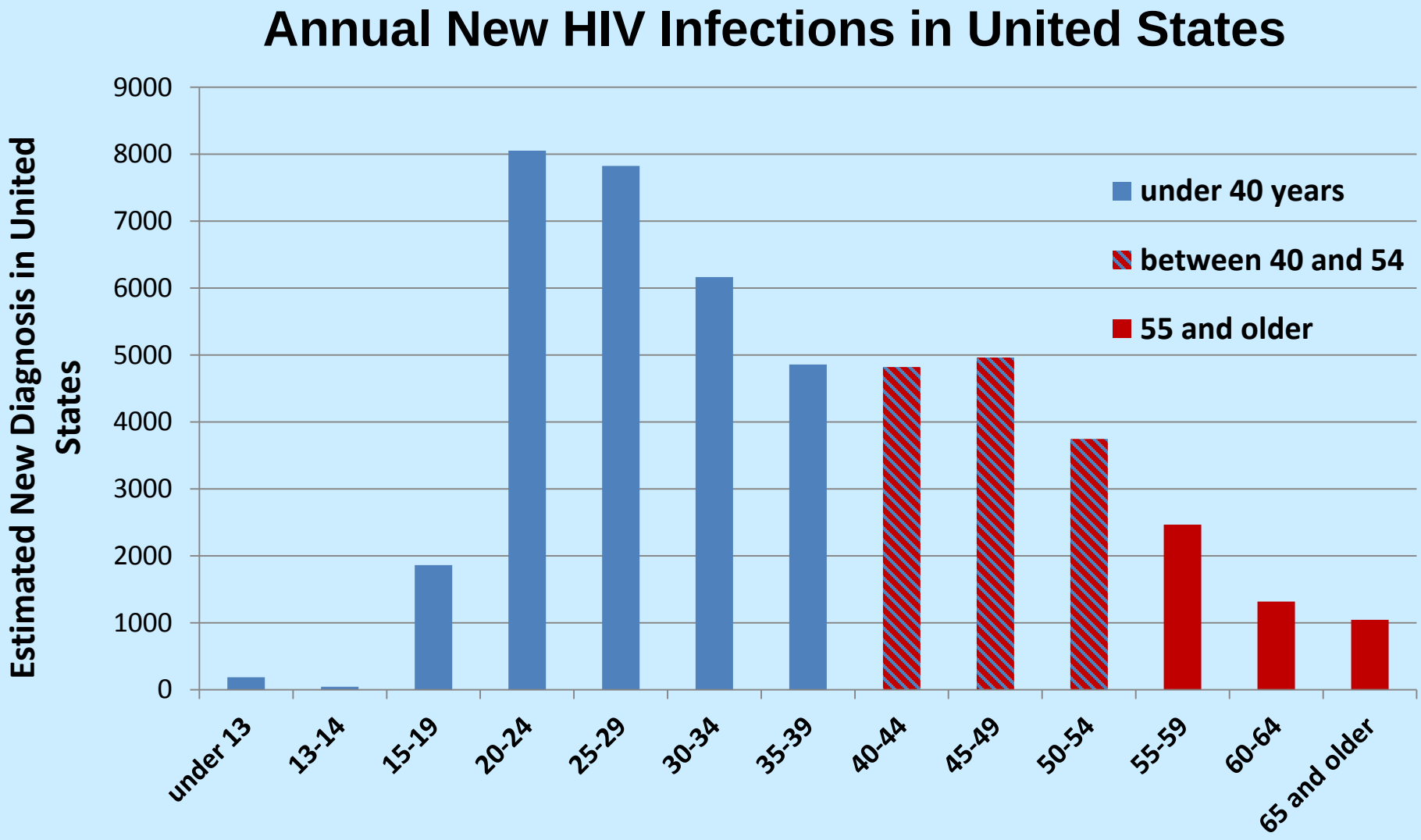
Reduced activation of emtricitabine and tenofovir in cervical and vaginal explants from postmenopausal donors

Melanie R Nicol[1], Lindsey Brewers[1], Angela DM Kashuba[2], Craig Sykes[2]

1. Department of Experimental and Clinical Pharmacology, University of Minnesota, Minneapolis, MN
2. Division of Pharmacotherapy and Experimental Therapeutics, University of North Carolina at Chapel Hill, Chapel Hill, NC

Background



Source: CDC. Diagnoses of HIV infection in the United States and dependant areas, 2013. *HIV Surveillance Report* 2015; vol.25.

Background: Biology of Menopause

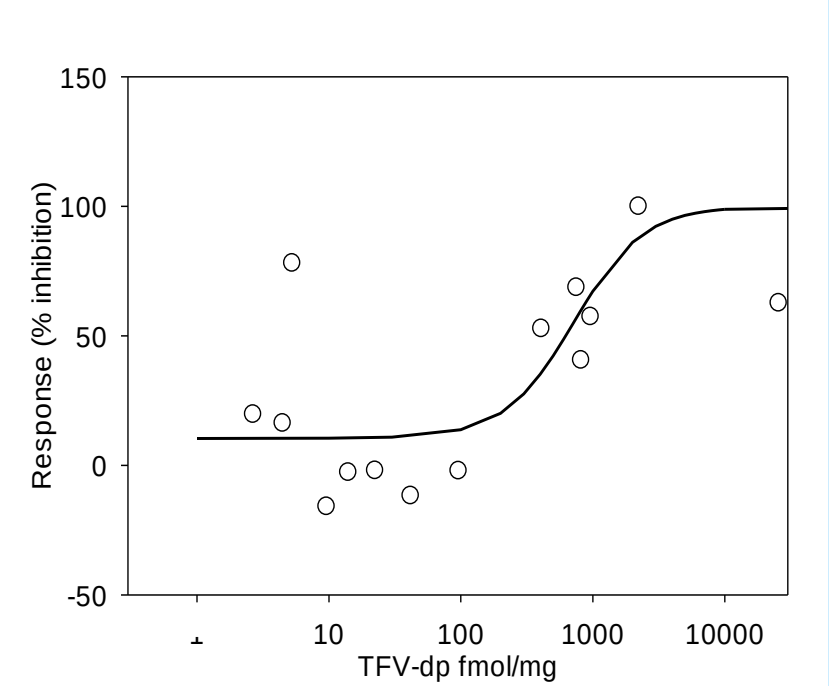
- Vaginal atrophy and epithelial thinning
- Changes in vaginal pH and microflora
- Decreased innate immunity
- Increased CCR5 expression

Potential for increased susceptibility

Farage, M. A. and H. I. Maibach (2011). Curr Probl Dermatol 40: 9-19.
Chappell, C. A., et al. (2015). Am J Obstet Gynecol 213(2): 204 e201-206.
Meditz, A. L., et al. (2012). J Acquir Immune Defic Syndr 59(3): 221-228.

Background

- Decreased tenofovir protection in postmenopausal cervical explants. (Thurman A *et al*. HIV R4P 2014. P44.11)
- Tenofovir metabolism to tenofovir diphosphate in vaginal explants is highly variable and significantly correlated with protection.



Nicol MR et al. *J Acquir Immune Defic Syndr.* 2015. 68(4):369-76.

Research Question: Does menopause affect the metabolism (activation) of nucleotide analogs in female genital tissues?

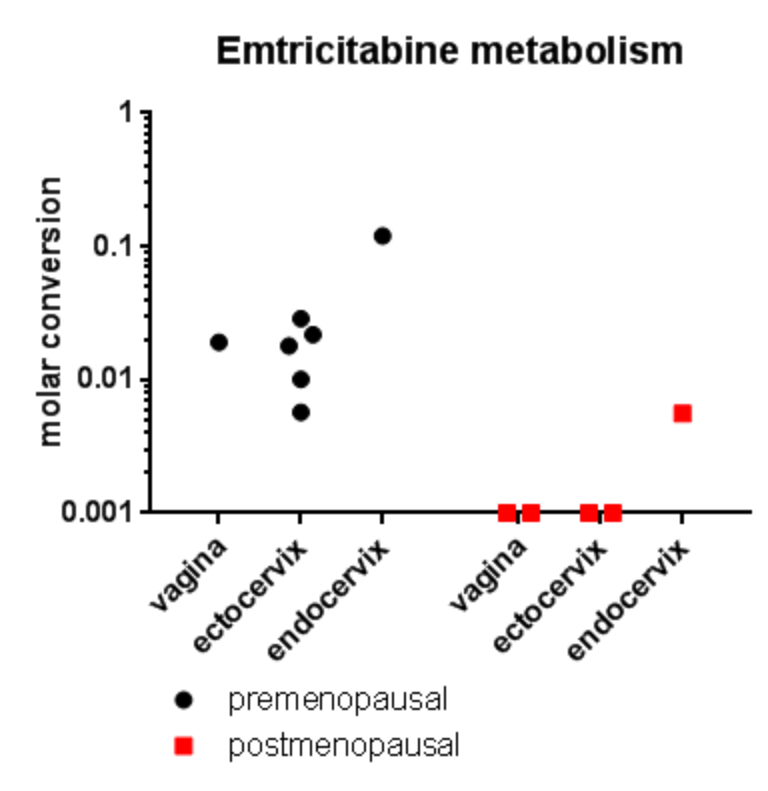
Methods

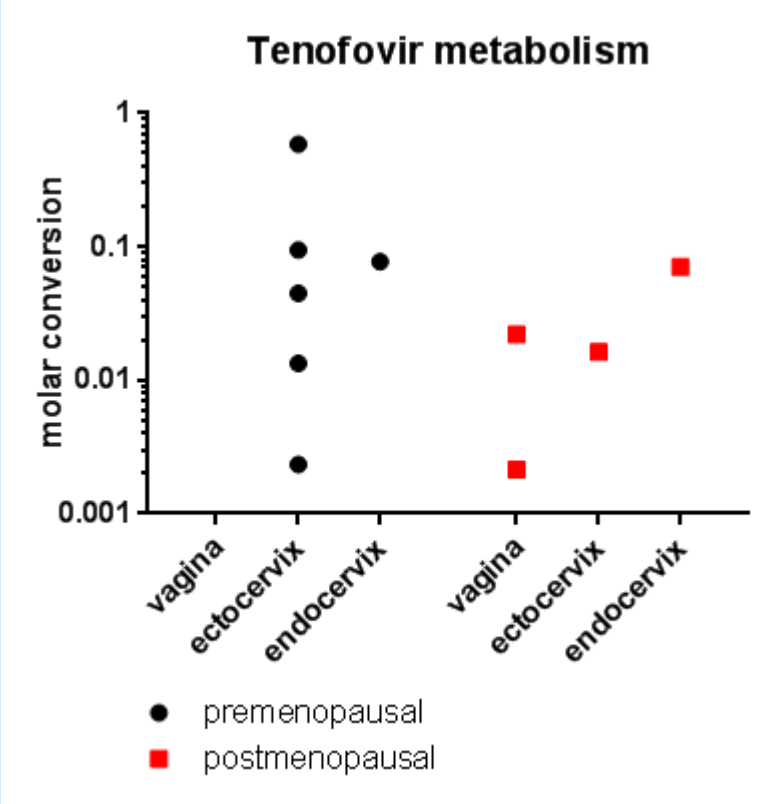
- Tissues from cadavers and women undergoing gynecologic surgeries
- 3mm explants from cervical and vaginal specimens
- Incubation in 100 µg/mL tenofovir or emtricitabine for 24 hours.
- Intracellular concentrations of TFV-dp or FTC-tp measured using LC-MS/MS
  - LLOQ 0.02 ng/mL
- Concentrations compared between groups using the Rank Sum Test

Results

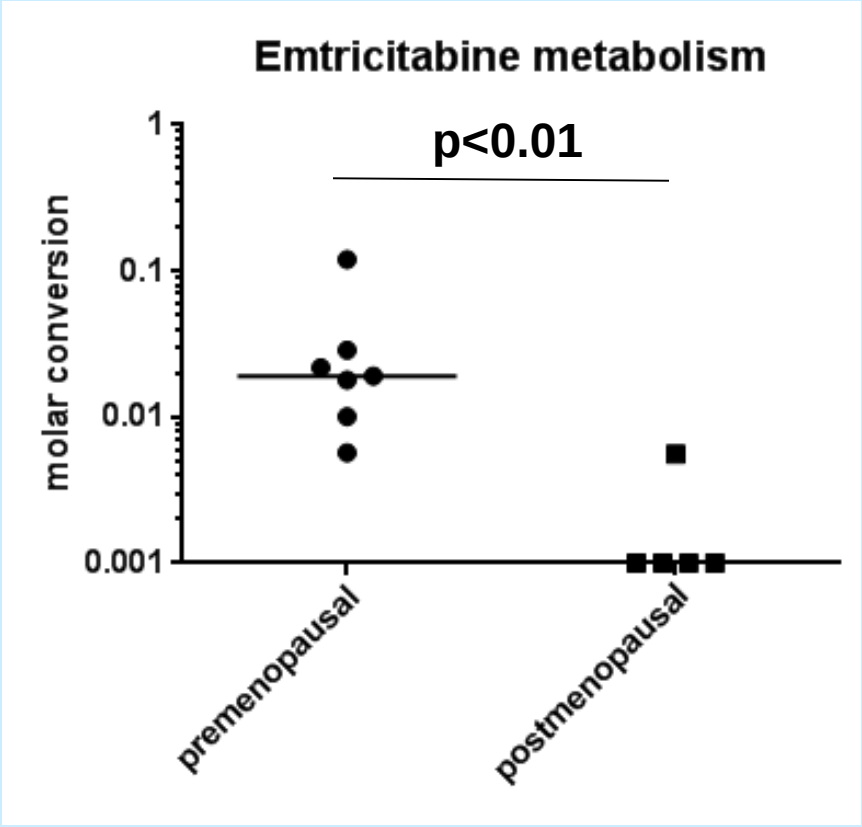
- 7 Pre menopausal (age range 32-51)
- 5 Post-menopausal (age range 52-68)
- FTC-tp was detectable in
  - 7/7 premenopausal explants
  - 1/5 postmenopausal explants
- TFV-dp was detectable in all (10) pre- and post-menopausal explants

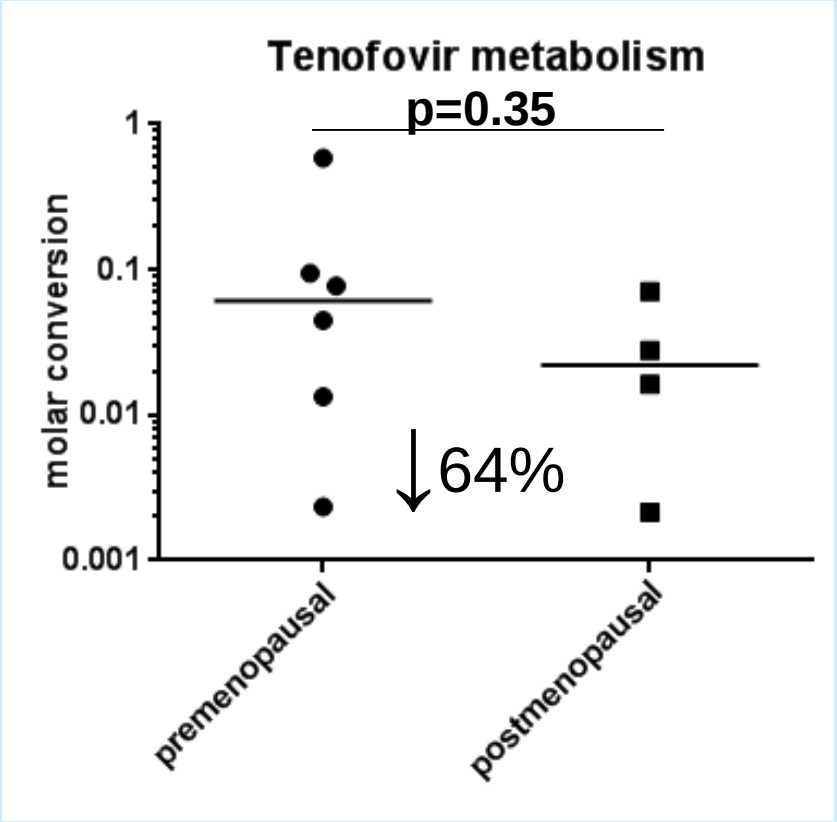
Results


Emtricitabine metabolism
molar conversion
1
0.1
0.01
0.001
vagina
ectocervix
endocervix
vagina
ectocervix
endocervix
premenopausal
postmenopausal


Tenofovir metabolism
molar conversion
1
0.1
0.01
0.001
vagina
ectocervix
endocervix
vagina
ectocervix
endocervix
premenopausal
postmenopausal

Results


Emtricitabine metabolism
p<0.01
molar conversion
1
0.1
0.01
0.001
premenopausal
postmenopausal


Tenofovir metabolism
p=0.35
molar conversion
1
0.1
0.01
0.001
64%
premenopausal
postmenopausal

Limitations and remaining questions

- Small sample size: could not assess for differences along the female genital tract
- Single concentration evaluated
- Did not measure endogenous dATP and dCTP
- No efficacy data

Next Steps

- Use *ex vivo* HIV challenge to determine if decreased concentrations confer decreased protection
- Examine expression of kinase/nucleotidase enzymes and transporters as potential mechanism

Conclusions

- Menopause was a significant predictor of intracellular nucleotide metabolism in cervical and vaginal explants.
- Postmenopausal tissues achieved lower concentrations of nucleotide active metabolites.
- Current pre-exposure prophylaxis strategies may have reduced efficacy in post-menopausal women.

Acknowledgements

- University of North Carolina Clinical Pharmacology and Analytical Chemistry Lab
- University of Minnesota Bionet Tissue Procurement
- National Disease Research Interchange
- NIH AIDS Reagent Repository