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Project Director HUNT, WILLIAM

Project Unit ECE

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Contract Entity GTRC

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IMMUNOASSAY...

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Closeout Action:	Y/N	Date Submitted
Final Invoice or Copy of Final Invoice	Y	
Final Report of Inventions and/or Subcontracts	N	
Government Property Inventory and Related Certificate	N	
Classified Material Certificate	N	
Release and Assignment	Y	
Other	N	

Comments

Distribution Required:

Project Director/Principal Investigator	Y
Research Administrative Network	Y
Accounting	Y
Research Security Department	N
Reports Coordinator	Y
Research Property Team	Y
Supply Services Department/Procurement	Y
Georgia Tech Research Corporation	Y
Project File	Y

Towards Chemically Specific Surface Acoustic Wave Sensors Utilizing Immunoassay Technology

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Over this initial period of the grant from US Customs, we have designed an array of SAW delay line oscillator devices, laid out the semiconductor mask and purchased the same from Nortel. The collection of designs includes low and high frequency implementations and a variety of sensor pad areas sizes. Fabrication of the devices using this set of three masks is currently underway. Further, we have established the protocol for the attachment of the antibody film on both gold and on the bare quartz substrate. To orient the antibody in its optimal position, protein A (obtained from Sigma Chemical Co., St. Louis, MO.) is first deposited on a gold surface. This provides a highly stable coupling between the antibody and gold. Absorption of protein A onto gold requires a hydrophilic gold surface, but freshly prepared gold becomes hydrophobic when exposed to air for an appreciable period of time. For this reason, gold covered quartz samples are used immediately after gold evaporation or are treated to ensure a suitable hydrophilic surface. Protein A is dissolved in a phosphate and acetate buffer solution at its isoelectric pH of 5.5 and at a concentration of 0.5 mg/ml. The gold quartz samples are immersed in the protein A solution which is agitated with a magnetic stirrer to create a flow system that promotes uniform protein deposition. The samples remain in the solution for 30 minutes and are removed and thoroughly air dried. A volume of antibody solution sufficient to cover the sample is then placed via a pipet on the surface which is then allowed to air dry. The result is an antibody layer which covers the gold film. In the first phase of this program we will continue to perfect this procedure to ensure that we have full coverage of the gold film, that the antibodies are oriented properly and there are a

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minimum of areas uncovered by antibodies on the gold film. These measurements of film coverage are being carried out by covalently labeling the antibody molecules with fluorescein. We have already employed fluorescein isothiocyanate to successfully label theophylline antibodies to study its adherence to quartz and polymer coated quartz surfaces. In the study with protein A and antibody, both proteins are being labeled with fluorescein to test each step of the process. Fluorescein labeling will allow preliminary visual inspection of the extent to which the surface is covered with protein A and antibody. A fluorescence microplate spectrometer will provide quantitative evaluation of the number of protein A and antibody molecules bound per surface area.

The activity of the theophylline antibodies will be tested using the binding of tritiated theophylline as an independent assay. Antibody activity will be first assayed in the liquid phase with tritiated theophylline in an aqueous buffer. The radioactivity will provide a rapid independent check of the binding of target molecules to the antibody on the sensor surface. The liquid phase activity can then be compared with antigen-antibody binding in gas phase using the SAW sensor.

Binding of antigens to the SAW sensor in the gas phase will be carried out using the INEL vapor phase testing system and we will be ready to begin this phase of testing within the next two weeks.

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