Biomaterial Interfaces
Room: Milo - Session BI-WeM

Biomaterials, Interfaces, and Cells
Moderator: Keith McLean, CSIRO, Australia

8:20am **BI-WeM2 Self-Assembly of Macroscopic RNA Membrane**, *Y. Park, H. Kim,* ***Jong Bum Lee***, University of Seoul, South Korea

DNA and RNA have gained attention as powerful materials for bio-nanotechnology. Although, a variety of DNA structures have been developed, structures based on RNA are extremely rare. Here, we developed the robust and free-standing RNA membrane using an enzymatic synthetic method. This macroscopic RNA structure was fabricated by following two steps, complementary rolling circle transcription (cRCT) and evaporation-induced self-assembly (EISA). In addition, properties of the membrane can be controlled by adjusting base-pairing of RNA strands and the concentration of template circular DNA. In this research, we fabricated three types of RNA membranes and used these membranes for controlled drug release systems.

8:40am **BI-WeM3 Cytocompatible Mineralization on Jurkat T Cell Surfaces with Titania Composites**, ***EunHyea Ko***, KAIST, Republic of Korea, *W.G. Youn,* KAIST*, I.S. Choi*, KAIST, Republic of Korea

The artificial shells of organic/inorganic materials on living cells would give new properties to the encapsulated cells. For example, the encapsulate cell could live against physical deformation and chemical hazards, and control the cell division. Also, functionalization would be easier than native cell surface. Recently, our paper said that the $(RKK)_4D_8$ peptide had both the TiO_2-inducing and cytocompatible units for *Chlorella* cell. However, due to the fragile property of mammalian cells, there are few studies on the mammalian cell encapsulation.

In this work, individual Jurkat T cells were encapsulated within peptide/TiO_2 composite shells by layer-by-layer assembly and bioinspired mineralization. The cell viability and shape were maintained during the encapsulation processes, and the division of the encapsulated cells was changed by the artificial TiO_2 shell. There are cluster of differentiation 3 (CD3) antigens on the Jurkat T cell surface. After encapsulation, anti-CD3 antibody was hindered to bind CD3 antigens on the cell. For the functionalization of cell surface, TiO_2 composites made it possible to anchor the ligands of interest to the shell. After formation of the TiO_2 shells on cell surfaces, the shells were functionalized via catechol chemistry in a cytocompatible fashion. We believe that these new properties on the Jurkat T cell surface could be apply to cell-based sensors and assays as well as for fundamental studies such as immunology.

9:20am **BI-WeM5 Selective Cell Adhesion to Surface Nanotopography**, ***Elena Liang***, *MN. Dickson, N. Vollereaux, AF. Yee*, University of California, Irvine

Understanding cell interactions with material surfaces is critical to the performance of medical devices. Of particular interest to our research, such understanding could lead to simple and durable ways to control cell adhesion without chemically modifying the surface of biomaterials used in implantable devices. Recently, it was found that the nanopillar structures on cicada wings are inherently antibacterial irrespective of surface chemistry (Ivanova *et al*. Small. 2012). Such nanostructures can eventually be incorporated on surfaces of medical devices, but first, we need to ensure that patient's own cells would not be adversely affected by these structures. Hu *et al*. showed that nanopillars of widely varying aspect ratios and surface energies had strong effects on cell morphology, discouraging cell spreading (Hu *et al.* 2010). Kong *et al*. discovered that human embryonic stem cells grown on nanopillar structures have a significantly reduced number of focal adhesions per cell and concordantly exhibit increased cell motility on the nanopillars (Kong *et al*. 2013). Based on these findings, we hypothesized that the pillar nanostructures on the cicada wing would prevent cells from adhering. To show this, we first created a library of nanostructures, beginning with a biomimetic cicada wing replicate. We molded a negative hPDMS stamp of the cicada wing and pressed the stamp into polymethylmethacrylate and polystyrene films to create the polymer replicates. We also fabricated pillar arrays of different spacings from commercially available silicon molds using nanoimprint lithography. To evaluate cell adhesion, we counted the number of fibroblasts adhering to flat polymer and the nanopillars, and we determined the number of focal adhesion sites from immunostaining for vinculin, a major protein in the focal adhesion complex. In addition, we examined cell morphology on the various surfaces. After 24 hours, we observed that the cells adopted different cell morphologies, possibly indicating changes in adhesion dynamics. Fibroblasts showed a spread-like morphology on the flat film while the cells on pillars were more equiaxed. Our study has shown that nanostructures in the 100-500 nm-size range do affect cell adhesion dynamics. We found that structure dimensions modulate the adhesion of cells, which may provide researchers a useful means of controlling cell adhesion on material surfaces.

9:40am **BI-WeM6 Polysaccharide Films at an Air/Liquid and a Liquid/Silicon Interface: Effect of the Polysaccharide and Liquid Type on their Physical Properties**, ***Cathy McNamee***, Shinshu University, Japan, *Y. Taira*, Tohoku University, Japan

Chitin and chitosan show biocompatibility, biodegradability, and non-toxity, and are therefore used in pharmaceutical and biomedical applications. The successful applications of chitin and chitosan require the ability to create well-defined films that display the required properties in the working environment. This ability requires an in-depth understanding on the physical properties of the films created using chitin or chitosan and the way to control these properties in different environments. The polysaccharide type, its conformation and packing in the film, and the surrounding liquids in the working environment contribute to the forces and friction of the system, which affect the properties of the polysaccharide films. We investigated the effect of the polysaccharide type, the subphase on which the chitin or chitosan Langmuir monolayers were prepared, and the liquid in which the properties of the transferred monolayers were measured on the physical properties of the polysaccharide films at an air/aqueous interface and at a liquid/silicon substrate interface, and the forces and friction of the polysaccharide transferred films when measured in solution against a silica probe.

Chitosan was modified with a silane coupling agent to make chitosan derived compounds with a low and a medium molecular weight. Chitin and the chitosan-derived compounds were used to make Langmuir monolayers at air/water and air/pH 9 buffer interfaces. The monolayers were transferred to silicon substrates via a Langmuir-Blodgett deposition, and the chitosan-derived compounds subsequently chemically reacted to the silicon substrates. Atomic Force Microscope force and friction measurements were made in water and in the pH 9 buffer, where the water and the pH 9 buffer acted as a good and a bad solvent to the polysaccharides, respectively.

The polysaccharide type affected the friction of the polysaccharide film, where the physically adsorbed chitin gave the lowest friction. The forces and friction of the polysaccharide films changed when the subphase on which the Langmuir monolayers were formed was changed or when the liquid in which the properties of the films adsorbed at the silicon substrate were being measured was changed. The friction increased significantly when the liquid was changed from water to the pH 9 buffer.

10:20am **BI-WeM8 Engineered Surfaces for Stem Cell Expansion**, ***Laurence Meagher***, CSIRO, Australia **INVITED**

Control over biomolecule-material and cell-material interactions is critical to the performance of designed surface coatings in a broad range of applications including cell culture materials, implantable biomedical devices and biosensors. Three key design features for materials used in the expansion of cells is that the materials should have very low non-specific protein adsorption, the coatings should be covalently attached to the substrate and should contain covalently attached, highly specific ligands to mediate cell attachment. For cell therapy applications, these materials should be able to function effectively in cell culture media which is chemically defined and animal product free (i.e. serum-free). We have developed a platform coating approach[1], which in one step, results in coatings with very low non-specific protein adsorption, i.e. no initial chemical functionalisation or priming steps are required. In addition, the coatings also contain functional groups onto which cell attachment ligands such as peptides can be chemically attached. The approach can be used to produce coatings on many different formats of interest, such as multiwall plates, tissue culture flasks and microcarrier particles. Microcarrier particles are particularly attractive for application in stirred tank and wavebag-type bioreactors

In this study we have prepared a number of synthetic polymer coatings using a platform grafting from approach to produce materials for the culture of cells. Coatings were formed using a grafting from approach from a monomer feed comprising 10 mole percent acrylic acid and 90 mole percent acrylamide. Coatings were found to be similar in composition to the monomer feed ratio, highly swelling. Characterisation was carried out using X-ray photoelectron spectroscopy and atomic force microscopy. Coupled to these coatings was a cyclic peptide (cRGDfK) which interacts in a highly specific manner with αvβ3 integrins only. These surfaces were found to be

highly suitable for the attachment and growth of murine L929 fibroblasts, bone marrow derived human mesenchymal stem cells (hMSCs) and human embryonic stem cells (hESCs). Furthermore, in the case of hMSCs the surfaces were used to expand the cells over three passages in three different media (two were serum free). The hMSCs were characterised by their ability to differentiate into adipocytes, osteocytes and chondrocytes as well as maintenance of cell surface markers typically used to define hMSCs.

References

[1.] Ameringer, T., Meagher, L., Thissen, H., Pasic, P., Styan, K., Process for Modifying a Polymeric Surface, WO 2014/000052 A1, 3January 2014

11:00am **BI-WeM10 Why Biointerface Science is Important for Stem Cell Research**, ***Kevin Healy***, University of California, Berkeley, USA **INVITED**

Highly regulated signals in the stem cell microenvironment such as ligand adhesion density, matrix stiffness and architecture, growth factor presentation and concentration have been implicated in modulating stem cell differentiation, maturation, and ultimately function. Therefore, it is desirable to have independent control over both the biochemical and mechanical cues presented to stem cells to analyze their relative and combined effects on stem cell function. Accordingly, we have developed a range of materials systems to study stem cell function. This presentation will discuss our progress in developing: 1) self-organizing human cardiac microchambers mediated by geometric confinement; and, 2) *in vitro* disease specific tissue models (e.g., 'organs on a chip') to be used for high content drug screening and patient specific medicine. Examples of how biointerface science is important in these applications will be highlighted. For example, in the former we used PEG-patterned polystyrene substrates to geometrically confine human pluripotent stem cell colonies and spatially present mechanical stress. Upon chemical modulation of the Wnt/b-catenin pathway, biochemical and biophysical cues synergistically induced self-organizing lineage specification and creation of a beating human cardiac micro-chamber confined by the pattern geometry. In the second theme, we employed microfabrication technologies to form cardiac and liver micro-tissues from patient-specific human induced pluripotent stem cells (hiPSC), to be used for high content drug screening and patient specific medicine. Ideally, the use of human disease specific tissues organized into a single integrated physiological system could have an enormous impact on the early screening of candidate drugs.

11:40am **BI-WeM12 Characterization of Tethered Phospholipid Bilayers by the Electrochemical Impedance Spectroscopy**, ***Gintaras Valincius***, *M. Mickevicius, T. Penkauskas*, Vilnius University, Lithuania

We discuss the characterization of tethered phospholipid bilayer membranes (tBLMs) utilizing the electrochemical impedance spectroscopy (EIS). An emphasis is put on applications of tBLMs in protein (peptide)/phospholipid membrane interaction studies. Such interactions modulate the dielectric properties and affect the integrity of phospholipid bilayer. Because of highly asymmetric structure tBLMs exhibit a unique EI response, which cannot be modeled by simple equivalent circuits consisting of capacitors and resistors. We discuss the special functions that describe the characteristic features of the EI spectra. Those analytical functions obtained by solving problem of voltage-current distribution at the interface take into account the structural and dielectric properties of tBLMs. Also, our analysis of the EIS provides the theoretical background for the utility of tBLMs as bioanalytical sensors for the membrane damaging agents, such as pore-forming toxins. We demonstrate that the magnitude and frequency of the negative of the impedance phase minimum, as well as the magnitude of impedance are the parameters indicative of the extent of the membrane damage, and may be used to estimate the defect density in bilayers, as well as the activity of the membrane damaging proteins and peptides. The precision of such estimates is highly dependent on the knowledge of the electric properties of the submembrane electrolyte reservoir separating phospholipid bilayer and solid surface. We present an algorithm, which allows to make estimates of the submembrane specific resistance. Clustering of the defects affects EI response in a unique way, which may be used for the qualitative analysis of the protein membrane interactions.

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