SELF-DRIVEN MICROFLUIDIC CHAMBERS FOR A PROTEIN MICROARRAY CELL-BINDING ASSAY

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Protein microarray technology has been developed for the parallel measurement of multiple interactions between protein binding partners. While these interactions are usually implemented between solid surface-bound and soluble molecules, the sample transport and characterisation technology has also been adapted for monitoring interactions between solid surface and cells in suspension. For such assays crucial steps are the controlled injection to, incubation on and washing of the cell suspension from the surface carrying the array features.

Since incubation and washing conditions were found to have significant effects on the outcome of the assay a microfluidic chamber system was developed to solve the effective filling and clearing the microarray surface by capillary forces. The microfluidic structure was fabricated by soft lithography technique in Polydimethylsiloxane (PDMS) using SU-8 epoxy based photoresist as moulding replica. Immunoglobulin subclasses were printed onto hydrogel-covered microarray slides, which were blocked afterwards. The sample chambers having height of 20µm were precisely aligned to the positions of a standard 16-pad microarray slide. Experiments with whole blood or suspensions of the monocytoid cell line U937 showed that cells were efficiently captured by the relevant microarray features. Subsequently non-adherent cells were smoothly removed from the microarray surface by washing buffer injected through the inlet port of the chamber and collected on conventional filter paper. The readout of the assay is the quantitation of bound fluorescently labeled cells by a microarray scanner.

A special cellular assay has been developed for monitoring immune cell adhesion and activation on protein microarrays consisting of antigens, antibodies and complexes thereof. Immune cells, such as monocytes and neutrophil granulocytes recognize antigen-bound antibodies – immune complexes – via immunoglobulin and complement protein receptors. These receptors trigger the binding of cells to the microarray surface.

We proved that the application of capillary driven microfluidic transport system for filling and exhaustion the sample chambers attached to microarrays can significantly improve the ease of handling and reproducibility of cellular binding assays.

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