



# Tricks in HIV's bag to counter vaginal or rectal microbicides

**Florian Hladik, MD, PhD**  
University of Washington  
Fred Hutchinson Cancer Research Center  
Seattle

# Contribution of the various HIV invasion routes to HIV cases worldwide

<b>Female genital tract</b>	<b>12.6 million</b>
<b>Male genital tract</b>	<b>10.2 million</b>
<b>Intestinal tract</b>	<b>7.3 million</b>
<b>Placenta</b>	<b>0.5 million</b>
<b>Blood stream</b>	<b>2.6 million</b>

# HIV vaccines



**April 23<sup>rd</sup>, 1984**



**Margaret Heckler  
Secretary of Health and Human Services**

**“We expect that a preventive HIV vaccine  
will be ready for testing within two years”**

# HIV microbicides



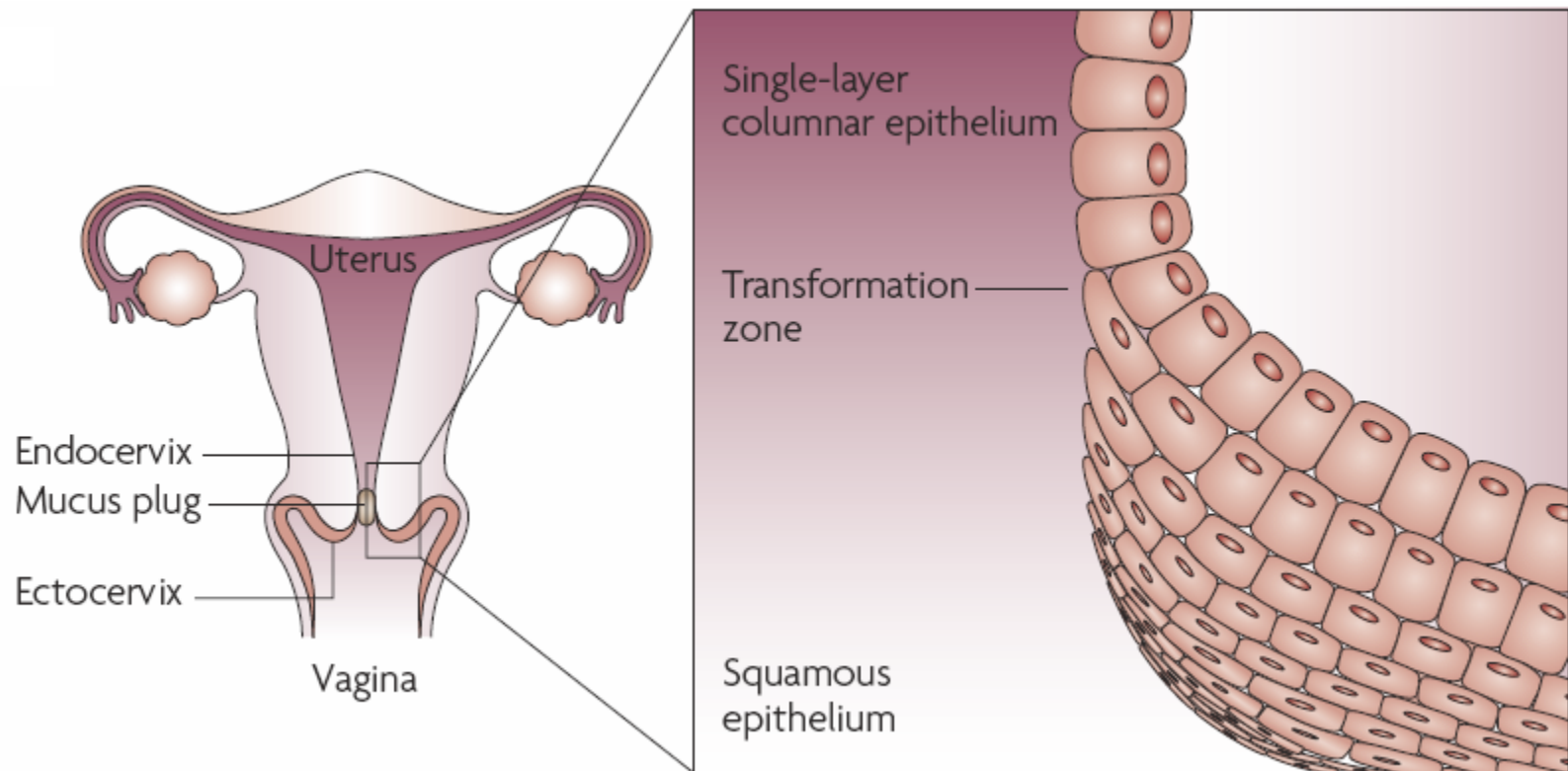
“

If you advise your husband to use a condom, he may beat you and send you away. Where do you go then?

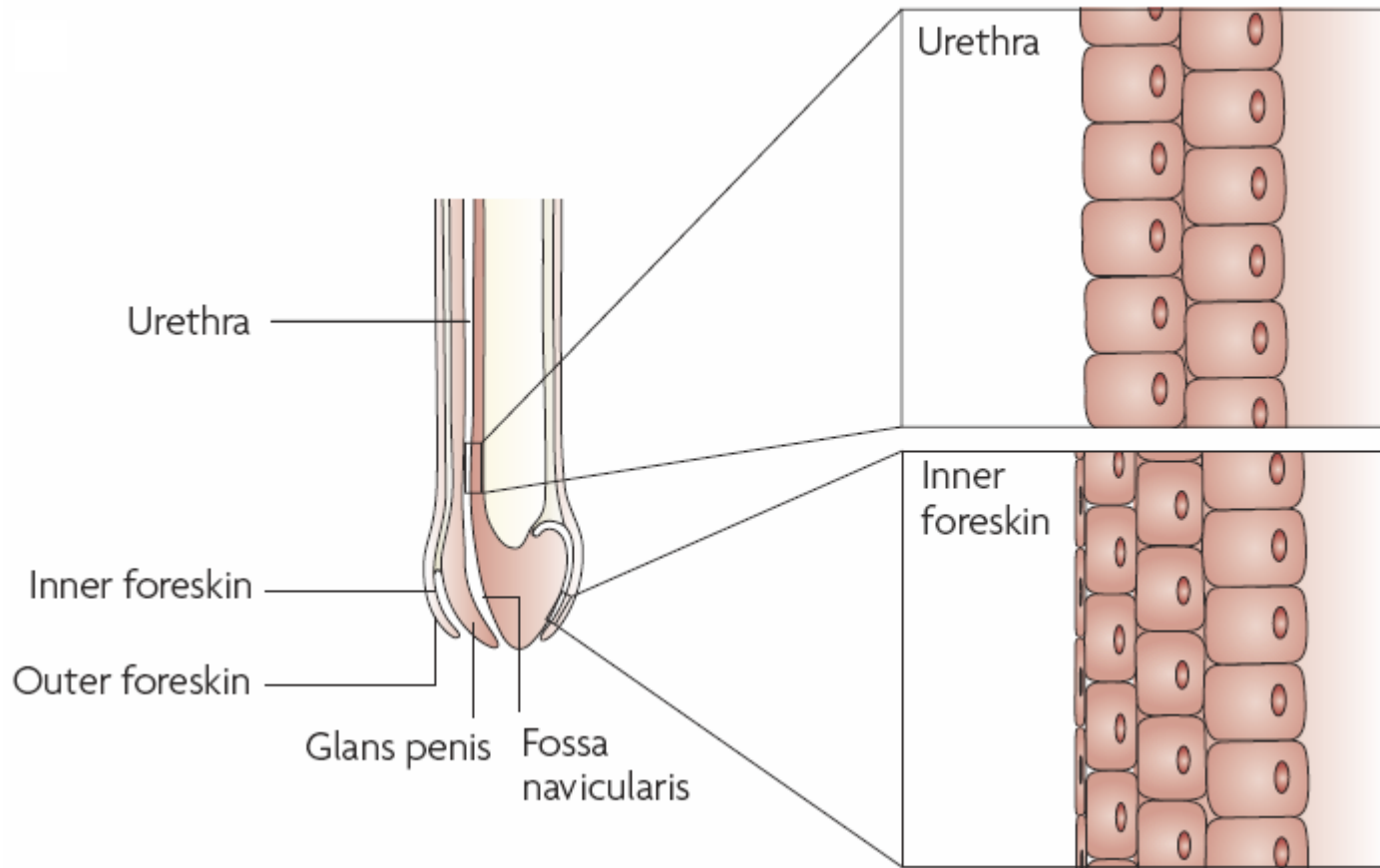
”

Rural woman  
Uganda

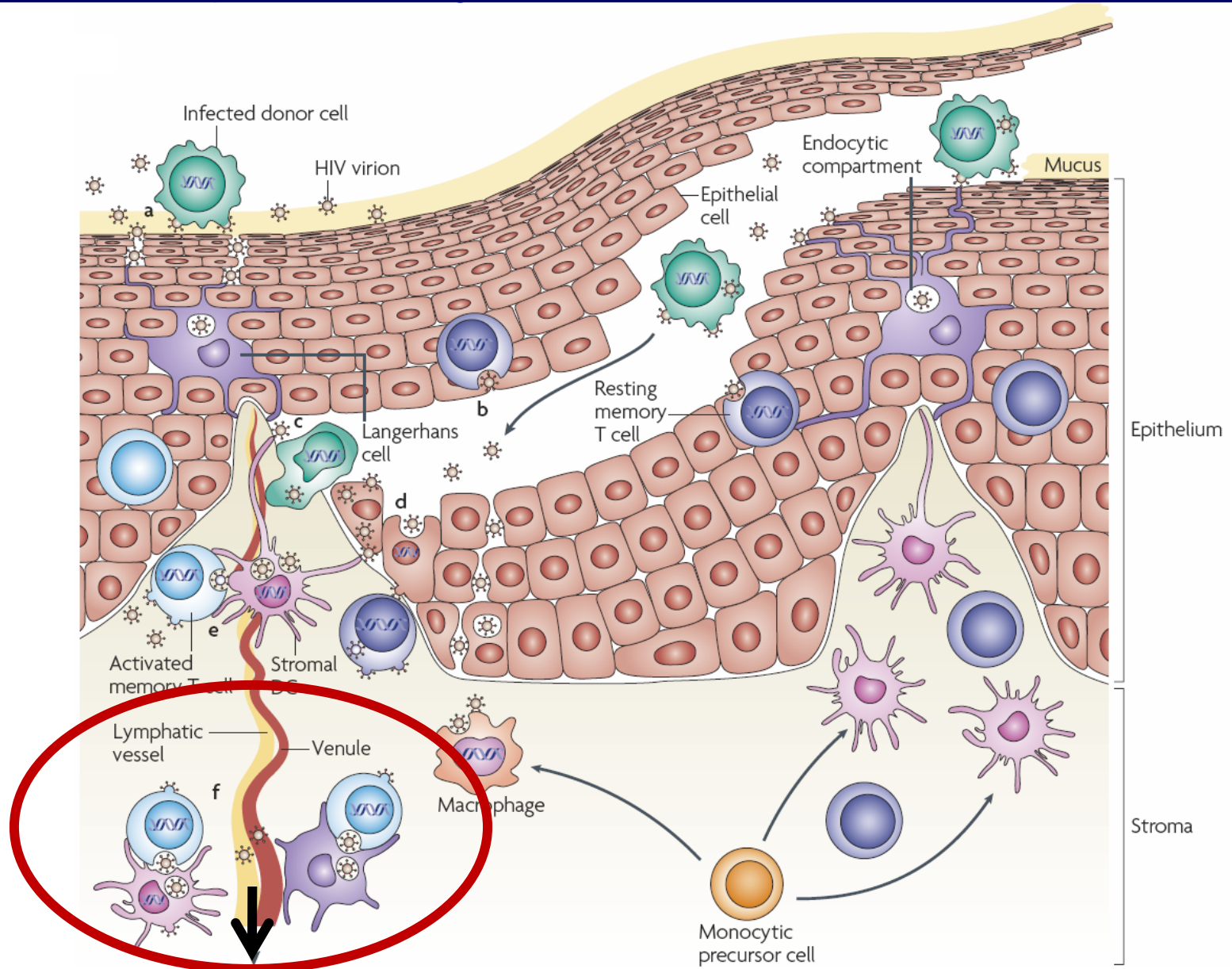
# Transmission sites in the female genital tract



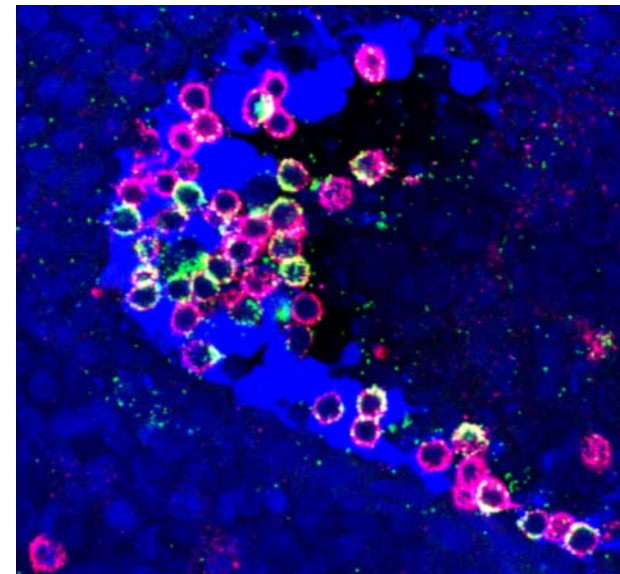
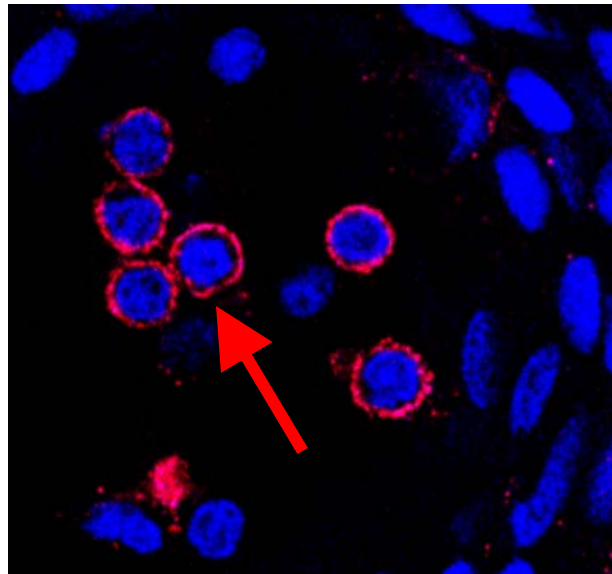
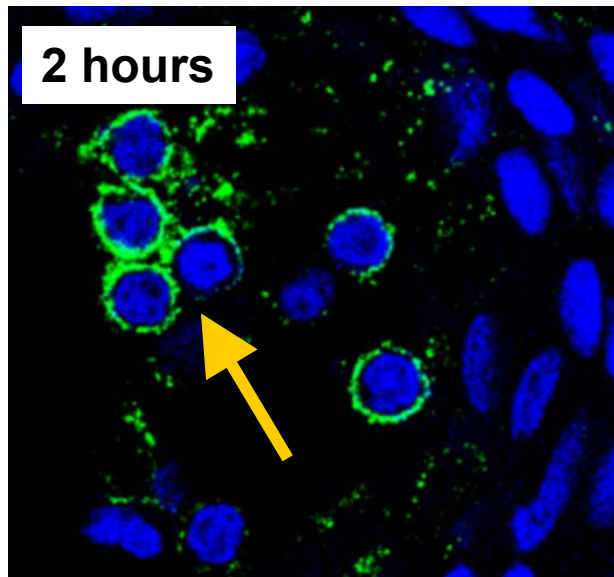
# Transmission sites in the male genital tract



# The mucosa – a multi-opportunity site for HIV



# HIV rapidly binds to intraepithelial vaginal T cells and productively infects them





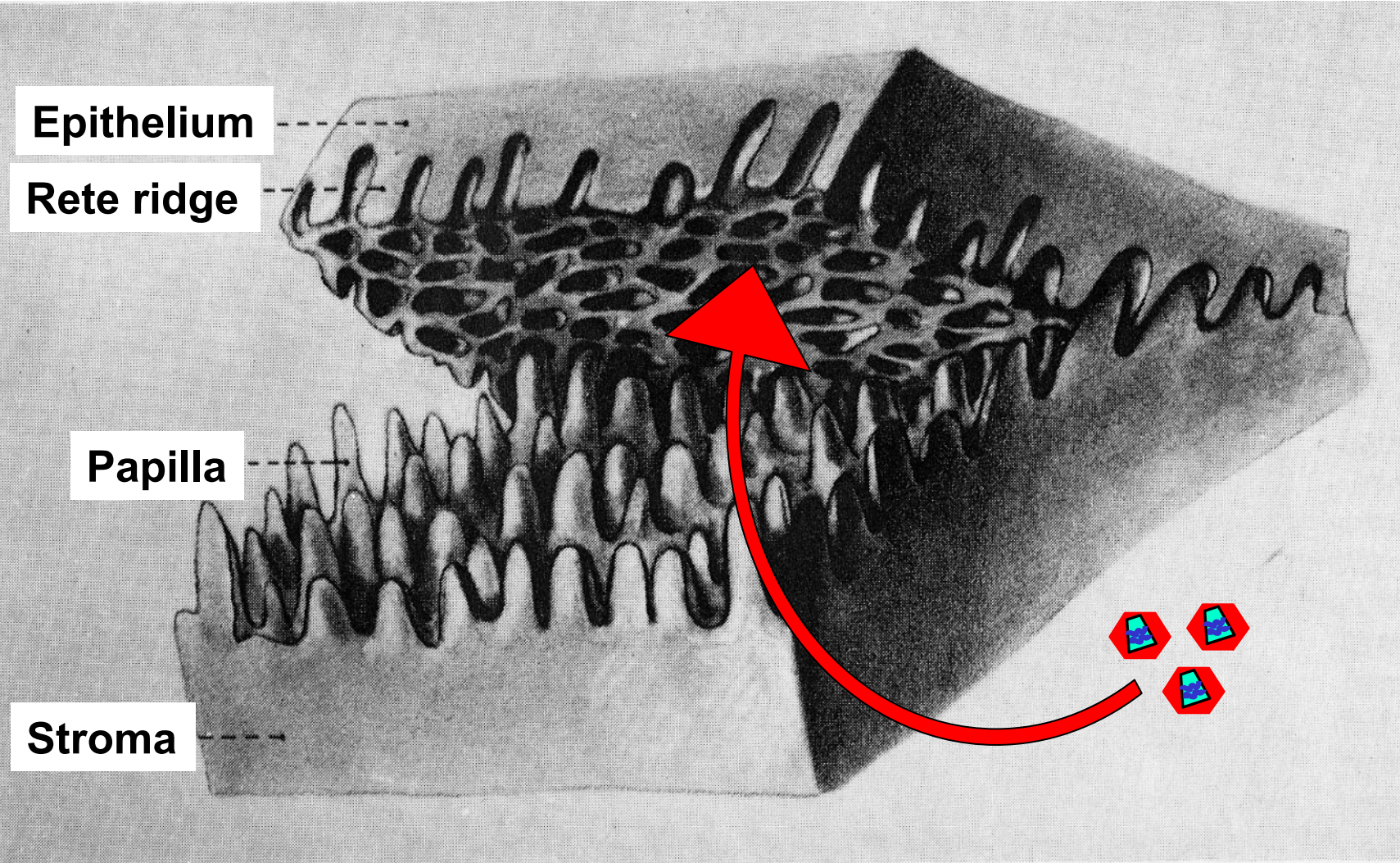
# HIV's tricks to counter microbicides

- 1. Disseminate beyond the reach of microbicide action**
- 2. Persist beyond the time of microbicide action**
- 3. Induce changes in the mucosa that pave the way for future infection**
- 4. Take advantage of co-existing STDs**
- 5. Acquire drug resistance**

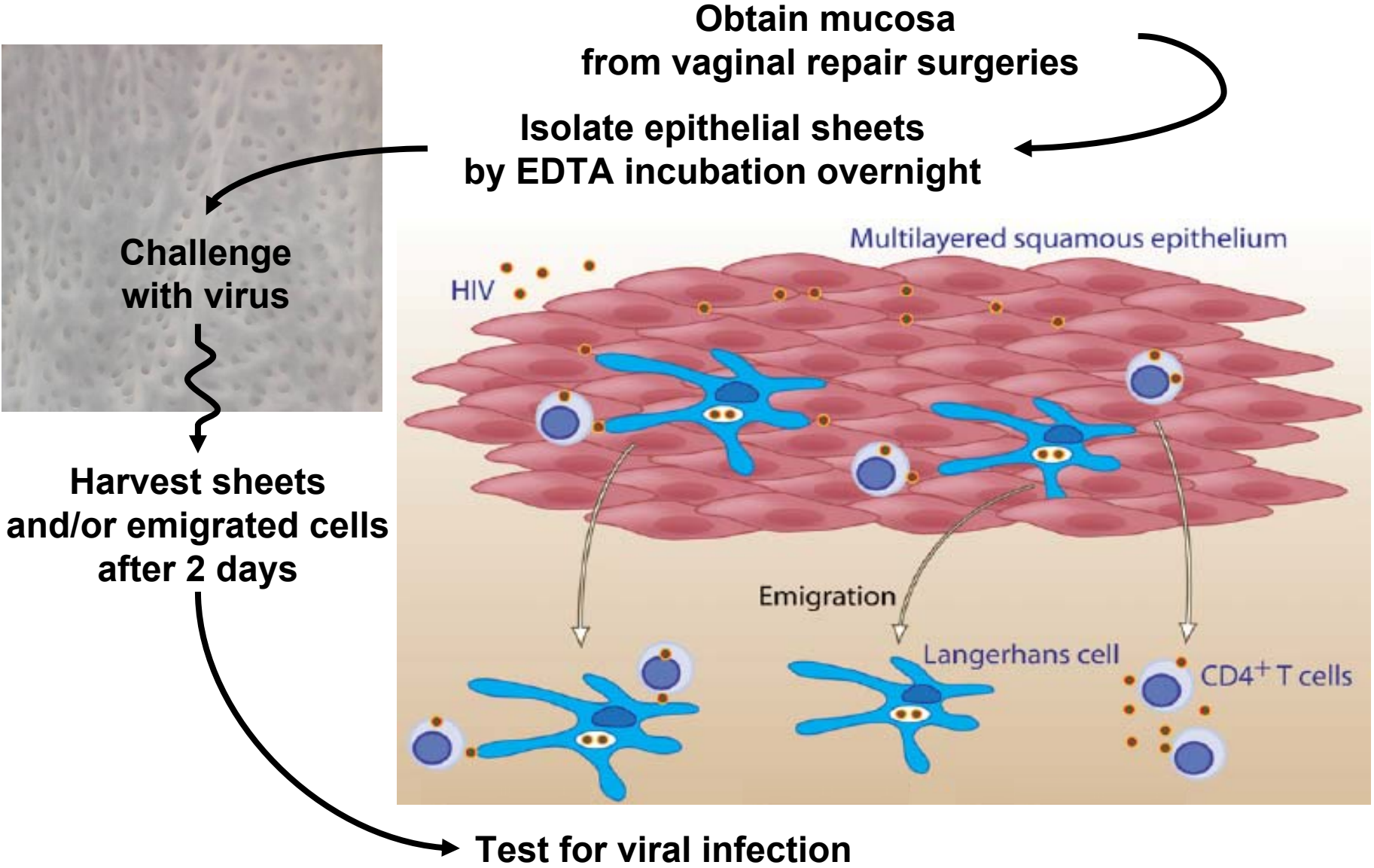
# **1. HIV dissemination beyond the reach of microbicide action**

- **Dissemination of HIV beyond the mucosa**
- **Insufficient mucosal penetration of the microbicide**

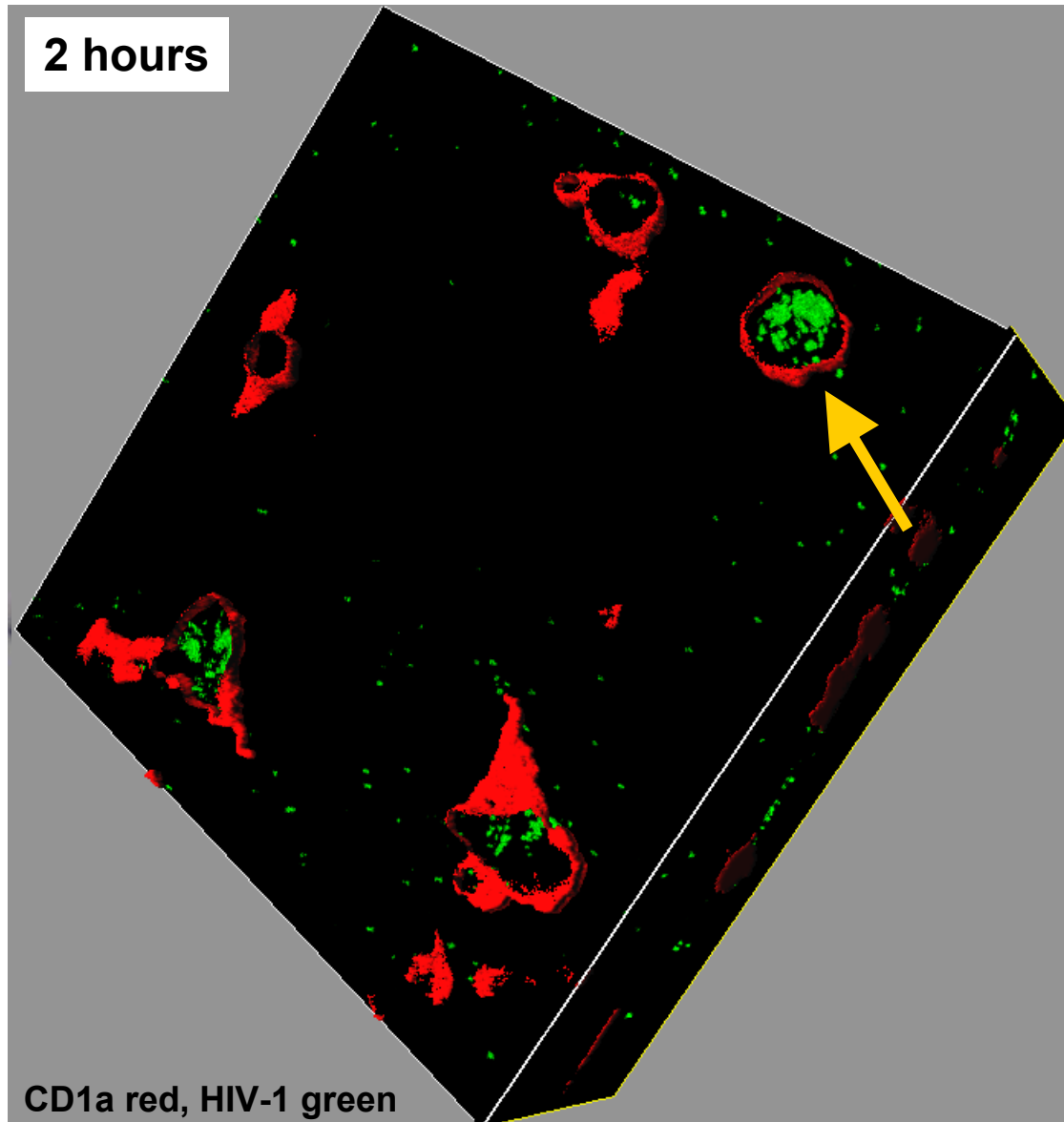
# Ex vivo HIV infection model of the vaginal epithelium



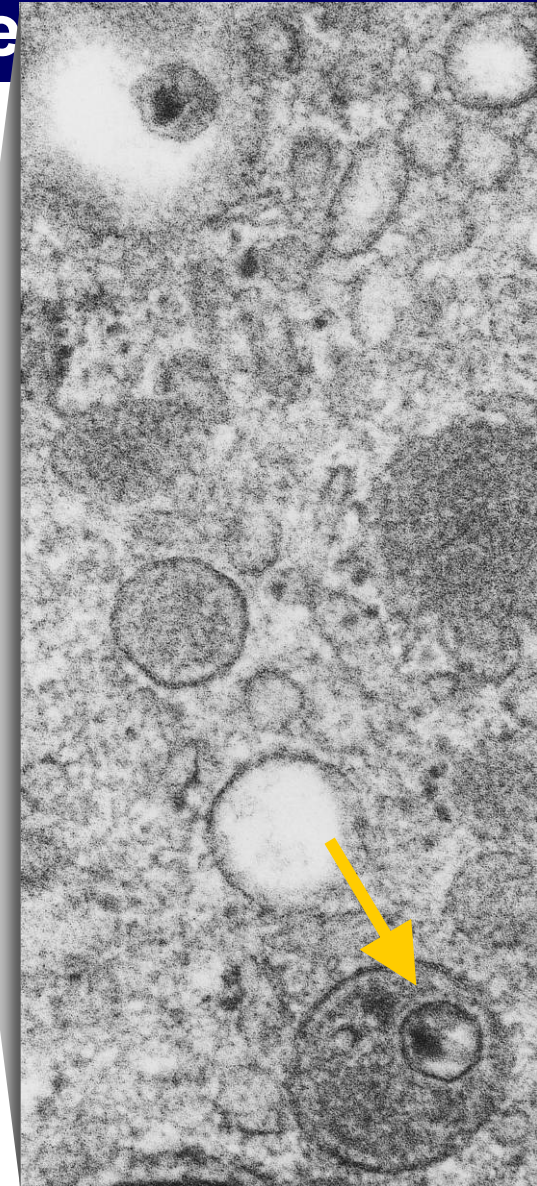
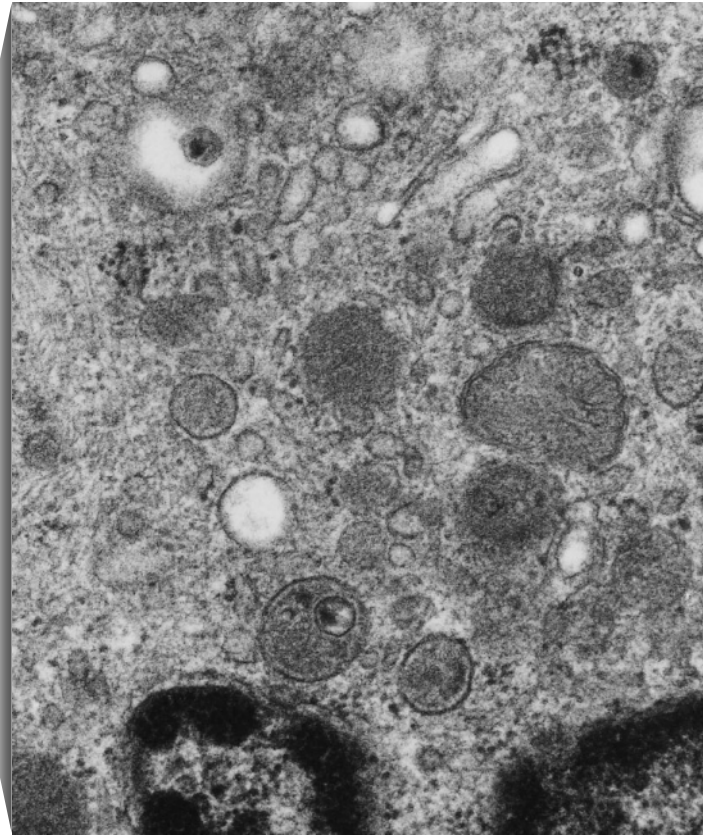
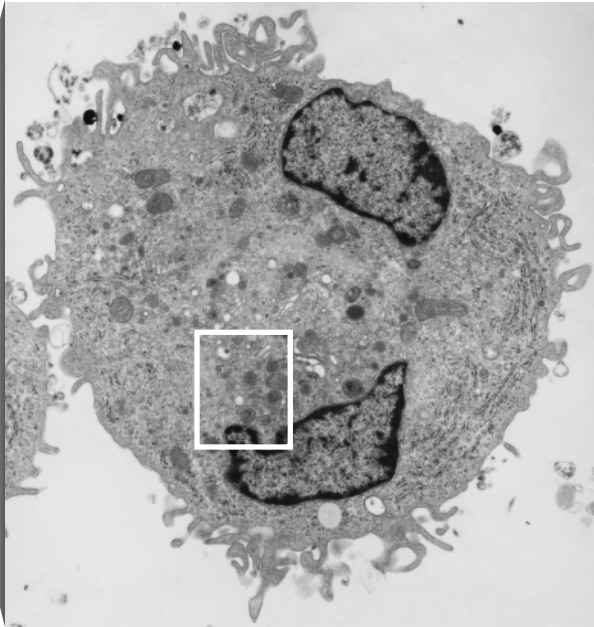
# Ex vivo HIV infection model of the vaginal epithelium



# HIV rapidly enters vaginal Langerhans cells

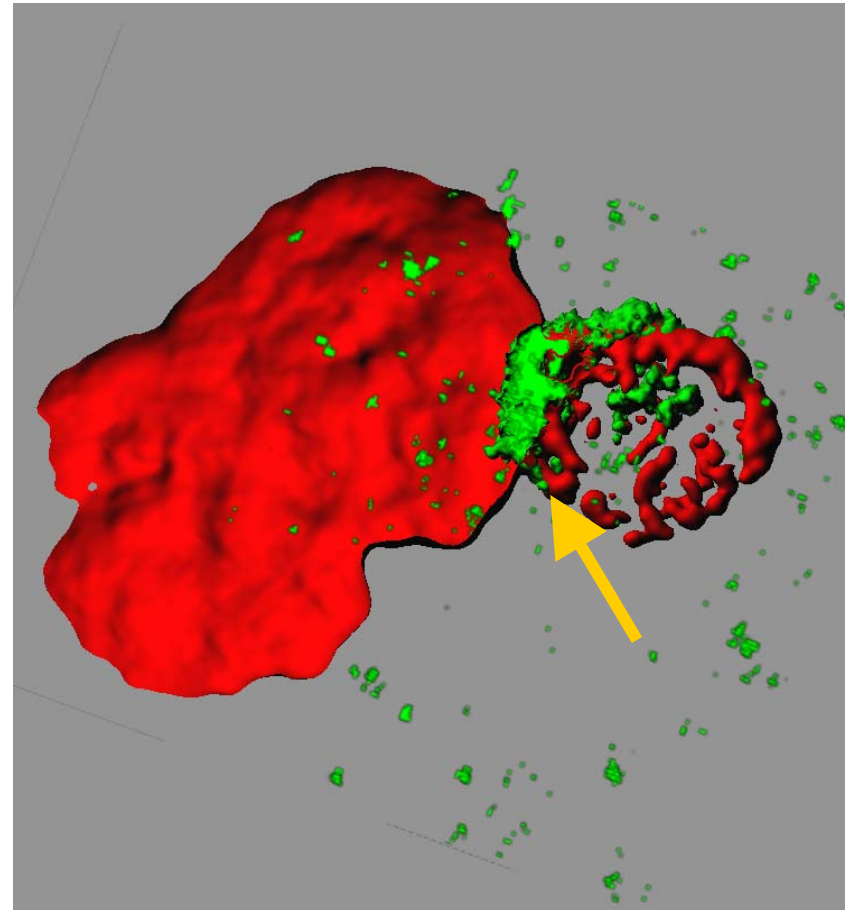
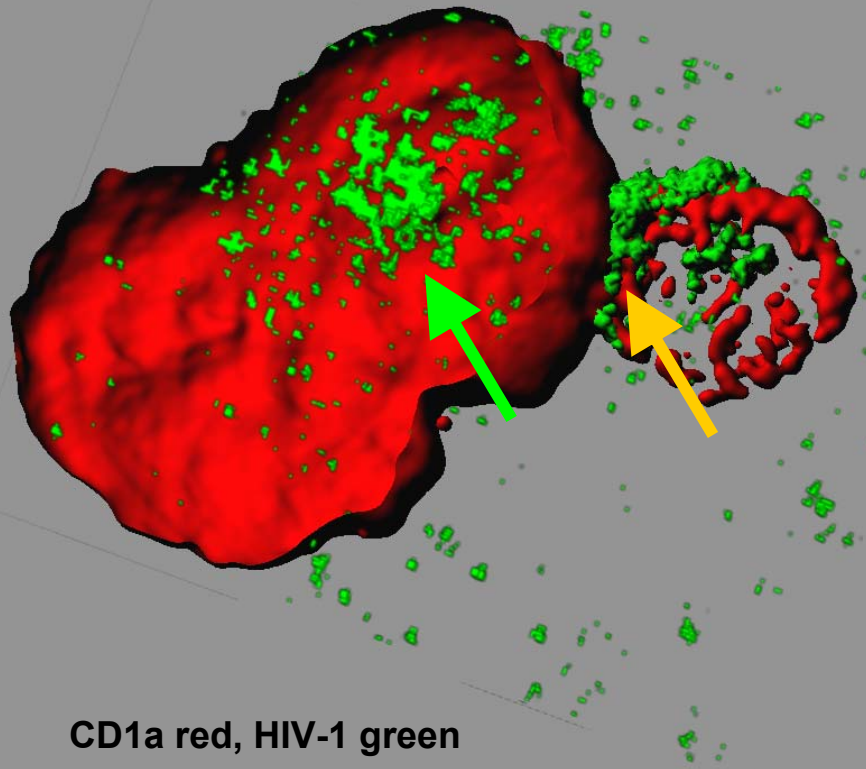


# Intact virions are frequently found in perinuclear endocytic compartments of Lange

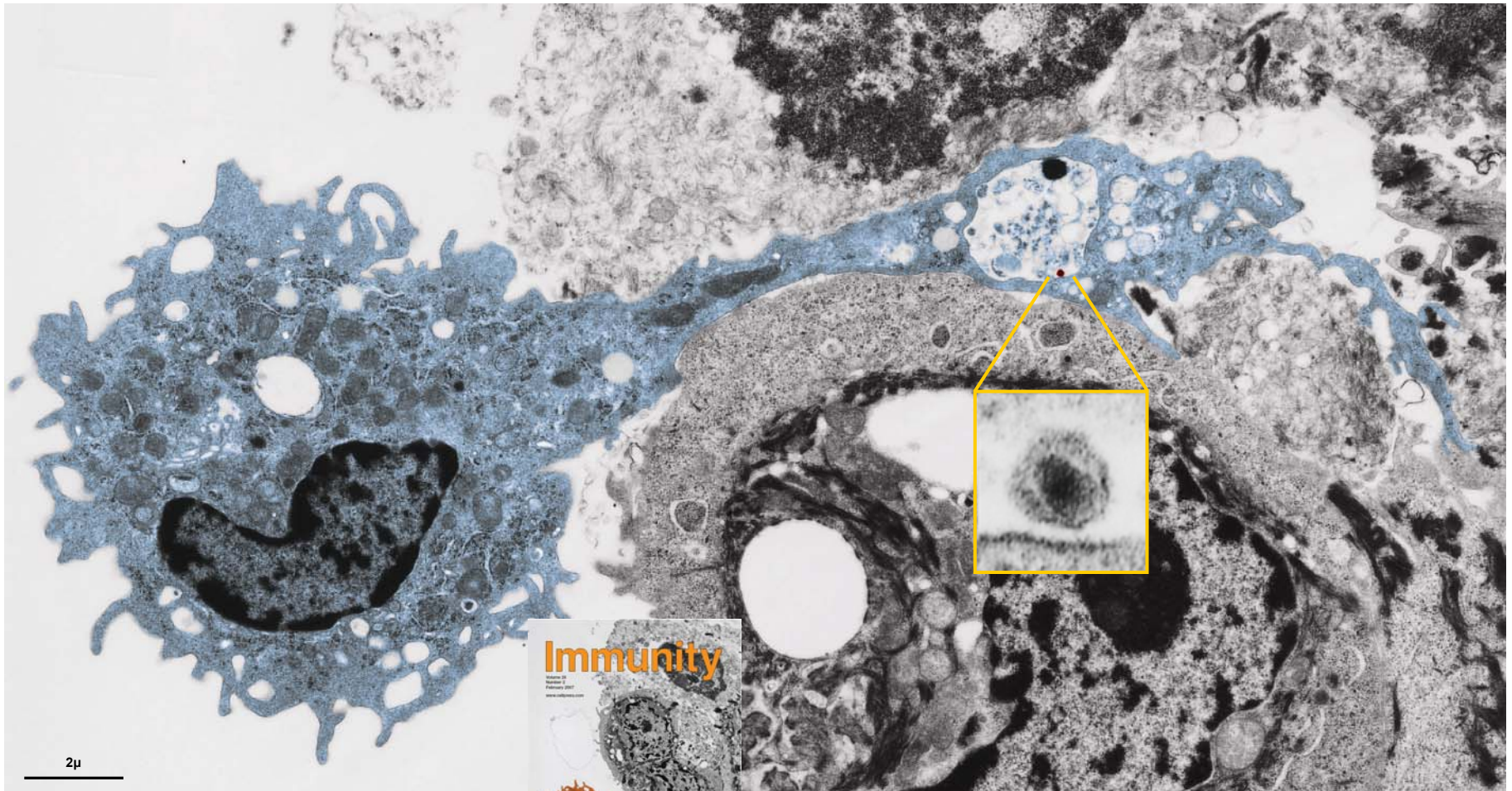


# Newly bound HIV-1 may concentrate along the LC-T cell junction (“infectious synapse”)

2 hours

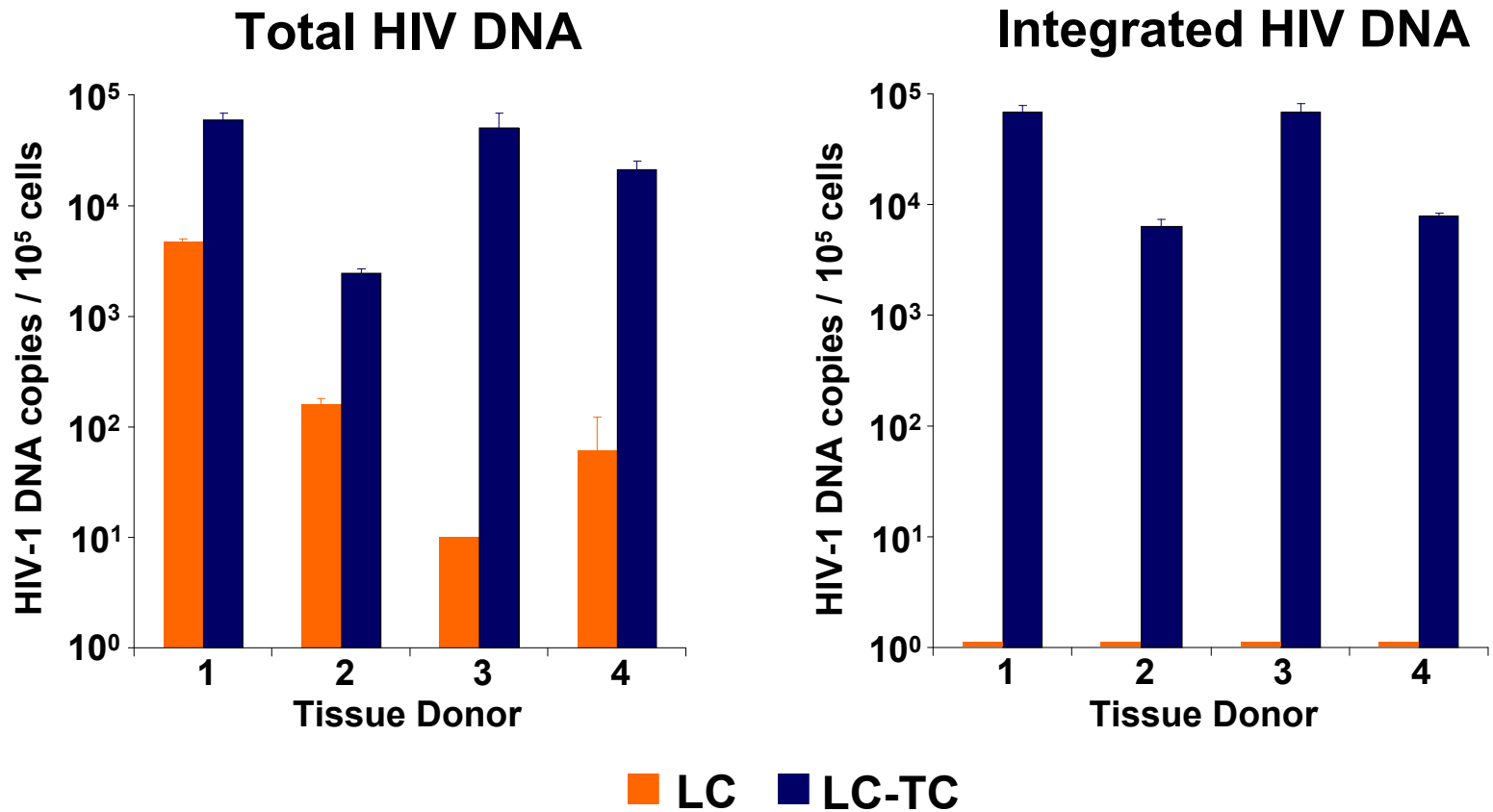


# Vaginal Langerhans cells carry internalized virions into the submucosa

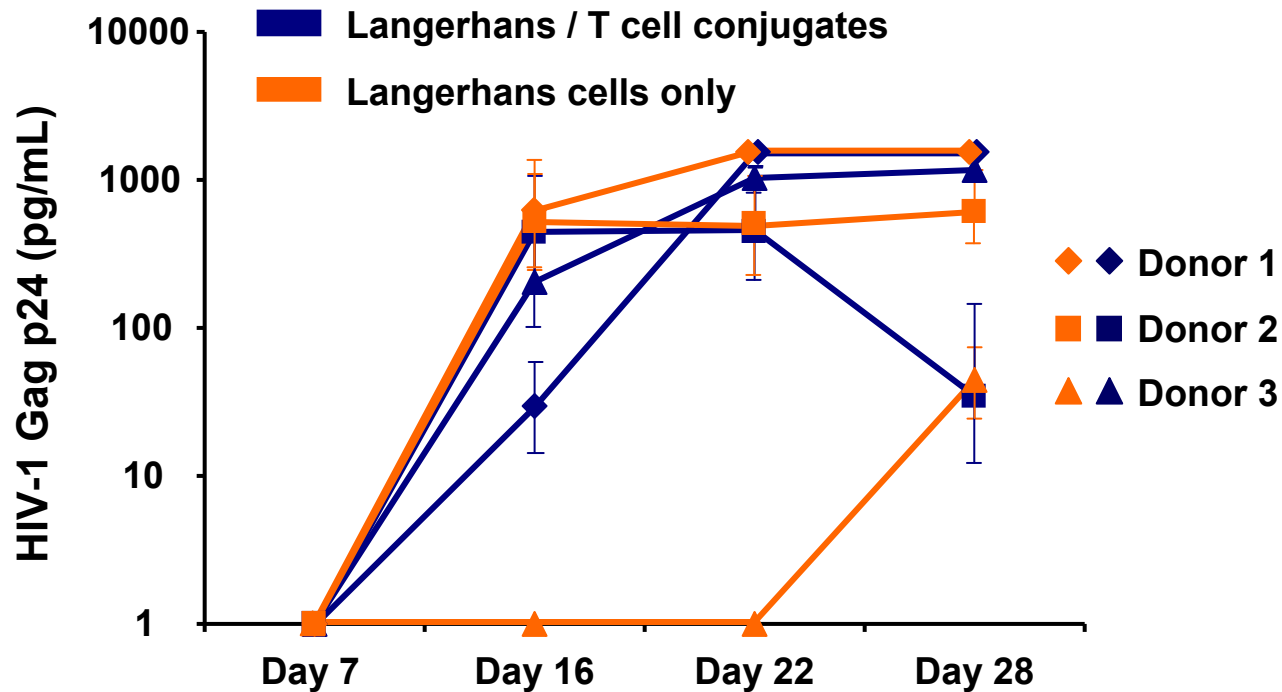




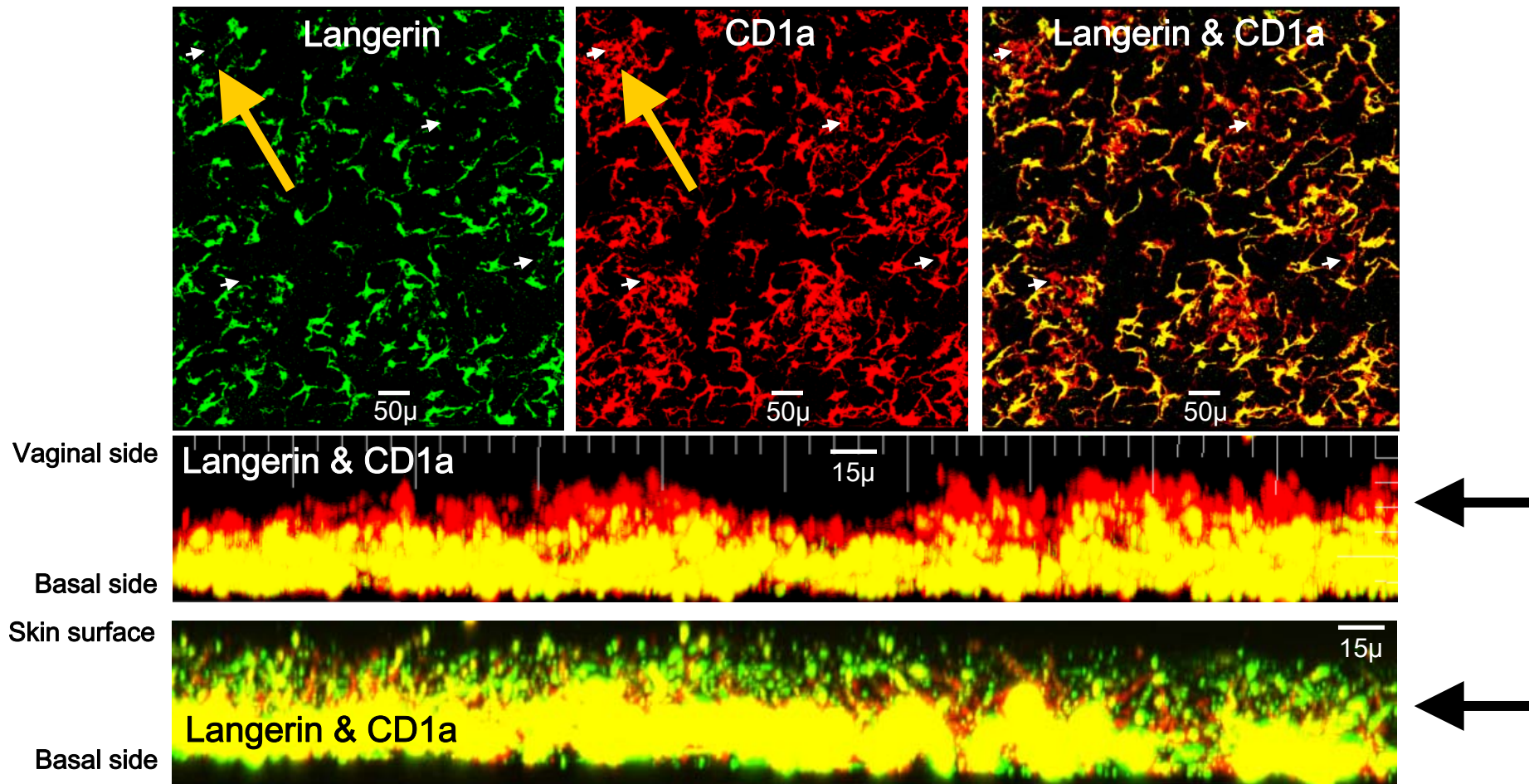
# Vaginal Langerhans are not productively infected



# Nevertheless, vaginal Langerhans cells pass infectious HIV-1 to T cells



# How does HIV bypass langerin-mediated degradation in vaginal Langerhans cells ?



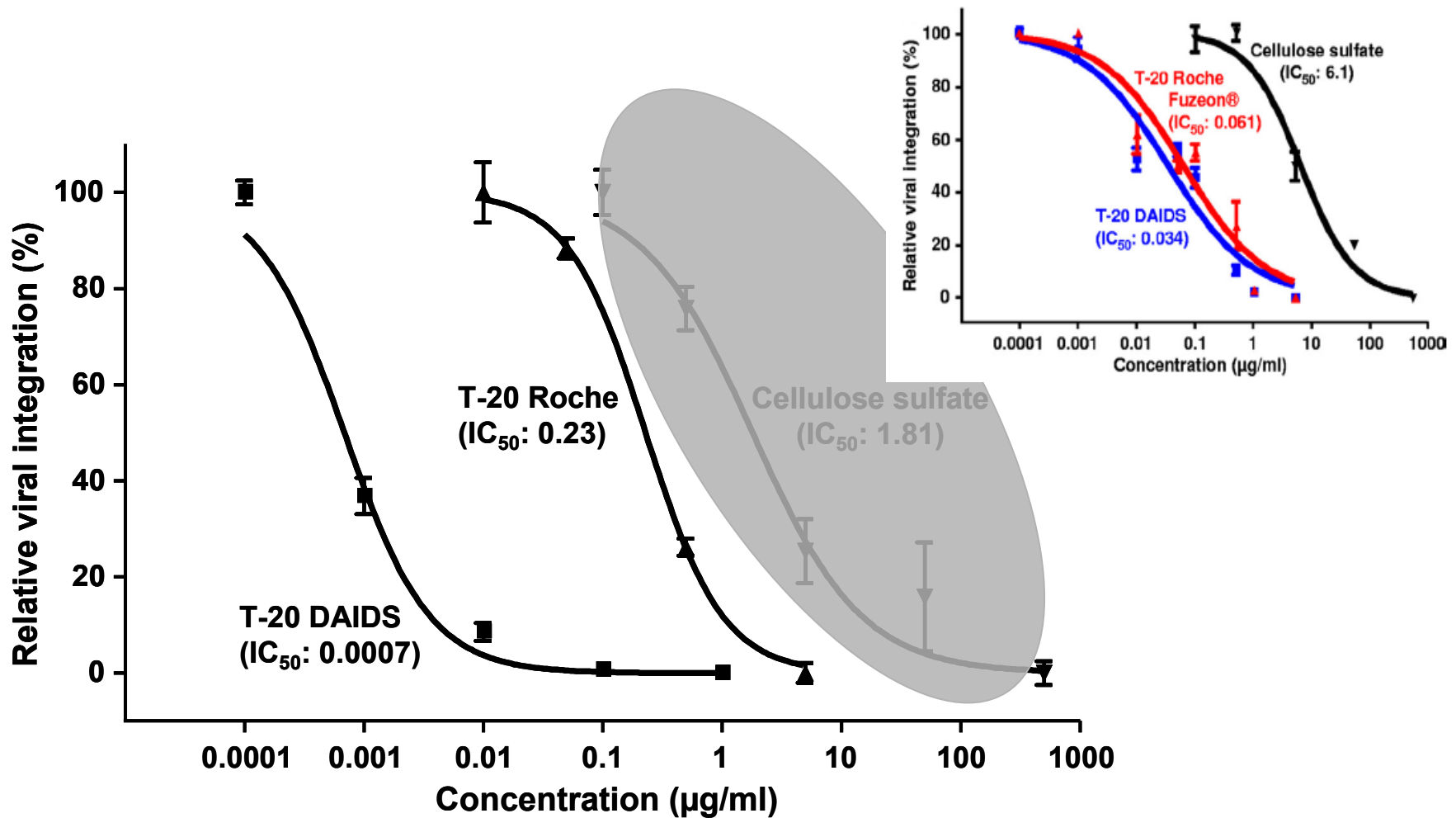
# **Dissemination of HIV beyond the mucosa**

## **Conclusion**

- **Prevention of productive infection of CD4<sup>+</sup> T lymphocytes residing in the mucosa is the most important goal**
- **However, HIV clearly has alternatives to straight-forward infection of local T cells**

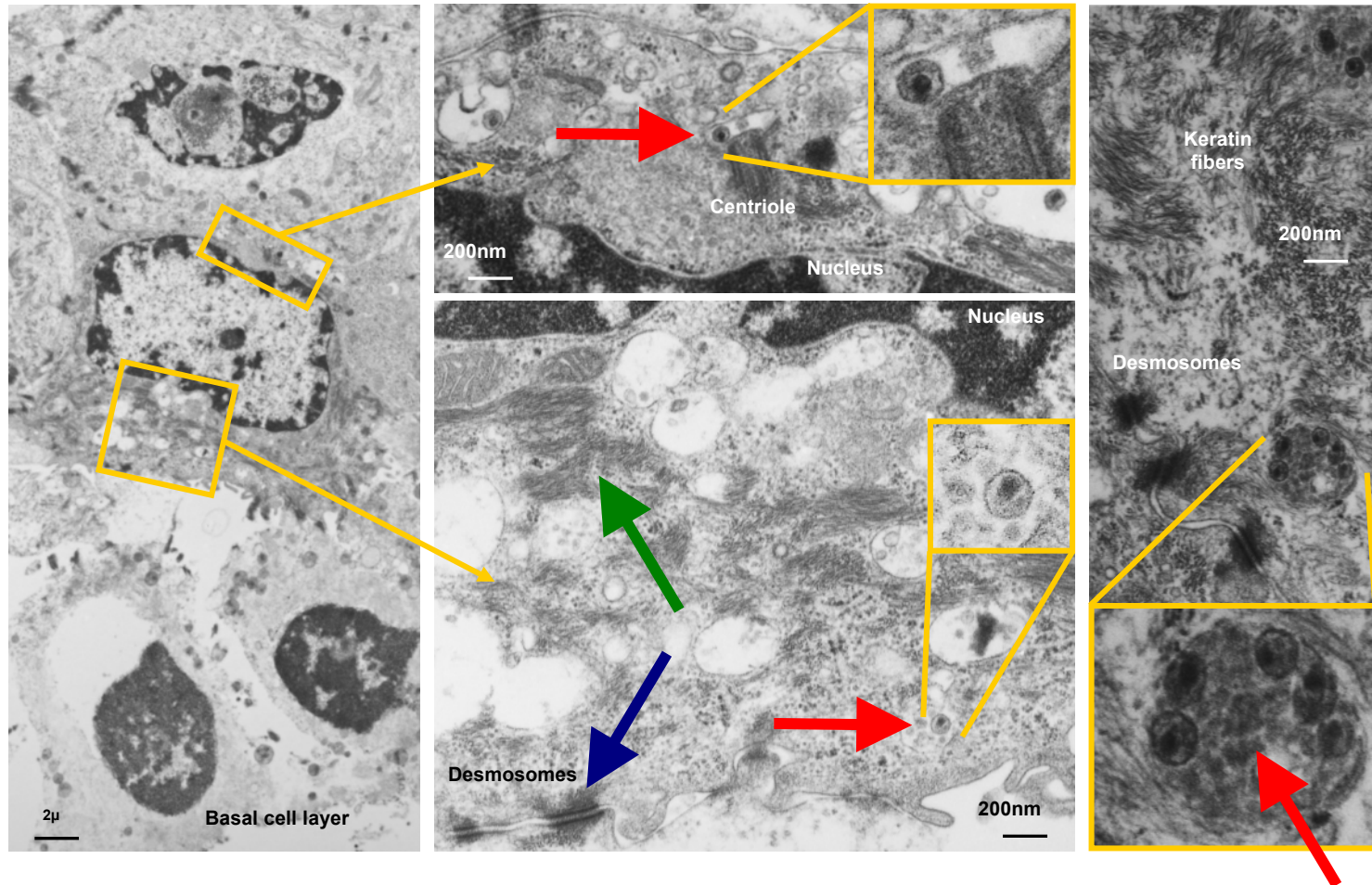
# Insufficient mucosal penetration of the microbicide

## An example: T-20 Fuzeon versus T-20 DAIDS



## 2. Local HIV persistence beyond the time of microbicide action

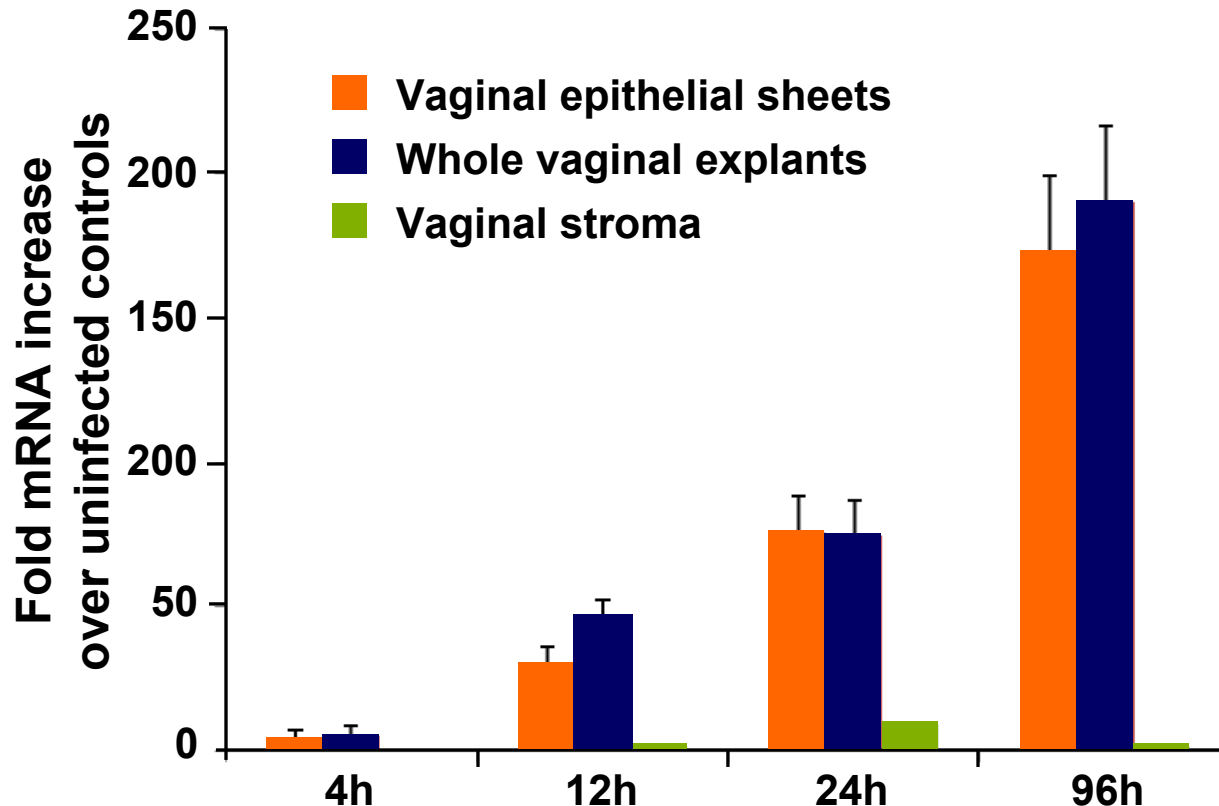
### HIV in vaginal epithelial cells



Unpublished results

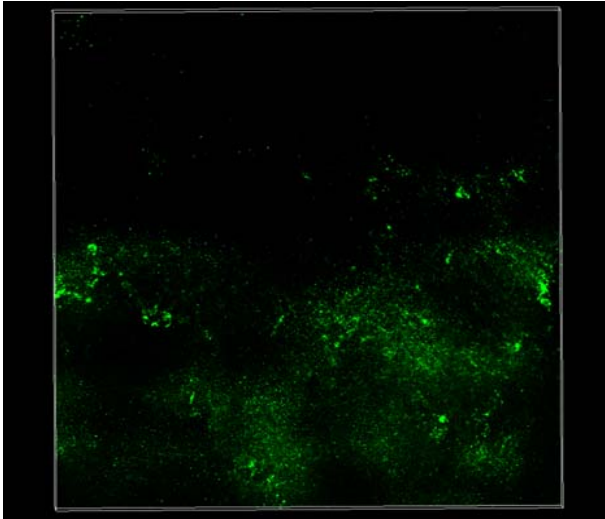
### 3. Infection-independent effects of HIV on the mucosa that pave the way for future infection

An example: HIV-1 triggers long-lasting expression of thymic stromal lymphopoietin (TSLP) in vaginal epithelium

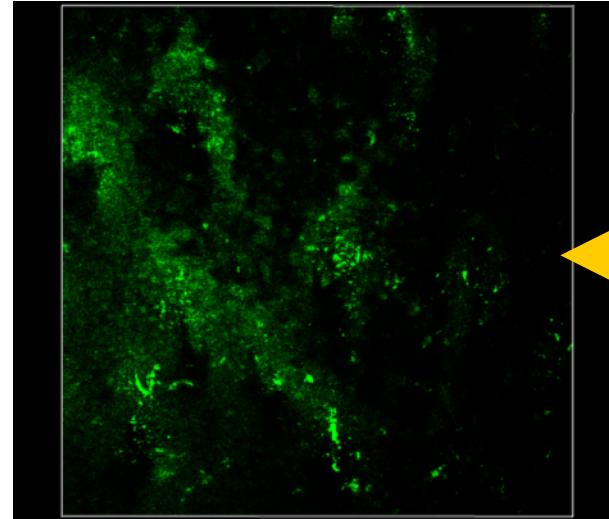


# HIV-1 triggers TSLP in vaginal epithelium

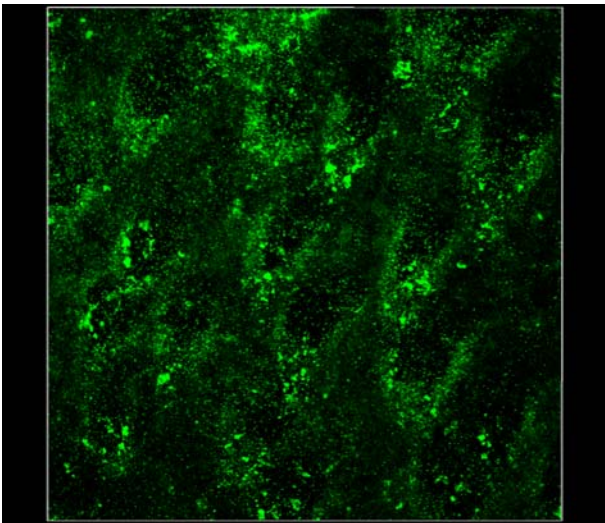
Baseline



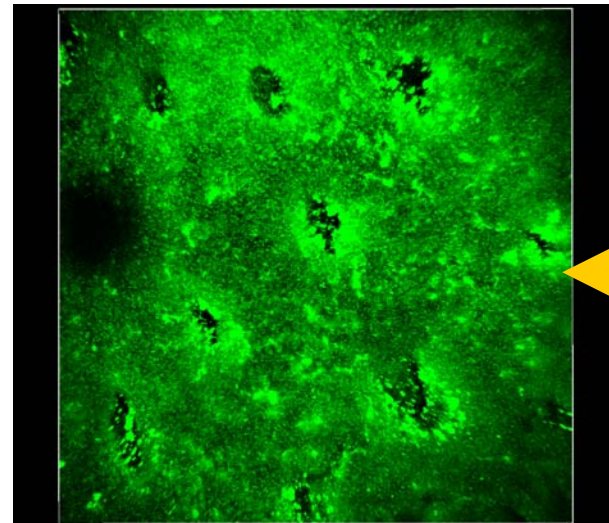
293T cell supernatant



Day 1



1 ng HIV-1 p24 /ml



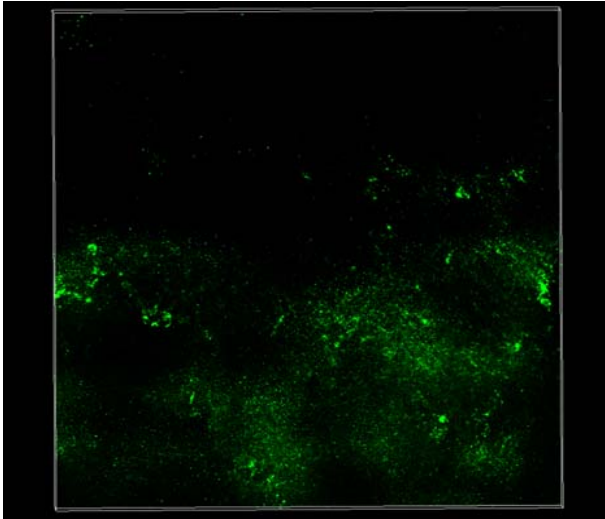
100 ng HIV-1 p24/ml

Unpublished results

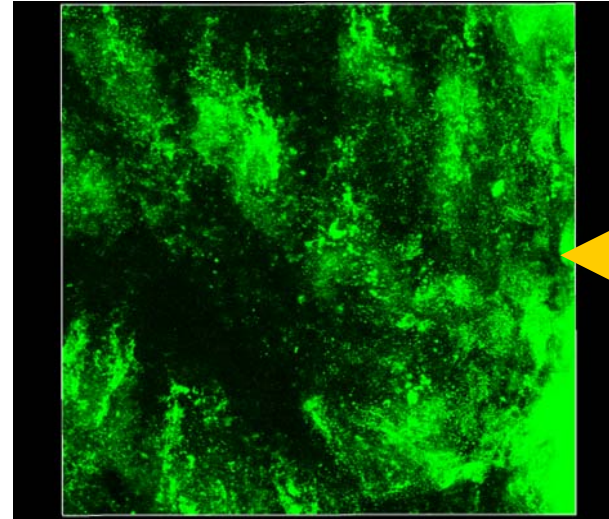


# HIV-1 triggers TSLP in vaginal epithelium

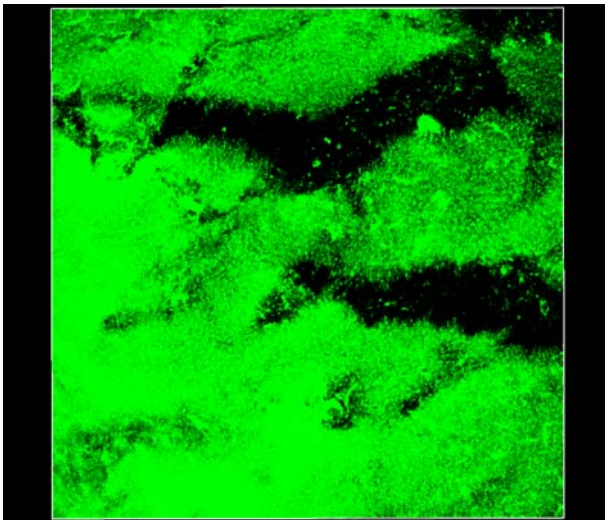
Baseline



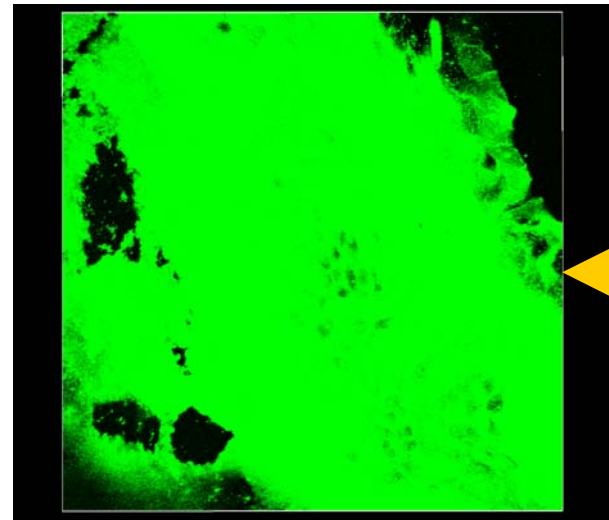
293T cell supernatant



Day 5



1 ng HIV-1 p24 /ml

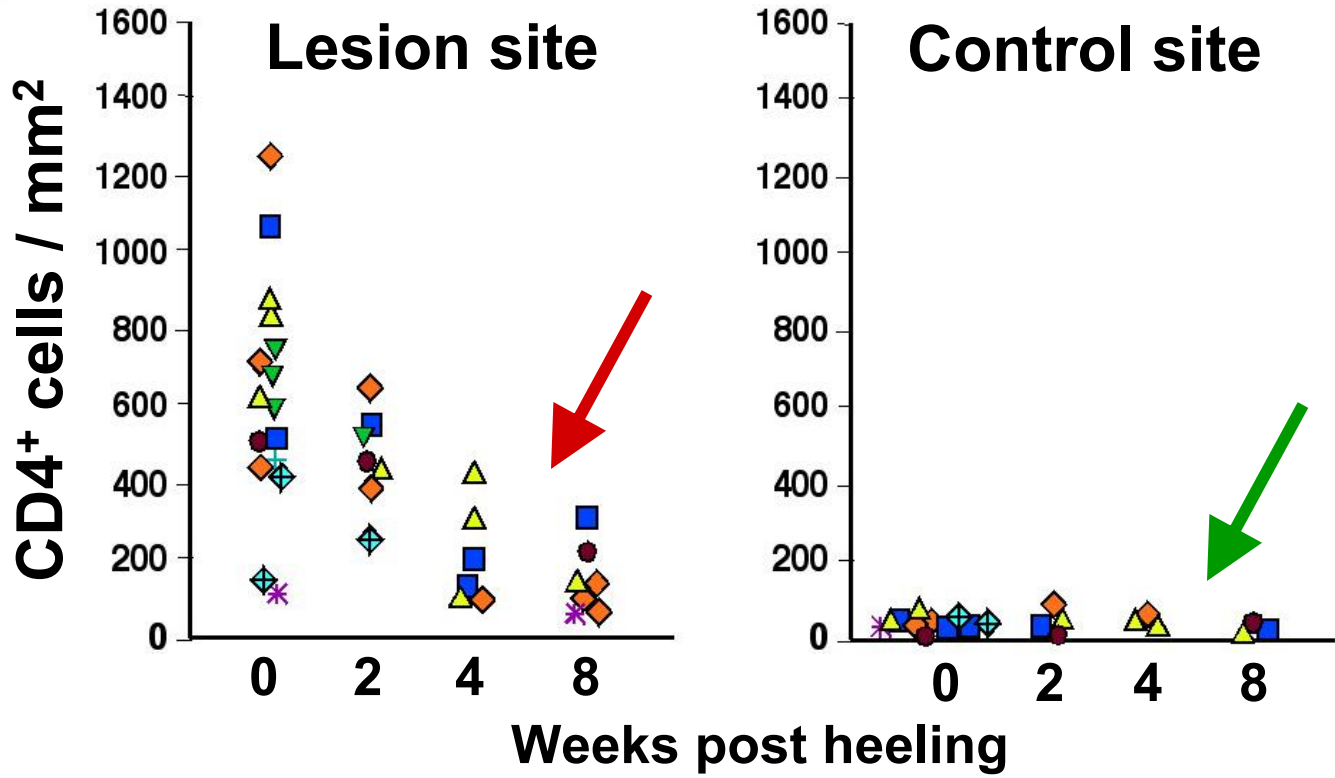


100 ng HIV-1 p24/ml

Unpublished results

# 4. Long-lasting effects of co-existing STDs

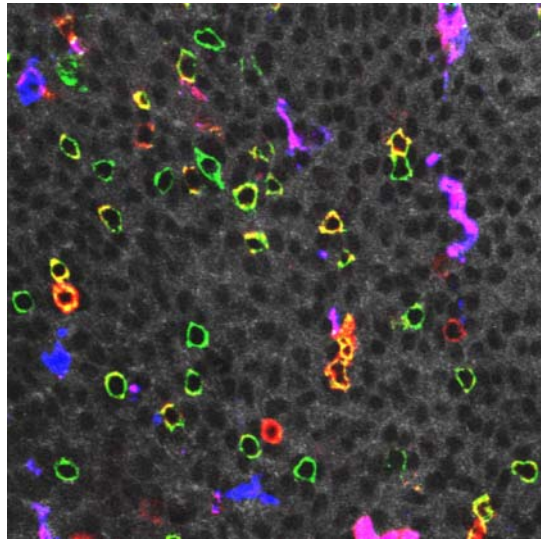
## Persistence of CD4<sup>+</sup> T cells in genital herpes simplex lesions after healing



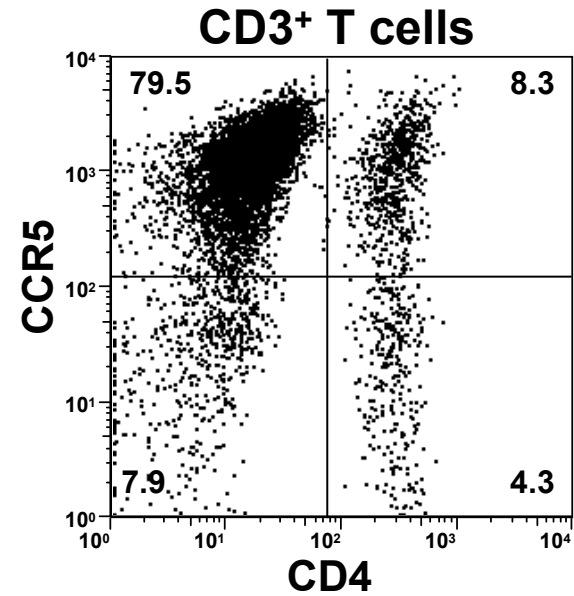
# Contribution of the various HIV invasion routes to HIV cases worldwide

<b>Female genital tract</b>	<b>12.6 million</b>
<b>Male genital tract</b>	<b>10.2 million</b>
<b>Intestinal tract</b>	<b>7.3 million</b>
<b>Placenta</b>	<b>0.5 million</b>
<b>Blood stream</b>	<b>2.6 million</b>

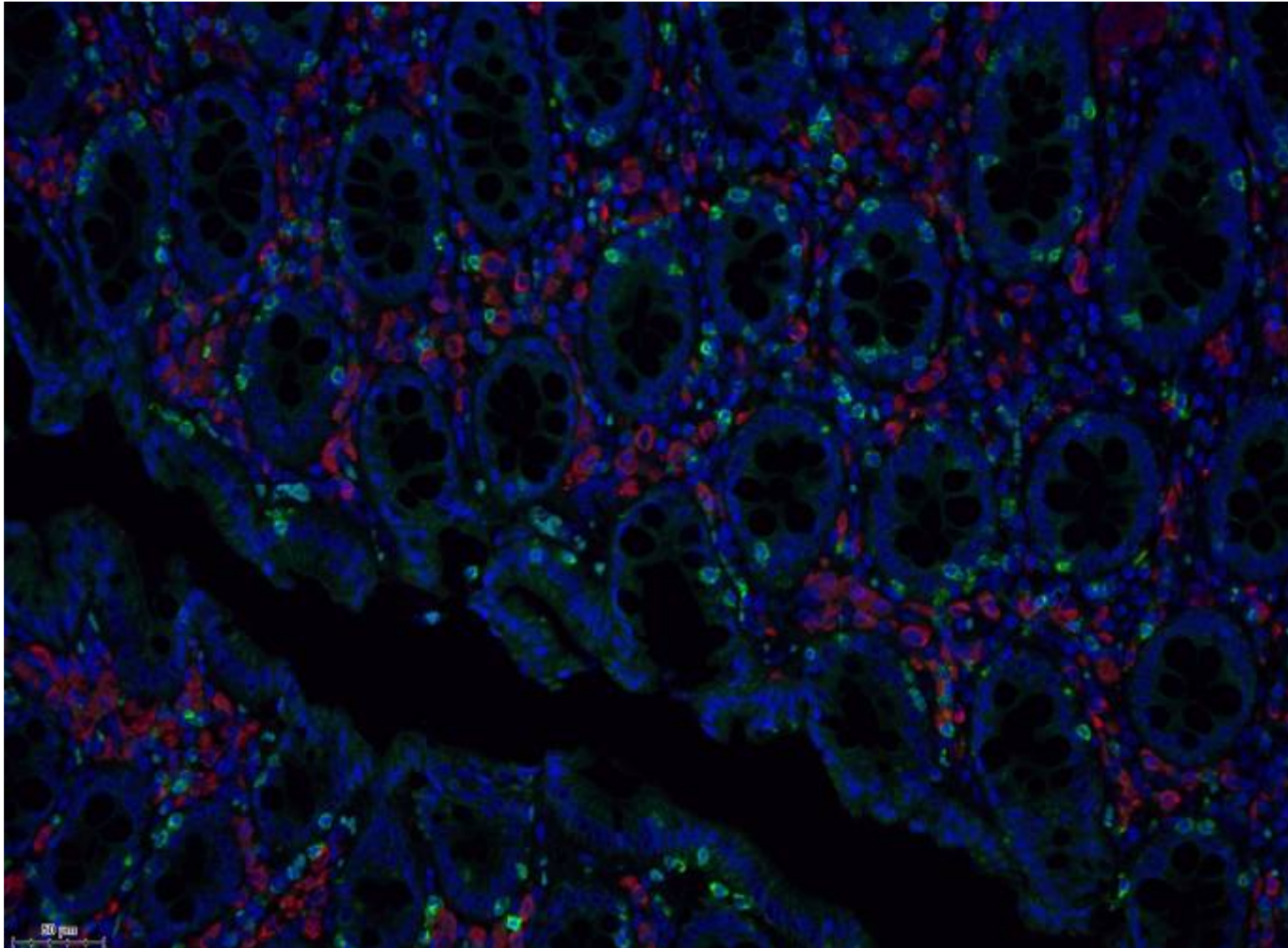
# CCR5 expression in the vaginal epithelium



CD3  CD1a  
CCR5



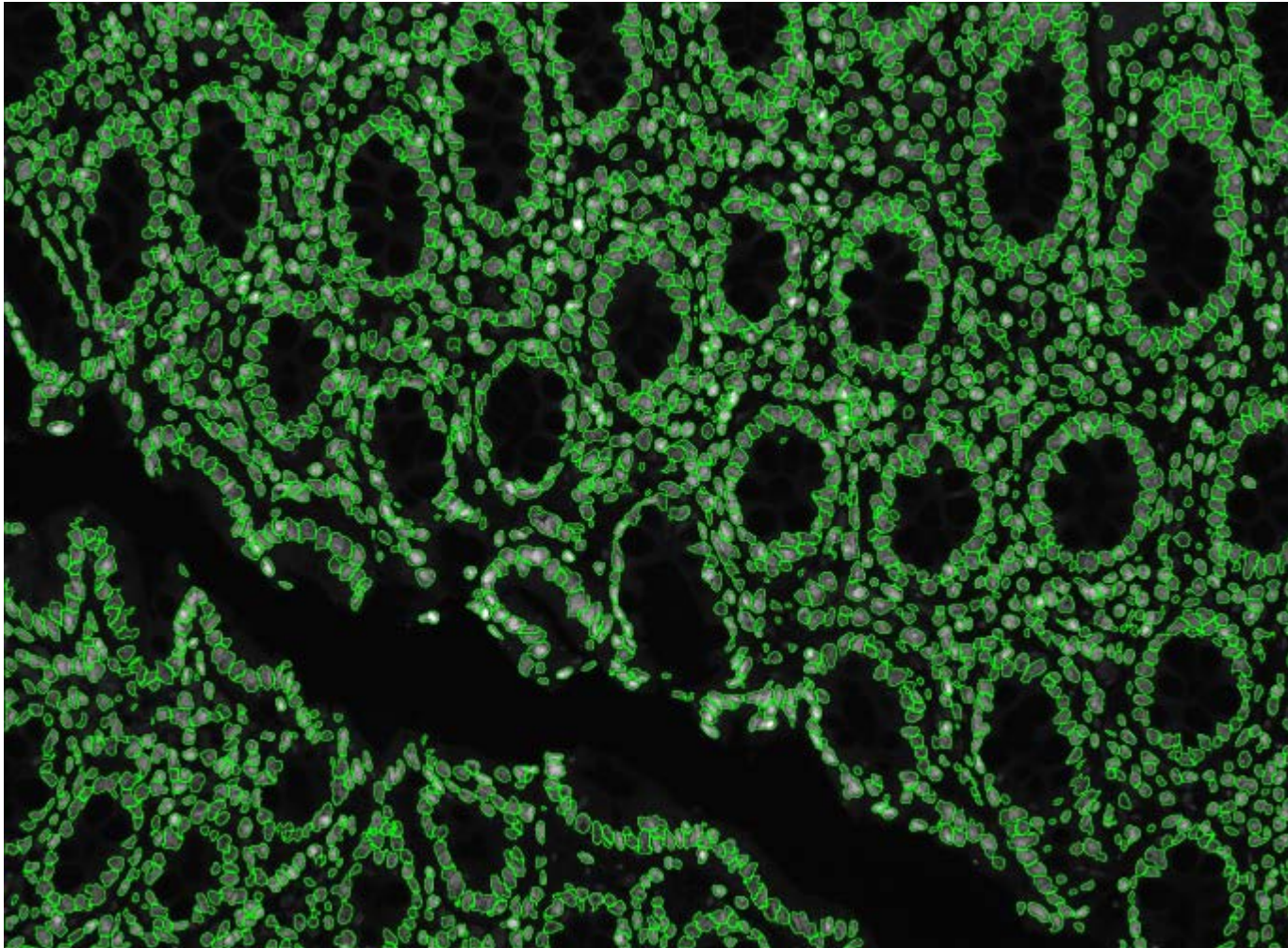
# CCR5 expression in the rectal mucosa



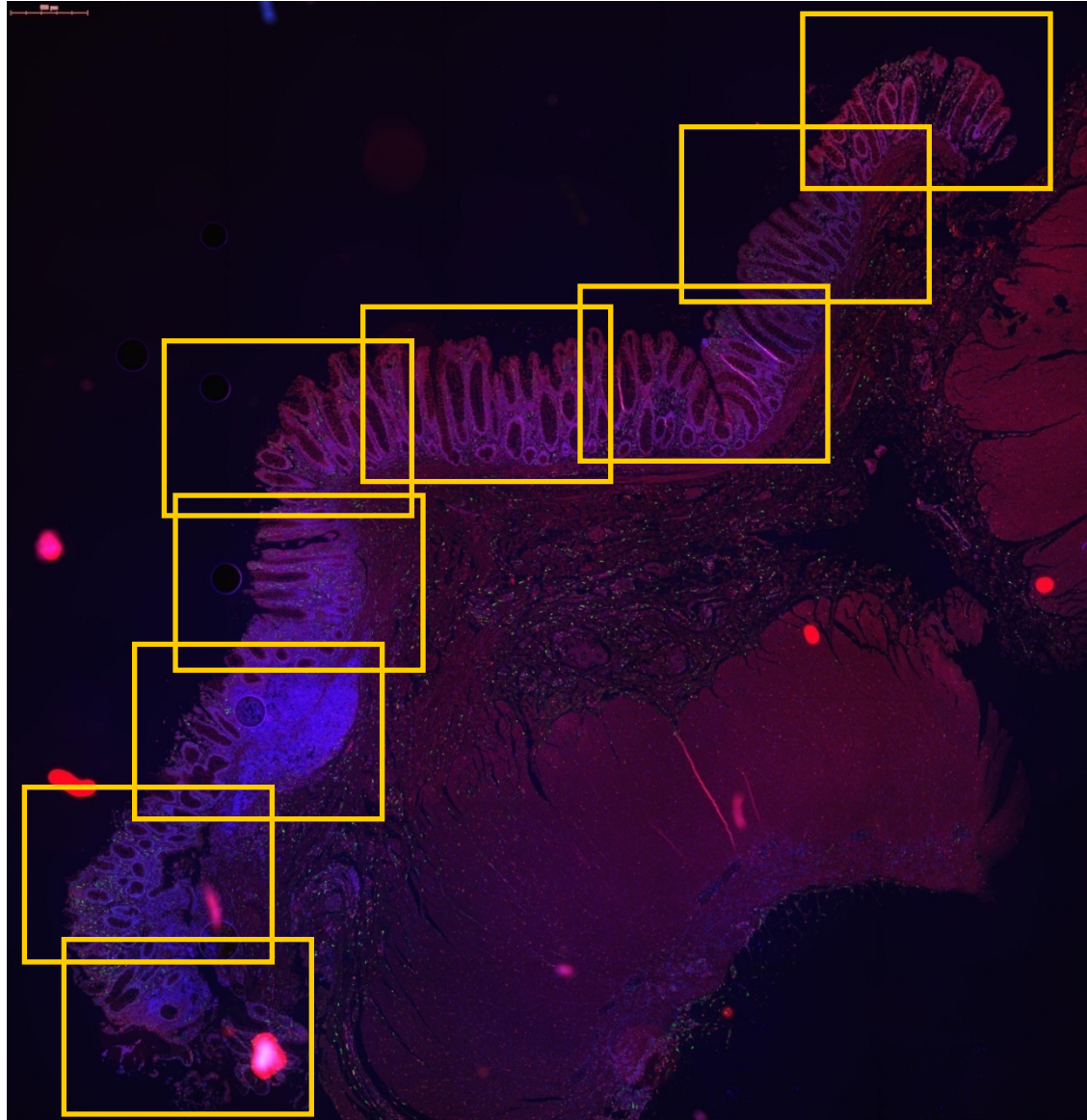
CCR5  
green

CD68  
red

# Computer-aided analysis of immunohistology

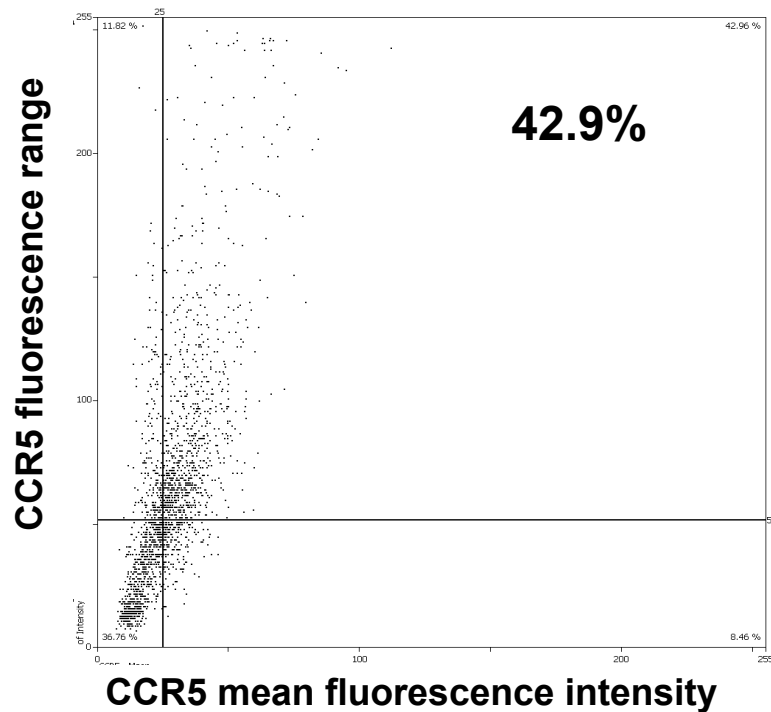


# Stitching individual images together

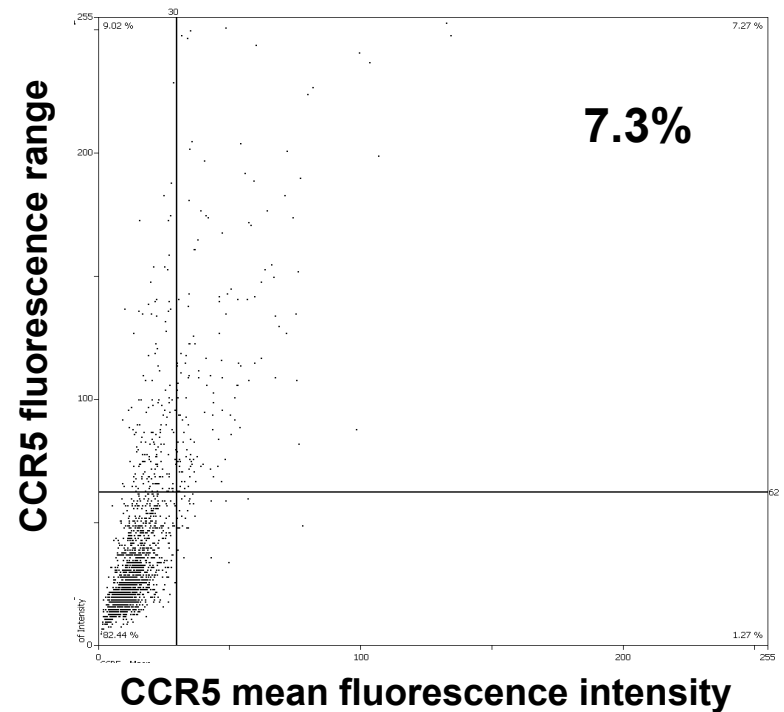


# CCR5 expression on rectal T cells and macrophages *in situ*

## CCR5 on CD3<sup>+</sup> T cells



## CCR5 on CD68<sup>+</sup> macrophages





# Conclusion and thoughts

- **Independent of the anatomical location, prevention of productive infection of CD4<sup>+</sup> T lymphocytes residing in the mucosa is the most important goal**
- **However, alternative routes of HIV entry, indirect effects of exposure to HIV and the influence of pathogenic co-factors on HIV infection afford the virus a potential edge and may vary between anatomical sites**

# Acknowledgements

Lamar Ballweber  
Allison Kreger  
Kimberly Smythe  
Barry Robinson

Julie McElrath

Jia Zhu  
Larry Corey

Jagannadha Sastry  
Danielle Fontenot

David Eschenbach  
Gretchen Lentz  
Michael Fialkow

Funding Agencies: NIH NICHD & NIAID