Real time in-situ monitoring of in-flow electroporation using impedance cytometry

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Introduction

Electroporation has become a widely adopted method for the intracellular delivery of exogeneous cargo. Its integration with microfluidics and further miniaturization has further consolidated its usefulness, offering more control, efficiency, and viability by performing electroporation on individual cells [1].

The characteristics of the applied electric pulse define the pore formation and are therefore of critical importance for the efficient delivery of cargo while avoiding negative side-effects (e.g. cytotoxicity) [2]. One possible method to further improve electroporation is through the implementation of a feedback loop, adjusting the pulse parameters based on electroporation outcome. However, current evaluation methods, like fluorescence microscopy, do not allow for such dynamic optimization scheme.

This works aims to monitor the in-flow electroporation process in real time using the impedance cytometry principle. The combination of electroporation and impedance cytometry has been shown before using macroscopic wire electrodes and a constriction channel geometry [3]. Our device further miniaturizes this process by using embedded electrodes inside a microfluidic channel, offering more control and sensitivity, paving the way for a dynamic optimization process.

Results and Discussion

The device consists of a straight microfluidic channel casted from PDMS (⌀ = 50 µm). The bottom of the channel is sealed with a (plexi)glass substrate which is outfitted with gold electrodes. Multiple electrode sets are included along the channel length to test the influence of electrode spacing (20 – 50 µm) and electrode widths (20 – 50 µm).

The layout of the electrode follows a typical coplanar impedance cytometry setup (Figure 1), while the excitation signal is a bipolar square wave typically used for electroporation. Using the property that a square wave is build-up of sinusoidal waves, lock-in amplifiers are used to extract the frequency components of interest.

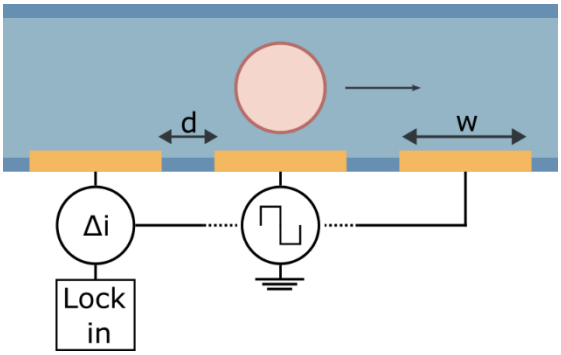

Figure 1: *Coplanar impedance cytometry setup with an electrode spacing d and electrode width w, and a square wave excitation signal. The current signals from the outer electrodes are subtracted from each other and outputted to a lock-in amplifier.*

The setup allows us perform impedance cytometry while simultaneously electroporating a cell and assessing electroporation performance in real time.

Conclusions

This work aims to develop a technique to simultaneously perform electroporation and impedance cytometry. The technique will provide the basis for a dynamic electroporation optimization scheme and enable in-situ monitoring to study the in-flow electroporation process.

References

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