

# Wednesday Morning, November 11, 2009

## Biomaterial Interfaces

Room: K - Session BI+AS+BM+MS-WeM

### Array-Based Sensors and Diagnostics: Grand Challenges

**Moderator:** D.W. Grainger, University of Utah, J. Shumaker-Parry, University of Utah

8:00am **BI+AS+BM+MS-WeM1 Design of Antibody Array-Based Sensors for Disease Proteomics: Grand Challenges, C. Wingren, Lund University, Sweden** **INVITED**

Antibody-based microarray is a new proteomic methodology setting a novel standard for analysing complex, non-fractionated proteomes. The first generation of antibody micro- and nanoarrays has already demonstrated its potential for generating detailed protein expression profiles, or protein maps, of human body fluids in health and disease, paving the way for new discoveries within the field of disease proteomics. The process of designing highly miniaturized, high-density and high-performing antibody array set-ups have, however, proven to be challenging. In this presentation, the key technological challenges that must be resolved in a cross-disciplinary manner before true global proteome analysis can be performed using antibody array-based sensors will be presented and discussed.

In this context, we have successfully designed a set of state-of-the-art recombinant antibody array technology platforms for high-throughput proteomics. In more detail, we use human recombinant single-chain Fv (scFv) antibody fragments, microarray adapted by molecular design as probes, displaying an outstanding on-chip functionality and stability. Uniquely, the platforms allows us to target both water-soluble as well as membrane proteins in a highly multiplexed and sensitive (pM to fM range) manner in complete, i.e. non-fractionated, directly labeled complex proteomes. Platforms compatible with a wide range of proteomes, including serum, plasma, urine, cell lysates, tissue extracts, intact cells etc, have been successfully designed. In addition, the first steps towards implementing label-free sensing (MS, MS-MS and SPRi) as well as designing self-addressable microarrays and miniaturized attovial-based nanoarrays as well as planar nanoarrays have been taken, clearly expanding the repertoire of technology platforms. The applicability of the platform(s) for differential high-content screening of clinical samples has been validated in a set of key applications within the field of oncoproteomics, autoimmunity, inflammatory diseases and allergy. The optimized antibody microarray technology platforms, as well as data from the screening analysis will be presented in context of the grand challenges the field experiences.

8:40am **BI+AS+BM+MS-WeM3 Development, Validation and Application of Q-Plex Array Technology, M. Groll, Quansys Biosciences** **INVITED**

The Quansys Q-Plex (multiplex ELISA) Array is a fully quantitative ELISA-based test where up to 25 distinct capture antibodies have been absorbed to each well of a 96-well plate in a defined array. This array is composed of 20 nanoliter spots with 350µm diameters and a pitch of 650µm between spots. Each spot represents a different distinct capture antibody population.

Using less than 30 µl of sample, up to 84 different samples can be assayed for all 25 unique analytes in less than 2.5 hours. Sensitivity is system dependent and typically ranges between 30 pg/ml to less than 1 pg/ml. All of the antibodies used in the Q-Plex arrays have been subject to a rigorous and comprehensive cross reactivity protocol and verified to be non-cross reactive with any other system on the array. Detection of this array is performed using the Quansys Q-View Imaging System. The image is then auto-processed using Quansys Q-View Software and concentrations for each analyte are output for the sample.

9:20am **BI+AS+BM+MS-WeM5 Drop on Demand Ink Jet Methods for Development and Manufacturing of Array Based Sensors and Diagnostics, T.C. Tisone, A.V. Lemmo, BioDot Inc.**

The development and manufacturing of array based formats requires the transfer of biomarker reagents to a carrier substrate which forms the basis of a sensor for executing a multiplexed assay for research and diagnostics applications. The typical volume range for these types of assays is in the range of 100 pL up to 1000 nL: which lies in the range of commercial drop on demand piezoelectric and solenoid drop on demand dispensers. This presentation will discuss aspects of the physics and chemistry of successful applications of drop on demand methods to provide quantitative and high throughput reagent transfer to sensor substrates suitable for both Development and Manufacturing. Issues of drop formation, drop/substrate

interactions and reagent/substrate interactions will be discussed. The agenda is to understand what role dispensing plays in the assay function.

10:40am **BI+AS+BM+MS-WeM9 New Molecular Strategies to Suppress Noise and Amplify Signal in Protein and DNA Microarrays, A. Chilkoti, Duke University** **INVITED**

This talk will highlight recent work from my laboratory that addresses new interfacial technologies to suppress noise (N) and amplify signal (S) leading to heterogeneous assays with extraordinarily high S/N. In the first demonstration, I will focus on the adventitious adsorption of proteins as the primary factor that controls the limit-of-detection (LOD) of protein microarrays and limits the measurement of analytes from complex mixtures such as serum or blood. I will show data on a new protein microarray assay where background adsorption is effectively eliminated through the use of a protein-resistant –nonfouling– polymer brush. These “zero background” protein microarrays were successfully used to quantify protein analytes in serum with femtomolar LOD and a dynamic range of six orders of magnitude of analyte concentration. These LODs are 100-fold lower when compared to the same protein microarrays spotted on a conventional polymer substrate that displays high binding capacity but significant adventitious protein adsorption. This study also provided the first demonstration of the interrogation of an analyte directly from undiluted, whole blood by a protein microarray with a LOD of ~15 fM. Next, I will summarize recent work in my laboratory on the development of a new isothermal fluorescence signal amplification and detection scheme that exploits the ability of terminal deoxynucleotidyl transferase (TdTase) to add up to 100 fluorescent nucleotides to the end of a short DNA tag with an exposed 3'-OH. I will show how DNA microarrays that are printed on the nonfouling polymer brush exhibit low background signal, yet allow on-chip fluorescence signal amplification, leading to DNA microarrays that exhibit a sub-picomolar LOD, which appears to be the lowest LOD reported for DNA microarrays, to date.

11:20am **BI+AS+BM+MS-WeM11 SwitchDNA Biosensors for the Label-Free Detection and Sizing of Protein Targets on a Chip, U. Rant, W. Kaiser, J. Knezevic, E. Pringsheim, M. Maruyama, P. Hampel, Technische Universitaet Munich, Germany, K. Arinaga, Fujitsu Laboratories Ltd., Japan, G. Abstreiter, Technische Universitaet Munich, Germany**

We introduce a chip-compatible scheme for the label-free detection of proteins in real-time that is based on the electrically driven conformation-switching of DNA oligonucleotides on metal surfaces. The switching behavior is a sensitive indicator for the specific recognition of IgG antibodies and antibody-fragments, which can be detected in quantities of less than 1 amol on the sensor surface. Moreover, we show how the dynamics of the induced molecular motion can be monitored by measuring the high-frequency switching response as well as by time-resolved fluorescence measurements. When proteins bind to the layer, the increase in hydrodynamic drag slows the switching dynamics, which allows us to determine the size of the captured proteins. We demonstrate the identification of different antibody fragments by means of their kinetic fingerprint. The switchDNA method represents a generic approach to simultaneously detect and size target molecules using a single analytical platform.

11:40am **BI+AS+BM+MS-WeM12 Nanomechanical Readout of DNA Microarrays, S. Husale, Rowland Institute at Harvard University, H.H.J. Persson, Stanford University, O. Sahin, Rowland Institute at Harvard University**

DNA microarrays have enabled high throughput analysis of gene-expression and genotyping. However, they still suffer from limited dynamic range and rely heavily on enzymatic manipulations and amplification to create detectable signals. Here we present application of a novel nanomechanical detection method to microarray analysis that may circumvent these disadvantages. It is based upon a modified atomic force microscope (AFM) that can map mechanical properties of surfaces at high speed and spatial resolution. Mechanical measurements can reliably discriminate single and double stranded DNA on a surface. Automated image analysis reveals hybridized molecules with single molecule precision, thus providing a digital measure of hybridization. This method can detect a broad range of target concentrations with a limit of detection in the low attomolar concentration range without any labeling, enzymatic manipulations, and amplification. We demonstrate the performance of this technique by measuring differential expressions of miRNAs in tumor samples, which has been shown to help discriminate tissue origins of metastatic tumors.

# Thursday Afternoon, November 12, 2009

## BioMEMS Focus Topic

Room: A8 - Session BM+MN+MS+TF+BI-ThA

## Advances in Microfluidics for BioMEMS

Moderator: G.W. Rubloff, University of Maryland

2:00pm **BM+MN+MS+TF+BI-ThA1** **Advances towards Programmable Matter**, *D. Erickson*, Cornell University **INVITED**

A dichotomy exists between the bottom-up self-assembly paradigm used to create regular structures at the nanoscale, and top-down approaches used to fabricate arbitrary structures serially at larger scales. The former of these enables rapid, highly parallel assembly but lacks critically important features of the latter such as the ability to arbitrarily direct the assembly location and perform error correction. We and our collaborators have recently proposed an alternative approach which combines these two based on dynamically programmable self-assembling materials, or *programmable matter*. The uniqueness of our approach is that it uses dynamically-switchable affinities between assembling components facilitating the assembly of irregular structures. In this talk I present an overview of our approach and detail some of the analytical and experimental advances towards a programmable matter system we have recently made. These include: the development of a multi-chamber microfluidic chip for improved far-field assembly, the demonstration of near-field inter-tile affinity switching using a thermorheological assembly fluid and the ability to enhance assembly in three dimensions using unique fluid-structure interactions.

2:40pm **BM+MN+MS+TF+BI-ThA3** **A Multilayered Microfluidic System with Buried Channels and Cell Compartmentalization for Engineering Heterogeneous Neural Networks**, *C. James, A. Greene, A. Schiess, G. Bachand*, Sandia National Laboratories, *M. Romero-Ortega*, University of Texas at Arlington

Current technology for engineering *in vitro* neural networks utilizes cell guidance cues that yield only temporary networks (< 1 month) as the cells rapidly diverge from their designed guidance cues during development of the culture. In addition, these engineered networks are typically comprised of homogeneous populations of neurons, thus the lack of multiple neuron types produces oversimplified networks that do not adequately represent *in vivo* networks. In addition, effective control over synaptic connections between different populations of neurons has not been demonstrated. Here, we describe a novel hybrid technology of multi-layered microfluidics with compartmentalized chambers containing multiple neuron types for engineering robust and complex neural networks with high resolution organization of synaptic connections. The device contains a first level of microfluidic channels etched 1-2 microns into the base glass substrate. These channels are fabricated with a novel process using a silicon nitride mask for hydrofluoric acid undercut etching to create buried microfluidic channels for robust containment and guidance of neurons. After the etching process, photoresist liftoff is performed to selectively adsorb poly-L-lysine (PLL) within the buried channels for improved neuron attachment and outgrowth at pre-defined locations. Polarity control of neurons is provided through a continuous set of guidance cues to promote axon development, while interrupted sets of guidance cues promote dendrite development. Current results show that axons and dendrites are positioned at predefined locations with a >65% accuracy. A second level of microfluidic channels and large (~mm) cell chambers are fabricated in polydimethylsiloxane (PDMS) from two-level SU-8 master molds. The base glass substrate and the PDMS substrate are aligned and bonded to create interconnects between channels in both substrates. These interconnects provide interaction regions for the development of synapses between growing neurites from cells in different chambers. We are currently applying this technology to engineer corticostriatal networks, an important region of the brain responsible for integrating multiple informational inputs crucial to complex decision-making in higher mammals. Specifically, we are using patch-clamp electrophysiology to track the development of synaptic memory in the form of long-term depression and potentiation (LTD/LTP) in these engineered networks.

3:00pm **BM+MN+MS+TF+BI-ThA4** **Vesicle Production on a Microfluidic Platform using pH Sensitive Block Copolymers**, *L.E. Brown*, The University of Sheffield, UK, *S.L. McArthur*, Swinburne University of Technology, Australia, *G. Battaglia, P.C. Wright*, The University of Sheffield, UK

The development of pH sensitive, biocompatible block copolymer vesicles has enabled the intracellular delivery of water soluble drugs and proteins.

Improving the encapsulation efficiency of the vesicles is now a critical parameter. Transferring the production method to a microfluidic device creates the potential to vary the encapsulation conditions and improve this efficiency. In this work, a flow focussing microfluidic device is used. The self assembly of PMPC-b-PDPA block copolymer vesicles is induced within the device by changing the pH of the flows within the microchannels. The use of pH shift eliminates the need for organic solvents currently required for glass capillary production methods. This enables the biocompatibility of the block copolymers to be maintained, an essential factor for their application as molecular delivery vehicles.

The flow focussing microfluidic device was produced through standard soft lithography techniques. A three-channel flow system is used with the copolymer in solution at pH6 in the central channel and aqueous buffered solutions flowing in the channels either side. The laminar flow conditions within the microfluidic device result in a pH gradient at the interfaces where the three channels meet and where the block copolymers self-assemble into vesicles. These vesicle formation processes have been imaged using confocal microscopy via FRET with a block copolymer containing both rhodamine and fluorescein isothiocyanate groups. Dynamic light scattering and TEM were used to confirm vesicle formation.

With 50nm to 250nm vesicles continuously being produced within the device it was then possible to investigate whether higher encapsulation efficiencies can be achieved using the microfluidic device. The protein myoglobin was introduced through the central channel along with the copolymer. Spectrophotometric analysis indicated the overall the efficiency of the encapsulation process within the device is not a significant improvement on the standard bulk methods currently used, involving sonication of the vesicle solution containing the molecule to be encapsulated. Despite this, the continuous nature of microfluidic devices, as well as the lack of organic solvents being used in the production process indicates that the development of these devices offers a viable alternative production method for polymer vesicles that may enable the increases in encapsulation efficiency to be achieved. Work is ongoing to achieve this using the same pH shift mechanism within a glass capillary microfluidic device.

3:40pm **BM+MN+MS+TF+BI-ThA6** **Integration of a Microfluidic Flow Cell Array with SPR Microscopy for In Situ Microarray Formation and Biomolecule Interaction Analysis**, *J. Liu, M. Eddings*, University of Utah, *A. Miles*, Wasatch Microfluidics, *B. Gale, J. Shumaker-Parry*, University of Utah

Analysis of biomolecule interactions based on surface plasmon resonance (SPR) microscopy provides a label-free approach to monitoring arrays of biomolecule interactions in real time. Typically the microarray sensing surface for these measurements is prepared *ex situ* and a single or few channel flow cell is used for the biomolecule interaction studies. The multiplexing nature then is derived from the microarray and the number of samples that can be run simultaneously is rather limited, diminishing the potential application for assays requiring a high-throughput approach due to a large number of samples. One example of this is the need to monitor for anti-drug antibodies from a large pool of patient samples during clinical trials of biotherapeutics. We demonstrate the capability of a multi-channel microfluidic flow cell array (MFCA) to expand the throughput capability when integrated with SPR microscopy. In addition, the MFCA provides an *in situ* approach to array fabrication that allows full characterization of the biomolecule immobilization process. We use the MFCA for delivery of sample solutions with continuous flow in 48 channels in parallel for rapid microarray creation and binding analysis while using SPR microscopy for real-time monitoring of these processes. Label-free measurement of antibody-antibody interactions demonstrates the capabilities of the integrated MFCA-SPR microscopy system and establishes the first steps of the development of a high-throughput, label-free immunogenicity assay. We demonstrate a limit of detection (LOD) of ~ 80 ng/ml for the particular antibody pair we studied. This LOD is ~6 times lower than the industry recommended immunogenicity assay detection limit. The high-throughput nature of the integrated system allows a large number of replicate experiments, including control experiments, to be performed simultaneously on the same sensor surface in a short time. The integrated system also will be applicable for more general high-throughput protein-array based analysis.

4:20pm **BM+MN+MS+TF+BI-ThA8** **Nanochannel Stretching of Nucleic Acids: Towards Epigenetic Analysis**, *D.E. Streng, S.-F. Lim, A. Karpusenka, J. Pan, J.A. Hook, R. Riehn*, NC State University

Nanochannels with a diameter of about 100nm<sup>2</sup> are a novel method for stretching DNA for genomic investigations. Such devices are implemented through standard nanolithography in fused silica. The elongation of DNA

results from an interplay of steric and entropic effects. Previous applications of nanochannel stretching included sizing, restriction mapping, and observation of transcription factor binding.

We show here that nanochannels can also be used to map the site-specific epigenetic state of DNA. In particular, we show here that the concept by nanoconfinement can be extended to chromatin, or DNA complexed to histones, and that the stretching is within the range expected from the de Gennes theory. We also demonstrate that the location-resolved cytidine methylation state of DNA can be mapped by specific fluorescent labeling. We will discuss the basic operation of these technique, and the application to artificial substrates with predefined epigenetic marks.

4:40pm **BM+MN+MS+TF+BI-ThA9 Microfluidic Models of Endothelial Cell Sprouting in Response to Biomechanical and Biochemical Microenvironments, A.M. Shamloo, S.C. Heilshorn, Stanford University**

A novel microfluidic device was designed in order to generate stable, quantifiable concentration gradients of biomolecules in a cell culture chamber for 2-D and 3-D studies of shear-sensitive cell types such as endothelial cells. Endothelial cells form the inner lining of blood vessels and initiate a critical step in angiogenesis (the sprouting of new blood vessels) during wound healing and cancerous tumor growth. Therefore, a deeper understanding of the critical biomechanical and biochemical factors regulating endothelial cell sprouting can lead to improved clinical therapies for a multitude of diseases. Concentration distribution of soluble growth factors inside the microfluidic cell culture chamber was determined by simulation and experiment, and the stability of the gradient was verified over multiple hours. This device allows independent tuning of the matrix rigidity, the growth factor absolute concentration, and the growth factor concentration gradient steepness within a single experimental platform. Sprout formation of dermal microvascular endothelial cells was studied within collagen gels of varying density (0.3 - 2.7 mg/mL, corresponding to shear moduli of 8 - 800 Pa) that contained stable gradients of soluble vascular endothelial growth factor (VEGF). These experiments revealed that endothelial sprouting into multi-cellular, capillary-like structures is optimized at an intermediate collagen matrix density ( $G' \sim 100$  Pa). At lower matrix densities, cells were more likely to lose their coordinated motion and migrate as individual cells through the matrix; while at higher matrix densities, the cells formed broad cell clusters that rarely elongated into a sprout. Sprout thickness directly correlated with matrix rigidity, with thicker and less frequent sprouts present in gels with the highest shear moduli. Intriguingly, our 3D experiments also found that endothelial sprouts alter their sensitivity to VEGF depending on the matrix density, suggesting a complex interplay between biochemical and biomechanical factors. As matrix stiffness increases, steeper VEGF gradients and higher VEGF absolute concentrations are required to induce directional sprouting. In more compliant gels, endothelial sprouts that originally misaligned were able to turn and properly reorient parallel to the VEGF gradient; however, this turning phenomenon was only rarely observed in stiffer gels. These results demonstrate that matrix stiffness is an effective factor in stabilization and orientation of endothelial cells during sprouting and suggests new anti-angiogenic strategies for potential cancer treatment as well as pro-angiogenic strategies for regenerative medicine scaffolds.

5:00pm **BM+MN+MS+TF+BI-ThA10 Plasma Polymerisation of PDMS for Microfluidic Applications, S. Forster, A.G. Pereira-Medrano, G. Battaglia, P.C. Wright, University of Sheffield, UK, S.L. McArthur, Swinburne University of Technology, Australia**

Polydimethylsiloxane (PDMS) has become the most popular material choice for a wide range of microfluidic bioengineering applications, including proteomics, protein separations and drug discovery and development. The reasons its popularity lie mainly in its highly advantageous fabrication requirements when compared to traditional materials such as glass and silicon. However, PDMS has some fundamental drawbacks, namely a lack of functionality present at the surface, high protein fouling and an inability to retain stable surface modification due to its motile hydrophobic monomer. These factors can lead to the loss of specificity and sensitivity in many bioassays. Due to this reason much work has been completed looking into surface modification of PDMS for such applications. Here an alternative method of stable surface modification of PDMS for many microfluidic applications through enhanced curing conditions and plasma polymerisation is shown. Stable and functional surface coatings have been achieved on bulk PDMS and within microfluidic channels. Bulk surfaces were characterised using a combination of XPS and ToF-SIMS, while coated micro-channels were tested using confocal microscopy and various assays. This methodology has been used in many applications and one area where it has proven extremely useful is in microfluidic proteomics where surface properties are of paramount importance due to the inherently small volumes and quantities associated with biological samples. Firstly, plasma polymer coated PDMS micro-

channels were utilised for on-chip IEF protein separations (i.e. separating proteins bases on charge) and showed reduced electrosmotic flow (EOF) and protein adsorption within the device. Secondly, a  $\mu$ IMER (micro-immobilised enzyme reactor) was produced using plasma polymer coated PDMS devices. The  $\mu$ IMER was then used in 'shotgun' protein digestion applications in conjunction with Mass Spectrometry where it was shown to have numerous advantages over untreated PDMS devices, as well as comparing favorably to published work on other  $\mu$ IMER systems. The device was used to digest single and multiple protein samples as well as complex membrane protein samples. Finally, successful covalent bonding of plasma polymer coated devices has led to the completion of polymer vesicle immobilisation within a microfluidic channel. Initial work looking at the immobilisation of polymer vesicles with an encapsulated digestive enzyme has shown to increase proteomic digestion efficiency. This success opens up the possibility of translating this technique into many potential microfluidic applications through the extensive versatility of encapsulation within polymer vesicles.

# Thursday Afternoon Poster Sessions

## BioMEMS Focus Topic

Room: Hall 3 - Session BM-ThP

## BioMEMS Poster Session

**BM-ThP1 A Novel Nanoporous Carbon Materials for Adsorption Gibberellins Acid from Solution.** *J. Li*, Hunan University, PR China and University of Florida, *J.-T. Xia, J.-H. Zhang*, Hunan University, PR China, *T. Liang*, University of Florida

Gibberellin acid (GA) is one kind of ubiquitous phytohormones that regulates various developmental processes of the plant growth. Monitoring and controlling the phytohormone is very important to ensure the efficient growth of crops to bring a high yield and quality production in agriculture or horticulture. Since the content of phytohormone in a plant is very low, and easily decomposed by heat, light, and oxygen, it is of considerable interest to the phytohormone research to prepare some specific adsorbents for adsorption of phytohormone molecules from the solution. A novel nanoporous carbon with tailored pore structure has been synthesized by dynamic carbonization with silica gel networks as a template, which was formed in sol-gel polycondensation using tetraethoxy silane (TEOS) as a silica precursor and sucrose as a carbon source. The pore structures of this kind of nanoporous carbon can be tunable to a high pore volume of 1.25 cm<sup>3</sup>/g and a large specific surface of 1744 m<sup>2</sup>/g, providing an high adsorption capacity about 6.8mg/g to the gibberellin acid (GA) in a solution within 9 h reaction time, and which is over 4~5 times higher than the adsorption of protein and starch in the same solution. It is indicated this nanoporous carbon materials has potential application as a novel adsorbent in the separation and purification of GA from the solution for its monitoring assay.

**Key words:** Nanoporous carbon; Adsorption; Gibberellin acid;

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**BM-ThP2 Boundary Slip and Nanobubble Study in Micro/Nanofluidics with Atomic Force Microscope.** *Y. Wang, B. Bhushan*, The Ohio State University

The boundary condition at the liquid-solid interface in micro/nano scale is an important issue in micro/nanofluidics systems. Recent studies have shown that the fluid velocity near solid surfaces is not equal to the velocity of the solid surface on hydrophobic surfaces, which is called boundary slip. The degree of boundary slip is evaluated by a slip length. Theoretical and experimental studies suggest that at the solid-liquid interface, the presence of nanobubbles is responsible for the breakdown of the no-slip condition. Nanobubbles are long lasting on hydrophobic surfaces, and movement and coalescence of nanobubbles are observed with higher scan loads during imaging with tapping mode AFM.

The slip length can be measured with both contact atomic force microscopy (AFM) and dynamic AFM methods. In the contact AFM method, the slip length is obtained by fitting the measured hydrodynamic force applied to a sphere as a function of separation distance between the sphere and solid surfaces when the sphere approaches the surfaces. In the dynamic AFM method, the amplitude and phase shift data of an oscillating sphere are recorded during approach to sample surfaces at low velocities. These data are then used to get the hydrodynamic damping coefficient to obtain the slip length. Until now, slip length has generally been studied on hydrophobic surfaces with AFM. The boundary slip properties of superhydrophobic surfaces are seldom studied. The impact of surface roughness on the obtained slip lengths also needs to be eliminated for superhydrophobic surfaces. Moreover, because the sphere should disturb the nanobubble during approach to sample surfaces in both the contact and dynamic AFM method, a new technique is needed to evaluate boundary slip. Regarding nanobubbles, the current studies mainly focus on their physical properties. The interaction between nanobubbles and the surfaces supporting them is seldom studied. More importantly, the relationship between nanobubble immobility and surface properties should be studied.

In this study, both contact and dynamic AFM methods have been applied to study the boundary slip on hydrophilic, hydrophobic, and superhydrophobic surfaces. A new AFM based technique is proposed to study boundary slip. Nanobubble movement and coalescence, as well as tip-bubble interaction, are studied in detail. The physical interaction between nanobubbles and the surfaces supporting them is investigated. Moreover, the relationship

between nanobubble immobility and surface properties of hydrophobic surface is revealed.

**BM-ThP3 Surface Plasmon Resonance Imaging of Carbohydrate Microarray: Kinetics, Surface and Solution Binding Affinity.** *A. Tyagi, M. Yan*, Portland State University

Oligosaccharides are increasingly being recognized as important partners in glycan-lectin binding and cellular signaling. Surface plasmon resonance (SPR) is a powerful tool for the real-time study of the specific interactions between biological molecules. We have developed an efficient surface coupling chemistry to probe carbohydrate-lectin interactions in an array format using SPR imaging. The coupling agent, a thiol-functionalized perfluorophenyl azide, PFPA-MUTEG, allows the covalent attachment of carbohydrates to gold surface by way of CH insertion reactions. The SPR chips were modified with mixed SAMs of PFPA-MUTEG and MDEG before the carbohydrate ligands were arrayed and immobilized. The carbohydrate array was composed of  $\alpha$ -1,3- $\alpha$ -1,6-D-mannotriose,  $\alpha$ -1,2-D-mannobiose, D-mannose, D-glucose and D-galactose, and the binding studies were carried out using Concanavalin A, a plant lectin that exhibits mannose-binding properties. The kinetic equilibrium constant (KA), adsorption coefficient (KADS) and solution equilibrium constant (KD) were obtained for each carbohydrate at different mixed SAM composition. The SAM containing 10% MDEG showed the highest sensitivity and the least non-specific adsorption. The KADS values for mannotriose, mannobiose and mannose were measured to be  $10.3 \pm 1.1 \times 10^6$ ,  $7.6 \pm 1.0 \times 10^6$ ,  $1.3 \pm 1.0 \times 10^6$ M<sup>-1</sup>, respectively.

**BM-ThP4 Chemical and Morphological Properties of Amino-Silane Coated Surfaces for DNA Purification.** *L. Marocchi, L. Lunelli, L. Pasquardini, C. Potrich, L.E. Vanzetti*, FBK-CMM, Italy, *G. Guella*, University of Trento, Italy, *C. Pederzoli*, FBK-CMM, Italy, *M. Anderle*, Provincia Autonoma di Trento, Italy

DNA purification and PCR amplification are a requirement for most genetic analysis. Combining these processes in a single micro device minimizes sample loss and contamination problems as well and reduces time and costs of analysis. Different strategies are available to perform DNA extraction on a chip. Here we exploited amino-coated silicon and pyrex surfaces as a tool for specific binding of DNA through the electrostatic interaction between amino groups and nucleic acids. Amino groups have been introduced on the surfaces via silanization carried out in wet condition [1] using three silanes carrying a different number of amino groups and different alkoxy groups (3-Aminopropyl)triethoxysilane (APTES), (3-Aminopropyl)trimethoxysilane (APTMS) and (3-[2-(2-Aminoethylamino)ethylamino]propyl-trimethoxysilane (AEEA)). The influence of different silanization conditions on surface properties, such as homogeneity and thickness of the silane layer, was also studied by changing solvents, concentration of silane solution and reaction temperature. The kinetic of hydrolysis of the alkoxy groups followed by oligomerization of aminosilanes was characterized by NMR measurements. Amino-coated surfaces were characterized by AFM, XPS and absorption spectroscopy to define their chemical and morphological properties. Multi-amino silane were found less prone to form uniform and tiny layers than mono-amino silanes, resulting less suitable for successive PCR amplification.

Finally, we analyzed the ability of treated surfaces to selectively adsorb/desorb genomic DNA with the aim to purify DNA from unwanted cellular components. Preliminary results suggest this strategy as very promising, permitting to obtain a considerable yield of purified DNA in short time.

[1] Fiorilli, S.; Rivolo, P.; Descrovi, E.; Ricciardi, C.; Pasquardini, L.; Lunelli, L.; Vanzetti, L.; Pederzoli, C.; Onida, B. & Garrone, E. (2008), *Journal of Colloid and Interface Science* **321**, 235-241.

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Pederzoli, C.: BM-ThP4, 4  
Pereira-Medrano, A.G.: BM+MN+MS+TF+BI-ThA10, 3  
Persson, H.H.J.: BI+AS+BM+MS-WeM12, **1**  
Potrich, C.: BM-ThP4, 4  
Pringsheim, E.: BI+AS+BM+MS-WeM11, 1

## — R —

Rant, U.: BI+AS+BM+MS-WeM11, **1**  
Riehn, R.: BM+MN+MS+TF+BI-ThA8, **2**  
Romero-Ortega, M.: BM+MN+MS+TF+BI-ThA3, 2

## — S —

Sahin, O.: BI+AS+BM+MS-WeM12, 1  
Schiess, A.: BM+MN+MS+TF+BI-ThA3, 2  
Shamloo, A.M.: BM+MN+MS+TF+BI-ThA9, **3**  
Shumaker-Parry, J.: BM+MN+MS+TF+BI-ThA6, 2  
Streng, D.E.: BM+MN+MS+TF+BI-ThA8, 2

## — T —

Tisone, T.C.: BI+AS+BM+MS-WeM5, **1**  
Tyagi, A.: BM-ThP3, **4**

## — V —

Vanzetti, L.E.: BM-ThP4, 4

## — W —

Wang, Y.: BM-ThP2, **4**  
Wingren, C.: BI+AS+BM+MS-WeM1, **1**  
Wright, P.C.: BM+MN+MS+TF+BI-ThA10, 3;  
BM+MN+MS+TF+BI-ThA4, 2

## — X —

Xia, J.-T.: BM-ThP1, 4

## — Y —

Yan, M.: BM-ThP3, 4

## — Z —

Zhang, J.-H.: BM-ThP1, 4