Operational Frequency Range Extension by Substrate Material Effect in a Microfluidics Impedance Cytometer

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ABSTRACT

In this work, we present a new method for extending a unique operational ac frequency range for a new designed microfluidic impedance cytometer. The substrate material of the device is found to affect the high-frequency limit applied to implement impedance sensing. First, we have designed the device with spatially-designed electrode pairs. 4 coplanar 20um electrodes were used and fabricated initially on glass substrates. Then in experiments and simulations, a frequency spectrum of fabricated device impedance is employed to specify original operational frequency range, which gives further information of non-ideal capacitive effect in high frequency part. Subsequently, we have experimentally demonstrated fabricated devices' abilities to differentiate bead properties under high operational frequency where different substrate materials like Polymethyl methacrylate(PMMA) are employed. These materials with much lower dielectric constant than glass substrate will show better barrier to allow signal passing through the channel instead of the substrate.

1. INTRODUCTION

Microfluidic flow cytometry provides a low cost and label-free technique for single cell analysis. Ayliffe et al. first demonstrated single-cell impedance measurements with a coplanar electrode pair for measuring red blood cells [1]. Later on Fuller et al. further characterized human peripheral blood granulocytes in multi-frequency manner too [2]. At the time, the concept of impedance spectra was proposed by Renaud et al to demonstrate a dependence of signal variation on operational frequencies while practicing bead type classification [3]. Low frequency is used for cell size differentiation while high one deals with intracellular properties. But a discovered frequency range limits every particle-analyzing device's capability under these frequencies. Non-ideal effects such as double layer or stray capacitance are the main reasons for the restriction.

Some work has dealt with double layer impedance by utilizing gold micro electrodes [3] and platinum electroplated ones [4]. However, in high frequency area often deteriorated by stray capacitance from small scaling of electrode design, there are few methods dealing with them for either at high cost or paying little attention. Wood et al. and Hierlemann et al. continually proposed to use an inductor circuit to create a resonance frequency to make channel impedance pure resistive [5-6]. However, only one specific resonance frequency in their work is available instead of a whole spectrum are for high-frequency sensing. Also the devices increase their complexity and cost by introducing an extra inductor circuit.

So in present work, a novel coplanar electrode is spatially designed and implemented on a glass substrate. A frequency range is first simulated with correspondent signal measurement data experimentally. Then, to enhance the device ability of higher frequency cell analysis, a substrate material PMMA substitution is further utilized. Finally, experiments indeed certify the simulation results of better sensitivity with PMMA-coated device.

2. EXPERIMENT

The particle-analyzing device can be fabricated as shown in the previous work [7]. Briefly, the polydimethylsiloxane (PDMS) channel is bounded on an oxygen-plasma treated glass substrate with the planar electrodes, which are evaporated by 20 nm chromium and 50 nm gold. The electrode design is defined by four 20 um width thin-film Au/Cr electrode geometry with a pitch of 20 um as shown in Fig. 1. An electrode A will be applied an 1V ac signal of different frequencies to perform cell impedance sensing and a lock-in amplifier will be responsible of input and output signal analysis where across signal means the signal measured from electrode C. The overall experimental setup is shown in Fig. 2. 6 and 10um polystyrene beads will be under test for verification. Then experimental data from PMMA-coated device will be compared with the original one.

To accomplish substrate material test, PMMA solution mixed with 5 % anisole are spin-coated on glass substrate and ready to be photoresist-coated with identical procedures mentioned before. they are bonded together after RIE treatment [8].

3. RESULTS AND DISCUSSION

The simulated impedance spectrum is first shown in Fig. 3. The result clearly shows the double layer impedance saliently blocks the detected signal at the frequency below 1MHz and the parasitic behavior begins to intensely dominate the sensing impedance at the frequency higher than 12MHz. The mid flat region between 1MHz and 12MHz represents the only channel resistance ideal for impedance sensing. Then in Fig. 4, 6um and 10um beads are respectively injected into microchannel experimentally to cause voltage differences under multi-frequency correspondent to the frequency range. Then same experimental results have been done with PMMA-coated device as shown in Fig.5. It clearly implies an extension for high frequency limit to nearly 50MHz in 10um beads and 15MHz in 6um beads.

4. CONCLUSION

The original device with glass substrate shows an original frequency range up to 12 MHz in 6um and 15 MHz in 10um beads. By substituting the substrate with PMMA-coated glass, the results indicate a successful extension for the frequency range up to 15MHz in 6um and 50MHz in 10um beads. PMMA with relatively low dielectric constant 2.6 gives a better impervious ability of preventing signal from going down in the substrate. Glass with that of 4.2 indicates a poorer resistivity of the signal where sensing electrode receive more than just electric line from the channel but from substrate. Overall, the proposed low dielectric constant PMMA enhances the impedance cytometer ability of exploring intracellular properties under much higher frequency region. Also, it implies the necessity of employing different kinds of substrate materials with lower dielectric constant for multi-frequency spectrum analysis.

5. REFERENCES

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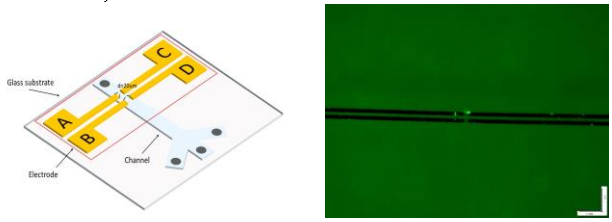

Fig. 1. (a) The overall electrode and channel design and (b) real electrode design under flueorescence with a 10um passing bead.

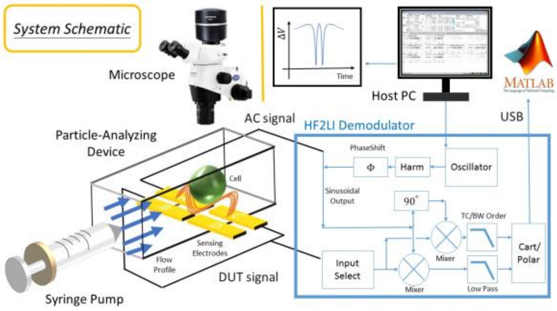

Fig. 2. The overall experimental setup.

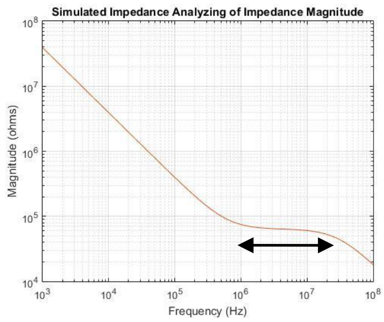

Fig. 3. Simulation of total analyzing impedance in channel from 1kHz o 100MHz without bead flow.

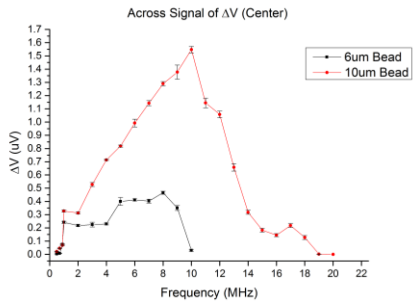

Fig. 4. Real across signal of 6,10um bead under multifrequencies.

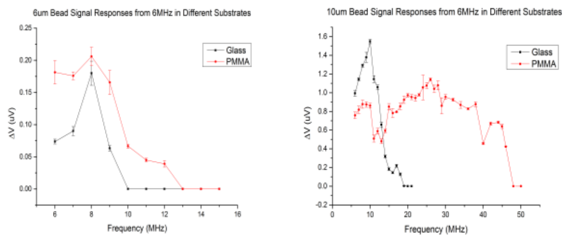

Fig. 5. (a) 6um Bead Signal Responses from 6MHz in Different Substrates and (b) 10um Bead Signal Responses.