ABSTRACT

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Title of Doctoral Thesis **Study of substances affecting permeability of the skin barrier**

The skin barrier plays a vital role in protecting the human body and enables mammals' life on dry land. The epidermis has the primary barrier function due to several cells' layers, which gradually differentiate to their final stage, the stratum corneum (SC). SC is formed by stratified keratinocytes (known as corneocytes) surrounded by a lipid matrix. This intercellular matrix consists of an approximately equimolar ratio of ceramides, free fatty acids and cholesterol. These are particular substances formed in the epidermis from their precursors during the keratinocyte's differentiation, and their arrangement into the multilamellar structure is essential for the impermeability of the skin barrier. However, some substances or factors can disrupt the skin barrier. It is usually an undesirable process of lipid disbalance resulting in disorders or diseases of the skin barrier. On the other hand, specific substances have been developed for a reversible disruption of the skin barrier (so-called enhancers) to allow drug penetration in (trans)dermal drug delivery. The aim of this work was to study both groups of these substances and their effect on the skin barrier using appropriate skin barrier models. Lipid models have been used for their advantage of easily modifiable structure, as well as the possibility to separate other biochemical processes that occur in the skin. These advantages allowed to study the effect of the ceramide precursor and the changing of the pH during the skin barrier formation. A new function of acidic pH in the skin barrier was discovered, more specifically, a direct influence of pH on the formation of the unique multilayer structure in the lipid matrix. Furthermore, interactions of skin barrier lipids with potential enhancers - dendrimers - were monitored using lipid models. It could be an interesting method for characterizing substances that interact with skin lipids in future use. Next, 3D cell models of the epidermis or skin were used. These models can be genetically modified to simulate skin diseases. Another advantage is the possibility to observe the effect of individual skin components on a more complex scale. It was prepared a reconstructed human epidermis with a defect in the synthesis of ω-O-acylceramide and using lipid formulations was observed effect of ω-O-acylceramide's delivery on skin barrier regeneration. Two types of models were compared, and a protocol for formulations application was developed. Reconstructed human skin models were prepared by colleagues in Germany to observe interactions between cells of the epidermis (keratinocytes) and dermis (fibroblasts). It was found that the presence of fibroblasts is essential for the differentiation of keratinocytes into the skin barrier, which was also achieved through SC lipids analyzes performed in our laboratory. The last model used in this work were ex vivo skin grafts. Thanks to its composition, it is the most complex model but without the possibility of being modified (like the previously described models). Nevertheless, it is often used for its barrier function. In this work, it was used for testing in our research group newly synthesized skin permeation enhancers. Advantageous properties of terpene esters to deliver cidofovir to the skin without adverse side effects were found. Furthermore, the effect of generation and concentration of dendrimers on (trans)dermal delivery of 5-fluorouracil was investigated. Surprisingly, the lower tested concentrations of the third generation of synthesized dendrimers appeared to be the most suitable.
Thus, new findings in the field of substances affecting the skin barrier were discovered, and at the same time, it was proved that the models have an irreplaceable role in their research. It should be mentioned that it is not possible to choose one universal model, but it is always necessary to adapt it to the intended research.