol outlines the specific requirements for implementation of this observational study at the clinical sites. For more specific information regarding the experimental testing planned using specimens collected in this study beyond what is already provided in Section 1.0, please refer to the CHAVI website at www.chavi.org.

2.0 STUDY OBJECTIVES AND DESIGN

2.1 Primary Objectives

- To determine the prevalence of acute HIV infection at the participating sites.
- To determine the characteristics of the transmitted virus in acute HIV-1 infection.
- To determine the host acquired and innate immune responses that contribute to virus control and/or protection against infection with HIV-1.
- To determine genetic factors that contribute to early virus control and/or protection from infection.
- To examine the clinical, laboratory, and behavioral characteristics of individuals at diverse sites with acute HIV infection and their sexual partners, and appropriate controls.

2.2 Study Design

This will be a multi-center, prospective, observational, cohort study following participants who meet the criteria outlined in Section 4.0 for one of four study groups:

Group 1: Acute HIV-1 infection

Group 1A: Sexual partners of members of Group 1

Group 2: Established HIV-1 infection

Group 3: HIV-1 negative

The clinical sites enrolling participants in this study are located in the following cities and countries: Blantyre and Lilongwe, Malawi; Durban, Klerksdorp, and Johannesburg, South Africa; Moshi and Arusha, Tanzania; Kampala, Uganda; and Chapel Hill and Durham, North Carolina, USA. Other clinical sites will be added as necessary.

Across all participating clinical sites, the study targets enrollment of approximately 1300 individuals, including approximately 600 individuals with acute HIV infection, approximately 600 sexual partners of individuals with acute HIV infection, approximately 50 individuals with established HIV-1 infection, and approximately 50 individuals who are HIV-1 negative. Study Groups 2 and 3 will serve as controls in order to be able to compare viral diversity and evolution, and cell-mediated immune responses.

Accrual into all study groups will require approximately 60 months and all participants will be followed for approximately 24 months. Accrual will be monitored throughout the recruitment period and adjustments will be made accordingly (*e.g.,* increase, decrease, or maintain current accrual goals) in order to maximize enrollment into all study groups. All adjustments to accrual goals will be made by agreement with the protocol team.

Additional Information Regarding Group 1A

Efforts will be made to recruit sexual partners of participants enrolled into Group 1 in a manner acceptable to local IRB/EC requirements. This group is referred to as Group 1A, and individuals enrolled in this group are not intended to contribute to the target numbers for the other three groups. Individuals in Group 1A will follow the Schedule of Evaluations consistent with their HIV infection status at study entry, and therefore, there is no separate Schedule of Evaluations for Group 1A (that is, these individuals will be either acutely infected, chronically infected, or HIV negative).

3.0 INCLUSION AND EXCLUSION CRITERIA

3.1 Inclusion Criteria

- Men and women aged $\geq$ 18 years at the time of screening.
- Able and willing to provide adequate information for locator purposes.
- Hemoglobin > 10.0 g/dL.
- Willing to receive HIV test results.
- Not intending to relocate out of the area for the duration of study participation and does not have a job or other obligations that may require long absences from the area.
- For Study Group 1, has acute HIV infection. (See Section 4.0 for definition).
- For Study Group 1A, is a sexual partner of a member of Group 1. (See Section 4.0 for definition).
- For Study Group 2, has established HIV-1 infection. (See Section 4.0 for definition).
- For Study Group 3, is HIV-1 negative. (See Section 4.0 for definition).

3.2 Exclusion Criteria

- Any prior or current use of antiretroviral therapy (ART). This does not apply to individuals screening for Group 1A. ART use for the prevention of perinatal transmission may be waived after prior consultation with the protocol team.
- Previous or current participation in a HIV vaccine study.
- Any condition that, in the opinion of the Investigator of Record, would make participation in the study unsafe, complicate interpretation of study outcome data, or otherwise interfere with achieving the study objectives.

4.0 SCREENING PROCESS FOR HIV STATUS DETERMINATION, AND DEFINITION OF STUDY GROUPS

Screening will take place in a step-wise manner, and may take place over one or more visits. Screening procedures and assessments may vary across sites due to different standards of care and local procedures, but will in general include determination of HIV infection status, obtaining locator information, obtaining limited demographic and HIV risk behavior information, and hemoglobin testing. At some sites, some or all of the elements of the HIV screening algorithm employed in this study is conducted as part of the local standard of care, and therefore consenting for such testing will not be necessary. However, for most sites, the HIV testing algorithm employed in this study is not the standard of care, and therefore consenting for HIV testing as outlined will be necessary. In addition, for sites based in North Carolina, documented evidence of HIV test results from laboratories external to their clinical site may be used as part of the documentation for entry into the study.

Participants will sign the site-specific screening consent before any study specific procedures or assessments are initiated.

For enrollment into the study groups, HIV infection status will be defined as outlined below, and will determine the visit schedule for participants.

Group 1 - Acute HIV-1 Infection:

One of the following three scenarios must occur within 45 days of the enrollment date in order to be considered an acute-HIV infection for enrollment into Group 1:

SCENARIO ONE:

For purposes of this protocol, acute HIV-1 infection for this scenario will be defined using 1) initial HIV test results, **PLUS** 2) additional HIV test results as outlined below. All testing outlined below must be performed for determination of acute HIV-1 infection, and for inclusion in the study database.

1) Initial testing with the following results:

A potential participant has a negative HIV antibody test using either two different rapid HIV tests performed simultaneously, or a standard EIA, AND a positive plasma p24 antigen or a positive nucleic acid test (NAT).

A participant may be enrolled in to the study based on these initial test results. Every effort should be made to perform the additional testing listed below prior to enrollment; however, this is not required.

2) Additional testing with the following results:

A. A negative Western blot or a second negative EIA on the initial or any subsequent sample. If performing an EIA, it must be a different test from what was used for the initial testing. In addition, if a subsequent sample is used for this testing, it should be obtained at the earliest time point possible following the initial sample.

OR

B. Concordant positive or discordant rapid tests (using two different rapid HIV tests), or a positive standard EIA, with a negative or indeterminate* Western blot using a subsequent sample. The subsequent sample used for this should be obtained at the earliest time point possible following the initial sample. (* An indeterminate Western blot will be defined as any visible band reactivity which does not meet the criteria for a positive result. A positive Western blot is defined as the presence of 2 or more of the HIV-1 specific Western blot bands: p24, gp41 and gp 120/160).

There may be cases where the additional HIV test results do not match what is outlined in "A" or "B" above. In such cases, email 001Acutes@CHAVI.org for further instructions.

SCENARIO TWO:

A potential participant has discordant rapid tests (using two different rapid HIV tests**), or** a positive standard EIA, **AND** a positive plasma p24 antigen or NAT, **AND** a negative Western blot. These tests results will be obtained from the initial screening sample, or on a sample drawn within 24 hours of the initial sample.

SCENARIO THREE:

A potential participant has discordant rapid HIV tests (using two different rapid HIV tests), **or** a positive standard EIA, **AND** a positive plasma p24 antigen or NAT, AND documentation within the previous 45 days of a negative HIV antibody test **(**via standard EIA, or two different HIV rapid tests).

Group 1A – Sexual partners:

Sexual partners of members of Group 1, defined as a person(s) who participated in a sexual act with a person enrolled in Group 1 (acute HIV infection), and the sexual act must have taken place within 12 weeks prior to the first time the member of Group 1 is identified as acutely infected with HIV. For example, if a participant enrolled in Group 1 (acute HIV infection) had their initial HIV screening test on March 15, a sexual partner of this participant must report that the sexual act with that person occurred within 12 weeks prior to March 15

Group 2 – Established HIV infection:

A positive HIV antibody test (two concordant rapid HIV tests or standard EIA) and a positive Western blot at any time point prior to study entry. HIV-1 culture, HIV-1 antigen, plasma HIV-1 RNA, or a second antibody test by a method other than an ELISA is acceptable as an alternative confirmatory test. Alternatively, if an EIA result is not available two HIV-1 RNA values > 2000 copies/ml at least 24 hours apart performed by a CHAVI-certified laboratory may be used to document infection.

Group 3 – HIV negative:

Within 30 days of the enrollment date:

- A negative HIV antibody test (two concordant rapid HIV tests or standard EIA) and a negative p24 antigen or NAT.
- An indeterminate HIV antibody test (including discordant rapid HIV tests) with a negative WB, and a negative p24 antigen or NAT.

4.1 HIV Counseling and Access to Care

All HIV testing will take place in the context of pre-test, risk-reduction, and post-test counseling. Regardless of their test results, individuals will be given condoms and information about safe sex practices. In addition, each person identified with HIV infection will be given information regarding access to local HIV care facilities per the local standard of care.

5.0 STUDY PROCEDURES

For each participant, independent written informed consent for screening and enrollment will be obtained before any procedures are initiated. The Schedules of Evaluations (Appendix I) provide the procedures, evaluations, and visit schedules for all study groups; not all procedures or evaluations will be performed for all study groups at each visit.

Tests and evaluations in support of the clinical care of participants but beyond the protocol requirements are not listed below, and may be managed throughout the study at

the discretion and judgment of the Investigator of Record, in accordance with local standards of care.

5.1 Administrative and Behavioral Procedures

- Locator information.
- Demographic information.
- Behavioral assessment.
- HIV pre-test, risk-reduction, and post-test counseling (for all participants at screening and for HIV negative participants throughout the study).
- HIV safe sex counseling (may be given to individuals or couples).

5.2 Clinical Procedures

- Directed history and physical exam.
- Blood collection.

  Note: The blood volume indicated in the Schedules of Evaluations and Procedures (Appendix I) can be collected if the participant has a hemoglobin value $\geq$ 10.0. However, if a participant presents with clinical signs and symptoms of a condition that may impact hemoglobin, the participant's hemoglobin should be tested prior to the scheduled blood draw. If the hemoglobin value is found between 7.5 and 9.9, the volume of each individual blood draw will be decreased by half until the hemoglobin values rises above 9.9 again. For example, if the protocol-specified blood draw volume is 100ml, but the participant has a hemoglobin value of 8.2, the draw volume will be reduced to 50ml. If the study participant's hemoglobin increases above 9.9 later in the study, the blood volume can be increased back to the original protocol-specified amount. If it is found that a participant has a hemoglobin value less than 7.5, no blood may be drawn for storage. In such cases, blood draws other than for storage should be prioritized as follows: 1) blood for testing necessary to assure the health of the participant and 2) protocol-mandated testing not to exceed testing for the requirements of the corresponding Schedule of Evaluations, e.g. HIV viral load, CD4+ cell count, and HIV antibody confirmation. Hemoglobin testing will be required at regular intervals throughout follow-up (refer to the Schedule of Evaluations for each study group).

- Optional samples: the following samples are considered optional for both the participating clinical site and the participant (see Section 5.4.2). The choices include:
    - Genital secretions (semen from men, sample from the endocervical canal from women)
    - Urine
    - Feces
    - Saliva
    - Breast milk from lactating women

5.3 Clinical Laboratory Evaluations

Laboratory evaluations to be conducted primarily at the clinical site laboratory:

- HIV-1 antibody testing: rapid or standard HIV testing (if rapid, use two simultaneous tests using two different types of kits per test kits listed in SSP).
- HIV p24 antigen or HIV NAT (when appropriate, use pooling strategy).
- Western blot.
- Hemoglobin (refer to Note above for "Blood collection", and to "NOTE 2" included on each Schedule of Evaluation)
- CD4+ cell count
- Blood plasma HIV-1 RNA PCR (to determine viral load in HIV-infected individuals).
- Specimen processing and shipment per SOPs/SSP.

5.4 Additional Procedures Regarding Group 1 and Group 3

For participants who are enrolled in Group 1, HIV testing should continue until there is a documented positive Western blot, or until Week 24, whichever occurs first. If by week 24 there is no positive Western blot, the participant will not be considered a "proven" acute HIV-1 infection, and the study team should be notified as soon as possible at 001Acutes@CHAVI.org for further instructions.

For participants who are enrolled in Group 3 and during the course of the study are identified as acutely or chronically infected with HIV, they should be offered to participate in the study according to the one of those groups, and consented accordingly.

If they refuse to do so, the study team should be notified as soon as possible at 001Acutes@CHAVI.org for further instructions.

5.5 Special Considerations

5.5.1 Flow and Testing of Specimens

As indicated above, the laboratory or laboratories associated with the clinical site will perform clinical testing that pertains directly to the health of the participant, including HIV testing, hemoglobin, CD4+ cell count, viral load, and any other testing as deemed necessary for the clinical care of the participant but not otherwise dictated by the protocol (*e.g.* STI testing). Clinical sites will throughout the course of the study process and ship specimens to the CHAVI Repositories located in Johannesburg, South Africa; and Durham and Chapel Hill, North Carolina for temporary storage. As directed by the CHAVI Scientific Leadership Group, the CHAVI Repositories will ship these specimens on an on-going basis to CHAVI investigators for experimental testing. In some cases where the technology exists, experimental testing may be performed at a clinical site laboratory with prior approval of the CHAVI Scientific Leadership Group.

5.5.2 Optional Specimens

Blood is the only specimen a participant must be willing to provide in order to participate in this study. It is at the discretion of each clinical site Investigator of Record and/or IRB/EC to determine whether to collect optional specimens, *i.e.* genital secretions, feces, urine, saliva, breast milk, as part of the study conducted at their site. It is understood that local IRB/ECs may likely impact this decision. The sample informed consents appended to this protocol are designed to allow sites to include in their site-specific informed consent forms the additional specimens deemed appropriate for their local population and clinical setting. Participants will have the option to provide these optional samples based on the choices included in the local informed consent forms.

5.6 HIV and Risk-Reduction Counseling, and Sexually Transmitted Infection and HIV Care

This study will enroll men and women who are either infected with HIV, or who may be at high risk for becoming infected with HIV or other STIs. As such, this study will provide both voluntary HIV counseling and testing (VCT) and risk-reduction and safe-sex counseling. The risk-reduction and safe-sex counseling may be administered to individuals or couples and will be provided in accordance with local standards and regulations. Condoms will also be provided at no cost to participants throughout the duration of their participation.

Participants requiring clinical care for the management of symptomatic STIs will either receive it at the participating study site, or will be referred for appropriate care. Similarly, HIV-infected individuals identified through screening, and participants who are infected or become infected with HIV during the study will be referred for appropriate HIV care (which may be under the management of the participating site).

This care will be consistent with the host country's national guidelines for care and treatment of people with HIV, and may include referral for possible enrollment into other available HIV treatment clinical trials or to other resources if available (*e.g.,* Global Fund, PEPFAR, etc.). It should be noted, however, that due to the specimen collection requirements of this study, co-enrollment into other studies will be made by the Investigator of Record on a case-by-case basis depending on the nature of the other studies.

5.7 Behavioral Risk Assessment

Participant risk behaviors will be assessed via standardized interviewer-administered questionnaires at regular intervals during the study. These assessments will include questions related to number of sexual partners and condom use, and will be used to describe the behaviors associated with HIV acquisition and transmission, and to guide study enrollment efforts.

In addition, during the screening process for the study, the clinical sites in Malawi will ask a subset of consenting individuals (approximately 1000 HIV positive and 1000 HIV negative individuals) a short, one-time only risk behavior questionnaire in order to gather more specific data related to genetic factors associated with HIV acquisition and transmission. Additional details related to this are outlined in Section 1.6.

5.8 Maximizing Participant Follow-Up

Once a person enrolls in this study, every reasonable effort should be made to follow him or her per the visit schedule included in Appendix I. Established systems for conducting follow-up visits at each site should be utilized to the extent possible. A site will make every effort to enroll, follow, and obtain samples from all enrolled participants according to the schedule of evaluations, while recognizing that each participant may not be able to return for all visits or provide all protocol-required samples, or undergo all protocol-dictated procedures. No individual should be terminated from the study based solely on his or her ability to return for study visits or provide samples. If a participant misses a visit, he or she may return outside of the visit window or at any future visit, and, thus, continue their participation in the study. For purposes of the study database only, unscheduled visits will be defined as interim visits and appropriate data will be collected. A protocol-required follow-up visit is considered missed when the participant does not complete any procedures prior to the start of the next visit window.

Sites must have written SOPs for retention, and should consider the following elements:

- Thorough explanation of the study visit schedule and procedural requirements during the informed consent process and re-emphasis at each study visit.
- Collection of detailed locator information during screening, and active review and update of this information at each subsequent visit.
- Use of mapping or other techniques to establish the location of participant residences and other locator venues.

- Use of appropriate and timely visit reminder mechanisms.
- Appropriate follow-up on missed visits.
- Mobilization of trained outreach workers or "tracers" to complete in-person contact with participants at their homes or other community locations.

5.9 Participant Withdrawal

Participants may voluntarily withdraw from the study for any reason at any time. The site Investigator of Record may withdraw participants from the study in order to protect their safety. Participants also may be withdrawn if the NIAID/NIH, Office of Human Research and Protection (OHRP), other regulatory authorities, or site IRBs/ECs terminate the study prior to its planned end date.

6.0 SAFETY MONITORING AND ADVERSE EVENT REPORTING

The study site team will monitor for and track serious adverse events related or possibly related to study procedures and/or to participation in the study. Such events that are unanticipated will be reported to the DAIDS CHAVI Program Officer at the same time as they are reported to IRB/ECs per their requirements and pre-established written procedures, as required by 45 CFR 46**.** No other adverse event reporting will be required for this observational study in which there is no intervention.

Limited information on social harms directly related to study participation will be collected on case report forms for entry into the study database. In addition, social harms judged to be significant by the Investigator of Record will be reported to the relevant IRB/ECs at least annually, or according to their individual requirements.

7.0 STATISTICAL CONSIDERATIONS

Samples collected during this study will be used to meet the CHAVI scientific objectives and specific aims as outlined in Sections 1.7 and 2.1 The enrollment target number for each Study Group is based on what is considered to be realistic and achievable at the current participating sites, and not based on statistical input as each of the planned studies is anticipated to be subject to different statistical considerations. Furthermore, all analyses will be exploratory and hypothesis-generating. Appropriate descriptive statistics will be used.

As described in Section 1.0, the work conducted by the CHAVI entails an ambitious research portfolio. An analysis plan will be developed for each research question. Included are two examples that show the types of analysis anticipated.

7.1 Host Genetics

In addition to the absence of large cohorts of patients monitored during the acute infection phase, genetic association studies of HIV disease to date have had two major

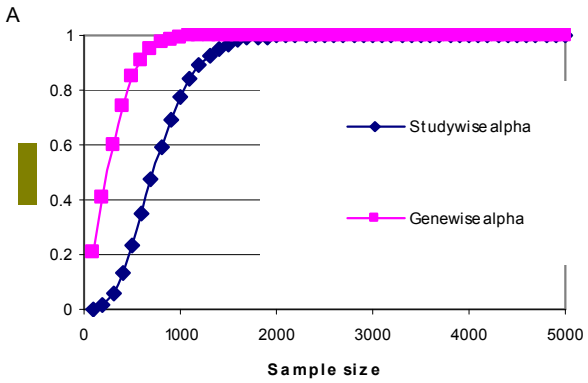
shortcomings. First, the number of genes examined in all genetic association studies has been very small (O'Brien & Nelson. 2004). Second, the representation of genetic variation in the genes under study has not been comprehensive, a point that is exemplified by studies showing specific genetic associations observed in one ethnic group, but not another, in spite of the presence of that polymorphism in both groups. Striking advances in knowledge of polymorphism in the human genome (*e.g.* www.hapmap.org) and methodologies for the selection of validated sets of tagging SNPs (Ahmadi, Weale, Xue, *et al*. 2005) allow for significantly more comprehensive genetic association studies than have previously been possible. Finally, host genetic studies in the HIV field are only now beginning to use the most contemporary standards for genetic analyses, such as formal checks for stratification.

Observed sample points will be used to statistically infer the full dynamic pattern of viral load (and other measures such as CD4+ counts) during the acute phase of infection. Then host genetic determinants will be searched for that influence viral control during these early stages of infection. Variants that influence viral load soon after the establishment of set point will also be studied.

To assess host genetic variation, Illumina genotyping platform will be used to genotype a set of putatively functional gene variants (e.g. coding polymorphisms) as well as haplotype tagging SNPs designed to represent all common variation in the genes of interest (see Ahmadi et al 2005 for explanation of this approach). This approach will be applied to approximately 500 key candidate genes. In addition, an Affymetrix 500,000 SNP chip will be used to carry out genome-wide association on a subset of the collected samples.

Power calculations have been performed for LD-based detection of variants that influence a quantitative trait. This may be for example the measured viral load at the establishment of set point, or it may be the inferred peak virus load during the acute phase of infection, or some other aspect of the dynamic pattern of viral buildup and decline during the acute infection phase. The extent to which any one genetic variant contributes to variation in HIV-related endophenotypes is unknown. Power is illustrated based on a 5% explanation of total phenotypic variance, consistent with some recent reports of single-locus association (Ma, Marmor, Zhong, Ewane, Su, & Nyambi. 2005). Note however that error associated with experimental measures of traits (such as viral load measurements) will further reduce power.

Two curves are presented, one for a gene-wise false positive rate of 5%, and one for a study-wise false positive rate of 5% (assuming 500 candidate genes studied, and comprehensive haplotype tagging). The variant is assumed to explain 5% of the total phenotype variation in the trait under question. As can be seen, there is moderate power for a quantitative variant of that effect size by 700 observations even after correcting for all tests in the experiment (lower curve). These calculations take into account the multiple testing that is entailed in the use of tagging SNPs in a haplotype based regression framework (Goldstein, Ahmadi, Weale, & Wood. 2003).



To identify trends (that is, significance for a genewise experiment) there is good power at 700 observations. Variants that fail to show formal significance after full correction but that show trends will be followed up with assessments of functional effects and assessment on related or identical phenotypes (where available) in independent cohorts.

7.2 Viral Diversity

This analysis will be conducted in two phases. An initial discovery phase will generate data for power and statistical calculations for each research study proposed. In the "discovery phase", viral genotypes and phenotypes and corresponding host immune responses will be studied in 60 participants with acute infection, 20 additional acutely infected participants together with their transmitting partners, and 40 matched contemporaneous chronic HIV-1 controls. For viral genetic analysis, a parallel computer cluster with Internet access has been set up at Los Alamos to enable CHAVI investigators to rapidly create robust maximum likelihood phylogenies using automated algorithms. The database includes an automated tree builder on a 36 node Pentium III Beowulf style cluster with a web interface. This cluster can build a preliminary maximum likelihood tree de novo from an alignment of 150 taxa with 6000 characters in approximately six hours. Based on these phylogenies, participants will be automatically assigned to one of the three categories using the following definitions: (i) highly bottlenecked infections in which no sequence has evolved more than 2% from the most recent common ancestral *env* (gp160) sequence in that individual; (ii) heterogeneous infections that have evolved more than 2%, but form a monophyletic group in phylogenetic trees including other regional samples; and (iii) heterogeneous infections varying by more than 2% of full length *env* nucleotide sequence but comprising multiple distinct lineages in phylogenetic trees.

By the end of the discovery phase, assuming 60 samples evenly divided between those with and without STIs and 20% heterogeneous samples overall have been acquired, power calculations indicate, for example, an 81% chance of detecting an 8-fold enrichment of heterogeneous infections among participants with STIs, at a 0.05 confidence level (Korber and Maldoon, unpublished). Over the subsequent years of the study, it can be established if there are relationships between viral diversity during acute infection and gender, route of transmission, sequence subtype, sexual behavior, as well as

viral loads in the blood and genital secretions of the donor. If 80 acutely infected participants and 20 sexual partners are identified, a subset could be identified based on timing of infection, sample availability, and initial viral genetic complexity for molecular studies of the evolving viral quasispecies and host immune responses. Pilot studies by CHAVI investigators (Wei, Decker, Wang, et al. 2003; Derdeyn, Decker, Bibollet-Ruche, et al. 2004) indicate that analysis of 20 acutely infected participants will be sufficient to obtain viral genetic and biological data, and host neutralizing antibody and cellular immunity data, for hypothesis generation and refinement, trial design, and power calculations for subsequent phases of the projects.

8.0 LABORATORY SPECIMENS AND BIOHAZARD CONTAINMENT

Specimens will be collected, some for testing at the local clinical laboratories, and some for processing and shipment only, including:

- Blood for HIV serology (HIV antibody (rapid or EIA), HIV NAT, HIV p24 antigen, Western blot), HIV-1 RNA PCR, CD4+ cell count, and hemoglobin.

- Blood (whole blood, plasma, serum, PBMC), and the option of genital secretions (semen and sample from endocervical canal), feces, urine, saliva, and breast milk for processing, short-term storage, and shipment.

Each study site will adhere to standards of Good Clinical Practice (GCP), and local standard operating procedures (SOPs) for proper collection, processing, labeling, transport, and storage of specimens at the local lab. Specimen collection, testing, and storage at the local clinical lab will be documented using a specimen tracking system outlined in the Study Specific Procedures (SSP) Manual, where procedures for shipment of specimens to the appropriate repositories for temporary holding will be outlined.

8.1 Quality Control and Quality Assurance Procedures

Clinical sites will be required to develop and maintain general laboratory quality control procedures, including SOPs for protocol-dictated testing, proper maintenance of laboratory testing equipment, and inventory of appropriate reagents. Samples will be requested for the purpose of QA/QC testing.

8.2 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 when drawing blood, handling samples, and shipping specimens for this study, as currently recommended by the US Centers for Disease Control and Prevention (CDC).

All infectious specimens will be transported using packaging mandated in the Code of Federal Regulations, 42 CFR Part 72. In addition, individual carrier guidelines (e.g. World Courier) will be referred for specific instructions. The SSP Manual will specify further instructions regarding shipping.

9.0 HUMAN SUBJECTS CONSIDERATIONS

9.1 Ethical Review

This protocol and the template informed consent forms (ICFs) contained in Appendices I will be reviewed and approved by the Division of AIDS (DAIDS) with respect to scientific content and compliance with applicable research and human subjects regulations. Prior to study implementation, each clinical site is responsible for modifying the template ICFs to be site-specific, and for gaining approval from all Institutional Review Boards/Ethical Committees (IRBs/ECs) that have jurisdiction over the study and/or site. Subsequent to initial review and approval, all responsible IRBs/ECs will review and re-approve the protocol and ICFs at least annually. All IRB changes will be reviewed by DAIDS before site initiation.

9.2 Informed Consent

Written informed consent will be obtained from each study participant (or a mark in the presence of an impartial witness for those who are illiterate). Participants will also be offered a copy of the informed consent to keep, and the study site will note this in the study participant's file. As stated in Section 9.1, each site is responsible for developing site-specific ICFs for local use based on the templates in Appendix II. These ICFs must contain the elements of informed consent according to GCP and any other applicable regulations, including the purpose of the study, the procedures to be followed, and the risks and benefits of participation. Written informed consent will also be obtained for long-term specimen storage and possible future testing.

Where applicable, the study site is responsible for translating the template forms into the local language(s). Study sites will also be asked to verify the accuracy of the translation by performing an independent back-translation.

9.3 Confidentiality

All study-related information will be stored securely at the study site. All participant information will be stored securely in areas with access limited to study staff. To maintain participant confidentiality, a coded number will identify all study-specific laboratory specimens, reports, study data collection, process, and administrative forms. Study sites that use local databases will secure the systems with password-protected access. A participant's study information will not be released without the written permission of the participant, except as necessary for monitoring by DAIDS and/or its contractors; representatives of CHAVI, or other government and regulatory authorities, and/or the site IRB/EC.

9.4 Study Discontinuation

The study may be discontinued at any time by NIAID/DAIDS, OHRP, in-country government or regulatory authorities, and/or the IRB/EC overseeing research at the study site.

10.0 ADMINISTRATIVE PROCEDURES

10.1 Study Activation

The DAIDS CHAVI Program Officer or designee will assure that study sites have the appropriate IRB/EC approval of protocol and informed consent forms prior to protocol initiation. The DAIDS CHAVI Program Officer will also make the decision when the protocol is final (i.e. final version 1.0, 2.0, etc.) for DAIDS.

Clinical sites may not begin screening and enrollment until they are given specific, written permission (referred to as site activation notification) to do so by the CHAVI Coordinating Center. Sites will be informed of the requirements they must fulfill in order to gain site activation and implement the study.

10.2 Study Coordination

Study implementation will be directed by this protocol as well as the SSP Manual. The SSP Manual will outline procedures for conducting study visits; collecting, processing, and shipping specimens; data and forms processing; management and reporting; and other study operations.

10.3 Study Monitoring

On-site study monitoring will be performed in accordance with DAIDS policies. Study monitors will visit the site to:

- Verify compliance with human subjects and other research regulations and guidelines.
- Assess adherence to the study protocol and local SOPs.
- Confirm the quality and accuracy of information collected at the study site and entered into the study database.
- Confirm the quality and compliance with GLP of specimen collection, storage, and shipping.

Site investigators will allow study monitors to inspect study facilities and documentation (*e.g.*, informed consent forms, clinic and laboratory records, source documents, and case report forms), as well as observe the performance of study procedures.

10.4 Protocol Compliance

The study will be conducted in full compliance with the protocol. The protocol will not be amended without prior approval by the Protocol Chair and DAIDS. All protocol amendments must be submitted to and approved by the relevant IRB(s)/EC(s) prior to implementation.

10.5 Maintenance of Essential Documents and Participant Files

The Investigator of Record will maintain and store in a secure manner complete, accurate, and current study records throughout the study. Study records include both essential documents (*e.g.,* the protocol) and documentation related to each participant screened and/or enrolled into the study (*e.g.,* ICFs, locator forms, case report forms (CRFs), and other source documents).

10.6 Use of Information and Publications

Publication of the results of this study will be governed by DAIDS and CHAVI policies. Any presentation, abstract, or manuscript generated by the clinical site Investigator of Record or their clinical staff will be submitted by the Investigator of Record to the CHAVI SLG for review prior to submission.

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APPENDIX I: SCHEDULE OF EVALUATIONS AND PROCEDURES

SCHEDULE OF EVALUATIONS AND PROCEDURES: Group 1: Acute HIV-1 Infection

	Screening[2]	Enrollment	Week 1	Week 2	Week 3	Week 4	Week 8	Week 12	Week 16	Weeks 24, 36, 48, 60, 72, 84, 96
Visit Window (days [d]/weeks [w])*			±3d	±3d	±3d	±3d	±7d	±2w	±2w	±4w
Administrative and Behavioral Procedures										
Screening consent	X									
Enrollment consent		X								
Locator information	X	X	X	X	X	X	X	X	X	X
Demographic information	X[6]	X								
HIV safe sex counseling	X	X	X	X	X	X	X	X	X	X
Sexual behavioral assessment		X						X		X
Sexual behavioral assessment (additional assessment for sites in Malawi only)	X									
HIV pre/post-test counseling	X									
Clinical Procedures										
Blood collection	X	X	X	X	X	X	X	X	X	X
Optional collection: Genital secretions, breast milk[5]		X	X	X	X	X	X	X	X	X
Optional collection: Feces, urine, saliva		X	X	X	X	X				
Medical history and physical exam		X						X		X
Clinical Laboratory Evaluations										
HIV-1 testing[3] (rapid or standard EIA, Western blot, HIV NAT [pooling or Individual], or HIV p24 antigen)	X	[X]	[X]	[X]	[X]	[X]	[X]	[X]	[X]	[X]
Hemoglobin[1]	X	X	[X]	[X]	[X]	[X]	[X]	X	[X]	X
CD4+ cell count[4]		X						X		X
HIV-1 RNA PCR (for viral load)		X	X	X	X	X	X	X	X	X
Specimen Processing										
Whole blood	X									
Plasma, Serum, PBMC		X	X	X	X	X	X	X	X	X
Genital secretions, breast milk		X	X	X	X	X	X	X	X	X
Feces, urine, saliva		X	X	X	X	X				
BLOOD VOLUME (mL)	10	100	60	60	60	60	60	75	75	75

* A week equals 7 days. // [ ] = brackets indicate to perform if necessary. // [1]Hemoglobin testing is required for study entry, and weeks 12, 24, 36, 48, 60, 72, 84, and 96. For all other visits, hemoglobin testing should be performed based on a clinical indication that would impact the amount of blood drawn, e.g. malaria, pregnancy, etc. // [2] Screening procedures will be dictated by site-specific practices. // [3] HIV-1 testing should follow the algorithm in Section 4.0. Once a participant is enrolled, HIV testing should continue until there is a documented positive Western blot, or until Week 24, whichever occurs first. If by week 24 there is no positive Western blot, the participant will not be considered a "proven" acute HIV-1 infection, and the study team should be contacted at 001Acutes@CHAVI.org. // [4] Perform CD4+ testing at enrollment, 24, 48, 72, and 96.//[5] Collect from lactating women up to 50 mL at up to 4 consecutive visits; the first collection can begin at any visit.// [6] US sites will NOT perform this procedure.

NOTE 1: This schedule should be followed for members of Group 1A if they are identified as acutely infected with HIV.

NOTE 2: Blood volume amounts listed are considered the maximum amounts, and are within the American Red Cross and South African National Blood Service guidelines of a maximum of 450 ml every 56 days (8 weeks). The blood volume indicated at each visit may be collected if the participant has a hemoglobin value $\geq$ 10.0. However, if a participant is found to have a hemoglobin value between 7.5 and 9.9, the volume of each individual blood draw should be decreased by half until the hemoglobin values rises above 9.9 again. If it is found that a participant has a hemoglobin value less than 7.5, no blood may be drawn for storage. In such cases blood draws other than for storage should be prioritized as follows: 1) blood for testing necessary to assure the health of the participant and 2) protocol-mandated testing not to exceed testing for HIV viral load, CD4+ cell count, and HIV antibody confirmation (if required).

SCHEDULE OF EVALUATIONS AND PROCEDURES: Group 2: Established HIV-1 Infection

	Screening[2]	Enrollment	Week 2	Week 4	Week 8	Week 12	Week 16	Week 24, 48, 72, 96
Visit Window (days [d]/weeks [w]) (a week equals 7 days)			±3d	±3d	±7d	±2w	±2w	±4w
Administrative and Behavioral Procedures								
Screening consent	X							
Enrollment consent		X						
Locator information	X	X	X	X	X	X	X	X
Demographic information	X[5]	X						
HIV safe sex counseling	X	X	X	X	X	X	X	X
Sexual behavioral assessment		X				X		X
Sexual behavioral assessment (additional assessment for sites in Malawi only)	X							
HIV pre/post-test counseling	X							
Clinical Procedures								
Blood collection	X	X	X	X	X	X	X	X
Optional collection: Genital secretions, breast milk[4]		X	X	X	X	X	X	X
Optional collection: feces, urine, saliva		X						
Medical history and physical exam		X				X		X
Laboratory Evaluations								
HIV-1 testing [3] (rapid or standard EIA, Western blot, HIV NAT [pooling or Individual], or HIV p24 antigen)	X							
Hemoglobin[1]	X	X	[X]	[X]	[X]	X	[X]	X
CD4+ cell count		X						X
HIV-1 RNA PCR (for viral load)		X	X	X	X	X	X	X
Other tests to fulfill the Specific Aims	X	X	X	X	X	X	X	X
Specimen Processing								
Plasma, serum, PBMCs		X	X	X	X	X	X	X
Genital secretions, breast milk		X	X	X	X	X	X	X
Feces, urine, saliva		X						
BLOOD VOLUME (mL)	10	60	60	60	75	75	75	75

[ ] = brackets indicate to perform if necessary.

[1]Hemoglobin testing is required at screening for study entry, and weeks 12, 24, 48, 72, and 96. For all other visits, hemoglobin testing should be performed based on a clinical indication that would impact the amount of blood drawn, e.g. malaria, pregnancy, etc.

[2] Screening procedures will be dictated by site-specific practices

[3] HIV-1 testing will be dictated by site-specific practices, but should follow algorithm in Section 4.0.

[4] Collect from lactating women up to 50mL at up to 4 consecutive visits; the first collection can begin at any visit.

[5] US sites will NOT perform this procedure.

NOTE 1: This schedule should be followed for members of Group 1A if they are identified as having established HIV infection.

NOTE 2: Blood volume amounts listed are considered the maximum amounts, and are within the American Red Cross and South African National Blood Service guidelines of a maximum of 450 ml every 56 days (8 weeks). The blood volume indicated at each visit may be collected if the participant has a hemoglobin value $\geq$ 10.0. However, if a participant is found to have a hemoglobin value between 7.5 and 9.9, the volume of each individual blood draw should be decreased by half until the hemoglobin values rises above 9.9 again. If it is found that a participant has a hemoglobin value less than 7.5, no blood may be drawn for storage. In such cases blood draws other than for storage should be prioritized as follows: 1) blood for testing necessary to assure the health of the participant and 2) protocol-mandated testing not to exceed testing for HIV viral load and CD4+ cell count.

SCHEDULE OF EVALUATIONS AND PROCEDURES: Group 3: HIV-1 Negative

	Screening[2]	Enrollment	Week 1	Week 2	Week 4	Week 8	Week 12	Weeks 24, 48, 72, 96
Visit Window (days [d]/weeks [w]) (a week equals 7 days)			±3d	±3d	±3d	±7d	±2w	±4w
Administrative and Behavioral Procedures								
Screening consent	X							
Enrollment consent		X						
Locator information	X	X	X	X	X	X	X	X
Demographic information	X[5]	X						
HIV safe sex counseling	X	X	X	X	X	X	X	X
Sexual behavioral assessment		X					X	X
Sexual behavioral assessment (additional assessment for sites in Malawi only)	X							
HIV pre/post-test counseling	X	X	X	X	X	X	X	X
Clinical Procedures								
Blood collection	X	X	X	X	X	X	X	X
Optional collection: genital secretions, breast milk[4]		X	X	X	X	X	X	X
Medical history and physical exam		X					X	X
Laboratory Evaluations								
HIV-1 testing[3] (rapid or standard EIA, Western blot, HIV NAT [pooling or Individual], or HIV p24 antigen)	X	X	X	X	X	X	X	X
Hemoglobin[1]	X	X	[X]	[X]	[X}	[X]	X	X
CD4+ cell count		X						X
Other tests to fulfill the Specific Aims	X	X	X	X	X	X	X	X
Specimen Processing								
Plasma, serum, PBMCs, genital secretions, breast milk		X	X	X	X	X	X	X
BLOOD VOLUME (mL)	10	60	60	60	60	75	75	75

[ ] = brackets indicate to perform if necessary.

[1]Hemoglobin testing is required at screening for study entry, and weeks 12, 24, 48, 72, and 96. For all other visits, hemoglobin testing should be performed based on a clinical indication that would impact the amount of blood drawn, *e.g.* malaria, pregnancy, etc.

[2] Screening procedures will be dictated by site-specific practices

[3] HIV-1 testing will be dictated by site-specific practices, but should follow algorithm in Section 4.0.

[4] Collect from lactating women up to 50mL at up to 4 consecutive visits; the first collection can begin at any visit.

[5] US sites will NOT perform this procedure.

NOTE 1: This schedule should be followed for members of Group 1A if they are identified as HIV-1 negative.

NOTE 2: Blood volume amounts listed are considered the maximum amounts, and are within the American Red Cross and South African National Blood Service guidelines of a maximum of 450mls every 56 days (8 weeks). The blood volume indicated at each visit may be collected if the participant has a hemoglobin value $\geq$ 10.0. However, if a participant is found to have a hemoglobin value between 7.5 and 9.9, the volume of each individual blood draw should be decreased by half until the hemoglobin values rises above 9.9 again. If it is found that a participant has a hemoglobin value less than 7.5, no blood may be drawn for storage. In such cases blood draws other than for storage should be prioritized as follows: 1) blood for testing necessary to assure the health of the participant and 2) protocol-mandated testing not to exceed testing for CD4+ cell count, and HIV antibody confirmation (if required).

APPENDIX II: SAMPLE INFORMED CONSENT FORMS

SAMPLE INFORMED CONSENT FORM

DIVISION OF AIDS, NIAID, NIH

CHAVI 001: Acute HIV-1 Infection Prospective Cohort Study, Version 2.0

SCREENING INFORMED CONSENT FORM

PRINCIPAL INVESTIGATOR: [*insert name*]
PHONE: [*insert number*]

INTRODUCTION:

You are being asked to volunteer for screening procedures to find out if you are eligible for the research study named above. This study is sponsored by the United States National Institutes of Health. The person in charge of the study at this site is [*insert name of Investigator of Record*]. Up to approximately 1300 people will participate in this research study in Africa and the United States.

The research study is for men and women who could get HIV, or already have HIV. HIV is the virus that causes AIDS. The screening procedures include questions and may include blood tests if you have not already had them. You will be told the results of all of your screening tests as soon as they are available.

Before you decide whether to undergo the screening procedures, we would like to explain the purpose of them, the risks and benefits to you, and what is expected of you. The study staff will discuss the information with you. They will answer any questions you may have. After the screening procedures have been fully explained to you, you can decide whether or not you want to participate. If you understand the tests and agree to participate, you will be asked to sign this consent form or make your mark in front of someone. You will be offered a copy of this form to keep.

Please note that:

- Your participation in the screening tests is entirely voluntary.
- You may decide not to take part or to withdraw from the screening process at any time without losing the benefits of your standard health care.
- You are only being asked to take part in the screening process at this time. Even if you agree to have the screening tests, you do not have to join the research study. If in the future you decide you do not want your specimens used for any of the testing you agree to, it is important that you let the study staff know.

PURPOSE OF THE SCREENING TESTS:

The purpose of the screening procedures is to find out if you are eligible for a research study. *[Institution]* is part of a group of scientists from all over the world doing research on how HIV works in the body and how the body tries to fight back. The purpose of this research study is to understand how people's infection fighting (immune) system reacts to HIV. The immune system is made up of the cells in a person's body that fight infection. In *[insert country],* the main way that people get infected with HIV is by having sex with someone who has HIV. We want to know how the immune system reacts if someone comes into contact with HIV in this way, whether or not they have gotten HIV. This might help us find out if a person's infection fighting cells can ever prevent someone from getting HIV. Some people may not be able to join the research study because of information found during the screening process.

After the screening process, you will find out if you are eligible for the research study. If you are eligible, the study staff will fully explain the research study to you and answer any questions you have. After the research study has been fully explained to you and if you decide to participate, you will be asked to sign another consent form.

[*Note to US sites: do not include this section since it is not relevant to your clinical setting]*: After the testing done today you may not be eligible for the study, or you may decide not to continue with the screening process. Either way, we will ask you if we can use part of the blood you gave today for the same reasons described above, including studying your genes. Genes, called DNA, tell us about the way your body is programmed to work. For instance a person that is very tall probably has different genes than a person who is very short, or a person whose ancestors come from a certain place probably has different genes compared to the genes of a person from a far off land. That is what makes each of us different. We will also be looking at your genes to study your body's reaction to infections, including how genes turn on or off. Understanding these "genetic" differences may help us understand how the human body responds to HIV, and to learn more on how to prevent HIV infection. You will not be told these results because they will not directly impact your health.

The researchers using the blood collected during the screening process are working with the NIH funded Center for HIV AIDS Vaccine Immunology (CHAVI), including the doctors at this *[insert clinic or hospital]*. They are also located at other institutions, or at companies that produce drugs to help people. The institutions and companies are located all over the world, so your samples may be sent to other countries. Some of the samples collected may be used to develop commercial products that will help people, like new drugs, vaccines, or tests. You will not be provided with financial compensation if a commercial product is made from research that used your samples. One or more of the institutions doing research using your samples will get the rights to any medicines, vaccines, or tests that might be made from research using your samples – this is called "intellectual property." The institutions or researchers that own this intellectual property may also get compensated from the sale of the medicines, vaccines, or tests that were made from research using your samples.

PROCEDURES:

If you agree to the screening process, you may be asked to come back to the clinic more than one time over the next couple of weeks.

During these visits:

- We will ask you where you live and how to find you.
- We will ask you questions about you like your age and your ethnic background, if you have any sexual partners who you would be willing to ask to bring in to the clinic for HIV testing. [*Malawi sites also include*: *And if you agree, we may ask you some additional questions about your sexual practices that may help researchers figure out how you may have come in to contact with HIV*.
- We will talk with you and your partner separately (if they are here with you) about HIV and we will provide you with information about how to protect yourself from getting HIV or preventing the spread of HIV. We will not share any information that you or your partner provide to us with the other (unless you request couples counseling). All information will be kept confidential.
- *[Include here any blood testing required for enrollment that has not already occurred through your standard of care at the clinical site, e.g. HIV testing, hemoglobin. If performing HIV*

testing and hemoglobin, add the following: We will draw some of your blood (no more than 10 mL, which is about 2 teaspoons [*change to local equivalent, if appropriate, also, if hemoglobin testing only, change to amount necessary*]). This blood will be tested to see if you are infected with HIV, and to measure the health of your blood. During the screening process we may need to test you more than once for HIV and hemoglobin. Before we draw your blood, we will talk with you about the HIV test, what it may mean to know your HIV status, and whether you are prepared to receive your HIV test result. Sometimes an HIV test is not clearly positive or negative. If this happens, we will test your blood again until we know the result for sure. We will tell you if your HIV test is positive or negative.

You may be eligible to participate in this study if you are infected with HIV or not.

If you have HIV, this clinic [*can or cannot*] provide you with drugs to prevent or treat infections related to HIV. However, the clinic [*can or cannot*] provide anti-HIV drugs. In order to receive anti-HIV drugs, you would have to buy the drugs from a private doctor or local pharmacy. [*This paragraph should be changed to be site-specific, indicating what kind of HIV-care can be normally provided by the clinic to HIV-infected individuals*].

RISKS and/or DISCOMFORTS:

If you participate in the screening, there are a few risks or discomforts you should know about.

If you have your blood drawn, you may feel discomfort, dizzy, or even faint when your blood is drawn. Redness, pain, swelling, bruising, or an infection may occur where the needle goes into your arm.

You may become embarrassed, worried, or anxious talking about HIV or sex, or discussing or waiting for your test results. Learning that you have HIV may make you worried or anxious. A trained counselor will help you deal with any feelings or questions you may have.

We will make every effort to protect your privacy and confidentiality while you are being screened for this study. Your visits here will take place in private. It is possible that others may learn of your participation here, and think you have HIV. Because of this, others may treat you unfairly. For example, you could have problems getting or keeping a job. You could also have problems being accepted by your family or community.

When tests are done on the samples there is a small but possible risk to your privacy. It is possible that if others found out information about you that is learned from tests (such as information about your genes) it could cause you problems with your family (having a family member learn about a disease that may be passed on in families or learning who is the true parent of a child) or problems getting a job or insurance, but this would be very rare.

POTENTIAL BENEFITS:

You may get no direct benefit from the screening procedures. However, you will receive counseling about HIV and information on your HIV status *[include only if HIV testing]*. You will receive information about how to prevent the spread of HIV and you will get free condoms. If you are infected with HIV, you will be told where you can receive health care, counseling, and other services, as well as information about other research studies.

ALTERNATIVES TO PARTICIPATION:

If you choose not to take part in the screening, it will have no effect on your regular health care at this clinic.

NEW FINDINGS:

You will be told of any new information learned during the course of the screening process that might cause you to change your mind about participating in the screening process.

REASONS WHY THE SCREENING TESTS MAY NOT BE COMPLETED:

You may not be eligible for the screening tests for the following reasons:

- The study is stopped or cancelled.
- Undergoing the screening tests would be harmful to you.
- You are not willing to find out your HIV test result.

COSTS AND COMPENSATION:

There is no cost to you for the screening. At the end of each visit, you will be given [*insert amount of money or incentive package to compensate participant for food, travel expenses, lost work time, etc. Be mindful of fluctuating amounts of your local currency, and therefore you may want to put a range of reimbursement.]*

CONFIDENTIALITY:

Efforts will be made to keep your screening records and test results confidential to the extent permitted by law. However, we cannot guarantee confidentiality. You will be identified by a code (a number), and personal information from your records will not be released without your written permission. You will not be personally identified in any publication about this study. However, your records may be reviewed, under guidelines of the United States Federal Privacy Act [*U.S. sites only];* the sponsor of the study (United States National Institutes of Health [NIH]), the [*insert name of site*] Institutional Review Board (IRB)/Ethics Committee (EC), study staff, study monitors, and [*insert applicable local authorities*].

Your blood will be kept in a secure place without your name. When other researchers are given your samples, they will not be given your personal information. The results of tests will not be included in your health records.

If during the course of these screening tests, we find out that you have [*insert all applicable reportable diseases (e.g., HIV)*], we must report it to [*insert the name(s) of the local health authorities*]. Although we must report that we have treated someone with [*insert all applicable reportable diseases*], your name will not be reported to the agency. [*Amend this paragraph to reflect the local requirements. If there are no local requirements to report communicable diseases, delete this paragraph.*]

RESEARCH-RELATED INJURY:

[*Site-specific: insert institutional policy*] It is unlikely that you will be injured as a result of having the screening procedure. If you are injured, the [*institution]* will give you immediate necessary treatment for

your injuries. You [will/will not] have to pay for this treatment. You will be told where you can get additional treatment for your injuries. If you require additional treatment, you will be told where you can get it because there is no program for monetary compensation or other forms of compensation for additional treatment either through this institution or the U.S. National Institutes of Health (NIH). You do not give up any legal rights by signing this consent form.

PROBLEMS or QUESTIONS:

For questions about this study or a research-related injury, contact:

[*insert name of the Investigator of Record or other study staff*]

[*insert telephone number and physical address of above*]

For questions about your rights as a research subject, contact:

[insert name or title of person on the Institutional Review Board (IRB) or other organization appropriate for the site]

[*insert telephone number and physical address of above*]

SIGNATURE PAGE: SCREENING INFORMED CONSENT FORM

CHAVI 001: Acute HIV-1 Infection Prospective Cohort Study, Version 2.0

If you have read this informed consent, or have had it read and explained to you, and understand the information, and you voluntarily agree to undergo the screening procedures for this research study, please sign your name or make your mark below.

_____ I agree to the screening procedures. I also agree to have samples of my blood used for additional testing related to HIV infection (including genetic testing).

_____ I agree to the screening procedures. I also agree to have samples of my blood used for additional testing related to HIV infection, but not genetic testing.

_____ I agree to the screening procedures, but do not agree to have samples of my blood used for additional testing related to HIV infection.

_____ I agree to the screening procedures. [*This option only for US sites]*

_____	_____
Participant Name (print)	Participant Signature and Date
_____	_____
Study Staff Conducting Consent Discussion (print)	Study Staff Signature and Date
_____	_____
Witness Name (print) (As appropriate)	Witness Signature and Date

SAMPLE INFORMED CONSENT FORM

DIVISION OF AIDS, NIAID, NIH

CHAVI 001: Acute HIV-1 Infection Prospective Cohort Study, Version 2.0

STUDY ENROLLMENT INFORMED CONSENT FORM – ACUTE HIV-1 INFECTION

PRINCIPAL INVESTIGATOR: [*insert name*]
PHONE: [*insert number*]

INTRODUCTION:

You are being asked to volunteer for the research study named above because you may have recently been infected with HIV. This study is sponsored by the U.S. National Institutes of Health. The person in charge of the study at this site is [*insert name of Investigator of Record*].

Before you decide whether or not to take part in this research study, you need to know the purpose, the possible risks and benefits to you, and what will be expected of you during the study. This consent form provides that information. The study staff will discuss the information with you. They will answer any questions you may have. After the study has been fully explained to you, you can decide whether or not you want to participate. Once you understand this study, and if you agree to take part, you will be asked to sign this consent form or make your mark in front of someone. You will be offered a copy of this form to keep.

Please note that:

- Your participation in this study is entirely voluntary.
- You may decide not to take part or to withdraw from this study at any time without losing the benefits of your or your partner's standard health care. In addition, if you withdraw from the study and decide you do not want the specimens already provided by you to be tested, it is important that you let the study staff know.

PURPOSE OF THE STUDY:

[Institution] is part of a group of scientists from all over the world doing research on how HIV works in the body. It is hoped that this research will help to make medicines to prevent the spread of HIV. HIV is the virus that causes AIDS. The purpose of this research study is to understand how people's infection fighting (immune) system reacts to HIV. Your body is made-up of the cells, proteins, and other chemicals that help to fight infection. We want to know how the body, including blood and other fluids from the body, works if someone has or comes into contact with HIV. We also want to examine the genes in your body, called DNA, since they might affect how your body responds to HIV. For instance, a person that is very tall probably has different genes than a person who is very short, or a person whose ancestors come from a certain place probably have different genes compared to the genes of a person from a far off land. No two persons in the world have exactly the same genes. That is what makes each of us different. We will also be looking at your genes to study your body's reaction to infections, including how genes turn on or off. Understanding these "genetic" differences may help us understand how the human body responds to HIV. The kind of testing described here is called experimental testing. You will not be told these results because they will not directly impact your health. We will do other testing in this study that is directly related to your health which will be shared with you.

The study will initially take place in Africa and the United States. Other locations may be added after the study has started. Up to approximately 1300 people will participate in this research study. The whole study will take up to 7 years to finish. Each person in the group you are being asked to join will be in the study for up to 2 years.

If you have a sexual partner not already enrolled in this study, we will ask you if you would be willing to ask your sexual partner(s) to come in for HIV testing and possible enrollment in the study. The information that you and your partner provide for the study will not be shared with the other to protect your confidentiality.

The research done on your samples will be reviewed by the United States National Institutes of Health.

Additional Information about the Experimental Testing in this Study:

The experimental testing being done in this study is by researchers working with the NIH funded Center for HIV AIDS Vaccine Immunology (CHAVI), including the doctors at this *[insert clinic or hospital]*. They are located at other institutions, or at companies that produce drugs to help people. The institutions and companies are located all over the world, so your samples may be sent to other countries. Some of the samples collected may be used to develop commercial products that will help people, like new drugs, vaccines, or tests. You will not be provided with financial compensation if a commercial product is made from research that used your samples. One or more of the institutions doing research using your samples will get the rights to any medicines, vaccines, or tests that might be made from research using your samples – this is called "intellectual property." The institutions or researchers that own this intellectual property may also get compensated from the sale of the medicines, vaccines, or tests that were made from research using your samples.

Samples to be Tested:

In order to be in this study you must agree to provide blood for testing that is directly related to your health, and for experimental testing. We would also like to test the following samples if you agree to it – [*list here the optional samples that your site will include in your site-specific consent based on the following choices: semen if you are a man, samples from your vagina if you are a woman, breast milk if you are breastfeeding, feces. urine, and saliva]*. You do not have to agree to the additional samples in order to be in the study. You will indicate your choices about these other samples at the end of this informed consent form. These other samples are for experimental testing only.

You will be told of the results of the tests that are done for your health, like results of HIV testing. The researchers do not plan to contact you or this study site with the results from any of the experimental tests. This is because the experimental tests are for research purposes only and are not directly related to your health.

Samples Leftover at the End of the Study:

Once all the experimental testing is done for this research study, there may be some leftover samples. We would like to use the leftover samples for future research that is not a part of this study, but would be used for HIV-related research only. Some of this may include genetic testing. An Institutional Review Board or Ethics Committee, which watches over the safety and rights of research participants, must approve any research studies using your samples. There is no time limit on how long these samples will be stored.

If you agree to have these samples stored for future research, they will be stored safely and securely in a storage facility. Only the people who work at the facility and approved researchers will have access

to your samples. The people who work at the facility will not have any information that identifies you. The approved researchers may be given information about you such as your age and sex, but they will not be given your name or any other information that identifies you. There is no time limit on how long your samples will be stored. At the end of this consent form you will indicate whether you agree to this future testing. If you do not agree to store your leftover samples, your samples will be destroyed once all the testing for this study has ended.

PROCEDURES:

If you agree to join this study, you will be asked to come back on a regular basis. Including your visit today, you will return to the clinic for at least 15 visits; however, we may ask you to return for additional visits if you are sick.

Study Visits:

During the first study visit, the study will be explained to you. You will have time to ask questions and discuss any concerns you may have with the study staff. The first visit will last up to 2 hours. All other visits will last up to 30-60 minutes.

During your first study visit (called the enrollment visit), the study will be explained to you. You will have time to ask questions and discuss any concerns you may have with the study staff. No procedures will be done to you until you have read and signed this informed consent form.

The following activities will happen at each visit, unless otherwise indicated.

- The study staff will ask for your age, ethnic background, and other information about you (enrollment visit only).
- The study staff will ask you where you live and how to find you.
- The study staff will ask if you have any health problems (every 3 months).
- The study staff will ask you questions about your sexual practices (every 3 months).
- You will receive a physical exam (every 3 months).
- Give blood as follows:
  - At the first visit, give 100 ml of blood, which is about 20 teaspoons [or local equivalent], at the next 5 visits, give 60 ml of blood at each visit, which is about 12 teaspoons of blood, and the remaining 9 visits, give 75 ml of blood, which is about 15 teaspoons of blood.
- The following will happen if you have agreed to give these samples:
  [*NOTE to Sites: include below <u>only</u> the optional samples you intend to collect*]
  - If you are female, the study staff will collect fluid from your vagina.
  - If you are a breastfeeding female, you will be asked to express up to 50 ml (10 teaspoons) [or local equivalent] of breast milk at up to 4 visits.
  - If you are male, you will be asked to give a semen sample.
  - Give urine, feces, and saliva *[sites to choose which ones]* at Enrollment, Week 1, 2, 3, and 4
- You will be given free condoms.
- You will receive counseling, and talk with study staff about ways to avoid passing HIV and other infection during sex.
- You will receive referrals for medical care and other services if you need them.

The study staff may need to examine you or do tests on you for your health at any of your visits in order to take care of you. We will tell you the results of any tests we do here at this clinic as soon as they become available. If we find any infections or other conditions during your physical examination or from

the laboratory tests we do here at this clinic, you will receive free treatment for the conditions to the extent possible, or you will be referred to care at [*insert local treatment facilities here*].

If you decide to leave the study before your planned study end date, you will be asked to have a last study visit that may include some or all the exams and tests listed above.

RISKS and/or DISCOMFORTS:

You may feel discomfort, dizzy, or even faint when your blood is drawn. Redness, pain, swelling, bruising, or an infection may occur where the needle goes into your arm.

You may feel discomfort during the examination of your vagina or penis.

You may become embarrassed, worried, or anxious when discussing your sexual practices and HIV. A trained counselor will help you deal with any feeling or questions you have.

We will make every effort to protect your privacy and confidentiality while you are in the study. Your visits here will take place in private. However, it is possible that others may learn of your participation here. Because of this, others may treat you unfairly or discriminate against you. For example, you could have problems getting or keeping a job. You could also have problems being accepted by your family or community.

If you choose to have leftover samples stored, there are few risks related to storing your samples. When tests are done on the stored samples there is a small but possible risk to your privacy. It is possible that if others found out information about you that is learned from tests (such as information about your genes) it could cause you problems with your family (having a family member learn about a disease that may be passed on in families or learning who is the true parent of a child) or problems getting a job or insurance, but this would be very rare.

POTENTIAL BENEFITS:

There may be no direct benefit to you from this study. However, you and others could benefit in the future from research done in this study, because we will learn more about HIV. The information we learn may help researchers understand better how to make vaccines or medicines to prevent or treat diseases like HIV.

Knowing early in the course of infection that you have HIV may help you to prevent passing the infection to others. You will also get physical exams, information about your health, and the opportunity to talk to counselors about your health, your feelings, and ways to prevent spreading HIV. If these exams show that you have a health problem, we will treat you to the extent possible, or we will tell you where you can go for treatment. You will also receive free condoms throughout the entire course of the study.

NEW FINDINGS

You will be told any new information learned during this study that might cause you to change your mind about staying in the study. You will be told when the results of the study may be available, and how to learn about them.

REASONS WHY YOU MAY BE WITHDRAWN FROM THE STUDY WITHOUT YOUR CONSENT

You may be removed from the study without your consent for the following reasons:

- The study may be discontinued at any time by the sponsor of the study (NIH), the U.S. Office of Human Research and Protection, the CHAVI Scientific Leadership Group, in-country government or regulatory authorities, and/or the IRB/EC overseeing research at the study site.
- The study staff feels that staying in the study would be harmful to you.
- Other administrative reasons.

ALTERNATIVES TO PARTICIPATION

If you choose not to participate in this study it will not impact the health care you can receive at this clinic. [*Sites to include/amend the following if applicable:* There may be other studies going on here or in the community that you may be eligible for. If you wish, we will tell you about other studies that we know about. There also may be other places where you can go for HIV counseling and testing. We will tell you about those places if you wish.]

COSTS AND COMPENSATION:

There is no cost to you for being in this study. [*Insert if this applies to your site*: At the end of each visit, you will be given [*insert amount of money, incentive package to compensate participant for food, travel expenses, lost work time, etc.* ***Be mindful of fluctuating amounts of your local currency****]*

CONFIDENTIALITY:

Efforts will be made to keep your study records and test results confidential to the extent permitted by law. However, we cannot guarantee confidentiality. You will be identified by a code (a number), and personal information from your records will not be released without your written permission. You will not be personally identified in any publication about this study. However, your records may be reviewed, under guidelines of the United States Federal Privacy Act [*U.S. sites only];* the sponsor of the study (United States National Institutes of Health [NIH]), the [*insert name of site*] Institutional Review Board (IRB)/Ethics Committee (EC), study staff, study monitors, and [*insert applicable local authorities*].

Blood and, if you agree, *[Sites to add here the optional samples intend to be collected: breast milk, semen, vaginal fluid samples, urine, feces, and saliva* collected during this study will be tested for experimental research purposes. These samples will be put into containers that do not have your name on them but that use a code to protect your privacy. The code can only be traced back to your study clinic; however, your name, where you live, and other personal information will be protected by the study clinic. When researchers are given your samples, they will not be given your personal information. If you agree to have any leftover samples stored for future research, these samples will be stored without your name and the results of any future tests will not be included in your health records.

[Include only if applicable – If during the course of the study we find out that you have [*insert all applicable reportable diseases (e.g., STI's, HIV)*], we must report it to [*insert the name(s) of the local health authorities*]. Although we must report that we have treated someone with [*insert all applicable reportable diseases*], your name will not be reported to the agency. [*Amend this paragraph to reflect the local requirements. If there are no local requirements to report communicable diseases, delete this paragraph.*]

[For U.S. sites only:] In addition to the efforts made by the study staff to keep your personal information confidential, a Certificate of Confidentiality has been obtained from the U.S. Federal Government for this study. This Certificate protects study staff from being forced to tell people who are not connected with this study, such as the court system, about your participation or information you give for study purposes. Even with the Certificate of Confidentiality, however, if the study staff learn of possible child abuse and/or neglect or a risk of harm to you or others, they will be required to tell the proper authorities. Having a Certificate of Confidentiality does not prevent you from releasing information about yourself and your participation in the study.

RESEARCH-RELATED INJURY:

The study staff will monitor your health closely while you are in this study. If you have any health problems between visits, please contact the study staff. If you have a medical emergency that requires immediate care, [insert site-specific instructions].

[Sites to specify institutional policy:] If you are injured as a result of being in this study, the [institution] will give you immediate necessary treatment for your injuries. You [will/will not] have to pay for this treatment. If you need additional treatment for your injuries you will be told where you can go because there is no program for monetary compensation or other forms of compensation for such injuries either through this institution or the U.S. National Institutes of Health (NIH). You do not give up any legal rights by signing this consent form.

PROBLEMS or QUESTIONS:

For questions about this study or a research-related injury, contact:

[*insert name of the Investigator of Record or other study staff*]

[*insert telephone number and physical address of above*]

For questions about your rights as a research subject, contact:

[insert name or title of person on the Institutional Review Board (IRB) or other organization appropriate for the site]

[*insert telephone number and physical address of above*]

SIGNATURE PAGE: STUDY INFORMED CONSENT FORM – ACUTE HIV-1 INFECTION

CHAVI 001: Acute HIV-1 Infection Prospective Cohort Study, Version 2.0

If you have read this informed consent, or have had it read and explained to you, and understand the information, and you voluntarily agree to participate in this research study, please sign your name or make your mark in the signature area at the bottom of this page. By providing your initials in the spaces below you may also agree to additional sample collection, genetic testing, or long-term storage.

____ I agree to allow genetic testing of my blood during the study.

____ I do not agree to allow genetic testing of my blood during the study.

I also agree to provide the following additional samples for experimental testing during the study:

[NOTE TO SITES: INCLUDE ONLY THE SAMPLES YOU INTEND TO COLLECT]:

Genital secretions:	____**including** genetic testing	____**excluding** genetic testing
Breast milk:	____**including** genetic testing	____**excluding** genetic testing
____Saliva	____Urine	____Feces

____I do not agree to provide any of the additional samples listed above.

I agree to allow the following leftover samples to be saved for future testing:

[NOTE TO SITES: THIS LIST SHOULD MATCH THE LIST ABOVE]

Blood:	____**including** genetic testing	____**excluding** genetic testing
Genital secretions:	____**including** genetic testing	____**excluding** genetic testing
Breast milk:	____**including** genetic testing	____**excluding** genetic testing
____Saliva	____Urine	____Feces

____I do not agree to allow any of my leftover samples to be saved for future testing.

_____	_____
Participant Name (print)	Participant Signature and Date
_____	_____
Study Staff Conducting Consent Discussion (print)	Study Staff Signature and Date
_____	_____
Witness Name (print) (As appropriate)	Witness Signature and Date

SAMPLE INFORMED CONSENT FORM

DIVISION OF AIDS, NIAID, NIH

CHAVI 001: Acute HIV-1 Infection Prospective Cohort Study, Version 2.0

STUDY ENROLLMENT INFORMED CONSENT FORM – ESTABLISHED HIV-1 INFECTION

PRINCIPAL INVESTIGATOR: [*insert name*]
PHONE: [*insert number*]

INTRODUCTION:

You are being asked to volunteer for the research study named above because you are infected with HIV. This study is sponsored by the U.S. National Institutes of Health. The person in charge of the study at this site is [*insert name of principal investigator*].

Before you decide whether or not to take part in this research study, you need to know the purpose, the possible risks and benefits to you, and what will be expected of you during the study. This consent form provides that information. The study staff will discuss the information with you. They will answer any questions you may have. After the study has been fully explained to you, you can decide whether or not you want to participate. Once you understand this study, and if you agree to take part, you will be asked to sign this consent form or make your mark in front of someone. You will be offered a copy of this form to keep.

Please note that:

- Your participation in this study is entirely voluntary.
- You may decide not to take part or to withdraw from this study at any time without losing the benefits of your or your partner's standard health care. In addition, if you withdraw from the study and decide you do not want the specimens already provided by you to be tested, it is important that you let the study staff know.

PURPOSE OF THE STUDY:

[Institution] is part of a group of scientists from all over the world doing research on how HIV works in the body. It is hoped that this research will help to make medicines to prevent the spread of HIV. HIV is the virus that causes AIDS. The purpose of this research study is to understand how people's infection fighting (immune) system reacts to HIV. Your body is made-up of the cells, proteins, and other chemicals that help to fight infection. We want to know how the body, including blood and other fluids from the body, works if someone has or comes into contact with HIV. We also want to examine the genes in your body, called DNA, since they might affect how your body responds to HIV. For instance, a person that is very tall probably has different genes than a person who is very short, or a person whose ancestors come from a certain place probably have different genes compared to the genes of a person from a far off land. No two persons in the world have exactly the same genes. That is what makes each of us different. We will also be looking at your genes to study your body's reaction to infections, including how genes turn on or off. Understanding these "genetic" differences may help us understand how the human body responds to HIV. The kind of testing described here is called experimental testing. You will not be told these results because they will not directly

impact your health. We will do other testing in this study that is directly related to your health which will be shared with you.

The study will initially take place in Africa and the United States. Other locations may be added after the study has started. Up to approximately 1300 people will participate in this research study. The whole study will take up to 7 years to finish. Each person in the group you are being asked to join will be in the study for up to 2 years.

The research done on your samples will be reviewed by the United States National Institutes of Health.

Additional Information about the Experimental Testing in this Study:

The experimental testing being done in this study is by researchers working with the NIH funded Center for HIV AIDS Vaccine Immunology (CHAVI), including the doctors at this *[insert clinic or hospital]*. They are located at other institutions, or at companies that produce drugs to help people. The institutions and companies are located all over the world, so your samples may be sent to other countries. Some of the samples collected may be used to develop commercial products that will help people, like new drugs, vaccines, or tests. You will not be provided with financial compensation if a commercial product is made from research that used your samples. One or more of the institutions doing research using your samples will get the rights to any medicines, vaccines, or tests that might be made from research using your samples – this is called "intellectual property." The institutions or researchers that own this intellectual property may also get compensated from the sale of the medicines, vaccines, or tests that were made from research using your samples.

Samples to be Tested:

In order to be in this study you must agree to provide blood for testing that is directly related to your health, and for experimental testing. We would also like to test the following samples if you agree to it – [*list here the optional samples that your site will include in your site-specific consent based on the following choices: semen if you are a man, samples from your vagina if you are a woman, breast milk if you are breastfeeding, feces. urine, and saliva]*. You do not have to agree to the additional samples in order to be in the study. You will indicate your choices about these other samples at the end of this informed consent form. These other samples are for experimental testing only.

You will be told of the results of the tests that are done for your health, like results of HIV testing. The researchers do not plan to contact you or this study site with the results from any of the experimental tests. This is because the experimental tests are for research purposes only and are not directly related to your health.

Samples Leftover at the End of the Study:

Once all the experimental testing is done for this research study, there may be some leftover samples. We would like to use the leftover samples for future research that is not a part of this study, but would be used for HIV-related research only. Some of this may include genetic testing. An Institutional Review Board or Ethics Committee, which watches over the safety and rights of research participants, must approve any research studies using your samples. There is no time limit on how long these samples will be stored.

If you agree to have these samples stored for future research, they will be stored safely and securely in a storage facility. Only the people who work at the facility and approved researchers will have access to your samples. The people who work at the facility will not have any information that identifies you. The approved researchers may be given information about you such as your age

and sex, but they will not be given your name or any other information that identifies you. There is no time limit on how long your samples will be stored. At the end of this consent form you will indicate whether you agree to this future testing. If you do not agree to store your leftover samples, your samples will be destroyed once all the testing for this study has ended.

PROCEDURES:

If you agree to join this study, you will be asked to come back on a regular basis. Including your visit today, you will be to return to the clinic for at least 10 visits; however, we may ask you to return for additional visits if you are sick.

Study Visits:

During the first study visit, the study will be explained to you. You will have time to ask questions and discuss any concerns you may have with the study staff. The first visit will last up to 2 hours. All other visits will last up to 30-60 minutes.

The following activities will happen at each visit, unless otherwise indicated:

- The study staff will ask for your age, ethnic background, and other information about you (enrollment visit only).
- The study staff will ask you where you live and how to find you.
- The study staff will ask if you have any health problems (enrollment, and 3, 6, 12, 18, and 24 month visits).
- The study staff will ask you questions about your sexual practices (enrollment, and 3, 6, 12, 18, and 24 month visits).
- You will receive a physical exam at enrollment, 3, 6, 12, 18, and 24 month visits.
- Give blood as follows:
  - At the first 3 visits, give 60 ml of blood, which is about 12 teaspoons [or local equivalent], and at all other visits, give 75 ml of blood, which is about 15 teaspoons [or local equivalent].
- The following will happen if you have agreed to give the following samples:
  [*NOTE to Sites: include below <u>only</u> the optional samples you intend to collect*]
  - If you are female, the study staff will collect fluid from your vagina.
  - If you are a breastfeeding female, you will be asked to express up to 50 ml (10 teaspoons) [or local equivalent] of breast milk at up to 4 visits.
  - If you are male, you will be asked to give a semen sample.
  - Give urine, feces, and saliva (*sites to choose which ones*) at enrollment only.
- You will be given free condoms.
- You will receive counseling, and talk with study staff about ways to avoid passing HIV and other infection during sex.
- You will receive referrals for medical care and other services if you need them.

The study staff may need to examine you or do tests on you for your health at any of your visits in order to take care of you. We will tell you the results of any tests we do at this clinic as soon as they become available. If we find any infections or other conditions during your physical examination or from the laboratory tests that we do here at this clinic, you will receive free treatment for the conditions to the extent possible, or you will be referred to care at [*insert local treatment facilities here*].

If you decide to leave the study before your planned study end date, you will be asked to have a last study visit that may include some or all the exams and tests listed above.

RISKS and/or DISCOMFORTS:

You may feel discomfort, dizzy, or even faint when your blood is drawn. Redness, pain, swelling, bruising, or an infection may occur where the needle goes into your arm.

You may feel discomfort during the examination of your vagina or penis.

You may become embarrassed, worried, or anxious when discussing your sexual practices and HIV. A trained counselor will help you deal with any feeling or questions you have.

We will make every effort to protect your privacy and confidentiality while you are in the study. Your visits here will take place in private. However, it is possible that others may learn of your participation here. Because of this, others may treat you unfairly or discriminate against you. For example, you could have problems getting or keeping a job. You could also have problems being accepted by your family or community.

If you chose to have leftover samples stored, there are few risks related to storing your samples. When tests are done on the stored samples there is a small but possible risk to your privacy. It is possible that if others found out information about you that is learned from tests (such as information about your genes) it could cause you problems with your family (having a family member learn about a disease that may be passed on in families or learning who is the true parent of a child) or problems getting a job or insurance, but this would be very rare.

POTENTIAL BENEFITS:

There may be no direct benefit to you from this study. However, you and others could benefit in the future from research done in this study, because we will learn more about HIV. The information we learn may help researchers better understand how to make vaccines or medicines to prevent or treat diseases like HIV.

By knowing early in the course of infection that you have HIV may help you to prevent passing the infection to others. You will also get physical exams, information about your health, and the opportunity to talk to counselors about your health, your feelings, and ways to prevent spreading HIV. If these exams show that you have a health problem, we will treat you to the extent possible, or we will tell you where you can go for treatment. You will also receive free condoms throughout the entire course of the study.

NEW FINDINGS

You will be told any new information learned during this study that might cause you to change your mind about staying in the study. You will be told when the results of the study may be available, and how to learn about them.

REASONS WHY YOU MAY BE WITHDRAWN FROM THE STUDY WITHOUT YOUR CONSENT

You may be removed from the study without your consent for the following reasons:

- The study may be discontinued at any time by the sponsor of the study (NIH), the U.S. Office of Human Research and Protection, the CHAVI Scientific Leadership Group, in-country government or regulatory authorities, and/or the IRB/EC overseeing research at the study site.
- The study staff feels that staying in the study would be harmful to you.

- Other administrative reasons.

ALTERNATIVES TO PARTICIPATION

If you choose not to participate in this study it will not impact the health care you can receive at this clinic. [*Sites to include/amend the following if applicable:* There may be other studies going on here or in the community that you may be eligible for. If you wish, we will tell you about other studies that we know about. There also may be other places where you can go for HIV counseling and testing. We will tell you about those places if you wish.]

COSTS AND COMPENSATION:

There is no cost to you for being in this study. Treatments available to you from the study for infections passed during sex will be given to you free of charge. At the end of each visit, you will be given [*insert amount of money, incentive package to compensate participant for food, travel expenses, lost work time, etc.* ***Be mindful of fluctuating amounts of your local currency****]*

CONFIDENTIALITY:

Efforts will be made to keep your study records and test results confidential to the extent permitted by law. However, we cannot guarantee confidentiality. You will be identified by a code (a number), and personal information from your records will not be released without your written permission. You will not be personally identified in any publication about this study. However, your records may be reviewed, under guidelines of the United States Federal Privacy Act [*U.S. sites only];* the sponsor of the study (United States National Institutes of Health [NIH]), the [*insert name of site*] Institutional Review Board (IRB)/Ethics Committee (EC), study staff, study monitors, and [*insert applicable local authorities*].

Blood and, if you agree, *[Sites to include here the optional samples intended to be collected: breast milk, semen, vaginal fluid samples, urine, feces, and saliva]* samples collected during this study will be tested for experimental research purposes. These samples will be put into containers that do not have your name on them but that use a code to protect your privacy. The code can only be traced back to your study clinic; however, your name, where you live, and other personal information will be protected by the study clinic. When researchers are given your samples, they will not be given your personal information. If you agree to have any leftover samples stored for future research, these samples will be stored without your name and the results of any future tests will not be included in your health records.

[Include only if applicable – If during the course of the study we find out that you have [*insert all applicable reportable diseases (e.g., HIV)*], we must report it to [*insert the name(s) of the local health authorities*]. Although we must report that we have treated someone with [*insert all applicable reportable diseases*], your name will not be reported to the agency. [*Amend this paragraph to reflect the local requirements. If there are no local requirements to report communicable diseases, delete this paragraph.*]

[For U.S. sites only:] In addition to the efforts made by the study staff to keep your personal information confidential, a Certificate of Confidentiality has been obtained from the U.S. Federal Government for this study. This Certificate protects study staff from being forced to tell people who are not connected with this study, such as the court system, about your participation or information you give for study purposes. Even with the Certificate of Confidentiality, however, if the study staff learn of possible child abuse and/or neglect or a risk of harm to you or others, they will be required to tell the proper authorities.

Having a Certificate of Confidentiality does not prevent you from releasing information about yourself and your participation in the study.

RESEARCH-RELATED INJURY:

The study staff will monitor your health closely while you are in this study. If you have any health problems between visits, please contact the study staff. If you have a medical emergency that requires immediate care, *[insert site-specific instructions*].

[Sites to specify institutional policy:] If you are injured as a result of being in this study, the [institution] will give you immediate necessary treatment for your injuries. You [will/will not] have to pay for this treatment. If you need additional treatment for your injuries you will be told where you can go because there is no program for monetary compensation or other forms of compensation for such injuries either through this institution or the U.S. National Institutes of Health (NIH). You do not give up any legal rights by signing this consent form.

PROBLEMS or QUESTIONS:

For questions about this study or a research-related injury, contact:

[*insert name of the Investigator of Record other study staff*]

[*insert telephone number and physical address of above*]

For questions about your rights as a research subject, contact:

[insert name or title of person on the Institutional Review Board (IRB) or other organization appropriate for the site]

[*insert telephone number and physical address of above*]

SIGNATURE PAGE: STUDY INFORMED CONSENT FORM
ESTABLISHED HIV-1 INFECTION

CHAVI 001: Acute HIV-1 Infection Prospective Cohort Study, Version 2.0

If you have read this informed consent, or have had it read and explained to you, and understand the information, and you voluntarily agree to participate in this research study, please sign your name or make your mark in the signature area at the bottom of this page. By providing your initials in the spaces below you may also agree to additional sample collection, genetic testing, or long-term storage.

____ I agree to allow genetic testing of my blood during the study.

____ I do not agree to allow genetic testing of my blood during the study.

I also agree to provide the following additional samples for experimental testing during the study:

[NOTE TO SITES: INCLUDE ONLY THE SAMPLES YOU INTEND TO COLLECT]:

Genital secretions:	____**including** genetic testing	____**excluding** genetic testing
Breast milk:	____**including** genetic testing	____**excluding** genetic testing
____Saliva	____Urine	____Feces

____I do not agree to provide any of the additional samples listed above.

I agree to allow the following leftover samples to be saved for future testing: ***[NOTE TO SITES: THIS LIST SHOULD MATCH THE LIST ABOVE]***

Blood:	____**including** genetic testing	____**excluding** genetic testing
Genital secretions:	____**including** genetic testing	____**excluding** genetic testing
Breast milk:	____**including** genetic testing	____**excluding** genetic testing
____Saliva	____Urine	____Feces

____I do not agree to allow any of my leftover samples to be saved for future testing.

_____	_____
Participant Name (print)	Participant Signature and Date
_____	_____
Study Staff Conducting Consent Discussion (print)	Study Staff Signature and Date
_____	_____
Witness Name (print) (As appropriate)	Witness Signature and Date

SAMPLE INFORMED CONSENT FORM

DIVISION OF AIDS, NIAID, NIH

CHAVI 001: Acute HIV-1 Infection Prospective Cohort Study, Version 2.0

STUDY ENROLLMENT INFORMED CONSENT FORM – HIV-1 NEGATIVE

PRINCIPAL INVESTIGATOR: [*insert name*]
PHONE: [*insert number*]

INTRODUCTION:

You are being asked to volunteer for the research study named above because you are HIV negative. This study is sponsored by the United States National Institutes of Health. The person in charge of the study at this site is [*insert name of Investigator of Record]*

Before you decide whether to participate, we would like to explain the purpose of the study, the risks and benefits to you, and what is expected of you. The study staff will discuss the information with you. They will answer any questions you may have. After the study has been fully explained to you, you can decide whether or not you want to participate. If you understand the study and agree to participate, you will be asked to sign this consent form or make your mark in front of someone. You will be offered a copy of this form to keep.

Please note that:

- Your participation in the study is entirely voluntary.
- You may decide not to take part or to withdraw from the study at any time without losing the benefits of your standard health care. In addition, if you withdraw from the study and decide you do not want the specimens already provided by you to be tested, it is important that you let the study staff know.

PURPOSE OF THE STUDY:

[*Institution*] is part of a group of scientists from all over the world doing research on how HIV works in the body. It is hoped that this research will help to make medicines to prevent the spread of HIV. HIV is the virus that causes AIDS. The purpose of this research study is to understand how people's infection fighting (immune) system reacts to HIV. Your body is made-up of the cells, proteins, and other chemicals that help to fight infection. We want to know how the body, including blood and other fluids from the body, works if someone has or comes into contact with HIV. We also want to examine the genes in your body, called DNA, since they might affect how your body responds to HIV. For instance, a person that is very tall probably has different genes than a person who is very short, or a person whose ancestors come from a certain place probably have different genes compared to the genes of a person from a far off land. No two persons in the world have exactly the same genes. That is what makes each of us different. We will also be looking at your genes to study your body's reaction to infections, including how genes turn on or off. Understanding these "genetic" differences may help us understand how the human body responds to HIV. The kind of testing described here is called experimental testing. You will not be told these results because they will not directly impact your health. We will do other testing in this study that is directly related to your health which we will share with you.

The study will initially take place in Africa and the United States. Other locations may be added after the study has started. Up to approximately 1300 people will participate in this research study. The whole study will take up to 7 years to finish. Each person in the group you are being asked to join will be in the study for up to 2 years.

If you have a sexual partner not already enrolled in this study, we will ask you if you would be willing to ask your sexual partner(s) to come in for HIV testing and possible enrollment in the study.

The research done on your samples will be reviewed by the United States National Institutes of Health.

Additional Information about the Experimental Testing in this Study:

The experimental testing being done in this study is by researchers working with the NIH funded Center for HIV AIDS Vaccine Immunology (CHAVI), including the doctors at this *[insert clinic or hospital]*. They are located at other institutions, or at companies that produce drugs to help people. The institutions and companies are located all over the world, so your samples may be sent to other countries. Some of the samples collected may be used to develop commercial products that will help people, like new drugs, vaccines, or tests. You will not be provided with financial compensation if a commercial product is made from research that used your samples. One or more of the institutions doing research using your samples will get the rights to any medicines, vaccines, or tests that might be made from research using your samples – this is called "intellectual property." The institutions or researchers that own this intellectual property may also get compensated from the sale of the medicines, vaccines, or tests that were made from research using your samples.

Samples to be Tested:

In order to be in this study you must agree to provide blood for testing that is directly related to your health, and for experimental testing. We would also like to test the following samples if you agree to it – [*list here the optional samples that your site will include in your site-specific consent based on the following choices: semen if you are a man, samples from your vagina if you are a woman, breast milk if you are breastfeeding]*. You do not have to agree to the additional samples in order to be in the study. You will indicate your choices about these other samples at the end of this informed consent form. These other samples are for experimental testing only.

You will be told of the results of the tests that are done for your health, like results of HIV testing. The researchers do not plan to contact you or this study site with the results from any of the experimental tests. This is because the experimental tests are for research purposes only and are not directly related to your health.

Samples Leftover at the End of the Study:

Once all the experimental testing is done for this research study, there may be some leftover samples. We would like to use the leftover samples for future research that is not a part of this study, but would be used for HIV-related research only. Some of this may include genetic testing. An Institutional Review Board or Ethics Committee, which watches over the safety and rights of research participants, must approve any research studies using your samples. There is no time limit on how long these samples will be stored.

If you agree to have these samples stored for future research, they will be stored safely and securely in a storage facility. Only the people who work at the facility and approved researchers will have access to your samples. The people who work at the facility will not have any information that identifies you. The approved researchers may be given information about you such as your age

and sex, but they will not be given your name or any other information that identifies you. There is no time limit on how long your samples will be stored. At the end of this consent form you will indicate whether you agree to this future testing. If you do not agree to store your leftover samples, your samples will be destroyed once all the testing for this study has ended.

PROCEDURES:

If you agree to join this study, you will be asked to come back on a regular basis. Including your visit today, you will return to the clinic for at least 10 visits; however, we may ask you to return for additional visits if you are sick. The enrollment visit will take up to 2 hours. The remaining visits will take up to 30-60 minutes.

Study Visits:

During your first study visit (called the enrollment visit), the study will be explained to you. You will have time to ask questions and discuss any concerns you may have with the study staff. No procedures will be done to you until you have read and signed this informed consent form.

The following activities will happen at each study visit based, unless otherwise indicated:

- The study staff will ask for your age, ethnic background, and other information about you (enrollment visit only).
- The study staff will ask you where you live and how to find you.
- The study staff will ask if you have any health problems (enrollment, and at the 3, 6, 12, 18, and 24 month visit).
- The study staff will ask you questions about your sexual practices (enrollment, and 3, 6, 12, 18, and 24 month visits).
- You will receive a physical exam at enrollment, and the 3, 6, 12, 18, and 24 month visit.
- Give blood as follows:
  - At the first 4 visits, give 60 ml of blood, which is about 12 teaspoons [or local equivalent], and at all other visits, give 75 ml of blood, which is about 15 teaspoons [or local equivalent].
- The following will happen if you have agreed to give the following samples:

[*NOTE to Sites: include below <u>only</u> the optional samples you intend to collect*]

- If you are female, the study staff will collect fluid from your vagina.
- If you are a breastfeeding female, you will be asked to express up to 50 ml (10 teaspoons) [or local equivalent] of breast milk at up to 4 visits.
- If you are male, you will be asked to give a semen sample.

- You will be given free condoms.
- You will receive counseling, and talk with study staff about ways to avoid passing HIV and other infection during sex.
- You will receive referrals for medical care and other services if you need them.

The study staff may need to examine you or do tests on you for your health at any of your visits in order to take care of you. We will tell you the results of any tests we do at this clinic as soon as they become available. If we find any infections or other conditions during your physical examination or from the laboratory tests that we do here at this clinic, you will receive free treatment for the conditions to the extent possible, or you will be referred to care at [*insert local treatment facilities here*].

If you decide to leave the study before your planned study end date, you will be asked to have a last study visit that may include some or all the exams and tests listed above.

IF YOU BECOME INFECTED WITH HIV WHILE PARTICIPATING IN THIS STUDY:

If you become infected with HIV during the study, we will ask if you are willing to continue your participation in the study. You will be asked to sign another informed consent form that will explain the additional tests and visits. If you are not willing to have these additional tests and visits, your participation in the study will end. Either way we will refer you to the available health related resources for people with HIV infection.

RISKS and/or DISCOMFORTS:

You may feel discomfort, dizzy, or even faint when your blood is drawn. Redness, pain, swelling, bruising, or an infection may occur where the needle goes into your arm.

You may feel discomfort during the examination of your vagina or penis.

You may become embarrassed, worried, or anxious when discussing your sexual practices, ways to protect yourself against or passing HIV, or discussing or waiting for your test results during the study. Learning that you have HIV or other infections passed through sex may make you worried or anxious. A trained counselor will help you deal with any feeling or questions you have.

We will make every effort to protect your privacy and confidentiality while you are in the study. Your visits here will take place in private. However, it is possible that others may learn of your participation here. Because of this, others may treat you unfairly or discriminate against you. For example, you could have problems getting or keeping a job. You could also have problems being accepted by your family or community.

If you chose to have leftover samples stored, there are few risks related to storing your samples. When tests are done on the stored samples there is a small but possible risk to your privacy. It is possible that if others found out information about you that is learned from tests (such as information about your genes) it could cause you problems with your family (having a family member learn about a disease that may be passed on in families or learning who is the true parent of a child) or problems getting a job or insurance, but this would be very rare.

POTENTIAL BENEFITS:

There may be no direct benefit to you from this study. However, you and others could benefit in the future from research done in this study, because we will learn more about HIV. The information we learn may help researchers better understand how to make vaccines or medicines to prevent or treat diseases like HIV.

During the study, you will receive physical exams, information about your health, and the opportunity to talk to counselors about your health, your feelings, and ways to prevent spreading HIV. If these exams show that you have a health problem, we will treat you to the extent possible, or we will tell you where you can go for treatment. You will also receive free condoms throughout the entire course of the study.

NEW FINDINGS

You will be told any new information learned during this study that might cause you to change your mind about staying in the study. You will be told when the results of the study may be available, and how to learn about them.

REASONS WHY YOU MAY BE WITHDRAWN FROM THE STUDY WITHOUT YOUR CONSENT

You may be removed from the study without your consent for the following reasons:

- The study may be discontinued at any time by the sponsor of the study (NIH), the U.S. Office of Human Research and Protection, the CHAVI Scientific Leadership Group, in-country government or regulatory authorities, and/or the IRB/EC overseeing research at the study site.
- The study staff feels that staying in the study would be harmful to you.
- Other administrative reasons.

ALTERNATIVES TO PARTICIPATION

If you choose not to participate in this study it will not impact the health care you can receive at this clinic. [*Sites to include/amend the following if applicable:* There may be other studies going on here or in the community that you may be eligible for. If you wish, we will tell you about other studies that we know about. There also may be other places where you can go for HIV counseling and testing. We will tell you about those places if you wish.]

COSTS AND COMPENSATION:

There is no cost to you for being in this study. At the end of each visit, you will be given [*insert amount of money, incentive package to compensate participant for food, travel expenses, lost work time, etc.* ***Be mindful of fluctuating amounts of your local currency*** *]*

CONFIDENTIALITY:

Efforts will be made to keep your study records and test results confidential to the extent permitted by law. However, we cannot guarantee confidentiality. You will be identified by a code (a number), and personal information from your records will not be released without your written permission. You will not be personally identified in any publication about this study. However, your records may be reviewed, under guidelines of the United States Federal Privacy Act [*U.S. sites only];* the sponsor of the study (United States National Institutes of Health [NIH]), the [*insert name of site*] Institutional Review Board (IRB)/Ethics Committee (EC), study staff, study monitors, and [*insert applicable local authorities*].

Blood and, if you agree, breast milk, semen and vaginal fluid samples collected during this study will be tested for research purposes. These samples will be put into containers that do not have your name on them but that use a code to protect your privacy. The code can only be traced back to your study clinic; however, your name, where you live, and other personal information will be protected by the study clinic. When researchers are given your samples, they will not be given your personal information. If you agree to have any leftover samples stored for future research, these samples will be stored without your name and the results of any future tests will not be included in your health records.

[Include only if applicable – If during the course of the study we find out that you have [*insert all applicable reportable diseases (e.g., HIV)*], we must report it to [*insert the name(s) of the local health authorities*]. Although we must report that we have treated someone with [*insert all applicable reportable diseases*], your name will not be reported to the agency. [*Amend this paragraph to reflect the local requirements. If there are no local requirements to report communicable diseases, delete this paragraph.*]

[For U.S. sites only:] In addition to the efforts made by the study staff to keep your personal information confidential, a Certificate of Confidentiality has been obtained from the U.S. Federal Government for this study. This Certificate protects study staff from being forced to tell people who are not connected with this study, such as the court system, about your participation or information you give for study purposes. Even with the Certificate of Confidentiality, however, if the study staff learns of possible child abuse and/or neglect or a risk of harm to you or others, they will be required to tell the proper authorities. Having a Certificate of Confidentiality does not prevent you from releasing information about yourself and your participation in the study.

RESEARCH-RELATED INJURY:

The study staff will monitor your health closely while you are in this study. If you have any health problems between visits, please contact the study staff. If you have a medical emergency that requires immediate care, [*insert site-specific instructions*].

[Sites to specify institutional policy:] If you are injured as a result of being in this study, the [*insert institution name*] will give you immediate necessary treatment for your injuries. You [will/will not] have to pay for this treatment. If you need additional treatment for your injuries you will be told where you can go because there is no program for monetary compensation or other forms of compensation for such injuries either through this institution or the U.S. National Institutes of Health (NIH). You do not give up any legal rights by signing this consent form.

PROBLEMS or QUESTIONS:

For questions about this study or a research-related injury, contact:

[*insert name of the Investigator Of Record or other study staff*]

[*insert telephone number and physical address of above*]

For questions about your rights as a research subject, contact:

[insert name or title of person on the Institutional Review Board (IRB) or other organization appropriate for the site]

[*insert telephone number and physical address of above*]

SIGNATURE PAGE: STUDY INFORMED CONSENT FORM – HIV NEGATIVE

CHAVI 001: Acute HIV-1 Infection Prospective Cohort Study, Version 2.0

If you have read this informed consent, or have had it read and explained to you, and understand the information, and you voluntarily agree to participate in this research study, please sign your name or make your mark in the signature area at the bottom of this page. By providing your initials in the spaces below you may also agree to additional sample collection, genetic testing, or long-term storage.

____ I agree to allow genetic testing of my blood during the study.

____ I do not agree to allow genetic testing of my blood during the study.

I also agree to provide the following samples for experimental testing during the study:

[NOTE TO SITES: INCLUDE ONLY THE SAMPLES YOU INTEND TO COLLECT]:

Genital secretions: ____**including** genetic testing ____**excluding** genetic testing

Breast milk: ____**including** genetic testing ____**excluding** genetic testing

____I do not agree to provide any of the additional samples listed above.

I agree to allow the following leftover samples to be saved for future testing: ***[NOTE TO SITES: THIS LIST SHOULD MATCH THE LIST ABOVE]***

Blood: ____**including** genetic testing ____**excluding** genetic testing

Genital secretions: ____**including** genetic testing ____**excluding** genetic testing

Breast milk: ____**including** genetic testing ____**excluding** genetic testing

____I do not agree to allow any of my leftover samples to be saved for future testing.

_____ _____
Participant Name (print) Participant Signature and Date

_____ _____
Study Staff Conducting Consent Discussion (print) Study Staff Signature and Date

_____ _____
Witness Name (print) (As appropriate) Witness Signature and Date