Determining critical physiological and drug specific parameters for enhancing a physiologically-based pharmacokinetics model for the female reproductive tract

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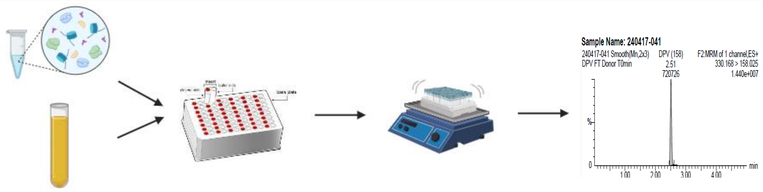
BACKGROUND

Our physiologically-based pharmacokinetics (PBPK) model can be utilized to predict drug exposure within the female reproductive tract (FGT). We aimed to expand our PBPK model to include fallopian tubes as well as enhance the sensitivity of this model. This work fills two gaps: 1) evaluation of gene expression of drug transporters and drug metabolizing enzymes (DMEs) in the FGT tissues including fallopian tubes, and 2) evaluation of drug specific parameters (biological fluid protein binding and solubility and tissue protein binding) for drugs representative of those under development as new non-hormonal contraceptive agents.

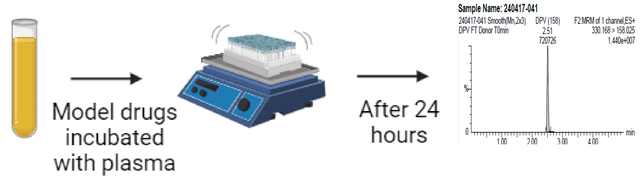
METHODS

Model drugs	Dapivirine	MK-2048	EFdA
Log P	5.6	1.62	-0.42

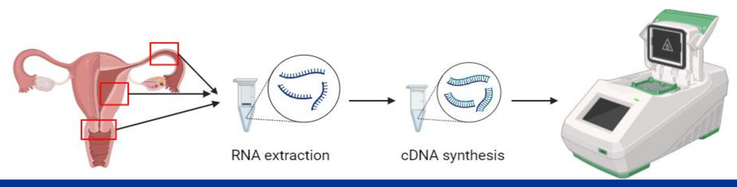
Protein Binding Studies



Solubility Studies



Gene Expression Studies



Genes of Interest	
Efflux Transporters	Pgp/*ABCB1/MDR1*, BCRP/*ABCG2*, MRP1/*ABCC1,* MRP4/*ABCC4,* MRP5/*ABCC5,* MRP7/*ABCC10*
Uptake Transporters	OCT2/*SLC22A2,* OCT3/*SLC22A3,* ENT1/*SLC29A1,* ENT2/*SLC29A3,* OATP-D /*SLCO3A1,* OAT3/*SLC22A8*
Phase I DMEs	CYP1A1, CYP1B1, CYP1A2, CYP2E1, CYP2C9, CYP2C19, CYP2B6, CYP2D6, CYP3A4, CYP2C8
Phase II DMEs	UGT1A1, UGT1A3, UGT1A4, UGT1A7, UGT1A8, UGT1A9, UGT1A10, UGT2B4, UGT2B7, UGT2B15, UGT2B17

Statistical Analysis: Outcomes were compared using one-way ANOVA, with *p<0.05, **p<0.01, ***p<0.001, ****p<0.0001

RESULTS

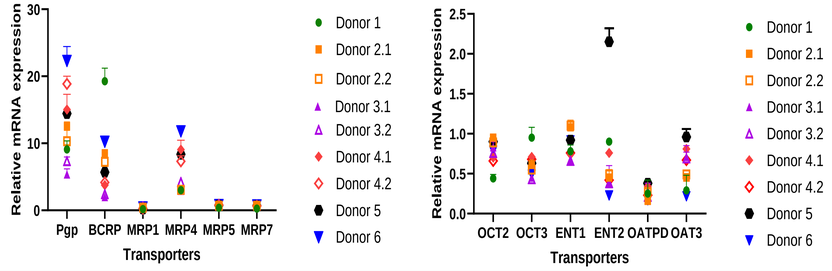
Plasma/CVF Protein Binding & Plasma Solubility

Model drugs	Plasma (%, mean±SD)	CVF (%, mean±SD)	Solubility (μg/mL, 24 hours, mean±SD)
Dapivirine	99.06±1.27 (n=6)	99.47±0.07 (n=3)	34.14 ± 1.04 (n=3)
MK-2048	96.12 ± 0.33 (n=6)	84.75±3.00 (n=4)	456.92 ± 38.59 (n=3)
EFdA	7.41 ± 4.73 (n=5)	13.58±5.07 (n=5)	1808.02 ± 67.36 (n=3)

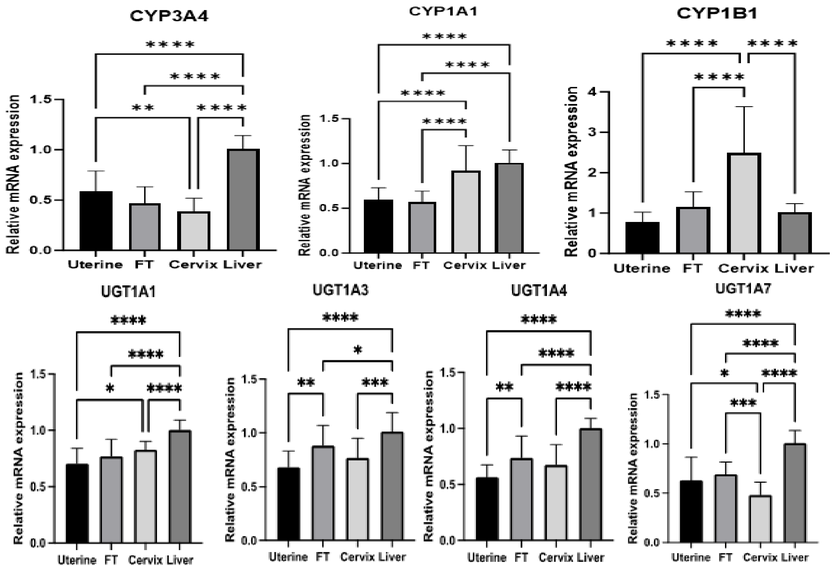
Tissue Protein Binding (%)

Model drugs	Cervical epithelium	Cervical stroma	Myometrium	Endometrium	Fallopian Tubes
Dapivirine	98.27±0.8		98.82±0.16	97.49±1.33	99.45±0.02
MK-2048	89.42±2.6	85.95±3.02	82.90±0.62	86.79±7.06	92.57±0.53
EFdA	13.28±4.1	8.01±7.81	17.97±10.06	24.98±1.22	7.18±7.33

Gene Expression – Drug Transporters (n=6)

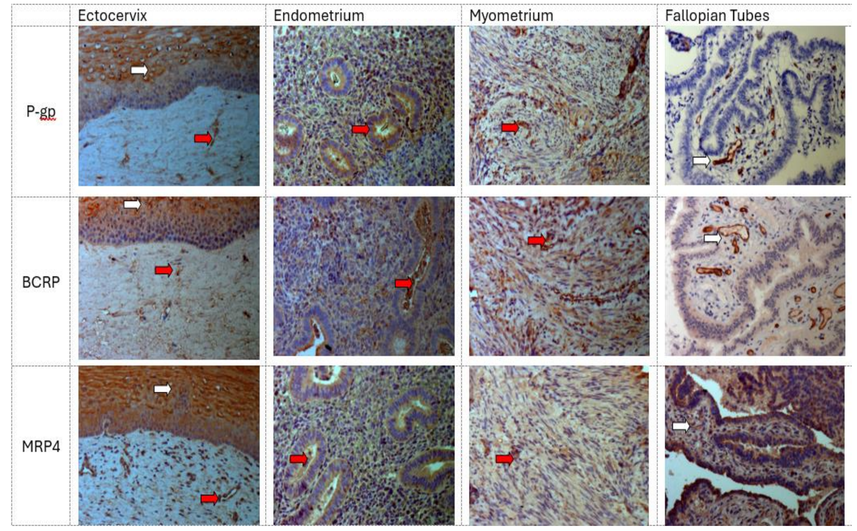


Representative Gene Expression – Drug Metabolizing Enzymes (n=6 for each tissue type)



RESULTS (Continued)

Immunohistochemistry Staining



Localization of P-gp, BCRP, and MRP4 proteins in human ectocervix, endometrium, myometrium, and fallopian tubes tissues. White arrows: epithelial cells and mucosal folds (fallopian tubes), and Red arrows : vascular endothelial cells. Magnification: 20× - 40x

SUMMARY & FUTURE DIRECTIONS

- ❑ Protein binding (human plasma, CVF, and tissues) was highest for the hydrophobic drug. Plasma solubility was greatest for the hydrophilic drug.
- ❑ Expression of P-gp, BCRP, and MRP4 efflux transporters was increased in human fallopian tubes compared to liver.
- ❑ 15/21 Phase I and II DMEs (highlighted red) tested had mRNA expression levels > 50% of that found in liver for all tissue types evaluated (human uterine, fallopian tube, and cervix).
- ❑ IHC localized P-gp, BCRP, and MRP4 transporters to the epithelium, submucosa, and/or the mucosal folds.
- ❑ Characterize and correlate permeability of the model drugs using *in vitro* and *ex vivo* systems
- ❑ These parameters will be used to expand our current PBPK model for the female reproductive tract to include the fallopian tubes.

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