ABSTRACT

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Title of doctoral thesis: Interactions of antiretroviral drugs with membrane transporters

Oral delivery is the most common, convenient, and economical form of drug administration. Absorption of orally administered drugs occurs mainly in the intestine. Intestinal absorption can be reduced by the activity of efflux drug ABC transporters, mainly p-glycoprotein (ABCB1) and breast cancer resistance protein (ABCG2), located in the apical membrane of the intestinal epithelium.

HIV-infected patients are dependent on lifelong pharmacotherapy, which includes a combination of three or more antiretroviral drugs. Hepatitis C (HCV) is a common co-infection of HIV. In addition, the HIV-positive population is aging, which is associated with burden of other comorbidities. This results in an indication of polypharmacy and thus an increased risk of drug-drug interactions. Many antiretroviral drugs used are substrates, inhibitors and /or inducers of ABCB1, so they might quantitatively affect the intestinal absorption of co-administered drugs (ABCB1 substrates), thereby affecting the efficacy/safety of treatment. As part of this dissertation, we focused on drug-drug interactions of anti-HIV and anti-HCV drugs on the intestinal ABCB1 transporter and on the introduction of an *ex vivo* model for analysis of the induction of intestinal ABCB1.

Using *in vitro* and *in vivo* methods, we have shown that abacavir is a substrate of intestinal ABCB1 and ABCG2 and that its intestinal barrier permeability can be increased by co-administration of rilpivirine. In the next phase of our research, we for the first time used a recently established *ex vivo* method based on the accumulation of rhodamine123 in precision-cut intestinal slices (PCIS) prepared from rat ileum and human jejunum and we assessed the potency of clinically relevant therapeutics to inhibit intestinal ABCB1. We tested, anti-HIV and anti-HCV drugs and we identified lopinavir, ritonavir, saquinavir and atazanavir as significant inhibitors.

In addition to inhibiting ABCB1, another important type of interaction is the induction of the ABCB1 transporter. Due to a number of shortcomings in the currently available models, the drug research and drug policy authorities do not provide any recommendations for testing the induction potential of ABCB1 *in vitro*. Therefore, in our further work we focused on the development of the PCIS methodology for the induction of intestinal transporters. We found that PCIS incubated for 48 hours retained intact morphology, ATP content, and full ABCB1 function. Rifampicin, a model ligand for the pregnane X receptor (PXR), significantly increased functional ABCB1 expression and *CYP3A4* gene expression.

Our results may help to elucidate the molecular basis of the described increase in the bioavailability of some ABCB1 substrates when they are co-administered with anti-HIV and / or anti-HCV drugs. We further demonstrated that the PCIS method allows to conduct inhibition and induction studies focused on ABCB1 parallelly, which makes this method interesting for preclinical studies. At the same time, this method could be very important for quantifying the contribution of intestinal ABCB1 and drug interactions in pre-systemic drug elimination.