Novel Drug Permeability Screening Platform Combining Microfluidics with LC-MS

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Membrane permeability of molecules is a key parameter for cellular drug absorption, but is also a challenge in drug discovery and development. Currently, mainly the parallel artificial membrane permeability assay (PAMPA) and Caco-2 cellular monolayers are used for drug permeability screening [1]. To overcome the shortcomings of the state-of-the-art, we use microfluidics, fluorescence microscopy, and liquid chromatography-mass spectrometry (LC-MS) to develop a novel platform consisting of droplet interface bilayers (DIBs), on a basis of water-in-oil droplet formation and spotting as previously developed in our lab [2, 3]. The membrane composition of DIBs can be tailored and their characteristics as an artificial bilayer between the compartments are more physiologically relevant than those of PAMPA. Together with the advantages of microfluidics, our assay operates with small sample amounts and shortened incubation times, therefore improving throughput.

We generated aqueous droplets of ~25 nL surrounded by a lipid monolayer with a microfluidic T-junction [3] and spotted them with micrometer-precision on a hydrophobically coated glass slide with hydrophilic spots in close proximity. Upon contact of donor and acceptor droplets, a lipid bilayer formed.

First, we optimized and evaluated the method with fluorescent dyes, which allowed kinetic measurements over time. We observed an increase in permeation velocity dependent on the temperature and the presence of cholesterol in the bilayers. In a second step, to measure permeation of non-fluorescent drugs, we developed a protocol that interfaces our platform with LC-MS. We were able to detect and quantify drug permeation around 30 times faster and with 8’000 times smaller compartments than PAMPA [4].

In conclusion, our herein descripted platform allows on-demand droplet generation, spotting, on-site investigation, and subsequent label-free LC-MS analysis, opening doors for permeability studies regarding (asymmetric) membrane composition. In future, we are aiming for embedding membrane proteins in DIBs and generating multi-compartmental artificial cell networks.

[1] M. Kansy, F. Senner, K. Gubernator, *J. Med. Chem.*, **1998**, *41*, 1007-1010.
[2] D. Hümmer, C. Bärtschi, R. Steinhoff, J. Förster, L. M. Blank, R. Zenobi, P. S. Dittrich, *Proceedings of MicroTAS 2017*, **2017**, 201-202.
[3] S. K. Küster, S. R. Fagerer, P. E. Verboket, K. Eyer, K. Jefimovs, R. Zenobi, P. S. Dittrich, *Anal. Chem.*, **2013**, *85*, 1285-1289.
[4] K. F. Hermann, C. S. Neuhaus, V. Micallef, B. Wagner, M. Hatibovic, H. E. Aschmann, F. Paech, R. Alvarez-Sanchez, S. D. Krämer, S. Belli, *Eur. J. Pharm. Sci.*, **2017**, *104*, 150-161.