Biomaterial Interfaces
Room: 317 - Session BI+MG-WeA

Design and Discovery: Biointerfaces
Moderator: Morgan Alexander, The University of Nottingham, UK

2:20pm **BI+MG-WeA1 Discovery of Materials for Stem Cell Control using Polymer Microarrays**, ***Morgan Alexander***, The University of Nottingham, UK

Polymer micro arrays have proven to be useful tools for the discovery of new synthetic materials which control cells.[1] This high throughput (HT) materials discovery approach is attractive because the paucity of understanding of the cell-material interface hinders the *ab initio* rational design of new materials.[2] The large number of polymer chemistries that can be investigated on a single polymer micro array act as a wide "net" in the search for materials that can achieve a certain cell response. Micro array *hits* are the starting point from which new materials may be developed.

Combinatorial acrylate libraries formed on standard glass slides were presented as a HT platform by Anderson and Langer of MIT.[3] To complement materials screening, we developed the approach of *HT surface characterisation* employing a range of analytical techniques in collaboration with the MIT group.[4] This surface characterisation step is necessary to directly relate the effect of the material on attached cells to the actual surface on which they sit, and to enable effective scale up from micro array to culture ware dimensions. Application of chemometrics, to handle the large amounts of complex data, reveals the importance of certain surface moieties, guiding the process of materials discovery and increasing our understanding of the cell-material interface.

We have applied this approach to the identification of materials which resist bacterial attachment and biofilm formation with application in the reduction of medical device centred infection.[5,6] In the mammalian cell field, we have identified materials which show promise as synthetic substrates for pluripotent stem cell culture.[7,8] These materials require pre-treatment with expensive proteins such as vitronectin, a constraint which limits their commercialisation.[9]

In this talk, screening of arrays with greater chemical diversity than ever before, incorporating up to 140 monomers[10], is reported which leads to the identification of materials which support pluripotent stem cell expansion without pre-treatment of the substrate with protein. Materials which support differentiation to mature cardiomyocytes which have potential application in in vitro toxicology screening have also been discovered.

1 Hook, A. L. **Biomaterials** (2010).

2 Kohn, J., **Nature Materials** (2004).

3 Anderson et al. **Nature Biotechnology** (2004).

4 Urquhart, A.et al. **Advanced Materials** (2007).

5 Hook, A. L. et al. **Nature Biotechnology** (2012).

6 Hook, A. L. et al. **Advanced Materials** *(*2013)

7 Mei, Y et al. **Nature Materials** (2010).

8 Saha, K. et al **PNAS** (2011).

9 Celiz, A. D et al. **Nature Materials** (2014).

10 Celiz, A.et al. **Biomaterials Science** (2014).

3:00pm **BI+MG-WeA3 Interfacial Force Field Parameterization in CHARMM for the Accurate Molecular Dynamics Simulation of Peptide Adsorption on High-Density Polyethylene**, ***Tigran Abramyan***, *J.S. Snyder, J.Y. Yancey, S.S. Stuart, R.A. Latour*, Clemson University

A fundamental molecular-level understanding of protein-surface interactions (PSIs) is crucial for many applications in biotechnology and bioengineering. All-atom molecular dynamic (MD) simulation methods hold great promise as a valuable tool for understanding and predicting PSIs. However, current MD force fields have not been validated for this application. In this study, adsorption free energy (ΔG_{ads}) of small TGTG-X-GTGT host-guest peptides (T = threonine, G = glycine, and X = variable amino acid residue) on a high-density polyethylene (HDPE) surface (110 crystalline plane) using the CHARMM force-field were calculated and compared with experimental results in order to find inaccuracies. In order to accurately calculate ΔG_{ads} in our simulation studies, advanced sampling methods such as umbrella sampling and replica-exchange MD were used to provide adequate conformational sampling of the peptides over the HDPE surface. Results revealed substantial discrepancies between the simulation and the experimental ΔG_{ads} values (i.e., differences exceeding 1.0 kcal/mol). To correct the adsorption behavior, an *in-house-developed* interfacial force-field (IFF) was incorporated into the simulation program with IFF parameters adjusted until satisfactory agreement with the experimental data set was achieved. Subsequent studies are planned to apply the tuned IFF to simulate the adsorption behavior of lysozyme and ribonuclease A proteins to HDPE, for which synergistically matched experimental studies have also been conducted to validate the developed method for protein-adsorption simulations.

3:20pm **BI+MG-WeA4 Degradable Silica Nanoshells for Ultrasonic Imaging and Therapy**, ***Alexander Liberman***, *C. Barback, R. Viveros, S.L. Blair, D. Vera, L. Ellies, R. Mattrey, W. Trogler, A.C. Kummel*, University of California at San Diego

As a safe alternative to intrasurgical guidewires and implantable radioactive seeds, gas-filled hollow Fe-doped silica particles have been developed, which can be used for ultrasound-guided surgery for multiple foci. The function of the Fe doping is to render the silica shells biodegradable. The particles are synthesized through a sol-gel method on a polystyrene template, and calcined to create hollow, rigid nanoshells. The Fe-doped silica shell is derived from tetramethyl orthosilicate (TMOS) and iron ethoxide, which forms a rigid, nanoporous shell upon calcination. The nanoshells are filled with perfluoropentane (PFP) vapor or liquid. The flourous phase is contained within the porous shell due to its extremely low solubility in water. *In vitro* studies have shown that continuous particle imaging time is up to approximately three hours non-stop. *In vivo* particle injection longevity studies have been performed in tumor bearing mouse models show signal presence with color Doppler imaging up to ten days post injection. To study biodistribution, nanoshells were functionalized with DTPA and radiolabeled with Indium-111 and then imaged by gamma scintigraphy over 72 hours. Scintigraphic imaging and gamma counting confirm that particles undergoing IV delivery to tumor bearing mice will passively accumulate in the tumors which may allow for tumor detection and therapuetic applications. Additionally, long term biodistribution studies in mice have shown a steady decrease in silicon content over the course of 10 weeks by inductively coupled plasma optical emmision spectroscopy (ICP-OES).

These silica shells break under acoustic excitation to release uncovered gas pockets which increase acoustic energy absorption and reduce acoustic cavitation threshold locally. Therefore they may also be employed as a sensitizing agent in high intensity focused ultrasound (HIFU) therapy. Traditional ultrasound agents which can be used as a HIFU senstizing agent pose several potential drawbacks such as poor *in vivo* persistence (minutes) and high risk during continuous perfusion. Preliminary *in vivo* HIFU ablation studies show that very few particles are needed in order to develop a sensitizing effect to HIFU thereby substantially reduce the amount of HIFU exposure necessary to achieve an ablative effect. It was found that nanoshells systemically adminstered to breast tumor bearing mice could be cavitated by HIFU 24 hours after administration. This mechanical cavitation caused liquification within the focal volume of the HIFU which contained the nanoshells within seconds of the HIFU application. This may potentially allow for a larger area to be ablated in less time with less power.

4:20pm **BI+MG-WeA7 An Encapsulation Technique for Adenovirus to Enhance Viral Gene Therapy**, ***Natalie Mendez***, *V. Herrera, L. Zhang, F. Hedjran, W. Trogler, S.L. Blair, A.C. Kummel*, University of California at San Diego

Oncolytic viruses (OVs) constitute a promising class of cancer therapeutics which exploit validated genetic pathways known to be deregulated in many cancers. To overcome an immune response and to enhance its potential use to treat primary and metastatic tumors, a method for liposomal encapsulation of adenovirus has been developed. The encapsulation of adenovirus in anionic 140-180nm diameter PEG containing non-toxic liposomes has been prepared by self-assembly of lecithin around the viral capsid. The encapsulated viruses retain their ability to infect cancer cells. Furthermore, an immunoprecipitation (IP) technique has shown to be a fast and effective method to extract non-encapsulated viruses and homogenize the liposomes remaining in solution. 76% of adenovirus plaque forming units were encapsulated and retained infectivity after IP processing. Additionally, encapsulated viruses have shown enhanced transfection efficiency up to 4X higher compared to non-encapsulated Ads. Extracting non-encapsulated viruses from solution may prevent an adverse in vivo immune response and may enhance treatment for multiple administrations.

4:40pm **BI+MG-WeA8 Sequential and Competitive Adsorption of Peptides at Pendant PEO Layers**, *X.W. Wu, M.R. Ryder, J.M. McGuire,* ***Karl Schilke***, Oregon State University

A more quantitative understanding of peptide entrapment and elution from otherwise protein-repellent polyethylene oxide (PEO) brush layers will provide direction for development of new strategies for drug storage and delivery. Here we describe criteria for peptide integration and structural change within the PEO brush, and discuss the reversibility of peptide entrapment with changing solvent conditions. For this purpose, three cationic peptides were used: the arginine-rich amphiphilic peptide WLBU2, the chemically identical but scrambled peptide S-WLBU2, and the non-amphiphilic homopolymer poly-*L*-arginine (PLR). Circular dichroism (CD) was used to record the adsorption and conformational changes of WLBU2 and S-WLBU2, and polyarginine peptides at PEO-coated silica nanoparticles. UV spectroscopy and a quartz crystal microbalance with dissipation monitoring (QCM-D) were used to quantify changes in the extent of peptide elution. Peptide conformation was controlled between disordered and α-helical forms by varying the concentration of perchlorate ion. We show an initially more ordered (α-helical) structure promotes peptide adsorption into the PEO layer. Peptide interaction with the PEO chains resulted in entrapment and conformational change that was irreversible to elution with changing solution conditions in the case of the amphiphilic peptide. In contrast, the adsorption and conformational change of the non-amphiphilic peptide was reversible. We also evaluated the effects of peptide surface density on the conformational changes caused by peptide-peptide interactions, and using CD, QCM-D, and UV spectroscopy, showed that these phenomena substantially affect the rate and extent of peptide elution from PEO brush layers. Specifically, for amphiphilic peptides at sufficiently high surface density, peptide-peptide interactions result in conformational changes which compromise their resistance to elution. In contrast, elution of a non-amphiphilic peptide is substantially independent of its surface density, presumably due to the absence of peptide-peptide interactions.

The sequential and competitive adsorption behavior of WLBU2, S-WLBU2 and PLR at pendant PEO layers was studied by optical waveguide lightmode spectroscopy (OWLS), time-of-flight secondary ion mass spectrometry (TOF-SIMS), CD and UV spectroscopy. Results strongly indicate that amphiphilic peptides are able to displace non-amphiphilic peptides that are adsorbed in PEO layers, while non-amphiphilic peptides cannot displace amphiphilic ones. In summary, peptides of high amphiphilicity are expected to dominate the competitive adsorption with less amphiphilic peptides in PEO layers.

5:00pm **BI+MG-WeA9 Moulding Cells and Materials in High Throughput**, ***Clemens van Blitterswijk***, *R. Truckenmuller, L. Moroni, N. Rivron, P. Habibovic, J. De Boer*, Maastricht University, The Netherlands **INVITED**

The interaction of cells and materials at their interface is crucial for the performance of devices that are applied in regenerative medicine.In general the approach to optimize interaction is characterized by a mechanistic low throughput research cycle where researchers try to move forward by improving performance based on fundamental insights and related small volume in vitro/in vivo experiments. Although this approach has successes it has its disadvantages. First as the field of regenerative medicine is young we currently lack fundamental insights into many of aspects that are relevant to our field.Second, the research cycle is slow, so if our experiments do not give the anticipated results we may lose several years.Third,the conventional approach only allows us to test a maximum of ca.10 experimental conditions in one cycle forces us to leave out many other possibly equally interesting, opportunities.

In our lab we are convinced on how influential surface geometry of material can be on cell behavior and in vivo response by recently inducing prominent bone formation in muscle tissue in large animals by modulating the biomaterial surface in the submicrometer range.The effects of these instructive materials are equivalent to the use of growth factors while no biological agents or cultivated cells were applied.As we have no complete insight in the underlying mechanism,a conventional low throughput mechanistic approach does not seem the method of choice for further optimizing this performance and applying it to other tissue types.

Therefore,we developed multiwell screening systems that allow us to test a selection of thousands of surfaces from a truly designed high throughput library of 150 million different surface features in a single run. We have shown that this method allows us to modify cell shape and function in a remarkable way,both as far as cell attachment,proliferation and differentiation are concerned.As the above topochip platform is focused on 2D single cell performance and actual tissues are 3D and multicellular we have developed alternative platforms that allow us the build 3D mesoscale complex tissues in the thousands,while we have also generated so called 2,5 D muliwell systems that present convex surface features. Applying such systems allowed us to demonstrate that the mechanism of function follows form not only holds for individual cells but equally for millimeter scale cell aggregates.We are currently applying these technology platforms to create deeper insights in formation of tissues for regenerative medicine by introducing very early(embryonic)tissues in these systems while actively collaborating with developmental cell biologists.

6:00pm **BI+MG-WeA12 The Influence of Structural Array of Polymorphic hIAPP fibrils to its Mechanical Properties**, ***HyunJoon Chang***, *M. Lee,* Korea University, Republic of Korea, *G. Yoon,* Boston University, *S. Na*, Korea University, Republic of Korea

Amyloid proteins are misfolded, denatured proteins that are responsible for causing several degenerative and neuro-degenerative diseases, such as type II diabetes, Alzheimer's disease, Huntington's disease, and so on. Determining the mechanical stability of these amyloids is crucial for understanding the disease mechanism, which will allow us to provide guidance in treatment. Furthermore, many research groups also recognized amyloid proteins as a functional biological materials that can be used in nano sensor, bacterial biofilms, coatings, etc. There have been many *in vitro* studies to determine the material characteristics via force spectroscopy methods, Atomic Force Microscopy and Optical Tweezers to exemplify. However, computational methods (e.g. Molecular Dynamics (MD) and Elastic Network Model) not only reveal the mechanical properties, but also provide a more in-depth information on the amyloids by visualizing the conformation. In this study, we have discovered the material properties of four different polymorphic structures of Human Islet Amyloid Polypeptide (hIAPP) by using MD simulations under tensile Steered Molecular Dynamics (SMD) conditions. Also, from our results, we have observed how these mechanical properties may differ in respect of their structural formation. This study will help us to take a step forward for treating degenerative disease and also establish a template for the functional biological materials.

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