Review Response Submission Form (Version 1.0)

1.0 Review Response Submission Form

You have received this form either during the administrative pre-review process or after formal review by the committee. This form allows you to respond to recommendations, stipulations, or other issues identified during this review process. Make the appropriate revisions to your submission and attach the new versions in the Revised Submission Materials section.

TIP: We recommend saving this form frequently to avoid loss of work due to being timed out of your session. To save the form, change all the drop down answers in the stipulations to "Yes," "No," or "N/A" and click "Save Form."

2.0 General Information

2.1 Principal Investigator:

[redacted]

2.2 Study Title:

HIV, EBV and HPV interaction with mucosal epithelium

2.3 Study Number:

10-03277

2.4 Study Alias:

H8597-30664-04

3.0 Stipulations and Comments

3.1 To address each stipulation, you need to update the Study Application, submission form or document to which the stipulation is linked. To do so, click "Add Revision" the first time you revise the item or click the component name if you have already added a revision. For help, click the Help section in the upper right-hand corner and read the "Responding to Requests for Submission Corrections" quick guide. Please also write your responses to each of the stipulations in the Details fields rather than at the end of the form. (The system keeps a history of stipulations and responses and it doesn't work if it's not used right.)

Stipulations that must be addressed:

Stipulation 1 out of 4:

Please delete the following text from section 20.1:

"A. Risks and Discomforts:
*1. **Describe the risks and discomforts** of any investigational or approved drugs, devices and procedures being used or assigned for study purposes. Describe the expected frequency of particular side effects. If subjects are restricted from receiving standard therapies during the study, please also describe the risks of those restrictions."*

Stipulation Type: **(Stipulation must be addressed)**

Do you accept this Stipulation?	○ **N/A** • **Yes** ○ **No**
Provide an explanation on how you addressed this Stipulation:	We deleted the indicated text from section 20.1

Stipulation 2 out of 4:

Description:

Section 22.1 - Select a source for the study data. Please refer to the previously approved application.

Stipulation Type: **(Stipulation must be addressed)**

Do you accept this Stipulation?	○ **N/A** • **Yes** ○ **No**
Provide an explanation on how you addressed this Stipulation:	We selected a source for the study data.

Stipulation 3 out of 4:

Description:

Section 23.2 - Please delete the response entered under "Describe the additional safeguards that have been included in the study to protect the rights and welfare of these subjects and minimize coercion or undue influence:". The previously approved application did not have a response for this section. Also, the information provided is already in the appropriate section of the Imedris application (section 25.1).

Stipulation Type: **(Stipulation must be addressed)**

Do you accept this Stipulation?	○ **N/A** • **Yes** ○ **No**
Provide an explanation	

addressed this Stipulation:	We deleted the indicated text from the section 23.2

Stipulation 4 out of 4:

Description:

Section 26.1 - Please select "Advertisements, notices, and/or media used to recruit subjects..."

Stipulation Type: **(Stipulation must be addressed)**

Do you accept this Stipulation?	○ **N/A** • **Yes** ○ **No**
Provide an explanation on how you addressed this Stipulation:	In the section 26.1 we selected above indicated text.

Comments That Must Be Addressed With Follow-up Deadlines:

No Stipulation entered.

Comments:

No Stipulation entered.

4.0 Unresolved Stipulations/Comments

4.1

No Stipulation is outstanding.

5.0 Revised Submission Materials

5.1 A copy of the materials you submitted most recently as part of this submission is attached. Click the green bar to access these items, make changes, and attach new or revised documents.

6.0 Response Comments

6.1 Additional comments about this response:

Initial Review Submission Packet (Version 1.1)

1.0 Initial Review Submission Packet

1.1 Today's Date:

08/24/2010

1.2 Study Title:

HIV, EBV and HPV interaction with mucosal epithelium

1.3 IRB #:

1.4 Principal Investigator:

[redacted]

1.5 * Lay summary (1 to 3 brief sentences):

Interaction of HIV and HIV-associated opportunistic viruses, including EBV and HPV with adult oral and genital mucosal and fetal oral and GI tact mucosal epithelium is not well understood. Therefore, our goal is to establish an ex vivo organ culture model from the adult and fetal mucosal epithelium, to study the molecular mechanisms of HIV, EBV and HPV interactions with the mucosal epithelium of the adult and fetus, and to determine the role of amniotic fluid and breast milk in HIV dissemination via the fetal epithelium.

1.6 * This submission is a:

- ○ New study (never been approved before)
- ◉ Currently approved study due for continuing review (also submit a Continuing Review application in iMedRIS)
- ○ Currently approved study that is being modified (also submit a Modification application in iMedRIS)
- ○ Currently approved study not due for continuing review with no modifications

1.7 For currently approved studies, provide the CHR approval number:

H8597-30664-04

1.8 Special processing instructions or information about the submission:

2.0 CHR Application Form

2.1 * Attach the IRB application you completed for this protocol:

Edit/ View	Version	Title
	1.1	Study Application (Version 1.1) - Attached

3.0 Consent Documents

3.1 * Attach the informed consent documents (consent/assent forms, consent scripts, and/or information sheets) for this protocol: When possible, attach Word documents instead of PDFs.

Version	Sponsor Version	Title	Category	Language	Expiration Date	Consent Outcome	View Document
1.0		Consent-saliva-HIV+		English	06/03/2012	Expired	136.54 KB
1.0		Consent-saliva-HIV-		English	06/03/2012	Expired	131.04 KB
1.0		Consent-oral biopsy-HIV+		English	06/03/2012	Expired	147.09 KB
1.0		Consent-oral biopsy-HIV-		English	06/03/2012	Expired	147.13 KB
1.0		Consent-cervical biopsy-HIV+		English	06/03/2012	Expired	146.61 KB
1.0		Consent-cervical biopsy-HIV-		English	06/03/2012	Expired	146.62 KB
1.0		Consent-blood-HIV+		English	06/03/2012	Expired	136.13 KB
1.0		Consent-blood-HIV-		English	06/03/2012	Expired	130.56 KB
1.0		Consent breast milk-		English	06/03/2012	Expired	127.18 KB
1.0		Consent AF-HIV+		English	06/03/2012	Expired	127.58 KB
1.0		Consent AF-HIV		English	06/03/2012	Expired	128.26 KB

4.0 Other Study Documents

4.1 Attach the other study documents (e.g. protocol, investigators brochure, recruitment materials, instruments, case report forms, study handouts or other miscellaneous documents):

Version	Sponsor Version	Title	Category	Expiration Date	Document Outcome	View Document

1.0		CHRP grant-research design			Approved	165.41 KB
1.0		NIH R21 grant-Research design section			Approved	155.66 KB
1.0		Research saliva-HIV+	Recruitment materials		Approved	75.53 KB
1.0		Research saliva-HIV-	Recruitment materials		Approved	74.83 KB
1.0		Research oral-HIV+	Recruitment materials		Approved	79.57 KB
1.0		Research oral-HIV-	Recruitment materials		Approved	79.19 KB
1.0		Research cervical-HIV+	Recruitment materials		Approved	80.28 KB
1.0		Research cervical-HIV-	Recruitment materials		Approved	80.27 KB
1.0		Research BM-HIV-	Recruitment materials		Approved	89.96 KB
1.0		Research blood-HIV+	Recruitment materials		Approved	76.80 KB
1.0		Research blood-HIV-	Recruitment materials		Approved	76.09 KB
1.0		Research AF-HIV-doc	Recruitment materials		Approved	75.36 KB

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1.0		Consent-cervical biopsy-HIV+		English	06/03/2012	Expired	146.61 KB
1.0		Consent-cervical biopsy-HIV-		English	06/03/2012	Expired	146.62 KB
1.0		Consent-blood-HIV+		English	06/03/2012	Expired	136.13 KB
1.0		Consent-blood-HIV-		English	06/03/2012	Expired	130.56 KB
1.0		Consent breast milk-		English	06/03/2012	Expired	127.18 KB
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1.0		Research blood-HIV-	Recruitment materials		Approved	76.09 KB
1.0		Research AF-HIV-doc	Recruitment materials		Approved	75.36 KB

Study Application (Version 1.1)

1.0 General Information

***Enter the full title of your study:**

HIV, EBV and HPV interaction with mucosal epithelium

***Enter the study alias:**

H8597-30664-04
* This field allows you to enter an abbreviated version of the Study Title to quickly identify this study.

2.0 Add departments

2.1 and Specify Research Location:

Is Primary?	Department Name
◉	**UCSF -** [redacted]

3.0 List the key study personnel: (Note: external and affiliated collaborators who are not in the UCSF directory can be identified later in the Qualifications of Key Study Personnel section at the end of the form)

3.1 *Please add a Principal Investigator for the study:

[redacted]

Select if applicable

☐ Department Chair ☐ Resident

☐ Fellow

If the Principal Investigator is a Fellow, the name of the Faculty Advisor must be supplied below.

3.2 If applicable, please select the Research Staff personnel

A) Additional Investigators

[redacted]

[redacted]

B) Research Support Staff

[redacted]

3.3 *Please add a Study Contact

[redacted]

The Study Contact(s) will receive all important system notifications along with the Principal Investigator. (e.g. The project contact(s) are typically either the Study Coordinator or the Principal Investigator themselves).

3.4 If applicable, please add a Faculty Advisor/Mentor:

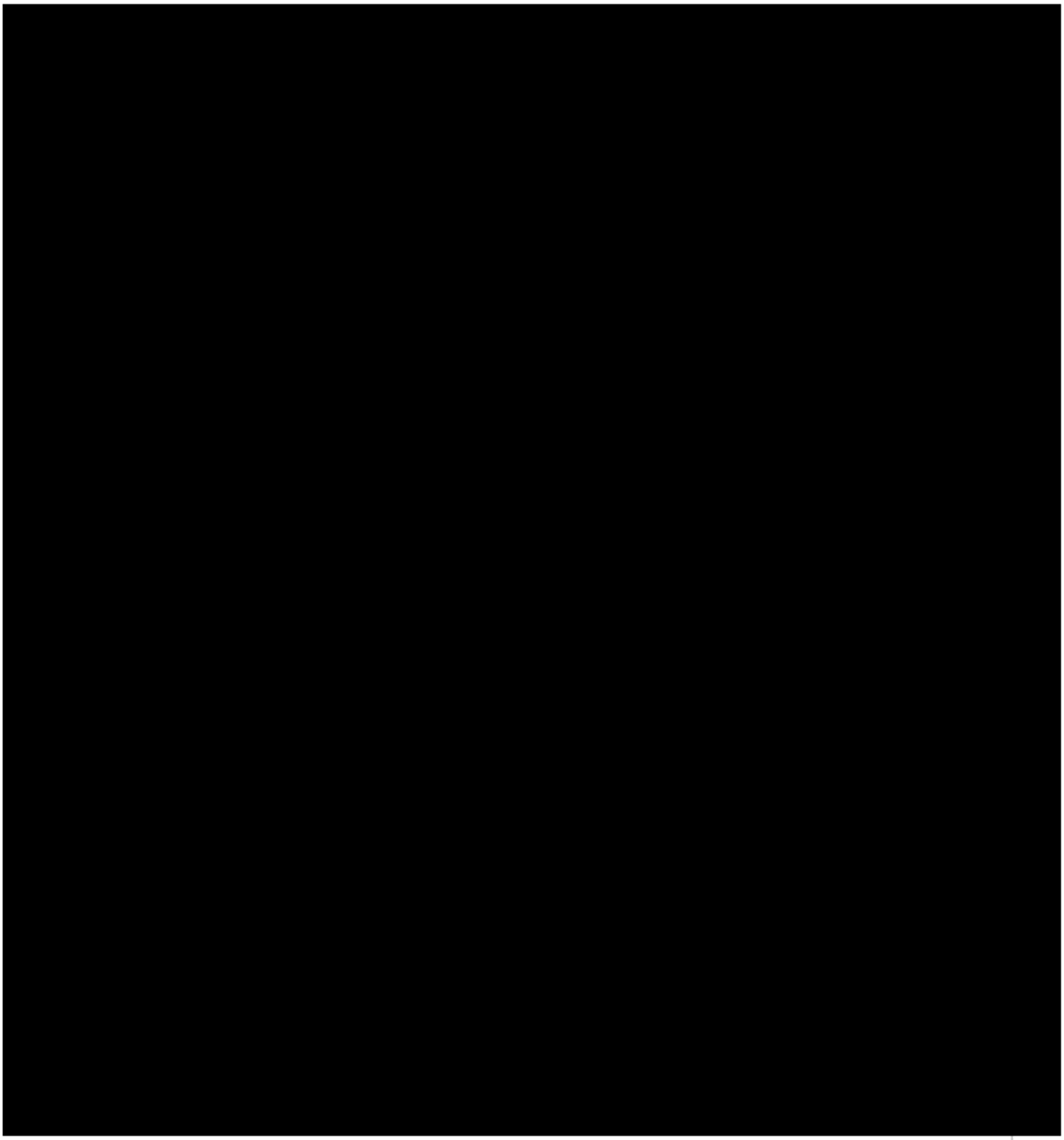
3.5 If applicable, please select the Designated Department Approval(s)

Add the name of the individual authorized to approve and sign off on this protocol from your Department (e.g. the Department Chair or Dean).

4.0 Qualifications of Key Study Personnel

4.1 List the study responsibilities and qualifications of any individuals who qualify as Key Study Personnel (KSP) by clicking the "Add a new row" button:

KSP Name	Description of Study Responsibilities	Qualifications
[redacted]		



5.0 Initial Screening Questions

5.1 * This study involves human stem cells, gametes or embryos:

- (•) No
- ( ) Yes, and requires CHR and GESCR review
- ( ) Yes, and requires GESCR review, but NOT CHR review

5.2 * This application involves a Humanitarian Use Device:

- (•) No
- ( ) Yes, and it includes a research component
- ( ) Yes, and it involves clinical care ONLY

5.3 * This is a CIRB study (e.g. the NCI CIRB will be the IRB of record):

- ( ) Yes (•) No

5.4 * This application includes a request to rely on another UC IRB to be the IRB of record:

○ Yes ◉ No

Note: If this request is approved, the CHR will **NOT** review and approve this study. Another UC campus will be the IRB of record.

6.0 Application Type

6.1 * This research involves:

◉ Minimal risk
○ Greater than minimal risk

6.2 * This application is:

◉ Full Committee
○ Expedited
○ Exempt

6.3 If you think this study qualifies for expedited review, select the regulatory category(ies) that the research falls under:

☐ Category 1: A very limited number of studies of approved drugs and devices
☐ Category 2: Blood sampling
☐ Category 3: Noninvasive specimen collection
☐ Category 4: Noninvasive clinical procedures
☐ Category 5: Research involving materials that were previously collected for either nonresearch or research purposes
☐ Category 6: Use of recordings
☐ Category 7: Low risk behavioral research
☐ Category 8: Renewal of inactive research protocols or protocols that are essentially complete
☑ Category 9: Renewal of other minimal risk research protocols

6.4 * This study involves:

◉ Subject contact (including phone, email or web contact)
○ No subject contact (limited to medical records review, biological specimen analysis, and/or data analysis)

7.0 Funding

7.1 Identify all the funding sources and their roles on the project:

View Details	Sponsor Name	Sponsor Type	Awardee Institution:	Contract Type:	Project Number	UCSF RAS System Award Number ("A" + 6 digits)
⊟	California HIV/AIDS Research Program	14	UCSF	Grant		A115176

Sponsor Name:	California HIV/AIDS Research Program
Sponsor Type:	14
Sponsor Role:	
Awardee Institution::	UCSF
Is Institution the Primary Grant Holder:	Yes
Contract Type:	Grant
Project Number:	
UCSF RAS System Award Number ("A" + 6 digits):	A115176
Grant Number for Studies Not Funded thru UCSF:	
Grant Title:	HIV tat- and gp120-facilitated HPV epithelial entry
PI Name: (If PI is not the same as identified on the study.)	
Explain Any Significant Discrepancy:	

⊟	NIH Natl Inst Dental & Craniofacial Res.	01	UCSF	Grant		A115176

Sponsor Name:	NIH Natl Inst Dental & Craniofacial Res.
Sponsor Type:	01
Sponsor Role:	Funding
CFDA Number:	
Grant/Contract Number:	
Awardee Institution::	UCSF
Is Institution the Primary Grant Holder:	Yes
Contract Type:	Grant
Project Number:	
UCSF RAS System Award Number ("A" + 6 digits):	A115176
Grant Number for Studies Not Funded thru UCSF:	
Grant Title:	HIV TRANSCELLULAR AND TRANSSYNAPTIC PENETRATION OF MUCOSAL EPITHELIUM
PI Name: (If PI is not the same as identified on the study.)	
Explain Any Significant Discrepancy:	

7.2 If you tried to add the sponsor in the question above and it was not in the list, check here:

○ Sponsor not in list

Only if your sponsor is not yet in the list, type the sponsor's name:

If the sponsor is not in the system, download the C&G Add Sponsor Form from Help link and attach it to this application. **Your study will not receive CHR approval until the sponsor and funding details have been added to your application.**

7.3 For Federally funded studies only, indicate which portion of your grant you will be attaching:

- [x] The Research Plan, including the Human Subjects Section of your NIH grant
- [ ] For other federal proposals (contracts or grants), the section of the proposal describing human subjects work
- [ ] The section of your progress report if it provides the most current information about your human subjects work

7.4 If this study has no sponsor, check all that apply:

- [ ] Unfunded student project
- [ ] Unfunded (miscellaneous departmental funding)
- [ ] Specific departmental funding

8.0 Statement of Financial Interest

8.1 * The Principal Investigator and/or one or more of the key study personnel has financial interests related to this study:

- ( ) Yes
- (•) No

If **Yes**, attach the **Disclosure of Investigators' Financial Interests Supplement** to this application.

9.0 Sites

9.1 Institutions (check all that apply):

- [x] UCSF
- [ ] Mt. Zion
- [ ] San Francisco General Hospital (SFGH)
- [ ] SF VA Medical Center (SF VAMC)
- [ ] Helen Diller Family Comprehensive Cancer Center
- [ ] Fresno (Community Medical Center)
- [ ] Blood Centers of the Pacific (BCP)
- [ ] Blood Systems Research Institute (BSRI)
- [ ] Gallo
- [ ] Gladstone
- [ ] Institute on Aging (IOA)
- [ ] SF Dept of Public Health (DPH)

9.2 Check all the other types of sites not affiliated with UCSF with which you are cooperating or collaborating on this project:

- [ ] Foreign Country

List:

- [ ] Other UC Campus
- [ ] Other institution
- [ ] Other community-based site

9.3 * This is a multicenter study:

- ( ) Yes (•) No

9.4 Check any research programs this study is associated with:

- [ ] Cancer Center
- [ ] Center for AIDS Prevention Sciences (CAPS)
- [ ] Global Health Sciences
- [ ] Immune Tolerance Network (ITN)
- [ ] Osher Center
- [ ] Positive Health Program

10.0 Study Design

10.1 Study design:

HIV mucosal transmission is a potentially important, poorly understood route of HIV infection that has global consequences. The oropharyngeal mucosal epithelium of the fetus/neonate and adult genital mucosa may serve as an efficient portal of entry for HIV, yet oral transmission of HIV among adults is rare (2, 10, 14, 15). The reasons for this difference are not well understood. The goal of our study is to investigate molecular mechanisms of resistance of adult oral epithelial cells and the susceptibility of fetal oral and adult genital epithelial cells for HIV infection. We hypothesize that resistance of adult oral epithelial cells for HIV infection is due to the expression of multiple anti-HIV innate molecules, including calprotectin, defensins, lactoferrin, secretory leukocyte protease inhibitor, thrombospondin, polyanionic albumins, acidic proline-rich proteins, and salivary agglutinins (12). These proteins may not be expressed or may not have anti-HIV function in fetal/neonatal oral and adult genital epithelial cells. These molecules are present in amniotic fluid and breast milk, however, their role in HIV infection with fetal/neonatal mucosal epithelium in not clear. Therefore we will investigate role of fetal mucosal epithelium in HIV transmission, as well as role of amniotic fluid and breast milk in HIV transmission via fetal oral mucosal epithelium. We will compare expression of innate proteins in the fetal and infant oral epithelia. We also will study HIV transmission via cervical mucosal epithelium, and expression of innate proteins in cervical epithelial cells.

It is well known that during the systemic HIV infection the innate immune and barrier functions of mucosal epithelium may be impaired (8, 13). This could be due to reactivation and dissemination of common oral pathogens such as EBV and HPV within the oral epithelium, which may lead to disruption of oral epithelium and lead to HIV transmission (1, 3-5, 7, 9). Therefore, our research will focus to investigate the molecular mechanisms of HIV, EBV and HPV interactions with mucosal epithelium.

10.2 Check all that apply:

- [ ] Phase I
- [ ] Phase II
- [ ] Phase III
- [ ] Phase IV

11.1 Hypothesis:

This study has a hypothesis:

(•) Yes ( ) No

If yes, state the hypothesis or hypotheses:

Hypothesis: A tissue explants of adult oral and cervical, and fetal andoropharyngeal, gastric and intestinal mucosal epithelia ex vivo will retain its natural tissue architecture, including the epithelium, lamina propria, and intraepithelial and submucosal immune cells. This model will be suitable to evaluate HIV-1 transmission from mother to fetus or neonate via mucosal epithelium.
- Hypothesis: Expression of HBD1, HBD2, HBD3 and SLPI in adult oral epithelium may be substantially higher than in infant and fetal oral epithelia, and this may determine transepithelial transmission of HIV via adult and infant/fetal epithelia.
- Hypothesis: HIV spread across the mucosal epithelium may occur by two parallel mechanisms: (i) cells may express one or more of the HIV co-receptors CXCR4, CCR5 and GalCer. Cell-free X4- or R5-tropic HIV from amniotic fluid, cervicovaginal secretions or breast milk may bind to appropriate HIV co-receptors and GalCer on the surface of the epithelium, initiating entry into and infection of epithelial cells by X4- and/or R5-tropic HIV strains; (ii) cell-associated X4- or R5-tropic HIV from amniotic fluid, cervicovaginal secretions or breast milk may infect the epithelium by mechanisms of cell-cell spread; and (iii) the interaction of cell-free or cell-associated HIV with the apical surface of the fetal/neonatal epithelium may initiate transcytosis of virions from apical to basolateral membranes of mucosal epithelial cells. These mechanisms may lead to dissemination of HIV infection into immune cells in the lamina propria, and therefore may cause systemic HIV/AIDS disease in the fetus and neonate. HIV resistance in adult oral epithelium could be due to absence of HIV receptors or high level of anti-HIV proteins expression.
Hypothesis: Amniotic fluid and breast milk contain C-C (MIP-1a, MIP-1band RANTES) and C-X-C (SDF-1) chemokines, and anti-viral innate moleculles (calprotectin, defensins), which may modulate HIV infection of the oral mucosal epithelium of the fetus/neonate: (i) C-C and C-X-C chemokines in amniotic fluid and breast milk may suppress cell-free HIV infection or transcytosis via mucosal epithelial cells; (ii) however, these chemokines may not affect GalCer-mediated HIV entry or transcytosis of HIV and cell-to-cell spread of virus from HIV-infected maternal cells into the mucosal epithelium of the fetus/neonate; (iii) HIV-positive pregnant woman may have a higher rate of upper and lower genital tract and breast infection/inflammation, which may lead to activation in the genital tract and in amniotic fluid and breast milk of pro-inflammatory cytokines, including tumor necrosis factor (TNF)-a, interferon (IFN)-g, interleukin (IL)-6 and IL-8. Increased levels of such cytokines may disrupt cellular junctions of the mucosal epithelium by apoptotic and /or non-apoptotic mechanisms and thereby impair their barrier function. This impairment leads to penetration of cell-free or cell-associated HIV via the paracellular space.
- Hypothesis: During HIV/AIDS disease, HIV tat protein is secreted into the circulation and binds to the b1 family integrinsof peripheral blood monocytes via tat's RGD motif. This binding activates integrin signaling and their clustering within the lipid rafts of monocyte membranes, facilitating EBV infection by interaction of EBV BMRF-2 with b1 integrins. HIV and/or EBV infected monocytes may migrate into adult oral mucosal epithelium and reduce its innate immune and barrier functions.
- Hypothesis: HIV-infected immune cells migrate into oral epithelium and secrete TNF-a, which in turns lead to disruption of epithelial cell junctions. HIV/TNF-a-mediated disassembly of cell junctions facilitates entry of HPV into the basal layers of epithelium.

11.2 List the specific aims:

Aim 1. Establish and characterize an ex vivo organ culture model from the oral and cervical of the adult, and oropharyngeal, gastric and intestinal mucosal epithelium of the fetus.
Aim 2. Analyze expression of HBD1, HBD2, HBD3 and SLPI in adult, infant and fetal oral epithelia.
Aim 3. Study mechanisms of HIV spread via the oral and cervical of the adult, and oropharyngeal, gastric and intestinal mucosal epithelium of the fetus.
Aim 4. Determine the role of amniotic fluid and breast milk in the interaction of HIV with the mucosal epithelium of the fetus.
Aim 5. Study the mechanisms of HIV-EBV co-infection of circulating monocytes and its role in dissemination of HIV and EBV infection within the oral mucosal epithelium.

Aim 6. Determine the role of HIV-mediated disruption of mucosal epithelial tight junctions in paracellular spread of HPV.

11.3 Statistical analysis:

Data will be analyzed by a study steering committee, which will be composed by [redacted]. It is estimated that 60 adult, 100 fetal and 20 infant tissues will investigated for this study. The 150 blood and saliva samples from HIV-positive with HAART, 150 from HIV-positive w/o HAART and 150 from HIV-negative will be examined for HIV and EBV infection. Fifteen amniotic fluid samples from HIV-negative and 4 from HIV-positive pregnant women will be investigated. Ten breast milk from HIV-negative woman will be analyzed. A three years cumulative enrollment is planned.
HIV-1 infection of the fetal mucosal epithelium will be evaluated in the presence or absence of breast milk or amniotic fluid by detection of HIV-1 signals. At least five tissue explants will be used for each breast milk sample and untreated control. As fetal epithelial tissues will be treated and untreated with breast milk, the statistical analysis will account for the paired nature of the data, i.e., HIV infection or transcytosis in tissue explants treated with breast milk compared with untreated controls. Quantitative data for statistical analysis of the number of cells containing HIV infection will be obtained by screening 30–50 fields per tissue section. We will test the null (no difference) hypothesis against the alternative hypothesis that there is a difference. With five tissue samples, using a paired *t* test with a 5% false-positive rate, we will have 80% power to detect a difference between treated and untreated samples.

11.4 * This is an investigator-initiated study:

(•) Yes ( ) No

11.5 This study has received scientific or scholarly review from (check all that apply):

- [ ] Cancer Center Protocol Review Committee (PRC) (Full approval or contingent PRC approval is required prior to final CHR approval for cancer-related protocols.)
- [ ] CTSI Clinical Research Center (CRC) advisory committee
- [ ] Departmental scientific review
- [x] Other:

Specify **Other**:

Committee on human research

If applicable, attach the **Departmental Scientific Review Form** at the end of the application.

12.0 Background

12.1 Background:

It is well documented that HIV oral transmission in adult population is rare, and oral mucosal epithelium is resistant for HIV infection (4, 6, 8). However, the oropharyngeal mucosal epithelium of the fetus/neonate may serve as an efficient portal of entry for perinatal and postnatal mother-to-child transmission (MTCT) of HIV (6). It was also well documented that HIV transmission via genital epithelium is substantially higher than the oral epithelium. MTCT of HIV may occur in utero before birth (prenatal), during labor and delivery (perinatal) or after birth (postnatal) (6). In prenatal MTCT, RNA/DNA and proteins were detected in placental trophoblasts, endothelial cells, and villous Hofbauer cells from 8 weeks of gestation, suggesting HIV transmission by the transplacental route (6). An alternative prenatal route could be transamniotic, where cell-free HIV virus and/or HIV-infected cells may penetrate into the amniotic sac and infect the fetus via the oropharyngeal cavity or gastrointestinal (GI) tract. A case report showed that HIV was detected in the amniotic fluid of HIV-positive women at 32 weeks gestation (6). Another showed that HIV-1 was detected in gastric aspirates of a 15-week-old fetus (6). Enzyme-linked immunosorbent assay (ELISA) analysis of HIV-1 p24 in samples of amniotic fluid from 10 HIV-positive women showed that 8 women were positive for p24 (6). Maiques et al.

amniotic fluid before HAART and in 3% of the fluid under HAART (6). Mohlala et al. showed that HIV-1 was not detected in the amniotic fluid of 23 HIV-positive women under HAART who had a normal pregnancy, and these data indicate that HAART treatment substantially reduces MTCT of HIV in utero (11). The solid evidences indicate that postnatal MTCT of HIV via breastfeeding, and HAART treatment does not efficiently inhibit postnatal MTCT (6). All above evidence clearly indicate that fetal/neonatal mucosal epithelium play critical role in HIV MTCT. The higher rate of genital HIV transmission also indicate that genital epithelium may play critical role in HIV spread. Therefore our research focuses on HIV interaction with adult oral and genital, and fetal oral and GI mucosal epithelium. An adult oral mucosal epithelium is resistant for HIV infection. However, during the HIV/AIDS disease multiple opportunistic infections, including EBV and HPV may reactivate and disseminate within the oral epithelium and reduce its protective function for HIV infection. Therefore we will study mechanisms of HIV, EBV and HPV dissemination within the adult oral mucosal epithelium.

12.2 Preliminary studies:

12.3 References:

1. **Boulter, A. W., N. Soltanpoor, A. V. Swan, W. Birnbaum, N. W. Johnson, and C. G. Teo.** 1996. Risk factors associated with Epstein-Barr virus replication in oral epithelial cells of HIV-infected individuals. Aids **10:**935-40.
2. **Casper, C., and E. M. Fenyo.** 2001. Mother-to-child transmission of HIV-1: the role of HIV-1 variability and the placental barrier. Acta Microbiol Immunol Hung **48:**545-73.
3. **Chou, L. L., F. Boustany, and D. Nathanson.** 1996. G to A hypermutation in env loop V2 region of HIV-1 in oral hairy leukoplakia. J Dental Res **75:**115.
4. **Greenspan, D., and J. S. Greenspan.** 1997. Oral manifestations of HIV infection. AIDS Clin Care **9:**29-33.
5. **Greenspan, D., J. S. Greenspan, N. G. Hearst, L. Z. Pan, M. A. Conant, D. I. Abrams, H. Hollander, and J. A. Levy.** 1987. Relation of oral hairy leukoplakia to infection with the human immunodeficiency virus and the risk of developing AIDS. J Infect Dis **155:**475-81.
6. **Hille, J. J., J. Webster-Cyriaque, J. M. Palefski, and N. Raab-Traub.** 2002. Mechanisms of expression of HHV8, EBV and HPV in selected HIV-associated oral lesions. Oral Dis **8 Suppl 2:**161-8.
7. **Lau, R., J. Middeldorp, and P. J. Farrell.** 1993. Epstein-Barr virus gene expression in oral hairy leukoplakia. Virology **195:**463-74.
8. **Leigh, J. E., K. Shetty, and P. L. Fidel, Jr.** 2004. Oral opportunistic infections in HIV-positive individuals: review and role of mucosal immunity. AIDS Patient Care STDS **18:**443-56.
9. **Lucht, E., P. Biberfeld, and A. Linde.** 1995. Epstein-Barr virus (EBV) DNA in saliva and EBV serology of HIV-1-infected persons with and without hairy leukoplakia. J Infect **31:**189-94.
10. **Minkoff, H.** 2003. Human immunodeficiency virus infection in pregnancy. Obstet Gynecol **101:**797-810.
11. **Mohlala, B. K., T. J. Tucker, M. J. Besser, C. Williamson, J. Yeats, L. Smit, J. Anthony, and A. Puren.** 2005. Investigation of HIV in amniotic fluid from HIV-infected pregnant women at full term. J Infect Dis **192:**488-91.
12. **Moutsopoulos, N. M., T. Greenwell-Wild, and S. M. Wahl.** 2006. Differential mucosal susceptibility in HIV-1 transmission and infection. Adv Dent Res **19:**52-6.
13. **Patton, L. L., and C. van der Horst.** 1999. Oral infections and other manifestations of HIV disease. Infect Dis Clin North Am **13:**879-900.
14. **UNAIDS.** 2004. UNAIDS, AIDS epidemic update. Geneva:UNAIDS/WHO.
15. **UNAIDS.** 2006. UNAIDS, AIDS epidemic update. Geneva:UNAIDS/WHO.

If you have a separate bibliography, attach it to the submission with your other study documents.

13.0 Sample Size and Eligibility

13.1 Number of subjects that will be enrolled at UCSF and affiliated institutions:

555

13.2 Total number of subjects that will be enrolled at all sites:

13.3 Estimated number of people that you will need to consent and screen here (but not necessarily enroll) to get the needed subjects:

600

13.4 Sample size calculation:

13.5 * Eligible age range(s):

- [ ] 0-6 years
- [ ] 7-12 years
- [ ] 13-17 years
- [x] 18+ years

13.6 Inclusion criteria:

The only inclusion criterion will be willingness to donate blood, oral and cervical tissue, amniotic fluid and breast milk samples and have a known HIV-status, which were documented negative HIV test in the past 3 months.

13.7 Exclusion criteria:

The only exclusion criteria for isolation of oral and cervical explants, and amniotic fluid and breast milk from adult individuals would be inflammation or bacterial infection, since these factor may increase of risk for subsequent bleeding. Also, the exclusion criteria for isolation of tongue and buccal explants would be if volunteers have allergy to anesthetic.

13.8 There are inclusion or exclusion criteria based on gender, race or ethnicity:

○ Yes ◉ No

If **yes**, please explain the nature and rationale for the restrictions:

14.0 Drugs and Devices

14.1 * Drugs or biologics will be studied under this application:

○ Yes ◉ No

14.2 * Medical devices will be studied under this application:

○ Yes ◉ No

14.3 Verification of IND/IDE numbers: If the sponsor's protocol does not list the IND/IDE number, you must submit documentation from the sponsor or FDA identifying the IND/IDE number for this study. Attach this documentation in the Other Study Documents section of the Initial Review Submission Packet.

15.0 Other Approvals and Registrations

15.1 This is a clinical trial:

○ Yes ◉ No

Clinical Trial Registration
"NCT" number for this trial:

15.2 * This study involves human gene transfer or recombinant DNA research:

○ Yes ◉ No

15.3 This study involves other regulated materials and requires approval and/or authorization from the following regulatory committees:

☐ Institutional Biological Safety Committee (IBC)

Specify BUA #:

8597-BU-01-INC

☐ Institutional Animal Care and Use Committee (IACUC)

Specify IACUC #:

☐ Radiation Safety Committee

Specify RUA #:

☐ Radioactive Drug Research Committee (RDRC)

Specify RDRC #:

☐ Controlled Substances

16.0 Procedures

16.1 List all study procedures, test and treatments required for this study:

1. Collection of adult oral and cervical mucosal tissue and establishment of tissue explants ex vivo. The tongue and buccal explants (30 for each, total 60) will be obtained from HIV-positive and –negative adult volunteers. Cervical tissue also will be collected from HIV-positive and –negative premenopausal women.
First, donor from oral biopsies will be treated with local anesthetic agent lidocaine (4.5 mg/kg) for 5-10 min. After confirmation of numbness the biopsy procedure will be performed. Biopsies of the oral and cervical mucosa containing epithelium and connective tissue will be obtained using 4-mm–diameter biopsy punches in [redacted] Cervical tissue will be collected from [redacted]

Immediately after biopsy the tissue will be placed into a tube with 2 ml of RPMI medium (explant medium) containing 10% heat-inactivated fetal bovine serum, 20 mM HEPES, 100 mM glutamine, 20 µg/ml gentamicin, 100 U/ml penicillin, 100 µg/ml streptomycin, and 50 ng/ml amphotericin-B. Before use the explants will be washed three times with 0.5 ml cold explant medium. Also will be taken blood 20 ml of blood from each individual who will donate tongue and buccal explants.

2. Collection of fetal tongue, tonsil, gastric and intestinal tissue explants
Fetal tongue, tonsil, gastric and intestinal tissue explants (total 100) will be collected from discarded fetal tissues obtained following an elective termination of pregnancy at [redacted] We will not have access the any identifying documents associated with fetal materials. The tongue, tonsil, gastric and intestinal tissue explants containing stratified mucosal epithelium and submucosal stromal tissue will be obtained from discarded fetuses at 18 to 22 weeks of gestation, because, in most cases, abortion later than 23 weeks of gestation is generally not be permitted or performed. The 5-mm pieces of tissue will be dissected from the dorsal surface of the fetal tongue, cheek and tonsil by surgical instrument. Similarly sized pieces will be dissected from the gastric mucosa and small and large intestine. All mucosal samples will include the epithelium and lamina propria with connective tissues. Immediately after biopsy isolation, the tissues will be placed into a tube with 2 ml of RPMI medium (explant medium) containing 10% heat-inactivated fetal bovine serum (FBS), 20 mM HEPES, 100 mM glutamine, 20 µg/ml gentamicin, 100 U/ml penicillin, 100 µg/ml streptomycin and 50 ng/ml amphotericin-B. Before use, the explants will be washed three times with 0.5 ml cold explant medium. To establish a polarized organ culture system, the explants, with the epithelial layer oriented on top, will be placed in the top chamber on a Transwell-clear filter insert with a pore size of 0.45 µm and a diameter of 6.5 mm (#3472, Costar Corp., Cambridge, Mass). Filter inserts with explant will be incubated at 37 C in a humidified atmosphere containing 5% CO_2, and one third of the medium from each chamber will be changed every other day.
3. Collection of infant buccal and tonsil tissue explants from cadavers. The buccal and tonsil mucosa in the area of buccal and tonsil, respectively will be lifted with forceps. A sharp scalpel will be used to excise a small piece of (about 5 mm) tissues. The tongue and tonsil tissue explants containing stratified mucosal epithelium will be obtained from infants (cadavers) under 1 year old. Immediately after biopsy isolation, the tissues will be fixed with 3% paraformaldehyde and dehydrate with sucrose gradient, will be used confocal microscopy immunodetection analysis. We will not have access the any identifying documents associated with infant materials.

4. Infection of tissue explants with HIV and/or EBV.
Tissue explants mounted in filter inserts will be infected from their mucosal surface. For HIV infection HIV-1 X4 tropic SF33 and R5 tropic SF162 viruses will be used. For each explant 10^5 TCID50 virions will be used. For EBV B95-8 strain will be used and each explant will be infected with 10^5 virions. HIV and EBV co-infection will be performed by simultaneous infection of HIV and EBV viruses. Also, the tissue explants will be infected with cell-associated HIV and EBV. The PBMC and purified B-lymphocytes will be infected with HIV and EBV, respectively, and after 3-5 days these cells will be added to the mucosal surface of tissue explants.
5. Collection of amniotic fluid and breast milk.
To examine the role of amniotic fluid and breast milk in HIV infection of fetal mucosal epithelium we will coleect amniotic fluid and breast milk. Volunteers will donate amniotic fluid and breast milk for these studies. Five ml amniotic fluid from 10 healthy HIV-seronegative and 4 HIV-positive pregnant women will be collected at 18 and 22 weeks of gestation. Amniotic fluid samples will be collected by transabdominal amniocentesis with a single needle insertion, which will take place at [redacted] About 5-20ml of amniotic fluid will be aspirated into the syringe and centrifuged for 10 min at 1000 r/min. Pellet containing amniotic cells will be saved for karyotype analysis, and 5 ml of supernatant will be used for our research. Amniotic fluid will be filtered through 22-µm pore filters and aliquoted in 500 µl and stored at –80 C for further use in our research. Fifty ml of breast milk from 20 healthy HIV-seronegative women will be collected during the first 2-3 month of breast-feeding. Prior to sample collection, 2-3 ml of milk will be removed from the breast, and the nipple and surrounding areas of the breast that will have contact with the pump (Lactation Care Inc., Newton MA) will be cleaned with an ethanol-infused sterilizing pad and completely dried. The bottle and tubes will be sterilized by boiling them, according to manufacturer instruction. After collection breast milk samples will be filtered through 0.8-µm pore filters and aliquoted in 500 µl and stored at –80 C for further use in our research. It is expected that the donors will be between the ages of 18 and 40 years.
6. Collection of salivaand blood samples from HIV positive and negative individuals.
To study the status of EBV and HIV co-infection (lytic and latent) in monocytes and lymphocytes from HIV-positive individuals we will collect blood and saliva samples from 150 HIV-positive patients with HAART, 150 HIV-positive w/o HAART and 150 HIV-negative healthy volunteers. We

and blood samples. Collection of blood samples will be at [redacted]

If you have a procedure table, attach it to the submission with your other study documents.

16.2 Interviews, questionnaires, and/or surveys will be administered or focus groups will be conducted:

○ Yes ◉ No

List any standard instruments used for this study:

Attach any non-standard instruments at the end of the application.

16.3 Conduct of study procedures or tests off-site by non-UCSF personnel:

○ Yes ◉ No

If yes, explain:

16.4 Sharing of experimental research test results with subjects or their care providers:

○ Yes ◉ No

If yes, explain:

16.5 * Specimen collection for future research and/or specimen repository/bank administration:

◉ Yes ○ No

16.6 Time commitment (per visit and in total):

1. For obtaining one biopsy sample per visit will be required about 30 min. Each individual will be seen only once. From some of biopsy donors will be collected about 20 ml blood samples. For obtaining one blood sample will be required about 10-20 min. Each individual will be seen only once.
2. For obtaining one blood sample will be required about 10-20 min. Each individual will be seen only once.
3. For obtaining one saliva sample will be required about 10-20 min. Each individual will be seen only once.
4.The time to obtaining one breast milk sample will be about 10 min.

16.7 Locations:

1.The oral biopsy tissues from the adult tongue, and buccal mucosa from normal healthy individuals and HIV-positive patients will be collected in [redacted]
Cervical tissue will be collected from HIV-negative and -positive premenopausal women at the [redacted] For obtaining one biopsy sample per visit will be required about 30 min. Each individual will be seen only once. From some of biopsy donors will be collected about 20 ml blood samples. For obtaining one blood sample will be required about 10-20 min. Each individual will be seen only once.
2. The oral autopsy tissues will be obtained from infants (cadavers) under 1 year old by [redacted]

2. The blood samples from HIV/AIDS patients and normal healthy volunteers will be collected in [redacted] For obtaining one blood sample will be required about 10-20 min. Each individual will be seen only once.
3. The saliva samples from HIV/AIDS patients and normal healthy volunteers will be collected in [redacted] For obtaining one saliva sample will be required about 10-20 min. Each individual will be seen only once.
4. The fetal tongue, tonsil, gastric and intestinal tissue samples from discarded fetus will be collected in [redacted] There is no direct patient contact for this portion of the study.
5. Theamniotic fluid samples from normal healthy individuals and HIV/AIDS patients will be collected in [redacted] There will be no additional time requirement per patient apart from what is done in routine clinical care.
6. The breast milk samples will be collected in breastfeeding healthy HIV-negative women at [redacted] The time to obtaining one breast milk sample will be about 10 min.
7. All HIV and/or EBV and/or HPV infection procedures with adult and fetal tissue samples in presence of amniotic fluid and breast milk will be performed in [redacted]

16.8 Describe the resources in place to conduct this study in a way that assures protection of the rights and welfare of participants:

All specimens will be de-identified and designated by study numbers. The researchers and tissue bank staff will not have access to this information, rather they will have only study numbers. This numbers will be destroyed as soon as the study in completed. All data required by the protocol will be recorded and entered into electronic database using the unique study number. The study team will review the electronic data for accuracy. Data clarification or corrections will be made electronically and the databases will be password secured.

To minimize the discomfort of the oral tissue biopsy and transabdominal amniocentesis procedures,a local anesthetic is provided, followed by as-needed pain medications after the procedure. Sterile technique is used to avoid infection, and bleeding is stopped using standard clinical procedures

17.0 Specimen Collection for Future Research and/or Specimen Repository/Bank Administration

(Note: This section replaces the old "Human Biologic Specimen Collecting and/or Banking for Future Research" supplement form. Please do not attach the old form to this application.)

17.1 Specimens are (check all that apply):

- [ ] Surplus clinical specimens from a diagnostic or therapeutic procedure
- [x] Specimens collected for research purposes only
- [ ] Other

If Other, explain:

17.2 Types of specimens:

- [x] Blood
- [x] Tissue (describe below):
- [ ] Existing/archival materials (name source below): --

Describe and/or name source:

17.3 Consent will be obtained via:

- ☐ Separate specimen banking consent form
- ☑ Specimen banking section within a main research study consent form
- ☐ Surgical consent form with tissue donation brochure

17.4 Specimens will ultimately be stored (check all that apply):

UCSF

- ☑ UCSF repository/bank being established under this protocol
- ☐ Existing UCSF specimen repository/bank with CHR approval

Provide the name of the bank and CHR approval number (if not being banked at UCSF under this protocol):

Outside Entity

- ☐ Cooperative group bank
- ☐ NIH
- ☐ Other university
- ☐ Industry sponsor
- ☐ Other

Specify to what institution, cooperative group or company specimens will be transferred:

17.5 Direct identifiers will be sent with specimens or shared with other researchers and/or outside entities:

- ○ Yes
- ◉ No
- ○ N/A – Specimens will not be shared with others

If **Yes**, which identifiers will be sent with specimens:

- ☐ Name
- ☐ Date of birth
- ☐ Social Security number
- ☐ Medical record number
- ☐ Address
- ☐ Phone number
- ☐ Email address
- ☐ Other dates (surgery date, clinic visit dates, etc.)

If **Yes**, provide a justification for sending direct identifiers with the specimens:

18.0 Establishing a Specimen Repository/Bank at UCSF

(Note: This section replaces the old "Human Biologic Specimen Collecting

and/or Banking for Future Research" supplement form. Please do not attach the old form to this application.)

18.1 The repository/bank is physically located at (list the address and room number for all locations):

[redacted]

18.2 Methods for maintaining confidentiality:

- [x] Samples are completely de-identified before being added to the bank/repository. There is no way to link the specimens back to the subjects.
- [ ] Samples are coded and researchers are able to link the specimens to specific subjects. However, future recipients will not receive direct identifiers with the specimens.
- [ ] Samples are stored with direct identifiers in the repository. However, future recipients will not receive direct identifiers with the specimens.
- [ ] Samples are coded and/or kept with direct identifiers in the repository. The bank/repository may release identifiers with specimens to researchers under special circumstances with prior IRB approval.

Explain under what circumstances identifiers may be released:

18.3 If the repository/bank maintains the identifiers, list the identifiers that will be maintained with the specimens:

- [ ] Name
- [ ] Date of birth
- [ ] Social Security number
- [ ] Medical record number
- [ ] Address
- [ ] Phone number
- [ ] Email address
- [ ] Other dates (dates of surgery, visit dates)

18.4 Clinical follow-up data will be linked to specimens:

- ( ) Yes
- (•) No

If **Yes**, provide duration of follow-up or indefinitely:

18.5 There is a formal specimen utilization review process:

- ( ) Yes
- (•) No

If **Yes**, describe the process:

18.6 Specimens banked at UCSF may be made available to (check all that apply):

- [x] UCSF researchers
- [ ] Non-UCSF researchers
- [ ] Industry

19.1 Study drug or treatment is available off-study:

- ○ Yes
- ◉ No
- ○ Not applicable

19.2 Describe the usual care or activities at UCSF (or study site) that are available to prospective subjects who do not enroll in this study:

This study does not involve any treatments and participation in the study is voluntary.

19.3 Describe other alternatives to study participation that are available to prospective subjects:

This study do not involve any treatment and do not have any significant impact on subject's concurrent or future care, the alternative may be not to participate.

20.0 Risks and Benefits

20.1 Risks and discomforts:

A. Risks and Discomforts:

Risk and discomforts for biopsy procedures of adult oral and cervical tissue samples.The only potential risks are those associated with donating tissue biopsy material will include discomfort, bleeding, and rarely, infection. Sterile technique is used to avoid infection and bleeding is stopped using standard clinical procedures.

Risk and discomforts for collection blood samples.The only potential risks are those associated with donating blood samples will be discomfort, bleeding, and rarely, infection. To reduce potential risk blood samples will be collected for from donors who weigh at least 110 pounds, and the amounts drawn may not exceed 40 ml. No more than three skin punctures will to be made in any single attempt to draw blood.Sterile technique is used to avoid infection and bleeding is stopped using standard clinical procedures.
Risk and discomforts for collection saliva.Collection of saliva has no potential risk or discomfort.

Risk and discomforts for collecting of amniotic fluid samples.
The potential risks are those associated with the transabdominal amniocentesis procedure, including miscarriage/pregnancy loss, preterm premature rupture of the membranes and fetal injury, discomfort, bleeding, inflammation and, rarely, infection. Sterile technique is used to avoid infection and bleeding is stopped using standard clinical procedures. To reduce other risks amniocenteses will be performed under continuous ultrasound guidance. The amniocenteses will only be performed for clinical purposes. It is not expected that participation in the study will have any impact on the risks associated with the clinical procedure as no additional fluid will be removed than would have for clinical purposes.

Risk and discomforts for collection breast milk.Collection of breast milk has minimum potential risk or discomfort, the only potential risks are those associated with un-optimized electrical pump. High suction setting of pump may cause discomfort and to minimize discomfort the setting of pump will be optimized and examined for each pumping kit and individual donor.

20.2 Steps taken to minimize risks to subjects:

To minimize the discomfort of the oral tissue biopsy and transabdominal amniocentesis procedures,a local anesthetic is provided, followed by as-needed pain medications after the procedure. Sterile technique is used to avoid infection, and bleeding is stopped using standard clinical procedures.

20.3 Benefits to subjects:

○ Yes ◉ No

If yes, describe:

20.4 Benefits to society:

Knowledge of EBV, HPV and HIV infection via the adult and fetal oral cavity has important biological and public health implications regarding transmission of infection from one individual to another.

20.5 Explain why the risks to subjects are reasonable:

The study risks on an individual level will be minimal (potential loss of privacy, risks associated with blood draws). Also, the risk of harm from oral biopsies is very low and the importance of the knowledge to be gained is very high. There are no significant risks from donating 10cc of breastmilk and the amniotic fluid volunteers would be obtaining the amniocentesis for clinical purposes anyway.

21.0 Data and Safety Monitoring Plan

21.1 Describe the plan for monitoring data and safety:

21.2 This study requires a Data and Safety Monitoring Board:

○ Yes
◉ No or not sure

If **yes**, press **SAVE and CONTINUE** to move to the next section of the application.

21.3 If No, provide rationale:

○ Social/Behavioral research
○ Phase I trial
○ Treatment IND/Compassionate Use Trial
◉ Other (explain below)

If **Other,** explain:

This project is not interventional study.

22.0 Confidentiality and Privacy

22.1 Study data are:

☐ Derived from the Integrated Data Repository (IDR)
☑ Derived from a medical record (identify source below)
☐ Added to the hospital or clinical medical record
☑ Created or collected as part of health care
☐ Used to make health care decisions
☐ Obtained from the subject, including interviews, questionnaires

- [ ] Obtained from records open to the public
- [ ] Obtained from existing research records
- [ ] None of the above

If **derived from a medical record**, identify source:

[redacted]

22.2 Plans for accessing subject information while maintaining privacy:

Advertisements will be placed (in UCSF campus, local newspapers and on the Internet) seeking volunteers to donate blood samples, tongue and buccal mucosal tissue, amniotic fluid and breast milk from HIV-negative individuals. Interested individuals will be asked to contac[redacted] who will describe the study. If the individual remains interested, he or she will come to the clinic where the purpose of the study and the procedures will again be described. The individual will be enrolled after his/her questions are answered and he/she reads and signs the informed consent forms. After signing of consent the PI will obtain information about participants.

22.3 Identifiers may be included in research records:

(•) Yes ( ) No

If **yes**, check all the identifiers that may be included:

- [x] Names
- [x] Dates
- [x] Postal addresses
- [x] Phone numbers
- [ ] Fax numbers
- [ ] Email addresses
- [x] Social Security Numbers*
- [x] Medical record numbers
- [x] Health plan numbers
- [ ] Account numbers
- [ ] License or certificate numbers
- [ ] Vehicle ID numbers
- [ ] Device identifiers or serial numbers
- [ ] Web URLs
- [ ] IP address numbers
- [ ] Biometric identifiers
- [ ] Facial photos or other identifiable images
- [ ] Any other unique identifier

* Required for studies conducted at the VAMC

22.4 Plans for maintaining privacy in the research setting:

All specimens will be de-identified and designated by study numbers. The researchers and tissue bank staff will not have access to this information, rather they will have only study numbers. This numbers will be destroyed as soon as the study in completed. All data required by the protocol will be recorded and entered into electronic database using the unique study number. The study team will review the electronic data for accuracy. Data clarification or corrections will be made electronically and the databases will be password secured.

22.5 Possible consequences to subjects resulting from a loss of privacy:

We do not see potential risks to reputation, insurability or other social risks that would occur as a result of participation n this study that is above that received in usual medical care.

22.6 Identifiable information might be disclosed as part of study activities:

○ Yes ◉ No

If **yes**, indicate where identifiable information may be released to:

- ☐ The subject's medical record
- ☐ The study sponsor
- ☐ The US Food & Drug Administration (FDA)
- ☐ Others (Specify below)
- ☐ A Foreign Country or Countries

If **Others**, specify:

22.7 Indicate how data are kept secure (check all that apply):

- ☐ Data are stored securely in My Research
- ☑ Data are coded; data key is destroyed at end of study
- ☐ Data are coded; data key is kept separately and securely
- ☐ Data are kept in a locked file cabinet
- ☐ Data are kept in a locked office or suite
- ☑ Electronic data are protected with a password
- ☑ Data are stored on a secure network
- ☐ Data are collected/stored using REDCap or REDCap Survey

22.8 Additional measures to assure confidentiality:

22.9 This study may collect information that State or Federal law requires to be reported to other officials or ethically requires action:

○ Yes ◉ No

Explain:

22.10 This study will be issued a Certificate of Confidentiality:

○ Yes ◉ No

23.0 Subjects

23.1 Check all types of subjects that may be enrolled:

- ☐ Inpatients
- ☐ Outpatients
- ☑ Healthy volunteers
- ☑ Staff of UCSF or affiliated institutions

23.2 Additional vulnerable populations:

- [ ] Children
- [ ] Subjects unable to consent for themselves
- [ ] Subjects unable to consent for themselves (emergency setting)
- [ ] Subjects with diminished capacity to consent
- [x] Subjects unable to read, speak or understand English
- [x] Pregnant women
- [ ] Fetuses
- [ ] Neonates
- [ ] Prisoners
- [ ] Economically or educationally disadvantaged persons
- [ ] Investigators' staff
- [ ] Students

Explain why it is appropriate to include the types of subjects checked above in this particular study:

Describe the additional safeguards that have been included in the study to protect the rights and welfare of these subjects and minimize coercion or undue influence:

24.0 Inclusion of Non-English Speaking Subjects

24.1 Indicate which method(s) you will use to consent non-English speaking subjects:

- [x] Preferred Method—Consent form and other study documents will be available in the subject's primary language Personnel able to discuss participation in the patient's language will be present for the consent process.
- [x] Short-Form—A qualified interpreter will translate the consent form verbally, and subjects will be given the Experimental Subject's Bill of Rights in their primary language, following instructions in Those Who do not Read, Speak or Understand English for required witnessing and signatures

24.2 Explain how you will maintain the ability to communicate with non-English speakers throughout their participation in the study:

Will have trained bilingual staff available for translation.

25.0 Inclusion of Pregnant Women, Fetuses, and/or Neonates

25.1 Review the regulatory categories and identify all sections of 45 CFR 46 Subpart B under which you believe the research falls and provide study-specific information showing why the research falls within those sections:

Category 46.204:
(i) For this study will be collected the amniotic fluid samples from HIV-positive and-negative pregnant women. These amniotic fluid samples will be collected after separation of fetal cells for karyotyping analysis, thus these samples will be otherwise discarded. HIV-infected women do not, routinely, obtain amniocenteses. Nonetheless, some of the HIV-infected pregnant patients do undergo amniocentesis in the setting of abnormal first or second-trimester genetic screening tests or advanced maternal age.
(ii) For this study, will be collected the tissue samples from aborted fetuses that are also discarded materials.

Since, above procedures do not have any interventions and do not cause risk to pregnant women

26.0 Recruitment

26.1 * Methods (check all that apply):

- [x] Study investigators (and/or affiliated nurses or staff) recruit their own patients directly in person or by phone.
- [ ] Study investigators recruit their own patients by letter. Attach the letter for review.
- [ ] Study investigators send a "Dear Doctor" letter to colleagues asking for referrals of eligible patients. If interested, the patient will contact the PI or the PI may directly recruit the patients (with documented permission from the patient). Investigators may give the referring physicians a study information sheet for the patients.
- [ ] Study investigators provide their colleagues with a "Dear Patient" letter describing the study. This letter can be signed by the treating physicians and would inform the patients how to contact the study investigators. The study investigators may not have access to patient names and addresses for mailing
- [x] Advertisements, notices, and/or media used to recruit subjects. Interested subjects initiate contact with study investigators. Attach ads, notices, or media text for review. In section below, please explain where ads will be posted.
- [ ] Study investigators identify prospective subjects through chart review. (Study investigators request a Waiver of Authorization for recruitment purposes.)
- [ ] Large-scale epidemiological studies and/or population-based studies: Prospective subjects are identified through a registry or medical records and contacted by someone other than their personal physician. (Study investigators request a Waiver of Authorization for recruitment purposes.)
- [ ] Direct contact of potential subjects who have previously given consent to be contacted for participation in research. Clinic or program develops a CHR-approved recruitment protocol that asks patients if they agree to be contacted for research (a recruitment database) or consent for future contact was documented using the consent form for another CHR-approved study.
- [ ] Study investigators list the study on the School of Medicine list of UCSF Clinical Trials website or a similarly managed site. Interested subjects initiate contact with investigators.
- [ ] Study investigators recruit potential subjects who are unknown to them through methods such as snowball sampling, direct approach, use of social networks, and random digit dialing.
- [ ] Other

If **Other**, explain:

26.2 How, when, and by whom eligibility will be determined:

Eligibility for the oral and cervical biopsy isolation for this research will be determined by [redacted] respectively. They will examine the volunteers for oral and cervical infection, allergy and inflammation before the biopsy procedure. The HIV-negative volunteers will be tested for HIV to verify their status.

26.3 How, when, where and by whom potential subjects will be approached:

Advertisements will be placed seeking volunteers to donate blood samples, oral and cervical mucosal tissue, amniotic fluid and breast milk. The advertisements will be posted around the UCSF and San Francisco General campuses, in local newspapers and on the Internet. All volunteers will be financially compensated for their time. Interested individuals will be asked to contact [redacted] who will describe the study. If the individual remains interested, he or she will come to the clinic where the purpose of the study and the procedures will again be described. The individual will be enrolled after his/her questions are answered and he/she reads and signs the informed consent forms.

1. The blood samples from HIV/AIDS patients and normal healthy volunteers will be collected in [redacted]

2.The biopsy tissues from the adult oral and cervical mucosa will be collected in [redacted]

3. The fetal tongue, tonsil, gastric and intestinal tissue samples from discarded fetus will be collected in [redacted]

4. The and amniotic fluid samples from normal healthy individuals and HIV/AIDS patients will be collected in [redacted]

5. The breast milk samples will be collected in breastfeeding healthy HIV-negative women at [redacted]

26.4 * Protected health information (PHI) will be accessed prior to obtaining consent:

○ Yes ◉ No

27.0 Informed Consent

27.1 * Methods (check all that apply):

- ☑ Signed consent will be obtained from subjects and/or parents (if subjects are minors)
- ☐ Verbal consent will be obtained from subjects using an information sheet or script
- ☐ Electronic consent will be obtained from subjects via the web or email
- ☐ Implied consent will be obtained via mail, the web or email
- ☐ Signed consent will be obtained from surrogates
- ☐ Emergency waiver of consent is being requested for subjects unable to provide consent
- ☐ Informed consent will not be obtained

27.2 Process for obtaining informed consent:

After advertisements any responded subject will be invited to office of [redacted] for blood samples study, to office of [redacted] for amniotic fluid study and [redacted] for oral tissues and [redacted] for cervical tissue samples study.

[redacted] will meet with each of interested subject in their office and describe them he purpose of the study and the procedures and answered their questions. If the individual remains interested he/she reads and signs the informed consent forms.

The fetal samples from discarded aborted materials will be collected at [redacted] Since, this study will use only discarded fetal materials, the mothers consent to the use of the fetal tissue in research will not be obtained by the researchers in this study. The fetal samples will be used for the research and will be banked in [redacted]

27.3 How investigators will make sure subjects understand the information provided to them:

Persons obtaining consent will ask potential participants to restate what they have understood at various steps along the informed consent procedure.

28.0 Financial Considerations

28.1 Subjects payment or compensation method (check all that apply):

Payments will be (check all that apply):

- [ ] Subjects will not be paid
- [ ] Cash
- [x] Check
- [ ] Gift card
- [ ] Other:

Specify **Other**:

28.2 Describe the schedule and amounts of payments, including the total subjects can receive for completing the study. If deviating from recommendations in Subject Payment Guidelines, include specific justification below.

The amount of payment for singly donation will be as follow:
$20 for 40 ml blood
$150 for one tongue tissue
$100 buccal tissue
$100 cervical tissue

28.3 Costs to Subjects: Will subjects or their insurance be charged for any study procedures?

- ○ Yes
- ◉ No

If **yes,** describe those costs below, and compare subjects' costs to the costs associated with alternative care off-study. Finally, explain why it is appropriate to charge those costs to the subjects.

29.0 CTSI Screening Questions

29.1 * This study will be carried out at one of the UCSF Clinical Research Centers (CRCs) or will utilize CRC services:

- ○ Yes
- ◉ No

29.2 This project involves community-based research:

- ○ Yes
- ○ No

29.3 This project involves practice-based research:

- ○ Yes
- ○ No

29.4 Please check other CTSI services below that you plan to utilize to conduct your research:

Guidance and Services:

- [ ] Biostatistics
- [ ] Study Design and Implementation
- [ ] Data Management
- [ ] Ethics
- [ ] Health Policy

- [ ] Bioinformatics Data Analysis
- [ ] Regulatory Knowledge
- [ ] THREDS The Health Record Data Service
- [ ] Community-Engaged Research
- [ ] Collaborating with Kaiser Researchers

Clinical Research Centers:

- [ ] Community Engagement (CE)

Funds to Innovate:

- [ ] Strategic Opportunity Support (SOS)

Training:

- [ ] Clinical & Translational Sciences Training (CTST)
- [ ] Career Advancement (CA)

CTSI Core Services:

- [ ] Animal/Preclinical
- [ ] Array
- [ ] Bioinformatics
- [ ] Biostatistics
- [ ] Cell Culture
- [ ] Clinical Services
- [ ] Epidemiology
- [ ] Flow Cytometry
- [ ] Human/Clinical
- [ ] Imaging
- [ ] Immunohistochemistry
- [ ] Islet Production
- [ ] Microscopy
- [ ] Molecular/Genomic
- [ ] Monoclonal Antibody
- [ ] Proteomics
- [ ] Resale Products
- [ ] Tissue

Study Application (Version 1.0)

1.0 General Information

***Enter the full title of your study:**

HIV, EBV and HPV interaction with mucosal epithelium

***Enter the study alias:**

H8597-30664-04
* This field allows you to enter an abbreviated version of the Study Title to quickly identify this study.

2.0 Add departments

2.1 and Specify Research Location:

Is Primary?	Department Name
◉	**UCSF -** [redacted]

3.0 List the key study personnel: (Note: external and affiliated collaborators who are not in the UCSF directory can be identified later in the Qualifications of Key Study Personnel section at the end of the form)

3.1 *Please add a Principal Investigator for the study:

[redacted]

Select if applicable

☐ Department Chair ☐ Resident

☐ Fellow

If the Principal Investigator is a Fellow, the name of the Faculty Advisor must be supplied below.

3.2 If applicable, please select the Research Staff personnel

A) Additional Investigators

[redacted]

[redacted]

B) Research Support Staff

[redacted]

3.3 *Please add a Study Contact

[redacted]

The Study Contact(s) will receive all important system notifications along with the Principal Investigator. (e.g. The project contact(s) are typically either the Study Coordinator or the Principal Investigator themselves).

3.4 If applicable, please add a Faculty Advisor/Mentor:

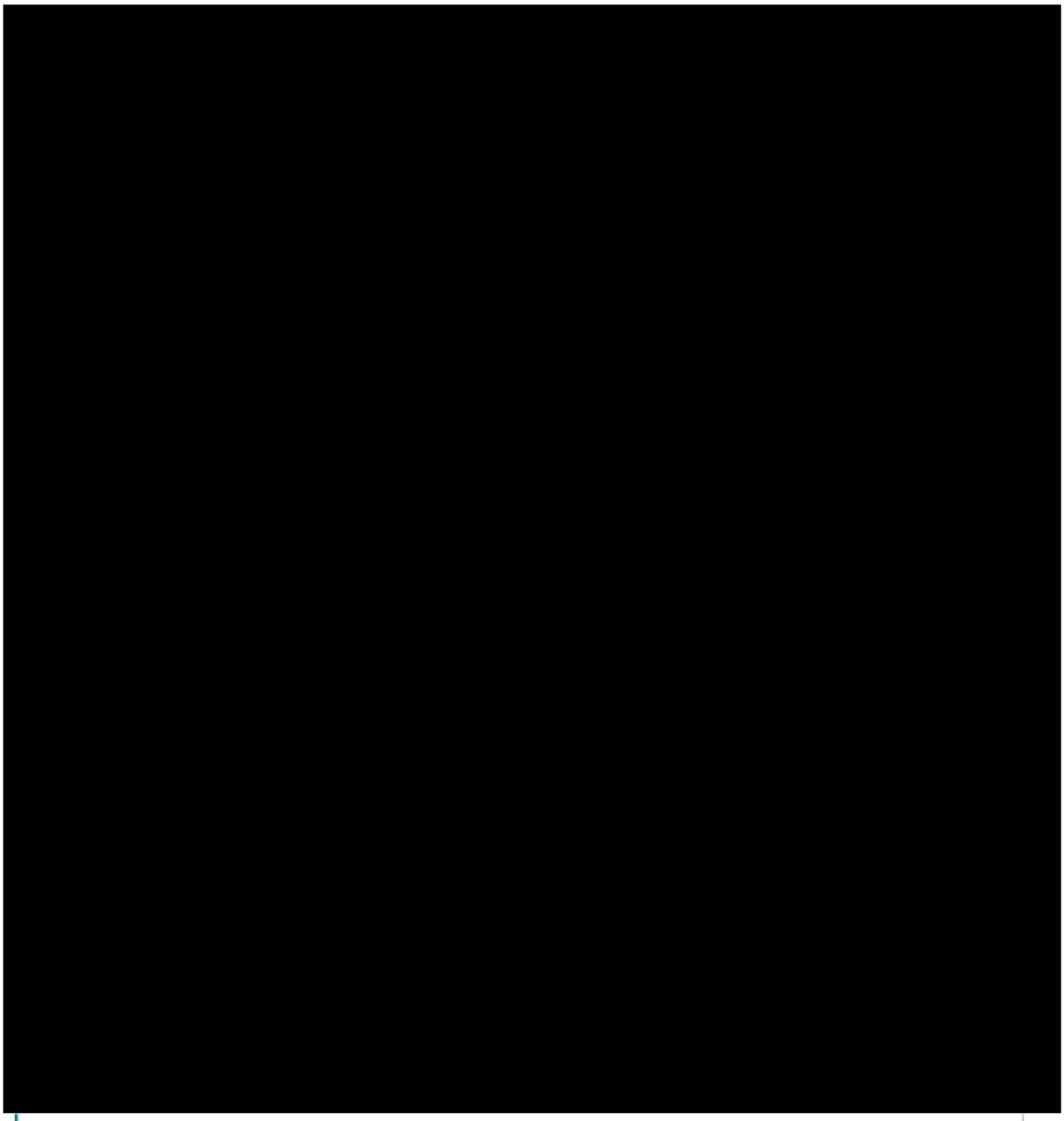
3.5 If applicable, please select the Designated Department Approval(s)

Add the name of the individual authorized to approve and sign off on this protocol from your Department (e.g. the Department Chair or Dean).

4.0 Qualifications of Key Study Personnel

4.1 List the study responsibilities and qualifications of any individuals who qualify as Key Study Personnel (KSP) by clicking the "Add a new row" button:

KSP Name	Description of Study Responsibilities	Qualifications
[redacted]		



5.0 Initial Screening Questions

5.1 * This study involves human stem cells, gametes or embryos:

- (•) No
- ( ) Yes, and requires CHR and GESCR review
- ( ) Yes, and requires GESCR review, but NOT CHR review

5.2 * This application involves a Humanitarian Use Device:

- (•) No
- ( ) Yes, and it includes a research component
- ( ) Yes, and it involves clinical care ONLY

5.3 * This is a CIRB study (e.g. the NCI CIRB will be the IRB of record):

- ( ) Yes (•) No

5.4 * This application includes a request to rely on another UC IRB to be the IRB of record:

○ Yes ◉ No

Note: If this request is approved, the CHR will **NOT** review and approve this study. Another UC campus will be the IRB of record.

6.0 Application Type

6.1 * This research involves:

◉ Minimal risk
○ Greater than minimal risk

6.2 * This application is:

◉ Full Committee
○ Expedited
○ Exempt

6.3 If you think this study qualifies for expedited review, select the regulatory category(ies) that the research falls under:

☐ Category 1: A very limited number of studies of approved drugs and devices
☐ Category 2: Blood sampling
☐ Category 3: Noninvasive specimen collection
☐ Category 4: Noninvasive clinical procedures
☐ Category 5: Research involving materials that were previously collected for either nonresearch or research purposes
☐ Category 6: Use of recordings
☐ Category 7: Low risk behavioral research
☐ Category 8: Renewal of inactive research protocols or protocols that are essentially complete
☑ Category 9: Renewal of other minimal risk research protocols

6.4 * This study involves:

◉ Subject contact (including phone, email or web contact)
○ No subject contact (limited to medical records review, biological specimen analysis, and/or data analysis)

7.0 Funding

7.1 Identify all the funding sources and their roles on the project:

View Details	Sponsor Name	Sponsor Type	Awardee Institution:	Contract Type:	Project Number	UCSF RAS System Award Number ("A" + 6 digits)
⊟	California HIV/AIDS Research Program	14	UCSF	Grant		A115176

Sponsor Name:	California HIV/AIDS Research Program
Sponsor Type:	14
Sponsor Role:	
Awardee Institution::	UCSF
Is Institution the Primary Grant Holder:	Yes
Contract Type:	Grant
Project Number:	
UCSF RAS System Award Number ("A" + 6 digits):	A115176
Grant Number for Studies Not Funded thru UCSF:	
Grant Title:	HIV tat- and gp120-facilitated HPV epithelial entry
PI Name: (If PI is not the same as identified on the study.)	
Explain Any Significant Discrepancy:	

⊟	NIH Natl Inst Dental & Craniofacial Res.	01	UCSF	Grant		A115176

Sponsor Name:	NIH Natl Inst Dental & Craniofacial Res.
Sponsor Type:	01
Sponsor Role:	Funding
CFDA Number:	
Grant/Contract Number:	
Awardee Institution::	UCSF
Is Institution the Primary Grant Holder:	Yes
Contract Type:	Grant
Project Number:	
UCSF RAS System Award Number ("A" + 6 digits):	A115176
Grant Number for Studies Not Funded thru UCSF:	
Grant Title:	HIV TRANSCELLULAR AND TRANSSYNAPTIC PENETRATION OF MUCOSAL EPITHELIUM
PI Name: (If PI is not the same as identified on the study.)	
Explain Any Significant Discrepancy:	

7.2 If you tried to add the sponsor in the question above and it was not in the list, check here:

○ Sponsor not in list

Only if your sponsor is not yet in the list, type the sponsor's name:

If the sponsor is not in the system, download the C&G Add Sponsor Form from Help link and attach it to this application. **Your study will not receive CHR approval until the sponsor and funding details have been added to your application.**

7.3 For Federally funded studies only, indicate which portion of your grant you will be attaching:

- [x] The Research Plan, including the Human Subjects Section of your NIH grant
- [ ] For other federal proposals (contracts or grants), the section of the proposal describing human subjects work
- [ ] The section of your progress report if it provides the most current information about your human subjects work

7.4 If this study has no sponsor, check all that apply:

- [ ] Unfunded student project
- [ ] Unfunded (miscellaneous departmental funding)
- [ ] Specific departmental funding

8.0 Statement of Financial Interest

8.1 * The Principal Investigator and/or one or more of the key study personnel has financial interests related to this study:

- ( ) Yes
- (•) No

If **Yes**, attach the **Disclosure of Investigators' Financial Interests Supplement** to this application.

9.0 Sites

9.1 Institutions (check all that apply):

- [x] UCSF
- [ ] Mt. Zion
- [ ] San Francisco General Hospital (SFGH)
- [ ] SF VA Medical Center (SF VAMC)
- [ ] Helen Diller Family Comprehensive Cancer Center
- [ ] Fresno (Community Medical Center)
- [ ] Blood Centers of the Pacific (BCP)
- [ ] Blood Systems Research Institute (BSRI)
- [ ] Gallo
- [ ] Gladstone
- [ ] Institute on Aging (IOA)
- [ ] SF Dept of Public Health (DPH)

9.2 Check all the other types of sites not affiliated with UCSF with which you are cooperating or collaborating on this project:

- [ ] Foreign Country

List:

- [ ] Other UC Campus
- [ ] Other institution
- [ ] Other community-based site

9.3 * This is a multicenter study:

- ( ) Yes (•) No

9.4 Check any research programs this study is associated with:

- [ ] Cancer Center
- [ ] Center for AIDS Prevention Sciences (CAPS)
- [ ] Global Health Sciences
- [ ] Immune Tolerance Network (ITN)
- [ ] Osher Center
- [ ] Positive Health Program

10.0 Study Design

10.1 Study design:

HIV mucosal transmission is a potentially important, poorly understood route of HIV infection that has global consequences. The oropharyngeal mucosal epithelium of the fetus/neonate and adult genital mucosa may serve as an efficient portal of entry for HIV, yet oral transmission of HIV among adults is rare (2, 10, 14, 15). The reasons for this difference are not well understood. The goal of our study is to investigate molecular mechanisms of resistance of adult oral epithelial cells and the susceptibility of fetal oral and adult genital epithelial cells for HIV infection. We hypothesize that resistance of adult oral epithelial cells for HIV infection is due to the expression of multiple anti-HIV innate molecules, including calprotectin, defensins, lactoferrin, secretory leukocyte protease inhibitor, thrombospondin, polyanionic albumins, acidic proline-rich proteins, and salivary agglutinins (12). These proteins may not be expressed or may not have anti-HIV function in fetal/neonatal oral and adult genital epithelial cells. These molecules are present in amniotic fluid and breast milk, however, their role in HIV infection with fetal/neonatal mucosal epithelium in not clear. Therefore we will investigate role of fetal mucosal epithelium in HIV transmission, as well as role of amniotic fluid and breast milk in HIV transmission via fetal oral mucosal epithelium. We will compare expression of innate proteins in the fetal and infant oral epithelia. We also will study HIV transmission via cervical mucosal epithelium, and expression of innate proteins in cervical epithelial cells.

It is well known that during the systemic HIV infection the innate immune and barrier functions of mucosal epithelium may be impaired (8, 13). This could be due to reactivation and dissemination of common oral pathogens such as EBV and HPV within the oral epithelium, which may lead to disruption of oral epithelium and lead to HIV transmission (1, 3-5, 7, 9). Therefore, our research will focus to investigate the molecular mechanisms of HIV, EBV and HPV interactions with mucosal epithelium.

10.2 Check all that apply:

- [ ] Phase I
- [ ] Phase II
- [ ] Phase III
- [ ] Phase IV

11.1 Hypothesis:

This study has a hypothesis:

(•) Yes ( ) No

If yes, state the hypothesis or hypotheses:

Hypothesis: A tissue explants of adult oral and cervical, and fetal andoropharyngeal, gastric and intestinal mucosal epithelia ex vivo will retain its natural tissue architecture, including the epithelium, lamina propria, and intraepithelial and submucosal immune cells. This model will be suitable to evaluate HIV-1 transmission from mother to fetus or neonate via mucosal epithelium.
- Hypothesis: Expression of HBD1, HBD2, HBD3 and SLPI in adult oral epithelium may be substantially higher than in infant and fetal oral epithelia, and this may determine transepithelial transmission of HIV via adult and infant/fetal epithelia.
- Hypothesis: HIV spread across the mucosal epithelium may occur by two parallel mechanisms: (i) cells may express one or more of the HIV co-receptors CXCR4, CCR5 and GalCer. Cell-free X4- or R5-tropic HIV from amniotic fluid, cervicovaginal secretions or breast milk may bind to appropriate HIV co-receptors and GalCer on the surface of the epithelium, initiating entry into and infection of epithelial cells by X4- and/or R5-tropic HIV strains; (ii) cell-associated X4- or R5-tropic HIV from amniotic fluid, cervicovaginal secretions or breast milk may infect the epithelium by mechanisms of cell-cell spread; and (iii) the interaction of cell-free or cell-associated HIV with the apical surface of the fetal/neonatal epithelium may initiate transcytosis of virions from apical to basolateral membranes of mucosal epithelial cells. These mechanisms may lead to dissemination of HIV infection into immune cells in the lamina propria, and therefore may cause systemic HIV/AIDS disease in the fetus and neonate. HIV resistance in adult oral epithelium could be due to absence of HIV receptors or high level of anti-HIV proteins expression.
Hypothesis: Amniotic fluid and breast milk contain C-C (MIP-1a, MIP-1band RANTES) and C-X-C (SDF-1) chemokines, and anti-viral innate moleculeS (calprotectin, defensins), which may modulate HIV infection of the oral mucosal epithelium of the fetus/neonate: (i) C-C and C-X-C chemokines in amniotic fluid and breast milk may suppress cell-free HIV infection or transcytosis via mucosal epithelial cells; (ii) however, these chemokines may not affect GalCer-mediated HIV entry or transcytosis of HIV and cell-to-cell spread of virus from HIV-infected maternal cells into the mucosal epithelium of the fetus/neonate; (iii) HIV-positive pregnant woman may have a higher rate of upper and lower genital tract and breast infection/inflammation, which may lead to activation in the genital tract and in amniotic fluid and breast milk of pro-inflammatory cytokines, including tumor necrosis factor (TNF)-a, interferon (IFN)-g, interleukin (IL)-6 and IL-8. Increased levels of such cytokines may disrupt cellular junctions of the mucosal epithelium by apoptotic and /or non-apoptotic mechanisms and thereby impair their barrier function. This impairment leads to penetration of cell-free or cell-associated HIV via the paracellular space.
- Hypothesis: During HIV/AIDS disease, HIV tat protein is secreted into the circulation and binds to the b1 family integrinsof peripheral blood monocytes via tat's RGD motif. This binding activates integrin signaling and their clustering within the lipid rafts of monocyte membranes, facilitating EBV infection by interaction of EBV BMRF-2 with b1 integrins. HIV and/or EBV infected monocytes may migrate into adult oral mucosal epithelium and reduce its innate immune and barrier functions.
- Hypothesis: HIV-infected immune cells migrate into oral epithelium and secrete TNF-a, which in turns lead to disruption of epithelial cell junctions. HIV/TNF-a-mediated disassembly of cell junctions facilitates entry of HPV into the basal layers of epithelium.

11.2 List the specific aims:

Aim 1. Establish and characterize an ex vivo organ culture model from the oral and cervical of the adult, and oropharyngeal, gastric and intestinal mucosal epithelium of the fetus.
Aim 2. Analyze expression of HBD1, HBD2, HBD3 and SLPI in adult, infant and fetal oral epithelia.
Aim 3. Study mechanisms of HIV spread via the oral and cervical of the adult, and oropharyngeal, gastric and intestinal mucosal epithelium of the fetus.
Aim 4. Determine the role of amniotic fluid and breast milk in the interaction of HIV with the mucosal epithelium of the fetus.
Aim 5. Study the mechanisms of HIV-EBV co-infection of circulating monocytes and its role in dissemination of HIV and EBV infection within the oral mucosal epithelium.

Aim 6. Determine the role of HIV-mediated disruption of mucosal epithelial tight junctions in paracellular spread of HPV.

11.3 Statistical analysis:

Data will be analyzed by a study steering committee, which will be composed by [redacted] It is estimated that 60 adult, 100 fetal and 20 infant tissues will investigated for this study. The 150 blood and saliva samples from HIV-positive with HAART, 150 from HIV-positive w/o HAART and 150 from HIV-negative will be examined for HIV and EBV infection. Fifteen amniotic fluid samples from HIV-negative and 4 from HIV-positive pregnant women will be investigated. Ten breast milk from HIV-negative woman will be analyzed. A three years cumulative enrollment is planned.
HIV-1 infection of the fetal mucosal epithelium will be evaluated in the presence or absence of breast milk or amniotic fluid by detection of HIV-1 signals. At least five tissue explants will be used for each breast milk sample and untreated control. As fetal epithelial tissues will be treated and untreated with breast milk, the statistical analysis will account for the paired nature of the data, i.e., HIV infection or transcytosis in tissue explants treated with breast milk compared with untreated controls. Quantitative data for statistical analysis of the number of cells containing HIV infection will be obtained by screening 30–50 fields per tissue section. We will test the null (no difference) hypothesis against the alternative hypothesis that there is a difference. With five tissue samples, using a paired *t* test with a 5% false-positive rate, we will have 80% power to detect a difference between treated and untreated samples.

11.4 * This is an investigator-initiated study:

(•) Yes ( ) No

11.5 This study has received scientific or scholarly review from (check all that apply):

- [ ] Cancer Center Protocol Review Committee (PRC) (Full approval or contingent PRC approval is required prior to final CHR approval for cancer-related protocols.)
- [ ] CTSI Clinical Research Center (CRC) advisory committee
- [ ] Departmental scientific review
- [x] Other:

Specify **Other**:

Committee on human research

If applicable, attach the **Departmental Scientific Review Form** at the end of the application.

12.0 Background

12.1 Background:

It is well documented that HIV oral transmission in adult population is rare, and oral mucosal epithelium is resistant for HIV infection (4, 6, 8). However, the oropharyngeal mucosal epithelium of the fetus/neonate may serve as an efficient portal of entry for perinatal and postnatal mother-to-child transmission (MTCT) of HIV (6). It was also well documented that HIV transmission via genital epithelium is substantially higher than the oral epithelium. MTCT of HIV may occur in utero before birth (prenatal), during labor and delivery (perinatal) or after birth (postnatal) (6). In prenatal MTCT, RNA/DNA and proteins were detected in placental trophoblasts, endothelial cells, and villous Hofbauer cells from 8 weeks of gestation, suggesting HIV transmission by the transplacental route (6). An alternative prenatal route could be transamniotic, where cell-free HIV virus and/or HIV-infected cells may penetrate into the amniotic sac and infect the fetus via the oropharyngeal cavity or gastrointestinal (GI) tract. A case report showed that HIV was detected in the amniotic fluid of HIV-positive women at 32 weeks gestation (6). Another showed that HIV-1 was detected in gastric aspirates of a 15-week-old fetus (6). Enzyme-linked immunosorbent assay (ELISA) analysis of HIV-1 p24 in samples of amniotic fluid from 10 HIV-positive women showed that 8 women were positive for p24 (6). Maiques et al.

amniotic fluid before HAART and in 3% of the fluid under HAART (6). Mohlala et al. showed that HIV-1 was not detected in the amniotic fluid of 23 HIV-positive women under HAART who had a normal pregnancy, and these data indicate that HAART treatment substantially reduces MTCT of HIV in utero (11). The solid evidences indicate that postnatal MTCT of HIV via breastfeeding, and HAART treatment does not efficiently inhibit postnatal MTCT (6). All above evidence clearly indicate that fetal/neonatal mucosal epithelium play critical role in HIV MTCT. The higher rate of genital HIV transmission also indicate that genital epithelium may play critical role in HIV spread. Therefore our research focuses on HIV interaction with adult oral and genital, and fetal oral and GI mucosal epithelium. An adult oral mucosal epithelium is resistant for HIV infection. However, during the HIV/AIDS disease multiple opportunistic infections, including EBV and HPV may reactivate and disseminate within the oral epithelium and reduce its protective function for HIV infection. Therefore we will study mechanisms of HIV, EBV and HPV dissemination within the adult oral mucosal epithelium.

12.2 Preliminary studies:

12.3 References:

1. **Boulter, A. W., N. Soltanpoor, A. V. Swan, W. Birnbaum, N. W. Johnson, and C. G. Teo.** 1996. Risk factors associated with Epstein-Barr virus replication in oral epithelial cells of HIV-infected individuals. Aids **10:**935-40.
2. **Casper, C., and E. M. Fenyo.** 2001. Mother-to-child transmission of HIV-1: the role of HIV-1 variability and the placental barrier. Acta Microbiol Immunol Hung **48:**545-73.
3. **Chou, L. L., F. Boustany, and D. Nathanson.** 1996. G to A hypermutation in env loop V2 region of HIV-1 in oral hairy leukoplakia. J Dental Res **75:**115.
4. **Greenspan, D., and J. S. Greenspan.** 1997. Oral manifestations of HIV infection. AIDS Clin Care **9:**29-33.
5. **Greenspan, D., J. S. Greenspan, N. G. Hearst, L. Z. Pan, M. A. Conant, D. I. Abrams, H. Hollander, and J. A. Levy.** 1987. Relation of oral hairy leukoplakia to infection with the human immunodeficiency virus and the risk of developing AIDS. J Infect Dis **155:**475-81.
6. **Hille, J. J., J. Webster-Cyriaque, J. M. Palefski, and N. Raab-Traub.** 2002. Mechanisms of expression of HHV8, EBV and HPV in selected HIV-associated oral lesions. Oral Dis **8 Suppl 2:**161-8.
7. **Lau, R., J. Middeldorp, and P. J. Farrell.** 1993. Epstein-Barr virus gene expression in oral hairy leukoplakia. Virology **195:**463-74.
8. **Leigh, J. E., K. Shetty, and P. L. Fidel, Jr.** 2004. Oral opportunistic infections in HIV-positive individuals: review and role of mucosal immunity. AIDS Patient Care STDS **18:**443-56.
9. **Lucht, E., P. Biberfeld, and A. Linde.** 1995. Epstein-Barr virus (EBV) DNA in saliva and EBV serology of HIV-1-infected persons with and without hairy leukoplakia. J Infect **31:**189-94.
10. **Minkoff, H.** 2003. Human immunodeficiency virus infection in pregnancy. Obstet Gynecol **101:**797-810.
11. **Mohlala, B. K., T. J. Tucker, M. J. Besser, C. Williamson, J. Yeats, L. Smit, J. Anthony, and A. Puren.** 2005. Investigation of HIV in amniotic fluid from HIV-infected pregnant women at full term. J Infect Dis **192:**488-91.
12. **Moutsopoulos, N. M., T. Greenwell-Wild, and S. M. Wahl.** 2006. Differential mucosal susceptibility in HIV-1 transmission and infection. Adv Dent Res **19:**52-6.
13. **Patton, L. L., and C. van der Horst.** 1999. Oral infections and other manifestations of HIV disease. Infect Dis Clin North Am **13:**879-900.
14. **UNAIDS.** 2004. UNAIDS, AIDS epidemic update. Geneva:UNAIDS/WHO.
15. **UNAIDS.** 2006. UNAIDS, AIDS epidemic update. Geneva:UNAIDS/WHO.

If you have a separate bibliography, attach it to the submission with your other study documents.

13.0 Sample Size and Eligibility

13.1 Number of subjects that will be enrolled at UCSF and affiliated institutions:

555

13.2 Total number of subjects that will be enrolled at all sites:

13.3 Estimated number of people that you will need to consent and screen here (but not necessarily enroll) to get the needed subjects:

600

13.4 Sample size calculation:

13.5 * Eligible age range(s):

- [ ] 0-6 years
- [ ] 7-12 years
- [ ] 13-17 years
- [x] 18+ years

13.6 Inclusion criteria:

The only inclusion criterion will be willingness to donate blood, oral and cervical tissue, amniotic fluid and breast milk samples and have a known HIV-status, which were documented negative HIV test in the past 3 months.

13.7 Exclusion criteria:

The only exclusion criteria for isolation of oral and cervical explants, and amniotic fluid and breast milk from adult individuals would be inflammation or bacterial infection, since these factor may increase of risk for subsequent bleeding. Also, the exclusion criteria for isolation of tongue and buccal explants would be if volunteers have allergy to anesthetic.

13.8 There are inclusion or exclusion criteria based on gender, race or ethnicity:

○ Yes ◉ No

If **yes**, please explain the nature and rationale for the restrictions:

14.0 Drugs and Devices

14.1 * Drugs or biologics will be studied under this application:

○ Yes ◉ No

14.2 * Medical devices will be studied under this application:

○ Yes ◉ No

14.3 Verification of IND/IDE numbers: If the sponsor's protocol does not list the IND/IDE number, you must submit documentation from the sponsor or FDA identifying the IND/IDE number for this study. Attach this documentation in the Other Study Documents section of the Initial Review Submission Packet.

15.0 Other Approvals and Registrations

15.1 This is a clinical trial:

○ Yes ◉ No

Clinical Trial Registration
"NCT" number for this trial:

15.2 * This study involves human gene transfer or recombinant DNA research:

○ Yes ◉ No

15.3 This study involves other regulated materials and requires approval and/or authorization from the following regulatory committees:

☐ Institutional Biological Safety Committee (IBC)

Specify BUA #:

8597-BU-01-INC

☐ Institutional Animal Care and Use Committee (IACUC)

Specify IACUC #:

☐ Radiation Safety Committee

Specify RUA #:

☐ Radioactive Drug Research Committee (RDRC)

Specify RDRC #:

☐ Controlled Substances

16.0 Procedures

16.1 List all study procedures, test and treatments required for this study:

1. Collection of adult oral and cervical mucosal tissue and establishment of tissue explants ex vivo. The tongue and buccal explants (30 for each, total 60) will be obtained from HIV-positive and –negative adult volunteers. Cervical tissue also will be collected from HIV-positive and –negative premenopausal women.
First, donor from oral biopsies will be treated with local anesthetic agent lidocaine (4.5 mg/kg) for 5-10 min. After confirmation of numbness the biopsy procedure will be performed. Biopsies of the oral and cervical mucosa containing epithelium and connective tissue will be obtained using 4-mm–diameter biopsy punches in the [redacted]

Immediately after biopsy the tissue will be placed into a tube with 2 ml of RPMI medium (explant medium) containing 10% heat-inactivated fetal bovine serum, 20 mM HEPES, 100 mM glutamine, 20 µg/ml gentamicin, 100 U/ml penicillin, 100 µg/ml streptomycin, and 50 ng/ml amphotericin-B. Before use the explants will be washed three times with 0.5 ml cold explant medium. Also will be taken blood 20 ml of blood from each individual who will donate tongue and buccal explants.

2. Collection of fetal tongue, tonsil, gastric and intestinal tissue explants
Fetal tongue, tonsil, gastric and intestinal tissue explants (total 100) will be collected from discarded fetal tissues obtained following an elective termination of pregnancy at [redacted] We will not have access the any identifying documents associated with fetal materials. The tongue, tonsil, gastric and intestinal tissue explants containing stratified mucosal epithelium and submucosal stromal tissue will be obtained from discarded fetuses at 18 to 22 weeks of gestation, because, in most cases, abortion later than 23 weeks of gestation is generally not be permitted or performed. The 5-mm pieces of tissue will be dissected from the dorsal surface of the fetal tongue, cheek and tonsil by surgical instrument. Similarly sized pieces will be dissected from the gastric mucosa and small and large intestine. All mucosal samples will include the epithelium and lamina propria with connective tissues. Immediately after biopsy isolation, the tissues will be placed into a tube with 2 ml of RPMI medium (explant medium) containing 10% heat-inactivated fetal bovine serum (FBS), 20 mM HEPES, 100 mM glutamine, 20 µg/ml gentamicin, 100 U/ml penicillin, 100 µg/ml streptomycin and 50 ng/ml amphotericin-B. Before use, the explants will be washed three times with 0.5 ml cold explant medium. To establish a polarized organ culture system, the explants, with the epithelial layer oriented on top, will be placed in the top chamber on a Transwell-clear filter insert with a pore size of 0.45 µm and a diameter of 6.5 mm (#3472, Costar Corp., Cambridge, Mass). Filter inserts with explant will be incubated at 37 C in a humidified atmosphere containing 5% CO_2, and one third of the medium from each chamber will be changed every other day.
3. Collection of infant buccal and tonsil tissue explants from cadavers. The buccal and tonsil mucosa in the area of buccal and tonsil, respectively will be lifted with forceps. A sharp scalpel will be used to excise a small piece of (about 5 mm) tissues. The tongue and tonsil tissue explants containing stratified mucosal epithelium will be obtained from infants (cadavers) under 1 year old. Immediately after biopsy isolation, the tissues will be fixed with 3% paraformaldehyde and dehydrate with sucrose gradient, will be used confocal microscopy immunodetection analysis. We will not have access the any identifying documents associated with infant materials.

4. Infection of tissue explants with HIV and/or EBV.
Tissue explants mounted in filter inserts will be infected from their mucosal surface. For HIV infection HIV-1 X4 tropic SF33 and R5 tropic SF162 viruses will be used. For each explant 10^5 TCID50 virions will be used. For EBV B95-8 strain will be used and each explant will be infected with 10^5 virions. HIV and EBV co-infection will be performed by simultaneous infection of HIV and EBV viruses. Also, the tissue explants will be infected with cell-associated HIV and EBV. The PBMC and purified B-lymphocytes will be infected with HIV and EBV, respectively, and after 3-5 days these cells will be added to the mucosal surface of tissue explants.
5. Collection of amniotic fluid and breast milk.
To examine the role of amniotic fluid and breast milk in HIV infection of fetal mucosal epithelium we will coleect amniotic fluid and breast milk. Volunteers will donate amniotic fluid and breast milk for these studies. Five ml amniotic fluid from 10 healthy HIV-seronegative and 4 HIV-positive pregnant women will be collected at 18 and 22 weeks of gestation. Amniotic fluid samples will be collected by transabdominal amniocentesis with a single needle insertion, which will take place at the [redacted] About 5-20ml of amniotic fluid will be aspirated into the syringe and centrifuged for 10 min at 1000 r/min. Pellet containing amniotic cells will be saved for karyotype analysis, and 5 ml of supernatant will be used for our research. Amniotic fluid will be filtered through 22-µm pore filters and aliquoted in 500 µl and stored at –80 C for further use in our research. Fifty ml of breast milk from 20 healthy HIV-seronegative women will be collected during the first 2-3 month of breast-feeding. Prior to sample collection, 2-3 ml of milk will be removed from the breast, and the nipple and surrounding areas of the breast that will have contact with the pump (Lactation Care Inc., Newton MA) will be cleaned with an ethanol-infused sterilizing pad and completely dried. The bottle and tubes will be sterilized by boiling them, according to manufacturer instruction. After collection breast milk samples will be filtered through 0.8-µm pore filters and aliquoted in 500 µl and stored at –80 C for further use in our research. It is expected that the donors will be between the ages of 18 and 40 years.
6. Collection of salivaand blood samples from HIV positive and negative individuals.
To study the status of EBV and HIV co-infection (lytic and latent) in monocytes and lymphocytes from HIV-positive individuals we will collect blood and saliva samples from 150 HIV-positive patients with HAART, 150 HIV-positive w/o HAART and 150 HIV-negative healthy volunteers. We

and blood samples. Collection of blood samples will be at [redacted]

If you have a procedure table, attach it to the submission with your other study documents.

16.2 Interviews, questionnaires, and/or surveys will be administered or focus groups will be conducted:

○ Yes ◉ No

List any standard instruments used for this study:

Attach any non-standard instruments at the end of the application.

16.3 Conduct of study procedures or tests off-site by non-UCSF personnel:

○ Yes ◉ No

If yes, explain:

16.4 Sharing of experimental research test results with subjects or their care providers:

○ Yes ◉ No

If yes, explain:

16.5 * Specimen collection for future research and/or specimen repository/bank administration:

◉ Yes ○ No

16.6 Time commitment (per visit and in total):

1. For obtaining one biopsy sample per visit will be required about 30 min. Each individual will be seen only once. From some of biopsy donors will be collected about 20 ml blood samples. For obtaining one blood sample will be required about 10-20 min. Each individual will be seen only once.
2. For obtaining one blood sample will be required about 10-20 min. Each individual will be seen only once.
3. For obtaining one saliva sample will be required about 10-20 min. Each individual will be seen only once.
4.The time to obtaining one breast milk sample will be about 10 min.

16.7 Locations:

1.The oral biopsy tissues from the adult tongue, and buccal mucosa from normal healthy individuals and HIV-positive patients will be collected in [redacted]
Cervical tissue will be collected from HIV-negative and -positive premenopausal women at the [redacted] For obtaining one biopsy sample per visit will be required about 30 min. Each individual will be seen only once. From some of biopsy donors will be collected about 20 ml blood samples. For obtaining one blood sample will be required about 10-20 min. Each individual will be seen only once.
2. The oral autopsy tissues will be obtained from infants (cadavers) under 1 year old by [redacted]

2. The blood samples from HIV/AIDS patients and normal healthy volunteers will be collected in [redacted] For obtaining one blood sample will be required about 10-20 min. Each individual will be seen only once.
3. The saliva samples from HIV/AIDS patients and normal healthy volunteers will be collected in [redacted] For obtaining one saliva sample will be required about 10-20 min. Each individual will be seen only once.
4. The fetal tongue, tonsil, gastric and intestinal tissue samples from discarded fetus will be collected in [redacted] There is no direct patient contact for this portion of the study.
5. Theamniotic fluid samples from normal healthy individuals and HIV/AIDS patients will be collected in [redacted] and [redacted] There will be no additional time requirement per patient apart from what is done in routine clinical care.
6. The breast milk samples will be collected in breastfeeding healthy HIV-negative women at [redacted] The time to obtaining one breast milk sample will be about 10 min.
7. All HIV and/or EBV and/or HPV infection procedures with adult and fetal tissue samples in presence of amniotic fluid and breast milk will be performed in [redacted]

16.8 Describe the resources in place to conduct this study in a way that assures protection of the rights and welfare of participants:

All specimens will be de-identified and designated by study numbers. The researchers and tissue bank staff will not have access to this information, rather they will have only study numbers. This numbers will be destroyed as soon as the study in completed. All data required by the protocol will be recorded and entered into electronic database using the unique study number. The study team will review the electronic data for accuracy. Data clarification or corrections will be made electronically and the databases will be password secured.

To minimize the discomfort of the oral tissue biopsy and transabdominal amniocentesis procedures,a local anesthetic is provided, followed by as-needed pain medications after the procedure. Sterile technique is used to avoid infection, and bleeding is stopped using standard clinical procedures

17.0 Specimen Collection for Future Research and/or Specimen Repository/Bank Administration

(Note: This section replaces the old "Human Biologic Specimen Collecting and/or Banking for Future Research" supplement form. Please do not attach the old form to this application.)

17.1 Specimens are (check all that apply):

- ☐ Surplus clinical specimens from a diagnostic or therapeutic procedure
- ☑ Specimens collected for research purposes only
- ☐ Other

If Other, explain:

17.2 Types of specimens:

- ☑ Blood
- ☑ Tissue (describe below):
- ☐ Existing/archival materials (name source below): --

Describe and/or name source:

17.3 Consent will be obtained via:

- ☐ Separate specimen banking consent form
- ☑ Specimen banking section within a main research study consent form
- ☐ Surgical consent form with tissue donation brochure

17.4 Specimens will ultimately be stored (check all that apply):

UCSF

- ☑ UCSF repository/bank being established under this protocol
- ☐ Existing UCSF specimen repository/bank with CHR approval

Provide the name of the bank and CHR approval number (if not being banked at UCSF under this protocol):

Outside Entity

- ☐ Cooperative group bank
- ☐ NIH
- ☐ Other university
- ☐ Industry sponsor
- ☐ Other

Specify to what institution, cooperative group or company specimens will be transferred:

17.5 Direct identifiers will be sent with specimens or shared with other researchers and/or outside entities:

- ○ Yes
- ◉ No
- ○ N/A – Specimens will not be shared with others

If **Yes**, which identifiers will be sent with specimens:

- ☐ Name
- ☐ Date of birth
- ☐ Social Security number
- ☐ Medical record number
- ☐ Address
- ☐ Phone number
- ☐ Email address
- ☐ Other dates (surgery date, clinic visit dates, etc.)

If **Yes**, provide a justification for sending direct identifiers with the specimens:

18.0 Establishing a Specimen Repository/Bank at UCSF

(Note: This section replaces the old "Human Biologic Specimen Collecting

and/or Banking for Future Research" supplement form. Please do not attach the old form to this application.)

18.1 The repository/bank is physically located at (list the address and room number for all locations):

[redacted]

18.2 Methods for maintaining confidentiality:

- [x] Samples are completely de-identified before being added to the bank/repository. There is no way to link the specimens back to the subjects.
- [ ] Samples are coded and researchers are able to link the specimens to specific subjects. However, future recipients will not receive direct identifiers with the specimens.
- [ ] Samples are stored with direct identifiers in the repository. However, future recipients will not receive direct identifiers with the specimens.
- [ ] Samples are coded and/or kept with direct identifiers in the repository. The bank/repository may release identifiers with specimens to researchers under special circumstances with prior IRB approval.

Explain under what circumstances identifiers may be released:

18.3 If the repository/bank maintains the identifiers, list the identifiers that will be maintained with the specimens:

- [ ] Name
- [ ] Date of birth
- [ ] Social Security number
- [ ] Medical record number
- [ ] Address
- [ ] Phone number
- [ ] Email address
- [ ] Other dates (dates of surgery, visit dates)

18.4 Clinical follow-up data will be linked to specimens:

- ( ) Yes
- (•) No

If **Yes**, provide duration of follow-up or indefinitely:

18.5 There is a formal specimen utilization review process:

- ( ) Yes
- (•) No

If **Yes**, describe the process:

18.6 Specimens banked at UCSF may be made available to (check all that apply):

- [x] UCSF researchers
- [ ] Non-UCSF researchers
- [ ] Industry

19.1 Study drug or treatment is available off-study:

- ○ Yes
- ◉ No
- ○ Not applicable

19.2 Describe the usual care or activities at UCSF (or study site) that are available to prospective subjects who do not enroll in this study:

This study does not involve any treatments and participation in the study is voluntary.

19.3 Describe other alternatives to study participation that are available to prospective subjects:

This study do not involve any treatment and do not have any significant impact on subject's concurrent or future care, the alternative may be not to participate.

20.0 Risks and Benefits

20.1 Risks and discomforts:

A. Risks and Discomforts:
1. Describe the risks and discomfortsof any investigational or approved drugs, devices and procedures being used or assigned for study purposes. Describe the expected frequency of particular side effects. If subjects are restricted from receiving standard therapies during the study, please also describe the risks of those restrictions.
Risk and discomforts for biopsy procedures of adult oral and cervical tissue samples.The only potential risks are those associated with donating tissue biopsy material will include discomfort, bleeding, and rarely, infection. Sterile technique is used to avoid infection and bleeding is stopped using standard clinical procedures.

Risk and discomforts for collection blood samples.The only potential risks are those associated with donating blood samples will be discomfort, bleeding, and rarely, infection. To reduce potential risk blood samples will be collected for from donors who weigh at least 110 pounds, and the amounts drawn may not exceed 40 ml. No more than three skin punctures will to be made in any single attempt to draw blood.Sterile technique is used to avoid infection and bleeding is stopped using standard clinical procedures.
Risk and discomforts for collection saliva.Collection of saliva has no potential risk or discomfort.

Risk and discomforts for collecting of amniotic fluid samples.
The potential risks are those associated with the transabdominal amniocentesis procedure, including miscarriage/pregnancy loss, preterm premature rupture of the membranes and fetal injury, discomfort, bleeding, inflammation and, rarely, infection. Sterile technique is used to avoid infection and bleeding is stopped using standard clinical procedures. To reduce other risks amniocenteses will be performed under continuous ultrasound guidance. The amniocenteses will only be performed for clinical purposes. It is not expected that participation in the study will have any impact on the risks associated with the clinical procedure as no additional fluid will be removed than would have for clinical purposes.

Risk and discomforts for collection breast milk.Collection of breast milk has minimum potential risk or discomfort, the only potential risks are those associated with un-optimized electrical pump. High suction setting of pump may cause discomfort and to minimize discomfort the setting of pump will be optimized and examined for each pumping kit and individual donor.

20.2 Steps taken to minimize risks to subjects:

To minimize the discomfort of the oral tissue biopsy and transabdominal amniocentesis procedures a local anesthetic is provided, followed by as-needed pain medications after the

procedure. Sterile technique is used to avoid infection, and bleeding is stopped using standard clinical procedures.

20.3 Benefits to subjects:

○ Yes ◉ No

If yes, describe:

20.4 Benefits to society:

Knowledge of EBV, HPV and HIV infection via the adult and fetal oral cavity has important biological and public health implications regarding transmission of infection from one individual to another.

20.5 Explain why the risks to subjects are reasonable:

The study risks on an individual level will be minimal (potential loss of privacy, risks associated with blood draws). Also, the risk of harm from oral biopsies is very low and the importance of the knowledge to be gained is very high. There are no significant risks from donating 10cc of breastmilk and the amniotic fluid volunteers would be obtaining the amniocentesis for clinical purposes anyway.

21.0 Data and Safety Monitoring Plan

21.1 Describe the plan for monitoring data and safety:

21.2 This study requires a Data and Safety Monitoring Board:

○ Yes
◉ No or not sure

If **yes**, press **SAVE and CONTINUE** to move to the next section of the application.

21.3 If No, provide rationale:

○ Social/Behavioral research
○ Phase I trial
○ Treatment IND/Compassionate Use Trial
◉ Other (explain below)

If **Other,** explain:

This project is not interventional study.

22.0 Confidentiality and Privacy

22.1 Study data are:

☐ Derived from the Integrated Data Repository (IDR)
☐ Derived from a medical record (identify source below)
☐ Added to the hospital or clinical medical record

- [ ] Used to make health care decisions
- [ ] Obtained from the subject, including interviews, questionnaires
- [ ] Obtained from a foreign country or countries only
- [ ] Obtained from records open to the public
- [ ] Obtained from existing research records
- [ ] None of the above

If **derived from a medical record**, identify source:

22.2 Plans for accessing subject information while maintaining privacy:

Advertisements will be placed (in UCSF campus, local newspapers and on the Internet) seeking volunteers to donate blood samples, tongue and buccal mucosal tissue, amniotic fluid and breast milk from HIV-negative individuals. Interested individuals will be asked to contact [redacted] who will describe the study. If the individual remains interested, he or she will come to the clinic where the purpose of the study and the procedures will again be described. The individual will be enrolled after his/her questions are answered and he/she reads and signs the informed consent forms. After signing of consent the PI will obtain information about participants.

22.3 Identifiers may be included in research records:

(•) Yes ( ) No

If **yes**, check all the identifiers that may be included:

- [x] Names
- [x] Dates
- [x] Postal addresses
- [x] Phone numbers
- [ ] Fax numbers
- [ ] Email addresses
- [x] Social Security Numbers*
- [x] Medical record numbers
- [x] Health plan numbers
- [ ] Account numbers
- [ ] License or certificate numbers
- [ ] Vehicle ID numbers
- [ ] Device identifiers or serial numbers
- [ ] Web URLs
- [ ] IP address numbers
- [ ] Biometric identifiers
- [ ] Facial photos or other identifiable images
- [ ] Any other unique identifier

* Required for studies conducted at the VAMC

22.4 Plans for maintaining privacy in the research setting:

All specimens will be de-identified and designated by study numbers. The researchers and tissue bank staff will not have access to this information, rather they will have only study numbers. This numbers will be destroyed as soon as the study in completed. All data required by the protocol will be recorded and entered into electronic database using the unique study number. The study team will review the electronic data for accuracy. Data clarification or corrections will be made electronically and the databases will be password secured.

22.5 Possible consequences to subjects resulting from a loss of privacy:

We do not see potential risks to reputation, insurability or other social risks that would occur as a result of participation n this study that is above that received in usual medical care.

22.6 Identifiable information might be disclosed as part of study activities:

○ Yes ◉ No

If **yes**, indicate where identifiable information may be released to:

- ☐ The subject's medical record
- ☐ The study sponsor
- ☐ The US Food & Drug Administration (FDA)
- ☐ Others (Specify below)
- ☐ A Foreign Country or Countries

If **Others**, specify:

22.7 Indicate how data are kept secure (check all that apply):

- ☐ Data are stored securely in My Research
- ☑ Data are coded; data key is destroyed at end of study
- ☐ Data are coded; data key is kept separately and securely
- ☐ Data are kept in a locked file cabinet
- ☐ Data are kept in a locked office or suite
- ☑ Electronic data are protected with a password
- ☑ Data are stored on a secure network
- ☐ Data are collected/stored using REDCap or REDCap Survey

22.8 Additional measures to assure confidentiality:

22.9 This study may collect information that State or Federal law requires to be reported to other officials or ethically requires action:

○ Yes ◉ No

Explain:

22.10 This study will be issued a Certificate of Confidentiality:

○ Yes ◉ No

23.0 Subjects

23.1 Check all types of subjects that may be enrolled:

- ☐ Inpatients
- ☐ Outpatients
- ☑ Healthy volunteers

☑ Staff of UCSF or affiliated institutions

23.2 Additional vulnerable populations:

☐ Children
☐ Subjects unable to consent for themselves
☐ Subjects unable to consent for themselves (emergency setting)
☐ Subjects with diminished capacity to consent
☑ Subjects unable to read, speak or understand English
☑ Pregnant women
☐ Fetuses
☐ Neonates
☐ Prisoners
☐ Economically or educationally disadvantaged persons
☐ Investigators' staff
☐ Students

Explain why it is appropriate to include the types of subjects checked above in this particular study:

Describe the additional safeguards that have been included in the study to protect the rights and welfare of these subjects and minimize coercion or undue influence:

Omit *minors, those unable to consent for themselves,* and *prisoners*(who are covered by separate Supplements); **for *pregnant women, fetuses, and neonates*, see section G below**):
Category 46.204:
(i) For this study will be collected the amniotic fluid samples from HIV-positive and-negative pregnant women. These amniotic fluid samples will be collected after separation of fetal cells for karyotyping analysis, thus these samples will be otherwise discarded. HIV-infected women do not, routinely, obtain amniocenteses. Nonetheless, some of the HIV-infected pregnant patients do undergo amniocentesis in the setting of abnormal first or second-trimester genetic screening tests or advanced maternal age.
(ii) For this study, will be collected the tissue samples from aborted fetuses that are also discarded materials.

Since, above procedures do not have any interventions and do not cause risk to pregnant women or life fetus this research does not fall within the sections 45 CFR 56 subpart B.

24.0 Inclusion of Non-English Speaking Subjects

24.1 Indicate which method(s) you will use to consent non-English speaking subjects:

☑ Preferred Method—Consent form and other study documents will be available in the subject's primary language Personnel able to discuss participation in the patient's language will be present for the consent process.
☑ Short-Form—A qualified interpreter will translate the consent form verbally, and subjects will be given the Experimental Subject's Bill of Rights in their primary language, following instructions in Those Who do not Read, Speak or Understand English for required witnessing and signatures

24.2 Explain how you will maintain the ability to communicate with non-English speakers throughout their participation in the study:

Will have trained bilingual staff available for translation.

25.0 Inclusion of Pregnant Women, Fetuses and/or Neonates

25.1 Review the regulatory categories and identify all sections of 45 CFR 46 Subpart B under which you believe the research falls and provide study-specific information showing why the research falls within those sections:

Category 46.204:
(i) For this study will be collected the amniotic fluid samples from HIV-positive and-negative pregnant women. These amniotic fluid samples will be collected after separation of fetal cells for karyotyping analysis, thus these samples will be otherwise discarded. HIV-infected women do not, routinely, obtain amniocenteses. Nonetheless, some of the HIV-infected pregnant patients do undergo amniocentesis in the setting of abnormal first or second-trimester genetic screening tests or advanced maternal age.
(ii) For this study, will be collected the tissue samples from aborted fetuses that are also discarded materials.

Since, above procedures do not have any interventions and do not cause risk to pregnant women or life fetus this research does not fall within the sections 45 CFR 56 subpart B.

26.0 Recruitment

26.1 * Methods (check all that apply):

- [x] Study investigators (and/or affiliated nurses or staff) recruit their own patients directly in person or by phone.
- [ ] Study investigators recruit their own patients by letter. Attach the letter for review.
- [ ] Study investigators send a "Dear Doctor" letter to colleagues asking for referrals of eligible patients. If interested, the patient will contact the PI or the PI may directly recruit the patients (with documented permission from the patient). Investigators may give the referring physicians a study information sheet for the patients.
- [ ] Study investigators provide their colleagues with a "Dear Patient" letter describing the study. This letter can be signed by the treating physicians and would inform the patients how to contact the study investigators. The study investigators may not have access to patient names and addresses for mailing
- [ ] Advertisements, notices, and/or media used to recruit subjects. Interested subjects initiate contact with study investigators. Attach ads, notices, or media text for review. In section below, please explain where ads will be posted.
- [ ] Study investigators identify prospective subjects through chart review. (Study investigators request a Waiver of Authorization for recruitment purposes.)
- [ ] Large-scale epidemiological studies and/or population-based studies: Prospective subjects are identified through a registry or medical records and contacted by someone other than their personal physician. (Study investigators request a Waiver of Authorization for recruitment purposes.)
- [ ] Direct contact of potential subjects who have previously given consent to be contacted for participation in research. Clinic or program develops a CHR-approved recruitment protocol that asks patients if they agree to be contacted for research (a recruitment database) or consent for future contact was documented using the consent form for another CHR-approved study.
- [ ] Study investigators list the study on the School of Medicine list of UCSF Clinical Trials website or a similarly managed site. Interested subjects initiate contact with investigators.
- [ ] Study investigators recruit potential subjects who are unknown to them through methods such as snowball sampling, direct approach, use of social networks, and random digit dialing.
- [ ] Other

If **Other**, explain:

26.2 How, when, and by whom eligibility will be determined:

Eligibility for the oral and cervical biopsy isolation for this research will be determined by [redacted] respectively. They will examine the volunteers for oral and cervical infection, allergy and inflammation before the biopsy procedure. The HIV-negative volunteers will be tested for HIV to verify their status.

26.3 How, when, where and by whom potential subjects will be approached:

Advertisements will be placed seeking volunteers to donate blood samples, oral and cervical mucosal tissue, amniotic fluid and breast milk. The advertisements will be posted around the UCSF and San Francisco General campuses, in local newspapers and on the Internet. All volunteers will be financially compensated for their time. Interested individuals will be asked to contact [redacted] who will describe the study. If the individual remains interested, he or she will come to the clinic where the purpose of the study and the procedures will again be described. The individual will be enrolled after his/her questions are answered and he/she reads and signs the informed consent forms.

1. The blood samples from HIV/AIDS patients and normal healthy volunteers will be collected in [redacted]

2.The biopsy tissues from the adult oral and cervical mucosa will be collected ir [redacted]

3. The fetal tongue, tonsil, gastric and intestinal tissue samples from discarded fetus will be collected in [redacted]

4. The and amniotic fluid samples from normal healthy individuals and HIV/AIDS patients will be collected in [redacted]

5. The breast milk samples will be collected in breastfeeding healthy HIV-negative women at [redacted]

26.4 * Protected health information (PHI) will be accessed prior to obtaining consent:

○ Yes ◉ No

27.0 Informed Consent

27.1 * Methods (check all that apply):

- [x] Signed consent will be obtained from subjects and/or parents (if subjects are minors)
- [ ] Verbal consent will be obtained from subjects using an information sheet or script
- [ ] Electronic consent will be obtained from subjects via the web or email
- [ ] Implied consent will be obtained via mail, the web or email
- [ ] Signed consent will be obtained from surrogates
- [ ] Emergency waiver of consent is being requested for subjects unable to provide consent
- [ ] Informed consent will not be obtained

27.2 Process for obtaining informed consent:

After advertisements any responded subject will be invited to office of [redacted] for blood samples study, to office of [redacted] for amniotic fluid study and [redacted] for oral tissues and [redacted] for cervical tissue samples study. [redacted] will meet with each of interested subject in their office and describe them he purpose of the study and the procedures and answered their questions. If the individual remains interested he/she reads and signs the informed consent forms.

The fetal samples from discarded aborted materials will be collected at the [redacted] Since, this study will use only discarded fetal materials, the mothers consent to the use of the fetal tissue in research will not be obtained by the researchers in this study. The fetal samples will be used for the research and will be banked in [redacted]

27.3 How investigators will make sure subjects understand the information provided to them:

Persons obtaining consent will ask potential participants to restate what they have understood at various steps along the informed consent procedure.

28.0 Financial Considerations

28.1 Subjects payment or compensation method (check all that apply):

Payments will be (check all that apply):

- [ ] Subjects will not be paid
- [ ] Cash
- [x] Check
- [ ] Gift card
- [ ] Other:

Specify **Other**:

28.2 Describe the schedule and amounts of payments, including the total subjects can receive for completing the study. If deviating from recommendations in Subject Payment Guidelines, include specific justification below.

The amount of payment for singly donation will be as follow:
$20 for 40 ml blood
$150 for one tongue tissue
$100 buccal tissue
$100 cervical tissue

28.3 Costs to Subjects: Will subjects or their insurance be charged for any study procedures?

- ○ Yes
- ◉ No

If **yes,** describe those costs below, and compare subjects' costs to the costs associated with alternative care off-study. Finally, explain why it is appropriate to charge those costs to the subjects.

29.0 CTSI Screening Questions

29.1 * This study will be carried out at one of the UCSF Clinical Research Centers (CRCs) or will utilize CRC services:

- ○ Yes
- ◉ No

29.2 This project involves community-based research:

- ○ Yes
- ○ No

29.3 This project involves practice-based research:

○ Yes ○ No

29.4 Please check other CTSI services below that you plan to utilize to conduct your research:

Guidance and Services:

- ☐ Biostatistics
- ☐ Study Design and Implementation
- ☐ Data Management
- ☐ Ethics
- ☐ Health Policy
- ☐ Bioinformatics Data Analysis
- ☐ Regulatory Knowledge
- ☐ THREDS The Health Record Data Service
- ☐ Community-Engaged Research
- ☐ Collaborating with Kaiser Researchers

Clinical Research Centers:

- ☐ Community Engagement (CE)

Funds to Innovate:

- ☐ Strategic Opportunity Support (SOS)

Training:

- ☐ Clinical & Translational Sciences Training (CTST)
- ☐ Career Advancement (CA)

CTSI Core Services:

- ☐ Animal/Preclinical
- ☐ Array
- ☐ Bioinformatics
- ☐ Biostatistics
- ☐ Cell Culture
- ☐ Clinical Services
- ☐ Epidemiology
- ☐ Flow Cytometry
- ☐ Human/Clinical
- ☐ Imaging
- ☐ Immunohistochemistry
- ☐ Islet Production
- ☐ Microscopy
- ☐ Molecular/Genomic
- ☐ Monoclonal Antibody
- ☐ Proteomics
- ☐ Resale Products
- ☐ Tissue

UNIVERSITY OF CALIFORNIA, SAN FRANCISCO
CONSENT TO DONATE SALIVA SAMPLES FOR FUTURE RESEARCH

Study Title: HIV, EBV and HPV interaction with mucosal epithelium
PI, [redacted],
Co-PI- [redacted]

This is a request that you donate specimens (saliva) for medical research. The researchers from the University of California at San Francisco will explain this research to you.

Medical research includes only people who choose to take part. Take your time to make your decision about participating. You may discuss your decision with your family and friends and with your health care team. If you have any questions, you may ask the researchers.

You are being asked to take part in this study because you are infected with HIV.

Why is this research being done?

We are performing this study so we can learn how oral transmission of HIV occurs from mother to children during pregnancy and through breastfeeding.

Oral transmission is a potentially important, poorly understood route of HIV infection that has global consequences. The oral cavity of the fetus/neonate may serve as an efficient portal of entry for mother-to-child transmission (MTCT) of HIV, yet oral transmission of HIV among adults is rare. The reasons for this difference are not well understood. Therefore, we need how oral transmission of HIV occurs from mother to children during pregnancy and through breastfeeding. Your doctor/researcher will be happy to discuss any questions you may have about your involvement in the study and we encourage you to ask his or her help if you are unclear about anything.
This study is funded by the National Institutes of Health

How many people will take part in this research?

About 300 people, 150 with HAART and 150 w/o HAART will donate saliva samples.

What will happen if I agree to donate my specimens?

If you agree to let researchers collect and store your specimens for future research, the following will happen:

- We will collect about 5 ml of saliva. We will use the donated samples in our research to study how oral transmission of HIV occurs from mother to children during pregnancy and through breastfeeding. We may give the specimen to the UCSF scientists only. Reports about any research will not be given to you or your doctor. Sometimes specimens are used for genetic research (about diseases that are passed on in families). Even if we use the specimen for genetic research, we will not

put the results in your medical record. Your specimen will be kept until it is used up or destroyed. It may be used to develop new drugs, tests, treatments or products. In some instances these may have potential commercial value. Your personal health information cannot be used for additional research without additional approval from either you or a review committee.

- Your specimen will be kept for indefinitely. If you decide later that you do not want your sample and information to be used for future research, you can tell us, and we will destroy any remaining identifiable sample and information.

What risks are involved with donating specimens for research?

- There is no risk for collection of saliva.
- Confidentiality: Participation in research may cause a loss of privacy, but information about you will be kept as confidential as possible. Your name will not be used in any published reports about this study.

What happens if I am injured because I took part in this study?

It is important that you tell your study doctor, [redacted], if you feel that you have been injured because of taking part in this study. You can tell the doctor in person or call to [redacted].

> **Treatment and Compensation for Injury:** If you are injured as a result of being in this study, treatment will be available. The costs of the treatment may be covered by the University of California, depending on a number of factors. The University does not normally provide any other form of compensation for injury. For further information about this, you may call the office of the Committee on Human Research at 415- 476-1814.

What are the benefits of donating specimens for research?

There will be no direct benefit to you from allowing your specimen to be used for research. However, we hope we will learn something that will help in the treatment of future patients.

What financial issues should I consider before donating?

You will not be charged for donating your specimen.

What alternatives do I have?

The alternative is not to participate in the study.

What are my rights if I take part in this study?

Taking part in this study is your choice. You may choose either to take part or not to take part in the study. No matter what decision you make, there will be no penalty to you and you will not lose any of your regular benefits. Leaving the study will not affect your medical care. You can still get your medical care from our institution.

In the case of injury resulting from this study, you do not lose any of your legal rights to seek payment by signing this form.

Who can answer my questions about the study?

You can talk to the researchers about any questions or concerns you have about this study. Contact the researcher(s), [redacted]

For questions about your rights while taking part in this study, call the office of the **Committee on Human Research**, UCSF's Institutional Review Board (a group of people who review the research to protect your rights) at **415-476-1814**

Consent

You have been given copies of this consent form and the Experimental Subject's Bill of Rights to keep.

PARTICIPATION IN RESEARCH IS VOLUNTARY. You have the right to decline to participate or to withdraw at any point in this study without penalty or loss of benefits to which you are otherwise entitled.

You will be asked to sign a separate form authorizing access, use, creation, or disclosure of health information about you.

If you wish to participate in this study, you should sign below.

Date	Subject's Signature for Consent
Date	Person Obtaining Consent