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## Development of a Plastic Membrane Containing Micro-hole(s) for a Potential Bio-Sensing Application

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A poly(methylmethacrylate) (PMMA) membrane containing single or several micro-holes (MHs) as a prototype of a simple sensing platform of a lab-on-a-chip device for a potential analysis of clinical samples has been developed. MHs with a diameter in the range of 20 to a few hundred  $\mu\text{m}$  were fabricated using laser ablation and mechanical micro-milling. Laser ablation enabled to cut MHs down to 20  $\mu\text{m}$  in diameter. However better reproducibility and MHs structural resolution was achieved with micro-milling. The MHs were characterised by optical and scanning electron microscopy as well as electrochemical impedance spectroscopy (EIS). Four probe EIS setup, with two electrodes placed on each side of a membrane, was adopted for monitoring of MH impedance. To investigate, if EIS could be used to sense the trapping of an analyte in the MHs, latex micro-beads of different sizes were tested for clogging MHs. For this purpose, the surface of MHs was rendered hydrophilic using air plasma, and the MH trapping exploited by capillary forces. The beads in the MH were cross-linked by using glutaraldehyde vapour to retain the stability of the MH-based sensor construction. The influence of the aspect ratio of MHs on MH impedance was evaluated when using PMMA membranes of different thicknesses. EIS measurements on a single 100  $\mu\text{m}$  diameter MH showed that the presence of beads in the MH considerably increased the impedance of the MH. When comparing the ratio of the impedance of the modified and unmodified MHs, the higher impedance ratio, i.e. the higher normalized response, was observed for the MHs in 500  $\mu\text{m}$  membrane than the ones in 250  $\mu\text{m}$  membrane. Thus, the lower MH aspect ratio enables the higher normalized response of MH-based impedance measurement. Finite element model simulations were performed using Comsol Multiphysics software to theoretically evaluate the electric current distribution and the sensitivity field of the EIS measurement through the MH(s), thus to help to understand the limitations and improvement possibilities of the MH-based EIS measurement setup.