Label-Free Detection of Gamma-Aminobutryric Acid Biomarker Using Dielectrophoresis and Absorption

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***Abstract*—Gamma-aminobutyric acid (GABA) is a potential biomarker for the detection of neurological disorders. In this work, we have investigated a method for label-free detection of GABA molecules.**

Keywords—Protein, Dielectrophoresis, Absorbance

I. Introduction

GABA (γ-aminobutryric acid) is an amino acid that acts as an inhibitory neurotransmitter in the brain and spinal cord. GABA is also produced by many orther organs, including insulin producing beta-cells in the pancreas; and stimulates beta-cell growth and inhibit pancreatic alpha cells [1]. Recent studies have demonstrated that one of the causes of the neurological disorders is due to imbalance of neurotransmitter molecules such as GABA [2]. For example, Parkinson disease patients had significantly higher levels of GABA molecules in the central nervous system than healthy individuals. Similarly, individuals suffering from Meningitis had lower GABA molecules in the central nervous system than the healthy individuals. This evidence show that GABA molecules can be used as a potential biomarker for detection and diagnosis of neurological disorders [2].

Recent biosensor development research has focused on developing devices for accurate detection of GABA molecules. For example, studies have used anti-GABA molecules attached on solid surfaces to immobilize GABA molecules. Subsequently, the electrical impedance methods have utilized to quantify the immobilized GABA molecules [1]. In addition, recently field-effect transistor based immunosensor was also developed to detect GABA molecules. In this study, anti-GABA molecules were attached on sensor surface and variation of conductance with GABA molecular concentration was used to detect GABA molecules from unknown samples [2]. These studies have resulted developing promising devices for potential use in diagnosis and screening. However, these methods require anti-GABA surfaces and complex detection methods (e.g., electrical impedance). Ideally, to be useful in screening and diagnosis, there must be a simple and low-cost device for quantifying GABA molecular levels. Our long-term goal is to develop a label –free (anti-GABA free) detection method. In this work, we have investigated a viable GABA detection method. We utilized dielectrophoretic (DEP) force of GABA molecules to concentrate them on the electrodes. We then used the optical absorbance to quantify the GABA molecules.

DEP force is a result of dielectrophoresis, which is produced when an electrically polarizable particle is placed in a non-uniform electric field [3]. By choosing the appropriate frequency, the direction of DEP force can be produced to concentrate the particle on the electrodes or away from the electrodes. DEP force has been extensively used to concentrate biomarker molecules (e.g., microRNA and protein) and cells (e.g., circulating tumor cells). In addition, DEP force of the cells has used to separate cells (e.g., cancer cells) from clinical samples [3]. We have used T-shaped interdigitated electrode (TIEs) arrays in experiments. The TIEs have been previously shown to produce large electric field gradients required for efficient capture of biomolecules by DEP force [3]. Figure 1 show an image of the device and an individual T-electrode.

II. Methods

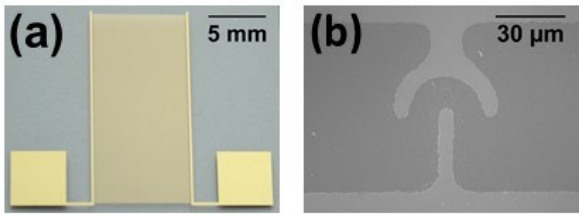

Figure 1: (a) Device and **(b)** electrodes used in GABA detection

As described earlier, in experiments, we utilized TIEs in conjunction with absorbance spectrums. Commercially available GABA molecules (Sigma

Aldrich, San Louis, MO) were used in experiments. Briefly, GABA molecules were suspended in diluted (.01x) phosphate buffered saline (PBS) and produced 100 mM solution. We then pipetted about 5 µL of the solution onto TIEs. A quartz cover slide (25.4 mmX25.4 mmX.2 mm) was placed over the GABA solution to ensure uniform distribution of sample over TIE. A potential of 10 Volts peak-to-peak was applied at a selected frequency for about 3 minutes. The TIE with sample was then immediately placed in a commercially available UV-Vis Spectrophotometer (Cary 60, Agilent Technologies, Santa Clara, CA) and recorded the absorption spectrum (200 -800 nm) for each sample. We then extracted the absorbance at 205, 270 and 280 nm. Selection of the absorption wavelength was based on the previously published studies [4]. Each experiment was repeated multiple times and calculated the average and its standard deviation. In this study, we were interested in finding the frequency dependent DEP force of GABA molecules. We conducted series of experiments and varied the frequency of the applied electric field from .1 -25 MHz and measured the absorption of each sample. Figure 2 illustrates the variation of absorption recorded at 205 nm with frequency of the applied frequency. Similar variation was shown in the absorption measured at 270 nm.

III. Results

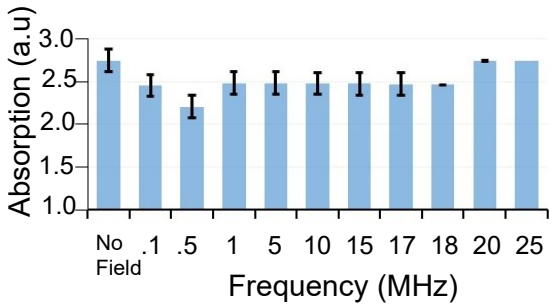

Figure 2: Variation of sample absorption with the frequency of the electric field

If there is a strong attractive DEP force produced on GABA molecules at a certain frequency, GABA molecules are concentrated near the electrodes. As a result, GABA concentration outside the electrodes is reduced and measured absorption value be less than the absorbance value recorded for the sample with no applied electric field. Moreover, comparison of absorption values with frequency provide information about the magnitude of the DEP force produced on GABA molecules. The lowest absorption was recorded at 500 kHz, and it could be due to the strongest DEP force produced on GABA at this frequency. In our previous study, we have investigated the DEP force of free and conjugated interleukin-2 (IL-2) molecules. We have found that at 200 kHz, a strong DEP force is produced on IL-2 molecules [5].

IV. Conclusion

In conclusion, we have demonstrated that integrated DEP force and absorption can be used to detect GABA molecules label free manner. Additional studies will require further fine tuning of the technology toward medical applications. Especially, detection of GABA in real patients' samples will be needed. We will further develop additional studies to detect GABA molecules in real patients' samples.

V. Acknowledgment

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