

Pioneering systems biology in clinical microbicide trials – MTN-007 and beyond

Florian Hladik
www.hlab.science

UW Medicine
DEPARTMENT OF OBSTETRICS
AND GYNECOLOGY

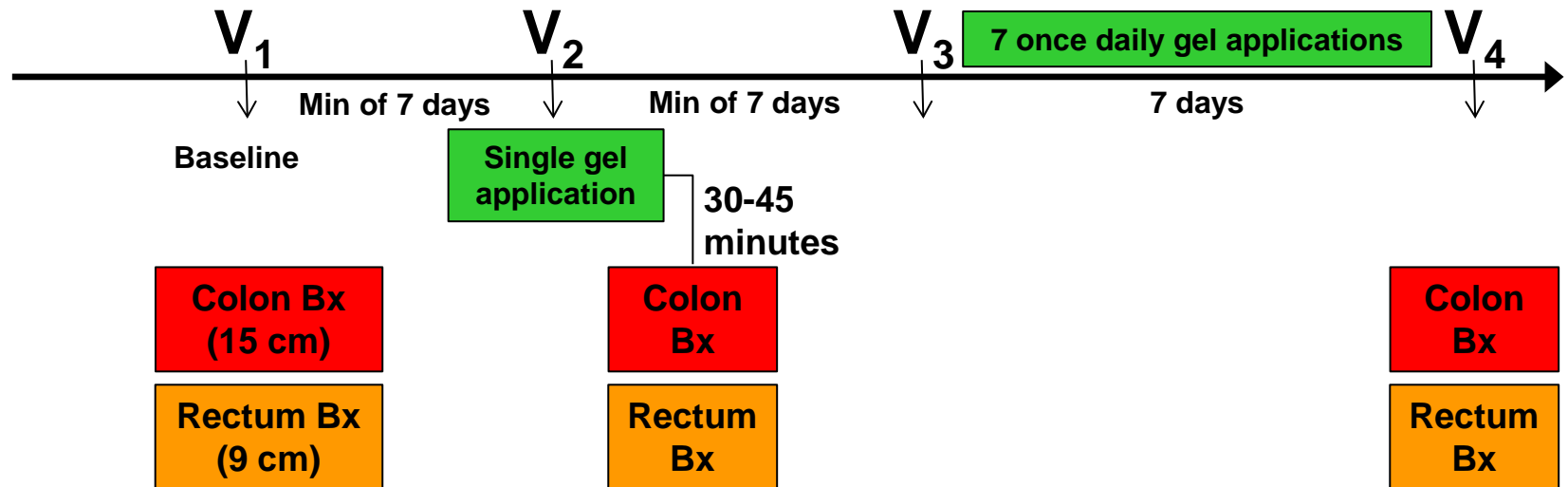
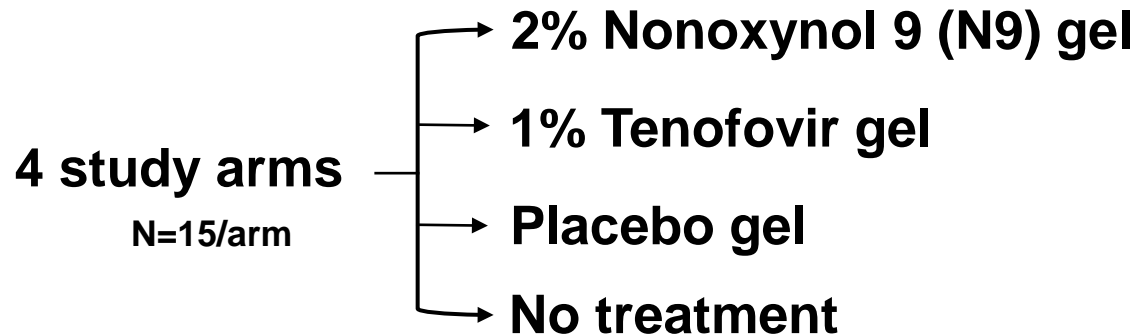


FRED HUTCH™
CURES START HERE

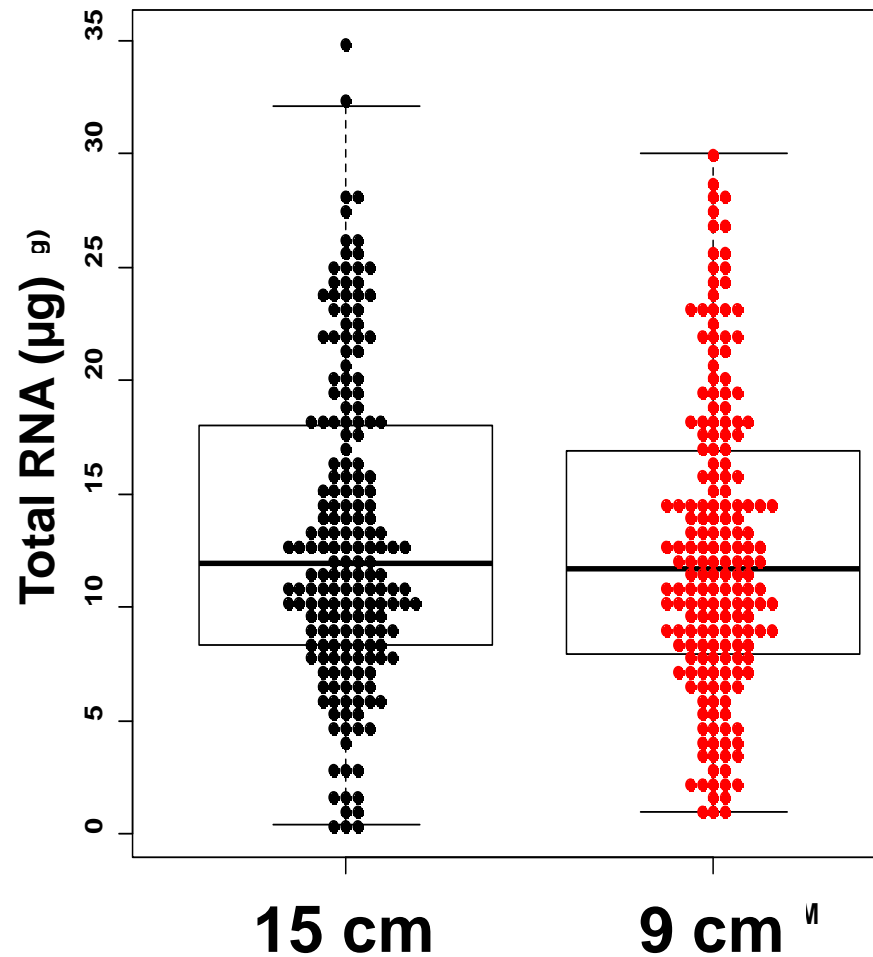
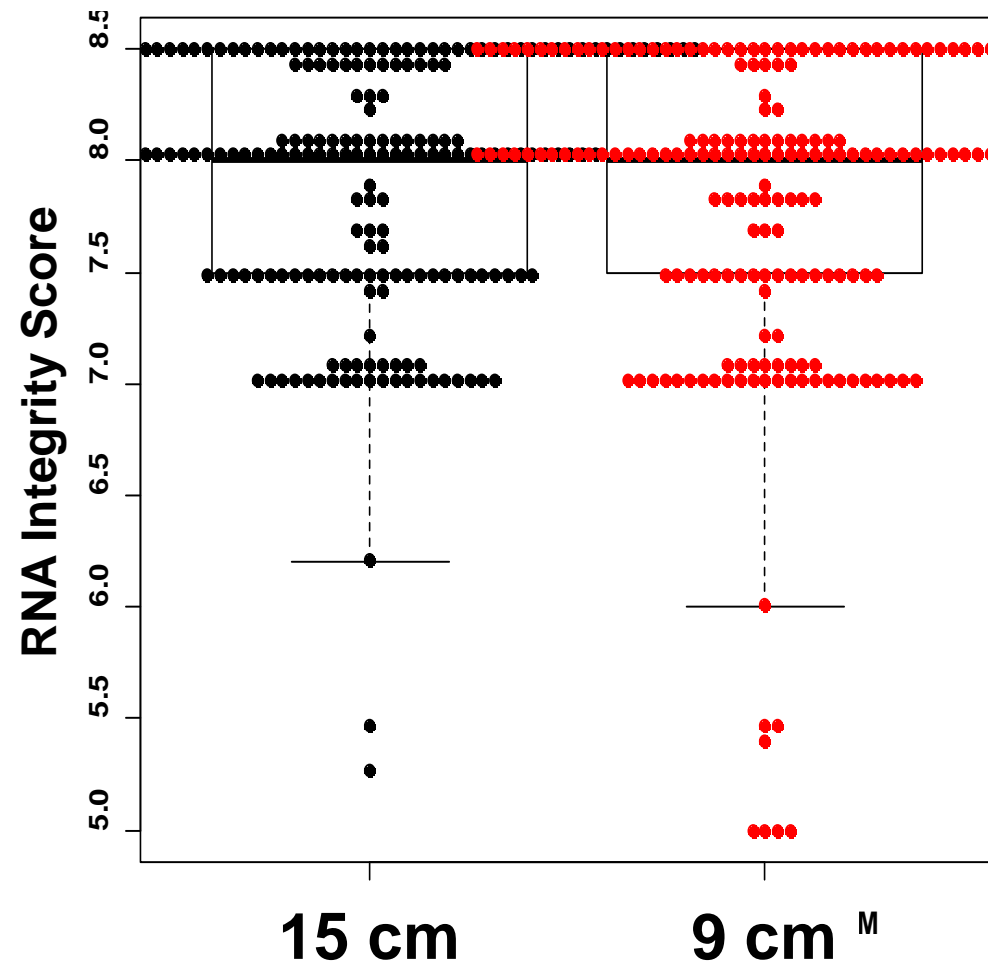
The ever interesting story of tenofovir gel

- I. Immunological effects of tenofovir gel**
- II. Potential longer-term effects and how to study them**
- III. Hypothetical ramifications beyond the microbicide field**

MTN-007 study design



MTN-007 rectal biopsies yielded excellent RNA quality and quantity

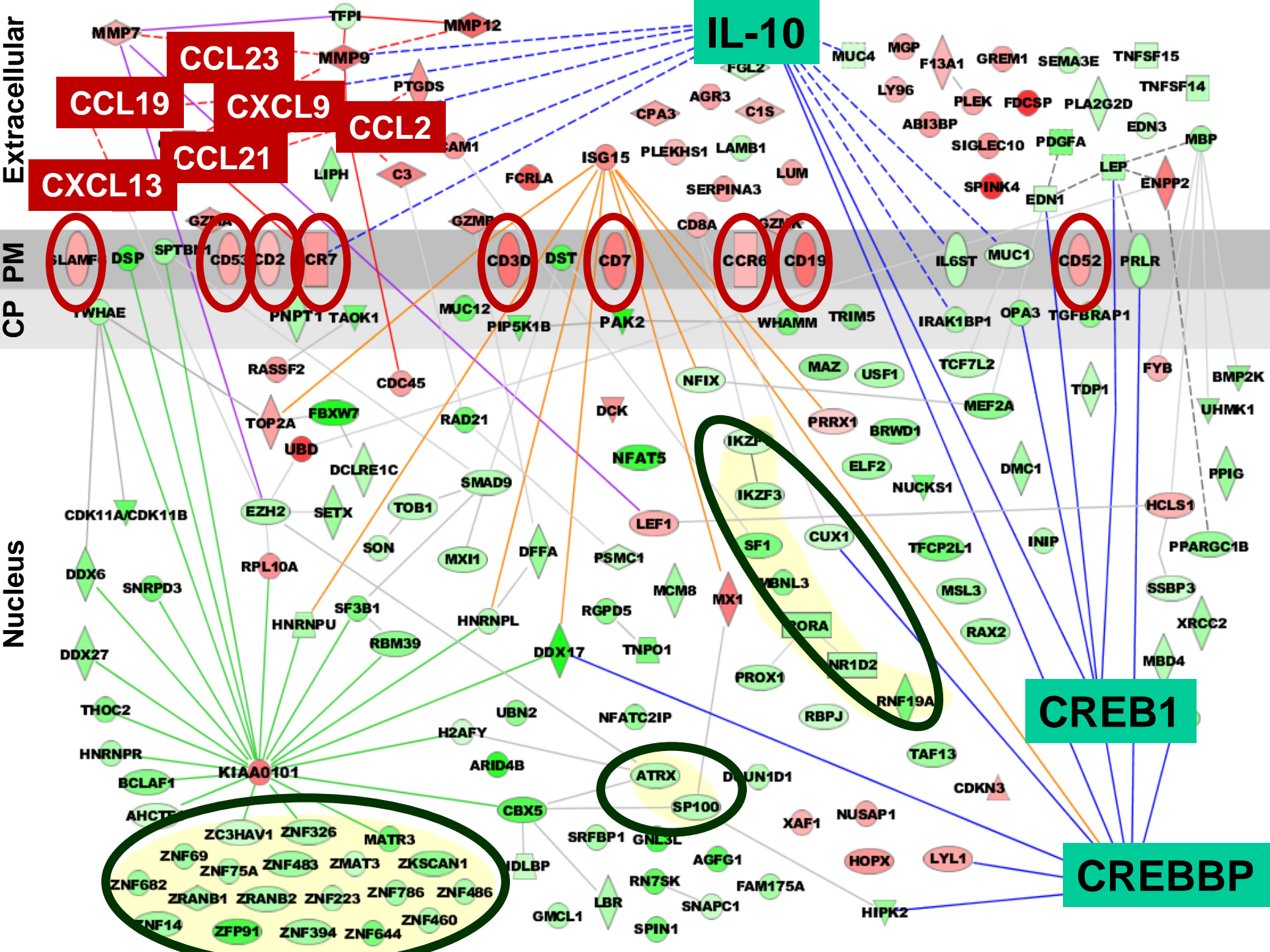


n=381

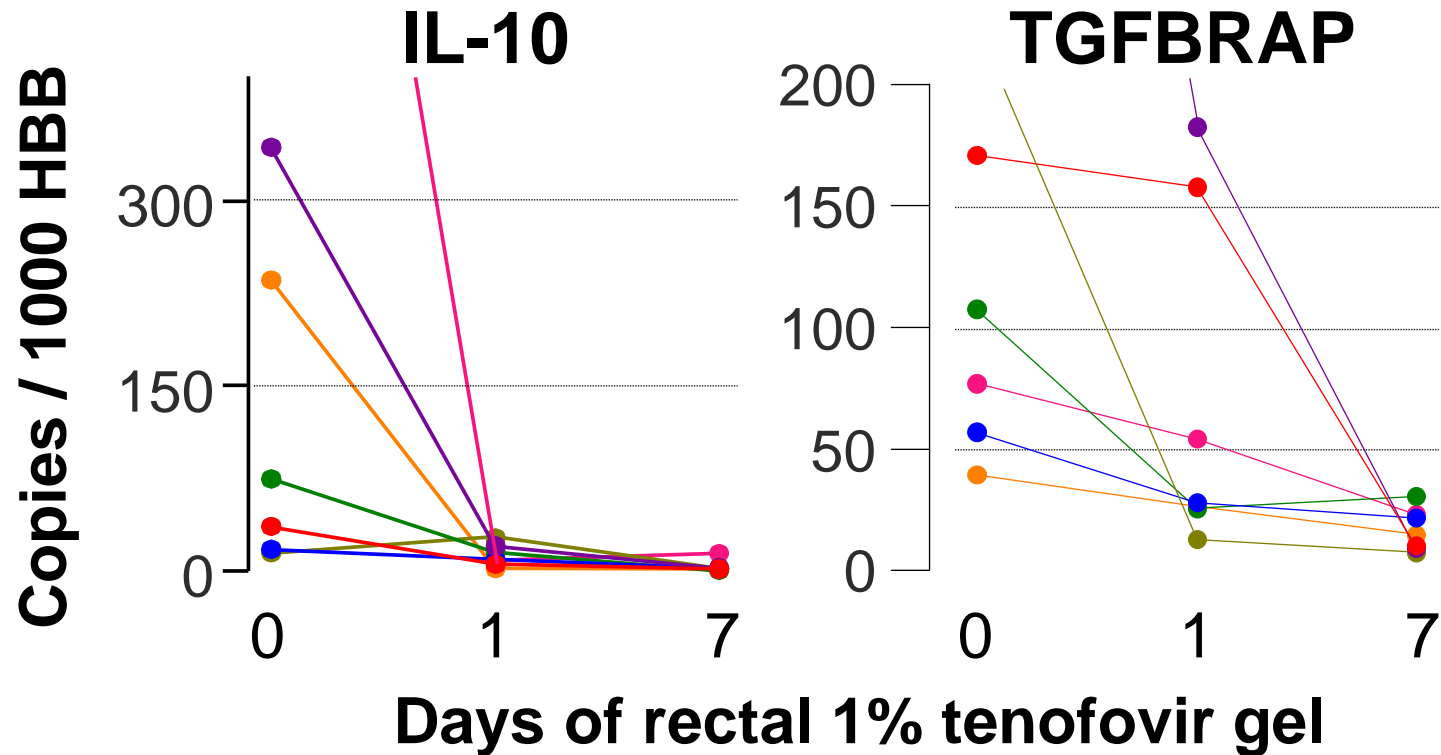
Number of genes changing expression after 7 days of treatment

	Up	Down
Nonoxynol-9	60	56
Tenofovir	137	505
HEC	12	4
No treatment	17	6

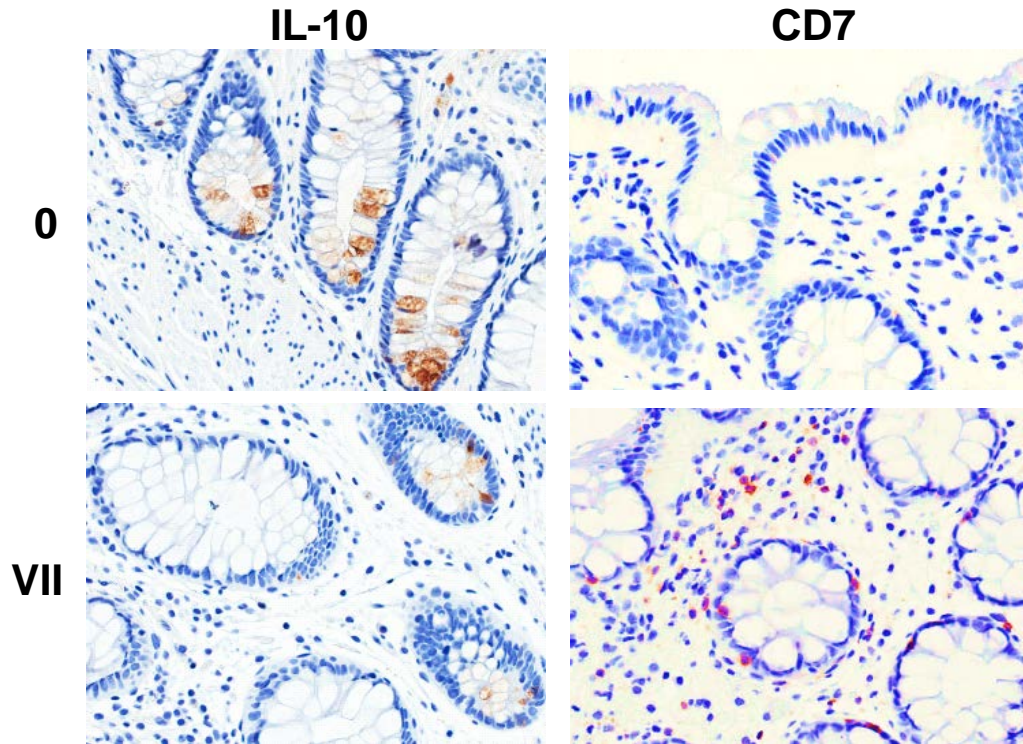
Consensus data across 8 study subjects



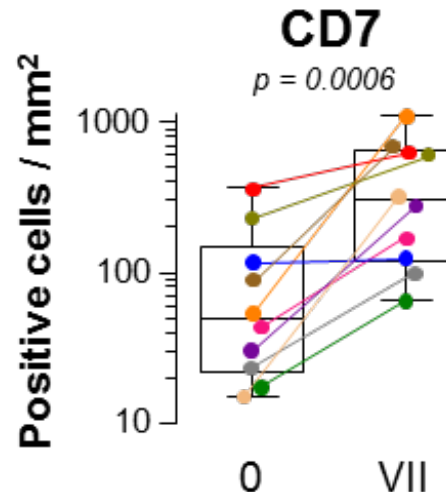
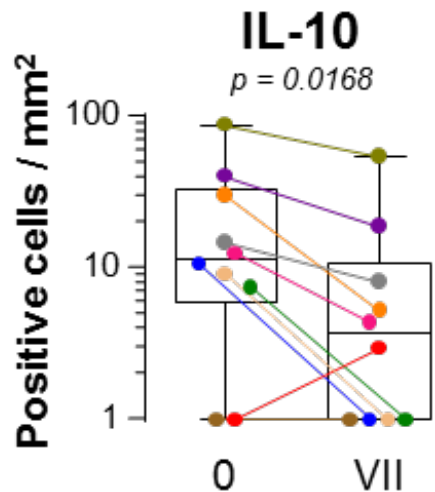
Tenofovir inhibits the anti-inflammatory arm of gut immunity



Confirmation by immunohistochemistry

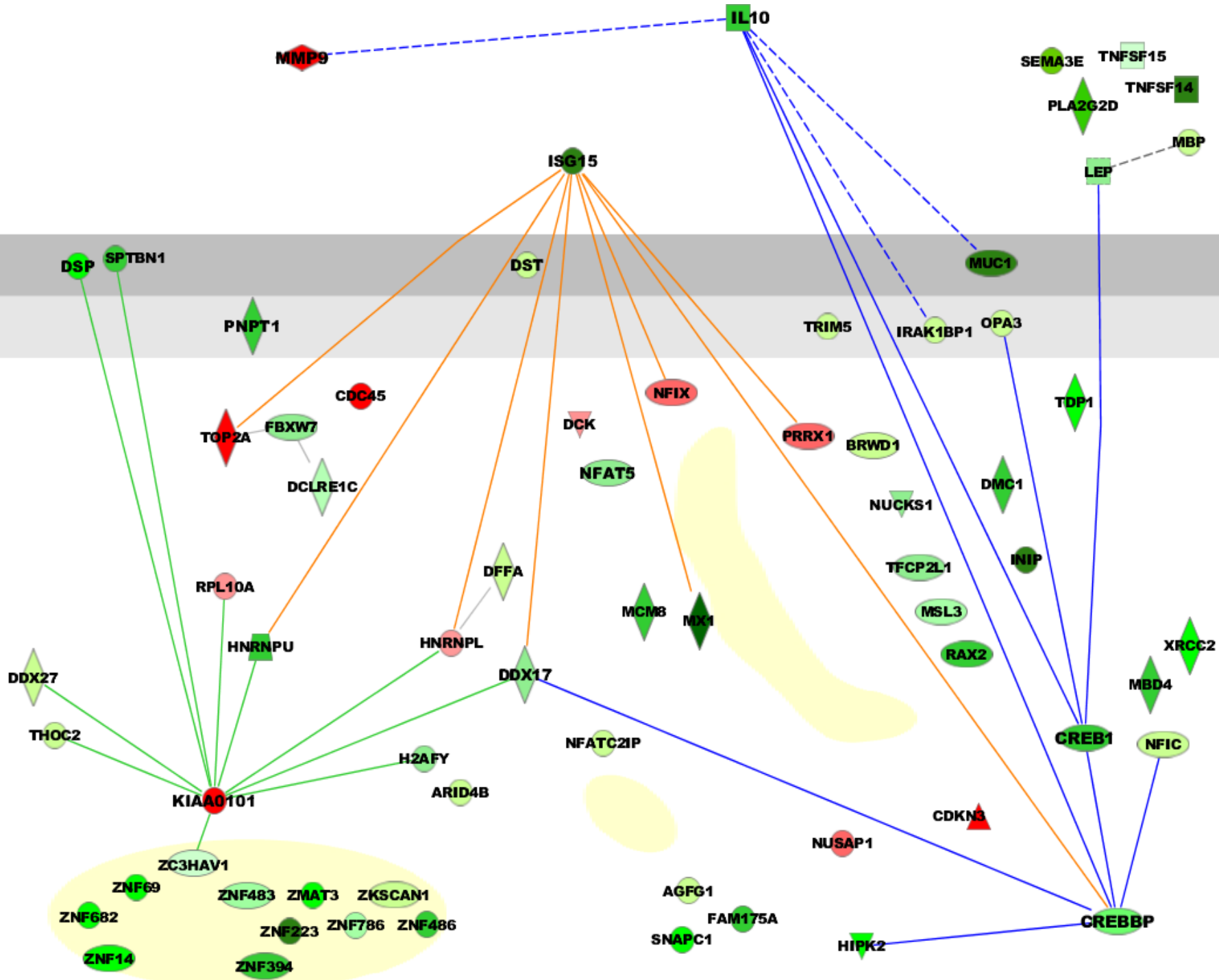


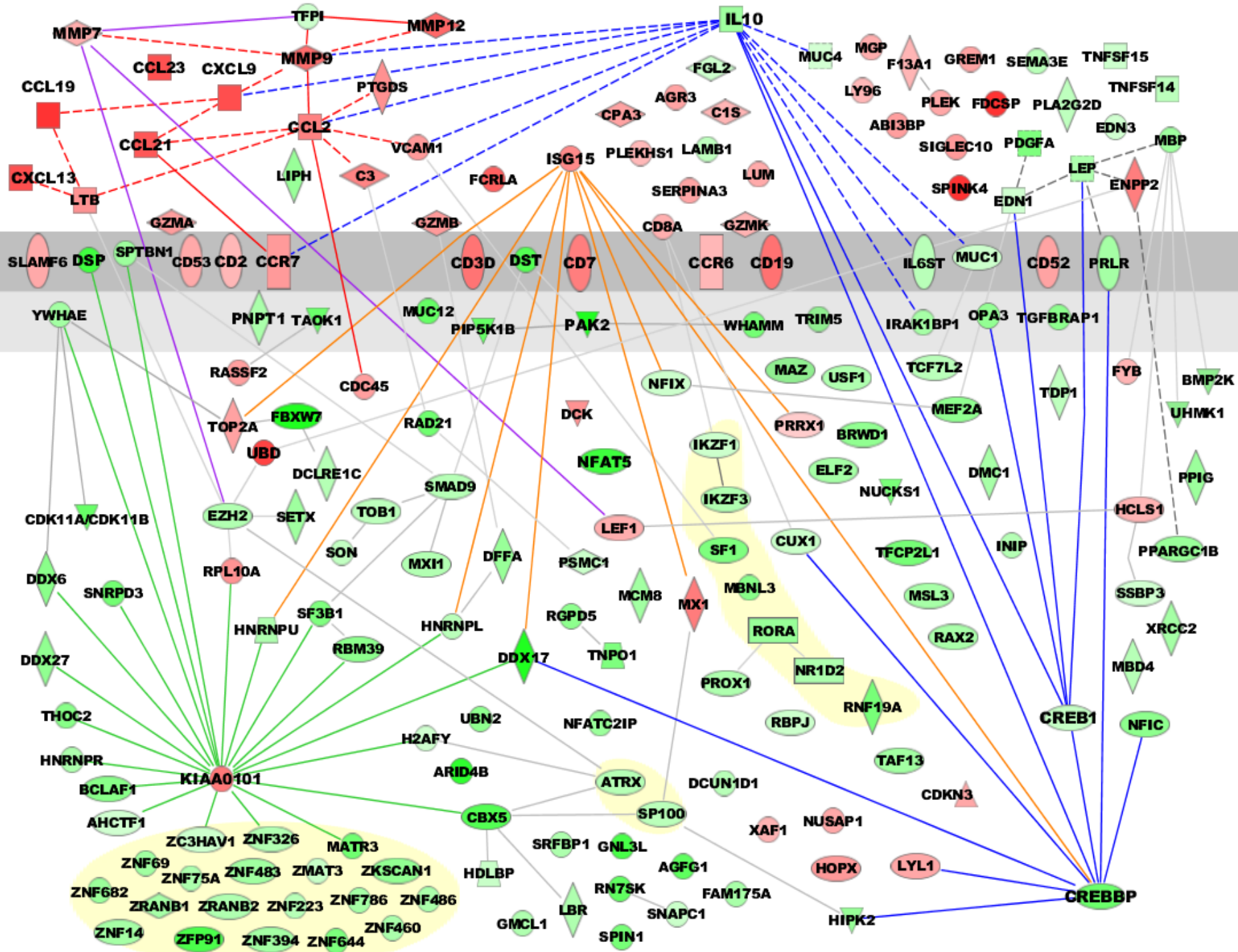
Rectal biopsies
in MTN-007

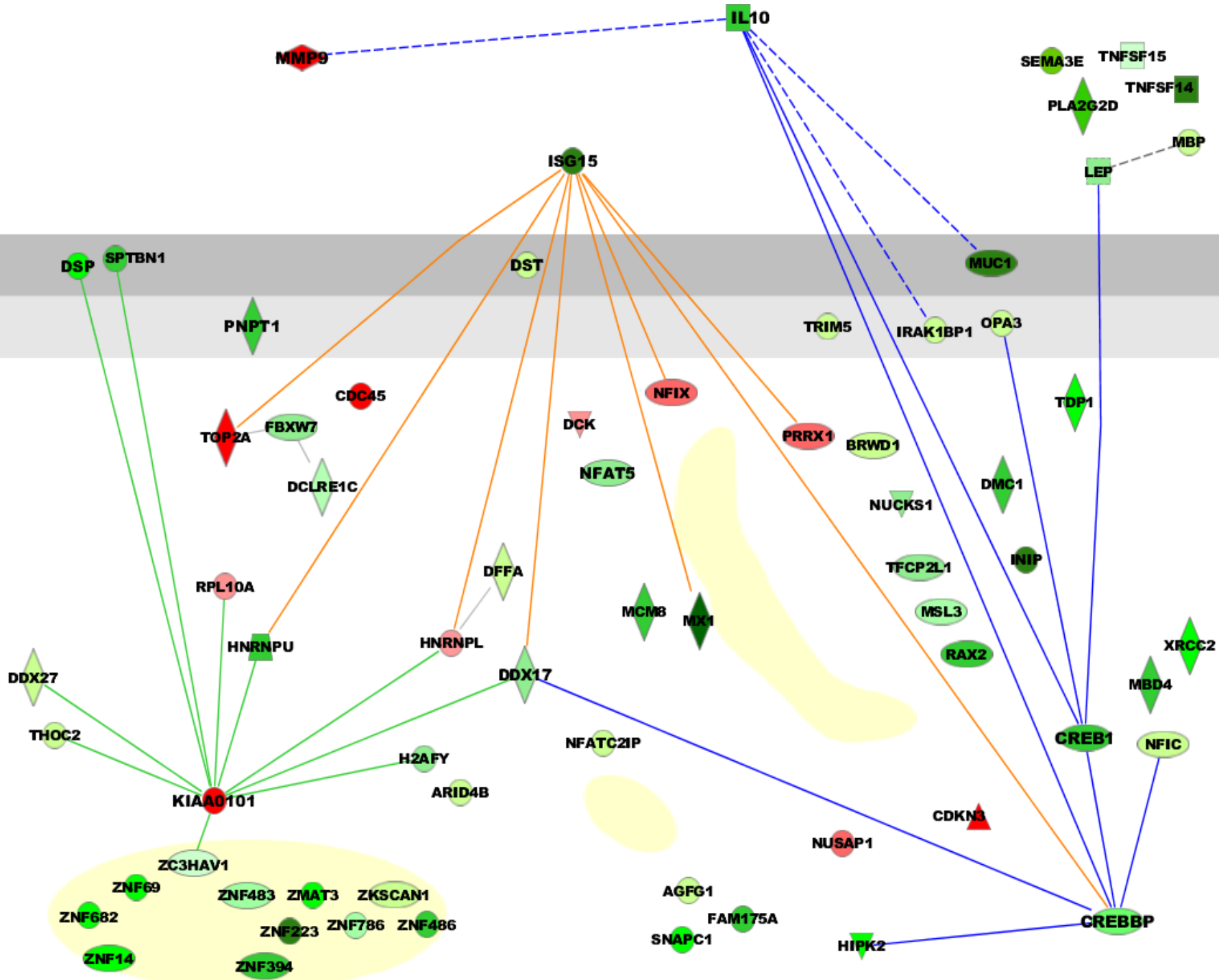


Do the immunological effects of tenofovir gel also occur in the genital tract?

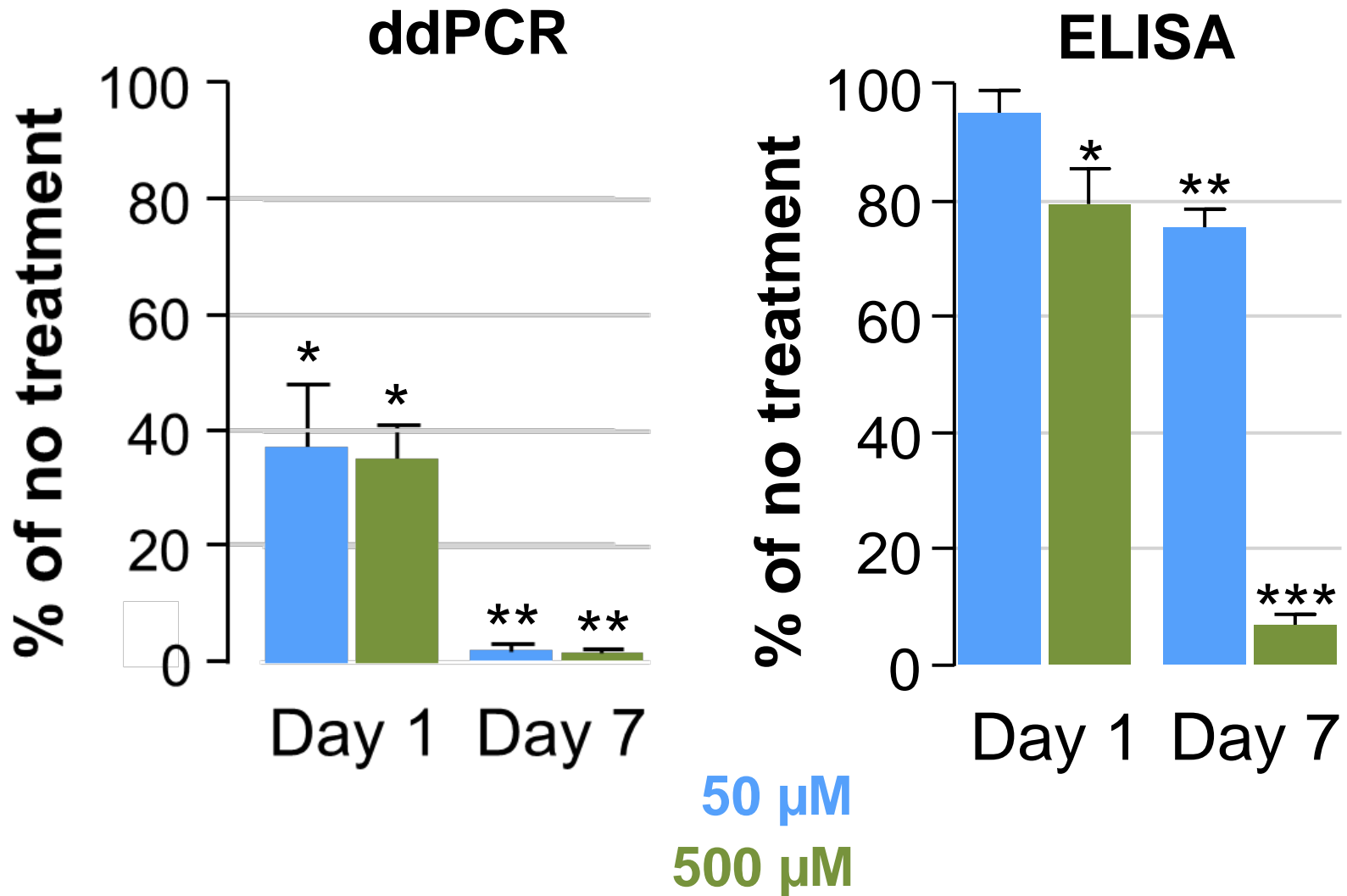
- Will be investigated in MTN-014, a vaginal-rectal cross-over study of 1% tenofovir gel**
- Was investigated *in vitro* with primary vaginal epithelial cells established from four healthy women**







IL-10 in vaginal epithelial cells



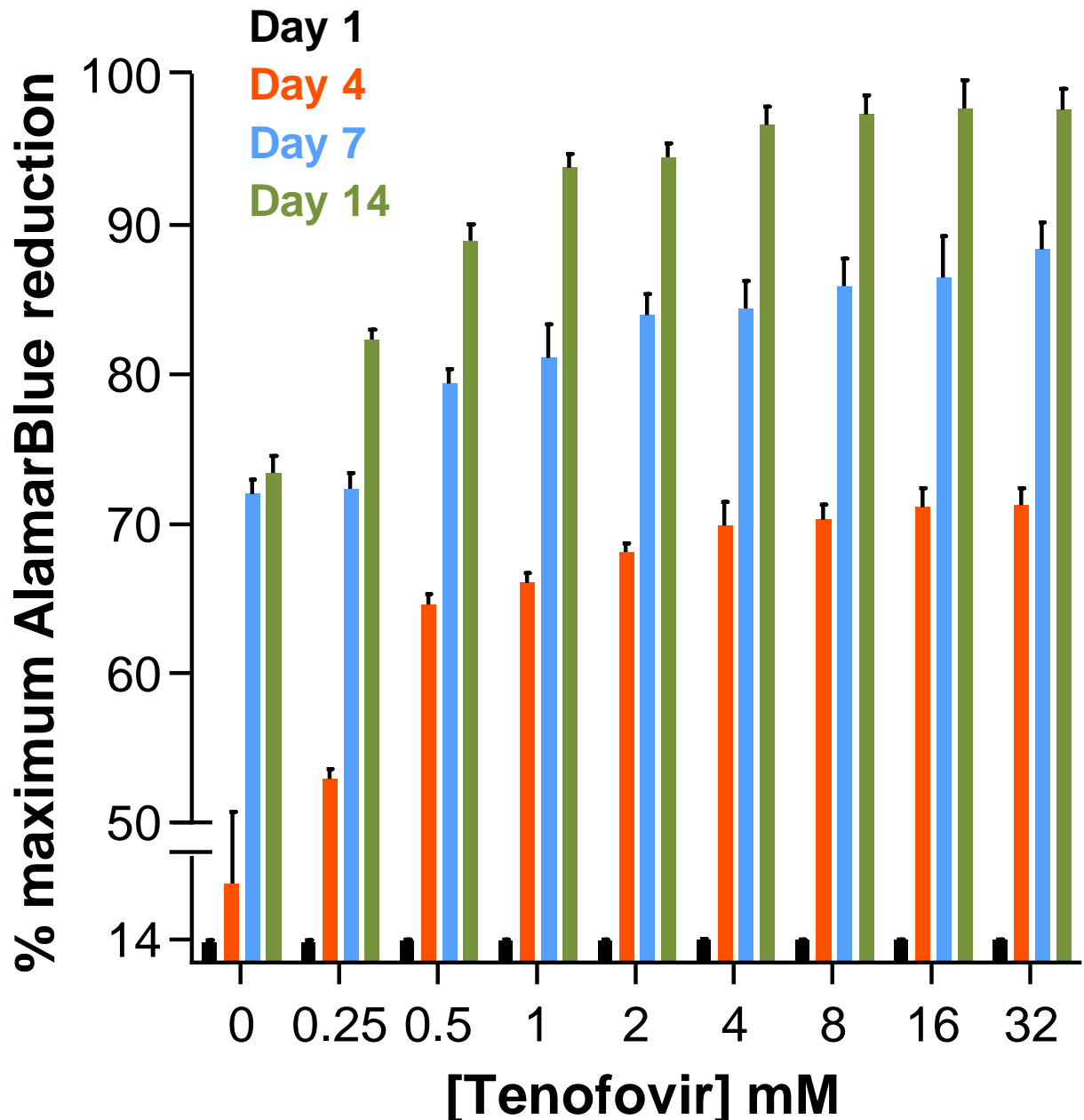
Olszak T et al. Protective mucosal immunity mediated by epithelial CD1d and IL-10. *Nature* 22;509:497-502 (2014)

The immunological effects of tenofovir are complex

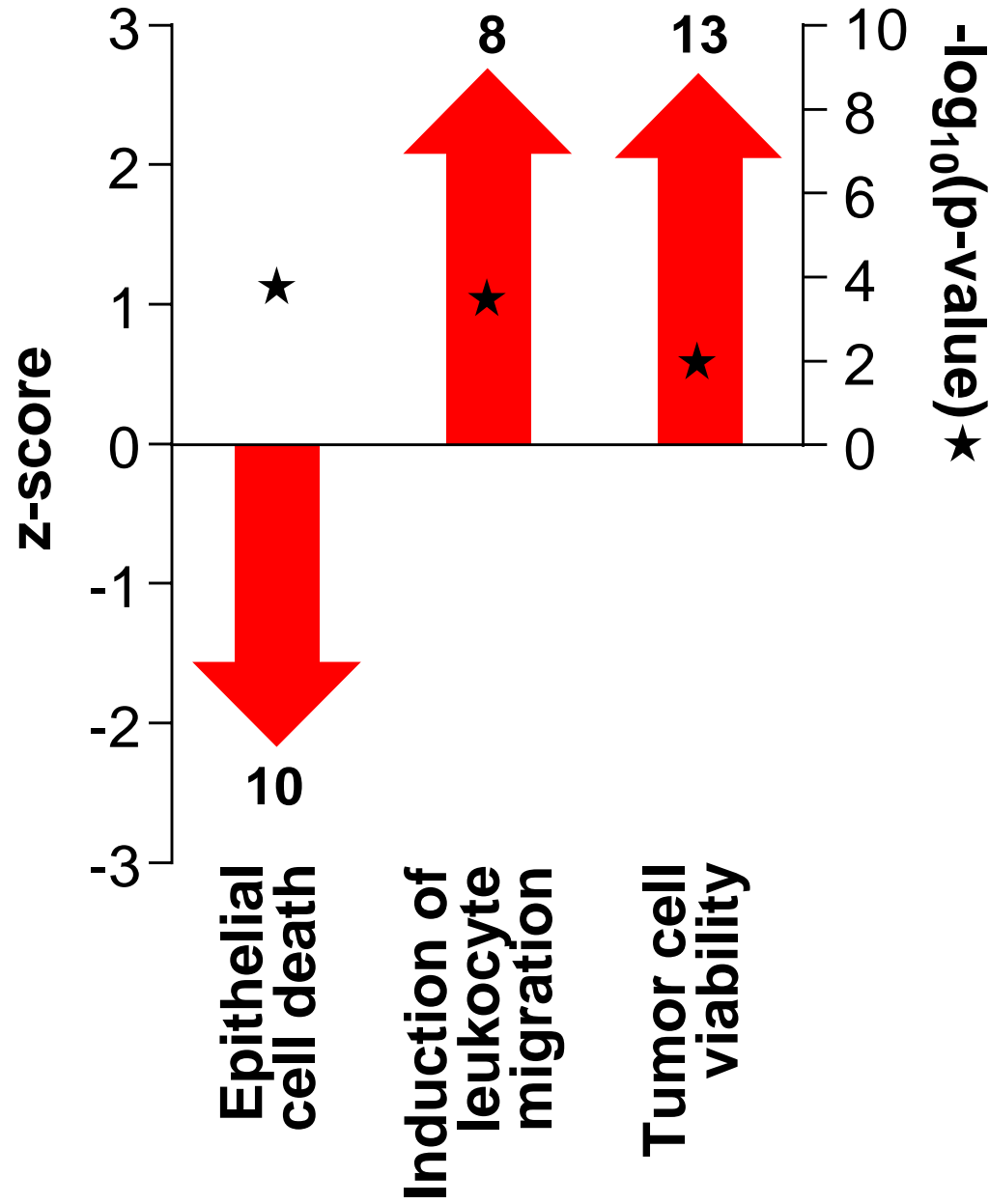
- Tenofovir is an immune modulator rather than stimulator in the mucosa
(“anti-anti-inflammatory”)
- The consequences of this property on HIV susceptibility remain unclear, but ongoing studies in CAPRISA 004 subjects suggest that in the presence of inflammation they may become clinically relevant (Jo-Ann Passmore)

II. Potential longer-term effects and how to study them

Tenofovir increases epithelial cell proliferation ...



... and generally enhances cell viability

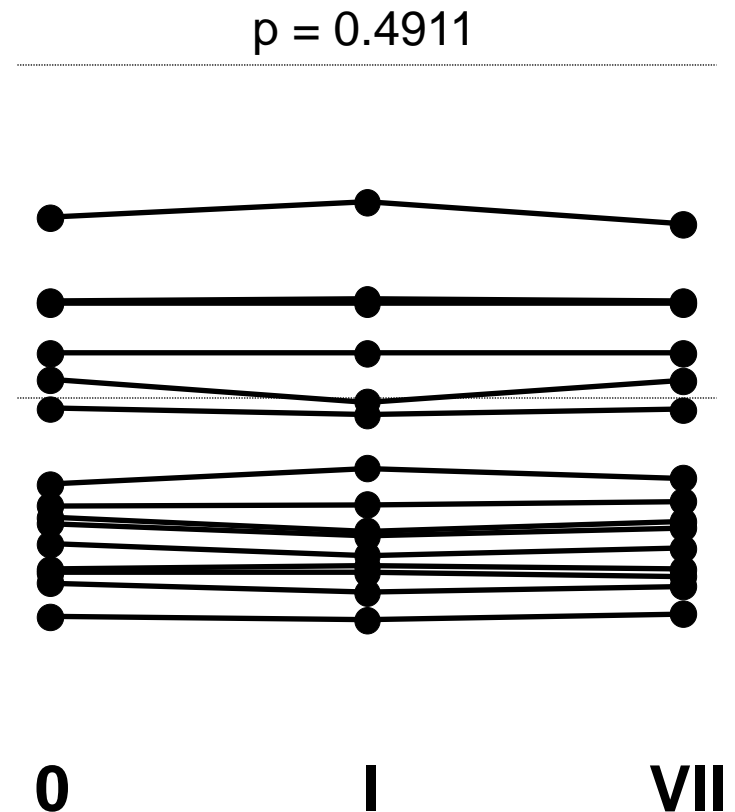
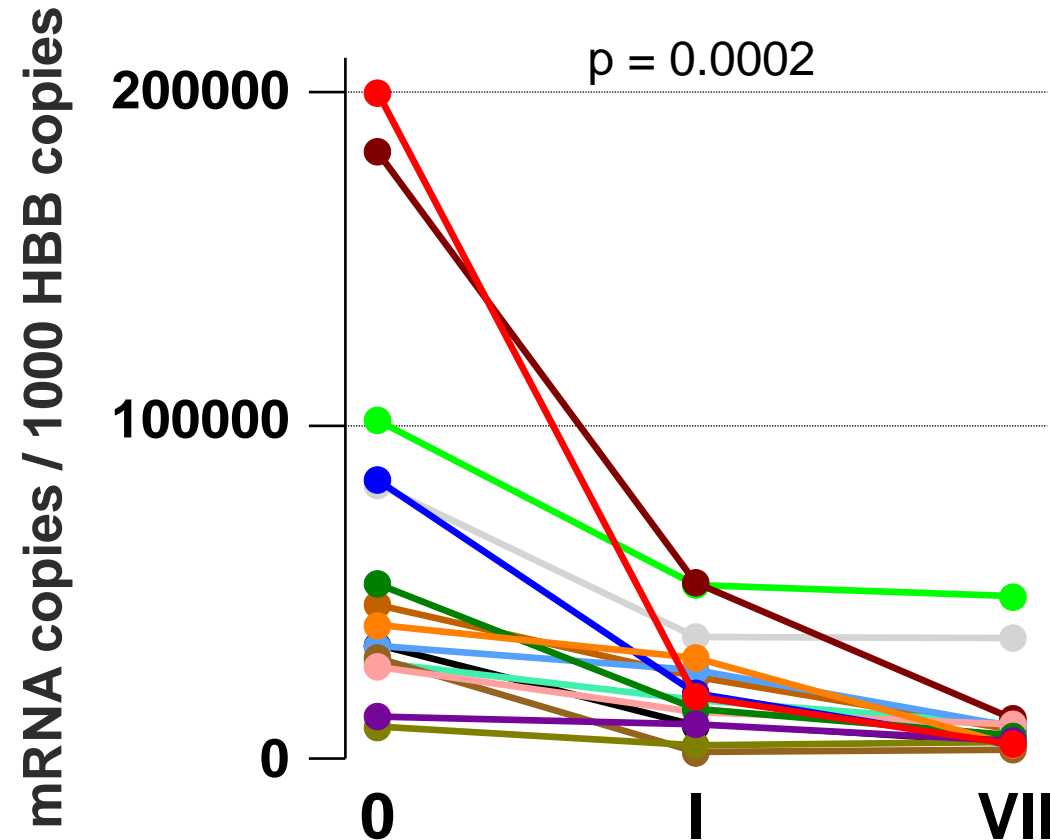


Tenofovir causes mitochondrial dysfunction

Mitochondrial ATP6 gene suppression

Tenofovir

Nonoxynol 9



AZT, another NRTI, was highly carcinogenic in topical rodent studies

Incidence of Vaginal Histopathologic Findings in Mice Given Zidovudine Intravaginally *(n = 50 / arm)*

Finding	Concentration (mg/ml)			
	EC	VC	5	20
Squamous cell carcinoma	0	0	2	13
Epithelial dysplasia	0	0	1	6

Note. EC, environmental control; VC, vehicle control.

Ayers KM et al.

Nonclinical toxicology studies with zidovudine: genetic toxicity tests and carcinogenicity bioassays in mice and rats.

Fundamental & Applied Toxicology 32:148-58 (1996)

Thus, carcinogenicity assessment of topical microbicides could be important, but is difficult

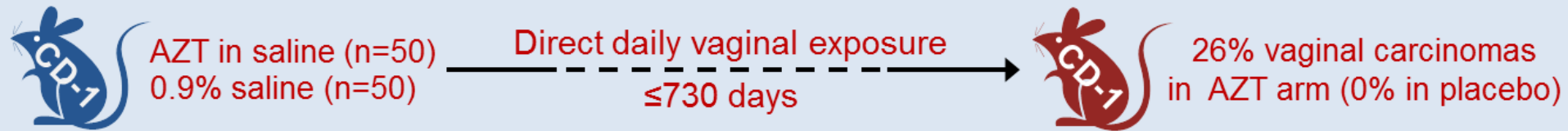
NIH/NIAID R01 AI116292 (05/01/15 – 04/30/20)

Systems and Carcinogenic Impact Assessment of Topical Microbicides on the Human Mucosa

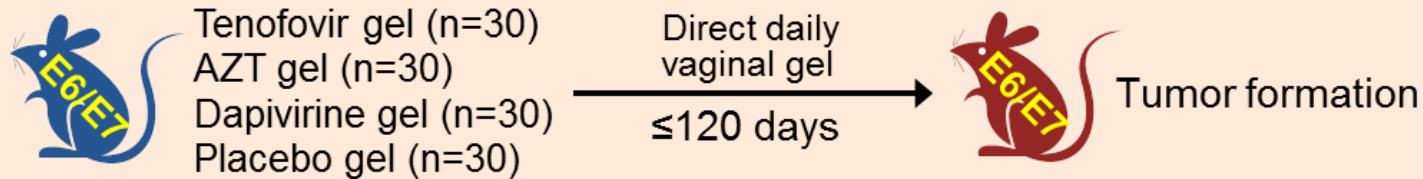
- Systems studies: MTN-014 and MTN-017**
- Carcinogenicity screening models**

Carcinogenicity screening of microbicides

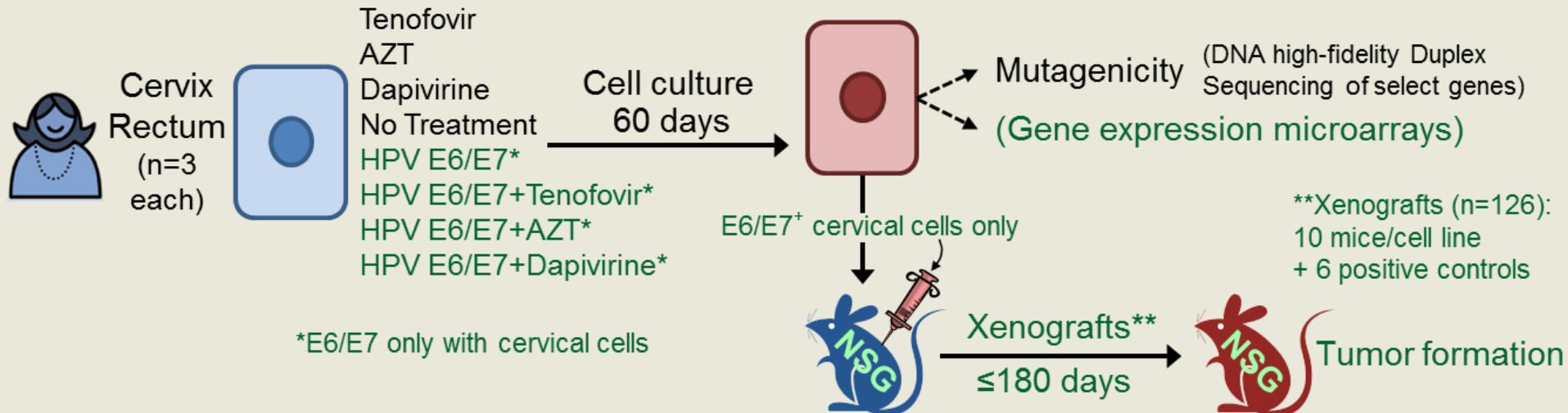
HISTORIC STANDARD MOUSE CARCINOGENICITY STUDY (excessively expensive)⁹



PROPOSED MOUSE CARCINOGENICITY STUDY

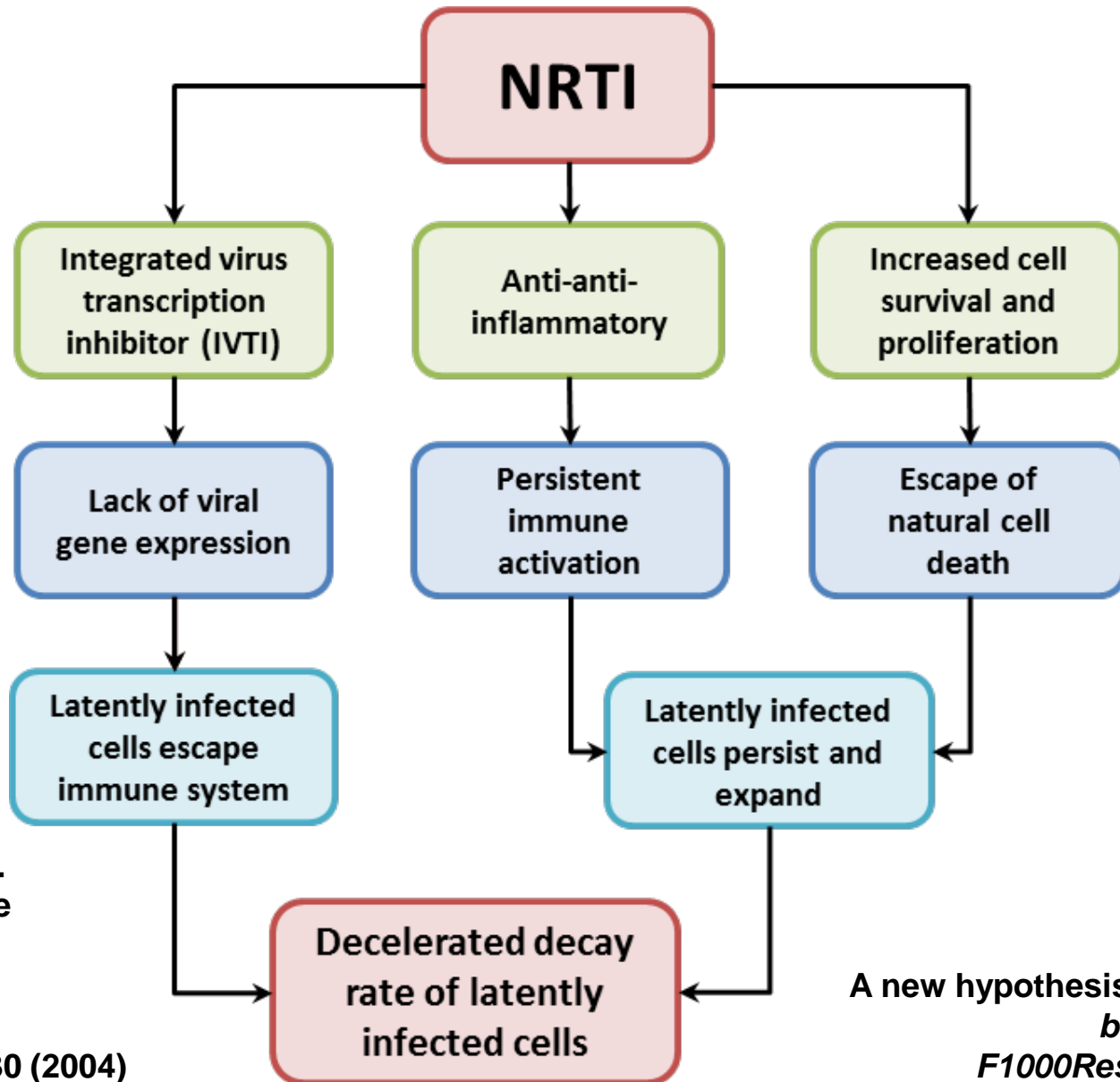


PROPOSED HUMAN CELL CARCINOGENICITY STUDY



III. Hypothetical ramifications beyond the microbicide field

Hypothesized effects of NRTIs on HIV latency



Saavedra-Lozano et al.
3TC and ABC decrease
p24 production from
HIV-infected resting
T cells after activation
J Virology 48:2825-2830 (2004)

Hladik F.
A new hypothesis on HIV cure
bioRxiv (2014)
F1000Research (2015)

Disclaimer

There is as yet no clinical evidence whatsoever for NRTI carcinogenicity or latency-prolonging effects

Acknowledgements

Ian McGowan
(MTN-007)

Lamar Fleming
Sean Hughes (Fred Hutch)

Julie McElrath
(Fred Hutch)

Adam Burgener
Blake Ball (Manitoba)

Craig Hendrix
(Johns Hopkins)

Raphael Gottardo
(Fred Hutch)

NIH / NIAID (MIG)
NICHD / NIMH

Study Participants
& Clinic Staff
MTN-007 Investigators