Review Response Submission Form (Version 6.0)

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 1:

Please delete all mention of cervical biopsy from the application (click 'Printer Friendly' to open the application in a separate window and then use Control + F to find all the places where cervical is mentioned).

Please attach the modified 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 have deleted all text related cervical biopsies

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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:**

Review Response Submission Form (Version 5.0)

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

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Stipulations that must be addressed:

Stipulation 1 out of 2:

Since cervical biopsies are no longer to be collected, will [redacted] and [redacted] still be part of this study?

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

Do you accept this Stipulation?	○ **N/A** ● **Yes** ○ **No**
Provide an explanation on how you addressed this Stipulation:	Dr. Palefsky and Dr.Chin-Hong will be part of the study, because we will investigate the cervical biopsies, which were collected in the past years. Drs Palefsky and Chin-Hong will be participated in data analysis.

Stipulation 2 out of 2:

Description:

Please delete all mention of cervical biopsy from the application (click 'Printer Friendly' to open the application in a separate window and then use Control + F to find all the places where cervical is mentioned).

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 have deleted all text related to cervical biopsy.

Comments That Must Be Addressed With Follow-up Deadlines:

No Stipulation entered.

Comments:

Stipulation 1 out of 1:

Description:

Guidance on "Responding to Requests for Submission Corrections" (stipulations) is located under the Help icon in the upper right-hand corner of the iRIS system. **Most stipulations are now linked to the corresponding document. Click "Add revision" next to the items in the stipulations list to create an editable version.** Please contact me, Jessica Phillips, at 415-476-3477 or at **jessica.phillips@ucsf.edu** if you have any questions.

Stipulation Type: **(Comments)**

Do you accept this Stipulation?	○ **N/A** • **Yes** ○ **No**
Provide an explanation on how you addressed this Stipulation:	We have follwed your recommendation and accordingly revised the application.

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:**

Continuing Review Submission Form (Version 4.2)

1.0

Continuing Review Form

To start with a copy of the last continuing review form (only if there is one):
1) Open the study through My Studies.
2) From the Study Management dashboard page, click the Continuing Review form link, check the box next to last year's form and click "Copy Form."
3) After you submit the form, click on the Continuing Review Due task on your home screen to remove the notification.
Please review the Quick Guide titled "Submitting Post-Approval Forms" in the system "Help" area for step-by-step instructions and screen shots.

1.1 Principal Investigator:

[redacted]

1.2 Study Title:

HIV, EBV and HPV interaction with mucosal epithelium

1.3 Study Number:

10-03277

1.4 Expiration Date:

06/04/2014

*If the study has expired, an explanation is required at the end of the form.

1.5 Lay Summary:

Interaction of HIV and HIV-associated opportunistic viruses, including EBV and HPV with oral and genital mucosal and GI tract mucosal epithelia is not well understood. Therefore, our goal is to establish an ex vivo organ culture model from oral, genital and GI tract mucosal epithelia, to study the molecular mechanisms of HIV, EBV and HPV mucosal transmission.

1.6 * This is a:

- ○ Continuing Review Only—no changes from last approval
- ◉ Continuing Review and Minor Modification
- ○ Continuing Review and Major Modification

1.7 * This application includes personnel changes:

1.8 * Has your study expired:

○ Yes ◉ No

If **Yes**, work on this study must stop **IMMEDIATELY.**

1.9 Outstanding Stipulations:

No Stipulation is outstanding.

2.0 Study Status

2.1 * Have any new risks been identified since the last continuing review (or since approval if this is the first continuing review):

○ Yes ◉ No

2.2 Current study status: (*choose one only*)

Study is open to accrual:

- ○ No participants have been enrolled and no additional risks have been identified.
- ○ Participants are currently receiving study intervention.
- ◉ Participants have been enrolled but none are currently receiving study intervention.
- ○ Ongoing medical record review/biological specimen analysis.

Study is closed to accrual:

- ○ Some participants are still receiving study intervention.
- ○ Study intervention is complete for all participants; research-related diagnostic tests or follow-up clinic visits are continuing.
- ○ Study intervention is complete or there was no intervention, and there is ongoing research-related follow-up contact with participants via questionnaires, phones calls, interviews, or mailings.
- ○ Study intervention is complete or there was no intervention and follow-up is limited to review of medical records or other records (no ongoing contact).
- ○ Study is in data analysis phase only.

2.3 Summary of results to date:

We have established polarized tonsil epithelial cells and polarized oriented tonsil tissue explants from children under 5 years old. Polarized cells and tissues were preincubated with chlorpromazine, nystatin, or amiloride, which are inhibitors of clathrin- and caveolin-mediated endocytosis and macropinocytosis, respectively. Polarized tonsil cells and tissues were also pretreated with antibodies against transmembrane protein T-cell immunoglobulin and mucin domain 1 (TIM-1), which is receptor for HIV-associated phosphatidylserine (PS). Then, dual tropic HIV-1_{SF33} was added to the apical surface of polarized cells and, after 4 h and 8 h, viral transmigration i.e., transcytosis was examined in the basolateral compartment by detecting HIV p24 using ELISA assay. HIV transmigration via mucosal tissue was evaluated by confocal immunofluorescence assay.

Analysis of HIV transmigration by inhibitors of clathrin- and caveolin-mediated endocytosis and macropinocytosis via polarized tonsil epithelial cells and tonsil tissue explants showed substantial

macropinocytosis in HIV transepithelial migration. We hypothesize that HIV envelope-associated PS may play a role in induction of macropinocytosis of virions. Tonsil epithelial cells expressed PS receptor TIM-1. Preincubation of tonsil epithelium with antibodies against TIM-1 substantially reduced (≈60%) HIV transcytosis, suggesting that interaction of virion-associated PS with TIM-1 of tonsil epithelium may lead to HIV macropinocytosis from the apical membranes of tonsil epithelial cells. Some tonsil epithelial cells containing HIV virions were stained with fluorescien labeled Sambucus nigra, a plant lectin that binds to M (microfold) cells, suggesting that tonsillar M cells may facilitate HIV transcytosis via tonsil mucosal epithelium.

Our data show that HIV macropinocytosis/transcytosis through infant tonsil epithelium may play a critical role in HIV MTCT. Interaction of HIV virion associated PS with TIM-1 may facilitate viral macropinocytosis/transcytosis. Interference of HIV-PS interaction with TIM-1 may substantially reduce the risk of HIV transmission via infant tonsil epithelium.

2.4 Brief summary of plans for the coming year:

For the coming year, we will begin to perform proposed experiments for Specific Aim 2. In this aim, we hypothesize that HIV-infected macrophages attach to the mucosal surfaces of fetal/infant oral and fetal intestinal epithelia by adhesion of LFA-1 of macrophages to ICAM-1 of epithelial cells. To test this hypothesis, we will study the role of LFA-1 and ICAM-1 in binding of HIV-infected macrophages to infant oral and intestinal mucosal epithelia. R5-tropic HIV-1_{SF170}-infected macrophages will be propagated and labeled with carboxyfluorescein diacetate succinimidyl ester (CFSE). Macrophages will then be incubated with function-blocking antibodies to LFA-1. In parallel experiments polarized oriented infant tonsil and fetal oropharyngeal and intestinal explants will be incubated with function-blocking antibodies to ICAM-1. Macrophages will then be added to the apical surfaces of tissue explants. Uninfected macrophages and explants without antibodies (or with isotype control antibodies) will serve as controls. At 4 h and 8 h, tissues will be fixed, sectioned and co-immunostained for HIV p24 (red) and pan-keratin (blue). CFSE-labeled HIV-positive macrophages (green) bound to the apical surface of the epithelium will be quantitatively analyzed. We expect that attachment of HIV-infected macrophages will be mediated by macrophage LFA-1 and epithelial ICAM-1.

3.0 Recent Literature

3.1 Summary of recent literature:

1. Tugizov, S.M., *et al.* Differential transmission of HIV traversing fetal oral /intestinal epithelia and adult oral epithelia. *Journal of Virology* **86**, 2556-2570 (2012).
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7. Nussenblatt, V., *et al.* Epidemiology and microbiology of subclinical mastitis among HIV-infected women in Malawi. *Int J STD AIDS* **16**, 227-232 (2005).
8. Semba, R.D. Mastitis and transmission of human immunodeficiency virus through breast milk. *Ann N Y Acad Sci* **918**, 156-162 (2000).
9. Semba, R.D. & Neville, M.C. Breast-feeding, mastitis, and HIV transmission: nutritional implications. *Nutr Rev* **57**, 146-153 (1999).
10. Bomsel, M. Transcytosis of infectious human immunodeficiency virus across a tight human epithelial cell line barrier. *Nat Med* **3**, 42-47 (1997).
11. Hocini, H. & Bomsel, M. Infectious human immunodeficiency virus can rapidly penetrate a tight human epithelial barrier by transcytosis in a process impaired by mucosal immunoglobulins. *Journal of Infectious Diseases* **179 Suppl 3**, S448-453 (1999).

4.0 Subject Accrual

4.1 Recruitment and enrollment activities:

New subjects enrolled since last continuing review

Locally:

11

For study overall:

11

Total subjects enrolled to date

Locally:

173

For study overall:

173

Total number of subjects that will be enrolled as specified in the Sample Size and Eligibility section of the CHR Application

Locally:

292

For study overall:

292

4.2 Did you exceed your approved enrollment numbers:

○ Yes ◉ No

Explain how this happened and what will be done to prevent this in the future:

4.3 Total refusals (individuals asked to be in the study but who chose not to participate) to date:

Locally:

none

4.4 Total withdrawals:

Locally:

none

Summarize the main reasons why subjects withdrew:

5.0 Significant Findings & Other Reportable Events

5.1 Did you report any 10-day reportable events, including complaints about the research, since the last continuing review (or since initial approval if this is the first continuing review):

○ Yes ◉ No

If **Yes,** provide a brief narrative summary of adverse events, protocol violations, safety information, and/or complaints over the past year so that CHR members can understand the overall trends:

5.2 Are you submitting any new or missed 10-day reportable events, including complaints, at this time (submit separately in system using the appropriate submission form):

○ Yes ◉ No

5.3 Plan for informing subjects of information that may affect willingness to continue participation:

If new risks have been identified since the last continuing review (or since initial approval if this is the first continuing review), explain how subjects were or will be informed of any new information and when the modification was approved if the notification already occurred:

Attach any additional documents (consent forms or contact letters) that will be used for this purpose at the end of the form.

5.4 * Does this study undergo formal on-site monitoring:

○ Yes ◉ No

5.5 * This study was audited by any external group or entity (i.e., sponsor, FDA) since its last renewal:

○ Yes ◉ No

Auditing entity:

5.6 If YES to either monitoring or auditing, describe any significant findings resulting from either the monitoring or auditing activities or state "None" if there were no significant findings:

5.7 Other reportable events:

If this is a biomedical **interventional study** did you have any **internal** (on-site) participant deaths determined to be *unrelated* to research participation?

○ Yes ◉ No

If **yes**, attach an **AE Summary Log** at the end of this form.

Is your study sponsor *requiring* that you forward any Study Sponsor **external** (off-site) Safety Reports *that are not otherwise reportable* under CHR guidelines:

○ Yes ◉ No

6.0 Updated Financial Disclosure

6.1 * Are there any changes in any financial interests related to this study or in any conflicts of interest of the PI or any other investigator:

○ Yes ◉ No

If **Yes**, the Conflict of Interest Advisory Committee (COIAC) office may contact you for additional information.

7.0 New Modifications

7.1 This modification is being submitted as a result of an adverse event report (AER), protocol violation or incident report, or publication of a new Investigator's Brochure (IB) or other safety data:

○ Yes ◉ No

Type of event:

○ Adverse event report
○ Protocol violation or incident report
○ New Investigator's Brochure
○ Other safety data

If **YES,** what date was the event submitted into the system on:

7.2 Proposed modifications:

The following modification was done in the revised application:
1. Collection of 15 cervical tissues was canceled and therefore sample size was reduced in the sections 15.1, 15.2 and 15.3 as follow:
15.1- 292
15.2- 292
15.3- 335

2. All related text to collection of cervical tissues were deleted in sections 12, 15 and 18.

3. Collection of saliva samples was canceled in previous renewal, however, the concent forms were approved. In this submission we did not attached the consent forms for saliva collection.

4. In the previous renewal we missed to attach the approved consent form for collection of oral (buccal) biopsies from HIV-negative donors, and in this renewal we attahced the missing consent form. We did not collect the buccal biopsies during last 1 year.

7.3 * This modification includes a change to enrollment targets:

◉ Yes ○ No

If **Yes**, be sure to update the subject numbers in the application (Section Samples Size and Eligibility).

7.4 Explain why these modifications are being made:

1. Collection of 15 cervical tissues was canceled as we do not study cervical tissues in our new R01 grant.

2. Due to canceling of cervical tissue collection all related text to cervical tissues were deleted in sections 12, 15 and 18.

3. Collection of saliva samples was canceled in previous renewal as we are completed this study. However, the concent forms were not deleted. In this submission we did not attached the consent forms for saliva collection.

4. In the previous renewal we missed to attach approved the consent form for collection of oral (buccal) biopsies from HIV-negative donors, and in this renewal we attahced the missing approved and unchanged consent form. We did not collect the buccal biopsies during last 1 year period.

7.5 * Does this modification require a change to the application form:

(•) Yes ( ) No

8.0 Revisions to the Application Form

8.1 Click the bar below to make revisions to the application form:

(Note: you are seeing this section because you either indicated that there are changes that affect the application or there are personnel changes that need to be made in the application.)

Edit/ View	Version	Title
	1.7	Study Application (Version 1.7) - Attached

9.0 Attach Consent Documents

9.1 Attach all consent documents you need approved with this continuing review: When possible, attach Word documents instead of PDFs.

Attach your consent forms following these instructions:

Approved Consent Documents with No Changes: All approved consent documents that you will continue to use must be attached. Click Select or Revise Existing. *If you have the currently approved consent form saved on your computer,* click Upload the Revised Consent. *If you need to download the approved consent form first,* click Download Document for Editing and then Upload the Revised Consent. Save your work. DO NOT SELECT THE CURRENTLY APPROVED VERSION OF THE CONSENT FORM. The CHR cannot stamp over last year's approval stamp and the submission will be returned to you.

Approved Consent Documents with Revisions: Click the Select or Revise Existing button. Click Upload the Revised Consent form and select the revised document from your computer. *If you need to download the current version of the consent form from iRIS first,* click Download Document for Editing and then Upload the Revised Consent after you've updated the document. Save your work.

For more information, click on the Help icon and read the "Revising Documents and Forms" quick guide.

New Consent Documents: Click on the Add a New Consent button and upload your new consent form.

Version	Sponsor Version	Title	Category	Language	Expiration Date	Consent Outcome	View Document

1.3.3		tonsil tissues		English	06/04 /2015	Void	265.89 KB
1.7		Consent-oral biopsy-HIV+		English	06/04 /2015	Void	202.82 KB
1.6		Consent-oral biopsy-HIV-		English	06/04 /2015	Void	203.40 KB
1.3		Consent-blood-HIV+		English	06/04 /2015	Void	168.83 KB
1.3		Consent-blood-HIV-		English	06/04 /2015	Void	162.63 KB
1.4		Consent breast milk-		English	06/04 /2015	Void	157.58 KB

9.2 Indicate which type(s) of consent documents are attached:

- ☐ New consent documents
- ☑ Active consent documents with NO changes
- ☐ Active consent documents with revisions
- ☐ No consent documents attached (explain below)

If no consent documents are attached, explain why not:

10.0 Attach Other Study Documents (AE Summary Log, revised documents, or new documents)

10.1 Upload AE Summary Log and/or any new or revised other documents here:

Attach the documents following these instructions:

Revised Documents: Follow these steps if you are revising currently approved documents or if you are submitting revised documents created by the study sponsor or lead site (e.g. revised protocols or investigator's brochures).

Click the Select or Revise Existing button. Click Upload the Revised Document and select the revised document from your computer. *If you need to download the current version of the document from iRIS first,* click Download Document for Editing and then Upload the Revised Document after you've updated the document. Save your work.

New Documents: Click on the Add Document button and upload your new document.

Reminder: If you are revising or adding new study documents, indicate that the continuing review includes modifications in question 1.6.

Approved Documents – No Changes: These documents do **not** need to be resubmitted during the continuing review.

Version	Sponsor Version	Title	Category	Expiration Date	Document Outcome	View Document
No Document(s) have been attached to this form.						

Continuing Review Submission Form (Version 4.1)

1.0

Continuing Review Form

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[redacted]

1.2 Study Title:

HIV, EBV and HPV interaction with mucosal epithelium

1.3 Study Number:

10-03277

1.4 Expiration Date:

06/04/2014

*If the study has expired, an explanation is required at the end of the form.

1.5 Lay Summary:

Interaction of HIV and HIV-associated opportunistic viruses, including EBV and HPV with oral and genital mucosal and GI tract mucosal epithelia is not well understood. Therefore, our goal is to establish an ex vivo organ culture model from oral, genital and GI tract mucosal epithelia, to study the molecular mechanisms of HIV, EBV and HPV mucosal transmission.

1.6 * This is a:

- ○ Continuing Review Only—no changes from last approval
- ◉ Continuing Review and Minor Modification
- ○ Continuing Review and Major Modification

1.7 * This application includes personnel changes:

- ○ Yes ○ No

1.8 * Has your study expired:

○ Yes ◉ No

If **Yes**, work on this study must stop **IMMEDIATELY.**

1.9 Outstanding Stipulations:

No Stipulation is outstanding.

2.0 Study Status

2.1 * Have any new risks been identified since the last continuing review (or since approval if this is the first continuing review):

○ Yes ◉ No

2.2 Current study status: (*choose one only*)

Study is open to accrual:

- ○ No participants have been enrolled and no additional risks have been identified.
- ○ Participants are currently receiving study intervention.
- ◉ Participants have been enrolled but none are currently receiving study intervention.
- ○ Ongoing medical record review/biological specimen analysis.

Study is closed to accrual:

- ○ Some participants are still receiving study intervention.
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- ○ Study intervention is complete or there was no intervention and follow-up is limited to review of medical records or other records (no ongoing contact).
- ○ Study is in data analysis phase only.

2.3 Summary of results to date:

We have established polarized tonsil epithelial cells and polarized oriented tonsil tissue explants from children under 5 years old. Polarized cells and tissues were preincubated with chlorpromazine, nystatin, or amiloride, which are inhibitors of clathrin- and caveolin-mediated endocytosis and macropinocytosis, respectively. Polarized tonsil cells and tissues were also pretreated with antibodies against transmembrane protein T-cell immunoglobulin and mucin domain 1 (TIM-1), which is receptor for HIV-associated phosphatidylserine (PS). Then, dual tropic HIV-1_{SF33} was added to the apical surface of polarized cells and, after 4 h and 8 h, viral transmigration i.e., transcytosis was examined in the basolateral compartment by detecting HIV p24 using ELISA assay. HIV transmigration via mucosal tissue was evaluated by confocal immunofluorescence assay.

Analysis of HIV transmigration by inhibitors of clathrin- and caveolin-mediated endocytosis and macropinocytosis via polarized tonsil epithelial cells and tonsil tissue explants showed substantial

macropinocytosis in HIV transepithelial migration. We hypothesize that HIV envelope-associated PS may play a role in induction of macropinocytosis of virions. Tonsil epithelial cells expressed PS receptor TIM-1. Preincubation of tonsil epithelium with antibodies against TIM-1 substantially reduced (≈60%) HIV transcytosis, suggesting that interaction of virion-associated PS with TIM-1 of tonsil epithelium may lead to HIV macropinocytosis from the apical membranes of tonsil epithelial cells. Some tonsil epithelial cells containing HIV virions were stained with fluorescien labeled Sambucus nigra, a plant lectin that binds to M (microfold) cells, suggesting that tonsillar M cells may facilitate HIV transcytosis via tonsil mucosal epithelium.

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2.4 Brief summary of plans for the coming year:

For the coming year, we will begin to perform proposed experiments for Specific Aim 2. In this aim, we hypothesize that HIV-infected macrophages attach to the mucosal surfaces of fetal/infant oral and fetal intestinal epithelia by adhesion of LFA-1 of macrophages to ICAM-1 of epithelial cells. To test this hypothesis, we will study the role of LFA-1 and ICAM-1 in binding of HIV-infected macrophages to infant oral and intestinal mucosal epithelia. R5-tropic HIV-1_{SF170}-infected macrophages will be propagated and labeled with carboxyfluorescein diacetate succinimidyl ester (CFSE). Macrophages will then be incubated with function-blocking antibodies to LFA-1. In parallel experiments polarized oriented infant tonsil and fetal oropharyngeal and intestinal explants will be incubated with function-blocking antibodies to ICAM-1. Macrophages will then be added to the apical surfaces of tissue explants. Uninfected macrophages and explants without antibodies (or with isotype control antibodies) will serve as controls. At 4 h and 8 h, tissues will be fixed, sectioned and co-immunostained for HIV p24 (red) and pan-keratin (blue). CFSE-labeled HIV-positive macrophages (green) bound to the apical surface of the epithelium will be quantitatively analyzed. We expect that attachment of HIV-infected macrophages will be mediated by macrophage LFA-1 and epithelial ICAM-1.

3.0 Recent Literature

3.1 Summary of recent literature:

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2. Tugizov, S.M., *et al.* HIV is inactivated after transepithelial migration via adult oral epithelial cells but not fetal epithelial cells. *Virology* **409**, 211-222 (2011).
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9. Semba, R.D. & Neville, M.C. Breast-feeding, mastitis, and HIV transmission: nutritional implications. *Nutr Rev* **57**, 146-153 (1999).
10. Bomsel, M. Transcytosis of infectious human immunodeficiency virus across a tight human epithelial cell line barrier. *Nat Med* **3**, 42-47 (1997).
11. Hocini, H. & Bomsel, M. Infectious human immunodeficiency virus can rapidly penetrate a tight human epithelial barrier by transcytosis in a process impaired by mucosal immunoglobulins. *Journal of Infectious Diseases* **179 Suppl 3**, S448-453 (1999).

4.0 Subject Accrual

4.1 Recruitment and enrollment activities:

New subjects enrolled since last continuing review

Locally:

11

For study overall:

11

Total subjects enrolled to date

Locally:

173

For study overall:

173

Total number of subjects that will be enrolled as specified in the Sample Size and Eligibility section of the CHR Application

Locally:

292

For study overall:

292

4.2 Did you exceed your approved enrollment numbers:

○ Yes ◉ No

Explain how this happened and what will be done to prevent this in the future:

4.3 Total refusals (individuals asked to be in the study but who chose not to participate) to date:

Locally:

none

4.4 Total withdrawals:

Locally:

none

Summarize the main reasons why subjects withdrew:

5.0 Significant Findings & Other Reportable Events

5.1 Did you report any 10-day reportable events, including complaints about the research, since the last continuing review (or since initial approval if this is the first continuing review):

○ Yes ◉ No

If **Yes,** provide a brief narrative summary of adverse events, protocol violations, safety information, and/or complaints over the past year so that CHR members can understand the overall trends:

5.2 Are you submitting any new or missed 10-day reportable events, including complaints, at this time (submit separately in system using the appropriate submission form):

○ Yes ◉ No

5.3 Plan for informing subjects of information that may affect willingness to continue participation:

If new risks have been identified since the last continuing review (or since initial approval if this is the first continuing review), explain how subjects were or will be informed of any new information and when the modification was approved if the notification already occurred:

Attach any additional documents (consent forms or contact letters) that will be used for this purpose at the end of the form.

5.4 * Does this study undergo formal on-site monitoring:

○ Yes ◉ No

5.5 * This study was audited by any external group or entity (i.e., sponsor, FDA) since its last renewal:

○ Yes ◉ No

Auditing entity:

5.6 If YES to either monitoring or auditing, describe any significant findings resulting from either the monitoring or auditing activities or state "None" if there were no significant findings:

5.7 Other reportable events:

If this is a biomedical **interventional study** did you have any **internal** (on-site) participant deaths determined to be *unrelated* to research participation?

○ Yes ◉ No

If **yes**, attach an **AE Summary Log** at the end of this form.

Is your study sponsor *requiring* that you forward any Study Sponsor **external** (off-site) Safety Reports *that are not otherwise reportable* under CHR guidelines:

○ Yes ◉ No

6.0 Updated Financial Disclosure

6.1 * Are there any changes in any financial interests related to this study or in any conflicts of interest of the PI or any other investigator:

○ Yes ◉ No

If **Yes**, the Conflict of Interest Advisory Committee (COIAC) office may contact you for additional information.

7.0 New Modifications

7.1 This modification is being submitted as a result of an adverse event report (AER), protocol violation or incident report, or publication of a new Investigator's Brochure (IB) or other safety data:

○ Yes ◉ No

Type of event:

○ Adverse event report
○ Protocol violation or incident report
○ New Investigator's Brochure
○ Other safety data

If **YES,** what date was the event submitted into the system on:

7.2 Proposed modifications:

The following modification was done in the revised application:
1. Collection of 15 cervical tissues was canceled and therefore sample size was reduced in the sections 15.1, 15.2 and 15.3 as follow:
15.1- 292
15.2- 292
15.3- 335

2. All related text to collection of cervical tissues were deleted in sections 12, 15 and 18.

3. Collection of saliva samples was canceled in previous renewal, however, the concent forms were approved. In this submission we did not attached the consent forms for saliva collection.

4. In the previous renewal we missed to attach the approved consent form for collection of oral (buccal) biopsies from HIV-negative donors, and in this renewal we attahced the missing consent form. We did not collect the buccal biopsies during last 1 year.

7.3 * This modification includes a change to enrollment targets:

◉ Yes ○ No

If **Yes**, be sure to update the subject numbers in the application (Section Samples Size and Eligibility).

7.4 Explain why these modifications are being made:

1. Collection of 15 cervical tissues was canceled as we do not study cervical tissues in our new R01 grant.

2. Due to canceling of cervical tissue collection all related text to cervical tissues were deleted in sections 12, 15 and 18.

3. Collection of saliva samples was canceled in previous renewal as we are completed this study. However, the concent forms were not deleted. In this submission we did not attached the consent forms for saliva collection.

4. In the previous renewal we missed to attach approved the consent form for collection of oral (buccal) biopsies from HIV-negative donors, and in this renewal we attahced the missing approved and unchanged consent form. We did not collect the buccal biopsies during last 1 year period.

7.5 * Does this modification require a change to the application form:

(•) Yes ( ) No

8.0 Revisions to the Application Form

8.1 Click the bar below to make revisions to the application form:

(Note: you are seeing this section because you either indicated that there are changes that affect the application or there are personnel changes that need to be made in the application.)

Edit/ View	Version	Title
	1.6	Study Application (Version 1.6) - Attached

9.0 Attach Consent Documents

9.1 Attach all consent documents you need approved with this continuing review: When possible, attach Word documents instead of PDFs.

Attach your consent forms following these instructions:

Approved Consent Documents with No Changes: All approved consent documents that you will continue to use must be attached. Click Select or Revise Existing. *If you have the currently approved consent form saved on your computer,* click Upload the Revised Consent. *If you need to download the approved consent form first,* click Download Document for Editing and then Upload the Revised Consent. Save your work. DO NOT SELECT THE CURRENTLY APPROVED VERSION OF THE CONSENT FORM. The CHR cannot stamp over last year's approval stamp and the submission will be returned to you.

Approved Consent Documents with Revisions: Click the Select or Revise Existing button. Click Upload the Revised Consent form and select the revised document from your computer. *If you need to download the current version of the consent form from iRIS first,* click Download Document for Editing and then Upload the Revised Consent after you've updated the document. Save your work.

For more information, click on the Help icon and read the "Revising Documents and Forms" quick guide.

New Consent Documents: Click on the Add a New Consent button and upload your new consent form.

Version	Sponsor Version	Title	Category	Language	Expiration Date	Consent Outcome	View Document

1.3.3		tonsil tissues		English	06/04 /2015	Void	265.89 KB
1.7		Consent-oral biopsy-HIV+		English	06/04 /2015	Void	202.82 KB
1.6		Consent-oral biopsy-HIV-		English	06/04 /2015	Void	203.40 KB
1.3		Consent-blood-HIV+		English	06/04 /2015	Void	168.83 KB
1.3		Consent-blood-HIV-		English	06/04 /2015	Void	162.63 KB
1.4		Consent breast milk-		English	06/04 /2015	Void	157.58 KB

9.2 Indicate which type(s) of consent documents are attached:

- ☐ New consent documents
- ☑ Active consent documents with NO changes
- ☐ Active consent documents with revisions
- ☐ No consent documents attached (explain below)

If no consent documents are attached, explain why not:

10.0 Attach Other Study Documents (AE Summary Log, revised documents, or new documents)

10.1 Upload AE Summary Log and/or any new or revised other documents here:

Attach the documents following these instructions:

Revised Documents: Follow these steps if you are revising currently approved documents or if you are submitting revised documents created by the study sponsor or lead site (e.g. revised protocols or investigator's brochures).

Click the Select or Revise Existing button. Click Upload the Revised Document and select the revised document from your computer. *If you need to download the current version of the document from iRIS first,* click Download Document for Editing and then Upload the Revised Document after you've updated the document. Save your work.

New Documents: Click on the Add Document button and upload your new document.

Reminder: If you are revising or adding new study documents, indicate that the continuing review includes modifications in question 1.6.

Approved Documents – No Changes: These documents do **not** need to be resubmitted during the continuing review.

Version	Sponsor Version	Title	Category	Expiration Date	Document Outcome	View Document
No Document(s) have been attached to this form.						

Continuing Review Submission Form (Version 4.0)

1.0

Continuing Review Form

To start with a copy of the last continuing review form (only if there is one):
1) Open the study through My Studies.
2) From the Study Management dashboard page, click the Continuing Review form link, check the box next to last year's form and click "Copy Form."
3) After you submit the form, click on the Continuing Review Due task on your home screen to remove the notification.
Please review the Quick Guide titled "Submitting Post-Approval Forms" in the system "Help" area for step-by-step instructions and screen shots.

1.1 Principal Investigator:

[redacted]

1.2 Study Title:

HIV, EBV and HPV interaction with mucosal epithelium

1.3 Study Number:

10-03277

1.4 Expiration Date:

06/04/2014

*If the study has expired, an explanation is required at the end of the form.

1.5 Lay Summary:

Interaction of HIV and HIV-associated opportunistic viruses, including EBV and HPV with oral and genital mucosal and GI tract mucosal epithelia is not well understood. Therefore, our goal is to establish an ex vivo organ culture model from oral, genital and GI tract mucosal epithelia, to study the molecular mechanisms of HIV, EBV and HPV mucosal transmission.

1.6 * This is a:

- ○ Continuing Review Only—no changes from last approval
- ◉ Continuing Review and Minor Modification
- ○ Continuing Review and Major Modification

1.7 * This application includes personnel changes:

- ○ Yes ○ No

1.8 * Has your study expired:

○ Yes ◉ No

If **Yes**, work on this study must stop **IMMEDIATELY.**

1.9 Outstanding Stipulations:

No Stipulation is outstanding.

2.0 Study Status

2.1 * Have any new risks been identified since the last continuing review (or since approval if this is the first continuing review):

○ Yes ◉ No

2.2 Current study status: (*choose one only*)

Study is open to accrual:

- ○ No participants have been enrolled and no additional risks have been identified.
- ○ Participants are currently receiving study intervention.
- ◉ Participants have been enrolled but none are currently receiving study intervention.
- ○ Ongoing medical record review/biological specimen analysis.

Study is closed to accrual:

- ○ Some participants are still receiving study intervention.
- ○ Study intervention is complete for all participants; research-related diagnostic tests or follow-up clinic visits are continuing.
- ○ Study intervention is complete or there was no intervention, and there is ongoing research-related follow-up contact with participants via questionnaires, phones calls, interviews, or mailings.
- ○ Study intervention is complete or there was no intervention and follow-up is limited to review of medical records or other records (no ongoing contact).
- ○ Study is in data analysis phase only.

2.3 Summary of results to date:

We have established polarized tonsil epithelial cells and polarized oriented tonsil tissue explants from children under 5 years old. Polarized cells and tissues were preincubated with chlorpromazine, nystatin, or amiloride, which are inhibitors of clathrin- and caveolin-mediated endocytosis and macropinocytosis, respectively. Polarized tonsil cells and tissues were also pretreated with antibodies against transmembrane protein T-cell immunoglobulin and mucin domain 1 (TIM-1), which is receptor for HIV-associated phosphatidylserine (PS). Then, dual tropic HIV-1_{SF33} was added to the apical surface of polarized cells and, after 4 h and 8 h, viral transmigration i.e., transcytosis was examined in the basolateral compartment by detecting HIV p24 using ELISA assay. HIV transmigration via mucosal tissue was evaluated by confocal immunofluorescence assay.

Analysis of HIV transmigration by inhibitors of clathrin- and caveolin-mediated endocytosis and macropinocytosis via polarized tonsil epithelial cells and tonsil tissue explants showed substantial

macropinocytosis in HIV transepithelial migration. We hypothesize that HIV envelope-associated PS may play a role in induction of macropinocytosis of virions. Tonsil epithelial cells expressed PS receptor TIM-1. Preincubation of tonsil epithelium with antibodies against TIM-1 substantially reduced (≈60%) HIV transcytosis, suggesting that interaction of virion-associated PS with TIM-1 of tonsil epithelium may lead to HIV macropinocytosis from the apical membranes of tonsil epithelial cells. Some tonsil epithelial cells containing HIV virions were stained with fluorescien labeled Sambucus nigra, a plant lectin that binds to M (microfold) cells, suggesting that tonsillar M cells may facilitate HIV transcytosis via tonsil mucosal epithelium.

Our data show that HIV macropinocytosis/transcytosis through infant tonsil epithelium may play a critical role in HIV MTCT. Interaction of HIV virion associated PS with TIM-1 may facilitate viral macropinocytosis/transcytosis. Interference of HIV-PS interaction with TIM-1 may substantially reduce the risk of HIV transmission via infant tonsil epithelium.

2.4 Brief summary of plans for the coming year:

For the coming year, we will begin to perform proposed experiments for Specific Aim 2. In this aim, we hypothesize that HIV-infected macrophages attach to the mucosal surfaces of fetal/infant oral and fetal intestinal epithelia by adhesion of LFA-1 of macrophages to ICAM-1 of epithelial cells. To test this hypothesis, we will study the role of LFA-1 and ICAM-1 in binding of HIV-infected macrophages to infant oral and intestinal mucosal epithelia. R5-tropic HIV-1_{SF170}-infected macrophages will be propagated and labeled with carboxyfluorescein diacetate succinimidyl ester (CFSE). Macrophages will then be incubated with function-blocking antibodies to LFA-1. In parallel experiments polarized oriented infant tonsil and fetal oropharyngeal and intestinal explants will be incubated with function-blocking antibodies to ICAM-1. Macrophages will then be added to the apical surfaces of tissue explants. Uninfected macrophages and explants without antibodies (or with isotype control antibodies) will serve as controls. At 4 h and 8 h, tissues will be fixed, sectioned and co-immunostained for HIV p24 (red) and pan-keratin (blue). CFSE-labeled HIV-positive macrophages (green) bound to the apical surface of the epithelium will be quantitatively analyzed. We expect that attachment of HIV-infected macrophages will be mediated by macrophage LFA-1 and epithelial ICAM-1.

3.0 Recent Literature

3.1 Summary of recent literature:

1. Tugizov, S.M., *et al.* Differential transmission of HIV traversing fetal oral /intestinal epithelia and adult oral epithelia. *Journal of Virology* **86**, 2556-2570 (2012).
2. Tugizov, S.M., *et al.* HIV is inactivated after transepithelial migration via adult oral epithelial cells but not fetal epithelial cells. *Virology* **409**, 211-222 (2011).
3. Mundy, D.C., Schinazi, R.F., Gerber, A.R., Nahmias, A.J. & Randall, H.W., Jr. Human immunodeficiency virus isolated from amniotic fluid. *Lancet* **2**, 459-460 (1987).
4. Jaspan, H.B., Robinson, J.E., Amedee, A.M., Van Dyke, R.B. & Garry, R.F. Amniotic fluid has higher relative levels of lentivirus-specific antibodies than plasma and can contain neutralizing antibodies. *J Clin Virol* **31**, 190-197 (2004).
5. Maiques, V., Garcia-Tejedor, A., Perales, A., Cordoba, J. & Esteban, R.J. HIV detection in amniotic fluid samples. Amniocentesis can be performed in HIV pregnant women? *Eur J Obstet Gynecol Reprod Biol* **108**, 137-141 (2003).
6. Willumsen, J.F., *et al.* Subclinical mastitis as a risk factor for mother-infant HIV transmission. *Adv Exp Med Biol* **478**, 211-223 (2000).
7. Nussenblatt, V., *et al.* Epidemiology and microbiology of subclinical mastitis among HIV-infected women in Malawi. *Int J STD AIDS* **16**, 227-232 (2005).
8. Semba, R.D. Mastitis and transmission of human immunodeficiency virus through breast milk. *Ann N Y Acad Sci* **918**, 156-162 (2000).
9. Semba, R.D. & Neville, M.C. Breast-feeding, mastitis, and HIV transmission: nutritional implications. *Nutr Rev* **57**, 146-153 (1999).
10. Bomsel, M. Transcytosis of infectious human immunodeficiency virus across a tight human epithelial cell line barrier. *Nat Med* **3**, 42-47 (1997).
11. Hocini, H. & Bomsel, M. Infectious human immunodeficiency virus can rapidly penetrate a tight human epithelial barrier by transcytosis in a process impaired by mucosal immunoglobulins. *Journal of Infectious Diseases* **179 Suppl 3**, S448-453 (1999).

4.0 Subject Accrual

4.1 Recruitment and enrollment activities:

New subjects enrolled since last continuing review

Locally:

11

For study overall:

11

Total subjects enrolled to date

Locally:

173

For study overall:

173

Total number of subjects that will be enrolled as specified in the Sample Size and Eligibility section of the CHR Application

Locally:

292

For study overall:

292

4.2 Did you exceed your approved enrollment numbers:

○ Yes ◉ No

Explain how this happened and what will be done to prevent this in the future:

4.3 Total refusals (individuals asked to be in the study but who chose not to participate) to date:

Locally:

none

4.4 Total withdrawals:

Locally:

none

Summarize the main reasons why subjects withdrew:

5.0 Significant Findings & Other Reportable Events

5.1 Did you report any 10-day reportable events, including complaints about the research, since the last continuing review (or since initial approval if this is the first continuing review):

○ Yes ◉ No

If **Yes,** provide a brief narrative summary of adverse events, protocol violations, safety information, and/or complaints over the past year so that CHR members can understand the overall trends:

5.2 Are you submitting any new or missed 10-day reportable events, including complaints, at this time (submit separately in system using the appropriate submission form):

○ Yes ◉ No

5.3 Plan for informing subjects of information that may affect willingness to continue participation:

If new risks have been identified since the last continuing review (or since initial approval if this is the first continuing review), explain how subjects were or will be informed of any new information and when the modification was approved if the notification already occurred:

Attach any additional documents (consent forms or contact letters) that will be used for this purpose at the end of the form.

5.4 * Does this study undergo formal on-site monitoring:

○ Yes ◉ No

5.5 * This study was audited by any external group or entity (i.e., sponsor, FDA) since its last renewal:

○ Yes ◉ No

Auditing entity:

5.6 If YES to either monitoring or auditing, describe any significant findings resulting from either the monitoring or auditing activities or state "None" if there were no significant findings:

5.7 Other reportable events:

If this is a biomedical **interventional study** did you have any **internal** (on-site) participant deaths determined to be *unrelated* to research participation?

○ Yes ◉ No

If **yes**, attach an **AE Summary Log** at the end of this form.

Is your study sponsor *requiring* that you forward any Study Sponsor **external** (off-site) Safety Reports *that are not otherwise reportable* under CHR guidelines:

○ Yes ◉ No

6.0 Updated Financial Disclosure

6.1 * Are there any changes in any financial interests related to this study or in any conflicts of interest of the PI or any other investigator:

○ Yes ◉ No

If **Yes**, the Conflict of Interest Advisory Committee (COIAC) office may contact you for additional information.

7.0 New Modifications

7.1 This modification is being submitted as a result of an adverse event report (AER), protocol violation or incident report, or publication of a new Investigator's Brochure (IB) or other safety data:

○ Yes ◉ No

Type of event:

○ Adverse event report
○ Protocol violation or incident report
○ New Investigator's Brochure
○ Other safety data

If **YES,** what date was the event submitted into the system on:

7.2 Proposed modifications:

The following modification was done in the revised application:
1. Collection of 15 cervical tissues was canceled and therefore sample size was reduced in the sections 15.1, 15.2 and 15.3 as follow:
15.1- 292
15.2- 292
15.3- 335

2. All related text to collection of cervical tissues were deleted in sections 12, 15 and 18.

3. Collection of saliva samples was canceled in previous renewal, however, the concent forms were approved. In this submission we did not attached the consent forms for saliva collection.

4. In the previous renewal we missed to attach the approved consent form for collection of oral (buccal) biopsies from HIV-negative donors, and in this renewal we attahced the missing consent form. We did not collect the buccal biopsies during last 1 year.

7.3 * This modification includes a change to enrollment targets:

◉ Yes ○ No

If **Yes**, be sure to update the subject numbers in the application (Section Samples Size and Eligibility).

7.4 Explain why these modifications are being made:

1. Collection of 15 cervical tissues was canceled as we do not study cervical tissues in our new R01 grant.

2. Due to canceling of cervical tissue collection all related text to cervical tissues were deleted in sections 12, 15 and 18.

3. Collection of saliva samples was canceled in previous renewal as we are completed this study. However, the concent forms were not deleted. In this submission we did not attached the consent forms for saliva collection.

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7.5 * Does this modification require a change to the application form:

(•) Yes ( ) No

8.0 Revisions to the Application Form

8.1 Click the bar below to make revisions to the application form:

(Note: you are seeing this section because you either indicated that there are changes that affect the application or there are personnel changes that need to be made in the application.)

Edit/ View	Version	Title
	1.6	Study Application (Version 1.6) - Attached

9.0 Attach Consent Documents

9.1 Attach all consent documents you need approved with this continuing review: When possible, attach Word documents instead of PDFs.

Attach your consent forms following these instructions:

Approved Consent Documents with No Changes: All approved consent documents that you will continue to use must be attached. Click Select or Revise Existing. *If you have the currently approved consent form saved on your computer,* click Upload the Revised Consent. *If you need to download the approved consent form first,* click Download Document for Editing and then Upload the Revised Consent. Save your work. DO NOT SELECT THE CURRENTLY APPROVED VERSION OF THE CONSENT FORM. The CHR cannot stamp over last year's approval stamp and the submission will be returned to you.

Approved Consent Documents with Revisions: Click the Select or Revise Existing button. Click Upload the Revised Consent form and select the revised document from your computer. *If you need to download the current version of the consent form from iRIS first,* click Download Document for Editing and then Upload the Revised Consent after you've updated the document. Save your work.

For more information, click on the Help icon and read the "Revising Documents and Forms" quick guide.

New Consent Documents: Click on the Add a New Consent button and upload your new consent form.

Version	Sponsor Version	Title	Category	Language	Expiration Date	Consent Outcome	View Document

1.3.3		tonsil tissues		English	06/04 /2015	Void	265.89 KB
1.7		Consent-oral biopsy-HIV+		English	06/04 /2015	Void	202.82 KB
1.6		Consent-oral biopsy-HIV-		English	06/04 /2015	Void	203.40 KB
1.3		Consent-blood-HIV+		English	06/04 /2015	Void	168.83 KB
1.3		Consent-blood-HIV-		English	06/04 /2015	Void	162.63 KB
1.4		Consent breast milk-		English	06/04 /2015	Void	157.58 KB

9.2 Indicate which type(s) of consent documents are attached:

- ☐ New consent documents
- ☑ Active consent documents with NO changes
- ☐ Active consent documents with revisions
- ☐ No consent documents attached (explain below)

If no consent documents are attached, explain why not:

10.0 Attach Other Study Documents (AE Summary Log, revised documents, or new documents)

10.1 Upload AE Summary Log and/or any new or revised other documents here:

Attach the documents following these instructions:

Revised Documents: Follow these steps if you are revising currently approved documents or if you are submitting revised documents created by the study sponsor or lead site (e.g. revised protocols or investigator's brochures).

Click the Select or Revise Existing button. Click Upload the Revised Document and select the revised document from your computer. *If you need to download the current version of the document from iRIS first,* click Download Document for Editing and then Upload the Revised Document after you've updated the document. Save your work.

New Documents: Click on the Add Document button and upload your new document.

Reminder: If you are revising or adding new study documents, indicate that the continuing review includes modifications in question 1.6.

Approved Documents – No Changes: These documents do **not** need to be resubmitted during the continuing review.

Version	Sponsor Version	Title	Category	Expiration Date	Document Outcome	View Document
No Document(s) have been attached to this form.						

Study Application (Version 1.7)

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]

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) at UCSF and affiliated sites ONLY by clicking the "Add a new row" button: NOTE: This information is required and your application will be considered incomplete without it.

[redacted]

5.0 Initial Screening Questions

5.1 * This study involves human stem cells (including iPS cells and adult 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 (e.g. buccal swabs, urine, hair and nail clippings, etc.)
- ☐ Category 4: Noninvasive clinical procedures (e.g. physical sensors such as pulse oximeters, MRI, EKG, EEG, ultrasound, moderate exercise testing, etc.)
- ☐ Category 5: Research involving materials (data, documents, records, or specimens) that were previously collected for either nonresearch or research purposes
- ☐ Category 6: Use of recordings (voice, video, digital or image)
- ☐ Category 7: Low risk behavioral research or research employing survey, interview, oral history, focus group, program evaluation, human factors evaluation, or quality assurance methodologies
- ☐ Category 8: Continuing review of previously approved full committee research that is essentially complete
- ☑ Category 9: Continuing review of research NOT involving an IND or IDE where the IRB has determined that the research poses no greater than minimal risk

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 sponsors and provide the funding details:

External Sponsor:

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	P0037598	

Sponsor Name:	California HIV/AIDS Research Program
Sponsor Type:	14
Sponsor Role:	Funding
CFDA Number:	
Grant/Contract Number:	

Awardee Institution::	UCSF
Is Institution the Primary Grant Holder:	Yes
Contract Type:	Grant
Project Number:	P0037598
UCSF RAS System Award Number ("A" + 6 digits):	
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		A121266

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):	A121266
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:	

Gift, Program, or Internal Funding (check all that apply):

- ☐ Funded by gift (specify source below)
- ☐ Funded by UCSF or UC-wide program (specify source below)
- ☐ Specific departmental funding (specify source below, if applicable)
- ☐ Unfunded (miscellaneous departmental funding)
- ☐ Unfunded student project

List the gift, program, or departmental funding source:

7.2 If you tried to add a sponsor in the question above and it was not in the list, follow these steps:

- **If funding has already been awarded or the contract is being processed by the Contracts and Grants or Industry Contracts unit, your sponsor is already in the system and the project has a UCSF RAS System Proposal or Award number. Check with your department's Research Services Analyst (RSA) to ask how the sponsor is listed in the UC sponsor list and what the Proposal or Award number is.**
- **If you need additional assistance, contact the Contracts and Grants Award Team at CGAwardTeam@ucsf.edu and list the sponsor in the box below.**

○ Sponsor not in list

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

If the funding is administered by the UCSF Office of Sponsored Research, your study will not receive CHR approval until the sponsor and funding details have been added to your application.

7.3 * This study is supported in whole or in part by Federal funding:

◉ Yes ○ No

If **yes**, indicate which portion of your grant you will be attaching:

- ☑ The Research Plan, including the Human Subjects Section of your NIH grant or subcontract
- ☐ 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
- ☐ The grant is not attached. The study is funded by an award that does not describe specific plans for human subjects, such as career development awards (K awards), cooperative agreements, program projects, and training grants (T32 awards)

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):

- ☑ UCSF
- ☐ Mt. Zion
- ☑ San Francisco General Hospital (SFGH)
- ☐ SF VA Medical Center (SF VAMC)
- ☐ Helen Diller Family Comprehensive Cancer Center
- ☐ Blood Centers of the Pacific (BCP)
- ☐ Blood Systems Research Institute (BSRI)

- [ ] Gallo
- [ ] Gladstone
- [ ] Institute on Aging (IOA)
- [ ] Jewish Home
- [ ] 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:

- [ ] Other UC Campus
- [x] Other institution
- [ ] Other community-based site
- [ ] Foreign Country

List the foreign country/ies:

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 Studies Involving Other Sites

10.1 UCSF is the coordinating center:

- ( ) Yes
- (•) No

If **Yes**, describe the plan for communicating safety updates, interim results, and other information that may impact risks to the subject or others among sites:

If **Yes**, describe the plan for sharing modification(s) to the protocol or consent document(s) among sites:

10.2 Check any other UC campuses with which you are collaborating on this research study:

- [ ] UC Berkeley
- [ ] UC Davis
- [ ] Lawrence Berkeley National Laboratory (LBNL)
- [ ] UC Irvine
- [ ] UC Los Angeles
- [ ] UC Merced
- [ ] UC Riverside

- [ ] UC Santa Barbara
- [ ] UC Santa Cruz

10.3 Are the above UC campuses requesting to rely on UCSF's IRB (check all that apply)?

- [ ] Yes (Attach the Notice of Intent to Rely on One UC IRB form in the Other Study Documents section)
- [ ] No (Complete IRB Approval Certification section)

11.0 IRB Approval Certification

(Note: This section replaces the old IRB Approval Certification Supplement form. Please do not attach the old form(s) to this application.)

11.1 IRB Approval Certification Sub-Form

Click "Add a new row" to enter information for a site. Click it again to add a second site and againand again for a third, a fourth, etc.

Outside Site Information

Non-UCSF affiliated site information:

Site name:

Cooperative Human Tissue Network (CHTN) ;http://www.chtn.nci.nih.gov/what-is/

Contact name:

[redacted]

Email:

[redacted]

Phone:

[redacted]

For Federally-funded studies only, corresponding FWA#:

*** The research at this site will be reviewed by:**

- ( ) The non-affiliated site's IRB or a private IRB
- ( ) The non-affiliated site is requesting UCSF to be the IRB of record for this study
- (•) The non-affiliated site is not engaged in the human subjects research and has provided a letter of support

If the other site's IRB approval letter is available now, attach it to

submit it once you receive it.

Or, if the other site is **not engaged** in human subjects research, attach the letter of support to your application.

12.0 Study Design

12.1 * 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/infant 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/infant 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 breast milk, however, their role in HIV infection with fetal/neonatal mucosal epithelium in not clear. Therefore we will investigate role of fetal/infant mucosal epithelium in HIV transmission, as well as role of breast milk in HIV transmission via fetal/infant oral mucosal epithelium. We have shown that HIV infection may disrupt mucosal epithelium leading paracellular penetration of Epstein-Barr virus (EBV) and human paplillomavirus (HPV). Uisng biopsy tissues from HIV-infected donors we will study the paracellular penetration of EBV and HPV.

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.

12.2 Check all that apply:

- ☐ Phase I
- ☐ Phase II
- ☐ Phase III
- ☐ Phase IV

13.0 Scientific Considerations

13.1 Hypothesis:

This study has a hypothesis:

◉ Yes ○ No

If yes, state the hypothesis or hypotheses:

Hypothesis: The HIV envelope contains PS, and interaction of virus-associated PS with the PS receptor TIM-1 at the fetal/infant oral and intestinal mucosal surfaces induces viral macropinocytosis. Interaction of HIV with heparan sulfate proteoglycan (HSPG) and

Both macropinocytosis and endocytosis of HIV facilitate transport of virions into intracellular vesicular/endosomal compartments, which subsequently deliver virus to basolateral membranes by transcytosis.

Hypothesis: HIV-infected monocytes/macrophages attach to the mucosal surfaces of fetal/infant oral and intestinal epithelia by binding of LFA-1 of monocytes/macrophages to ICAM-1 of oral epithelial cells. Secretion of TNF-a from HIV-infected monocytes/macrophages activates MCP-1 in oral/intestinal epithelium, facilitating migration of HIV-infected monocytes/macrophages into the epithelium.

Hypothesis: HBD-2 and/or -3 binds to HSPG, which mediates HBD binding to HIV gp120, leading to inactivation of HIV within the vesicular/endosomal compartments of fetal/infant oral and intestinal epithelial cells. Recombinant HBD-2 and -3 containing protein trunsduction signals are efficiently taken up into infant oral and intestinal epithelia, and spread of HBDs within mucosal epithelial tissues inhibits or substatially reduces HIV MTCT.

Hypothesis: In HIV infected individulas the tight junctions of mucosal epithelium disrupted, leading to paracellular penetration of EBV and HPV into epithelium.

13.2 * List the specific aims:

Aim 1. Establish and characterize an ex vivo organ culture model from the oral of the adult, and oropharyngeal, and intestinal mucosal epithelium of the fetus, and tonsil epithelium of infants.
Aim 2. Analyze expression of HBD1, HBD2, HBD3 and SLPI in adult, fetal and infant epithelia.
Aim 3. Study mechanisms of HIV spread via the oral of the adult, and oropharyngeal and intestinal mucosal epithelium of the fetus and tonsil epithelium of infant.
Aim 4. Determine the role of breast milk in the interaction of HIV with the mucosal epithelium of the fetus and infant.
Aim 5. Study the mechanisms of HIV-mediated tigh junction disruption and its role in HPV and EBV paracellular penetration.

13.3 Statistical analysis:

Data will be analyzed by a study steering committee, which will be composed by [redacted] and [redacted] It is estimated that 30 adult, 50 fetal and 50 infant tissues will be investigated for this study. Fifteen breast milk from HIV-negative woman will be analyzed. A five years cumulative enrollment is planned.
HIV-1 infection of the mucosal epithelium and breast milk will be evaluated in the presence of HIV by immunostaining and ELISA assays, respectively. At least five samples will be used for each experiment. The statistical analysis will account for the paired nature of the data. 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.

13.4 * This is an investigator-initiated study:

(•) Yes ( ) No

13.5 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**:

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

14.0 Background

14.1 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. examined HIV in the amniotic fluid of 366 pregnant women, and HIV was detected in 17% of the 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 HIV may disrupt epithelial tight junctions leading to facilitate EBV and HIV paracellular penetration. Therefore we will study mechanisms of HIV, EBV and HPV dissemination within the adult oral mucosal epithelium.

14.2 Preliminary studies:

Our preliminary results show the following: (ii) HIV via mucosal epithelium is initiated by macropinocytosis and clathrin- and caveolin-mediated endocytosis. (iii) The HIV envelope contains PS, and infant oral epithelium expresses the PS receptor TIM-1, which may play a critical role in viral macropinocytosis. (iv) Infant oral epithelium expresses ICAM-1, which may bind macrophage LFA-1, facilitating binding of HIV-infected macrophages to mucosal surfaces. TNF-a induces MCP-1 expression in infant tonsil keratinocytes, which may initiate migration of HIV-infected monocytes/macrophages into epithelium. (v) Infant oral epithelial cells do not express the anti-HIV innate proteins HBD-2 and -3. Recombinant HBD-2 and/or -3 internalized into polarized infant oral cells bind indirectly to HIV gp120 and inactivate virus during its transcytosis. HIV tat containing a PTD signal rapidly penetrates into oral epithelium, suggesting that generation of HBDs containing a PTD signal could facilitate their penetration into epithelium. (vi) Infant tonsil epithelium contains M cells, which may play an important role in HIV transcytosis and MTCT.

14.3 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

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.

15.0 Sample Size and Eligibility

15.1 Number of subjects that will be enrolled at UCSF and affiliated institutions (locally):

292

15.2 Total number of subjects that will be enrolled at all sites (for study overall):

292

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

235

15.4 Sample size calculation:

This is an ongoing project and the first CHR application was submitted in 2007 for NIH R01 and UCSF intramural grants with the initial sample size of 586. However, the NIH R01 was not funded and we were not able to perform some studues. Therefore, so far we have colleced 162 samples, we are currently analyzing data from these samples.

We are continuing this project under new NIH R01 grant. For this study will be used 50 tonsil tissues from young children (between 0.5 to 5 years old). We also will use 50 fetal tissue explants (25 oropharyngeal and 25 intestinal), which will be collected from aborted (discarded) fetuses between 18 and 24 weeks of gestation. Next, we will use 15 oral (buccal) biopsies from adult donors (18+ years old). Finally, we will collect breast milk from 15 donors. Will be propagated small tissues explants from each tissue, and each experiment will be performed using a set of

independent donors. HIV transmission through mucosal epithelium will be examined by applying of virus with and without breast milk from the surface of epithelium and detection of virus in the lamina propria.

Total sample size = 292, estimated sample szie= 335
162 samples are already collected and currently under investigation

Anticipated sample size for next 5 years:
Tonsil tissues from tonsillectomy/surjical material= 50
Fetal tissues from discarded fetuses= 50
Oral (buccal) biopsy =15
Breasr milk= 15

We will use total of 50 tonsil tissues from children, 50 tissues from fetus (25 oropharyngeal and 25 intestinal), 15 oral tissues and 15 breast milk samples from adults. These numbers have chosen based on nature of treatment and controls.

15.5 * Eligible age range(s):

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

15.6 Inclusion criteria:

The only inclusion criterion will be willingness to donate oral tissues, and breast milk samples.

15.7 Exclusion criteria:

The only exclusion criteria would be inability to provide informed consent and having a contraindication for a biopsy procedure, e.g., bleeding diathesis or infection and inflammation.

15.8 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:

16.0 Drugs and Devices

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

- ( ) Yes
- (•) No

16.2 * Investigational medical devices or in vitro diagnostics will be used OR approved medical devices or in vitro diagnostics will be studied under this application:

- ( ) Yes
- (•) No

16.3 * A Non-Significant Risk (NSR) determination is being requested for an investigational device:

○ Yes ◉ No

16.4 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.

17.0 Other Approvals and Registrations

17.1 * This is a clinical trial:

○ Yes ◉ No

Clinical Trial Registration
"NCT" number for this trial:

17.2 * Data from this study will be submitted to NIH for Genome-Wide Association Studies (GWAS):

○ Yes ◉ No

17.3 * This study involves vaccines produced using recombinant DNA technologies:

○ Yes ◉ No

17.4 * This study involves human gene transfer (NOTE: Requires NIH Recombinant DNA Advisory Committee (RAC) review prior to CHR approval):

○ Yes ◉ No

17.5 * The study protocol requires radiological procedures (e.g. CT scans, x-rays) or exposes subjects to radiation:

○ Yes ◉ No

17.6 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

18.0 Procedures

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

1. Collection of adult oral tissue and establishment of tissue explants ex vivo.
The buccal explants will be obtained from HIV-positive and –negative adult volunteers. 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 mucosa containing epithelium and connective tissue will be obtained using 4-mm–diameter biopsy punches in [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.

For verification of HIV-negative status 20 ml of blood sample will be taken from arm vein of each donor. HIV test will be performed by HIV diagnostic lab at [redacted] If HIV test shows positive results, [redacted] will contact donor by phone to discuss this issue and will refer for treatment and care. [redacted] will inform HIV positive result by name of donor to the Department of Public Health and tissue of HIV-positive donor will be removed from the experiments.

2. Collection of fetal oropharyngeal and intestinal tissue explants
Fetal ropharyngeal gastric and intestinal tissue explants 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 tissues 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 oropharyngeal area by surgical instrument. Similarly sized pieces will be dissected from the small 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 0.4 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 tonsil tissue samples from 0.5-5 years old children.
Collection of discarded tonsil tissue samples from HIV-negative children under 5 years old after routine tonsillectomy. A small tissue samples with about 5 -5 cm will be collected from surgical materials after routine tonsillectomy. 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.

4. Collection of breast milk.
To examine the role of breast milk in HIV infection of fetal/infant mucosal epithelium we will collect breast milk. Fifty ml of breast milk from 15 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

5. Infection of tissue explants with HIV, HPV 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 HPV infection will be used HPV 16 pseudovirions (1 ng/per biopsy). 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 HPV-16 pseudovirions.

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

18.2 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.

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

○ Yes ◉ No

If yes, explain:

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

○ Yes ◉ No

If yes, explain:

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

◉ Yes ○ No

18.6 Time commitment (per visit and in total):

1. For obtaining one oral biopsy sample per visit will be required about 30 min.
2. For obtaining one tonsil sample per visit will be required about 1 h.
3.The time to obtaining one breast milk sample will be about 10 min.

18.7 Locations:

1.The oral biopsy tissues from the adult buccal mucosa from normal healthy individuals and HIV-positive patients will be collected in [redacted] Contact person: [redacted]
2. The discarded fetal oropharyngeal and intestinal tissue samples from discarded fetus will be collected in [redacted]
3. The discarded infant tonsil tissues will be collected in [redacted] Contact person, [redacted]

4. The breast milk samples will be collected in breastfeeding healthy HIV-negative women at [redacted] Contact person: [redacted]
5. All HIV and/or EBV and/or HPV infection procedures with tissue samples in presence or absence of breast milk will be performed in [redacted] Contact person, [redacted]

18.8 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 tissue biopsy 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

19.0 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.)

19.1 Specimens are (check all that apply):

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

If Other, explain:

19.2 Types of specimens:

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

Describe and/or name source:

Oral biopsy tissues and breast milk will be collected

19.3 Consent will be obtained via:

- [ ] Separate specimen banking consent form
- [x] Specimen banking section within a main research study consent form
- [ ] Surgical consent form with tissue donation brochure

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

- [x] 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:

19.5 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:

20.0 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.)

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

[redacted]

20.2 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.

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:

20.3 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)

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

○ Yes ◉ No

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

20.5 There is a formal specimen utilization review process:

○ Yes ◉ No

If **Yes**, describe the process:

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

- ☑ UCSF researchers
- ☐ Non-UCSF researchers
- ☐ Industry

21.0 Alternatives

21.1 Study drug or treatment is available off-study:

- ○ Yes
- ◉ No
- ○ Not applicable

21.2 * Is there a standard of care (SOC) or usual care that would be offered to prospective subjects at UCSF (or the study site) if they did not participate:

○ Yes ◉ No

If yes, describe the SOC or usual care that patients would receive if they choose not to participate:

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

21.3 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.

22.0 Risks and Benefits

22.1 * Risks and discomforts:

A. Risks and Discomforts:

Risk and discomforts for biopsy procedures of adult oral 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 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.

22.2 Steps taken to minimize risks to subjects:

To minimize the discomfort of the oral tissue biopsy 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.

22.3 Benefits to subjects:

○ Yes ◉ No

If yes, describe:

22.4 Benefits to society:

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

22.5 Explain why the risks to subjects are reasonable:

The study risks on an individual level will be minimal (potential loss of privacy). 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.

23.0 Data and Safety Monitoring Plan

23.1 Describe the plan for monitoring data and safety:

This is not an interventional trial and no data safety monitoring board is needed.

23.2 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.

23.3 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.

24.0 Confidentiality and Privacy

24.1 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.

24.2 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.

24.3 Study data are:

- ☐ Derived from the Integrated Data Repository (IDR) or The Health Record Data Service (THREDS) at SFGH
- ☑ 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 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:

[redacted]

24.4 Identifiers may be included in research records:

(•) Yes ( ) No

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

- [x] Names
- [ ] Dates
- [ ] Postal addresses
- [ ] Phone numbers
- [ ] Fax numbers
- [ ] Email addresses
- [ ] Social Security Numbers*
- [ ] Medical record numbers
- [ ] 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

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

( ) Yes (•) No

If **yes**, indicate to whom identifiable information may be disclosed:

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

If **Others**, specify:

24.6 Indicate how data are kept secure and protected from improper use and disclosure (check all that apply): NOTE: Whenever possible, do not store subject identifiers on laptops, PDAs, or other portable devices. If you collect subject identifiers on portable devices, you MUST encrypt the devices.

- [ ] Data are stored securely in My Research
- [x] 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
- [x] Data are kept in a locked office or suite
- [x] Electronic data are protected with a password
- [x] Data are stored on a secure network

24.7 Additional measures to assure confidentiality and protect identifiers from improper use and disclosure, if any:

All study numbers will be destroyed as soon as the needed data are transcribed.

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

○ Yes ◉ No

Explain:

HIV positive test results will be reported to the Department of Public Health.

24.9 This study will be issued a Certificate of Confidentiality:

○ Yes ◉ No

25.0 Subjects

25.1 Check all types of subjects that may be enrolled:

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

25.2 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:

For this study will be collected the tonsil tissue samples from children under 10 years old, which are undergo to routine tonsilectomy procedures. The surgical tissues are discarded materials. Since,
tonsilectomy procedures do not have any interventions and do not cause risk to children.

Also, will be collected oropharyngeal and intestinal tissues 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.

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:

26.0 Inclusion of Children in Research

(Note: This section replaces the old "Inclusion of Children and Minors in Research" supplement form. Please do not attach the old form to this application.)

26.1 This study will enroll children who can legally consent for themselves:

○ Yes ◉ No

If **yes,** explain why they can consent for themselves in the research setting:

If you will **ONLY** be enrolling children who can legally consent for themselves, press **SAVE and CONTINUE** to skip the rest of this section.

26.2 Select all the regulatory categories that apply:

- ☑ No greater than minimal risk (45 CFR 46.404, 21 CFR 50.51)
- ☐ Greater than minimal risk but presenting prospect of direct benefit (45 CFR 46.405, 21 CFR 50.52)
- ☐ Greater than minimal risk (though only a minor increase over minimal risk) and no prospect of direct benefit but likely to yield generalizable knowledge about the subjects disorder or condition (45 CFR 46.406, 21 CFR 50.53)
- ☐ Research not otherwise approvable which presents an opportunity to understand, prevent, or alleviate a serious problem affecting the health or welfare of children (45 CFR 46.407, 21 CFR 50.54)

Explain why the research in this study falls under the above category or categories:

For this study will be collected the tonsil tissue samples from children under 10 years old, which are undergo to routine tonsilectomy procedures. The surgical tissues are discarded materials. Since,
tonsilectomy procedures do not have any interventions and do not cause risk to children.

Also, will be collected oropharyngeal and intestinal tissues 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.

26.3 Parental permission or waiver:

- ☑ Parental permission will be obtained
- ☐ Waiver of parental permission is requested: Parental permission is not a reasonable requirement
- ☐ Waiver of parental permission is requested: The waiver meets the provisions for a waiver of consent set forth in 45 CFR 46.116, Subpart A

If you are requesting a **waiver of parental permission**, explain why the study meets the regulatory criteria for this waiver:

26.4 Assent of children or waiver:

- ☐ Assent of children old enough to provide assent will be obtained
- ☑ Waiver of assent is requested: Children cannot be consulted or the research has prospect of

direct benefit only available in the study

☐ Waiver of assent is requested: The waiver meets the provisions for a waiver of consent set forth in 45 CFR 46.116, Subpart A

If you are requesting a **waiver of child's assent**, explain why the study meets the regulatory criteria for this waiver:

Children under 10 years old are too young to be consulted.

26.5 Documentation of permission and assent (select all that will be used):

☑ Permission form addressed to the parents
☐ Simplified assent form addressed to the child, 7-12 years old (parents get separate form)
☐ Assent form addressed to the child, 13 years and older (for subjects and parents)
☐ Assent form addressed to the child, 13 years and older (parents get separate form)

Check one:

◉ One parent's signature will be obtained
○ Two parents' signatures will be obtained

If this study is approvable under .404 or .405 and you plan to get permission from only one parent, explain why you think one parent's permission is sufficient:

26.6 This study may enroll wards of the state:

○ Yes ◉ No

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

27.1 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:
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.

28.0 Recruitment

28.1 * Methods (check all that apply):

☑ 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

- [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:

28.2 * How, when, and by whom eligibility will be determined:

After obtaining of informed consent eligibility of donors for the oral biopsies will be determined by [redacted] and [redacted] respectively. HIV-negative donors should provide evidence of HIV negative test. Alternatevily, HIV negative status of donors will be verified by standard HIV lab testing in the [redacted] Twenty ml of blood sample will be taken from arm vein for HIV testing.

The tonsil tissues will be collected from discarded surgical materials. After obtaining of informed consent [redacted] will perform tonsilectomy from HIV-negative children and discarded tonsil tissues will be provided to [redacted] laboratory. In the lab [redacted] will verify HIV negative status of donors by tesing of tonsil lymphocytes for HIV using ELISA p24, immunofluorescence and and Western blot assays.

After obtaining of informed consent the breast milk will be collected by HIV-negative donors and provided to [redacted] will verify HIV negative status of breast milk by tesing it for using ELISA p24, immunofluorescence and and Western blot assays.and lymphocytes from tonsil tissue will be tested for HIV using ELISA p24 assay.

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

Advertisements will be placed seeking volunteers to donate oral mucosal tissue, and breast milk. The advertisements will be posted around the UCSF and San Francisco General campuses, in local newspapers and on the Internet. Volunteers donating oral tissues will be financially compensated for their time. Interested individuals will be asked to contact [redacted] and [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 biopsy tissues from the adult oral mucosa will be collected in [redacted]

2. The fetal oropharyngeal and intestinal tissue samples from discarded fetus will be collected [redacted]

3. The infant tonsil tissues will be collected from discarded surgical materailas in [redacted]

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

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

(•) Yes ( ) No

29.0 Waiver of Consent/Authorization for Recruitment Purposes

(Note: This section partially replaces the old "Request for Waiver of Consent/Authorization for Minimal Risk Research or for Screening for Recruitment" supplement form. Please do not attach the old form to this application.)

This section is now required when study investigators (and/or affiliated nurses or staff) recruit their own patients directly.

29.1 * Study personnel need to access protected health information (PHI) during the recruitment process and it is not practicable to obtain informed consent until potential subjects have been identified:

(•) Yes

If **no**, a waiver of consent/authorization is NOT needed.

29.2 * A waiver for screening of health records to identify potential subjects poses no more than minimal risk to privacy for participants:

(•) Yes

If **no**, a waiver of authorization can NOT be granted.

29.3 * Screening health records prior to obtaining consent will not adversely affect subjects' rights and welfare:

(•) Yes

If **no**, a waiver of authorization can NOT be granted.

29.4 * Check all the identifiers that will be collected prior to obtaining informed consent:

- [x] Names
- [x] Dates
- [ ] Postal addresses
- [ ] Phone numbers
- [ ] Fax numbers
- [ ] Email addresses
- [ ] Social Security Numbers*

Medical record numbers
- [ ] 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
- [ ] None

Note: HIPAA requires that you collect the minimum necessary.

29.5 * Describe any health information that will be collected prior to obtaining informed consent:

HIV status of donor

Note: HIPAA requires that you collect the minimum necessary.

29.6 * Describe your plan to destroy the identifiers at the earliest opportunity consistent with the research or provide a health or research justification for retaining the identifiers, or indicate and explain that retention is required by law:

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.

30.0 Informed Consent

30.1 * 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

30.2 * Process for obtaining informed consent:

After advertisements any responded subject will be invited to office of [redacted] for oral tissues.
[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

The infnat tonsil samples from discarded materials will be collected at [redacted] She will meet with parents of child in her office and describe them the purpose of the study and the procedures and answered their questions. If the parents remains interested he/she reads and signs the informed consent forms.

30.3 * 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.

31.0 Financial Considerations

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

Payments will be (check all that apply):

- ☐ Subjects will not be paid
- ☐ Cash
- ☑ Check
- ☐ Gift card
- ☐ Other:

Specify **Other**:

31.2 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:

$100 buccal tissue

31.3 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.

32.0 CTSI Screening Questions

32.1 * This study will be carried out at one of the UCSF Clinical Research Services (CRS) units or will utilize CRS services:

○ Yes ◉ No

32.2 This project involves community-based research:	
○ Yes ◉ No	
32.3 This project involves practice-based research:	
○ Yes ◉ No	

Study Application (Version 1.6)

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]

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) at UCSF and affiliated sites ONLY by clicking the "Add a new row" button: NOTE: This information is required and your application will be considered incomplete without it.

KSP Name	Description of Study Responsibilities	Qualifications
[redacted]		

5.0 Initial Screening Questions

5.1 * This study involves human stem cells (including iPS cells and adult 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 (e.g. buccal swabs, urine, hair and nail clippings, etc.)
- ☐ Category 4: Noninvasive clinical procedures (e.g. physical sensors such as pulse oximeters, MRI, EKG, EEG, ultrasound, moderate exercise testing, etc.)
- ☐ Category 5: Research involving materials (data, documents, records, or specimens) that were previously collected for either nonresearch or research purposes
- ☐ Category 6: Use of recordings (voice, video, digital or image)
- ☐ Category 7: Low risk behavioral research or research employing survey, interview, oral history, focus group, program evaluation, human factors evaluation, or quality assurance methodologies
- ☐ Category 8: Continuing review of previously approved full committee research that is essentially complete
- ☑ Category 9: Continuing review of research NOT involving an IND or IDE where the IRB has determined that the research poses no greater than minimal risk

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 sponsors and provide the funding details:

External Sponsor:

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	P0037598	

Sponsor Name:	California HIV/AIDS Research Program
Sponsor Type:	14
Sponsor Role:	Funding
CFDA Number:	
Grant/Contract Number:	

Awardee Institution::	UCSF
Is Institution the Primary Grant Holder:	Yes
Contract Type:	Grant
Project Number:	P0037598
UCSF RAS System Award Number ("A" + 6 digits):	
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		A121266

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):	A121266
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:	

Gift, Program, or Internal Funding (check all that apply):

- ☐ Funded by gift (specify source below)
- ☐ Funded by UCSF or UC-wide program (specify source below)
- ☐ Specific departmental funding (specify source below, if applicable)
- ☐ Unfunded (miscellaneous departmental funding)
- ☐ Unfunded student project

List the gift, program, or departmental funding source:

7.2 If you tried to add a sponsor in the question above and it was not in the list, follow these steps:

- **If funding has already been awarded or the contract is being processed by the Contracts and Grants or Industry Contracts unit, your sponsor is already in the system and the project has a UCSF RAS System Proposal or Award number. Check with your department's Research Services Analyst (RSA) to ask how the sponsor is listed in the UC sponsor list and what the Proposal or Award number is.**
- **If you need additional assistance, contact the Contracts and Grants Award Team at CGAwardTeam@ucsf.edu and list the sponsor in the box below.**

○ Sponsor not in list

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

If the funding is administered by the UCSF Office of Sponsored Research, your study will not receive CHR approval until the sponsor and funding details have been added to your application.

7.3 * This study is supported in whole or in part by Federal funding:

◉ Yes ○ No

If **yes**, indicate which portion of your grant you will be attaching:

- ☑ The Research Plan, including the Human Subjects Section of your NIH grant or subcontract
- ☐ 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
- ☐ The grant is not attached. The study is funded by an award that does not describe specific plans for human subjects, such as career development awards (K awards), cooperative agreements, program projects, and training grants (T32 awards)

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):

- ☑ UCSF
- ☐ Mt. Zion
- ☑ San Francisco General Hospital (SFGH)
- ☐ SF VA Medical Center (SF VAMC)
- ☐ Helen Diller Family Comprehensive Cancer Center
- ☐ Blood Centers of the Pacific (BCP)
- ☐ Blood Systems Research Institute (BSRI)

- [ ] Gallo
- [ ] Gladstone
- [ ] Institute on Aging (IOA)
- [ ] Jewish Home
- [ ] 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:

- [ ] Other UC Campus
- [x] Other institution
- [ ] Other community-based site
- [ ] Foreign Country

List the foreign country/ies:

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 Studies Involving Other Sites

10.1 UCSF is the coordinating center:

- ( ) Yes
- (•) No

If **Yes**, describe the plan for communicating safety updates, interim results, and other information that may impact risks to the subject or others among sites:

If **Yes**, describe the plan for sharing modification(s) to the protocol or consent document(s) among sites:

10.2 Check any other UC campuses with which you are collaborating on this research study:

- [ ] UC Berkeley
- [ ] UC Davis
- [ ] Lawrence Berkeley National Laboratory (LBNL)
- [ ] UC Irvine
- [ ] UC Los Angeles
- [ ] UC Merced
- [ ] UC Riverside

- [ ] UC Santa Barbara
- [ ] UC Santa Cruz

10.3 Are the above UC campuses requesting to rely on UCSF's IRB (check all that apply)?

- [ ] Yes (Attach the Notice of Intent to Rely on One UC IRB form in the Other Study Documents section)
- [ ] No (Complete IRB Approval Certification section)

11.0 IRB Approval Certification

(Note: This section replaces the old IRB Approval Certification Supplement form. Please do not attach the old form(s) to this application.)

11.1 IRB Approval Certification Sub-Form

Click "Add a new row" to enter information for a site. Click it again to add a second site and againand again for a third, a fourth, etc.

Outside Site Information

Non-UCSF affiliated site information:

Site name:

Cooperative Human Tissue Network (CHTN) ;http://www.chtn.nci.nih.gov/what-is/

Contact name:

[redacted]

Email:

[redacted]

Phone:

[redacted]

For Federally-funded studies only, corresponding FWA#:

*** The research at this site will be reviewed by:**

- ○ The non-affiliated site's IRB or a private IRB
- ○ The non-affiliated site is requesting UCSF to be the IRB of record for this study
- ◉ The non-affiliated site is not engaged in the human subjects research and has provided a letter of support

If the other site's IRB approval letter is available now, attach it to

submit it once you receive it.

Or, if the other site is **not engaged** in human subjects research, attach the letter of support to your application.

12.0 Study Design

12.1 * 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/infant 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/infant 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 breast milk, however, their role in HIV infection with fetal/neonatal mucosal epithelium in not clear. Therefore we will investigate role of fetal/infant mucosal epithelium in HIV transmission, as well as role of breast milk in HIV transmission via fetal/infant oral mucosal epithelium. We also will study HIV transmission via cervical mucosal epithelium, and expression of innate proteins in cervical epithelial cells. We have shown that HIV infection may disrupt mucosal epithelium leading paracellular penetration of Epstein-Barr virus (EBV) and human paplillomavirus (HPV). Uisng biopsy tissues from HIV-infected donors we will study the paracellular penetration of EBV and HPV.

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.

12.2 Check all that apply:

- ☐ Phase I
- ☐ Phase II
- ☐ Phase III
- ☐ Phase IV

13.0 Scientific Considerations

13.1 Hypothesis:

This study has a hypothesis:

◉ Yes ○ No

If yes, state the hypothesis or hypotheses:

Hypothesis: The HIV envelope contains PS, and interaction of virus-associated PS with the PS receptor TIM-1 at the fetal/infant oral and intestinal mucosal surfaces induces viral

galactosylceramide initiates endocytosis of HIV into fetal/infant oral and intestinal epithelium. Both macropinocytosis and endocytosis of HIV facilitate transport of virions into intracellular vesicular/endosomal compartments, which subsequently deliver virus to basolateral membranes by transcytosis.

Hypothesis: HIV-infected monocytes/macrophages attach to the mucosal surfaces of fetal/infant oral and intestinal epithelia by binding of LFA-1 of monocytes/macrophages to ICAM-1 of oral epithelial cells. Secretion of TNF-a from HIV-infected monocytes/macrophages activates MCP-1 in oral/intestinal epithelium, facilitating migration of HIV-infected monocytes/macrophages into the epithelium.

Hypothesis: HBD-2 and/or -3 binds to HSPG, which mediates HBD binding to HIV gp120, leading to inactivation of HIV within the vesicular/endosomal compartments of fetal/infant oral and intestinal epithelial cells. Recombinant HBD-2 and -3 containing protein trunsduction signals are efficiently taken up into infant oral and intestinal epithelia, and spread of HBDs within mucosal epithelial tissues inhibits or substatially reduces HIV MTCT.

Hypothesis: In HIV infected individulas the tight junctions of mucosal epithelium disrupted, leading to paracellular penetration of EBV and HPV into epithelium.

13.2 * 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, and intestinal mucosal epithelium of the fetus, and tonsil epithelium of infants.
Aim 2. Analyze expression of HBD1, HBD2, HBD3 and SLPI in adult, fetal and infant epithelia.
Aim 3. Study mechanisms of HIV spread via the oral and cervical of the adult, and oropharyngeal and intestinal mucosal epithelium of the fetus and tonsil epithelium of infant.
Aim 4. Determine the role of breast milk in the interaction of HIV with the mucosal epithelium of the fetus and infant.
Aim 5. Study the mechanisms of HIV-mediated tigh junction disruption and its role in HPV and EBV paracellular penetration.

13.3 Statistical analysis:

Data will be analyzed by a study steering committee, which will be composed by [redacted] and [redacted] It is estimated that 30 adult, 50 fetal and 50 infant tissues will be investigated for this study. Fifteen breast milk from HIV-negative woman will be analyzed. A five years cumulative enrollment is planned.
HIV-1 infection of the mucosal epithelium and breast milk will be evaluated in the presence of HIV by immunostaining and ELISA assays, respectively. At least five samples will be used for each experiment. The statistical analysis will account for the paired nature of the data. 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.

13.4 * This is an investigator-initiated study:

(•) Yes ( ) No

13.5 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:

Committee on human research

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

14.0 Background

14.1 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. examined HIV in the amniotic fluid of 366 pregnant women, and HIV was detected in 17% of the 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 HIV may disrupt epithelial tight junctions leading to facilitate EBV and HIV paracellular penetration. Therefore we will study mechanisms of HIV, EBV and HPV dissemination within the adult oral mucosal epithelium.

14.2 Preliminary studies:

Our preliminary results show the following: (ii) HIV via mucosal epithelium is initiated by macropinocytosis and clathrin- and caveolin-mediated endocytosis. (iii) The HIV envelope contains PS, and infant oral epithelium expresses the PS receptor TIM-1, which may play a critical role in viral macropinocytosis. (iv) Infant oral epithelium expresses ICAM-1, which may bind macrophage LFA-1, facilitating binding of HIV-infected macrophages to mucosal surfaces. TNF-a induces MCP-1 expression in infant tonsil keratinocytes, which may initiate migration of HIV-infected monocytes/macrophages into epithelium. (v) Infant oral epithelial cells do not express the anti-HIV innate proteins HBD-2 and -3. Recombinant HBD-2 and/or -3 internalized into polarized infant oral cells bind indirectly to HIV gp120 and inactivate virus during its transcytosis. HIV tat containing a PTD signal rapidly penetrates into oral epithelium, suggesting that generation of HBDs containing a PTD signal could facilitate their penetration into epithelium. (vi) Infant tonsil epithelium contains M cells, which may play an important role in HIV transcytosis and MTCT.

14.3 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

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.

15.0 Sample Size and Eligibility

15.1 Number of subjects that will be enrolled at UCSF and affiliated institutions (locally):

292

15.2 Total number of subjects that will be enrolled at all sites (for study overall):

292

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

235

15.4 Sample size calculation:

This is an ongoing project and the first CHR application was submitted in 2007 for NIH R01 and UCSF intramural grants with the initial sample size of 586. However, the NIH R01 was not funded and we were not able to perform some studues. Therefore, so far we have colleced 162 samples, we are currently analyzing data from these samples.

We are continuing this project under new NIH R01 grant. For this study will be used 50 tonsil tissues from young children (between 0.5 to 5 years old). We also will use 50 fetal tissue explants (25 oropharyngeal and 25 intestinal), which will be collected from aborted (discarded) fetuses between 18 and 24 weeks of gestation. Next, we will use 15 cervical and 15 oral (buccal) biopsies

propagated small tissues explants from each tissue, and each experiment will be performed using a set of explants from the same biopsy and will be repeated at least 3 times with biopsies obtained from independent donors. HIV transmission through mucosal epithelium will be examined by applying of virus with and without breast milk from the surface of epithelium and detection of virus in the lamina propria.

Total sample size = 292, estimated sample szie= 335
162 samples are already collected and currently under investigation

Anticipated sample size for next 5 years:
Tonsil tissues from tonsillectomy/surjical material= 50
Fetal tissues from discarded fetuses= 50
Oral (buccal) biopsy =15
Breasr milk= 15

We will use total of 50 tonsil tissues from children, 50 tissues from fetus (25 oropharyngeal and 25 intestinal), 15 oral tissues and 15 breast milk samples from adults. These numbers have chosen based on nature of treatment and controls.

15.5 * Eligible age range(s):

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

15.6 Inclusion criteria:

The only inclusion criterion will be willingness to donate oral, tonsil and cervical tissues, and breast milk samples.

15.7 Exclusion criteria:

The only exclusion criteria would be inability to provide informed consent and having a contraindication for a biopsy procedure, e.g., bleeding diathesis or infection and inflammation.

15.8 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:

16.0 Drugs and Devices

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

- ( ) Yes (•) No

16.2 * Investigational medical devices or in vitro diagnostics will be used OR approved medical devices or in vitro diagnostics will be studied under this application:

- ( ) Yes (•) No

16.3 * A Non-Significant Risk (NSR) determination is being requested for an investigational device:

○ Yes ◉ No

16.4 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.

17.0 Other Approvals and Registrations

17.1 * This is a clinical trial:

○ Yes ◉ No

Clinical Trial Registration
"NCT" number for this trial:

17.2 * Data from this study will be submitted to NIH for Genome-Wide Association Studies (GWAS):

○ Yes ◉ No

17.3 * This study involves vaccines produced using recombinant DNA technologies:

○ Yes ◉ No

17.4 * This study involves human gene transfer (NOTE: Requires NIH Recombinant DNA Advisory Committee (RAC) review prior to CHR approval):

○ Yes ◉ No

17.5 * The study protocol requires radiological procedures (e.g. CT scans, x-rays) or exposes subjects to radiation:

○ Yes ◉ No

17.6 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

18.0 Procedures

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

1. Collection of adult oral tissue and establishment of tissue explants ex vivo.
The buccal explants will be obtained from HIV-positive and –negative adult volunteers. 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 mucosa containing epithelium and connective tissue will be obtained using 4-mm–diameter biopsy punches in [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.

For verification of HIV-negative status 20 ml of blood sample will be taken from arm vein of each donor. HIV test will be performed by HIV diagnostic lab at UCSF Parnassus campus. If HIV test shows positive results, [redacted] will contact donor by phone to discuss this issue and will refer for treatment and care. [redacted] will inform HIV positive result by name of donor to the Department of Public Health and tissue of HIV-positive donor will be removed from the experiments.

2. Collection of fetal oropharyngeal and intestinal tissue explants
Fetal ropharyngeal gastric and intestinal tissue explants 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 tissues 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 oropharyngeal area by surgical instrument. Similarly sized pieces will be dissected from the small 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 0.4 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 tonsil tissue samples from 0.5-5 years old children.
Collection of discarded tonsil tissue samples from HIV-negative children under 5 years old after routine tonsillectomy. A small tissue samples with about 5 -5 cm will be collected from surgical materials after routine tonsillectomy. 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.

4. Collection of breast milk.
To examine the role of breast milk in HIV infection of fetal/infant mucosal epithelium we will collect breast milk. Fifty ml of breast milk from 15 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-

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.

5. Infection of tissue explants with HIV, HPV 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 HPV infection will be used HPV 16 pseudovirions (1 ng/per biopsy). 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 HPV-16 pseudovirions.

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

18.2 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.

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

○ Yes ◉ No

If yes, explain:

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

○ Yes ◉ No

If yes, explain:

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

◉ Yes ○ No

18.6 Time commitment (per visit and in total):

1. For obtaining one oral or cervical biopsy sample per visit will be required about 30 min.
2. For obtaining one tonsil sample per visit will be required about 1 h.
3.The time to obtaining one breast milk sample will be about 10 min.

18.7 Locations:

1.The oral biopsy tissues from the adult buccal mucosa from normal healthy individuals and HIV-positive patients will be collected in [redacted] Contact

person: [redacted] Cervical tissue will be collected from HIV-negative and -positive premenopausal women at [redacted] Contact person: [redacted]
2. The discarded fetal oropharyngeal and intestinal tissue samples from discarded fetus will be collected in [redacted]
3. The discarded infant tonsil tissues will be collected in [redacted] Contact person, [redacted]
4. The breast milk samples will be collected in breastfeeding healthy HIV-negative women at [redacted] Contact person: [redacted]
5. All HIV and/or EBV and/or HPV infection procedures with tissue samples in presence or absence of breast milk will be performed in UCSF Parnassus campus, [redacted] Contact person, [redacted]

18.8 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 tissue biopsy 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

19.0 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.)

19.1 Specimens are (check all that apply):

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

If Other, explain:

19.2 Types of specimens:

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

Describe and/or name source:

Oral and cervical biopsy tissues and breast milk will be collected

19.3 Consent will be obtained via:

- [ ] Separate specimen banking consent form
- [x] Specimen banking section within a main research study consent form

☐ Surgical consent form with tissue donation brochure

19.4 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:

19.5 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:

20.0 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.)

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

[redacted]

20.2 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:

20.3 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)

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

- ○ Yes
- ◉ No

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

20.5 There is a formal specimen utilization review process:

- ○ Yes
- ◉ No

If **Yes**, describe the process:

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

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

21.0 Alternatives

21.1 Study drug or treatment is available off-study:

- ○ Yes
- ◉ No
- ○ Not applicable

21.2 * Is there a standard of care (SOC) or usual care that would be offered to prospective subjects at UCSF (or the study site) if they did not participate:

○ Yes ◉ No

If yes, describe the SOC or usual care that patients would receive if they choose not to participate:

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

21.3 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.

22.0 Risks and Benefits

22.1 * 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 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.

22.2 Steps taken to minimize risks to subjects:

To minimize the discomfort of the oral and cervical tissue biopsy 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.

22.3 Benefits to subjects:

○ Yes ◉ No

If yes, describe:

22.4 Benefits to society:

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

22.5 Explain why the risks to subjects are reasonable:

The study risks on an individual level will be minimal (potential loss of privacy). Also, the risk of harm from oral and cervical 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.

23.0 Data and Safety Monitoring Plan

23.1 Describe the plan for monitoring data and safety:

This is not an interventional trial and no data safety monitoring board is needed.

23.2 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.

23.3 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.

24.0 Confidentiality and Privacy

24.1 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.

24.2 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.

24.3 Study data are:

- ☐ Derived from the Integrated Data Repository (IDR) or The Health Record Data Service (THREDS) at SFGH
- ☑ 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 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:

[redacted]

24.4 Identifiers may be included in research records:

- (•) Yes ( ) No

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

- [x] Names
- [ ] Dates
- [ ] Postal addresses
- [ ] Phone numbers
- [ ] Fax numbers
- [ ] Email addresses
- [ ] Social Security Numbers*
- [ ] Medical record numbers
- [ ] 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

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

- ( ) Yes (•) No

If **yes**, indicate to whom identifiable information may be disclosed:

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

If **Others**, specify:

24.6 Indicate how data are kept secure and protected from improper use and disclosure (check all that apply): NOTE: Whenever possible, do not store subject identifiers on laptops, PDAs, or other portable devices. If you collect subject identifiers on portable devices, you MUST encrypt the devices.

- [x] 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
- [x] Data are kept in a locked office or suite
- [x] Electronic data are protected with a password
- [x] Data are stored on a secure network
- [ ] Data are collected/stored using REDCap or REDCap Survey

24.7 Additional measures to assure confidentiality and protect identifiers from improper use and disclosure, if any:

All study numbers will be destroyed as soon as the needed data are transcribed.

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

- ○ Yes
- ◉ No

Explain:

HIV positive test results will be reported to the Department of Public Health.

24.9 This study will be issued a Certificate of Confidentiality:

- ○ Yes
- ◉ No

25.0 Subjects

25.1 Check all types of subjects that may be enrolled:

- [ ] Inpatients
- [ ] Outpatients
- [x] Healthy volunteers
- [ ] Staff of UCSF or affiliated institutions

25.2 Additional vulnerable populations:

- [x] 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
- [x] 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

For this study will be collected the tonsil tissue samples from children under 10 years old, which are undergo to routine tonsilectomy procedures. The surgical tissues are discarded materials. Since,
tonsilectomy procedures do not have any interventions and do not cause risk to children.

Also, will be collected oropharyngeal and intestinal tissues 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.

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:

26.0 Inclusion of Children in Research

(Note: This section replaces the old "Inclusion of Children and Minors in Research" supplement form. Please do not attach the old form to this application.)

26.1 This study will enroll children who can legally consent for themselves:

○ Yes ◉ No

If **yes,** explain why they can consent for themselves in the research setting:

If you will **ONLY** be enrolling children who can legally consent for themselves, press **SAVE and CONTINUE** to skip the rest of this section.

26.2 Select all the regulatory categories that apply:

- ☑ No greater than minimal risk (45 CFR 46.404, 21 CFR 50.51)
- ☐ Greater than minimal risk but presenting prospect of direct benefit (45 CFR 46.405, 21 CFR 50.52)
- ☐ Greater than minimal risk (though only a minor increase over minimal risk) and no prospect of direct benefit but likely to yield generalizable knowledge about the subjects disorder or condition (45 CFR 46.406, 21 CFR 50.53)
- ☐ Research not otherwise approvable which presents an opportunity to understand, prevent, or alleviate a serious problem affecting the health or welfare of children (45 CFR 46.407, 21 CFR 50.54)

Explain why the research in this study falls under the above category or categories:

For this study will be collected the tonsil tissue samples from children under 10 years old, which are undergo to routine tonsilectomy procedures. The surgical tissues are discarded materials. Since,
tonsilectomy procedures do not have any interventions and do not cause risk to children.

Also, will be collected oropharyngeal and intestinal tissues 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.

26.3 Parental permission or waiver:

- ☑ Parental permission will be obtained
- ☐ Waiver of parental permission is requested: Parental permission is not a reasonable requirement
- ☐ Waiver of parental permission is requested: The waiver meets the provisions for a waiver of consent set forth in 45 CFR 46.116, Subpart A

If you are requesting a **waiver of parental permission**, explain why the study meets the

26.4 Assent of children or waiver:

- [ ] Assent of children old enough to provide assent will be obtained
- [x] Waiver of assent is requested: Children cannot be consulted or the research has prospect of direct benefit only available in the study
- [ ] Waiver of assent is requested: The waiver meets the provisions for a waiver of consent set forth in 45 CFR 46.116, Subpart A

If you are requesting a **waiver of child's assent**, explain why the study meets the regulatory criteria for this waiver:

Children under 10 years old are too young to be consulted.

26.5 Documentation of permission and assent (select all that will be used):

- [x] Permission form addressed to the parents
- [ ] Simplified assent form addressed to the child, 7-12 years old (parents get separate form)
- [ ] Assent form addressed to the child, 13 years and older (for subjects and parents)
- [ ] Assent form addressed to the child, 13 years and older (parents get separate form)

Check one:

- (•) One parent's signature will be obtained
- ( ) Two parents' signatures will be obtained

If this study is approvable under .404 or .405 and you plan to get permission from only one parent, explain why you think one parent's permission is sufficient:

26.6 This study may enroll wards of the state:

( ) Yes (•) No

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

27.1 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:
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.

28.0 Recruitment

28.1 * 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.

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:

28.2 * How, when, and by whom eligibility will be determined:

After obtaining of informed consent eligibility of donors for the oral and cervical biopsies will be determined by [redacted] and [redacted] respectively. HIV-negative donors should provide evidence of HIV negative test. Alternatevily, HIV negative status of donors will be verified by standard HIV lab testing in [redacted] Twenty ml of blood sample will be taken from arm vein for HIV testing.

The tonsil tissues will be collected from discarded surgical materials. After obtaining of informed consent [redacted] will perform tonsilectomy from HIV-negative children and discarded tonsil tissues will be provided to [redacted] laboratory. In the lab [redacted] will verify HIV negative status of donors by tesing of tonsil lymphocytes for HIV using ELISA p24, immunofluorescence and and Western blot assays.

After obtaining of informed consent the breast milk will be collected by HIV-negative donors and provided to [redacted] will verify HIV negative status of breast milk by tesing it for using ELISA p24, immunofluorescence and and Western blot assays.and lymphocytes from tonsil tissue will be tested for HIV using ELISA p24 assay.

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

Advertisements will be placed seeking volunteers to donate oral and cervical mucosal tissue, and breast milk. The advertisements will be posted around the UCSF and San Francisco General campuses, in local newspapers and on the Internet. Volunteers donating oral and cervical tissues 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 biopsy tissues from the adult oral and cervical mucosa will be collected in [redacted] and in [redacted]

2. The fetal oropharyngeal and intestinal tissue samples from discarded fetus will be collected [redacted]

3. The infant tonsil tissues will be collected from discarded surgical materailas in [redacted]

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

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

(•) Yes ( ) No

29.0 Waiver of Consent/Authorization for Recruitment Purposes

(Note: This section partially replaces the old "Request for Waiver of Consent/Authorization for Minimal Risk Research or for Screening for Recruitment" supplement form. Please do not attach the old form to this application.)

This section is now required when study investigators (and/or affiliated nurses or staff) recruit their own patients directly.

29.1 * Study personnel need to access protected health information (PHI) during the recruitment process and it is not practicable to obtain informed consent until potential subjects have been identified:

(•) Yes

If **no**, a waiver of consent/authorization is NOT needed**.**

29.2 * A waiver for screening of health records to identify potential subjects poses no more than minimal risk to privacy for participants:

(•) Yes

If **no**, a waiver of authorization can NOT be granted.

29.3 * Screening health records prior to obtaining consent will not adversely affect subjects' rights and welfare:

(•) Yes

If **no**, a waiver of authorization can NOT be granted.

29.4 * Check all the identifiers that will be collected prior to obtaining informed consent:

☑ Names

- [x] Dates
- [ ] Postal addresses
- [ ] Phone numbers
- [ ] Fax numbers
- [ ] Email addresses
- [ ] Social Security Numbers*
- [ ] Medical record numbers
- [ ] 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
- [ ] None

Note: HIPAA requires that you collect the minimum necessary.

29.5 * Describe any health information that will be collected prior to obtaining informed consent:

HIV status of donor

Note: HIPAA requires that you collect the minimum necessary.

29.6 * Describe your plan to destroy the identifiers at the earliest opportunity consistent with the research or provide a health or research justification for retaining the identifiers, or indicate and explain that retention is required by law:

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.

30.0 Informed Consent

30.1 * 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

30.2 * Process for obtaining informed consent:

After advertisements any responded subject will be invited to office of [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 the [redacted]

The infnat tonsil samples from discarded materials will be collected at [redacted] She will meet with parents of child in her office and describe them the purpose of the study and the procedures and answered their questions. If the parents remains interested he/she reads and signs the informed consent forms.

30.3 * 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.

31.0 Financial Considerations

31.1 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**:

31.2 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:

$100 buccal tissue
$50 cervical tissue

31.3 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.

32.0 CTSI Screening Questions

32.1 * **This study will be carried out at one of the UCSF Clinical Research Services (CRS) units or will utilize CRS services:**

○ Yes ◉ No	

32.2 This project involves community-based research:

○ Yes ◉ No	

32.3 This project involves practice-based research:

○ Yes ◉ No	

Outside Site Information (Version 1.1)

1.0 Outside Site Information

1.1 Non-UCSF affiliated site information:

Site name:

Cooperative Human Tissue Network (CHTN) ;http://www.chtn.nci.nih.gov/what-is/

Contact name:

[redacted]

Email:

[redacted]

Phone:

[redacted]

1.2 For Federally-funded studies only, corresponding FWA#:

1.3 * The research at this site will be reviewed by:

- ○ The non-affiliated site's IRB or a private IRB
- ○ The non-affiliated site is requesting UCSF to be the IRB of record for this study
- ◉ The non-affiliated site is not engaged in the human subjects research and has provided a letter of support

If the other site's IRB approval letter is available now, attach it to the application. If the IRB approval letter is not yet available, submit it once you receive it.

Or, if the other site is **not engaged** in human subjects research, attach the letter of support to your application.