Optimization and Bioconjugation of Silicon Nanowire Biosensors for Cancer Marker Detection

Jacob Sadie

Electrical Engineering, Clemson University

NNIN REU Site: Nanotechnology Research Center, Georgia Institute of Technology, Atlanta, GA
NNIN REU Principal Investigator(s): Dr. James Meindl, Electrical Engineering, Georgia Institute of Technology
NNIN REU Mentor(s): Ramasamy Ravindran, Electrical Engineering, Georgia Institute of Technology
Contact: jake.sadie@gmail.com, james.meindl@mirc.gatech.edu, rravindran@gatech.edu

Introduction:

Silicon nanowire (SiNW) biosensors are highly sensitive nanoscale field-effect transistors. Because the channel width and height are on the nanometer scale, minor environmental alterations elicit obvious changes in the transistor's conductivity [1].With proper bioconjugation techniques, the presence of bound molecules in buffer solutions will alter the surface charge on the nanowires, changing the conductivity of the wire and indicating the sensing event [2]. Label-free methods are more cost and time-effective than labeled procedures, so we will focus our work on successful label-free procedures for ultimately detecting cancer markers [1]. Our process is a top-down fabrication technique including electron-beam lithography for the patterning of silicon nanowires. This project focused on optimizing the fabrication process by (a) adjusting the electron-beam resist application and etching procedures, (b) modifying the annealing procedures before and after electrode deposition, and (c) determining optimal conditions for pH sensing and protein bioconjugation.

Procedure:

Fabrication. The fabrication process began with electron-beam lithography (EBL) using negative resist hydrogen silsesquioxane (HSQ) spun on a silicon-on-insulator (SOI) wafer. The pattern was an array of sixteen nanowires. After lithography, we developed the resist with resist developer MF-319. Next, dry plasma etching with a standard oxide etcher (SOE) removed both silicon and HSQ to expose the silicon nanowires (SiNW) and buried oxide layer (BOX). To optimize the resist procedure, the initial thickness of HSQ was found using an automated film thickness measuring system. By varying the initial thickness (~ 30-40 nm) and etch time (14-20 sec) and then using a profilometer to measure the final thickness, the etch rates of HSQ, silicon, and the BOX could be found. Next, an oxygen anneal in a rapid thermal processor (RTP) reduced damage caused by the etching process and grew an oxide layer on the SiNWs. Our wafers underwent optical lithography (NR9-1500PY resist) and electron-beam evaporation to pattern the aluminum electrode contacts. A final forming gas anneal was performed in the RTP to ensure contact between the SiNWs and aluminum contacts. The time for this anneal was varied to determine the most effective procedure. A final passivation step exposed the tips of the aluminum electrodes and a small rectangular region around the nanowire array to prepare for wet testing.

Testing. Dry testing involved a three-point probe, where the source and drain were two electrodes on either end of the SiNW and the chuck acted as a back-gate. We used a semiconductor parameter analyzer to measure the drain current versus source drain voltage at varying gate voltages as well as the transconductance of the nanowires. To perform wet testing we attached glass wells around the exposed nanowire array using crystal bond. Then, buffer solutions at different pH levels were transferred in and out with a micropipetter. A third test was also performed, which plotted the transient drain current as buffer solutions in the well were changed.

Functionalization. The final procedure for protein conjugation required silicon nanowire functionalization with 3-aminopropyltriethoxysilane (3-APTES) [3]. We then bonded fluorophore-tagged (FITC) proteins to the 3-APTES molecules using a 2-(*N*-morpholino) ethanesulfonic acid (MES) buffer solution, a cross linker, and an activator.

Results:

Resist Optimization. We were able to characterize the etch rates of HSQ, silicon, and BOX in our given plasma process (Figure 1). The most important etch rate determined was the HSQ etch rate. Previously, the initial HSQ thickness of the process was approximately 80 nm, which meant that even after a buffered oxide etch (BOE) dip, HSQ may have remained

Spin	Average HSQ Height After EBL (nm)	Standard Deviation (nm)
1	41.6	0.9
2	33.1	1.4

Spin	Average Etch Rates (nm/s)			Standard Deviation (nm/s)		
	Si	BOX	HSQ	Si	BOX	HSQ
1	5.9	0.9	1.6	0.0	0.1	0.1
2	5.9	1.0	1.5	0.0	0.3	0.3
Cumulative	5.9	1.0	1.6	0.0	0.2	0.2

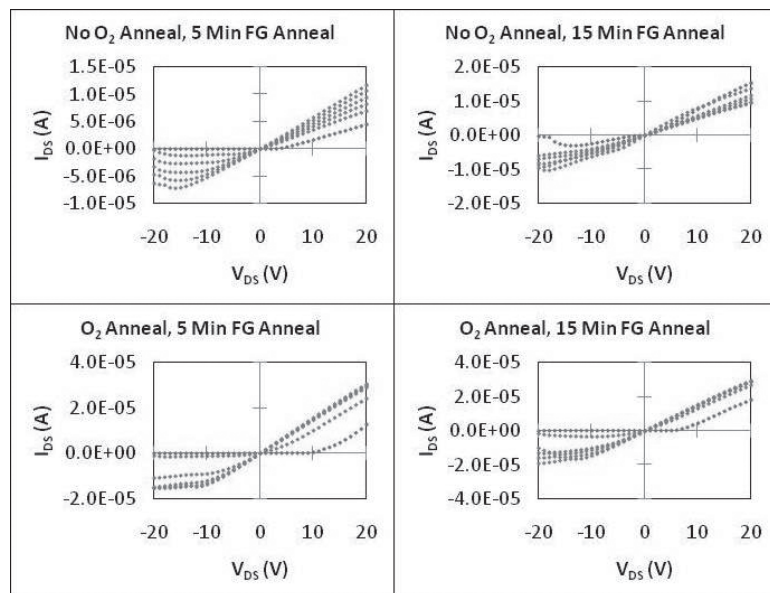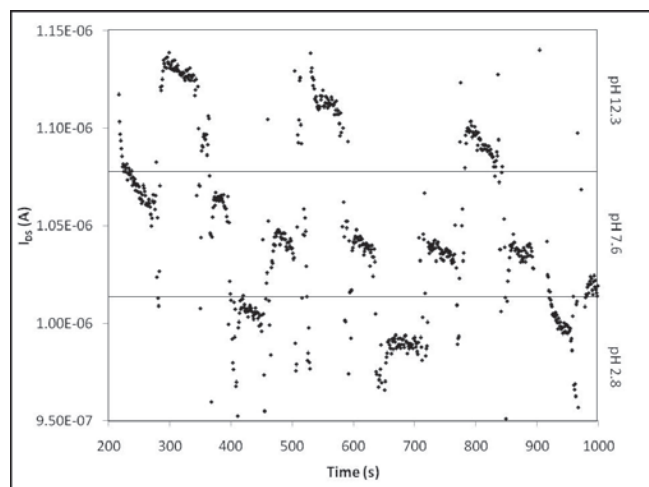*Figure 1: Silicon, buried oxide, and HSQ etch rates.*

Figure 2, above: Anneal optimization results.

Figure 3, below: Transient pH test results.

on the SiNW and prevented bioconjugation. Now, we have optimized initial HSQ thickness to be approximately 40 nm, allowing the plasma etch to nearly remove all of the HSQ and ensuring complete removal after a short dip in BOE.

Anneal Optimization. Our tests altered the two anneal procedures to determine the most effective process before and after metallization. Results seen in Figure 2 reveal that the initial oxygen anneals prior to metallization proved necessary, as the drain current range was doubled as a result of this anneal. In addition, we determined that there is no significant difference between a five or fifteen minute post-metallization anneal with FG. Finally, energy dispersive spectroscopy (EDS) indicated that thirty minute FG anneals would sometimes cause the aluminum to diffuse the length of the SiNW, effectively creating short circuits.

Potential of Hydrogen (pH) and Bioconjugation. Figure 3 indicates that our chips can sense and function over a range of pH levels. However, there was only an approximately 15% change in current over our large pH range rather than reported current changes of about 50% over similar pH ranges [3]. Finally, our bioconjugation techniques proved successful as seen in Figure 4. When performing a photobleaching process with confocal microscopy, we confirmed the binding of proteins to a silicon dioxide substrate.

Future Work:

The next steps will be to apply the bioconjugation techniques we have begun to the SiNW array in order to begin protein sensing experiments. Finally, because label-free methods are the ultimate goal, we will begin researching methods for label-free bioconjugation and testing.

References:

[1] Stern E, "Label-free sensing with semiconducting nanowires" (PhD dissertation, Yale University, 2007) pp. 39-93.

[2] Stern E, Vacic A, Reed MA, "Semiconducting Nanowire Field-Effect Transistor Biomolecular Sensors," IEEE Transactions On Electron Devices, vol. 55, pp. 3119-3130, Nov. 2008.

[3] Cui Y, Wei QQ, Park HK, Lieber CM, (2001) "Nanowire nanosensors for highly sensitive and selective detection of biological and chemical species," Science 293:1289–1292.

Figure 4: Bioconjugated and photobleached SiO_2 substrate.