Topic No. 10- Lab on Chip
Oral

Decoupling sensitivity and dynamic range: electrokinetically driven microfluidic impedance immunosensor with 3D antibody immobilization

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Purpose: Detection and quantification of biomolecules is a central topic in research and clinical diagnostics. However, current assays are expensive, have a long sample-to-answer time and a long hands-on time. These limitations suggest the need for a new kind of analytical device. Microfluidic chips with integrated electrodes that incorporate electrochemical impedance spectroscopy (EIS) to detect biomolecules are an attractive option for label-free detection. However, in the classical design of this sensor, the surface area of the electrode limits the number of binding sites and thus the dynamic range of the sensor. Furthermore, the size of the electrodes influences the sensitivity, generally defined as the magnitude of response to a step change in the input, since larger electrode surface results in a smaller change of impedance upon binding. The sensitivity and dynamic range of the sensor are thus coupled and inversely related to each other. That is, sensors with a high sensitivity have a small dynamic range, while sensors with a large dynamic range have a low sensitivity. The goal of this work is to develop a microfluidic impedance immunosensor that decouples the sensitivity and dynamic range by capturing the biomolecules in a 3D matrix and separating the detection electrodes from the capture sites. The sensitivity of this sensor is in theory determined by the cross sectional area of the microfluidic channel in which the biomolecules are captured, while the length of this channel determines the dynamic range.

Methods: A model based on electrical circuit theory was developed, and did demonstrate the decoupling of sensitivity and dynamic range for the proposed device concept. To validate this model and experimentally test the hypothesis of decoupling, a two-layer microfluidic device with integrated electrodes was fabricated by hot embossing cyclo-olefin polymer (COP) microfluidic chips, e-beam evaporation of gold electrodes on COP substrates and solvent assisted thermal bonding. Antibodies for bovine serum albumin (BSA) were incubated with N-hydroxysuccinimide acrylate to introduce a polymerizable vinyl group to the antibodies, and subsequently co-polymerized into a linear polyacrylamide capture gel. Prior the use, this gel was injected into the microfluidic device. Electrokinetic injection was utilized to inject discrete volumes of sample containing fluorescently labeled BSA protein into the capture gel while observing the protein capturing using a fluorescent microscope. Change in electrical impedance of the capture gel upon binding with proteins was subsequently measured in an integrated 4-electrode setup.

Results: A low-cost microfluidic device with integrated electrodes was successfully fabricated using scalable production processes. Fluorescence microscopy images confirmed that the synthesized 3D capture gel containing anti-BSA successfully captured electrokinetically injected BSA. The integrated 4-electrode configuration could distinguish between conductivities of buffer solutions with varying concentrations of BSA, and change in impedance was detected upon capturing of proteins. Bubble formation occurred on the integrated electrodes during electrophoresis, due to bipolarity of the electrodes under the applied electric field. Bubble formation was reduced by placing the sensing electrodes in side-channels, thus shielding them from the electric field during the electrokinetic injection. This work demonstrates the many aspects involved in integrating multiple technologies and serves as a first step towards creating a microfluidic immunosensing platform in which sensitivity and dynamic range are decoupled.

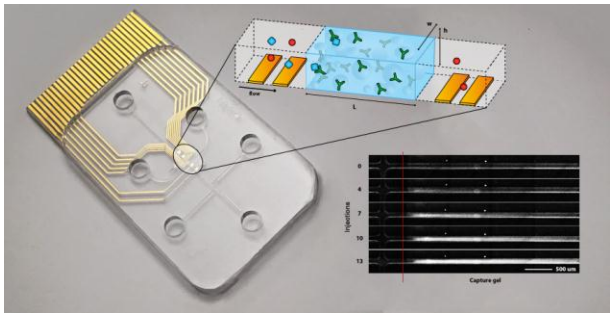

Figure 1:Picture of the fabricated 2-layer microfluidic device and a schematic illustration of the capture and detection volume inside the chip. The inset shows successful capturing of fluorescently labeled BSA in an anti-BSA 3D capture gel after repeated (4,7,10,13) electrokinetic injection of a sample containing 0.2µM BSA