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Study of Novel Nanodevices for Cancer Biomarker Detection

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Abstract: ***A multi-dimension theoretical study of novel nanodevices for application in cancer biomarker detection is presented in this work. Nanoscale 1D bio FETs, microfluidic chips, and nanopore devices are covered here. Computer simulation of Si bio FETs has been carried out where selectivity of double-gate FET biosensors are investigated for a number of significant cancer biomarkers, namely, TOP2A, CK19, HER2, S100P, and EGFR. At first, individual selectivity of the biomarkers are analyzed in their respective biological test environments and then chi-square test along with Bayesian analysis is performed to study the possibility of parallel detection of these biomarkers in a common environment. Molecular dynamics has been used to simulate the bio-environment for nanopores. Finally, the paper presents COMSOL simulation of a practical microfluidic lab-on-a-chip (LOAC) and a device prototype development process via 3D printing technology.***

Keywords: Nanoscale 1D bio FETs, Nanobiosensor, Silicon Nanopore, Cancer Biomarker, Microfluidic chips

1. Introduction

1.1. Motivation

Recent trends in health science and biomedical research indicate a continuous need for technologies that allow for the achievement of accurate measurements and consistent results [1], [2]. Given the stochastic nature of the experiments studied in the biomedical realm, it is important to reduce errors and inconsistencies that arise from poorly prepared environments. Most chemical and biomedical tests and experiments are conducted inside laboratories with a large number of expensive equipment [2]-[4]. It is therefore very difficult to conduct environmental or medical tests in rural areas. The specimen to be tested would have to be transported to a lab that has the required equipment, which could be geographically distant. In many cases, the specimen must be preserved under specific environmental conditions while being transported and the possibility of sample deterioration or contamination always exists. These difficulties can be overcome if such experiments could be conducted using small, inexpensive, easy-to-use portable devices. The semiconductor industry allows for the integration of many advanced instrumentation and analysis circuits in a relatively small area [5].

Moreover, many biochemical experiments use large quantities of analyte samples, catalysts, and detectors manly due to limited sensitivity of available detection schemes [6]. Field-effect devices and synthetic nanopores along with other devices fabricated using semiconductor technology, can provide enhanced sensitivity in detection scheme due to the high level of purity in the design and the extremely small dimensions thereby reducing the need for large sample requirements [7]. Therefore, the idea of integrating the biochemical domain to that of microelectronics and integrated circuits seems quite appealing. Moreover, early detection of some lethal diseases is quite difficult with currently available technology due to their limitation of detecting analytes at extremely low concentration. Due to their extraordinary sensitivity, label-free detection, and potential for the integration with classical microelectronic technology, one-dimensional, nanoscale sensors have attracted a huge interest in the field during the last decade [8]. The fundamental reasons for driving to the nano materials/devices are based on the two major properties: (i) they are compatible in size with most biological entities like DNA, protein and viruses and (ii) thus they can be the ideal transducer between biomolecules and measurement instruments and that (iii) their high surface-to volume ratio (S/V) makes them sensitive to the changes in their ambient conditions.

Cancer is one of the leading causes of premature morbidity and mortality in the developed world, accounting for up to 12% of all deaths [9]. At present, cancers are often diagnosed late in the course of the disease since available diagnostic methods are not sufficiently sensitive and specific. For example, although the prognosis for lung cancer patients is poor with 5-year survival rates being less than 10%, the 5-year survival rate increases dramatically to 52% only if patients are diagnosed sufficiently early in the disease process and treated promptly by surgery [10]. Thus, the ideal goal of screening for various cancers is to detect the disease at an early phase when it is curable. The need for diagnosis of early stage cancers has prompted research into methods of screening. So far, the detection of cancers in the clinic has been relied on the detection of biomarkers (proteins or nucleotides indicating the presence of specific cancers in human body) with laboratory tests such as enzyme linked immuno sorbent assay (ELISA) [11]. Despite considerable advances in protein detection, the current ELISA-based detection methods have several drawbacks: (i) the incubation time is several hours or even up to a day, (ii) the limit of detection is around nano-Molar down to pico-Molar regime

at their best, which is still not enough for the early detection and screening of cancers, (iii) inherent auto fluorescence or optical absorption of biological samples contaminate its fluorescent or colorimetric signal, and (iv) the cost-inefficient pre-processing of samples since the tagging of antibodies with fluorescent dye is required.

To resolve the aforementioned issues, various aspects of biosensors and interaction of biosensors with different cancer biomarkers has been explored extensively. A biosensor is an analytical device that integrates a biological sensing element (e.g., an enzyme or antibody, DNA, proteins, etc.) with a physical (e.g., optical, mass, or electrochemical) transducer, whereby the active interaction between the bio-recognition molecules and the target is translated into a measurable electrical signal. A biosensor has basically two components: a receptor and a transducer [12]. The receptors are responsible for the selectivity of the sensor (e.g., enzymes, antibodies, and lipid layers). The transducer translates the physical or chemical change by recognizing the analyte and relaying it through a signal. Figure 1 describes a typical biosensor configuration that detects the target analyte with receptors. The device incorporates a bimolecular sensing element with a traditional transducer. The biological-sensing element recognizes selectively a particular biological molecule through specific adsorption, a reaction, or other chemical or physical processes, and the transducer converts the result of this recognition into a quantifiable signal. Common transduction techniques are based on optical, electrochemical, or electro-optical signals; this variety offers many opportunities to tailor biosensors for specific applications.

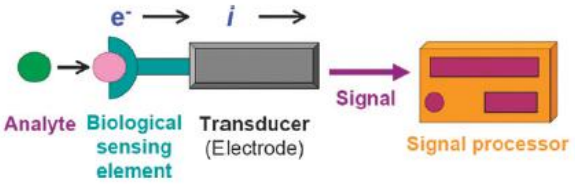

Figure 1: Conversion of the biological signal to electrical signal by a typical biosensor configuration.

1.2. Contribution

Selectivity property of the nanobiosensors, as it is one of the prominent properties for biochemical systems, was explored and a theoretical model has been proposed. Signal to noise ratio of target molecule was calculated for each of the targeted biomarkers. For example, it is desired to apply diagnosis for breast cancer; hence the target molecule is CK19. It is expected that no other proteins or enzymes are absorbed by the sensor device. If selectivity is not good then other bio-entities will be absorbed in large numbers and reliability of the detected signal will decrease. This is a huge problem for a biosensor array. So it has been a target to find optimum ways to improve the selectivity by blocking parasitic molecules without hampering the sensitivity. Biochemical system incorporates fluidic channels. So, a microfluidic system was simulated via finite element analysis modeling fluid flow as laminar. Adsorptions of biomolecules and signal detection limits were explored for six target biomarkers.

Nanopore based sensor is relatively newer technology and shows great promise. With the current surge of research in the field of personalized medicine, a synthetic Silicon Nitride (Si_3N_4) nanopore using molecular dynamics was constructed. Presence of biomarkers has been detected via their translocation through nanopore.

Finally, a prototype of a practical biosensor device has been designed and developed by 3D printing technology.

1.3. Paper Organization

The reminder of the paper is ordered as follows. Section 2 reviews the related works in the field of biosensors. Section 3 extensively describes the simulation of selectivity property of double gate bio-FET sensors and proposes a scheme for parallel detection of cancer biomarkers. Section 4 discusses the detection of a cancer biomarker through a synthetic nanopore system by molecular dynamics. In section 5, a practical microfluidic chip is simulated and the development process of the prototype of that simulated chipis presented. Section 6 concludes the paper.

2. Literature Survey

The most common ways to implement the signal transducer is to use electronic devices and monitor the changes in their conductance because of the interaction with biomolecules nearby. Ions or biomolecules (such as DNA or proteins) have their own net charge in electrolyte solution, and their electrical interaction with transducers allows us to detect them [13]. In 1970, Bergveld first suggested that the electronic pH sensing can be achieved with ion-sensitive field-effect transistors (ISFETs), promising the microscale, integrated biosensors for multiplexed detection of target molecules [14], [15]. It has been demonstrated that silicon nanowire (SiNW) biosensors [16] or carbon nanotubes (CNTs) [17] offers greatly enhanced sensitivity in the electronic biosensing compared to the conventional planar ISFETs. The powerful and most important benefits of SiNW sensors are the possibility of label-free, multiplexed, and real-time detection. The basic mechanism of nanowire sensors is based on the principle of field-effect transistors (FETs). To design biosensors, charged species binding on the surface of the SiNWsis analogous to applying a gate voltage. By observing the change of conductance, the binding of targets to probe molecules can be detected on the Si surface. CNTs also could be considered an ideal material for sensing applications since every atom in a single-walled carbon nanotube (SWNT) is located on the surface, leading to extreme sensitivity to the surrounding environment.

Introduction of target molecules causes a change in conductance across the nanowire. The conductance returns to its initial level after rinsing with buffers [18]. The current detection sensitivity of SiNW/CNT is in the range of femto-Molar [16], which are several orders of magnitude more sensitive than a conventional ELISA assay. The major drawback of electronic detection is, however, the screening effect of charged target molecules due to the presence of their counter ions in solution [19]. Avoiding the electrolyte screening effect requires additional processing costs such as dilution of buffer solutions [16], introduction of electro diffusion flow in electrolyte before detection [20].

Optical biosensors provide vast advantages over conventional analytical techniques. The selectivity of the bio-sensing element provides the opportunity for the implementation of highly specific devices to analyze in real-time, without the necessity of vast sample pretreatment or large sample volumes. Because of the unique optical properties of light-emitting semiconductor nanocrystals and Quantum Dots (QDs) have been used largely as biomolecular detection tools, which conferred advantages over traditional fluorophores such as organic dyes [21], [22]. Gold nanoparticles (GNPs) based optical sensors have been used to detect environmental pollutants including heavy metals, toxins, and other pollutants [23]. Fluorescent graphene-based materials have received increasing attention in recent years [24] due to their excellent biocompatibility, chemical inertness and low toxicity.

Magnetic particles have been also used for many years in biological assays. A wide variety of biological species–such as cells, proteins, antibodies, pathogens, toxins, and DNA–can be labeled by attaching them to superparamagnetic microbeads [25]. Naval Research Laboratory (NRL) implemented the first prototype magnetoresistive biosensor named Bead Array Counter (BARC) [25]. Another method that has achieved considerable success is based on magnetic resonance (MRI/NMR), which involves using magnetic nanoparticles as proximity sensors [26].

As a fourth-generation DNA sequencing technology, nanopore-based sequencers have the potential to sequence the entire human genome reliably and quickly for less than $1000, and possibly for even less than $100. This technology used the single-molecule techniques which allow us to further study on the interaction between DNA and protein, as well as between protein and protein. First nanopore paper was published in PNAS [27] in 1996, and from then nanopore-based detection of single molecules has appeared as one of the most effective and powerful sequencing technologies. The noteworthy advantages of nanopores include ultra-long reads (104-106 bases), label-free, low material requirement, and high throughput. Each of these advantages simplifies the experimental process greatly and can be easily used for DNA sequencing applications. The nanopore approach has been identified as a key player for the fourth-generation low-cost and rapid DNA sequencing technology. Biological nanopores have been used widely in disease diagnosis, single-molecule detection, and DNA sequencing. Recent advances in nanotechnology have facilitated the rise of solidstate nanopore sensors [28], [29]. In combination with other devices such as a FET, these synthetic nanopores can be integrated on a circuit chip, which provides the potential for portable and miniature DNA sequencing devices. More recently, to take advantages of the features of both solid-state and biological nanopores, hybrid nanopores have been proposed [30]. Nanopore DNA sequencing technology is developing rapidly. An instrument based on nanopore technology that sequences DNA at the scale of a single molecule is currently available on the market though it has a very high error rate (over 90%) [31].

Microfluidics is a multidisciplinary field of technology that links different disciplines of sciences including chemistry, physics, biochemistry, micro-technology, biotechnology, nanotechnology, and engineering [32]. As the surface-to-volume ratio is larger, microfluidic devices are more portable which is important for on-site testing. There are three types of microfluidics: (i) droplet-based; (ii) continuous-flow; and (iii) digital microfluidics. Recently, a significant demand and effort in merging biosensors into lab-on-a-chip (LOAC) technology using microfluidics systems has been demonstrated [33], [34] which add numerous benefits to the biosensor technology [35]. The integration of biosensors with microfluidic systems offers an integrated and miniaturized alternative to the traditional repetitive laboratory methods [35], [36], as it offers significant reduction in sample, reagent, energy consumption [37], [38], and waste production [39]. Due to the small size of micro-systems, a single microfluidic biosensor can perform full analysis [40] including continuous sampling, sample separation and mixing [41] and pre-concentration and treatment [42].

3. FET Nanobiosensors

3.1. Numerical Approach

For numerical analysis “Biosensor Lab” [43] was used. Target receptor size was taken as 2e-07 cm whereas parasitic molecule size was set as 1e-07cm. Target biomarker molecule concentration was set as 0.001×standard concentration (µm).

Parasitic molecule concentration was taken as 1 µm. The pH of buffer solution was set to 4. A schematic of a DGFET-based biosensor with fluid gate immersed in electrolyte and backgate to modulate the current in Si body is shown in figure 2.

The cancer biomarkers [44] taken into consideration for this part of the analysis are related to lung and breast cancer types such as Topoisomerase (DNA) II alpha (TOP2A), Keratin 19 (CK19), Erb-b2 receptor tyrosine kinase 2 (HER2/ERBB2), protein KIAA1522 (KIA1522), S100 calcium binding protein P (S100P), and Epidermal growth factor receptor (EGFR) [45]. Ck-19 is widely applied as post-operative diagnostic marker of papillary thyroid carcinoma [46]. Many studies have depicted that HER2 is also present in other several malignancies, including ovarian cancer, colorectal cancer, prostate cancer, lung cancer, and particularly, for prognosis of gastric and Gastroesophageal cancer [47]. HER2 protein overexpression, amplification, and mutation are also found in endometrial cancer, head and neck squamous cell cancer, oesophagealcancer, gall bladder and urinary tract cancer [48]. TOP2A is found in lymphoma, testis cancer etc. S100P is used as biomarker for pancreatic and colon cancer while also being a target biomarker for rare bile duct cancer [49]. KIAA1522 is being thoroughly investigated to find correlation with other cancers.

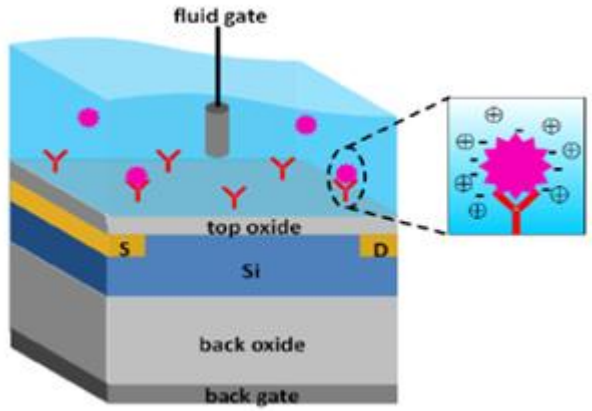

Figure 2: A schematic of DGFET-based biosensor. Fluid gate (FG) immersed in electrolyte as well as the back gate (BG) can control the current in Si body. The receptors to target molecules are immobilized on the top oxide so that the target capture modulates Si electrostatics and shifts the threshold voltage.

The first step included functionalization of the surface of the sensor with receptor molecules to the desired target. In this step, the solution holding the receptor molecules was introduced to the sensor surface. Afterwards, the receptor molecules are diffused inside the fluidic volume to finally attach to the surface of the sensor at random positions. Generally, steric hindrance due to the finite size of receptor molecules prevented overlap between adjacent attachments. This effect, coupled with the random nature of receptor attachment, caused fragmentation of the available surface area for subsequent adsorption of receptors and lead to voids of varying sizes on the sensor surface. Surface conjugation was allowed to proceed for a certain (often insufficient) incubation time, which resulted in a receptor surface density N_0 and an associated distribution of voids. These voids eventually allowed adsorption of parasitic molecules and dramatically reduced the Selectivity of label-free bio-sensing. This first step arbitrated the density of receptors N_0, as well as the distribution of open voids of size r at time t, V (r, t), on the sensor surface.

In the second step, the "receptor functionalized sensor" was introduced to a solution containing target biomolecules as well as other parasitic molecules (at time t=0). The target molecules diffused through the solution and finally reached the sensor surface. The sensor response was dictated by the net number of molecules (target or otherwise) captured on the surface as shown in Figure 3.

3.2. Mathematical Quantification for Selectivity

A basic consideration for parallel detection of multiple target molecules is defined by the "Selectivity" of the sensor technology (Figure 3) which quantifies the ability of a sensor to detect the desired target via "lock-and-key" principle in the presence of a host of almost similar molecules which is called interfering or parasitic molecules.

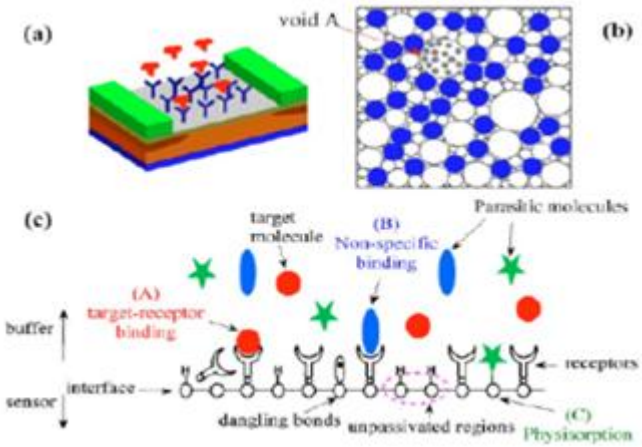

Figure 3: Biosensor system. (a) Schematic diagram of the sensor including receptors functionalized to the surface. (b) Top view of the biosensor shown in figure (a). Solid dots represent the receptor molecules. The receptors are attached with random sequential manner which introduce voids (open circles) of varying sizes on sensor surface over which parasitic adsorption can occur (illustrated as shaded dots in the void A). (c) Cross-section of a sensor system illustrating the various components that contributes towards Selectivity.

Selectivity can be quantified in "signal to noise ratio" (SNR) [50].

$$SNR \equiv \frac{S}{N} (1)$$

The SNR predicted by Eq. 1 is in addition to the noise that arises from statistical fluctuations in the density of captured molecules and ion concentration [51]. To estimate SNR, we needed to evaluate both signal and the noise components. Apart from the parameters like reaction constants and target molecule densities, the signal and noise components are entirely determined by two parameters - N_0 and N_p. N_0 completely determines the signal component, while N_p and N_0 are required for prediction of noise component.

If we consider SNR due to physiorption of parasitic molecules,

$$SNR \approx \frac{\sigma_T k_T \rho_T N_0}{\sigma_P k_P \rho_P N_P} (2)$$

Where-
σ_T = charge of target molecule
σ_P= charge of parasitic molecule
k_T = normalized reaction constant of receptor-target molecule
k_P = normalized reaction constant for physiorption of parasitic molecules on sensor surface
ρ_T= concentration of target molecule concentration
ρ_P= concentration of parasitic molecule concentration
N_0= density of target receptors
N_p=density of parasites

It was observed that label-free Selectivity of 1 ppb (parts per billion) with SNR>1 is possible with $N_0 > 2\times10^{12}$ per cm^2, an achievable receptor density. This result provided an estimate for SNR of biosensors in the presence of physisorption of parasitic molecules and suggested that label-free sensors for electrical detection of biomolecules might be viable even in the presence of parasitic molecules at a much higher

concentration provided sufficient incubation times are allowed. It turned out that incomplete surface functionalization with $N_0<2\times10^{12}$ per cm^2 would rapidly erode SNR and make the technology irrelevant for parallel detection.

3.3. Result Analysis

The previously developed mathematical parameter, Signal to Noise ratio (SNR) were calculated for the aforementioned biomarkers and SNR as a function of receptor density have been presented in Figure 4 only for CK19, as an example. It can be seen from the SNR results, an excess of 10^7 is achieved with DGFET sensor with receptor density greater than $2x10^{12}$ per cm^2, as predicted. This actually paved the way for an intuitive SNR result for a microarray of 6 sensors in a buffer solution expecting to detect all the 6 focused biomarkers and resulted in a scheme for parallel detection.

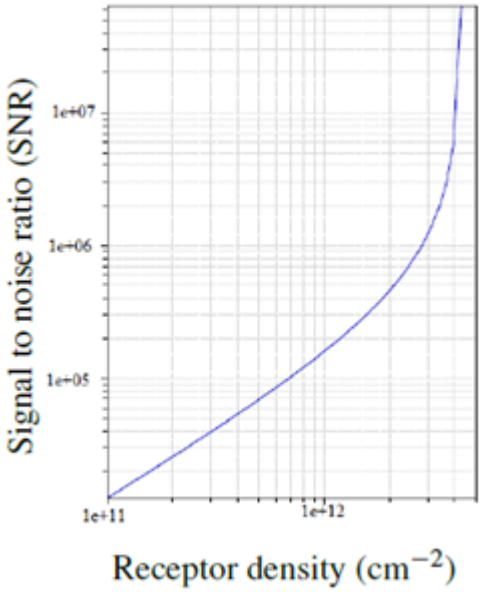

Figure 4: SNR of CK19 (charge =+31 at pH=4)in presence of biomolecule of net charge +10.

3.4. Parallel Detection Scheme

Based on the analysis from the previous section, a scheme for parallel detection method is described in this section. In preparing the biological environment for solution - a biological buffer solution was added to keep the solution environment to a fixed pH. Then the target molecule in solution was added to the biosensing system and electronic response of FET system was recorded. The buffer solutions mostly used to achieve pH conditions are shown in Table 1 [52].

For the DGFET sensor in back-gate operation fluid gate bias was taken as 1V for highest pH sensitivity [53], as presented in Figure 5. Back gate voltage V_{BG} was taken as0.2V for low power operation and more importantly for low sub-threshold swing for operation in high sensitivity region [54].

Table 1: Bio-buffer solutions for different pH

Buffer solution used	Amount (mL)	Solution pH
Na_2HPO_4+ 0.1 M citrate	10.2 + 39.8 19.3 + 30.7 25.7 + 24.3 32.1 + 17.9 43.6 + 6.4	3 4 5 6 7
0.2MHCL + 0.2M sodium barbital	17.5 + 50 2.5 + 50	8 9

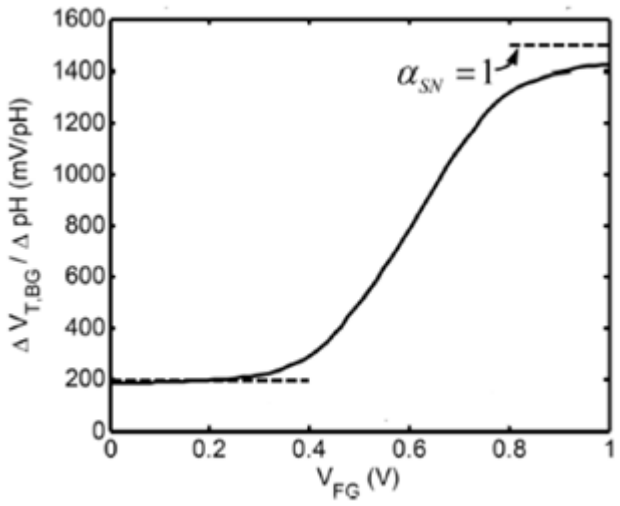

Figure 5: pH sensitivity curve of DGFET sensor with respect to fluid gate voltage (VFG); y-axis quantitatively denotes sensitivity- change of VBG with change in pH. α_{SN} is a factor depending on the bias conditions at the top and bottom surface of the device.

Finally, Form the sequence data of the biomarkers - their standard weight and standard solution molar concentration were calculated with bioinformatics tools [53], [55] and shown in Table 2.

Table 2: Resulting analyte concentrations of biomarkers

Biomarker	Weight (Kilo Dalton)	Standard solution (mM)	Isoelectric point(zero charge protein pH)
CK19	44.12	22.67	5.057
HER2	23.45	42.64	7.511
TOP2A	176.8	5.66	8.606
S100P	10.4	96.15	4.757
EGFR	134.3	7.45	6.312
KIAA1522	113.08	8.84	9.852

4. Nanopore Based Biosensors

4.1. Numerical Approach

Resistive-pulse sensors for molecular analytes [56] use a nanopore in a synthetic or biological membrane as the sensor element. This method is called stochastic sensing [56], [57] and involves mounting the membrane having the nanopore between two electrolyte solutions, applying a trans-membrane potential difference, and measuring the resulting ion current flowing through the electrolyte filled nanopore. When the analysts enter and translocate the nanopore, it rapidly blocks the ion current, ensuing a upward current

pulse. An artificial nanopore embedded in a mechanically and chemically robust synthetic membrane [58]-[60] is structured micro-lithographic methods to bore the nanopore onto a Si_3N_4 membrane.

As bio-molecules are big in sizes, the system demands a lot of computational power to be properly simulated. In this case, NAMD [61] software was used for this particular task. A crystalline membrane from its unit cell with the basis of eight nitrogen atoms and six silicon atoms was built. The unit cell information was contained in a Protein Database (PDB) file. Figure 6 shows unit cell and nanopore.

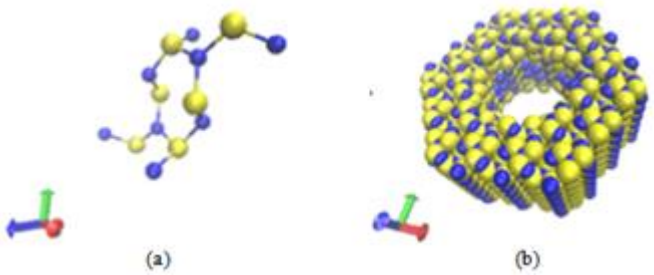

Figure 6: (a) Si_3N_4 unit cell and (b) Final nanopore structure.

To perform MD simulation with PME electrostatics, the total charge of the simulated system needed to be adjusted to zero. The charges on all of the nitrogen atoms were tuned by the equation

$$q_N = -\frac{q_i N_i}{N_N} \quad (3)$$

Where, q_i and N_i are the charge and number of each species, respectively. For this pore, the q_N adjustment is generally less than 2% times of its absolute value, and for most purposes it is negligible.

The formula for calculating dielectric constant is-

$$\kappa = 1 + \frac{\Delta p}{\varepsilon_0 EV} \quad (4)$$

Where Δp is the magnitude of the difference in the dipole moment between identical systems with and without an electric field, E is the magnitude of the applied electric field, and V is the volume of the system dielectric material [62]. Volume of the hexagonal prism was calculate the by-

$$V = \frac{3\sqrt{3}}{2} R^2 I_z \quad (5)$$

Where R is radius of the hexagon and l_z is the prismheight. Obtaining R and l_z from membrane bound file, volume was found to be 2348 $\dot{A}^3$and energy as 16.0 kcal/(mol$\dot{A}$e).

As all biological systems rely on water to function, nanopore was solvated by water to simulate the proper environment. In addition, ions, resulting from added salt, facilitated measurements of small currents in nanopore systems by substantially increasing the conductivity of the solution.

4.2. Result Analysis

Ionic current, a macroscopic quantity that gave insight into nanoscale processes. E3 Ubiquitin [63] is a biomarker for Melanoma which was translocated through the nanopore. The combined system was simulated with an applied voltage and the ionic current from the trajectory was calculated. The constraint files were generated using the parameters that reproduced the experimental dielectric constant. Temperature was raised from 0 to 295 K at constant volume. Equilibrating operation was done at constant pressure and Langevian thermostat. 20 V was applied at constant volume.

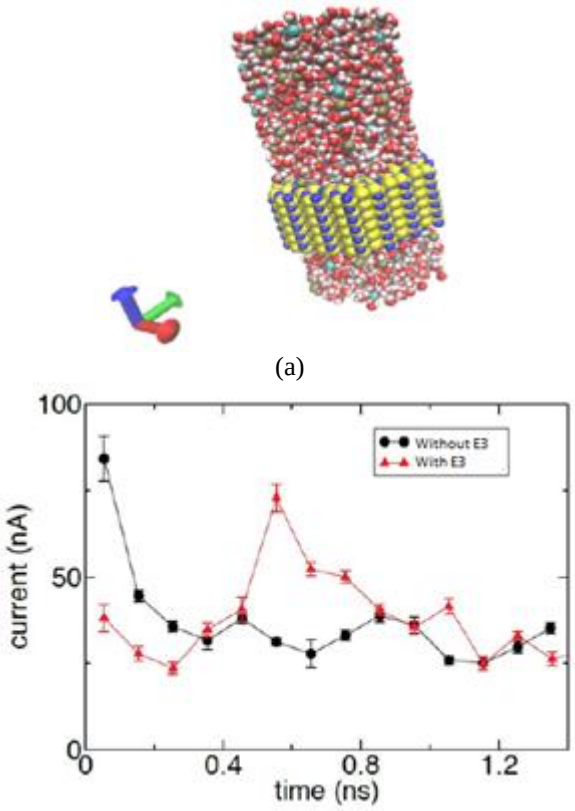

Figure 7: Translocation E3 ubiquitin, (a) combined pore+Ubiquitin system in the ionic solution environment, (b) Comparison of current between nanopore and pore+E3 system.

The current graph during Ubiquitin translocation is shown in Figure 7. From the upward deflection of current magnitude and time duration, presence of E3 Ubiquitin was detected.

Developing a Practical Biosensor

4.3. Numerical Approach

In this subsection, a hands-on micro purification chip (MCP) biosensor was simulated via COMSOL [64], as shown in Figure 8. The model is a arrangement of mass balance, analyte binding represented as reaction on the pillar surface, convection, and diffusion. Navier-Stokes equation was applied to account for pressure driven flow inside the micro-channel. The boundary conditions were concentration, c_0 at the micro-channel inlet, and initial flow velocity. At the outlet, the boundary conditions were atmospheric pressure and convective flux. All constants for the model were matched for prostate specific antigen (PSA) binding from a physiological solution.

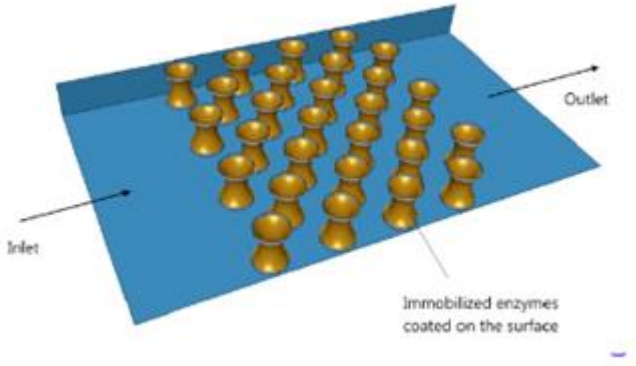

Figure 8: Micro flow cell with pillars coated with enzymes, designed to detect a specific biomolecule

4.4. Result Analysis

In this study, analyte accumulation pulse amplitude (mole/m^3) was varied, keeping inlet injection velocity constant at 2e-4 m/s with 4 pillars across each row to obtain the variation in fraction of occupied active surface sites or surface fraction P, as shown in Figure 9. The fluid flow regime was modeled as laminar flow (fluid flowed in parallel layers, with no disruption between the layers).

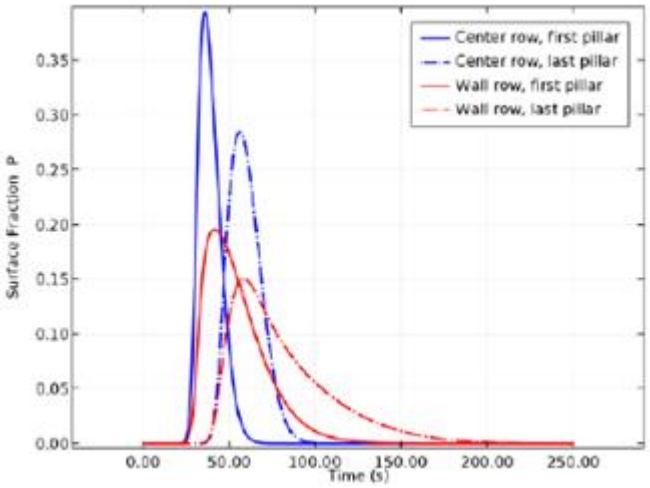

Figure 9: Surface fraction, P for analyte injection 450 mole/m^3

As seen from the Figure 10, the velocity distribution of the flow field caused pillars near the wall to reach their maximum adsorption level at a later time compared to pillars in the center of the stream. Pillars near the wall also took longer to release adsorbed analyte. The position of a pillar in a row also had an effect on the maximum adsorption level and the time at which it reached. As perceived from Figure 10, with the increment in analyte molecule injection, the occupied sites also increased, translating into an increase of target molecule capture which was quite an instinctive outcome for the device. The biosensor model presented here paved the way for fabrication of the device.

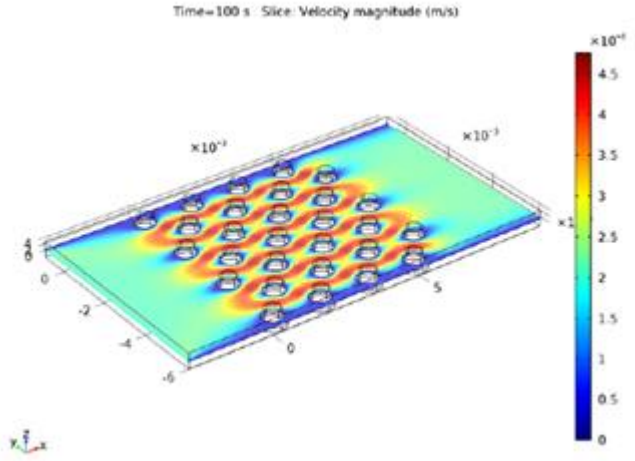

Figure 10: The velocity magnitude of the laminar flow field in the biosensor flow cell.

4.5. Fabrication of the Simulated Device

A lab-on-a-chip (LOAC) is a scaled down device that integrates one or several analyses onto a single chip which are usually done in a laboratory. The basis of the lab-on-a-chip's vision is to assimilate thousands of biochemical operations that could be done by splitting a single drop of blood collected from the patient in order to get a precise diagnosis of potential biomarker onto a single chip. Microfluidic know-hows used in lab-on-a-chip devices enable the fabrication of millions of micro-channels, each measuring micrometers, on a single chip (Figure 11). A significant demand and effort in merging biosensors into lab-on-a-chip (LOAC) technology using microfluidic systems has been demonstrated in [65], [66]. In this work, a very basic prototype was built. Our proposed device increased the selectivity based on the mass of the fluidic system. Its resolution was in the order of micrometer range. The CAD design of the device is done in "Blender" [67]. A wireframe design and a solid body design are shown Figure 12.

Figure 11: Zoomed view inside the LOAC (Lab-On-A-Chip) microfluidic chamber.

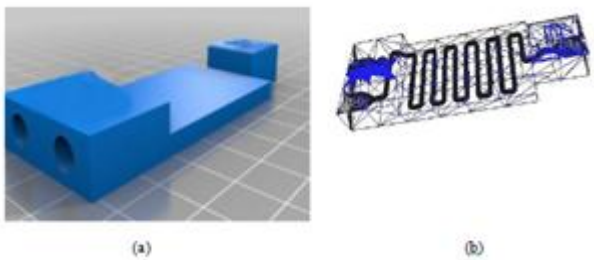

Figure 12: CAD design of the prototype LOAC, (a) Flat plate solid body, (b) Wireframe Design.

Inspired from the result of numerical analysis, a lab-on-a-chip was 3D printed, as shown in Figure 13. Resolution was 100 micron, material was taken as Transparent PLA, and DreamForge printer was used to complete the job. Though the device is just a prototype, it supported the findings of the numerical analysis of the previous sections.

Figure 13: The prototype LOAC- 3D printed.

5. Conclusion

Detailed numerical analysis has been conducted for double gate FET (DGFET) sensors with biomarkers and a mathematical model has been developed and parallel detection scheme has been proposed. Using a 4x4 array of micro enzyme rod, the absorption of biomarkers in the laminar flow region of analyte fluid has been observed. A Si_3N_4 nanopore based sensor has been developed using molecular dynamics and change in current vs. time graph for E3 ubiquitin has been observed. By taking some feature of the signal (magnitude and time duration) and comparing those with previous template data, the biomarkers can be detected. Finally, the prototype of a practical biosensor has been built which can differentiate between microfluids by using the differences of their mass.

6. Future Scope

The work can be extended to advanced research level. The methods proposed here can be modified for better performance. Some of the scopes for future work are mentioned below-

- This work can be extended to other biomarkers of cancer and a more generalized method can be developed for increasing selectivity.
- Biosensors with high K dielectric materials like HfO2, ZrO2 and TiO2 instead of SiO2 used here or even without any oxide dielectric material can be experimented with.
- Biosensors based on P type materials can be researched for application of CMOS circuits in large biosensor array.
- In COMSOL laminar flow simulation a 2D rectangular shaped array has been used. In place of this, hexagonal or other geometry can be experimented with to observe the improvement of adsorption of analyte signal.
- In nanopore based system instead of E3 Ubiquitin, other biomarkers can be used to observe their current vs time graph for their detection and a detailed generalized method can be developed.
- The prototype LOAC device can be developed further for clinical and commercial grade.

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