ABSTRACT

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Title of diploma thesis: ***In vitro* Methods for the Prediction of Blood Brain Barrier Penetration**

This thesis deals with the correlation between two *in vitro* models simulating the blood-brain barrier (HEB, hematoencephalic barrier) and their comparison in terms of practical use. These are the PAMPA (Parallel Artificial Membrane Permeability Assay) method and the MDCK (Madin-Darby Canine Kidney) cell line, which are models for potential central nervous system (CNS) penetration screening. Within this work, a set of sixteen standard drugs were measured. The procedure was similar in both methods in order to obtain information on the amount of test substances passing through the membrane from the donor portion of the plate to the acceptor. The concentration in the donor portion was measured by UV-VIS spectrophotometry. The main difference between these methods is the membrane through which the substances penetrate. In the case of PAMPA, a lipid solution that has been isolated from pig brain (PBL, polar brain lipid) is used. This lipid simulates the phospholipid membrane of the brain capillary endothelium. In the MDCK model, the membrane is a monolayer of MDCK cells that are grown on a microporous membrane located between the donor and acceptor portions of the system. The detected donor concentration was converted to permeability coefficients and the methods were compared with each other and also with *in vivo* permeability. For thirteen drugs, the results from both methods corresponds to the *in vivo* situation. It has been found that the best correlation is for substances that pass through HEB by passive diffusion. In the PAMPA model, a wrong prediction was found for substances using active transport mechanisms, which the MDCK model largely eliminated. Although the MDCK model contains active transporters, they do not all have the same substrate specificity as in the HEB. In addition, some transporters present in the HEB are missing. Therefore, the MDCK model erroneously determines the transmission of substances that are subject to transporters that are specific to HEB. The PAMPA method is fast, cheap and suitable for screening larger sets of substances compared to the MDCK model. A partial goal was to determine the effect of dimethylsulfoxide (DMSO), which was used as a co-solvent in sample dissolution. The DMSO concentrations used were found not to affect the reliability of the PAMPA method.

Key words: blood brain barrier, *in vitro,* PAMPA, MDCK cell, DMSO