Abstract

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Title of diploma thesis: Effect of dexamethasone on ABCB1 and CYP3A4 expression in human precision-cut intestinal slices

Dexamethasone is a corticosteroid that has anti-inflammatory, immunosuppressive, and anti-allergic effects. The mechanism of dexamethasone action involves its ability to bind to intracellular glucocorticoid receptors, which are present in many types of cells, including intestinal mucosal cells. Upon binding to these receptors, it translocates to the nucleus of the cells and affects gene expression. This mechanism also affects the expression of genes important for the metabolism and transport of xenobiotics in the intestinal mucosa. The most important such genes include *ABCB1*, an important intestinal transporter, and *CYP3A4*, a significant biotransformation enzyme. Their localization in the wall of the small intestine can significantly affect the absorption of orally administered drugs. Studying drug interactions with this transporter and biotransformation enzyme is important for safe and effective pharmacotherapy.

To determine whether dexamethasone affects the expression of these genes in the intestinal barrier, we used the method of precision-cut tissue slices. Precision-cut intestinal slices (PCIS) represent a mini-model of the intestinal barrier and contain all types of cells in the studied tissue. This method allows the study of the behavior of the intestinal barrier in real-time and in a physiological environment, which can contribute to a better understanding of the mechanisms that affect the pharmacokinetics of drugs. However, there is limited information on the use of this technique in studying gene expression induction.

First, we focused on the effect of dexamethasone on the viability of intestinal slices during incubation. Then we investigated whether dexamethasone affects the expression of *ABCB1*, *CYP3A4*, and glucocorticoid receptor in our chosen model of the intestinal barrier. The last goal was to determine whether any effect on gene expression affects the transport of the model substrate rhodamine123 through *ABCB1* in unaffected and affected intestinal slices.

We incubated the intestinal slices with dexamethasone at concentrations of 50 μM and 100 μM for up to 48 hours. The known inducer rifampicin was used as a positive control at a concentration of 30 μM. Gene expression was evaluated by quantitative polymerase chain reaction (qPCR).

Measurements of the ATP concentration show that dexamethasone has no negative effect on tissue viability. Its effect on CYP3A4 and ABCB1 was observed for both studied concentrations. A significant increase was observed for CYP3A4 at 24 and 48 hours and ABCB1 at 48 hours. Glucocorticoid receptor expression was below the control level at all times and in all groups, including 50 μM dexamethasone and 30 μM rifampicin. Only at 100 μM of dexamethasone after 12 hours was the expression slightly above the control level. Intracellular rhodamine123 concentrations were not affected by induction in most cases. Only dexamethasone 100 μM decreased the concentration, but this was only in one experiment.