Tuesday Morning, October 21, 2008

BioMEMS Topical Conference
Room: 309 - Session BM+MN+BI+BO-TuM

MEMS/NEMS for Biology and Medicine
Moderator: E. Meng, University of Southern California

8:00am **BM+MN+BI+BO-TuM1 Microfluidic Systems for Cell Growth and Analysis**, ***K.F. Jensen***, *A. Adamo, L. Ye, Z. Zhang*, Massachusetts Institute of Technology **INVITED**

We present microfluidic systems for cell growth, including instrumented microbioreactors in which the main process parameters (e.g., optical density, dissolved oxygen and pH) are monitored optically and controlled. The system accommodates bioreactors in different operational modes, batch and continuous. The systems are disposable and consist of layers of poly(methyl methacrylate) for structural integrity and poly(dimethyl siloxane) (PDMS) layers for aeration. We also combine cell growth with analysis of protein responses underlying cell signaling. Analysis of these potentially fast transient events requires very short treatment times and well-controlled and reproducible stimulus conditions. Consequently, such pathways can be difficult to probe reproducibly with conventional laboratory techniques that are susceptible to small fluctuations in manual handling – in particular at short times. Microfluidic systems provide for reproducible and automated analysis with excellent control over experimental conditions. We describe microfluidic based methods for investigating signaling pathway ways of adherent cells with the overall aim of controlling cell culture, cell stimulation, and the subsequent protein analysis. The devices, which are fabricated in PDMS by soft lithography, enable dynamic studies of cell signaling by taking advantage of the equivalence between distance travelled along a microfluidic channel and treatment time. They perform all the necessary steps needed in stimulus-signal response analysis of signaling pathways by a fluorescent immunocytochemical assay including cell culture, cell stimulus, cell fixation, and antibody analysis. Average cell population data are obtained by scanning and imaging the entire device, while high resolution microscopy moving along the channel allows responses to be collected at the single cell level. Finally, we present microfluidic devices for quantitative microinjection of macromolecules and nanoparticles into living cells. These approaches overcome limitations with traditional manual manipulation of microinjection needles.

8:40am **BM+MN+BI+BO-TuM3 High-Throughput pMHC Microarrays for Characterizing Diverse T Cell Populations**, ***M. Paulaitis***, Ohio State University and Johns Hopkins University, *C. Yue, N. Guzman*, Ohio State University, *J. Schneck, M. Oelke*, Johns Hopkins School of Medicine **INVITED**

We are developing protein microarrays for rapidly scanning and screening diverse T cell populations to characterize human adaptive immune responses. An important early molecular recognition event that triggers an immune response is the interaction of a T cell receptor (TCR) on the surface of the T cell with its complementary major histocompatibility complex (MHC) on the surface of antigen-presenting cells. This interaction is mediated by a small peptide (the antigen) 8-10 amino acids in length bound to the MHC, such that the amino acid sequence of the peptide antigen determines the specificity of the TCR/peptide-MHC (pMHC) interaction. Surface plasmon resonance studies of TCR/pMHC interactions have established that the overall range of binding affinities of stimulatory pMHC ligands is low relative to that for anti¬body-antigen interactions. Yet, these interactions have remarkably high specificity/sensitivity leading to T cell activa¬tion and different immune responses depending on the nature of the peptide. Individual T cells are also characterized by a unique TCR; therefore, pMHC microarrays printed with peptides having different amino acid sequences serve to distinguish T cells by their characteristic TCR/pMHC interactions. In addition, co-printing antibodies against cytokines secreted by the captured T cells enables an antigen-specific functional analysis of T cell activation across this population. We show that pMHC microarrays can selectively capture and enumerate antigen-specific T cells in diverse populations at high sensitivity, and that this information provides insights into the general principles governing early molecular recognition events in human immune responses. Results on the functional diversity of the human immune response will also be presented. This work is supported by the National Science Foundation (BES-0555281) and the National Institute of Allergy and Infectious Diseases of the National Institutes of Health (1R21AI077097-01).

9:20am **BM+MN+BI+BO-TuM5 Tunable Microeddies for Microfluidics: Non-contact Single-cell Trapping using Gentle Fluid Flow**, ***B.R. Lutz****, J. Chen, D.T. Schwartz,* University of Washington*, D.R. Meldrum*, Arizona State University

Cells that normally live in suspension typically exhibit strong biological responses to physical contact. Microfluidic devices have been very successful for studying single adherent cells in controlled chemical environments, but tools for manipulating single cells in suspension are extremely limited. We developed a non-contact microfluidic single-cell trap that creates strong trapping forces using only gentle fluid flow. The traps are based on steady streaming flow, which is the steady flow generated when oscillating fluid interacts with any boundary that causes the fluid to turn (e.g., obstacles, cavities, bends). Steady streaming was first identified over a century ago, but its remarkable ability to trap cells was not known. A key feature of this approach is that traps are insensitive to differences in cell shape, cell density, and fluid medium. We demonstrate the ease of trapping for bubbles, spheres, rod-like debris, non-spherical motile phytoplankton, macrophages, and monocytes in different fluid media. The approach is remarkably simple to implement and control, in fact, early work used hand-built flow channels and a home stereo amplifier. The flow is created by audible-frequency fluid oscillation in a microchannel containing a cylindrical post. The back-and-forth motion creates four eddies around the cylinder, and each eddy traps a cell and holds it in place at a predictable location within the fluid. We use capture and release of swimming phytoplankton to estimate the trap strength; strong trapping forces capable of holding the strongest swimmers are easily generated (>30 picoNewtons), while gentle shear conditions in the traps are comparable to arterial blood flow. By using flow to displace trapped spheres under different conditions, we determine a simple scaling relationship that quantitatively describes the trapping force for common cell sizes (5-50 microns). The traps withstand net flows as large as 1 cm/second, which enables medium exchange and chemical treatment of single cells in suspension. Posts can be arrayed with little effect on trapping behavior, providing the potential for high-throughput screening of suspension cells based on dynamic measurements. The combination of strong, tunable trapping forces and gentle trapping environment makes this an appealing new alternative for manipulating single cells in microfluidic devices.

9:40am **BM+MN+BI+BO-TuM6 Automated on-Chip Rapid Microscopy, Phenotyping, and Screening of C. elegans**, ***H. Lu***, Georgia Institute of Technology

Microscopy, phenotyping, and visual screens are critical methods frequently applied to model organisms in combination with genetics. Although widely used, these techniques for multicellular organisms have mostly remained manual and low-throughput. We report the complete automation of sample handling, high-resolution microscopy, phenotyping, and screening of C. elegans using a custom-designed microfluidic system. The engineered system, coupled with customized software, enables high-throughput diffraction-limited imaging and sorting of samples with no human intervention with any microscopy setup. The robustness and automation of our system relies greatly on integrated closed-loop control software as well as engineered hardware design of the microchip. The chip has six salient features that ensure a consistent and reliable operation for an extended period of time. First, it automatically self-regulates the loading of nematodes by a simple passive loading-regulator design. Constant pressure drives the flow, so that no feedback or intervention is necessary for the microchip to allow one and only one animal to occupy the imaging area at a time. Second, the setup automatically positions the samples in an identical position in the chip, so as to minimize the travel of the motorized stage and thereby reduce the processing time and increase the throughput. Third, the device has an integrated local temperature control system whereby animals are cooled to ~4 °C and completely immobilized briefly (~ a few seconds) for imaging and manipulation without the use of anesthetic drugs. Cooling provides an alternative to anesthetics, potentially minimizing the adverse developmental effects. Fourth, the microchip and the setup are compatible with any standard microscopy setup with no modification necessary, including simple compound epifluorescence microscopy, as well as more expensive multiphoton or confocal microscopy. Fifth, the microchip has no permanent small features (<20 µm), and therefore is easy to fabricate, less likely to be clogged by debris, and can operate very robustly. Lastly, losses through our system are minimal (~3%,) and the device design is gentle on the animals as the viability of all the sorted animals is ~100%. We show that compared to standard manual operation, time for phenotyping and visual screens can be reduced by ~2 orders of magnitude in our system with no human intervention, which has not been demonstrated before. Moreover, we show the ability to perform multiple sensitive and quantitative genetic

screens with real biological samples based on cellular and subcellular features with over 95% accuracy per round.

10:40am **BM+MN+BI+BO-TuM9 BioMEMS Challenges and Opportunities – A Department of Defense Perspective**, *D. Polla,* Defense Advanced Research Projects Agency*,* ***S. Barker***, System Planning Corporation **INVITED**

Considerable progress has been made over the last 15 years in realizing a great variety of BioMEMS devices and systems. The field of BioMEMS can be approximately subdivided into three categories: (1) bioanalytical systems, (2), surgical systems, and (3) therapeutic systems. All three areas have numerous commercial and defense applications, but in many cases progress is inhibited by fundamental scientific and technological challenges. This paper presents the authors' perspective on the top 10 challenges facing BioMEMS today. Bioanalytical systems, which are also commonly referred to as "lab-on-a chip," have not realized their full potential for numerous reasons: (1) Autonomous sample processing with minimal human intervention has yet to be achieved. (2) Sample clean-up and pre-processing pose significant challenges that often limit the performance of a bioassay. (3) The ability to take a biological sample and obtain a result or set of results is still a long process, often taking several hours; obtaining a microfluidics-based PCR bioassay result in less than one minute persists as a grand challenge for the BioMEMS community. (4) The development of size-scaled microinstruments for bioanalysis presents an enormous opportunity toward the realization of remote site-derived information that can be conveniently communicated to a physician's office and correlated with a patient's stored medical record. The potential of surgical MEMS has often meant "micro-invasive" surgery that provides significant benefit to the patient. (5) But non-invasive surgery enabled by MEMS has the potential for providing even better patient outcomes. (6) Both sensors and actuators with the capability for more accurately and more reliably reproducing the skill of a surgeon's hands still need to be realized. (7) Developments that enable in vivo imaging of cells and organs using MEMS devices may also play an important role in enabling more effective precision surgeries. Therapeutic systems based on MEMS technology have yet to be made smart. (8) This means effectively integrating sensors, electronics, and actuators in a controlled feedback system designed to provide therapy only when the body needs it. These systems are often implantable and are limited by (9) battery size and lifetime. And finally, (10) neural prosthesis represents an exciting new domain where MEMS may provide an effective interface between nerves and electronics.

11:20am **BM+MN+BI+BO-TuM11 CD Based Sample Preparation and Pathogen Screening**, ***M.J. Madou***, Unversity of California, Irvine **INVITED**

We have demonstrated the feasibility of a multiplexed microfluidic CD apparatus for sample preparation of a wide variety of clinical samples and the subsequent detection of viruses, bacteria and fungi through fast DNA hybridization on the same platform. As the CD slowly rotates, a free moving magnetic disc in a lysis chamber is moved back and forth in the radial direction by the magnetic force of stationary magnets located below the rotating CD. The movement of the magnetic disk causes mechanical shear that disrupts cell membranes. This CD does not only process multiple samples simultaneously, but can also be used for the centrifugal precipitation of solids from each sample liquid. After precipitation, the resulting clarified liquid is transferred through a solid phase extraction membrane to capture the DNA. This step is followed by subsequent automated washing, elution, and detection by hybridization and fluorescence detection on an embedded DNA array. Recent sample to answer results and modeling of ice valves and coriolis valves will be detailed.

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