SYMPOSIUM BM04

Biomaterials for Regenerative Engineering
November 27 - November 29, 2018

Symposium Organizers
Josephine Allen, University of Florida
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* Invited Paper

SESSION BM04.01: Biomaterials for Regeneration of Tissues I
Session Chairs: Guillermo Ameer and Gulden Camci-Unal
Tuesday Morning, November 27, 2018
Sheraton, 2nd Floor, Independence West

8:00 AM *BM04.01.01
Bioresorbable Electronic Materials for Wireless Neuroregenerative Therapy John A. Rogers; Northwestern University, Evanston, Illinois, United States.

Peripheral nerve injuries commonly result in lifelong disability. Even the most advanced surgical procedures and pharmaceutical treatments have limited ability to improve clinical outcomes. Intraoperative electrical stimulation performed at the site of nerve repair is a well-established treatment that can accelerate and improve overall rates of functional recovery. Clinical utilization of electrical stimulation has, however, been limited to the intraoperative period, wherein injured tissue is physically accessible. This talk describes bioresorbable electronic materials and devices that allow for non-pharmacologic neuroregenerative therapy via prolonged post-operative electrical stimulation throughout the healing process, enabling substantially improved outcomes in nerve regeneration and functional recovery compared to the existing intraoperative mode. An essential characteristic of these implantable systems is that they undergo complete dissolution and elimination from the body via natural biochemical processes over timescales matched to operational requirements and without adverse biological effects. The result thereby eliminates the need for secondary surgical extraction and associated risks to the patient and to site of the nerve repair. This type of bioresorbable technology represents a new vehicle for the delivery of non-pharmacologic bioelectric and neuroregenerative therapies in a variety of clinical settings, and a significant paradigm shift in the treatment of critical nerve injuries with limited potential for sensorimotor recovery.

8:30 AM *BM04.01.02
Elastomeric Polymers for Microfabrication of Organs-on-a-Chip Milica Radisic; Univ of Toronto, Toronto, Ontario, Canada.

Recent advances in human pluripotent stem cell (hPSC) biology enable derivation of essentially any cell type in the human body. However, limitations related to cell maturation, vascularization, cellular fidelity and inter-organ communication still remain.
Here, biological wire (Biowire) technology will be described, developed to specifically enhance maturation levels of hPSC based cardiac tissues, by controlling tissue geometry and electrical field stimulation regime (Nunes et al Nature Methods 2013). We will describe new applications of the Biowire technology in engineering a specifically atrial and specifically ventricular cardiac tissues, safety testing of small molecule kinase inhibitors, potential new cancer drugs, and modelling of left ventricular hypertrophy using patient derived cells.

For probing of more complex physiological questions, dependent on the flow of culture media or blood, incorporation of vasculature is required, most commonly performed in organ-on-a-chip devices. Current organ-on-a-chip devices are limited by the presence of non-physiological materials such as glass and drug-absorbing PDMS as well as the necessity for specialized equipment such as vacuum lines and fluid pumps that inherently limit their throughput. An overview of two new technologies, AngioChip (Zhang et al Nature Materials 2016) and inVADE (Lai et al Advanced Functional Materials 2017) will be presented, that overcome the noted limitations and enable engineering of vascularized liver, vascularized heart tissues and studies of cancer metastasis. These platforms enable facile operation and imaging in a set-up resembling a 96-well plate. Using polymer engineering, we were able to marry two seemingly opposing criteria in these platforms, permeability and mechanical stability, to engineer vasculature suitable for biological discovery and direct surgical anastomosis to the host vasculature.
Finally, to enable minimally invasive delivery of engineered tissues into the body, a new shape-memory scaffold was developed that enables delivery of fully functional tissues on the heart, liver and aorta through a keyhole surgery (Montgomery et al Nature Materials 2017).

9:00 AM BM04.01.03
Acellular PCL Scaffolds Laden with Fibroblast/Endothelial Cell-Derived Extracellular Matrix for Bone Regeneration Radoslaw Junka and Xiaojun Yu; Stevens Institute of Technology, Hoboken, New Jersey, United States.

Biological scaffolds derived from decellularized tissues function as tissue remodeling templates during bone regeneration. This isolated extracellular matrix (ECM) provides a structural framework that regulates adherence, migration, proliferation, and differentiation of bone residing cells and those in surrounding tissues. Nonetheless, decellularization protocols, like the ones used in isolation of demineralized bone matrix (DBM), require use of acids and other harsh chemicals that render osteoinductive proteins in DBM denatured. Also, lack of vascular cues in DBM presents another impediment to bone healing, and results in non-unions from poor vascularization of the regenerating tissue. To address these limitations, we used tissue engineering approach and tested the regenerative capacity of decellularized ECMs derived from sequential cultures of fibroblasts and endothelial cells grown on polycaprolactone (PCL) fibers. We hypothesized that this vascular ECM would enhance osteoblast proliferation, differentiation, and matrix deposition in vitro. The bottom-up strategy eliminated competition between cell types with varying proliferation rates and allowed for ECM remodeling. ECMs from decellularized cultures were evaluated via methylene and Coomasie blue stains, and their protein and DNA content was quantified. Staining also revealed that endothelial cells grown on fibroblast ECM (Fibro/Endo) form networks resembling capillaries. These structures stained positively for endothelial markers CD31 and vWF. Analysis of SEM images indicated changes in morphology and preferential attachment of endothelial cells to PCL fibers laden with fibroblast ECM. Osteoblasts grown on this Fibro/Endo ECM yielded higher proliferation rates at each time point during 28 day culture. Significantly higher ALP activity in these cultures suggest better capacity of osteoblasts to form bone tissue and higher degree of differentiation. The color area and intensity of Alizarin Red staining of Fibro/Endo cultures revealed uniform and greater calcium deposits than in cultures with only single type of ECM. Relative expression of osteoclacein and osteopontin between culture conditions was compared via immunostaining. Successive culture/decellularization cycles enriched the ECM product, which in turn significantly enhanced proliferation and differentiation of osteoblasts in vitro. Thus, addition of vascular ECM cues to biological scaffolds might lead to improved bone healing rate in vivo. The use of this hybrid ECM can expanded to other combinations with additional cell types, which might further improve regeneration of tissues.

Acknowledgements:
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9:15 AM BM04.01.04
Influence of Cerium Oxide Nanoparticles on the Properties of Gelatin-Alginate Scaffold for Bone Tissue Engineering Shiv D. Purohit[1], Rakesh Bhaskar[2], Hemant Singh[1], Indu Yadav[1], Mukesh K. Gupta[2] and Narayan C. Mishra[1]; [1]Polymer and Process Engineering, Indian Institute of Technology Roorkee, Roorkee, India; [2]Biotechnology and Medical Engineering, National Institute of Technology Rourkela, Rourkela, India.

It has been observed that polymeric scaffolds loaded with cerium oxide nanoparticles (CeONP) hold great potential for tissue engineering applications. In this study, nanocomposite scaffolds (NCS) have been fabricated by freeze drying of aqueous mixture of the CeONP, gelatin and alginate, with a goal of obtaining CeONP incorporated porous biocompatible scaffolds for bone tissue engineering applications. Further, influence of varying concentration of CeONP, on the scaffold properties, was evaluated in terms of mechanical, biodegradation, cell attachment and cell proliferation properties of the scaffold. Field emission scanning electron microscopy images of the NCS revealed presence of interconnected pores. The NCS was highly porous with porosity ranging from 82-89%. The CeONP covered the surface of the composite matrix and made the surface of the NCS rougher. Compressive strength of the NCS was found to be significantly higher than the gelatin/alginate scaffolds which are not having any CeONP. This may be due to the CeONP present in the NCS. High % swelling (~640%) of the NCS indicates its hydrophilicity. Slow biodegradation (~26% in 30 days) indicates its suitability for bone regeneration. In vitro cell culture studies, by seeding MG-63 osteoblast-like cells over NCS and performing cell attachment studies, MTT assay, Environmental Scanning Electron Microscopy of cell-scaffold construct and Giemsa staining, showed an enhancement in cell attachment, proliferation and adhesion as compared to the gelatin/alginate scaffold which are not having any CeONP: this indicates the influence of the CeONP of such enhancement in the scaffold properties which, in turn, can enhance bone regeneration process ultimately. Thus, it could be stated that the incorporation of CeONP to gelatin-alginate, or in other words, the CeONP incorporated composite scaffold has vital importance for applications in bone tissue-engineering in future regenerative therapies.

9:30 AM OPEN DISCUSSION

9:45 AM BM04.01.06
***In Vivo* Bioresorbability and Tissue Reaction of Hydroxyapatite/Collagen– (3-Glycidoxypropyl)Trimethoxysilane Injectable Bone Paste** Taira Sato[1], Yuki Shirosaki[2], Sho Oshima[3, 4], Yoshihisa Koyama[3], Mamoru Aizawa[1] and Masanori Kikuchi[3]; [1]Department of Applied Chemistry, Meiji University, Kawasaki, Japan; [2]Department of Materials Science, Kyushu Institute of Technology, Kitakyushu, Japan; [3]National Institute for Materials Science, Tsukuba, Japan; [4]Major in Industrial Science, Ibaraki University, Hitachi, Japan.

Injectable self-setting bone pastes are a user-friendly bone void filler in comparison to dense, porous and granular ones, because pastes are applied for minimal invasive surgeries and are shaped easily to fit bone defects. However, surgeons desire biodegradable bone paste strongly because present bone pastes are very low biodegradable and brittle and a risk to be a cause of secondary bone fracture. Presently available biodegradable bone void fillers in Japan are β-tricalcium phosphates, carbonated apatite and hydroxyapatite/collagen bone-like nanocomposite (HAp/Col). The HAp/Col demonstrates good viscoelasticity and excellent bioresorbability with bone formation ability by incorporating into bone remodeling process. We focused on the HAp/Col as a base material for novel injectable bone cement. In the previous report, the HAp/Col biodegradable self-setting pastes were prepared by a mixing of the HAp/Col powder and aqueous solution of (3-glycidoxypropyl)trimethoxysilane (GPTMS), which is a setting agent by cross-linking of collagen and forming siloxane network by self-condensation. The HAp/Col-GPTMS bone pastes implanted into porcine tibia were resorbed and replaced by newly formed bone within 12 weeks. In this study, the pastes were implanted in the rat tibia and the biological tissue reaction and the absorption behavior of the pastes up to 4 weeks after transplantation were investigated in detail.
The HAp/Col (80/20 in mass ratio) powder at 100 μm or less in particle size was prepared by ball-milling of the HAp/Col compact prepared by a uniaxial pressing after synthesis by the simultaneous titration method. Aqueous solutions of GPTMS as 1.0 and 10 % in volume were prepared by mixing of GPTMS in distilled water followed by 1-hour hydrolyzation of GPTMS. They were mixed at the powder/liquid ratio of 1.00 g/cm^3, optimal for the anti-washout property, for 3 min and molded into cylindrical shape with a 2.0 mm in diameter and 2.0 mm in height. The shaped pastes were then incubated for 72 h to be hardened. The hardened pastes were then implanted in 2.0-mm hole defect of proximal SD rat tibia (male, 8 weeks old). At 1, 2 and 4 weeks after the implantation, bioresorption behavior and biological tissue responses of the pastes were investigated by the X-ray μ-computed tomography (μ-CT) and histological observations. All animal tests were authorized by NIMS animal committee (49-2016-3).
The μ-CT observations demonstrated the residual volume of the pastes tended to decrease with the implantation period. The residual volume of the paste with 1.0-% GPTMS significantly decreased between 2 and 4 weeks that is considered to be an osteoclastic resorption. Although no drastically decrease was observed for the paste with 10-% GPTMS, it is expected to be substituted completely with newly formed bone deduced from the results of pig test. In addition, the results of histological observations will be presented on a podium.

10:00 AM BREAK

10:30 AM *BM04.01.07

Bioactive Microrods for the Attenuation of Chronic Cardiac Fibrosis Long V. Le[1], Priya Mohindra[1], Qizhi Fang[2], Rich Sievers[2], Michael Mkrtschjan[4], Brenda Russell[5], Randall J. Lee[2] and Tejal Desai[3, 1]; [1]UC Berkeley-UCSF Graduate Group in Bioengineering, Berkeley, California, United States; [2]Department of Medicine, University of California, San Francisco, San Francisco, California, United States; [3]UCSF Bioengineering and Therapeutic Sciences, San Francisco, California, United States; [4]Department of Bioengineering, University of Illinois at Chicago, Chicago, Illinois, United States; [5]Department of Physiology and Biophysics, University of Illinois at Chicago, Chicago, Illinois, United States.

Coronary artery disease is the leading cause of death in the United States, with over 700,000 myocardial infarctions (MI) occurring each year. While effective strategies have been developed to reduce mortality rates from the acute event, there remain significant challenges to preventing the formation of fibrous scar tissue, which leads to reduced cardiac function and eventual heart failure. Here, we demonstrate the fabrication of hyaluronic acid (HA)-based microrods with tunable size, shape, and stiffness for the attenuation of chronic cardiac fibrosis. These microrods modify the mechanical properties of the tissue microenvironment and modulate the fibrotic phenotype through mechanotransduction pathways. Additionally, HA is a naturally occurring material that is bioresorbable and exhibits several wound healing properties, making it a promising material for this microtopography-based approach. We show that fibroblasts closely interact with these microrods *in vitro and in vivo*, leading to dramatic changes in proliferation, collagen expression and fibrotic phenotype. NIH-3T3s and primary NVRFs attached to HA microrods and often conform to the rod geometry. When grown in the presence of HA microrods, fibroblasts have reduced expression of collagen I and aSMA, suggesting that HA microrods may be able to attenuate the conversion of fibroblasts to the active myofibroblast that is responsible for the overproduction of scar tissue. When injected into the myocardium of a rat infarct model, HA microrods improved cardiac function at 6 weeks post infarct. We have previously shown that PEG-based microrods are able to decrease the loss of cardiac function caused by MI, increasing the % change in EF from -11% to -3%. Impressively, HA-based microrods are able to increase EF by nearly +10%, suggesting that the rods are not only able to reduce deterioration of function, but restore function to the injured heart as well. This may be due to HA being bioresorbable and able to elicit additional biochemical effects through HA signaling pathways as elucidated on our studies. We also show demonstrate that the biochemical conjugation of the microrods with angiogenic peptides can further enhance the ability of this material to promote tissue regeneration. Such materials based approaches can be used in a variety of disease settings including vascular and musculoskeletal, opening up new therapeutic approaches for tissue regeneration.

11:00 AM *BM04.01.08

Platform Technologies for Engineering Functional Composite Tissues Warren Grayson; Johns Hopkins University, Baltimore, Maryland, United States.

Regenerative Engineering strategies have unprecedented potential to restore function to large musculoskeletal injuries. The talk will be used to describe methods developed together with collaborators to treat critical-sized segmental bone defects and volumetric muscle loss. Our lab has developed promising 3D-printing scaffold technologies that combine poly-ε-caprolactone (PCL) with decellularized bone (DCB) matrix to make composite PCL-DCB scaffolds. We have demonstrated the osteoinductive properties of PCL-DCB scaffolds by assessing the *de novo* bone formation when seeded with adipose-derived stromal/stem cells and stromal vascular fraction (SVF) and implanted in murine critical-sized cranial defects. Murine models of critical-sized bone defects are also useful in understanding the role of vascularization in bone regeneration. We have recently demonstrated its use in the application of a novel multimodality imaging platform for acquiring *in vivo* images of microvascular architecture, microvascular blood flow and tracer/cell tracking via intrinsic optical signaling, laser speckle contrast and fluorescence imaging. We are currently using these techniques to explore the impact of rapid, early-stage revascularization on bone healing. In considering translation to humans, however, the complex loading regimens in the craniofacial skeleton make the design of clinically applicable scaffolds inherently more complex than those used to treat murine calvariae. To address this, we are developing algorithms for designing scaffolds with heterogenous porous structures tailored to the expected physiologic loads.
To treat volumetric muscle loss, our team is developing a novel biomaterial-based approach to overcome scarring and induce regeneration of densely vascularized muscle in non-healing, skeletal muscle defects. In particular, we have demonstrated the capacity of novel electrospun fibrin hydrogel scaffolds seeded with myoblasts to regenerate the structure and function of damaged muscle. Myoblast-seeded scaffolds enabled remarkable muscle regeneration with high myofiber and vascular densities after 2 and 4 weeks, mimicking that of native skeletal muscle (while acellular scaffolds lacked muscle regeneration). Both myoblast-seeded and acellular scaffolds fully recovered muscle contractile function to uninjured values after 2 and 4 weeks. In ongoing studies, we are evaluating the potential for tuning the myogenic impact by varying the mechanical properties of the scaffolds. I will also describe our ongoing work on modulating the biochemical characteristics of the scaffold to stimulate neural infiltration and the formation of functional neuromuscular junctions within regenerated skeletal muscle tissues.

11:30 AM BM04.01.09

Tissue Origami for Template-Guided Mineralization Gulden Camci-Unal; University of Massachusetts Lowell, Lowell, Massachusetts, United States.

Due to disease, degeneration, trauma, and aging, bone loss occurs in the body. Although there have been remarkable improvements in development of functional bone scaffolds, it remains difficult to fabricate porous and biocompatible constructs in physiologically relevant sizes (cm-scale). Herein we developed biomineralized origami-inspired paper scaffolds in three-dimensions (3D). To our knowledge, this work is the first demonstration that paper can be used as a 3D construct to induce template-guided mineralization by osteoblasts.
In this work, we used the principles of origami to fabricate free-standing paper scaffolds in cm-scale. Because paper is an extremely flexible material that can easily be cut, creased, and folded to form 3D structures, the scaffolds were easily fabricated in a variety of different geometries. This feature can potentially be useful in generation of constructs for patient-specific applications especially for patients who have defects of irregular sizes and shapes. After sterilizing the constructs, they were seeded with osteoblasts in a collagen matrix. The samples were cultured up to 21 days and mineralization was evaluated using various assays including colorimetric assays, immunocytochemistry, high-resolution imaging (SEM), and micro-computed tomography (micro-CT). We also performed *in vivo* subcutaneous implantation experiments in a rat model.
In this project, we generated paper scaffolds in different shapes, sizes, and configurations (mm-cm scale). Due to its porous structure, paper allowed for transport of oxygen and nutrients across its thickness. Paper scaffolds supported a homogenous distribution of cells within their 3D structures. In our experiments, proliferation of osteoblasts increased until day three and then decreased. Hydroxyapatite content of the samples indicated that there was a progressive increase in the amount of hydroxyapatite in the paper scaffolds over 21 day of culture period. We used SEM to visualize the deposition of mineral clusters, and EDAX to calculate the ratio of calcium to phosphate. Our *in vivo* experiments demonstrated that paper scaffolds did not cause inflammation. The paper implants integrated with the existing tissue strongly and rapidly vascularized.
To sum up, we have shown that origami-inspired tissue engineering is useful for template-guided mineralization. We obtained partially mineralized scaffolds in various 3D geometries. The osteoblasts deposited calcium phosphate in these scaffolds and induced template-guided mineralization. Our approach used paper, a readily available material, as the cell culture scaffold. Paper has great potential to tackle the limitations of traditional scaffolds including cost, availability, accessibility, porosity, flexibility, and ease of fabrication. In the future, paper-based scaffolds could potentially guide and accelerate bone repair using patient specific cells.

11:45 AM BM04.01.10
Cryogenically Electrospun Fibrous Sponge Scaffolds as Stromal Extracellular Matrix for Salivary Gland Regeneration Pujhitha Ramesh[1], Natalya Tokranova[1], L.P. Madhubhani Hemachandra[1], Deirdre Nelson[2], Yubing Xie[1], Susan Sharfstein[1], Melinda Larsen[2] and James Castracane[1]; [1]Colleges of Nanoscale Science and Engineering, SUNY Polytechnic Institute, Albany, New York, United States; [2]Department of Biological Sciences, University at Albany, State University of New York, Albany, New York, United States.

Extracellular matrix (ECM) topography, composition, and stiffness vary across different tissues in the human body. Scaffold-based regenerative strategies must emulate native ECM of the region of interest and be conducive to cell function and differentiation. Healthy soft-tissue ECM has low kPa range of stiffness. Images of decellularized soft tissue ECM (dstECM) have shown that the matrix has little backbone material, a fibrous backbone thickness of ~ 1 µm and pore sizes of 10-30 µm. Currently, ECM-mimicking scaffolds of interest are nanofiber mats, sponges, hydrogels and nanofiber-hydrogel composites. While nanofiber mats have fibrous topography, they have impenetrable pores and fail to mimic either the 3D topography or stiffness of soft tissue ECM. Hydrogels allow tunable stiffness in the kPa range for soft tissue scaffolds, however, they lack an insoluble backbone to mechanically support cells. Existing hybrid nanofiber-hydrogel scaffolds and sponges do not adequately represent the topography seen in dstECM. In this work, we overcame these limitations of current scaffolds by fabricating structures with minimal fibrous backbone and pore size very similar to dstECM using an emerging technique called cryogenic electrospinning (CE).
CE is different from traditional electrospinning in that the collector plate is maintained at less than 0°C. This promotes ice crystal growth between deposited fibers, which can be subsequently lyophilized to produce air pores. This allows scalable 3D growth, increased porosity, reduced scaffold density and kPa-range bulk stiffness. CE has mostly been explored with synthetic polymers dissolved in organic solvents, producing loosely packed nanofibers. In our work, we adopt a greener approach by electrospinning hydrogel materials and ECM proteins to emulate native ECM, with water as the solvent. The topography of our fabricated scaffolds is dramatically different from CE scaffolds reported in literature, due to material and solvent choices and has a 3D fibrous, honeycomb-like backbone, highly interconnected pores that facilitate cell penetration, with pore sizes around 20 µm, and a spongy bulk, strikingly similar to dstECM. We explored collagen, a major structural component of native ECM, and elastin, a pliable protein that will accommodate the push-pull forces of migrating cells, together with alginate and/or polyethylene glycol, hydrogel materials that will act as soft cushions, as biomaterials for CE. We expect these composite fibrous sponge scaffolds to boost growth and optimal function of stromal cells, such as mesenchymal stem cells (MSC), for therapeutic use. MSCs in fibrous sponges may recapitulate a regeneration supportive microenvironment for epithelial cells, leading to improved MSC-based treatments for salivary hypofunction in patients suffering from Sjögren's syndrome, diabetes, or side effects of radiation therapy.

SESSION BM04.02: Biomaterials for Regeneration of Tissues II
Session Chairs: Josephine Allen and Junji Fukuda
Tuesday Afternoon, November 27, 2018
Sheraton, 2nd Floor, Independence West

1:30 PM *BM04.02.01
A Multicellular Tissue Model for Vascularized Osteogenesis Esmaiel Jabbari; University of South Carolina, Columbia, South Carolina, United States.

There is a close correlation between vascularization and bone formation in endochondral ossification as maximum extent of bone formation follows maximum levels of VEGF expression. This suggests that osteogenesis and vascularization are coupled by spatiotemporal regulation of paracrine signaling in which the invading vascular endothelial cells secrete osteogenic morphogens to stimulate cell differentiation and bone formation. The objective of this work was to develop a tissue model to investigate the effect of spatial patterning of mesenchymal stem cells and endothelial progenitor cells and spatiotemporal delivery of osteogenic and vasculogenic morphogens on vascularized osteogenesis in a 3D culture system. To achieve the objective, a 3D co-culture system was developed consisting of a cell-adhesive, degradable polyethylene glycol matrix with gelatin methacrylate-filled microchannels for patterning of human mesenchymal stem cells (MSC) and endothelial progenitor cells (EPC). MSC were encapsulated in the matrix and a combination of MSC+EPC were encapsulated in the microchannels. Self-assembled polyethylene glycol nanogels (PEG NG) were synthesized for timed delivery of BMP-2 and VEGF morphogens. The osteogenic BMP-2 was conjugated to 21-day release NG and added to the MSC-laden matrix. The vasculogenic VEGF was conjugated to 5-day release NG and added to the MSC+EPC-laden microchannels. The 3D tissue model was cultured in osteogenic-vasculogenic medium. At each time point, the tissue model was evaluated for osteogenesis and vasculogenesis by biochemical, mRNA, and protein analysis.
Groups included MSC/EPC patterned tissue model without BMP-2/VEGF (None), with dissolved BMP-2/VEGF, and with BMP2-NG/VEGF-NG. Osteogenic control group was MSC encapsulated in degradable PEG gel with BMP-2 or BMP2-NG. Vasculogenic control group was MSC+EPC encapsulated in gelatin methacrylate with VEGF or VEGF-NG. Based on the results, the extent of vascularized osteogenesis was higher in patterned cellular constructs compared to un-patterned constructs. Further, timed-release of VEGF and BMP-2 in the patterned cellular constructs significantly enhanced the extent of vascularized osteogenesis compared with the direct addition of VEGF and BMP-2. We further discovered that the spatial patterning of MSC and EPC and the spatiotemporal of BMP-2 and VEGF sharply increased the expression of vasculogenic factors bFGF and PDGF and osteogenic factor TGF-β in the tissue constructs. The results suggest that osteogenesis and vasculogenesis are coupled by localized secretion of paracrine signaling factors during bone formation.

2:00 PM BM04.02.02
Dentinogenic Peptide Hydrogels for Pulpal Regeneration Peter Nguyen[1], William Gao[1], Biplab Sarkar[1], Zain Siddiqui[1], Saloni Patel[1], Emi Shimizu[2], Saul Weiner[2] and Vivek Kumar[1]; [1]New Jersey Institute of Technology, Newark, New Jersey, United States; [2]Rutgers, The State University of New Jersey, Newark, New Jersey, United States.

Endodontic root canal therapy is one of the most common clinical procedures to treat infected dental pulp. This non-regenerative treatment removes the dental pulp as well as the vascular and nerve tissues and replaces them with elastomeric composites, such as gutta-percha. The resulting tooth is devitalized and fragile, which may require additional intervention within 3 years. Our self-assembling peptide hydrogels (SAPHs) aim to establish a regenerative solution to this problem by replacing the inert material used in endodontic therapy with materials that promote dental pulp regeneration. In this work, we have used solid-phase peptide synthesis to create dentinogenic self-assembling peptides that form hydrogels under physiological pH and ionic strength. Physical characterization of these hydrogels, using circular dichroism, atomic force microscopy, and scanning electron microscopy, revealed that our peptides formed β sheet nanofibers, which in turn are non-covalently crosslinked to create robust hydrogels. We demonstrated the thixotropic nature of these hydrogels through oscillatory rheometry, and further verified their injectability and *in situ* reassembly into strong hydrogels through *in vivo* subcutaneous injection studies. In both *in vitro* and *in vivo* studies, we were able to show the efficacy of our SAPHs to support and promote the

proliferation of dental pulp stem cells. Additionally, in our *in vivo* studies we observed the infiltration of blood vessels into our hydrogels, suggesting their ability to provide a suitable environment for dental pulp regeneration. The goal of these SAPHs is to provide an improved regenerative alternative to conventional endodontic therapy.

2:15 PM BM04.02.03
3D Self-Foldable Silk-Based Nanoladder Scaffold for Directional Axonal Outgrowth and Functional Regeneration After Spinal Cord Injuries Yimin Huang and Chen Yang; Boston University, Boston, Massachusetts, United States.

Neurons are naturally encompassed by a network in a highly aligned manner. After spinal cord injury (SCI) in the central nerve system (CNS), the organized extracellular matrix (ECM) within the spinal cord is profoundly disrupted, which causes axonal regeneration over injury sites challenging due to lack of orientational guidance. Because of the nature of the spinal cord, bioengineered scaffold in a three-dimensional (3D) format is of great importance for the functional recoveries after injuries. Herein, we report a self-foldable 3D silk-based nanoladder scaffold to mimic the hierarchic structure of the spinal cord with no spatial constraints, comparable mechanical properties, controllable biodegradation rate and sustainable growth factor release. In this study, we fabricated a silk-based nanoladder film with the integration of two scales, micron-meter fibers, and nanoprotrusions. We have proved that micron-meter fibers can provide directional guidance to the regenerated axons, while nanoscale protrusions can serve as mechanical cues to stimulate neurite outgrowth and synapse formation. We further developed the 3D self-foldable nanoladder by coupling the hydrophobic silk nanoladder film with a hydrophilic thermal expanding hydrogel layer. By controlling the biodegradation rate of the 3D nanoladder, a sustainable release of growth factors embedded in the silk film was achieved to trigger the axonal regeneration after injuries. We further applied organotypic spinal cord tissue slices as the ex-vivo injured model to demonstrate an enhanced axonal regeneration and functional connection between two slices placed in a distance of 2-3 mm. In all, we suggest that 3D silk-based nanoladder can serve as a grafting bridge to guide axonal regenerations to desired targets for functional reconnections after SCI.

2:30 PM BREAK

3:00 PM *BM04.02.04
Citrate Chemistry and Biology for Orthopedic Engineering Jian Yang; The Pennsylvania State University, University Park, Pennsylvania, United States.

Leveraging the multifunctional nature of citrate in chemistry and inspired by its important biological roles in human tissues, a class of highly versatile and functional citrate-based biomaterials has been developed. Citric acid, historically known as an intermediate in the Krebs cycle, is a multifunctional, nontoxic, readily available, and inexpensive cornerstone monomer used in the design of citrate-based biomaterials. In addition to the convenient citrate chemistry for the syntheses of a number of versatile polymers that may be elastomeric, mechanically strong and tough, injectable, photocrosslinkable, tissue adhesive, bioimaging/biosensing-enabled, and/or electrically conductive, citric acid also presents inherent anti-bacterial, anti-clotting, angiogenic characteristics and modulates cellular energy levels leading to facilitated stem cell differentiation, which make citrate biomaterials ideal for a number of medical applications. We have attained a comprehensive new understanding of the citrate roles on osteo-phenotype progression and identified a new mechanism pertaining to the metabolic regulation of citrate to elevate cell energy status for bone formation, referred to as citrate metabonegenic regulation. This previously unexplored citrate metabonegenic regulation has allowed us to design new biomaterials to meet the dynamic biological, biochemical, and biophysical needs in bone regeneration. In this presentation, a methodology for the design of biomimetic citrate biomaterials and their applications in regenerative engineering, drug delivery, bioimaging and biosensing will be discussed with a focus on orthopedic engineering.

3:30 PM *BM04.02.05
Multifunctional Biomaterials Containing Amino Acid Based Segments for Tissue Regeneration and Efficient Transfection of Primary Human Cells Andreas Lendlein[1, 2]; [1]Institute of Biomaterial Science, Helmholtz-Zentrum Geesthacht GmbH, Teltow, Germany; [2]Institute of Chemistry, University of Potsdam, Potsdam, Germany.

Modern medicine requires biomaterials combining multiple functions such as degradability, stimuli-responsivity, cell instructivity or carrier capabilities for bioactive molecules. Complex polymer network architectures are a versatile molecular design for integrating different functions in one material system. Such networks often contain physical netpoints for adjusting mechanical properties or implementing stimuli-sensitivities. For this purpose macromolecules are potentially equipped with chain segments being able to exhibit strong physical interactions.
Here amino acid based oligomeric segments are built either from *L*-lysine diissocyanate or from morpholindiones.
Pure oligodepsipeptides, alternating copolymers of an α-amino acid and an α-hydroxy acid, have been selected as a hydrophobic block in segmented polymers in order to achieve strong physical interactions for stabilizing nanoparticles during their formation [1] or for providing high formstability to thermoplastic elastomers. Degradable triblock copolymers having a central oligodepsipeptide block have shown great potential as transfection agent combining high transfection capability with low toxicity [1]. Depsipeptide based multiblock copolymers are suitable for creating soft actuators with excellent performance in shape stability and reversible strain [2].
L-lysine-based oligoureas are incorporated as dangling side chains or crosslinking segments in gelatine based polymer networks. In architectured gelatin-based hydrogels (ArcGel) the local elastic modulus was adjustable independently from the macroscopic compression modulus by the molar ratio of *L*-lysine diissocyanate to freely available amino groups in gelatin. The dynamic alteration of cellular microenvironments accommodating mesenchymal stem cells is studied during degradation. Along with the degradation-related pore growth cell migration and differentiation were followed. The potential of ArcGels for a purely material-induced regeneration was demonstrated in a critical femur defect [3] and a cranial defect [4] in rat models.

References
[1] W. Wang, T. Naolou, N. Ma, Z. Deng, X. Xu, U. Mansfeld, C. Wischke, M. Gossen, A. T. Neffe, A. Lendlein, Biomacromolecules 2017, 18, 3819-3833.
[2] W Yan, T. Rudolph, U. Noechel, O. Gould, M. Behl, K. Kratz, A. Lendlein, Macromolecules Article ASAP, DOI: 10.1021/acs.macromol.8b00322
[3] A.T. Neffe, B. F. Pierce, G. Tronci, N. Ma, E. Pittermann, T. Gebauer, O. Frank, M. Schossig, X. Xu, B. M. Willie, M. Forner, A. Ellinghaus, J. Lienau, G. N. Duda, A. Lendlein, Adv. Mater 2015, 27, 1738-1744.
[4] P. Lohmann, A. Willuweit, A.T. Neffe, S. Geisler, T. Gebauer, S. Beer, H. Coenen, H. Fischer, B. Hermanns-Sachweh, A. Lendlein, N. J. Shah, F. Kiessling, K.-J. Langen, Biomaterials 2017, 113, 158-169.

4:00 PM BM04.02.06
Nanofibrous Scaffolds with Both MSC-Laden Cell Fibers and Growth Factor-Loaded Fibers for Tissue Regeneration Huihua Li, Haoran Sun and Min Wang; Department of Mechanical Engineering, The University of Hong Kong, Hong Kong, Hong Kong.

Tissue engineering scaffolds with biomimetic nanofibrous topography can facilitate the regeneration of tendon and ligament which are fibrous connective tissues composed of specific fibroblasts and aligned nanofibrous extracellular matrix (ECM). As a versatile and effective method to produce nanofibers, electrospinning has been extensively investigated to make nanofibrous scaffolds. Apart from physical cues, biological signaling molecules such as growth factors, are also often used in tissue engineering. Meanwhile, using mesenchymal stem cells (MSCs) for tissue regeneration has many advantages. Among various types of growth factors, basic fibroblast growth factor (bFGF) is a typical biomolecule that upregulate gene expression of tendon and ligament-specific ECM proteins and hence promote the proliferation and differentiation of MSCs toward fibroblasts. Therefore, the combination of incorporating MSCs in a scaffold and having controlled delivery of bFGF in the scaffold should provide a good strategy for tendon/ligament regeneration. MSCs can be encapsulated in fibers via cell electrospinning developed in our group and the fibers can be aligned parallelly in scaffolds to simulate the cell distribution in native tendon/ligament tissue. In this study, multilayered scaffolds consisting of cell fibers and bFGF-containing nanofibrous membranes were fabricated. Bone marrow-derived MSCs were encapsulated in cell fibers while bFGF was incorporated in nanofibrous membranes for its controlled delivery to promote MSC differentiation. Medical grade poly(lactic-co-glycolic acid) (PLGA) was employed for fabricating bFGF-containing fibers in scaffolds. Cell fibers were made from Na-alginate solutions and contained MSCs. The Na-alginate was crosslinked into Ca-alginate using a $CaCl_2$ solution. In in vitro experiments, the constructs with bFGF-containing PLGA nanofibers and MSC-laden cell fibers were cultured for up to 21 days. In day 1, a sodium citrate solution was dripped onto scaffolds to disrupt the crosslinked Ca-alginate cell fibers to release to MSCs. The viability and proliferation of MSCs were studied using LIVE/DEAD assay and MTT assay. On Day 1 and Day 3, the cell viability was all above 90%. MSCs proliferated well during the culture period up to 21 days. The MSCs in scaffolds with encapsulated bFGF proliferated faster than the control group and elongated notably, indicating that MSCs cultured under bFGF release had fibroblasts-like differentiation. The morphology and structure of scaffolds before and after different culture periods were investigated using SEM. Mechanical properties of the constructs were also examined.

4:15 PM BM04.02.07
Growth Factor-Laden Microparticles Incorporated into Polylactic Acid/Apatite Composite Scaffolds for Tooth Regeneration Ali Salifu, John D. Obayemi, Vanessa O. Uzonwanne and Winston O. Soboyejo; Worcester Polytechnic Institute, Worcester, Massachusetts, United States.

Growth factors such as bone morphogenetic protein 2 (BMP2) have been found to stimulate the odontogenic differentiation of mesenchymal stem cells (MSCs) of the dental pulp. This is particularly important for the regeneration of the dentin tissue of the tooth. However, the short half-life and poor distribution of growth factors like BMP2 may present problems with high cost and inconvenience due to the need for repeated dose injections to sustain tissue regeneration and healing. Consequently, strategies that combine slow, sustained release of BMP2 with three dimensional (3D) bioactive scaffolds are important for the differentiation of the MSCs into the odontoblast lineage to accelerate dentinogenesis. Herein, we present the results of in vitro studies of the controlled release of BMP2 from gelatin methacrylate (GelMA) microparticles to human dental pulp stem cells (hDPSCs) growing on 3D polylactic acid (PLA)/apatite composite scaffolds. First, 3D printing is utilized to fabricate PLA scaffolds, which are then surface-coated with biomineralized apatite particles synthesized via a bioinspired mineralization process. This involves alternate immersion of the PLA scaffolds in solutions of calcium nitrate and potassium phosphate dibasic to form a precipitate that is mineralized into apatite at physiological pH. The architectural, microstructural, and mechanical properties of the resulting 3D PLA/apatite composite scaffolds are then determined. Second, GelMA photopolymer is synthesized from porcine gelatin and methacrylic anhydride. After that, BMP2-laden GelMA microparticles are fabricated using an oil-in-water emulsion technique and ultraviolet photocrosslinking. Subsequently, a scaffold/microparticle hybrid construct is produced by the attachment of the BMP2-laden microparticles to the 3D PLA/apatite composite scaffolds. The microstructure of the GelMA microparticles and the release profiles of BMP2 from the microparticles are characterized prior to and following attachment to the PLA/apatite scaffolds and the differences are highlighted. Finally, the effect of the controlled release of BMP2 on hDPSC cells growing on the PLA/apatite scaffolds is investigated. This is done by assessing hDPSC cell proliferation, odontogenic differentiation, and extracellular matrix production and mineralization. The implications of the results for tooth regeneration are then discussed.

SESSION BM04.03: Poster Session I: Biomaterials for Regenerative Engineering
Session Chairs: Josephine Allen, Guillermo Ameer, Gulden Camci-Unal and Junji Fukuda
Tuesday Afternoon, November 27, 2018
8:00 PM - 10:00 PM
Hynes, Level 1, Hall B

BM04.03.01
Mussel-Inspired Hydrogel-Based on Polyphenol Oxidation for Wet Tissue Adhesion and Immune Modulation Su-Hwan Kim, Kyungmin Kim and Nathaniel S Hwang; Seoul National University, Seoul, Korea (the Republic of).

Nature inspired chemistry, and small molecules have led to the development in the field of the material science and biomedical engineering as it exhibited unique physicochemical properties. Recently, polyphenols extracted from green tea have been widely investigated, due to their intrinsic properties such as anti-inflammation and radical scavenging. Interestingly, a 1,2,3-trihydroxyphenyl group in epigallocatechin gallate (EGCG) could mimic mussel inspired chemistry through oxidative reactions, and generate tissue adhesive nature. In this paper, we report a tissue adhesive and immune modulation hydrogel inspired by the mussel chemistry and polyphenol. We conjugated tyramine (HA_T) and EGCG (HA_E) into hyaluronic acid (HA), and the hydrogel (HA_TE) was fabricated by an oxidative reaction using tyrosinase from *Streptomyces avermitillis* (SA_Ty). With strong oxidative nature of EGCG, the HA_TE hydrogel can be fast formed in a few seconds. We compared HA_TE hydrogel with commercial products (cyanoacrylate and fibrin glue) in the aspects of tissue adhesive and sealants. In the lap shear and burst pressure test, HA_TE exhibited the highest tissue adhesiveness regardless of wetness compared to commercial products. When HA_TE was applied as tissue adhesive into mouse wound closure, and it successfully closed wound and recovered damaged tissue. Additionally, due to EGCG naturally possesses anti-inflammation and minimize host recognition, HA_TE hydrogel produced little inflammatory cytokines in vivo that are comparable to PBS group. This demonstrates that polyphenol based hydrogel might provide a robust platform in the field of both material science and translational medicine.

BM04.03.02
Large-Scale Preparation of Hair Follicle Germ (HFG) Using Microfabricated PDMS Spheroid Chips for Hair Regenerative Medicine Chisa Yoshimura[1], Tatsuto Kageyama[1], Keiichiro Kasai[2] and Junji Fukuda[1]; [1]Yokohama National University, Yokohama, Japan; [2]Shonan Beauty Clinic, Tokyo, Japan.

Hair regenerative medicine is a new approach for the treatment of hair loss caused by aging, diseases, injury, and medical treatments. Hair follicle morphogenesis is triggered by reciprocal interactions between hair follicle germ (HFG) epithelial and mesenchymal layers. Recent studies have revealed

that HFGs can be fabricated *in vitro* by integrating two respective aggregates of epithelial and mesenchymal cells. *This approach showed promising results for hair regenerative medicine, but preparing a large number of HFGs remains challenging, particularly considering that hundreds of thousands of HFGs are necessary for a single patient.* In this study, we developed a method for the large-scale preparation of HFGs *in vitro* via the self-organization of cells. *We mixed epithelial and mesenchymal cells in a culture medium and then seeded them onto an oxygen permeable polydimethylsiloxane (PDMS) spheroid chip, which has hemispherical wells with a diameter of 1 mm and a density of 100 wells/cm^2. The cells initially formed a randomly distributed single aggregate, but then were spatially separated from each other and exhibited typical morphological features of a HFG after three days of culture. Interestingly, oxygen supply through the bottom of the spheroid chip was crucial for the spontaneous formation of HFGs and subsequent hair shaft generation.* The generated hair follicles also entered the hair cycle through the rearrangement of follicular stem cells. Unlike previous approaches, this spontaneous HFG formation *in vitro* facilitated the preparation of a large number of cell aggregates (~5000 aggregates/plate). We further optimized the HFG culture conditions for human hair cells. In particular, gene expressions related to hair morphogenesis and generation were significantly increased by increasing fibroblast growth factor (FGF-2) concentration (from 0 to 100 ng/mL). Hair shaft pigmentation was observed by transplantation of HFGs including hair pigment cells. Hair shafts that were generated showed typical morphological features, such as hair cuticles and hair growth cycle. *This simple HFG preparation approach may provide a promising strategy for advancing hair regenerative medicine.*

BM04.03.03

Oxygenating Bioinks for Organ-Like Cell Density Constructs Benjamin Dalisson[2], Huaifa Zhang[3] and Jake Barralet[1]; [1]Division of Orthopaedics, Department of Surgery, Faculty of Medicine, McGill University, Montreal, Quebec, Canada; [2]Faculty of Dentistry, McGill University, Montreal, Quebec, Canada; [3]Mechanical Engineering, University of Ottawa, Ottawa, Ontario, Canada.

In tissue engineering and bioplotting, the major limitation to building large constructs with physiological cell densities is the poor diffusion of oxygen and nutrients. Organs and many tissues have cell densities in the range 1-5x 10^8 cells/ml, yet bioinks can only sustain up 25x10^6 cells/ml. Oxygen concentration in culture medium is one of the major limiting factors for cell survival and its concentration is about 30 times lower than glucose. This limits the tissue models that can be printed. Furthermore, upon implantation cell survival is dependent on revascularization rate and limit the potential applications *in vivo*. Using oxygen releasing microparticles we designed a bioink system capable of prolonging cell survival at high cell density (2x10^8 cells/mL), mimicking physiological organ cell densities.

Oxygen releasing microparticles (OμP) were produced by phase separation method using polycaprolactone (Mw 80000, Aldrich, USA) and calcium peroxide (Aldrich, USA). Particles size measurement was confirmed using field emission scanning electron microscope (FE-SEM, FEI Inspect F-50, USA). The bioink was prepared by mixing 10% (w/v) of OμP in a 1% alginate solution. Oxygen release was measured in 250μL of bioink crosslinked with 0.1M calcium chloride solution and immersed in 1mL PBS at 37°C using an AL300 oxygen sensor (OceanOptics via Gamble Technologies, Canada). Organ-like high density cell culture was performed by seeding a high density (4x10^8 cells/mL) by combining 125μL CHO cells and 125μL of 2% alginate solution alone or containing 20% w/v OμP. The mixture was then extruded through a 20G needle in a 0.1M calcium chloride solution to form 20μL beads. Cells were then cultured for 48h. Cell viability was assessed using MTT.

Scanning electron microscopy revealed that the size of the microparticles ranged from 200μm to 5μm. Oxygen measurements indicated that the dissolved oxygen content of the bioink was kept at 41 ± 5% for 72h. Bioinks with organ-like cell densities without OμP had a cell viability of 27 ± 19% and 13 ± 4.8% at 24 and 48h respectively, whereas with OμP viability was significantly higher at both times; 87 ± 18% and 63 ± 20% at 24 and 48h respectively ($N=9$, $p<0.001$).

Lack of oxygen can be detrimental to cell function and survival. In tissues the diffusion of oxygen around a capillary is reported to be around 200μm. By incorporating OμP to a bioink it was possible to create organ-like density construct (2mm thick) with a high viability. This new approach to bioinks may enable printing of more complex and physiologically relevant tissue models.

BM04.03.04

Application of Ultrasound and DEFINITY® Microbubbles for Drug Delivery *In Vitro* Alina Karki, Emily Giddings, Mercedes Rincon and Junru Wu; University of Vermont, Colchester, Vermont, United States.

Ultrasound assisted by ultrasound contrast agents, microbubbles is one of the promising non-viral approach for the therapeutic application such as drug delivery and gene delivery. Using ultrasound, the rapid oscillation of the gas encapsulated micron- sized bubbles on the nearby cells may generate the shear stress on their cell membranes, and thus, produce nanometer size holes temporarily. The specific drugs could be delivered inside the cells via these holes. This process is called sonoporation. We have performed in- vitro experiments using this technique to deliver siRNA molecules inside the mouse and human T-cells with the help of DEFINITY® microbubbles. We have optimized the ultrasound parameters (like frequency, intensity, duty cycle, time of ultrasound excitation), concentration of DEFINITY® microbubbles and siRNA per sample to provide efficient siRNA drug delivery in human and mice T-cells.

BM04.03.05

Dynamic Modulation of Hydrogels for Mechanical Modulation of Cells in a Reversible Manner Yashoda Chandorkar, Arturo Castro Nava, Tamas Haraszti, Marcel Van Dongen, Jens Koehler, Hang Zhang, Ahmed Mourran, Martin Moeller and Laura De Laporte; DWI – Leibniz-Institute for Interactive Materials e.V., Aachen, Germany.

The extra-cellular matrix (ECM) conveys different biochemical, mechanical and structural cues to cells. These signals are highly orchestrated in space and time. Precise instructions from the ECM dictate cell fate processes, such as proliferation, differentiation and migration. The ECM exerts mechanical forces on cells, which are sensed by cells through different mechanisms, and are translated into biological outcomes. However, these mechanisms are not well understood. One of the main limitations in deciphering this language of forces on cells has been the lack of *in vitro* systems, which can generate forces on cells that mimic natural stresses.

The present-day methods, which attempt to apply such forces on cells, include single cell manipulation techniques that are highly invasive and although very cell-selective, do not mimic natural stresses. Other techniques rely on the use of flexible elastomers, which better replicate natural stretches, but do not provide user-defined cell selectivity. Therefore, it remains a challenge to develop a system for manipulating cells with mechanical forces, which are precisely controlled in space and time domains.

Here we demonstrate a novel hydrogel system, which can reversibly apply precise, user-defined mechanical forces on selected cells in a cell population. Our approach comprises a smart ECM-mimic hydrogel system, which responds to a light trigger. This causes reversible local deformations of the cell growth substrate and leads to the generation of mechanical forces on cells. These forces are transient and can be controlled at a sub cellular and sub-

population scale, in a wide range of time scales (up to ms), with pre-defined directionality.
Such a system for opto-mechanical stimulation of cells is an effective tool for investigating how repeated actuation of a soft hydrogel affects cells. This is experimentally demonstrated in a case study using fibroblast cells to show the proof-of-principle of the concept.
The dynamic hydrogel swelling/shrinking closely replicates the stretches experienced by soft tissues in the body during activities, such as movement, growth etc. We believe that this system bridges the gap between single cell manipulation techniques and cell sheet deformation techniques. This system shows great potential in fields of 'mechano-diseases' and in understanding cell-ECM interactions.

BM04.03.06
Improved Antibacterial Properties of Titanium Implants After Acid Etching and Atomic Layer Deposition Paria Ghannadian, James W. Moxley and Thomas Webster; Northeastern University, Boston, Massachusetts, United States.

Despite the progress tissue engineering has made in the development of improved biomaterials, inhibiting bacterial infection has not been a central focus to date. Infection is a leading cause of implant failures with many agencies (such as the Centers for Disease Control) predicting more deaths from bacteria than all cancers combined by 2050. Gram-negative bacteria are naturally resistant to numerous treatments and are difficult to kill due to their robust and hydrophobic outer lipopolysaccharide membrane which helps to prevent the flow of antibiotics or drugs into the cell. Moreover, due to extensive antibiotic use, gram-positive Staphylococcus aureus has evolved to a methicillin-resistant strain, which can overcome other classes of antibiotic treatments. The development of an implant capable of reducing bacterial growth (without resorting to the use of antibiotics which causes antibiotic resistant bacteria) would be an effective way to improve implant success. Recently, scientists have been investigating novel materials and techniques to meet growing orthopedic tissue engineering needs. The first step in implant infection is bacterial adhesion, which can potentially result in the formation of antibiotic resistant biofilms for some species. Bacterial adhesion, growth, and subsequent biofilm formation on surfaces are particularly resistant towards the body's defense mechanisms and antibiotic treatments, which can cause implant rejection. Multiple substrate properties, including chemical composition, hydrophobicity, and surface roughness, are believed to be of significance in the bacterial attachment process. In this study, multiple titanium samples were etched with different concentrations of nitric acid (10N or 12N) for varying durations (60 or 90 minutes), followed by a consistent and extended heat treatment (400 for one hour) for all samples. As a comparison to these samples, which were modified through conventional acidic etching treatment, another group of titanium samples were prepared by coating with 25 nm of titanium dioxide at 200 for approximately 4 hours through an atomic layer deposition (ALD) technique. To assess the potential effect of both approaches on inhibiting bacterial adhesion, and thus conferring antibacterial properties, samples were cultured with Staphylococcus aureus and colony forming unit (CFU) assays were conducted. ALD treatment, in comparison to conventional acidic etching treatment, demonstrated reduced bacterial density. As such, ALD treatment may pose a promising way to inhibit the growth of infectious bacterial populations, on a vast variety of surfaces and materials, without the need for antibiotics.

BM04.03.07
Bio-Plotting Facial Cartilage Replacements Raymond Oliver[1], Michelle Griffin[2], Peter Butler[2] and Chawisa Deesomboon[1]; [1]School of Design, Northumbria University, Newcastle upon Tyne, United Kingdom; [2]Division of Surgery and Interventional Science, Centre for Nanoscience and Technology, London, United Kingdom.

Several diseases include cancer, skin diseases, inflammatory conditions, trauma and congenital deformalities cause ear and nose defects that require reconstuction. Due to the wide patient population that this affects, nose reconstruction creates a huge social and economic burden. Each year, 1/6000 children are born with a small or missing ear, a condition called microtia. This devastating facial disfigurement causes high physical, social and mental burden for both the child and parent. Current surgical reconstruction involves harvesting tissue from elsewhere in the body, to recreate the cartilage framework of the ear and the nose and then implanting the framework beneath the skin. These techniques cause pain, are limited by tissue availability, can fail and have potential wound-healing complications.
Several synthetic and biological materials have been considered for the reconstruction of the nose and ear but with high levels of infection, unnatural look and feel, they are not considered an acceptable alternative. Synthetic materials are promising candidates to provide the mechanicalproperties and support for the constructs. However, biological materials are useful as they have good biocompatibility and can support tissue formation, which synthetic materials often lack. The incorporation of patient's own cells within the construct can also enhance the biocompatibility of the implant material.
Additive three dimensional bio plotting, a modified form of Fused Deposition Modelling (FDM) has now allowed synthetic and biological biomaterials to be combined to create organ replacements. In addition, to being able to create more complex shapes which better mimic the native tissue, they can be manufactured specific to the patient. Bioplotting also allows the direct printing of cells with the material to create a biocompatible and functional implant.
The work described in this paper represents a new approach to ear and nose reconstruction using 3D-Bio-plotting. We have tested several combinations of biological hydrogels and synthetic polysaccharide composite materials to act as the replacement for the cartilage framework of the ear and the nose.
We describe the concepts being developed from 3D to 4D biofabrication using a high precision Bioplotter robot (Envisiontec) to ensure we are creating accurate patient focused auricular and nasal relacements. The bioplotter has proven capable of printing several materials sequentially in very precise locations with and without cells incorporated in the material. Now, in the second stage of our current programme, we are exploring suitable combinations of synthetic and biological material for nose and ear reconstruction, printing of the patient's own cells within the biological component of the material will be optimised. The ability of the cells to survive, grow and support tissue formation is driven by novel laminar(low shear) flow mixing to ensure maximum stem cell survival.

BM04.03.08
Peptide-Based Polyelectrolyte for Neural Tissue Engineering Wei-Fang W. Su, Chia-Yu Lin, Jia-Shing Yu and Shy-Chyang Luo; National Taiwan Univ, Taipei, Taiwan.

Neural tissue engineering has emerged as a potential technology to cure neural damages. Although various synthetic polymers with good biocompatibility and biodegradability are adopted as candidate materials for scaffolds, most of them require incorporation of biomolecules or conductive materials to promote the growth of long axon. Here we propose a peptide-based polyelectrolyte which is conductive and contains neurotransmitter of glutamic acid. The designed copolymer of poly(γ-benzyl-L-glutamate) and poly(L-glutamic acid) sodium salt ($PBGA^-Na^+$) is electrospun into 3D scaffold with aligned fibers. Neuron-like rat phaeochromocytoma (PC12) cells are cultured on the scaffolds to evaluate cell proliferation and differentiation. The results show with both electrical and biochemical cues, the polyelectrolyte $PBGA^-Na^+$ gives longer axon outgrowth and higher differentiation ratio compared with the neutral copolymer of poly(γ-benzyl-L-glutamate) and poly(L-glutamic acid) (PBGA).

BM04.03.09
Graphene Oxide as a Drug Carrier for Delivery of Zoledronate in Methabolic Bone Disease and Secondary Bone Cancer Treatment Sepideh Tavakoli and Duygu Ege; Bogazici University, Istanbul, Turkey.

In this study, Zoledronic acid (ZOL), a type of nitrogen containing bisphosphonate, was loaded on graphene oxide (GO) particles to increase the particle size of the drug-nano-carrier complex which reduces drug filtration by the kidney and consequently, increases drug circulation time and its tumor uptake. The conjugation between ZOL and GO occurs via π-π stacking and hydrogen bonding interactions, and therefore, the drug may be gradually released from GO in physiological conditions which eliminates the need to apply high doses of the drug. Loading and release profile of ZOL on GO particles was investigated by using UV-Vis spectroscopy. Samples with different concentrations of 0.025-1.25 mg/ml of ZOL were loaded on 0.2 mg/ml GO. UV analysis showed that the maximum loading happens at ZOL to GO ratio of 1:0.2. This loading was obtained when 1 mg/ml of ZOL was initially loaded on 0.2 mg/ml of GO nanoparticles. The drug and drug carrier complexes were characterized using FTIR, AFM, and UV-vis spectroscopy. Cell culture studies were carried out with MCF-7 breast cancer cells for three dosages of ZOL, ZOL-GO and GO. Cell migration was assessed using Bio-Coat cell migration chambers and cell proliferation was investigated by alamarBlue assay. Cell viability was evaluated by staining dead cells with propidium iodide (PI) and live cells with acridine orange (AO). Overall, the characterization results confirm loading of ZOL on GO nanoparticles and cell studies results show that GO conjugated ZOL complexes are promising to reduce MCF-7 breast cancer cells migration, proliferation and viability.

BM04.03.10

Bioinspired Mineral-Organic Bioresorbable Bone Adhesive Alina Kirillova, Cambre N. Kelly, Natalia von Windheim and Ken Gall; Department of Mechanical Engineering and Materials Science, Duke University, Durham, North Carolina, United States.

Bioresorbable bone adhesives have potential to revolutionize the clinical treatment of the human skeletal system, ranging from the fixation and osseointegration of permanent implants to the direct healing and fusion of bones without permanent fixation hardware.[1] With sufficient strength bioresorbable bone adhesives could ultimately become an ideal means for fixing bone fractures instead of conventional plates, nails, pins and screws used today.[2] Despite the evident clinical need, there are currently no bioresorbable bone adhesives in clinical use that can form a bond to bone in a wet environment strong enough to bear clinical loads and sustainable enough to allow fracture healing.[1]
Inspired by the sandcastle worm that creates a protective tubular shell around its body by gluing together sand grains and shell fragments underwater using a proteinaceous adhesive, we introduce a novel mineral-organic bone adhesive (aka Tetranite®) that cures in minutes in an aqueous environment and provides high bone-to-bone adhesive strength. The new bioresorbable material is measured to be more adhesive than both bioresorbable calcium phosphate and poly(methyl methacrylate) bone cements, which are standards of care in the clinic today. Osteointegration and bioresorbability of the bone adhesive are demonstrated over a 52-week period in a critically-sized distal femur defect in rabbits. Based on its unique capabilities, Tetranite is the first in a new class of biomaterials, which may spark innovative clinical treatments and revolutionize procedures in which bone regeneration or fixation is critical for treatment.

[1] Farrar, D. F. Bone adhesives for trauma surgery: A review of challenges and developments. *International Journal of Adhesion and Adhesives* **2012**, *33*, 89-97.
[2] Heiss, C.; Kraus, R.; Schluckebier, D.; Stiller, A.-C.; Wenisch, S.; Schnettler, R. Bone Adhesives in Trauma and Orthopedic Surgery. *European Journal of Trauma* **2006**, *32*, 141-148.
[3] Kirillova, A.; Kelly, C.; Von Windheim, N.; Gall, K. Bioinspired Mineral-Organic Bioresorbable Bone Adhesive. *Advanced Healthcare Materials* **2018**, DOI: 10.1002/adhm.201800467.

BM04.03.11

Silicon-Based Nanoneedles to Guide and Regulate Stem Cell Behaviour Hyejeong Seong[1], Stuart G. Higgins[1, 2, 3], Spencer W. Crowder[1], Julia Sero[2], Jelle Penders[1], Charlotte Lee-Reeves[2], James Armstrong[1] and Molly Stevens[1, 2, 3]; [1]Department of Materials, Imperial College London, London, United Kingdom; [2]Department of Bioengineering, Imperial College London, London, United Kingdom; [3]Institute of Biomedical Engineering, Imperial College London, London, United Kingdom.

In recent years, surfaces with nano/microscale topography have been widely used to control stem cell behaviour. These patterned substrates possess fascinating qualities that render them more valuable than conventional flat surfaces in many bio-applications, such as neuronal differentiation, biosensing, tissue engineering and DNA analysis. Among these, nanopillar and nanoneedle structures have been extensively investigated because they are beneficial in terms of increasing enhancing cell adhesion and growth, and ability to penetrate cells for facilitating drug/biomolecules delivery.
We have co-developed high-aspect ratio, porous silicon nanoneedles made by electrochemical wet etching for *in vitro* and *in vivo* manipulation of cell behaviour[1,2]. These structures are highly biocompatible and can be used to directly interact with the cell membrane, cytoskeleton, and nucleus of primary human cells, generating distinct responses at each of these cellular compartments[3].
Moreover, we have recently developed a new generation of non-porous nanoneedles using a deep reactive ion etching process. Our new system provides high chemical stability in cell culture media, making it suitable for the long-term investigation of stem cell fates and differentiation at a nanoneedle interface. Furthermore, by systematically tuning the sharpness of the nanoneedles, we could precisely probe their effect on cellular mechanotransduction. The structural effect on cell morphology, alignment, and gene-level expression was observed with scanning electron microscopy, immunofluorescence, and real-time polymerase chain reaction. Our findings provide an ideal framework for manipulating and exploiting stem cell behaviour for longer periods, as a means for understanding cell-material interfaces and differentiation capacity of stem cells. Moreover, we used focused ion beam scanning electron microscopy to determine the critical sharpness required to achieve close interaction between the surface and the cell membrane. We expect elucidating the interfaces between nanoneedles and cells to enable new applications in bioengineering, especially in the sensing and monitoring of live cell cultures via 3D-structured electronic devices.

[1] C. Chiappini *et al.*, *ACS Nano* 2015, 9, 5500-5509
[2] C. Chiappini *et al.*, *Nat. Mater.* 2015, 14, 532-539
[3] C. S. Hansel *et al.*, 2018, *In revision*

BM04.03.12

Magnetic Isolation of Exomes Using Fe/Au Nanowires—Towards an Improved Early Detection of Cancer Zohreh Nemati Porshokouh, Daniel Shore, Kelly Makielski, Joseph Um, Rhonda Franklin, Jaime Modiano, Bethanie J. Stadler and Mohammad Reza Zamani Kouhpanji; University of Minnesota, Saint Paul, Minnesota, United States.

Early detection of cancer plays an important role in successful treatment. Therefore, there is an urgent need for more effective and less toxic biomarkers to detect cancer. Cancer cells use exosomes to survive and metastasize to other tissues. Exosomes are small vesicles released to blood by cells, and they can deliver proteomic and genetic information unique to each cell. Therefore, isolating the exosomes secreted by cancer cells can provide us valuable information about the state of a tumor. Since every cell in the body releases exosomes, separating those coming from cancer cells can be a cumbersome task. Current techniques to isolate exosomes are time consuming and costly. Hence, our aim in this study is to use magnetic nanowires (MNWs) to magnetically isolate cancer cells' exosomes in an efficient way through a simple blood biopsy and a magnetic stand.

In this work, we have used Fe/Au segmented MNWs to separate exosomes released by osteosarcoma cancer cells. These MNWs have been functionalized with PEG, and their concentration has been optimized in order to improve their capture and internalization by the cancer cells. We have observed by TEM that most of the MNWs end up inside the lysosomes in cancer cells. Once inside the cells, these MNWs tend to be broken into smaller pieces that can be released inside the exosomes. This way, by using a magnetic stand, we can easily and efficiently separate only the cancer cells' exosomes, since they contain segments of magnetic nanowires.

In addition, our Fe/Au segmented MNWs can also be used as customized radio frequency identification (cRFID) labels. MNWs have magnetic (Fe) and nonmagnetic (Au) segments which resemble barcodes, and their structure can be engineered (e.g., by changing the length of each segment) to produce different cRFID signatures. Hence, distinct nanowires can be attached to different types of cancer cells in order to distinguish between the exosomes derived from each type, thereby further improving the efficacy of our blood biopsies.

BM04.03.13
Integration of Phase-Change Materials with Electrospun Nanofibers for Promoting Neurite Outgrowth Under Controlled Release of Biological Effectors Jiajia Xue and Younan Xia; The Wallace H. Coulter Department of Biomedical Engineering, Georgia Institute of Technology, Atlanta, Georgia, United States.

Electrospun fibrous scaffolds have shown great promise in promoting axon regeneration to improve repair of peripheral nerve defect. Especially, when the fibers are collected as uniaxially aligned arrays, the growth of neurites can be guided and accelerated. Despite the progress, it remains a challenge to place temporally and spatially controlled delivery of biological effectors such as growth factors from the electrospun fibrous scaffold. Such a requirement can be met by integrating electrospun fibers with a controlled release system based upon a stimuli-responsible material. We have developed a temperature-regulated system for the on-demand release of nerve growth factor to promote neurite outgrowth. The system was based upon microparticles fabricated using co-axial electrospray, with the outer solution comprised of a phase-change material (PCM) and the inner solution containing the payloads. When the temperature was kept below the melting point of the PCM, there was no release due to the extremely slow diffusion through a solid matrix. Upon increasing the temperature to slightly pass the melting point, the encapsulated payloads could be readily released from the melted PCM. By leveraging the reversibility of phase transition, the payloads were released in a pulsatile mode through on/off heating cycles. When the PCM microparticles (co-loaded with nerve growth factor and a near-infrared dye) were sandwiched between two layers of electrospun fibers, the nerve growth factor could be released on-demand upon photothermal heating with a near-infrared laser. The nerve growth factor was released with well-preserved bioactivity and stimulated the extension of neurites from spheroids of PC12 cells. By choosing different combinations of PCM, biofactor, and scaffolding material, this controlled release system can be applied to a wide variety of biomedical applications.

BM04.03.14
Biocompatible and Bioadhesive Lectin Conjugated Liposomes as Drug Carriers for the Management of Oral Ulcerative Lesions Sashini Wijetunge; University of Massachusetts Lowell, Lowell, Massachusetts, United States.

Oral ulcerative lesions are a painful side effect of cancer chemo and radiation therapy. The current clinical management of this condition requires multiple classes of drugs administered through topical formulations. However, these formulations need frequent dosing for optimal therapeutic effect which is inconvenient and leads to patient non- compliance. In this study, we prepare wheat germ agglutinin conjugated liposomes (WGA- liposomes) as a bioadhesive nanocarrier that can potentially encapsulate multiple classes of drugs, show fast binding to oral cells and localized sustained drug delivery. Fluorescence studies demonstrated that WGA- liposomes can rapidly bind to cells (within 1 min) and have a significantly higher binding (p<0.05) compared to the original liposomes. Studies with model drug amoxicillin encapsulated WGA- liposomes revealed sustained *in vitro* drug release over several days and potent antimicrobial activity against *Streptococcus mutans* in an oral cell- bacteria co- culture system. Fluorescence studies on liposome release showed that the WGA- liposomes stayed in oral cells for 48h after which it was cleared from cells. A significant reduction in oral cell damage in bacteria pre- infected oral cells after treatment with amoxicillin encapsulated WGA- liposomes compared to the untreated cells was observed through cell viability studies. Cell viability studies also showed that oral cells are not significantly damaged in the presence of WGA- liposomes indicating a potentially biocompatible nanocarrier. These results point to the great potential of WGA- liposomes as a drug delivery vehicle to effectively treat oral ulcerative lesions with reduced dosing frequency.

BM04.03.15
Lego Scaffold—3D Platforms for Reprogramming Cellular Behaviour Fabrizio A. Pennacchio, Angela Langella, Giulia Iaccarino, Fabio Caliendo, Velia Siciliano and Francesca Santoro; Istituto Italiano di Tecnologia, Napoli, Italy.

Reprogramming cellular functions through the design, fabrication and use of engineered platforms that mimic the physiological cellular environment is a major goal of cell engineering.
Indeed, it has been widely demonstrated that the use of 3D systems, compared to 2D, is crucial for a more physiological relevant study of cellular systems, since the third dimension could differently and strongly affect diverse cell functions[1]. However, the precise engineering of 3D systems often results challenging, consequently limiting the control over cellular fate.
Here, we report the fabrication of 3D instructive platforms that modulate cellular behaviour in terms of cellular polarization, membrane curvature and uptake capability. By means of two-photon polymerization (2PP) technology, we processed a commercial biocompatible photoresist for fabricating a cage-like 3D structure capable to entrap cells. We then investigated the cell-material interaction and the effect of different micro-topographies (grooves) on cellular response. To evaluate the effect of such topographies on cellular membrane curvature, we took advantage of the SEM/FIB technology and ultra-thin plasticization (UTP) of cells, which gives the opportunity to directly observe cell-material sections with nanometric resolution [2]. We thus gathered important informations on the relations between membrane curvature and caveolae formation, known to be fundamental in endocytosis processes[3].
Moreover, by functionalizing our structures with fluorescent nanoparticles (NPs) we were able to observe how different topographies modulate the cellular uptake by evaluating NPs internalization with confocal microscopy.
Our results clearly show that by modulating cellular membrane curvature through specific topographical micro-features, it is possible to tune cellular membrane curvature and, thus, the cellular uptake capabilities. Such results could then give new guidelines for the design of innovative and more efficient delivery systems based on 3D scaffold-like devices.

1. Pampaloni, F., Reynaud, E. G. & Stelzer, E. H. K. DOI: 10.1038/nrm2236 NAT REV CELL BIO (2007).
2. Santoro, F. *et al.* DOI: 10.1021/acsnano.7b03494 ACS NANO (2017).
3. Zhao, W. *et al.* doi: 10.1038/nnano.2017.98. NAT NANOTECHNOL (2017).

BM04.03.16
Tunable Visible Light Polymerization of Poly (Ethylene Glycol) Hydrogels for Post-Polymerization Modulation of Material Properties Katherine

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Synthetic hydrogels, such as those formed with multifunctional poly(ethylene glycol) (PEG) or poly(vinyl alcohol) (PVA) macromers, are of great interest for a variety of biological applications. The high degree of property control that these materials afford, including mimicking the elasticity or 'stiffness' of tissues in the human body, makes them particularly useful as biomimetic, multidimensional culture environments for hypothesis testing in studies of disease and regeneration. Traditionally, control of synthetic hydrogel mechanical properties has been achieved with polymer concentration, molecular weight, or reactive group stoichiometry. Recently, the rate of hydrogel formation also was demonstrated as an effective handle for controlling mechanical properties. For example, the rate of hydrogel formation by oxime chemistry was controlled using pH, where the resulting differences in mechanical properties were determined to arise from differences in network heterogeneity (e.g., defects) that depended on the rate of gelation (Zander et al, Advanced Materials, 2015). Inspired by this, in this work we investigated if a rate-based approach for controlling mechanical properties could be used with photoinitiated (lithium acylphosphinate, LAP) synthetic thiol-ene hydrogels (PEG-8-norbornene, PEG-2-thiol) and the resulting defects exploited for temporal property modulation. Specifically, we established a system for hydrogel formation using different doses of visible light (455nm LED, 70-90 mW/cm^2, 1-10 min) to tune the mechanical properties. We confirmed dependence of hydrogel mechanical properties on factors beyond polymer concentration, including light intensity and exposure time. Elasticity, measured by dynamic mechanical analysis (DMA), indicated that, for precursor solution of the same composition, elasticity increased with both increasing light intensity and exposure time. To better understand the source of defects contributing to differences in hydrogel mechanical properties, end group conversion during hydrogel formation was monitored with magic angle spinning (MAS-NMR) and correlated with mechanical properties over the polymerization time. Through these comparisons, both reduced end group conversion and looping were determined to contribute to differences in mechanical properties observed at different rates of hydrogel formation. Control of end group conversion subsequently was exploited to stiffen hydrogels post-polymerization by covalent incorporation of a secondary thiol-ene network using photopolymerization (365nm, 10 mW/cm^2). In sum, we have demonstrated the high level of property control afforded by this visible light polymerization system and the potential utility of this approach for post-polymerization modulation of material properties. This method of modulating properties is promising for studying cell response to dynamic stiffness in three-dimensional culture, with applications in the study of cancer progression and wound healing.

BM04.03.17
Microcapsule Sensors for *In Situ* Monitoring of pH in Microenvironment Sangmin Lee, Chan Ho Park and Shin-Hyun Kim; Korea Advanced Institute of Science and Technology, Daejeon, Korea (the Republic of).

In-situ monitoring of pH is of great importance in biomedical fields as pH affects activities of enzyme and drug and is a symptom of certain diseases. It is known that the microenvironment of cancer cells is weakly acidic due to the secretion of lactic acid through anaerobic respiration. Therefore, pH can be an effective indicator for cancers. However, it is very difficult to use conventional litmus papers or pH-meters for measurement of local pH in cellular environments. To provide an injectable, implantable, suspendable platform of pH sensors, we suggest a microcapsule-type sensor that is composed of the pH-responsive optical sensor in the core and semipermeable polymer in the shell. As a template to produce microcapsules, monodisperse water-in-oil-in-water (W/O/W) double-emulsion droplets are prepared using a capillary microfluidic device. The innermost water phase contains molybdenum disulfide (MoS_2) nanosheets whose surfaces are grafted by pH-responsive polymers with a fluorescent group at the distal end. As the middle oil phase, a photocurable resin of polysiloxanes modified with methacrylate is used. The double-emulsion drops are irradiated by ultraviolet, which leads to the polymerization of the resin, forming a semipermeable solid shell. The pH-responsive polymer that links the MoS_2 nanosheets and fluorescent groups are designed to show a drastic conformation change in the range of pH 6.0-7.4. At physiological condition of pH 7.4, the pH-responsive polymer is collapsed so that the fluorescent groups are brought to the optical quencher of MoS_2, yielding a weak fluorescence due to the Forster resonance energy transfer (FRET). By contrast, at cancer microenvironment with pH 6.3, the pH-responsive polymer is highly extended, increasing fluorescent intensity. As the pH sensors are encapsulated by a semipermeable shell, they are free from dilution with physiological fluids and adhesion of proteins and lipids, thereby maintaining the sensing performance in a physiological environment. The microcapsule sensors can be injected, implanted, and suspended in any target volumes, which enables the in-situ monitoring of pH in the microenvironment where the microcapsules are located.

BM04.03.18
Piezoelectric Performance and Biocompatibility of $(Ba,Ca)(Zr,Ti)O_3$ Ceramics for Biomedical Applications Kara K. Poon[1], Matthias Wurm[2], Mari-Ann Einarsrud[1], Rainer Lutz[2] and Julia Glaum[1]; [1]Department of Materials Science and Engineering, Norwegian University of Science and Technology (NTNU), Trondheim, Norway; [2]Department of Oral and Maxillofacial Surgery, Friedrich-Alexander-Universität Erlangen-Nürnberg, Erlangen, Germany.

The replacement of bone tissue in surgical interventions with artificial materials is a standard procedure in current clinical practice, however, it is a substantial surgery with high patient morbidity and high healthcare costs. It is therefore desirable to develop artificial bone materials which induce controlled, guided and rapid healing to improve patient recuperation and which allow for a stable fixation between bone and implant for immediate loading.

Interest in piezoelectric ceramics for biomedical applications has risen in recent years, due to the need for biocompatible materials with active functionalities. Several studies have proposed that osteogenic regeneration may be improved with the application of electrical stimuli. Barium titanate doped with calcium and zirconium, $(Ba,Ca)(Zr,Ti)O_3$, are a class of lead-free piezoelectric ceramics which generate electric surface potentials under a mechanical load due to its non-centrosymmetric crystal structure. This class of piezoelectric ceramics may serve as bioactive bone replacement materials. However, the biocompatibility of BCZT is not well established.

In the present study, we investigate the suitability of BCZT as an artificial bone replacement material. Several compositions of bulk BCZT ceramics were synthesised via solid-state synthesis, including a morphotropic phase boundary composition (MPB) and several other tetragonal compositions. The MPB composition was chosen for its expected high piezoelectric performance, and the tetragonal compositions for the different stabilities in piezoelectric performance upon mechanical loading. Piezoelectric properties were determined for all compositions. The biocompatibility of the BCZT ceramics was investigated via cell proliferation and viability studies. We determined the BCZT ceramics to be compatible with human osteoblast cells and endothelial cells, thus giving encouragement into the further study of BCZT as bioactive bone replacement materials.

BM04.03.19
Adhesion Effect of Elastin-Like Polypeptide-Supplemented Composite Cement on the Tooth Sun-Young Kim[1] and Hyun-Jung Kim[2]; [1]Seoul National University, Seoul, Korea (the Republic of); [2]Kyung Hee University, Seoul, Korea (the Republic of).

I. Objectives
Elastin-like polypeptide (ELP) has a variety of application in biomedical field. ELPs are composed of repeats of the pentapeptide Val-Pro-Gly-Xaa-Gly; the guest residue Xaa can be any amino acid except Pro. We have tried to improve the mechanical property of dental cement by ELP supplementation in previous studies. Specially, we found the supplementation of ELP increased the adhesion ability of composite cement to tooth. Here, the objective of this

study was to investigate the adhesion effect of ELP supplementation on the tooth between the ELP-supplemented dental cement and tooth surface.

II. Materials & Methods
ELP genes either with or without octaglutamic acid termination were genetically engineered: V125 and V125-E8. Pure ELPs were gathered through a series of protein synthesis process using E.coli through gene transformation, expression, protein purification. 10 wt% ELP solutions were then prepared. The crown of human third molar without caries and restorations was horizontally sectioned to have 2mm thickness by high-speed sawing machine (Buehler). Two holes in tooth specimen were made 4.1 mm in height and 1.4 mm in diameter.
0.3 ratio in liquid/powder were prepared: cement + 60mL of either DW, V125, or V125-E8. Mixed cements as given ratio, were loaded at tooth cavities (20 cavities x 3 subgroup) and set in 37°C incubator for 7 days under 100% humidity. Push-out strength of samples were measured in universal testing machine (SHIMADZU, Japan) at a cross-head speed of 1mm/min with compressive mode. Data were analyzed using Two-way ANOVA and Bonferroni's post-hoc test at 95% significance level.
0.3 ratio in liquid/poweder were prepared same as push-out strength test. The composite cement was loaded in rheometer (Ta instrument Co., DE) and the viscoelastic property was measured for 1 hour.
Tooth disk of 2 mm in thickness was prepared and the 1 mm cavity was prepared. Same liquid/powder ratio of composite cement was prepared and filled in the cavity. The sample was stored in 100 % humidity for 2 days and in PBS solution for 1 -2 weeks more. Vertical section through half was performed with diamond saw and polished serially from 500 grit to 2400 grit. Dried sample was gold-coated and observed by SEM (Hitachi, Tokyo, Japan).

III. Results
V125-E8 group showed the highest push out strength significantly ($p<0.001$). V125 group showed lower strength than V125-E8 group, but higher push-out strength than DW group ($p<0.001$). V125-E8 groups showed significantly less viscosity and high flow compared to other groups. V125-E8 group showed a narrower gap between composite cement and dentin and composite tag in dentinal tubule while other group showed wider gap and no tag inserted in dentinal tubule.

IV. Conclusion
ELP-supplemented dental cement has higher push-out strength than DW-mixed MTA. V125-E8 showed the highest adhesion ability to tooth. The increased adhesion ability might be due to rheologic property through low viscosity and high flowability.

BM04.03.20
Patterning Type I Collagen Fiber Alignment and Geometry Using 3D Printing Bryan A. Nerger, Pierre-Thomas Brun and Celeste Nelson; Princeton University, Princeton, New Jersey, United States.

Type I collagen forms fibrous viscoelastic networks that comprise a dominant fraction of the extracellular matrix (ECM). Through interactions with single cells and tissues, networks of type I collagen can be remodeled to form anisotropic networks, which are observed in biological processes as diverse as branching morphogenesis and metastatic invasion. However, replicating the structure of anisotropic collagen networks *ex vivo* is challenging because current collagen fiber alignment techniques are often limited to unidirectional alignment over small mm-scale areas or in thin films. Here, we adapt three-dimensional (3D) microextrusion printing as a technique to fabricate anisotropic networks of type I collagen with tunable fiber alignment and geometry. Using collagen-Matrigel and collagen-Ficoll inks, we 3D-printed collagen fiber networks with 30-40% of the fibers oriented in the printing direction. Collagen fiber alignment increased with increasing printing speed and decreasing nozzle diameter, which suggests that shear and extensional flows in the conical nozzle are responsible for collagen alignment. Molecular crowding and substratum chemistry also affected the extent of collagen fiber alignment. Changing the concentration of Matrigel in the collagen-Matrigel ink allowed the geometry of collagen fibers to be tuned. By modifying the aforementioned parameters, we 3D-printed complex patterns of collagen fiber alignment and geometry that were separated by sharp interfaces. 3D-printed networks of type I collagen have great potential for studies that assess the role of collagen fiber alignment in developmental biology and tissue engineering.

BM04.03.21
Rational Design of Antimicrobial Peptide Nanofibers Biplab Sarkar[1], Steven Park[2], Peter Nguyen[1], Zain Siddiqui[1], Michael McGowan[1], David Perlin[2] and Vivek Kumar[1]; [1]New Jersey Institute of Technology, Newark, New Jersey, United States; [2]Public Health Research Institute, Rutgers, The State University of New Jersey, Newark, New Jersey, United States.

Natural antimicrobial peptides are crucial components of the host innate immune system against invading pathogens. Inspired by these natural peptides, we designed a set of cationic amphiphilic peptides that can self-assemble into injectable hydrogels. Our self-assembled system employs non-covalent forces to yield hierarchical assembly of these materials capable of disrupting cell membranes of pathogens, such as bacteria (e.g. *Pseudomonas aeruginosa*) and fungi (e.g. *Fusarium solani*). The self-assembly of the nanofibrous hydrogel was characterized with spectroscopic techniques as well as high-resolution microscopy techniques such as atomic force microscopy and scanning electron microscopy. We tested the self-assembling peptide hydrogels were tested for compatibility with stromal cells with *in vitro* cell culture. Moreover, we subcutaneously implanted the hydrogels and observed no systemic toxicity. We screened the antimicrobial platform against a spectrum of pathogens that ware associated with nosocomial infections. In addition, we determined the mechanism through which the nanofibers disrupt the pathogen colonies. The hydrogel can be either be applied as a topical antibiotic or can be integrated into medical devices, such as catheters and grafts. In addition, due to their shear-thinning viscoelastic properties, these hydrogels can be syringe aspirated and injected directly onto or into a target site. The rational design of the nanofiber should provide researchers and clinicians a viable platform to build therapies against multi-drug resistant pathogens that threaten to complicate even the most routine surgical procedures.

BM04.03.22
Biodegradable pH-Activated Polymeric Nanoparticle Modulate Lysosomal Acidification and Autophagy in Parkinson's Disease Jialiu Zeng[1], Andrew Martin[1], Orian Shirihai[2], Han Xue[1] and Mark Grinstaff[1]; [1]Boston University, Boston, Massachusetts, United States; [2]University of California, Los Angeles, Los Angeles, California, United States.

We have developed a novel polymeric pH-activable, acidifying nanoparticle (acNP) that restores the pH of compromised lysosomes to rescue autophagic flux and cellular function in neurons (PC-12 cells) under exposure to either 1-methyl-4-phenylpyridinium (MPP+) toxin or 6-hydroxydopamine (6-OHDA). Parkinson's disease (PD) occurs in 13 per 100,000 people in the population, and about 60,000 new cases are identified each year. It is characterized by the accumulation of alpha synuclein (a-syn) within Lewy bodies and neurites of the nervous system in the form of amyloid fibrils. Recent studies have indicated that perturbations in the autophagy-lysosome pathway, especially impaired lysosomal acidification that mediate the degradation of a-syn may play a role in its pathogenesis. Therefore, targeting lysosomal acidity represent a new target for therapeutic development. Although some studies have demonstrated that genetic restoration of autophagy can inhibit the development of PD, to date no effective therapeutic approach has been developed. In this study, we designed an acidic nanoparticle (acNP) that contains caged acid which can be released upon moderate pH changes to enable controlled acidification of the impaired lysosomes under lipotoxicity. The non-cytotoxic acNPs are taken up into the lysosomes of PC-12 cells, rescue cell death caused by MPP+ and 6-OHDA neuro-toxins, restore lysosomal acidity and decrease the accumulation of autophagic proteins LC3II and p62 expression

levels, indicating an overall rescue of autophagic flux. The acNPs also decreased a-syn accumulation in the PC-12 cells, effectively improving the function of PC-12 cells. These results established a primary causative role of impaired lysosomal acidification on the de-regulation of autophagic flux and cellular function in neurons, and the acNPs are of potential therapeutic interest for neuro-degenerative pathologies associated with lysosomal acidity impairment, such as PD and Alzheimer's disease.

BM04.03.23
Characterization of Crosslinked Gelatin 2D Films for Cell-Based Sensing Applications Gaurav D. Kulkarni, Soumen Das and Santanu Dhara; School of Medical Science & Technology, IIT Kharagpur, Kharagpur, India.

In cell-based sensing, use of 2D films supporting the growing cells is an ongoing area of research. The real-time application of any biomaterial primarily requires it's assessment concerning physical, mechanical and, biological characterizations. Impedance sensing has proved to be a low cost, label-free and, simple approach allowing real-time monitoring of cell growth. Evaluation of electrical performance of biomaterial is important for studying the bioimpedance of cell-based system in 2D biomaterial microenvironment. The detected information provides growth dynamics at the cellular level that is useful for tumour diagnosis by comparing the electrical properties of normal and cancerous cells. While gelatin based biomaterial is developed in authors' lab for tissue engineering, a thorough investigation of different properties of this biomaterial is reported here. Gelatin is a well-studied biomaterial for its non-antigenicity, the presence of Arginyl-Glycyl-Aspartic acid (RGD) motif and, for simple processing. Dissolution resistance in cell culture medium was overcome by chemical crosslinking in 1-Ethyl-3-(3-dimethylaminopropyl) carbodiimide/ N-Hydroxysuccinimide (EDC/NHS) and glutaraldehyde (GTA). The films after modification were tested using Ninhydrin assay for crosslinking index, rheological properties for change in gel strength and cytocompatibility of HaCaT cells for biocompatibility evaluation of 2D crosslinked gelatin films. AC conductivity was evaluated using dielectric constant of the material measured by capacitance method. The crosslinking index for EDC and GTA crosslinked films reached almost 80% at concentrations of about 200 mM and 50 mM respectively. Films crosslinked with GTA displayed more increase in storage modulus values compared to EDC films. The biocompatibility of EDC films was superior to GTA films due to cytotoxicity effects of GTA release. Cell proliferation rate on both types of films was slower with EDC group found to be more biocompatible than GTA. The AC conductivity for GTA films was higher than EDC films. The maximum conductivity obtained using capacitive method was 2 $S.m^{-1}$. The concentrations of EDC and GTA which can be suggested for crosslinking gelatin films based on this study are 100 mM and 50 mM for cell-based sensing applications.

BM04.03.24
Ultra-Low Fouling Biocompatible Coatings for Medical Devices Doreen Chan[2], Eneko Axpe[1], Anton Smith[1], Lyndsay Stapleton[3] and Eric A. Appel[1]; [1]Department of Materials Science & Engineering, Stanford University, Stanford, California, United States; [2]Department of Chemistry, Stanford University, Stanford, California, United States; [3]Department of Bioengineering, Stanford University, Stanford, California, United States.

Non-specific adhesion of proteins, cells, and micro-organisms presents huge problems in the medical industry, instigating the occlusion, contamination, and malfunction of biomaterial devices. Several "gold-standard" non-fouling coatings have been developed based on poly(ethylene glycol) and zwitterionic poly(meth)acrylates; however, these coatings suffer from instability and short lifetimes. In contrast, polyacrylamides exhibit remarkable stability and thus increased lifetime, making polyacrylamides ideal candidates for surface coatings on devices that come into contact with biological fluids. Polyacrylamides are a broad class of polymers, but have been poorly studied as non-fouling coatings, driving the need for the development of novel ultra-low fouling materials that are biocompatible. Towards this aim, we have screened several hundred acrylamide copolymer hydrogels by studying their non-fouling properties to identify top-performing materials that resist protein absorption and blood platelet adhesion. Further, we have examined the biocompatibility of top-performing formulations as coatings on a variety of substrates in the subcutaneous space in mice. These materials demonstrate resistance to biofouling and the foreign body response for a month in vivo. This technology can be easily applied to biomaterial devices such as neural implants, synthetic vascular grafts, and infusion catheters to prevent fouling, improving device function and lifetime.

BM04.03.25
Molecularly Mobile Surfaces with Heparin-Binding Proteins for Improving Functions of Hepatocyte-Derived Cells Yoshinori Arisaka and Nobuhiko Yui; Institute of Biomaterials and Bioengineering, Tokyo Medical and Dental University, Tokyo, Japan.

Polyrotaxane (PRX) is a supermolecule with many cyclic molecules (e.g., cyclodextrins) threaded onto an axis polymer (e.g., polyethylene glycol) and has a potential of freely sliding and rotational motions of the cyclic molecules along the chain as molecular mobility. In recent years, we clarified the relationship between the cell differentiation of mesenchymal stem cells (MSCs) and the molecular mobility on PRX coated surfaces (PRX surfaces). The highly mobile surfaces were preferable to adipogenic differentiation, whereas the less mobile surfaces induced their osteogenic differentiation [1]. For the further advancement of the control of cellular behaviors, we designed growth factor-tethered PRX surfaces. Previously, we have succeeded in tethering positively charged heparin-binding growth factors onto sulfonated-PRX surfaces via electrostatic interaction. The surfaces enhanced osteoblast differentiation by the mobility of PRX and tethering of growth factors [2]. Based on the surface design, we attempted to improve hepatic functions *in vitro* of hepatic-derived cells (HepG2) in the present study. In particular, a sulfonated-PRX triblock copolymer, which consists of a sulfated-PRX as a middle block segment and poly(benzyl methacrylate) as both-terminal segments, was coated to a polystyrene substrate by a simple casting method. Subsequently, heparin-binding epidermal growth factor-like growth factor (HB-EGF) as a survival factor for hepatocytes was tethered on a sulfated-PRX surface. As a control surface, HB-EGF was adhered on a non-sulfonated PRX surface by hydrophobic interactions. For assessing mechanical-signalings by the molecular mobility, initial adhesion and subcellular localization of Yes associated protein (YAP) which is an essential transducer of mechanical signals was analyzed by microscopic observation. After 24 h in culture, the number of adhered cells on sulfonated surfaces was almost the same as that on non-sulfonated PRX surfaces, regardless of the molecular mobility. Although highly mobile surfaces suppressed nuclear localization of YAP, less mobile surfaces transferred YAP into the nucleus. These results indicate that the YAP activity can be regulated by the molecular mobility independently of the initial adhesiveness. It had reported that the YAP nuclear localization led to dedifferentiate hepatocytes and impaired hepatic functions [3]. Highly mobile surfaces would be suitable to maintain or enhance the functions. In order to evaluate the functions, albumin secretion from HepG2 on surfaces was quantified. Highly mobile sulfonated PRX surfaces with tethered HB-EGF induced the highest secretion of albumin among all culture conditions. These results strongly suggest the synergistic effect of the mobility of PRX and tethering of HB-EGF via electrostatic interactions.
[1] Adv Healthcare Mater. 2015;4(2):215-222. [2] J Biomater Sci polym ed. 2017;28(10-12):974-985. [3]Cell. 2014;157(6):1324-1338.

BM04.03.26
Synthesis of Selenium-Incorporated Alpha-Tricalcium Phosphate and Evaluation of Its Cement-Type Reactivity Bersu Bastug[1, 2] and Caner Durucan[1, 2]; [1]Metallurgical and Materials Engineering, Middle East Technical University, Ankara, Turkey; [2]BIOMATEN Center of Excellence in Biomaterials and Tissue Engineering, Middle East Technical University, Ankara, Turkey.

Alpha-tricalcium phosphate (α-TCP) is a promising hard tissue analog due to its cement-type hydraulic conversion to the mineral component (calcium deficient hydroxyapatite, CDHAp) of natural bone tissue. A variety of different inorganic and organic agents can be incorporated to α-TCP in order to enhance its therapeutic and regenerative properties. In this study, an ionic incorporation - selenium (Se) - was accomplished to impart anti-carcinogenic

property to α-TCP. Se-incorporated α-TCP (α-TCP:Se) was obtained by a solid-state reaction of custom synthesized Se-incorporated monetite ($CaHPO_4$:Se) and calcium carbonate ($CaCO_3$) precursors at 1200 °C for 2 h. The effect of Se-incorporation on crystal structure of α-TCP and on its' cement type hydraulic reactivity at 37 °C was investigated in detail by crystallographic analysis (Rietveld refinement) and by isothermal calorimetry, respectively. At low amount of Se-incorporation (< 5 wt.%), α-TCP remains phase pure, however relatively higher amount of Se addition (10 wt.%) leads to formation of β-polymorph of TCP lacking the cement-type setting behavior. The SEM examinations showed that the morphological properties of α-TCP remain unaffected after doping with Se. However, Se-incorporation occurs at a limited extent, lower than the theoretically expected values as revealed by spectroscopical analyses. Se-incorporation does not change the reticulated needle/plate like morphology of the CDHAp, which is characteristic to cement-type hydration and hardening. Se-incorporated TCPs fully convert to CDHAp, but Se-incorporation changes the reaction kinetics and mechanistic path for α-TCP to CDHAp cement conversion and higher amount of Se addition slows down the reaction kinetics. The results imply that an optimal Se amount is critical the preserve intrinsic cement nature/behavior of α-TCP.

BM04.03.27

Nanocomposite for Wound Dressing Applications Obtained by 3D Printing Mayra E. Garcia-Sanchez, Mariana I. Garay Barragan, Christian R. Moya-Garcia, Fabio A. Gonzalez Sanchez, Jorge A. Perez and Ines Jimenez Palomar; Research and Development, Inmateriis S.A. de C.V., Guadalajara, Mexico.

Infection in exposed wounds is one of the main factors affecting wound healing. Thus, antimicrobial engineered biomaterials have been addressed in clinical applications for regenerative medicine [1]. Wound dressings used in the treatment of diabetic foot ulcers, affliction causing 85 % of non traumatic lower limb amputations, may prevent infections and possible amputations providing an environment to improve wound healing [2]. Herein, a 3D printed novel nanocomposite with tissue regeneration and antimicrobial properties constituted by a bacterial cellulose (BC) membrane and a biocompatible polymer matrix was produced.
BC presents attractive applications, especially in the medical area due to its high degree of crystallinity, purity, reticulated conformation, biodegradability and biocompatibility [3]. For this research, BC was obtained from mango pulp, an industrial waste used in the formulation of alternative culture media. In addition, BC membrane was supported with biocompatible polymers (polycaprolactone and poly(vinyl alcohol)) at varying ratios and functionalized with grapefruit seed extract to enhance antimicrobial properties and durability. Physico chemical properties of the nanocomposite were analyzed by SEM, TGA, FTIR, tensile testing and water holding capacity techniques. *In vitro* assays consisted of evaluating biocompatibility, viability, antimicrobial and antifungal activity. *In vivo* studies comprised the analysis of intracutaneous reactivity, cytotoxicity, and mutagenic activity.
As BC membranes exhibit biocompatibility properties, high mechanical resistance in wet conditions, high capacity for hydration and permeability of liquids and gases; in addition to the null or low irritation that causes to the skin, these membranes are a useful material in the care and regeneration of cutaneous wounds [4]; complementing the BC membrane with biopolymers and an antimicrobial agent to create a nanocomposite suggests the biocompatibility and bioactivity behavior may increase. Hence, the developed nanocomposite may be used in diverse regenerative engineering applications such as wound dressings.
[1] Campoccia, D., Montanaro, L., & Arciola, C.R. (2013). A review of the clinical implications of anti–infective biomaterials and infection resistant surfaces. Biomaterials, 34(33), 8018–8029.
[2] Solway, D. R., Clark, W. A., & Levinson, D. J. (2011). A parallel open–label trial to evaluate microbial cellulose wound dressing in the treatment of diabetic foot ulcers. International wound journal, 8(1), 69–73.
[3] Huang, Y., Zhu, C., Yang, J., Nie, Y., Chen, C., & Sun, D. (2014). Recent advances in bacterial cellulose. Cellulose, 21(1), 1–30.
[4] Keshk, S. M. A. S. (2014). Bacterial Cellulose Production and its Industrial Applications. Bioprocessing and Biotechniques, 4(2), 1–10.

BM04.03.28

Integrated Design Scheme Based on Mechano-Biology and Level Set Based Topology Optimization for Bone Scaffolds Mehmet S. Aydin[1, 2], Busra Kuloglu[1], Calga Sipahi[1], Berkay Cayir[1] and Gullu Kiziltas[1, 2]; [1]Sabanci University, Istanbul, Turkey; [2]Sabanci University Nanotechnology Research and Application Center, Istanbul, Turkey.

One of the key requirements of scaffolds is the balance between mechanical function and mass transport to aid biological delivery and tissue regeneration. Computational topology design and Solid Free-Form Fabrication efforts made it possible to create scaffolds with controlled architecture. The level-set based approach unlike standard topology optimization methods can overcome issues such as high computational demand and local minima problems resulting in a more efficient and generalized synthesis effort and hence better performing designs of tissue-engineering scaffolds. Here we develop a computational design tool based on the integration of the level set method and mechano-regulatory models for optimizing scaffolds based on desired multi-functionality including elasticity, diffusivity, and permeability as well as tissue differentiation. First, for the level set method, computational models are implemented using COMSOL Multiphysics which provides the opportunity to build an FEA (Finite Element Analysis) model where various boundary value problems are coupled and studied at the same time to reach an optimally performing tissue. Response of the scaffold is analyzed using solid mechanics, general form PDE and fluid flow modules. These modules are integrated to solve respective governing equations simultaneously. Level-set method is performed by utilizing Hamilton-Jacobi equation in the general form PDE module. A initial level-set surface is updated according to the sensitivity of the desired metrics with respect to material parameters. The change in the level-set surface is automatically reflected on the scaffold structure and an optimum structure with desired stiffness, porosity and diffusivity is obtained by finding an optimum level-set surface. As a second capability, the level set based topology scheme is integrated to a self-healing simulation capability, via a mechano-regulatory model developed to mimic tissue regeneration. For the topology optimization design method the level-set method is employed, where the design process starts with an initial geometry that satisfies physical constraints. At each time step, this geometry is improved based on sensitivity analysis results until convergence is reached. Results of both the mechanoregulatory and the topology optimization methods validate well-known benchmark design problems in literature. One design that resulted from the integrated framework on both is successfully fabricated using non-solvent-induced phase separation and 3D printing. Characterization studies using micro-CT and mechanical testing are underway. Finite element method integrated to the level-set based topology optimization is proven to be among the most computationally efficient and generic design tool for solving non-intuitive tissue engineering problems. Hence, the proposed design framework, when implemented with corresponding physical models, is equally applicable to other hard and soft tissue designs.

BM04.03.29

A Microinvasive Toolkit for Chronic Multimodal Deep Brain Interfacing Khalil Ramadi[1, 2] and Michael J. Cima[3, 2, 1]; [1]Harvard-MIT Health Sciences & Technology, Cambridge, Massachusetts, United States; [2]Koch Institute for Integrative Cancer Research, Massachusetts Institute of Technology, Cambridge, Massachusetts, United States; [3]Department of Materials Science, Massachusetts Institute of Technology, Cambridge, Massachusetts, United States.

Brain pathologies often arise from the dysfunction of specific neural circuit nodes. Interrogation of these nodes is a primary goal of neuroscience research. Reliable targeting of these microstructures can be difficult, however. Nodes are often small (sub-mm) and irregularly shaped. Current approaches employ stereotactic mapping and rely on large (>300μm) guide cannulas to ensure minimal deflection of probes during insertion. Such techniques can result in extensive glial scarring. This can substantially modify the local microenvironment to be investigated and limit chronic viability of implants. This is especially the case for fluidic targeting and drug delivery. Acutely inserted needles used for drug microinfusions (100nl-2μl) are large (23-28G), causing

insertion trauma and backflow of infusate.

We present a toolkit for manufacturing multimodal neural probes (termed Miniaturized Neural drug Delivery systems (MiNDs)) for use in both small and large animal models. Probes can be customized according to desired functionalities. Here we show in vivo functionality of MiNDs containing fluidic and electrical functionalities and an MRI-compatible MiNDs with 2 fluidic channels in a 200µm footprint. We also report the ability to independently insert and steer individual 60µm fibers of various materials, allowing access to multiple brain sites from a single burr hole. We characterize the mechanical insertion properties of different probe sizes and materials, elucidating the various advantages of each. The microinvasive footprint of the probes limits gliosis and enhances neural regeneration, allowing for chronic viability and functionality up to 1 year post-implantation.

High sensitivity MicroPET was used to characterize distinct infusion dynamics of nanoliter fluid infusions through chronic implants, emphasizing the ability to finely tune volume to target brain microstructures. We supported this by inducing volume-dependent behavior modulation in rodents with unilateral stimulation of GABA circuitry in the substantia nigra. These techniques are readily transferable to other laboratories seeking to develop custom neural probes for multimodal chronic neural interfacing. MiNDs is a powerful tool for the dissection of deep brain microstructures in small and large preclinical models.

BM04.03.30

Polymerized siRNA Delivery System for Kras-Mutant Ovarian Cancer Minju Kim[2, 1, 4], So Jin Lee[3], Sun Hwa Kim[1], Thomas Roberts[4] and Ick Chan Kwon[2, 1]; [1]Korea Institute of Science and Technology, Seoul, Korea (the Republic of); [2]KU-KIST Graduate School of Converging Science and Technology, Seoul, Korea (the Republic of); [3]Samyang Biopharmaceuticals, Seongnam-si, Korea (the Republic of); [4]Cancer Biology, Dana Farber Cancer Institute, Boston, Massachusetts, United States.

Cancer is an unconquered disease affecting millions of patients a year. To overcome the critical drawback of conventional therapies, targeted-therapy has gained attention as next generation strategy to cure cancer patients with less toxicity. Among many designs, biological material based nucleic acid as drug agent was demonstrated by numerous groups from early 2000s. Small interfering RNA, or siRNA, is non-coding short RNA molecules capable of regulating protein expression using RNA interference machinery. Utilizing siRNA as biological drug to repair cancerous metabolism caused by mutated protein has resulted in positive outcome with anti-cancer effects in pre-clinical experiments and clinical trials. In fact, siRNA therapeutics by Alnylam Pharmaceuticals is FDA-approved for clinical use for respiratory syncytial virus infections. However, the delivery of siRNA has been major hurdle in translating the efficacy in various cancer patients. Here, we incorporated nanoparticle to enhance the delivery of siRNA to target tumor sites under systemic administration. Bio-compatible and bio-degradable glycol chitosan was self-assembled with polymerized siRNA to form stable delivery complex to tumor sites to downregulate oncogenic expression. Phosphoinositide 3-kinase (Pi3K) and Ras are the most commonly activated oncogenic pathways in solid malignancies and have interdependent relationship via feedback loop system. Many previous reports have shown that dual-inhibition of Pi3K and Ras pathway has therapeutic effects in various cancer models. Though use of multi-drug achieves simultaneous inhibition of specific pathways, undesired side-effects and resistance occasionally occur from collision of pharmacokinetics of small molecule inhibitors. In addition, currently none of Ras inhibitor is potent enough for pathway inhibition, instead secondary inhibitor on downstream protein, MEK, is more commonly used. Here, we used siRNA therapeutics in collaboration with pan-Pi3k inhibitor to demonstrate anti-tumor effects in ovarian cancer. Ovarian cancer cell line with PTEN deficiency and $Kras^{G12D}$ mutation were obtained from spontaneous tumor model to examine synergistic effect of Pi3k and Ras inhibition. In cellular level, GDC and siKRAS impeded the activity of Pi3k activity and expression of Ras, respectively. When both pathways were simultaneously inhibited, cell proliferation and migration significantly delayed. In allograft ovarian cancer model, tumor sizes were critically reduced when both Pi3k and Ras were inhibited together. The immunoblot and immunohistochemistry results verified that that dual-inhibition impeded tumor growth and induced apoptosis. Here, we demonstrate that combination inhibition of Pi3k and Ras using different treatment strategies is an exceeding alternative over conventional single-agent treatments.

BM04.03.31

A Novel Approach for Local Delivery of Drugs for Treatment of Spinal Metastasis Elie Akoury, Pouyan Ahangar, Bardia Barimani, Michael Weber and Derek Rosenzweig; McGill University Health Centre - Research Institute, Montreal, Quebec, Canada.

Introduction: Bone metastases are the most common bone tumor, and they are often derived from solid tumors of different organs including the prostate, lung and breast. Current non-surgical therapies focus on chemotherapy and bone preservation, with Doxorubicin (Dox) and Zoledronate (Zol) being some of the most commonly used drugs. These drugs are usually systemically delivered to patients, and they can cause multiple adverse effects. Interestingly, our group has investigated the potential of local delivery of zoledronate to the site of bone metastasis and has shown reduced tumor-induced osteolysis compared to systemically treated xenograft animals. Over the past decades, researchers adopted new methods for drug delivery and explored the use of nanoparticles and porous materials as an effective method for delivering anti-cancer drugs directly to tumor sites. Our aim is to develop controlled-release carriers that locally delivers Zol or Dox in a patient-specific manner for the treatment of bone metastasis.
Methods: Testing drug release from nanobeads: fluorescent Zol or Dox was incubated with mesoporous silica nanoparticles. Aliquots of drug-containing supernatant were taken daily, and fluorescent drug was measured using a plate reader. Alternatively, we have also been testing LAY-FOMM 60 nanoporous 3D printable material, for releasing Dox or Zol over a sustained period of time. Testing the effect of direct Zol and Dox treatment on a prostate cancer cell line and prostate cancer-induced bone metastasis cells: cells were seeded, incubated with non-fluorescent Zol or Dox and assessed for proliferation, metabolic activity, migration and invasion using commercially available kits.
Results: Nanoparticles and 3D printed LAY-FOMM scaffolds take-up and release effective doses of Zol or Dox over time. The cancer cell line and patient-derived bone metastases cells treated directly with Zol or Dox show significantly reduced cell proliferation, metabolic activity, migration and invasion. In ongoing experiments, Zol or Dox loaded nanoparticles and LayFomm scaffolds are being tested on a prostate cancer cell line and patient-derived cells.
Conclusions: Nanoparticles or 3D-printed constructs releasing drugs could constitute a therapeutic promise to combat metastatic spine tumors. Nanoparticles can be integrated into commercial bone cements to develop a bioactive bone graft following bone tumor resection to deliver localized drug facilitating bone stability and healing while preventing tumor recurrence. Likewise, 3D printed scaffolds can be made to perfectly fit patients' defect sites while at the same time be loaded with chemotherapeutics to block cancer recurrence and promote bone repair.

BM04.03.32

Development of Biodegradable Polyurethane Elastomers with Chain Regulators for Bone Tissue Engineering Betul S. Yagci[1] and Eda Ayse Aksoy[1, 2]; [1]Faculty of Pharmacy, Department of Basic Pharmaceutical Sciences, Hacettepe University, Ankara, Turkey; [2] Institute of Science, Department of Polymer Science and Technology, Hacettepe University, Ankara, Turkey.

Bone injuries necessitate the use of scaffold-based tissue engineering approaches and development of hard tissue supports and innovative biomaterials are important. In this study it is aimed to develop novel biodegradable polyurethane elastomers as based bone regenerative films. For this purpose, firstly synthesis of polyurethane prepolymers were studied and condensation polymerization between polycaprolactone diol and hexamethylene diisocyanate monomers were carried out. Monomer ratios, catalyst amount, synthesis time and temperature were changed and obtained prepolymers were characterized according to their chemical properties, average molecular weights and solubility in solvent systems. During the second step of polymerization with chain regulators were integrated in to diisocyanate-terminated polyurethane prepolymer via their difunctional groups. As chain regulator agent organic and inorganic compounds and an amino acid molecule were used. The chemical, thermal, viscoelastic and surface free energy of chain regulated polyurethane films were investigated by FTIR, DSC, TGA, DMA and goniometer, respectively. The bioactive chain regulator containing polyurethanes had shown of urethane, urea hydrogen bonds and provided all the transitions of viscoelastic behavior. Biodegradation behaviors were examined in enzymatic and oxidative media in 75 days period. The bioactive chain regulator containing polyurethanes have showed surface erosion type of biodegradation and over 20 % and 60 % weight loss were recorded gravimetrically in oxidative and enzymatic medias respectively. Significant and positive findings were obtained for the developed biodegradable polyurethane elastomers as bone regenerative film.

Acknowledgement: This study is financially supported by Hacettepe University BAP Carrier Transition Project TKG-2017-15635.

SESSION BM04.04: Hydrogel-Based Biomaterials
Session Chairs: Josephine Allen and Gulden Camci-Unal
Wednesday Morning, November 28, 2018
Sheraton, 2nd Floor, Independence West

8:00 AM *BM04.04.01
Nano- and Microfabricated Hydrogels for Regenerative Engineering Ali Khademhosseini; Department of Bioengineering, Chemical Engineering, Radiology, University of California, Los Angeles, Los Angeles, California, United States.

Engineered materials that integrate advances in polymer chemistry, nanotechnology, and biological sciences have the potential to create powerful medical therapies. Our group aims to engineer tissue regenerative therapies using water-containing polymer networks, called hydrogels, that can regulate cell behavior. Specifically, we have developed photocrosslinkable hybrid hydrogels that combine natural biomolecules with nanoparticles to regulate the chemical, biological, mechanical and electrical properties of gels. These functional scaffolds induce the differentiation of stem cells to desired cell types and direct the formation of vascularized heart or bone tissues. Since tissue function is highly dependent on architecture, we have also used microfabrication methods, such as microfluidics, photolithography, bioprinting, and molding, to regulate the architecture of these materials. We have employed these strategies to generate miniaturized tissues. To create tissue complexity, we have also developed directed assembly techniques to compile small tissue modules into larger constructs. It is anticipated that such approaches will lead to the development of next-generation regenerative therapeutics and biomedical devices.

8:30 AM *BM04.04.02
Decellularized Extracellular Matrix Based Hydrogels for Regenerative Engineering Karen L. Christman; University of California, San Diego, La Jolla, California, United States.

The extracellular matrix (ECM) is nature's scaffold, and in recent years, researchers have isolated these scaffolds for tissue engineering applications by removing all of the cellular components, a process called decellularization. These scaffolds are known to promote cell influx, regeneration, and healing in a variety of tissues, and their degradation products have angiogenic, chemoattractant, and antimicrobial properties, as well as promote cell proliferation. By removal of the cellular antigens, these scaffolds are considered biocompatible, and xenogeneic sources can be used. While these scaffolds retain the native ECM structure, they are not amenable to minimally invasive, injectable procedures. We have developed a variety of injectable ECM derived hydrogels that self-assemble to form porous, nanofibrous scaffolds once injected in vivo or brought to physiological conditions *in vitro*. These ECM based scaffolds have been shown to increase tissue specific differentiation and maturation of a variety of progenitor and stem cells *in vitro*, and are showing promise *in vivo* in several tissues including the myocardium and skeletal muscle. This talk will cover the recent progress with these materials.

9:00 AM BM04.04.03
Self-Assembling Peptide Hydrogel for Enhanced Cholesterol Uptake Biplab Sarkar, Peter Nguyen, Zain Siddiqui and Vivek Kumar; New Jersey Institute of Technology, Newark, New Jersey, United States.

Blocking proprotein convertase subtilisin/kexin type 9 (PCSK9) is an important therapeutic target for lowering circulation of low density lipoprotein (LDL) particles, which can improve cardiovascular health. We have developed a self-assembling peptide hydrogel that can bind PCSK9, leading to increased uptake of LDL particles in the liver cells. The cholesterol-lowering therapeutic hydrogel may be locally applied/implanted for sustained release. The platform is based on beta-sheet nanofibers that are ionically crosslinked into thixotropic hydrogels. We have characterized the hierarchical self-assembly of the peptide through microscopy and spectroscopy. Crucially, *in vitro* and *in vivo* experiments conducted provide important insight into the efficacy of the hydrogel. The injectable/implantable hydrogel may lead to a general delivery platform to combat and prevent cardiovascular conditions associated with high blood pressure.

9:15 AM BM04.04.04
Molecular Gradients in Conducting Polymer Films Generated via Hydrogel-Mediated Electrodeposition for Tissue Engineering Applications Fereshtehsadat Mirab and Sheereen Majd; University of Houston, Houston, Texas, United States.

Gradients of biological molecules play a key role in a number of cellular functions such as proliferation and differentiation. Surfaces that present such molecular gradients are thus, attractive platforms for controlling cellular behavior and for tissue regeneration. The aim of this study is to create gradients of biomolecules on films of conducting polymers (CPs) as versatile substrates for studying and controlling cells. CPs, a group of organic polymers with tunable physical and chemical properties, have become increasingly popular for a wide range of biomedical applications including tissue engineering and

bioelectronics. We previously developed a facile and efficient method of hydrogel-mediated electropolymerization for fabrication of patterned films of a biocompatible CP, polypyrole (Ppy) functionalized with biomolecules. Herein, we extend the scope of this technique and produce gradients of biomolecules incorporated into films of Ppy. To this end, an agarose hydrogel stamp containing CP precursors, e.g. pyrrole monomer and polystyrene sulfonate (as dopant), is prepared via replica molding and loaded with a solution of fluorescent-tagged molecules (e.g. dextran tetramethylrhodamine). Diffusion of fluorescent molecules through agarose hydrogel is utilized to generate distinct molecular gradients at different time points. These gradients are then incorporated into Ppy films by placing the hydrogel in contact with a gold-coated substrate following by electrodeposition of Ppy using a current density of 0.3-0.9 mA/cm^2 for 60-240 sec. The resultant gradients are characterized by fluorescence microscopy and Energy Dispersive X-ray Spectroscopy (EDS) analysis and are compared to theoretical model of diffusion. The fluorescence imaging of both the hydrogel stamp and the corresponding deposited CP film demonstrate that the profile of molecular gradient within the hydrogel is time-dependent. These results are further confirmed using EDS analysis. The present approach can be effectively applied to generate CP films of various geometries with controlled molecular gradients for guiding cellular behavior for tissue engineering purposes.

9:30 AM BM04.04.05
Designing Degradable Poly(Ethylene Glycol) Hydrogels for Improved Cartilage Tissue Engineering with Combined Experimental and Computational Methods Margaret C. Schneider[1], Stanley Chu[1], Shankar Lalitha Sridhar[2], Franck J. Vernerey[2] and Stephanie J. Bryant[1]; [1]Chemical and Biological Engineering, University of Colorado Boulder, Boulder, Colorado, United States; [2]Mechanical Engineering, University of Colorado Boulder, Boulder, Colorado, United States.

Photoclickable thiol:norbornene poly(ethylene glycol) (PEG) hydrogels are promising for *in situ* delivery of chondrocytes to promote tissue regeneration in focal cartilage defects. This platform offers a high degree of tunability through the introduction of thiolated matrix components and growth factors, achieving a range of mechanical properties and degradation behaviors with spatiotemporal control offered by photo-polymerization. In particular, degradation is critical for allowing the transition from hydrogel to neo-tissue as cells secrete new extracellular matrix (ECM) molecules. Using a combined experimental and computational approach to design degradable hydrogels, we have identified two mechanisms that facilitate the transition from gel to neo-tissue: 1) interactions between the cell and polymer precursors, which reduce crosslinking near the cell and 2) cell clusters, which enable early localized tissue growth and eventual connection to form neo-tissue. This work aimed to identify the interplay of cell clusters and heterogeneous crosslink density and its influence on neo-cartilage growth in PEG hydrogels.

We defined three parameters: R_d, the distance over which the crosslink density varies from the cell membrane to the bulk gel; F, the volume fraction of clusters relative to the total construct volume; and c_f, the ratio of cluster cell density to the background cell density. Computationally, we identified that gels with high F and c_f led to interconnected ECM prior to reaching reverse gelation due to an overlapping R_d. Experimentally, we developed methods to induce cell clusters via agitation pre-encapsulation. The ratio of clusters to single cells was controlled pre-encapsulation in hydrolytically degradable gels enabling the cluster size to be varied while holding F and c_f constant. When cultured for 4 weeks, there was no difference in ECM deposition measured by sulfated glycosaminoglycans (sGAGs) and collagen as a function of cluster size. Moreover, the modulus decreased from ~20 kPa to ~5 kPa indicating an overall loss in gel without sufficient matrix production. However, the gel was expected to reach reverse gelation prior to 4 weeks indicating that the little ECM produced had formed enough of a connected network to maintain the construct, which was supported by histology imaging for collagen and sGAGs. The clinical viability of the constructs was tested over longer times in *in vivo* experiments by placing cellular gels subcutaneously in nu/nu mice. TGFβ3 was immobilized in gels to help support and maintain the chondrogenic phenotype. With a 9 week culture there was a ~2x increase in the modulus of the gels compared to a 25% decrease in *in vitro* gels.

To conclude, we combined experimental and computational methods to identify that heterogeneous distribution of cells and crosslinks contribute to the transition from hydrogel to neo-tissue. We are able to induce clusters and design gels that promote tissue formation.

9:45 AM BM04.04.06
Integration of Gold Nanorod and Agarose Hydrogel for Controlling *In Vitro* Neurite Outgrowth Nari Hong and Yoonkey Nam; KAIST, Daejeon, Korea (the Republic of).

To control the position of neurons and connection of their neurites on culture substrates, various cell-attractive or cell-repellent materials have been used for micropatterning techniques. However, when neurons are cultured on an engineered substrate fabricated by conventional patterning methods, it is difficult to change their patterned distribution again during the cultivation. In this study, we utilized gold nanorods (GNRs) that has the property of photothermal effect in the near-infrared (NIR) region to fabricate thermoplasmonic interface on a culture substrates. By integrating this interface with a micropatterning method using thermosensitive agarose hydrogel, we demonstrate the manipulation of neurite outgrowth even after the neuronal cultures formed confined networks. GNRs, which was synthesized by seed-mediated method, were immobilized on glass culture substrates through the electrostatic binding. Following by poly-D-lysine coating on the thermoplasmonic interface for neuron attachment, agarose hydrogel was patterned using micro-molding in capillary method to fabricate micro-sized square wells. On the patterned substrate, E18 hippocampal neurons were seeded and they connected each other only within individual agarose wells because of the repellency of the hydrogel for cell adhesion. At one or two weeks after cell seeding, agarose hydrogel was successfully melted by localized heat of GNRs under NIR illumination. Through the micro-sized channel produced by the melted agarose, the neurites extended from neuronal networks in agarose wells. We also show that the heat generated from GNRs can be used for inducing localized neurite damage by applying photothermal stimulation to neurites directly. NIR beam was focused on the neurites that grew along the melted agarose channel and local damage was observed at the illuminated spot. The developed method that can manipulate neurite outgrowth by photothermal stimulation even during the cultivation is expected to be useful for studying in vitro model of nerve injury and regeneration.

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10:00 AM BREAK

10:30 AM *BM04.04.07
Sliding Hydrogels with Tunable Molecular Mobility and Ligands as 3D Niche for Accelerating MSC-Based Cartilage Regeneration Xinming Tong[1] and Fan Yang[1, 2]; [1]Department of Orthopedic, Stanford University, Stanford, California, United States; [2]Department of Bioengineering, Stanford University, Stanford, California, United States.

Hydrogels are attractive matrices for delivering mesenchymal stem cells (hMSCs) for cartilage repair given its injectability and tunable properties. However, conventional hydrogels often result in slow cartilage formation due to the time needed to degrade and remodel the original hydrogel network.

Our group has recently reported sliding hydrogels with molecular mobility as a novel 3D cell niche, which is characterized by mobile crosslinks and biochemical ligands, and enable cells to reorganize biochemical ligands and protrude in 3D hydrogels. The goal of this study is to develop sliding hydrogels with tunable mobility and biochemical ligands to evaluate the effect of introducing molecular mobility and varying biochemical ligand types on modulating cartilage formation by MSCs in 3D.
PEG-based sliding hydrogels with compressive modulus ~10kPa were synthesized and fabricated as we recently reported. To facilitate the cartilage matrix production and deposition by MSC in 3D, MSCs were encapsulated in sliding hydrogels crosslinked by MMP cleavable peptide (CGPQGIWGQC) to allow cell-mediated degradation. To assess the effect of varying ligand types in sliding hydrogels on MSC chondrogenesis, two types of biochemical ligands (1 mM) were incorporated into sliding hydrogels including CRGDS and N-cadherin mimicking peptide HAVDIGGGC. Molecular mobility in non-degradable sliding hydrogels substantially accelerated and enhanced cartilage matrix deposition by MSCs in 3D. Incorporation of cell-mediated degradation synergized with molecular mobility, and further enhanced neocartilage formation throughout the hydrogels. Compared to no ligand control, CRGDS significantly inhibited the chondrogenesis of hMSCs, as shown by downregulations of cartilage markers as well as decreased sGAG deposition in 3D. While previous study has suggested that HAVDI incorporation into hyaluronic acid-based hydrogels enhanced chondrogenic differentiation and cartilage matrix production, our results showed HAVDI incorporation at 1 mM did not significantly impact the speed and amount of cartilage formation by MSCs in 3D. Future studies will further examine the effects of varying the types and density of mobile ligands on modulating chondrogenesis in 3D. Our results validate sliding hydrogels as promising matrices for enhancing stem cell-based cartilage regeneration, and provide a materials tool to uncover the novel role of molecular mobility in modulating stem cell differentiation and tissue formation in 3D.
Acknowledgements: The authors would like to thank NIH R01DE024772, California Institute for Regenerative Medicine (Grant #TR3-05569) and National Science Foundation CAREER award program (CBET-1351289) for funding.

11:00 AM *BM04.04.08
Centrifugally Synthesized Functional Hydrogel Microparticles for Biomedical Applications Hiroaki Onoe; Keio University, Kanagawa, Japan.

Biofunctional hydrogel microparticles have played a central role in biomedical fields involving drug delivery systems, scaffold materials for in vitro tissue culture and tissue encapsulation for in vivo implantation. Here I introduce recent progress on simple centrifuge-based techniques for synthesizing microparticles (typical diameter: ~100 μm) composed of functional hydrogels. A pre-gel solution was introduced into a pulled glass thin capillary (tip diameter: ~100 μm) that was fixed in a centrifugal tube and ejected by centrifugal force (1000-3000 G) applied by a table-top centrifuge to form microdroplets of the pre-gel solution. The ejected microdroplets were solidified into hydrogel microparticles by crosslinking using ionic reaction, temperature or UV irradiation. The feature of this particle synthesizing technique is simple, easy-to-use and cost-effective and is applicable to encapsulation of tiny amounts of precious samples in the microparticles. This easiness of the particle synthesis procedures could enable to access to this technique for non-specialists of microfluidic engineers including biological scientists and medical doctors. A basic mechanism of the particle synthesis relies on droplet generation from the tip of the glass capillary, and the transition of jetting and dripping phenomena in the droplet generation determines the uniformity and diameter controllability of the generated hydrogel microparticles. The variations of the functional hydrogel microparticles were demonstrated such as homogeneous, core-shell and Janus particles composed of alginate, collagen or pNIPAM for the applications of cell encapsulation, 3D co-culture, and drug release.

11:30 AM BM04.04.09
Effect of Varying the Molecular Weight of Hyaluronic Acid Based Hydrogels on 3D Angiogenesis Models Samir F. Hossainy[1, 2], Shane A. Browne[2] and Kevin E. Healy[1, 2]; [1]Materials Science and Engineering, University of California, Berkeley, Berkeley, California, United States; [2]Bioengineering, University of California, Berkeley, Berkeley, California, United States.

One of the major challenges of stem cell transplantation therapy is improving survival after transplantation and enabling engraftment with host tissue to prolong the therapeutic window. Hyaluronic Acid (HyA)-based hydrogels provide a promising scaffold for cell engraftment and angiogenesis, due to tunable mechanical and biological properties. Accordingly, we have developed HyA hydrogels to assess the effects of adhesion ligand presentation, matrix metalloproteinase (MMP) labile crosslinkers, material moduli, and endogenously synthesized growth factor sequestering capacity on transplanted stem cell survival. Previous studies show that HyA hydrogels support vascular network formation via optimized ligand density, heparin-mediated growth factor presentation and MMP–mediated degradation kinetics [1,2]. To further assess the mechanical tunability of the hydrogel system, we altered the molecular weight of HyA macromers to modulate cell survival, vessel formation, and degradation. In this study, we quantified the mechanical and degradation effects associated with using different number-averaged molecular weights (M_n) of HyA hydrogels.

HyA hydrogels were synthesized using previous methods [1]. Briefly, HyA derivative containing grafted acrylate groups (AcHyA) was generated by reacting sodium hyaluronate of three different M_n's (60kD, 500kD and 1MD) with acryloxysuccinimide. Separately, thiolated heparin (Heparin-SH) was synthesized, along with AcHyA containing RGD cell binding motif (AcHyA-RGD). Hydrogels were made by crosslinking a precursor solution of AcHyA, AcHyA-RGD, Heparin-SH and growth factors with bis-cysteine containing MMP cleavable peptide. Cell assays were conducted using encapsulated vascular cells derived from human induced pluripotent stem cells in precursor solution. Stiffness and gelation time of crosslinked hydrogels were determined using time sweeps from oscillatory rheometry. Degradation of crosslinked hydrogels in Type I collagenase solution was measured by determining mass loss of swelled samples at 24-hr time intervals.

Varying M_n of HyA hydrogels led to changes in viscoelastic properties (G', G") and degradation rates. Between the highest and lowest M_n's, we found gelation times of 2.65 +/- 0.77 min for 1MD samples and 7.45 +/- 1.9 min for 60kD. Crosslinked 1MD hydrogels had G' of 1451 +/- 160 Pa, while 60kD showed G' of 260 +/- 48 Pa. Furthermore, higher M_n hydrogels degraded more slowly; initial studies showed that 1MD hydrogels took 14 days to fully degrade, while 60kD took 4 days. Consequently, these changes in moduli, gelation and degradation times affected the behavior of encapsulated vascular cells, as observed in initial 3D culture studies. Future studies will explore how the resultant variation in stiffness and viscosity will affect vascular network formation in more complex 3D angiogenesis models.

[1] Jha A, et al. (2015). Biomaterials 47:1-12.
[2] Jha A, et al. (2016). Biomaterials 89:136-147.

11:45 AM BM04.04.10
Long-Term Maintenance of an Mesenchymal Stem Cell Niche Through Self-Assembly of Injected Monodisperse Hydrogel Particles into a Microporous Scaffold Jaekyung Koh[1], Donald Griffin[2], An-Chieh Feng[3], Thomas Horn[1], Michael Margolis[1], Hamed Haddadi[1], Tatiana Segura[5], Philip Scumpia[3] and Dino Di Carlo[1, 4]; [1]Department of Bioengineering, University of California, Los Angeles, Los Angeles, California, United States; [2]Department of Biomedical Engineering, University of Virginia, Charlottesville, Virginia, United States; [3]David Geffen School of Medicine, University of California, Los Angeles, Los Angeles, California, United States; [4]Jonsson Comprehensive Cancer Center, University of California, Los Angeles, Los Angeles, California, United States; [5]Biomedical Engineering, Duke University, Durham, North Carolina, United States.

Mesenchymal stem cell (MSC) therapies hold promise for numerous intractable diseases arising from their immunosuppressive and tissue repair properties. However, systemic administration, the most common delivery method in clinical settings, suffers from poor survival and engraftment of transplanted stem cells. Moreover, loss of stem cell properties of the engrafted cells due to the unique environment in regenerating tissue can decrease therapeutic effects. Local delivery of MSCs using biomaterials has shown promising results. Ideal materials should deliver cells in a minimally invasive manner, enhance viability during and after transplantation as well as creating a microenvironment that stimulates self-renewal and expansion. Naturally-derived extracellular matrix (ECM) components, including Matrigel, have been applied but their high variability potentially impacts the reproducibility of the therapy. Artificial hydrogels with a high degree of tunability enable reliable and precise control of biophysical properties. However, the nanoscale porosity confines cells, thus interfering with migration, proliferation and cell-cell communication. Introduction of microscale porosity (e.g. through introduction of porogens) often compromises the ability to modulate mechanical properties and injectability of the hydrogel scaffold independently.
Here, we describe the development of an injectable microporous stem cell niche using microfluidically-generated monodisperse modular hydrogel particles. Delivered with MSCs, these particles are enzymatically assembled *in situ*, generating a highly-controlled interconnected microscale pore space, where cells can easily migrate and proliferate, leading to enhanced survival of transplanted cells. Moreover, the properties of the particles, including stiffness, degradability and cell binding motif concentration, can be modulated independently. By modulating these properties, the microporous scaffold can mimic the natural ECM for high maintenance of stem cell properties.
The scaffold made from these monodisperse (CV < 5%) particles yielded increased expansion and retention of embedded MSCs *in vitro* and *in vivo*. The diffusivity through the scaffolds for small molecules (0.3kDa) and large proteins (70kDa) was 70% and 40% respectively of that through pure buffer. Bone-marrow-derived C57BL/6 mouse MSCs incorporated in the scaffold expanded at a rate 7-fold faster than cells in the nanoporous scaffolds over the course of two weeks. The retention of subcutaneously injected MSCs *in vivo* was 4~8 times higher than with the nanoporous scaffold or PBS over a two-week period. We also demonstrated that scaffolds with 2.5kPa modulus and 2.5mM RGD concentration particles showed the highest retention of stemness of MSCs. Therefore, this new class of injectable microporous biomaterial should accelerate the development of stem cell therapies by creating an injectable microporous niche that enhances function.

SESSION BM04.05: Polymeric Biomaterials for Regenerative Engineering I
Session Chairs: Guillermo Ameer and Junji Fukuda
Wednesday Afternoon, November 28, 2018
Sheraton, 2nd Floor, Independence West

1:45 PM *BM04.05.01
Nanoengineered Biomaterials for Regenerative Medicine Akhilesh K. Gaharwar; Texas A&M University, College Station, Texas, United States.

Two-dimensional (2D) nanomaterials have gained unprecedented attention due to their unique atomically thin, layered, and well-defined structure. As the dimensions of 2D nanomaterials are only a few nanometers thick, they interact with biological moieties in a unique way and have raised exciting questions about their interactions with cellular components. We have used next-generation sequencing technology (RNA-seq) to understand the effect of a synthetic 2D nanomaterials (nanosilicates) on human stem cells at the whole transcriptome level. Our results identify more than 4,000 genes that are significantly affected, and several biophysical and biochemical pathways that are triggered by nanosilicates treatment. This approach in understanding nanosilicates-cell interactions, illustrates how change in transcriptomic profile can predict downstream effects following nanomaterial treatment. Based on our transcriptomic data, we will demonstrate the application of nanosilicates towards bone and cartilage tissue engineering. The high surface area and charged characteristics of 2D nanomaterials is leveraged for sustained and prolonged delivery of pro-angiogenic molecules to stimulate angiogenesis. We have also evaluated the application of nanosilicates in the emerging field of 3D bioprinting to print complex organ and tissue.

2:15 PM BM04.05.03
Hybrid Nanogels with Self-Integrated Plasmonic Nanoparticles for Light-Induced Molecular Delivery Seungki Lee, Jung A Kwon, Yunjeong Lee, Hyo Sil Kim, Chang Min Jin and Inhee Choi; University of Seoul, Seoul, Korea (the Republic of).

Nanoscaled hydrogel (nanogels) have drawn much attention as one of the promising materials in developing drug delivery system. Owing to superior water swelling property, nanogels are profitable to load large amounts of molecules including drugs, growth factors, and genes. Recently, many efforts have been made to develop hybrid nanogels which exhibit multi-functions and stimuli-responsive characteristics as well as encapsulation of the molecules. Herein, we present a novel method for synthesizing light (or heat)-responsive hybrid nanogels composed of biocompatible polymers, plasmonic gold nanoparticles, and thermo-responsive linkers (e.g., N-isopropyl acrylamide or N-vinylcaprolactam). We systemically characterize the physicochemical properties including size, shape, surface functionality, and light and thermo-responsive properties. By using the optimized hybrid nanogels, we demonstrate light-induced releases of the encapsulated molecules via photothermal-conversion effect of the embedded gold nanoparticles. Furthermore, we achieve the successful light-induced molecular delivery into the live cells. We envision that the proposed light-responsive hybrid nanogels would be beneficial materials in developing new drug delivery systems and further applying to regenerative engineering.

2:30 PM BREAK

3:30 PM *BM04.05.04
Optical Cell Manipulation Using Biomaterial-Based Photodegradable Hydrogels Shinji Sugiura; Biotechnology Research Institute for Drug Recovery, National Institute of Advanced Industrial Science and Technology, Tokyo, Japan.

We have developed a photocleavable crosslinker for preparing photodegradable hydrogels by means of a one-step mixing reaction between the crosslinker and biocompatible polymers. We could control the chemical property of the photodegradable hydrogel by choosing the base polymer. We could prepare the biomaterial-based photodegradable hydrogels, on which cells adhered, and in which cells were encapsulated. So far, we have demonstrated cell separation from three-dimensional culture environment, cell micropatterning, and perfusion culture in the microfluidic devices. These studies will create new opportunities as novel the cell manipulation technologies in science and industry.

4:00 PM *BM04.05.05
Promoting Endogenous Tissue Repair with Materials Tatiaba Segura; Duke University, Durham, North Carolina, United States.

We believe that all tissues in the body have the capacity to repair through local stem or progenitor cells, but that due to unfavorable environmental

conditions during the normal healing process they are not able to do so. We investigate the engineering of materials to "unlock" the regenerative capacity of damaged or diseased tissue to promote repair. Our general strategy is to combine our biomaterials engineering with designing materials that promote the formation of a space filling vascular plexus that could serve as part of a reparative niche directly at the wound site. In addition, our materials are engineered to modulate the immune system to decrease scarring and remove inhibitors to regeneration. The idea is that this vascular plexus would lay the groundwork for the recruitment of endogenous stem cells located in the local tissue surrounding the damaged area and immune modulation would generate an environment that would foster repair rather than scaring.

4:30 PM BM04.05.06

Stress/Strain and Degradation Tests on Nanofibrous Scaffold for Cardiovascular Regeneration Amanda Kennell, Anthony Brayer and Andrei Stanishevsky; University of Alabama in Birmingham, Birmingham, Alabama, United States.

Cardiovascular disease-related mortalities are predicted to rise to 23.3 million per year worldwide by 2030 [1] with the main cause being cardiovascular diseases (CVD) [2]. The current procedures to correct CVD are time consuming, because a surgeon needs a healthy artery to perform bypass surgery which is not readily accessible. However, if hospitals had a healthy blood vessel scaffold in storage the patient's recovery time would decrease. Such a scaffold must be biodegradable, easily manufactured, and environmentally safe.
To make this scaffold, AC electrospinning is used to create nanofibrous sheets (NFs) from a base polymer solution (fish skin gelatin, FSG). Additions of polysaccharides (Chitosan and Cellulose) are added in to strengthen the scaffold. These NFs are then thermally crosslinked and placed in synthetic body fluid (SBF). In increments, over a period of 3 weeks, the NFs sheets are removed to test their degradation and strength.
Preliminary data has shown these NFs to last for two weeks in SBF at 37°C. These NFs were able to withstand a range of applied forces from 30 to 70mN at 80%-100% elastic deformation which translates into 0.15-0.85MPa. To increase the NFs strength a larger percentage of polysaccharides is being used. These NFs will further be seeded with endothelial cells.
These NFs meet the requirements for the blood vessel scaffold. They are a "green" scaffold (made from an aqueous solution), easily manufactured (AC electrospinning is a "high yield" technique, 15-30g/h productivity), and "biodegradable". It is expected that these nanofibrous tubular scaffolds will help reduce the mortality rate, patient recovery time, and need for organs at a lower price and higher acceptance rate. Further studies *in vitro* and *in vivo* are planned to evaluate the feasibility of the approach.
[1] C.D. Mathers, of global mortality and burden of disease from 2002 to 2030. PLoS Med 3, e442, 2006.
[2] E. J. Benjamin, S. S. Virani. P. Muntner., of Heart Disease and Stroke Statistics—2018 Update p. e247 (American Heart Association, Texas, 2018)

4:45 PM BM04.05.07

Short-Term Evaluation of Antithrombogenecity of a PMPC-Grafted PEEK Mechanical Heart Valve in a Porcine Aortic Valve Replacement Model Yusuke Kambe[1], Atsushi Mahara[1], Kyoko Fukazawa[2], Yihua Liu[1], Kazuhiko Ishihara[2] and Tetsuji Yamaoka[1]; [1]Department of Biomedical Engineering, National Cerebral and Cardiovascular Center Research Institute, Suita, Japan; [2]Department of Materials Engineering, School of Engineering, The University of Tokyo, Bunkyo-ku, Japan.

Improved antithrombogenecity of mechanical valves is desired to decrease the risk of thromboembolism and thrombosis and to reduce the dosage of anticoagulation with a vitamin K antagonist (e.g., warfarin). For several mechanical valves, design-derived features are responsible for their improved antithrombogenecity. However, it remains unclear whether material-derived features provide a practical level of antithrombogenecity of mechanical valves. Here we studied the effect of a bileaflet valve made of poly(ether ether ketone) (PEEK) with a poly(2-methacryloyloxyethyl phosphorylcholine) (PMPC)–grafted surface (PEEK-*g*-PMPC). PEEK has a benzophenone-like structure in its unit, where radicals can be generated by photoirradiations. Thus, PMPC, which is a well-known antithrombogenic polymeric material, was directly graft-polymerizaed on the PEEK valve surface by a 27-mW/cm^2 ultraviolet light irradiation at 60^{o}C for 30 min. Surface characterization, such as Fourier-transform infrared analysis, X-ray photoelectron spectroscopy, and transmission electron microscopy, showed the existence of a 250-300-nm-tihck PMPC layer on the PEEK-g-PMPC valve surface. Porcine aortic valve replacements were conducted using neither an anticoagulant nor an antiplatelet agent and the animals were observed up to 26 h. It was shown that the PEEK-*g*-PMPC valve opened and closed normally with an allowable transvalvular pressure gradient. Unlike an untreated PEEK valve, no thrombus formed on the PEEK-*g*-PMPC valves on gross anatomy examination in addition to the absence of traveling thrombi to the kidney and lung tissues. This improved antithrombogenicity of the valve was attributed to the PMPC layer on the valve surface. Therefore, material (PEEK-*g*-PMPC)–derived antithrombogenecity appeared to decrease the risk of thromboembolism and thrombosis for patients with mechanical valves. However, further studies are required to improve the antithrombogenecity of the PEEK-*g*-PMPC valve because fibrous fouling was still observed on the leaflet. The authors thank Prof. Hiroshi Tanaka, Prof. Kenji Minatoya, Dr. Sachiro Kakinoki, and Dr. Masayuki Kyomoto for their assistance.

SESSION BM04.06: Poster Session II: Biomaterials for Regenerative Engineering
Session Chairs: Josephine Allen, Guillermo Ameer, Gulden Camci-Unal and Junji Fukuda
Wednesday Afternoon, November 28, 2018
8:00 PM - 10:00 PM
Hynes, Level 1, Hall B

BM04.06.01

Enhanced Cell Adhesion on N-Cadherin Modified Graphene Oxide Stimulates Neuronal Growth and Intracellular Transport Ellen Qin[1], Mikhail Kandel[1], Evangelos Liamas[2], Zhengyu Zhang[2], Martha U. Gillette[1], Gabi Popescu[1], Deborah Leckband[1] and Hyunjoon Kong[1]; [1]University of Illinois at Urbana Champaign, Urbana, Illinois, United States; [2]University of Birmingham, Edgbaston, United Kingdom.

In the nervous system, cell-cell contacts are predominantly mediated via the homophilic interactions between N-cadherins on adjacent cells. Recombinant N-cadherin molecules are often attached to cell culture substrates via physical adsorption to recapitulate the neural environment. However, those methods typically involve more complicated surface chemistries or result in poor cell adhesion. To address this issue, we hypothesized that the additional noncovalent interactions of graphene derivatives with proteins would enhance the specific activity of immobilized recombinant N-cadherin. Results showed a substantial enhancement of neural cell adhesion on recombinant N-cadherin physisorbed on graphene oxide (GO) and reduced graphene oxide (rGO) relative to N-cadherin on glass, with improvements in the microstructure and physiological activity of neural networks. Biophysical measurements showed the mean adhesive force between AFM tips and N-cadherin coated GO or rGO flakes was ten times higher than that measured with N-cadherin physisorbed to uncoated glass. The latter behavior correlated with increased dendritic arborization and neural network formation on the GO- or rGO substrates, relative to cells on glass. Furthermore, intracellular mass transport was higher along the neurites on N-cadherin coupled to GO- or rGO-coated glass, as visualized using spatial light interference microscopy. The results of this study will be broadly useful for recreating active neural tissues *in*

vitro and for improving our understanding of the development, homeostasis, and physiology of neural networks.

BM04.06.02
Proliferation Acceleration of Mesenchymal Stem Cells on Nanostructured Surfaces Hyeji Park and Jin Seok Lee; Sookmyung Women's University, Seoul, Korea (the Republic of).

Biophysical properties of the microenvironment, such as nanotopography, modulate proliferation and differentiation of stem cells. Cells directly probe and respond to the physicomechanical properties of their extracellular environment through adhesion complexes. Many previous studies have been investigated cell behavior such as cell adhesion, spreading area, cell proliferation, and migration using various topographical cues and the results have indicated topographical confinement by nanostructured architecture affects cellular behavior significantly. Up to date, overall control of cellular response has been carried out by cell isolation in the structure through geometry. However, there is little research about the effect of surface topography on cellular behavior. It is essential to study the surface topography by controlling the size, shape and density of the nanostructure.
In this study, we investigated the effects of the nanostructured silica bead arrays on cellular behavior, particularly proliferation. Human mesenchymal stem cells (hMSCs) were selected to investigate the nanotophological effects on cell proliferation. In the first step, the silica beads were synthesized by Stöber Method by controlling the amount of reagents or injection rates. The silica bead arrays can be achieved by utilizing a rubbing method, which is simple and fast, to obtain a monolayer of silica beads with various diameter sizes from 200 nm to 1900 nm. In the second step, the hMSCs were seeded onto the flat glass and silica bead arrays and then cultured for 24 hours to figure out the nanotopological effect on the cellular behavior. The results have shown that the cell spreading areas and proliferation on the different nanotopological substrates were significantly different.
This paper will discuss how the silica bead arrays influence hMSC cellular behavior and optimize cell proliferation within the system. This study could be extended to serve as a model for stem cell therapy in terms of cell proliferation.

BM04.06.03
Skin-Penetrating Peptide-Based Micelle for Transdermal Drug Delivery Do Hyun Bae[1], Yong Ho Kim[2, 1], Jin-Chul Kim[3] and Ki Sung Kang[4]; [1]Department of Biomedical Engineering, Sungkyunkwan University, Suwon-si, Korea (the Republic of); [2]SKKU Advanced Institute of Nano Technology (SAINT), Sungkyunkwan University, Suwon-si, Korea (the Republic of); [3]Natural Products Research Institute, Korea Institute of Science and Technology, Gangneung-si, Korea (the Republic of); [4]College of Korean Medicine, Gachon University, Seongnam-si, Korea (the Republic of).

Transdermal drug delivery has been extensively studied to overcome the enzymatic degradation of drugs delivered via oral administration because it not only passes metabolic digestion passage, but also has a possibility to sustain the release of drugs. However, low permeability of stratum corneum inhibits deep penetration of drugs into target area and reduces percutaneous adsorption of drugs. Therefore, a new carrier system that can efficiently transport drugs into the stratum corneum would greatly enhance transdermal drug administration. Here, we demonstrated a formation of skin-penetrating peptide-based micelle for efficient transdermal drug delivery. Skin-penetrating peptides (SPPs) can be used as an excellent transdermal drug transporter due to its biocompatibility and exceptional cell-penetrating ability. We examined various cell-penetrating peptides (CPP) that adapt adsorptive-mediated transcytosis to verify their skin-cell permeability and identified one CPP that showed high cellular uptake inside the dermal papillae cells. The newly discovered SPP was further modified by conjugating an aliphatic chain at the N-terminal of the peptide to induce self-assembly for lipopeptide-based micelle formation. The micelle was founded to achieve enhanced thermodynamic stability compared to SPPs only, which suggests that it can maintain its construction in biological condition. The self-assembled micelle could securely encapsulate the minoxidil, a drug for hair regeneration, and increased cellular uptake of the drug into the dermal papillae cells. We believe that our new transdermal drug delivery system can be applied in the field of cosmetics and pharmaceuticals to increase skin administration efficiency of functional components.

BM04.06.04
RF Coupling of Interdigitated Electrode Array on Aerogels for *In Vivo* Nerve Guidance Applications Jacob Hadley, Jack Hirschman, Bashir Morshed and Firouzeh Sabri; University of Memphis, Memphis, Tennessee, United States.

Aerogels are light-weight porous materials that can tolerate the processing steps required for designing and creating an interdigitated electrode (IDE) array using sputter coating with a shadow mask where the aerogel can be utilized as a substrate. Previous studies have shown the biostability and biocompatibility of polyurea crosslinked silica aerogels both in vivo and in vitro and have demonstrated the potential use of aerogels in biomedical applications. In vitro studies have shown that in the presence of an applied electric field neurites regeneration rate was greater on crosslinked silica aerogels than on tissue culture petridish that served as control. Currently, Epineural suturing and nerve grafting are the gold standards for surgical reconstruction of severed nerves. However, because these techniques rely on passive mechanisms for reapproximating the distal and proximal terminals they often lead to partial or limited recovery leaving room for improvement. The present study investigates the feasibility of a wireless aerogel –based electrically-stimulating implant intended for nerve repair applications. Here the authors report on a transcutaneous RF coupling between a primary coil (external) and a secondary coil (internal) connected to an IDE array consisting of eleven interdigitated fingers, created on a silica aerogel substrate. The coupling strength was tested both in air and in an animal model, as a function of distance and will be reported. Multiple primary coil geometries were tested and their coupling efficiencies were evaluated. Results report on the optimum coil geometry and spacing and their efficacy in a cadaver model.

BM04.06.05
Nanostructured Biomaterials for Functional Neurons Debika Debnath[2], Krishnan Gopal Jain[1], Manu Dalela[1], Sonali Rawat[1], Amtoj Kaur[1], Neha Kaushik[1], Ankarao Kalluri[2], Bhushan Dharmadhikari[3], Prabir Patra[2] and Sujata Mohanty[1]; [1]Stem Cell Facility, All India Institutes of Medical Sciences, New Delhi, India; [2]Biomedical Engineering, University of Bridgeport, Bridgeport, Connecticut, United States; [3]Electrical Engineering, University of Bridgeport, Bridgeport, Connecticut, United States.

We fabricated Polycaprolactone-graphene (PCL-G) biomaterials scaffolds by stretching graphene dispersed viscoelastic PCL solution uniaxially under an applied voltage with an aim to provide microenvironment in-vitro for differentiation and proliferation of functional neurons from mesenchymal stem cells (MSCs). Varied amount of graphene (.005 .01, .05 wt percentage of graphene) was dispersed in PCL matrix for scaffold formation. We induced MSCs to differentiate into neurons by taking advantages of topological and electrical effect of scaffold, and biochemical effect of FGF2 and Oxysterol. SEM images of PCL-G scaffolds were taken to study the effect of the scaffolds on cell morphology. We performed confocal imaging to confirm differentiation of MSCs into neurons. Confocal images of Vinculin and FAK on MSC grown on scaffolds were observed. We measured Contact angle of water on scaffolds to determine hydrophobicity and hydrophilicity of scaffold.: Calcium ions being the most important ions for regulating biological processes we did the Ca+ ion imaging . Cell culture and in vitro cell adhesion studies showed that PCL-G scaffolds was most effective for promoting stem cells adhesion and spreading. Microscopic images show that the PCL-G fibers are structurally similar to ECM proteins like collagen, laminin and fibrils. This work also demonstrates the key role of graphene in aligning neurons. With its optimum dose as .05 wt percentage as filler in composite scaffolds significantly provides permissive surfaces for protein and cell adhesion as well as electrically stimulate axonal growth that increases over all biological responses. Thus, we envisaged that such a platform could serve as a powerful tool for developing future therapies for any diseases and injuries of the spinal cord.

BM04.06.06
Pulsed Laser Deposition and Biocompatibility of Titanium Nitride Coatings Meenakshi Singh[1] and Svitlana Fialkova[2]; [1]STEM Early College at NC A&T, Greensboro, North Carolina, United States; [2]Mechanical Engineering, North Carolina Agricultural and Technical State University, Greensboro, North Carolina, United States.

The purpose of this study is to evaluate the effect of Titanium Nitride (TiN) thin films deposited on magnesium substrates using a pulsed laser deposition method. The application of a TiN coating on magnesium implants has the potential to remove the necessity for a second surgery for patients undergoing the process of healing. The coating developed has been found to help reduce the degradation rate of the magnesium implant such that the Mg would remain stable for the time required for healing before beginning to degrade. Magnesium was chosen as a substrate due to its biocompatible and biodegradable properties. TiN was chosen due to its stellar properties of high melting point, good diffusion barrier, high hardness and good electrical conductivity, and scattered reports in the literature about its biocompatibility. The crystallographic orientation and surface morphology of the films were studied using X-ray diffraction (XRD) and scanning electron microscope (SEM). The hydrophilic nature of the films was investigated using contact angle measurements. Preliminary results on the biological behavior of the TiN coated Mg substrates suggest that TiN is a biocompatible material and has great promises in biological applications.

BM04.06.07
Magnetic Nanoparticles as a Therapeutic Biomaterial in Magneto–Ultrasonic Hyperthermia Arkadiusz Jozefczak, Katarzyna Kaczmarek and Tomasz Hornowski; Institute of Acoustics, Adam Mickiewicz University, Poznan, Poland.

In medicine, a controlled increase in temperature up to 41–45°C is called hyperthermia. It induces heat in cancer cells which leads to their weakening. Weakened cells are therefore more susceptible to radiotherapy or chemotherapy. The heating can be induced by means of ultrasonic waves or magnetic field. The effectiveness of ultrasound therapy can be significantly improved by using so-called sonosensitizers, for example, magnetic nanoparticles that locally increase the attenuation of the ultrasonic wave [1]. Magnetic particles can also be used for selective induction of heat by an externally applied alternating magnetic field (AC) [2]. Recently, there has been a great interest in the application of multimodal thermal treatments. Magnetic and ultrasonic hyperthermia may work synergistically to produce a more efficient treatment. This sonomagnetic therapy is a promising new technique based on the synergistic interactions of ultrasound and AC magnetic field. The presence of magnetic nanoparticles also improves contrast in both ultrasonic and magnetic resonance imaging, which facilitates control of temperature during hyperthermia therapy. Magnetic nanoparticles can act as theranostic nanoparticles.
In the study, we have focused on evaluating the influence of magneto–ultrasonic heating on phantom temperature in the presence of superparamagnetic iron oxide nanoparticles (SPION). The experiments are performed with the use of agar tissue mimicking phantoms doped with magnetic sonosensitizers. Integrated treatment by means of simultaneous application of a focused ultrasound wave and alternating magnetic field (bimodal sonomagnetic hyperthermia) leads to a higher temperature increase, which enables more precise control over the heating process. Magneto–ultrasonic heating creates very innovative, promising approach which has an application potential to treat cancer at a lower SPION concentration. We demonstrate that bimodal stimulation of nanoparticles provides better heating efficiency.
References:
[1] K. Kaczmarek, T. Hornowski, M. Kubovcíkova, M. Timko, M. Koralewski, A. Jozefczak, Heating Induced by Therapeutic Ultrasound in the Presence of Magnetic Nanoparticles, ACS Applied Materials & Interfaces, 10 (2018) 11554-11564.
[2] A. Jozefczak, B. Leszczynski, A. Skumiel, T. Hornowski, A comparison between acoustic properties and heat effects in biogenic (magnetosomes) and abiotic magnetite nanoparticle suspensions, J. Magn. Magn. Matter., 407 (2016) 92-100.

BM04.06.08
Antibacterial Coating on Contact Lens Using Polyethylenimine Based Polymers Suresh Kumar Raman Pillai, Yogesh Shankar Vikhe, Zheng Hou, Sheethal Reghu and M. B. Chan-Park; Nanyang Technological University, (NTU) Singapore, Singapore, Singapore.

Microorganisms such as bacteria that grow on the surface of contact lenses cause irritation to the eyes. These infections that are difficult to treat and can cause to vision loss. Besides the availability of new contact lens materials and better cleaning solutions, the risks of the above conditions remains the same for many years. Silver coating have been commonly studied, but the material can leach out of the lenses and their antimicrobial effect fades over time. In this report, we have synthesized PEI-PEGMA polymer and used for the application of an antimicrobial coating on contact lenses in order to prevent bacterial infection and reduce the risk of extended wear. Different types of failures such as (1) reduction of coating adherence to the substrate of contact lens and (2) disintegration of interface between coating and substrate of contact lens exists. To improve coating formulations to resist failures caused by the applied normal load with fingers of users, monomer (3-Acrylamidopropyl) trimethyl ammonium chloride (AMPTMA) or [2-(Methacryloyloxy) ethyl] dimethyl-(3-sulfopropyl) ammonium hydroxide (SBMA) was added to the antimicrobial polymer PEI-PEGMA.

Two different coating strategies were adopted to form PEI-PEGMA-AMPTMA or PEI-PEGMA-SBMA polymeric film on the surface of the contact lens. In the first method, the silicone contact lenses were treated with ozone to activate the surface and form the peroxide group. Various concentrations of PEI-PEGMA and AMPTMA/SBMA dissolved in DI water and degassed the solution using Argon for 45 min. The contact lenses were immersed into the solution and the radical initiative polymerization reaction was activated by the addition of Ammonium iron(II)sulfate. The polymerization reaction was carried out at 37°C for 24 hrs. The lenses were thoroughly washed with DI water and isopropanol (IPA) before doing the antibacterial testing. Antibacterial activity of the coated lenses were tested with 1x 10^7 CFU of Methicillin-resistant Staphylococcus aureus (MRSA). The coated contact lenses were found to be effective for antibacterial activity with killing rate > 99.9 % and log reduction of 3.20. Cell viability for the coated lenses were found to be 80 % using MTT assay protocol. In the second method, the radical initiation was activated thermally. The lenses were treated with ozone to form peroxide group on the surface of contact lens. Different concentrations of PEI-PEGMA and AMPTMA/SBMA dissolved in 90 % DI water and 10 % IPA mixture. The polymer solution was degassed for 45 min and immersed the lenses into the solution and polymerization reaction was carried out at 90 °C for 3 hrs. The coated lenses were washed several times with DI water and IPA and tested for antibacterial activity. The coated contact lenses show antibacterial activity for MRSA with killing rate >99.9 % and log reduction of 3.98.

BM04.06.09
Arterial Revascularization by Peptide-Modified Acellular Graft Evaluated in Long-Term Miniping and Goat Transplantation Model Atsushi Mahara[1], Kentaro Kojima[1, 2], Maria Munisso[1], Yoshiaki Hirano[2] and Tetsuji Yamaoka[1]; [1]National Cerebral and Cardiovascular Center Research Institute, Suita, Japan; [2] Kansai University, Suita, Japan.

Acellular blood vessels are promising materials as a tissue-engineered graft. However, the tissue is mainly composed of collagen, and there is a risk to induce the blood coagulation and the rupture by matrix degradation after transplantation. Therefore, revascularization of the acellular tissue by autologous cells is required. In our previous work, we succeeded in proving a good patency of tissue-engineered acellular small diameter long bypass graft (inner

diameter of 2 mm and the length of 20-30 cm) by modification with the REDV peptide, which is known as an integrin α4β1 ligand, in minipig femoral-femoral bypass model [1]. After transplantation for one day, the surface was covered with cells. The cells formed cell layer after three days, and the cells were stained with anti CD34 and Flk-1 antibodies. After 7 days transplantation, 94% and 99% of cells expressed CD31 and CD34, respectively. Moreover, 53% and 62 % of cells expressed CD105 and Flk-1, respectively. Moreover, we previously reported that the luminal surface at not only center part of the graft but also near the anastomotic site was covered with the endothelial-like progenitor cells. Although the reconstruction of the media layer is also important for arterial revascularization, it was not clearly illustrated in the previous report. In this study, we evaluated the revascularization process including not only endothelial but also vascular smooth muscle layer by long-term minipig and goat transplantation model.
Ostrich carotid artery was decellularized, and the luminal surface was modified with the peptide [1]. To evaluate the response for 3, 12 months, the graft was transplanted into the femoral and median artery of miniping and goat as orthotopic graft, respectively. After experimental periods, the center part of the graft was extirpated. Luminal surface was evaluated by histological staining using HE, vWE, as well as primary antibody against CD31, CD34, CD105, and Flk-1. For evaluation of the medium layer, the tissue was stained with antibody against α-smooth muscle actin (αSMA)
After three months, the luminal layer expressed vWF, CD31 and CD105 but not CD34 and Flk-1. The αSMA positive cells existed in media layer after 3 months transplantation. When the graft was transplanted to goat median artery for 12 month, vWF positive cell layer was observed on the luminal surface, and αSMA positive cells existed as same as the results for three month transplantation. Although the cell density in media layer was sparse as compared with a native blood vessel, regenerative vessels having intima and media layer was reconstructed by the graft. From these results, we found that the graft could regenerate the arterial vascular tissue in three months.
Mahara A., et al. Biomaterials 58 (2015) 54-62.

BM04.06.10
Sub-Compartmentalized Microreactors as Cell Implants for Conducting Enzimatic Cascade Reactions Maria Godoy Gallardo, Cédric Labay and Leticia Hosta-Rigau; Department of Micro- and Nanotechnology, Technical University of Denmark, Kgs. Lyngby, Denmark.

Malfunction at cellular level is the primary cause for many diseases and, as such, creating micro- and nanoreactors that can act intracellularly to substitute for missing or lost cellular function, will add a novel paradigm in biomedicine.Those micro/nanoreactors acting as cell implants are also known as artificial organelles and are expected to be particularly useful for enzyme replacement therapy since, malfunctioning enzymes is the main cause for organelle dysfunction.
However, the delivery of enzymes has been proven so far very challenging and, thus, so far it has shown very limited success. The reason being that enzymes are very fragile entities prone to degradation in the bloodstream. This fact makes them difficult to reach the target site, which is usually a diseased cell, and remain active for a reasonable amount of time. Thus, the creation of artificial organelles encapsulating enzymes that will protect them first from degradation in the bloodstream and then from the intracellular degrading contents, will greatly diminish current limitations of enzyme replacement therapy.
Biological organelles operate by conducting multiple sets of (enzymatic) reactions with high specificity and accuracy without cross-contamination. They can separate multiple functions while protecting internal contents thanks to compartmentalization. This compartmentalization can be clearly seen in the Golgi apparatus or the mitochondria, where several enzymes for the citric acid cycle are situated in the intermembrane space.
Thus, herein, with the aim to design a highly-sophisticated artificial organelle we present a carrier containing multiple compartments consisting of polymeric capsules entrapping thousands of liposomes and gold nanoclusters. Liposomes are chosen as the subcompartments since, by means of their lipid bilayer which makes them biomimetic, are ideal candidates to encapsulate fragile biomolecules such as enzymes protecting them from misfolding or denaturation. The polymer carrier shell possesses the structural integrity and it is able to protect the liposomes from degradation and the gold nanoclusters, thanks to their fluorescent properties, allow us to detect and tract the artificial organelles inside the host cell.
We demonstrate preservation of functionality of our artificial organelles by encapsulating two different enzymes within different liposomes and conducting an enzymatic cascade reaction inside the cell. In particular, artificial organelles loaded with the enzymes glucose oxidase (GOx) and horseradish peroxidase (HRP) are internalized by macrophages. Next, upon incubation with β-D-glucose, it gets converted by GOx into β-D-gluconolactone and hydrogen peroxide. The latter is utilized by HRP to convert the Amplex Red probe into the fluorescent product resorufin which is detected by fluorescent spectroscopy to confirm that the enzymatic cascade reaction has successfully taken place.

BM04.06.11
In-Flow Preparation of Collagen Sheets with Tunable Fibril Alignment for the Engineering of Arterial Substitutes That Recapitulate Blood Vessel Microstructure David Miranda-Nieves[1, 3], Shashi Malladi[2], Daniel Wong[3], Constantine Tarabanis[3], Carolyn Haller[3], Axel Guenther[2] and Elliot Chaikof[S, 1]; [1]Massachusetts Institute of Technology, Boston, Massachusetts, United States; [2]University of Toronto, Toronto, Ontario, Canada; [3]Harvard University, Boston, Massachusetts, United States.

Lower extremity peripheral arterial disease (PAD) affects up to 15% of the population over 65 years old. The number of endovascular and bypass operations has doubled in the past decade, yet outcomes after surgical and catheter-based interventions remain compromised with low patency rates. Tissue-engineering strategies have been explored as alternatives; however, no clinically available graft exists. The main limitation of most approaches is failure to recapitulate the blood vessel microstructure, and, as a consequence, native physiological properties. Considering that the arterial wall is a circumferentially aligned fibrous matrix, the capacity to create highly structured, oriented structures is critical for the generation of physiologically responsive arterial substitutes.

Over the years, various methods have been designed to influence the organization of self-assembling collagen fibrils, including the use of magnetic fields and fiber spinning. However, none of these approaches represent a one-step, scalable technique for the generation of collagen films with precise control over fibril orientation. Here, we present a solution that leverages molecular crowding and hydrodynamic focusing to fabricate ultrathin collagen sheets with tunable fibril alignment and mechanical properties, and our initial attempts to engineer arterial substitutes using these sheets.

Monomeric rat-tail tendon collagen was dissolved in 10 mM HCl, and injected into the middle layer of a multilayered, PDMS-based microfluidic device at varying flow rates. As sheath flow, a polyethylene glycol (PEG) solution was used. The presence of PEG caused molecular crowding, which triggered the gelation of the collagen solution. Hydrodynamic focusing was achieved by modifying the flow rates of the two solutions.

Extruded collagen sheets had thicknesses of 3-8μm and widths of 15-30 mm. The degree of alignment and compaction of the collagen fibrils was controlled, with up to 40% of fibers aligned within ± 5° of one another, and up to 95.5% of compaction. As a result, ultimate tensile strengths of 1.25-13MPa, Young's moduli of 1.3-130MPa, and strains to failure of 15-35% were achieved. Molecular alignment of the collagen sheets induced preferential alignment of vascular smooth muscle cells (vSMC), maintained cellular expression of phenotypic markers, and guided active film contraction.

Efforts involving the controlled assembly of the ultra-thin, robust, anisotropic collagen sheets seeded with vSMC have yield arterial constructs with circumferential collagen fibrils and high density of vSMC that closely recapitulate native blood vessel microstructure. Biomechanical characterization has revealed that the constructs can withstand high pressures and tensile strengths. Ultimately, we believe that these biological and mechanical properties will translate to the mimicry of physiologic responses in vivo.

BM04.06.12
DMD Printed Scaffold for Vascularized Tissue Roya Samanipour[1, 2] and Mina Hoorfar[2]; [1]Harvard-MIT Division of Health Sciences and Technology, Massachusetts Institute of Technology, Boston, Massachusetts, United States; [2]University of British Columbia, Vancouver, British Columbia, Canada.

In this paper, we developed a high resolution stereolithography-based bioprinter (DMD printing) to fabricate micron size of vasculature using CT images. The development of *in-vitro* highly organized/vascularized three-dimensional (3D) complex constructs is of great importance in tissue engineering [1]. While several microfabrication strategies (ranging in micromolding, photolithography, stereolithography, and bioprinting,) have been used, these technologies lack the spatial control for the formation of complex vascular networks [2]. Current 3D bioprinting techniques cannot provide the required printing resolution, throughput, and complex and biomimetic microarchitectural features. Here, we developed a DMD (Digital Mirror Device) 3D printing technology platform capable of rapidly fabricating tissue constructs with smooth features and high cell viability. DMD 3D printing was developed to fabricate variety of microscale resolution vascularized tissue. Blended prepolymer solutions consisting of varying ratios of GelMA (synthesized by the method described by Cha et al [3]) and polyethylene glycol diacrylate (PEGDA, M_n 700, Sigma Aldrich) with LAP photoinitiator (Sigma Aldrich) were tested to optimize the polymer composition for printing. To ensure proper material properties needed to withstand subsequent perfusion, mechanical testing of the different prepolymer formulations was performed using a CellScale MicroSquisher system (Waterloo, ON, Canada). The optimized polymer composition of 7.5 wt % GelMA+ 10 wt % PEGDA 700+ .25 LAP v/v % were used to 3D print 310 um microchannels with fine structural features. Afterwards, Human umbilical vein endothelial cells (HUVECs, Lonza, Portsmouth, NH) were injected into printed microchannel to create enthetolial microchannel construct. To create a uniform monolayer of cells, the whole construct was manually rotated every 1.5 hours after the HUVECs-laden pre-polymer solution was injected into the channel. The seeded cells into the printed structure were cultured for 14 days. The F-actin/Dapi was performed on cultured samples on days 7 and 14. The CD31/Dapi was performed on the cultured samples on days 7 and 14. These results show that the endothelial cells proliferated and uniformly covered the microchannel. PrestoBlue assay was performed to assess proliferation on days 1, 3, 7,10 and 14.
References:
[1] "3D bioprinting for vascularized tissue fabrication." R. Dylan, J. Jia, M. Yost, R. Markwald, and Y. Mei. Annals of biomedical engineering 45, 1, (2017).
[2] "Microfluidic techniques for development of 3D vascularized tissue.", H. Anwarul, A. Paul, N. E. Vrana, X. Zhao, A. Memic, Y. S. Hwang, M. R. Dokmeci, and A. Khademhosseini. Biomaterials 35, 26, (2014).
[3]"Microfluidics-Assisted Fabrication of Gelatin-Silica Core–Shell Microgels for Injectable Tissue Constructs.",
C. Cha, J. Oh, K. Kim, Y. Qiu, M. Joh, S. R. Shin, X.Wang, G. Camci-Unal, K.Wan, R. Liao, A. Khademhosseini, Biomacromolecules, 15, 283, (2014).

BM04.06.13
Block Copolymer Nanoparticles Remove Biofilms of Drug-Resistant Gram-Positive Bacteria by Nanoscale Bacterial Debridement M. B. Chan-Park; Nanyang Technological Univ, Singapore, Singapore.

Bacterial biofilms are the main cause of chronic infections and typically 1000-fold more resistant to conventional antibiotics and/or antimicrobial agents. Hence, many antibiotics, natural antimicrobial peptides and synthetic antimicrobial agents fail to eradicate biofilms. Besides, various antibiofilm agents such as metallic or inorganic nanoparticles have shown the ability to remove biofilm, but they are usually suffered from the problem of toxicity and limited life span. In this paper, we developed a novel antibiofilm polymeric nanoparticles which showed excellent preformed biofilm dispersal ability with non-hemolytic *in vitro* and low *in vivo* cytotoxicity. This polysaccharide-based polymer can self-assemble into nanoparticles which can effectively remove biofilms of multi-drug resistant/clinically relevant Gram-positive bacteria (*i.e.* Methicillin-resistant *Staphylococcus aureus* (MRSA), Vancomycin-Resistant *Enterococci* (VRE) and *Enteroccocus faecalis* (OG1RF)), with efficacy superior or comparable to that of current standard antibiotics. Interestingly, the antibiofilm property of these nanoparticles is not come from the antibacterial effect, but a new mechanism which we term “nanoscale bacterial debridement”. The nanoparticles can diffuse into biofilms and complex with bacterial surface. Further, its hydrophilic polysaccharide corona enhances the solvation of the bacteria/nanoparticle complex leading to detachment of bacteria from biofilm. Our *in vivo* data also shows the nanoparticles can remove the MRSA biofilm by 3.7 order of log reduction, compared to 2.1 order achieved by vancomycin antibiotic. Overall, this novel biofilm removal approach provides exciting opportunities for eradication of multi-drug resistant biofilm and which further may have widespread applications.

BM04.06.14
Short Aromatic Peptide Carriers for the Targeted Delivery of Cranberry Extracts Yasaman Hamedani, Elvira Lou Evangelista, Catherine Neto and Milana Vasudev; University of Massachusetts Dartmouth, Dartmouth, Massachusetts, United States.

In recent years, drug delivery using nanoparticles has gained attention due to their ease of synthesis, high effectiveness due to ease of uptake in cells, increased half-life in systemic circulation and the ability to target certain organs or group of cells. Such nanoparticles can be fabricated from natural or synthetic biopolymers, and lipids. Peptide-based nanoparticles are potential candidates as nanocarriers for the enclosure of various drugs. Peptides have complex structures and biological recognition abilities. Hydrophobic interactions, hydrogen bonding and p-p stacking all lead to formation of stable structures via self-assembly. Depending on the conditions for synthesis, peptides can self-assemble into various structures such as nanotubes, nanofibers and nanospheres. In this study, we have demonstrated the self-assembly of tripeptides to form nanospheres through a process called electrospraying. By optimizing the electrospraying conditions, we were successfully able to form hollow peptide nanospheres which are suitable candidate for delivery of various drugs. Moreover, cationic amphiphilic peptide sequences were designed to form peptide-based micelles for encapsulating various therapeutics drugs. Scanning electron microscopy (SEM) as well as Transmission Electron Microscopy (TEM) techniques have been utilized to investigate the morphology of the fabricated spheres/micelles. Chemical characterization of the samples were performed using Fourier transform Infrared (FT-IR) and Raman spectroscopy. Cranberry extracted compounds such as flavonoids/polyphenols and triterpenoids are amongst the natural therapeutic compounds which recently have gained considerable attentions due to their effectiveness in prevention of cardiovascular, carcinogenic, neurodegenerative and immune diseases. These natural therapeutic compounds are not stable in the biological environment and are likely to be excreted from the body, therefore sufficient therapeutic concentrations are not available at the organs of interest. In this study, we have shown the targeted delivery of these natural compounds to colon cancer cells, by enclosing them in peptide-based nanocarriers. For this purpose, we have studied the encapsulation of these therapeutic agents and their *in vitro* and *in vivo* release behavior as well as the biocompatibility and biodegradability of peptide-based carriers.

BM04.06.15
Delivery of Growth Factors via Bijels-Derived Hybrid Hydrogels Haoran Sun and Min Wang; Department of Mechanical Engineering, The University of Hong Kong, Hong Kong, Hong Kong.

Bicontinuous interfacially jammed emulsion gels (“bijels”) can be used as templates to fabricate bijels-derived structures, which maintain the bicontinuous internal structure of bijels and hence possesses unique properties for various applications. Bijels-derived structures are potential delivery vehicles for the controlled release of growth factors (GFs) in tissue engineering. To apply bijels-derived structures in tissue engineering, the biocompatibility requirement must be met but most existing bijels-derived structures are not biocompatible. Using biocompatible materials such as hydrogels may provide solutions for

solving the problems. Bioactive molecules, particularly GFs, have been used in tissue regeneration. Vascular endothelial growth factor (VEGF) and platelet derived growth factor (PDGF) are often used in the regeneration of tissues such as skin and gastrointestinal tract. Controlled delivery of GFs through appropriate vehicles can promote tissue regeneration. This study investigated the fabrication and characteristics of hybrid hydrogels consisting of biocompatible materials made via bijels for VEGF and PDGF delivery. A modified solvent induced phase separation process was used to fabricate bijels-derived hybrid hydrogel membranes. A ternary liquid mixture was made by adding pure ethanol, hexanedioldiacrylate (HDA), 2-hydroxy-2-methylpropiophenone (HMP), deionized water, Ludox TMA (silica nanoparticle suspension) and CTAB in ethanol A glass plate was immersed in the ternary mixture for forming a mixture film on its surface. It was taken out and then immersed in a water bath to form bijels membranes through phase transition. A high-intensity UV light was applied to cure HDA and harden the bijels structure. Then bijels films were immersed in VEGF- or PDGF-containing Na-alginate solutions, taken out and immersed in $CaCl_2$ solution for crosslinking, forming GF-containing bijels-derived hybrid hydrogel membranes. The bicontinuous microstructure in membranes could be clearly seen under SEM. UV-cured HDA polymer and crosslinked CA-alginate hydrogel formed two continuous phases in the membrane. The diameter of the bicontinuous structure (channel size) could be adjusted by UV-curing time and other parameters. In *in vitro* experiments of as-fabricated hybrid hydrogel membranes, human dermal fibroblasts (HDF) were cultured on membranes whose biocompatibility was assessed using LIVE/DEAD assay. At Day 3, fibroblasts proliferated on membranes and nearly no cell death was observed. MTT assay was used to evaluate cell proliferation on membranes. Results showed good cell proliferation after 1, 2, 3-day culture. Following the established test protocol, the *in vitro* release behavior of VEGF and PDGF for bijels-derived membranes was studied. Steady and sustained releases of VEGF and PDGF were both seen within test durations. This study demonstrates the high potential of bijels technology in the tissue engineering field.

BM04.06.16

Gradient Micro-Topography for Morphological Control and Synapse Formation Ryan McNaughton, Yuda Huo, Guicai Li, Hengye Man and Xin Zhang; Boston University, Boston, Massachusetts, United States.

In the present study, we demonstrate a rapid and low-cost method to prepare biomaterials with anisotropic, gradient micro-ridge/groove arrays having variable local pattern width. It was anticipated this study will demonstrate the ability of gradient micro-topographies to influence preferential neuronal adhesion and maturation, dendritic tree expansion, and synaptic network formation, providing a comprehensive understanding of the neuron-microenvironment interaction for the design of neuroregenerative devices.

The surface topography of biomaterials with specific spatial structure mimic the physical microenvironment of neurons, regulating their orientation and neurite outgrowth. However, most topographical structures used in neuron culture are fabricated with single geometries. This necessitates bulk fabrication, which is both inefficient and laborious, while also increasing potential contamination. A single gradient micro-topographical device with variable geometries of a single orientation begins to ameliorate these issues, although few studies have examined the influence of linear micro-ridge/groove structures on synaptic network formation and dendritic tree structure.

The physicochemical properties of the prepared gradient micro-ridge/groove array were characterized by analyzing the surface morphology and wettability. The patterning structure and dimensions were confirmed to be intact through scanning electron microscopy and viable for cell contact. Additionally, cell adhesion was increased through surface treatment with poly-l-lysine and laminin. The chemical composition of the surface modification was verified through FTIR-ATR spectroscopy. Uniformity of the surface treatment was verified through fluorescent intensity of FITC-poly-l-lysine. Cell experiments were subsequently carried out using primary rat hippocampal neurons.

Immunofluorescent images of neurons in culture for 14 days revealed smaller pattern widths regulate neuron growth and increase orientation along the microtopography direction. Neurons cultured on these substrates demonstrate a preference to attach in areas of smaller pattern width. Additionally, cell somas located within a 5µm groove demonstrate heightened aspect ratios. Dendrites of these neurons extended less when introduced to a linear topography, decreasing their coverage area with respect to decreasing geometry. Preferential axon maturation was found to grow more within the groove as opposed to on top of each ridge. Finally, the density of synapses formed and relative protein expression significantly decreased in the presence of topography, with large increases when the geometry becomes smaller than the neuron soma. Thus, the effect of gradient micro-topography on neuron behavior was achieved and systematically understood by a one-step screening on a single integrated chip, lending itself to potential advances in the design of neuroregenerative micro-devices.

BM04.06.17

Engineering Titanium Substrate by Atomic Layer Deposition for Dental Pulp Stem Cells Proliferation and Differentiation Studies Ya-Chen Chuang[1, 2], Likun Wang[1], Marcia Simon[3] and Miriam Rafailovich[1]; [1]Materials Science & Engineering, Stony Brook University, The State University of New York, Stony Brook, New York, United States; [2]ThINC Facility, Advanced Energy Center, Stony Brook, New York, United States; [3]Oral Biology & Pathology, Stony Brook University, The State University of New York, Stony Brook, New York, United States.

Stem cells are sensitive to both chemical and mechanical changes in the environment, which proliferation and differentiation depend on three main factors: the type of stem cell, the underlying scaffold, and the signaling molecules added. It has been shown that stem cells isolated from the dental pulp (dental pulp stem cells (DPSCs)) can differentiate and express markers of odontoblasts, osteoblasts, adipocytes or neuronal cells when they are grown in specific inducing media. However, the external chemical inducers such as steroids can cause adverse side effects such as hyperglycemia and a weakened immune system in clinical studies. Therefore, we focus on engineering the underlying substrates to induce DPSCs differentiate along the desired pathway without external chemical inducers added. Titanium, a material used as dental implant, has shown to promote its osseointegration with specific surface treatment to manipulate its surface roughness and topography. Herein, we introduce a new method to fabricate titanium substrate by atomic layer deposition (ALD), which deposits a homogeneous 2~3nm thickness of titanium on silicon wafer substrates. Due to surface chemistry changed, DPSCs have shown to proliferate well on ALD titanium substrates compared to bare silicon surface. At week 4, biomineralization were characterized by SEM/EDS and Raman spectroscopy. RT-PCR was also used to identify odontogenic and osteogenic differentiation markers. The results showed that biomineralized deposits (Ca/P) along with collagen fibers were observed on ALD titanium substrates, and RT-PCR results showed that osteocalcin (OCN) was upregulated from week 2 to week 4 but Dentin Sialophosphoprotein (DSPP) expression remained low over 4 weeks. It suggests that DPSCs growing on ALD titanium surface might induce them to differentiate along osteogenic pathway.
The ALD method provides a fast and easy process to coat a homogeneous thin layer of titanium on the substrate, where only surface chemistry changes but which roughness and topography remain the same. This method could be a potential application to coat a thin layer on titanium on any biomaterial to further promote stem cells differentiation and proliferation.

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BM04.06.18

Substrate Mechanics in Combination with Nanoparticles Effects on Dental Pulp Stem Cells Proliferation and Differentiation Ya-Chen Chuang[1, 2], Chung-Chueh Chang[2], Marcia Simon[3], Miriam Rafailovich[1], Samantha Ying[4] and Mindy Li[5]; [1]Materials Science and Engineering, Stony Brook University,

Stony Brook, New York, United States; [2]ThINC Facility, Advanced Energy Center, Stony Brook, New York, United States; [3]Oral Biology & Pathology, Stony Brook University, The State University of New York, Stony Brook, New York, United States; [4]South Side High School, Hempstead, New York, United States; [5]Princeton High School, Princeton, New Jersey, United States.

Stem cells are sensitive to both chemical and mechanical changes in the environment, which proliferation and differentiation depend on three main factors: the type of stem cell, the underlying scaffold, and the signaling molecules added. It has been shown that stem cells isolated from the dental pulp (dental pulp stem cells (DPSCs)) can differentiate and express markers of odontoblasts, osteoblasts, adipocytes or neuronal cells when they are grown in specific inducing media. In our previous study, we have shown that monodisperse polybutadiene (PB) can be used to produce biocompatible flat thin films with different surface mechanics by simply altering the film thicknesses where surface chemistry remains the same. We have also shown that DPSCs can sense and adjust their cell mechanics accordingly to the underlying substrate mechanics. In addition, without the addition of inducing media, dexamethasone, biomineralized deposits and up-regulation of osteocalcin (OCN) gene marker were observed on hard PB surfaces. In contrast, extremely low level of biomineralized deposits and OCN were observed on the softer PB surfaces. On the other hand, the rise of nanotechnology also promotes the study on the effects of nanoparticles (NPs) on stem cell and shows that nanoparticles can also offer a means of regulating cell function. However, stem cells are extremely sensitive to the extracellular signals where the stimuli from substrate mechanics and NPs should be studied simultaneously. Hence, in this study, we want to investigate how DPSCs proliferate and differentiate when both substrates mechanics and NPs cues were involved. Briefly, TiO_2 NPs (0.1 mg/mL) were added post-plating onto soft and hard PB substrates after DPSCs fully attached. Cell proliferation and cell mechanics were measured at week 1 by hemocytometer and shear modulation force microscopy (SMFM). At week 4, biomineralized deposits were characterized by SEM/EDS and Raman spectroscopy. RT-PCR was also used to identify odontogenic and osteogenic differentiation markers. The results showed that with TiO_2 NPs added, collagen fibers along with biomineralization were deposited on the substrates, and it showed up-regulation of OCN gene at the later stage of differentiation process no matter the substrate is soft or hard. The results suggest that TiO_2 NPs overwrite substrate mechanics effect and dominate DPSCs differentiation in this system, which could be a potential application for nanoparticles using as stem cell differentiation inducer.

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BM04.06.20
Non-Invasive Transdermal Photomedicine Using Hyaluronic Acid Derivatives Ki Su Kim; Department of Organic Materials Science and Engineering, Pusan National University, Busan, Korea (the Republic of).

A variety of drug delivery systems have been investigated for high therapeutic efficacy via easy administration. Among them, transdermal delivery is presented as an attractive alternative to needle-based drug delivery because of patient preferences. Although transdermal microneedles are less invasive promising alternatives, needle–free topical delivery without involving physical damage to the natural skin barrier is still sought after as it can further reduce needle–induced anxiety and is simple to administer. However, this long–standing goal has been elusive since the intact skin is impermeable to most macromolecules. Also, the depth of light penetration in skin in photomedicine is a serious constraint on clinical applications. Here, we show an efficient, noninvasive transdermal delivery using Hyaluronic acid (HA) derivatives as carrier.
For vaccine delivery, following topical administration in the skin, a model vaccine ovalbumin (OVA) and HA (HA–OVA conjugates) penetrated into the epidermis and dermis in murine and porcine skins, as revealed by intravital microscopy and fluorescence assay. Topical administration of HA–OVA conjugates significantly elevated both humoral and mucosal antibodies, with peak levels at four weeks. An OVA challenge at week eight elicited strong immune–recall responses. With pretreatment of the skin using non–ablative fractional laser beams as adjuvant, strong immunization was achieved with much reduced doses of HA–OVA.
In addition, we also developed implantable light–delivery devices using HA. With this light delivery system, we successfully demonstrated the facilitated photochemical tissue bonding (PTB) using hyaluronic acid (HA)–rose bengal (RB) conjugate and upconversion nanoparticle (UCNP). The UCNP emitting red and green light in the skin tissue by skin-penetrating near infrared (NIR) laser illumination could activate the RB dye and crosslink the collagen, inducing skin repair and deep tissue wound healing. Furthermore, hyaluronate–hollow gold nanosphere–adipocyte–targeting peptide (HA–HAuNS–ATP) conjugates will be presented for the photothermal ablation of adipose tissues.

BM04.06.21
Enhancing Biocompatibility of Tantalum via Anodization for Orthopedic Applications Ece Uslu[1, 2] and Batur Ercan[1, 2]; [1]Metallurgical and Materials Engineering, Middle East Technical University, Ankara, Turkey; [2]BIOMATEN, Middle East Technical University, Center of Excellence in Biomaterials and Tissue Engineering, Ankara, Turkey.

Tantalum and its alloys have been investigated as the next generation of orthopedic implant materials in the last decade. Being a valve metal, tantalum forms a naturally occurring stable oxide layer approximately 3-5 nm on its surface at ambient conditions and this layer both prevents heavy ion release from the metal and provides a natural barrier for implant corrosion. In fact, due to its chemically inert nature, tantalum has the highest corrosion resistance of all metals used in orthopedic applications. Tantalum also exhibits higher fatigue properties compared to the currently-used implant materials. Despite having ideal properties for orthopedic applications, bioinert nature of tantalum surfaces, which limits osseointegration with the juxtaposed tissue, is the leading problem to be addressed before its widespread use in implants. To overcome this problem, surface modification of tantalum within nanoscale could be a potential remedy to enhance its bioactivity.

Anodization is an electrochemical process which produces oxide based nanofeatured surfaces on various metals. It gained popularity in the last decade due to its versatility in controlling biomaterial surface topography. In literature, it was shown that anodized nanostructured surfaces having different morphologies, topographies and feature sizes enhanced bone cell adhesion, proliferation and cellular functions in orthopedic applications. Specifically, anodized titanium and its alloys were well characterized to enhance cellular functions *in vitro*. However, there is very limited data on the anodization of tantalum for orthopedic applications.

In this study, tantalum samples were anodized using 1M H_2SO_4+ 3.3 wt % NH_4F, 1:9 (v/v) concentrated and aqueous HF/H_2SO_4 solutions to obtain oxide based nanofeatures on its surface. Upon anodization, 4 different surface morphologies, namely nanodimple, nanotubular, nanoporous and nanocoral, were successfully obtained on tantalum surfaces. Furthermore, anodization duration (1min-4hr) and voltages (10-80V) were fine-tuned to control feature sizes between 25 to 140 nm for the nanodimple, nanocoral and nanoporous morphologies. Topographical investigations indicated higher nanophase surface roughness on anodized surfaces compared to as-received tantalum. Anodized samples also expressed enhanced surface hydrophilicity independent of the morphology and feature size. To investigate biocompatibility of the samples, osteoblast (ATCC CRL-11372) adhesion and proliferation were examined up to 7 days of culture. Results indicated enhanced cellular functions on nanodimple, nanocoral and nanoporous morphologies compared to non-anodized surfaces. Furthermore, immersing these samples into simulated body fluid up to 1 month showed enhanced bioactivity of these surfaces compared to non-anodized tantalum. In conclusion, surface modification of tantalum via anodization could be a potential way to enhance biocompatibility of tantalum for orthopedic applications.

BM04.06.22
Suppression of Platelet Adhesion on Decellularized Vascular Graft by High Density REDV Peptide Immobilization Kentaro Kojima[2, 1], Atsushi Mahara[2], Yoshiaki Hirano[1] and Tetsuji Yamaoka[2]; [1]Department of Biochemical Engineering, Kansai University, Suita, Osaka, Japan; [2]Department of Biomedical Engineering, National Cerebral and Cardiovascular Center Research Institute, Suita, Osaka, Japan.

[Purpose] Suppression of thrombus formation is required for graft patency of small caliber blood vessels. In our previous work, we succeeded in proving the good patency of the small-diameter long bypass graft by modifying the luminal surface with a bioactive peptide [1]. The peptide consists of REDV and POG repetitive sequence which are the integrin $\alpha_4\beta_1$ ligand and collagen binding sequence, respectively. We found that the REDV modified surface captured endothelial progenitor cells and inhibited the blood coagulation during initial contact with the heparinized blood. However, platelets were adhered on the REDV modified surface when platelet-rich plasma (PRP) was statically incubated with the surface. The REDV density was 8.4×10^{-6} molecules/nm^3 when the REDV was immobilized via POG binding sequence. Therefore, the surface would not be fully covered by the peptide, and we assumed that the platelet adsorption was caused by exposed collagen. In this study, we developed the REDV peptide-conjugated silane coupling agents (PCSi) for high density REDV immobilization to decellularized vascular graft. The platelet adhesion on the PSCi modified surface and EPC binding affinity were evaluated.
[Method] REDV peptide was conjugated with the 3-(triethoxysilyl) propyl isocyanate (PCSi). The 0.02 - 10.0% PCSi solution was dropped into decellularized ostrich carotid artery and incubated under vacuum condition. The modification was evaluated by EDS and FT-IR. The density was quantified with ^{125}I-labeled PCSi. PRP was isolated from mini-pig arterial blood, and the tissues were incubated with the PRP for 1 hour. Human EPCs were seeded on the surface, and cell binding was evaluated by WST-8 assay.
[Results] Si signal and Si-O-Si band around 1020 cm^{-1} in EDS and FT-IR were observed after PCSi treatment, and the REDV immobilized density was 8.4×10^{-3} molecules/nm^3, which is almost 1000 times higher than that immobilized by the previous method using POG sequence. Platelets scarcely adhered on the PCSi treated tissue, but EPCs were adhered and spread out on the surface. These results suggested that the REDV was immobilized at a high density via PCSi, and the surface suppressed platelet adsorption and promoted the EPCs adhesion.
[References] 1. A.Mahara et al., Biomaterials, **58**, 54-62, 2015.

BM04.06.23
Biodegradable Hollow Capsules for the Efficient Delivery of Therapeutic Molecules and Nanoparticles Isabel Gessner, Eva Krakor, Sven Saniternik and Sanjay Mathur; University of Cologne, Cologne, Germany.

For the interconnection between a synthetic material and living matter, hollow mesoporous silica capsules (HMSC) have recently gained intense attention as drug delivery vehicles due to their biocompatibility, high loading capacity and sufficient stability in biological milieu. Compared to most of the published data, which mainly focus on the formation of spherical mesoporous silica particles, in this work, a simple and reproducible synthesis of ellipsoid-shaped HMSC (aspect ratio ~ 2) *via* a hematite template assisted route is presented. Hollow structures were formed through, coating of solid templates with a silica sol followed by acidic leaching of the core material. The porosity of prepared capsules was demonstrated by gas sorption studies, revealing mesoscopic pores around 4 nm and a high surface area of 308.8 m2/g. Both, cell uptake studies as well as cell viability assays, revealed the high biocompatibility of HMSC. Moreover, the successful cellular internalization was proven by confocal microscopy using human cervical cancer (HeLa) cells. The suitability of HMSC for drug delivery applications was tested by loading antibiotic (ciprofloxacin) and anticancer (curcumin) compounds. A pH dependent drug release over several days under physiological conditions at 37° C was demonstrated (UV-vis spectroscopy) in both cases, which showed the versatility of HMSC in transporting hydrophilic as well as hydrophobic drugs. Ciprofloxacin-loaded HMSC were additionally evaluated towards gram negative (*E.coli*) bacteria to clearly demonstrate a complete bacterial growth inhibition over 18 hours using particle concentrations of 10 µg/ml. Besides the delivery of therapeutic molecules, metallic nanoparticles such as Cu or Ag were incorporated whereby a controlled leaching of metal ions demonstrated their usefulness for antibacterial applications. Additionally, hollow capsules slowly degraded into non-toxic molecules which is of crucial importance considering their application as biocompatible and clearable drug transporter.

BM04.06.24
Bacterial Cellulose Growth in Three-Dimensional Micrometric Molds—Production, Characterization and Biomedical Applications Prospects Adriano J. Otuka[1], Rafael R. Domeneguetti[2], Moliria Santos[1], Sidney Ribeiro[2] and Cleber Mendonca[1]; [1]Photonics Group, São Carlos Institute of Physics, University of São Paulo - USP, São Carlos, Brazil; [2]Laboratory of Photonic Materials, Institute of Chemistry, São Paulo State University - UNESP, Araraquara, Brazil.

Bacterial cellulose (BC) has emerged as an interesting candidate to fabricate advanced biomaterials, aiming applications in the tissue engineering, such as, artificial skin for burns treatment and wound healing. This biopolymer exhibits a nanofibrous porous network highly moldable, with high strength, low density, high biocompatibility, and which can be easily functionalized with drugs or other biological agents for selective applications. In this work, we fabricate several polymeric microenvironments with distinct geometries, to evaluate and study the growth dynamics of bacterial cellulose. The microenvironments were produced by two-photon polymerization using a Ti:Sapphire laser oscillator, centered at 780 nm, operating at a repetition rate of 86 MHz and delivering 100 fs pulses. The polymeric resin used in the microenvironments is composed by two acrylate monomers, *tris(2-hydroxyethyl) isocyanurate triacrylate* and *dipentaerythritol pentaacrylate*, which are combined to provide hardness to the structure and a smoothed surface after the polymerization, preventing deformations on the final structure. These monomers are mixed with an acylphosphine oxide photoinitiator, *ethyl-2,4,6-trimethylbenzoyl phenylphosphinate*, an organic compound responsible to generate free radicals after laser pulse irradiation. The laser beam is focused through a microscope objective (10X, NA=0.25) into the sample and scanned in the x-y direction by a pair of movable mirrors, while the sample's axial (z) positioning is performed by a motorized stage. The photopolymerization experiment can be monitored in real time, by an illumination source and a CCD camera. After complete fabrication of the molds and sterilization, bacteria *Gluconacetobacter xylinus* (ATCC 23760) are inoculate into the microenvironments, receiving all the necessary conditions for their development. We evaluate the bacterial cellulose growth for one week, monitoring the samples in specific times (hourly and daily). The formed biofilms were characterized morphologically and structurally by scanning electron microscopy (SEM), infrared spectroscopy (IRS) and Raman spectroscopy (RS). The structure and composition of grown bacterial cellulose in the microenvironments are similar than those grown in macro systems. The results obtained in this work open new opportunities for tissue regenerative engineering studies, as well as, show other possibilities to evaluate drug delivery mechanisms due to selective permeability of the formed biofilms.

BM04.06.25
Brain HDL-Mimetic Nanomaterials Designed to Mitigate Microglia-Mediated Neuroinflammation in Alzheimer's Disease Jinhwan Kim, Song Ih Ahn and YongTae Kim; Department of Mechanical Engineering, Georgia Institute of Technology, Atlanta, Georgia, United States.

Microglia are the innate immune cells of the brain that mediate opposing deleterious pro-inflammatory and protective anti-inflammatory functions in Alzheimer's Disease (AD). Multiple evidences showed microglia-mediated neuroinflammation has been recognized as a prominent manifestation of the AD brain. As disease-modifying treatments for AD are lacking, specific inhibitors of pro-inflammatory microglial functions with high brain bioavailability are desperately needed. Kv1.3 is a microglial potassium channel that regulates membrane potential and pro-inflammatory functions and is highly expressed

by amyloid beta plaque-associated microglia in human AD brains. However, the delivery of the inhibitor into the brain remains ineffective due to the blood brain barrier (BBB) that limits the bioavailability of therapeutic molecules. One possible route to deliver drug molecules across the BBB is to leverage physiological transport of natural molecules into the brain. Here, we present engineered bioinspired materials mimicking brain high-density lipoprotein (HDL) with apolipoprotein E3 (apoE3) and microfluidic approaches to testing the delivery of a Kv1.3 inhibitor effectively across the BBB in physiologically relevant BBB-on-a-chip device.
We engineered apoE3-based HDL-mimetic nanoparticles (eHNP-E3) in a controllable and reproducible manner using a microfluidic synthesis technology. Lipids derived from 1,2-dimyristoyl-sn-glycero-3-phosphocoline (DMPC) were mixed through a series of microvortices in defined ratios with human recombinant apoE3. A ShK223 peptide (ShK), an effective Kv1.3 blocker, was conjugated with cholesterol to anchor the peptide on the surface of eHNP-E3. Our microfluidic platform creates a series of controlled microvortices that enabled the rapid assembly of precursors (DMPC and apoE3) into discoidal structures of eHNP-E3 at a Reynolds number (Re = 50), followed by the incorporation of ShK into the nanoparticle (eHNP-E3-ShK). The size of eHNP-E3-ShK was approximately 18 – 20 nm as demonstrated by DLS and TEM. Successful cellular internalization of eHNP-E3-ShK was also monitored in microglia, suggesting the potential biological function as Kv1.3 channel blocker of this nanoparticle at the subcellular level. We are currently examining the biological function of eHNP-E3-ShK both in our microengineered human BBB-on-a-chip model and in the 5XFAD model.
In summary, we demonstrated successful synthesis and physicochemical characterization of eHNP-E3-ShK and are testing the biological functions for the delivery of a therapeutic molecule into the brain. We are evaluating the biological activities of our nanoparticle targeting anti-inflammatory effects on microglia in vitro and in vivo.

BM04.06.26
Neuron-Encapsulated Self-Foldable Graphene for Graft-Electrode Interface Koji Sakai, Tetsuhiko Teshima, Hiroshi Nakashima and Yuko Ueno; NTT Basic Research Laboratories, Nippon Telegraph and Telephone Corporation, Kanagawa, Japan.

In the field of neuronal transplantation therapy, there has been a strong demand for a neuron-electrode interface to make it possible to monitor the process by which a graft is integrated into a host neuronal circuit. Since cell migration and glial scar formation lead to a loss of contact between an electrode and a graft, it is technically difficult to monitor the sequential changes in the circuit between the graft and the host tissue. Fabricating a graft by encapsulating neurons with a self-foldable and biocompatible electrode is a potent strategy for the maintenance of electrode-graft contact, because the graft can be enclosed and fixed by the folded electrode. The encapsulation of neurons with self-foldable metal films has been intensively studied, and close contact between encapsulated neurons and film has been achieved. However, conventional metal films are not applicable with this approach because 1) metal film has insufficient biocompatibility and optical transparency, and 2) the enclosed structure only allows partial connections between the encapsulated graft and the host tissue. In this study, we developed a self-foldable graphene film that has high biocompatibility, optical transparency and permeability with high electro-conductivity. Micro-pores patterned on the film allowed axonal passage, thus providing connections between the graft and host tissue.
The self-foldable graphene film with micro-pores was composed of monolayer graphene and parylene thin film laminated on a sacrificial layer of calcium alginate. Following the dissociation of the sacrificial layer, the graphene-parylene bilayer film was spontaneously rolled up to form a tubular structure by the π-π stacking interaction between these layers. To keep neurons ($\varphi > 10$ mm) inside the micro-roll and allow only their axons ($\varphi < 2$ mm) passage, an array of 8 mm micro-pores was photo-lithographically formed. We seeded primary hippocampal neurons and induced self-folding. The neurons were successfully kept inside the micro-roll. Furthermore, we confirmed that the micro-pores allowed the passage of axons outside the pore-patterned tube, unlike with plane tubes. Time-lapse images show that encapsulated neurons extend neurites through the side wall of the pore-patterned tube onto the bottom of the dish, while neurites grow only from the end of a plane tube. In addition, staining results indicate that Tau-1-positive axons grow more extensively from the pore-patterned tube than from the plane tube. MAP2-positive dendrites and cell bodies were localized within both types of tube, indicating that neurons were kept within the tubes. These results show that the self-folding of our graphene film can be applied to neuron encapsulation and that micro-pores pass axons selectively, providing a pathway for connections between the graft and host tissue. This encapsulation technique with self-foldable graphene film is promising as a tool for realizing a reliable neuronal graft-electrode interface.

BM04.06.27
Inert Metal/Degradable Metal Hybrid Stent Enabling Spontaneous and Controllable H_2O_2 Generation for Antirestenotic Functionality Hyunseon Seo, Jimin Park, Yu-Chan Kim and Myoung-Ryul Ok; Korea Institute of Science and Technology, Seoul, Korea (the Republic of).

Significant advances in design of biocompatible metals allowed development of bare metallic stents (BMSs) utilizing these metals for the treatment of vascular diseases. However, restenosis, caused by abnormal accumulation of smooth muscle cells (SMCs) near BMSs, has hindered BMSs to have confidence in their clinical efficacy and safety, and continuously demanded a new type of the stent. Drug eluting stents (DESs) suggested pharmacological methods to tackle with restenosis, however, anti-restenotic agents not only inhibit SMCs but also adversely affect vascular endothelial cells (VECs) which are essential for recovery of blood vessels by inducing re-endothelialization after stent implantation. Therefore, a different strategy to selectively inhibit the accumulation of SMCs is highly required for the stent application.
Here, we suggest a new inert metal (NiTi)/biodegradable metal (Mg-Zn alloy) hybrid stent utilizing hydrogen peroxide (H_2O_2) spontaneously generated from the galvanic coupling of two metals as a SMCs-inhibitive agent. H_2O_2, one of reactive oxygen species (ROS) *in vivo*, can be easily formed near the surface of NiTi stent body through oxygen-reduction-reaction (ORR) as degradable Mg-Zn alloys work as sacrificial electron sources. The amount of H_2O_2 generation can be controlled *via* the engineering of biodegradation kinetics through Mg-Zn alloy design. We confirm that the optimized amount of H_2O_2 released from Mg-NiTi connected system selectively inhibited proliferation and function of smooth muscle cells (SMCs) without harming vascular endothelial cells (VECs).
Furthermore, for proving high feasibility of our Mg-NiTi stent to be applied *in vivo*, we introduce surface engineering of Mg thin film coated on the NiTi stent. Because the stent requires to be significantly expanded after implanted to the vessel, interfacial stability of Mg-NiTi interfaces is highly important. Through investigation of delamination phenomenon of Mg thin film coated on the NiTi stent by using both experimental and simulational (finite element analysis) stretching test, we optimize coating pattern of Mg that can stably maintain the adhesion with NiTi stent even after highly deformed condition. Our achievement offers a new insight on development of metallic stents by proposing a simple but novel approach to solve restenosis, which has been a major constraint on clinical application of the stent.

BM04.06.28
A New Hydrogel System Encapsulated Single Red Blood Cell for Transfusion Mingjie Fan, Yueqi Zhao, Ruikang Tang and Ben Wang; Zhejiang University, Hangzhou, China.

The blood groups severely restrict blood transfusion owning to the antigens on red blood cells (RBCs) recognized by the immune system, which result in vast loss of mismatching blood or shortage of matching blood, sometimes occurrence of significant influence following transfusion in emergency. The red blood cell (RBC) membrane is architecturally complex and is characterized by significant biochemical diversity. The glycoprotein on the surface of RBC determined the blood type, while the different blood type lead to huge obstacle during clinical blood transfusion, especially in natural hazard, terrorist attack and warfare acute transfusion. Here we constructed a cell surface engineering system for hydrogel encapsulated single RBC, which based on the hydrogel formation by enzymatic crosslinking. Using biocompatible anchors for memberane, enzyme which could catalyze for the formation of a layer of

hydrogel shell was introduced onto cell surface, and encapsulation of a single RBC by hydrogel shell was produced. We made a system of mTG-gelation hydrogel system which can shield cell surface antigen, more importantly, the physical properties, biological functions, tissue distributions and in vivo biocompatibility of the encapsulated RBCs were similar to those of the native RBCs, indicating a promising application for development using an enzyme catalyzed hydrogel system. These results may provide a new feasible solution for related research and applications for emergency blood transfusion.

BM04.06.29
A Robust Thin Film with a Selective Antibacterial Property for Infection-Resistant Medical Application Goro Choi, Eunjung Lee and Sung Gap Im; Chemical and Biomolecular Engineering, Korea Advanced Institute of Science and Technology, Daejeon, Korea (the Republic of).

Rapid advances in medical and healthcare system brought about a great attention in antibacterial surface coating since bacterial infection of biomedical device are emerging as an urgent issue. Device-associated infections (DAI) have become a serious issue due to the increased risk of infectious diseases requiring hospitalization. Changing the surface properties of medical devices is a simple and direct approach to prevent DAI and intense research efforts have been focused on this aspect. Several surface modification methods such as non-adhesive, antibiotic releasing, silver-coated, or direct killing have been developed and evaluated to prevent bacterial infection on implant surfaces. However, these methods contain inherent limitations such as complicated, laborious procedures, the lack of controllability, cytotoxicity, and leaching problem.
Herein, we propose a new strategy for anti-infective surfaces made of a cross-linked ionic polymer film to achieve dual functionality that kill the bacteria while at the same time favor the survival of mammalian cells. A one-step polymerization process, termed initiated chemical vapor deposition (iCVD) process could generate a cross-linked ionic polymer film from 4-vinylbenzyl chloride (VBC) and 2-(dimethylamino) ethyl methacrylate (DMAEMA) monomers in vapor phase. Especially, the deposition process produced a polymer network with quaternary ammonium crosslinking sites, providing the surface with ionic moiety with excellent contact-killing antibacterial property. This method possesses substrate compatibility, which enable the ionic polymer film coating on various materials of medical implants. Moreover, the ionic polymer-deposited surfaces supported the healthy growth of mammalian cells while selectively inhibited the bacterial growth in co-culture models without any detectable cytotoxicity. Thus, the cross-linked ionic polymer-based antibacterial surface developed in this study can serve as an ideal platform for biomedical application requiring highly sterile environment.

BM04.06.30
Labeling and Specifying the Magnetically Aligned Collagen Fibrils for Hyperthermia Cancer Treatment Mohammad Reza Zamani Kouhpanji, Daniel Shore, Joseph Um, Yali Zhang, Rhonda Franklin and Bethanie J. Stadler; University of Minnesota Twin Cities, Minneapolis, Minnesota, United States.

Collagen is the major component of connective tissues and accounts for one-third of all proteins in the human body. The fibril nature of the collagen, especially collagen type I, provides advantages in biological studies, such as imaging and cancer treatment, such as hyperthermia and drug delivery. In these applications and many others, labeling and identifying the cells in order to distinguish and separate the healthy and unhealthy cells is a crucial task to not damage healthy cells while treating the unhealthy ones.
In this work, different ferromagnetic nanowires were electrodeposited, characterized and incorporated into collagen fibrils. The nanowires were crosslinked with the collagen fibrils to achieve one-step bi-directional alignment of the collagen fibrils as verified using optical microscopy techniques. A new technique for radio-frequency identification (RFID) was demonstrated by placing the aligned collagen matrix onto a coplanar waveguide under a fixed microwave frequency while sweeping the magnetic field. The ferromagnetic resonance (FMR) of the nanowires provided distinct signatures for each collagen matrix. Finally, an alternating magnetic field was applied to control and manipulate the temperature of the matrices and investigate the possibility of hyperthermia cancer treatment.
Specifically, variation in the trends of FMR absorption vs. applied field at 240GHz were successfully measured. Engineered effective fields included shape anisotropy of the nanowire/matrix and the dipole moments that the nanowires apply to each other. Minimum detectable concentration of the nanowires in aligned collagen fibrils was quantified to be within a few percent, resulting in optimization of the magnetic properties of aligned collagen fibrils as well as labeling process. Furthermore, the hyperthermia experiments showed that aligned nanowires can proved specific absorption rates up to 1600W/g of ferromagnetic metal. These values are sufficient for both cryowarming of preserved tissues and cancer treatment using aligned collagen fibrils prior to denaturization of the collagen fibrils.

BM04.06.31
Effect of Microporous Structure on the Mechanics and Permeability of Polymer Films Angelica Rose Galvan, Kendell M. Pawelec and Jeff Sakamoto; Mechanical Engineering, University of Michigan–Ann Arbor, Ann Arbor, Michigan, United States.

There are approximately 700,000 peripheral nerve injuries per year, yet there are few technologies to repair damage to nerve tracts > 3cm. Porous poly ε-caprolactone (PCL) multi-channeled scaffolds are a technology that demonstrates nerve regeneration following traumatic nerve injury. However, further enhancing nerve regeneration requires tuning the microstructure within the scaffold architecture to improve nerve regeneration.

The size, shape and interconnectivity of pores in the scaffold walls affect the mechanics and permeability, which are important for suturability, patient mobility, nutrient diffusion and cell adhesion. The ideal scaffold is robust enough to maintain structural integrity during implantation, but also compliant enough to prevent microchannels from collapsing. The scaffold's porous microstructure is determined by the type of porogen (e.g., NaCl), or the soluble particle that acts as a template for pores.

In order to investigate how microstructure affects scaffold mechanics, characterization was conducted on PCL films with 70 vol% porosity fabricated with porogen (NaCl), with particle sizes ranging from (10 – 60 μm), obtained via ball-milling and roller-milling. The tensile elastic moduli of the films were analyzed and compared. It was found that the elastic modulus decreases as the particle size decreases from 2.40 ± 0.34 to 1.06 ± 0.15 MPa for 62.2 and 9.2 μm particles, respectively. This correlation is likely due to better packing with smaller porogen size.

Permeability tests were also performed to characterize how the varying porogen sizes affected pore interconnectivity, which in turn influences cell adhesion and nutrient diffusion. The permeability decreases as the particle size decreases, from 9.91 x 10^{-13} m^2 at 62.2 μm to 2.53 x 10^{-13} m^2 at 9.2 μm, due to a reduction in the size of the pore interconnections and increased tortuosity in films with smaller porogen.

Characterizing the effect of scaffold microstructure on mechanics and permeability can help us predict how these properties will affect cell proliferation and adhesion, leading to improved scaffolds for nerve repair.

BM04.06.32
Enhancing Biocompatibility of Calcium Carbonate Particles for Biomedical Applications Çağatay Mert Oral[1, 3], Derya Kapusuz[2] and Batur Ercan[1, 3]; [1]Middle East Technical University, Ankara, Turkey; [2]Metallurgical and Materials Engineering, Gaziantep University, Gaziantep, Turkey; [3]BIOMATEN, Middle East Technical University, Center of Excellence in Biomaterials and Tissue Engineering, Ankara, Turkey.

Calcium carbonate ($CaCO_3$) is a widely occurring biomineral synthesized by marine creatures. In nature, $CaCO_3$ mainly exists in its anhydrous forms, namely vaterite (unstable), aragonite (metastable) and calcite (stable). Though it is possible to synthesize these anhydrous polymorphs for biomedical applications, it still remains a challenge to fine-tune physical and chemical properties of $CaCO_3$ particles due to different stabilities of anhydrous polymorphs and their complex crystallization behavior. This situation is especially critical for vaterite particles, which have been proposed for bone cement and drug delivery applications, because of their unstable characteristics.

In our study, pH values and $[Ca^{2+}]$:$[CO_3^{2-}]$ ratios of precursor solutions were altered and their effects on the polymorph, morphology and size of $CaCO_3$ particles were investigated. At low pH values, spherical and ellipsoidal vaterite particles were synthesized, whereas cuboidal and flower-like calcite particles were obtained at high pH values. Importantly, transformations from vaterite to calcite were observed at different pH values depending on $[Ca^{2+}]$:$[CO_3^{2-}]$ ratio of the precursor solutions. In addition, average particle size was constantly decreased from micron to submicron sizes with decreasing pH values. Since ethylene glycol concentration was considered as one of the critical factors determining $CaCO_3$ particle properties, control experiments were performed to distinguish the effects of ethylene glycol concentration and pH values of precursor solutions. pH was observed as the dominating factor controlling $CaCO_3$ particle properties as opposed to findings in literature, although ethylene glycol concentration was also influential on some of the $CaCO_3$ particle properties. Since bone cell functions differ depending on physical and chemical properties of particles they interact with, the synthesized $CaCO_3$ particles were also investigated *in vitro* using human osteoblasts (bone cells). Results showed that none of the synthesized $CaCO_3$ particles exhibited any toxic effect upon their interaction with osteoblasts up to 5 days of culture, while polymorph morphology altered bone cell functions.

To conclude, pH and $[Ca^{2+}]$:$[CO_3^{2-}]$ ratio of precursor solutions were shown as effective variables on polymorph, morphology and size of $CaCO_3$ particles without effecting their biocompatible characteristics. In contrast to findings in literature, pH, an ignored variable in most $CaCO_3$ particles synthesis protocols, was found to be the dominating factor as opposed to ethylene glycol concentration of the precursor solutions. *In vitro* tests also showed that bone cells performed their cellular functions without any compromise to their viability.

BM04.06.33

Exploiting Inherent Instability of 2D Black Phosphorus for Controlled Phosphate Release from PLGA/BP Composite Nanofibres Negin Kamyar[1], Ryan D. Greenhalgh[5], Tatiana R. Nascimento[2], Eliton S. Medeiros[2], Peter D. Matthews[3], Liebert P. Nogueira[4], Håvard J. Haugen[4], David J. Lewis[1] and Jonny J. Blaker[1]; [1]School of Materials, University of Manchester, Manchester, United Kingdom; [2]Department of Materials Engineering, Federal University of Paraíba, João Pessoa, Brazil; [3]School of Chemical & Physical Science, Keele University, Newcastle, United Kingdom; [4]Department of Biomaterials, University of Oslo, Oslo, Norway; [5]Department of Physics, Cambridge University, Cambridge, United Kingdom.

Black phosphorus (BP) is a two-dimensional (2-D) semiconductor with a tuneable direct band gap and highly anisotropic properties. It is inherently unstable and degrades into phosphate ions in aqueous media via oxidation [1]. Whilst the current paradigm with 2D materials leans toward stabilisation, in this study, we do the opposite and explore the use of 2D BP as a source of phosphate ions by exploiting its inherent instability for controlled phosphate ion release. Liquid exfoliated BP [2] was incorporated into degradable poly (lactide-*co*-glycolide) (PLGA) fibres via solution blow spinning [3,4], forming a flexible 3-D nanocomposite with a continuous open-fibre structure. With increasing BP concentration, the average fibre diameter increased by 43%, which we attribute to changes in the precursor solution properties including surface tension and viscosity. Raman spectroscopy along with ICP-AES confirmed the incorporation of BP into the nanocomposite. By increasing the initial loading of BP there was an increase in the BP optical phonon mode intensity in Raman spectra. ICP-AES was used to quantify exact BP loading and demonstrated that modifying the initial loading of BP in the PLGA fibres permitted tuneable release rates of phosphate ions over 8 weeks *in vitro*. Hence, the release rate of phosphate ions from PLGA-BP nanocomposite fibres can be controlled by compositional tuning of BP and lactide to glycolide ratio in the PLGA. Such nanocomposites have advantages over conventional bioactive glasses as they do not exhibit brittle behaviour which imparts great potential for non-load-bearing bone tissue applications and flexible therapeutic implants.

References:

1. J. R. Brent, A. K. Ganguli, V. Kumar, D. J. Lewis, P. D. McNaughter, P. O'Brien, P. Sabherwal and A. A. Tedstone, *RSC Advances*, 2016, 6, 86955-86958.
2. J. R. Brent, N. Savjani, E. A. Lewis, S. J. Haigh, D. J. Lewis and P. O'Brien, *Chemical Communications*, 2014, 50, 13338-13341.
3. R. D. Greenhalgh, W. S. Ambler, S. J. Quinn, E. S. Medeiros, M. Anderson, B. Gore, A. Menner, A. Bismarck, X. Li, N. Tirelli and J. J. Blaker, *Journal of Materials Science*, 2017, 52, 9066-9081.
4. E. L. G. Medeiros, A. L. Braz, I. J. Porto, A. Menner, A. Bismarck, A. R. Boccaccini, W. C. Lepry, S. N. Nazhat, E. S. Medeiros and J. J. Blaker, *ACS Biomaterials Science & Engineering*, 2016, 2, 1442-1449.

BM04.06.34

Hybrid Mats for Wound Dressing Applications from "Green" Renewable Resources Adnan Memic[1], Tuerdimaimaiti Abudula[1], Lassaad Gzara[1], Giovanna Simonetti[4], Ahmed AlShahrie[1], Numan Salah[1], Pierfrancesco Morganti[5], Angelo Chianese[4], Afsoon Fallahi[2], Ali Tamayol[3] and Sidi Bencherif[2]; [1]King Abdulaziz University, Jeddah, Saudi Arabia; [2]Northeastern University, Boston, Massachusetts, United States; [3]University of Nebraska–Lincoln, Lincoln, Nebraska, United States; [4]University of Rome, Sapienza, Rome, Italy; [5]Nanoscience Centre, MAVI Sud, Aprilia, Italy.

In the local treatment of both chronic and acute wounds it is crucial to prevent infections, control the removal of exudates and create a moist environment to allow for skin healing. To address these challenges it is necessary to develop the next generation of wound dressings. Chitin and lignin are bio-waste resulting from byproducts of crustacean crusts and plant biomass that have recently been proposed for bioengineering applications. However, their weak mechanical properties need. To accomplish this we fabricated hybrid mats composed of a chitin–lignin (CL)-based sol–gel mixture and elastomeric poly (glycerol sebacate) (PGS) using a standard electrospinning approach. Obtained results showed that PGS could be coherently blended with the sol–gel mixture to form a nanofibrous scaffold exhibiting remarkable mechanical performance and improved antibacterial and antifungal activity. The developed hybrid fibers showed promising potential in advanced biomedical applications such as wound care products. Ultimately, recycling these sustainable biopolymers and other bio-wastes alike could propel a "greener" economy.

BM04.06.35

Antimicrobial Properties of Hydroxyapatite Nanoparticles Doped with Magnesium and Platinum (HA/Mg/Pt NPs) Produced by Solvothermal Method Carlos Paucar[1], Jeniffer Caballero[1], Claudia P. Garcia[1], Garcia Carlos[3] and Asunción Fernandez[2]; [1]National University of Colombia, Medellín, Colombia; [2]Departamento Química Inorgánica, Universidad de Sevilla, Sevilla, Spain; [3]Universidad del Sinú, Montería, Colombia.

For bactericidal effects, several alternatives have been studied, including the use of metal nanoparticles (MNPs), and some inevitable problems have been found like toxicity and low biocompatibility. From human-inspired systems, the antibacterial efficiency of the hydroxyapatite nanoparticles depends strongly on the type of composites and nanoparticles size. Various types of hydroxyapatite nanoparticles and their derivatives have received much attention

for its antibacterial potential effect, including magnesium oxide nanoparticles (MgO NPs). The purpose of this research is to produce by solvothermal method, a biocompatible antimicrobial compound of nanoparticles of hydroxyapatite doped with magnesium and platinum (HA/Mg/Pt NPs).

The solvothermal method was implemented from $Ca(NO_3)_2 .4H_2O$; and $(NH_4)_2HPO_4$. . All solutions were stirred until pH 11 with ammonium hydroxide solution. Next, the solutions were added slowly into the mixed oil phase of cyclohexane; with a suitable stoichiometric ratio of cetyltrimethylammonium bromide (CTAB- surfactant) and polyethylene glycol 600 (PEG 600-cosurfactant) with continuous stirring. The milk suspension was heated in a hydrothermal system at 170°C for 12h, followed by cooling to room temperature. After the hydrothermal reaction, the obtained precipitate was dried at 60°C for 14h to remove organic matter and resultant material was calcined at 600°C for 2h. Antibacterial activity of as synthesized nanopowders was tested by the standard agar disk-diffusion method. Bacterial suspensions (E.coli – S. aureus) were prepared by growing a single colony overnight in Luria–Bertani broth with a turbidity of 0.5 McFarland standards. Then, filter paper discs (about 6 mm in diameter), containing 14 mg of as synthesized nanopowders dissolved in 1 mL distilled water, are placed on the Mueller–Hinton agar plates. Plates were incubated at 37°C for 24 h in a bacteriological incubator, and the zone of growth inhibition was measured using a digital Vernier. As result of this research, Hydroxyapatite doped with magnesium and platinum was successful synthetized by solvothermal method as shown by TEM. Structural characterization indicate magnesium substitution, FTIR analysis of the HAMg nanoparticles gives broader spectra compared to pure HA and crystallite size of HA decreased. Further, results of antibiogram method showed that nanoparticles of HA/Mg/Pt are resistant material against gram negative bacterial. However, several factors affect these results, develop antibiogram method correctly is important, samples should be sonicated for 2 hours before testing and increment the concentration of the samples. In other study, zone inhibition was bigger (~8.00 mm±0,05 mm) at a concentration of 28 mg/ml. Another consideration is enhance the synthesis parameters for obtain nanoparticles with smallest size, different morphology, greater surface area and narrow distribution range of particle size.

SESSION BM04.07: Polymeric Biomaterials for Regenerative Engineering II
Session Chairs: Gulden Camci-Unal and Junji Fukuda
Thursday Morning, November 29, 2018
Sheraton, 2nd Floor, Independence West

8:00 AM *BM04.07.01
Degradable Polyphosphazenes—Development of First, Second and Third Generational Polymers Kenneth S. Ogueri[4, 3], Harry R. Allcock[1, 5] and Cato T. Laurencin[2, 3, 4]; [1]Chemistry, The Pennsylvania State University, State College, Pennsylvania, United States; [2]Orthopedic Surgery, University of Connecticut Health Center, Farmington, Connecticut, United States; [3]Institute for Regenerative Engineering, University of Connecticut Health Center, Farmington, Connecticut, United States; [4]Materials Science and Engineering, University of Connecticut, Storrs, Connecticut, United States; [5]Materials Science and Engineering, Pennsylvania State University, State College, Pennsylvania, United States.

The design of advanced biomaterials with a wide range of properties has been fueled by new fields such as regenerative engineering, defined as the convergence of advanced materials science, stem cell science, physics, developmental biology and clinical translation for the regeneration of complex tissues. The complexity and demands of this innovative approach have inspired the synthesis of new polymeric materials that can be tailored to meet application-specific needs. Polyphosphazenes are composed of an inorganic backbone with alternating phosphorus and nitrogen atoms. Each phosphorus atom bears two substituents, with a wide variety of side groups available for property optimization. Polyphosphazenes have been investigated as potential biomaterials for regenerative engineering. The polymers have received a great deal of attention due to their outstanding synthetic flexibility and their ability to exhibit a wide range of properties.
Polyphosphazenes for use in biomedical tissue regeneration applications have evolved as a class to include different generations of degradable polymers. The 1st generation of polyphosphazenes for tissue regeneration entailed the incorporation of hydrolytically active side groups such as imidazole, lactate, glycolate, glucosyl, or glyceryl groups. These side groups were selected based on their ability to sensitize the polymer backbone to hydrolysis, which allowed them to break down into non-toxic small molecules that could be metabolized or excreted. The 2nd generation of polyphosphazenes developed for regenerative applications consisted of polymers with amino acid ester side groups. When blended with PLAGA, it showed the feasibility of neutralizing the acidic degradation products of PLAGA but formed mostly partially miscible blends. To overcome the miscibility issue, third generation degradable polyphosphazenes have been designed by incorporating dipeptide side groups which impart significant hydrogen bonding capability on the polymer for the formation of completely miscible polyphosphazene-PLAGA blends. The 3rd generation exhibits a unique degradation behavior by which the polymer blend changes from a film to an assemblage of microspheres with interconnected porous structures upon degradation. Our current work seeks to broaden the development of generational degradable polyphosphazenes with high strength and in situ pore-forming ability for musculoskeletal applications.

8:30 AM BM04.07.02
Investigating the Effects of Substrate Mechanical Patterns on Proliferation and Differentiation of Dental Pulp Stem Cells Ya-Chen Chuang[4, 1], Chung-Chueh Chang[1], Jessica Hofflich[4], Grace Liu[2], Albert Zhu[3], Marcia Simon[5] and Miriam Rafailovich[4]; [1]ThINC Facility, Stony Brook University, Stony Brook, New York, United States; [2]Bayview Secondary School, Richmond Hill, Ontario, Canada; [3]Our Lady of Lourdes High School, Poughkeepsie, New York, United States; [4]Materials Science and Engineering, Stony Brook University, Stony Brook, New York, United States; [5]Oral Biology and Pathology, Stony Brook University, Stony Brook, New York, United States.

Stem cells isolated from the dental pulp (dental pulp stem cells (DPSCs)) were shown to differentiate and express markers of odontoblasts, osteoblasts, adipocytes or neuronal cells when they sense the environmental changes such as the underlying scaffold and the signaling molecules added. In our previous study, we have shown that monodispersed polybutadiene forms a convenient biocompatible scaffold, to which the cells can adhere without additional coatings. Furthermore, we showed that the substrate modulus obeyed a continuously differential function where the film modulus could be varied by more than an order of magnitude simply by changing the film thickness. DPSC plated on these PB substrates were able to adjust their moduli in response to the film thickness, obeying the same functional form as the PB films. Yet, despite the continual change in cell hardness, an abrupt change occurred for substrates moduli greater than 2.3 MPa when large amounts of biomineralized deposits were observed after 28 days.
RT-PCR analysis indicated that the substrate mechanics induced differentiation of the cells without any additional chemical inducers. Florescent immunohistochemically staining indicated that all the cells in the tissue that formed expressed OCN, a protein necessary for biomineralization and an indicator for osteogenic differentiation, on thin, hard PB films, while no OCN was found in cells on the softer, thick PB films. Hence direct contact with the hard substrate was only required for one of the layers, and the effect was propagated further into the films.
Scaffolds with mechanical patterns surface patterns were then produced with length scales ranging from the macro to the nanoscale. The patterns were produced simply by imprinting in the Si wafer and adjusting the film thickness, without chemical cross linkers introduced into the PB films. Hence the influence of purely mechanical heterogeneity could be probed. In the case of microscale patterns, the results indicate that the influence of the substrate mechanics is communicated within the tissue via cell-cell contact. In the case where both hard and soft patterns were present, in a manner with enabled

cell-cell contacts to form between patterns, no biomineralization was observed. If cell-cell contacts were prevented, differentiated and non-differentiated cells were able to coexist within a single culture, where the phenotype was governed by the substrate mechanics. This study is important when applying printed scaffolds as dentin/tooth regenerative biomaterials, which surface is rough and the mechanics is not homogeneous.

8:45 AM BM04.07.03
Bioactive Aligned Conducting Polymer Nanofibers Using Laminin-Derived Biomolecules for Neuritogenesis Milad Khorrami, Zhilin Guo, Mohammad Reza Abidian and Anthony M. Kisucky; University of Houston, Houston, Texas, United States.

Peripheral nerve transection occurs commonly in traumatic injury, causing deficit distal to the injury sites. Many studies have been devoted to promote nerve conduits for nerve injury repair including fabrication of hollow tubes or fibers. However they often fail due to short and slow regeneration over long gap. An ideal nerve conduit for regeneration should provide physical and/or biochemical guidance cue to direct the neural axons. To that end, conducting polymer have been widely used for neural applications due to 1) facile functionalization process with biomolecules to tune biological response, 2) soft mechanical properties that mimics the tissue properties and 3) the ability to transduce mixed ionic and electronic conductivity.

In this research, we investigated the fabrication of bioactive aligned conducting polymer nanofibers (CPN) to potentially direct the extension of neural processes. The fabrication process includes (1) electrospinning of poly (l-lactic acid) (PLLA) template nanofibers from homogeneous solutions of 3% (w/w) PLLA and 2% BTEAC (w/w) dissolved in chloroform at 100 kV/m electrical field and rotation rate of 1500 RPM , (2) electrochemical polymerization of laminin-doped poly(3,4-ethylenedioxythiophene) (PEDOT) from 0.02M PEDOT and 6.17µM laminin-derived peptide (DEDEDYFQRYLI) in water/acetonitrile (1:1) solution with charge density of 600mC/cm^2. Scanning electron microscopy showed that the size of PLLA fibers was 302.65±101.66 nm diameter (n=100). The X-ray photoelectron spectroscopy results have proved the presence of laminin-derived peptide on the surface of CPN. We characterized the electrical properties (i.e. impedance and capacity of charge transfer) of the bioactive CPN. We will use rat dorsal ganglions to study the effect of laminin-doped aligned CPN on growth of neuronal cells and neurite outgrowth. Future study will also focus on creating a gradient of human laminin on the surface of aligned CPN as guidance for axonal growth.

9:00 AM BM04.07.04
Enhancing Oseointegration on Biodegradable Polymer Scaffolds with ALD Deposition of Titania Kuan-Che Feng[1], Adriana Pinkas-Sarafova[1], Likun Wang[1], Ya-Chen Chuang[1], Linxi Zhang[1], Chung-Chueh Chang[2, 1], Marcia Simon[1] and Miriam Rafailovich[1]; [1]Stony Brook University, Stony Brook, New York, United States; [2]Advanced Energy Center, ThINC Facility, Stony Brook, New York, United States.

Fused deposition modeling (FDM) is a rapidly growing method for device fabrication. The technique is inexpensive and the product, such as bone inserts, dental devices, can be printed directly from CT scans or impressions, and hence specifically tailored for the individual. However, from the former study shows that the cell did not attach well on the FDM printed surface due to the roughness of FDM printed surface and hydrophobicity of the polylactic acid (PLA).

In this study we produced FDM printed scaffolds that were then coated with titanium dioxide via the atomic layer deposition (ALD) method. TiO_2has been shown in numerous studies to enhance osseointegration. Hence by this technique one can produce scaffolds that are at once biodegradable, and yet support osteogenic or odontogenic differentiation. In order to probe this concept we plated dental pulp stem cells on these scaffolds, incubated for 28 days, in media with glycerol phosphate, but without the commonly used induction factor, dexamethasone. The culture was then harvested for qRT-PCR and the surfaces were imaged with scanning electron microscopy. Cell mobility, proliferation, and differentiation were studied and significant differences in both biomineralization and differentiation were observed between coated and uncoated surfaces.

9:15 AM BM04.07.05
Mechanical Performances of Engineered Polymer Scaffolds Ozlem Yasar[1] and Ozgul Yasar-Inceoglu[2]; [1]City University of New York, Brooklyn, New York, United States; [2]Mechanical Engineering, California State University, Chico, Chico, California, United States.

There are more than 114,000 people are waiting in the waiting list for an organ transplantation and on average 20 people die each day waiting for a transplant. In recent years, tissue engineering has brought to the attention to do organ/tissue regeneration as an alternative way to the organ transplantation. Success rate of tissue regeneration strongly depend on the accurate scaffold fabrication. In this research, scaffolds were fabricated with Poly(ethylene glycol) diacrylate (PEGDA) and 2,2-dimethoxy-2-phenylacetophenone (DMPA). PEGDA is a biocompatible polymer that can be easily cured in the room temperature. DMPA is used as a photoinitiator, which starts the polymerization reaction when it interacts with the UV light. The mechanical characterization of PEGDA and DMPA mixture have been considered in some range in tissue engineering field. However, it is not fully studied.

In this research, firstly, PEGDA was diluted with water to prepare 20% and 40% PEGDA and 80% PEGDA. With these different PEGDA concentrations, cylindrical samples were prepared with photolithography process. On the other hand, 0.02% (w/v), 0.06% (w/v), and 0.1% (w/v) photoinitiator-solvent mixtures were prepared to alter the DMPA concentration. Then, cylindrical samples with altered DMPA concentrations were also prepared with photolithography process. After that, compression tests for all the cylindrical samples that were prepared with different PEGDA and DMPA concentrations performed with the Instron 3369 universal testing machine. Our results indicate that, as the PEGDA concentrations increased, compressive strength of the hydrogels also increased and PEGDA concentration had significant effects on elastic modulus and ultimate strength. Average ultimate strengths for 20%, 40% and 80% PEGDA concentrations were in the order of 1 MPa, 1.5 MPa, and 4.5 MPa, respectively. Our results related to the effect of photo-initiator on mechanical properties of engineered scaffolds showed that as the DMPA concentration was increased, ultimate strengths were decreased. For 0.02% (w/v), 0.06% (w/v) and 0.1% (w/v) DMPA, average ultimate strengths were obtained in the order of 6 MPa, 5 MPa and 4 MPa, respectively. Thus, these results showcases, mechanical properties of PEGDA based hydrogels can be controlled by changing the water amount as well as the photoinitiator concentration.

9:30 AM BM04.07.06
Adhesive Polymer Composite Wound Dressings—Silver and Silica for Antimicrobial and Hemostatic Applications John L. Daristotle[1], Lung W. Lau[2], Joseph Hunter[1], Shadden T. Zaki[3], Leopoldo Torres Jr[1], Priya Srinivasan[2], Aristotelis Zografos[3], Omar B. Ayyub[4], Anthony D. Sandler[2] and Peter Kofinas[4]; [1]Fischell Department of Bioengineering, University of Maryland, College Park, Maryland, United States; [2]Sheikh Zayed Institute for Pediatric Surgical Innovation, Joseph E. Robert Jr. Center for Surgical Care, Children's National Medical Center, Washington, District of Columbia, United States; [3]Department of Materials Science and Engineering, University of Maryland, College Park, Maryland, United States; [4]Department of Chemical and Biomolecular Engineering, University of Maryland, College Park, Maryland, United States.

Wound closure devices prevent excessive blood loss and exposure to pathogens, whether for internal or external use. Conformal, adhesive dressings improve upon conventional wound closure devices such as sutures by minimizing collateral damage to tissue and providing a leak-proof seal. In this work, we demonstrate how solution blow spinning (SBS) can be used to spray-deliver polymer composite dressings in situ--and how the composite element delivers advanced functionality, either by (1) releasing silver ions to prevent infection or (2) by incorporating silica particles to reduce coagulation time. Both approaches utilize a body temperature-responsive biodegradable polymer blend of poly(lactic-co-glycolic acid) and poly(ethylene glycol) (PLGA/PEG) with minimal cytotoxicity.

While silver salts are frequently used as a source of antimicrobial silver ions (Ag^+), their elution in commercial external wound care products is immediate and therefore their solubility is used to control Ag^+ availability, often leading to exposure to unnecessarily high and cytotoxic levels. In approach (1), silver nitrate ($AgNO_3$) is dissolved into the PLGA/PEG blowspinning solution, establishing consistent loading and elution profiles: The PLGA/PEG/Ag fibers release Ag^+ quickly in a controlled manner over 24 hours, and they continue to release Ag^+ over 30 days. $AgNO_3$ concentration was tuned to 1 mg/mL to produce a dressing that inhibits microbial growth but does not affect cell viability. In a porcine burn graft donor model, PLGA/PEG/Ag can be applied once and requires few, if any, reapplications during the course of healing, and produces no delay in healing compared to a conventional polyurethane dressing. Because it is biodegradable, the polymer is incorporated into the scab and can be removed after the wound bed is reepithelialized and no longer requires a dressing.

Polymers can also be used internally as surgical sealants to prevent fluid leaks and promote hemostasis by occlusion of blood flow. However, uncharged synthetic polymers lack a mechanism to trigger the coagulation cascade. Approach (2) incorporates silica particles into PLGA/PEG to trigger the coagulation cascade via the “glass effect”, which takes advantage of the strongly negative charge present on bare silica. Composite sealants cause citrated blood to clot in scenarios where PLGA/PEG alone does not. The effect is also size-dependent: composites containing 20 nm silica particles cause blood to clot 25% faster composites with 620 nm particles. The composite sealant was tested in a liver laceration model, and achieved near-complete hemostasis within 15 minutes, while PLGA/PEG did not. Additionally, silica particles increase adhesion and can be used to modify the stiffness and extensibility of the sealant.

9:45 AM BM04.07.07
Megakaryocyte Membrane-Wrapped Nanoparticles to Target Hematopoietic Stem Cells Jenna Harris[1], Erica Winter[2], E. Terry Papoutsakis[3, 2] and Emily S. Day[4, 1]; [1]Materials Science and Engineering, University of Delaware, Newark, Delaware, United States; [2]Biological Sciences, University of Delaware, Newark, Delaware, United States; [3]Chemical & Biomolecular Engineering, University of Delaware, Newark, Delaware, United States; [4]Biomedical Engineering, University of Delaware, Newark, Delaware, United States.

Delivery of therapeutic cargo to hematopoietic stem cells (HSCs) is a challenging problem whose solution could transform the treatment of many diseases, ranging from autoimmune disorders to hematological malignancies [1]. HSCs are multipotent cells that can differentiate into all blood cell types in the body, so delivering agents that promote lineage-specific differentiation of HSCs could transform medical practice. However, delivering therapeutic cargo to HSCs *in vivo* is extremely difficult given that HSCs reside in bone marrow and are notoriously difficult to transfect. To overcome this challenge, we have developed a biomimetic nanoparticle (NP) platform that enables targeted cargo delivery to HSCs.

We hypothesized that polymeric poly(lactic-co-glycolic acid) (PLGA) NPs coated with membranes derived from megakaryocytes (Mks) could enable specific recognition of HSCs for targeted cargo delivery. This hypothesis was based on prior work that showed megakaryocytic microparticles (extracellular vesicles that bud off Mk cells) could specifically bind and enter HSCs *in vitro* [2,3]. Here, we provide the results of our *in vitro* studies, which demonstrate the synthesis of Mk membrane-wrapped PLGA NPs (MkNPs) and validate that these MkNPs can effectively bind and enter HSCs to deliver their cargo.

To create MkNPs, ~100 nm diameter DiD-loaded PLGA NPs were co-extruded with empty membrane vesicles derived from Mk cells, which were labeled with PKH26, using an Avanti Mini Extruder. DLS and TEM measurements showed that MkNPs were monodisperse and spherical, with wrapped NPs having a diameter 10-20 nm larger than bare NPs. Bare NPs had a zeta potential of -48.55 mV, and the charge of MkNPs (-19.18 mV) was similar to that of membrane vesicles (-20.93 mV), indicating successful wrapping. Successful wrapping was further confirmed by stability in PBS, as MkNPs placed in PBS at 4°C did not swell, while bare NPs swelled from 120 nm to 600 nm in <1 h. Flow cytometric analysis of CD41, an outer membrane marker, on MkNPs versus membrane vesicles confirmed through similar expression levels that the membranes were right-side out on the MkNPs. Confocal imaging showed that CD34+ HSCs take up MkNPs within 24 hours of incubation, and super-resolution microscopy confirmed that PHK26 and DiD signals colocalize within HSCs. The MkNPs preferentially interact with the uropod region of HSCs, similar to what has been reported for megakaryocytic microparticles [2,3].

In summary, PLGA NPs can be wrapped with Mk-derived membranes, allowing the resultant MkNPs to bind HSCs to deliver their cargo. These data support continued development of MkNPs for HSC manipulation.

1. Riviere I. Blood 2012; 119: 1107:1116. 2. Jiang J, et al. Blood 2014; 124: 2094-2103. 3. Jiang J, et al. J. Control Release 2017; 247: 1-18.

10:00 AM BREAK

10:30 AM BM04.07.08
Wood-Derived Nanocellulose for Biofabrication and Biomedical Applications Binbin Zhang[2, 1], Chunlin Xu[3], Xiaoju Wang[3], Fang Cheng[3], Wenyang Xu[3], Paul J. Molino[1], Markus Bacher[4], Thomas Rosenau[4], Stefan Willför[3] and Gordon Wallace[1]; [1]University of Wollongong, Wollongong, New South Wales, Australia; [2]Yokohama National University, Yokohama, Japan; [3]Åbo Akademi University, Turku, Finland; [4]University of Natural Resources and Life Sciences, Vienna, Austria.

Cellulose materials have shown great potential for biomedical applications owing to their intrinsic characteristics, such as biocompatibility, hydrophilicity, porosity, and tunable mechanical properties[1]. Wood-derived nanocellulose, coming from the most abundant biomass on earth, is naturally low cost and suitable for mass production. Compared to widely studied and commercially developed bacterial nanocellulose[2], wood-derived nanocellulose supports easy post-production processing, both mechanically and chemically, making it a favorite candidate for applications such as 3D bioprinting[3].
In this study, we used cellulose nanofibrils (CNF) prepared from bleached birch kraft pulp, chemically oxidized using 2,2,6,6-Tetramethylpiperidinyloxy or 2,2,6,6-Tetramethylpiperidine 1-oxyl (TEMPO) and mechanically treated[4]. This CNF consisted of fibers with a dimension of ca. 5 nm in width and hundreds of nanometers in length and had a consistency of 1 w/v% with 1.14 mmol/g negative charge attributed to carboxylic acid groups from TEMPO-oxidization. The prepared CNF demonstrated excellent rheological properties in demand for 3D printing, in terms of high yield stress and shear thinning properties. Herein, we report for the first time 3D printed scaffolds with high resolution features printed using a single CNF component.
A secondary cross-linking approach was also investigated to further enhance and adjust the stability and mechanical properties of the 3D printed CNF

scaffolds. CNF scaffolds were first cross-linked through ionic interaction between Ca^{2+} and carboxylic groups on the fiber surface of CNF, which provided a fast and *in situ* mechanism for 3D printing. The printed CNF scaffolds were then chemically cross-linked by 1,4-butanediol diglycidyl ether (BDDE) to form irreversible covalent bonds.
A series of characterizations were performed on the biofabricated CNF scaffolds, including rheology, swelling ratio, mechanical tests and cell culture, etc. The CNF scaffolds had demonstrated long term stability in PBS and a high ratio for water re-adsorption after freeze-drying. The compressive Young's modulus and shear modulus were in the suitable range for skin wound healing and the cell studies showed positive results in supporting human dermal fibroblast cells adhesion and proliferation.

[1] J. Appl. Polym. Sci. 2015, 132, 41719.

[2] Appl. Microbiol. Biotechnol. 2015, 99, 2491.

[3] ACS Sustainable Chem. Eng. 2018, 6, 5, 5663.

[4] Cellulose 2014, 21, 2587.

10:45 AM BM04.07.09
Aligned Conducting Polymer Nanotubes for Precisely Triggered Release of Proteins Mohammadjavad Eslamian and Mohammad Reza Abidian; University of Houston, Houston, Texas, United States.

Conducting polymers actuators are one of the most promising materials for development of controlled drug delivery systems, owing to their outstanding capability to reversibly change their volume during electrochemical process. We previously demonstrated the precise release of drugs such as dexamethasone from conducting polymer nanotubes. The fabrication process involved electrospinning of drug-loaded biodegradable nanofibers on microfabricated electrodes, followed by electrochemical deposition of conducting polymers on microelectrodes and around the electrospun nanofibers. Poly(3,4-ethylenedioxythiophene) (PEDOT) is one of the most versatile conducting polymers employed in the field of polymer electronics, owing to its superior electrical conductivity and chemical stability. Soluble growth molecules such as nerve growth factor (NGF) provide trophic support for neurons and are vital for axonal growth. The goal of this research is to develop a nanoscale device for precise delivery of NGF. To accomplish this task, NGF is encapsulated in aligned poly (lactic-co-glycolic acid) (PLGA) nanofibers via emulsion electrospinning process on gold coated silicon-based electrodes. Then, a thin layer of PEDOT is galvanostatically deposited on the electrodes and around the electrospun nanofibers at the current density of 1 mA/cm^2 for 1 min to form NGF-loaded aligned PEDOT nanotubes. The outer diameter of PEDOT nanotubes is 440±91 nm. The release behavior of NGF from PEDOT nanotubes is investigated by electrical actuation of PEDOT nanotubes in phosphate-buffered saline (PBS) using enzyme-linked immunosorbent assay (ELISA). The electrical actuation will be performed using cyclic voltammetry (CV) at the potential range of -0.8 V to +0.4 V at different scan rates, including 10, 50, 100, and 200 mV/s, and for different numbers of CV cycles (up to 100 cycles). Finally, the NGF release rate will be assessed as a function of scan rates and CV cycles. The results of this study may have impact for development of NGF delivery platforms for modular growth of axons in both central and peripheral nervous systems.

11:00 AM BM04.07.10
Solvent-Free Preparation of Porous Poly(L-Lactide) Microcarriers for Cell Culture and Bone Tissue Engineering Mirasbek Kuterbekov[1, 2], Paul Machillot[2], Pierre Lhuissier[3], Catherine Picart[2], Alain M. Jonas[1] and Karine Glinel[1]; [1]BSMA, IMCN, Université catholique de Louvain, Louvain-la-Neuve, Belgium; [2]IMBM, LMGP, Communauté Université Grenoble Alpes, Grenoble INP, CNRS, Grenoble, France; [3]SIMAP, Communauté Université Grenoble Alpes, Grenoble INP, CNRS, Saint Martin d'Heres, France.

One of the main challenges in regenerative medicine is the development of appropriate biomaterial constructs that can guide cell behavior. Microcarriers are particularly attractive in this regard due to their small size. This feature allows them to circumvent many of the issues facing more traditional monobloc constructs such as low nutrient-metabolite exchanges and uneven cell infiltration into the bulk while still being suitable for dynamic culturing and engineering of large-scale tissue grafts [Declercq *et al.* Biomaterials 2013]. Porous microcarriers made of synthetic biodegradable polymers have the added advantage of a larger surface area, improved mass transfer and the tunability of their bioactive properties.
Currently, most methods for the production of porous polymeric microcarriers rely on creating emulsions, which require potentially toxic organic solvents and relatively complex setups [Silva *et al.* J. Tissue Eng. Regen. Med. 2007]. Alternatives limiting the use of organic solvents have been proposed; however, such methods often fail to replicate the tunable microcarrier morphologies that can be obtained with the emulsion-based methods. This in turn hinders the wider adoption of microcarriers due to concerns over safety to human health and potential limits in large-scale production.
To address this issue, we developed a novel organic-solvent-free approach for the production of porous poly(L-lactide) (PLLA) microcarriers. The method is based on the isothermal spherulitic crystallization of PLLA in its blend with polyethylene glycol (PEG). Resulting PLLA spherulites are easily recovered as microcarriers by simple removal of water-soluble PEG. Independent control and tunability of microcarrier size and porosity were demonstrated, with higher crystallization temperatures leading to larger sizes and higher PLLA content resulting in lower levels of porosity. Moreover, microcarriers were shown to support not only the long-term proliferation of murine myoblast and human adipose stromal/stem cells (hASC) but also differentiation of hASCs toward osseous tissues. Furthermore, while no significant differences were observed during cell proliferation on microcarriers of two different porosities, microcarriers of lower porosity induced a stronger hASC osteogenic differentiation, as evidenced by higher alkaline phosphatase enzymatic activity and matrix mineralization. Consequently, the proposed organic-solvent-free method for the fabrication of biocompatible porous PLLA microcarriers represents an innovative methodology for ex vivo cell expansion and its application in stem cell therapy and tissue engineering.

11:15 AM BM04.07.11
The Role of Dissipation in Developing Hydrogels for Functional Tissue Engineering Naser Nasrollahzadeh and Dominique P. Pioletti; EPFL Lausanne, Lausanne, Switzerland.

Hydrogels as hydrated polymeric structures are promising biomaterials for biomedical applications. In particular, hydrogels inherent analogy with soft tissues (water-swollen solid network) has made them an interesting candidate as regenerative substitute for damaged tissues. However, these synthetic equivalents usually lack the desired functional properties as load bearing biomaterials. Apart from biological key features, a demanding set of mechanical properties including stiffness, elasticity, strength, dissipation, permeability, toughness, and fatigue resistance is required to sustain load. Indeed, an ideal biomaterial might have different characteristics corresponding to physiological stress/strain range for the target load bearing tissues. In case of cartilage for instance, a fairly stiff and tough biomaterial, contrary to soft but highly stretchable and tough, is required to avoid a long period of in vitro culture in bioreactor before implantation. The insufficient dissipative capacity is known as the main reason for low toughness and inability to resist defect growth during loading. Therefore, combining simultaneously high dissipative capacity and stiffness can lead to a load-bearing and tough biomaterial. Yet, there is no guarantee for fatigue resistance performance of the merely stiff and dissipative hydrogels. Indeed, the different sources of dissipation must be well

designed for a robust behavior under fatigue load.
By following the sacrificial bonds principle, a single network (SN) system can be developed via a dual crosslinking strategy. Indeed, SN hydrogels created by block copolymers and ionic cross-linking systems demonstrated high stiffness and partial fatigue resistance owing to simultaneous incorporation of soft and stiff bonds. Our lab has recently focused on synthesizing poly [2-hydroxyethyl methacrylate] (PHEMA) hydrogels with tunable dissipative capacity. Crosslinked PHEMA based hydrogels present interesting properties for designing SN systems. In one hand, different reversible bonds including chains entanglement, hydrogen bonding and hydrophobic associations can be formed throughout the network intrinsically. On the other hand, the stiffness and dissipation level can be significantly enhanced if the network crosslinked by short length ethylene glycol dimethacrylate (EGDMA) molecule.
In this study, we show that by combining physical reversible soft bonds with stiff covalent bonds in p(HEMA-co-EGDMA) based hydrogels, different sources of flow independent dissipation can be designed. In parallel, by controlling the morphological architecture of the hydrogel, different extent of fluid frictional drag dissipation and load support are achievable. We will show that a careful combination of these two sources of dissipation can lead to a system presenting optimized stiffness, dissipation and fatigue resistance performances.

11:30 AM BM04.07.12
Analyzing the *In Vitro* Viability of Novel Gelatin-Pluronic® F127 Hybrid Hydrogels as Cell Barrier Membranes for Guided Bone Regeneration Following Periodontitis Juyi Li[2], Kevin Chen[1], Joon Young Lee[3], Aaron Sun[4] and Miriam Rafailovich[2]; [1]Mira Costa High School, Manhattan Beach, California, United States; [2]Stony Brook University, Stony Brook, New York, United States; [3]Seoul International School, Seongnam City, Korea (the Republic of); [4]Ed. W Clark High School, Las Vegas, Nevada, United States.

Periodontitis, or conventionally “Gum Disease,” begins with the infection, and subsequent inflammation, of gingival tissue and is currently the leading cause of tooth loss in the United States. Though there exists a wide array of methods for treatment of periodontitis, one of the most effective is guided bone regeneration (GBR). GBR consists of applying a barrier membrane to separate inflamed gingival tissue from bone, restricting invasion and allowing regeneration of osteoblasts. Current barrier membranes, however, do not fulfill all the desired properties of high biocompatibility, cell impermeability, and, in particular, high mechanical strength. As such, the following study synthesized novel Gelatin-Pluronic F127 hybrid hydrogels, thoroughly analyzing their *in vitro* viability as potential cell barrier membranes for use in GBR. Rheological analysis demonstrated high mechanical strength as hybrid hydrogels’ elastic moduli drastically increased with increasing percentages of the chemical cross-linker microbial transglutaminase (mTG). The surface of the hybrid gels was visualized with laser microscope to show topographic changes among different crosslinking density. Cytotoxicity tests were first conducted to show the biocompatibility of hybrid gels. To investigate levels of cell adherence, confocal microscopy was performed on hybrid hydrogels plated with human dermal fibroblasts, which demonstrated significantly reduced cell attachment as compared to pure gelatin. Cell impermeability was further investigated by observing cell migration from gelatin gel to hybrid gel compared with from gelatin to gelatin, with a control setup consisting of Gelatin / Gelatin / Gelatin and an experimental setup of Gelatin / Hybrid / Gelatin. Human dermal fibroblasts plated on gelatin gels migrated through the middle gelatin, but, particularly at the highest mTG concentration, were unable to migrate through middle hybrid gels, showing hybrid hydrogels’ impermeability to cells. Our findings, identifying *in vitro* high mechanical strength, cell impermeability, and biocompatibility, point to novel Gelatin-Pluronic F127 hybrid hydrogels as promising biomaterials for use as GBR cell barrier membranes in treatment of periodontitis.

[1]Bhatnagar, Divya, et al. "Rheological characterization of novel HA-Pluronic thermoreversible hydrogels." *Journal of Chemical and Biological Interfaces* 1.2 (2013): 93-99.

11:45 AM BM04.07.13
Marine Polysaccharide Based Immunomodulatory Hydrogel for Type-1 Diabetes Md Lutful Amin, Damia Mawad and Charles C. Sorrell; School of Materials Science and Engineering, University of New South Wales, Sydney, New South Wales, Australia.

Type-1 diabetes is an autoimmune disease affecting ~90,000 children each year. Destruction of the insulin-producing islets by the autoactivated immune system leads to type-1 diabetes. Implantation of cells from other sources cannot reverse the condition owing to the immune response as the cells remain in contact with an activated immune system. In previous works, encapsulation of islets inside a hydrogel has shown promise but the cells were not protected from small inflammatory cytokines. Therefore, advanced functionality in a carrier with immunomodulatory capacity is a strategic goal for islet implantation. Marine-sulphated polysaccharides possess structures similar to those of extracellular matrix polysaccharides and have immunomodulatory properties. In the present work, fucoidan and three carrageenans (with different numbers of sulphate groups) were functionalised with methacrylate groups for photopolymerisation; the number of functional group in the polysaccharide chain was controlled precisely. The toxicity of the polysaccharides was evaluated by the cell growth inhibition assay on fibroblast L929 cells and all of them were found noncytotoxic. Fucoidan treatment resulted in a significant decrease in the LPS-stimulated expression of CD86, a costimulatory molecule essential to exert a full immune response to implanted islets, on PMA-differentiated THP1 cells; this response was comparable to that of IL-10, an anti-inflammatory cytokine. Fucoidan had a protective effect on THP1 cells from LPS- and IFN-mediated growth inhibition. Fucoidan also decreased the LPS-stimulated production of nitric oxide. For islet encapsulation, methacrylated polyvinyl alcohol (PVA) was synthesised and used as the base polymer and the marine polysaccharides were incorporated in order to support cell survival and impart immunomodulatory properties. Mouse insulinoma cell line, MIN6 cells were encapsulated in hydrogels (20 wt%) by UV photopolymerisation. Cell viability was assessed over a period of 28 days by live/dead assay, revealing a positive correlation between the number of sulphate groups in the disaccharide units and cell survival. Cell viability was found to be maximal in the fucoidan hydrogels that remained unaffected over this period. The protective effect of the hydrogels from inflammatory cytokines was evaluated by incubating the encapsulated islets in a medium containing inflammatory cytokines (IL-1β, TNF-α, & IFN-γ) for 48 h. Cell viability in the fucoidan hydrogels remained unaffected, which indicates the protective role of fucoidan, whereas decreased cell viability was observed in other hydrogels. The results show that marine polysaccharides with immunomodulatory properties were screened successfully. The principal finding is that fucoidan favours islet survival inside hydrogels and is effective in reducing the immune response, suggesting that fucoidan hydrogels can be used for *in vivo*islet implantation for the treatment of type-1 diabetes.

SESSION BM04.08: Polymeric Biomaterials for Regenerative Engineering III
Session Chairs: Josephine Allen, Guillermo Ameer and Vivek Kumar
Thursday Afternoon, November 29, 2018
Sheraton, 2nd Floor, Independence West

1:30 PM BM04.08.01
Photo-Responsive Peptide Hydrogels for Tailorable Epitope Presentation Zain Siddiqui[1], Peter Nguyen[1], Biplab Sarkar[1], David Sabatino[2] and Vivek Kumar[1]; [1]New Jersey Institute of Technology, Newark, New Jersey, United States; [2]Seton Hall University, South Orange, New Jersey, United States.

Self-assembling peptides can form nanofibrous hydrogels that can deliver cells, small-molecule drugs, and growth factors to potentiate phenotypic modulation. Here we show a modification of this platform that may tailor the *in vitro* and *in vivo* phenotype of a cellular niche via caged short pro-apoptotic sequences. We demonstrate the dose-dependent cell viability *in vitro*and show sequence dependent cellular infiltration *in vivo*. Although cytocompatible cellular carriers are promising delivery vehicles, potential for maladaptive responses, such as teratoma formation, are potential roadblocks for clinical translation. We designed and assembled sacrificial scaffolds that can be tuned on-demand. Peptides with cell adhesive RGD or pro-apoptotic WEWT moieties were introduced to create cell-adhesive and cell-arresting hydrogels. Culture of fibroblasts and stem cells demonstrated signaling efficacy of the hybrid peptides. Photolabile caging of adhesive and arresting groups show remarkable potential for triggerable adhesion, proliferation, or apoptosis. "Caging/uncaging" of cellular signaling domains can act as an "on/off" switch in cell-loaded 3D hydrogels. Photo-patterning of cell-loaded gels yielded well-resolved niches of specific cells. Subcutaneous implants of the hydrogel showed biocompatibility through cellular infiltration and lack of a fibrous capsule around the implant. Light-activated switching of cellular behavior and phenotype could be broadly useful for construction of new photo-responsive scaffolds with encoded signals for controlling cellular outcome. Multicomponent tissue-engineered scaffolds based on this technology may be crucial next steps for our design paradigm.

1:45 PM BM04.08.02
Self-Expanding and Biodegradable Porous Scaffolds Based on Magnesium-Hydroxide and Hydrocolloids for Bone Tissue Engineering Domenic T. Cipollone[1], Cerasela Dinu[2] and Konstantinos Sierros[1]; [1]Mechanical and Aerospace Engineering, West Virginia University, Morgantown, West Virginia, United States; [2]Chemical and Biomedical Engineering, West Virginia University, Morgantown, West Virginia, United States.

The repairing and regeneration of large bone defects is a significant clinical challenge and has garnered considerable research attention over the past few decades. Typical methods for bone regeneration include allografts (from patient to patient), autografts (from one site to another), or metal implants. While useful, they present limitations in regards to availability, donor site morbidity, and the risk of disease transfer. Moreover, metal scaffolds may require a secondary surgery for removal of the implant. Therefore, the controlled engineering of biodegradable, biocompatible, porous, and mechanically robust scaffolds is crucial.
This work reports on the fabrication and characterization of self-expanding, biodegradable, porous magnesium-hydroxide hydrocolloid scaffolds for use in bone tissue regeneration. In particular, the proposed interconnected porous network is engineered through *in-situ* hydrogen gas generation, thus providing the framework for a biodegradable and osteoconductive scaffold, while offering control over the pore size distribution and the resulting mechanical properties. The engineered foam is shown to be self-expanding in confined spaces, thus able to fill irregular and complex geometries. Furthermore, the use of magnesium-hydroxide, in combination with cross-linked hydrocolloids, aims to provide a solution to the high corrosion rates typical of magnesium scaffolds currently utilized. Surface morphology and porosity of the scaffolds are characterized though micro-computed tomography, scanning electron microscopy, and porosimetry, while uniaxial compression tests are used to study the resulting mechanical properties. *In-vitro* studies are then used to determine cell viability and bioactivity of the scaffolds. It is found that the total porosity and pore size distribution of the scaffolds may be tailored through control of the magnesium-solvent reaction and resultant hydrogen generation. Moreover, this leads to control over the scaffold's relative density and compressive modulus. It is believed that this work may hold the key for the development of next generation - low-cost, highly porous, biodegradable, and self-expanding - scaffolds for use in bone tissue engineering and regeneration.

2:00 PM BM04.08.03
Biodegradable Nitric Oxide (NO) Storage and Delivery Hyaluronic Acid-Based Nanofibers—Potent Applications for Tissue Engineering and Regenerative Medicine Kihak Gwon and Jae Ho Shin; Department of Chemistry, Kwangwoon University, Seoul, Korea (the Republic of).

Nitric oxide (NO) is one of the smallest pharmaceutical gas molecules, which mediates versatile physiological processes including stem cell regulation, angiogenesis, immune response, vasodilation, blood pressure regulation, antibacterial property, and wound healing. These several physiological functions have motivated researchers to develop various NO delivery systems for therapeutic applications. Recently, electrospun nanofibers as a NO carrier have received a great interest because of their facile functionalization, tunable mechanical properties, and large effective surface areas. In our previous studies, we have developed a series of polymethylmethacrylate (PMMA)-based NO-releasing nanofibers where NO donor *N*-diazeniumdiolate-modified aminoalkoxysilane and silyl-modified PMMA are covalently bound *via* sol-gel chemistry, displaying tunable NO storage amount and release kinetics. However, PMMA-based NO-releasing nanofibers are difficult to be degraded in the physiological milieu, restricting its potential *in vivo* applications. Our recent work has, thus, aimed to develop biodegradable, NO-releasing hyaluronic acid (HA)-based nanofibers. The NO donor *N*-diazeniumdiolates are covalently bound to the HA backbone *via* use of appropriate conjugate chemistry. Various chemical compositions (e.g., HA concentration and NO donor amount) and electrospinning conditions (e.g., applied potential and flow rate) are tuned to control fiber diameter, degradability, and NO release properties (e.g., maximum flux, total NO release amount, and half-life time). In addition, cell proliferation and migration controlled by such NO-releasing nanofibers and its cytotoxicity are evaluated.

2:15 PM BM04.08.04
Fabrication of 3D Scaffolds Based on Nano-Biomimetic Collagen Hybrid Constructs for Skin Tissue Engineering Abolfazl Akbarzadeh[1], Soodabeh Davaran[2], Ebrahim Mostafavi[1] and Azizeh Rahmani Del Bakhshayesh[2]; [1]Northeastern University, Boston, Massachusetts, United States; [2]Tabriz University, Tabriz, Iran (the Islamic Republic of).

Three dimensional (3D) biodegradable and biomimetic porous scaffolds are ideal frameworks for skin tissue engineering. In this study hybrid constructs of 3D scaffolds were successfully fabricated by freeze-drying method from combinations of the type I collagen (Col), and synthetic poly (lactic acid) (PLLA) or polycaprolactone (PCL). Four different groups of 3D porous scaffolds including PCL, PCL-Col, PCL-PLLA, PCL-PLLA-Col were fabricated and systematically characterized by HNMR, FT-IR and SEM. Adipose tissue derived mesenchymal stem cells (AT-MSCs) were seeded in all scaffolds and the viability, proliferation and adhesion of the cells were investigated using MTT assay and scanning electron microscopy (SEM). The results showed that scaffolds containing Col, particularly PCL-PLLA-Col scaffold, with pore sizes close to 400nm and sufficiently interconnected, have significantly greater potential ($p<0.01$) for encouraging AT-MSCs adhesion and growth. The PCL-PLLA provided a mechanically stronger mesh support and the type I Col microsponges encouraged excellent cell adhesion and tissue formation. The scaffold with the best properties could be an appropriate functional candidate for preparation of artificial skin constructs.

2:30 PM BREAK

3:00 PM BM04.08.05
Hydrogel with Tunable Degradability for Tissue Engineering Applications—Characterization of Morphological Changes Using Cryo-SEM Bonhye Koo[1], Soyon Kim[2], Jiwen Zheng[1] and Min Lee[2]; [1]U.S. Food and Drug Administration, Silver Spring, Maryland, United States; [2]University of California, Los Angeles, Los Angeles, California, United States.

Photopolymerizable hydrogels are commonly used as tissue engineering scaffolds. Under visible blue light irradiation (VBL), methacrylated glycol

chitosan (MeGC) can be photopolymerized with riboflavin (RF, vitamin B2 derivative) and applied to support the encapsulated cell proliferation and deposition of extracellular matrix. MeGC alone, however, is not a perfect candidate for a scaffold due to its relatively slow degradation rate. This is critical for a tissue engineering scaffold since it might hinder cell recruitment and delay tissue regeneration. Thus, there is strong impetus to develop a degradable tissue engineering scaffold to allow subsequent tissue regeneration and/or the release of encapsulated bioactive molecules.
In this report, we designed hydrogels with tunable degradability by incorporating lysozyme (Lys) with various concentrations (0, 0.1, and 1 mg/mL) in MeGC, based on its ability to degrade chitosan by cleaving the polysaccharide backbone. A suitable characterization technique is required to demonstrate the degree of hydrogel degradation. Cryogenic scanning electron microscopy (cryo-SEM) allowed to preserve the native structure of hydrogels in a frozen hydrate state and enabled direct imaging of the mesh structure of hydrogels degraded at atmosphere at 37 °C at a nanometer resolution. Our cryo-SEM micrographs of three hydrogels clearly showed differences in size of the mesh structure and the size change from day 0 to day 10. At day 0, the average pore size of the mesh structure was 38.6, 36.7, and 37.9 nm respectively for MeGC, Lys0.1 and Lys1. Their size changed to 67.6, 86.4, and 408.6 nm respectively for MeGC, Lys0.1 and Lys1 after 10 days. In addition, the maximum pore size increased up to 1,200 nm for Lys1 at day 10. This supported that MeGC hydrogel became degradable with lysozyme and the degree of degradation was controllable by adjusting the amount of lysozyme.

3:15 PM BM04.08.06
Fabrication of Composite Bone Scaffolds with Controlled Multi-Scale Porosity Using Non-Solvent Induced Phase Separation Based 3D Printing Mehmet S. Aydin[1, 2], Gullu Kiziltas[1, 2] and Busra Kuloglu[1]; [1]Sabanci University, Istanbul, Turkey; [2]Sabanci University Nanotechnology Research and Application Center, Istanbul, Turkey.

Bone fracture is a widespread injury associated with individual disability and loss of social productivity resulting in very high treatment costs. Well-designed scaffold implants are good alternatives in bone tissue engineering known to result in effective healing. An ideal bone scaffold should be biocompatible, porous, interconnected and strong, i.e. multi-functional. Therefore, composite materials stand out as desired material candidates. Advances in the design and production of porous composite scaffolds took place owing to solid free-form fabrication (SFF) techniques with mechanical and biological functions tailorable to specific bone defects. Aim of this study is to develop an effective SFF based technique capable of printing composites with well controlled macro-micro porosities making primarily use of commercial 3D printers. Similar features are displayed by novel designs obtained via topology optimization [Hollister, S. J., Nature Materials, 2003; Aslan, O. S. and Kiziltas, G., Proc. of TERMIS-EU, 2013]. Poly(ε-caprolactone) (PCL) was chosen as the scaffold polymer, which is FDA approved. A variety of SFF techniques have been developed to produce controlled porous PCL scaffolds. Also, incorporation of bioactive, stiff inorganic materials into PCL polymer led to significant enhancements in mechanical properties, bioactivity, and bone regeneration ability. However, only a few attempts have been made to create scaffolds with macro-micro porosity, despite their potential to more closely mimic the hierarchical architecture of native bones. Recently, non-solvent-induced phase separation (NIPS) seems capable of producing these scaffolds with multi-scale porosity if integrated into the 3D printing based process. Using this method, here we develop scaffolds with multi-scale porosity (< 10μm and >100 μm) where, PCL pellets are dissolved in the THF solvent and mixed with various amounts of HA powders. Resulting solution is extruded and deposited in ethanol bath at RT. Microporous PCL/HA composite filaments are created via exchange of the solvent and the non-solvent (EtOH). Printing parameters were optimized in order to match the designed scaffold geometry. Morphological parameters such as porosity and connectivity of scaffolds were analyzed via micro-CT and SEM. FTIR, TGA and mechanical testing for tensile and compressive strength were carried out. Biological in-vitro tests for cytocompatibility and apatite-forming ability are underway to measure cell attachment, proliferation and growth. Initial results show that using NIPS and commercial printing, scaffolds displaying both macro and micro-porosity (%10-20) were successfully fabricated. Further characterization should prove that these scaffolds are bio-compatible, strong and well-designed. The capability to directly manufacture novel designs should open up new possibilities for other applications demanding multi-functional scaffolds, thereby allowing effective patient specific treatment.

3:30 PM BM04.08.07
3D Woven Metallic Lattices as Bio-Scaffolds with Hydroxyapatite Coatings Ju Xue[1], Ashley Farris[2], Yunfei Wang[1], Cristina Romany[1], James Guest[3], Warren Grayson[2], Shoji Hall[1] and Timothy P. Weihs[1]; [1]Materials Science and Engineering, Johns Hopkins University, Baltimore, Maryland, United States; [2]Biomedical Engineering, Johns Hopkins University, Baltimore, Maryland, United States; [3]Civil Engineering, Johns Hopkins University, Baltimore, Maryland, United States.

There is a strong need for the next generation of bio-scaffolds that combine biologically activated coatings with porous and biodegradable substrates. Here we present studies of 3D woven metallic lattices that are designed using topology optimization to enhance fluidic permeability and mechanical stiffness and are coated uniformly with hydroxyapatite (HAp) to improve bioactivity and osteointegration. The ultimate goal is to weave and coat Mg alloy wires. Here we present initial results describing successful coating of HAp on 304 stainless steel weaves and a study of in vitro corrosion of Mg alloy wires of various chemistries and diameters.

304 stainless steel wires with ~200 diameters were woven into parts with dimensions of 3.6mm x 36mm x 500mm using a 3D weaving machine. Weave samples were then electrochemically coated with HAp using an aqueous solution containing and . Various coating parameters were explored to offer a systematic understanding of the deposition process. The HAp coatings are distributed relatively uniformly across the multiple layers of the weave, suggesting that ion flux is high during deposition and local depletion zones within the 3D weaves are avoided. The coatings consist of “nano flakes” or crystals, and X-ray diffraction (XRD), energy dispersive spectroscopy (EDS), and Raman spectroscopy data confirm that the coating is HAp under the optimized deposition conditions. An in vitro study was then conducted to compare coated and uncoated scaffolds. Live/dead stain and PicoGreen DNA/MTS assays were employed to characterize the overall cell viability as well cell number/metabolism. Post-mortem scanning electron microscopy (SEM) was utilized to investigate to the morphology of the cells on coated and uncoated samples.

Prior to weaving with Mg-based wires, immersion testing was conducted on a range of Mg alloy wires using a modified-simulated body fluid (m-SBF) at 36.5 celsius degree. Weight loss and pH changes are reported as a function of time for multiple chemistries and wire diameters. The corrosion products are relatively uniform along the lengths and diameters of the wires, and they are depleted in Mg as expected. Sites of more intense corrosion appear to correlate with the as-drawn microstructures and cracking within the corrosion product is attributed to the development of tensile stresses within the product. The initial in vitro results suggest that corrosion rates are high and need to be minimized through judicious choices of Mg alloys and wire drawing and annealing parameters when choosing Mg-based wires for 3D weaving.

3:45 PM BM04.08.08
Multi-Responsive Self-Healing Gels as Cellular Delivery Vehicles Aderito J. Amaral and George Pasparakis; University College London, London, United Kingdom.

The development of materials that mimic aspects of self-healing is of paramount importance as it could solve a number of problems in the biomedical field. Interesting self-repair properties have emerged by suitable chemical strategies in bulk materials, which include the incorporation of healing agents in the form of nanoparticles/capsules, or the introduction of dynamic chemical bonds as structural elements. In this regard, remotely controlled healing strategies that allow for on-demand repairing of the material at the site of interest are of utmost importance.[1]

In this work, we propose the construction of remotely healable and transiently malleable soft gels based on the dynamic, albeit covalent, boronate ester bonds formation. We designed a system comprising a synthetic thermoresponsive boronic acid copolymer that is crosslinked with poly(vinyl alcohol) to form hydrogels within seconds under physiological conditions. These constructs can further be impregnated with colloidally stable polyvinylpyrrolidone-coated gold nanoparticles, which render the gels optically active without compromising their mechanical properties significantly. The gel nanocomposites can undergo a rapid and reversible gel-sol transition owing to the disruption of the boronate ester crosslinks by thermal (heating above 37 °C) or optical stimuli (irradiation with green light) that are spatiotemporally confined at the area of interest. The reconstituted gels exhibit shear-thinning behaviour, excellent and fast healing properties, even without the application of any external stimuli. It was also demonstrated that the constructs were able to encapsulate living cells and release them without affecting their viability.[2]
We believe that these materials constitute a versatile biocompatible platform for the construction of remotely healable (soft) gels as cell capture and release systems or soft fillers of biological cavities for tissue engineering.

1. Amaral, A.J.R. and Pasparakis, G., *Polym. Chem.*, 2017, 8, 6464
2. Amaral, A.J.R., Emamzadeh, M. and Pasparakis, G., *Polym. Chem.*, 2018, 9, 525

4:00 PM BM04.08.09
Gene Delivery from Injectable, Hyaluronic Acid-Based Hydrogels with *In Situ*-Forming Macropores Arshia Ehsanipour, Tommy Nguyen, Tasha Aboufadel, Phillip Cox, Chris Walthers, Weikun Xiao and Stephanie Seidlits; University of California, Los Angeles, Los Angeles, California, United States.

Injectable biomaterial scaffolds are ideal for minimally invasive treatments for conditions where the area of injury is difficult to access or ill-defined in structure. Tunable *in situ* forming hydrogels have been developed to facilitate regeneration by providing structural support, introducing mechanical cues, and delivering cells, gene vectors, or soluble signals in a controlled manner. However, injectable scaffolds have limited macrostructural control compared to their implantable counterparts, leading to limited cell infiltration or transduction by gene vectors.

Currently, macroporous injectable scaffolds have been produced using microparticles which can either be 1) annealed *in situ* to provide porous space between particles, or 2) encapsulated within an *in situ* forming gel and degraded to leave behind pores. We investigated the efficacy of both techniques for improving cell infiltration and *in vivo* transduction from viral vectors compared to nonporous controls.

In the first system, particles are formed by water-in-oil emulsion from thiolated hyaluronic acid (HA) and maleimide-terminated poly(ethylene glycol) (PEG). These particles are produced with an excess of thiol moieties to allow for *in situ* disulfide-crosslinking between particles to form gels from microporous annealed particles (MAP). Void space between particles allows for loading of viral vectors and infiltration of cells.

In the second system, encapsulated degradable PEG microparticles (PEG-MPs) act as degradable porogens which can be injected alongside an *in situ* forming hydrogel composed of cross-linked HA and PEG. Thiolated HA and vinyl sulfone-terminated PEG are mixed before injection, forming hydrogels *in situ* within an hour. PEG-MPs degrade rapidly *in vivo*, providing a pore template which facilitates cell infiltration

We demonstrate that the delivery of firefly luciferase-over expressing virus through PEG-MPs or MAP gels both enhance cell infiltration and viral transduction compared to nonporous gels. Infiltrated distance of cells almost doubled in PEG-MP conditions and penetrated the entirety of MAP gels, compared to roughly 100µm in nonporous gels. Bioluminescence due to transduced cell expression of firefly luciferase similarly increased in PEG-MP gels by 1.6x and in MAP gels by 3.3x, relative to nonporous controls. These results show the potential for injectable macroporous gel strategies to facilitate cell infiltration and gene delivery relative to nonporous hydrogels.

4:15 PM BM04.08.10
Biomimetic Mineralized Collagen Scaffolds with Antimicrobial Peptide Coating for Osteogenic Applications Zhou Ye, Xiao Zhu, Christine Lui, Yipin Qi and Conrado Aparicio; Minnesota Dental Research Center for Biomechanics and Biomaterials, University of Minnesota, Minneapolis, Minnesota, United States.

Bone and tooth are organic-inorganic composites with hierarchical nanostructures. The major organic component is type I collagen and the major inorganic component is hydroxyapatite (HAp). Biomimetic mineralized collagen scaffolds with similar compositions and structures have been developed for bone and tooth tissue engineering. A good example is bio mineralization using the polymer-induced liquid-precursor (PILP) process, which shows a great promise in clinical applications. Mineralized collagen scaffolds present strong mechanical properties due to the aligned intrafibrillar HAp crystals and good biocompatibility and osteogenesis. However, the fabricated extracellular matrix also provides an excellent environment for microbial adhesion and biofilm development. Thus, infection could be significant and cause the failure of bone and tooth repair or regeneration.
The use of antimicrobial peptides (AMPs) is a promising approach as they have broad-spectrum antimicrobial activity with low bacterial resistance, as opposed to antibiotics. GL13K is an AMP derived from a salivary protein, which has shown low cytotoxicity and notable antimicrobial activity against gram-positive and gram-negative bacteria and biofilms. In this work, we incorporated GL13K within biomimetic mineralized collagen scaffolds and studied its antimicrobial and osteogenic activity. The collagen gels were mineralized by PILP process for different days and characterized by scanning electron microscope (SEM) and transmission electron microscopy (TEM). The degree of mineralization was quantified by thermogravimetric analysis (TGA) to study the effect of mineral content on the GL13K coating. The distribution of the coated GL13K was investigated by confocal fluorescence microscopy with fluorescent-labeled peptides. Water contact angle analysis revealed that GL13K coating significantly increased the hydrophobicity of the mineralized collagen scaffolds. To study the coating stability, we monitored the released GL13K in solution by mass spectroscopy. To evaluate the antimicrobial activity, *Streptococcus gordonii* was incubated in the scaffolds for 24 hours and the viable cell number was compared using microbial ATP assays. The attached bacteria was also stained by LIVE/DEAD assays and imaged by confocal fluorescence microscopy. Osteogenic activity was evaluated by the adhesion, proliferation and differentiation of mesenchymal stem cells in the fabricated scaffolds. In this work, we introduced antimicrobial peptides in biomimetic mineralized collagen scaffolds. These scaffolds showed strong antimicrobial acivity and osteogenic potential. We also evaluated the specific interactions of collagen and HAp with the antimicrobial peptide so that we will be able to control and further study the effects of each component on the biological properties to optimize their antimicrobial and osteogenic activity.

8:00 PM - 10:00 PM
Hynes, Level 1, Hall B

BM04.09.01
Phenotype and Gene Expression of Human Mesenchymal Stem Cells in Response to Varied Mechanical Environment in 3D GelMA Hydrogel Models Cultured *In Vitro* Kayla Barton, Tayler Laycox, Ella Bonfield, Sara Hopper and Jason W. Nichol; Endicott College, Beverly, Massachusetts, United States.

Mechanical stiffness has been shown previously to direct stem cell differentiation and phenotype in multiple in vitro studies, however, many of these studies use 2D surfaces rather than more physiological 3D environments, as well as 3D encapsulation in materials where stiffness and concentration cannot be independently altered. As such, knowledge of the expression profiles of stem cell differentiation in response to 3D mechanical environment alone is unclear. Biomimetic materials should mimic the extracellular matrix (ECM) and the complex architecture of native tissues to be successful *in vitro* models that give valid cues as to how these cells and tissues would perform *in vivo*. The most popular biomimetic scaffold material for these purposes are hydrogels and we have chosen to work with gelatin methacrylate (GelMA) which has been extensively characterized in numerous models and with various cell types. The degree of methacrylation of GelMA can be controlled to vary the crosslinking density of the resultant hydrogel, allowing for variation of the mechanical properties independent of concentration, while further modulation of stiffness can be effected by altering the gel concentration. Furthermore, the authors and others have demonstrated that GelMA is a versatile and suitable hydrogel for culture of many cell types with positive cellular attachment and natural degradation properties, within a mechanical stiffness range typically up to 30-50 kPa. For these studies, multiple formulations of GelMA were used to study human mesenchymal stem cell (hMSCs) elongation, morphology, proliferation and gene expression using the TruSeq RNA stem cell expression panel over 14 days of static culture. It was hypothesized that the varying elastic moduli of the hydrogels in both the composite and gelMA hydrogels would result in morphological and expression differences typical of what is seen in 2D systems. The data showed that hMSCs elongation occurred early (day 1-4) in both the medium and high stiffness hydrogels with continued growth at day 14 but limited elongation of the hMSCs in the low stiffness hydrogels over 14 days. The morphological differences of the hMSCs between the low, medium, and high hydrogels could indicate the beginning of elastic modulus regulated differentiation. More thorough analysis of cell morphology and RNA expression profiles are still ongoing, but appear to validate the use of this model system both to study stem cell differentiation, as well as to provide an open transcriptome reference data set for other studies

BM04.09.02
Behavior and Phenotype of Cancer Cell Lines in Response to Varied Mechanical Environment in 3D GelMA Hydrogel Models Cultured *In Vitro* Kayla Barton, Tayler Laycox, Ella Bonfield, Sara Hopper and Jason W. Nichol; Endicott College, Beverly, Massachusetts, United States.

Normal, healthy cells will react in response to changes in their mechanical environment typically in an attempt to return their surroundings to homeostatic conditions. In cancerous cells this response is varied, which is one reason why the extracellular matrix (ECM) and mechanical stiffness play key roles in cancer cell phenotype and tumor formation/progression. It is known that cancer cells originating from tissues of different stiffnesses will sense and respond to their environment differently, which could yield important information in better understanding cancer cell physiology. Gelatin methacrylate (GelMA) is a UV-crosslinkable hydrogel that has been shown to be effective in the 2D and 3D culturing of cells in a wide variety of applications including cancer models. Cells can easily bind to GelMA 2D surfaces and within 3D structures, and can proliferate, elongate, and remodel their surroundings due to the presence of natural binding and enzymatic degradation sites in the gelatin backbone. GelMA is a highly elastic material with mechanical stiffnesses demonstrated in the 1 to 30 kPa range through variation of gel concentration and degree of methacrylation. Recently, we have been able to reliably produce hydrogels with compressive moduli below 1 kPa to better mimic the native environment of healthy soft tissues such as breast tissue. It has been established that normal breast tissue has a compressive modulus of roughly 200 Pa, whereas precancerous regions have a modulus of roughly 600 Pa and cancerous regions can be as high as 1-2 kPa, while other tissues have different, but analogous, stiffness ranges as well. The major aims in these studies were to create robust GelMA hydrogels in the 3 mechanical ranges (normal, precancerous, tumor) for breast and other tissues, and once validated to investigate the differences in behavior, morphology and gene expression of cancer cells encapsulated in these hydrogels. Gene expression profiles for normal and cancer specific phenotype will be investigated to determine the relative role in cell behavior in response to changes in stiffness. Initial phenotypic results suggest that cancer cells are less likely to spread, elongate and proliferate when at physiological stiffness, as compared to super-physiological stiffness more on the order of tumor tissues.

BM04.09.03
Doping of Carbon Nanodots with Silver Nanoclusters for Saving Cells from ROS Induced Nanotoxicity Bodhisatwa Das[1, 2] and Santanu Dhara[2]; [1]Biomedical Engineering, Rutgers University, Piscataway, New Jersey, United States; [2]SMST, Indian Institute of Technology, Kharagpur, Kharagpur, India.

Silver nanoparticles are explored for many advanced biological applications including the development of antimicrobial surfaces on implants, SERS imaging, nanotherapeutics, biosensing etc. However, recent research findings suggest silver nanoparticles provide blockade of differentiation of mesenchymal stem cells (MSCs), especially into osteogenic lineage through the generation of ROS. These studies suggest that application of silver nanoparticles in orthopedic implants should be prohibited. In the current study, carbon nanodots(CND) supported silver clusters (AgC) is explored as a remedy to solve this problem. The nanostructure was synthesized in a microwave irradiation induced rapid method and characterization was conducted via UV-Vis spectroscopy, fluorescence spectroscopy, HRTEM, XRD, FTIR, Raman spectroscopy, DLS, AFM, and XPS. Fluorescence spectrum showed a quantum yield of 0.25 while Raman spectroscopy showed rapid amplification of CND specific peaks implicating significant SERS property. Further *in vitro* biocompatibility (MTT) and bio-imaging capability was assessed culturing Wharton's Jelly-derived MSCs. In this study, its efficacy as *in-situ* cellular oxidative stress scavenger is also studied using NBT and DCFH-DA assay. Via ALP assay, alizarin red staining, cell membrane nanoindentation studies, PCR analysis and immunocytochemistry for osteoblast-like gene expression it was confirmed that AgCs can control silver nanoparticle-induced inhibition of osteogenic differentiation *in vitro*. Further, *in* vivo implantation *of AgC-Gelatin-MSC* composite in rodent model showed comparable ectopic osteogenic differentiation potential. Thus AgCs are not only considered to be a dual mode bio-imaging nanoprobe but also a remedy to the silver-induced ROS generation and osteogenic differentiation blockade of MSCs.

BM04.09.04
Enhancement of Hydrophobic Drug Loading in Polymeric Nanoparticles Using a Coaxial Turbulent Jet Mixer Hyeon-Woo Han and Jong-Min Lim;

Soonchunhyang University, Asan-si, Korea (the Republic of).

The development of targeted nanoparticle platforms can indeed open a new age of well-design and tunable release of drug that would revolutionize the field of pharmaceutics. A range of formulation parameters and nanoparticle physicochemical properties has been studied for preclinical evaluations during the development of therapeutic agent loaded targeted polymeric nanoparticles. Although conventional bulk nanoprecipitation methods were widely adopted to prepare polymeric nanoparticles in laboratory scale due to its simplicity and versatility, loading of hydrophobic drug that can maintain uniform NP size distribution is limited. When initial loading of hydrophobic drug is increased to enhance the final loading in nanoparticles, formation of large hydrophobic drug aggregates is inevitable in conventional bulk nanoprecipitation method. Here, we demonstrated a simple and versatile methods to enhance the loading of hydrophobic drug in polymeric nanoparticles, which were synthesized in coaxial turbulent jet mixer by rapid nanoprecipitation. In this work, we used docetaxel as a model hydrophobic drug and PLGA-PEG as a biodegradable nanoparticle matrix. To enhance the docetaxel loading, we systematically studied the following three steps during the nanoparticle preparation. First, we could remove docetaxel aggregates, which were originated from a high initial loading of docetaxel, by adding one more centrifuge step prior to nanoparticles washing in conventional bulk synthesis method. Second, we could enhance the drug loading as well as explore the possibilities of scaling up the nanoprecipitation process by using a coaxial jet turbulent mixer. Finally, we enhanced the drug loading further with the addition of organic co-solvent that would trap the docetaxel and make it easier to be encapsulated in the hydrophobic PLGA core of the PLGA-PEG nanoparticles. The novel strategy has strong possibility to reduce the gap between nanoparticle formulation prepared in academic laboratories and that in pharmaceutical industry thanks to enhanced hydrophobic drug loading (up to 6.5%) in polymeric nanoparticles and extremely high production rate (up to 3.15 kg/d).

BM04.09.05
Self-Assembled Nanoconstructs Modified with Amplified Aptamers Inhibited Tumor Growth and Retinal Vascular Hyperpermeability via Vascular Endothelial Growth Factor Capturing Yeong Mi Lee and Won Jong Kim; POSTECH, Pohang, Korea (the Republic of).

Here, nanoconstructs consisting of a DNA-amplified aptamer with a biocompatible polymer backbone for capturing target biomolecules are presented. First, the polymer–DNA nanoconstructs were prepared by hybridization of two complementary single-stranded DNAs that were each conjugated to a dextran polymer backbone. The designed polymer–DNA amplified aptamer nanoconstructs (PA-aNCs) were then prepared by utilizing polymer–DNA nanoconstructs conjugated with an aptamer (PA-NCs) using a rolling circle amplification reaction to amplify the aptamer. These PA-aNCs were successfully applied to alleviate tumor growth and vascular endothelial growth factor (VEGF)-induced retinal vascular hyperpermeability in vivo through the highly effective capture of human VEGF as a target molecule. These PA-aNCs could be used as therapeutic agent for anti-VEGF therapy by efficiently capturing human VEGF.
Keywords:
anti-VEGF therapy; antitumor therapy; DNA nanoconstructs; polymer−DNA conjugates; retinal vascular hyperpermeability

BM04.09.06
Sterilization by Gamma Radiation of Hydroxyapatite with Brazilian Native Propolis Antonio M. Scatolini, Silvana M. Pugine, Luci C. Vercik, Andres Vercik, Mariza P. Melo and Eliana C. Rigo; University of Sao Paulo, Pirassununga, Brazil.

The aim of this work was to evaluate the possible bactericidal activity the hydroxyapatite (HA) powder containing extracts of green and red propolis, before and after sterilization by gamma radiation (Cobalt 60) with a load of 25 KGy. Ethanol extracts of green (GP) and red (RP) propolis were obtained in 80% alcohol solution. Green and red extracts (8 mg/mL) were incorporated into the material at 10% (w/v) via spray drying, obtaining HA-GP and HA-RP samples. Powder characterization was done by X–ray diffraction (XRD), scanning electron microscopy (SEM) and Fourier Transform Infrared Spectroscopy (FTIR). The cytotoxicity of the materials was determined by the neutral red uptake method. The antimicrobial activity was evaluated by the minimal inhibitory concentration (MIC) and the minimal bactericidal concentration (MBC) against Staphylococcus aureus (S. aureus). The characterization of the powders before and after the sterilization showed crystalline structure and apparently spherical morphology, indicating a decrease in the degree of agglomeration with the addition of propolis, regardless of whether they were sterilized or not. FTIR indicated the presence of functional groups characteristic for HA and propolis. The cytotoxicity assay before and after sterilization showed similar values for IC 50 (concentration reflecting 50% of cell viability) for the HA-GP and HA-RP samples. Higher bactericidal activity was observed for samples without sterilization by gamma radiation. The obtained results in this work suggest that the sterilization process of HA powders with propolis influenced the antimicrobial capacity, however, without any signal indicating that these materials could not be used in medical devices with antimicrobial agent, regarding the requirements of maximum values for the IC 50 index.

BM04.09.07
Graphene Oxide as an Inhibitor of the Interactions Between Nucleoside Diphosphate Kinase (NDPK) and G Proteins Rohit Kanaparthy and Isaac Macwan; Univ of Bridgeport, Bridgeport, Connecticut, United States.

During a heart failure, due to the lack of calcium ion homeostasis in cardiomyocytes, the systolic and diastolic functions are affected leading to a higher amount of nucleoside diphosphate kinase (NDPK) enzyme in the sarcolemma membrane. This inhibits the synthesis of second messenger cyclic adenosine monophosphate (cAMP) that regulates the calcium ion balance for normal functioning of the heart. In a dependent pathway, NDPK normally phosphorylates the stimulatory guanosine diphosphate, $GDP_{(s)}$ to a guanosine triphosphate, $GTP_{(s)}$, on the heterotrimeric (α, β and γ subunits) guanine nucleotide – binding protein (G protein) resulting in the stimulation of the cAMP formation. However, in case of a heart failure, an increased quantity of NDPK also reacts with the inhibitory $GDP_{(i)}$, which is converted to a $GTP_{(i)}$ resulting in the inhibition of the cAMP formation. Typically the βγ dimer of the G protein binds with hexameric NDPK – B/C complex and receives the phosphate at the residue His266 (histidine ID 266) from His118 of NDPK – B. Previous studies have stated that any enzymatic activity can be inhibited by graphene oxide (GO), which is an oxidized carbon allotrope. In this work, the interactions between NDPK – B and – C are quantified in the presence and absence of GO with respect to the binding site His118. These results are further utilized to simulate the interactions between the heterotrimeric $GDP_{(i)}$ and NDPK – B/C with and without GO to explore the binding events between His118 (on NDPK) and His266 (on Gβ). The system was modelled using visual molecular dynamics (VMD) and four 100ns all – atom simulations were carried out using nanoscale molecular dynamics (NAMD). The co-ordinate files for the enzyme and the G proteins were acquired from the pdb database and GO flake (10Å X 9Å) having a chemical structure ($C_{10}O_1(OH)_1(COOH)_{0.5}$) was modeled using nanotube builder in VMD and further modified using TCL scripting and VegaZZ software. CHARMM (Chemistry at HARvard Macromolecular Mechanics) force field is used for modeling and simulating the environment. The temperature is maintained at 300K using Langevin Thermostat and the pressure of 1atm is set through Nose – Hoover Langevin piston

barostat with a period of 100ps and a decay rate of 50ps. It is found that GO is capable of suppressing the interactions between NDPK and G proteins, where it specifically blocks the His118 binding site on NDPK, thereby preventing it to come in contact with His266 on Gβ. Based on the quantification of the van der Waals, electrostatics and conformational energies, it is seen that the affinity of NDPK to GO is higher compared to its affinity to the G proteins. Root mean square deviation (RMSD) and center of mass analysis agrees well with the energetics and the secondary structure analysis of the NDPK and G protein showed only a minor structural arrangement in the protein secondary structure indicating favorable interactions between NDPK and GO.

BM04.09.08

Anti-Tumour Property of Pyrrole Doped Electrospun Pcl Fibrous Scaffold—A Novel Breast Cancer Therapy Ahmet E. Meydan, Ahsen Seyrek and Mehmet Mutlu; TOBB University of Economics and Technology, Ankara, Turkey.

In this study, antitumor biomaterial fabrication targeting the cancer cells without affecting healthy cells was aimed. In accordance with this purpose, heterocyclic pyrrole ring which has antitumor property was doped at 5%, 10%, and 20% (w:w) concentrations to PCL scaffolds via electrospinning. The average fiber diameter of PCl, 5%, 10%, and 20% pyrrole doped scaffolds are measured 1.907 ± 0.286 µm, 1.393 ± 0.177 µm, 0.882 ± 0.218 µm, and 1.100 ± 0.285 µm, respectively, with homogenous, bead free and continuous fiber formation. According to the MTT assay, with 11.045% increase, the highest cell viability was observed in PCL in reference of TCP. Besides, less cell viability was seen in pyrrole doped PCL (PdPCL) scaffolds with increase in pyrrole concentration that resulted in 17.357%, 24.457%, and 36.489% decrease, respectively. According to the DAPI staining results which supported MTT assay data, highest cell viability was observed on PCL against to decrease in PdPCL scaffolds accordance with increase in pyrrole concentration. As a consequence of this research, first time in literature, antitumour property of pyrrole doped into electrospun fiber matrix was achieved. With this study, a novel approach to cancer treatment methods is developed by supporting different biomaterials with pyrrole.

BM04.09.09

pH-Activable Polymeric Nanoparticles Modulate Lysosomal Acidification and Autophagy in Beta Cells Jialiu Zeng[1], Kevin Smith[1], Orian Shirihai[2] and Mark Grinstaff[1]; [1]Boston University, Boston, Massachusetts, United States; [2]University of California, Los Angeles, Los Angeles, California, United States.

We have developed a novel polymeric pH-activable, acidifying nanoparticle (acNP) that restores the pH of compromised lysosomes to rescue autophagic flux and cellular function in pancreatic beta cells (INS1 cells) under lipotoxicity. In beta cells, chronic exposure to high levels of fatty acids (lipotoxicity) leads to an inhibition of autophagic flux and subsequent cellular dysfunction, which has been recently associated with impaired lysosomal acidification and elevated lysosomal pH. Therefore, restoration of lysosomal pH is essential in alleviating the block in autophagy and promote proper cellular quality control and function. In this study, we designed an acidic nanoparticle (acNP) that contains caged acid which can be released upon moderate pH changes (pH 6.0) to enable controlled acidification of the impaired lysosomes under lipotoxicity. Rhodamine labelled acNPs demonstrate dose dependent uptake into lysosomes of INS1 cells. pH-activation of acNPs in dysfunctional lysosomes at pH 6.0 environment demonstrate acidification of lysosomes and restored lysosomal pH with minimal cytotoxicity. acNPs also increased lysosomal cathepsin enzyme activity, and decreased both autophagic proteins LC3II and p62 levels, indicating a rescue of lysosomal function and autophagic flux due to restoration of lysosomal acidity. Additionally, acNPs restored glucose-stimulated insulin secretion that is reduced in INS1 cells and mouse islets under lipotoxicity. These results indicate that acidifying lysosomes with acNPs improved lysosomal function and autophagic flux in INS1 cells under lipotoxicity, and are of therapeutic interest for pathologies associated with lysosomal acidity impairment such as type II diabetes and non-alcoholic fatty liver disease (NAFLD).

BM04.09.10

Synthesis and Characterization of Non-Cell-Adhesive Gelatin/Pluronic Hybrid Hydrogels Juyi Li, Clement Marmorat, Miriam Rafailovich and Marcia Simon; Stony Brook University, Stony Brook, New York, United States.

Non-cell-adhesive biohydrogels stand important roles in many in vivo applications. In the case of periodontitis, a biohydrogel non-cell-adhesive material could isolate the soft gum tissue from the hard bone and promote selective growth of both tissues on each side of the membrane. Many shortcomings are associated with the use of current materials like polytetrafluoroethylene including degradability, workability, cost or cytotoxicity. In this study, we introduce a novel cost-effective material, a hybrid biohydrogel of gelatin and Pluronic F127, strong enough to withhold mechanical degradation during the surgical procedure but yet degradable *in vivo*, non-cytotoxic, porous to promote the diffusion of nutrients and physiological fluids while remaining anti-adherent to gingival fibroblasts, would be ideal for this type of application. The hybrid gels were crosslinked via microbial transglutaminase (mTG). Hybrid gels with different ratio of gelatin, F127 and mTG were synthesized. Rheological properties of those gels were determined by rheometer and the surface and side cut section of those gels were observed by laser scanning microscope and scanning electron microscope. UV-vis and FT-IR spectroscope were used for structure characterization of the hybrid gels. Fibroblast with green fluorescent were seeded on the surface of hybrid gels to determine cell adhesive ability. Our results showed the synthesized hybrid gels preserve the mechanical stability of gelatin-based hydrogels, while also exhibiting excellent workability and non-cell-adhesive properties of Pluronic F127.

BM04.09.12

Antibiotic Eluting Contact Lenses for the Treatment of Bacterial Keratitis Liangju Kuang[1], Lokendrakumar C. Bengani[1], Amy Ross[1], Daniel S. Kohane[2] and Joseph B. Ciolino[1]; [1]Department of Ophthalmology, Massachusetts Eye and Ear Infirmary, Harvard Medical School, Boston, Massachusetts, United States; [2]Departments of Anesthesia and Surgery, Boston Children's Hospital, Harvard Medical School, Boston, Massachusetts, United States.

Bacterial keratitis is one of the leading causes of corneal blindness. The current standard of care involves an intensive regimen of hourly antibiotic drops to eradicate the infection. A major unmet need for the treatment of bacterial keratitis is a sustained approach of drug delivery to improve treatment efficacy and patient adherence. Contact lenses are emerging as a promising platform for controlled ocular drug delivery and potential release of drugs to the back of the eye. We report here an antibiotic-eluting therapeutic contact lens (TCL) that incorporates a thin drug-polymer film within the periphery of a contact lens using standard materials, which enables the sustained release of drugs at therapeutic rates, while allowing unimpeded vision through a central aperture in the lens. Antibiotic–polymer films were encapsulated in methafilcon via ultraviolet light polymerization and then lathed into contact lenses. Various UV polymerization conditions were examined to achieve the suitable physical properties of TCLs. The physical properties and morphology of antibiotic-eluting TCLs were characterized. Fourth-generation fluoroquinolones were selected because of their broad spectrum antibacterial activities and high potency. The physiochemical interactions between fluoroquinolones and polymers in the drug-polymer films were characterized by Fourier-transform infrared spectrometer. *In vitro* drug release was evaluated under infinite sink conditions. TCLs enabled the sustained release of besifloxacin and moxifloxacin for

more than 24 hours in clinically relevant mass. The release of gatifloxacin from TCLs was also achieved in a temporally controlled manner but was faster than that of besifloxacin and moxifloxacin. Additionally, *in vitro* drug release profiles were tuned by polymer characteristics (hydrophobicity, hydrophilicity, and molecular weight), polymer/drug ratios, polymer/plasticizer ratio, and polymer ratios in the blend film. The antibiotic-eluting TCL may be used as a convenient alternative for extended ocular drug delivery with translational potential for the treatment of bacterial keratitis.

BM04.09.13

Self-Assembled Peptide Nanotubes for Neural Cell Proliferation Prathyushakrishna Macha, Vikas Soni, Maricris Mayes and Milana Vasudev; University of Massachusetts Dartmouth, Dartmouth, Massachusetts, United States.

Self-assembly, a process that assembles molecules into ordered structures can form numerous structures in various conditions. Self-assembled structures when made from biomolecules such as DNA and peptides, form functional and biocompatible nanostructures such as nanotubes, fibers, spheres, which could be used in different biomedical applications. We have synthesized aromatic dipeptide-based nanotubes using dityrosine and tryptophan-tyrosine through solution-phase self-assembly (SPSA) and eco-friendly plasma enhanced chemical vapor deposition (PECVD). Insights into the self-assembly process and driving forces involved were obtained using quantum chemical computational methods at different levels of theories like dispersion-corrected density functional and Moller-Plesset perturbation.

The morphological features of SPSA and PECVD nanotubes were studied using confocal and scanning electron microscopes. These nanotubes were characterized using thermally and spectroscopically using differential scanning calorimetry and thermogravimetric analysis, Fourier transform infrared spectroscopy, liquid chromatography-mass spectroscopy, and Raman scattering, and circular dichroism spectroscopy, and powder x-ray diffraction. The cytotoxicity and biological interactions of these nanotubes with rat adrenal pheochromocytoma (PC-12) and human bone marrow neuroblasts (SH-SY5Y) cells were studied using MTT (3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide), dopamine-enzyme linked immunosorbent assay and real-time polymerase chain reaction (q-PCR). We observed substantial differences in proliferation and expression between the cells grown on tissue culture treated and nanotubes coated surfaces.

BM04.09.14

Creating Rechargeable Anti-Thrombotic Surfaces via Enzyme Mediated Ligation Hyun Ok O. Ham[1, 2, 3], Madhukar S. Patel[1, 2, 3], Carolyn Haller[1, 2, 3], Erbin Dai[1, 2, 3], David R. Liu[4], Jian Liu[5] and Elliot Chaikof[1, 2, 3]; [1]Harvard Medical School, Boston, Massachusetts, United States; [2]Wyss Institute, Boston, Massachusetts, United States; [3]Beth Israel Deaconess Medical Center, Boston, Massachusetts, United States; [4]Harvard University, Cambridge, Massachusetts, United States; [5]University of North Carolina at Chapel Hill, Chapel Hill, North Carolina, United States.

Foreign body reactions at the blood-material interface frequently result in implant failure. Current engineered anti-thrombotic coatings are challenged by bioactive surface breakdown resulting in device-associated complications. This study aims to develop a system for generating ‘rechargeable’ surfaces utilizing reversible bond formation between bioactive molecules and modified implant surfaces through enzyme-mediated ligation. In this scheme, reversibility allows for stripping of degraded coatings with subsequent surface recharging.

Variants of laboratory evolved *Staphylococcus aureus* sortase A (eSrtA) were generated for reversible transpeptidation of two anti-thrombotic molecules (recombinant thrombomodulin [TM] and heparin oligosaccharide [HS]) on implantable medical devices. Two eSrtA variants were designed to specifically recognize different peptide tags available on therapeutically active TM and HS. Using eSrtA, repeated charging/ stripping of TM was conducted *in vitro* in whole blood, and subsequent generation of activated protein C (aPC) was demonstrated. Similarly, chemo-enzymatically synthesized ultra-low molecular weight heparin oligosaccharide (HS), which binds to antithrombin III that can inhibit factor Xa activities involved in the blood coagulation cascade reaction, was repeatedly immobilized and stripped utilizing reversible enzyme ligation. Within an hour of incubation on pentaglycine-modified surfaces, eSrtA variants could selectively charge/strip of TM and HS in a sequential or simultaneous manner. Alternating charging/stripping of TM/HS on the surface and thereby switching on/off of aPC generation and anti-Fxa inhibition from the surface was also validated. For a translational application, reactions were subsequently performed and confirmed on the luminal side of implantable vascular catheters.

The ability to regenerate bioactive coatings using eSrtA variants is a means by which engineered implant surfaces can be functionally preserved. Furthermore, using this system to perform site-specific co-immobilization of different anti-thrombotic moieties on vascular catheters and selectively regenerate bioactivity has potential to decrease long-term implant related morbidity through targeted action on distinct parts of the coagulation cascade.

BM04.09.15

Internal Design and Fabrication of Tissue Scaffolds Ozlem Yasar[1], Ozgul Yasar-Inceoglu[2] and Joyce Tam[1]; [1]City University of New York, Brooklyn, New York, United States; [2]Mechanical Engineering, California State University, Chico, Chico, California, United States.

In the field of tissue engineering, design and fabrication of precisely patterned, highly porous scaffolds/matrixes are required to guide overall shape of tissue growth and replacement. Although Rapid Prototyping fabrication techniques have been used to fabricate the scaffolds with desired design characteristics, controlling the interior architecture of the scaffolds has been a challenge due to CAD constrains. Moreover, large thick tissue scaffolds have reported limited success primarily due to the inability of cells to survive deep within the scaffold. Without access to adequate nutrients, cells placed deep within the tissue construct die out, leading to non-uniform tissue regeneration. This study aims to overcome these design and fabrication limitations. In this work, research has been expanded to design of scaffolds which have inbuilt micro scale fluidic networks. In this procedure, inbuilt channels serve as material delivery paths to provide oxygen and nutrients for the cells. First of all, negative of a cylindrical shape with a single channel was designed with AutoDesk Inventor and printed with a 3D printer to be used as a mold. Then, 3D printed mold was filled out with Poly(ethylene glycol) diacrylate (PEGDA) which is a photo-curable solution to fabricate the cylindrical hydrogel. Once PEGDA was exposed to UV light with the wavelength of 365nm, polymerization completed in about 3 minutes. After that, the same procedure was repeated for cylinders with two and three channels respectively. Then, their mechanical characterization tests were done to compare the compressive strengths of the scaffolds that has different internal architectures. Our preliminary results indicate that, 3D printing and polymerization techniques can be used together to control the interior architectures as well as the compressive strengths of scaffolds.

BM04.09.16

Antibacterial Properties of 2D Black Phosphorus Nanosheets for Regenerative Engineering Emre Firlar[1, 2], Laura Alzate[3], Ramin Rojaee[2], Reza Shahbazian-Yassar[2] and Tolou Shokuhfar[1]; [1]Bioengineering, University of Illinois at Chicago, Chicago, Illinois, United States; [2]Mechanical and Industrial Engineering, University of Illinois at Chicago, Chicago, Illinois, United States; [3]Biomedical Engineering, EIA University, Envigado, Colombia.

2D black phosphorous is an emerging new material with implications for regenerative engineering. Bacterial infection during implant surgeries has been a major concern for the long-term stability of the installed implant due to the health and cost burdens on the patients. Osseointegration between the implant and tissues will deteriorate due to the presence of bacteria in addition to the spread of bacteria through-out the body. Therefore, novel biocompatible nanomaterials should be developed to fight against the bacteria. Black phosphorus (BP), which is biocompatible, has been proven to be effective in this regard, but mainly with irradiation with near infrared light. Extensive irradiation of photons to emit singlet oxygen will worsen the effectiveness of BP and cause hypoxia to the cells. Thus, independent irradiation strategies should be developed to kill the bacteria using BP. In this work, *E. coli* and *M. magneticum* were cultured and the effectiveness of BP was tested for their antibacterial properties through fluorescence imaging, cell counting and transmission electron microscopy (TEM).
BP, in the presence of water, forms a phosphate layer. Further reaction with oxygen from water might constitute dangling bonds, which might protect the underneath layers. Further reaction with water results in the formation of weak H_3PO_4 acid. The reaction of H_3PO_4 with the bacteria can kill the bacteria by releasing a protein and leading to acidification of the cytoplasm and rupture to the bacteria's wall.
Chemical exfoliation of BP was carried out to have BP nanosheets. By monitoring the O-K edge via electron energy loss spectroscopy, we observed that BP forms an oxide layer when transferred from IPA to water. During antibacterial tests, E. *Coli* was grown in LB broth and scraped onto agar plates and then single colonies from the agar plate were cultured on a freshly prepared LB broth. For colony counting tests, BP was added to the bacteria culture and sat for 6-20 hours where they were then scraped onto agar plates. Cell counting test was carried out by comparing the BP treated, bleach treated and untreated E. *Coli*. The effect of BP on the bacteria viability was also monitored via fluorescence microscopy. It was observed that BP reduces the viability of both *E. coli* and *M. magneticum*. The interaction of BP with *E. coli* was further investigated by conventional TEM experiments, specifically, interaction of BP with bacterial cell membrane and the changes in the ultrastructure of bacteria was observed.

BM04.09.17
The Effects of Graphene in Different Morphologies of Polymer on Dental Pulp Stem Cells Linxi Zhang[1], Kuan-Che Feng[1], Chung-Chueh Chang[2], Marcia Simon[3] and Miriam Rafailovich[1]; [1]Materials Science and Engineering, Stony Brook University, The State University of New York, Stony Brook, New York, United States; [2]Advanced Energy Center, Stony Brook University, The State University of New York, Stony Brook, New York, United States; [3]Oral Biology and Pathology, Stony Brook University, The State University of New York, Stony Brook, New York, United States.

Graphene and graphene-based materials have been developed and widely used in tissue regeneration engineering, due to their excellent physical properties. Many studies have shown that graphene can control and accelerate multi-lineage differentiation of stem cells *in vitro*. Graphene are commonly used, in most studies, as the substrates which have direct contact with cells. In general cells do not interact directly with substrates. Rather, extracellular matrix proteins secreted by the cells adsorb first, coating the substrates, enabling cell adhesion. Hence as was previously shown, the response of the cells on these substrates will depend on the conformation of the adsorbed ECM. Therefore, in order to comprehend the mechanism through which graphene affects stem cell differentiation, it is important to understand the influence of graphene on ECM protein structures. We have previously shown that the differentiation of dental pulp stem cells is greatly affected by the substrate morphology of poly(4-vinylpyridine) (P4VP). It provides a perfect platform to study the the additional effect of graphene on the cells. We found that graphene can be easily distributed into this system by electrospinning and spin coating process. The cell behavior and biomineralization and differentiation of DPSCs on the graphene-containing scaffolds are determined by multiple techniques, including SEM, Raman and RT-PCR. The results show that the cell-secreted ECM structure and biomineralization on different scaffolds are affected by the addition of graphene in the original polymer matrices.
The research was supported in part by the NSF (Inspire Award #1344267) program.

BM04.09.18
Biomimetic, 3D Hydrogels to Investigate Effects of Microenvironment Biophysical Properties on Patient-Derived Glioblastoma (GBM) and Endothelial Cells (ECs) Alireza Sohrabi, Jesse Liang and Stephanie Seidlits; University of California, Los Angeles, Los Angeles, California, United States.

Introduction: This study investigated how the mechanical microenvironment of glioblastoma (GBM) tumors may affect morphology and phenotype of patient-derived GBM and brain endothelial cells (ECs) – both critical components in the perivascular invasive niche.
Materials and Methods: Hydrogels were fabricated from thiolated hyaluronic acid (HA-SH) (0.5 w/v%) and 4-arm PEG-SH (varied), 8-arm PEG-norbornene (varied). Gelation occurred upon exposure to UV light (365 nm, 4 mw/cm^2) for 15 s in the presence of a cytocompatible photoinitiator LAP (Lithium Pehnyl (2, 4, 6-trimethylbenzoyl) phosphinate, 0.025 w/v%). Patient-derived gliomaspheres or single dissociated ECs were mixed with hydrogel precursors prior to gelation. Phase contrast images were acquired every 3 days to monitor cell migration. After 9 days, 3D cultures were fixed, and cells visualized with CellMask™ Green (ThermoFisher) and Hoescht (nuclei). Cell survival following encapsulation was evaluated using a Live/Dead Assay (Life Technologies). A Leica LSP5 confocal microscope was used to image 3D cultures. Storage moduli (G') of hydrogels were measured using shear rheometer (TA DHR-2) with an 8-mm flat plate geometry.
Results and Discussion: Hydrogel stiffness (G') was controlled by varying Thiol content to achieve a range of 150–1500 Pa, which are representative of the normal brain and tumor microenvironments, respectively. Gliomaspheres and ECs cultured showed comparable survival 7 days post-encapsulation in both soft and stiff gels. Gliomaspheres cultured in stiff hydrogels remained did not migrate away from spheroids, while cells encapsulated in soft hydrogels exhibited robust migration.
Our previous results showed that ECs residing in GBM perivasculature differentially express high amount of integrin-binding sialoprotein (IBSP). Survival of ECs in hydrogels baring different biophysical properties is an appropriate platform to study the effect of stiffness on ECs protein expression.
Conclusion: Patient-derived GBM and ECs were successfully cultured in 3D HA hydrogels mimicking biophysical properties of brain or GBM tumor tissue. GBM cells cultured in stiffer environments were unable to migrate, but upon culture in soft environments were found to migrate up to 200 µm away from seeded spheroids within 9 days of cultures.

BM04.09.19
Tissue Engineered Scaffolds—Next Generation 3D Bone Mimetic Cancer Metastasis Testbeds Kalpana Katti, MD Shahjahan Molla, Sumanta Kar and

Dinesh R. Katti; North Dakota State University, Fargo, North Dakota, United States.

Tissue engineering through use of scaffolds is a very promising technology for replacement of tissues In regenerative medicine. A unique nanoclay based scaffold that enables mineralization of hydroxyapatite inside nanoclay galleries provides structural support and enables growth, proliferation and differentiation of human mesenchymal cells to form hierarchical mineralized collagen and ECM formation mimicking bone which is useful for regenerative medicine. In addition, it is known that breast cancer and prostate cancer have the propensity to metastasize to bone in the human body. The cancer at this stage of metastasis is incurable. Here we describe the use of tissue engineered bone as a humanoid testbed to create cancer tumors in vitro. We also demonstrate that this engineered test-bed duplicates the last stage of cancer metastasis as indicated by the gene expression and immunocytochemistry analysis of the tumors generated in the testbed. We also report the use of unique nanoindentation methodology to obtain elastic properties of tumors as they grow at the humanoid metastasis stage inside the bone scaffolds. FTIR experiments are also conducted during progression of tumor at metastasis and unique signatures of the DNA and protein contents during cancer progression are reported. Nanomechanical experiments on live tumors during their evolution and growth is related to the gene expression studies to bring mechanobiology as a new biomarker of the cancer progression. The engineering test-bed can be used for personalized medicine as well as a screening tool for new anti-cancer drugs. Regenerative medicine thus provides unique opportunities to evaluate cancer metastasis.

BM04.09.21

Mechanical Behavior of Collagen Hydrogel Network Through Tensile Testing on Water Surface and Microstructure Analysis Jieung Kim[1], Sangmin Lee[3], Hyunjoon Kong[2], Taek-Soo Kim[3] and Dongchan Jang[1]; [1]Nuclear and Quantum Engineering, Korea Advanced Institute of Science and Technology, Yuseong-gu, Korea (the Republic of); [2]Chemical and Biomolecular Engineering, University of Illinois at Urbana-Champaign, Urbana, Illinois, United States; [3]Mechanical Engineering, Korea Advanced Institute of Science and Technology, Yuseonggu, Korea (the Republic of).

Collagen is at once a main structural material and the most abundant protein in vertebrates. Collagen hydrogel reflects mechanical behavior of human tissues and is the most commonly used scaffold material and extracellular matrix in tissue engineering. As a structural material, it bears and loads stress and affects cell behavior depending on mechano-sensitivity of each cell type. Thus, providing prediction of cell response and mechanical reliability of collagen hydrogel presents significant challenge, so it is essential to understand mechanical properties of aforementioned material. However, soft collagen hydrogels were only tested shear, compressive and rheological properties because they have instability by gravitational forces and dehydrating issues at existing tensile testing methods. In addition, polymer network structure of collagen hydrogel varies under applied stress with showing non-linear stress-strain relationship and mechanism for the non-linearity was not proven yet. To predict hydrogels' mechanical behavior completely, previously unavailable tensile data is required, and an alternative methodology is necessary to collect tensile properties of collagen hydrogel. In this study, we aim at investigating the mechanical behavior of collagen hydrogels under quasi-static tensile stress by suggesting a solution for tensile testing of soft collagen hydrogel and providing its mechanical response to tensile stress. Tensile testing on water surface imparts hydrating and free-standing environment to collagen hydrogels. Moreover, microstructure analysis of collagen hydrogels using SEM, and nano-scale 3D X-ray tomography demonstrates the mechanism of shown tensile properties. As the fiber diameter increases and the crosslink density decreases with decreasing gelation temperature, the tensile modulus due to the network structure change increases. In addition, the higher the collagen concentration, the more the crosslink density and the elastic modulus increase. After the first regime of mechanical behavior, the entropic deformation of the collagen network is completed and the enthalpic deformation of the network begins. These results illustrate that collagen hydrogels respond in complex manners to tensile stress, either as network or as fiber, which shows non-linear stress-strain relationship. Furthermore, we propose a new approach for tensile testing method for soft and hydrated materials that can be applied for bioengineering field.

BM04.09.22

Sphingosine-1-Phosphate-Bound High-Density Lipoprotein-Mimetic Nanoparticles Engineered to Protect Endothelial Functions in Cardiovascular Disease Hyun-Ji Park, Jiwon Yom, Yoshitaka J. Sei and YongTae Kim; Georgia Institute of Technology, Atlanta, Georgia, United States.

Sphingosine-1-phosphate (S1P) is a natural signaling sphingolipid known to have important roles in regulating vascular and immune systems. In human plasma, S1Ps are associated with high density lipoprotein (HDL) of apolipoprotein A1 (ApoA1) and apolipoprotein M (ApoM) and leads to pleiotropic functions through specific receptors such as S1PR. Specifically, S1P-bound HDL (HDL-S1P) regulates several key biological functions in vascular system, such as endothelial nitric oxide (NO) production and vasodilation. Plasma S1P levels are reportedly lower in patients with coronary artery disease, indicating that S1P is involved in the pathogenesis of atherosclerosis. However, the pathophysiological and molecular mechanisms leading to such disease-associated shifts in HDL-S1P remain elusive due to multiple reported arguments of the biphasic, pro- and anti-atherosclerotic properties. Here, we leverage our *in vitro* human carotid artery-mimicking model system to study the biological function of S1P using engineered HDL-mimetic nanoparticles (eHNPs). Our advanced microvortex technology enhanced assembly of hydrophobic lipids (DMPC and/or S1P) and hydrophilic proteins (ApoA1 and ApoM) to continuously produce homogenous discoidal eHNPs, such as eHNP-A1, eHNP-A1-S1P, and eHNP-A1-M-S1P with the size range of 10 to 15 nm, which is consistent to the native HDL. These eHNPs were endocytosed to human carotid artery-derived cells such as human carotid artery endothelial cells and smooth muscle cells. Moreover, the uptake efficiency of eHNP-A1 was enhanced by incorporating S1P into the eHNP-A1 (eHNP-A1-S1P). More interestingly, we successfully reproduced natural carotid arterial functions such as endothelial NO production and vasodilation using our *in vitro* cardiovascular model system and investigated the physiological functions of eHNPs under pathophysiological conditions such as TNF-α stimulation or disturbed flow. This study helps to understand the mechanisms of endothelial effect of HDL and S1P and provides clues to improve HDL-based clinical studies of cardiovascular diseases. Our integrated approach using the designed eHNP complex and *in vitro* cardiovascular model system reported in this study are promising prescreening alternatives to costly animal model studies to study the structural and functional heterogeneity of HDL. Moreover, this study potentially leads to novel therapeutic measures that can prevent the progression of cardiovascular diseases such as atherosclerosis and coronary artery diseases.

BM04.09.23

Cardioprotective Effects of *Asparagus racemosus* Against Doxorubicin Induced Cardiotoxicity in Rats Manisha Chatterjee, Raj Kumar Goel, Manish Saini, K.K. Saxena, Pinki Vishwakarma and Monica Sharma; Lala Lajpat Rai Memorial Medical College, Meerut, India.

Recently, herbal remedies have raised considerable research attention as they are safer, less expensive with fewer side effects than synthetic drugs. Here we report, the cardioprotective potential of medicinal plant *Asparagus racemosus* against cardiotoxicity induced by anticancer drug Doxorubicin. The

comparison was done with the standard cardioprotective drug (Carvedilol) in albino rats. For this experimental study, the albino rats were divided into four different groups namely Control group (administered pellet diet and tap water for 21days), Toxic control group(in addition to pellet diet and tap water this group was administered Doxorubicin in a single dose intraperitoneally on 21st day), and two Test groups (they were administered aqueous extract of *Asparagus racemosus* in doses 250mg/kg and 500mg/kg respectively for 21 days, followed by Doxorubicin in a single dose intraperitoneally on 21st day) and a Standard group (treated with Carvedilol per orally for 21days followed by Doxorubicin in a single dose intraperitoneally on 21st day). All animals were sacrificed 48 hours after Doxorubicin administration. Blood samples were collected and different pathological tests like CK-MB, LDH, SGOT, and SGPT were performed. It was observed that the serum levels of CK-MB, LDH, SGOT, and SGPT were raised significantly in the Toxic control group, whereas these values were raised but to a lesser extent in the test group treated with *Asparagus racemosus* in dose of 250mg/kg, and these were within the normal limits in the test group treated with *Asparagus racemosus* in dose of 500mg/kg which was comparable to the standard group treated with Carvedilol. The histopathological tests of the dissected heart also confirm these observations. Collectively, these data indicate that *Asparagus racemosus* pretreatment could alleviate doxorubicin-induced cardiotoxicity

BM04.09.24
CHANNELMAT—Controlling Mechanotransduction in Porous Biomaterials Mohammadreza Taale, Katharina Siemsen, Christine Arndt, Fabian Schutt, Rainer Adelung and Christine Selhuber-Unkel; University of Kiel, Kiel, Germany.

Cells respond to external mechanical stimuli through a biological process called mechanotransduction. Mechanotransduction has great impact on cellular proliferation, migration and differentiation, as well as on cell adhesion. Likewise, diseases such as cancer and cardiac dysfunctions are also related to cellular mechanotransduction. Here we show data on controlling cells by an innovative 3D material that serves as a platform for controlling mechanotransduction by mimicking natural 3D cellular environments. Our material contains microporous structures represented by micron-sized channels that are embedded in a soft hydrogel matrix. For controlling mechanotransduction, the stiffness of the matrix is well-defined by the amount of cross-linker in the hydrogel. In addition we modify the hydrogel such that its surface is equipped with cell adhesion ligands, e.g. RGD and collagen. As the pores in the material provide a large and spatially controlled cell-surface contact area of up to 80%, the mechanical properties of the hydrogel environment will have large impact on the cells. As an additional feature, we can also control the conductivity of the porous hydrogels by the targeted addition of carbon nanomaterials such as CNTs and graphene. We here show first data on the biocompatibility of the materials, on cell growth in the materials and how we can control the properties of cells by our materials.

BM04.09.25
Efficient Excision of Proviral HIV-1 Genome in Human Primary Cells with Cell Penetrating TALEN Nanocapsules Ming Zhao, Jing Wen and Yunfeng Lu; University of California, Los Angeles, Los Angeles, California, United States.

A critical step during human immunodeficiency virus (HIV) infection occurs when the complementary viral DNA is integrated into the host genome. This so-called “HIV provirus” becomes transcriptionally silent and sustains as a latent viral reservoir that can escalate new infections upon self-reactivation. Though the present anti-retroviral therapies could effectively suppress active virus replication, the latent HIV-1 reservoirs remain intractable and a tremendous risk to infected individuals. Therefore, a novel therapy that could abrogate proviral DNA is highly desirable but challenging.
Genome-editing technologies whereby cellular genomic information is readapted have become increasingly utilized recently. Targets for gene modification, including the HIV-1 genome itself, have been investigated as a potential therapy for HIV-1 disease using the clustered regularly interspaced short palindromic repeat (CRISPR)/Cas9 endonuclease system and the transcription activator-like effector nucleases (TALEN), yet the off-target cleavage of the CRISPR/Cas9 remains a concern by many scientists. In contrast, TALEN possesses significant accuracy and robust nuclease activity for the targeted sequence. So far, cell-penetrating peptide (CPP) mediated TALEN delivery has been applied to knock down the HIV-1 co-receptor, CCR5, yet the working concentration of TALEN protein was excessively high (~μM). Likewise, transfection of TALEN mRNA abolishes HIV-1 proviral function drastically in T cell lines, which, however, is ineffective in disrupting proviral DNA in primary T cells.
Herein, we report a proof-of-concept study that TALEN protein under the guidance of a pH-sensitive nanocapsule could be effectively delivered into the nucleus. The cationic surface charge and the acid-labile crosslinkers of the nanocapsule enable high-efficient transmembrane transportation, cargo release and avoidance of endosome/lysosome sequestration. As a result, the delivered TALEN demonstrated good gene-editing capability under an ultra-low working concentration. In particular, HIV-1 latent reservoirs in primary T cells were efficiently excised with this strategy, thereby providing a useful suggestion for developing anti-HIV drugs, particularly against integrated HIV genome in primary cells.

BM04.09.26
Hierarchical Decoration of Eggshell Membrane with Polycaprolactone Nanofibers to Fabricate a Bilayered Scaffold for Full-Thickness Cutaneous Wound Healing Preetam Guha Ray, Pallabi Pal and Santanu Dhara; BMTE, School of Medical Science and Technology, Indian Institute of Technology Kharagpur, Kharagpur, India.

The modern era of skin tissue engineering focuses on engineering and fabrication of scaffolds that can closely mimic the extracellular matrix and its microenvironment so as to facilitate cell adhesion and proliferation. The objective of the present study is to design and fabricate a porous, mechanically stable, biocompatible and non-immunogenic bilayered scaffold for wound healing applications. In order to do so, microfibrous eggshell membrane (ESM) along with nanofibers of Polycaprolactone (PCl) was deployed. In addition to a rich source of collagen, ESM also contains several growth factors and GAGs, essential for the regeneration process. A homogeneous blend of PCl was electrospun onto the ESM to decorate it with randomly arranged nanofibers (200 nm diameter), followed by cross-linking the same with microfibers (2μm diameter) using NHS/EDC coupling (EpN). The surface topography of the 110 μm thick bilayered scaffold was explored using FE-SEM and AFM, while the porosity was evaluated using BET analysis. The spectroscopic analyses using FT–IR and XPS reveal successful crosslinking between the nanofibers of PCl and microfibers of ESM in the scaffold which corroborates to FESEM and AFM results. The matrix exhibits considerable enzymatic degradation after 27 days, 70% wettability, tensile strength of 15 MPa and substantial anti–microbial activity. In addition, MTT, Rhodamine–DAPI assay and FESEM studies demonstrates excellent cell adhesion and proliferation of human dermal fibroblast (hDF) cells. Furthermore, the samples presented enhanced wound healing characteristics when grafted over a full thickness wound on a rat model. Moreover, the histopathological examination of the treated wounds at different time intervals revealed fast re-epithelization and collagen deposition in the extracellular matrix. EpN implanted wounds demonstrated complete wound closure within 14 days. Excellent physico-chemical properties and superior wound healing efficacy demonstrated by micro/ nano architectured EpN mats make it a potential wound healing matrix for clinical skin regeneration.

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Terminal Autoclave Sterilization—Impact on Cryogel Properties and Injectability Sidi Bencherif[1, 2, 3], Pierre Villard[1], Mahboobeh Rezaeeyazdi[1], Kasturi Joshi Navare[1], Colombani Thibault[1] and Adnan Memic[1, 4]; [1]Department of Chemical Engineering, Northeastern University, Boston, Massachusetts, United States; [2]Harvard John A. Paulson School of Engineering and Applied Sciences, Harvard University, Cambridge, Massachusetts, United States; [3]Sorbonne University, UTC CNRS UMR 7338, Biomechanics and Bioengineering (BMBI), University of Technology of Compiègne, Compiègne, France; [4]Center of Nanotechnology, King Abdulaziz University, Jeddah, Saudi Arabia.

While effective sterilization is crucial for clinical utilization of biomaterials, maintaining both their structural and biological properties post-processing remain a challenge. One of the most popular terminal sterilization methods is heat sterilization and more particularly autoclaving. Autoclaves use highly pressurized steam heated up to 134°C which is detrimental to many biomaterials including polymeric hydrogel scaffolds. Three-dimensional scaffolds are attractive for many tissue engineering applications due to their unique properties such as high-water content, tunable mechanical properties and biocompatibility. However, hydrogels typically get sanitized not sterilized, especially in an academic setting, as they are unable to survive the autoclave sterilization process.

Recently, we have developed a new class of more robust hydrogels with unique properties. These cryopolymerized hydrogels or cryogels do not only possess large and interconnected pores, and mechanical robustness sustaining up to 90% deformation, but also shape memory properties allowing their injection through conventional needles. A series of injectable cryogels prepared with various biopolymers have been autoclaved and tested not only for their degrees of sterilization but also for their physico-chemical integrity (i.e. injectability, physical properties, and retention of their intrinsic biological properties). Overall, our preliminary results suggest that unlike conventional hydrogels, injectable cryogels are resilient to the aggressive nature of steam sterilization and are well suited as safe and non-invasive biomaterials for a number of biomedical applications.

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BM04.09.28

Advanced Drug Delivery Platform—Silica Nanoparticles Engineered with Nanoscale Surface Roughness Hao Song and Chengzhong Yu; The University of Queensland, Brisbane, New South Wales, Australia.

Recent advances in nanotechnology have greatly boosted the development of drug delivery systems, while the key challenge still lies in the rational design and fabrication of safe and efficient nano-carries. It is noteworthy that the delivery performance could be possibly maximized by custom-designed nano-carriers considering the configuration and surface textures of both cargo biomolecules and target cell/environment. Here, we showcase our recent progress on the development of silica-based advanced delivery platform by engineering the surface textures at the nanoscale. First, through a biomimetic approach, silica nanopollens with an intrinsic spiky surface are fabricated.[1] The nanoscale surface roughness exhibits interesting surface properties at the microscale, enabling the nanoparticles with strong adhesion toward the hairy bacterial surface, thus leading to an efficient antimicrobial enzyme delivery. Then this novel delivery system is well extended for gene delivery.[2] Distinct from small drug or protein molecules, plasmid DNA possesses unique rope-like loop structures. The spiky nanotopography is here further demonstrated with great advantages at the nanoscale, acting as hooks to entangle the DNA loops and protect the gene molecules sheltered in the spiky layer against nuclease degradation. These spiky nanoparticles show excellent gene transfection efficacy, especially under harsh enzymatic conditions. The intriguing properties enabled by this unique nanoscale rough feature have been further explored in our recent studies. This rough nanoparticle-based novel delivery platform is of significant potential in practical biomedical translations, such as antibiotic-free animal feed and DNA vaccines.

BM04.09.29

pH Sensitive Nanoparticles to Study the Biofilm Interfacial Microenvironment Padryk Merkl and Georgios A. Sotiriou; Karolinska Institutet, Stockholm, Sweden.

Implant associated infections caused by biofilms are a major cause of implant rejection. One promising strategy to help combat biofilms are “smart” anti-biofilm release surfaces. These surfaces are often loaded with antibiotics or other antimicrobials and respond to the creation of a microenvironment characteristic of biofilms by releasing their cargo. One such characteristic biofilm microenvironment is low pH, whereas in healthy tissue the pH should lie close to 7.4 at the biofilm substrate interface the pH can decrease to 5 and below. These heterogeneous pH environments have been studied previously using electrodes and optical devices. However, here a novel ratiometric sensor which relies on the same processes for sensing as for antimicrobial release is presented in order to optically probe the interfacial biofilm surface pH. Calcium phosphate nanoparticles doped with europium were synthesised and directly deposited onto silicon wafer chips by flame spray pyrolysis, the deposited film was then further stabilised by in situ annealing. These phosphorescent nanoparticles exhibit pH dependent dissolution, with an associated decrease in luminescence intensity which provides a sensor response and known pH buffers were used to draw a calibration curve. A panel of clinically relevant biofilm forming bacteria of both the gram-negatives and gram-positives were selected to measure their interfacial pH. Both gram-negatives and gram-positives demonstrated low pH environments, however, differences between bacterial strains and species were observed. These pH responsive silicon chips can therefore be used to guide the development of anti-biofilm surfaces to target particular bacterial infections.

BM04.09.31

Engineering Vascularized Cardiac Tissue Using a Combination of Electrospinning and 3D-Bioprinting Technologies Ebrahim Mostafavi and Thomas Webster; Northeastern University, Boston, Massachusetts, United States.

Cardiovascular disease is one of the most leading causes of mortality in the USA. Fabrication of 3D large-scale cardiac tissue constructs with functional vascularization has been a great challenge in engineering tissues suitable for repairing injured heart tissue. To address this challenge, here a new combinatory approach of electrospinning and 3D bioprinting techniques is employed to make a functional cardiac tissue. First, we engineered highly elastic blood vessel made of gelatin methacryloyl (GelMA) and poly(e-caprolactone) (PCL) by using electrospinning technique. The engineered tube was then endothelialized *in vitro* by using human umbilical vein endothelial cells (HUVECs) and perfused by culture media to form a functional blood vessel. In the next step, and a cell-laden GelMA-based hydrogel was 3D printed around the engineered GelMA/PCL vessel to form a vascularized cardiac tissue. Also, the effect of incorporation of Iron Oxide (Fe_2O_3) magnetic nanoparticles (MNPs) into both electrospun fibers as well as 3D printing GelMA bioink will be investigated to find out its influence on the cell growth, proliferation as well as the cardiac tissue regeneration.

Our results revealed that the mechanical properties of the engineered GelMA/PCL vessels are comparable with small diameter porcine blood vessels. Moreover, the fabricated vessels by electrospinning are suturable and depicted high suture retention strength. The 3D printed cell-laden GelMA hydrogels exhibited high cell viability over 5 days (>90%) of culture as captured from fluorescent microscopy images using a commercial Live/Dead assay. The metabolic activity of 3D printed cell-laden GelMA hydrogel also increased consistently during 5 days of culture as measured by using PrestoBlue assay. In addition, the number of cells increased around 3-fold from day 1 to day 5, confirming cell proliferation within 3D printed hydrogels.

BM04.09.32

Silver Nanowire/Chitosan Nanocomposite Scaffolds for Tissue Engineering with Enhanced Durability Dilara Aydin[1], Eda Ayse Aksoy[2], Sevda Senel[1], Husnu E. Unalan[3] and Doga Doganay[3]; [1]Pharmaceutical Technology, Hacettepe, Ankara, Turkey; [2]Hacettepe University, Ankara, Turkey; [3]METU, Ankara, Turkey.

We report on a successful scaffold based tissue engineering approach that aims to facilitate maximal cell seeding efficiency and subsequent cell proliferation using durable scaffolds with interconnected porosity with high surface area per volume and enhanced mechanical strength, durability. Among the biopolymers chitosan, which is a biodegradable, biocompatible cationic polysaccharide exerting bioactive properties (eg.antimicrobial, anti-inflammatory, hemostatic immunostimulatory etc) provides several advantageous as tissue engineering scaffold. Previously we have shown antimicrobial activity of nanocomposite films of chitosan and silver nanowires (Doganay, 2017). In this study, silver nanowire/chitosan nanocomposite scaffolds were prepared via freeze-drying method and their durability were enhanced with αβ-glycerophosphate. Morphology and chemical nature of the nanocomposites were characterized by X-ray Diffraction Spectroscopy (XRD), Scanning Electron Microscopy (SEM) and Fourier Transform Infrared Spectroscopy (FTIR). Mechanical strength, compression modulus, bioadhesion, porosity and water holding capacity of nanocomposite scaffolds were investigated. The scaffolds were observed to retain 35 to 65-fold water while maintaining their form and integrity. The nanocomposite scaffolds showed equilibrium swelling index compared to chitosan scaffold. Further mechanical analysis demonstrated that the incorporation of αβ-glycerophosphate enhanced the elastic modulus and tensile strength values of prepared scaffolds. These and other results obtained in this work revealed that the nanocomposite scaffolds can be mechanically and biofunctionally improved through tailoring the chitosan/glycerophosphate ratio and the amount of incorporated silver nanowires.

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Synthesis and Characterization of Biocompatibility Enhanced Poly(Caprolactone-Co-Glycidylmethacrylate) Shape Memory Polymer Ki Bum Kim, A Ram Heo, Ji Hwan Park and Woo Soon Jang; Future Bio Works Co., Seoul, Korea (the Republic of).

Shape-memory polymers (SMPs) are polymeric smart materials that have ability to return from a deformed state to their original shape by an external stimulus. Polycaprolactone (PCL) has properties of biocompatibility and biodegradable, therefore having a wide use in medical field such as drug delivery materials and biological tissue engineering. To make a shape – memory property in PCL, we proceed synthesizing poly(caprolactone-co-glycidylmethacrylate) (PCL-co-PGMA) which is network structure polymer by copolymerizing glycidylmethacrylate (GMA) with caprolactone (CL). PCL-co-PGMA was characterized by GPC, 1H-NMR. In addition, we controlled ratio of GMA in copolymer to set a melting temperature which is showing shape-memory property and DSC was performed.
The crossinked poly(caprolactone-co-glycidylmethacrylate) network were prepared by photocrosslinking with ultraviolet (UV) irradiation and using Irgacure 2959 by as the photoinitiating agent. The resultant crosslinked polymer was then characterized by DSC and dynamic mechanical analysis (DMA). Nitric oxide (NO) has many physiological functions such as vasodilation, neurotransmission, and angiogenesis in the human body. Especially, the nonthrombogenic properties of vascular surfaces are primarily attributed to NO generated from endothelial cells that line the inner walls of all blood vessels and Nitric oxide derived from endothelial cells regulates blood flow and pressure and inhibits platelet activation and aggregation under normal conditions.
To enhance biocompatibility of shape memory polymer, Nitric oxide was introduced on the surface of shape memory polymer. After that, that was evaluated by cell toxicity test.

BM04.09.34

Preparation and Characterization of Poly(Glycerol-Sebacate-Stearic Acid) for Shape Memory Polymer Bo Keun Lee, Seol Jang and Woo Soon Jang; Future Bio Works Co., Seoul, Korea (the Republic of).

For past few decades, a new type of smart material, Shape Memory Polymer (SMP), has developed rapidly. Shape memory polymers can respond to changes in external conditions such as temperature, pH, and ionic strength. However, such as non-absorbable biomedical polymers can undergo undesirable reactions due to the physical or chemical properties of the surrounding biological components that can persist in vivo for a long period of time to cause toxic reactions in vivo. Therefore, these polymers may need to be removed from the body as a second surgery.
Poly (glycerol-sebacate) (PGS) is a biocompatible and biodegradable elastomer that is used in a wide range of biomedical applications to manufacture microfluidic devices, vascular scaffolds, and other micro-tissue engineering systems. Polymers containing glycerol and sebacic acid have already been approved for medical use. While the porous form is preferred for the support to permit intracellular growth, PGS degradation has been studied primarily in non-porous form. Some papers describe their biocompatibility and biodegradation, but no studies have been presented on the shape memory effect of these polymers. The purpose of this study was to investigate shape memory properties, porosity and biocompatibility.
Another way to strengthen this polymer is to impart new functionality such as poly (glycerol sebacate) -stearate (PGSS). PGSS based block copolymer with glycerol, sebacic acid and stearic acid by bulk polymerization. PGSS was successfully synthesized and characterized by ^{1}H-NMR, GPC and DSC. The DSC was used to study the reversible movement required for temporary shape storage of shape memory materials. PGSS performed in vitro biocompatibility studies.
Characterization of PGSS is important when evaluating polymerization and future use of this biomaterial. The results confirm that PGSS is a shape memory material with a recovery rate of 99.5% or more. PGSS has also been found to be porous and biocompatible. Thus demonstrating that this material can be designed as a potential biomaterial in other applications where it interacts with pharmaceutical and biological systems.

BM04.09.35

Magnetic Hyperthermia Induced Liposomal Doxorubicin Release for Thermo-Chemotherapy Shan Zhao, Samuel Klein, James Petryk, Catalina Spatarelu, Fridon Shubitidze, Zi Chen and Jack Hoopes; Dartmouth College, Hanover, New Hampshire, United States.

The development of a targeted drug delivery system is an emerging approach to increase the targetability of anticancer agents. One of the most promising technologies to achieve high specificity is using a liposomal delivery system. In this work we developed a magnetic liposomal delivery system that combines magnetic nanoparticles (mNPs), the doxorubicin (Dox) and the application of an external alternating magnetic field (AMF). The mNPs are encapsulated in the hydrophilic core of liposomes and the Dox is embedded in the hydrophobic bilayer. Once the liposome is delivered into the tumor, then the AMF is activated. The AMF transfers the electromagnetic energy to the mNPs, which in return generates heat and increases the temperature locally in the surrounding bilayer. When the temperature reaches the liposome melting temperature T_m, the Dox is released. To further increase the liposome's targetability, in addition to the AMF-mNPs triggered Dox release approach, we have used a non-invasive, non-toxic external static magnetic field (SMF) to target the liposomes to the tumors. This combined AMF-SMF feature could reduce the non-specific side-effects and toxicity of encapsulated drugs, while ensuring a targeted, safe, high percentage Dox release and accumulation at the tumor site.
An in vitro release experiment at 37ºC showed that more than 80% of the encapsulated Dox was retained in liposomes after 6 hrs in PBS. Under the exposure of AMF (641 Oe, 170 kHz) for 10 mins, liposomes of 10 ug/ml displayed 80% of Dox release, indicating the high efficiency of this triggering technique. Mice inoculated with B16 murine melanoma cell, intradermal, rear limb, were assessed for Dox release and treatment efficacy when the tumors reached 120 mm^3. Our preliminary in vivo study indicated that the external magnetic field demonstrated a notable increase of mNP when the STM was applied at the tumor site 1 hour post-injection. The levels and distribution of Fe in tissue are examined via histopathology, TEM and ICP-MS. The concentrations of Dox in these biological samples (supernatants of tissue homogenates and plasma) are determined using the fluorescence spectrophotometer. This multimodal technology will allow for a safe, targeted, increase in Dox accumulation at the tumor site and ultimately an improvement in the therapeutic ratio and efficacy of Dox with radiation or other possible combinations of therapies.

BM04.09.36

Supercritical Carbon Dioxide Functionalization of Biomaterials with Antimicrobial Molecules Guillaume Nonglaton[1, 4], Clémentine Darpentigny[2, 1, 4], Bastien Michel[1, 4], Pierre R. Marcoux[1, 4], Julien Bras[3, 4] and Bruno Jean[2, 4]; [1]CEA-Leti, Grenoble, France; [2]CERMAV-CNRS, Grenoble, France; [3]Grenoble INP-CNRS, Grenoble, France; [4]Université Grenoble Alpes, Grenoble, France.

In a context where the need for innovative medical devices is increasing and the environmental issue is becoming a priority, the objective of this study was to develop "active" wound dressings with antimicrobial properties using a bio-inspired strategy and an eco-friendly solvent: supercritical carbon dioxide (SC-CO2).
First, two different types of materials were studied: 1) nanocellulose-based aerogels or membranes and 2) polypropylene (PP), polyethylene terephthalate (PET) or polyamide (PA) plastic sheets as standard polymers used in biomedical applications. We choose to use nanocellulose derived from the biomass as biocompatible building blocks. Nanocellulose particles are a class of very promising bio-based material. In the biomedical field, their relatively low cost, low toxicity and biocompatibility have made them very attractive. They were assembled to prepare aerogels or surfaces with structural parameters controlled by the preparation process and the nanocellulose properties.
Then, the different materials were functionalized in SC-CO2 in an attempt to respect green chemistry principles. SC-CO2, considered as a non-toxic, eco-friendly solvent, could be used to impart antimicrobial properties to the nanocellulose structures without affecting the fragile porous structures nor the thermos-sensitive compounds. Two strategies of functionalization of the materials were tested: impregnation with synthetic antibiotics or extract molecules from essential oil and chemical derivatization of the materials. In order to enhance their bioavailability, their solubility in SC-CO2 was studied upon exposure to various supercritical conditions. The bioactive molecules were then incorporated inside the nanocellulose matrices via an impregnation process or covalent grafting.
The detailed characterization of the structure materials and their chemical functionalization have been carried out using advanced technologies. The antimicrobial activity of the nanocellulose-based materials was assessed and correlated to the structure and chemistry. For impregnated molecules, the drug release kinetics and the zone of inhibition of growth were analyzed. For structures where antimicrobial agents were grafted, the contact killing activity was assessed and log-D reduction was measured against two bacteria and a eukaryote.
Nanocellulose membranes and aerogels that exhibit lightweight and high specific surface areas were compared to determine the structure-functionalization-activity relationship. Impregnation
The mode of action and the lifespan of the newly designed antimicrobial materials were studied according to the incorporation process of bioactive molecules in supercritical carbon dioxide. Both impregnation and grafting can be used to impart antimicrobial properties and extend the shell-life of the bioactive wound dressing. These bioactive materials will be used for the design of external wound dressings and implantable medical devices.

BM04.09.37

Eradication of HT-29 Colorectal Adenocarcinoma Cells by Controlled Photorelease of CO from a CO-Releasing Polymer (photoCORP-1) Triggered by Visible Light Through an Optical Fiber-Based Device Miguel N. Pinto, Indranil Chakraborty, Cosme Sandoval and Pradip K. Mascharak; University of California, Santa Cruz, Santa Cruz, California, United States.

Carbon monoxide (CO), known for its toxicity, is a gaseous signaling molecule (gasotransmitter) endogenously produced through heme catabolism. CO is known to interact selectively with the soft metal centers of heme proteins and indirectly modulate other non-heme-containing targets through signaling pathways. This gasotransmitter participates in a variety of roles in mammalian pathophysiologyand has been implicated in oxidative stress, cell proliferation, and apoptosis. In addition, CO has been shown to sensitize cancer cells to known chemotherapeutics, as well as induce dose-dependent eradication of malignant cells. The main difficulty with using CO as a therapeutic is its site-specific delivery in a controlled and sustained manner. Direct inhalation of CO requires continuous exposure to relatively high levels of this gas, lacking specificity and promoting negative effects. An alternate approach to deliver CO to tissue is to use CO-releasing molecules (CORMs). Photoactivated CORMs (photoCORMs), with a few exceptions, are metal carbonyl complexes that release CO upon illumination, allowing for the delivery of this gas with spatial and temporal control. Efforts have been made to impart desirable characteristics to these CO donors, such as visible light sensitivity, solubility, stability, and low toxicity. A wide variety of carriers have been used to deliver CO to biological targets. However, desirable delivery materials should retain the photoCORM or the CO-spent product, thus avoiding possible adverse side-effects. In this account, we describe a novel visible light active polyHEMA-based CO-releasing polymer (photoCORP-**1**). This hydrogel contains a manganese photoCORM ($[Mn(CO)_3(qbt)(4\text{-vpy})]CF_3SO_3$(**1**)) co-polymerized to a hydroxyethyl methacrylate/ethylene glycol dimethacrylate (HEMA/EGDMA) backbone via use of a 4-vynilpyridine ancillary ligand. This covalent attachment ensures that the photoCORM as well as the CO-spent product remain inside of the bulk polymeric material. This robust yet flexible CO-releasing hydrogel can be molded, cast, or cut into any shape or form. Its high transparency and gas permeability allow for the rapid and sustained release of CO upon illumination with low power visible light. The release of CO can be visually tracked by the loss of the color arising from **1**. A fiber optic-based catheter using a photoCORP-**1**tip (CO-catheter) was constructed for use in light inaccessible cavities. The reported CO-catheter was successfully used to promote apoptosis in human colorectal

adenocarcinoma (HT-29) cells by triggering CO photorelease under low power visible light. The easy application/removal of the CO-catheter from the malignant site circumvents toxicity arising from the photoCORM/photo-products while allowing for the delivery of controlled and sustained does of CO at the target site. The use of this CO-releasing material coupled to a current endoscope can be used deliver required doses of CO to a colonic target.

BM04.09.38

pH-Triggered Tumor Targeting Polymeric Micelles Loading with Dimeric Drug for FRET-Traceable Drug Release Xing Guo[1, 2], Lin Wang[1], Kayla Duval[1], Jing Fan[3], Shaobing Zhou[2] and Zi Chen[1]; [1]Dartmouth College, Hanover, New Hampshire, United States; [2]Southwest Jiaotong University, Chengdu, China; [3]City College of New York, New York, New York, United States.

Trans-activating transcriptional activator (TAT), a cell-penetrating peptide, is extensively used for facilitating cellular uptake and nuclear targeting of drug delivery systems. However, the positively charged TAT peptide strongly interacts with serum components and undergoes substantial phagocytosis by the reticuloendothelial system, causing a short blood circulation in vivo. In this work, an acid-active tumor targeting nanoplatform DA-TAT-PECL is developed to inhibit the nonspecific interactions of TAT in the bloodstream. 2,3-dimethylmaleic anhydride (DA) is used to convert the TAT's amines to carboxylic acid; the resulting DA-TAT is conjugated to poly(ethylene glycol)-poly(ε-caprolactone) (PEG-PCL, PECL) to get DA-TAT-PECL. After self-assembly into polymeric micelles, they are capable of circulating in the physiological condition for a long time and promoting cell penetration upon accumulation at the tumor site and deshielding the DA group. Moreover, camptothecin (CPT) is used as the anticancer drug and modified into a dimer $(CPT)_2$-ss-Mal, in which two CPT molecules are connected by a reduction-labile maleimide thioether bond. The Förster resonance energy transfer signal between CPT and maleimide thioether bond is monitored to visualize the drug release process, and effective targeted delivery of antitumor drugs is demonstrated. This pH/reduction dual-responsive micelle system provides a new platform for high fidelity cancer therapy.

BM04.09.39

Comparative Study of Ampicillin-Loaded Melanin-Polycaprolactone Nanofibers Prepared by Single-Needle and Co-Axial Electrospinning Methods Gözde Kabay[1, 2], Gizem Ak[3], Gizem Kaleli Can[1] and Mehmet Mutlu[3]; [1]Biomedical Engineering Division, TOBB University Of Economics and Technology , Ankara, Turkey; [2]Biological Systems Engineering Department, University of Wisconsin–Madison, MADISON, Wisconsin, United States; [3]Biomedical Engineering Department, TOBB University of Economics and Technology, Ankara, Turkey.

Melanin, natural consistent of human body, has attracted great attention in the last few years by being amorphous biopolymer with semiconducting, biodegradable, biocompatible and non-toxic, antioxidant and abundant biopolymer derived from natural sources [1–3]. Despite its great potential and ease on extraction, it has been rarely studied in pharmaceutical and biomedical fields. So that, in this study, it is decided to use natural melanin nanoparticles by incorporating into PCL solution to achieve controlled release of the ampicillin from electrospun membranes produced by coaxial and single-needle electrospinning.

In the experimental setup, 10% (w:v) PCL was mixed with appropriate amounts of extracted melanin (0,001-0,1 g/ml) and ampicillin (0,01 g/ml) to prepare single-needle electrospun membranes (S). For core-shell structures fiber production, 0,01 g Amp was mixed with 10% (w:v) PCL solution and used as a core solution, whereas 4% (w:v) PCL solution combined with melanin (0,001-0,1 g/ml) for a shell coverage, in turn. After spinnable solutions reached the nozzle tip, they were exposed to high voltage of 12 kV, while tip-to collector distance was kept at 9 cm and nanofibers were produced. Drug release amounts of PCL-amp core (C), PCL-Mel-Amp single-needle and coaxially electrospun membranes (S and CS) were measured by Ultraviolet-Visible Spectroscopy at the wavelength of 203 nm and corresponded drug release profiles were analyzed.

For C membrane, 82% of the ampicillin was released within the first hour indicating burst release. The burst release can be due to the lower compatibility of ampicillin inside PCL fibers or the accumulation of the drug on fibers' surface during spinning. Besides, in the presence of melanin, slower initial burst release compared to C was observed for the S membrane, which showed a burst release of 37%. In the same period, the release rate of the drug was only 7% for the CS membranes. It states that, more controlled drug release was achieved for CS nanofibrous membrane compared to the S membrane even if the melanin addition was carried out for both processes. This result would be attributed to the shell layer coverage that introduced an additional diffusional barrier.

All in all, we first-time evaluated melanin-polycaprolactone composite as a drug nanocarrier, both in coaxial and monolithic forms. The findings of this work demonstrated that, the drug release from Mel-PCL matrix could be expanded to other drugs, which have low compatibility within a carrier matrix.

References

[1] P. A. Riley: Melanin. *Int. J. Biochem. Cell Biol.* **29**(11), 1235 (1997).
[2] M. Araújo, R. Viveiros, T. R. Correia, I. J. Correia, V. D. B. Bonifácio, T. Casimiro, and A. Aguiar-Ricardo: Natural melanin: A potential pH-responsive drug release device. *Int. J. Pharm.* **469**(1), 140 (2014).
[3] B. Larsson and H. Tjälve: Studies on the mechanism of drug-binding to melanin. *Biochem. Pharmacol.* **28**(7), 1181 (1979).

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Phase-Separation Induced High-Order All-Aqueous Emulsions in Microfluidics Youchuang Chao[1, 2], Sze Yi Mak[1, 2] and Ho Cheung Shum[1, 2]; [1]HKU-Shenzhen Institute of Research and Innovation (HKU-SIRI), Shenzhen, Guangdong 518000, China; [2]Department of Mechanical Engineering, The University of Hong Kong, Pokfulam Road, Hong Kong, China.

Multiphase emulsion drops have wide applications such as in drug delivery and food industry because of their ability to encapsulate active ingredients and to release them in a controllable time. Recently, high-order multiphase emulsion drops have been successfully obtained by mass-transfer induced phase separation in microfluidic devices [1,2]. However, the ternary systems used in these studies always involve organic-solvents, such as oil and polar solvent, and few of them focus on the formation of high-order all-aqueous emulsions which are oil-free and highly biocompatible. Therefore, we propose a robust approach to form high-order multiple all-aqueous emulsions in a glass-based microfluidic device [3]. The proposed method is essentially based on the phase separation induced by the osmolality difference between the drop and the continuous phases. By varying the initial concentration of the drop phase, we also successfully achieve all-aqueous emulsions with different complexity and further summarize the complexity of the droplets into a phase diagram. Our method is simple, and the fabricated high-order all-aqueous emulsions drops could be templated as biocompatible capsules which are capable of

encapsulating and releasing active components upon trigger.
References:
[1] Haase, M. F., & Brujic, J. (2014). *Angew. Chem., Int. Ed.*, 53(44), 11793-11797.
[2] Moerman, P. G., Hohenberg, P. C., Vanden-Eijnden, E., & Brujic, J. (2018). *Proc. Natl. Acad. Sci. USA*, 115(14), 3599-3604.
[3] Chao, Y., Mak, S. Y., Rahman, S., Zhu, S., & Shum, H. C. (2018). *Small*, 1802107.

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Utilization of P4VP and Graphene to Differentiate Dental Pulp Stem Cells into Functional Neurons John Chen[1], Zaiff Khan[1], Linxi Zhang[2], Kuan-Che Feng[2], Rebecca Isseroff[2, 1], Marcia Simon[3] and Miriam Rafailovich[2]; [1]Lawrence High School, Cedarhurst, New York, United States; [2]Dept. of Materials Science and Chemical Engineering, Stony Brook University, The State University of New York, Stony Brook, New York, United States; [3]Stony Brook University School of Dental Medicine, Stony Brook, New York, United States.

Currently, the central nervous system is unable to heal effectively by itself, leading to a search for ways to regenerate or grow neurons for replacement. Dental Pulp Stem Cells (DPSCs), easily acquired from extracted wisdom teeth, are multipotent stem cells that can transform into osteoblasts, cardiac, and neuron cells and can thus provide cells resulting in an autologous implant. P4VP, a polymer shown to retain cell morphology, can help cells adhere to a substrate, potentially eliminating the need for polyornithine. Graphene, known for its electrical conductivity, may prove useful in the differentiation of DPSCs into neurons, since neurons communicate through the electrical impulses of the synapses. This research compares the effects of thin film substrates composed of different combinations of P4VP, graphene, and polyornithine; as well as the effectiveness of these substrate combinations electrospun into fibers, on DPSCs and their potential differentiation into neural cells.

Four experimental substrates were created: P4VP and P4VP + graphene thin films, as well as P4VP and P4VP + graphene electrospun into fibers. Two sets of each substrate were created; one set was coated with polyornithine and the other set was not coated. A positive control of tissue culture plastic was also plated with DPSCs.

Optical microscopy displayed that DPSCs grown on flat substrates of P4VP and P4VP+graphene grew into longer cells than those grown in the positive control. In addition, they had formed branches of axiom-like structures from day 14, even without the use of polyornithine, suggesting that P4VP thin films allow cells to adhere on their own to the substrate. However, electrospun fibers did not show differences in cell growth from the positive control. Confocal Microscopy conducted after 21 days of culture confirmed that the cells directly attached to the P4VP and P4VP+graphene substrate had elongated, while the cells not attached were more round, but both types showed high confluency. Also, cells plated on electrospun fiber samples had the same shape and confluency as flat film samples, suggesting that fibers had similar effects as thin films on DPSC growth into nerve-shaped cells. No visible changes were seen with the addition of graphene. Further results were acquired with Scanning Electron Microscopy (SEM) and Real-Time Polymerase Chain Reaction (RT-PCR), determining whether the DPSCs were starting to undergo differentiation into neurons and thus showing promise that P4VP can provide a suitable scaffold for neuron cell development.

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Micropatterning of TiO_2 Nanotubes for High-Throughput Study Qiaoling Huang[1, 2], Yanran Li[1], Yuanjun Dong[1], Ping Mu[1], Xiang Yang Liu[3, 1] and Changjian Lin[1]; [1]Xiamen University, Xiamen, China; [2]John A. Paulson School Of Engineering And Applied Sciences, Harvard University, Cambridge, Massachusetts, United States; [3]National University of Singapore, Singapore, Singapore.

TiO_2 nanotubes (TNTs) have attracted extensive attention by virtue of the similarity of their highly ordered nanotubular structure to that of cortical bone. Recent studies have indicated that titanium nanotubes are superior to pure titanium in terms of bone regeneration, blood compatibility and corrosion resistance. However, the optimal properties of TNTs for certain applications remain mystery. For example, the nanotube dimension ranges from several nanometers to hundreds of nanometers. The existing literature reports biological responses to confined dimensions owing to the technique difficulties in evaluating hundreds of TNTs with different properties at the same time. In this study, TNT gradients with different properties were fabricated and applied for high-throughput study. Results showed that TNT gradients provide facile platforms for high-throughput screening of biological responses, including cell responses, protein adsorption, bacterial adhesion, and et al.

References

[1] P. Mu, Y. Li, Y. Zhang, Y. Yang, R. Hu, X. Zhao, A. Huang, R. Zhang, X.Y. Liu, Q. Huang, C. Lin, High-throughput Screening of Rat Mesenchymal Stem Cell Behavior on Gradient TiO2 Nanotubes, *ACS Biomaterials Science & Engineering*, **4** (2018) 2804-2814
[2] .R. Song, Y. Zhang, Q. Huang, Y. Yang, L. Lin, J. Liang, R. Hu, G. Rui, C. Lin, Facile Construction of Structural Gradient of TiO2 Nanotube Arrays on Medical Titanium for High Throughput Evaluation of Biocompatibility and Antibacterial Property, *ACS Applied Bio Materials*, (2018). Just Accepted.

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Mechanical Properties of Drawn Electrospun Polycaprolactone (PCL) Nanofibers Katie Sun[1, 2] and Vince Beachley[2]; [1]Rutgers, The State University of New Jersey, New Brunswick, New Jersey, United States; [2]Rowan University , Glassboro, New Jersey, United States.

Introduction: Polycaprolactone (PCL) is a bioresorbable polymer that has favorable structural, mechanical, thermal, and chemical properties which provide the necessary strength, rheology, porosity, biocompatibility, and flexibility to fulfill the role as a biological scaffold in musculoskeletal tissue engineering and repair. Significantly, PCL nanofibers have historically proved to be able to sustain large amounts of elongation before break. The method of electrospinning provides highly linear and aligned scaffolds like that of natural tissues, produces a favorable surface area to volume ratio, and protects the scaffold's ability to provide a mechanical strength comparable to that of natural tissue. Thus, post-draw of PCL nanofibers after electrospinning produces a product with mechanical properties that are correlated with strain rate.

Methods: An electrospinning device designed by our lab utilizes an adjustable frame and aluminum tape tracks controlled by a motor system to both electrospin PCL nanofibers while also post-drawing them to a desired draw ratio and improving alignment. The process of electrospinning 18wt% polycaprolactone (PCL) is protected and humidity-controlled in a polycarbonate enclosure. The electrospinning draw ratio was set at 2 for the PCL fibers.

A scanning electron microscope (SEM) was utilized to analyze the fibers' diameters, densities, and cross-links. The mechanical properties were tested under strain rates of 0.5mm/min, 5mm/min, and 50mm/min for nanofibers laid on a 10mm by 10mm testing square.

Results: The electrospun PCL nanofibers exhibited many trends after mechanical testing. The young's modulus decreased with increased fiber number and with increased strain rate. The ultimate tensile strength increased with increased fiber number and increased more gradually with increased strain rates. The toughness shows increases correlated to fiber number and a strongly positive and steep correlation with a 0.5mm/min strain rate. The 5mm/min and 50mm/min testing rates and toughness show constant toughness regardless of fiber number. The ultimate tensile strain of the fibers varies little and thus is independent of testing strain rates.

Conclusion: Electrospinning while post-drawing proved to create PCL nanofibers that exhibit an inverse negative correlation to young's modulus but direct positive correlation to ultimate tensile strength, toughness, and ultimate tensile strain. The increase in testing strain rate decreased the average-per-strain-rate young's modulus, ultimate tensile strength, toughness, and ultimate tensile strain. This is due to the higher testing strain rates causing rapid plastic deformation. This prevents the PCL polymer chain alignments from accommodating new charge repulsions due to the elongation and movement of PCL fibers during mechanical testing, leading to failure. The data collected indicates that if further mechanical testing strain rates are tested, the same correlations will be exhibited.

BM04.09.45

Influence of Exposure to TiO_2 Nanoparticles on *Staphylococcus aureus* Infection Fan Yang[1], Justin Zhou[2], Vincent Zhang[3], Jonathan Goldschlag[4], Ethan Winkler[4] and Miriam Rafailovich[1]; [1]Stony Brook University, Stony Brook, New York, United States; [2]Patchogue-Medford High School, Medford, New York, United States; [3]Sachem High School East, Farmingville, New York, United States; [4]Hebrew Academy of the Five Towns and Rockaway, Cedarhurst, New York, United States.

Titanium dioxide (TiO_2), commonly used in paints, toothpaste, sunscreen, cosmetics, pharmaceutics, and food additives, has been extensively studied for its anti-cancer and anti-bacterial applications when irradiated with UV light, but there is minimal data on its relative safety for normal human cells. In a previous study, HeLa cell exposed to TiO_2 nanoparticles showed an increased susceptibility to bacterial infection partially attributed to inhibition of enzymatic activity involved in membrane cholesterol distribution.[1] Since HeLa cells are an immortalized cell line derived from tumor tissue, they may not be representative of cells found in healthy human tissue. To investigate the influence on healthy tissue, we chose to focus on skin tissue, since skin is the first barrier to contact with various toxins. Dermal fibroblasts and keratinocytes were plated on tissue culture plastic for 24 hours and then exposed for another 24 hours to 0.1mg/ml of TiO_2. The results indicate that an increased susceptibility to bacterial infection is also present in healthy primary tissue cells.
Since cells are also influence by their environment, we also investigated the role of the substrate on the toxicity to TiO_2 particles, as well as bacterial infection. Preliminary results indicate increased toxicity of the particles, when the cells are cultured on gelatin, and in particular collagen, which is commonly present in the skin tissue.

[1] Xu, Yan, et al. "Exposure to TiO_2 nanoparticles increases *Staphylococcus aureus* infection of HeLa cells." *Journal of nanobiotechnology* 14.1 (2016): 34.

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Preparation and Performance of Polycaprolactone Nanocarrier for the Controlled Release of Interferon-α for the Treatment of Malign Melanoma Enes Celik, Gözde Kabay and Mehmet Mutlu; Biomedical Engineering, Ankara, Turkey.

Melanoma is a type of skin cancer, results in higher mortality rates mostly in metastatic states which constitutes 80% of skin cancer deaths (Miller, A.J., et all. 2006). Because of being such a wild type of cancer, various therapeutic methods such as surgery, radiation therapy, chemotherapy, immunotherapy and targeted therapy are used to decrease the mortality rates. However, none of these procedures are enabled completely to remove the problems such as toxicity and efficacy. To overcome these problems, controlled drug release technology remains as a plausible approach.

Nanofibrous scaffolds designed for drug delivery purpose has various advantages compared to conventional drugs such as high loading and entrapment capacity, protection of therapeutic agent, controlling the release of the drug in a timely manner and accordingly preservation from any toxic effect. Several controlled drug release profiles such as sustained, burst, and delayed can be obtained by using electrospun nanofibrous membranes as carriers (Hu, X., et all, 2014) . The ability to adhere the scaffold at the site of infection is another advantage of these systems compared to other drug delivery methods, which would be beneficial for the skin cancer types (Akduman, C., et all, 2016).

Based on the clinical problem defined above and the previous reports, we hypothesized that the good biocompatibility and biodegradability of polycaprolactone (PCL) makes it promising nanocarrier for drug release applications. Therefore, we developed a drug release platform combination of PCL and interferon-alpha-2B (INFα-2B) which is a therapeutic agent approved by the US Food and Drug Administration (FDA) and recently being used for the treatment of metastatic melanoma.

In the experimental setup, electrospun membranes combined of PCL and INFα-2B were produced by single and coaxial electrospinning setups. 10% (w/v) PCL was combined with INFα-2B by mixing with ratios of 5%,10% and 20% (w/w). System parameters such as applied voltage and tip-to-collector distance were adjusted to 12 kV and 9 cm, respectively. Flow rates were varied between 0,5 and 1,0 ml/h for core and shell solution, in turn. The physical characterization of the electrospun membranes were achieved SEM and TEM analysis and chemical characterizations were carried out by FTIR spectroscopy. Moreover, in vitro drug release profile of INF α-2B from the electrospun membranes was sketched by calculating the concentration of INF α-2B released into the phosphate buffered saline solution (PBS, pH: 7.4) with UV–vis spectrophotometer (Hitachi U-5100, Japan) at 214 nm (Sultanova, Z., et all, 2016).

We showed that, coaxially electrospun INFα-2B loaded PCL nanofibrous membranes can be used to treat melanoma disease by providing controlled release of INFα-2B. The accurate adjustment of the drug release behavior of INFα-2B-loaded PCL nanofibers against tumor cells is currently under investigation.

BM04.09.47

Characterization of Thermoreversible Hydrogels from Multiblock Poloxamers and Hybrid Hydrogels for an Application as Cell Barrier Layer Juyi Li[1], Erica Inyoung Choi[3], Christina Tong[2] and Miriam Rafailovich[1]; [1]Stony Brook University, Stony Brook, New York, United States; [2]Fairview High School, Boulder, Colorado, United States; [3]St. Paul's School, Concord, New Hampshire, United States.

Periodontitis is a highly prominent issue in dental health today and the current solutions such as Guided Bone Regeneration (GBR), or the use of a barrier membrane to separate the alveolar bone and gums, have many shortcomings. These barrier layers can be composed of hydrogels, 3D cross-linked polymeric networks that are used for regenerative medicine; hydrogels are showing very promising applications in the biomedical field due to their biocompatibility and unique properties. Our previous work showed a promising hybrid gel synthesized with gelatin and poloxamer F127. Polymerized multiblock poloxamers maintain the thermo-reversibility but with an improved mechanical property. This study focused on characterization of the multiblock poloxamers PF127, PF108, and PF98 and synthesis/characterization of hybrid gels using these poloxamers to evaluate their potency. Rheology was used to characterize the poloxamer solutions and hybrid hydrogels, revealing that PF108 had a significantly higher elastic modulus compared to the other gels. The laser microscope imaging showed a unique, branching fiber structure of the PF108 hybrid gel, while the structures of the other hybrid gels displayed gelatin mesh networks. The PF108 hybrid hydrogel also showed significantly lower surface roughness. This is associated with decreased cell attachment, thus, gelatin-PF108 hybrid gel may possess promising characteristics to serve as a better cell barrier layer.

[1]Jiang, Jun, et al. "Rheology of thermoreversible hydrogels from multiblock associating copolymers." *Macromolecules*41.10 (2008): 3646-3652.

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Novel Collagen and Elastin Interlaced Composites for Heart Valve Tissue Engineering (HVTE) Sonia Iftekhar[1], Colleen Lopez[2], Jan Czernuszka[1] and Carolyn Carr[2]; [1]Department of Materials, University of Oxford, Oxford, United Kingdom; [2]Department of Physiology, Anatomy and Genetics, University of Oxford, Oxford, United Kingdom.

Valvular Heart Disease (VHD) is an incurable disease that affects 2.5% of the population in the USA, this includes both congenital and acquired forms of VHD. Valve replacement is the most likely treatment when the valve is severely damaged resulting in stenosis and/or regurgitation. Prostheses used to replace damaged valves are predominantly of two types, biological or mechanical. Both types are temporary non-regenerative solutions that have drawbacks, these include a limited life-time and the prerequisite to take anticoagulants.

A tissue engineered substitute has the potential to overcome such limitations and could form a fully functional and viable heart valve, this is particularly advantageous for infants suffering from congenital VHD.

Heart valves are active, self-repairing tissues that can withstand tremendous magnitudes of pressure due to their unique architecture. They have an interconnected tri-layer structure made up of three distinct zones. Type I collagen and elastin are the two major components of the extracellular matrix (ECM). Elastin is a non-collagenous protein that is present in the form of a three-dimensional network with interconnected collagen fibres, together they provide the ability to bear high loads as well as be flexible and resilient.

Designing a scaffold to mimic the structure and function of native heart valves requires bioactive materials that can easily be processed and fabricated into scaffolds. Naturally based biomaterials are the most promising candidates to be used as the basis for rapidly regenerative scaffolds.

A combined microstructural, micromechanical and stem cell based approach is proving to be the most successful in terms of reproducibility, rapid generation of new ECM and changes to stiffness. Our work follows a bioinspired approach, where we have designed novel scaffolds using ECM components to mimic the interconnected architecture of the heart valves closely. These unique compositions are the first step towards designing tri-layer structures to replicate the entire heart valve structure.