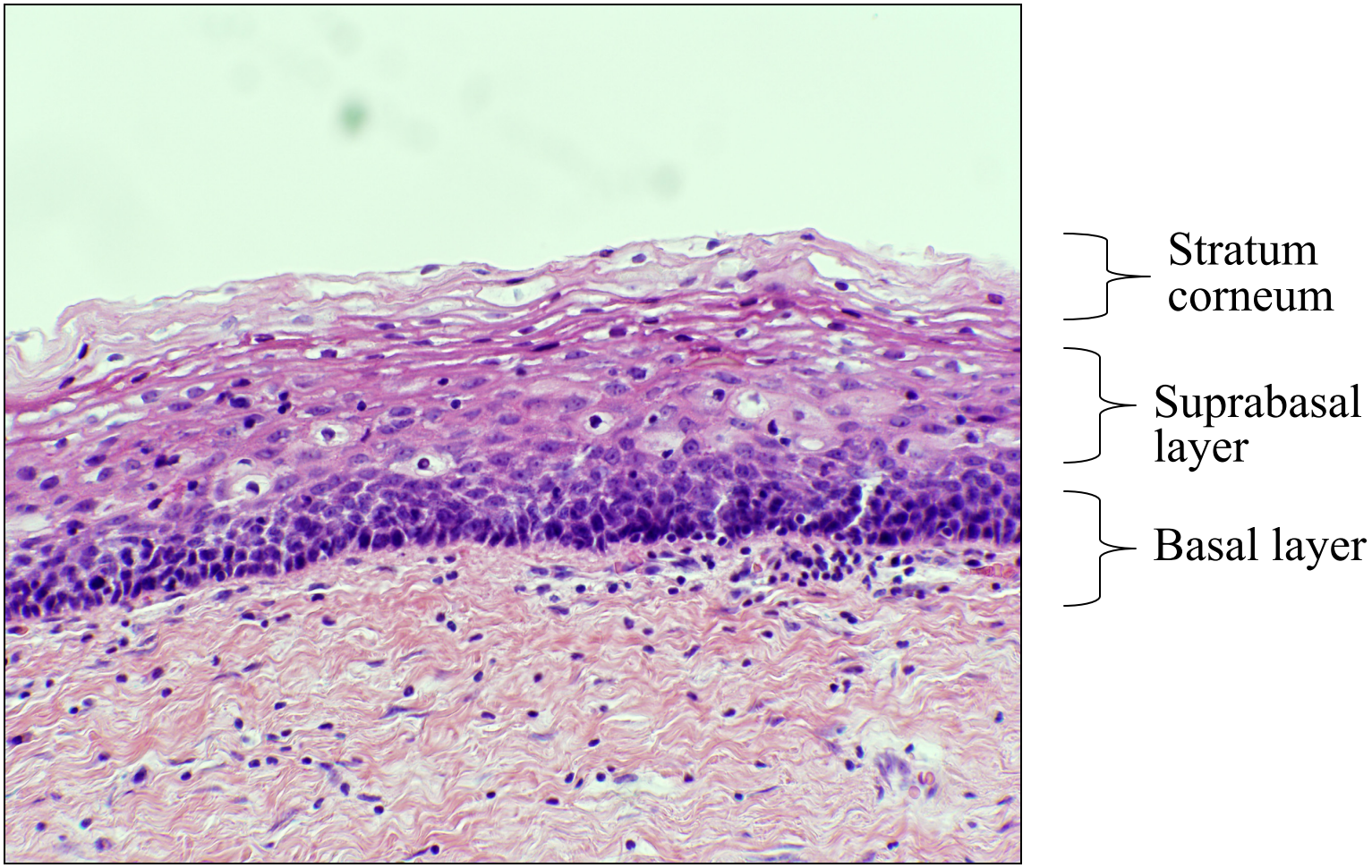
Vaginal Stratum Corneum: Demilitarized Zone or Battlefront?

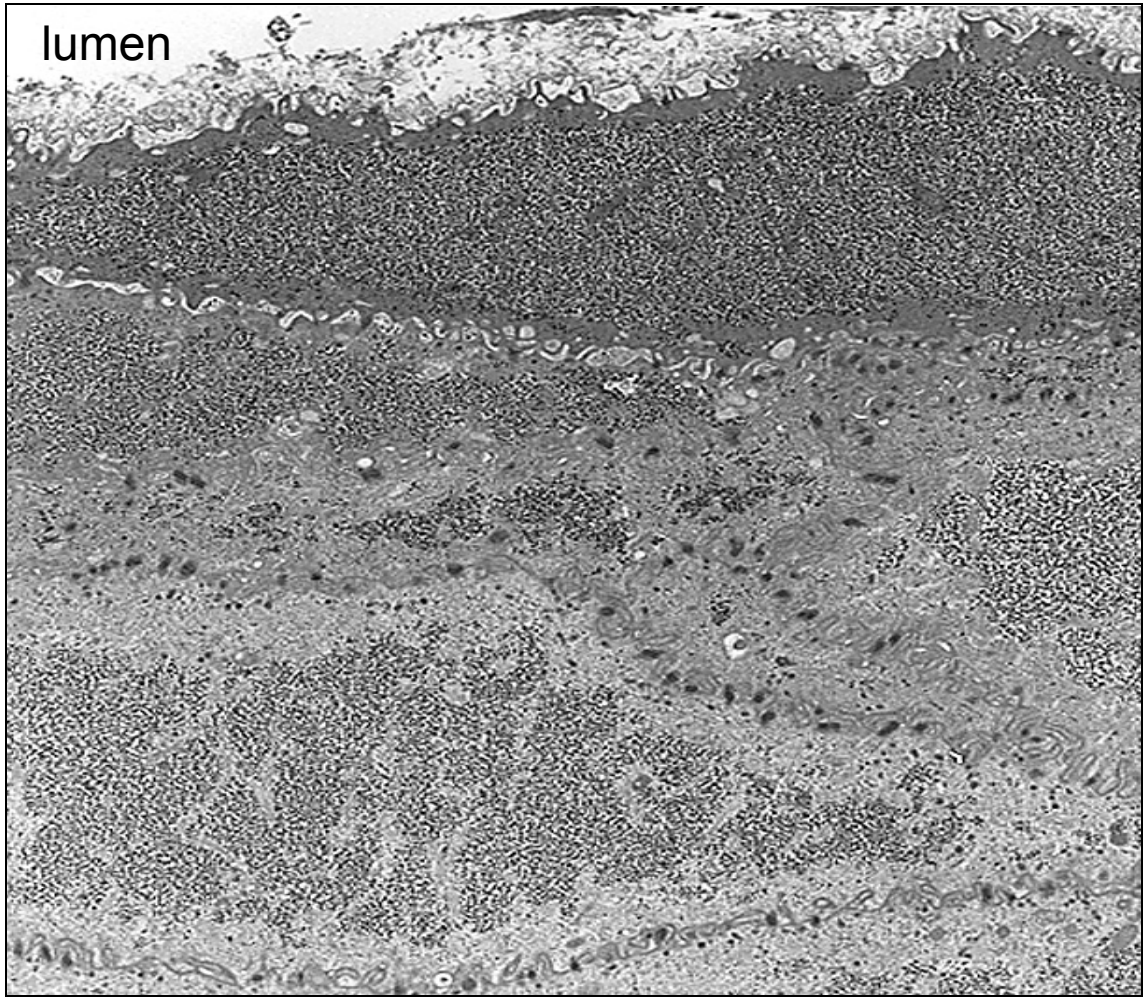
Deborah Anderson

Boston University School of Medicine
Boston, MA, USA

Vaginal Epithelium

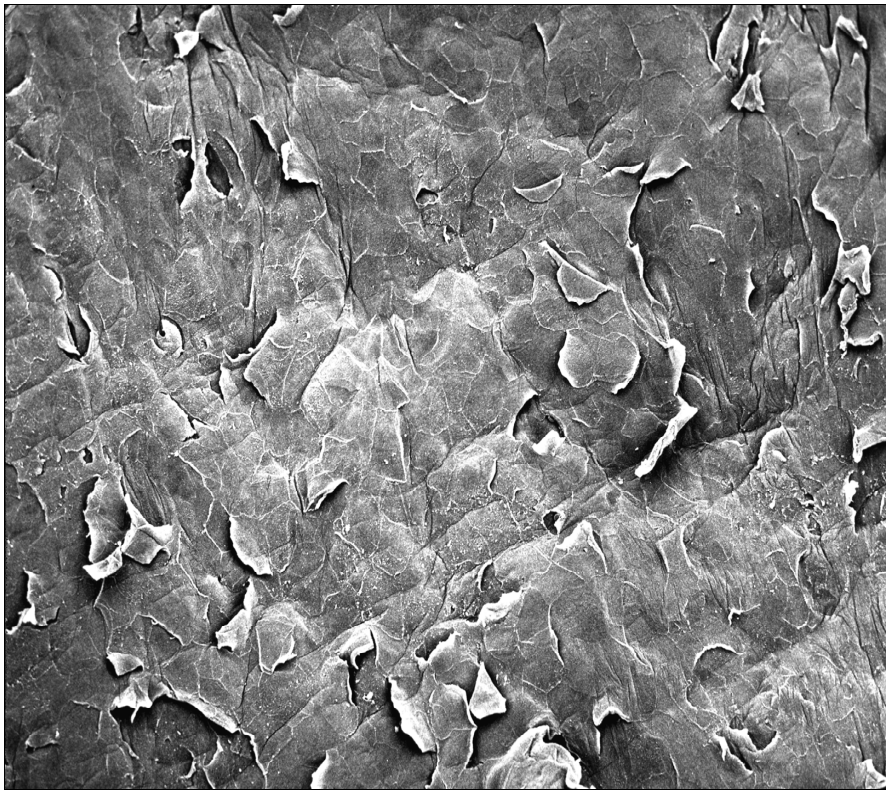


Epithelial Cells in Vaginal Stratum Corneum (Corneocytes)

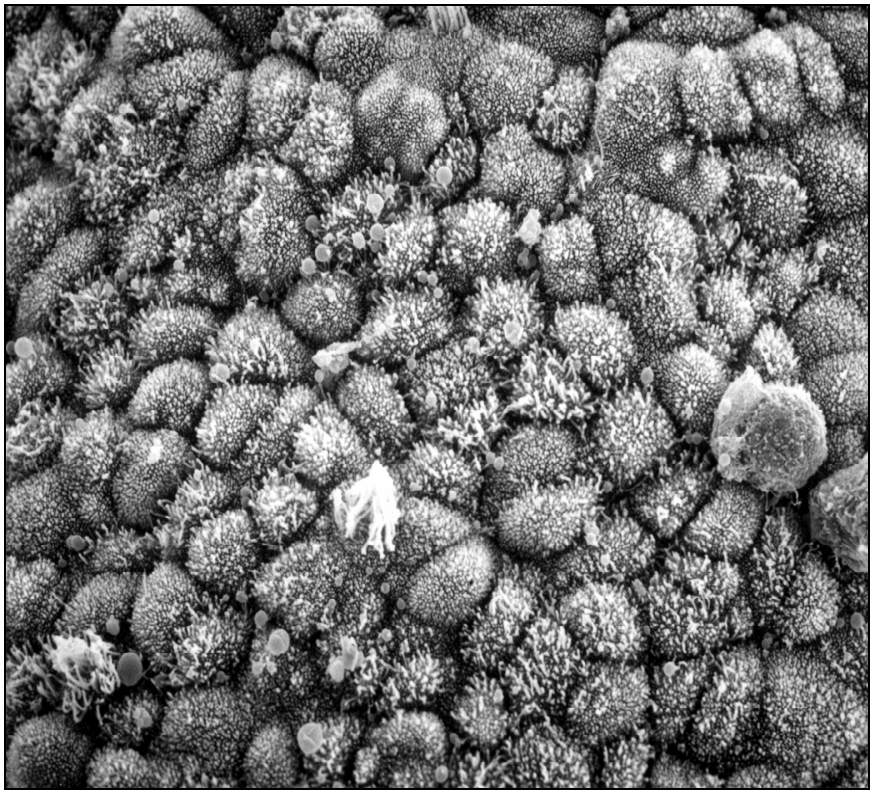


- lose nuclei and organelles
- glycogen-filled
- no tight junctions
- continuous turnover of apical cells

Vagina and Endocervix: Comparison of Apical Surfaces



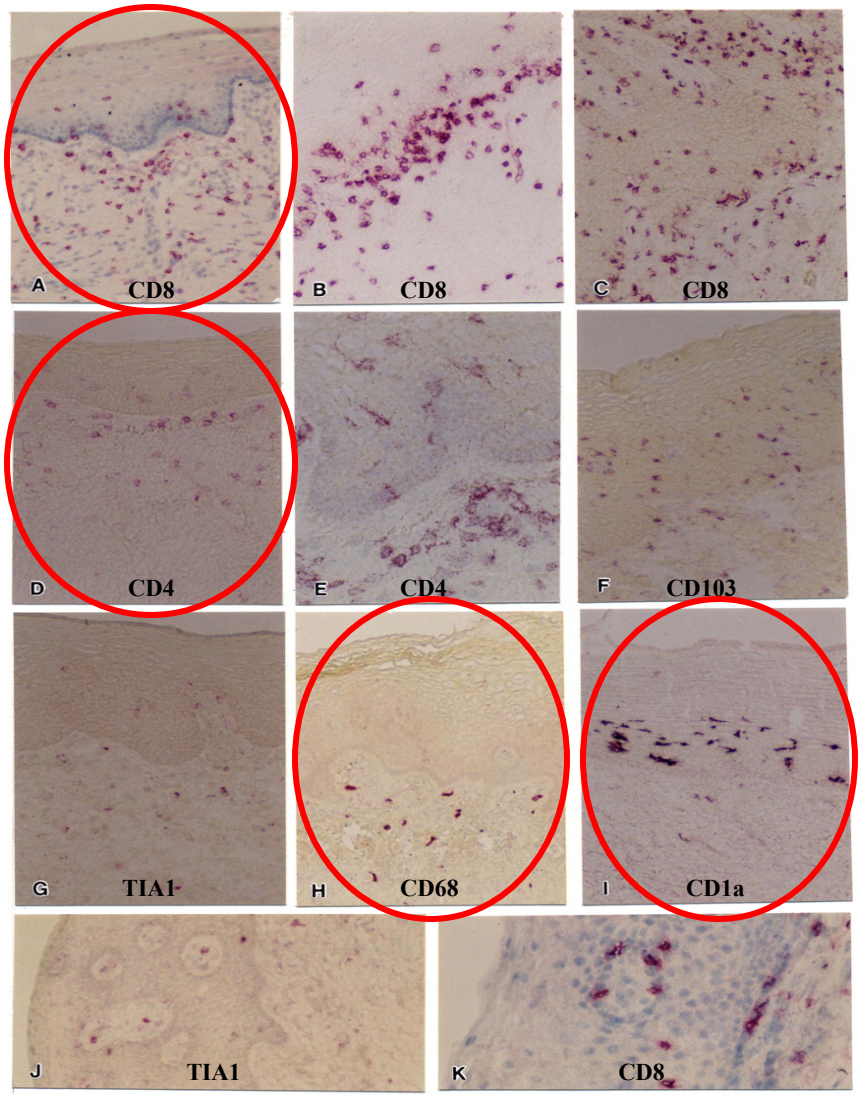
Vagina



Endocervix

Stratum Corneum: Demilitarized Zone?

Immune Cells in the Vaginal Epithelium

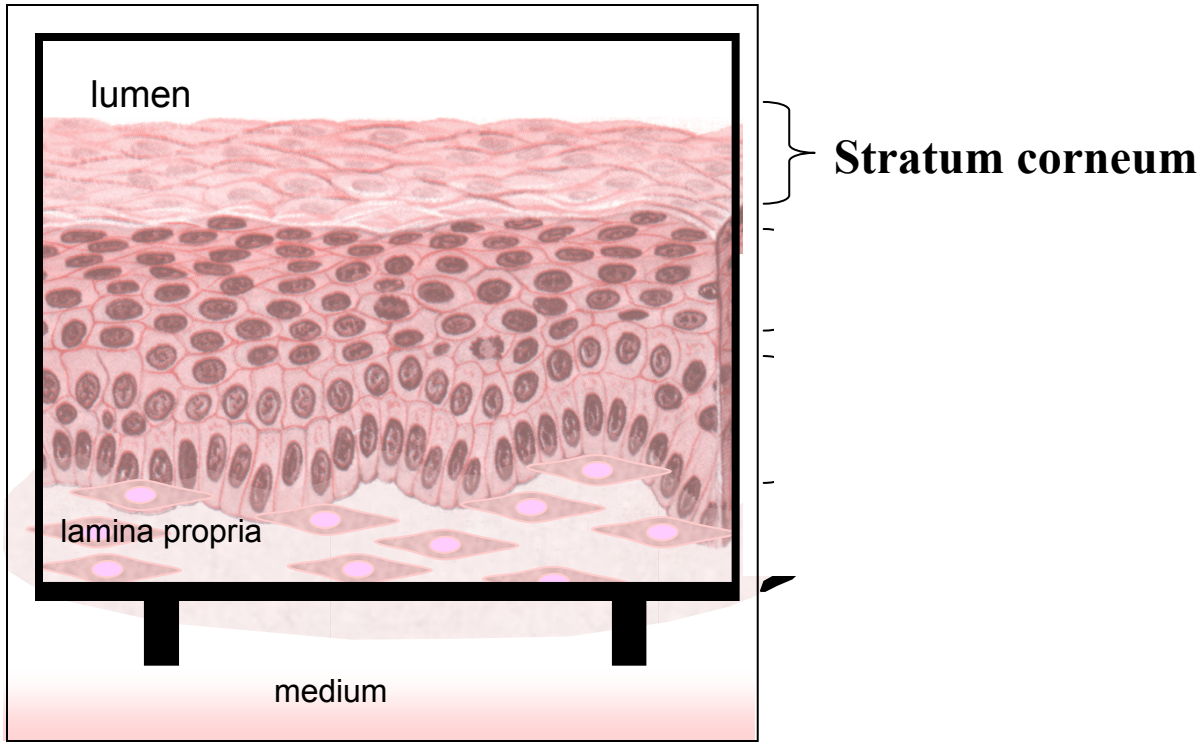


Few immune cells are present in the healthy SC

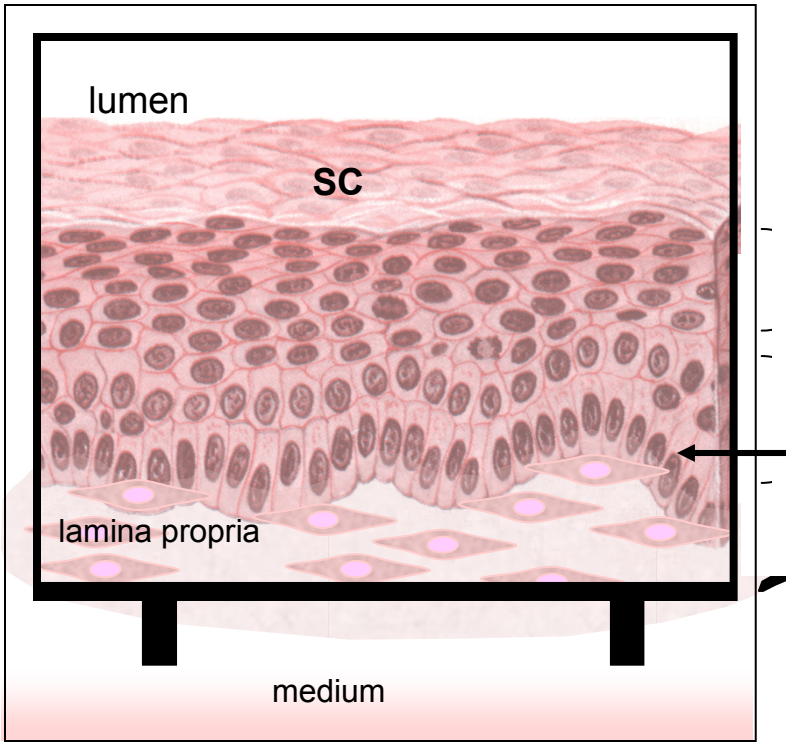
Pudney J, Quayle AJ, and Anderson DJ
Biol Reprod 2005;73:1253-1263

Does the vaginal stratum corneum prevent stimulation of basal epithelial cells by products of luminal bacteria?

MatTek VEC culture



MatTek VEC culture



Basal epithelial cells express TLRs 1/2, 3, 5

(notably they do not express TLR4, the LPS receptor)

Primary and immortalized cultures of vaginal basal epithelial cells produce proinflammatory cytokines and chemokines when stimulated with TLR agonists

Il-6

Il-8

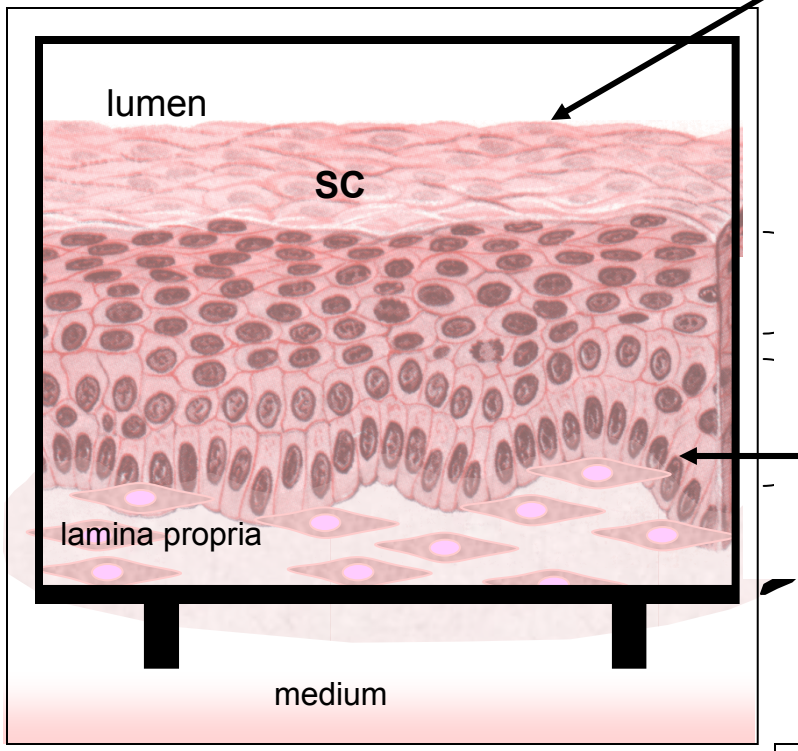
TNF-alpha

RANTES

MIP-1alpha

These products recruit and activate immune cells

MatTek VEC culture



TLR agonists added to **apical** surface

FSL

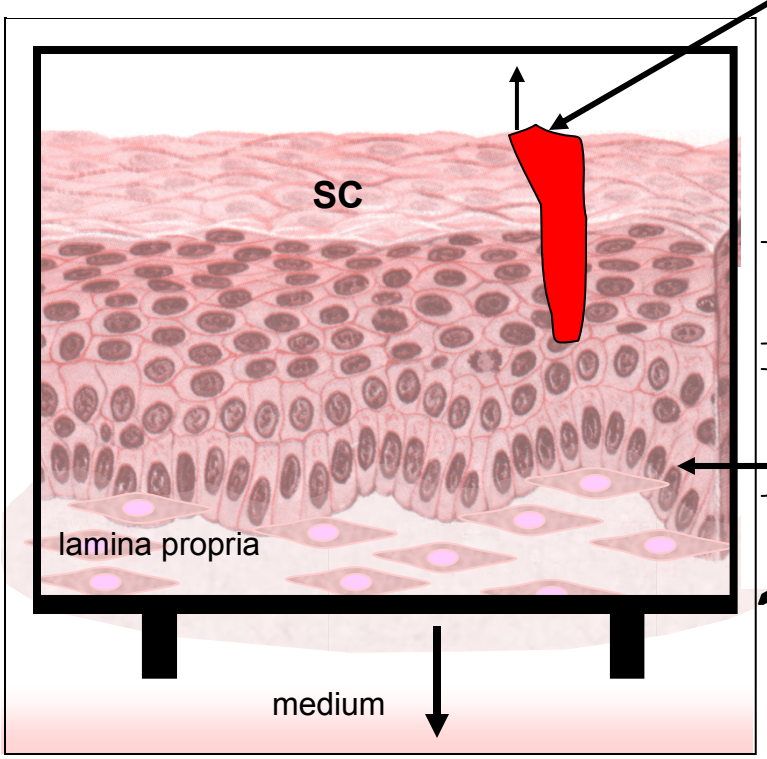
Poly (I:C)

Flagelin

Basal epithelial cells express TLRs 1/2, 3, 5

No cytokines detected in apical or basal supernatants

MatTek VEC culture



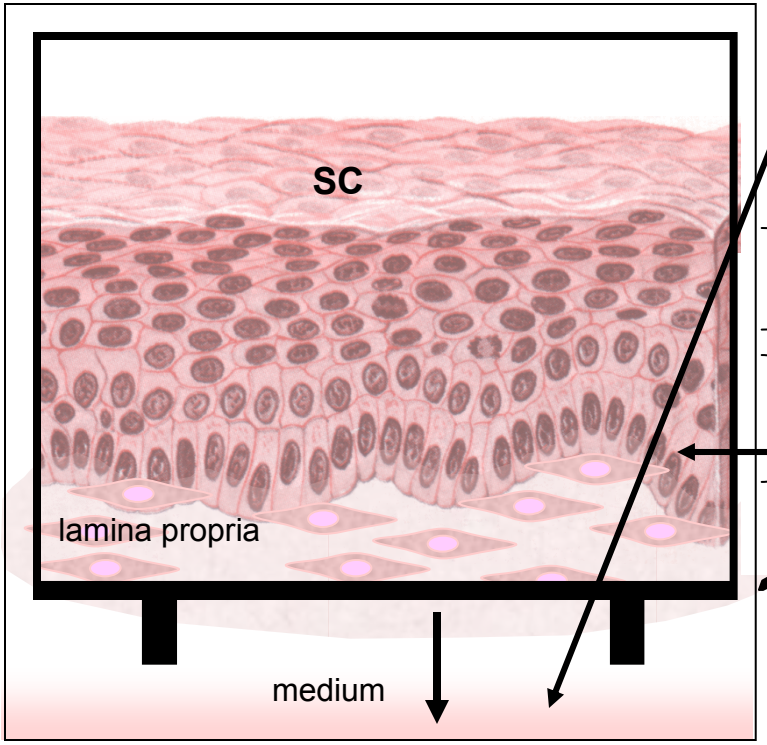
TLR agonists added to **scarified** surface

FSL
Poly (I:C)
Flagelin

Basal epithelial cells express TLRs 1/2, 3, 5

Cytokines detected in **apical and basolateral** supernatant

MatTek VEC culture



TLR agonists added to **basolateral** surface

FSL
Poly (I:C)
Flagelin

Basal epithelial cells express TLRs 1/2, 3, 5

Cytokines detected in **basolateral** supernatant

Evidence that the healthy intact vaginal stratum corneum is a passive protective barrier

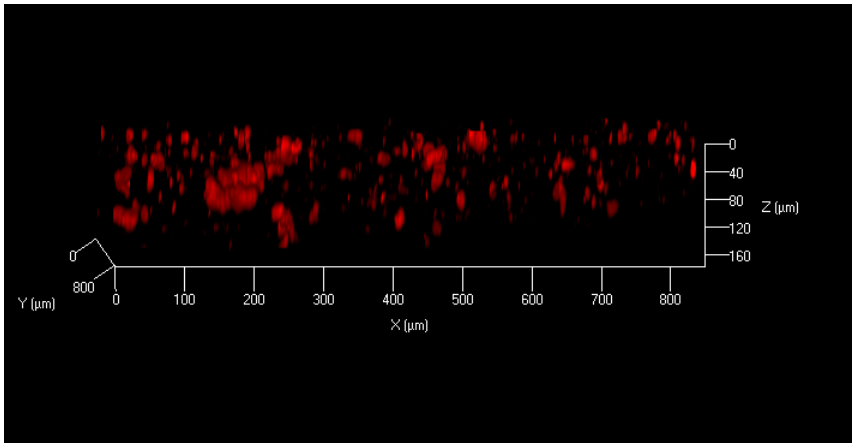
- Does not contain many immune cells

- Protects basal epithelial cells from stimulation with products from endogenous luminal bacteria

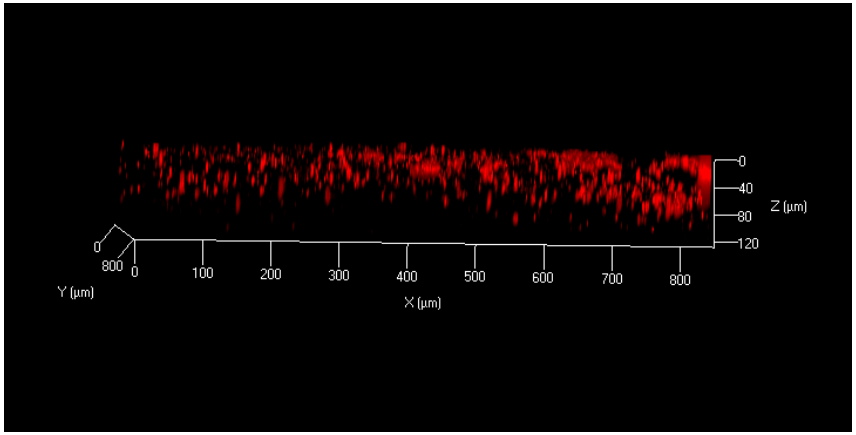
Stratum Corneum: Battlefront

Can pathogens infiltrate the stratum corneum?

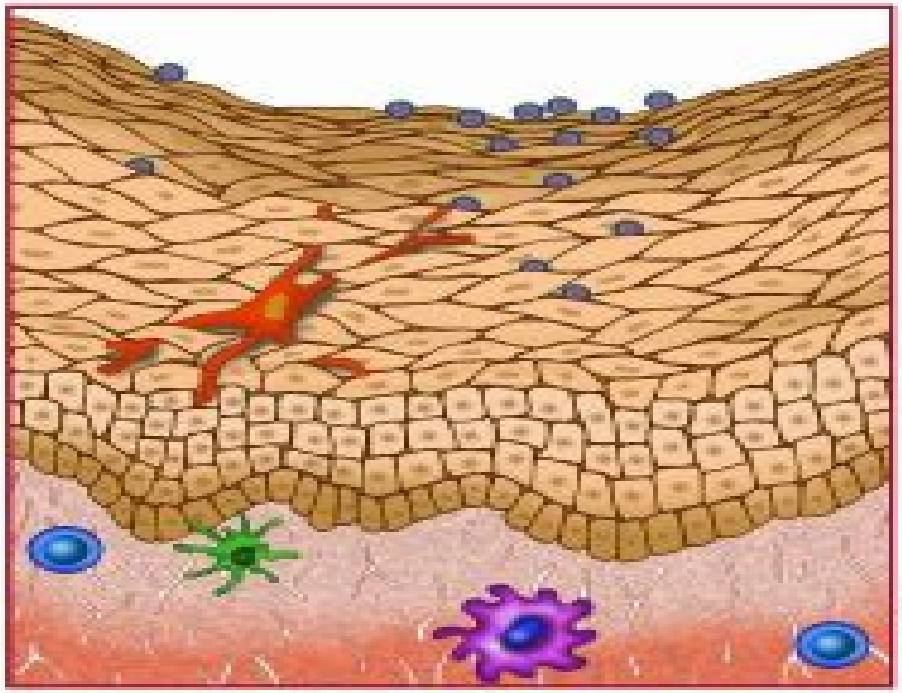
Infiltration of Stratum Corneum by Inert Beads



0.1um (virus)



1um (bacterium)



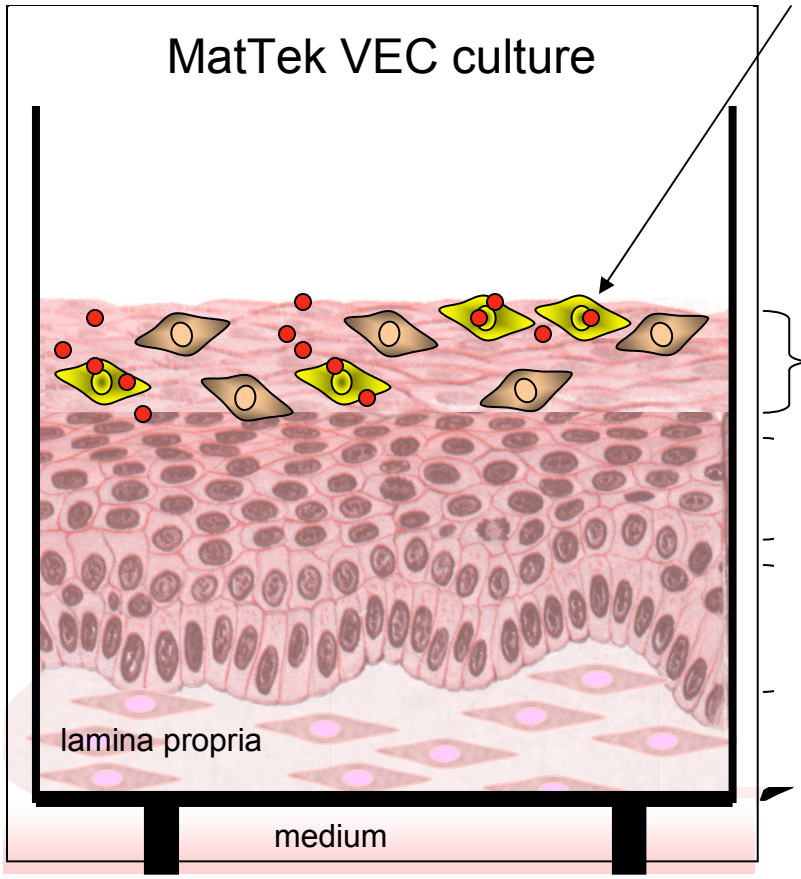
HIV enters the vaginal stratum corneum where it may encounter Langerhans cells and transmit infection.

Thomas Hope 2009

Question:

How long does HIV remain viable (infectious) in the stratum corneum?

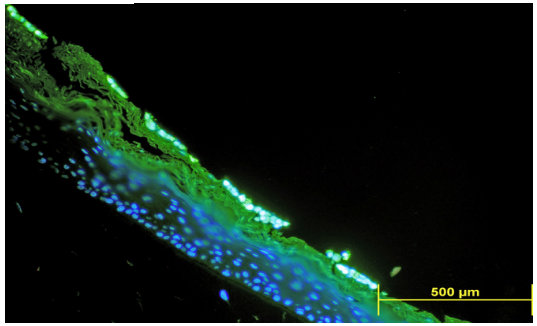
Assessment of HIV infectivity in the Stratum Corneum



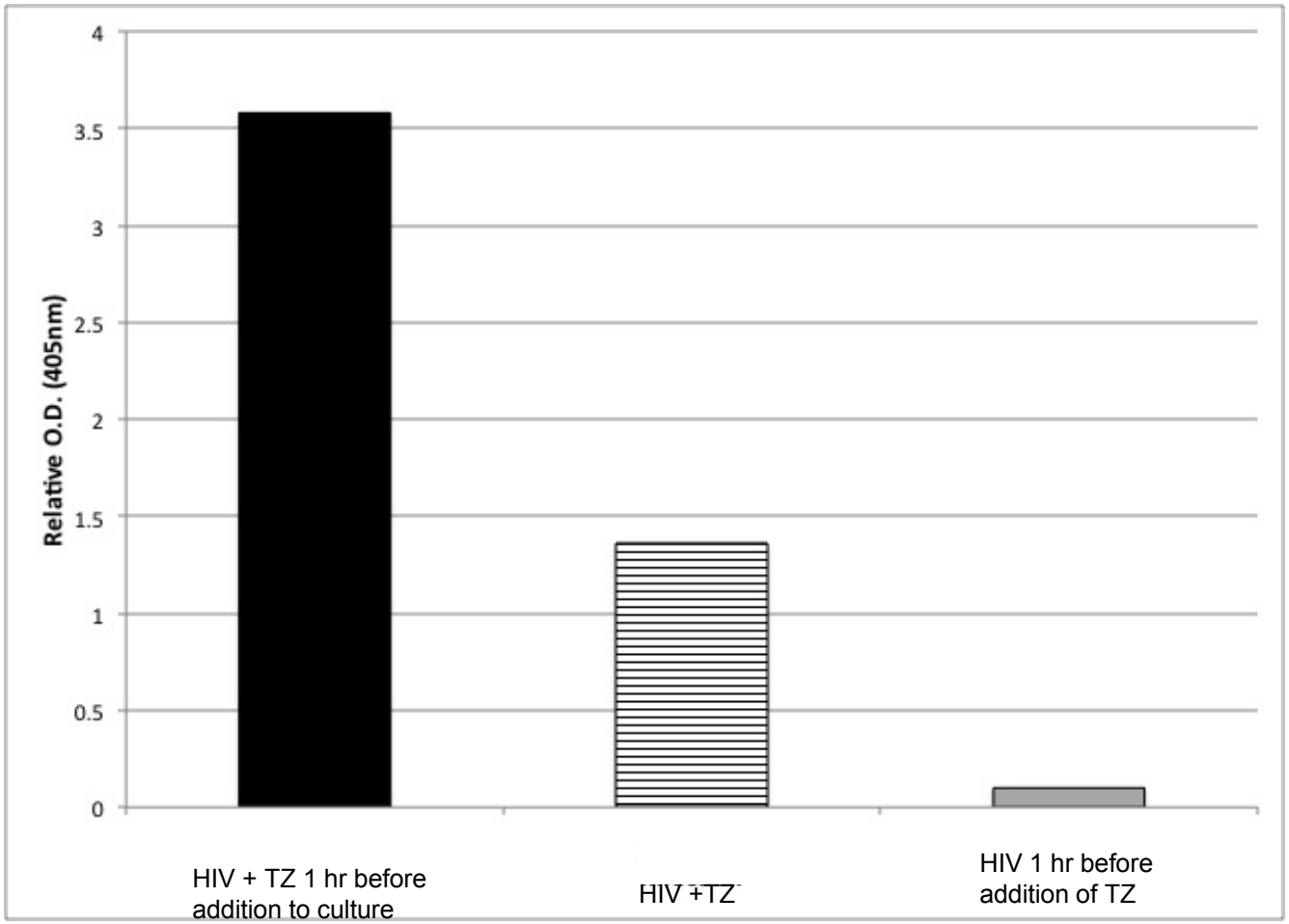
TZM-bl (HIV indicator) cells were added to the apical surface of VEC cultures to assess infectious HIV.

Stratum corneum

TZM-bl cells on SC

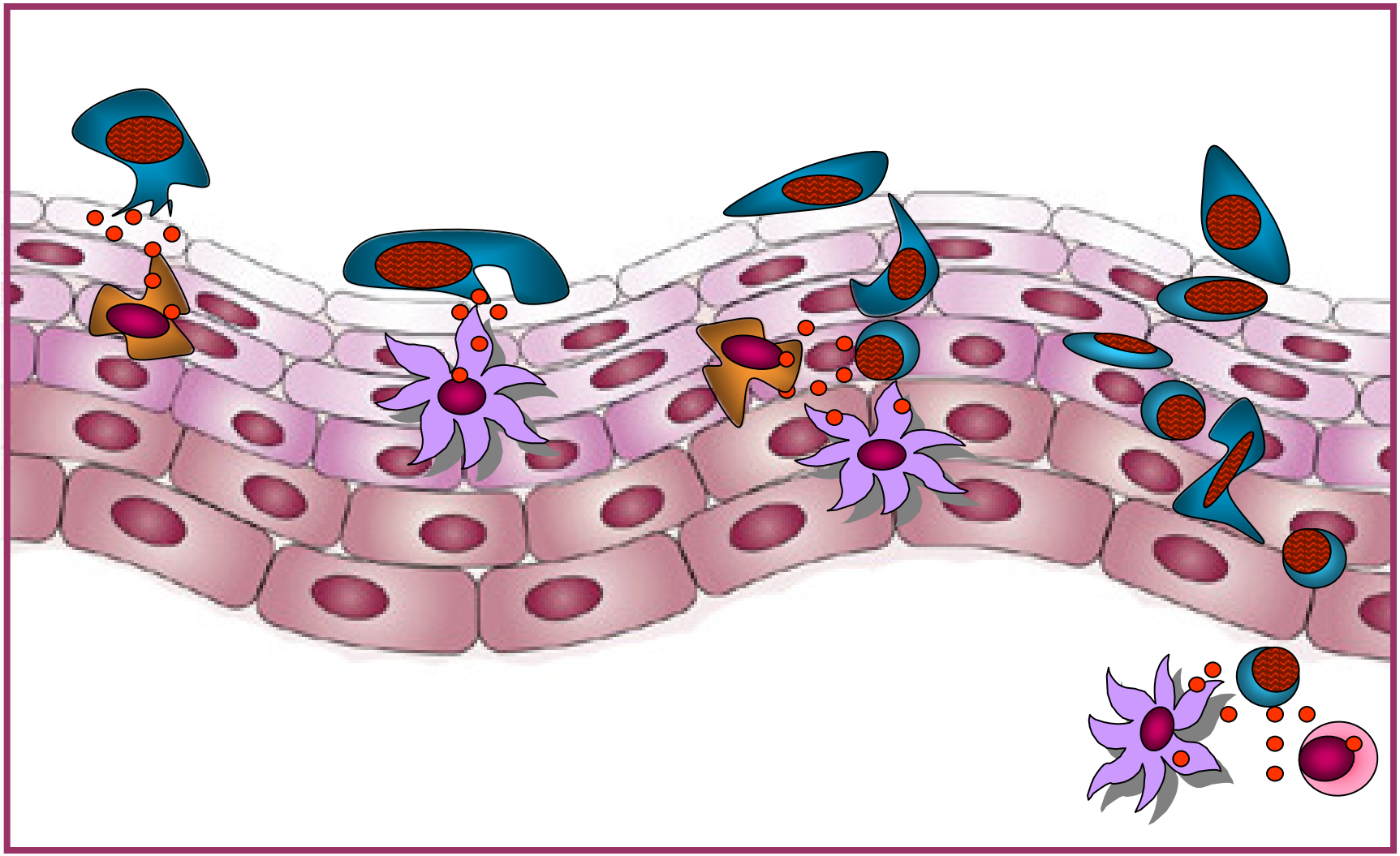


HIV Infection of TZM-bl cells on Vaginal Surface

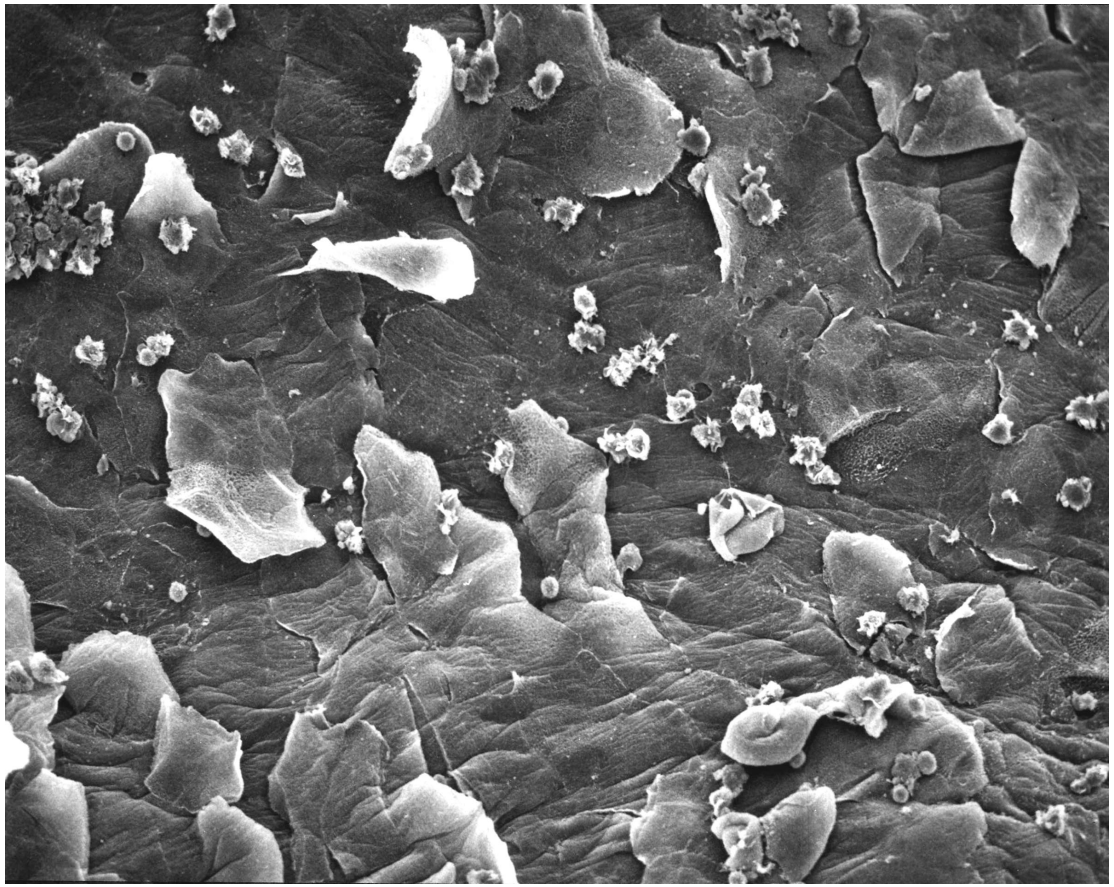


Can HIV-infected cells infiltrate the stratum corneum from the apical side?

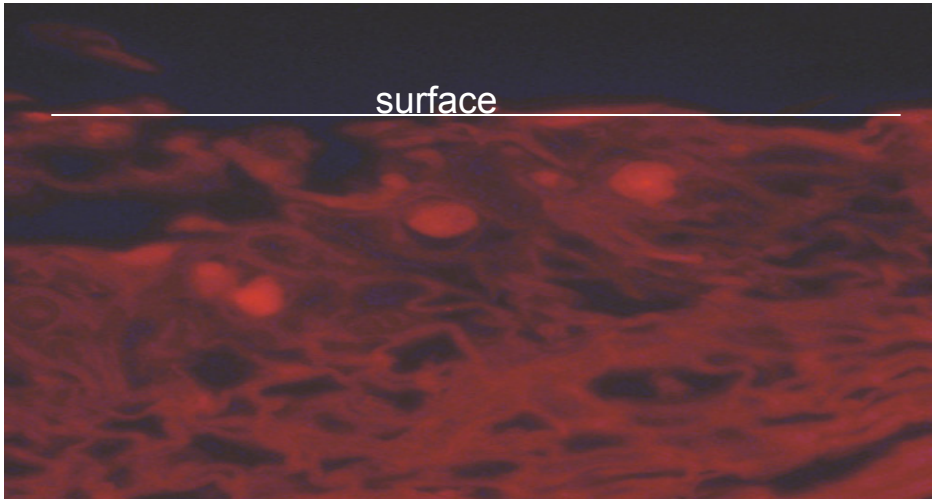
Cell-Associated HIV Transmission



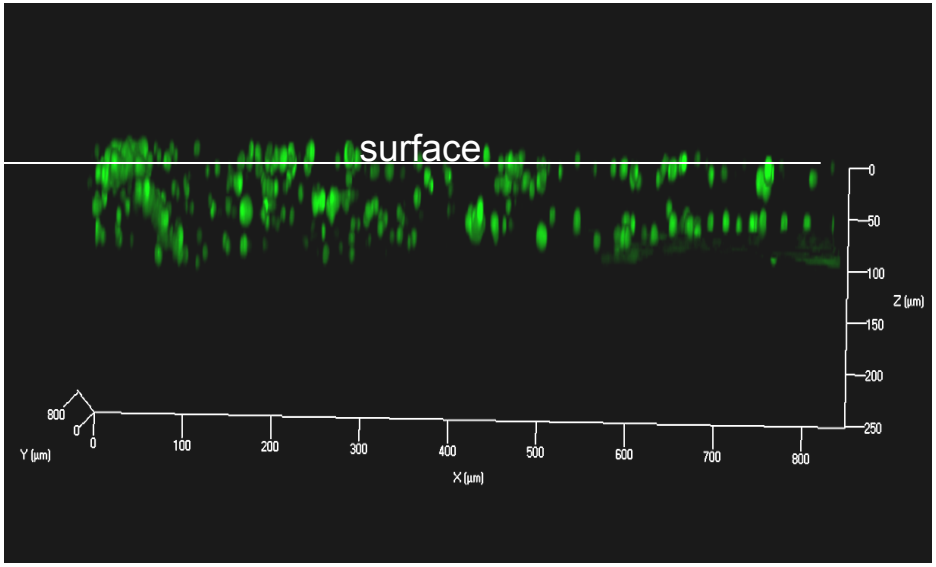
PBMCs Adhere to Vaginal Stratum Corneum



Apical to basal leukocytic infiltration of the the vaginal stratum corneum



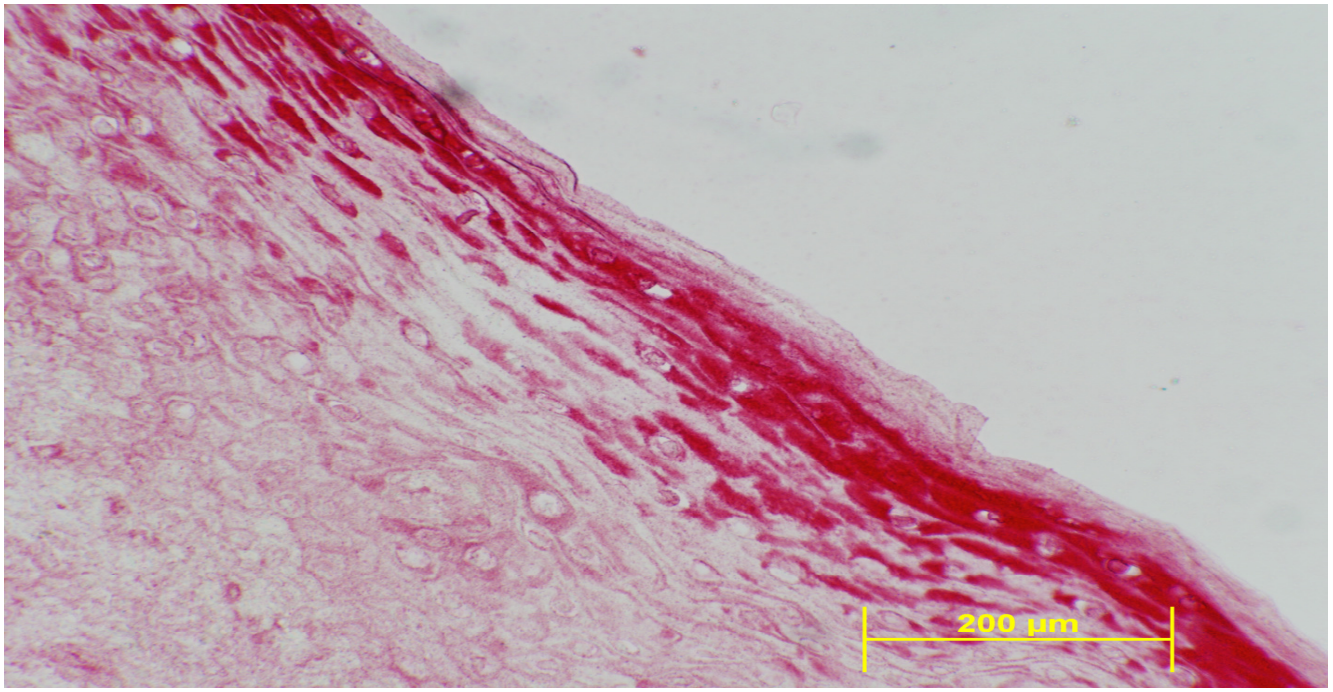
Macrophages



Seminal leukocytes

Does the vaginal stratum corneum retain soluble immunological mediators?

Igs in the Vaginal Stratum Corneum



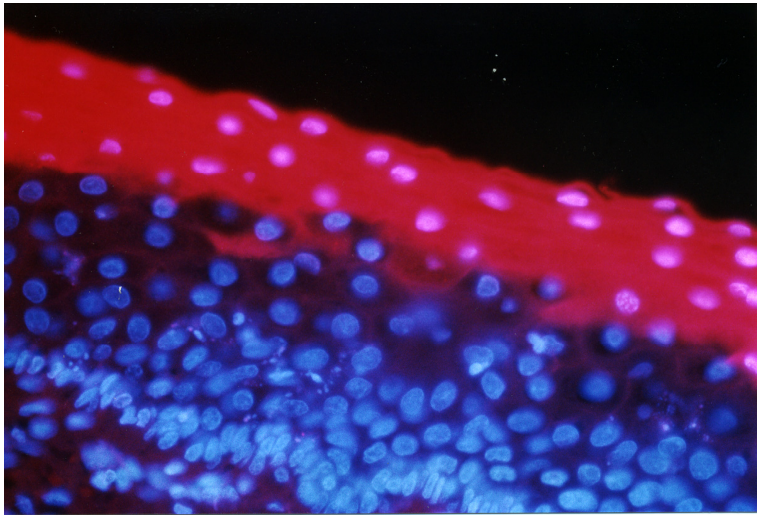
IgG

IgA

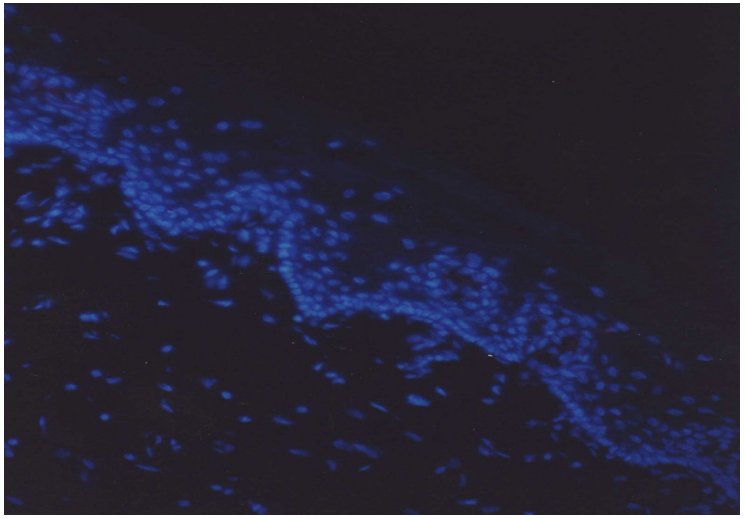
IgM

- **Does the SC absorb Igs from the lumen?**
- **Do antibodies retain their activity in the SC?**
- **Time course of Ig retention in the SC?**

IgG Uptake by Apical Vaginal Epithelial Cells



IgG-Cy3



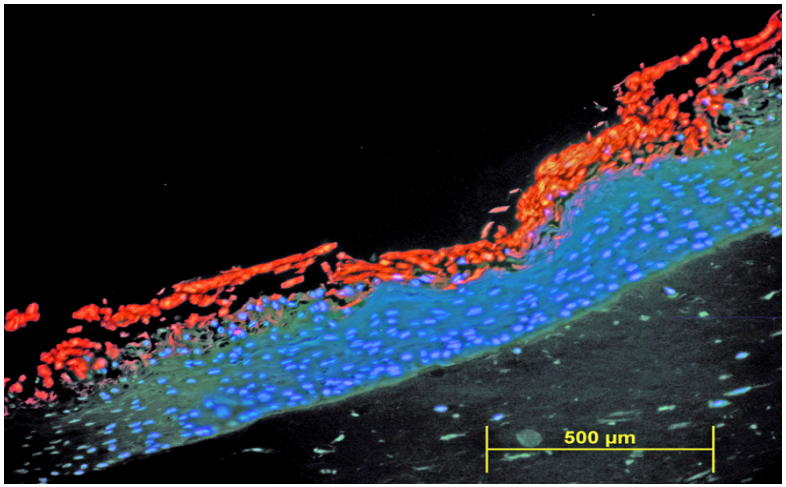
Negative Control

Evidence that Ig Uptake is not receptor mediated

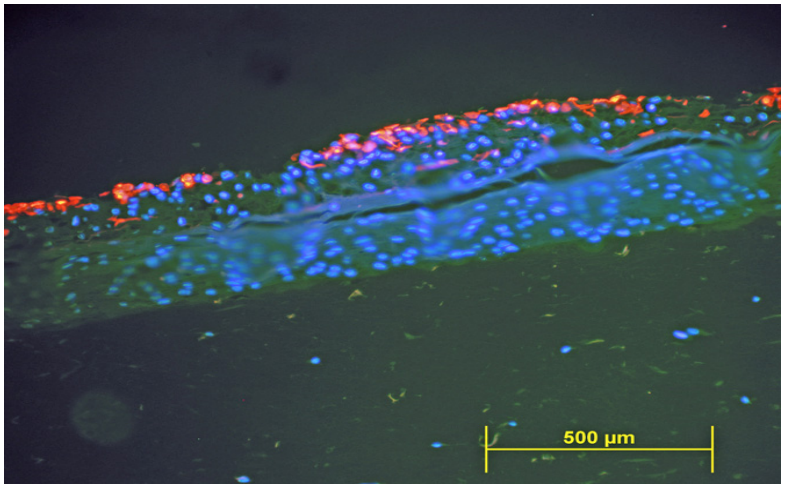
- Absorption and retention occurs at 4ºC
- Absorption and retention of IgY
- Ig receptors not detected in stratum corneum
- The stratum corneum absorbs and retains other types of immune mediators
  - lysozyme
  - type-1 defensins

Do antibodies retain activity in the stratum corneum?

Retention of HSV-Cy3 mab by vaginal stratum corneum

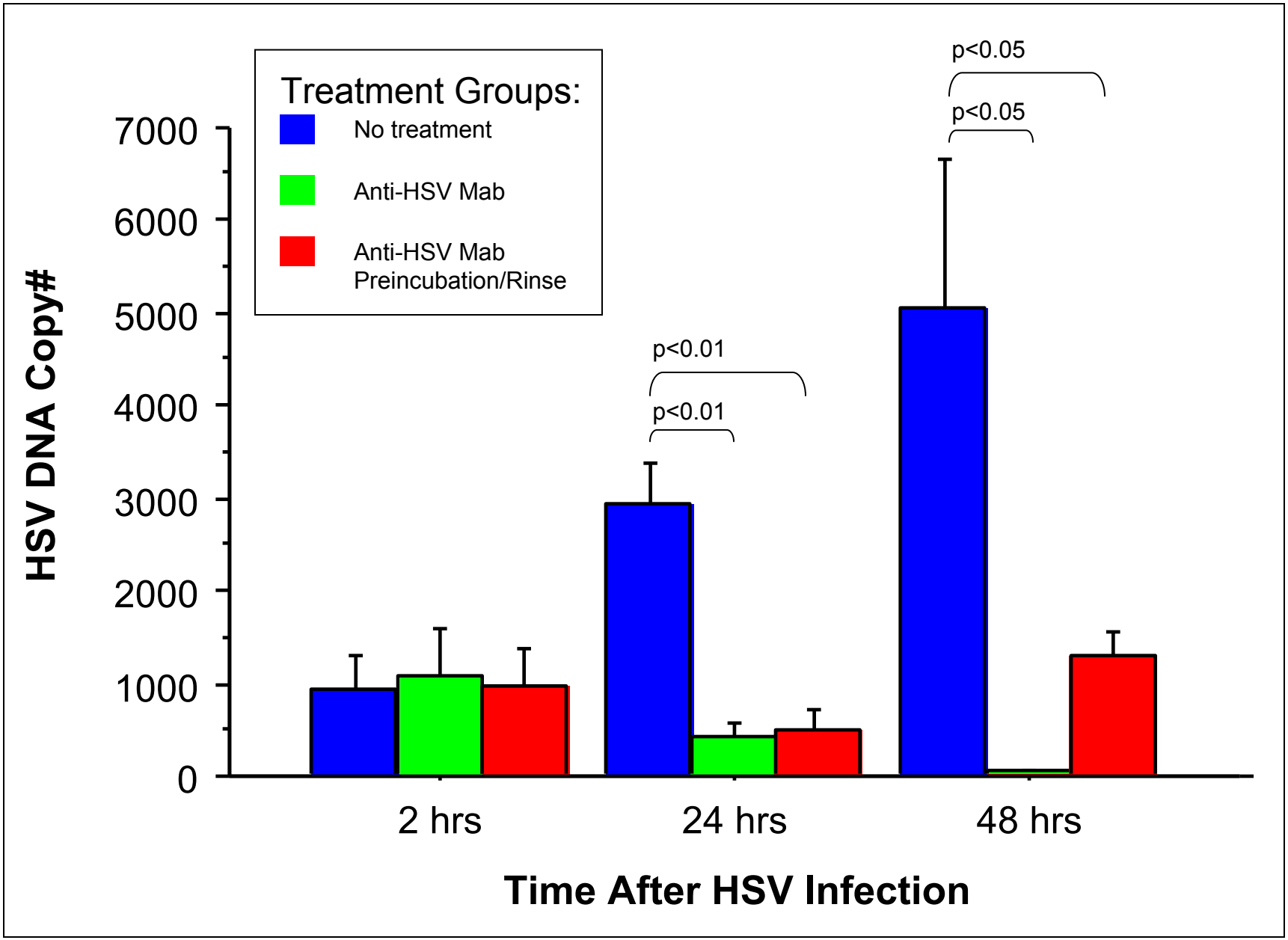


After 1 hour

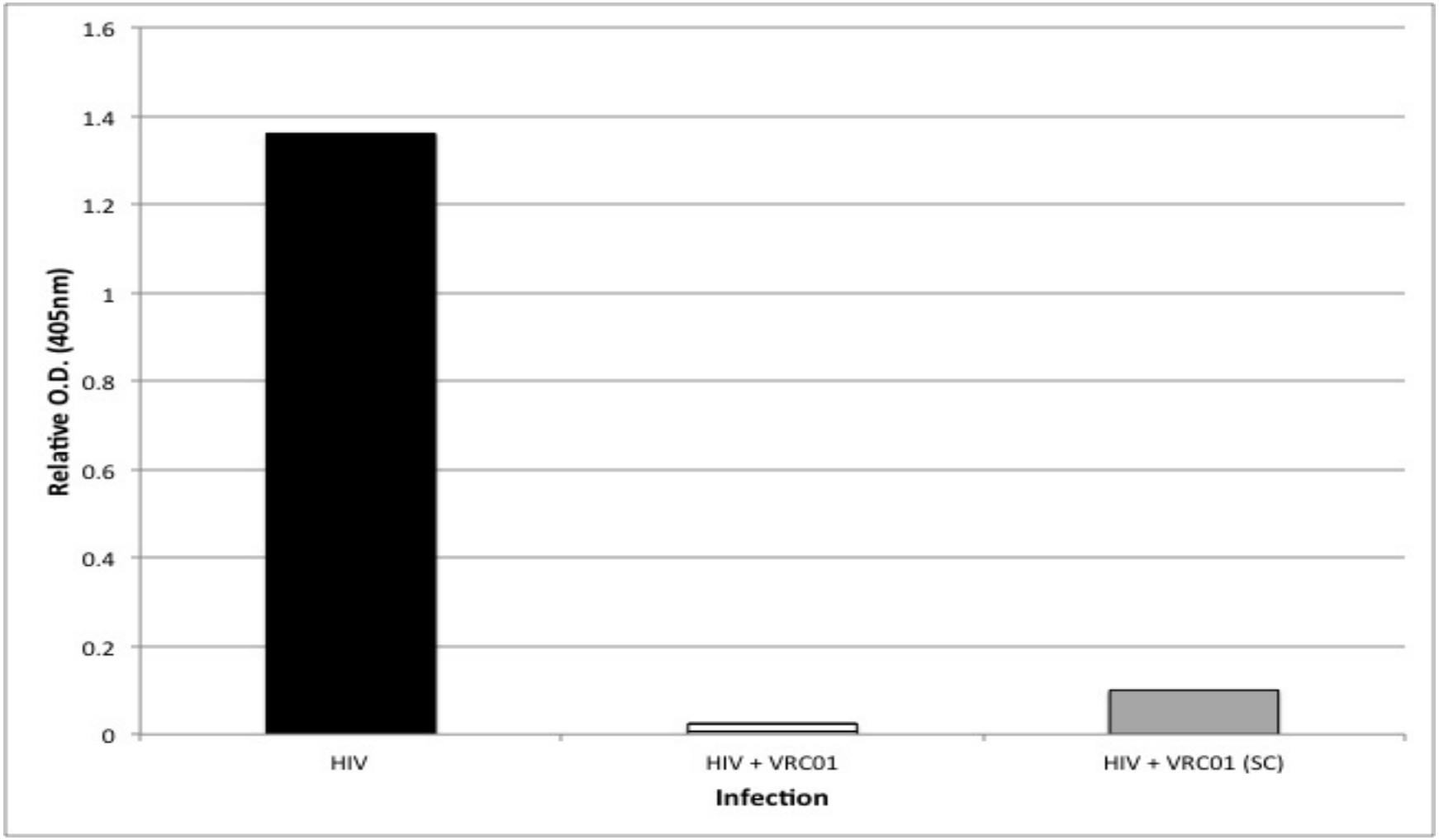


After 12 hours

Suppression of HSV-2 Infection *in vitro* with Anti-HSV Mab



VRC01 HIV neutralizing mab blocks HIV infection of TZM-bl cells in vaginal stratum corneum



Vaginal Stratum Corneum Battlefront Summary

Cell-free and cell-associated pathogens (eg. HIV) can enter the SC

Soluble immunological mediators and leukocytes provide immune defense in the stratum corneum

Implications for microbicide research

- Assess concentrations, activity and half-life of microbicides in SC layer

- Design microbicides to fortify the SC

  -monoclonal antibodies

Acknowlegements

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- Ken Mayer
- Manish Sagar
- Kevin Whaley

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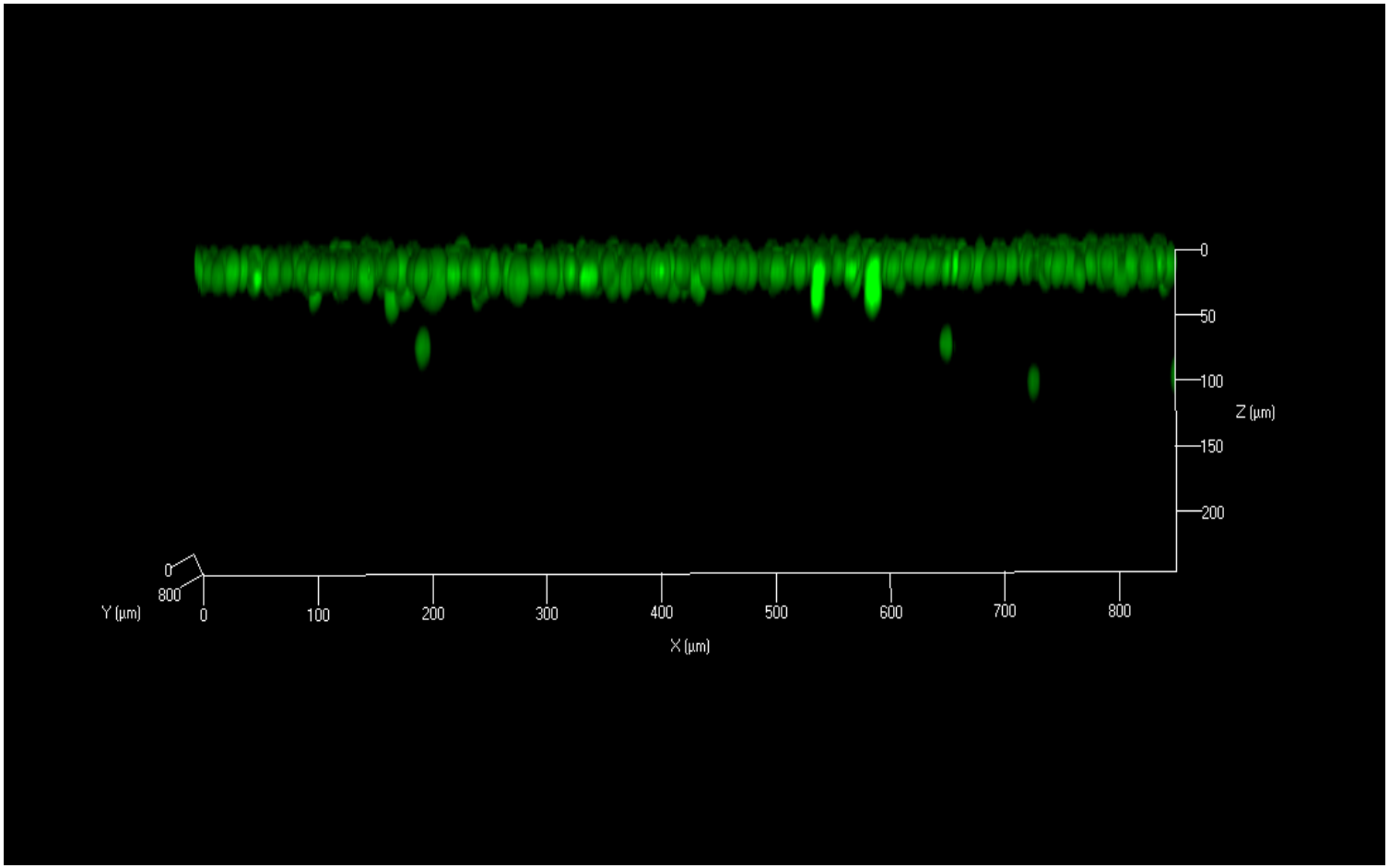
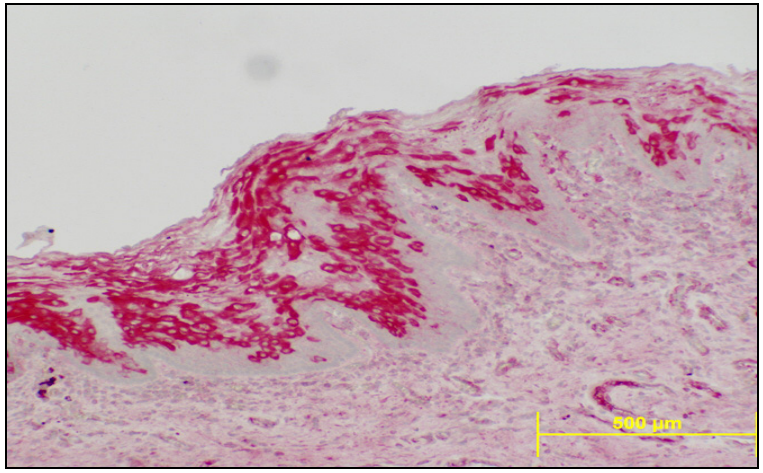
IPCP-HTM: Monoclonal Antibody Multipurpose Microbicides

ICAM antibody blocked leukocyte infiltration into vaginal epithelium

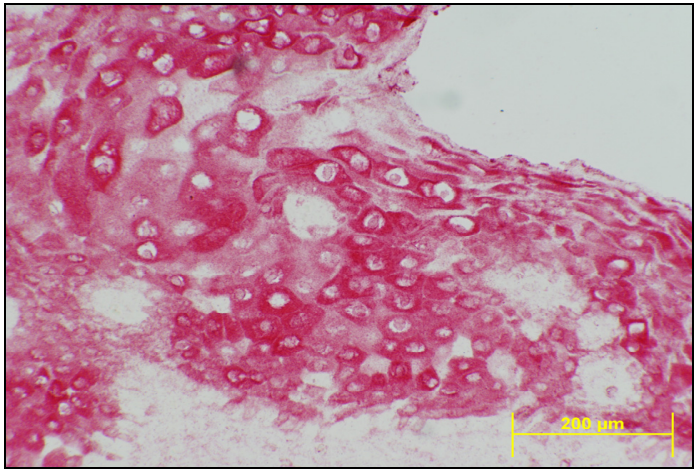


Ig deposits in human vaginal epithelium

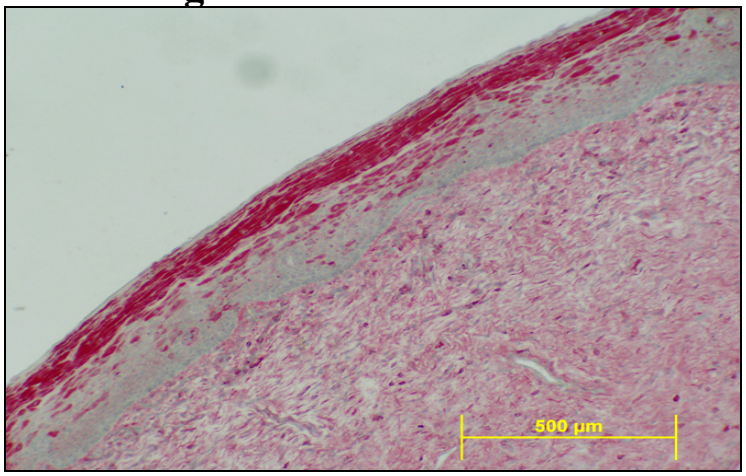
Intracellular IgA in superbasal layer



Intracellular IgG in superbasal layer



IgG in stratum corneum



Intercellular IgG