

HIV Prevention Trials Network

CLARIFICATION #1 HPTN 036, Version 2.0: HIV Prevalence, Incidence, and HSV-2 Prevalence Among High-Risk MSM in Lima, Peru

27 August 2002

Summary of and Rationale for Clarification

FINAL Version 2.0 of the HPTN 036 protocol, dated 23 July 2002, specifies that a repeat serology will be performed if HIV infection is confirmed at the Enrollment Visit. This algorithm does not match the standard HIV antibody testing algorithm employed by the Peruvian Ministry of Health, nor does it match the HIV antibody testing algorithm for Follow-up Results Visits in the protocol, both of which do not require a repeat serology. The protocol team intended to strike the repeat serology line from the protocol in an earlier iteration. The study team at the HPTU at Impacta and Via Libre have been following the Ministry of Health guidelines to confirm HIV infection since the initiation of the study, and have not been performing a repeat serology.

Implementation

The HPTU will submit this clarification to the appropriate Institutional Review Boards (IRB) and Ethics Committees.

This clarification is pertinent to the specifications of protocol Section 4.2.

1. Under the heading “If HIV infection is confirmed, the participant is ineligible for the cohort study,” the following line has been struck:
“Repeat serology (ELISA and Western blot) for confirmation.”

HPTN 036

HIV prevalence, incidence and HSV-2 prevalence among high-risk MSM in Lima, Perú

A Study of the HIV Prevention Trials Network

Sponsored by:

Division of AIDS
US National Institute of Allergy and Infectious Diseases
US National Institutes of Health

Protocol Chair:

Connie Celum, MD, MPH

Protocol Co-Chair:

Jorge Sanchez, MD, MPH

Final Version 2.0
23 July 2002

HIV prevalence, incidence and HSV-2 prevalence among high-risk MSM in Lima, Perú

TABLE OF CONTENTS

PROTOCOL TEAM ROSTER	4
INVESTIGATOR SIGNATURE PAGE	5
PROTOCOL SUMMARY	6
1 INTRODUCTION	7
1.1 Background.....	7
1.2 Rationale.....	9
2 STUDY OBJECTIVES AND DESIGN	10
2.1 Primary Objectives.....	10
2.2 Secondary Objective.....	11
2.3 Study Design.....	11
3 STUDY POPULATION	12
3.1 Inclusion Criteria.....	12
3.2 Exclusion Criteria.....	13
3.3 Participant Withdrawal.....	14
4 STUDY PROCEDURES	14
4.1 Screening Visit.....	14
4.2 Enrollment Visit.....	15
4.3 Follow-up Visits.....	16
4.4 Follow-up Results Visits	17
4.5 Interim Contacts and Visits.....	19
5 STATISTICAL CONSIDERATIONS	19
5.1 General Design.....	19
5.1.1 Primary Endpoints.....	19
5.1.2 Secondary Endpoints.....	19
5.2 Accrual, Follow-up and Sample Size	19
5.3 Recruitment and Retention.....	21
5.4 Data Monitoring and Analysis	22
6 HUMAN SUBJECTS CONSIDERATIONS	23
6.1 Ethical Review.....	23
6.2 Informed Consent.....	23
6.3 Confidentiality	24
6.4 Incentives.....	24
6.5 Communicable Disease Reporting Requirements.....	24
6.6 Study Discontinuation	24
7 LABORATORY SPECIMENS AND BIOHAZARD CONTAINMENT	24
8 ADMININSTRATIVE PROCEDURES	25
8.1 Study Coordination.....	25
8.2 Study Monitoring.....	25

8.3 Protocol Compliance..... 25
8.4 Investigator's Records..... 26
8.5 Use of Information and Publications..... 26

REFERENCES 27

APPENDICES

I. Schedule of Events 28
II. Study Informed Consent Form 1 of 5
III. HIV Informed Consent Form..... 1 of 3
IV. Blood Sample Storage Consent Form 1 of 5

LIST OF ABBREVIATIONS AND ACRONYMS

AIDS	Acquired Immunodeficiency Syndrome
DAIDS	Division of AIDS
HIV	Human Immunodeficiency Virus
HPTN	HIV Prevention Trials Network
IRB	Institutional Review Board
NIAID	National Institute of Allergy and Infectious Diseases
NIH	National Institutes of Health
NMCRD	Naval Medical Research Center Detachment
PROCETSS	National STD/AIDS Control Program
PPS	Prevention Preparedness Study
SDMC	Statistical and Data Management Center
STD	Sexually transmitted disease
US	United States

HPTN 036

HIV prevalence, incidence and HSV-2 prevalence among high-risk MSM in Lima, Perú

PROTOCOL TEAM ROSTER

Connie Celum MD, MPH	Protocol Team Chair	University of Washington HPTU Cabrini Medical Tower, Box 359927 901 Boren Avenue , Suite 1300 Seattle, WA 98104 ccelum@u.washington.edu
Jorge Sanchez, MD, MPH	Protocol Team Co-Chair	Impacta Salud y Educación Grimaldo del Solar 805 Lima 18 - Perú jorges@hot.com
Stephen Tabet, MD, MPH	Co-investigator (Clinical investigator)	University of Washington HPTU Cabrini Medical Tower, Box 359927 901 Boren Avenue , Suite 1300 Seattle, WA 98104 stabet@u.washington.edu
Jose (Toti) Sanchez, MD, MPH	Co-investigator (Epidemiologist)	U.S. Naval Medical Center Research Detachment, Unit Number 3800 American Embassy APO AA 34031-3800 sanchez@nmrcd.sld.pe
W. L. H. Whittington, PhD	Co-investigator (Epidemiologist)	University of Washington Center for AIDS and STDs, Box 359931 325 Ninth Ave Seattle, WA 98104
Martina Morris, PhD	Co-investigator (Sociologist)	University of Washington School of Social Work Savery Hall, Rm 221
King Holmes, MD, PhD	Co-investigator	University of Washington Center for AIDS and STDs, Box 359931 325 Ninth Ave Seattle, WA 98104
Aldo Lucchetti, MD	Clinical Coordinator (Perú)	Impacta Salud y Educación Grimaldo del Solar 805 Lima 18 – Perú
Pedro Goicochea, MSc, MA	Cohort Coordinator (Perú) Co-Investigator (Perú)	Impacta Salud y Educación Grimaldo del Solar 805 Lima 18 – Perú pgoicoch@impacta.org
Erin Lennon	Peru HVTU and HPTU Study Coordinator (Seattle)	University of Washington UW HPTU Cabrini Medical Tower, Box 359927
Lourdes Kusunoki, MD	Study Coordinator (Perú)	Impacta Salud y Educación Grimaldo del Solar 805 Lima 18 – Perú lkusunoki@impacta.org

HPTN 036
HIV prevalence, incidence and HSV-2 prevalence among high-risk MSM in Lima, Perú
A Study of the HIV Prevention Trials Network

Sponsored by:

Division of AIDS (DAIDS)
US National Institute of Allergy and Infectious Diseases (NIAID)
US National Institutes of Health (NIH)

I, the Principal Investigator, agree to conduct this study in full accordance with the provisions of this protocol. I agree to maintain all study documentation for a minimum of five years from the end of the study, unless directed otherwise by the HPTN CORE. Publication of the results of this study will be governed by HPTN and DAIDS policies. Any presentation, abstract, or manuscript will be made available by the investigators to the HPTN Manuscript Review Committee and DAIDS for review prior to submission.

I have read and understand the information in this protocol and will ensure that all associates, colleagues, and employees assisting in the conduct of the study are informed about the obligations incurred by their contribution to the study.

Name of Principal Investigator (Domestic)

Signature of Principal Investigator

Date

Name of Principal Investigator (International)

Signature of Principal Investigator

Date

HPTN 036
HIV prevalence, incidence and HSV-2 prevalence among high-risk MSM in Lima, Perú

PROTOCOL SUMMARY

- Design:** 1) Prospective cohort study with a six-month accrual period and 12 months of follow-up for enrolled HIV-uninfected participants.
2) Cross-sectional study of risk behaviors, partnership status, and HSV-2 serostatus of HIV-infected men who have sex with men.
- Population:** HIV-uninfected and HIV-infected men who have sex with men (MSM).
- Study Duration:** Accrual will require six months and HIV-uninfected participants will complete one year of follow-up; therefore, the entire study should be completed within approximately 18 months.
- Primary Objectives:** (a) Determine prevalence, incidence, and risk factors for syphilis, HSV-2, and HIV among high-risk HIV-uninfected MSM.
(b) Among HSV-2 seropositive men, evaluate informed consent procedures, eligibility criteria, and willingness to participate in a trial of daily suppressive acyclovir for HIV prevention
(c) Identify effective follow-up strategies for high-risk MSM in Lima to achieve $\geq 90\%$ retention at 12 months.
- Primary Endpoints:** (a) HIV incidence and risk factors for HIV infection
(b) HSV-2 prevalence, incidence and association with incident HIV infection.
(c) Risk characteristics, knowledge about HSV-2, and willingness to participate in daily suppressive acyclovir trial among HSV-2 seropositive MSM
(d) Number of participants retained at the 12 months visit
- Secondary Objectives:** (a) Measure the use of condoms, barriers, and facilitators to enhance condom use with male and female partners
(b) Characterize the sexual networks of MSM, and the proportion of HIV-infected and HIV-negative men who are sexually active with both men and women
- Secondary Endpoints:** (a) Prevalence of condom use with male and female partners, and barriers and facilitators to condom use
(b) Concurrency, mixing, and sexual behavior with male and female partners in the prior 6 months among HIV-positive and HIV-negative men

1 INTRODUCTION

1.1 Background

MSM are an important component of the HIV epidemic in many countries in the Caribbean and in Central and South America, such as Mexico, Columbia, Venezuela, and Perú. The male:female ratio of reported AIDS cases in 1997 ranged from 6.75 in Mexico to 5.13 in Andean countries, including Perú in which the ratio is 3, to 2.42 in Central America. [PAHO 1998] Reported AIDS cases stratified by risk group for South and Central America for 1997 indicate that MSM comprised the largest risk group (40-45% of AIDS cases) in Andean countries and Mexico and the second largest group in Brazil. Even though the epidemic does not currently appear to be primarily heterosexual in Central and South America, MSM could represent an important “bridge” to the heterosexual population [Tabet 1996, Tabet 2001, Sanchez 1996, Carceres 1997].

The HIV epidemic and the Sentinel Surveillance System in Perú

During the first 19 years of the HIV epidemic in Perú, the number of reported AIDS cases has steadily increased with a cumulative 10,403 cases reported as of June 2000. Almost all reported AIDS cases in Perú have been acquired through sexual transmission, the majority by homosexual transmission with a gradual increase in the proportion of heterosexually-acquired cases. To effectively monitor the HIV seroprevalence in Perú, in 1998 the National STD/AIDS Control Program (PROCETSS) of the Ministry of Health of Perú implemented a Sentinel Surveillance system among pregnant women, female sex workers (FSW) and men who have sex with men (MSM). The cities with the largest known female sex workers and MSM populations were selected to be included in the sentinel surveillance. Seroprevalence surveys of postpartum women have been conducted yearly since 1996, in which over 3000 samples are tested annually in Lima, and a minimum of 300 samples in smaller cities. The Sentinel Surveillance system for MSM and FSWs was started in 1998 and includes screening for HIV by ELISA (confirmed by Western blot), and syphilis by RPR. Participants are recruited by active outreach referral by MSM and female sex worker peer educators and during their regular visits to STD reference centers [Holmes 2000].

In both the 1998 and 2000 sentinel surveillance, the highest HIV and syphilis prevalence for MSM was observed in Lima (with 11% HIV and 14% syphilis, respectively) and Iquitos (13% and 26%, respectively in year 2000). Consistently, in all cities surveyed, HIV seroprevalence was higher for MSM (9.0% overall among the 3795 MSM surveyed in 2000) compared to FSWs (1.2% among the 5093 FSWs surveyed in 2000). In January 2001, the less sensitive HIV EIA was performed on HIV-infected sera from MSM identified in the 2000 sentinel surveillance, using the Organon-Teknika Dilvicon™ assay [Rawal 1999] performed by Dr. Chip Sheppard and colleagues from the HVTN Central Laboratory, and analyzed according to the methods of Janssen et al [Janssen 1998]. Among the HIV-positive MSM in the 2000 sentinel surveillance, the estimated HIV incidence was 7.2% among MSM in Lima and 2.9% in the 8 medium-sized cities included in the 2000 sentinel surveillance with the highest estimated HIV incidence in Iquitos (5.7%).

HIV incidence, STDs, and risk behaviors among MSM in Lima

Drs. Tabet and Sanchez conducted a cross-sectional study of 459 MSM in Lima during 1996-97 that showed considerable heterogeneity in self-identified sexual identity (ie homosexual, “moderno”, “heterosexual”, “woman”, transvestite) which correlated with HIV and STD prevalence. The highest syphilis seropositivity and HIV prevalence was observed among transvestites (32% and 43% among transvestites, respectively) and self-identified homosexual men (18% and 15% HIV and syphilis, respectively). A substantial minority (26%) of these men reported sex with both men and women, 11% of whom were HIV-infected [Tabet 2001].

Another important finding from this study and another longitudinal study of female sex workers in Lima by Dr. Sanchez [Sanchez 1998], was that both MSM and FSWs reported that they felt uncomfortable with seeking care at the regional health centers due to feeling stigmatized by providers. Focus groups of MSM from Lima in 2000 indicated that the patient advocates and peer educators have been instrumental in making the health centers more friendly and acceptable to these core groups. This is a significant issue in enhancing health-seeking behavior for core groups who report having been discriminated against by clinics. One of the keys to the success of the studies of MSM in Lima has been availability of HIV and STD services at our collaborating non-governmental clinic, “Via Libre”, and the use of MSM who serve as “patient advocates” at Ministry of Health clinics.

To provide additional data on the epidemiology of HIV and STDs among MSM and in an effort to provide more widespread HIV and STD services to MSM, Dr. Sanchez initiated a HIV and STD screening program for MSM in five clinics in Lima in 1998. A subset of the almost 8000 MSM who were tested and HIV and STDs between 1998 and 2000 in Lima underwent a more intensive behavioral evaluation to characterize risk behaviors among HIV-infected and HIV-uninfected men at screening. Risk factors for incident HIV were characterized among a cohort of the highest risk HIV-negative MSM called “Alaska”. Eligibility criteria for the cross-sectional study included both HIV-infected and HIV-uninfected men who reported sex with a man in the prior year. Additional eligibility criteria for the cohort study of high-risk HIV-negative men included any of the following criteria: ≥ 5 sex partners in the past 6 months, HIV-infected sex partner, STD in the past year, no condom use with last anal sex, or commercial sex work. The funding for the MSM cohort was from the Peruvian Ministry of Health, a grant from the University of Washington Fogarty program, and USAID AIDS-HELP. The focus of those efforts was on reaching the largest number of MSM for surveillance and services and resources were inadequate for retention, particularly for lower socioeconomic MSM (i.e., transvestites and male sex workers) whom often do not have phones or adequate locating information.

STDs were found to be very prevalent among the HIV-infected and HIV-uninfected MSM who have been screened in Lima since October 1998. The prevalence of urethritis (based on a syndromic diagnosis) was 7.6%, syphilis seropositivity was 15.1% (18% of whom had non-treponemal test titers of $\geq 1:16$, representing possible early infectious syphilis), and rectal chlamydia prevalence was 4.0% by culture (an additional 2.5% were positive by PCR among the first 426 HIV-uninfected men in the prospective cohort). By type-specific HSV Western blot, 92% of 105 HIV-infected in the cross-sectional study and 49% of 171 age-matched HIV-uninfected MSM in the prospective cohort were HSV-2 seropositive.

HIV-uninfected MSM who had anal sex in the past year, exchanged sex for money or drugs, had an STD in the past year, or sex with an HIV-infected man were enrolled in a prospective cohort. As of March 2000, 1972 HIV-uninfected MSM were enrolled in the prospective cohort. Among 928 HIV-uninfected MSM with ≥ 1 follow-up visits over an average of 308 (± 131) days follow-up, 28 seroconverted for an observed HIV incidence of 3.3/100 person-years ($CI_{95}=2.1-4.5$). STDs represent a significant risk factor for incident HIV infection; seroconverters were more likely to report symptoms or a diagnosis of urethritis in the last six months prior to study enrollment (O.R. 4.3, 95% CI 1.3-12), anal ulcers (O.R. 4.4, 95% CI 1.04-14.3), and proctitis (O.R. 5.3, 95% CI 0.9-19.6) [Sanchez 2001].

Data from the baseline interviews indicate that bisexual men may serve as “bridge” populations for transmission of STDs and HIV to women. Bisexuality is common among high-risk MSM; 30% of HIV-uninfected MSM, 18% of men with recent HIV infection, and 11% of chronically HIV-infected MSM reported recent sex with ≥ 1 woman in the past 6 months. Among HIV seronegative men “bridgers” (defined as men who acknowledged sex with both men and women during the past six months) had an average of 3.1 female partners, were infrequent condom users (25% reported “always” using condoms in the prior 6 months), and 8% reported

an HIV-infected male partner in the past 6 months. “Bridgers” were less likely to report receptive anal sex with a male partner (25% compared to 84% of “non-bridgers”) but those who did report receptive anal sex reported a median of 2 male partners, and 67% reported inconsistent condom use with receptive anal sex in the prior 6 months.

We have identified a low frequency of consistent condom use and reasons for not using condoms; 40% and 34% of the first 1148 HIV-uninfected MSM enrolled in the cohort reported not using a condom with their last episode of insertive anal sex and receptive anal sex, respectively, with a casual male sexual partner. The three major reasons cited for not using condoms with casual male partners were that: 1) they wanted to use a condom but did not have one available when they had sex (18%), 2) they were intoxicated (19%), or 3) they seldom use condoms (30%). Reasons as to why condoms were not regularly used were not systematically recorded in terms of acceptability, partner’s willingness, and other reasons.

1.2 Rationale

HIV incidence is high (>3%) among MSM in Lima- Perú, and bacterial STDs and HSV-2 are prevalent in both recently and chronically-HIV-infected MSM as well as HIV-uninfected MSM. Bisexual HIV-infected MSM could be an important “bridge” transmitting HIV and other STDs to women, a variation on bridging patterns that our colleague, Dr. Martina Morris has described among gay men in New York (Morris 1995) and in heterosexual partnerships in Thailand (Morris 1996) and Uganda (Morris 1997, Konde-Lule 1997).

Interventions to reduce both HIV and STDs are urgently needed among MSM in Perú. Given these associations and the high prevalence of STDs among MSM in Lima, prevention of HIV infections is partially dependent on innovative methods to control bacterial and viral STDs among high-risk HIV-uninfected MSM. The previous cohort of MSM enrolled in 1998-2000 documented an important need for acceptable and accessible HIV counseling and testing and STD services for MSM, in which over 8,000 MSM were tested in Lima from October 1998-June 2000. The previous cohort had inadequate resources for retention activities, a critical element for the successful conduct of clinical trials. In addition, limited behavioral data on sexual behavior and partnerships of HIV-infected MSM were collected, of particular interest for the behavioral intervention being developed for HIV-infected MSM with recent bacterial STDs and determining the feasibility of recruiting high-risk female partners of HIV-infected bisexual men for HIV vaccine trials (e.g, HVTN protocol 501).

The data from this prevention preparedness study is critical for future HIV prevention and vaccine trials that will be conducted in Perú. Potential HPTN studies to be conducted in Perú include: a) daily suppressive acyclovir among high-risk HIV-negative HSV-2 seropositive MSM, b) a hybrid STD intervention with improved pharmacy and clinic -based syndromic STD treatment as well as core group interventions for FSW and MSM in medium-sized cities outside Lima, given that estimated HIV incidence is 2.9% overall among 3100 MSM from 8 medium-sized cities (based on less sensitive ELISA results in year 2000 Sentinel Surveillance samples), and c) a brief behavioral intervention for HIV-infected MSM with STD incidence as a primary outcome, currently under development by the HPTN STD and Behavioral working groups. Peru will also be a major site to recruit high-risk MSM for HIV vaccine efficacy trials, including HVTN protocol 501.

In order to prepare for the HPTN HSV-2 suppressive intervention, this pre-trial preparedness cohort will focus on HSV-2 rather than bacterial STDs and will provide data on HSV-2 prevalence and incidence, correlates of HSV-2 seropositivity, and the association of HSV-2 as a risk factor for incident HIV infection. The study will also provide additional data on operational aspects of the acyclovir study, including assessing potential eligibility criteria, willingness to participate and characteristics of those willing to participate, clinical recurrence

rates, use of acyclovir among HSV-2 seropositive MSM, and methods of counseling and obtaining informed consent for the acyclovir trial. The study design for the HPTN HSV-2 suppressive trial will include HIV-discordant heterosexual couples (of whom at least one member is HSV-2 seropositive) in Africa and India and high-risk HSV-2 seropositive MSM from Peru and the U.S. Peru will be expected to recruit 700-800 HIV-negative HSV-2 seropositive MSM for the HSV-2 suppressive trial.

This preparedness study will also enable us to further develop a peer-driven behavioral intervention for MSM as part of a community-randomized STD/HIV intervention trial. The peer outreach workers utilized by Dr. Sanchez in the previous MSM studies are similar to the peer diffusion model found to be effective in accessing high-risk injection drug use networks [Broadhead 1998]. However, in spite of the success of recruiting MSM for STD and HIV testing and condom promotion, critical information is needed on barriers and facilitators to condom use and sexual behavior with male and female partners if an MSM intervention is conducted in Perú. In particular, given the previous studies that indicate a high proportion of MSM are actively bisexual, sexual network analyses will provide relevant data on the proportion of MSM with HIV and other STDs who are bisexual, their sexual behaviors and condom use patterns with both male and female partners. These data will be valuable in designing the behavioral intervention for HIV-positive MSM in the domestic-international HPTN intervention as well as the capacity to identify high-risk female partners of HIV-positive bisexual men for HIV vaccine and other prevention studies.

This cohort study will provide an opportunity to focus on retention strategies, a key component for successful conduct of large clinical trials of vaccine and non-vaccine prevention strategies. The previous cohort conducted by Drs. Sanchez and Celum had very limited resources, which were primarily directed towards recruitment, STD and HIV counseling and testing. Drs. Celum and Sanchez will collaborate on intensive efforts to retain this new cohort, adapting successful strategies in the Seattle site of the EXPLORE study, and identifying new approaches with the peer educators to retain study participants without phones and with limited locator information.

If the acyclovir trial to suppress HSV-2 infection for HIV prevention and/or the behavioral intervention for MSM are implemented before the one year follow-up of the MSM cohort is completed, there will be a transition to screen and enroll eligible MSM from the cross-sectional study of HIV-infected MSM and the prospective cohort of HIV-uninfected MSM. Men in the cohort study who are not eligible for a prevention clinical trial will no longer be followed, and the pretrial preparedness trial will be terminated to enable the site to devote resources to implementation of the new trial(s). The informed consent form states that participants will be followed on a quarterly basis until the study is terminated for a maximum period of 12 months.

2 STUDY OBJECTIVES AND DESIGN

2.1 Primary Objectives

- Determine prevalence, incidence and risk factors for syphilis, HSV-2, and HIV among high risk HIV-uninfected MSM.
- Among HSV-2 seropositive MSM, evaluate informed consent procedures, eligibility criteria, and willingness to participate in a trial of daily suppressive acyclovir for HIV prevention
- Identify effective follow-up strategies for high-risk MSM in Lima to achieve $\geq 90\%$ retention at 12 months.

2.2 Secondary Objectives:

- Measure the use of condoms and facilitators to enhance condom use with male and female partners
- Characterize the sexual networks of MSM, and the proportion of HIV-infected and HIV-uninfected MSM who are sexually active with both men and women

2.3 Study Design

High-risk MSM will be recruited by trained MSM peer educator/outreach workers, as utilized successfully for the previous MSM cohort in Lima (called the “Alaska cohort”) who will refer MSM to one of two study sites (Impacta and Via Libre clinics). After determining eligibility using an eligibility check-list and obtaining informed consent, all MSM will be counseled and tested for HIV, HSV -2, and syphilis at the screening visit. They will be given the results of their syphilis test at the same visit. Men will then be interviewed about risk behaviors in the past six months, utilizing computer-assisted technology. Trained counselors will provide risk reduction counseling according to locally accepted standards. Depending on HIV prevalence in the men screened to reach the goal of 250 HIV-negative MSM for the prospective cohort, 75-100 HIV-infected men will be assessed for HSV-2 serostatus, risk behaviors, and current partnership status at the screening visit. A prospective cohort of 250 high-risk HIV-uninfected MSM in Lima, Perú will be enrolled, based on eligibility determination at the enrollment visit. The cohort will be followed at 3 month intervals to determine the incidence of and risk factors for prevalent and incident HIV and HSV-2, and to evaluate the effectiveness of retention strategies.

At the enrollment visit, HIV and HSV-2 results will be provided. A study clinician will counsel HIV-infected men and HSV-2 seropositive MSM about the meaning of their positive test results and will conduct a brief clinical history with a standardized questionnaire (about clinical manifestations of herpes, use of acyclovir, and willingness to be in the future acyclovir-HSV-2 suppression trial). Counseling materials about HSV-2 (in Spanish) will be piloted and information participants determine necessary to make informed decisions about participation in the HSV-2 suppression trial will be obtained. HIV-uninfected MSM who meet eligibility criteria (described below) will be enrolled into a one-year prospective cohort study with follow-up visits every three months, for a total of up to four follow-up visits. HIV-infected men will be referred for care and counseling. Men who appear to be recently infected at baseline will be identified, based on a reactive standard ELISA and non-reactive less sensitive ELISA, which will be performed at the US NMRCDC lab (using the Organon-Teknica Dilvicon™ assay with the US NMRCDC under the CDC IND). Men who are recently infected (based on the less sensitive ELISA) and documented HIV seroconverters during the study will be offered enrollment into an early HIV infection natural history study. (Appendix I contains an explanation of the activities at each visit.)

The eligibility checklist will be interviewer-administered to provide “real-time” determination of eligibility. Data Fax forms specific to this protocol will be used for the eligibility checklist, case report forms for enrollment and follow-up visits, and laboratory results for the Peru preparedness study. The use of the core Data Fax forms will provide the Peru site experience in DataFax and enable SCHARP to monitor recruitment, retention, and incidence. The screening, baseline, and follow-up questionnaires will be administered by Computer Assisted Self-Administered (CASI) technology. Dr. Tony Rossini (of the UW CFAR and SCHARP) has piloted CASI technology in the current MSM cohort in Perú and has found high acceptability of the CASI technology.

The baseline and follow-up questionnaires have been developed by Drs. Celum, Sanchez, Whittington, and Morris. These instruments will collect data on socio-demographic characteristics, sexual behaviors, STD history, and use of drugs or alcohol with sex. In the baseline questionnaire, three additional modules will address sexual networks, willingness to participate in HIV vaccine or prevention trials, including the acyclovir trial, and any possible acute retroviral syndrome symptoms. This instrument has been adapted from the previous Perú MSM questionnaire, but has a greater focus on frequency of condom use with male and female partners, reasons for not using condoms, and behaviors with partners of different HIV serostatus. The instrument is already programmed in CASI (in Spanish), based on incorporation of many of the questions from the Spanish version of the EXPLORE questionnaire; changes and additional questions for the Peru preparedness study have also been programmed.

Study data will be collected and entered into a local database. Data will be transferred to the Seattle HPTU site at periodic intervals during accrual and study follow-up. Quality assurance of the data will be provided by SHCARP (for the data collected by Data Fax). The local site data manager, in conjunction with the Seattle HPTU data manager, will monitor the quality and completeness of the behavioral questionnaires. The Seattle HPTU will oversee data management in order to facilitate quality assurance as well as data analysis.

3 STUDY POPULATION

Lima is the capital of Perú and the largest city in the country. Its harbor, Callao, thirty minutes driving from downtown Lima, is the major port town in Perú and one of the largest along the Pacific coast in South America. Together, Lima and Callao concentrate nearly 36% of the population of Peru, estimated to be 26,748,972 inhabitants.

A two-tiered recruitment strategy will be employed. Approximately 500 men will be screened at baseline with the goal of enrolling 250 eligible HIV-uninfected MSM into the prospective cohort and identifying 75 - 100 HIV-infected men during screening for the behavioral and HSV-2 serologic assessment. Screening will end once the target of 250 HIV-negative MSM for the cohort study has been reached. Recruitment will be performed at two health centers (Asociación Impacta Salud y Educación, Asociación and VÍA LIBRE). Subjects to be included in the baseline questionnaire will be men over 18 years of age, who have had at least one episode of homosexual activity (defined as anal intercourse, either insertive or receptive) during the last 12 months and who have not previously tested positive for HIV. Due to (1) the sociological diversity of this population, (e.g., socioeconomic status, social & sexual networking, self-identified sexual identity [e.g., gay/bisexual/transvestite], and commercial sex work); and (2) the resulting differences in HIV and HSV-2 seroprevalence, an effort will be made to represent such heterogeneity in the sample through targeted recruitment at different venues.

Only HIV-uninfected men who practice high-risk behaviors during the past year will be invited to participate in the prospective cohort study, based on one or more of the following: more than 5 partners in the last 3 months, no condom use with the most recent episode of anal intercourse, exchanging sex for money in the past six months, current STD or self-report of an STD in the past 6 months, or an HIV-infected partner in the past 6 months). These recruitment strategies and eligibility criteria were used in the previous MSM cohort study in Lima, in which HIV incidence was 3.3/100 person-years. Although recruitment of larger numbers of participants would be feasible, sample size estimates indicate this is a sufficient sample to achieve the primary goals of the study, and enable a focus on retention.

3.1 Inclusion Criteria

The aims of the study are to assess HSV-2 prevalence, sexual networks and behaviors of HIV-infected and HIV-uninfected MSM; and to enroll a prospective cohort of the highest risk HIV-uninfected MSM to determine HSV-2 prevalence, HIV incidence, risk factors for incident HIV

infection, and effective retention strategies. Therefore, eligibility will be ascertained in a two-step process:

3.1.1 Inclusion criteria determined at Screening Visit (Visit 0):

Persons must meet the following criteria in order to be eligible for the initial screening visit and brief screening questionnaire:

- Men age 18 years and older
- A man who has engaged in anal intercourse, insertive or receptive, with another man in the past 12 months.
- Able and willing to provide written informed consent for HIV testing and study participation.

3.1.2. Inclusion criteria determined at Enrollment Visit (Visit 1):

To recruit the cohort of high-risk HIV-uninfected MSM, behavioral eligibility criteria from the previous cohort will be utilized, as they have effectively recruited a cohort with HIV incidence of 3.3/100 person-years from 1998-2000. At the Enrollment Visit, men must meet the following criteria in order to be eligible for the prospective cohort study:

- Available for 12 months of study participation.
- Able and willing to provide adequate information for locator purposes (as defined by local site standard operating procedures).
- HIV-seronegative by licensed ELISA at screening.
- Have engaged in high-risk sexual behavior defined as:
 - more than 5 partners in the last 3 months, or
 - no condom use with the most recent episode of anal intercourse, or
 - exchanged sex for money in the past 6 months, or
 - currently has an STD or had a self-reported STD in the past 6 months, or
 - had an HIV-infected partner in the past 6 months.

3.2 Exclusion Criteria

Persons who meet any of the following criteria will be excluded from the study:

- Have an obvious psychological/psychiatric disorder that would preclude provision of informed consent or otherwise contraindicate study participation.

3.3 Participant Withdrawal

Once a participant has enrolled in the study, the study site will make every reasonable effort to retain him for 12 months of follow-up. Retention rates of at least 90 percent at 12 months are

targeted for this one-year cohort study. However, participants may withdraw from the study for any reason at any time. The investigator also may withdraw participants from the study if they are unwilling or unable to comply with required study procedures. Participants also may be withdrawn if the sponsor or regulatory authorities terminate the study prior to its planned end date. Every reasonable effort will be made to complete final HIV testing of participants who terminate from the study prior to their last scheduled follow-up visit, and study staff will record the reason(s) for all withdrawals from the study in participants' study records.

4 STUDY PROCEDURES

4.1 Screening Visit (Visit 0)

Written informed consent for study screening will be obtained prior to the conduct of any screening procedures. Potential study participants will undergo eligibility screening (see Section 3.1) by an interviewer. Eligibility will be determined based on participant responses to an interviewer-administered eligibility checklist. Eligibility for the prospective cohort related to HIV serostatus will be ascertained via HIV ELISA testing from the screening visits; test results will be provided at the Enrollment Visit in approximately 7-14 days allowing adequate time for confirmatory Western blots to be run on sera reactive by HIV-1 ELISA.

The following procedures will be performed at each Screening Visit:

- Ascertain participant identity and assign Participant ID number.
- Explain the purpose of the visit and the informed consent and eligibility determination processes.
- Determine eligibility, using Screening Eligibility Checklist (interviewer-administered)
- Obtain written informed consent for study participation (see Appendix II).
- Collect participant contact and locator information.
- Administer Screening Questionnaire with CASI, which includes Acute Retroviral Syndrome Symptoms Questionnaire.
- Deliver HIV and STD pre-test and risk reduction counseling; obtain written informed consents for HIV testing (see Appendix III) and stored blood for future testing (see Appendix IV).
 - ⇒ HIV and STD counseling includes encouraging, training in, and negotiation of condom use with partners and provision of condoms. Counselors will also discuss risk reduction by discussion of HIV serostatus and risks with new and existing partners, reduction of number of sexual partners, and the importance of prompt treatment for STDs.
- Obtain 20 cc of venous blood for syphilis testing, HSV-2 ELISA testing (by type-specific HSV ELISA from MRL), HIV ELISA test, and confirmatory HIV Western Blot (if ELISA is positive).
- Deliver syphilis test results and post-test counseling;
 - ⇒ If syphilis is diagnosed,

- Refer for treatment or treat according to Peruvian Ministry of Health guidelines.
- Document all referrals.
- Provide study site contact information and instruct the participant to contact the study site for additional information or counseling, if needed, prior to the Enrollment Visit.
- Schedule Enrollment Visit to occur in 7-14 days.
- Complete and enter all required data collection for ms.

4.2 Enrollment Visit (approximately 7-14 days following Screening Visit, also known as Day 0)

During the Enrollment Visit, the participant's HIV test results will be disclosed and HIV post-test counseling will be delivered. Participants who test HIV-uninfected and meet eligibility criteria will be enrolled in the prospective cohort study.

The following procedures will be performed at the Enrollment Visit:

- Confirm participant identity and ID number.
- Update locator information.
- Provide result of HSV-2 ELISA;
 - ⇒ If HSV-2-positive,
 - Counsel about HSV-2 natural history, symptoms, link between HSV-2 and HIV transmission, and upcoming acyclovir trial.
 - Administer Genital Herpes Acyclovir Trial Questionnaire.
- Provide HIV post-test counseling and disclose results of HIV antibody test.
 - ⇒ If HIV infection is **confirmed**, the participant is **ineligible** for the cohort study:
 - Repeat serology (ELISA and Western blot) for confirmation.
 - Administer Enrollment HIV+ Questionnaire.
 - Administer complete physical exam.
 - Refer participant to appropriate medical and psychosocial services, and other available research studies.
 - If the less sensitive HIV-1 EIA (using Organon-Teknika Diluviron™ assay at US NMRCDC under the CDC IND) is non-reactive and the standard HIV-1 EIA and Western blot are reactive, the participant will be counseled that their infection may be recent, and told about the upcoming early HIV infection natural history study.
 - Complete and submit required data collection forms.

⇒ If HIV status is **indeterminate**:

- Counsel about significance of indeterminate HIV result (possible seroconversion versus cross-reactive antibodies)
- Perform re-testing (both ELISA and Western blot).
- Schedule visit to occur in 7-14 days. [Eligibility for the prospective study will be based on follow-up serologic testing to determine whether the participant is persistently indeterminate, in which case he will be offered enrollment, or an HIV seroconverter, in which case he will be offered enrollment into the early HIV infection natural history study]
- Complete and submit required data collection forms.

⇒ If HIV antibody is **negative**,

- Counsel about test results and provide risk reduction counseling.
- Confirm eligibility for prospective cohort and willingness to be followed for up to 12 months (using Enrollment Eligibility Checklist).
- Administer Enrollment STD Symptoms Questionnaire.
- Genital and rectal exam by study clinician for men who report STD symptoms.
 - ⇒ If participant has a symptomatic STD, such as urethral discharge or genital ulcer, refer for treatment or treat syndromically according to Peruvian Ministry of Health guidelines.
- Schedule Follow-up Visit to occur on study Day 91 (± 14 days).
- Provide study site contact information and instruct the participant to contact the study site for additional information about the study, HIV counseling and/or HIV testing, if needed, prior to the first Follow-up Visit.
- Confirm locator information
- Complete and submit required data collection forms

4.3 Follow-up Visits (Days 91, 182, 274, and 365)

Four quarterly Follow-up Visits will take place in the year following enrollment in the study. These visits are targeted to take place 91, 182, 274, and 365 days from the participant's study enrollment date, plus or minus 14 days. The following procedures will be performed at each visit:

- Confirm participant identity and ID number.
- Update locator information.

- Administer Follow-Up Questionnaire by CASI, which includes Acute Retroviral Syndrome Symptoms Questionnaire and Follow-up STD Symptoms Questionnaire.
- Genital and rectal exam by study clinician for men who report STD symptoms.
 - ⇒ If participant has a symptomatic STD, such as urethral discharge or genital ulcer, refer for treatment or treat syndromically according to Peruvian Ministry of Health guidelines.
- Deliver HIV and syphilis pre-test and risk reduction counseling as in Section 4.2. (*and at day 365 only, for previously HSV-2-uninfected participants at enrollment only*), deliver HSV-2 pre-test counseling.
- Obtain written informed consent for HIV ELISA test (Appendix IV).
- Obtain 20 cc of venous blood for syphilis testing, HIV ELISA test and confirmatory HIV Western Blot (if required) and *at day 365 only and for previously HSV-2-uninfected participants at enrollment only*, HSV-2 ELISA testing (i.e. MRL) to determine HSV-2 seroincidence.
- Deliver syphilis test results and post-test counseling; refer participant to local healthcare, social service, and/or other providers, if needed, and document all referrals.
 - ⇒ If syphilis is confirmed by positive RPR (and, if no history of treatment in the past year):
 - Refer for treatment or treat according to Peruvian Ministry of Health guidelines.
 - Document all referrals.
- Refer participant to local healthcare, social service, and/or other providers if needed; document all referrals.
- Schedule Follow-up Results Visit to occur in 7-14 days.
- Schedule next Follow-up Visit to occur on study day 182, 274, or 365 (± 14 days).
- Reiterate study site contact information and instruct the participant to contact the study site for additional information about the study, HIV counseling, and/or HIV testing, if needed, prior to the next scheduled visit.
- Complete and enter all required data collection forms.

4.4 Follow-Up Results Visit (7-14 days following each Follow-up Visit)

The following procedures will be performed at each Follow-up Results Visit:

- Confirm participant identity and ID number.
- Update locator information.

- Deliver HIV test results and post-test counseling; refer participant to local healthcare, social service, and/or other providers, if needed, and document all referrals.

- *At day 372 only*, provide result of HSV-2 ELISA, *if performed at Day 365*.
 - ⇒ If HSV-2 seropositive,
 - Counsel about HSV-2 natural history, symptoms and upcoming acyclovir trial.
 - Administer Genital Herpes Acyclovir Trial Questionnaire.
 - Obtain permission to contact participant regarding possible future studies. If HPTN acyclovir trial has been initiated, determines eligibility and interest in participating in acyclovir study.

- Provide HIV post-test counseling and disclose results of HIV ELISA-antibody test.
 - ⇒ If HIV ELISA and Western blot are **positive**:
 - Counsel about condom use and partner disclosure and referral.
 - Administer Follow-up HIV+ Questionnaire.
 - Refer participant to appropriate medical and psychosocial services.
 - Refer participant to HPTN early HIV infection natural history study.
 - Administer complete physical exam.
 - Complete and submit required data collection forms.
 - ⇒ If HIV status is **indeterminate**, based on Western Blot, the same procedures as in Section 4.3 will be followed
 - Obtain confirmatory HIV Western blot testing from local lab.
 - ⇒ If HIV antibody is **negative**,
 - Counsel about test results and provide risk reduction counseling.
 - Reiterate study site contact information and instruct the participant to contact the study site for additional information about the study, HIV counseling, and/or HIV testing, if needed, prior to the next scheduled visit.
 - Confirm locator information
 - Confirm schedule for next Follow-up Visit.
 - Complete and enter all required data collection forms.

4.5 Interim Contacts and Visits

Interim contacts and visits may be conducted at participant request at any time during the study. Interim HIV/STD counseling and testing should be provided as needed in response to participant reports of potential exposure to HIV or STD, or STD symptoms. All interim contacts and visits will be documented in participants' study records.

5 STATISTICAL CONSIDERATIONS

5.1 General Design

This is a study of HIV and HSV-2 prevalence and a prospective cohort of HIV and HSV-2 seroincidence, for which accrual of 250 HIV-uninfected participants will be completed over the course of six months or less. Follow-up assessments will be completed three, six, nine, and twelve months from the time of enrollment.

5.1.1 Primary Endpoints

Consistent with the primary study objectives, the following primary endpoints will be assessed:

- HIV incidence and risk factors for HIV
- HSV-2 prevalence, incidence, and association with incident HIV infection.
- Risk characteristics, knowledge about HSV-2, and willingness to participate in daily suppressive acyclovir trial for HIV prevention among HSV-2 seropositive MSM
- Number of participants retained at the 12 months visit

5.1.2 Secondary Endpoints

- Prevalence of condom use with male and female partners, and barriers and facilitators to condom use
- Concurrency, mixing, and sexual behavior with male and female partners in the prior six months among HIV-infected and HIV-uninfected MSM

5.2 Accrual, Follow-up, and Sample Size

MSM ages 18 years and older will be invited to participate in a study aimed at obtaining information for the design of future HIV prevention and vaccine trials. Recruitment will utilize the following methods:

- Recruitment from existing cohort (called "Alaska") for HIV-uninfected MSM who meet eligibility criteria for new cohort. We will enroll a maximum of 250 men from the Alaska cohort, and of those, will enroll previous Alaska cohort members who meet our eligibility criteria, agree to one year follow-up, provide locator information, and had at least one follow-up visit during the Alaska study.
- Street-based contacts by peer educators in selected areas of the city
- Flyers in businesses and NGOs that cater to MSM

- Snowball referral
- Recruitment in HIV testing centers and health facilities associated with the program
- Advertisements and announcements in the press

The site will screen approximately 500 men, of whom approximately 100 are anticipated to be HIV-infected (based on previous seroprevalence studies) and target accrual of 250 HIV-uninfected study participants within a six-month accrual period. This timeline is realistic given the site’s past recruitment performance and the enrollment of high-risk HIV-uninfected MSM from the existing cohort of approximately 2000 HIV-uninfected MSM in Lima. The site will target retention of 95 and 90 percent of enrolled participants through 6 and 12 months of follow-up, respectively.

Assessment of the primary study outcomes related to accrual and retention will not require statistical analysis. However the precision of study estimates of HIV seroincidence depends on the number of participants enrolled and retained in the study. Enrollment of a cohort of 250 MSM will provide a relatively precise estimate of HIV incidence, depending on the observed incidence rate (I), and 12 month retention (R) rates of 80% or 90%. The following table depicts the half-width of the 95% confidence interval by varying HIV incidence and retention rates with 250 MSM in the cohort:

A = 250		
	R = 80%	R = 90%
I = 2%	1.34	1.28
I = 5%	2.10	2.01
I = 8%	2.64	2.53

Evaluation of willingness to participate in the HSV-2 suppressive trial will depend on descriptive techniques. Based on preliminary data, we estimate that 55% (~550) of HIV-seronegative men and 90% of the approximately 200 HIV-infected MSM identified at screening will be seropositive for HSV-2 antibodies. HSV-2 seropositive men will be asked about their willingness to participate in a trial of daily suppressive acyclovir for HIV prevention; demographics and risk behaviors of those willing and not willing to participate in the acyclovir suppressive trial (i.e., theoretical “acceptors” and “refusers”) will be compared.

We will also conduct analyses to assess baseline demographic and risk behaviors of men lost-to-follow-up. We will have adequate power, assuming a 90% retention rate, to compare the baseline demographic and risk behavior characteristics of the approximately 450 who are retained to the 50 lost-to-follow-up over the course of one year, focusing on attributes such as whether participants have a home or mobile phone, previously was an Alaska cohort member, and attended the monthly raffles and participant appreciation events.

The primary purpose of the sexual network data obtained at the baseline visit is to characterize the proportion of HIV-infected and HIV-uninfected MSM who are bisexual, their sexual behavior, and pattern of condom use with both male and female partners. The small number of participants will preclude some subgroup analyses (i.e. risk behaviors of transvestites or male sex workers compared to other MSM). However, we can compare high-risk activities between groups (bisexual versus exclusively homosexual men) Sexual network analyses will allow us to both characterize HIV-infected and uninfected men with regard to their sexual activities, and to contextualize participant sexual behaviors within partnerships. Specifically, we will evaluate participants’ sexual mixing patterns, condom use, and partner concurrency to ascertain the likelihood of transmission of HIV and STDs. These analyses, particularly in context of the participant’s HIV and STD status, will inform future prevention studies and recruitment strategies targeted for specific high-risk subgroups.

5.3 Recruitment and Retention

Recruitment will utilize the methods summarized in section 5.2. Trained and supervised peer educators will provide information, flyers, and invite potential participants to the 2 clinics (Impacta and Via Libre) to be screened for the study. The sites will target accrual of 250 study participants within six months or less. This timeline is realistic given the site's past recruitment performance and the roll-over of eligible high-risk HIV negative MSM from the existing "Alaska" cohort in Lima.

The sites will target retention of 95 and 90 percent of enrolled participants through 6 and 12 months of follow-up, respectively. Extensive locator information will be collected, including address, phone number, phone number of close contacts of study participants, employer, and frequently attended social venues, as well as participant preferences for how they would like to be contacted.

In order to achieve this high retention, the following new approaches will be implemented:

- The research objectives, rationale, and priority of high retention has been communicated to all the Peru HPTU and HVTU staff, and will be reinforced through staff training and evaluation.
- The most effective peer educators from the Alaska cohort have been selected to be outreach workers and retention specialists for the preparedness cohort. They have been extensively trained in the importance of high retention rates for all HPTN and HVTN studies.
- The following Seattle HPTU staff have conducted site visits in 2000-01 with the Peru staff to implement effective retention strategies from Seattle, adapted for Peru: Jerry Galea, MSW (study coordinator for the Seattle EXPLORE study with over 700 participants, which has over 90% 1 year retention), Dennis Torres (Seattle HPTU/HVTU community educator who has worked with the Lima MSM peer educators in recruitment and retention strategies), Rachael McClennen, MPH (Seattle-based coordinator for HPTN and HVTN studies in Peru), and Drs. Celum, Tabet, and McElrath. Jerry Galea is currently working in Lima with the Impacta staff to ensure a clean dataset and to finalize the CASI questionnaire. Rachael McClennen, and Niles Eaton have conducted another site visit prior to the initiation of the preparedness cohort.
- HPTU counselors will be trained in informed consent procedures to ensure that potential participants understand the nature of the research and the importance of retention to the study objectives.
- Only men who will provide locator information and agree to one year follow-up will be recruited and enrolled in the new cohort.
- A modest monetary incentive will be provided to cover transportation costs, which was cited as a barrier to study visits by some Alaska participants.
- The Seattle and Peru HPTU data managers have designed a data management system that will provide visit windows, reminders, and locator information on a weekly basis to the Peru site.
- A dedicated retention specialist has been designated in Lima, who will be trained in remote follow-up procedures for men who agree to off-site visits. Home visits will be made by the outreach worker or retention specialist to participants who fail to attend their appointments.
- Several focus groups were conducted with "Alaska" participants who recommended a number of retention strategies, including a participant forum, dances, volleyball tournaments, skill-building seminars (eg computer skills), which have been implemented.

- Evening and Saturday hours will be available for those who cannot make weekday appointments.
- A 1-800 phone line is being established for appointments which will allow cohort members to ask for new appointments, change appointment times, and ask for interim services, regardless of whether they have a phone.
- Reminder phone calls will be made 1 week and 1 day before appointments.
- Appointment reminders, birthday, and Christmas cards will be sent to cohort members.
- The waiting rooms at Impacta and Via Libre will have written materials and videos related to HIV and STD prevention and gay men's health.

5.4 Data Monitoring and Analysis

Accrual and follow-up rates and adverse events (primarily social harms such as discrimination resulting from study participation) will be monitored closely by the study team. Since this is an observational study in which participant will not receive any investigational agents, no adverse event or serious adverse event reporting will be undertaken. Drs. Celum and Sanchez and key study staff in Seattle and Lima will have every two week conference calls and regular email communication to monitor study implementation.

The investigators have programmed the questionnaire in CASI which will facilitate timely data management and QCs and optimize risk behavior reporting. The Seattle HPTU site has expertise in CASI programming (Dr. Tony Rosini and Jerry Galea, MSW) and have piloted the CASI technology in Peru. Mr. Galea has collaborated with Ms. Alice Fisher of SCHARP who programmed the EXPLORE questionnaire in CASI. In addition, much of the CASI questionnaire utilizes the existing CASI programming for the EXPLORE questionnaire (in Spanish).

Case Report Forms (CRFs) and study samples will be sent from Via Libre to Impacta on a daily basis via Impacta's vehicle. Upon arrival to Impacta, the CRFs will be checked by the Study Coordinator for any errors or missing information before sending to DataFax. In the event of a correction or addition needing to be made, she will communicate by phone with the staff at Via Libre to correct the error. All CRFs will be maintained at the Impacta site. Prior to each monitoring visit, the CRFs will be sent to each site to enable the monitor to review them along with the other study records.

Close cooperation between the site investigators, NIAID Representative, Protocol Coordinator, Biostatistician, Data Managers, and other study team members will be necessary in order to track study progress, respond to queries about study implementation, and address other issues in a timely manner. Bi-weekly conference calls between the Seattle and Peru core staff have been implemented to ensure close communication, augmented by frequent e-mail communication. Rates of accrual, follow-up, and protocol compliance will be monitored closely by the study team. Representatives of the HPTN CORE and SDMC also will evaluate these rates on a regular basis. If unexpected concerns arise, they will be addressed according to DAIDS and HPTN standard operating procedures.

Data analysis will be performed by Dr. Whittington, with Dr. Morris overseeing the analysis of the sexual network data. Corresponding to each of the study objectives and outcomes, the following primary data analyses will be performed:

- The incidence (and 95% confidence intervals) of HSV-2 infection will be calculated by dividing the number of HSV-2 seroconversions by the total number of person-years of followup.

- The incidence (and 95% confidence intervals) of HIV infection will be calculated by dividing the number of HIV seroconversions by the total number of person-years of followup.
- Univariate and multivariate analysis will be performed for risk factors for prevalent and incident HIV and HSV-2. Multivariate models to fit the data will be tested using survival analysis in STATA statistical software package, which can fit time-dependent covariates.
- Frequency of willingness to participate in HSV-2 suppression trial will be described among MSM who test positive for HSV-2 antibodies. Demographics and levels of risk taking of theoretical “acceptors” and “refusers” will be compared.
- Prevalence and incidence of HSV-2 among HIV seroconverters will be analyzed, adjusted for sexual exposure (e.g. number of episodes of unprotected receptive and insertive anal sex, and number of partners in the previous three months).
- Participant retention for each study follow-up interval, and across all four study visits, will be calculated. The baseline demographics and HIV risk behaviors of men completing and not completing scheduled follow-up will be compared. The denominator for these calculations will be the total number of participants enrolled in the study. The numerator will include all participants who complete a Follow-up Visit during the interval and/or are known to have become HIV-infected or to have died during the study.
- Odds ratios will be used to estimate the relative risk of HIV, HSV-2 and syphilis between HIV-discordant and concordant partnerships, and between participants with or without concurrent sexual partners. Generalized estimating equation methods will be used to control for possible correlations among multiple partners of a given participant [Liang 1998].

6 HUMAN SUBJECTS CONSIDERATIONS

6.1 Ethical Review

This protocol, the informed consent forms contained in Appendices II and III— and any subsequent modifications — will be reviewed and approved by DAIDS and the Institutional Review Boards of the University of Washington (MPA # M-1183), the US Naval Medical Research Center’s Institutional Review Board (FWA #00000152), Impacta (FWA #00001491), and Via Libre (ICPA # T-5104) with respect to scientific content and compliance with all applicable research and human subjects regulations.

The protocol, site-specific informed consent form, participant education, outreach, and recruitment materials, and other requested documents — and any subsequent modifications — also will be reviewed and approved by these Institutional Review Boards.

Subsequent to initial review and approval, each IRB will review the protocol at least annually. The investigator will make safety and progress reports to these IRBs at least annually, and within three months of study termination or completion. These reports will include the total number of participants enrolled in the study, the number of participants who completed the study, number of HIV seroconverters, all changes in research activities, and all unanticipated problems involving risks to human subjects or others.

6.2 Informed Consent

Written informed consent will be obtained from each study participant (or the parents or legal guardians of participants who cannot consent for themselves). The HPTN CORE will review all site-specific informed consent forms and approve them for use according to DAIDS policies; study site staff may not begin obtaining informed consent from study participants until receiving HPTN CORE approval of the forms, in the form of confirmed site registration to begin study operations. Consent forms will be developed in Spanish by the Peru HPTU staff and back-translated into English by a fluent Spanish/English speaker (per UW IRB guidelines).

Each participant (or his parent or legal guardian) will be provided with a copy of his informed consent forms if he is willing to receive them. Study staff will document the informed consent process as described in the study-specific procedures manual and in the participant record.

6.3 Confidentiality

All study-related information will be stored securely at the study site. All participant information will be stored in locked file cabinets in areas with limited access. All laboratory specimens, reports, study data collection, process, and administrative forms will be identified by a coded number only to maintain participant confidentiality. All records that contain names or other personal identifiers, such as locator forms and informed consent forms, will be stored separately from study records identified by code number. All local databases will be secured with password-protected access systems. Forms, lists, logbooks, appointment books, and any other listings that link participant ID numbers to other identifying information will be stored in a separate, locked file in an area with limited access.

Participants' study information will not be released without the written permission of the participant, except as necessary for monitoring by NIAID and/or its contractors (e.g., the DAIDS monitoring contractor), representatives of the HPTN CORE and/or SDMC, and US or Peruvian regulatory authorities.

6.4 Incentives

Participants will receive HIV/STD post-test counseling, and up to 100 free condoms will be provided free to the men at each quarterly visit. Men will be reimbursed 20 Peruvian soles (approximately \$5.00) per visit to cover transportation expenses.

6.5 Communicable Disease Reporting Requirements

Peruvian law dictates that doctors must report the number of cases of HIV/STDs, but not the names of cases, to the Ministry of Health. Study staff will report the number of HIV/STD cases to the Ministry of Health. Participants will be made aware of all applicable reporting requirements during the study informed consent process.

6.6 Study Discontinuation

The study may be discontinued at any time by NIAID, the HPTN, and US or Peruvian government.

7 LABORATORY SPECIMENS AND BIOHAZARD CONTAINMENT

The Impacta Laboratory will perform the HSV-2 ELISA and syphilis RPR, and process serum samples to send to US NMRCDC for the HIV-1 ELISA and Western blot. Impacta laboratory is in the process of getting Proficiency Testing from the College of American Pathologists for HIV-1 ELISA and Western

blot. After getting Proficiency Testing, HIV-1 ELISA and Western blot will be done at the Impacta Laboratory. The local laboratories will also aliquot a serum sample for storage from participants who consent to storage for future testing. Once daily, a courier from Impacta will transport specimens from the local laboratories (Impacta and Via Libre) to US NMCRD, and will pick up print-outs of laboratory results for each of the two clinic sites. The local laboratories participating in the Peru HPTN and HVTN studies will be coordinated and supervised by Rosa Galvan, microbiologist, who was responsible for the laboratory aspects of the Peru sentinel surveillance system and the Alaska MSM cohort in Lima.

The technicians at local laboratories (at Impacta and Via Libre clinics) and NMCRD have been trained in universal precautions and biohazard containment. Appropriate blood and secretion precautions will be employed by all personnel, including personnel at US NMCRD, in the drawing of blood and shipping and handling of all specimens for this study, as currently recommended by the US Centers for Disease Control and Prevention.

The HVTN Central Laboratory conducted a site evaluation of the Peru HPTU and HVTU local laboratories and NMCRD laboratory in January 2001, in conjunction with the training on the less sensitive EIA.

8 ADMINISTRATIVE PROCEDURES

8.1 Study Coordination

Study implementation will be directed by this protocol as well as a common study-specific procedures manual (with site-specific sections as appropriate). This manual will outline procedures for conducting study visits, collecting and submitting study data, and other study operations. Study case report forms have been developed by the protocol team. Data will be faxed via DataFax to the SDMC.

We will utilize CASI, based on a successful pilot of CASI among MSM in Perú in August 2000. Data will be transferred to the Seattle HPTU site by internet transfer and cleaned. Data will be stored at both Impacta and the Seattle HPTU on tape. Quality control reports and queries will be routinely sent back to the site for verification and resolution.

Close cooperation between the protocol chairs, site investigator, NIAID Representative, protocol coordinator, biostatistician, data managers, and other study team members will be necessary in order to track study progress, respond to queries about proper study implementation, address issues in a timely manner, and assure consistent case management, documentation, and information sharing. Rates of accrual, follow-up, and protocol compliance will be monitored closely by the protocol chairs in the every other week conference calls and email communication, and will be communicated to the representatives of the HPTN CORE.

A common study laboratory manual will be followed to standardize specimen collection, preparation, processing and shipping. Oversight for laboratory procedures (including QA/QC) will be provided by the HPTN Central Laboratory.

8.2 Study Monitoring

On-site study monitoring will be performed in accordance with HPTN 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, study-specific procedures manual, and locally-accepted HIV counseling practices; and confirm the quality and accuracy of information collected at the study site and entered into the study database. The site investigator will allow study monitors and officials to inspect study facilities and documentation (e.g., informed consent forms, clinic and laboratory records, other source documents, and case report forms), as

well as observe the performance of study procedures. The investigator also will allow inspection of all study-related documentation by authorized representatives of the HPTN CORE, SDMC, NIAID, and US and Peruvian regulatory authorities. A site visit log will be maintained at the study site to document all visits.

8.3 Protocol Compliance

The study will be conducted in full compliance with the protocol. With the exception of modifications required to eliminate immediate participant safety concerns, the protocol will not be amended without prior written approval by the Protocol Chair or designee; protocol amendments requiring IRB approval must be submitted to and approved by the relevant site IRBs/ECs and the HPTN CORE prior to implementing the amendment.

8.4 Investigator's Records

The study site investigator will maintain, and store in a secure manner, complete, accurate, and current study records throughout the study. The investigator will retain all study records for at least five years after the completion of the study, unless directed otherwise by the HPTN CORE. Study records include administrative documentation — including site registration and initiation documents and all reports and correspondence relating to the study — as well as documentation related to each participant screened and/or enrolled in the study — including informed consent forms, locator forms, data collection forms, notations of all contacts with the participant, and all other source documents.

8.5 Use of Information and Publications

Publication of the results of this study will be governed by DAIDS policies. Any presentation, abstract, or manuscript will be made available by the investigator to the HPTN Manuscript Review Committee, and DAIDS for review prior to submission.

REFERENCES

1. Pan American Health Organization, World Health Organization. AIDS Surveillance in the Americas. 1998.
2. Tabet S, de Moya EA, Holmes KK, et al. Sexual behaviors and risk factors for HIV infection among men who have sex with men in the Dominican Republic. AIDS 1996; 10: 201-6
3. Tabet S, Sanchez J, Goichoea P, et al. HIV in men who have sex with men (MSM): Potential bridging population to heterosexuals. In prep.
4. Sanchez J, et al Gender differences in sexual practices and sexually transmitted infections among adults in Lima, Peru. Am J Public Health 1996; 86: 1098-1107
5. Carceres CF, Rosasco AM. The correlates of safer behavior among homosexually active men in Lima. AIDS 1997; 11 (S1): S53-9.
6. Holmes KK, Celum C, Ghee AE. Evaluation of the USAID project "AIDS HELP in Peru." Submitted by the Synergy Project. TvT Associates, Inc and University of Washington, 2000 (available at tvt@tvtassoc.com)
7. Sanchez J, Zuckerman R, Whittington W, et al. Intersecting epidemics of HIV and STDs among MSM in Peru. Submitted to 14th meeting of the Int'l Society for STD Research (ISSTD), June 2001.
8. Morris M, Zavixca J, Dean L. Social and sexual networks: Their role in the spread of HIV/AIDS among young gay men. AIDS Education and Prevention, 1995; 7S: 24-35.
9. Morris et al. Bridge populations in the spread of HIV/AIDS in Thailand. AIDS 1996; 10: 1265-71
10. Morris M, Kretzschmar M. Concurrent partnerships and the spread of HIV. AIDS 1997; 11: 641-8
11. Konde-Lule JK, Sewankumba N, Morris M. Adolescent sexual networking: HIV transmission in rural Uganda. Health Transit Rev 1997; 7S: 89-100.
12. Rawal BD, Degula C, Lebedeva M, et al. Equivalence of Dilutional "Vironostika" EIA (DILVIRON) with 3A11 LS-EIA for detecting early HIV-1 infection in clinical and epidemiological studies. In prep
13. Janssen RS, Satten GA, Stramer SL, et al. New testing strategy to detect early HIV-1 infection for use in incidence estimates and for clinical and prevention purposes. JAMA 1998; 280: 42-48.
14. Broadhead R, Heckathorn D, Weakliem D, et al. Harnessing peer networks as an instrument for AIDS prevention: Results from a peer-driven intervention. Pub Health Reports 1998; 113(S1): 42-57
15. Liang, KY, Zeger SL. Longitudinal data analysis using generalized linear models. Biometrika. 1986; 73:13-22.

Appendix I

HPTN 036

HIV prevalence, incidence and HSV-2 prevalence among high-risk MSM in Lima, Perú

SCHEDULE OF EVENTS

PROCEDURE	Screening Visit	Enrollment Visit	Follow-up Visits	Follow-up Results Visits
Assign/confirm participant ID number	X	X	X	X
Determine eligibility for Screening Visit	X			
Obtain informed consent for study participation	X			
Collect/update locator information	X	X	X	X
Administer Screening Questionnaire	X			
Administer Follow-up Questionnaire			X	
Deliver HIV/STD pre-test counseling	X		X	
Obtain informed consent for HIV testing	X		X	
Administer STD Symptoms Questionnaire	X		X	
Genital and rectal exam if STD symptoms reported	X		X	
Blood draw for HIV ELISA testing	X		X	
Blood draw for syphilis testing	X		X	
Blood draw for HSV-2 ELISA (MRL)	X		X ^ψ	
Deliver syphilis result and post-test counseling	X		X	
Deliver HIV/STD (except syphilis) result and post-test counseling		X		X ^π
Administer Acyclovir Trial Questionnaire		X ^{&}		X ^{&}
Determine eligibility for cohort participation		X ^π		
Administer Retroviral Symptoms Questionnaire		X		X
Refer or provide treatment for syphilis ^θ		X		X
Provide contact information and instructions	X	X	X	X
Schedule next visit/confirm schedule	X	X	X	X
Complete and enter data collection forms	X	X	X	X

^ψ At month 12 only and only if HSV-2 uninfected at screening

^π Refer to HIV early infection study if recently infected at enrollment visit (based on nonreactive less sensitive ELISA and reactive standard ELISA) or an HIV seroconverter at follow-up visits

[&] For those who test HSV-2 positive

^θ For those who test positive for syphilis