Biomaterial Interfaces Division
Room: 108 - Session BI-MoA

Sensors and Fluidics for Biomedical Applications
Moderator: E. Reimhult, University of Natural Resources and Life Sciences, Austria

2:00pm **BI-MoA1 Three-dimensional Microfluidic Flow Cell Array Integrated with SPR Microscopy for Multi-channel Bioassays**, ***J.S. Shumaker-Perry***, University of Utah **INVITED**

The expanding development and implementation of biotherapeutics to treat a wide range of diseases, including autoimmune diseases and cancer, have increased the need for immunogenicity assessment of these therapies. Immunogenicity is an immune system response of a patient to a drug. Anti-drug antibodies (ADAs) produced during the immune response may cause serious adverse effects such as reducing drug efficacy, altering pharmacokinetics, causing infusion reactions including anaphylaxis and serum sickness, and neutralizing native proteins. The complexity of immunogenicity analysis presents challenges in identifying and characterizing the neutralizing activity of ADAs. We have integrated a three-dimensional microfluidic flow cell array (MFCA) with surface plasmon resonance microscopy (SPRM) for a multi-channel, array-based approach to immunogenicity assessment. The microfluidic device provides 48 separate flow channels that can be used simultaneously for biomolecule immobilization and subsequent array-based biomolecule interaction analysis. Because the biomolecules can be immobilized *in situ*, exposure to harsh environments can be avoided, a major benefit for protein immobilization. In addition, the biomolecule immobilization process can be monitored in real time by SPR microscopy. This versatile, multi-channel biomolecule interaction analysis platform is being developed for ADA assessment which will benefit from the ability to implement controls, identify optimal assay conditions, and obtain detailed data about the nature of the biomolecule interactions. Characterization of the integrated system and initial investigations related to ADA assessment will be presented.

2:40pm **BI-MoA3 Transport Properties of Proteins and Quantum Dots in Nanochannels in Multi-Gated Field-Effect-Transistor Configuration**, ***L. Tribby***, University of New Mexico, *F. Van Swol*, Sandia National Laboratories, *C.F. Ivory*, Washington State University, *S.M. Han*, University of New Mexico

The use of nanofluidic architectures as a means of concentrating and separating biomolecules, nanoparticles, and other small species of similar size scale may prove useful in developing new bioseparation and detection technologies. Recognizing this potential, a variety of nanofluidic devices have emerged that utilize enhanced electrokinetic control of fluid and molecular/particle motions at these scales. In our study, we have fabricated an array of slit-like nanochannels (100 nm w x 400 nm d x 15 mm l) in a multi-gated field-effect-transistor configuration, using interferometric lithography and conventional top-down fabrication techniques. Our main objective in developing such a dynamically controllable separation platform is to further increase our ability to rapidly concentrate and separate proteins (or nanoparticles) that have low abundance or require long separation time by conventional methods. In order to produce effective separation strategies, we have first experimentally characterized electrokinetic transport properties of proteins and nanoparticles within our device. Based on this characterization and understanding, we report a technique to form highly concentrated protein bands in our nanochannels. We will also report observable differences in electrokinetic mobility for semiconductor nanocrystals in aqueous solutions whose surface is functionalized with organic ligands to assume different charges. These results and their implications towards nanofluidic separation techniques will be further discussed.

3:00pm **BI-MoA4 High Sensitivity Recessed AlGaN/GaN HFET Protein Sensors**, ***X. Wen***, *H. Kim, P. Casal, S. Lee, W. Lu*, The Ohio State University

We have demonstrated the improvement of sensitivity by biasing the AlGaN/GaN HFET biosensors into the subthreshold regime. To bias the channel into the subthreshold regime, V_g=~-4 V was. The gate voltage causes ions in the physiological buffer solution to move and result in measurement noise. To avoid side effects from a high gate voltage, we adopted the gate recess process to shift the subthreshold gate voltage to zero/near zero volt and retain high sensitivity.

The AlGaN/GaN heterostructure used in this study has a 23 nm thick undoped AlGaN barrier. The recession of AlGaN barrier was conducted with an Oxford Plasmalab 100 system. A two-step recession process was used. The first step uses BCl_3 to etch the AlGaN barrier and the second step uses a combination of $Cl_2/N_2/O_2$ to passivate the etched surface. To achieve zero/near-zero subthreshold gate voltage, we recessed devices with the first step duration to be 50, 55, 60, 65, and 70 sec respectively. We also fabricated diodes with the Schottky area recessed with the same conditions to check the material property changes. The measured C-V characteristics show that the threshold voltage shifts along the positive direction with the increase of etching time. With 70 sec etching time, the threshold voltage of Ni diodes achieve subthreshold regime at Va=0 V. Extracted from C-V curves, the etched depths are 12.0, 11.6, 11.2, 9.7, and 8.8 nm respectively. AFM measurements of both recessed and original AlGaN surfaces show that the surface smoothness is improved after recession processes.

The fabricated AlGaN/GaN HFET biosensors have a recessed gate of 100 um (L) × 2 mm (W). To detect streptavidin, the AlGaN surface was modified by silanization and biotinylation. The measured Id-Vg curves of AlGaN/GaN HFETs with (a) recession time of 65 sec, and without any recession. There are two major differences: 1) the threshold voltage of recessed device shifted to around 0 V; and 2) the off-state current of recessed device is 1-2 orders lower. The threshold voltage promises high sensitivity when the gate is floating or biased at a very low voltage. The decrease of the off-state current indicates that signal-to-noise ratio is improved. The difference between the threshold voltage of diodes and HFETs mainly because of 1) the diode Schottky metal is Ni and the HFET gate is PBS and 2) the HFET is treated with oxygen plasma for surface modifications. The detection of 25 pg/ml (473 fM) SA solution in 1X PBS shows that the drain current is decreased by 22.7% because SA carry negative charges in PBS solution. The sensitivity is increased by one order compared to our previously published results with non-recessed devices.

3:40pm **BI-MoA6 AlGaN/GaN HFETs for DNA Sensing: Charge Layer Distance Dependence**, ***Y. Wang***, *W. Lu*, The Ohio State University

AlGaN/GaN heterojunction features two-dimensional electron gas which is highly sensitive to proximal charges and excellent stability against chemicals. These unique advantages imply the potential of AlGaN/GaN based field effect transistor (FET) in facilitating various biological and chemical studies. We have previously reported the detection of hybridization between probe DNAs and fully complementary target DNAs with AlGaN/GaN HFET biosensors. To further improve the sensitivity, comprehensive understanding of the working principle is necessary. In this work, we focus on a systematical investigation on the effects of the distance between the charged layer and the sensing surface, i.e., the distance from target DNA biomocules to the AlGaN surface. We immobilized a monolayer of single-strand probe DNAs modified with thiol groups on the Au-coated active gate surface of an AlGaN/GaN HFET. Different target ssDNA molecules were designed so that distance between the hybridization sites and the AlGaN surface varied. Experimental results show an explicit relationship between the detection sensitivity and the charge layer distance.

Six types of synthesized 25-mer oligonucletides (Allele) solutions were prepared, including a probe DNA, a fully complementary target DNA, a 21-bp complementary DNA, a 13-bp complementary DNA, a 9-bp complementary DNA and a mismatched DNA. The amount of charges is assumed identical for DNAs with a constant length. The distance of the charge layer can be well controlled by varying the hybridization sites of target DNAs to the probe DNAs. For example, the 21-bp complementary DNA is 4-bp further away from the surface than the fully complementary DNA.

We compared the I_{DS}-V_{DS} characteristics of devices before and after hybridization. A decrease of drain current was noticed, revealing the binding of negatively charged DNAs. More importantly, the decreases in I_{DS}-V_{DS} demonstrated a clear dependence of charge layer distance by an observation of the current change ratio, defined by $\Delta I_{DS} / I_{DS}$, (gate bias: -1 V): 16.1% for fully complementary target DNA (0.1 μ mol), 14.9% for 21-bp complementary DNA, 10.9% for 13-bp complementary DNA, 6.89 % for 9-bp complementary DNA, and 0.75% for mismatch DNA, which was comparably insignificant. The current change decreases with the distance between the target DNA and sensing surface, although the amount of charges for the target DNA is constant. These results demonstrate that the detection sensitivity is significantly dependent on the charge layer distance. Theoretical analysis for varying charge layer distances of target DNAs will also be studied and compared with the experimental results.

4:00pm **BI-MoA7 Effect of Analyte Flow Rate on the Sensitivity of Microcantilever Biosensors**, ***R.P. Desikan***, *C.W. Van Neste, T.G. Thundat*, University of Alberta, Canada

The past decade has witnessed the use of microcantilevers as mechanical transducers of molecular recognition and for the development of miniaturized and sensitive biochip platforms. Microcantilever based biosensors can be based on either mass adsorption or surface stress variation. The potential to operate a microcantilever sensor in liquid medium with extreme sensitivity makes it an ideal choice for the development of biological sensors. Selective detection is obtained by immobilizing receptor molecules on one side of the cantilever. Microcantilever based biological sensors predominantly operate in a liquid environment; this is done in order to retain the functionality of the biomolecules immobilized on the surface. Typically reference cantilevers serve to observe and analyze the effect of non specific interactions and fluid flow rate from specific biomolecular interactions. We have additionally observed that the interaction of analytes on the functionalized surface of the cantilever is influenced by the varying the flow rate of the solution used. The surface stress observed due to the adsorption of analyte molecules on to the receptors on cantilever surface in static condition is much higher compared to a dynamic condition where analyte molecules are allowed to flow across the cantilever surface with the help of a flow control system. Here we address the effect of flow rate on the biomolecular adsorption kinetics of the system and how it affects the sensitivity of Microcantilever based biological sensors.

4:20pm **BI-MoA8 Fabrication of Nanowire FETs for pH Sensing**, ***C. D'Emic***, *S. Zafar, A. Afzali, B. Fletcher, T. Ning, M.A. Guillorn, D.-G. Park*, IBM T.J. Watson Research Center

Sensors for measuring pH are very important for understanding reactions of biological species such as proteins, enzymes and cells. While traditional sensors based upon such techniques as infrared spectroscopy, fluorescence and others have low sensitivity and slow response time, more recent nanowire field effect transistor sensors offer improved sensitivity and response time due to smaller size and increased surface areas. [1, 2]

We have fabricated nanowire FET sensors using conventional CMOS semiconductor processes. The nanowires were patterned by electron beam lithography and reactively ion etched into 30 nm thick silicon on insulator (SOI) substrates. The gate sensing surface is comprised of a hafnium oxide/ silicon dioxide stack covering the nanowire, while the source/drain regions are comprised of boron activated SOI with nickel-platinum silicide contacts. The resulting 16 nm wide nanowire devices show high sensitivity for pH measurements. The FET drain current increases by a factor of eight per unit change in pH, while the subthreshold slope is ~ 77 mV/decade. The sensors operate at a reduced sensing voltage of 0.5 V, making them promising candidates for low power, bio-medical applications.

[1] E. Stern, J. Klemic, et al., Nature, 445 (2007) 519

[2] S. Zafar, C. D'Emic, et al, to be published

4:40pm **BI-MoA9 La^{3+} doped TiO_2 Nano-engineered Platforms for Biosensor**, ***R.R. Pandey***, Centre for Cellular and Molecular Biology, India, *K.K. Saini,* National Physical Laboratory, India, *M. Dhayal*, Centre for Cellular and Molecular Biology, India

The use of nanotechnology tools has opened new opportunities to explore analytical applications of the nano-engineered materials which attracted great attention due to their unique morphology, extraordinary physical and chemical properties towards development of biosensors to facilitate the improvement of the selectivity and sensitivity of the current methods. In this work, the importance of semiconducting La^{3+} doped TiO_2 a metal-oxide-based nanostructure platform is highlighted for biosensors platforms. La^{3+} doped TiO_2 nanostructure having nanometer-scale inner-core cavity which were exposed to the outer surface with different oxidation states having possibilities for redox-activity can make them attractive for sensing uses. Therefore, the use of La^{3+} doped TiO_2 for the development of electrochemical sensors will be discussed. These platforms has been characterized by XRD, XPS, FTIR, SEM, cyclic voltametry to determine structure, surface chemistry and electron transfer characteristics for biosensor applications. Cholesterol oxidase immobilized onto La^{3+} doped TiO_2-based nanostructured surfaces exhibited a pair of well-defined and quasireversible voltammetric peaks in CV measurements. We will also discuss the potential prospect of these surfaces as low cost stable platforms for biomedical diagnosis.

5:00pm **BI-MoA10 Spray Deposition of Functional Antibody Films**, ***J. Figueroa***, *S. Magana, D. Gomez, D.V. Lim, R. Schlaf*, University of South Florida

Antibody films for the use in biosensors and assays are usually deposited via wet-chemical attachment methods. The presented experiments demonstrate that pneumatic spray deposition of antibody thin films from aqueous solution yields films of similar sensitivity and durability without special surface treatments and attachment chemistries. The experiments were performed using a commercially available low flow nebulizer in combination with a syringe pump and a substrate rotation stage to homogenize the coating.

In the experiments E. coli O157:H7 antibody was deposited on cleaned microscopy glass slides without any other pre-treatment. Standard wet-chemically prepared silanized glass slides using the avidin-biotin attachment scheme were also prepared for direct comparison of sensitivity and longevity of the spray based substrates. After incubation with GFP-labeled E. coli O157:H7 cells (ATCC 35150) ranging from 10^4-10^6 CFU/ml the slides were rinsed and AF647-labeled detector antibody was added and incubated. After rinsing and drying the slides were interrogated with a 635 nm laser and visualized using a CCD camera. Slides were also visualized by epifluorescence microscopy to examine antibody patterns and determine E. coli capture efficiencies.

The results of the experiments demonstrate that there is little difference between spray and standard protocol wet-chemically prepared substrates. This indicates that antibody films can be prepared via physisorption without complex attachment chemistries, and that antibodies can directly attach to glass slides, while retaining their functionality.

5:20pm **BI-MoA11 Microfluidic Extraction of Human Chromosomal DNA from Single Cells**, ***J. Topolancik***, *H.C. Tian, C.B. Wallin, D.R. Latulippe, J.J. Benítez, B.R. Cipriany, P.J. Murphy, P.D. Soloway, H.G. Craighead*, Cornell University

Genome-wide analysis of single cells is important in life science research and modern medicine in applications ranging from cancer diagnosis to understanding tissue development. Microfluidic devices have been explored as a promising platform for single cell studies, providing superior handling of minute sample and reagent volumes in engineered microstructures. Isolation of nucleic acids from biological samples is an essential step of every type of genetic analysis. While numerous extraction methods have been explored, it remains rather challenging to isolate and analyze genomic DNA from small cell populations and individual cells. Traditional microfluidic devices utilize solid phase extraction (SPE), a method based on binding of DNA to chemically functionalized solid phase matrices for separation of nucleic acids from cell lysates. The binding affinity is sensitive to factors such as pH, temperature, and buffer composition which must be controlled, often dynamically, to minimize DNA losses. Even when the extraction process is optimized, it is difficult to ensure that all of the DNA fragments are adsorbed on the solid phase matrix and that the whole genome is represented in the purified extracts. An appreciable fraction of genomic DNA is often lost during the purification process when the cell debris is washed away. Additional DNA losses can be caused by incomplete elution. State-of-the-art microfluidic devices for DNA separation from cell lysates exhibit rather modest extraction efficiencies of 60-85%. This is sufficient for genetic analysis of cell populations because multiple copies of every gene are present in the extract, which statistically guarantees complete genome coverage, but such losses are hardly acceptable when single-copy genes in a single cell need to be investigated. This work describes a valveless two-port microfluidic device for highly-efficient isolation and fluorescent analysis of DNA contents of single cells. Long strands of human chromosomal DNA released from the cell by chemical lysis loop around PDMS micropillars and are physically retained while the remaining cellular contents are washed away under hydrodynamic flow. DNA fragmentation is minimized by operating at low flow rates. Hydrodynamic entrapment of DNA in non-functionalized obstacle arrays allows separation of very large genomic DNA from cell debris and components such as proteins and membrane fragments as well as from much smaller mitochondrial DNA and RNA. The purified DNA was subsequently released from the device by enzymatic fragmentation with restriction endonucleases under continuous flow and collected for fragment-size analysis and evaluation of the extraction efficiency. Fluorospectrometric measurements indicate that the microdevice extracts >95% of genomic DNA, which outperforms all alternative microchip-based extraction methods.

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