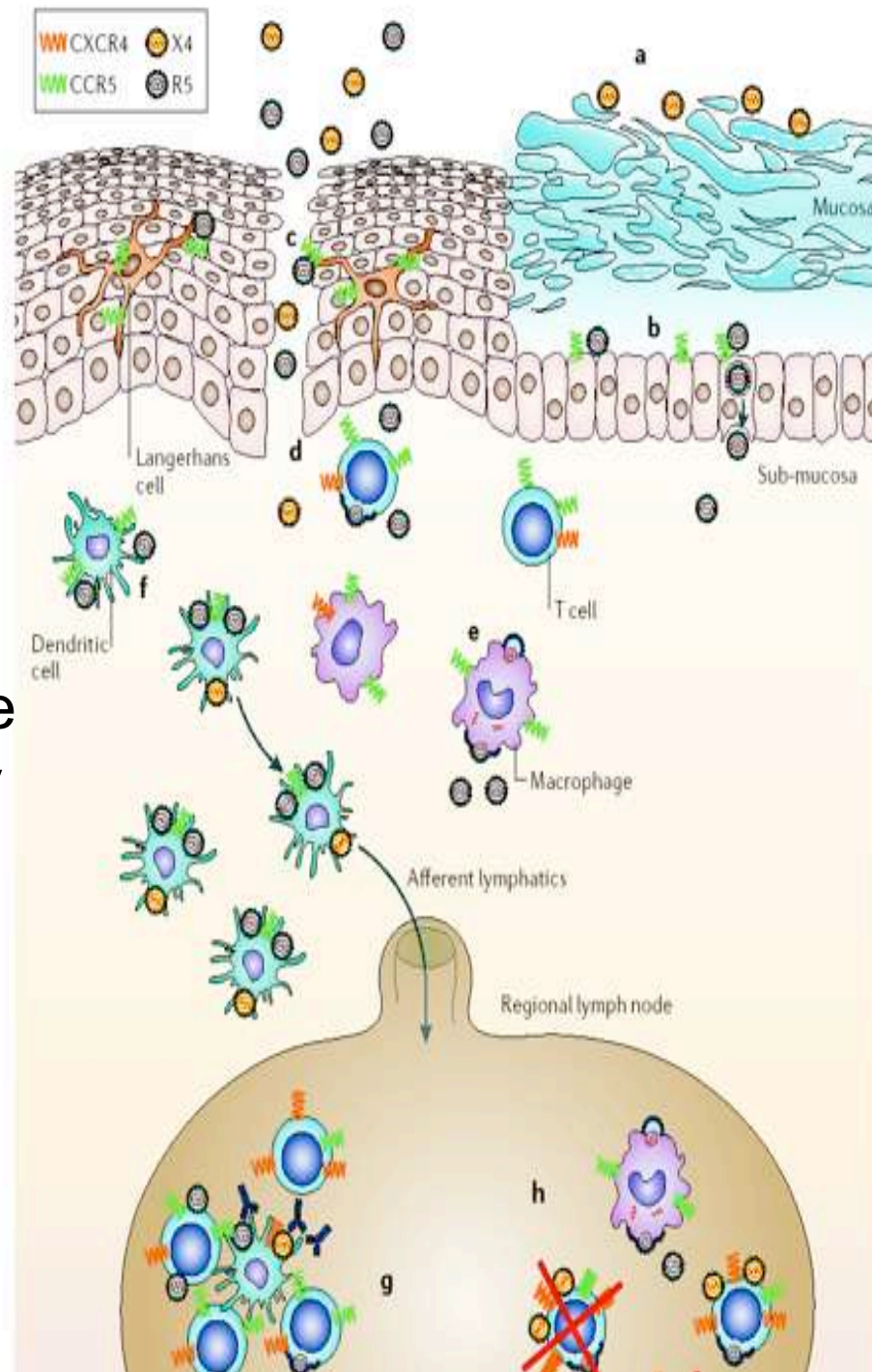


Development of functional antibody assays

Do non-neutralizing antibodies have any role to play in HIV prevention?



The right assay requires the right question

Virus Association with Endocervix

background



background

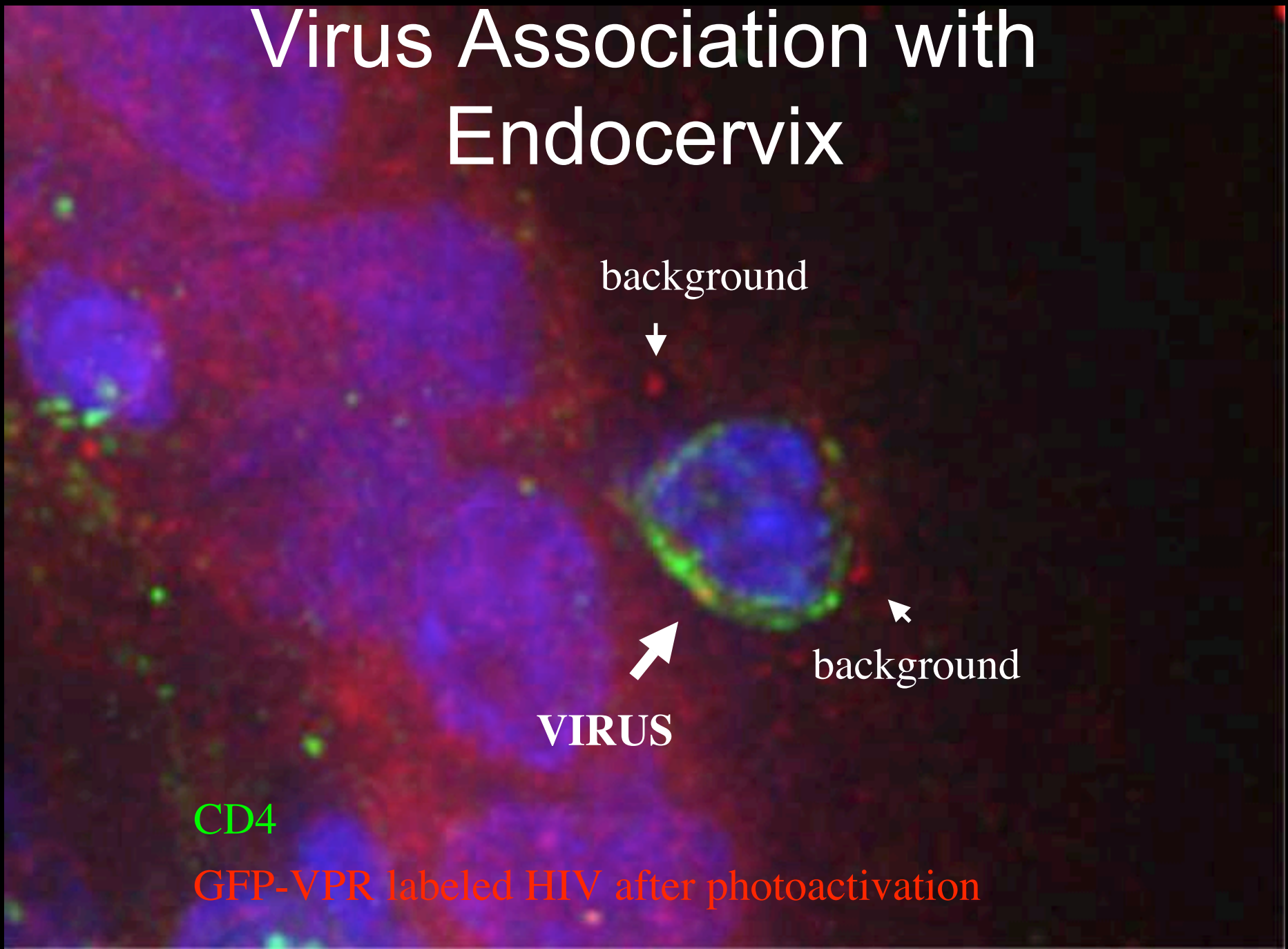


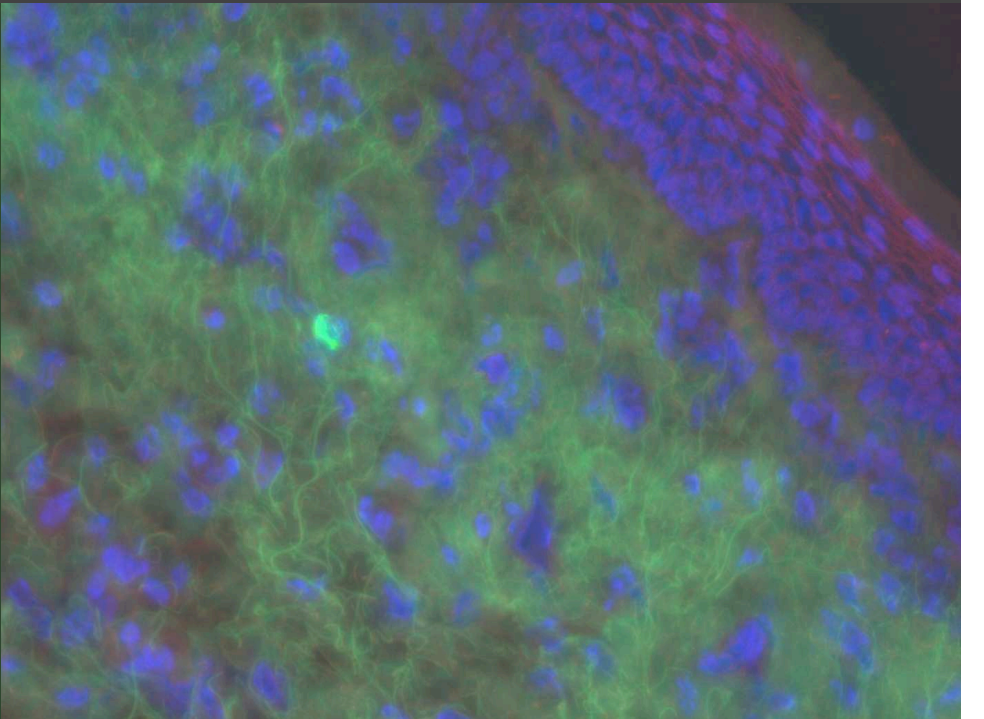
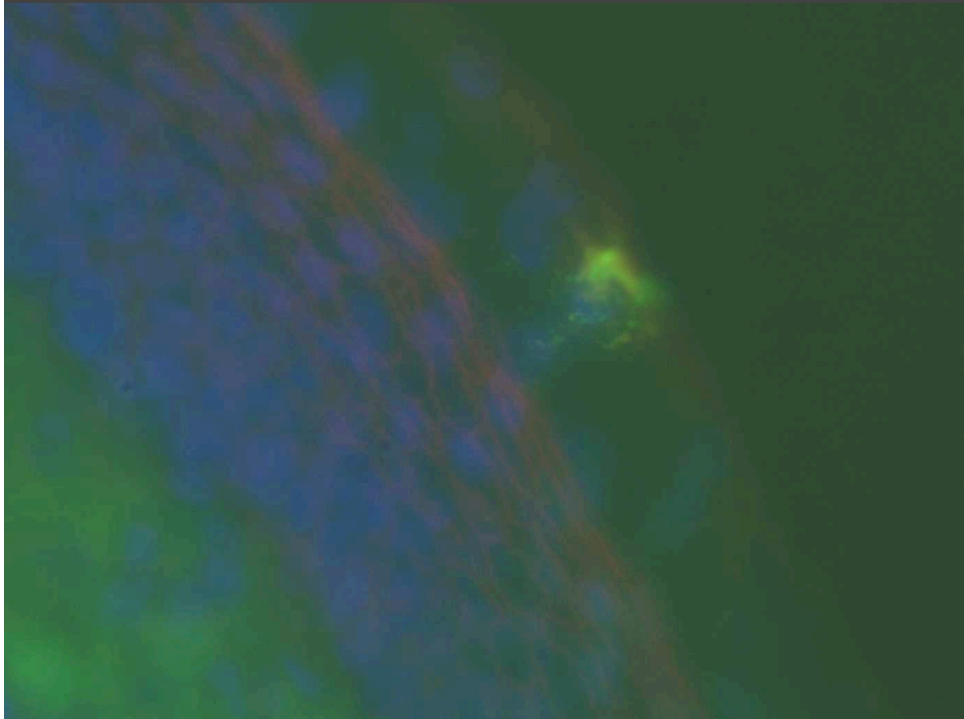
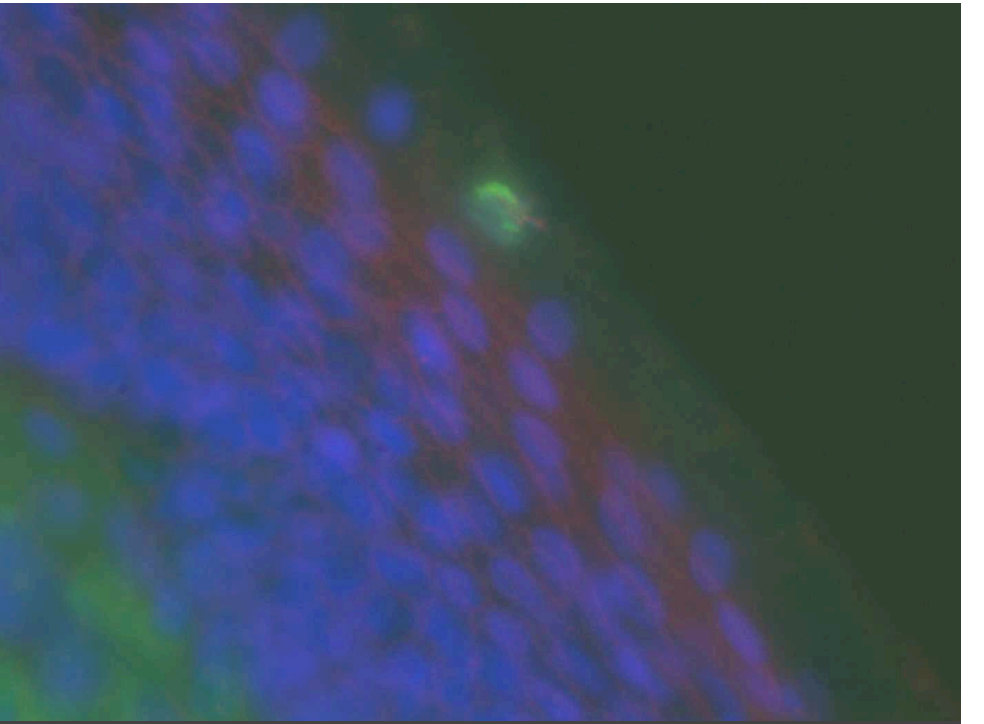
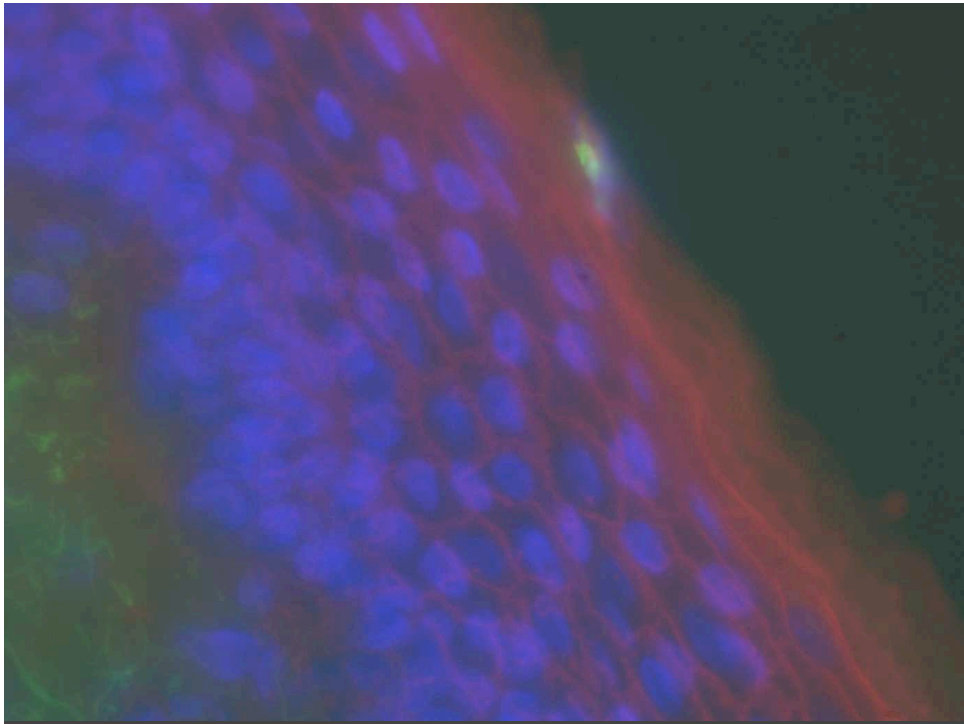
VIRUS



CD4

GFP-VPR labeled HIV after photoactivation





Questions to Address:

Binding Antibodies and HIV: Antibody Type and Function

Current Questions:

What are the functional abilities of the different binding antibodies?

IgG1 and IgG3 are predominant subclass response to viral infection. IgG2 (anti-gp41) may correlate with CD4 help and LTNP status.

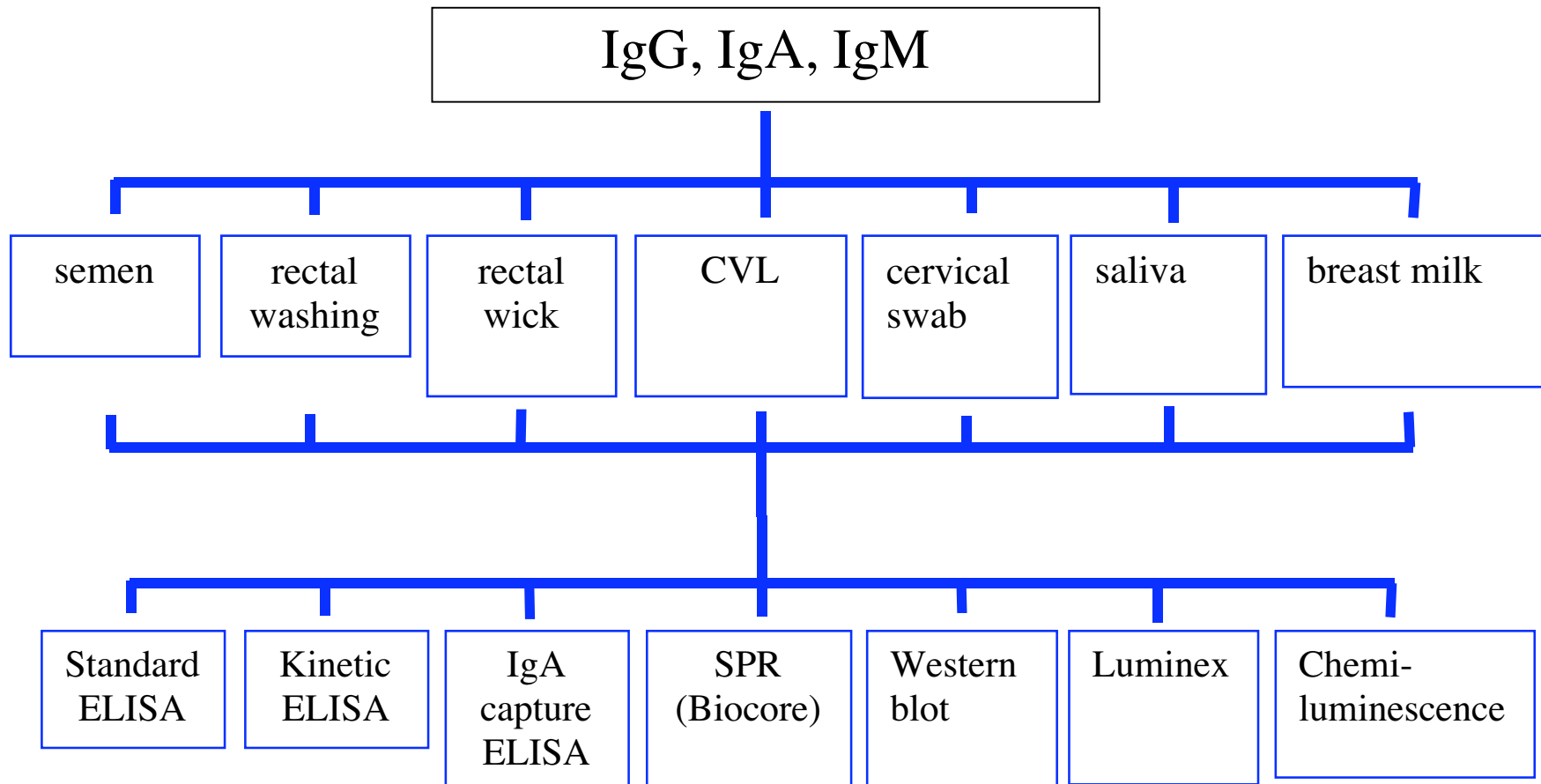
IgG3 may correlate with antibodies that have neutralizing activity. IgG1 may correlate with antibodies that have ADCC.

sIgA may prevent transcytosis/induce intra-epithelial .

What concentrations, types and specificity of antibodies are present at mucosal sites (vaginal, rectal, seminal, saliva) compared to serum/plasma?

Are protective responses different to those that modulate viral set-point and disease progression

Optimization of mucosal humoral assays.



Sample size, inhibitory factors, sensitivity, hormonal status

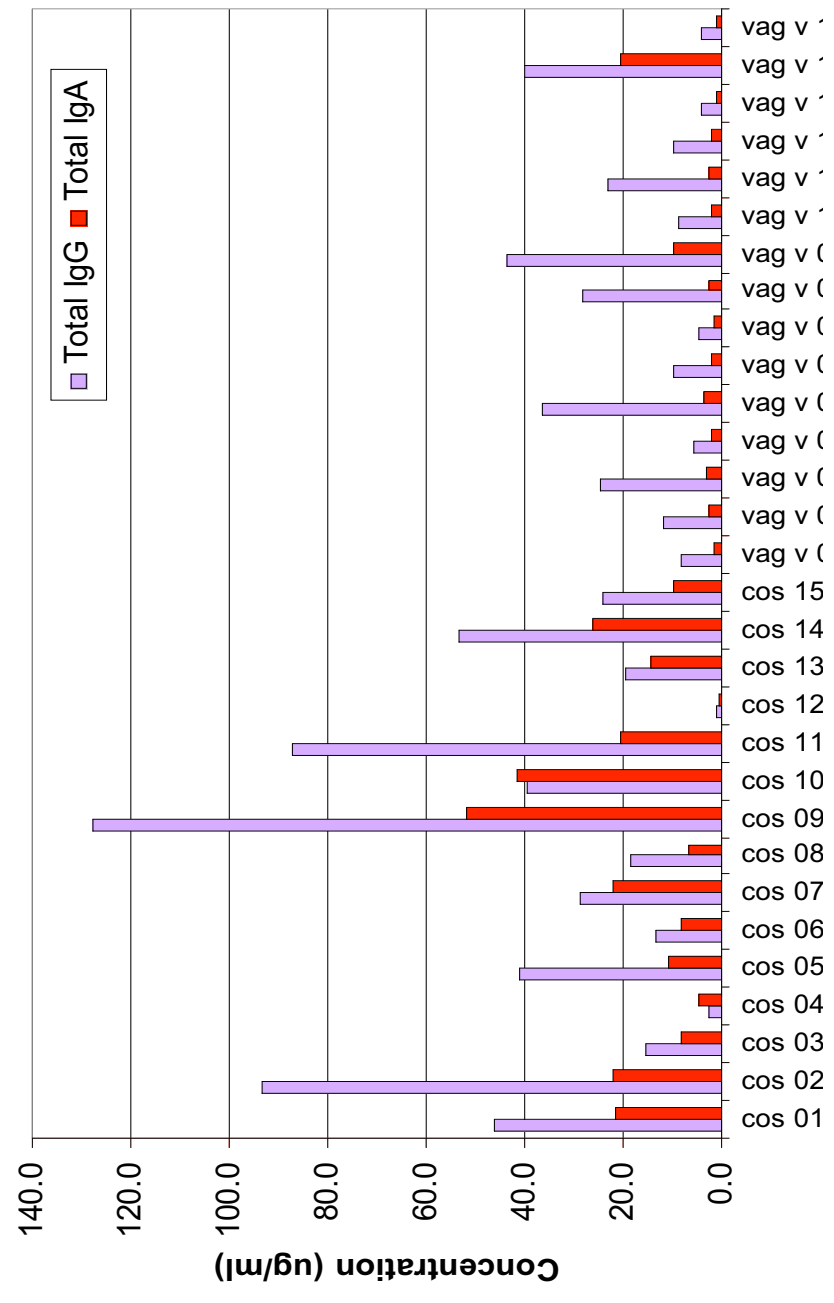
Comparative Characteristic Features of the Immune Systems of the Genital and Intestinal Tracts

	Genital tract	Intestinal tract
Dominant Ig isotypes	IgG \times IgA	IgA \gg IgG
Hormonal regulation	+++	- ?
Inductive site for local and generalized immune responses	- to +	+++
Effector site	++	+++
Intranasal immunization	++	+
Expression of homing receptors on lymphocytes and ligands on endothelial cells	α4β1	α4β7 MAd CAM-1
Contribution of antibodies from the circulation	+++ (~ 50%)	< 1%

Jiri Mestecky

IgG levels are higher than IgA in cervico-vaginal samples: higher levels of recovery from cervical os

Immunoglobulin levels in Weck-cel cervico-vaginal samples



Probability of Detection of HIV-1-neutralizing Antibodies in External Secretions

- Serum levels of total IgG

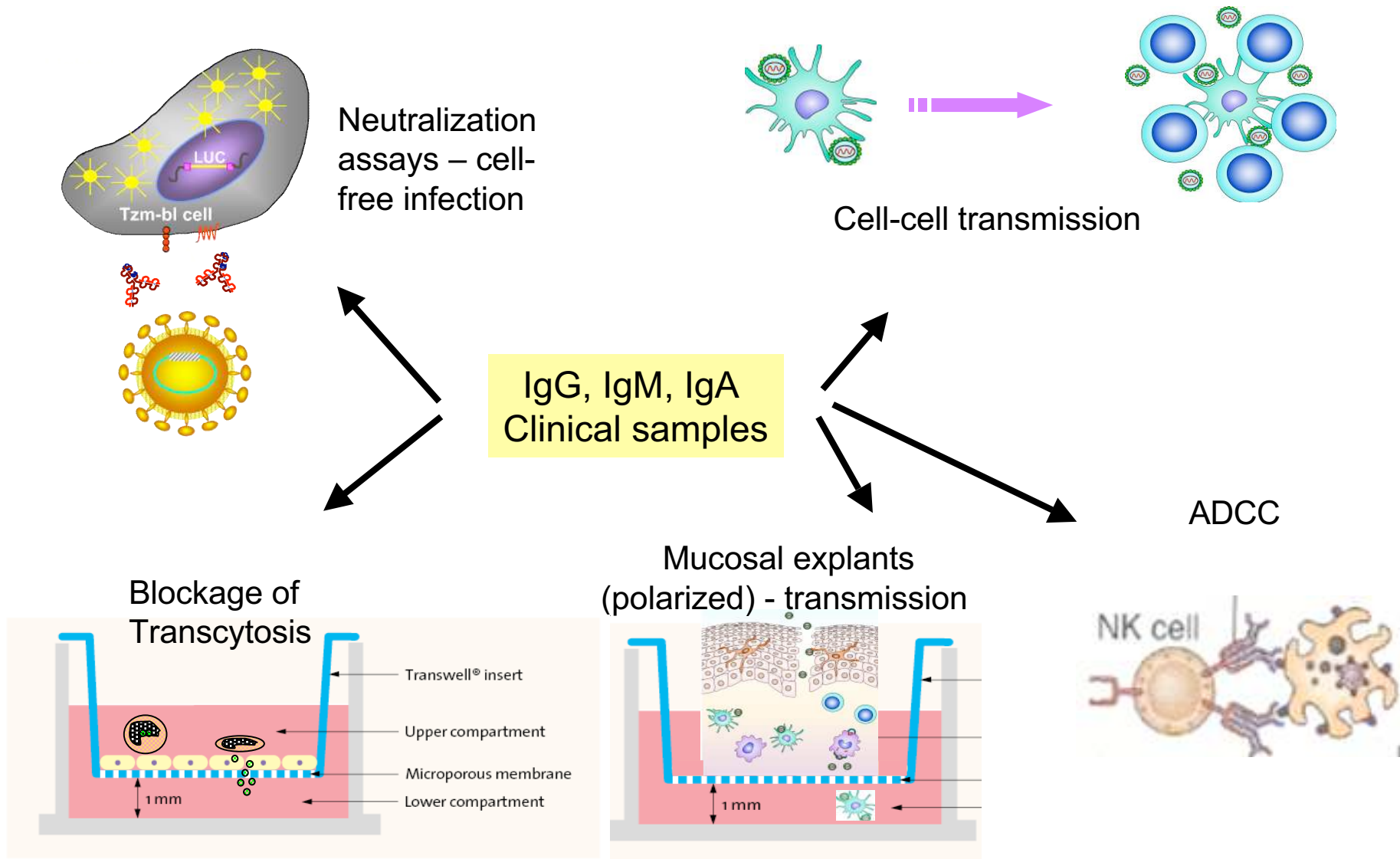
15,500 μ g/ml - neutralizing Ab 1:100 = \sim 155 μ g/ml

- External secretions - levels of total IgG in μ g/ml

cervico-vaginal fluid	108 \pm 30
semen	62 \pm 19
rectal wash	5 \pm 2
urine	14 \pm 5
parotid saliva	2 \pm 0.5

**Probability of detecting of functional characteristics?
(transcytosis, cell-cell transmission, DC uptake etc)**

Assay development for functional analysis of antibody responses.



Future directions

- The relative role of antibodies against transcytosis, DC-T cell, T cell-T cell, Mo-T cell and those that induce ADCC, C' activation in transmission/control/pathogenesis require better definition.
- IgA and IgG transport across epithelial cells needs to be better understood – including effectiveness of polymeric Ig receptor and FcRn, and effect of pH on transport.
- Vaginal IgA and IgG transport and concentration in women following sexual arousal needs to be defined.
- Deficiency in HIV-specific IgA in infected individuals needs to be confirmed and understood at a mechanistic basis – tolerance, inhibition of Ig switch
- Mucosal immunology in primate models with SHIV and SIV, particularly those using mucosal challenges, needs to be studied.
- The relative role of IgG, IgA and IgM in prevention of transmission needs to be evaluated in primate models
- Techniques to further characterize T and B cell responses from mucosal sites need to be further developed and standardized.