Tuesday Afternoon, November 1, 2011

Biofabrication and Novel Devices Focus Topic
Room: 105 - Session BN-TuA

Biofabrication Methods and Devices

Moderator: L. Gamble, University of Washington

2:00pm **BN-TuA1 Microengineered Hydrogels for Stem Cell Bioengineering and Tissue Regeneration**, ***A. Khademhosseini***, Brigham and Women's Hospital, Harvard Medical School, MIT, and Harvard University **INVITED**

Micro- and nanoscale technologies are emerging as powerful tools for controlling the interaction between cells and their surroundings for biological studies, tissue engineering, and cell-based screening. In addition, hydrogel biomaterials have been increasingly used in various tissue engineering applications since they provide cells with a hydrated 3D microenvironment that mimics the native extracellular matrix. In our lab we have developed various approaches to merge microscale techniques with hydrogel biomaterials for directing stem cell differentiation and generating complex 3D tissues. In this talk, I will outline our work in controlling the cell-microenvironment interactions by using patterned hydrogels to direct the differentiation of stem cells. In addition, I will describe the fabrication and the use of microscale hydrogels for tissue engineering by using a 'bottom-up' and a 'top-down' approach. Top-down approaches for fabricating complex engineered tissues involve the use of miniaturization techniques to control cell-cell interactions or to recreate biomimetic microvascular networks within mesoscale hydrogels. Our group has also pioneered bottom-up approaches to generate tissues by the assembly of shape-controlled cell-laden microgels (i.e. tissue building blocks), that resemble functional tissue units. In this approach, microgels were fabricated and seeded with different cell types and induced to self assemble to generate 3D tissue structures with controlled microarchitecture and cell-cell interactions.

2:40pm **BN-TuA3 Nanoscale Architectures for Probing Cell Mechanics**, ***S. Wind***, *M. Schvartzman, M. Palma, M. Biggs, T. Fazio, R. Piqueras Jover, M. Sheetz*, Columbia University

The physical properties of a cell's environment are important factors in determining cell behavior and ultimately, phenotype. Two key factors that have been associated with major changes in cell morphology and behavior are (1) spatial organization of extracellular matrix (ECM) molecules and (2) rigidity. In order to understanding how cells sense these factors at the nanoscale and how these factors affect cell function, we have developed new nanofabricated surfaces in which these physical characteristics of the ECM are simulated.

The first type of surface combines nanoimprint lithography with selective biofunctionalization to precisely control the placement and geometric arrangement of integrin binding sites. The binding sites consist of sub-10 nm metallic nanodots functionalized with ECM binding ligands, designed so that each site can accommodate only a single integrin molecule. Cell spreading and motility assays were performed using 3T3 fibroblasts on arrays in which binding site spacing, density and number were independently varied. Cell spreading efficiency was markedly enhanced for clusters comprising at least 4 liganded sites spaced $\leq$ 60 nm apart, with little or no dependence on global density. This points to the existence of a minimal matrix adhesion unit defined in space and stoichiometry.

A second type of surface consists of elastomeric substrates with locally variable rigidity. We have found that exposure of poly(dimethylsiloxane) (PDMS) to an electron beam alters the rigidity of the elastomer, with the modulus of the exposed regions increasing with the applied electron dose. In addition to planar surfaces, pillared substrates can be patterned with no measurable change to the pillar dimensions. Immortalized mesenchymal stem cells plated on soft PDMS surfaces patterned in this manner displayed a distinct preference for the more rigid, exposed regions, forming focal adhesion nearly exclusively there. Furthermore, focal adhesion formation diminished significantly as the size of the exposed features was reduced below 1 μm, indicating that there is a length scale for cellular rigidity sensing, with the critical length in the range of a few hundred nanometers.

By adapting the tools of nanomanufacturing to cellular systems, we are able to define important parameters that can control aspects of cell function and behavior and will help identify conditions under which these functions may be altered. Potential applications range from therapeutic treatments that block metastasis to the development of new adoptive immunotherapies, as well as the development of new guidelines for the design of tissue scaffolds that can optimize healing without scarring.

3:00pm **BN-TuA4 Production of Functionalized 3D Micro Environment for Cell Culture**, ***J. Nowak***, *D. Mehn, P. Colpo, M. Zurn, T. Martin, F.J. Rossi*, European Commission, JRC Institute for Health and Consumer Protection, Italy

One of the main challenges for the robust *in-vitro* studies is to obtain adaptable 3D culture systems that may mimic the tissue environment. Unfortunately the universal condition used in 2D cell culture techniques may hinder the full functionality of cells and generate misleading results.

Fabrication of firm and flexible micro-structures from organic polymers offers benefits for making smart 3D environments capable of driving cell behavior and surpassing the limitations of the 2D systems. These 3D bio-scaffolds can be employed to study various aspects of cell biology. Furthermore upon functionalization with the extra-cellular matrix proteins or signaling molecules they can be used as platforms for governing stem cell differentiation into the specialized cell types.

Here we present the straightforward approach to generate 3D bio-scaffolds that can facilitate cell growth under controlled geometrical and chemical conditions.

The technique involves UV cross-linking of the polymeric precursors to create the micro-well structures. The geometrical features of the structures are obtained by introducing a physical mask in contact with a liquid precursor, therefore restricting the region of the polymerization. We used PDMS mold as a physical mask to direct the polymerization of the PEG-DA and epoxy based polymers. However the technique can be used with various UV-sensitive polymeric materials.

The chemical and geometrical properties of the structures were characterized by XPS and microscopic techniques.

The features of the scaffolds lead to the development of a geometrically defined neuronal network when applied as platforms in a primary-neuron culture. Cell morphology and expression of the neuronal markers were characterized by fluorescent microscopy.

Authors Index

Bold page numbers indicate the presenter