Ion-Sensitive Field-Effect Transistor-Based Biosensor for PSA Antigen Concentration Measurement Using Microfluidic System

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***Abstract*— Prostate cancer is one of the most common cancers in men. Prostate-Specific Antigen (PSA) is an important biomarker in the diagnosis of prostate cancer. In the present paper, an Ion-Sensitive Field-Effect Transistor (ISFET) is introduced, employing microfluidic technology to detect PSA antigens efficiently. PSA antigen is our analyte in this sensor. Due to the PSA antigen's acidity and sensor sensitivity to the hydrogen ions and pH index, absorbing the hydrogen ion by the OH receptor on the sensor surface modulates the drive current and the device's threshold voltage. The electroosmotic flow is induced inside the microchannel by applying a voltage to the electrodes on the walls of the microchannel. Consequently, turbulence in the fluid flow in the channel has occurred that effectively moves the intended analytes toward the sensor surface. The biosensor performance is investigated by the simulations carried on in COMSOL. Our simulation results indicate that the proposed structure facilitates rapid detection and measurement of PSA concentration.**

Index Terms— ISFET, Prostate-Specific Antigen, Microfluidic, Electro-kinetics, Electroosmotic, Biosensor.

I. INTRODUCTION

Nowadays, biosensors are used in various applications such as the pharmaceutical industry, chemical industry, food industry, medical diagnosis, environmental monitoring, and production of health products to identify different biological molecules. The most common use of biosensors is in medical diagnoses and laboratory sciences. Significant improvements have been reported in silicon-based biosensors, including optical and field-effect transistor-based biosensors (ISFETs) [1]. Biological analysis of humans is an important technique for health monitoring. The values of the chemical parameters like pH, pNa, glucose, potassium, and calcium, along with the physical data of activity, body temperature, and the heartbeat, can effectively indicate someone's health condition. In addition, ISFET sensors are used to analyze human body sweat and plasma [2].

Ion-Sensitive Field-Effect Transistors (ISFETs) and Extended-Gate Field-Effect Transistors (EGFETs) are the first biosensors introduced. Employing biological materials as surface receptors, they exhibit more reliable detection of biological and chemical species [3]. One of the main advantages of the FET-based biosensors is their potential ability to be miniaturized while scaling other types of sensors (such as optical and surface acoustic wave sensors) has fundamental limitations. ISFET is a potentiometric sensor in which the field effect is created by the presence of electric charges at the electrolyte-insulator interface. ISFET-based biosensors are promising candidates for biological and chemical fluid analysis applications and laboratory diagnoses [4]. In addition to the ease of miniaturization, ISFETs can work at high temperatures [5]. Like MOSFET, ISFET is a three-terminal device. Still, in ISFET, the metal gate is replaced by an ion-sensitive membrane, electrolyte solution, and reference electrode, where the gate voltage is applied [6]. Although ISFETs exhibit high accuracy and fast response [7], they suffer from weak chemical stability [8].

ISFETs can measure the concentration of cancer biomarkers and detect cancer [9]. Cancer is a global health threat causing millions of deaths every year. The statistics reveal that prostate cancer is the world's second most common cancer in men [10]. Early detection of prostate cancer is of particular importance. One of the effective methods of diagnosing and treating this disease is its biomarker diagnosis. Currently, the best biomarker employed for this detection is a prostate-specific antigen (PSA) [11]. PSA is a protein made by prostate cells. Prostatic secretions are slightly acidic, with a pH of around 6.4. PSA is secreted by both healthy cells and prostate cancer cells. The normal level of PSA in human blood is about 4 ng/ml, but in patients with prostate cancer, this amount increases. Since about 30% of men with this type of cancer have PSA levels in the range of 4.1-9.9 ng/ml, accurately measuring PSA levels is critical [12].

In the present study, an Ion-Sensitive Field-Effect Transistor (ISFET) based biosensor is simulated to measure PSA concentration. Microfluidic technology is employed to improve the efficiency of the sensor. In section 2, the ISFET theory is

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introduced, and in section 3, the microfluidic and electroosmotic flow theory is explained. The simulation approach is presented in section 4, and the simulation results are presented and discussed in sections 5 and 6. Finally, section 7 concludes the paper.

II. The ISFET theory

The ISFET operation is explained with the use of site-binding theory. To form the binding sites, the oxide layer surface is covered by hydroxyl groups (OH^-), which can be positively or negatively charged by absorption of H^+ or by losing H^+, respectively, depending on the concentration of hydrogen ions in the electrolyte. Accordingly, the insulating surface is charged with surface charge density depending on the hydrogen ion concentration (pH) of a solution [13]. The PSA concentration can also be measured by absorbing hydrogen ions on the sensor surface covered by OH^- [14]. An Ag electrode is employed as the reference electrode in sensor structure [15]. Fig.1 represents a three-dimensional scheme of the ISFET sensor in the COMSOL software framework.

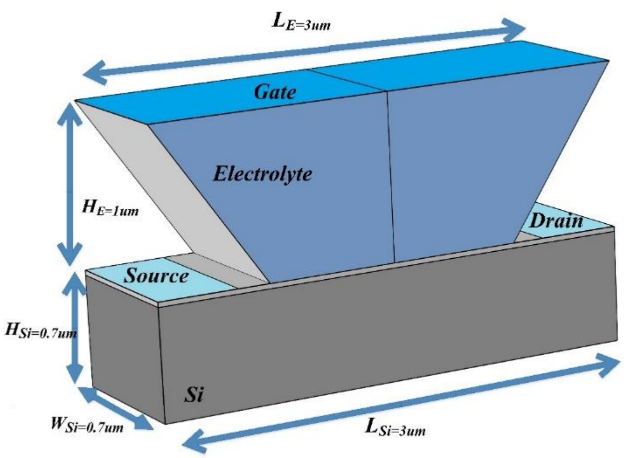

Fig1. Schematic 3D view of ISFET sensor.

When electrolyte enters between the reference electrode and the oxide layer, two potentials are induced at the reference electrode (E_{ref}) and solution-oxide interface (named interfacial potential $\psi_0 + \chi^{sol}$), where ψ_0 is the surface potential, which is a function of solution pH, and χ^{sol} is the surface dipole potential of solvent. The threshold voltage for ISFET is given by [16],

$$V_{TH(IS)} = E_{ref} - \psi_0 + \chi^{sol} - \phi_{si} - \frac{Q_{ox} + Q_{ss} + Q_B}{C_{ox}} + 2\phi_f \qquad (1)$$

where φ_{si} is the semiconductor work function, Q_{ox} is the oxide charge, Q_{ss} is the semiconductor surface charge at the insulator-semiconductor interface, Q_B is the depletion region charge, C_{ox} is the oxide capacitance, and φ_f is the Fermi potential. The potential surface changes with the absorption of hydrogen ions on the oxide layer surface, consequently affecting the device's performance. The value of surface potential is determined by [17],

$$\psi_0 = \chi^{sol} + \frac{\rho_e}{C_{stern}} \qquad (2)$$

Drain current of ISFET in the non-saturated region of operation may be expressed the same as that of MOSFET as,

$$I_{DS} = \frac{W}{L}\mu C_i \left[(V_G - V_{TH})V_{DS} - \frac{V_{DS}^2}{2} \right] \qquad (3)$$

where μ is the mobility of electrons in the inverted channel for an n-channel transistor, C_i is the capacitance per unit area of the gate, and W and L indicate the channel width and length of ISFET, respectively[18].

III. The Microfluidic and Electroosmotic Flow Theory

Research has been conducted to employ microfluidic technology in these sensors to increase the efficiency of ISFET sensors. Microfluidic technology is widely used in biosensors [19] and provides advantages such as less measure time, high sensitivity, transportability, and integration of laboratory methods in one device (lab-on-chip technology) [20]. Electrokinetics is an efficient technique to control liquids and samples in microfluidic systems. This technique is used to separate samples and create turbulence within the microchannel. By applying a driving potential to the microchannel's surface electrodes, the state of motion of liquid, which contains biomolecules, is changed. The velocity of the fluid in this electroosmotic flow is given by [21].

$$u_s = -\frac{\varepsilon_f \zeta}{U} E \qquad (4)$$

where ε_f is the dielectric constant, U is the viscosity constant, and E is the electric field. Electroosmotic flow is also used in micromixers, where fluid-induced turbulence leads to mixing operation in microchannel [22]. The fluid exhibits laminar flow without applied voltage with a small Reynolds number. In this situation, the absorption of target ions to the sensor surface is weakened; consequently, the sensor's response time and sensitivity are degraded. Inducing turbulence in the microchannel enhances the analyte absorption and improves the sensor efficiency. The Damkohler number is a dimensionless scale to study mass transfer rate [23].

$$Da = \frac{k_{on}\theta_0}{(D/h)} \qquad (5)$$

where Da is the Damkohler number, k_{on} is the reaction rate, θ_0 is the number of free receptors for antibody-antigen combination, D is the molecular diffusion coefficient, and h is the channel height. According to this equation, achieving a desirably high mass transfer rate requires a high reaction rate of the target ions and sufficient molecular diffusion.

IV. The ISFET sensor simulation

To predict the sensory operation of ISFET, we employ the COMSOL simulation framework. The sensitivity and conductivity of the sensor are evaluated in different modes. The substrate of the structure defined in the simulation environment has 3µm length and 0.7µm height, and the electrolyte cavity has 1µm height and 1.6µm length. The gate length is 3µm. The physical models employed to simulate ISFETs are the same as those used for simulating MOSFETs. These models can describe and evaluate the electric charge concentration and their interactions, concentration of different species, and diffusion coefficient of ions. The microchannel that is simulated in COMSOL, has 25µm height and 100µm length, and two electrodes with 20µm length are located under the channel 10µm apart from each other. The sensor is located under the lower wall of the microchannel. This structure is indicated in Fig. 2.

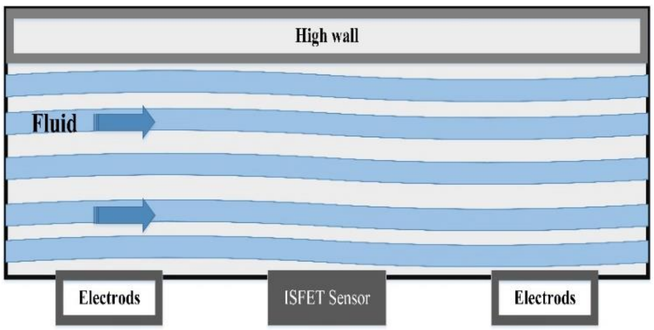

Fig 2. A schematic view of the microchannel and the locations of electrodes and ISFET sensor.

The incompressible Navier-Stokes and electrostatic physics are utilized to investigate the microchannel's fluid behavior. Navier-Stokes and continuity equations are used to analyze fluid motion in the presence of an electrical field, and electrostatic physics refers to the applied voltage to microchannel surface electrodes. The other boundaries of the channel are assumed insulated.

V. Results of ISFET Sensor Simulation

The current flows when applying a voltage to the reference electrode and increasing its value to a certain level, called threshold voltage. The threshold voltage changes according to the PSA value. Increasing PSA means decreasing pH value and the threshold voltage increases. While decreasing of PSA value leads to a decrease in the threshold voltage. Fig. 3. shows the transfer characteristics of ISFET for different values of PSA.

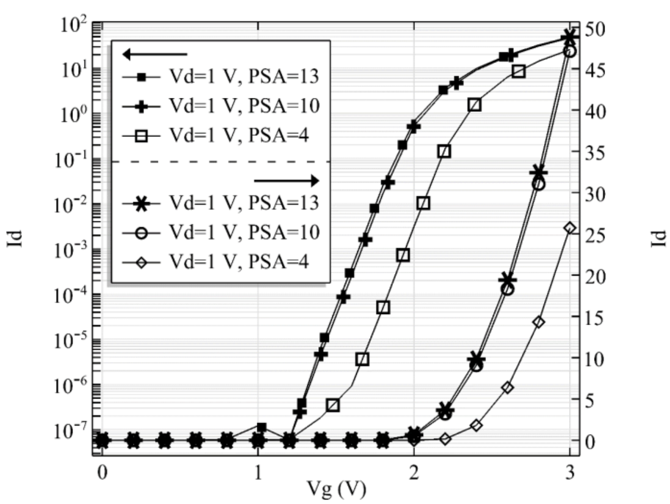

Fig 3. Transfer characteristics of ISFET for different values of PSA in logarithmic (left) and liner (right) scales.

Fig. 4 (a) shows the sensitivity curve for the ISFET PSA sensor operating in the constant current mode. A feedback circuit adjusts the gate voltage, so the drain current is maintained at a constant set point. The resulting gate voltage (the sensor's output) as a function of the PSA value (the sensor's input) is plotted in the figure. In Figure 4(b), the changes in the drain current for different PSA levels are depicted. The applied gate and drain voltages are kept constant to obtain this curve.

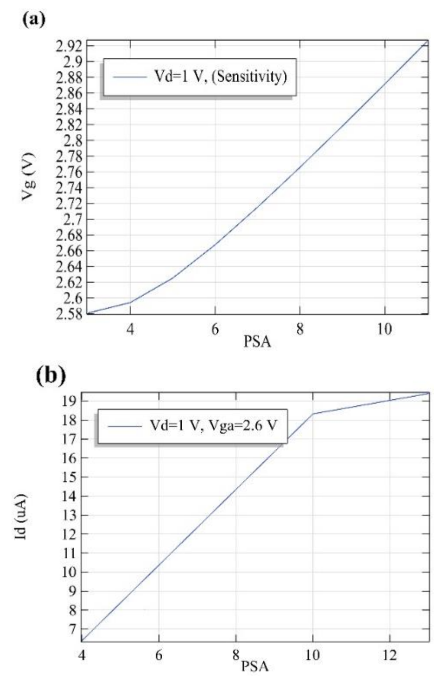

Fig. 4 (a) Sensitivity characteristics of ISFET sensor at a given drain current, and (b) variation of drain current for different PSA levels at a given bias point.

VI. Results of Microchannel Simulation

The simulation of the microchannel is carried out in two modes, with and without applying a voltage to the surface electrodes. As Fig. 5. indicates, the fluid flows without applying a voltage to the electrodes in a laminar state. It is shown in the figure that the fluid velocity is in layered distribution, and it flows faster in the microchannel center (red indicates higher speed and blue indicates lower speed). Applying an ac voltage with a frequency of 0.1Hz to the electrodes induces a turbulence state within the microchannel. The turbulence state changes over time, as is shown in Fig. 6. The higher the turbulence in the microchannel is, the more the analyte is pushed toward the sensor surface, and consequently, the adsorption time reduces.

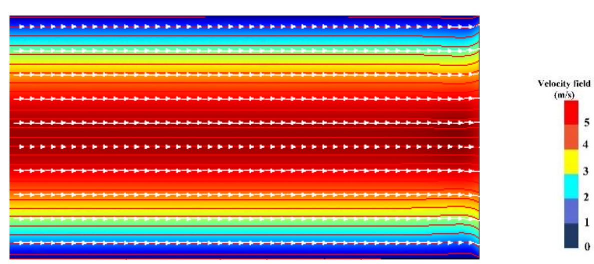

Fig 5. Fluid state in the microchannel without applying a voltage to the surface electrodes.

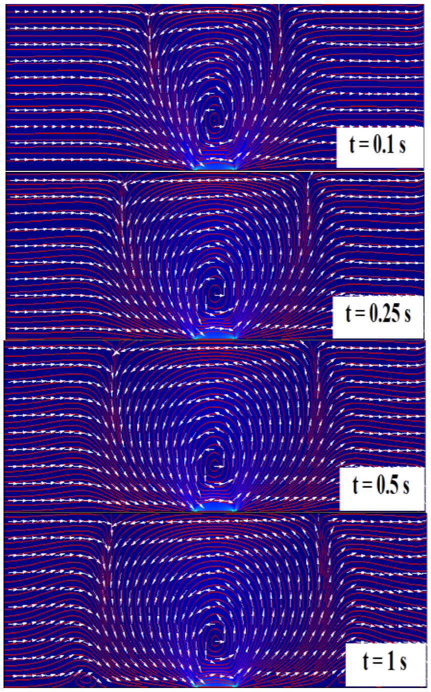

Fig 6. Changes in the turbulence state within the microchannel over time.

The required voltage to induce the most suitable turbulence depends on the microchannel dimensions, the electrode's distance from each other, fluid properties, and the initial velocity value.

VII. Conclusion

An ISFET-based biosensor for evaluation of PSA level is introduced and simulated in this work. Since the sensitivity and response time of this sensor depends on the proximity of analytes to the sensor surface, we have designed a microchannel on the sensor surface in which the electroosmotic flow is induced by applying external voltage. Adsorption of target ions on the ISFET surface modulated the channel's conductivity and changed the device's threshold voltage. Our simulation results have shown that the proposed structure facilitates rapid detection and measurement of PSA concentration.

REFERENCES

[1] Majji, Sankararao, Chandra Sekhar Dash, and Asisa Kumar Panigrahy. "Ion Sensitive Field Effect Transistor as a Bio-compatible Device: A Review." 2022 International Conference on Electronics and Renewable Systems (ICEARS). IEEE, 2022.

[2] Poghossian, Arshak, and Michael J. Schöning. "Recent progress in silicon-based biologically sensitive field-effect devices." Current Opinion in Electrochemistry 29 (2021): 100811.

[3] Ma, Xiaohao, et al. "Recent advances in ion‐sensitive field‐effect transistors for biosensing applications." Electrochemical Science Advances (2022): e2100163.

[4] Wadhera, Tanu, et al. "Recent advances and progress in development of the field-effect transistor biosensor: A review." *Journal of Electronic Materials* 48.12 (2019): 7635-7646.

[5] Mehta, Aditya, et al. "Machine Learning Techniques for Performance Enhancement of Si 3 N 4-gate ISFET pH Sensor." *2020 IEEE 17th India Council International Conference (INDICON)*. IEEE, 2020.

[6] Wang, Hui, and Naiyun Tang. "A modified TCAD simulation model for a-InGaZnO based ISFETs on GaAs substrate for pH sensing applications." *Materials Research Express* 8.9 (2021): 095901.

[7] Acar, Gizem, et al. "An ISFET Sensor-Integrated Micromixer for pH Measurements." *2020 21st International Conference on Thermal, Mechanical and Multi-Physics Simulation and Experiments in Microelectronics and Microsystems (EuroSimE)*. IEEE, 2020.

[8] Cao, Shengli, et al. "ISFET‐based sensors for (bio) chemical applications: A review." Electrochemical Science Advances: e2100207.

[9] Pindoo, Irfan Ahmad, and Sanjeet K. Sinha. "Increased sensitivity of biosensors using evolutionary algorithm for biomedical applications." *Radioelectronics and Communications Systems* 63.6 (2020): 308-318.

[10] Akbari jonous, Zahra, et al. "An electrochemical biosensor for prostate cancer biomarker detection using graphene oxide–gold nanostructures." *Engineering in Life Sciences* 19.3 (2019): 206-216.

[11] Crulhas, Bruno P., et al. "Electrochemical aptamer-based biosensor developed to monitor PSA and VEGF released by prostate cancer cells." *Analytical and bioanalytical chemistry* 409.29 (2017): 6771-6780.

[12] Arneth, Borros M. "Clinical significance of measuring prostate-specific antigen." *Laboratory Medicine* 40.8 (2009): 487-491.

[13] Abdolkader, Tarek M., et al. "ISFET pH-sensor sensitivity extraction using conventional MOSFET simulation tools." International Journal of Chemical Engineering and Applications 6.5 (2015): 346.

[14] Sant, William, et al. "On-line monitoring of urea using enzymatic field-effect transistors." Sensors and Actuators B: Chemical 160.1 (2011): 59-64.

[15] Cazalé, Arnaud, et al. "Study of field-effect transistors for the sodium ion detection using fluoropolysiloxane-based sensitive layers." Sensors and Actuators B: Chemical 177 (2013): 515-521.

[16] Sinha, Soumendu, et al. "Fabrication, characterization and electrochemical simulation of AlN-gate ISFET pH sensor." Journal of Materials Science: Materials in Electronics 30.7 (2019): 7163-7174.

[17] Dutta, Jiten Ch. "Ion sensitive field-effect transistor for applications in bioelectronic sensors: A research review." 2012 2nd National Conference on Computational Intelligence and Signal Processing (CISP). IEEE, 2012

[18] Muangsuwan, Wannaporn, et al. "Development of an immunoFET biosensor for the detection of biotinylated PCR product." Heliyon 2.10 (2016): e00188.

[19] . Madec, Morgan, et al. "Environment for modeling and simulation of biosystems, biosensors, and lab-on-chips." IEEE Transactions on Electron Devices 66.1 (2018): 34-43.

[20] Jiang, Yuting, et al. "Transient electroosmotic slip flow of fractional Oldroyd-B fluids." Microfluidics and Nanofluidics 21.1 (2017): 7.

[21] Bazant, Martin Z., and Todd M. Squires. "Induced-charge electrokinetic phenomena: theory and microfluidic applications." Physical Review Letters 92.6 (2004): 066101.

[22] Hu, Guoqing, Yali Gao, and Dongqing Li. "Modeling micropatterned antigen-antibody binding kinetics in a microfluidic chip." Biosensors and Bioelectronics 22.7 (2007): 1403-1409.