SYMPOSIUM W
Biological and Bio-Inspired Materials and Devices
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* Invited paper
The fabrication of novel biomaterials through molecular self-assembly: Shuguang Zhang, Center for Biomedical Engineering, Massachusetts Institute of Technology, Cambridge, Massachusetts.

Two complementary strategies can be employed in the fabrication of molecular biomaterials. In the "top-down" approach, biomaterials are generated by stripping down a complex entity into its component parts. This contrasts with the "bottom-up" approach, in which materials are assembled molecule by molecule and in some cases even atom by atom to produce novel supramolecular architectures. The latter approach is likely to become an integral part of nanomaterials manufacture and requires a deep understanding of individual molecular building blocks, their structures, assembling properties and dynamic behaviors. A key element in molecular fabrication is chemical complementarity and structural compatibility, both of which confer the weak and noncovalent interactions that bind building blocks together. Significant advances have been achieved at the interface of nanomaterials and biology, including the fabrication of nanofiber materials for three-dimensional cell cultures. The results of such work have demonstrated the potential to produce functional materials where both organization and specific biological function are required, e.g., sensors, adaptable materials, which lives in geothermal hot springs and grows at temperatures of up to 86 degrees Celsius and pH 2-9. Structural data and genetic engineering tools have allowed the creation of chaperonin mutants that bind biomolecules or inorganic nanoparticles. These chaperonins show that the chaperonins are interacting with the intramolecular folding events and secondary structure, in addition to forming of viral arrays. We find that when the chemical templates have dimensions comparable to the size of the virus, they tend to spontaneously form close-packed structures. Using these templates as crystals oriented normal to the surface can be produced in a glassy PS matrix in films with thicknesses several micrometers thick. Here, we show the chaperonin-driven self-assembly of the P(S-b-EO) diblock copolymer thin film cast from an aqueous solution of chaperonin and polymer. AFM images of the resulting thin films with and without chaperonin show domains oriented normal to the polymer. The combination of order from the self-assembly properties of diblock copolymers with the genetic adaptation of proteins opens up new possibilities of producing multifunctional materials and the functional components of devices where both organization and specific biological function are required, e.g., sensors, adaptable materials, medical implants, and biocompatible devices.

Successful solutions to many problems in science and technology have come by extracting design or strategy from biology, and applying it in a non-biological context. The use of biomimetic approaches is particularly well suited when designing self-assembling functional systems, because life from single cells to complex, multicellular organisms demonstrates an enormous number of successful functional designs, and because living systems assemble themselves. There are two reasons for studying self-assembly. First, self-assembly is centrally important for life. Biological systems are sustained as a result of self-organization. Understanding life therefore requires - among other things - a study of self-assembly. Second, self-assembly can generate ordered aggregate domains ranging in size from the molecular to the macroscopic. These structures often cannot be generated by any other procedure. In the past, self-assembly has been best known as a synthetic strategy in the molecular size regime. New examples of its application to nanoscale microcomponent components are now beginning to emerge. As a consequence, self-assembly is becoming increasingly important as a strategy for the formation of useful, nano- and micro-scale structures. This talk discusses the characteristics of self-assembly in living systems and reviews self-assembled functional systems designed according to biological principles.

The combination of polymers and proteins to form hierarchically structured multifunctional materials with the processability of polymers while retaining biological function of the protein is being studied. A unique chemistry resulting from the mixing of these two disparate self-assembling systems has been found. The materials utilized were an asymmetric diblock copolymer of polystyrene (PS) and polyethyleneoxide (PEO) denoted P(S-b-EO) and a double ring structure-forming protein from a class of heat shock proteins known as chaperonins. Solvent casting has been shown to be a viable and rapid route by which arans of nanoscopic PEO domains oriented normal to the surface can be produced in a glassy PS matrix in films with thicknesses several micrometers thick. The combination of order from the self-assembly properties of diblock copolymers with the genetic adaptation of proteins opens up new possibilities of producing multifunctional materials and the functional components of devices where both organization and specific biological function are required, e.g., sensors, adaptable materials, medical implants, and biocompatible devices.
In biological systems, dynamic nanometer scale structures self-assemble with sufficient precision that their structures are regular at the level of multibillionth crowded molecules. They do this in a highly controlled manner in a noisy environment full of other proteins. To cope with the demands of controlled and precise assembly they have evolved a number of sophisticated control mechanisms. The mechanisms include: well-controlled linear assembly pathways, the use of substructure assembly to improve fidelity, controlled conformational switching during assembly, staged assembly, and the use of templates or jigs to assist in form determination. These principles and paradigms are well illustrated in the polymer pathway of the kinesin ATPase complex. In this talk, a series of vignettes drawn from experimental studies of the assembly of complex biological systems, primarily phage, which serve to illustrate these general principles will be presented.

11:15 AM W1.7/O1.7
Hongyou Fan, Kai Yang, Kevin Malloy, Sigmon Thomas and Jeff Brinker; Sandia National Laboratory, Albuquerque, New Mexico.

Nanocrystals exhibit size-dependent physical and optical properties that are of considerable interest. These properties and their potential applications in catalysis, biolabeling, and microelectronics and optics. Currently, nanocrystals are synthesized in organic solvents. Here we report the synthesis of a new ordered nanocrystal (NC) array through surfactant self-assembly of water-soluble NC/silica thin films mesophases during spin-coating. The intermediate NC-micelles are water-soluble and of interest for bio-labeling. The robust, 3-D NC mesophase solids are of interest for development of collective optical and electronic phenomena, and, importantly, for the integration of nanocrystal arrays in device architectures. Integration of a MOS capacitor using such an ordered NC/silica oxide demonstrated charge storage on the gold nanocrystals and discharge behavior dominated by electron transport within the ordered gold nanocrystal array. Temperature dependent device V caracteristic and electron tunneling behavior have been observed. Sandia National Laboratory is a multiprogram laboratory operated by Sandia Corporation, a Lockheed Martin Company, for the United States Department of Energy under contract DE-AC04-94AL85000.

11:30 AM W1.8/O1.8
Conjugated Polymer/Silica Nanocomposites with Tunable Mesostructure.
Byron McCaughey1, Chris Costello2, Donghai Wang1,1 J Eric Hampsey1, Chaojun Li3, C Jeffrey Brinker1 and Yunfeng Lu1; 1Chemical Engineering, Tulane University, New Orleans, Louisiana; 2Chemistry, Tulane University, New Orleans, Louisiana; 3Sandia National Laboratories, Albuquerque, New Mexico.

Conjugated polymer-silica nanocomposites have been extensively researched because they show enhanced conductivity, mechanical strength, processability, environmental stability, and unique optical properties. Our research focuses on the synthesis of a new multifunctional nanocomposite containing conjugated poly(2,5-thienylene ethynylene) (PTE)/silica nanocomposites with tunable mesostructure. The synthesis approach involves surfactant-induced self-assembly and co-organization of a monomer/polymer and palladium-based catalyst within a poly(silicon acid) matrix. Surfactant choice and self-assembly conditions created hexagonal, lamellar, or cubic silica mesophases. Subsequent polymerization initiates crosslinking the ordered nanocrystal array. Temperature dependent device V caracteristic and electron tunneling behavior have been observed. Sandia National Laboratory is a multiprogram laboratory operated by Sandia Corporation, a Lockheed Martin Company, for the United States Department of Energy under contract DE-AC04-94AL85000.

1:30 PM W2.1
Proton conductive membrane synthesized from biological molecular hybrid.
Iancu Hanea and Masanori Yamada; EEL, AIST, Tsukuba, Ibaraki, Japan.

Proton conductive membrane is a very important materials for functional electrochemical devices such as fuel cells, battery, electrochromic device and sensors. In particular, temperature tolerant as anhydrous proton conductor has been attracted much attention for application to advanced polymer electrolyte fuel cells operated above 100 °C, where the water ion exchange membrane is dried to lose proton conductivity. Here, in this presentation, the anhydrous proton conducting polymer membrane has been synthesized from biological molecular hybrid materials. The chitin phosphate polymer is mixed with uracil/ribose or uracil/ribose/methanol (PTE) molecules to form acid/base hybrid materials. The materials show proton conductivity exceeding 1 mS/cm under non-humified condition up to the 150 °C, which can be potentially applied to the PEMFC membrane operated under high temperature and non-humified condition. The conductive mechanism different from water molecules vehicle or Groththus has been suggested.

1:45 PM W2.2
Bio-inspired and non-lithographic formation of nanodevices.
Harold Craighead, Cornell University, Ithaca, New York.

Observing biological systems at the nanoscale can motivate new device designs and inspire new material processing concepts. We have been adapting lithographic fabrication approaches for patterning self-assembled biomaterials for integration with electronic and optical devices. This may provide the basis for new sensors and diagnostic devices. We have used a simple polymer lift-off process, for example, to create high spatial resolution patterns in lipid bilayers, resembling a cell membrane, containing active receptor molecules. Conversely we have been adapting non-lithographic approaches for creating functional devices, using methods that in some ways emulate the way living systems create complex structures. In a simple example of the bio-inspired approach, we have used a scanning electrodeposition process for creating both biological and inorganic nanoscale devices. For example, we have deposited individual conducting nanofibers to act as high sensitivity chemical sensors with high spatial resolution. We have used a similar non-lithographic approach to create nanofluidic systems and nanoelectro-mechanical devices.

2:15 PM W2.3
Engineered Microchannels for Active Nanomaterials Assembly.
Andrew K. Boal, Joseph M. Bauer, Susan B. Rivera, Ronald P. Manginell, George D. Bachand and Bruce C. Bunker; Biomolecular Materials and Interfaces, Sandia National Labs, Albuquerque, New Mexico.

Our current research is focused on the development of high motor protein integrated microfluidic devices for the active assembly of nanomaterials. These systems, based on surface bound kinesin motor proteins, have been developed to propel microtubules (MTs) bearing nanoparticle cargo, require the fabrication of a microfluidic channel environment capable...
of both directing overall microtubule motion and providing surfaces able to off-load nanoparticle cargo in predetermined locations. To address both of these concerns, we have designed channels with gold walls deposited on oxide surfaces of silicon wafers. Microtubule movement within a channel is largely decided by microtubule-wall collision events, which are in turn highly governed by the chemical nature of the wall surface. Typically, a microtubule-wall collision can lead to one of three scenarios: (1) in environments where kinesin is adsorbed to the channel wall as well as the floor, microtubules almost always move out of the channel, (2) devices where kinesin is placed on the channel wall and collisions mostly lead MT redirection within the channel, and (3) devices where the walls do not adsorb kinesin and also redirect the MTs. To optimize the performance of our devices with regards to microtubule guidance, we have investigated a variety of albumin thioles to selectively form monolayers on the gold walls and their effectiveness at resisting motor protein absorption and directing MT motion has been evaluated. Applications of these devices range from the manipulation of nanomachines using active MTs in conjunction with chemically active walls and surfaces will also be discussed.

2:30 PM *W3.4
Bio-inspired Periodic Micromask Arrays with Integrated Pore Structures Created by Multi-beam Interference Lithography. Shu Yang1, Gang Chen1, Chaitanya K. Ullal2, Mischa Megens3, Yong-Jin Han1, Ronen Rapaport1, Edwin L. Thomas1, Chada Ruengruglikit1, Bell Laboratories, Lucent Technologies, Murray Hill, New Jersey; 2Materials Science and Engineering, Massachusetts Institute of Technology, Cambridge, Massachusetts; 3Food Science, Rutgers, The State University of New Jersey, New Brunswick, New Jersey; 4Philips Research Laboratories, Eindhoven, Netherlands.

Nature has developed strategies that give biological processes and structures exquisite control on multiple length scales. Inspired by the discovery of light-sensitive brittlestars, which have microlens arrays in conjunction with chemically active surfaces will also be discussed.

3:30 PM *W3.1
Templating of Magnetic and Semiconducting Materials a la Microchannel Flow, and Patterning focus explores the efficacy of TiO2 as a photocatalyst for oxidation of alkane and the PEG-bl-PPS-bl-PEG sulfone. The advent of novel characterization and patterning methods allows for manipulation of these polymers and bio-molecules. We have employed ultraviolet (UV) energy, EC-OWLS (electrochemical optical waveguide lightmode) and microchannel flow, and photocatalysis to control sulfide adsorption via light to pattern surfaces. When PEG-bl-PPS-bl-PEG is oxidized, it loses its strong chemisorbed bond to gold and is easily removed from a gold surface. Initial homogenous oxidation studies were performed on PEG-bl-PPS-bl-PEG coated gold surfaces. The PPS polymer backbone, characterized with XPS, changed from a sulfide to a sulfone state upon exposure to UV energy generated in the presence of oxygen. Homogeneous oxidation was also achieved with EC-OWLS on PEG-bl-PPS-bl-PEG coated ITO (indium tin oxide). Further studies were performed to locally oxidize the material. To this end, we first employed a macro-capillary cell setup to perform solution-based electrochemical oxidation of a PEG-bl-PPS-bl-PEG coated gold substrate. Polarization tests were conducted using the micro-capillary electrochemical cell setup in both aqueous and organic environments and staining XPS confirmed chemical oxidation. This oxidation has also been achieved with microchannel fluidic patterning with PDMS (polydimethylsiloxane) stamps on gold. Our present patterning focus explores the efficacy of TiO2 as a photocatalyst for oxidation of alkane and the PEG-bl-PPS-bl-PEG sulfone.

Several commercial exogenous surfactants such as Survanta, Infasurf and Curosurf are all successfully used for lung surfactant replacement therapy to treat RDS (Respiratory Distress Syndrome) although their compositions are quite different. Making use of Langmuir films, a plausible in vitro model for the lung environment we identify common possible differences between commercial surfactants and lung surfactant film. The surface pressure versus trough area isotherms show that all the films meet the requirements regarded as first criteria to define a good surfactant: high collapse pressure and efficient respreading. However, further characterization highlights striking discrepancies. Surface morphologies, as seen by Brewster angle and fluorescence microscopy, are very different and their evolution upon compression and expansion as well. Phase transitions and respreading behavior is also seen to relate to the surface viscosity and thus to the composition.

4:00 PM *W3.3
Biocompatible Sulfur Containing Polymers for Surface Passivation and Patterning. Jane P. Bearinger1, Samuel Terrettta2, Roger Michel2, Christine A. Orme2, Marcus Textor2 and Jeffrey A. Hubbell2, 1MTP/ CMS, LLNI, Livermore, California; 2ETH, Zurich, Switzerland; 3EPFL, Lausanne, Switzerland.

Biocompatible polysulfides have been designed to overcome existing size-heuristics limitations of micromachined devices and use as coatings in the formation of biodiagnostic and bioanalytical devices. Specifically, triblock copolymers of poly(propane sulfide) and poly(ethylene glycol) (PEG-bl-PPS-bl-PEG) were synthesized to stably chemisorb to gold via the PPS domain and present a hydrophilic passivating surface via the PEG domain. The copolymers were tested for stability and the ability to resist adsorption of proteins and attachment of dermal cells. X-ray photoelectron spectroscopy (XPS) and surface plasmon resonance (SPR) results indicate that PEG-bl-PPS-bl-PEG applied to gold renders surfaces more resistant to oxidation than anethanes, as well as resistant to both protein and cells. Fibroblast cell activity was also reduced on PEG-bl-PPS-bl-PEG compared to Curosurf. When adsorbed, it loses its strong chemisorbed bond to gold and is easily removed from a gold surface. Initial homogenous oxidation studies were performed on PEG-bl-PPS-bl-PEG coated gold surfaces. The PPS polymer backbone, characterized with XPS, changed from a sulfide to a sulfone state upon exposure to UV energy generated in the presence of oxygen. Homogeneous oxidation was also achieved with EC-OWLS on PEG-bl-PPS-bl-PEG coated ITO (indium tin oxide). Further studies were performed to locally oxidize the material. To this end, we first employed a macro-capillary cell setup to perform solution-based electrochemical oxidation of a PEG-bl-PPS-bl-PEG coated gold substrate. Polarization tests were conducted using the micro-capillary electrochemical cell setup in both aqueous and organic environments and staining XPS confirmed chemical oxidation. This oxidation has also been achieved with microchannel fluidic patterning with PDMS (polydimethylsiloxane) stamps on gold. Our present patterning focus explores the efficacy of TiO2 as a photocatalyst for oxidation of alkane and the PEG-bl-PPS-bl-PEG sulfone.

4:45 PM W3.4
The Role of Protein Organization on Synthetic Surfaces on the Behavior of Attached Cells. Djordje Nikolic and Jeffrey D. Carbeck; Chemical Engineering, Princeton University, Princeton, New Jersey.

Control over the organization of proteins on surfaces on microscopic length scales is important in the development of biosensors and protein micro-arrays, as well as in the organization and control of growth of cells on surfaces. Protein conforming and protein patterns is finding widespread use in studying the functional effects of cell adhesion to substrates, usually using patterns similar to the size of a cell (10 - 50 μm). When cells adhere to surfaces, they form discrete adhesive sites called focal contacts that range in size from less than 0.1 nm to approximately 1 to 2 μm. To determine how the distribution of...
focal adhesions regulates cell behavior requires patterning on length scales much smaller than the diameter of a single cell. We are developing several strategies to organize proteins that direct cell adhesion on these length scales. In one approach, we have developed a new method for the tailoring of proteins on surfaces, based on patterning of 2 μm colloids functionalized with proteins, which provides control over protein organization on multiple length scales. We used surfaces functionalized in this way to form patterned arrays of cells and to study the role of adhesive contacts on the behavior of anchorage-dependent cells. To separate effects of protein organization from topology, we have developed a new method that combines evaporative silanization with photolithography to produce regularly spaced 2 μm islands of the cell adhesion protein fibronectin on silicon wafers. Several templates with different spacing between islands have been made (1, 2, 4, 8 μm). The separation between islands was chosen to bracket the average distance between adhesive sites naturally formed by cells on homogeneous coatings of fibronectin. Thus, we can determine the optimal distribution of adhesive sites for activating cells. Preliminary results suggest that the cell size may be inversely proportional to the distance between islands.

SESSION W4: Poster Session I: Biological and 
Bio-Inspired Materials and Devices 

W4.1 Biological Functionalization of Carbon Nanotubes. 
Ranjani Sirdeshmukh1, Kousik Sivalakumar2 and Balaji Panapachatisan; 
Department of Electrical and Computer Engineering, University of 
Delaware, Newark, Delaware.

Carbon nanotubes are known for the exceptional electrical and 
mechanical properties. The size dependant properties 
of nanomaterials have made them very attractive to develop highly 
sensitive sensors and detection systems. This is especially true in 
biochemical sciences, where the efficiency of a detection system would 
reflect on the size of the detector and the sample required for 
detection. At approximately 1 to 10 nm wide, and approximately 1.5 
and 2 μm long, the use of carbon nanotubes as sensors in biological 
systems would greatly increase the speed and accuracy of detection and 
diagnostics, for a highly reduced sample size. If binding proteins 
to the surface of nanotubes could vary the surface states on them, 
then it would result in varied electrical and optical properties, which 
would be very sensitive to the reactions occurring on the surface. It 
has been seen that proteins have a higher affinity to adhere to 
metallic surfaces. When metallic nanoparticles such as silver and 
platinum are electrochemically-deposited on the surface of nanotubes, 
silver and platinum nanowires are formed. Due to the small sizes of these 
nanowires (diameters ranging from 10 to 100 nm, and lengths of approximately 1.5 μm), the surface effects due to proteins and the actions of the cell surface receptors could be just as prominent. In this paper, we show the deposition of a fluorescent dye, fluorescein 
isothiocyanate (FITC) conjugated to an antibody with a 
concentration of 10 μL of 1 μg/mL antibody in 500 μL of Phosphate 
buffed saline on the surface of the silver and platinum nanowires and 
carbon nanotubes through fluorescence and reflection images 
taken using a multiphoton confocal microscope. The results from the 
images suggests the strong interaction or binding of the antibodies to 
the surfaces of the nanowires and nanotubes which lends promise for 
this technique for applications as drug delivery and detection vehicles in 
the area of biomedical nanotechnology. We are currently studying the 
fluorescence intensity as a function of the concentration of proteins 
on the surface of the nanowires and nanotubes and this will be 
presented at the conference. Detection, through the occurrence of 
fluorescence, and diagnostics using this technique could bring the 
sample size down to single-cell measurements, and could hold 
impressive potential in the area of biomedical nanotechnology.

W4.2 Photochemistry of self-assembled protein cages. 
Zachary B Varpnco1,2,3, Jesse Mosolf1,2,3, Dan Ensign1,2,3, Michelle 
Flenniken1,2,3, Debbie Wills1,2,3, Mark Young1,2,3 and Trevor 
Douglas1,2,3; 1Chemistry and Biochemistry, Montana State University, 
Bozeman, Montana; 2Department of Plant Sciences, Montana State 
University, Bozeman, Montana; 3Center for BioInsipired Nanomaterials 
(CBIN), Montana State University, Bozeman, Montana.

We have utilized the self-assembled protein cages from ferritin, 
ferritin-like proteins, and viral capsids, as size constrained reaction 
environments for nanomaterials synthesis. The nanoparticles are 
formed using either a photochemical reduction or via oxidative 
hydrolysis of precursor ions. The resulting transition metal oxide and 
metallic nanomaterials are monodispersed and have dimensions 
commensurate with the inner diameter of the protein cages. We 
have characterized the physical properties of these materials, and in 
particular the photocatalytic activity of both the unmineralized and 
mineralized protein cages. The chemical plasticity of these protein 
cages towards chemical and biological manipulations makes these highly 
versatile materials for imparting function by design.

W4.3 Microtubule Templated Synthesis of Inorganic 
Nanomaterials. Andrew K. Bou1, Thomas J. Headley, Ralph G 
Tissot and Bruce C. Bunker; BioMolecular Materials and Interfaces, 
Sandia National Labs, Albuquerque, New Mexico.

Protein microtubes (MTs) of polymerized tubulin have been 
used as templates for the biominetic synthesis of metal oxide and 
metal sulfide nanomaterials. MTs off a unique biomolecular scaffold 
as they have a cylindrical core, high aspect ratio (25 nm in diameter and 1-1000 nm 
long), and can be dynamically grown on a selected region on a substrate. As 
a first example of MT-based template synthesis of nanomaterials, we have 
prepared iron oxide coated MTs. Exposure of MTs to anaerobic aqueous solutions of Fe2 added to MT solutions: low amounts of Fe2 yielded 10-30 nm thick layers of nanomagnetic iron oxides. Higher amounts of Fe2 yielded MTs coated with crystalline layers of FeOOH up to 250 nm thick. On the micron scale, these coated MTs were observed to form large, amorphous bundles. Since iron oxide nucleation occurs relatively independently on the MT surface, it is likely that the highly negatively charged MT surface is responsible to the formation of the observed structures. Progress has also been made towards the synthesis of both metal sulfides and metal oxide coated MTs, formed by co precipitation on the MTs. These bionanomaterials are being used in biological sciences, where the efficiency of a detection system would 
be inversely proportional to the distance between islands.

W4.4 Deposition of Patterned Calcium Carbonate Film using a 
Binary Surfactant System for Drug Detoxification 
Applications. Javier Gutierrez, Laurie Gower, Debra Lush, Vishal 
Patel and Brad Carey; Materials Science and Engineering, University 
of Florida, Gainesville, Florida.

In the United States alone, over 300,000 patients enter the emergency 
room each year due to prescription drug overdose complications. 
In fact, the leading method of suicide is via overdose of amitriptyline, a 
"micro-sponge" that will absorb the overdosed drug. Then, through 
hydrolysis of precursor ions. The resulting transition metal oxide and 
mineralized protein cages. The chemical plasticity of these protein 
cages towards chemical and genetic manipulations makes these highly 
versatile materials for imparting function by design.

W4.5 Semiconductor Nanocrystals Arrayed on Cellulose and 
Cellulodextrins. Jun Feng1, Yong-Hyun Kim1, Shengbai Zhang2, 
Shiyun Ding3, Melvin P. Tucker3, Garry Rumbles2 and Michael E. 
Himmel1; 1National Bioenergy Center, National Renewable Energy 
Lab, Golden, Colorado; 2Basic Science Center, National Renewable 
Energy Lab, Golden, Colorado.

We are investigating potentially useful interactions between cellulese 
and semiconductor particles and nanocrystals. Cellulose has a unique microcrystalline structure composed of repeating units of 
cellulobiose stabilized by interchain hydrogen bonding. Native cellulose has 
distinctly hydrophobic (1,0,0) and a hydrophilic faces (1,1,0). Plant 
cellulose tends to form 10 to 100 micron size amorphous bundles and 
bacteria cellulose is observed on organized lower plants. In 
preliminary work, we have discovered that both cellulese nanoparticles 
and cellulose fibers demonstrate strong attraction for certain kinds of 
semiconductor nanocrystals, including TOPO-(CdSe)ZnS quantum 
dots (QDs). An initial indication of this interaction was the observation that cellulose exposure caused dispersion of QDs in water, 
yielding photoluminescent cellulose. Finding ways to overcome the 
hydrogen bound network of the cellulose crystal is challenging, 
however, cellulose microfibrils (small bundles of cellodextrins) may be 
useful for arraying QDs. We have also examined the interactions 
with other nanomaterials,
between cyclodextrins (CDs) and (CdSe)ZnS QDs. Cyclodextrins have ring structures with discrete dimensions and the inner lumen of CDs in hydrophobic regions, which can accommodate the 8 x 12 chains of the TOPO molecules used to passivate (CdSe)ZnS QDs during growth. We attribute the CD-sized dispersion of TOPO-QDs in water to overall entropic benefits afforded from hydrophobic shielding of the QDs. We also found that CD-QD solutions demonstrate a sizable 15-nm red shift. To develop an understanding for this red shift, we have performed first-principles density functional theory calculations that show that cyclodextrin hydrogen bonds to Cd-QD surface hydroxyl groups and form coordinate bonds with Zn, which explains the decrease in band gap observed experimentally. We also present AFM and SEM images of QDs arrayed on bacterial cellulose and cyclodextrins.

**W4.6 Direct Patterning of Membrane-Derivatized Colloids with in situ UV-Ozone Photolithography. Cheng-Han Yu1, Atul N. Parikh1 and Jay T. Groves1,** 1Department of Chemistry and Physical Biosciences Division, Lawrence Berkeley National Laboratory, University of California, Berkeley, California; 2Department of Applied Science, University of California, Davis, California.

A novel lithography method has been developed for patterning biological materials, e.g., lipid bilayer membranes. 3D patterns of the fluid membranes supported on silica beads have been achieved by in situ ultraviolet photolithography. Deep UV (185-254nm) generates ozone and reactive radicals, which chemically degrade and remove lipids with high spatial resolution, even in entirely aqueous environment. In this technique, we utilize backside UV illumination through the substrate, which also serves as microfabricated photomasks. Membrane-coated colloidal particles are allowed to settle gravitationally onto this substrate, in which arrays of patterns are in registry with surface pits that position the particles. This process is easy 2/3-batch photolithography as well as with standard bottom-up techniques. Micron-resolution membrane patterns on the colloidal particle surface are readily observed by epifluorescence microscopy. The removal of lipid bilayer was confirmed by the ability to refill with small unilamellar vesicles (SVV) of different lipid compositions, or proteins which bind to bare silica surface. The uniqueness of this photopatterning technique is that it occurs entirely in liquid environment and does not require development steps. Geometrically asymmetric structures on colloidal particles are prepared by presentation of biological signals (e.g. cell surface signaling molecules) to live cell cultures. They also create new possibilities to understand the general construction of 2D colloidal materials. In addition, the organization of them can be modified by adding biological polymerizable molecules in the membrane composition. This may give certain contributions to development of new biomechanical systems.

**W4.7 Bio-inspired Crystal Growth Induced by Novel Organic Compounds. Nicholas Bryan Dundale and Bridig R Heywood,** School of Chemistry & Physics, Keele University, Keele, Staffordshire, United Kingdom.

It is now well established that, to design materials with useful properties (e.g. precisely controlled particle size and morphology) it is highly beneficial to borrow processes and concepts from living and biological systems, and exploit similar principles in the manufacture of synthetic materials. Living organisms have developed extremely efficient materials which exhibit a finely-balanced compromise between desirable properties, such as low density and high mechanical strength. This study continues previous work by investigating the effect of various novel organic compounds (e.g. novel calixarene derivatives) not unlike those potentially found in organic systems, on the crystallisation of compounds including barium sulphate and calcium carbonate. Of interest in this study will be the usual properties of crystals, including morphology, particle size and uniformity. The effect of the concentration and substituent chain length of the ligand is investigated. More specifically, one key aspect for discussion will be that of the, relatively poorly understood, crystallographic phenomenon of twinning. Large scale multiple twinning has been observed in several of the experiemits conducted. Twinning occurs when two crystals are intergrown in a symmetrical manner, brought about, for example, by the sharing of a mirror plane or rotational axis. Twinning is thus explainable, for example, by the sudden reversal of layers in the ionoc packing (ABCBA) in the same mirror plane, the same way as can be demonstrated. More twinning is little understood, although it is clear here that the presence of additives favours this process. This study will attempt to suggest means by which this twinning may occur.

**W4.8 Effects of Materials Properties on Cell Culture. Bing Shi1, Aaron Fairchild1, Zenith Kleine2, Tom Kahn2 and Hong Liang1,** 1Mechanical Engineering, University of Alaska Fairbanks, Fairbanks, Alaska; 2Chemistry and Biochemistry, University of Alaska Fairbanks, Fairbanks, Alaska.

Biomaterials have been widely used in artificial joints, such as total hip and knee joint replacements. Their design, texture, properties, and biomechanical performance are of great importance. In this research, we investigate effects of materials properties, such as wettability and texture of selected materials on cell culture. We conduct cell culture experiments on conventional materials such as glass, polyurethane, and polyvinyl alcohol. The surface hydrophilicity and cell adhesion relationships are discussed in this presentation.

**W4.9 Reflective Interferometric Detection of Tag-Free Biomolecules. Jinghui Lu1, Benjamin Miller2 and Lewis Rothberg1,** 1Chemical Engineering, University of Rochester, Rochester, New York; 2Dermatology, University of Rochester, Rochester, New York.

Sensitive and selective schemes to detect biomolecules are important enabling tools in medicine, environmental monitoring and biological research. The vast majority of sensing instruments are based on fluorescent tagging of molecules in the sample under investigation, a time-consuming and expensive process. In addition, the fluorescent readout schemes currently available require relatively expensive imaging systems and detectors. Simplification of the chemistry to avoid tagging of analytes and development of possible inexpensive readout systems is a realistic possibility. Here, we present a reflective interferometric detection technique that can be used to detect tag-free QDs arrayed on bacterial cellulose and cyclodextrins. We describe here a novel lithography method based on destructive interference of reflected light that is sensitive, quantitative and has a simple format. The reflective interferometric detection technique is able to discriminate surface thickness changes as low as a monolayer change on a silicon waffer, and requires no preprocessing.

**W4.10 Glyceria Jaws: A Biocomposite of Metals, Melanin and Proteins. Dana Novak1, Helga Lichtenegger2, John Harrell3, Nelle Slack4, Galen Stucky2 and Herbert Waite1,** 1BioMolecular Science and Engineering, University of California, Santa Barbara, Santa Barbara, California; 2Materials Science and Testing, Vienna University of Technology, Wien, Austria; 3Chemistry and Biochemistry, University of California, Santa Barbara, Santa Barbara, California; 4Materials, University of California, Santa Barbara, Santa Barbara, California.

Glyceria is a marine polychaete worm equipped with four syringe-like jaws to inject venom into its prey. The jaws have unusually high resistance to wear, especially when considering their low density and high mineralization. Unmineralized zinc and both mineralized and unmineralized forms of copper are present in Glyceria jaws. We have determined that Glyceria jaws contain melanin as a major component. Melanin-composes approximately one third of the jaws by mass. In this work we explore the mechanical and structural ramifications of melanin in the jaws. The interaction of melanin with jaw proteins and metals is also examined. Glyceria jaws are about 50% copper by weight, which contain about 2% Si, 1% Fe and 1% Mn. Certain proteins may have even higher histidine contents. Given their role in other organisms, histidine-rich proteins are likely to interact with zinc and copper in Glyceria jaws. In this work we identify and characterize metal-binding jaw proteins and investigate their contributions to material properties of the jaws.

**W4.11 Observation of nano surface structures of various plant leaves with ultra water-repellency. Osamu Takai1, Yunying Wu1, Masao Kouno2, Hiroyuki Sugimura2 and Yasushi Inoue2,** 1Center for Integrated Research Science and Engineering, Nagoya University, Nagoya, Japan; 2Department of Materials Processing Engineering, Nagoya University, Nagoya, Japan.

Studies on some plant leaves revealed that the super-hydrophobic property was independent of the shapes of nano-scale asperities but mainly affected by these nanostructure although the surface of these leaves consists of both nano- and microstructures. The results from the natural world provide a guide for constructing artificial
super-hydrophobic surfaces with nano-scale fine roughness by Field Ion Beam (FIB). The water contact angle of such artificial surface was mainly affected by the presence of phase separated nanostructures. The results of more air trapped in the pores between higher height asperity if the fine-rough surface was made up of nano- (or submicro-) scale feature. This work is supported by JSPS - RFTIP913101 and ASTF.

W4.12 Macroscopic Spherical Assemblies from Charged Polypeptides and Small Multivalent Counterions. Brendan John McKenna1, Henrik Birkedal1, H. Bartl2, Timo Berg1, Deming2,1, Galen D. Stucky1,2,1. Chemistry, UC-Santa Barbara, Santa Barbara, California; 2Materials, UC- Santa Barbara, Santa Barbara, California. Micrometer-sized spherical vesicles have been found to assemble from homopolymer electrolytes and small, multi-charged counterions in water. In contrast to previous efforts, these vesicles do not use preformed templates, do not require block copolymers, and do not necessarily employ nanoparticles. We have investigated the requirements for vesicle formation with regards to both components of the assembly. We have found self-assembly to occur with 3 different polypeptides and a variety of counterions, all of which require a minimum number of charged groups to promote supramolecular crosslinking. Two of the polypeptides are formed by amino acids with positively charged side chains while the remaining polymer is anionic. Assemblies made with positively-charged amino acids can be stabilized by adding an outer layer of silica. We show how the assembly process is controlled by pH and how, in consequence, the pH of the charged organic groups can be used to reliably predict sphere formation. By varying the nature of the small counterions, we have determined the requirements for spherical vesicle formation. The results have been further investigated using confocal microscopy and fluorescent labeling of the different components. The mode of assembly and the chemical interactions leading to assembly are discussed.

W4.13 2D Colloidal Arrays of Monodisperse Phase Separated Membrane Domains Formed in a Supported Bilayer. Yoshihiko Kizuka1,2,3, Aaron Rozovsky2,1, and Jay T. Groves1,2,1. Chemistry, University of California, Berkeley, California; 2Physical Bioscience Division, Lawrence Berkeley Laboratory, Berkeley, California. In cellular membranes, lateral phase separation is widely believed to be involved in a variety of cellular processes, such as intracellular sorting of membrane proteins or immune cell recognition. Phase-separated domains, rich in cholesterol and sphingolipids, sometimes called rafts, are currently of interest in medicine and biology. Quantitative study of membrane lateral phase separation and its role in cellular processes requires precisely defined model membrane systems. We have recently introduced a variety of supported membrane structures formed by the rupture of giant unilamellar vesicles onto conventional supported membranes (supported intermembrane junction). These systems exhibit a number of phenomena not observed in isolated systems. Most notably, phase-separated structures can form in an environment that allows for free movement and interactions and facilitates imaging analysis. In general, phase-separated domains collide and coalesce without boundary conditions, under certain conditions of stabilized superstructures can form. These include monodisperse ordered arrays of 1 μm diameter rafts and lamellar stripe patterns. The planar configuration enables interferometric imaging of membrane topography with nanometer precision. Characteristics of superstructures and applications to cell membrane studies will be discussed.

W4.14 Calcium Oxalate Precipitation at Phase Separated Phospholipid Langmuir Monolayers. Daniel R. Tatham and Issa O. Benitez, Department of Chemistry, University of Florida, Gainesville, Florida. Calcium oxalate and calcium phosphate are the principal crystalline materials found in urinary stones. The inorganic crystals are always mixed with an organic matrix composed of carbohydrates, lipids and proteinaceous materials that account for about 2% of the total mass, although a much larger percentage of the total volume. To better understand the how the stone forms we have studied the interactions between the organic and crystalline components. We have previously performed a series of studies of calcium oxalate precipitation at an interface provided by phospholipid Langmuir monolayers that serve as models for the phospholipid domains within membranes. We observed that the Langmuir monolayers can effectively catalyze the precipitation of calcium oxalate monohydrate (COM) and that the identity of the monolayer has a strong influence on the rate of crystal formation. The present study investigates the role of phase boundaries on COM precipitation by studying phase-separated Langmuir monolayers. Brewster angle microscopy is used to monitor COM growth at monolayers in LC/LE coexistence and LE/LC coexistence. The results of these separate experiments are also studied. COM precipitation is enhanced at monolayers containing a single lipid in phase coexistence, where the phase boundary is dynamic. Phase separated mixtures of different lipids show an enhancement in crystal formation due to the presence of a boundary.

W4.15 A 1.7-Kilobase Single-Stranded DNA that can be Folded into a Regular Octahedron. William M. Shih1,2,3, Joel D. Quispe4 and Gerald F. Joyce2,1,3. Chemistry, The Scripps Research Institute, La Jolla, California; 2Molecular Biology, The Scripps Research Institute, La Jolla, California; 3The Skaggs Institute for Chemical Biology, The Scripps Research Institute, La Jolla, California; 4Cell Biology, The Scripps Research Institute, La Jolla, California. Rigid DNA-based nanostructures may play a role in the construction of miniaturized devices through their ability to direct the assembly of materials on the subnanometer to micrometer scale. A key property of DNA - its ability to be amplified exponentially by polymerases - facilitates both the large-scale cloning production of individual sequences and the directed evolution of sequence lineages toward optimized behaviors. Previous examples of three-dimensional geometric DNA objects, however, are not amenable to cloning because they contain topologies that prevent copying by polymerases. Here we show the design and synthesis of a 1,065-nucleotide, single-stranded DNA molecule that is readily amplified by polymerases and that, in the presence of five 40mer oligonucleotides, can be folded into a regular octahedron framework by a simple denaturation-renaturation procedure. The resulting octahedron, approximately 22 nanometers on a side, was visualized by cryo-electron microscopy and shown to have the predicted structure. No eleven base-pair sequence appears twice in the octahedron. Thus each part of the octahedron is uniquely addressable by the appropriate sequence-specific DNA binder. Inorganic and organic double templates were used to fabricate silica nanotubes with nanochannels perpendicular to the shells. Needle-like CaCO3 nanoparticles, synthesized by a high gravity reactive precipitation method, were used as inorganic and organic surfactant templates. Field emission scanning electron microscopy (FESEM) and transmission electron microscopy (TEM) were employed to characterize the nanotubes and nanochannel structures. It can be concluded that the length and diameter of the nanotube can be controlled by the inorganic inorganic CaCO3 nanorods and templates and the nanochannels in the shells can be tuned by different surfactant micelles. The nanotubes with nanochannels perpendicular to the shells have a potential application in chemical bio-catalyst, bio-separation, and drug delivery.

W4.16 In Situ Protein Directed Silica Biomimeralization. Diana D. Glinos1, Francisco Rodriguez1, Rajesh R. Naik2 and Morley O. Stone1. 1Engineering Science Department, Trinity University, San Antonio, Texas; 2Materials Directorate, Air Force Research Laboratories, Wright-Patterson AFB, Ohio. Numerous examples of nanopatterning and nanostructure are commonly found in nature, most apparent in the marine diatom and sponge. The diatom cell walls are considered as a paradigm for the controlled production of nanostructure silica. Peptides, such as silicateins, silafins and poly-L-lysine are capable of precipitating silica from a silane precursor. The biomimetic formation of silica nanotemplates using polypeptides allows control of morphology at the structural level desired for advanced applications. Here we demonstrate the deposition of silica nanotemplates onto conductive surfaces using electrophoresis. The exposure of the protein-coated surface to a silane precursor resulted in the formation of a network of plate-like silica structures. The silica morphologies are distinctly different from those observed by a variety of different surfactant micelles. The silica nanotemplates utilize different silica precursors, allowing for the potential application in chemical bio-catalyst, bio-separation, and drug delivery.
Reconstructing the growing interest in biomimetic chemistry as a powerful approach to combine complexity and functionality in new materials [1], the design of versatile nanoparticle–based superstructures has recently evolved into a dynamic discipline where biological concepts and entities are playing a crucial role. In particular, the emerging field of nano-electronics faces challenges, such as the anisotropic organization of spherical nanoparticles, which could be addressed by using biological entities as structuring building blocks. In this respect, we will show how self-assembled cylindrical particles of wild-type and recombinant tobacco mosaic virus (TMV) can be used as organic templates for the controlled deposition and organization of Pt, Au, or Ag nanoparticles [2]. Chemical reduction of [PtCl6]2- or [AuCl4]­­­ complexes at anodic over potential rise to the formation of a thin nanofilm that is anisotropic and highly oriented. The number of encapsulated nanoparticles increased when Ag benoate rather than Ag nitrate was used due to its reduced supersaturation. The control of the orientation of the surface-templating effect of the channel wall carboxylates of TMV resulted in nucleation and constrained growth of discrete quantum dots by molecular engineering of the internal and external organization of spherical nanoparticles, which could be addressed by biomimetic synthesis of a range of composite materials with practical applications. The potential of using engineered polypeptides such as DNA and proteins as ‘building blocks’ for the construction of functional nano-structures via self-assembly offers many advantages including homogeneous ‘building blocks’ population, self assembly mediated by bio-recognition and a sound basis of structural and biochemical data. The use of proteins as ‘building blocks’ is particularly attractive due to the large variety of shapes and sizes. Here we describe the use of protein crystals, obtained by methods routinely employed for X-ray diffraction studies, as templates for the preparation of novel composite materials. A unique family of nano-structured materials comprised of three dimensional highly ordered alternating arrays of biological moiety (the protein) and synthetic or metallic moieties embeded within crystal cavities may be thus created. Two kinds of ‘filling’ will be described: crosslinked organic–binding polypeptides could be genetically engineered to introduce functionality at the outer surface has allowed us to pattern the inner surface of the protein cage we can exert control over the surface-templating effect of the channel wall carboxylates. Techniques to have a specific recognition moiety for desired materials surfaces and to have a specific recognition moiety for desired materials surfaces. Our results demonstrates feasibility of this approach, paving the way to the fabrication and use of a new family of protein-based composite materials.
material that were ordered at the nano-scale and micron-scale. Periodic domains continuously propagated over a centimeter length scale, which was verified using various optical and electron microscopy techniques. Anti-strepavidin viruses, which could specifically bind to strepavidin previously conjugated with many nanomaterials, were also used to modulate nanomaterials in the self-assembly virus system. Resultantly, composite films had chiral anisotropic C structure due to the helical surface of the M13 virus. Additionally, 20 micrometer diameter fibers were fabricated with liquid crystalline virus suspension using wet-spinning process which mimicked spider’s spinning process. After blending with highly soluble polyvinyl pyrrolidone, nanoscale diameter fibers were fabricated using the electrospinning process. This approach to aligning nanomaterials in a genetically-engineered M13 virus-based liquid crystal system has several advantages. Monodisperse biopolymers (M13 virus) of specified lengths can be easily prepared by molecular cloning techniques. By genetic selection of a peptide recognition moiety, one can easily modulate and align different types of nanomaterials in 3D ordered structures. We anticipate that our approach using recognition as well as a liquid crystalline self-assembly system of engineered viruses may provide new pathways to organize electronic, optical, and magnetic materials.

SESSION W6: Calcium Phosphates as Biomaterials - Thursday Afternoon
Chair: William J. Landis Wednesday Morning, April 14, 2004 Room 3003 (Moscone West)
10:30 AM *W6.1

Human dentin consists of collagen matrix reinforced with apatite (35% organic and 45% mineral) similar in its nanostructure to bone and cementum (apatite). The organic component is responsible for improving the ductility and fracture toughness and mainly consists of collagen type-I in the form of fibrils. The distribution of minerals inside (intrafibrillar) and between the fibrils (extrafibrillar) defines the magnitude of storage and dissipation of structural loading. We have used atomic force microscopy (AFM) and micro-Raman spectroscopy to investigate the kinetics and structural and mechanical property changes during demineralization of human dentin collagen fibrils. For intrafibrillar demineralization studies, single dentin collagen fibrils were isolated. Collagen fibrils were imaged on a glass slide in real-time while immersed in trypsin for 2h and subsequently in 10% citric acid. Structural changes of collagen fibrils, including axial periodicity, diameter, and gap height were determined. We found that gap-overlap depth gradually increased with time (initial rate 0.35 nm/sec), linearly with the square root of the time interval. For extrafibrillar demineralization, we produced recombinant amelogenin rM179 adsorbed onto silica and determined the maximum adsorption sites of rM179 on synthetic hydroxyapatite crystals by means of Langmuir model for protein adsorption. The adsorption experiments were followed by optical wave light spectroscopy (OWLS). The used model surfaces are polyelectrolyte multilayer onto apatite crystals was confirmed by determining the adsorption isothersm of rM179 on synthetic hydroxyapatite crystals by means of Langmuir model for protein adsorption. The adsorption affinity of amelogenin onto apatite and the maximum adsorption sites were measured. The adsorption affinity of amelogenin onto apatite and the maximum adsorption sites were measured. The adsorption affinity of amelogenin onto apatite and the maximum adsorption sites were measured. The adsorption affinity of amelogenin onto apatite and the maximum adsorption sites were measured. The adsorption affinity of amelogenin onto apatite and the maximum adsorption sites were measured. The adsorption affinity of amelogenin onto apatite and the maximum adsorption sites were measured. The adsorption affinity of amelogenin onto apatite and the maximum adsorption sites were measured. The adsorption affinity of amelogenin onto apatite and the maximum adsorption sites were measured. The adsorption affinity of amelogenin onto apatite and the maximum adsorption sites were measured. The adsorption affinity of amelogenin onto apatite and the maximum adsorption sites were measured.

11:00 AM *W6.2

The outermost covering of vertebrate teeth, enamel, is itself a unique composite bioceramic material that is the hardest tissue in the vertebrate body. The dental enamel is formed by ameloblasts, whose name is derived from the Greek word for "to make new," implying a dynamic tissue. Ameloblasts produce the enamel matrix which consists of collagen and calcium phosphates (CaP). Amelogenins constitute more than 90% of the secretory stage enamel matrix proteins. The assembly of amelogenin protein into nanospheres has been postulated to be a key factor in the stability of enamel extracellular matrix framework, which provides the scaffolding for the mineralized apatite crystals to grow. Amelogenin self-assembles into nanospheres that mimic the microenvironment similar to natural tissue. Here we present a method to prepare calcium phosphate/collagen nanocomposite coating by electrochemical deposition in aqueous solutions. The coating was prepared on pre-cleaned silicon wafer or polished titanium plate by controlling the electrolyte pH at 4.9 to 5.5. The electrolyte was prepared with the ion concentration of calcium and phosphate of 200mM and 240mM respectively. The coated films contained 25 wt% of collagen and formed the electrolyte. After deposition, the coating was washed by distilled water to remove any excess electrolyte and dried in a vacuum oven.

11:45 AM W6.4
Calcium phosphate/collagen bone-like nanocomposite coating by electrochemical co-deposition, Yuwei Pan, Ke Duan and Rizhi Wei, Department of Materials Science, University of British Columbia, Vancouver, British Columbia, Canada.

A combination of making better biomaterials for tissue repair is to mimic the micro-environment similar to natural tissue. Bioceramic bone-like composite of collagen and calcium phosphates (CaP) has been reported to have high potential for bone repair. Here we present a method to prepare calcium phosphate/collagen nanocomposite coating by electrochemical deposition in aqueous solutions. The coating was prepared on pre-cleaned silicon wafer or polished titanium plate by controlling the electrolyte pH at 4.9 to 5.5. The electrolyte was prepared with the ion concentration of calcium and phosphate of 200mM and 240mM respectively. The coated films contained 25 wt% of collagen and formed the electrolyte. After deposition, the coating was washed by distilled water to remove any excess electrolyte and dried in a vacuum oven.
water and fixed immediately by Kanowsky fixative then gradient dehydrated and critical point dried. The macro- and microstructure of the coating containing denatured collagen fibers was examined by SEM. Fluorescent micrograph taken under the excitation of blue light, where the glutaraldehyde fixed collagen has strong green fluorescence emission, showed the collagen-CaP coating has high porosity 3D structure with pore sizes ranging from 1 to 2 mm. In the demineralized coating, collagen keeps a similar three-dimension framework. The collagen fibers are as small as 70 nm in diameter. On partly demineralized sample, about 50 nanometer sized crystals were found located along the fibres. These observations indicate that the fibrillation of collagen and mineralization are formed almost spontaneously. XRD spectrum indicates the coating is a DCPD like mineral at low pH and OCP like mineral at higher pH above 5.3. Cathode electrode reaction causes the increase of pH value locally and the supersaturation of the calcium phosphate. Soluble type I collagen molecules are able to self-assemble into collagen fibrils intrigued by increased pH. At the meantime, the calcium phosphate supersaturated locally near to the cathode will easily nucleate on the self-assembled collagen framework to form nano-CaP/collagen composite coating. In summary, electrochemically prepared coating has the morphology of nano-sized collagen fibrils with CaP nano-crystals. This composite coating is similar to the natural bone in structure, composition, and formation process. This study on the co-deposition of collagen and CaP points out a new technique of co-depositing other charged proteins or polyelectrolytes with CaP by a biomimetic way. This work is funded by NSERC. CHIR. R. Wang is an incumbent of the William and Millie Matheson postdoctoral fellowship. References: 1. Du C, Cui FZ, Zhang W, Feng QL, Zhu XD, de Groot K, J. Biomater. Mater. Res. 50 (4): 518-527, 2000 2. Roessler S, Born R, Scharnweber D, Worch H, Sewing A, Dard M, J. Mater. Sci.-Mater. in Med. 12 (10-12): 871-877, 2001 3. Shirkanzadeh, M. J. Mater. Sci. Mater. in Med. 9:67-72, 1998
Spider dragline silks are renowned for their high strength (ca. 1 GPa) and exceptional toughness (energy to break 150-200 MJ m⁻³), and as a consequence there is considerable interest in the production of genetically engineered materials based on the protein sequence designs of these silks. In addition to the physical properties of dragline silks, there is a significant descriptive appeal of the silk-making process. However, relatively little is known about the material properties of these structures. While nanoindentation is now commonly used to probe the mechanical properties of mineralized tissues, most such studies have used indentations that are deep relative to lamellar dimensions and have not reported sample surface roughnesses. Here, we report the mechanical properties of lamellar bone were examined. The effects of surface roughness and maximum nanoindentation load on the measured mechanical properties of lamellar bone were determined. The mechanical properties of lamellar and interlamellar bone in two samples of differing surface roughness were determined at multiple maximum nanoindentation loads. At low loads, the indentation modulus of the lamellar bone was approximately 20% greater than that of the interlamellar bone, while at high loads the measured properties of both layers converged to a composite value. As indentation depth increased relative to surface roughness, the variability of the interlamellar properties decreased substantially, while that of the lamellar properties remained constant. Relatively shallow indentations made on smooth surfaces revealed significant differences in the properties of lamellar and interlamellar bone that support microstructural observations of lamellar bone as more mineralized than interlamellar bone.
materials were designed as bone mimics, and it was necessary to determine whether the mechanical properties of the composite material were similar to those of bone tissue. We also wanted to test the crystalline mineral phase separately as a secondary method of determining the relative amounts of amorphous and crystalline mineral phase. We performed mechanical tests using a Hysitron Triboindenter apparatus, and each sample was loaded under uniaxial compression and evaluated for the surface modulus and hardness values of the composite materials. We tested both the hydrated and dry samples. Then, we measured the time-dependent mechanical properties of the materials using the Hysitron Triboindenter module. In creep experiments, constant load was maintained on the sample, while the material was allowed to continue displacing underneath the load. Data from these experiments yielded characteristic time constants that will be used to construct viscoelastic material models of the composite mineralized gel system.

4:30 PM W8.7

Nanowiring Enzymes to Carbon Nanotube Probes,
Charles Patrick Collier, Maria Jose Espandian, Ian Ross Shapiro and Vern Garrett Bittner; Chemistry, California Institute of Technology, Pasadena, California.

The observation of spectroscopic signals in response to mechanically induced changes in biological macromolecules can be enabled at an unprecedented level of resolution by coupling single-molecule manipulation/sensing using carbon nanotube probes with single-molecule fluorescence imaging. Proteins, DNA and other biomolecules can be attached to nanotubes to give highly specific single-molecule probes for the investigation of intracellular dynamics, the assembly of hybrid biological and nanoscale materials and the development of molecular electronics. Recent advances in nanotube fabrication and Atomic Force Microscope (AFM) imaging with nanotube tips have demonstrated the potential of these tools to achieve high-resolution images of single molecules. In addition, proof-of-principle demonstrations of nanotube functionalization and attachment of single molecules to these probes have been successfully made. Improved techniques for the growth and attachment of single wall carbon nanotubes, as well as novel and well-characterized tools for AFM imaging are being developed. This work serves as a foundation towards development of single-molecule sensors and manipulators on nanotube AFM tips for a hybrid atomic force microscope that also has single-molecule fluorescence imaging capability. An individual single-wall carbon nanotube attached to an AFM tip can function as a structural scaffold for nanoscale device fabrication on a scanning probe. Such a probe can have a novel role, to trigger specific biochemical reactions or conformational changes in a biological system with nanometer precision. The consequences of these perturbations can be read out in real time by single-molecule fluorescence and/or AFM sensing. For example, electrical wiring of single redox enzymes to carbon nanotube scanning probe tips allows for observation of electrochemical control of single enzymatic reactions, by monitoring fluorescence from a redox-active cofactor or the formation of fluorescent products. Enzymes nanowired to carbon nanotube tips may enable environmental control of biological stimulus-response with high spatial resolution, including product-induced signal transduction.

4:45 PM W8.8

Regular, low density cellular structures: rapid prototyping, numerical simulation, mechanical testing. Jürgen Stampfl1, Arithth Pisapán1, Martin M. Seyer1, Mathias H. Luxner1, Heinz E. Petermann1, Alexander Woess2 and Peter Fratzl2; 1Inst. of Materials Science and Testing, Vienna University of Technology, Vienna, Austria; 2Max Planck Institute for Colloids and Interfaces, Potsdam, Germany; 3Inst. of Lightweight Materials and Structures, Vienna University of Technology, Vienna, Austria.

Cellular solids form the basis of many biological and engineering structures. Most models use the apparent density and the mechanical properties of the bulk material as the main parameter for the prediction of the mechanical properties of such structures. In this work the influence of the architecture of periodic cellular solids with identical apparent density is investigated numerically and experimentally. Using computer aided design techniques, structures with 8x8x8 base cells are designed and fabricated. The physical prototypes which are tested experimentally are made from thermosetting polymers in order to avoid collapse under compression load. Various RP techniques are compared with respect to their suitability for the fabrication of cellular materials. The used technique must be able to shape these structures with sufficient accuracy in order to maintain the variation of the geometrical and mechanical properties within one batch of structures with identical architecture. After analyzing the elastic and visco-elastic properties of the bulk material, the fabricated structures are tested using a Hysitron Triboindenter apparatus and the surface modulus and hardness values of the composite materials are evaluated. The strain on the material is held constant, while the reduction in stress is measured as function of time as the material relaxes and micro and nanostructural rearrangements dissipate energy. In creep experiments, constant load was maintained on the sample, while the material was allowed to continue displacing underneath the load. Data from these experiments yielded characteristic time constants that will be used to construct viscoelastic material models of the composite mineralized gel system.

9:00 AM W9.0/5.2

Fabrication and Evaluation of Uniformly Sized Nanoporous Alumina for Human Osteoblast Cell Culture. Eric Leary Swan, Ketul Popat and Tejal A Desai; Biomedical Engineering, Boston University, Boston, Massachusetts.

Bone tissue engineering requires the ability to regulate cell behavior through precise control over substrate topography and surface chemistry. Aluminum oxide, or alumina, has been extensively employed as a substrate for bone cell seeding in dental and orthopedic implant applications. However, the current techniques do not allow...
precise surface topography and orientation of the porous material. A new method of producing alumina has been developed to improve osteoblast adhesion and proliferation. A two-step micromolding process has been optimized for fabrication of hexagonally arrayed nanoporous alumina membranes. This method allows for the formation of uniformly sized pores in the range of 30 to 80 nm diameter determined by anodization voltage. Nanoporous membranes display uniform pore density and pore size, which is suggested by scanning electron microscopy (SEM). From this process, a pure, uniform alumina membrane with through holes and specific control of nanostructure was produced. In order to test the compatibility of this porous scaffold with osteoblast adhesion, proliferation, morphology, and matrix production were tracked for various pore sizes and compared to amorphous aluminum oxide. Growth and adhesion results were evaluated by cell counting and microscropic imaging, while matrix production was quantified by enzymatic assays. Also, alumina surfaces were modified by cell adhesion peptides, and osteoblast growth was compared to the unmodified membranes. Nanoporous alumina can be produced with highly defined pores of constant size and density and provides a stable platform for osteoblast culture that is easily tailored to optimize growth and function. The alumina membranes show promise for employment as a substrate for dental or orthopaedic implants.

9:15 AM W9.3/05.3
Use of Soft Lithography for Multi-layer MicroMolding (MMM) of 3-D PCL Scaffolds for Tissue Engineering. Yang Sun, Nicholas Ferrell and Derek Hansford, Biomedical Engineering Center, The Ohio State University, Columbus, Ohio.

It is desirable that 3-D scaffolds for tissue engineering have precisely controlled geometries due to their improvement of cellular adhesion and functionality. Surface features smaller (~100 μm) than the typical cell dimensions have been shown to have significant effects on cell behavior and cell-surface interactions. In this paper, a soft lithography technique was used to fabricate PDMS stamps of repetitive groove and grid patterns with feature sizes of 5 μm width, 5μm depth, and 45μm wide spaces. Several methods were compared for the fabrication of 3-D multi-layer polycaprolactone (PCL) scaffolds with these patterns. Spin coating and oxygen plasma were combined to build 3-D scaffolds with PDMS stamps of the groove pattern. The resultant scaffolds had good alignment and connection between layers; however the upper layer collapsed due to the poor mechanical stability. Second, the micromolding in capillaries (MIMIC) technique was used to deliver the polymer into the small grooves by capillarity; however the resultant lines were discontinuous and not able to form layers. Finally, a new multi-layer micromolding (MMM) method was developed and successfully applied in a grid pattern to fabricate 3-D scaffolds. Proper heating and stamping parameters were identified that allowed the successful demonstration of the process on the thermoplastic PCL polymer. Scanning electron microscopy (SEM) characterization showed that the multi-layered scaffolds had high porosity and precisely controlled 3-D structures. Initial cell seeding experiments showed that the micropatterned scaffolds enhanced cellular attachment and proliferation, and encouraged cellular growth into the scaffold structure.

9:30 AM W9.4/05.4
Design and Fabrication of a Constant Shear Microfluidic Network for Tissue Engineering. Jeffrey T Borenstein1, Mohammad H Zargarotoori H1,2, David G Yoon1,2, Eli T Weinberg1,2, and Joseph P. Vacanti1,2,3,4, 1Biomedical Engineering Center, Draper Laboratory, Cambridge, Massachusetts; 2Department of Mechanical Engineering, Massachusetts Institute of Technology, Cambridge, Massachusetts; 3Department of Pediatric Surgery, Massachusetts General Hospital, Boston, Massachusetts; 4Department of Surgery, Harvard Medical School, Boston, Massachusetts.

Recent progress in microfabrication of biodegradable materials has resulted in the development of a three-dimensional construct suitable for use as a scaffold for engineering blood vessel networks. These networks are designed to replicate the critical fluid dynamic properties of physiological systems such as the microcirculation within a vital organ. Ultimately, these 3D microvascular constructs will serve as a framework for population with organ-specific cells for applications in organ assist and organ replacement. This approach for tissue engineering highly engineered designs and microfabrication technology to assemble cells in three-dimensional constructs that have physiological values for properties such as mechanical strength, oxygen, nutrient and waste transport, and fluidic parameters. This work focuses on the three-dimensional network with appropriate values for blood flow velocity, pressure drop and hemorheology distribution have been designed and fabricated using replica molding techniques, and populated with endothelial cells for long-term cell culture. The fluidic characteristics of these models are the shear stress exerted by blood flow on the endothelial cells lining the walls of the vessel; a key parameter that is known to initiate a cascade of mechanotransduction phenomena whereby mechanical shear forces yield biological responses that govern the cellular function. In this work, we report the design and construction of three-dimensional microfluidic constructs for tissue engineering that have uniform wall shear stress throughout the network. This type of control over the shear stress offers several advantages over earlier approaches, including more uniform and reversible, more rapid achievement of confluent coatings, and better control over endothelial cell behavior for in vitro and in vivo studies.

10:15 AM *W9.5/05.5
Nanobioreactors for Microfluidic Cell Engineering at Nanofabricated Scales. Alc Prokop1,2, Zdenka Prokop1, David Keifer Schaffer1, Eugene Kozlov2, John P Wilkso4, David E Cliffe1 and Franz J Baadenhaver3,1, 1NanoDelivery, Inc, Nashville, Tennessee; 2Chemical Engineering, Vanderbilt University, Nashville, Tennessee; 3Mechanical Engineering, Vanderbilt University, Nashville, Tennessee; 4Physics and Biomedical Engineering, Vanderbilt University, Nashville, Tennessee; 4Chemistry, Vanderbilt University, Nashville, Tennessee; 5Biomedical Engineering, Vanderbilt University, Nashville, Tennessee.

Micronanotextured cell-culture environments, i.e., Nanobioreactors (NBRs) for growing and maintaining populations of few cultured mammalian cells in volumes three orders of magnitude smaller than in standard environments would lead to major advances in a number of areas. The small NBR volume would reduce the time required for disease models, enable parallel monitoring of the environment of multiple cells. Closed-loop adjustments of the environment, e.g., pH and ionic concentrations, could be added to maintain homeostasis. For a noninvasive monitoring of metabolic activity and adhesion peptides, and osteoblast growth was compared to the unmodified membranes. Nanoporous alumina can be produced with highly defined pores of constant size and density and provides a stable platform for osteoblast culture that is easily tailored to optimize growth and function. The alumina membranes show promise for employment as a substrate for dental or orthopaedic implants.

Use of Soft Lithography for Multi-layer MicroMolding (MMM) of 3-D PCL Scaffolds for Tissue Engineering. Yang Sun, Nicholas Ferrell and Derek Hansford, Biomedical Engineering Center, The Ohio State University, Columbus, Ohio.

It is desirable that 3-D scaffolds for tissue engineering have precisely controlled geometries due to their improvement of cellular adhesion and functionality. Surface features smaller (~100 μm) than the typical cell dimensions have been shown to have significant effects on cell behavior and cell-surface interactions. In this paper, a soft lithography technique was used to fabricate PDMS stamps of repetitive groove and grid patterns with feature sizes of 5 μm width, 5μm depth, and 45μm wide spaces. Several methods were compared for the fabrication of 3-D multi-layer polycaprolactone (PCL) scaffolds with these patterns. Spin coating and oxygen plasma were combined to build 3-D scaffolds with PDMS stamps of the groove pattern. The resultant scaffolds had good alignment and connection between layers; however the upper layer collapsed due to the poor mechanical stability. Second, the micromolding in capillaries (MIMIC) technique was used to deliver the polymer into the small grooves by capillarity; however the resultant lines were discontinuous and not able to form layers. Finally, a new multi-layer micromolding (MMM) method was developed and successfully applied in a grid pattern to fabricate 3-D scaffolds. Proper heating and stamping parameters were identified that allowed the successful demonstration of the process on the thermoplastic PCL polymer. Scanning electron microscopy (SEM) characterization showed that the multi-layered scaffolds had high porosity and precisely controlled 3-D structures. Initial cell seeding experiments showed that the micropatterned scaffolds enhanced cellular attachment and proliferation, and encouraged cellular growth into the scaffold structure.

Design and Fabrication of a Constant Shear Microfluidic Network for Tissue Engineering. Jeffrey T Borenstein1, Mohammad H Zargarotoori H1,2, David G Yoon1,2, Eli T Weinberg1,2, and Joseph P. Vacanti1,2,3,4, 1Biomedical Engineering Center, Draper Laboratory, Cambridge, Massachusetts; 2Department of Mechanical Engineering, Massachusetts Institute of Technology, Cambridge, Massachusetts; 3Department of Pediatric Surgery, Massachusetts General Hospital, Boston, Massachusetts; 4Department of Surgery, Harvard Medical School, Boston, Massachusetts.

Recent progress in microfabrication of biodegradable materials has resulted in the development of a three-dimensional construct suitable for use as a scaffold for engineering blood vessel networks. These networks are designed to replicate the critical fluid dynamic properties of physiological systems such as the microcirculation within a vital organ. Ultimately, these 3D microvascular constructs will serve as a framework for population with organ-specific cells for applications in organ assist and organ replacement. This approach for tissue engineering highly engineered designs and microfabrication technology to assemble cells in three-dimensional constructs that have physiological values for properties such as mechanical strength, oxygen, nutrient and waste transport, and fluidic parameters. This work focuses on the three-dimensional network with appropriate values for blood flow velocity, pressure drop and hemorheology distribution have been designed and fabricated using replica molding techniques, and populated with endothelial cells for long-term cell culture. The fluidic characteristics of these models are the shear stress exerted by blood flow on the endothelial cells lining the walls of the vessel; a key parameter that is known to initiate a cascade of mechanotransduction phenomena whereby mechanical shear forces yield biological responses that govern the cellular function. In this work, we report the design and construction of three-dimensional microfluidic constructs for tissue engineering that have uniform wall shear stress throughout the network. This type of control over the shear stress offers several advantages over earlier approaches, including more uniform and reversible, more rapid achievement of confluent coatings, and better control over endothelial cell behavior for in vitro and in vivo studies.

Nanobioreactors for Microfluidic Cell Engineering at Nanofabricated Scales. Alc Prokop1,2, Zdenka Prokop1, David Keifer Schaffer1, Eugene Kozlov2, John P Wilkso4, David E Cliffe1 and Franz J Baadenhaver3,1, 1NanoDelivery, Inc, Nashville, Tennessee; 2Chemical Engineering, Vanderbilt University, Nashville, Tennessee; 3Mechanical Engineering, Vanderbilt University, Nashville, Tennessee; 4Physics and Biomedical Engineering, Vanderbilt University, Nashville, Tennessee; 4Chemistry, Vanderbilt University, Nashville, Tennessee; 5Biomedical Engineering, Vanderbilt University, Nashville, Tennessee.

Micronanotextured cell-culture environments, i.e., Nanobioreactors (NBRs) for growing and maintaining populations of few cultured mammalian cells in volumes three orders of magnitude smaller than in standard environments would lead to major advances in a number of areas. The small NBR volume would reduce the time required for disease models, enable parallel monitoring of the environment of multiple cells. Closed-loop adjustments of the environment, e.g., pH and ionic concentrations, could be added to maintain homeostasis. For a noninvasive monitoring of metabolic activity and adhesion peptides, and osteoblast growth was compared to the unmodified membranes. Nanoporous alumina can be produced with highly defined pores of constant size and density and provides a stable platform for osteoblast culture that is easily tailored to optimize growth and function. The alumina membranes show promise for employment as a substrate for dental or orthopaedic implants.
11:00 AM W0.7/05.7
Challenges in Engineering and Biologically-Inspired Hydrogel ECMs for Tissue Engineering. Kevin E. Healy, Materials Science & Engineering, Bioengineering, University of California at Berkeley, Berkeley, California.

A critical problem limiting the field of tissue engineering is the lack of engineering design rules to guide the synthesis and fabrication of artificial extracellular matrices (ECMs) or scaffolds. To address this issue, we have created artificial ECMs that are environmentally responsive and may be sensitive to several factors, including pH, temperature, proteins, and mechanical forces. Our current approach is to create modular hydrogel ECMs where different properties of the matrix can be manipulated independently, thus creating a system where parametric analysis of the effect of hydrogel properties on cell proliferation and differentiation is possible. For example, we have synthesized and characterized the physical properties of semi-interpenetrating polymer networks (sIPNs) consisting of a thermoresponsive N-isopropylacrylamide-co-acrylic acid network and an thermo-responsive N-isopropylacrylamide-co-acrylic acid network [p(NIPAam-co-AAc)]. To impart biomimetic character into the hydrogels, the AA groups on the linear chains have been functionalized with peptides containing the RGD and other sequences. The system allows for easy synthesis of admixtures of peptide sequences while maintaining the mechanical properties of the matrix. Therefore, studies addressing the effect of ligand type and density, in the context of matrices with various mechanical properties, can be easily performed. These peptide-modified p(NIPAam-co-AAc) hydrogels with protease degradable crosslinks serve as useful tools for studying cell-material interactions within three-dimensional structures and have the potential for use in tissue engineering applications. In addition, the synthetic strategy we have employed allows for easy control of mechanical and chemical properties of the matrix allowing parametric analysis of the effect of these properties on tissue development both in vitro and in vivo.

11:30 AM W0.8/05.8
A Novel System for Self-Assembly of Muscle-MEMS Devices. Jianzhong (Jeff) Xi, Jacob Schmidt and Carlo Montemagno, Bioengineering, UCLA, Los Angeles, California.

As microcomponents in engineered systems, biological muscles have unique advantages such as large force transduction, utilization of bioelectrical fuel source, and single cell, cells, over non-biologic actuators for biomedical engineering applications. Successful self-assembly of muscles with inorganic fabricated structures and electronics promises the capability of precisely characterizing muscles’ mechanical properties and fabrication self-assembled extracellular autonomous structures powered by ubiquitous glucose. However, the use of extracted muscle tissue from animals on these devices is impractical and inefficient, as the tissues must be dissected and incorporated into each device by hand with crude interfaces between the biological tissue and inorganic materials. Integration of muscle with fabricated structures would be optimally achieved through self-assembling muscle cells on MEMS. The construction of self-assembled muscle-MEMS structures is not limited by the stringent requirement to spatially direct the cell growth, control the tight connection of these differentiated structures with MEMS structures, and enable the cells and the integrated hybrid to be free to move. Soft photolithography techniques are extensively employed to pattern the growth of a variety of cell types and investigate their interaction with substrate in the micrometer level. However, these techniques are only suitable for patterning static cells on a surface, so a novel system of spatially patterning the contractile cells must be developed to enable the cells and the integrated hybrid to move. Here we present a novel system of self-assembling myocytes on MEMS devices. This system has shown its capability of spatially and selectively directed growth and differentiation of myocytes into single muscle bundles in situ, attachment of these functional bundles to MEMS structures, and the controlled partial release of the resultant hybrid devices. A novel force transducer capable of in situ characterization of the mechanical properties of muscle at both tissue and single-cell levels has been fabricated using this system. The mechanical properties of neonatal ventricular myocytes 1-3-day-old Sprague-Dawley rats (NRVMs), such as substrate-induced stress (2-5 kPa) and Young’s modulus (40 kPa), have been measured using this force transducer. This force transducer has also been used to perform dynamic studies of myocytes. Mechanical and dynamic characterization of healthy muscle cells will contribute to better understanding of cardiac tissue physiology and further engineering of functional cardiac tissue constructs. Our force transducer has shown the ability to achieve this goal. Furthermore, using this system, we have also created the first self-assembled muscle-powered microbots. The studies of the characteristics of these microbots will be also reported.

11:45 AM W0.9/05.10
Biobionic Processing of a Biodegradable, Segmented-Polyurethane for Use in Tissue Engineering Devices. Danielle N. Rockwood1, Jean S Stephens2, John F Rabolt1, Kimberly Woodhouse3 and Joanna Promstein2, 1Materials Science and Engineering, University of Delaware, Newark, Delaware; 2Chemical Engineering and Applied Chemistry, University of Toronto, Toronto, Ontario, Canada.

A segmented-polyurethane has been synthesized using an amino acid-derived diisocyanate and a phenylalanine-based chain extender. Contrary to many polyurethanes used for tissue engineering applications, this polymer is biodegradable and should prove to be biocompatible. In addition, the segmented nature of the polyurethane allows for elastomeric behavior thus providing the mechanical properties required to respond to physiological stresses. Compared with these advantageous physical properties, the chemical architecture of this polymer unites the necessary functional components (e.g. hydrolyzable groups to promote in vivo degradation) to satisfy the many of the desirable requirements for a tissue engineering construct. The goal of many tissue engineering devices is to closely mimic natural systems. In the case of tissue constructs, the extracellular matrix (ECM) contains protein fibers that range in diameter from a few microns to nanometer scale. In order to mimic the ECM architecture, electrospinning has been used to create membranes of nanometer scale polyurethane fibers. The nature of the electrospinning process is such that a range of fiber diameters and surface morphologies can be produced depending on the choice of processing protocols. In addition, Raman spectroscopy has been used to ensure the conformational integrity of the polymer before and after processing. Our overall goal is to seed cardiomyocyte cells on these electrospun membranes and hope that these cells will autonomously synthesize their own proteins. Over time, the polyurethane construct will be bioresorbed and the cells will create their own ECM. I. S. Megolski, J. Stephens, D. B. Chase and J. F. Rabolt, Macromolecules 35, 8456 (2002)
has been used to increase the rate of catalysis as well. These studies enabled the synthesis of self-assembling "biomimetics" that incorporate the biological sequences identified for catalysis, yielding new structure-directing catalysis of polymerization. The silicatins and their biomimetic counterparts catalyze structure-directing synthesis from a wide range of precursors, yielding inorganic silica (organosilicon, organosilica, silica) from silicic acid solutions in vitro and also control the following the addition of silicic acid. Remarkably, the precipitate structures are the intricately shaped, silicified cell walls of diatoms, enabling the synthesis of semiconductors. Interaction with the template-like silaffin-IA and LCPA. When natSil-2 and natSil-lA (or LCPA) are silicateins also catalyze and structurally direct the hydrolysis and silicateins and their biomimetic counterparts catalyze nanostructure-directed synthesis are under investigation for optical and electronic applications, sensors, energy transducers, cosmetics and pharmaceuticals.

2:00 PM W10.2
The Biochemistry of Silica Nanofabrication in Diatoms.
Nils Kroeger, Nicole Poulsen, Manfred Samper and Rainer Denthann; Biochime I, University of Regensburg, Regensburg, Germany.

The biological formation of inorganic materials with complex form (biominerals) is a widespread phenomenon in nature, yet the molecular mechanisms underlying biomineral morphogenesis are not well understood. Among the most fascinating examples of biomineral structures are the intricately shaped, silica-filled cell walls of diatoms, which exhibit species-specific nanopatterns (see figure). Bioinspired nanofabrication of silica is an extremely rapid process that is accomplished under mild physiological conditions, thus exceeding the capabilities of present-day materials engineering. Elucidating the molecular mechanisms of silica nanofabrication in diatoms is therefore expected to open up new synthetic routes to nanostructured silica materials. In search of the biological molecules that control silica nanomorphogenesis, a thorough biochemical analysis of the diatom biosilica-associated organic components was undertaken. This led to the discovery of novel organic molecules termed silaffins and long-chain polyamines (LCPA), which are able to drastically influence silica formation. Silaffins are complex proteins/peptides that are heavily posttranslationally modified. These modifications include phosphoamino acids and lysine residues carrying oligo-N-methyl-propyleneimine chains, which represents a novel type of amino acid modification. LCPA contain even longer chains of N-methyl-propyleneimine and lysine residues linked to putrescine residues via a long-chain hydrophobic peptide backbone. Native silaffin-1A (natSil-1A) and LCPA, respectively, not only highly accelerate silica formation from a silicate solution in vitro but also control the structure of the silica formed, generating nanostructured silica nanoparticles. Spherical particles of 10-100 nm in diameter have indeed been shown to constitute diatom biosilica, yet natSil-1A and LCPA are unable to create the porous nanopatterns that are typical for diatom biosilicas, suggesting that additional components are required. Recently, a novel silaffin molecule termed natSil-2 was discovered that lacks an intrinsic silica formation activity, but is able to modulate the activities of silaffin-1A and LCPA. When natSil-2 and natSil-1A (or LCPA) are combined, porous silica structures are precipitated within minutes following the addition of silicate. Remarkably, the precipitate displays pore sizes in the range 100-1000 nm, which is characteristic for diatom biosilicas. Biophysical measurements indicate that silaffin natSil-2 is incorporated into the silica formation in this way and the system is mediated by a self-assembled organic matrix of silaffins.

2:15 PM W10.3
Biological Aspects of Periodic Mesoporous Organosilicas.
Kai Landskron 1, Benjamin D Hatton 1,2, Doug D Perovic 2 and Geoffrey A Ozin 1; 1: Department of Chemistry, University of Toronto, Toronto, Ontario, Canada; 2: Department of Materials Science and Engineering, University of Toronto, Toronto, Ontario, Canada.

Organic-inorganic nanocomposites play a significant role in biological systems, e.g. in bones, teeth and shells. Periodic mesoporous organosilicas (PMOs) are synthesized by templating and integrating inorganic and organic components on the nanometer length scale and hence can be seen as biomimetic materials. Herein, we introduce a new PMO consisting of interconnected [Si(CH3)]3 rings of SiO2(CH2)2 tetrasil氧bridging units. This PMO can be functionalized with polymeric or oriented film morphologies.

3:00 PM W10.4
Shape, Size and Morphology Control of Inorganic Crystals with Self-Assembled Monolayers.
Yong-Jin Han and Joanna Aizenberg; Bell Labs, Murray Hill, New Jersey.

Self-assembled monolayers (SAMs) provide simple, yet sophisticated surfaces to mimic highly ordered organic surfaces used in the process of biominalization. A careful selection of organic molecules with an appropriate surface (i.e., H3OCH(OH)(CH3)2COOH) nucleation and growth of oriented calcite crystals and provide opportunities to study the formation of inorganic crystals assisted by organic molecules. We have elucidated the process of calcite crystal formation on SAMs while studying interactions between groups of calcite crystals and their final shapes, sizes and morphologies. We have concluded that the orientation of crystals plays a critical role in controlling their final shape, size and morphology by manipulating the ability of a crystal to interact with additives such as proteins and ions during nucleation and growth. In this presentation, we report our experimental results demonstrating how underlying organic molecules control and mold the final shape, size and morphology of biominerals from their initial nucleation during biominalization.

3:30 PM W10.5
Biomimetic Surfaces Mimicking Fibronectin Matrix Assembly.
Jason R. Capadona1, David M Collard1 and Andres J Garcia1; 1: School of Chemistry and Biochemistry, Georgia Institute of Technology, Atlanta, Georgia; 2: Woodruff School of Mechanical Engineering, George Institute of Technology, Atlanta, Georgia; 3: Petit Institute for Bioengineering and Bioscience, Georgia Institute of Technology, Atlanta, Georgia.

Cell adhesion to synthetic materials is vital to numerous biotechnological and biomedical applications, including biomaterials, tissue engineering, and in vitro culture substrates. Cell-material interactions are mediated by specific binding of cellular receptors to adsorbed proteins or engineered motifs on the surface of the material. We have previously shown that the quantity, structure, and function of adsorbed fibronectin (FN) are dynamically influenced by the orientation of FN polymorphs of these materials (e.g., anatase and rutile) is controlled by the underlying substrate. In an effort to increase integrin-mediated cell adhesion to synthetic surfaces, numerous groups have incorporated bio-recognition sites onto the material's surface, such as the RGD pentapeptide. Although these strategies do increase cell adhesion to synthetic materials, the biological activity of these short peptides is significantly lower than that of the complete protein. A critical characteristic of extracellular matrices is the assembly of supramolecular structures. For example, FN matrix assembly modulates cell cycle progression and differentiation. Using mixed self-assembled monolayers (SAMs) of alkanethiols on gold, we created a model nonfouling surface that can selectively tether peptides at a controlled density. We have engineered a biomimetic surface that incorporates a short peptide (FN-13) that has been shown to induce FN matrix assembly in several cell lines when added to tissue media. SAMs presenting this peptide tethered FN to a controlled density. We have concluded that the peptide sequence FN-13 is a biomimetic surface that promotes FN matrix assembly in an oncostat-like cell line. This biomimetic surface provides a promising strategy to the engineering of biomimetic supports.

3:45 PM W10.6
Semiconductor Surfaces with Nanometer Features Composed of TAT Peptides.
Youngman Cho and Albena Ivanisevic; Purdue University, West Lafayette, Indiana.

Atomic force microscopy (AFM) was used to fabricate well-defined peptide templates onto silicon and porous silicon surfaces via Dip-Pen Nanolithography (DPN). DPN is a powerful technique to write specific organic and/or inorganic molecules onto a surface with an AFM tip. In this work, DPN was used to construct arrays of peptides with nanometer features. Prior to depositing a thin peptide modification procedure was carried out. Clean surfaces were silanized to terminate them on amino groups, and subsequently reacted with a heterobifunctional cross-linker. TAT peptides (e.g. CGS6G3KRRGQRR) which exhibit rapid uptake in cells, were
Biological factors that influence calcium oxalate crystallization in plant cells. Mary Alice Webb, Botany and Plant Pathology, Purdue University, West Lafayette, Indiana.

Many plants accumulate crystals composed of calcium oxalate. These crystals occur in different sizes, shapes, and distributions that characterize particular groups of plants. Differing crystal shapes often reflect specific functions within plants. Calcium oxalate crystals can serve as varieties of functions, including storage of calcium, structural reinforcement, and protection against pests. Synthesis of these crystals by plants is a regulated biological process, similar to other types of biomineralization such as bone and tooth formation in animals. However, it is not known how plants control crystal growth to fashion different shapes of crystals from calcium oxalate. We are interested in identifying and characterizing biological factors that influence synthesis of calcium oxalate crystals in plants. We are studying crystal formation in grape plants, which produce unique needle-shaped crystals of calcium oxalate in a highly specialized morphology, termed a raphide. In grape we have shown that raphides develop along with specialized cells within compartments formed by biological membranes. Each of these cells, as it develops, constructs a highly organized bundle consisting of several hundred raphides. Raphide-forming cells are distributed throughout the plant, and the same crystal morphology and bundle organization are duplicated over and over again in each specialized cell. Our research has focused on identifying cellular factors influencing the fabrication of raphides and organization of raphide bundles within cells. We are interested in identifying genes that encode proteins, or genes involved in this process. We have used two different approaches to identify candidate proteins that may mediate raphide development. In one approach we isolated raphides from grape leaves and generated antibodies that recognized raphide-associated proteins (RAPs). We then used the antibodies to clone grape genes putatively encoding these RAPs. Using this approach we have identified and characterized a protein potentially involved in regulating crystal formation, as well as a molecular motor that may participate with other cellular structures in moving and organizing raphides within developing cells. Studies of these proteins and their putative functions are in progress. We are currently developing a second approach specifically targeting discovery of cellular factors that may control that crystal growth. With this approach we can assay a complex mixture of proteins and detect specific proteins within the mixture that may be active in modifying crystal growth. We expect that the same approach can be applied to identify functionally active proteins in a wide range of other mineralizing tissues.

4:30 PM **W10.8**

Modulation of Calcium Oxalate Crystallization by Biological Molecules. Roger Quiño,1 Chris Orme,1 John Hoyer,2 George Nancollas3, Salvador Zepeda4,1, Ana-Cindy5 and James De Yoreo1.1Chemistry and Materials Science, LLNL, University of California, Livermore, California; 2The Children’s Hospital of Philadelphia, University of Pennsylvania, Philadelphia, Pennsylvania; 3Chemistry Department, University at Buffalo, Amherst, New York; 4Department of Chemical Engineering and Materials Science, University of California, Davis, California; 5Department of Geological and Atmospheric Sciences, Iowa State University, Ames, Iowa.

Biological control of calcium oxalate crystallization is a crucial aspect of urolithiasis since the majority of human kidney stones are primarily composed of calcium oxalate monohydrate (COM) crystals. The mechanisms responsible for modification of COM by biological molecules normally present in urine have not been previously defined at the molecular level. In a report herein we present the first molecular-scale view of COM modulation by the aspartic acid-rich protein, osteopontin (OPN) and two synthetic 27 amino acid peptides, containing 21 aspartic acids and either serine or glycine spacers. Our atomic force microscopy studies showed that crystal habit and growth kinetics are controlled through anisotropic step pinning on crystal faces. However, the protein, OPN, modulated growth by its effects on steps only on the (010) face, while the peptides controlled growth on steps of both the (-101) and (010) face. The peptide with serine spacers showed much stronger growth modification than the peptide with glycine spacers. These findings were supported by our kinetic studies using the constant composition method that showed comparable relative potencies. In this presentation, the implication of these findings to insights concerning biological control of stone disease will be discussed.

4:45 PM **W10.9**

Probing Crystal Growth and Adhesion of Calcium Oxalate Crystal Surfaces: Toward an Understanding of Kidney Stone Formation. Michael David Ward, Jeffrey A. Wesson, Xiao Xia Sheng and Taesung Jung. Chemical Engineering and Materials Science, University of Minnesota, Minneapolis, Minnesota; Nephrology Division, Department of Veteran Affairs Medical Center and the Medical College of Wisconsin, Milwaukee, WI.

Kidney stones are biomineralized crystal aggregates, most commonly containing calcium oxalate monohydrate (COM) crystals as the primary constituent. Notably, in vitro studies have suggested that anionic molecules or macromolecules with substantial anionic functionality (e.g., carboxylate) play an important role in crystal aggregation and crystal attachment to renal epithelial cells. In an effort to elucidate these in vivo processes and the role of crystal surface structure and macromolecules in the regulation of kidney stone disease, in situ atomic force microscopy (AFM) has been used to obtain real-time in situ images of crystal growth on various different faces of calcium oxalate crystals. This approach allows direct visualization of crystal surface morphology, which can be interpreted in terms of the bulk crystal structure based on the unambiguous assignment of the growth features (e.g., steps, terraces) that are accessible with AFM. For example, dynamic AFM imaging reveals dramatically different morphologies for the (100), (010), and (12-1), and real time acquisition provides characterization of the growth modes and rates of growth along different crystallographic directions. These measurements also provide in situ determination of the influence of solute concentration and molecular or macromolecular additives on those rates. We also have used AFM to measure directly the adhesion forces between tip-immobilized molecules and the calcium oxalate surfaces in aqueous media. These measurements reveal the role of different functional groups on adhesion and support an important role for the carboxylate group in processes responsible for kidney stone formation. Furthermore, the adhesion forces depend upon the crystal face (e.g., COM [010]), which may be involved in the process. These force measurements can be performed in the presence of soluble macromolecules, including native urinary macromolecules (e.g., osteopontin), producing profiles of adhesion suppression/enhancement and suggesting a feasible methodology for identifying the most important crystal surface-macromolecule combinations related to stone formation. Building on recent reports from our group [1,2], this presentation will illustrate that the combination of dynamic imaging of crystal growth on various calcium oxalate crystal faces and direct force measurements on those faces can lead to insights that are expected to produce a comprehensive structural model for the interaction of biomolecules with calcium oxalate crystals. This model can be used to understand the role of atomic forces in the formation and dissolution of kidney stones.

**SESSION W11: Poster Session II - Biosensors and Tissue Engineering**

Chairs: William J. Landis, Christine Orme and Razi Wang

Thursday Evening, April 15, 2004

8:00 PM

Salons 8-8 (Marriott)

**W11.1**

Hierarchical order in PEG-peptide Block Copolymers Containing Bioinert Molecules. Ian W. Hamley, Valeria Castellote1, Oleksandr 0. Mykhaylyk2, Harm-Anton Klok2 and Annette Roesler3.1Chemistry, University of Leeds, Leeds, United Kingdom; 2Institut des Matériaux, EFPL, Lausanne, Switzerland; 3Max Planck Institute for Polymer Research, Mainz, Germany.

Nature has evolved ways to create exquisite nanostructures using biopolymers in a range of structural and functional materials. Mannkind now wishes to exploit such optimized architectures in normally designed nanomaterials and peptides in increasingly emerging attention as building blocks for design of self-assembled structures. The ultimate goal is the engineering of nanostructure via combinations of these biologically-derived or biologically-inspired
materials with appropriate synthetic polymer components. This talk will be concerned with nanostructured block copolymers in which the recognition properties and responsiveness of peptide sequences are modulated by incorporation of poly(ethylene glycol) (PEG) blocks in diblock and triblock architectures. Characterization of nanostructures in solutions and in the solid state has been performed using small-angle X-ray scattering and electron microscopy. In solution, we find PEG-coated fibrils, which aggregate into tangles resembling those formed by β-amyloid peptides. In the solid state, we studied the influence of PEG crystallization on microphase-separated structures containing β-sheet peptide domains. By combining peptide and synthetic polymer blocks, unprecedented control over hierarchical order is demonstrated.

W11.2 In-situ Formation of Biphasic Calcium Phosphate Bioceramics from Goniopora Exoskeleton. Murugan Ramalingam 1, S. Ramakrishna 1 and T.S. Sampath Kumar 2, 1NUS Nanoscience & Nanotechnology Initiative and Division of Bioengineering, National University of Singapore, Singapore; 2Dept. Metallurgy & Materials Engineering, Indian Institute of Technology, Chennai, India.

Biomaterials derived from natural resources have recently been recognized as human health care substitutes. Calcium phosphate-based bioceramics, in particular hydroxyapatite (HA) and beta-tricalcium phosphate (beta-TCP) are widely practiced clinically as bone substitutes due to their chemical composition similarity with human calcified tissues. Recently, beta-Ca3(PO4)2-based calcium phosphate (BCP) bioceramics have been attracted for many biomedical applications owing to their controlled biodegradation, balancing of more stable phase of HA and more soluble phase of beta-TCP upon implantation. The BCP can be prepared from various calcium and phosphorous precursors in desired phases and shapes depending upon clinical requirements. In this study, we report the in-situ formation of BCP bioceramics from goniopora exoskeleton containing β-sheet domains in solutions and in the solid state has been performed using thermal, pH and ion stimuli. The solubility of BCP was conducted in Hanks medium at pH 7.4 under in-vitro physiological conditions and indicating that the solubility of BCP lays in between the resorption levels of HA and beta-TCP. This study explores the ways to utilize our natural marine resources into value added clinical materials in particular BCP, which can be used in osseous defect areas.

W11.3 Modulation of Reversible Color Switch of Polydiacetylene Supramolecules for Sensing Matrix. Dong June Ahn 1, Tai Young Kim 1, Sang Hoon Lee 1, Doo Ho Yang 1 and Jong-Man Kim 2, 1Department of Chemical & Biological Engineering, Korea University, Seoul, South Korea; 2Department of Chemical Engineering, Hangyang University, Seoul, South Korea.

Polydiacetylene-based supramolecules are interesting biomimetic materials in view of application to chemical and biological sensors. These supramolecules can be the unique in changing color from initial blue to red upon specific binding events, caused by shortening of delocalization length of π-electrons along diacetylenic backbones. Various binding events including viruses, toxins, glucose, and ionic interactions have been reported detectable. However, most of the polydiacetylene-based chemosensors reported to date function via hydrogen-bonding in color change should be useful for designing reversible colorimetric sensors. In this presentation, we report a novel technique to modulate the colorimetric reversibility of the polydiacetylene supramolecules. Weakening the hydrogen-bonding strength by ion binding to the supramolecular surfaces makes the assemblies work irreversibly. By contrast, successive enhancement of the hydrogen-bonding by ion desorption completely recovers their colorimetric reversibility. This modulation technique enables one to capture snap shots of the reversibly-working polydiacetylene supramolecular matrix potentially used for continuous monitoring.


Inspired by the water-repellent behavior of the micro- and nano-structured plant surfaces, superhydrophobic materials, with a water contact angle larger than 150 degree, have received a lot of research attentions recently. It has been suggested that contamination oxidation and current conduction can be inhibited on such superhydrophobic surfaces, and the flow resistance in the microfluidic channels can also be reduced using super water-repellent materials. However, to fully utilize the properties of nanoscale surface structures, it is necessary to investigate the relationship between the nanostructure and the water repellent behavior on surfaces, and to fabricate the nanostructured surfaces with desired surface hydrophobicity. We have designed and fabricated tunable superhydrophobic surfaces using a combination of nanosphere lithography and plasma etching. It has been found that the water contact angle on these surfaces can be systematically tuned from 135 to 170 degree by tuning the diameters of poly(ene-yne) nanospheres using oxygen plasma. The water contact angles measured on these surfaces can be modeled by the Cassie's formulation without any adjustable parameter.
Biometric growth of bone-like apatite on nano-fibrous scaffolds. Guoqiang Wang and Peter X. Ma; Biomedical Engineering, University of Michigan, Ann Arbor, Michigan.

One of the challenges for bone tissue engineering scaffolds is the ability to integrate with the host tissue. To achieve this, we incorporated nano-sized hydroxyapatite (nHAP), or/and coated a layer of bone-like apatite on the pore surfaces throughout the scaffolds in contact with physiological environment. Nano-fibrous polymer scaffolds and their nHAP composite scaffolds in a supra-fluid (SBF). Bone-like apatite crystal growth became detectable on and in between nano-fibrous network of the polymer scaffolds after 6 days of incubation. The deposited apatite particles reached a few hundred micrometers in diameter, without interference from inter-connected macro-pores were prepared by the combination of a phase separation technique and a porogen sphere leaching process. In vitro calcification of the scaffolds was investigated by incubating pre-fabricated nano-fibrous scaffolds in a sucrose and calcium chloride (CaCl2) solution. A twenty-two day incubation in SBF led to a uniform apatite layer formation, covering all inner pore wall surfaces, and a mass increase of about 50%. Interestingly, it seemed that the maximum particle size did not increase after the initial incubation period, but the number of particles increased with incubation time. The scaffolds maintained the interconnected macro-porous structure, which is important for cell migration and mass transport. Pre-incorporation of nHAP particles into polymer scaffolds (even at a low content of 10% wt/wt) induced significantly greater amounts of apatite formation as compared to pure polymer scaffolds. In addition, polymer/nHAP composite scaffolds eliminated the 6-day lag time for apatite deposition, which was observed with pure polymer scaffolds. The results indicated that nHAP incorporation promoted in vitro calcification, and therefore, may also have the ability to improve mineralized new bone tissue formation. The nHAP incorporated scaffolds not only increased the crystallinity of the deposited hydroxyapatite, but also enhanced protein adsorption capacity. The demonstrated bioactivity of nHAP, together with well-controlled macro and nano structures, makes the novel nano-composite scaffolds promising candidates for bone tissue engineering.

Block Copolymer-Based Biomembranes Functionalized with Energy Transduction Proteins. Dean He, Benjamin Chu and Carlo D. Montemagno; UCLA, Los Angeles, California.

Block copolymer-based membrane technology represents a versatile class of nanoscale materials in which biomolecules, such as membrane proteins, can be reconstituted. Among its many advantages over conventional lipid-based membrane systems, block copolymers can mimic natural cell biomembrane environments in a single chain, which enables large-area membrane fabrication using methods like Langmuir-Blodgett deposition, or spontaneous protein-functionalized nanoscale formation. Furthermore, the single chain composition of the copolymers can be easily tailored to yield a wide variety of properties that can be functionalized, for example, variations to the block lengths, chemical composition, and hydrophilic/hydrophobic properties can be incorporated into the synthesis scheme to yield a plethora of application-specific materials. The membraneforming polypeptide chain, Halobacterium Halobium, serves as a light-actuated proton pump that develops proton gradients towards the demonstration of coupled functionality with other membrane proteins to effect ATP production, or production of electricity. Using quantum dot-labeled, engineered protein constructs, we have demonstrated large-scale insertion of membrane proteins into block copolymer Langmuir-Blodgett films. Following the absorption of the hybrid protein/polymer membrane to the conductive fluoropolymer Nafion, and towards the fabrication of a multicomponent system capable of facilitating coupled protein functionality, we have demonstrated measurable pH changes based upon light-actuated proton pumping. Light-actuated activity across the protein functionalized membrane was found in a TMOS sol-gel matrix has also been observed using impedance spectroscopy. Initial data has suggested a significant pH change of up to 1.56 in a volume of 100 microliters and surface area of 0.317 square centimeters, a level that is capable of powering a number of electrochemical reactions. The C-length of the initiating core had little effect on degradation properties, the latter of which is desirable in load bearing applications. Surface erosion behavior in these polypeptides was verified by mass loss of pressed pellets as a function of time. The progression of the degradation front within the polymer pellets was analyzed using Scanning Electron Microscopy. The erosion behavior in these polymers was further confirmed by following the release of a water-soluble dye from microspheres. Atomic Force Microscopy tapping mode imaging was used to study the changes in the surface characteristics of thin films as a function of degradation. Three parameters were modified in our experiments: initiating core length; lactide/glycolide chain length; and the diacid linker used to connect lactide/glycolide segments. In general, in polymers with lactide chain lengths > 20 repeat units and molecular weights > 55K, the C-length of the initiating core had little effect on degradation kinetics. Surprisingly, for molecular weights 55K or lower, polymers with longer initiating cores degraded slower than polymers with shorter initiating cores. This could be explained by the observation that polymers with longer initiating cores also exhibited lower glass transition (Tg) temperatures. We observed that polymers with shorter lactide repeat units degraded faster than ones with longer lactide blocks even though the former had twice the molecular weight. This is consistent with a surface erosion mechanism as increasing the lactide chain length should increase hydrophobicity and hence reduces surface availability. The system appears that the surface erosion behavior is driven by a combination of hydrophobicity of the diacid linker and long-range order in the
prominently been an issue that is counterintuitive is that increasing the diacid linker length, which should increase hydrophilicity and reduce scaffold penetrability, results in a faster degradation. This suggests that in novel polyglycolyester degradation is strongly influenced by entropic considerations, which are driven by chain flexibility. Understanding the effects of changing the different components of the scaffold can help in designing scaffolds that precisely control degradation rate. Additionally, we will compare our findings to custom-designed surface eroding polymers for specific applications.

**W11.11**

Surface modified diamond field effect transistors for enzyme immobilized biosensors. Kwangsoon Song1, Hirofumi Kanazawa,2, Yusuke Nakamura,1, Syota Kawamura,1,2, Munenori Degawa,1,2, Hitoshi Umezawa,1,2 and Hiroshi Kawarada1,2.

Diamond meets the requirements of robust biosensing devices because of its wide potential window, chemical-physical stability and biocompatibility. Although research on chemical and biological modifications on diamond surfaces has begun, the sensing region and transducing region necessary for biosensing have not been realized. Here, we introduce enzyme biosensors using electrolyte-solution-gate field-effect transistors (SGFETs) made of polycrystalline diamonds as transducers. Diamond SGFETs are expected to have high sensitivity, because the channel can be directly exposed to electrolyte solution not using membrane or passivation layer on channel surface. To form the sensing sites on diamond SGFETs, glucose oxidase (GOD) was immobilized on the channel surface. GOD-sensitive SGFETs operate based on the biocatalyzed decomposition of glucose by GOD. From this biocatalyzed decomposition, the pH changes induced by a spontaneous hydrolysis of D-glucono-6-lactone to gluconic acid decreasing pH near GOD immobilized electrode surface can be accompanied with a SGFET threshold voltage of SGFETs immobilizing GOD on channel surface shifted by approximately 30 mV/decade and drain current changed by approximately 40 μA. Since the polycrystalline diamond wafers are easily obtained, diamond SGFETs may be suitable biosensors and transducers in microelectronics for integrated biosensing and signal processing.

**W11.12**

Directed Osteoblast Adhesion at Metal Particle Boundaries: Promises for Nanophase Metals. Thomas Jay Webster, Jeremiah U Ejiofor and Brian Ward; Biomedical Engineering, Purdue University, Lafayette, Indiana.

Increased functions of osteoblasts (bone-forming cells) have been demonstrated on nanophase compared to conventional ceramics (specifically, alumina, titania, and hydroxyapatite), polymers (such as poly-lactic-glycolic acid and polyurethanes), metal nanofibers, and composites thereof. Nanophase materials are materials that simulate dimensions of constituent components of bone since they possess particle sizes on the order of 100 nm. However, to date, interactions of osteoblasts on nanophase compared to conventional metals remain to be elucidated. For this reason, the objective of the present in vitro study was to design, fabricate, and evaluate osteoblast adhesion on nanophase compared to conventional metals (specifically, Ti, Ti6Al4V, and CoCrMo). Results of this study provided the first evidence of increased osteoblast adhesion on nanophase compared to conventional metals. Moreover, directed osteoblast adhesion was observed preferentially at metal particle boundaries. It is speculated that since more particle boundaries were created through the use of nanophase compared to conventional metals, increased osteoblast adhesion resulted. Results of increased subsequent functions (such as proliferation, deposition of calcium-containing mineral, etc.) on nanophase metals will also be presented. Thus, this in vitro study suggests that nanophase metals have intriguing properties to enhance the efficacy of orthopedic implants.

**W11.13**

Collagen-inspired nano-fibrous poly(L-lactic acid) scaffolds for bone tissue engineering created from reverse solid freeform fabrication. Yuwei Chen1,2,3,4 and Peter X. Ma1,2,3.

Biomedical Engineering, University of Michigan, Ann Arbor, Michigan; 1Biological and Materials Sciences, University of Michigan, Ann Arbor, Michigan; 2Macromolecular Science and Engineering Center, University of Michigan, Ann Arbor, Michigan.

Tissue engineering aims to resolve the problems of tissue and organ donor shortages. One common technique uses a biodegradable scaffold that acts as an extracellular matrix (ECM) to support tissue regeneration. Cells are incorporated into the porous scaffold, and the cell-scaffold composite is cultivated to promote the formation of tissue throughout entire matrix. Recently, research has focused on engineering the internal structure of scaffolds from the micrometer to the nanometer level. Also, it has been suggested that the nano-fibrous architecture and the high surface area/volume ratio provided by the nanofibrous proteins in the ECM consequently affects cell migration, proliferation, and differentiation. Thus, it is important that the scaffold imitates the cells’ natural ECM environment until the host cells can repopulate and reorganize a new matrix. Here, we use computer-aided design (CAD) fabrication method to create highly-controlled macroporous structures in nano-fibrous poly(L-lactic acid) (PLLA) scaffolds. By using a computer-aided design (CAD) program to create a negative template for scaffold, the three-dimensional (3-D) mold is created on a 3-D printer using a wax. After the template is printed, a solution of PLLA in tetrahydrofuran (THF) is cast into the mold, and is subsequently transferred to a freezer to rapidly induce thermal phase separation until gives the nano-fibrous morphology. After phase separation, the THF is exchanged with water, and the wax is leached out with organic solvents. Water is then exchanged with the solvents, and the sample is frozen and lyophilized to yield a 3-D nano-fibrous scaffold with a uniform fiber mesh throughout the entire matrix. Fiber diameters in these scaffolds are around 150 nm, similar to type I collagen, and the densities of the fiber meshes can be altered by changing the polymer concentration. Macroscopically, the 3-D printer allows feature sizes down to 250 microns, and allows the user to create features with various geometries. In these scaffolds, interconnected macroporous structures (> 100 microns) can be created to allow for a uniform cell distribution during the seeding process, and guide cell migration and overall tissue formation in three dimensions. Also, the ability to create the macroporous regions of the scaffold a priori allows for the fabrication of scaffolds with anisotropically porous structures which potentially could be useful in implants where different tissue densities (e.g. trabecular and cortical bone) are common. Preliminary results also indicate that MC3T3-E1 osteoblasts attach and proliferate on these scaffolds. By having the ability to control the macroporous architecture, internal porosity, and external shape of the scaffold, this 3D fabrication phase separation technique has great potential to design and create ideal scaffolds for bone tissue engineering.

**W11.14**

Kinetin of controlled apatite growth aided by denin matrix protein 1. Gen He,1,2,3,4, David Schultz5, David Cookson,3,4, Jianjun Hao6 and Anne George1,2,3,4,5.


Bone and dentin mineralization requires controlled mineral deposition and multi-scale self-assembly into hierarchically ordered biocomposites with unique mechanical properties. Body fluid is supersaturated with respect to calcium phosphate, however, mineral deposition is only restricted to mineralized tissues and only initiated in the gap region of the type I collagen fibrils during bone and dentin formation. Clinical investigations and transgenic animal models emphasize the critical roles of bone/dentin specific acidic matrix proteins in mineral nucleation, growth, and organization. Here we present comprehensive studies on the regulatory roles of a bone/dentin specific protein, namely dentin matrix protein 1 (DMP1), in the nucleation and growth of collagen/calcium phosphate particles in the absence of mineralization with 10 mM NaCl, 10 mM HEPES, 2.5 mM CaCl2, 1 mM KH2PO4, pH 7.4 at room temperature was studied by time-resolved synchrotron small-angle X-ray scattering (SAXS), atomic force microscopy, and high resolution transmission electron microscopy (HREM). It was found that polydispersed spheroidal calcium phosphate nanoclusters emerged from the solution underwent simultaneous aggregation and deposition as apatite crystals. However, the growth and aggregation of the calcium phosphate particles were inhibited in the presence of DMP1 and the particle size was restricted below 30 nm. With time, these particles were stabilized in the solution, indicated by a significantly stronger scattering intensity compared to that with the controls, which are buffers with no additional additive or with 10 mM concentration of serum albumin. AFM analysis demonstrated that DMP1 self-assembled calcium phosphate clusters into nanorods with uniform size at 20±10 nm. On the other hand, when immobilized on a glass surface, DMP1 actively entraps calcium phosphate clusters from solution. The nucleated amorphous calcium phosphate precipitates ripe and nanocrystals form. Subsequently, these expand and coalesce into microparticle crystals elongated on the c-axis direction. Characterization of the functional domains in DMP1 demonstrated that intermolecular assembly of acidic clusters into a beta-sheet template was essential for the observed mineral nucleation. In conclusion, the present study proposes that acidic proteins such as DMP1 plays a dynamic role in the initiation of mineralization, facilitating mineral precipitation from physiological buffer and transforming into apatite of definite size and shape by spatially and temporally regulated nucleation, orientation, and organization.
W11.15 Calcium phosphate crystal phase formation on surfaces of collagen films. Marcela Silva1, Julio Garcia，则1, and Antonio Sombra2.1 Departamento de Quimica Organica e Inorganica, UFC, Fortaleza, CE, Brazil; 2 Departamento de Fisica, UFC, Fortaleza, CE, Brazil; 3 Departamento de Bioquimica, UFC, Fortaleza, CE, Brazil.

Considerable efforts have been made in the last years to improve the biocompatibility of materials and devices used for biomedical applications. The interest is to tailor the surface properties in such a way that a favorable interaction of the surface modified material and biological systems is achieved. Examples are the reduction of the non-specific binding of blood proteins to surfaces which are important for the blood compatibility of materials, and the control of adhesion of living cells to solid inorganic substrates (e.g. surfaces of field effect transistors) through surface attached polymer monolayers. In the present study, we report the growth of calcium phosphate crystals on collagen film. The method consists of two steps. First, the films had been soaked in a Ca(OH)2 or CaCl2 aqueous solution solution and second, the film is soaked in a Na2HPO4 aqueous solution (pH 10). The physical and chemical characteristics of the composites were tested. IR spectroscopy, X-rays and SEM analysis showed the calcium phosphate crystal phase formation on film surfaces, which was attributed to the catalytic effect of collagen molecules and the process of nucleation and crystal growth from released Ca2+ ions. This result provides a guiding principle for obtaining apatite - organic polymer composite composites. This composite is expected to have an analogous structure to that of natural bone.

W11.16 Abstract Withdrawn

W11.17 Interaction Between Titanium Implant Surface and Hydrogen Peroxide in Biologically Relevant Environments. Julie J. Mucyo1,2, Joanna M. McKittrick2, John A. Frangos1,3, and Christine A. Orme1. 1Chemistry and Materials Science, LLNL, Livermore, California; 2Materials Science and Engineering Program, University of California, San Diego, La Jolla, California; 3La Jolla Bioengineering Institute, La Jolla, California.

Titanium is a widely used material for load bearing and dental implants. Over the decades of use, it has been seen that titanium implants are able to be accepted by the body and in fact become integrated into surrounding bone tissue, osseointegrate. Titanium implants are inherently covered with a layer of titanium oxide, titania, that is a stable passive layer in an aqueous environment. Hydrogen peroxide has been shown to improve the biocompatibility of titanium surfaces with its high oxidative potential.


Calcium phosphate cements are used as bone substitutes for orthopedic and craniofacial applications. Addition of their injectability, biocompatibility, osteoconductivity, and similarity to the inorganic component of bone provides key advantages over other bone mineral substitutes. However, the application of these cements is limited by poor tensile strength properties. In addition, we recently reported that calcium phosphate cements are susceptible to accelerated cracking under both fatigue and moisture-assisted stress corrosion conditions. In the present work, we report the effect of introducing an organic second phase on the fracture behavior of hydroxyapatite cement. The study focuses on serum albumin, a prevalent blood protein that can be mixed with cement prior to surgical application. The addition of albumin can also be supplemented with biologically active growth-stimulating proteins such as those in the transforming growth factor-β (TGF-β) family. Results showing the effect of albumin additions on strengthening as well as on cracking rates in simulated physiological environments are reported. The microstructural changes that occur with the addition of albumin are also considered. The observed trends are used to develop mechanistic models that describe the kinetics of cracking in calcium phosphate cements both with and without organic second phases. Implications on load-bearing applications for calcium phosphate cements are discussed.

W11.19 Using Electrostatic Forces to Shape Materials for Biomedical Applications. Cheryl Lynn Casper1,2, Jean S. Stephens1,3, Nori Yamaguchi1,4, William Yang5, Cindy Farach-Carson2,6, Kristi L. Kick1,4, D. Bruce Chao7,8, and John F. Rabolt1,4. 1Materials Science and Engineering, University of Delaware, Newark, Delaware; 2Biomedical Sciences, University of California, San Diego, La Jolla, California; 3Dupont Central Research and Development, Experimental Station, Wilmington, Delaware; 4Delaware Biotechnology Institute, Newark, Delaware.

Electrospinning is a technique that applies an electric field to a polymer solution in order to produce nanometer to micrometer diameter fibers. The fibers are able to interact with collagen and the proteins that occur with the addition of albumin are also considered. The mechanical properties of the fibrous network make electrospun fibers a desirable choice for a wide range of biomedical applications. When electrostatic forces are used to shape polymers in solution, a number of variables influence the integrity of the polymer chain and its conformation as it is subjected to medium-large (≥ 500 V-cm) electric fields. Concerns exist that architectural changes may occur in these electrospun biopolymers as a result of the electrospinning process. The importance of this issue is compounded when the materials are subjected to the electric field and are biocompatible in nature. Recent work has investigated the post electrospinning changes in the mechanical properties of electrospun collagen fibers. Changes induced by electrospinning and treatment of the electrospun fibers with hydrogen peroxide were observed. The final mechanical properties of the electrospun fibers may be used to optimize the electrospinning process to achieve the desired mechanical properties.

W11.20 Influence of Short-Chain Alcohols on the Mechanical Properties and Area/Molecule of Fluid-Phase Lipid Bilayers. Hung Van Ly and Margie Longo. Chemical Engineering and Material Science, UC Davis, Davis, California.

The application of lipid membranes play an important role in the stability, permeability, and deformability of natural cells and drug-delivery liposomes. We show through micropipette aspiration of giant unilamellar liposomes that short-chain alcohols (methanol, ethanol, propanol, and butanol) can decrease the elastic moduli, toughness, and thickness of the fluid-phase bilayer. We propose the observed changes are caused by decreases in the interfacial tension of the bilayer. We verify this hypothesis by first determining the interfacial tension of the bilayer from elasticity measurements and showing how it decreases with increasing alcohol concentration. Secondly, from the decrease in the interfacial tension in the bilayer we predict the increase in the area per lipid molecule, and its value compares favorably to the area per molecule measurement obtained by flow-pipette micropipette aspiration technique. The area of the fluid-phase membrane is observed to decrease with increasing alcohol concentration. Furthermore, the alcohol-induced interfacial tension reduction will be related to surface adsorption at the bilayer-water interface. We propose that surface adsorption of small molecules, and subsequent drastic modification in mechanical and barrier properties of membranes, impacts a number of areas including: alcoholic fermentation, drug-delivery, cell preservation, and cell viability.

W11.21 Abstract Withdrawn


Loss of tissues and internal organs is usually caused by trauma and/or degenerative changes. While small defects may heal spontaneously, critical-size defects require augmentation to heal. In clinical practice autogenous tissues are used to promote healing. Limited availability of tissues calls for development functional structural scaffolds which might potentially be used instead of autogenous tissues and organs. Optimally, the scaffolds should have an interconnected porous structure allowing for the flux of nutrients and the ingrowth of cells, extracellular matrix and blood vessels, and be bioresorbable or
biodegradable to allow the implanted matrix to be replaced by new tissue. The surface of the pore walls should support the attachment and proliferation of cells involved in the regeneration of the given tissue. Biodegradable polymers are candidate materials for scaffold and can be transformed into scaffolds with various geometrical forms such as membranes or sponges for example. This report presents a systematic preparation of films from new biodegradable polyurethanes and/or their composites with nanosize hydroxyapatite or tricalcium phosphate crystals. The scaffolds support the attachment and growth of articular chondrocytes, osteoblasts, human mesenchymal stem cells and myoblasts. Possible applications of these scaffolds are for the treatment of large bone defects, defects of articular cartilage and nerves and cardiovascular tissues, to mention just a few. Structural scaffolds from biodegradable polyurethanes promote healing of critical-size defects of bones, and mono-, bis- and tricortical defects in the ilium. Elastomeric properties of the polyurethane scaffolds allow the application of load and shear to cell-scaffold constructs, which enhance the attachment and proliferation of chondrocytes. The pore structure and degradation rate of polyurethane scaffolds can be controlled by changing the material composition and synthesis conditions. Increasing the content of the hydrophilic segment in the polymer chain facilitates mineralization of scaffolds designed for cancellous bone graft substitutes.

*W11.23* Biomimetic Apatite/Aamelogenin Coating on Titanium and its Effects on Osteogenic Gene Expression. Chuang Du,1 G. B. Schneider,2 R. Zacharias,2 C. Abbott1, D. Sebold2, C. Stanford2 and Janet Moradian-Oldak1.1 University of Southern California, Los Angeles, California; 2University of Iowa, IA, Iowa.

The promotion of osteogenic cell differentiation by bioactive molecules is a central issue in bone tissue engineering. Biomimetic coating provides a promising vehicle for delivering such bioactive molecules. The potential of amelogenin protein to facilitate osteogenesis has been documented. We have used a recombinant mouse amelogenin RM170 to fabricate biomimetic apatite/amelogenin coating on titanium surfaces. Our studies were aimed to evaluate the incorporation of the protein in the coatings and their effect on osteogenic gene expression. Apatite coating on ultrasonically cleaned titanium was induced in a two-step process following a slight modification of the technique reported by Wen & Moradian-Oldak 2003 (JBIM 42A:483-490). The uniformity of protein distribution was evaluated by fluorescent labeling. The modification of mineral morphology by the protein was investigated with scanning electron microscopy. Gene expressions of Cbfa1, osteocalcin, collagen type I, BSPHL and alkaline phosphatase of Human Palatal Embryonic Mesenchymal cells cultured on the coatings were quantitatively analyzed by Real Time RT-PCR. The buildup of hydroxyapatite or tricalcium phosphate crystals. The scaffolds support adhesion and proliferation of chondrocytes. The pore structure and degradation rate of polyurethane scaffolds can be controlled by changing the material composition and synthesis conditions. Increasing the content of the hydrophilic segment in the polymer chain facilitates mineralization of scaffolds designed for cancellous bone graft substitutes.

**W12.1** Primary osteoblasts adhesion onto RGD functionalized and crosslinked polyelectrolyte multilayers films. Catherine Picard,1 Rene Elsam,1 Pierre Schanze2, Benoit Frach1 and Jean-claude Vogel1.1 INSERM U505, Strasbourg, France; 2Paragone, Strasbourg, France; 1Institut Charles Sadron, CNRS, Strasbourg, France; 4Laboratoire de Chimie Bioorganique, CNRS, Strasbourg, France.

The adhesion of primary osteoblastic cells on top of biocompatible polyelectrolyte multilayers (PEM) films was investigated for native films and for films functionalized by effects of poly(L-glutamic acid) and deposited on top of poly(L-lysine)/poly(L-glutamic acid) (PLL/PGA), PLL/Poly(alginate), and PLL/Poly(galacturonic) films. The buildup of the film and the adsorption of the PGA-RGD could be followed by Optical Waveguide Lightmode Spectroscopy and by Atomic Force Microscopy. The mechanical stimuli was achieved by crosslinking the film with a water soluble carbodiimide in combination with N-hydroxysuccinimide. Fourier Transform Infrared Spectroscopy evidenced the conversion of amine and carboxylic groups into amide groups. The Alkaline Phosphatase activity test was used to assess the primary osteoblasts adhesion and proliferation on top of the different films over a period of ten days in culture. Whereas the native films are poorly adherent, the RGD-functionalized ones are extremely attractive to cells with a increase adhesion and proliferation of a four-fold impact. Additionally, films were successfully crosslinked thereby becoming much more attractive to cells than the native ones. More interestingly, combining the RGD effect and crosslinking effect lead to a very good cell adhesion and proliferation. We found that the cells did not react similarly on the different types of films investigated: they were either most sensitive to the chemical stimuli, mechanical stimuli, or both stimuli combined.

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**SESSION W12:** Engineering Cell Structure and Adhesion

**Chair:** Thomas Webster  
**Friday Morning, April 16, 2004**  
**Room 2002 (Moscone West)**

**8:30 AM W12.1**

**Primary osteoblasts adhesion onto RGD functionalized and crosslinked polyelectrolyte multilayers films.** Catherine Picard,1 Rene Elsam,1 Pierre Schanze2, Benoit Frach1 and Jean-claude Vogel1.1 INSERM U505, Strasbourg, France; 2Paragone, Strasbourg, France; 1Institut Charles Sadron, CNRS, Strasbourg, France; 4Laboratoire de Chimie Bioorganique, CNRS, Strasbourg, France.

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**9:00 AM W12.2**

**Stable, Nanoscale Glycosphingolipid Films for Use in Sensing Applications.** Rory Stine1, Cara-Lynne Schengrund2 and Michael Vincent Pishko1.1 Chemical Engineering, Pennsylvania State University, University Park, Pennsylvania; 2Biochemistry and Molecular Biology, Hershey Medical Center, Penn State Univ., Hershey, Pennsylvania.

We have developed a means of producing thin, oriented lipid monolayers which are stable under repeated washing and which may be useful in biosensing or surface-coating applications. Glycosphingolipids (GSLs) such as GM1 were used as a representative lipid for this process. Initially, a mixed self-assembling film composed of octanethiol and hexadecanethiol was constructed on a clean gold surface. This highly hydrophobic surface was then brought into contact with a thin GSL layer that had been deposited at the air/water interface of a monomolecular film of GSL in hexane on top of a layer of water, leading to a GSL layer on the water with the fatty acid portion of the molecule facing upwards. The GSL layer, now deposited on the gold surface, was then heated to cause interconverting of the fatty acid and sphingolipid chains, and cooled to form a highly stable film which withstanded repeated rinsing and solution exposure. Presence and stability of the film was confirmed via ellipsometry, FTIR, and QCM, with an average overall film thickness of 10 nm. The orientation of the lipid film, with the polar head group at the air/substrate interface, may be used to coat a substrate surface in a manner that could impart a specific functionality to the interface. This could be ideal for sensing applications if a fatty acid with a specific binding ligand as its head group were used. The GM1 ligand may be used as such, with the saccharide head group showing a high degree of affinity for cholera toxin.

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**10:00 AM W12.3**

**An electronic retinal interface for single cell stimulation.** Neville Z. Mehentsi1, Greg S. Tsien2, Harvey A. Fishman2 and Stacey F. Bent3.1 Chemical Engineering, Stanford University, Stanford, California; 2Electrical Engineering, Stanford University, Stanford, California; 3Ophthalmology, Stanford University, Stanford, California.

While clinical results of visual prostheses to date have been encouraging, there remain many challenges before useful vision can be achieved. Current retinal prostheses primarily use microelectrode arrays to locally depolarize groups of neurons in a field-effect manner. An amplified signal from an array of photodiodes is commonly used to power the microelectrodes, which due to their large size are spaced at a low density relative to the cells layered in the central retina. Since current implants are located up to 100 microns or more from the cells they will be stimulating, both resolution and power requirements for the devices are adversely affected. A major goal of a retinal prosthesis that would restore useful vision is to connect microelectrodes to individual retinal neurons, thus providing a high-resolution interface with minimal power requirements. To address some of these challenges, we have used cell micropatterning technologies, such as microcontact printing, to direct the growth of retinal neurites to individual microelectrodes to achieve single cell stimulation. Rat retinal ganglion cell (RGC) neurites and purkinje cells were patterned through retinal dissection and immunopanning techniques. A laminin micropattern was aligned on a microelectrode array, and RGCs seeded on the array extended neurites along the pattern to contact individual electrodes. Threshold current and charge densities for cell stimulation were measured, and found to be an order of magnitude lower than those found for equidistant cell bodies that were not patterned toward a microelectrode. Since it is unclear what the governing electrical requirements for cell stimulation through micropatterned electrodes are, future work will be pursued to investigate these parameters and their effects. Electrode arrays with different geometries were microfabricated, and spatial and electrical requirements for cell stimulation through micropatterned neurites were characterized using fluorescent imaging techniques. Both physical and chemical approaches to cell patterning were
We have prepared synthetic cells in which a lipid bilayer membrane encapsulates a proteolytic functional mimic of the cytoplasm. This synthetic cytoplasm is comprised of two chemically dissimilar polymers, such as polyethylene glycol and dextran, at a few weight percent in water. Under appropriate conditions of temperature and concentration, these polymer solutions exist as aqueous two phase systems, in which each phase is enriched in one of the polymers. In our case, liposomes are prepared above the phase transition temperature for the polymer solution and then cooled to initiate phase separation within the liposomes. Biomolecules such as proteins and nucleic acids were localized to subvolumes within these synthetic cells by partitioning into one of the encapsulated phases. These structures are exciting in that they enable for the first time the interior volume of synthetic cells to be structured, and biomolecules to be partitioned reversibly among the encapsulated microphases. Ultimately, we hope to control biomolecule association and activity through manipulation of local concentrations within these synthetic cells.

In the design of microscale devices, there is considerable interest in the integration of biological components capable of performing complex functions. Proteins, for example, could serve as molecular motors or scaffolds, their natural ability to assemble into complex structures and transform chemical potential into mechanical force could be exploited in a device context. Living cells are capable of complex transformation processes such as metabolic reactions that are useful for the biogenesis of catalysis or bioremediation of toxins. They can transduce signals in response to detection of soluble analytes, and therefore have use in the design of biosensing devices. All of these potential applications of biological components require their integration into a synthetic device environment. To achieve this, methods for controlling the interface between the biological molecule or cell and the surrounding material are paramount. This has motivated us to develop new methods for coating material surfaces with synthetic substrates that are compatible with biological components, and conversely, for decorating cells with new chemical structures that permit their attachment to material surfaces. New technologies for interfacing cells and materials will be presented in this talk.

Microbubbles stabilized by a lipid/emulsifier monolayer shell are important for a variety of reasons in fundamental and engineering science. The shell composition inspired by naturally occurring microbubbles and recently innovated by methods of rational design, can be engineered to serve an array of functions. Our results obtained from electrochemical, optical and fluorescence microscopy on lipid monolayer-coated microbubbles and Langmuir monolayer-coated micro bubbles demonstrate the relationship between composition, microstructure and transport properties of the lipid shell and provide insight into microbubble stability. We show that the monolayer shell reduces surface tension and impedes gas transport. The stabilization mechanism is determined by the phase state of the monolayer acyl chain region. Surface tension is maintained at the equilibrium value for expanded phase lipids and completely diminished for condensed phase lipids. The shell resistance to gas permeation is only significant below that required for lipid shedding. The size and shape of the crystalline lipid domains are controlled by lipid hydrophobic chain length and quench rate; a rich shell microstructure is observed.

In recent years membrane constituted proteins in supported pattern bilayers have been used to visualize intercellular communication between cells. Different lipid compositions can be micro-patterned in supported bilayers and long range lateral mobility required for activation of cellular response may be controlled. Here we report a micro-pattern domain of the supported bilayer with the GPI-linked ectodomain of neurologin (NLG), a post synaptic membrane protein known to bind to neurolgin and induce synapse formation. We show that neuroligin expressing cells bind specifically to the supported bilayer that contains NLG, and that by controlling NLG pattern we can direct the cell binding and the synapse formation to specific location on our substrate. This micro-scale patterning of functional membrane-associated proteins is the first step toward cell based technological applications.

In millions of years of evolution, biological systems have developed abilities unmatched by manmade technologies. Membrane proteins are diverse and highly versatile components of these systems, having functions ranging from pores and pumps to sensors and transducers and more. Because of their high level of functionality, compact size, and common environment of lipid bilayer membranes, the possibilities of engineering devices functionalized by membrane proteins are very attractive. The functional lifetimes of these proteins may be extended from hours to years through incorporation into polymeric membranes, allowing their properties to be fully exploited and resulting in new classes of materials. Our initial work has centered on the creation, study, and characterization of the biomimetic polymer membranes. Several techniques allow the insertion of protein into the membranes. We have compared the protein insertion processes between natural lipid bilayers and our polymer monolayers and begun studies of protein function and diffusion within the membranes. As a benchmark, we have been employing a model system of voltage-gated pore proteins, which have electrically controllable pores. I will present our recent work in engineering this hybrid protein/polymer system. I will report on the progress of this work, the characterization of the membranes, protein insertion processes, and the yield and functionality of the composite. What we learn in this work is being applied to other proteins, such as the mechanosensitive channel protein MscL and the piezoelectric protein prestin, directed toward compact devices for mechanical sensing and acoustic sensing and transduction. We are developing a new family of active materials which derive their functional properties from membrane proteins. The development of this protein/polymer system will apply to most membrane proteins with some variation. This implies that once the learning curve necessary for successful production, insertion, orientation, and operation of a particular protein in an engineered environment has been successfully mastered, repeating the process will be somewhat easier for other proteins, facilitating a "plug-and-play" approach to protein membrane engineering.
binder and a foaming agent are dry-mixed and molded. The molded powder is then heated in a three-step thermal treatment that includes foaming through the decomposition of the foaming agent, followed by the crystallization and sintering of the polymer and the ceramic matrix. This step promotes partial solid-state diffusion and creates intergranular contacts between the sub-micron layers of hydrogels containing bound enzymes, which seed and their proliferation and differentiation rates, as well as their sequences of enzymes, substrates and inhibitors can be used to form proteins, biopolymers and synthetic polymers have been systematically altered to incorporate biological signals encoding degradation, controlled release, and tissue regeneration. Information garnered from both experimental and theoretical testing of these materials will be used to develop future generation materials that can serve as bioactive artificial extracellular matrices for regenerative medicine.

2:30 PM W13.4
Blomorphic separation gels as an inorganic growth medium.
Scott B. J. Oliver, Department of Chemistry, SUNY-Binghamton, Binghamton, New York.

Polycrylamide gels are commonly used to separate biomolecules, based on their highly ordered porous structure and their ability to bind and release these components in a controlled manner. Unlike most other gels, this system is based on control over the polymer network, which allows for faster mineralization on HA. This process is quite versatile and flexible and permits the adjustment of the final microstructural parameters of the foams. Cellular viability assays demonstrated the absence of cytotoxicity effects on macrophages in contact with porous titanium produced through the process described above, as well as in the presence of high quantities of the processing residuals. The aim of this study is to assess the potential of these highly porous titanium foams (< 81% porous) as an osteoconductive material. This has been achieved through a study of the morphology of MC3T3-E1 subclone 14 osteoblasts (ATCC, USA) adhering to the surface of titanium foams with three different pore sizes ranging from 136 to 434 µm after an incubation of 3 hours at 37°C. The cells are imaged with a scanning electron microscope after having been fixed with 2.5% glutaraldehyde in 1.0M sodium cacodylate, dehydrated in serial ethanol and amyl acetate baths and critical point dried. This analysis has demonstrated that cells adhere strongly to the titanium matrices because they have a spread out flat morphology and have formed extensive extracellular matrix networks throughout the three-dimensional scaffolds. These observations are compared to those observed on non-porous titanium discs with varying surface roughness. To complement this morphological study, the adhesion behavior of these pre-osteoblasts to porous titanium are quantified at different times from 1 to 18 hours after having been seeded and their proliferation and differentiation rates, as well as their capacity to produce mineralized extracellular matrix are investigated. These data suggest that the three-dimensional porous titanium scaffold is a promising osteoconductive material for bone tissue engineering applications.

REFERENCES:
Nanotechnology embraces a system whose core of materials is in the range of nanometric scales through several hundred nanometres, is referred to as "nanomaterial" by different experts, the commonly accepted concept refers nanomaterials as those materials with the basic structural unit specifically in the range of 1-100 nm (nanostructured). Crystaline solids with particle sizes 1-100 nm (nanofibers). Nanotechnology thus creates materials and products that can be synthesized in bulk, at several conventional terms in mechanical, electrical, catalytic, and optical properties. However, despite their promise to mimic the surface roughness, cells experience in vivo, the use of nanophase materials in biological applications may be limited by negative aspects. The present report in in vitro study was, therefore, to determine whether when added to a polymer composite, nanophase compared to conventional ceramics enhance functions of osteoblasts (or bone-forming cells). Results from this study provided the first evidence that functions (specifically, adhesion, synthesis of alkaline phosphatase, and deposition of collagen-containing mineral) of osteoblasts are enhanced on three-dimensional poly-lactic-co-glycolic acid (PLGA) composites containing nanophase compared to conventional titania (70:30 wt % PLGA:titania). Since the chemistry, material phase, porosity (%), and pore size of the composites were similar, this study implies that the nanostructured surface features created by adding a nanophase compared to conventional ceramic, was the key parameter responsible for the enhanced functions of osteoblasts. In this manner, this study adds another novel property of nanophase ceramics: their ability to enhance osteoblast functions in vitro in three-dimensional composite form. For this reason, nanomaterials demand further attention as orthopedic tissue engineering materials.

3:30 PM W13.7
Enzyme-Inspired Self-Assembling Peptide Amphiphile Nanofibers. Hannah Storrie1 and Samuel I. Stupp2,3; 1Department of Chemistry, Northwestern University, Evanston, Illinois; 2Department of Materials Science, Northwestern University, Evanston, Illinois; 3Feinberg School of Medicine, Northwestern University, Chicago, Illinois.

Mimicking the action of enzymes in self-assembling materials is a new frontier in biomaterials design. Enzyme mimics would provide access to chemically active materials capable of providing specific signals and substrates to cells in vitro and in vivo. Peptide amphiphiles provide a robust system for the introduction of epitopes required for enzyme activity. We have chosen to explore mimics of alkaline phosphatase (ALP) for biomimetic applications. The active site of ALP contains two zinc ions, one bound via two histidine residues and an aspartic acid residue and the other bound via two aspartic acid residues and one histidine residue. Additionally, an arginine residue and a serine residue are required to coordinate the ligand in the binding pocket. To mimic this protein structure, we have designed a novel PA that contains a branched peptide group with a terminal atom for zinc binding and a second arm for ligand binding coupled to an aliphatic tail by a crosslinkable cysteine tetramer region that allows the self-assembled structure to function across a broad pH range. PA's form gels at high concentrations (<1% w/v) and provide a three-dimensional scaffold for cell growth that is similar to the extra-cellular matrix. We have shown ion-dependent self-assembly of branched PA's in the presence of zinc ions and confirmed histidine-zinc interactions by NMR, which demonstrates a specific interaction of zinc ions with the PA, and indicates its ability to act as a structural mimic of ALP in vitro.

3:45 PM W13.8
In Vitro And In Vivo Tests Of Hydroxyapatite-Gelatin Nanocomposites For Bone Regeneration: A Preliminary Report. Chin-Chung Ko1, Ying-Lien Wu2, R A Narayanan2, and Wei-Shou Hu1; 1Oral Science, University of Minnesota, Minneapolis, Minnesota; 2Department of Materials Science, University of Minnesota, Minneapolis, Minnesota.

A biomimetic process has been developed to fabricate hydroxyapatite-gelatin (HAP-GEL) nanocomposites for bone regeneration (Chang and Ko et al. 2003). We hypothesize that this newly developed HAP-GEL is osteoconductive and is suitable for tissue engineered scaffolds. This preliminary study is aimed to characterize cell affinity and osseous regeneration of the HAP-GEL.

The HAP-GEL was synthesized according to the procedures described in the previous publication. The attachment and proliferation of human fetal calvarial osteoblasts on HAP-GEL nanocomposites were evaluated for three different gelatin contents (2g, 3g, and 4g). The cells were seeded onto each disc and incubated at 34 degrees Celcius in 5% CO2 air atmosphere. At different time points of cultivation, cells were stained with Fluorocrome to determine their viability and morphology. The cell proliferation, cells were detached at Days 1, 4, and 7 by trypsinization and counted. For in vivo tests, HAP-GEL rods were implanted into the proximal femur of Sprague-Dawley rats (Animal Protocol ID#2212A8902). After one month of the implantation, the femurs were harvested and fixed in 10% formalin. The decafcilined HAP-GEL-bone sections were prepared through the EXAKT microgrinding system and stained with Stevenel's Blue and Van Gieson. The formation was assessed by bright red. Four hours after attachment, most cells appeared round in all discs; cell spreading was observed after 24 hours. The highest gelatin content supported a significantly higher cell growth than the others at 7 days. This is a critical building support satisfactory attachment, spreading, and growth. In vivo results showed excellent interfacial bone regeneration. No necrotic tissues were found. In conclusion, the HAP-GEL not only mimics the biochemistry and nanostructures of bone but also provides the first evidence that functions (specifically, adhesion, synthesis of alkaline phosphatase, and deposition of collagen-containing mineral) of osteoblasts are enhanced on three-dimensional poly-lactic-co-glycolic acid (PLGA) composites containing nanophase compared to conventional titania (70:30 wt % PLGA:titania). Since the chemistry, material phase, porosity (%), and pore size of the composites were similar, this study implies that the nanostructured surface features created by adding a nanophase compared to conventional ceramic, was the key parameter responsible for the enhanced functions of osteoblasts. In this manner, this study adds another novel property of nanophase ceramics: their ability to enhance osteoblast functions in vitro in three-dimensional composite form. For this reason, nanomaterials demand further attention as orthopedic tissue engineering materials.

4:00 PM W13.9

Nanoparticulate systems are being developed for use in pharmaceutical and industrial controlled release applications. In the United States alone, over 300,000 patients enter the emergency room each year due to complications from overdose of prescription drugs. In fact, the leading methods for detoxification are via overdose of antipsychotic, popular anti-depressant. There currently exists no quick and effective method to detoxify these patients. The goal here is to synthesize "soft" emulsion particles coated with an inorganic shell with tailorable porosity and degradation properties, which when introduced to the bloodstream-intravenously, act as drug "sponges" for patients overdosed on these lipophilic drugs. This is done via a biologically inspired mineralization process of surface-induced deposition of calcium carbonate coatings templated onto charged emulsion particles. The experimental technique includes the addition of ammonium carbonate into a combination of oil-in-water microemulsion, calcium chloride, magnesium chloride, and a highly acidic polymer. Nanoparticles with diameters of about 200 nm have successfully been synthesized. Current experiments on these particles are directed towards in vitro testing of drug uptake capabilities and templating porosity of the inorganic shell using binary surfactant systems in order to achieve molecular filtration.

4:15 PM W13.10

This presentation describes the development of a minimal model of hemostasis. Hemostasis is a complex functional system that consists of 80 coupled biochemical reactions that involve biomolecules and cells, both in solution and on surfaces. Hemostasis is responsible for repairing damaged blood vessels and controlling bleeding. It maintains blood in a fluid, clot-free state under normal conditions, but creates a localized solid clot in response to vascular damage. The complexity of hemostasis is associated with a finely tuned self-regulation, essential for its function. This self-regulation is believed to be the basis of two essential features of hemostasis: i) threshold response: there is no response to small regions of internal vascular damage present throughout the circulatory system, but full response to substantial damage of a blood vessel. ii) local response: a clot formed in the region of substantial damage is confined to that region. A simple model of hemostasis was developed. Using this model, only three non-biochemical reactions were required to create a functional biomimetic microfluidic system. Over the past 200 years, when damaged. Some of the predictions of the model were successfully tested with human blood. This biomimetic system suggests that the function of hemostasis is highly dependent on the geometry of the junctions between vessels, and suggests a hypothesis that hemostasis influenced the evolution of mammalian vascular networks.

4:30 PM W13.11
Biomimetic Encapsulation of Enzymes. Rajesh R Naik1, Heather R Luckniff2, Jim C Spain3 and Morley O Stone3; 1MLPLP, Bdgl 651, Air Force Research Laboratory, Dayton, Ohio; 2Airbase Technologies Division, Air Force Research Laboratory, Tyndall Air Force Base, Florida.

The successful use of enzymes for applications in catalysis and sensors is dependent on the host material used for immobilization of the enzymes. One of the most widely used methods for immobilizing the enzymes.
Enzymes are sol-gel silica encapsulation. Entrapment of biomolecules by the sol-gel process involves the hydrolysis of the alkoxysilane precursor by water, acid or base catalysis, to form hydroxy derivatives. The biomolecule prepared in a buffered solution (pH 7) is subsequently added to the alcohol derivatives or sol, and gelation is initiated. The gel is slowly aged at low temperature over a period of several weeks. The sol-gel method is a generic immobilization technique used to entrap biomolecules. Here we describe the entrapment of enzymes in a silica matrix using a biomimetic approach. The entrapment process is a one-pot procedure wherein the silica matrix is biologically synthesized in the presence of the enzyme to be encapsulated.

Biohybrid Architectures from Amphiphilic Macromolecules.
Jeroen Cornelissen, Gerald Metselaar, Irene Reynhout, Dennis Vriezema, Alan Rowan and Roeland Nolte, Organic Chemistry, University of Nijmegen, Nijmegen, Netherlands.

Biohybrid Architectures from Amphiphilic Macromolecules: In Nature structural information is transferred in a hierarchical fashion from the smallest building blocks, i.e. amino and nucleic acids, to the complex architectures formed by them. These principles can be applied also in the formation of peptide derived polysiocyanides. Polymers of isocyanides have a helical conformation in which side group n is more or less on top of side group (n + 4). The distance between these side chains is very well suited for the formation of beta-sheet-like hydrogen bonds between the peptide groups present in these side chains. Both in the formation and their final structure these polysiocyanopeptides resemble the properties of proteins. The stereochemical information present in the monomer is transferred to the stable helical conformation of the polymer and in certain cases, i.e. acid catalyzed polymerization, the polymerization of optically active monomers is highly stereoselective. If a macromolecular initiator, such as polystyrene, is used for the isocyanide polymerization, an amphiphilic rod-coil diblock copolymer is obtained. In a selective solvent for the isocyanide segment, these macromolecules self-organize in a selection of morphologies. In water the aggregate formation is, among other factors, dependent on the ratio between the two blocks and the interaction between like blocks. This results next to micellar and bilayer type aggregates, in the formation of superhelices. In a slight modification these types of block copolymers give large and stable vesicular aggregates. These are able to encapsulate enzymes that stay active in the inner compartment of the vesicle, while substrate molecules still can diffuse through the polymer membrane. This results in a new type of microreactor. Systems are recently developed, where the hydrophilic segment of the macromolecules consists of a protein or an enzyme. Three different approaches were applied in connecting the biomacromolecule with the hydrophobic polystyrene segment: i.e. covalent, by cofactor reconstitution and by using the biotin-streptavidine couple.