Uptake And Efflux Of Abacavir And Tenofovir By Human Renal Transporters: A Mechanism For Drug-Drug Interactions?

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In 2008 two million new cases of HIV were reported world-wide, however death rates are decreasing due to advances in drug therapy (World Health Organisation, 2008). Highly active anti-retroviral therapy (HAART) has helped to reduce viral loads and increase survival time. Tenofovir, a nucleotide reverse transcriptase inhibitor, has been associated with acute and chronic nephrotoxicity, attributable in part to transport-mediated drug-drug interactions (Zimmermann et al., 2006). Determination of the mechanism(s) of uptake and efflux of anti-viral drugs could help to predict drug-drug interactions, resulting in clinical benefits for patients.

Uptake transporters investigated were the human organic anion transporters (hOATs) -1 and -3, and the human organic cation transporters (hOCTs) -1 and -2. The efflux transporters studied were the human multi-drug resistance proteins (hMRPs) -2 and -4. Transiently transfected HEK293 cell lines expressing hOATs, stably transfected HEK293 cells expressing hOCTs and commercially available Sf9 inside-out vesicles expressing hMRPs were used to determine transport specificity. The transfected cell lines or Sf9 inside-out vesicles were incubated with substrates for 5 minutes at 37 °C, washed with PBS or transport buffer and any internalised substrate measured by scintillation counting, fluorescence detection or LC-MS-MS. p-Aminohippurate (PAH) and tetraethylammonium bromide (TEA), were used as probe substrates for hOATs and hOCTs, while estradiol-β-glucuronide (EβGlu) and 5(6)-carboxy-2,7-dichlorofluorescein (CDF) were probe substrates for hMRPs.

PAH, TEA, CDF and EβGlu were selective substrates for hOAT1, hOCT1, hMRP2 and hMRP4 respectively. PAH is a better substrate for hOAT1 than for hOAT3. CDF, at 10 μM, is a probe substrate for hMRP2, but not hMRP4. Tenofovir, 10 μM, was transported by hOAT1 at 197-fold greater amount than in untransfected control cells (p<0.001) with a Km of 400 ± 80 μM and a Vmax of 5500 ± 400 pmol/min/mg. A 3-fold increase in uptake of tenofovir was observed with hOCT2 compared with control cells. Tenofovir showed ATP-dependent efflux by hMRP4 which was 2-fold greater than the efflux with AMP. Abacavir was not a good substrate for any of the renal transporters investigated, but was taken up by hOCT1 by a 4-fold greater amount than control cells.

In conclusion tenofovir is actively excreted by hOAT1, and possibly hOCT2, on the basolateral membrane of renal cells and effluxed by hMRP4 on the apical border. This correlates with in vivo renal clearance data for tenofovir (243 mL/min) compared with abacavir (approximately 10 mL/min). Therefore inhibition of hOATs by concurrently administered drugs may result in increased plasma concentrations of tenofovir, while hMRP inhibition could result in cellular accumulation and toxicity.

References:
