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Steady-state TDF/FTC in Genital, Rectal, and Blood Compartments in Males vs Females

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Abstract

Background
Despite the widespread use of tenofovir disoproxil fumarate and emtricitabine (TDF/FTC) for treatment and prevention of HIV infection, gaps in knowledge remain regarding the pharmacokinetics (PK) of the intracellular active moieties, tenofovir-diphosphate (TFV-DP) and emtricitabine-triphosphate (FTC-TP), at steady-state concentrations (Css) in different compartments in the body. The purpose of this study was to describe the steady-state PK of TFV-DP and FTC-TP in genital, rectal, and blood compartments in HIV-positive and negative males and females.

Methods
HIV-positive and negative adults were enrolled in an intensive PK study of daily TDF/FTC for 30 days. Peripheral blood mononuclear cells (PBMC) were obtained at first dose, days 3, 7, 20, and 30. Rectal biopsies and genital sampling were performed once each per participant, at staggered visits. Females underwent a cervical brush collection and males provided one semen sample. Cervical cells, seminal leukocytes, and rectal mononuclear cells were isolated, counted, assessed for viability, and lysed. First order PK was used to determine average steady-state values (Css) in each compartment for TFV-DP and FTC-TP. Comparisons were made between the genders and according to HIV status using unpaired t-tests, only for PBMC data given limits on sample sizes.

Results
Thirteen females (10 HIV negative) and 24 males (11 HIV negative) were included. Available samples from female participants included 13 cervical, 10 rectal, and 253 PBMC, and those from males included 18 seminal, 20 rectal, and 414 PBMC. Steady-state was reached in all compartments for both TFV-DP and FTC-TP within 30 days. Css values are shown in the table. TFV-DP and FTC-TP Css in PBMC did not differ significantly by gender or HIV-status, although females trended toward higher concentrations of TFV-DP in PBMC (p=0.08). Females also appeared to have higher TFV-DP in rectal cells. Genital cell concentrations at steady-state were ~10-fold greater in females compared with males for both TFV-DP and FTC-TP.

Table 1. Css of TFV-DP and FTC-TP in PBMC, genital, and rectal compartments in males and females (mean or point estimate, 95% confidence interval)

		Male [n=18]	Female [n=13]
Cervix	TFV-DP fmol/10^6 cells	89 (73.7 to 106.1)	108 (93.6 to 122.4)
	FTC-TP pmol/10^6 cells	5.5 (4.3 to 6.7)	5.6 (4.4 to 6.7)
Genital	TFV-DP fmol/million cells	11 (3 to 36)	150 (90 to 562)
	FTC-TP pmol/million cells	0.4 (0.2 to 0.7)	5.9 (2.3 to 9.5)
Rectal	TFV-DP fmol/million cells	1049 (559 to 1966)	3294 (992 to 10936)
	FTC-TP pmol/million cells	1.4 (0.5 to 1.9)	1.4 (0.3 to 2.6)

Conclusions
These findings illustrate differential drug penetration at steady-state in genital, rectal and blood compartments, which is relevant for prevention of HIV acquisition and suppression of HIV replication within potential reservoirs of the body. Females trended to have higher concentrations in these compartments relative to males. This information provides concentration ranges to help inform dose-response relationships for the prevention and treatment of HIV infection.

Background

Gaps in knowledge remain regarding the pharmacokinetics (PK) of the intracellular active moieties, tenofovir-diphosphate (TFV-DP) and emtricitabine-triphosphate (FTC-TP), at steady-state concentrations (Css) in different compartments in the body. Describing drug concentrations in these compartments may be relevant for prevention of HIV acquisition and suppression of HIV replication within potential reservoirs of the body.

Objective

To characterize Css of TFV-DP and FTC-TP in PBMC, cervical cells, seminal leukocytes, and rectal mononuclear cells in HIV positive and negative males and females.

Methods

HIV positive and negative males and females participating in “Cell-PrEP” (ClinicalTrials.gov NCT01040091) took daily TDF/FTC (+efavirenz in the HIV positive group) for 30 days.

First dose (day 1) and steady-state (day 30) PBMC samplings included 5 collections after an observed dose (approximately 1, 2, 4, 8, 24 hours post dose). Day 3, 7, and 20 visits included 3 collections surrounding an observed dose (approximately 0, 2, 8 hours post dose).

Rectal biopsies and genital sampling occurred only one time per subject on one of the visits between day 1 and day 30. Rectal biopsies took place approximately 2 hr post dose on one “on-drug” visit in all study subjects. Endocervical brush samples occurred at 2 hr post dose on one “on-drug” visit for female participants. Male subjects were asked to bring a semen sample from home at an assigned study visit. The subject recorded the time the sample was produced on a study form.

Rectal tissue was collected from participants by a gastroenterologist during sigmoidoscopy. The tissue was enzyme-digested prior to isolation of rectal mononuclear cells from the strained cell suspension, and RBC were lysed. Cervical cells were mechanically removed from the brush and RBC were lysed. Semen cells were isolated with ficoll.

Rectal mononuclear cells, cervical cells, and seminal leukocytes were isolated, assessed for viability, and lysed. Intracellular drug quantification was achieved with validated LC-MS/MS methodology. Results other than PBMC were corrected for viability.

PBMC, rectal mononuclear cell, and genital cell steady-state estimates were derived from 1st order equations using a 2-stage approach for PBMC (individual fits calculated in first stage, average taken of individual steady-state estimates in second stage) or naive-pooled fit for rectal and genital samples (all data pooled prior to fit) in GraphPad Prism. Concentrations of TFV-DP in rectal and genital samples were natural log transformed prior to fitting, reported Css estimates and 95% confidence intervals were backtransformed.

Results

Thirteen females (10 HIV negative) and 27 males (11 HIV negative) participated in the study. All but six participants, one female and five males, completed all study visits. Available samples from female participants included 13 cervical cell, 12 rectal mononuclear cell, and 253 PBMC. Samples from males included 18 seminal leukocyte, 23 rectal mononuclear cell, and 414 PBMC.

Results from unpaired t-tests found no statistically significant difference between steady-state TFV-DP concentrations in males vs females PBMC however females trended toward higher levels (p=0.06).

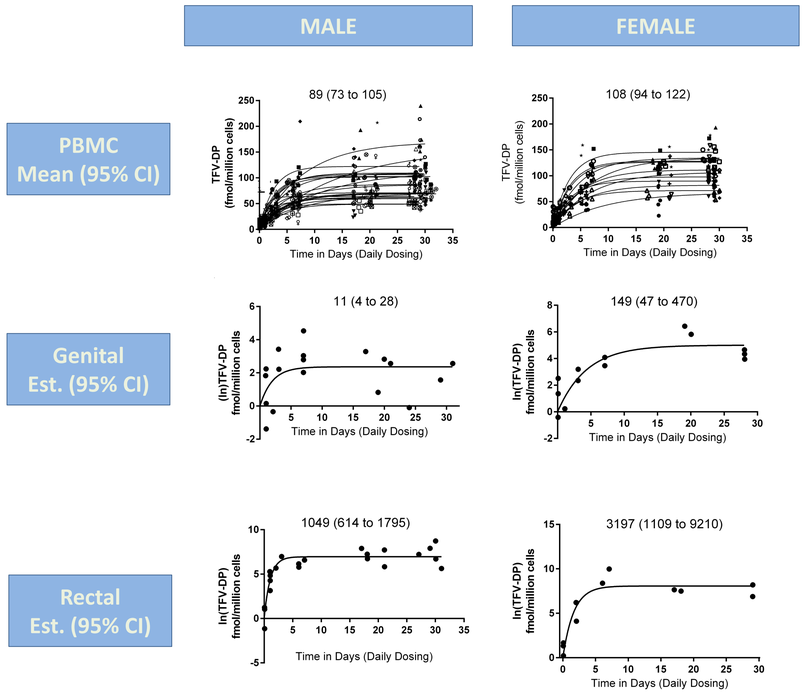
No differences were observed between steady-state concentrations of FTC-TP in PBMC in males vs females (p=0.88).

No differences were detected in steady-state concentrations of TFV-DP (p=0.38) and FTC-DP (p=0.97) in PBMC according to HIV status.

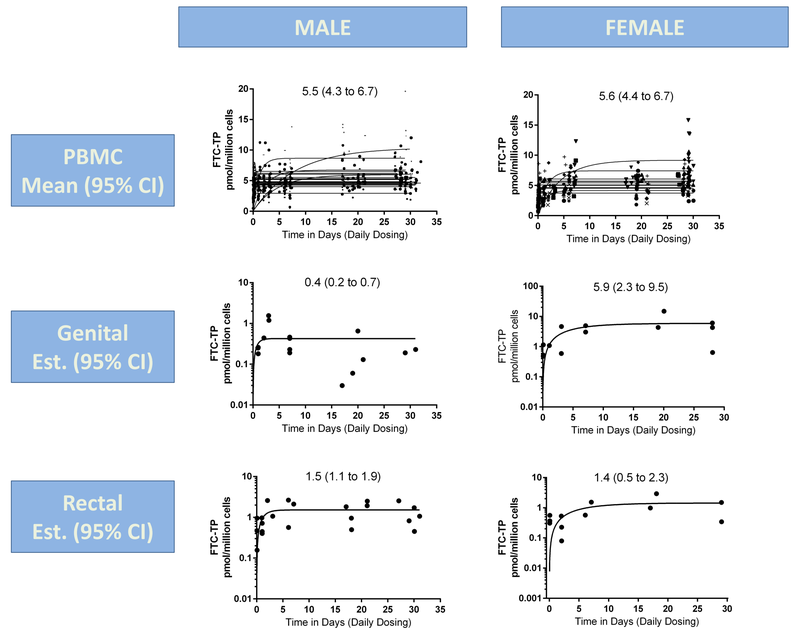
Participant Demographics

	Male	Female	Total
N (%)	27 (67.5%)	13 (32.5%)	40
HIV-positive HIV-negative N (%)	16 11	3 10	19 (47.5%) 21 (52.5%)
Age (yrs) Median (range)	35.5 (20 – 49)	28 (22 – 52)	31 (20 – 52)
Race/Ethnicity N (%)	Caucasian: 11 Afr-American: 11 Hispanic: 5	Caucasian: 8 Afr-American: 5 Hispanic: 0	Caucasian: 19 (47.5%) Afr-American: 16 (40%) Hispanic: 5 (12.5%)
Weight(kg) Median (range)	79 (56.5 - 124.4)	81.2 (60 – 113.2)	81.1 (56.5 – 124.4)

TFV-DP Css Estimates (fmol/million cells)



FTC-TP Css Estimates (pmol/million cells)



Conclusions

TFV-DP and FTC-TP demonstrated differential drug penetration at steady state in blood, rectal, and genital compartments in HIV positive and negative males and females, with rectal TFV-DP Css 10-fold higher than those observed in blood and genital compartments.

Women trended toward higher steady state concentrations of TFV-DP in PBMC compared to males however a preliminary analysis using unpaired t-tests revealed no statistically significant differences in Css of TFV-DP and FTC-TP in PBMC according to HIV status or gender. A more extensive analysis is underway using a multivariable regression and a linked plasma-intracellular pharmacokinetic model.

These findings provide steady-state concentration ranges that inform dose-response relationships for the prevention and treatment of HIV.

Acknowledgments and References

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