SYMPOSIUM O

Materials Inspired by Biology

April 21 – 25, 2003

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* Invited paper

TUTORIAL

ST O: TISSUE ENGINEERING
Monday, April 21, 2003
10:00 a.m. - 5:00 p.m.
Salon 1/2 (Marriott)

The rapidly expanding field of tissue engineering has capitalized upon the design and synthesis of materials that elicit specific cellular responses while maintaining desired physical properties. Accordingly, this tutorial is designed to introduce the materials scientist to the important roles that materials play in the engineering of new tissues and organs. Participants will be introduced to currently used materials and potential areas of development. The biological molecules and processes that must assume a central role in material design and application will then be covered, and specific applications for novel materials in both soft and hard tissue engineering will be discussed.

Instructors:
David Mooney, University of Michigan
Alyssa Panitch, Arizona State University
William Landis, Northeastern Ohio University

SESSION O1: TISSUE ENGINEERING AND BIOMATERIALS
Chairs: Laurie B. Gower and Kristi L. Kiick
Tuesday Morning, April 22, 2003
Franciscan I (Argent)

8:30 AM *O1.1
BIO-INSPIRED MORPHOGENS AND BIOMIMETIC BIOMATERIALS FOR REGENERATIVE MEDICINE.
A. Hari Reddi, Center for Tissue Regeneration and Repair, Department of Orthopaedic Surgery, University of California, Davis, Sacramento, CA.

Regenerative medicine is the science of design and manufacture of parts for functional restoration of damaged tissues due to cancer, disease and trauma. Morphogenesis is the developmental cascade of pattern formation, body plan establishment and differentiation of tissues culminating in adult form. Regeneration in general, recapitulates, embryonic morphogenesis. Thus, the principles of morphogenesis can be applied to tissue engineering for regenerative medicine and surgery. The three-key ingredients for tissue engineering are inductive morphogens, responding stem cells, and extracellular matrix materials. Therefore bioactive morphogens can be integrated into materials for functional restoration by tissue engineering. A morphogen is a morphogenetic protein signal that acts on responding stem cells. Morphogens induce a sequential multistep cascade. Bone morphogenesis is induced by bone morphogenetic proteins (BMPs). BMPs play a role in pattern formation, cell differentiation, maintenance and regeneration of tissues. BMPs are pleiotropic and acts on chemotaxis, mitosis and differentiation of progenitor stem cells. There are nearly twenty BMPs. BMPs have actions beyond bone in development of teeth, heart, kidney, eye, skin, brain. Thus, BMPs may be called body morphogenetic proteins. Stem cells are primordial cells with unlimited replicative potential and can be programmed by morphogens such as BMPs. Extracellular matrix is the native scaffolding material that can be used to deliver morphogens such as BMPs for tissue engineering of bone. Biomimetic materials mimic the properties of extracellular matrix materials such as collagens, proteoglycans and glycoproteins. BMPs bind to collagens I & IV, heparan sulfate and heparin. BMPs bound to collagen acts as a composite biomaterial to initiate bone formation and the shape can be molded an appropriate template. In studies with BMPs bound to hydroxyapatite granules and discs indicated the role of geometry. The geometry of the carrier matrix is critical for optimal tissue engineering. Recombinant human BMP2, BMP4 and BMP7 can induce new bone formation. Collagen particles smaller than 44 μm are feeble in bone induction compared to coarse 420 μm. Gene therapy approaches using genes for morphogenesis such as BMPs allows a sustained, prolonged secretion of gene products. Thus, we can translate today's basic research into tomorrow's treatment for patients using techniques of integrating morphogens into biomaterials for regenerative medicine.

9:00 AM O1.2
BIOCOMAPTIBLE SELF-ASSEMBLED NANOSTRUCTURES AND CHARACTERIZATION OF THE ABIOTIC/BIOTIC INTERFACE.
Helen K. Baca, Carlee Ashley, Tamara Hartenberger, University of New Mexico, Albuquerque, NM; Darren Dunphy, Sandia National Laboratories; C. Jeffrey Brinker, Department of Chemical and Nuclear Engineering/Center for Micro-Engineered Materials, University of New Mexico and Sandia National Laboratories, Albuquerque, NM.

A single living cell incorporates the abilities to respond to an external stimulus through signal recognition, amplification and transduction. Immobilizing whole cells in a porous, nanostructured silica host matrix through biocompatible evaporation induced self-assembly (EISA) allows the cells to be protected, sustained and confined in a buffered environment while remaining accessible for nutrient, analyte and waste transport requirements. Although templated self-assembly of inorganic mesophases has been well investigated, the incorporation of immobilized whole cells presents unique hurdles, including toxic by-products formed during polymerization of silica precursors and detergent properties of typical amphiphilic template molecules. We used a several-pronged approach to provide the biocompatible materials and processing conditions necessary to maintain cell viability while retaining careful control over the microstructure of the inorganic host. Biocompatible templates, buffered sols and multiple reservoir processing conditions allowed us to immobilize genetically engineered Saccharomyces cerevisiae cells in a nanostructured silica host and to detect an environmental stimulus through expression of green fluorescent protein. Using phospholipid templates as amphiphilic structure-directing agents during EISA results in a variety of ordered mesophases, depending on the molar ratio of lipid to silica and the ratio of the lipid's head group surface area to hydrophobic tail volume. Incorporating yeast cells into the inorganic matrix, however, strongly influences the self-assembly pathway. When cells are added to the sol, both the molar ratio of lipid to silica required to form a nanostructured phase, and the morphology of the phase, changes. Adding a fluorescently labeled lipid and using latex and silica beads as hydrophobic and hydrophilic models during self-assembly, allowed us to see the preferential localization of lipid around the cells, consistent with both the cells having a structure-directing role through interaction with lipid templates, and the lipid forming a protective barrier around the cells.

9:15 AM O1.3
MECHANICAL AND DELAMINATION BEHAVIOR OF SOFT TISSUES: STRATUM CORNEUM AND OTHER SOFT TISSUES.
Kenneth S. Wu, Department of Mechanical Engineering, Stanford University, Stanford, CA; William W. van Osdol, ALZA Corporation, Mountain View, CA; Reinhold H. Dauskardt, Department of Materials Science and Engineering, Stanford University, Stanford, CA.

The mechanical and fracture behavior of soft tissues is often crucial to their function, with the underlying cellular and extracellular structures being optimized for the required properties. For example, the outermost layer of skin, or stratum corneum (SC), provides mechanical protection and a controlled permeable barrier to the external environment. Previous studies have demonstrated that temperature, hydration, and the application of topical agents influence the mechanical properties of SC. This prior art has focused primarily upon in-plane properties of the SC. We present a mechanics approach to study the delamination resistance and mechanical behavior of human SC tissue. In particular, mechanical behavior in the direction normal to the skin surface, which has received virtually no attention, is reported. Fracture mechanics-based cantilever-beam experiments together with stress-separation tests were used to determine the debond energy and cohesive strength characteristics of human SC. The influences of hydration, temperature, and lipid-extraction upon SC tissue were explored, and the highly anisotropic nature of SC mechanical and fracture behavior will be discussed with reference to the underlying cellular structure. The techniques developed have been extended to characterize the mechanical behavior of other soft tissues, for example, vascular tissue, and results will be presented.

9:30 AM *O1.4
EFFECT OF MECHANICAL FORCE ON THE DEVELOPMENT OF A NOVEL TISSUE-ENGINEERED MODEL OF HUMAN MIDDLE PHALANGES. William Landis, Robin Jacquet, Jennifer Hillyer, Northeastern Ohio Universities, College of Medicine, Dept of Biochemistry and Molecular Pathology, Rootstown, OH; Shinichi Asamura, Noritaka Isogai, Kinki Univ School of Medicine, Dept of Plastic and Reconstructive Surgery, Osaka, JAPAN.

The growth in length and development of vertebrate long bones are considered to be mediated in part by effects of mechanical force on these tissues. To test this concept on novel tissue-engineered models of human middle phalanges, constructs were subjected to mechanical force and examined to determine its effect on the structure and biochemical nature of the bones. As previously documented (N. Isogai et al., J. Bone Joint Surg., 1999), constructs were composed of chondrocytes and periosteum obtained from shoulders of young calves. Chondrocytes were seeded on polyglycolic acid sheets. A chondrocyte-seeded sheet was sutured to each end of periosteum wrapped about a polyglycolic acid/poly-L-lactic acid scaffold molded to the shape of a human middle phalanx. Constructs were placed without further treatment in the dorsum of nude (athymic) mice. After up to 40 weeks of such implantation, constructs developed features common to bone in vivo. In present studies, proximal and

distal ends of some constructs were tethered with narrow silicon strips and sutures to the neck and hind region, respectively, of mice while other constructs remained untethering in additional animals. In this manner, tethered constructs were designed to be directly affected by mechanical forces generated during ambulation of mice and untethered constructs were thought to be relatively unaffected. After implantation for 20 weeks, mice were sacrificed and implants were dissected and analyzed. Tethered compared to untethered middle phalanx models were found to be larger; have greater volumes of proximal and distal cartilage and higher content of proteoglycans, and possess more developed bone in central construct regions. These data support results demonstrating that mechanical forces alter bone growth and development and suggest that mechanical effects enhance the structure and composition of bone for application in tissue-engineering of impaired mineralized tissue.

10:30 AM *O1.5
MOLECULAR MECHANISMS OF BIOADHESION.
Deborah Leckband, University of Illinois, Dept. of Chemical and Biomolecular Engineering, Champaign-Urbana, IL.

Bioadhesion is essential to all living systems. Tissue engineering, wound healing, and even preventing the biofouling of implant materials involve controlling bio-adhesion. In living systems, cell adhesion is the result of the interplay between specific receptor-mediated cell-surface or cell-cell attraction and other nonspecific repulsive forces. Subtle changes in this balance can promote or impede cell adhesion. An example of how this force balance can significantly alter biological function is the protein neural cell adhesion molecule (NCAM). This is one of the most abundant cell adhesion proteins in the brain. It mediates cell adhesion and cell-cell signaling, and it promotes neurite outgrowth during brain development. However, early in development, the protein is modified by long, linear carbohydrate chains, which have a significant impact on the biological function of NCAM. The molecular mechanism by which the protein mediates cell attachments as well as the mechanism by which the carbohydrate modification alters NCAM function were both investigated by direct force measurements. Force measurements of NCAM-NCAM adhesion, absent the carbohydrate modification, reveal how the molecular mechanism of bio-adhesion is linked to the architecture of this protein. Furthermore, the directly measured effect of the carbohydrate derivatization on intermembrane potentials provides molecular insights into how these carbohydrate polymers alter the ability of NCAM to mediate cell attachment. These direct force measurements provide molecular level insights into the regulation of bioadhesion in vivo, and also generate molecular level design rules for promoting and regulating intercellular interactions in vitro.

11:00 AM O1.6
SELECT, ENHANCED OSTEOBLAST ADHESION ON NANOSTRUCTURED POLY-LACTIC-CO-GLYCOLIC SURFACES.
Karen S. Ellison†, Rachael L. Price, Derick C. Miller, and Thomas J. Webster, †Department of Biological Systems Engineering, University of Idaho, Moscow, ID; Department or Biomedical Engineering, Purdue University, West Lafayette, IN.

Every year, almost 500,000 people worldwide receive hip implants, more than 500,000 people require reconstruction of bone because of injuries or defects, and 16 million Americans become toothless, many whom need strengthening of their jawbones for attaching artificial teeth. Unfortunately, many of these implants clinically fail and thus require revision surgeries to remove the failed implant. One specific area of failure is the material's surface where bonding between the implant and juxtaposed bone, i.e. osseointegration, should occur. If the osseointegration is insufficient, the implant-bone interface will be unable to support physiological loading conditions. Functions of osteoblasts (bone-forming cells) increase osseointegration, while functions of competitive cell lines (fibroblasts, chondrocytes and smooth muscle cells) inhibit osseointegration through the formation of soft, not hard, tissue. Previous studies have shown that osteoblast function is increased on carbon fibers with a nano-structured (dimensions 100 nm or less) compared a conventional structured (dimensions greater than 100 nm) surface. The objective of the present study was to determine if the proactive properties of nanophase carbon fibers could be transferred to polymers pertinent for tissue engineering applications. For this reason, in vitro cell adhesion experiments were conducted with osteoblasts and three competitive cell lines (fibroblasts, smooth muscle cells, and chondrocytes) on poly(lactic-co-glycolic acid) (PLGA) films created from molds of nano-structured and conventional-structured carbon fiber compacts. Results revealed that osteoblast adhesion increased while competitive fibroblast and chondrocyte adhesion decreased on PLGA with a nanophase surface when compared to the conventional PLGA form. Competitive smooth muscle cell adhesion was not affected by the surface structure of the PLGA. These results suggest for the first time that PLGA used in implantable devices should possess a nanophase surface, which would enhance select osteoblast adhesion possibly leading to increased performance of an orthopedic implant, thus reducing the likelihood of failure.

11:15 AM O1.7
RECOMBINANT PROTEIN-CO-POLY(ETHYLENE GLYCOL) NETWORKS AS SYNTHETIC EXTRACELLULAR MATRIX.
Simone C. Rizzi, Institute for Biomedical Engineering, Dept of Materials, ETH and University of Zurich, SWITZERLAND; Sven Halstenberg, Program in Neurosciences and Department of Psychology, Stanford University, Stanford, CA; Franz E. Weber, University Hospital Zurich, Dept of Cranio-Maxillofacial Surgery, Zurich, SWITZERLAND; Hugo G. Schmökel, Clinic for Small Animals, Department of Veterinary Medicine, University of Bern, SWITZERLAND; Jeffrey A. Hubbell, Institute for Biomedical Engineering, Dept of Materials, ETH and University of Zurich, SWITZERLAND.

Our aim is to develop biologically active hydrogels as matrices for tissue regeneration consisting of recombinant derived proteins and polyethyleneglycol-divinylsulfone (PEG-divinylsulfone). These matrices are formed under physiological conditions by Michael-type addition between the thiols of the cysteine residues, representing designed cross-linking sites in the protein backbone, and the vinylsulfone groups of the PEG (Mw = 6000). In order to mimic key features of the extra-cellular matrices, the proteins were designed to promote cellular adhesion and to undergo enzymatic degradation. Specifically, the monomeric unit of the recombinant expressed protein constructs consists of 15 amino acid of human fibrinogen (α-chain, residue L^{94}-Y^{108}), including a cell adhesion RGD-sequence, a plasmin degradable site, and 11 amino acids corresponding to an MMP cleavable sequence existing in human collagen(α1(I)-chain, residue G^{950}-V^{960}): MGSSHHHHHHSSGLVPRGSHMEEGGCHGG-(LRGDFSSANNRDNTYEEGGCGGEEGPQGIAGQRGVIDSCHGG)$_n$ EEG. Additionally, a second similar construct was produced representing the negative control protein where the cleavage sites for plasmin and MMP are inactive: MGSSHHHHHHSSGLVPRGSHM-EEGGCHGG(LRGDFSSANNGDNTYEEGGCGGEEGDQGIA-GFPGSIDSCHGG)$_n$ EEG. For each system two protein polymers were successfully expressed in E-Coli (30-20 mg/L culture): A dimer (ca.11.5 kDa) and a tetramer (ca.20 kDa). The produced hydrogels have shown to promote cell spreading after 4h in serum-free medium, which was comparable to fibrin gels. The specificity of the incorporated RGD-sequence in the protein backbone was confirmed by the addition of soluble cyclic-RGD. Biochemical assays revealed the specificity of the MMP degradable site and have shown significant differences in the degradation kinetic when hydrogels were subjected to plasmin. In vitro (human fibroblasts) and in vivo (cranial bone regeneration of critical defect size in rats) have shown evident differences in cell migration and bone regeneration in the degradable and non-degradable systems (dimeric proteins and PEG-divinylsulfone). Our results demonstrate the feasibility of designing biomaterials with precise biological properties by combining recombinant DNA technology and polymer science.

11:30 AM *O1.8
DESIGN AND SYNTHESIS OF BIOINDUCTIVE HYDROGELS.
Jeffrey T. Koberstein, Kirsten Ostenberg, Lucy Vojtova, Danielle Lewis, Columbia Univ, Dept of Chemical Engineering, New York, NY.

There is growing interest in the development of polymeric biomaterials that are bioinductive in nature, that is, that promote a specific biological response when placed in vivo. This presentation describes our approach toward reaching this goal: the synthesis of functional hydrogels with controlled molecular structure on the nanoscale that can be decorated with tissue response modifiers including growth factors and adhesion ligands. The particular application discussed is the creation of functional hydrogel coatings for glucose biosensors that inhibit fibrous encapsulation by promoting neovascularization. For this purpose, a novel macromonomer route to functional hydrogels is described that employs atom transfer radical polymerization to prepare hydrogel macromonomers. The macromonomers can be radiation cured to produce hydrogel networks with well-controlled molecular weights between crosslinks. Proper selection of monomers allows for photopatterning of carboxylic acid functionality in the hydrogels to serve as sites for conjugation with peptides and proteins. Mscromonomer-based hydrogels provide potential solutions to problems associated with the use of conventional monomers for applications such as encapsulation of cells and drug delivery vehicles, and wound healing materials.

SESSION O2: NOVEL BIOMIMETIC AND BIOINSPIRED POLYMERS I
Chairs: Kristi L. Kiick and James L. Thomas
Tuesday Afternoon, April 22, 2003
Franciscan I (Argent)

1:30 PM *O2.1
MUSSEL ADHESION: DEFINING THE MOLECULAR PLAYERS. J. Herbert Waite, Univ of California, Marine Science Institute, Santa Barbara, CA.

Marine mussels are only one of a diverse group of animals that exploit DOPA (3, 4-dihydroxyphenyl-L-alanine) for adhesion. Mussel adhesion is mediated by a holdfast structure known as the byssus. DOPA's role in adhesion is implicated by two trends: 1) the proportion of DOPA increases sharply as one progresses down the byssus towards the substratum, and 2) proteins with the highest DOPA content, i.e. mefp-3 and -5, are detected at the plaque-substratum interface by laser desorption ionization mass spectrometry. One of these, mefp-3, is typically present and expressed by mussels in numerous (>30) variant forms of primary structure and post translational modification. Mefp-5, however, is only detected in plaques collected from high energy surfaces such as stainless steel and glass. This suggests that mussels may tailor adhesion to surface type and begs the question as to what surface features are being recognized and what the sequence motifs of molecular recognition are. Defining the role of DOPA in adhesion is a priority but complicated by its involvement in 2-electron oxidations, reverse dismutation and metal coordination.

2:00 PM *O2.2
MUSSEL ADHESIVE PROTEIN MIMETIC POLYMERS: ADHESIVE AND NONADHESIVE STRATEGIES FOR MEDICAL APPLICATIONS. Phillip B. Messersmith, Jeffrey L. Dalsin, Bi-Huang Hu, Bruce P. Lee, Biomedical Engineering Dept and Institute for Bioengineering and Nanoscience in Advanced Medicine, Northwestern University, Evanston, IL.

Certain marine organisms secrete remarkable protein-based adhesive materials for adherence to the mineral, metal, and wood surfaces upon which they reside. For example, mussel adhesive proteins (MAPs) contain L-3,4-dihydroxyphenylalanine (DOPA), an amino acid that is believed to be responsible for the adhesive characteristics of MAPs. Although the exact role of DOPA in these proteins is not known, recent evidence suggests that interfacial adhesion to substrates is generally believed to be due to chemical interactions between DOPA and functional groups at the surface of the solid substrate. Numerous research groups have attempted to take advantage of the adhesive nature of DOPA by synthesizing biomimetic polymers containing DOPA or mussel adhesive protein analogs. In this presentation I will describe our recent efforts to exploit the adhesive qualities of DOPA to prepare novel biomaterials for medical and other applications. For this purpose, we have synthesized conjugates of DOPA and biocompatible polymers such as poly(ethylene glycol) (PEG). The synthesis and physical properties of these polymers will be described, and the results of preliminary adhesion experiments presented. Finally, I will also describe a new approach to preparing nonfouling surfaces that takes advantage of DOPA-containing polymers. A variety of material surfaces (metals, metal oxides, semiconductors) can be rendered nonfouling by exploiting DOPA residues as anchors for nonfouling polymers like PEG. For example, exposure of a metal (e.g. gold) surface to a solution of DOPA-PEG polymer results in significantly reduced protein and cell adsorption to the surface. Characterization of the modified surfaces by XPS, TOF-SIMS and other methods indicates that the nonfouling polymer is anchored onto the surface by the DOPA containing peptide. Obvious applications of this strategy include protein and cell-resistant surfaces for medical applications (biosensors, coagulation-resistant surfaces, etc.), however there is considerable potential use of this strategy for nonmedical applications as well.

2:30 PM O2.3
FROM THE SPIDER TO THE WEB: BIOMIMETIC PROCESSING OF PROTEIN POLYMERS AND COLLAGEN. J.S. Stephens and J.F. Rabolt, Department of Materials Science and Engineering, University of Delaware, Newark, DE; D.B. Chase, Central Research and Development, Dupont, Wilmington, DE.

The properties and materials that are applicable for tissue engineering devices have been defined as those that encourage cell growth and proliferation with little toxic effects to the body. Both bioderived and commodity materials have been investigated for tissue engineering devices, and the main areas of investigation have been porosity, surface area, roughness, and the passage of nutrients in and out of the structure. Our goal is not to replace these materials, but investigate a processing technique where these materials can be produced with the desired properties (mechanical, physical, chemical) for the application as tissue engineering devices. Electrospinning is a novel fiber formation process that readily produces nanodiameter fibers. We have been investigating structure/property/process relationships of electrospun fibers to build a stronger fundamental understanding of the process. This approach has allowed us to build a virtual database of the processing conditions needed to create materials with specific properties desirable for tissue engineering applications. By varying the processing parameters it is possible to produce unique nanoscale features, nanoporous fibers and nanowebs. The size, shape, and distribution of pores can be controlled by changing processing protocols. Through a judicious choice of the processing parameters we have been able to create webs of nanofibers (5 - 25 nm) from collagen, spider silk, nylon, and denatured collagen. Both the nanoporous surface texture and the nanoweb morphology increases the surface area of the fiber mats making them excellent candidates for tissue engineering scaffolds. Currently cell growth on these electrospun materials is being investigated in order to determine how differences in the properties (orientation, surface texture, fiber diameter) of the fibers affect cell growth. Investigations are also being carried out to explore how the sterilization processes and cell growth media affect the structural integrity of the electrospun materials as compared to the bulk material.

2:45 PM O2.4
THE SYNTHESIS OF GENETICALLY ENGINEERED MOLECULAR INTERCONNECT CANDIDATES FOR USE IN MICROELECTRONICS. S. Higashiya, S.C. Ngo, K.S. Bousman, X. Jin, N. Rana, E.T. Eisenbraun, R.E. Geer, A.E. Kaloyeros, J.T. Welch, University at Albany, Albany, NY.

Nanoscale molecular interconnects for use in electronic device construction may be based on self-assembly paradigms found in biological systems. When based on synthetic peptide architectures, these materials may be accessible via the methods pioneered by Tirrell. The peptide architecture is especially attractive in light of established rubrics for predicting self-assembly and the availability of a variety of functional motifs that can be employed to decorate the candidate interconnect molecules. Repetitive polypeptides designed for use as molecular interconnects were biologically generated from artificial coding sequences constructed by a novel block copolymerization technique based on joined multimers. Synthetic DNA sequences were oligomerized in the presence of appropriately designed endonuclease restriction site containing sequences. The resultant oligomers bearing the appropriate restriction sites were cloned into plasmid vectors. The multimerized repetitive DNA sequences were recovered as oligomer units by digestion using type IIs restriction endonucleases. The oligomer units were used for longer multimer construction or block copolymerization with a second multimer unit. The DNA sequences so constructed were cloned into expression vectors along with the restriction sites. The encoded polypeptides were over expressed in an Escherichia coli host using a conventional T7 system. This construction method allows multimerization and block copolymerization of flexibly constructed joined DNA sequences with adjustable endonuclease restriction site adapters for expression and functionalization of the resulting polypeptides. We have constructed repetitive DNA sequences and expressed the corresponding repetitive polypeptides such as (GAGAGAGYGAGAGAGF)n, designed to assume a beta pleated sheet structure where the aromatic residues may be arrayed to promote charge transport. Isolated polypeptides have been analyzed and are under evaluation as molecular interconnects.

3:30 PM *O2.5
SELF-ORGANIZATION AND SUPER-STRUCTURE FORMATION OF BIOPOLYMER-INSPIRED SYNTHETIC COPOLYMERS. Claus D. Eisenbach, Institute of Applied Macromolecular Chemistry, University of Stuttgart, Stuttgart, GERMANY.

The structural and functional properties of biological systems originate from the specific primary structure of the biomacromolecule which determines the secondary, tertiary and finally the quarternary structure of the biosystem. The mimicing of such building principles in purely synthetic polymers requires a controlled architecture of macromolecules which is most conveniently achieved by a step-by-step synthesis involving protective group chemistry. We have studied polyurethane-based segmented block copolymers containing precisely structured segments capable of forming nanoscopic microdomains consisting of either chain-extended or symmetrically chain-folded segments (with thermal morphology control) or even assemblies of double-helical bipyridine/Cu(I)complexes. The versatility of the microphase morphology and their potential for realizing novel bio-inspired polymer materials will be illustrated and discussed.

4:00 PM O2.6
Abstract Withdrawn.

4:15 PM O2.7
FOLDABLE HYBRID POLYMERS AS BIOSENSORS. Alex Li and Wei Wang, Washington State University, Department of Chemistry and Materials Research Center, Pullman, WA.

Foldable polymers with alternating single strand DNA and planar conjugated organic perylene units were found to self-organize into loosely folded nanostructures. Upon heating, the loosely folded

structures become more ordered as evidenced by pi-stacking in the perylene segments. The folding and unfolding processes driven by the molecular interactions of adjacent perylenes were monitored in both aqueous and organic solutions. Heat-promoted folding, or inverse temperature behavior, which originates from positive enthalpy changes, was only observed in water. Therefore, we attributed this inverse temperature dependence to hydrophobic effects rather than pi-pi molecular orbital overlaps between the perylene planes. These findings shed light on the design of new thermophiles in protein engineering as well as the construction of macromolecular-based nano-devices with actuator and sensory properties.

4:30 PM *O2.8
ORGANIZING DOMAINS IN COMPOSITE SYSTEMS: SYNTHESIS, PROCESSING, AND FUNCTION. Galen D. Stucky, Timothy J. Deming, Daniel E. Morse, Herb Waite, University of California, Santa Barbara, Depts of Chemisty & Biochemistry, Materials, and Life Sciences.

One of the most fascinating underlying aspects of the biogenesis of materials is the space/time definition of structure, function, and morphology at multiple length scales from complex mixtures of reactants and accessible processes. On the "bioinspired" benchtop synthesis and processing side, it means being able to make an organized continuum of multidomain components by molecular assembly and design. Of particular interest are some of the strategies used by nature to make possible the closely coupled assembly and processing of the inorganic domains, which once formed, generally have the deeper thermodynamic potential energies. Amorphous phase or nanoparticle assembly of the inorganic component enables space/time definition of the multi-compositional inorganic domains with built in functionality. As in biomineralization, an essential part of this organization is composite domain definition using electrolyte, hydrophobic/hydrophilic, or hydrogen bonding. This talk will selectively describe some biomineralization observations and the related use of kinetic control, competing processes, non-equilibria phenomena, multiphase media and organic/inorganic interfaces to synthesize composite materials that have patterned structural and functional properties.

SESSION O3: NOVEL BIOMIMETIC AND BIOINSPIRED POLYMERS II

Chairs: Kristi L. Kiick and Laurie B. Gower
Wednesday Morning, April 23, 2003
Franciscan I (Argent)

8:30 AM *O3.1
IT DEPENDS ON HOW YOU READ IT: NOVEL MACROMOLECULES VIA ALTERNATIVE TRANSLATIONS OF THE GENETIC CODE. David A. Tirrell, Pin Wang, Inchan Kwon, Division of Chemistry and Chemical Engineering, California Institute of Technology, Pasadena, CA.

We have developed three approaches to the synthesis of proteins containing novel amino acids. In the first approach, we replace every copy of one of the natural amino acids by an analogue, in effect building proteins from an altered set of twenty starting materials. This approach is most useful when one is interested in changing the overall physical properties of the protein. A second method, which has also been implemented successfully by Schultz and coworkers, allows site-specific incorporation of a single copy of an amino acid analogue in response to a stop codon. Such methods are useful in probing protein structure and function. The third approach, developed most recently, uses mutant transfer RNAs to break the degeneracy of the genetic code, and offers the prospect of a protein chemistry based on a substantially expanded set of amino acid building blocks. This lecture will describe the most important elements of each of these strategies as well as some thoughts on the design of wholly artificial proteins with potential application in biotechnology and materials science.

9:00 AM *O3.2
ENVIRONMETNALLY RESPONSIVE MATERIALS CONSTRUCTED VIA PEPTIDIC MOLECULE FOLDING AND SELF-ASSEMBLY. Darrin Pochan, Lisa Pakstis, Bulent Ozbas, Dept. of Materials Science and Engineering; Joel Schneider, Karthikan Rajagopal, Dept. of Chemistry and Biochemistry, University of Delaware, Newark, DE; Andrew Nowak, Timothy Deming, Department of Materials, UCSB, Santa Barbara, CA.

We are exploring new methods of materials construction via aqueous molecular self-assembly, specifically peptidic molecule self-assembly. By using peptidic molecules in the self-assembly design process, one can take advantage of inherent biomolecular attributes, namely secondary structure and intramolecular folding events, in addition to more traditional self-assembling molecular attributes such as amphiphilicty, to define hierarchical material structure and consequent properties. The self-assembled nature of the resultant material imparts beneficial rheological properties (e.g. shear thinning, self-healing) for ease of processing. Intramolecular folding events impart an environmental responsiveness in the materials (e.g. drastic viscoelastic changes with changes in pH). The utility in material design with two general classes of molecule, block copolypeptides and β hairpin peptides, will be discussed. Hydrogels, with unique nano- and microstructure, vs. membrane suspensions can be built with block copolypeptide amphiphiles relative to the polyelectrolyte character of the hydrophilic block (ionic = gel, nonionic = membrane). pH responsive hydrogels can be constructed with small peptides that must intramolecularly fold in order to self-assemble into a gel scaffold. The molecular design principles that underlie the observed material properties (morphological, rheological, and basic cell-level biological) will be discussed.

9:30 AM O3.3
REVERSIBLE SELF-ORGANISATION OF HYBRID BLOCK COPOLYMERS MEDIATED BY PROTEIN FOLDING MOTIFS. Harm-Anton Klok and Guido W.M. Vandermeulen, Max Planck Institute for Polymer Research, Mainz, GERMANY; Christos Tziatzios, Dieter Schubert, Institut fuer Biophysik, University of Frankfurt, GERMANY; Ruth Duncan, Centre for Polymer Therapeutics, Cardiff University, UNITED KINGDOM.

The ability of amphiphilic block copolymers to self-organise into vesicles or micelles or to form hydrogels is widely explored for the development of novel materials for drug delivery or tissue engineering applications. Generally, the self-assembly of such block copolymers is driven by unspecific hydrophobic interactions. As a result, control over nanoscale supramolecular organisation is limited and structure and properties of the materials can only be manipulated to a small extent. In this contribution, the synthesis, self-organisation and properties of a new class of polyethyleneglycol (PEG)-b-peptide block copolymers are presented. The peptide segments are derived from protein folding motifs and can mediate self-assembly not only via unspecific hydrophobic interactions, but also via specific and/or directed hydrogen-bonding and coulombic interactions, which may allow enhanced control over supramolecular organisation and macroscopic properties in comparison with the amphiphilic block copolymers mentioned above. As an example, PEG-b-peptide copolymers based on a tetrameric coiled-coil motif will be presented. These block copolymers are prepared via solid phase synthesis and are obtained by coupling appropriately end-functionalised polyethyleneglycols to the N-terminal amine group of the peptide block. Circular dichroism, dynamic light scattering and analytical ultracentrifugation experiments indicate that the self-assembly properties of the peptides are retained upon conjugation to a PEG block and that the block copolymers self-organise into discrete, mostly tetrameric, supramolecular aggregates. These results demonstrate that the ability of peptide sequences to self-assemble hierarchically into well-defined higher-order structures can be transferred onto hybrid block copolymers. Since most polymers are amorphous, or at most semi-crystalline, the combination of protein folding motifs with synthetic polymers may allow the development of hierarchically organised polymeric materials with unprecedented levels of structural control. Furthermore, preliminary biological experiments show that the PEG-b-peptide copolymers neither are haemolytically active nor are significantly cytotoxic, which offers good prospects in view of possible biomedical applications.

9:45 AM O3.4
PEPTIDE-BASED HYBRID MATERIALS: STRUCTURE AND FUNCTIONALITY OF β-HAIRPIN POLYMERS. Jan C.M. van Hest, Dennis Lowik, Lee Ayres, Marjolijn Roeters, Jurgen Smeenk, Nijmegen University, Dept. of Organic Chemistry, Nijmegen, THE NETHERLANDS.

The β-hairpin conformation is one of the main peptide folds that recently has drawn much attention from scientists out of different fields of research, because of its involvement in both functional and structural natural processes. It has been recognised that β-hairpins are major contributors in protein-protein interactions by displaying oligopeptide epitopes at the turn positions. Plague formation by prions is a result of extensive β-sheet stacking. Furthermore, from a structural point of view, β-sheets are the elements that induce strength in silkworm and spider silk. In our research the β-hairpin conformation plays a pivotal role. On a fundamental level we want to increase our insight in its folding process and influence its stability. We aim at increasing the level of stability of β-hairpin oligopeptides by introducing fluorine-fluorine interactions via unnatural amino acids. Another way of stabilization is by modifying the N and C terminal ends of an oligopeptide, making it susceptible for anchoring into a liposomal bilayer. In this respect we can stabilize β-hairpin conformations that play an important role in protein-protein recognition. The second part of our research aims at translating the

properties of natural silk into hybrid materials. In this investigation we produce oligopeptide monomers, containing β-hairpin amino acid sequences. These monomers are polymerized into block copolymers by the controlled radical polymerization technique ATRP. This allows us to construct polymer structures with similar composition of amorphous and β-sheet domains as normally found in silk. An extension of this investigation is to use protein engineering. In this case we obtain full control over the build-up of the β-sheet structure. Triblock copolymers can then be prepared by modification of the β-sheets with polymer chains at both the C and N terminus. These materials serve not only a purpose as silk-mimetics but their application as nanotechnology building blocks is also pursued.

10:30 AM *O3.5
MECHANICS OF PROTEIN-BASED MATERIALS. D.W. Urry, University of Minnesota, Twin Cities Campus, BioTechnology Institute, St. Paul, MN, and Bioelastics Research Ltd., Birmingham, AL.

Design, preparation and characterization of elastic-contractile model proteins made possible two fundamental developments in protein mechanics: (1) That entropic elasticity of model proteins arises due to damping of internal chain dynamics on extension (even of a single chain). (2) That contraction/relaxation resulting from hydrophobic association/dissociation results from an interaction energy, called an apolar-polar repulsive free energy of hydration, due to competition for hydration between hydrophobic (apolar) and polar (e.g., charged) species constrained by sequence to coexist along a chain molecule. These two distinct physical processes of entropic elasticity and hydrophobic association become interlinked during diverse protein functions that include performance of energy conversions extant in biology. The intensive variables of the free energy efficiently interconverted by these interlinked physical processes include mechanical force, temperature, pressure, chemical potential, electrochemical potential, and electromagnetic radiation in the visible and ultraviolet light frequency range and in the dielectric permittivity frequency range from kHz to GHz. Inspired by repeating peptide sequences of the mammalian elastic fiber, more than one thousand protein-based polymers were designed, prepared and characterized with control of sequence and with the potential for any one of twenty amino acid residues in each position. Specifically, protein-based polymers utilizing compositional variations of the repeating peptide sequence, (Gly-Val-Gly-Val-Pro)n, called elastic-contractile model proteins, with appropriate replacement of a Val residue become capable of performing energy conversions that sustain living organisms. By direct observation, the amount of hydrophobic hydration determines the temperature for hydrophobic association, Tt; energy inputs that change the amount of hydrophobic hydration change the value of Tt. Analysis of hydrophobic-induced pKa shifts demonstrates the apolar-polar repulsive free energy of hydration to occur even in the totally unfolded state. Analysis of isometric contraction data, whether thermally- or chemically-driven increase in force at fixed length, eliminates changes in solvent entropy as contributing to entropic elastic force.

11:00 AM *O3.6
SALT STABLE HYDROGELS FOR BIOMEDICAL APPLICATIONS. Tim Deming, Andrew Nowak, Victor Breedveld, University of California, Materials Department, Santa Barbara, CA; Darrin Pochan, Lisa Pakstis, Bulent Ozbas, University of Delaware, Materials Science Department, Newark, DE.

Salt stable copolypeptide hydrogels have been synthesized by transition metal mediated polymerization of amino acid N-carboxyanhydrides (NCAs). These hydrogels are diblock amphiphilic copolymers of hydrophilic, charged segments of poly(L-lysine HBr) or poly(L-glutamic acid sodium salt), and helical, hydrophobic segments of poly(L-leucine). While many of these samples are able to form strong gels in deionized water at concentrations as low as 0.25 wt% polymer, stability in salt or buffer solutions was found to be only achieved at moderately higher polymer concentrations ($\sim$3.0 wt%). We have been able to adjust relative copolymer compositions and molecular weights to optimize hydrogel strength and polymer solubility for salt concentrations up to 0.5 M NaCl, as well as in aqueous buffers of varying pH, and cell growth media. These materials are unique since they do not collapse in high ionic strength media, even though gel formation is contingent upon the presence of highly charged polyelectrolyte segments. The remarkable properties of these hydrogels make them excellent candidates for use as scaffolds in biomedical applications, such as tissue regeneration.

11:30 AM O3.7
STABLE AND REVERSIBLE ARTIFICIAL PROTEIN HYDROGELS FORMED THROUGH SYNERGETIC EFFECT OF LEUCINE ZIPPER SELF-ASSEMBLY AND DISULFIDE BONDS AND THEIR MICROPATTERNING. Wei Shen, Rob G.H. Lammertink, Julia A. Kornfield, David A. Tirrell, California Institute of Technology, Dept of Chemical Engineering, Pasadena, CA.

Hydrogels were constructed from triblock artificial proteins consisting of two leucine zipper end-blocks and a hydrophilic random coil midblock. Self-assembly of the leucine zipper domains leads to a reversible network, which can be switched on near physiological pH and switched off at higher pH. To stabilize the hydrogels against dissolution, one cystein residue was introduced within each leucine zipper domain. The tertiary structure of the self-assembled leucine zippers brings these cystein residues into proximity, promoting the formation of disulfide bonds. Hydrogels anchored by disulfide bonds are able to retain integrity in aqueous buffer near neutral pH for more than one week, while hydrogels without cystein residues dissolve within several hours. The insoluble hydrogels retain a reversible response to pH, indicating the synergetic effect of leucine zipper self-assembly and disulfide bonds in forming the stable hydrogels. This reversibility allows us to generate micropatterns of these hydrogels using contact printing. An elastomeric PDMS mold was wetted with the protein solution at high pH and brought into contact with an aminated surface. Rehydrating the dried patterned protein in aqueous buffer near physiological pH leads to patterned swollen hydrogels. The electrostatic interaction between the negatively charged glutamic acid residues in the random coil midblock and the positively charged amine groups on the surface anchors the gels and suppresses swelling in the lateral direction, while significant swelling in the vertical direction was observed. This anisotropic swelling facilitates retention of the original micropatterns, which was confirmed by optical microscopy and AFM imaging.

11:45 AM O3.8
POLYSACCHARIDE-BASED SELF-ASSEMBLING HYDROGELS. Brandon L. Seal and Alyssa Panitch, Harrington Department of Bioengineering, Arizona State University, Tempe, AZ.

We have created a novel hydrogel system that mimics an extracellular matrix environment and also can sequester growth factors, peptides and drugs with affinity for sulfated polysaccharides. These physical gels use heparin to coordinate heparin-binding peptides, which have been conjugated to multiarm poly(ethylene glycol). Additionally, soluble factors can be added to the gels and released at rates dependent on relative heparin affinity. A heparin-binding peptide (HBD1) based on the heparin-binding domain from antithrombin III was synthesized and conjugated to 4-arm poly(ethylene glycol) tetravinyl sulfone (VSPEG; MW 10,000) such that four HBD1 molecules were bound to each VSPEG molecule. Heparin (18 kD avg. MW) was added to VSPEG-HBD1 in a 2:1 ratio to create 10% hydrogels. Using a rheometer with a 2 cm acrylic parallel plate, dynamic mechanical testing was performed on gels formed at 25°C by applying a 1 Pa oscillatory stress throughout a 1-100 rad/s frequency sweep. Upon the addition of heparin, hydrogels formed immediately. Mechanical studies showed that the VSPEG-HBD1 and heparin mixtures had gel-like properties between 10 and 100 rad/s. Other peptides (HBD2 and HBD3) with affinities to heparin lower than that of HBD1 also were synthesized and fluorescently labeled. Preliminary release studies were performed by forming 10% gels composed of VSPEG-HBD1 and adding soluble HBD2 or HBD3. The gels were washed several times with PBS, and the extracted PBS was examined for the presence of the fluorescent tag. These diffusion studies showed that the gels were able to sequester heparin-binding peptides and release the peptides at different rates based upon heparin affinity. Thus, the hydrogels in this study can be useful in drug delivery. These studies show that peptides based on natural protein domains can retain biological activity and also direct material assembly by interacting with extracellular matrix molecules.

SESSION O4: BIOMINERALIZATION AND HARD TISSUE BIOMIMETICS
Chairs: Laurie B. Gower and Kristi L. Kiick
Wednesday Afternoon, April 23, 2003
Franciscan I (Argent)

1:30 PM *O4.1
MULTILEVEL CONTROL OF CALCITE CRYSTALLIZATION USING SELF-ASSEMBLED MONOLAYERS. Yong-Jin Han, Joanna Aizenberg, Nanotechnology Division, Bell Laboratories, Lucent Technologies, Murray Hill, NJ.

Modern technologies require innovative methods for controlled fabrication of inorganic materials with complex form. Precise localization of particles, their nucleation density, size and morphology are important, but not easily controlled parameters that affect the performance of these materials. We show that micropatterned self-assembled monolayers provide sites for ordered nucleation, and make it possible to control various aspects of crystallization with high

precision. The power of this approach is its ability simultaneously to control the nanostructure of the nucleation site and to induce the near-surface gradients of ions. Crystallization results in the formation of large-area, high-resolution inorganic replicas of the underlying organic substrates.

2:00 PM *O4.2
WHEN IS TEMPLATE DIRECTED MINERALIZATION REALLY TEMPLATE DIRECTED? Elaine DiMasi, Brookhaven National Laboratory, Physics Department, Upton, NY; V.M. Patel, M.J. Olszta, M. Sivakumar, G.R. Sivakumar, Y.P. Yang, and L.B. Gower, University of Florida, Department of Materials Science and Engineering, Gainesville, FL.

Biogenic mineralization is recognized to depend upon the organic molecules present, not only as they are generally incorporated into organic-mineral composite materials, but because the organic species affect crystallization on many levels, including control of crystal size, polytype, morphology, and even the stabilization of amorphous phases in minerals which would otherwise be crystalline. It is by now taken for granted that a well ordered organic film can play the role of an atomic-scale template, determining the crystal structure that nucleates at the film. But only recently have in-situ structural probes, such as synchrotron x-ray scattering, been available to study film and template structures at early growth times and monitor these structures as mineralization proceeds. We will describe surface sensitive synchrotron x-ray studies of calcium carbonate crystals and films nucleating at monolayers assembled on water. In our experiments, a number of purely kinetic factors may be distinguished from the effect of the presumed atomic-scale template. In particular we focus on amorphous calcium carbonate thin films, nucleating at fatty acids from supersaturated solution containing 25-100 μg/ml concentrations of poly(acrylic acid). We will show that the chemical environment works together with the template to determine film growth rate, and the subsequent mineralization or, in some cases, *dissolution* of the amorphous film. Diffraction from the crystallized films provides no evidence for preferential orientation of the mineral relative to the monolayer template. Instead, kinetic factors dominate mineralization in this system, in contrast to many assumptions often applied to biomimetic mineralization of calcium carbonates.

2:30 PM O4.3
UNIQUE OPTICAL DESIGNS IN BIOLOGICAL SYSTEMS. Vikram C. Sundar and Joanna Aizenberg, Nanotechnology Division, Bell Laboratories, Lucent Technologies, Murray Hill, NJ.

The increasing technological requirement for a new generation of optical devices with novel architectures, tunability and tailored properties, provides a sharp stimulus to the academic pursuit of optical systems in organisms. Transport of light in biological organisms is affected through the development of complex light guiding systems. This talk focuses on a series of such organisms that have developed hybrid inorganic-organic or single crystal light elements. For example, investigations into the skeletal structure of brittlestars have provided an interesting insight into biologically formed, calcitic microlens arrays, which are designed to minimize the effects of spherical aberration and birefringence. Correlations between the focusing ability of these lens arrays and the location of optically sensitive receptors is extended to other organisms, and an effort is made to understand the structure-function relationships in these complex systems. We believe that further studies of biological systems will increase our understanding of how organisms evolved their sophisticated optical structures and will provide new materials concepts and design solutions for optical technology.

2:45 PM O4.4
SEA URCHIN MINERALIZED TISSUE. S.R. Stock, Institute for Bioengineering and Nanosience in Advanced Medicine, Northwestern University, Chicago, IL.

Sea urchin ossicles are structural analogs of mammalian bones and serve as a model biomineral system. Sea urchins employ as wide a range of composite reinforcement strategies as are seen in engineering composites, and, studied as materials, teeth (and other ossicles) from different echinoid families illustrate combinations of reinforcement parameters and toughening mechanisms providing good functionality. Studying ossicles from different sea urchin families, therefore, is one method of probing the composite design space available to sea urchins, and this offers important guidance for engineering of structural tissue. Such a study is the subject of this presentation. The multi-mode x-ray investigation employs microCT, both synchrotron and laboratory sources, phase contrast radiography and transmission microbeam diffraction mapping; voxels (volume elements) approaching 1 μm**3 can be interrogated noninvasively in millimeter sized samples. The results focus on sea urchin teeth, pyramids (jaws) and spines and serve to illustrate how this sort of integrated approach might be applied to bone.

3:30 PM *O4.5
COPPER AND ZINC HARDEN POLYCHAETE WORM JAWS. Helga C. Lichtenegger, Michael H. Bartl, Galen D. Stucky, Dept. Chem., University of California, Santa Barbara, CA; J. Herbert Waite, Dept. Cell. Mol. Biol., University of California, Santa Barbara, CA; Th. Schöberl, Erich Schmid, Inst. for Material Science, Austrian Academy of Sciences, Leoben, AUSTRIA; Stuart Stock, IBNAM, Northwestern University, Chicago, IL.

Biological tissues exhibit a variety of strategies to obtain hardness and durability. One of the most unusual strategies is found in polychaete jaws where the transition metals copper and zinc play a structural role (1,2). The jaws of the marine polychaete worm Glycera have recently been found to contain the copper-based biomineral atacamite in form of long, mineralized fibers that are oriented along the outer shape of the jaw and reinforce the needle-like jaw tip. The mineral was shown to enhance local hardness and stiffness of the material (3). In the jaws of a different polychaete worm species, Nereis, nanoindentation experiments have shown that reinforcement is achieved through zinc. Zinc in Nereis jaws, however, occurs in non-crystalline form. One possible mechanism would be the binding of zinc ions by the histidine-rich protein, thus building additional cross-links and enhancing the hardness of the protein matrix. An analogous mechanism may be present in Glycera jaws, in addition to the reinforcement by mineralized fibers containing atacamite: X-ray Absorption Near Edge Structure (XANES) shows that in Glycera jaws part of the copper resides in a chemical environment different from the mineral atacamite. This part of copper could be directly integrated in the protein scaffold. Such an additional reinforcement of the protein matrix could explain the extraordinary abrasion resistance of Glycera jaws despite their very low degree of mineralization (3). X-ray microtomography shows that the 3D distribution of zinc in Nereis jaws resembles that of copper in Glycera jaws, further suggesting an analogous function of zinc and copper in both polychaete worm species. 1. G.W. Bryan & P.E. Gibbs (1980) J. mar. biol. Ass. U.K. 60, 641-654. 2. P.E. Gibbs & G.W. Bryan (1980) J. mar. biol. Ass. U.K. 60, 205-214. 3. H.C. Lichtenegger, Th. Schöberl, M. Bartl, J.H. Waite & G.D. Stucky (2002) Science 298, 389-392.

4:00 PM O4.6
MODULATION OF CALCIUM OXALATE CRYSTALLIZATION BY PROTEINS AND SMALL MOLECULES INVESTIGATED BY *IN SITU* ATOMIC FORCE MICROSCOPY. S. Roger Qiu and C.A. Orme, Department of Chemistry and Materials Science, Lawrence Livermore National Laboratory; A.M. Cody, Department of Geological and Atmospheric Sciences, Iowa State University; A. Wierzbicki, Department of Chemistry, University of South Alabama; J.R. Hoyer, The Children's Hospital of Philadelphia, University of Pennsylvania; George Nancollas, Department of Chemistry, SUNY Buffalo; J.J. De Yoreo, Department of Chemistry and Materials Science, Lawrence Livermore National Laboratory.

Calcium oxalate monohydrate (COM) is a source of pathogenesis in humans where it causes kidney stone disease. Although a great deal of research has been carried out on the modulation COM by proteins and small molecules, the basic mechanism has not yet been understood. In this study, in situ atomic force microscopy (AFM) was used to monitor the COM surface under controlled growth conditions both from pure solutions and those doped with citrate and osteopontin (OPN) in order to determine their effects on surface morphology and growth dynamics at the molecular level. It is found that COM grows on complex dislocation hillocks. In pure solution, while growth on the (010) face is isotropic, hillocks on the ($\bar{1}$01) face exhibit anisotropic step kinetics. Steps of [$\bar{1}$0$\bar{1}$] and $< 120 >$ orientation are clearly delineated with the [$\bar{1}$0$\bar{1}$] being the fast growing direction. When citrate is added to the solution, both growth rate and morphology are drastically changed on ($\bar{1}$01) face, especially along the [$\bar{1}$0$\bar{1}$] direction. This results in isotropic disc-shaped hillocks a shape that is then reflected in the macroscopic growth habit. In contrast, no large growth changes were observed on the (010) facet. At the same time, molecular modeling predicts an excellent fit of the citrate ion into the ($\bar{1}$01) plane and a poor fit to the (010) face. Here we propose a model that reconciles the step-specific interactions implied by the AFM results with the face-specific predictions of the calculations. Finally, we present the results of doping with aspartic acid as well as OPN, an aspartic acid rich protein and a powerful inhibitor of COM growth. The AFM results show that OPN inhibits growth on the ($\bar{1}$01) face through a step pinning mechanism. The implications of the findings to the field of medicine will also be addressed.

4:15 PM O4.7
CHEMICAL PRE-TREATMENT OF TITANIUM SUBSTRATE: EFFECT ON APATITE COATING ADHESION. Ramin Rohanizadeh, Masly Harsono, Racquel Z. LeGeros, Calcium Phosphate Research Laboratory, Department of Biomaterials & Biomimetics, New York University College of Dentistry, New York, NY.

Plasma-sprayed HA coating combines the bioactivity of calcium phosphate (Ca-P) and the strength of the metal implants. However, this coating was shown to have variable composition, principally in the crystalline hydroxyapatite (HA)/amorphous calcium phosphate (ACP) phases ratio Chemical deposition offers an alternative method of depositing bioactive Ca-P coating at lower temperatures. Initial studies showed that such coatings are not adherent to the substrate. The aim of this study was to improve the adhesion and the coverage of the apatite coating by chemical pre-treatment of the substrate. Materials and Methods. Forty commercially pure Ti discs were polished and ultrasonically cleaned in distilled H2O, acetone, and ethanol. The discs were divided into four groups: Group 1: control; Group 2: heated for 90 min at 900°C; Group 3: treated with a solution containing 50 mM CaCO3 and 1.7% H3PO4 (37C for 24 hours) and heated for 90 min at 900°C. Group 4: treated with 100 mM CaCO3 and 3.4% H3PO4 and heat-treated (same procedure as group 3). All samples were subsequently immersed in a supersaturated calcium phosphate solution for two weeks at 37C. The apatite coating on the Ti surface was analyzed using scanning electron microscopy, X-ray diffraction and FT-IR. The adhesion between coating and substrate was measured using an adhesive tape and Image Analysis system. Results. Ti substrate treatment with a Ca-containing phosphoric acid solution followed by heat-treatment resulted in the formation of Ca2P2O7 at low acid concentration (group 3) and both Ca2P2O7 and TiP2O7 at high acid concentration (group 4). The formation of TiP2O7 and Ca2P2O7 improved both the adhesion and coverage of apatite coating obtained from chemical deposition. Conclusions. Formation of Ca-P/Ti compounds allows the formation of an adherent apatite coating from a metastable calcium phosphate solution.

4:30 PM O4.8
A POTENTIAL MECHANISM BY WHICH SILICON INCREASES THE RATE OF BONE APPOSITION TO SILICON-SUBSTITUTED HYDROXYAPATITE IN VIVO. A.E. Porter, N. Patel, S.M. Best, W. Bonfield, Dept of Materials Science and Metallurgy, University of Cambridge, Cambridge, UNITED KINGDOM.

Hydroxyapatite (HA), which resembles bone mineral, has achieved significant application as a bone graft material in a range of medical and dental applications. The incorporation of silicon into hydroxyapatite (HA) has been shown to have the potential to significantly increase the rate of bone apposition to HA bioceramic implants. Furthermore, it has been suggested that the bioactivity of HA ceramics is dependent on its microstructure, and in particular on the relative number and type of defect structures. In this study, high-resolution transmission electron microscopy (HR-TEM) was used to compare dissolution of phase pure HA and silicon-substituted hydroxyapatite (Si-HA) in an ovine model and to relate these observations to ultrastructural differences in the ceramics materials. HR-TEM imaging pre-implantation suggested a marked difference in grain boundary structure between Si-HA and HA. In particular, significantly more sub-grain triple-junctions per unit area were observed in Si-HA. Dissolution of the ceramic grains followed the order 1.5 wt% Si-HA> 0.8 wt% Si-HA > pure HA, at six and twelve weeks post-implantation. High-resolution, energy-filtered TEM (EF-TEM) confirmed dissolution to occur principally at grain-boundaries and sub-grain triple-junctions in the Si-HA. Our study reports for the first time a relationship between increased bone apposition and increased dissolution rate of the Si-HA grains. If increasing silicon-substitution does change the defect type in HA, this could be a potential mechanism by which silicon increases the solubility of HA and the subsequent rate at which bone apposes HA ceramics.

4:45 PM O4.9
UNDERSTANDING THE BIOMINERALIZATION PROCESS OF CALCIUM CARBONATE USING SYNTHETIC MICRO- AND MACROTEMPLATES. Parayil Kumaran Ajikumar, Rajamani Lakshminarayanan, Valiyaveettil Suresh, Singapore-MIT Alliance, Department of Chemistry, National University of Singapore.

Biomineralization is a fascinating process in which hard tissues such as bones and shells are formed through the template-assisted mineralization of calcium salts. Many elegant approaches were reported by other research groups towards understanding the molecular mechanism of biomineralization. Our research efforts are focused on understanding the role of multifunctional water-soluble polymer additives and the role of functional groups on the surfaces of macrotemplates such as thin films and fibers. The talk will focus on our recent results in controlling the morphology of crystals as well as thin film deposition of calcium salts on various substrates.

SESSION O5: AMPHIPHILIC MEMBRANES AND TEMPLATES
Chairs: James L. Thomas and Laurie B. Gower
Thursday Morning, April 24, 2003
Franciscan I (Argent)

8:30 AM *O5.1
A NANOSTRUCTURED, SELF-ASSEMBLED, BIOCOMPATIBLE DRUG DELIVERY SYSTEM. Joseph Zasadzinski, Cecile Boyer, Cara Evans, University of California, Department of Chemical Engineering, Santa Barbara, CA; Edward Kisak and Bret Coldren, Advanced Encapsulation, Inc., Santa Barbara, CA.

Vesicle-based drug delivery is hardly new, however, it is experiencing a renaissance due to new applications, such as gene therapy, new methods of enhancing vesicle circulation in the bloodstream by steric stabilization, efficient methods of drug loading in vesicles, and novel self-assembly approaches to more complex lipid membrane structures. Using vesicles as drug delivery vehicles inherently improves the circulation time of the drug; the drug is released slowly from the vesicle during circulation, making sustained drug release possible. Our work involves a optimizing and extending drug release through manipulation of the basic vesicle structure by encapsulating vesicles within another lipid bilayer membrane, a structure we call a vesosome. Preliminary results suggest that drug release in serum from conventional unilamellar vesicles is dominated by degradation of the bilayer by various lipases and lytic agents in blood. For example, hydrophilic carboxyfluorescein is released in 1 hour in serum at 37°C, while in isotonic saline, equivalent release occurs over weeks. This bilayer degradation causes premature release of many drugs that would benefit from extended, controlled release in vivo. However, in the vesosome, two distinct bilayers in series must be degraded prior to drug release carboxyfluorescein release from vesosomes is extended for approximately 100 hours. However, from the exterior, our vesosome is identical to conventional drug delivery liposomes and should have identical circulation times, drug loading characteristics, biocompatibility and safety. Our encapsulation process also provides a new method of biocompatible colloid stabilization in physiological environments for potential use as imaging and contrast agents. The multicompartment structure may allow for the co-encapsulation of imaging agents and drugs.

9:00 AM O5.2
A SUPPORTED LIPID BILAYER SYSTEM FOR OBSERVING AND USING OBSTRUCTED LATERAL DIFFUSION. Timothy V. Ratto†; Wan-chen Lin, Marjorie L. Longo, University of California, Davis, Biophysics Graduate Group and Department of Chemical Engineering and Materials Science; †currently at Lawrence Livermore National Lab.

Proteins, macromolecules, and lipid domains are believed to hinder molecular lateral diffusion in cellular membranes. We have constructed a well-characterized model membrane system to better understand how obstacles in lipid bilayers obstruct diffusion and for use in controlling the confinement of mobile molecules. We utilize supported bilayers composed of mixtures of 1,2-dilauroylphosphotidylcholine (DLPC) and 1,2-distearoylphosphotidylcholine (DSPC). Because these lipids are immiscible and phase separate at room temperature, a novel quenching technique allowed us to construct fluid DLPC bilayers containing small (∼50 nm) disk-shaped gel-phase DSPC domains that acted as obstacles to lateral diffusion. Our experimental setup enabled us to analyze samples with atomic force microscopy and exactly characterize the size, shape, and number of gel-phase domains before measuring the obstacle-dependent diffusion coefficient. Lateral obstructed diffusion was found to be dependent on obstacle area fraction, size, and geometry. We find that at solid-phase area fraction between ∼35% and 70% (the percolation threshold), diffusion is anomalous at short times and becomes normal at longer times as predicted by theory and Monte Carlo simulations.

9:15 AM O5.3
TRANSPORT PROPERTIES AND SURFACE MORPHOLOGY OF THE LIPID SHELL ENCASING A MICROBUBBLE.
Mark A. Borden, Gang Pu, and Marjorie Longo, UC Davis, Davis, CA.

Micron-scale bubbles (microbubbles) are currently used in contrast-assisted ultrasound, drug and gene delivery, and blood substitution. Lipid-coated microbubbles are composed of a gaseous core and crystalline lipid/emulsifier monolayer shell that stabilizes the microbubble against rapid dissolution and coalescence. We modified Epstein and Plesset's bubble dissolution model to include a term for the lipid shell resistance to gas permeation. The lipid monolayer resistance was determined as a function of lipid acyl chain length (12 to 24 carbons) by fitting the model to experimentally determined radius-time plots. The resistance increased monotonically with chain length by two orders of magnitude (1 to 500 s/cm). Shedding of excess lipid occurred in a quasi-continuous manner for lipids close to their main phase transition temperature. For more condensed lipids, the shell exhibited buckles and folds in between discrete shedding events. We proposed a qualitative mechanism of the lipid shedding process that involves zippering on the monolayer to form bilayer at a critical point. Phase, buckling, and collapse behavior of the lipid/emulsifier

system is further scrutinized by fluorescence microscopy of microbubbles and Langmuir monolayers and by Langmuir isotherms.

9:30 AM O5.4
PHOTOELECTRIC RESPONSE OF SELF ASSEMBLED FILMS OF BACTERIO-RHODOPSIN PURPLE MEMBRANE INTEGRATED ONTO SILICON SURFACE. D. Ricceri, G. Scicolone, O. Di Marco, S. Conoci and S.Coffa, Si Optoelectronics, Bio-and Nano-Systems, Corporate R&D, STMicroelectronics, Catania, ITALY; B. Pignataro, Dipartimento di Scienze Chimiche, Universita di Catania, Catania, ITALY.

The purple membrane (PM) protein bacteriorhodopsin (BR) is one of the most widely studied biomaterials. Indeed, the long-term stability against thermal, chemical and photochemical degradation along with photoelectric and photochromic properties has made this biomolecule particularly attractive for many optical and optoelectronic applications [1, 2, 3]. In these applications the preparation of well oriented PM films by using practical and simple assembling methods are crucial points to obtain efficient and economically mass-produce PM-based devices. This study reports on the photoelectrical response of bacterio-rhodopsin thin films prepared by self-assembling (SA). Self-assembled monolayers (SAMs) of 3-aminopropyltrimethoxysilane (APTS) on oxidized Silicon (100) were used as template for the electrostatic absorption of BR. The morphological properties of PM films were inspected by Scanning Electron Microscopy (SEM) and Scanning Force Microscopy (SFM) highlighting the presence of densely packed films ~500 nm thick with randomly distributed PM crystal structures ~0.5-1 mm in diameter and ~45 Å in thickness [1]. Reflectance Uv-vis spectra on these films revealed the typical BR absorption at 570 nm. In order to measure the photoelectric response a sandwich-type structure consisting of Pt/PM films/Silicon was set up. By using eother a laser monochromatic light (532 nm, 10 mW) or environmental light illuminations chopped at 0.35Hz, photoelectric responses were detected. Differential light-on and light-off photocurrent signals of about ten mA/cm2 and tens of nA/cm2 were obtained upon laser light irradiation and environmental light exposition, respectively. These results strongly suggest that the PM films have a significant orientation. The above photoelectric response coupled with the simple SA method employed are an ideal starting point to develop PM-based silicon integrated devices having efficient photoelectric responses and requiring few processing steps. [1] J.A. He et al. Adv. Mat., 1999, 11, 435-446 [2] D. Haronian et al., Appl. Opt., 1991, 30, 597-608 [3]H. Takei et al. Appl. Opt., 1993, 30, 500-509.

9:45 AM O5.5
MECHANICAL PROPERTIES AND AREA/MOLECULE OF FLUID LIPID BILAYERS IN ALCOHOL/WATER MIXTURES: THE ROLE OF ADSORPTION AND INTERFACIAL TENSION. Hung V. Ly and Marjorie L. Longo, University of California, Dept of Chemical Engineering and Material Science, Davis, CA.

Short-chain alcohols, like other solvents, can dramatically alter the normal metabolism and behaviors of cells, leading to anesthesia or loss of viability. Presumably, the mechanism occurs through modification of membrane properties and protein conformations. We show here specifically that alcohols (methanol, propanol, ethanol, and butanol) decrease the elastic moduli and toughness, and increase the area/molecule of fluid-phase membrane. The technique utilized was micropipette aspiration of giant unilamellar vesicles. We propose the observed changes are caused by decreases in the interfacial tension of the bilayer. We verify our hypothesis by first determining the interfacial tension of the bilayer from elasticity measurements and showing how it decreases with increasing alcohol concentration. Secondly, we predict the increase in the area per lipid molecule for the decrease in the interfacial tension and its value compares favorably to the area per molecule measurement obtained by the flow-pipette micropipette aspiration technique. The alcohol-induced interfacial tension reduction is related to surface adsorption in addition to bulk interfacial tension reduction.

10:30 AM *O5.6
COATING AND ENCAPSULATION TECHNOLOGIES USING LIPID MONOLAYERS AND BILAYERS: MATERIAL STRUCTURE AND PROPERTIES OF THESE BIOLOGICALLY INSPIRED MATERIALS. David Needham, Department of Mechanical Engineering and Materials Science, Duke University, Durham, NC.

The lipid bilayer membrane is a truly remarkable engineering material, –it surrounds every cell on the planet providing a mechanical, chemical, and electrical encapsulating barrier for the cell. It also acts as a 2 dimensional solvent for the protein and other lipidic components of the cell membrane. From a materials engineering perspective, we have used micromanipulation techniques, applied to giant lipid vesicles, cells and monolayers on gas microparticles, to characterize lipid-based capsular and coating structures. Each of the three basic modes of deformation, (dilational, shear and bending), have been evaluated in terms of elastic moduli and viscous coefficients. Using a suction pipet, a single lipid vesicle can be aspirated and manipulated, and several mechanochemical experiments can be performed that characterize: membrane area expansion, tensile failure and bending for solid and liquid membranes; yield shear and shear viscosity for solid phase membranes; adsorption, uptake and desorption of various membrane-binding and membrane-soluble components; membrane water permeability coefficient; transmembrane pore formation due to the action of an electric field or uptake of macromolecular polymers; and thermal bilayer transitions. The ability to manipulate individual and pairs of vesicles has also allowed: the measurement of intermembrane adhesion energy that results from the cumulation of several attractive and repulsive colloidal potentials, including van der Waals, and polymer attraction, and hydration, electrostatic, undulation, and polymer-steric repulsions; the fusion between two vesicles to be observed as a result of electroporation and defect formation due to the inclusion of non-bilayer phospholipids; and adhesion mediated by specific receptor-ligand bonds to be quantified, even down to the level of single molecular contacts. Similarly, free standing lipid monolayers can be formed on gas microbubbles in both solid and liquid states. A grain microstructure is evident, and has been shown to correlate with yield shear and shear viscosity.

11:00 AM O5.7
ORIENTED CRYSTALLIZATION OF CALCITE SINGLE CRYSTALS UNDERNEATH MONOLAYERS OF AMPHIPHILIC MACROCYCLES. Marc Fricke and Dirk Volkmer, Faculty of Chemistry, AC-1, University of Bielefeld, GERMANY.

Crystallization of inorganic solids at organized surfaces is an important process in biomineralization and crystal engineering. Nucleation and growth at the interface is often very specific and result in a particular crystal morphology or polymorph. In recent years Langmuir monolayers and self-assembled monolayers (SAMs) have been used as 2D crystallization templates which induce highly oriented crystal growth. Parameters like electrostatic potential, hydrogen bonding and interfacial molecular recognition events including geometrical lattice matching and stereochemical complementarity were discussed as crucial factors in this context. To mimic structural aspects of the interaction between acidic proteins and biogenic calcite in calcified tissues (i.e. mollusk shells), we employ amphiphilic oligoacids based on macrocyclic calix[n]arene moieties as biologically inspired supramolecular templates for epitaxial crystal growth. We have observed growth of uniformly (012) oriented calcite single crystals underneath structurally dissimilar monolayers of tetracarboxy-resorc[4]arenes and tetracarboxy-calix[4]arenes at low surface pressures. In order to address factors like non-specific electrostatic interactions and dipole potential to interfacial crystal growth, we compare the template properties of the oligoacids and the corresponding uncharged ethylester. Monolayers are spread on aqueous subphases and the resulting isotherms are analysed in terms of possible supramolecular packing arrangements. The simultaneously recorded surface potential is related to the crystallization events. The dynamic macroscopic monolayer structure and the calcium carbonate crystal growth is characterized by means of Brewster angle microscopy (BAM). The growth of uniformly oriented calcite ($CaCO_3$) single crystals underneath the monolayers is monitored in situ by (polarization) optical microscopy. The orientation of calcite crystals with respect to the monolayer is determined by means of X-ray diffraction, scanning electron microscopy and optical microscopy. Further information is gleaned from single crystal X-ray crystallography of the amphiphilic ligands.

11:15 AM O5.8
RECONFIGURABLE HYDROPHOBIC/HYDROPHILIC SURFACES BASED ON SELF-ASSEMBLED MONOLAYERS. Joanne Deval, Teodoro Umali, Esther Lan, Bruce Dunn, Chih-Ming Ho, University of California at Los Angeles, Dept of Mechanical and Aerospace Engineering and Dept of Materials Science and Engineering, Los Angeles, CA.

The development of bio-microelectromechanical systems (MEMS) for a new generation of biomedical devices is based on micromachining techniques to form micron-scale channels and reaction chambers. With large surface to volume ratios, however, surface properties play a major role in determining device characteristics. We present a strategy to reconfigure a surface from hydrophobic to hydrophilic, with the ability to render the hydrophilic surface resistant to non-specific protein adsorption. The reversible nature of the surface is made possible by using deposition and removal of biomolecules on self-assembled monolayers (SAMs) and is fully adaptable to MEMS devices. This reversibility was achieved by avidin adsorption on a hydrophobic SAM and subsequently removing the protein using detergent. Moreover, the avidin-coated surface can be made further resistant to non-specific protein adsorption by the addition of biotinylated PEG. The ability to functionalize surfaces of silicon, SiO2, and gold provides the basis for controlling fluid flow, mixing

and separation in micromachined structures.

11:30 AM *O5.9
SUPRAMOLECULAR ASSEMBLY AT INTERFACES: USING AN INTERFACE TO DIRECT THE STRUCTURE OF INORGANIC MATERIALS. Daniel R. Talham, Department of Chemistry, University of Florida, Gainesville, FL.

Many of the proposed applications of nanoscale inorganic materials are likely to require positioning the structures at interfaces. Therefore, synthetic approaches that use a surface to organize or direct the final structure should be important. Lessons from how biology creates and organizes inorganic minerals at organic interfaces can be used to guide materials synthesis. This presentation will review our recent efforts to use an organic interface as a structure-directing element in the formation of molecular and inorganic network assemblies. Two approaches for synthesizing inorganic materials at interfaces will be discussed. One approach is to use preformed organic monolayers to template nucleation and growth of inorganic crystals. Examples of relevance from materials synthesis to pathology will be discussed. In the second approach, monolayers of inorganic networks are fabricated directly at an interface. Amphiphilic metal complexes confined to the air/water interface react with ions in the subphase to form two-dimensional networks. Only amorphous colloidal products are formed in analogous homogeneous reactions, illustrating the structure-directing role of the interface. One application of these monolayer networks is for templating magnetic thin films.

SESSION O6: COLLOIDAL AND NANOSTRUCTURED MATERIALS
Chairs: Laurie B. Gower and James L. Thomas
Thursday Afternoon, April 24, 2003
Franciscan I (Argent)

1:30 PM *O6.1
BIOMIMETIC MINERALIZATION OF TYPE-I COLLAGENOUS MATRICES. M.J. Olszta, M. Sivakumar, E.P. Douglas, L.B. Gower, University of Florida, Department of Materials Science and Engineering, Gainesville, FL.

While researchers are edging closer to a complete understanding of the mechanisms surrounding naturally mineralizing collagen, such as bones and tendons, the ability to recreate these structure is still far beyond the grasp of modern science. All attempts at nucleating a mineral component intrafibrillarly within a collagenous matrix result in simple surface nucleation. We hypothesize that in bone formation, in order achieve intrafibrillar mineralization, an amorphous, liquid-phase, mineral precursor to calcium phosphate is generated outside the collagen fibrils and is drawn by capillary action into the gaps and grooves of the collagen matrix. This secondary liquid phase then subsequently crystallizes as the waters of hydration are driven off, providing the highly mineralized composite that constitutes the nanostructured architecture of bone. We have preliminary data that demonstrates this type of precursor for calcium phosphate (CaP) is possible, and can be drawn into the interstices collagen substrates. Concurrently, we have examined in greater detail mineralization with a calcium carbonate mineral precursor, in which this polymer-induced liquid-precursor (PILP) mechanism was first observed. Using a variety of type I collagenous substrates, including bovine Achilles tendon, naturally mineralizing turkey tendon, sponges, foams and thin films, we have observed non-equilibrium morphology calcite embedded within individual collagen fibers and fibrils. Additionally, calcite disks were observed to preferentially lie perpendicular to the long axis of the fibers with a banded pattern on the order of $\sim$170 - 300 μm. When the composite was deproteinated with a dilute hypochlorite solution, the banded patterns remained while the collagen fibers were removed. The calcite disks spanned the entire diameter of the collagen fibers, indicating that intrafibrillar mineralization was achieved. We have also demonstrated the ability to sequentially mineralize high surface area collagen sponges in order to produce highly mineralized $CaCO_3$-collagen composites for potential application as biomimetic, load-bearing, bioresorbable, bone-graft substitutes.

2:00 PM *O6.2
ORGANIZATION OF CELL-ADHESION PROTEINS AT SURFACES ON SUB-CELLULAR LENGTH SCALES USING COLLOIDAL PARTICLE ASSEMBLY. Nathaniel J. Gleason and Jeffrey D. Carbeck, Princeton University, Department of Chemical Engineering, Princeton, NJ.

The organization of adhesion proteins within the extracellular matrix can play an important role in determining the response of cells. Attempts to replicate features of the extracellular matrix in synthetic materials have focused on the control of protein organization either on the length scale of a whole cells (10-100 microns) or of single adhesion receptors ($\sim$1 nanometer); intermediate length scales have largely been neglected. We have developed a technique for creating tunable, hierarchical patterns of proteins using two-dimensional arrays of colloidal particles assembled on surfaces. This technique gives control over protein arrangement on length scales ranging from the size of individual particles (20 nm - 2 microns) through the size of micropatterned arrays of particles, created with soft lithography (microns to centimeters). We show that arrays of colloidal particles presenting the cell adhesion protein fibronectin (FN) supported the attachment and spreading of fibroblast cells. The behavior of adherent cells was found to be dependent on particle density; cell attachment, spreading, morphology and cytoskeletal organization changed as the particle spacing was varied from a sparse arrangement to close-packed arrays. Colloidal particle arrays were also patterned on substrates using soft lithography. The organization of cells attached to patterned particles was defined by the geometry of the particle arrays. Finally, we demonstrate the formation of more complex surfaces composed of multiple particles presenting different proteins. Surfaces functionalized with protein-coated particles will be useful for forming patterned arrays of cells and for studying the effects of substrate organization on the behavior of anchorage-dependent cells. This method also provides a general strategy for controlling the organization of proteins on multiple length scales.

2:30 PM O6.3
BIOMIMETIC SYNTHESIS OF LARGE ARRAYS OF EXTENDED AND ORIENTED NANOSTRUCTURES. Zhengrong R. Tian, Jun Liu, James A. Voigt, Bonnie Mckenzie, Sandia National Laboratories, Albuquerque, NM.

Extended and oriented nanostructures are abundant in nature. Biomaterials like seashells use organic molecules to achieve precise control of the orientations and morphologies on multiple length scales that are most absent in synthetic materials. Here we discuss the synthesis of large arrays of extended and oriented ZnO nanostructures and morphology control by tailoring the surface chemistry of the crystals. This method produced large arrays of oriented columns and layer structures showing remarkable similarity to the growth morphology of nacreous aragonite in red abalone sea-shells. A uniform coating of ZnO nanoparticles was first applied to the substrate as the nucleation seeds. Large arrays of uniform ZnO nanorods were formed in dilute aqueous solutions at low temperatures. Furthermore, we were able to control the crystal growth behavior using surface modification agents. By selectively adsorbing citrate ions to the fast growing (002) planes of the ZnO nanorods, the aspect ratios of the oriented nanorods, and the microstructure of the materials were varied over a wide range. A high citrate concentration caused a morphology transition from rod-like crystals to plate-like crystals, and produced ZnO bilayer structures containing one layer of oriented columns, and another layer of oriented nanoplates. This morphology, and the rod-to-plate transition, are commonly observed in sea-shells. The same approach is also used to prepare oriented self-assembled silicates with a wide range of morphologies.

2:45 PM O6.4
FUNCTIONAL MESOPOROUS SILICA AS INORGANIC MEMBRANES TOWARD MIMICKING BIOLOGICAL ION CHANNELS. Nanguo Liu[a], Solomon Yilma[b], C. Jeffrey Brinker[a,c], and Vitaly Vodyanoy[b]. [a]Department of Chemical and Nuclear Engineering and Center for Micro-Engineered Materials, University of New Mexico; [b]Department of Anatomy, Physiology and Pharmacology, Auburn University; [c]Sandia National Laboratories.

Biological ion channels which are 0.2-2 nm in diameter and 1-2 nm in length show high ion selectivity, random switching between open and close states, and inhibition by some ions. Because of these unique functions, they have many potential applications in biosensors, molecular switches, molecular devices, nanoreactors etc. Channel proteins carry negative and positive charges that are very important in the electrodiffusion process of ion transport. Modeling and experiments indicate that small charged pores in general might exhibit transport behaviors similar to those of biological ion channels. To mimic the structure of biological ion channels in a robust synthetic material, we used evaporation induced self-assembly (EISA) to create highly ordered mesoporous silica membranes. Direct synthesis and post-synthesis grafting techniques were employed to derivatize the pores with -COOH or -NH_2 groups which can carry positive or negative charges, respectively, enabling control of both surface charge and charge density under physiologically buffered conditions. An ordered 3-D mesophase was developed to guarantee the existence of trans-membrane pores. Pore sizes of different mesostructured silica membranes, characterized by surface acoustic wave (SAW) based N_2 sorption isotherms, were in the range 1.5 to 7 nm in diameter, depending on the surfactant template, which is commensurate with that of biological ion channels. The trans-membrane currents characterized by a patch clamp amplifier technique exhibited a similar behavior to biological ion channels and were affected by the magnitude and sign of the surface charges.

3:30 PM *O6.5
BIOMOLECULES AS A SCAFFOLDS FOR MATERIALS CHEMISTRY. Vicki Colvin, Josh Faulkner, Mary Turner, and Katy Bosworth, Department of Chemistry, Rice University; George Phillips, Jr., Department of Biology, University of Wisconsin, Madison; Tianwei Lin and Jack Johnson, Scripps Institute.

Protein crystals are large arrays of biomolecules with extensive porous volume. Typically used for biological structure determination, we show that they are useful substrates for chemistry. By cross-linking adjacent subunits, crystals are stabilized well enough to be handled without loss of diffraction. Cross-linked crystals are ideal substrates for deposition of metal or ceramic nanostructures.

4:00 PM O6.6
BIOINSPIRED DESIGN OF MULTILAYERS. M. Huang, V. Thompson[a], D. Rekow[a], Z. Suo, and W.O. Soboyejo, The Princeton Materials Institute and The Department of Mechanical and Aerospace Engineering, Princeton University, Princeton, NJ. [a]College of Dentistry, New York University, New York, NY.

This paper integrates measured properties of the mechanical properties of enamel, dentin and the dento-enamel-juctiuon into a finite element framework for the design of damage tolerance dental multilayered structures. Nano-indentation methods are used to measure the elastic-plastic and viscoelastic properties of the different components of natural teeth. The measured properties are then incorporated into finite element model of tooth structure. These models idealize natural teeth as multilayered structure. They also consider the elastic and viscoelastic deformation, and the cracking that can occur in enamel, dentin and the dento-enamel-junction. The insight developed from the models is used to guide the identification of materials and layer geometries that can result in optimal crown performance.

4:15 PM O6.7
ULTRA-THIN BIOCOMPATIBLE COATINGS FOR BIOMIMETIC NANOSTRUCTURED POLYMER SURFACES. Niels B. Larsen, David Selmeczi, Jiang Wei, Peter Kingshott, Stephan Mosler, Risoe National Laboratory, Danish Polymer Centre, Roskilde, DENMARK; Nikolaj Gadegaard, Mathis Riehle, Adam Curtis, Chris Wilkinson, Glasgow University, Centre for Cell Eng., Glasgow, UNITED KINGDOM; Hans Griesser, Uni. South Australia, Ian Wark Res. Inst., Mawson Lakes, AUSTRALIA.

Polymer surfaces presenting nanostructured topography and chemistry that mimics nature are of great interest for tissue and cell engineering. We have recently demonstrated the feasibility of producing large numbers of thermoplastic polymer objects possessing nanostructured topography that mimics the sub-50 nanometer scale characteristics of fibrillar collagen. The general cell incompatibility of thermoplastic materials calls for reproducible and high-throughput methodologies for inducing biocompatibility while still presenting the underlying nanometer scale topography. We have pursued both wet chemical methods, plasma treatment in non-polymerizable gasses, and plasma-assisted polymerization to achieve this on a range of different thermoplastic materials. Our results, to be presented in this paper, show that all three methodologies are capable of inducing cell compatibility but that wet chemistry often covers the underlying topography at the nanometer scale while plasma treatment of some polymeric materials tends to be unstable in aqueous environments. Plasma-assisted polymerization currently appears to be the most promising candidate for application of stable, cell compatible, nanometer thick chemical coatings that presents the nanotopography of their supports faithfully.

4:30 PM *O6.8
INKJET PRINTING AS A MIMIC FOR BIOLOGICAL GROWTH. Paul Calvert, University of Arizona, Dept. of Materials Science and Eng, Tucscon, AZ.

Many biological tissues form by extracellular deposition of polymers and minerals adjacent to a layer of cells. Soluble reagents are secreted and then interact in the surrounding matrix to form insoluble materials. This process does have to be highly controlled in order to allow structures to develop to full density in a zone which extends away from the cell surface. An inkjet printer delivers drops of solution with a diameter of about 50 microns, similar to the size of a cell. It should be possible to mimic some biological growth processes by the sequential printing of a series of solutions, including precursors of structural polymers, modifying catalysts and inorganic salts. As these drops arrive at the substrate and dry, layers of about 100nm thickness are formed. Rapid diffusion between layers allows chemical reactions to occur between sequential drops. This approach has been applied to printing gels, self-assembling pairs of polymers, particles and minerals. Comparing this approach with what we know of tissue growth brings out the potential importance of timing of secretion, diffusion rates and reaction rates in biological growth.

SESSION O7: POSTER SESSION
MATERIALS INSPIRED BY BIOLOGY
Chairs: James L. Thomas and Laurie B. Gower
Thursday Evening, April 24, 2003
8:00 PM
Golden Gate (Marriott)

O7.1
MOLECULAR DYNAMICS STUDY OF SURFACE-TETHERED 1-THIAHEXA (ETHYLENE OXIDE) CHAINS TERMINATED BY METHYL GROUPS: HELIX FORMATION AND THERMAL DISORDER. Joseph B. Hubbard[a] and Raymond D. Mountain, Physical and Chemical Properties Division; Curtis W. Meuse[a]. [a]Biotechnology Division, National Institute of Standards and Technology, Gaithersburg, MD.

We present the results of a molecular dynamics study of a set of 225 surface-tethered 1-thiahexa(ethylene oxide) chains terminated by methyl groups. The detailed morphology of such monolayers (on gold) is currently believed to play a major role in conferring resistance of the surface to non-specific protein adsorption. We find that spontaneous helix formation and finer details of helix morphology are sensitive to the partial charge assignments ascribed to oxygen atoms and to the methylene groups. The effects of varying surface coverage as well as chain-surface interaction strength indicate, in accord with infrared spectroscopic studies, the presence of a set of approximate 7/2 helical structures oriented normal to the surface, even though thermal disorder clearly precludes a description based on the concept of a perfect crystalline monolayer. Thermal fluctuations in chain morphology in the vicinity of the terminal methyl groups lead to the exposure of oxygen to the external environment. We also find that the persistence of compact helix-containing domains at partial surface coverages results in the formation of well-defined cavities or void regions which expose the bare surface, even in the presence of strong chain-surface attractive interactions.

O7.2
PDMS SURFACES DERIVATIZED WITH NON-STICK PHOSPHOLIPID MEMBRANES. K. Scott Phillips and Quan Cheng, University of California, Dept of Chemistry, Riverside, CA.

Materials that show superior mechanical, optical and chemical properties are desirable for construction of novel biosensors and microchips. Rapid prototyping methods with polydimethylsiloxane (PDMS) yield promising materials that are moldable, transparent, annealable, and surface modifiable. Intricate micro-array patterns can be created in three dimensions at low cost. The main drawback of PDMS is the native hydrophobicity of the surface that causes sample loading problems and non-specific protein adsorption. Attempts to create a hydrophilic surface have included grafting, PEMS, blocking cocktails and trialkylchlorosilane monolayers. We report here two novel methods using vesicle fusion techniques that result in long-standing hydrophilic properties of PDMS blocks. Both methods require a short plasma cleaning step to generate reactive functional groups prior to surface modification. In the first approach, bilayers composed of egg phosphatidylcholine self-assemble directly onto the PDMS surface through vesicle fusion. Equilibrium contact angle measurements showed that the modified surface maintained a contact angle of 31° when rehydrated with water up to two hours after bilayer modification, and an angle of $<40°$ for up to two days. Unmodified PDMS surfaces returned to an angle of 80° within only 30 minutes and 93° after two hours. Fluorescence after photobleaching (FRAP) experiments, aimed at understanding the fluidity of the bilayers on PDMS, will be discussed. The second method uses hybrid membranes composed of octadecyltrichlorosilane (OTS)/PC monolayers, similar to those adopted in developing supported bilayer membranes. Optimum SAM deposition and vesicle fusion times were obtained by using SPR to monitor the change in thickness of OTS and lipid layers. A high resolution confocal fluorescence scanner was used to quantitate bilayer stability and non-specific adsorption of fluorescent proteins in microchannels. Preliminary work on using the modified PDMS to develop a protein toxin biosensor with electrochemical detection will be discussed.

O7.3
PERIODICALLY PATTERNED CALCIUM CARBONATE THIN FILMS FORMED THROUGH SELF-ORGANIZATION PROCESSES. Ayae Sugawara, Takeshi Ishii, Takashi Kato, Univ of Tokyo, Dept of Chemistry and Biotechnology, School of Engineering, Tokyo, JAPAN.

Organisms produce elaborate inorganic/organic nanocomposites such as shells, pearls, bones, and teeth. Their controlled, hierarchical

structures provide remarkable strength and/or optical properties. From the viewpoint of materials chemistry it is of interest to synthesize such regular structures through self-organization processes by using inorganic-organic interactions as seen in biomineralization processes. However, the preparation of controlled structures on submicrometer length scale by self-organization methods is difficult. We synthesized thin film crystals of calcium carbonate with a homogeneous thickness by bio-inspired processes, that is, using cooperative effects of water-insoluble and soluble polymers. For example, calcium carbonate thin films, about 800 nm thick, are formed on chitosan solid matrix from the solution containing poly(acrylic acid). Here we report on the spontaneous formation of periodically patterned calcium carbonate thin films. Hydrophobized pullulan containing six cholesterol groups per 100 glucose units (CHP) was employed as a new solid matrix on which calcium carbonate was crystallized. The CHP matrix was prepared by the spin coating of its solution on glass substrate. It was immersed in calcium chloride solution containing poly(acrylic acid). Calcium carbonate crystallization was induced by slow diffusion of ammonium carbonate vapor into calcium chloride solution at room temperature. Surprisingly, calcium carbonate thin film crystals with patterned surface were developed spontaneously on CHP matrix. Scanning electron and atomic force microscope observations reveal that periodic concentric ring patterns, about 700 nm pitch, are formed on the surface of the thin film crystals. Since the periodic patterns serve as diffraction gratings, the thin films appear to be iridescent. At present it is not clear why such patterns are formed on CHP matrix. Further study on the relationships between the structure of the CHP matrix and the calcium carbonate pattern is in progress.

O7.4
CONTROL OF NUCLEATION AND GROWTH OF PATTERNED CALCIUM CARBONATE FILMS ON SELF-ASSEMBLED MONOLAYERS (SAMS) VIA A POLYMER-INDUCED LIQUID-PRECURSOR PROCESS. Yi-yeoun Kim, Laurie B. Gower, Univ of Florida, Dept of Materials Science and Engineering, Gainesville, FL.

In mimicking the biological processes of mineralization, organic materials can be used both as a template for the controlled nucleation of inorganic crystals, as well as a process-directing agent to transform the solution crystallization into a precursor process. In the latter case, soluble acidic macromolecules can lead to a polymer-induced liquid-precursor (PILP) process, which could provide a means for accomplishing the deposition of mineral films because the liquid-phase precursor can be manipulated and shaped into a variety of non-equilibrium crystal morphologies under low-temperature and aqueous-based conditions. Our approach to patterning thin inorganic films is to use the PILP process to direct the deposition of precursor phase on SAM templates. Self-assembled monolayers (SAMs) patterned by micro-contact printing are able to act as synthetic templates for biomimetic mineralization, mimicking the role of biological organic matrices, which confine spatially the deposition of inorganic minerals, as well as template the nucleation and growth of the crystals. Therefore, we believe that cooperation of the functionalized endgroup of the SAMs and acidic macromolecules provides a useful biomimetic model system for investigating crystal nucleation and growth, and phase transformations and morphology of mineral phases. In our previous report, we demonstrated that the mineral phase, calcium carbonate, can be deposited onto specific areas templated with self-assembled monolayers of alkanethiolate on gold under constrained conditions via the PILP process. In this report, the focus is on controlling the nucleation and growth of patterned calcium carbonate films, which exhibit quite different behavior depending on numerous factors, such as impurity concentration, surfactants with different functionalized groups, pattern shape and dimension, polyelectrolyte concentration, and mineral solution supersaturation level. We believe the PILP process, in combination with the appropriate template, offers a viable means for mimicking the complex structures found in biominerals.

O7.5
A QUANTITATIVE UNDERSTANDING OF THE EFFECT OF ANTIFREEZE PROTEINS ON ICE CRYSTALLIZATION. Ning Du and X.Y. Liu, National University of Singapore, Dept of Physics, SINGAPORE.

Antifreeze proteins allow certain organisms to survive the cold winter. The one of the best known examples to date comes from polar and subpolar fish. There are several commercial applications of these antifreeze proteins based on their effectiveness in inhibiting ice growth, such as maintaining texture in frozen food, improving storage of blood, tissues and organs, and protecting the crops from freezing. Although much work has been published on the structure and behavior of antifreeze proteins, (1-3) there remains a lack of consensus on some of the fundamental issues to do with antifreeze action.(4) In nature, freezing in micro-sized water droplets occurs very frequently, such as hail and snow in climatic systems as same as freezing in micro-sized biological systems. For better understanding the antifreeze mechanism of the antifreeze proteins on ice crystallization in similar system, a so called "controlled ice nucleation in a micro-sized water droplet" technique has been developed by us.(5-7) This technique enabled us to quantify the effect of antifreeze proteins on ice nucleation kinetics. To explore the role of impurities or proteins on ice crystallization, we will tackle this issue based on a newly published model.(8) The effects on nucleation can be classified into (a) the interfacial nucleation barrier lowering effects and (b) the kink integration effect. The second effect includes that the effect of desolvation of additives or impurities from the adsorption sites of embryo surface, and the induced pre-ordering of liquid at the substrate-fluid interface. Antifreeze proteins inhibit ice nucleation not only by adsorbing on the surface of ice, which suppresses the interface epitaxial effect but also by adsorbing on the surface of foreign particle, which enhancing the desolvation energy barrier. The quantification of these effects will allow us to identify and design the most efficient nucleation promoters and inhibitors. References (1) Liou, Y.C., Tocilj, A., Davies, P.L. & Jia, Z. Mimicry of ice structure by surface hydroxyls and water of a b-helix antifreeze protein, Nature 406,322-324 (2000) (2) Sidebottom, C. et al. Heat-stable antifreeze protein from grass, Nature 406, 256 (2000) (3) Graether, S.P. et al. b-helix structure and ice-bonding properties of a hyperactive antifreeze protein from an insect, Nature 406, 325-328 (2000) (4) Charles A. Knight, Adding to the antifreeze agenda, Nature 406, 249-251 (2000) (5) Du Ning, X.Y. Liu, Controlled ice nucleation in microsized water droplet, Appl. Phys. Lett. 81, 445-447 (2002) (6) Elizabeth A. Shack, Protein promotes nucleation in water microdroplets, in MRS bulletin 27 (8), pp. 586 (2002) (7) Ed Gerstner, The little chill, in Research Highlights of Nature Physics Portal and Nature Materials Update 18 July 2002 (8) X.Y. Liu, Heterogeneous nucleation or homogeneous nucleation? J Chem. Phys. 112, 9949-9955 (2000).

O7.6
CRYSTAL MODIFIERS: USING SIMPLE ORGANIC MOLECULES TO MANIPULATE GROWING CALCIUM CARBONATE. Steven R. Dickinson and K.M. McGrath, Otago Univ, Dept of Chemistry, Dunedin, NEW ZEALAND.

The past decade has seen a noticeable rise in the profile of biomineralisation and synthetic inorganic/organic composites. This has seen the establishment of many new synthetic pathways in the formulation of unique materials. Examples include compressed organic monolayers as templates for crystallisation and surfactant mediated transport and eventual fusion of small crystallites. By unveiling the mechanistic pathways to biomaterial formation utilised by organisms, materials that mimic in some way these natural analogues have been prepared. However, full replication is in most cases beyond our synthetic capabilities. Increased knowledge of the fundamental processes underlying biomineralisation will aid greatly our ability to design and synthesise useful and novel materials. Calcium carbonate is one of the predominate minerals deposited by organisms, its use ranging from full skeletal support to gravity reception. The organisms have the ability to manipulate and control both the polymorphic form and the final morphology of the crystal, over a range of different length scales, resulting in a plethora of shapes and structures. The work presented here outlines an investigation into the effects of -OH moiety containing molecules on the nucleation and growth of unseeded calcium carbonate crystals. The crystals were grown on silica in an aqueous environment. A series of simple organic molecules containing -OH groups including alcohols and saccharides were used as crystal modifiers. It was found that by using short chain alcohols to control solution viscosity a new morphological form of calcite, known as a hopper, could be grown. This crystal is transient and thermodynamically unstable, hence reaction conditions must be carefully regulated for the form to be expressed. By restricting the -OH moieties into defined positions using saccharides, control over the polymorphic ratio was obtained. In general, saccharides promote calcite growth. However, it was observed that for a very narrow range of sugar concentrations this trend, of calcite stabilisation, could be reversed; with the kinetically favoured vaterite becoming prevalent.

O7.7
POLYMER-MEDIATED CRYSTAL GROWTH: A STUDY OF DESIGN CRITERIA. Nicholas B. Dinsdale, Brigid R. Heywood, Keele University, School of Chemistry & Physics, UNITED KINGDOM; W. James Feast, Joanna L. Megson, IRC, Durham University, UNITED KINGDOM.

It is well documented that ordered organic systems can facilitate the oriented nucleation of crystals to give control of polymorph identity and crystal size and yield novel habits and morphologies. Moreover, it has been argued that the control of crystallisation in these systems is effected by the organic compounds functioning as templates, mimicking particular crystal facets and transferring structural

information to the nascent crystal. One of the issues to be explored in this context is the chemical design of the organic species. In order to establish the relative importance of various criteria, a series of novel polymers were produced using ring opening metathesis polymerisation to control molecular weight. These polymers were then functionalised with a selection of reagents to allow for an exploration of such issues as their consensus polarity and relative hydrophobicity/hydrophilicity. Other factors to be investigated were the identity and activity of selected pendant functional groups and their positional density. All polymers were then assayed for their ability to direct the crystallisation of calcium carbonate. The results demonstrate that crystal formation (including polymorphism, habit and morphology) can be specifically regulated by varying the molecular design of a polymer. The impact upon the crystal can be regulated by subtle variations with crystal orientation being dictated by the stereochemical motif generated by an appropriately functionalised polymer matrix.

O7.8
AMELOGENIN INDUCES BIOMINERALIZATION AT SPECIFIC PH. Stefan Habelitz, Sally J. Marshall, Mehdi Balooch, Grayson W. Marshall University of California, Department of Preventive and Restorative Dental Sciences, San Francisco, CA; Wu Li and Pamela K. DenBesten, University of California, Department of Growth and Development, San Francisco, CA.

Proteins of the extracellular matrix (ECM) organize and control the calcification of enamel with a nanoscale precision, which exceeds the capabilities of human engineering, and generate a 95% mineralized tissue of extraordinary toughness consisting of the largest biological hydroxyapatite crystals organized in a remarkable microarchitecture. The use of these proteins for in-vitro biomineralization opens new pathways for materials synthesis with structural design on the molecular level at ambient temperatures and pressures. We studied nucleation and crystallization of apatite on bioactive glass-ceramic surfaces in the presence of recombinant human full-length amelogenin (rH174), which is the major component of the ECM of the developing enamel. The glass-ceramic substrates contains rod-like oriented fluoroapatite crystals (FAp) with 1 μm diameter. Topographic images by atomic force microscopy, revealed (001) and (hk0) planes of FAp at about 1 to 5 nm below the glass-level on polished samples. Substrates were immersed into 400 μL of buffered solutions at 37°C and pH between 6.5 and 9. Without the presence of rH174, FAp crystals grew in 24 h up to 20 nm above the glass matrix. The final height increased linearly with the pH, according to an increase in the degree of saturation. In the presence of 0.4 mg/ml rH174, the (001)-faces of FAp grew to a similar height as observed without protein. Only at pHs around 8, non-linear behavior of crystal growth was observed. (001)-faces of FAp grew up to 400 nm height and thus up to 20 times higher then in protein-free solutions. Protein was found to bind specifically to FAp but not to the surrounding glass matrix. The (hk0)-faces of apatite grew only about 30 to 60 nm, but showed aligned spherical structures, presumably amelogenin nanospheres. Nanoindentations on the biomineralized layers showed elastic moduli and hardness values of 78.9 $\pm$8.3 and 2.4 $\pm$0.3 GPa, respectively, which closely resembled the properties of natural human enamel. The high specifity of amelogenin for FAp combined with an increased activity in biomineralization at a certain pH range is assumed to be the result of structural changes of the protein induced by surface charges. We conclude that a specific ionic environment might be required to promote amelogenin guided crystal growth and biomineralization. Support: NIH/NIDCR P01-DE09859, PO1-DE11526, UCSF Academic-Senate, UCSF-DDCF12.

O7.9
THE INFLUENCE OF CADMIUM AND ZINC ON THE BIOMIMETIC MINERALIZATION OF DICALCIUM PHOSPHATES IN SEMI SOLID MEDIUM. G.R. Sivakumar, Matt Olszta, Laurie B. Gower, University of Florida, Department of Material Science and Engineering, Gainesville, FL; S. Narayana Kalkura, Crystal Growth Centre, Anna University, Chennai, INDIA.

The nucleation, growth and dissolution of dicalcium phosphates ($CaHPO_4$-DCP, $CaHPO_4.2H_2O$-DCPD) play an important role in the biological mineralization process as well as the setting of a variety of calcium phosphate cements for orthopedic and dental uses. The influence of inorganic ionic species (cadmium and zinc) on the nucleation, growth, morphology and microstructure of the DCP and DCPD crystals were investigated in a semi solid medium using a single diffusion method. Incorporation of cadmium ions in the crystallizing medium led to a change in the size and microstructure of the DCP, and further, it favored the formation of curved DCP crystals. A slight variation was observed in the length and breadth of the DCPD crystals at a higher concentration of cadmium ions. Presence of zinc ions in the medium favored the primary agglomeration of DCPD crystals and changed the unit cell dimensions of DCPD. Presence of zinc ions altered the surface morphology, size and microstructure of the DCP crystals. The size of spherulitic DCP crystallized in the presence of cadmium was found to be larger than that of DCP crystallized in the presence of zinc ions. The compositional analysis by inductively coupled plasma atomic emission spectroscopic analysis (ICPAES) revealed that the amount of cadmium and zinc ions increased with an increase of these ions in the crystallizing medium. These preliminary studies might be used to understand the mechanism involved in the transformation of dicalcium phosphates to hydroxyapatite, the mineral present in bone, teeth and kidney stones.

O7.10
INTRAFIBRILLAR MINERALIZATION OF COLLAGEN WITH HYDROXYAPATITE USING A LIQUID-PHASE MINERAL PRECURSOR. M. Sivakumar, M. Olszta, F. Amos, and L.B. Gower, Dept of Material Science and Engineering, University of Florida, Gainesville, FL.

The synthesis of complex inorganic forms which mimic natural structures offers exciting avenues for the chemical construction of macrostructures and a new generation of compositionally and structurally related bone analogs for tissue engineering. Synthetic bone replacement materials such as calcium phosphate are now widely used in hard tissue repair and regeneration. In this study, in situ mineralization of calcium phosphate and the role of acidic macromolecules (biopolymers) on initial nucleation, crystallization and physico-chemical properties of calcium phosphates under physiological condition has been studied. The novelty of our approach is that calcium phosphate crystals are prepared by a Polymer-Induced Liquid-Precursor (PILP) mineralization process using acidic biomimetic polymers, such as poly(aspartic acid) and poly(vinyl phosphonic acid), to induce a liquid-phase precursor to calcium phosphate mineral. Initial observation by optical and electron microscopy studies shows that nano-size PILP droplets are formed within the aqueous solution. The mineral formed by this process was characterized by various techniques, such as XRD, FT-IR, EDS and electron diffraction. It is evident from the XRD and FT-IR data that the crystal structure and characteristic groups (phosphate and hydroxyl) present in the final solidified products are the hydroxyapatite phase of calcium phosphate, which was further confirmed from energy dispersive spectrum and electron diffraction data. Work is in progress to mineralize these calcium phosphate crystals in the presence of various type-I reconstituted collagen substrates (i.e. foams and sponges), and the initial data confirms the calcium phosphates are mineralized within the collagen fibrils as well as on the surface. Our goal is to mimic the nanostructured architetecture produced in naturally mineralizing animal and human hard tissues (i.e. aligned and oriented HA nanocrystals embedded within collagen fibrils), which we hypothesize forms via a PILP process, in which the liquid phase mineral precursor is drawn into the holes zone of collagen fibrils via capillary action.

O7.11
INFLUENCE OF DESIGNER PEPTIDES AND NOVEL PROTEINS ON IN VITRO NUCLEATION AND MORPHOLOGY OF CALCIUM CARBONATE CRYSTALS. Suresh Valiyaveettil, Lakshminarayanan Rajamani, Boon Tee Ong, Ajikumar Parayil, R. Manjunatha Kini, National University of Singapore, Department of Chemistry, Singapore, SINGAPORE.

Calcium based hard tissues play important functions such as skeletal support, protection of soft tissues, in many organisms. Many elegant research are carried out to understand the molecular mechanism of such tissue generation in nature. We are interested in establishing a clear mechanism in which proteins or peptides play an important role in nucleation and morphology control of calcium carbonate crystals. The talk will focus on our recent results on proteins extracted from goose eggshells and the peptides designed based on the known sequence of such eggshell matrix proteins.

O7.12
BIOMIMETIC SYNTHESIS OF DIFFERENT POLYMORPH THIN FILMS OF CALCIUM CARBONATE VIA A POLYMER-INDUCED LIQUID-PRECURSOR (PILP) PROCESS. Lijun Dai, Laurie B. Gower, University of Florida, Dept. of Materials Science and Engineering, Gainesville, FL.

In nature, biomineralization process usually involves both water soluble and insoluble matrix, selectively forming mineral layers of some polymorph. We believe these polymorph controls in nature are via a polymer induced liquid precussor process in which both the soluble and insoluble polymers play their impotant roles in controlling polymorph and morphology of the minerals. In order to achieve mineral film of certain polymorph with high purity, we systematically examined the following factors include the time when the substrate is introduced into crystallization solution, the sequence of addition of reactants, different type of substrates, degree of supersaturation, stoichometry ratio of CO_3^{2-}/Ca^{2+}, polyacrylic acid concentration.

We have been able to selectively synthesize calcite, vaterite, aragonite, or monohydrocalcite thin films by changing these factors. X-ray diffraction, FTIR, optical microscopy, SEM and TEM were used to monitor and characterize the dynamic changes of these biomimetic mineralization processes. Both vapor diffusion and dropwise addition method were used to synthesize thin films of calcium carbonate in the presence of different organic substrates, including PVA, gelatin, surface modified PVA, and PVA/Gelatin composites, by a polymer-induced-liquid-precursor-(PILP) process. The polymeric inducers include polyaspartate and polyacrylic acid, as well as specially designed polypeptide additives. By changing polymeric additive concentration and rate of increasing supersaturation (via vapor diffusion of ammonium carbonate), relatively high fractions of aragonite or vaterite thin films are deposited on PVA substrates. The gelatin substrate was less able to stabilize vaterite than PVA. A composite of PVA and gelatin was found to preferentially induce aragonite. By introducing magnesium ion into the mineralization solution (e.g. Mg^{2+}/ Ca^{2+} equal 0.8:1) in the presence of PVA substrate, high magnesium-bearing calcite tablets were formed first, followed by monohydrocalcite thin films. By elevating the Mg^{2+}/ Ca^{2+} ratio to 2:1, a continuous thin film of pure monohydrocalcite forms on the PVA substrate, even in the absence of polymeric additive. By immobilization of polyaspartate onto PVA surfaces, which mimicks the sandwich structure of the organic matrix of nacre, an aragonite thin film was formed, which contains at least 95% aragonite. As an alternative method for raising the supersaturation, a drop-wise addition of calcium chloride or ammonium carbonate was utilized in the presence of polyacrylic acid in order to systematically examine the controlling factors that dictate the polymorph and morphology of thin films deposited on PVA or surface modified PVA substrates.

O7.13
IN-VITRO CRYSTALLIZATION OF NEWBERYITE AND STRUVITE CRYSTALS IN THE PRESENCE OF INORGANIC TRACE ELEMENTS. G.R. Sivakumar, Laurie B. Gower, University of Florida, Department of Material Science and Engineering, Gainesville, FL; S. Narayana Kalkura, Crystal Growth Centre, Anna University, Chennai, INDIA.

The in-vitro studies on newberyite ($MgHPO_4.3H_2O$) and struvite $MgNH_4PO_4.6H_2O$) crystals have received considerable attention as a crystalline constituent of human non-metabolic urinary stones. Multifaceted newberyite crystals were grown from the decomposition of struvite crystals in a colloidal medium in the presence of calcium, cadmium and zinc ions at physiological pH (7.4) and temperature (37°C) by a controlled and slow chemical reaction method. The presence and absence of ammonia in the crystals were studied using fourier transform infra-red analysis (FT-IR). Further struvite and newberyite were distinguished from each other through their weight loss using thermal analysis (TGA/DTG and DTA) The brushite ($CaHPO_4.2H_2O$) crystals were found to crystallize along with struvite and newberyite crystals at higher concentration of calcium ions. Presence of cadmium in the crystallizing medium uniformly enhanced the growth rate along different faces of the newberyite and struvite crystals. Agglomerated newberyite was found to grow in the presence of zinc ions, and its presence reduced the growth rate along (001) face of the struvite crystals. The inductively coupled plasma atomic spectroscopic technique was employed to quantitatively confirm the presence of calcium, cadmium and zinc ions in the crystalline lattice of the newberyite and struvite crystals. Single crystal x-ray diffraction technique was employed to calculate the lattice parameters of the crystals grown in the presence of calcium, cadmium and zinc ions.

O7.14
ZEBRAFISH, A NEW MODEL FOR STUDYING COLLAGEN MINERALIZATION AT MOLECULAR AND GENETIC LEVEL. Y. Zhang, F.Z. Cui, X.M. Wang, Q. L. Feng, Biomaterials Group, Department of Materials Science and Engineering, Tsinghua University, Beijing, CHINA.

Collagen mineralization is a fundamental process for the formation of bones and teeth. Great interest has been simulated for its promising application in the fields of tissue engineering and biomaterials. However, it is a complicated process that involves multi-molecular recognitions and interactions between collagen and non-collagen proteins, protein-crystals, crystal-cell surface. The understanding of the collagen mineralization is far from clear. In order to study the collagen mineralization at molecular and genetic level, choosing suitable model is of the first importance. Here we will report our results of studying collagen mineralization using a new model, Zebrafish (Danio rerio), an emerging model of development riding the next wave of genome-wide exploration, which provides a single opportunity to investigate the underlining molecular processes for vertebrate-specific collagen mineralization. A host of mutations that disrupt the development of organs and tissues in the zebrafish will help identify the genes and molecular processes that guide vertebrate collagen mineralization. Transmission Electron Microscope (TEM), thermal and nano-indentation analyses of the skeleton of zebrafish show that this bony fish has a collagen-hydroxyapatite system with same features as human bone mineralization, but a relatively simple structure. A kind of skeletal mutation, Stöpsel dtl28d zebrafish was found to be with defective collagen fibrils and abnormal mineralization. The defects involve abnormality of diameter and organization of collagen fibrils and alteration of mineralization in terms of mineral crystallinity, mineral phase, Ca/P molar ratio, mineral size, and precipitated location of minerals. Further study using nanoindentation disclosed that the bone of zebrafish becomes brittle after stp gene-mutation. All these features show an interesting similarity to those corresponding problems in human osteogenesis imperfect (OI), suggesting that zebrafish is not only a good model system for studying bone mineralization at up to gene level but might also be a promising model for studying human being related bone diseases.

O7.15
COLLOIDAL PREPARATION OF γ-Fe_2O_3@Au [CORE@SHELL] NANOPARTICLES. Jiye Fang, Eun Young Shin and Charles J. O'Connor, Univ of New Orleans, Dept of Chemistry/AMRI, New Orleans, LA; Jibao He and Deborah Grimm, Tulane University, CIF, New Orleans, LA.

To date development of (magnetic-core)@(gold-shell) structured nanocomposites have attracted more attention due to the requirement of advanced manipulation in bio-system. Presently, most related groups are concentrating their efforts on exploring the fabrication technique based on magnetic metals as the core materials. In order to skip the possible oxidation problem (by air and by $Au^{(}3+)$) related with magnetic metals, we alternatively proposed a new design using γ-Fe2O3 nanocrystals as core candidates and demonstrated the preparation of (γ-Fe_2O_3-core)@(Au-shell) nanoparticles employing a colloidal microemulsion technique. γ-Fe_2O_3 core nanocrystals (12-14 nm in size) were prepared and separated through a modified high temperature organic colloidal approach based on T. Hyeons work. We subsequently transferred the γ-Fe_2O_3 nanocrystals (core materials) into our recently established waterless-microemulsion system containing alcohol-octane-nonionic surfactant. $HAuCl_4$ was pre-dissolved into degassed alcohol phase and the formation of Au layer was assisted by an UV irradiation for 48 hours (254 nm, 560 watt). The preliminary characterization reveals that we have progressively achieved this design and successfully gained (γ-Fe_2O_3-core) @ (Au-shell) nanoparticles. The Au/Fe composition ratio was determined by EDS, and the core@shell structure was clearly exhibited by TEM image. Our further investigation also indicates that the wavelength of the UV-Vis absorption on these (γ-Fe_2O_3-core) @ (Au-shell) nanoparticles is dependent on the percentage of Au coated.

O7.16
SYNTHESIS OF CALCIUM CARBONATE COATED NANOPARTICLES FOR DRUG DETOXIFICATION. Debra Lush, Vishal Patel, Allison Kurz, Piyush Sheth, Javier Gutierrez, and Laurie Gower, Department of Materials Science & Engineering, University of Florida, Gainesville, FL.

Nanoparticulate systems are being developed for use in pharmaceutical and industrial controlled release applications. In the Unites States, over 300,000 patients enter the emergency room each year due to complications from overdose of prescription drugs. In fact, the leading method of suicide is via overdose of amitriptyline, a 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 blood 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 solution of calcium chloride, magnesium chloride, and a highly acidic polymer. Stearic acid and oil are dispersed in water as emulsion particles, which are then coated with a mineral shell of calcium carbonate to yield the particles of interest. Particles on the order of 1 to 3 μm have successfully been synthesized, with shells of 200 to 500 nm thickness. Current experiments on these particles are directed at further characterization and optimization of the calcium carbonate shells, in vitro testing of drug uptake capabilities, synthesis of similar core-shell particles on the nanoscale dimension using microemulsion particles, and templating porosity of the inorganic shell through binary surfactant systems.

O7.17
Abstract Withdrawn.

O7.18
PEG-LIPIDS, OLIGO(ETHYLENE GLYCOL) SURFACTANTS AND PLURONIC BLOCK COPOLYMERS ENHANCE ULTRASONIC PERMEABILIZABILITY OF LIPOSOMES. Hung-yin Lin and James L. Thomas, Department of Chemical Engineering, Columbia University, New York, NY.

Poly(ethylene glycol) (PEG) coated liposomes are used to prolong the lifetime of microcapsules in drug delivery systems. In this study, the susceptibility of PEG-containing liposomes to permeabilization by ultrasound at 20 kHz was investigated. Liposomes containing PEG-lipids, or control liposomes composed of phosphatidylcholine (with and without cholesterol) showed low responsiveness to very low levels of ultrasound. However, above a threshold intensity of about 2 W/cm^2 PEG-containing liposomes showed up to a ten-fold enhancement in permeabilization, as measured by fluorescent dye leakage, compared with controls. Studies with low concentrations of ethylene-glycol in the liposome buffer (Triton X-100, X405 and Tween 20, 80) showed that these surfactants are also able to dramatically increase susceptibility to insonation, at concentrations that caused no observable increase in permeability in the absence of ultrasound. Pluronic copolymers with PEO blocks of differing sizes were also found to sensitize liposomes to insonation. Pluronic P105 was also incorporated into the liposomes during formation and extrusion, and these liposome-polymer complexes retained high ultrasound responsivity. Interestingly, the ultrasound responsivity of the Pluronic-liposome complex increased significantly with temperature. This temperature effect may indicate a change in the disposition of the polymer in the membrane as the hydrophobicity of the PPO block increases. Enhanced ultrasound-mediated release may prove useful for targeted drug delivery applications. (Support by The Whitaker Foundation #RG-99-0427.)

O7.19
SURFACE PLASMON RESONANCE SPECTROSCOPIC STUDY ON PORE-FORMING BEHAVIOR OF STREPTOLYSIN O (SLO) ON SUPPORTED PHOSPHOLIPID BILAYERS. Thomas Wilkop, Danke Xu, Quan Cheng, Department of Chemistry, University of California, Riverside, CA.

Streptolysin O (SLO) is a four domain protein toxin permeabilising cell membranes. Oligomeric transmembrane pores are formed after monomer units of the toxin bind to cholesterol imbedded in the cell membrane and assemble into ringlike structures with diameters of up to 30 nm. Supported lipid membranes offer a convenient system for the study of protein toxins with the toxin/membrane interaction being detectable in several ways. The lipid bilayer structure has to fulfill a variety of criteria such as membrane fluidity, stability, high homogeneity and high surface coverage to successfully mimic the toxins' natural target, the living cell and to provide reproducible results. We have investigated two membrane systems on gold substrates consisting of a self-assembled hexyl thioctate (HT) layer, with a fused vesicle layer of DMPC on top. In system A the DMPC is incubated for 30 minutes at 37 degrees C with SLO, which leads to perforated vesicles, as observed by transmission electron microscopy, that fuse onto the HT. In system B the intact DMPC vesicles undergo membrane-forming fusion with SLO subsequently binding and inserting itself into the membrane. A complete characterization of the membrane system is performed using cyclic voltammetry, impedance spectroscopy and mostly surface plasmon resonance (SPR) spectroscopy. The stepwise analysis of the SPR spectra based on the theoretical multilayer reflectivity model and the Fresnel equations allows us to determine the thickness, dielectric constant, extinction coefficient and the short term adhesion stability of the individual layers. The binding kinetics of the toxin to the membrane was studied, and an estimate of the insertion depth of the SLO in case of incomplete permeation was derived. Combined with electrochemical studies, which provided permeability data, a comprehensive understanding of the toxin insertion and the mechanism of the vesicle fusion was obtained.

O7.20
BIOSURFACTANT PRODUCED UNDER NONASEPTIC FERMENTATION FOR SOLUBILIZING CHLORINATED SOLVENTS. Cumaraswamy Vipulanandan, Shyh-Yau Wang, Univ of Houston, Dept. of Civil & Environmental Engineering, Houston, TX.

A biosurfactant was produced in continuously stirred batch reactor using the Flavobacterium sp., which was isolated from a contaminated field sample. The biosurfactant was produced from used-vegetable oil under nonaseptic fermentation condition and then precipitated with a 1 N NaOH solution and was used in this investigation. The biosurfactant had a CMC of 0.7 g/L and lowered the surface tension to below 29 dynes/cm. Surface tension of the biosurfactant solution showed greater dependency on the pH as compared to SDS and Triton X-100. The interfacial tension at the biosurfactant solution-chlorinated solvent (methylene chloride and trichloroethylene (TCE)) interface was reduced to below 5 dynes/cm at a biosurfactant concentration of 4 g/L. The biosurfactant was effective in emulsifying both methylene chloride and trichloroethylene (TCE). Emulsification of methylene chloride and TCE were observed below the interfacial tension of 11 and 8 dynes/cm, respectively. Biosurfactant also increased the solubility of methylene chloride and TCE and the performance of the biosurfactant was comparable to chemical surfactants, sodium dodecyl sulfate (SDS) and Triton X-100. Changes in bonding characteristics during solubilization were investigated using the FTIR.

O7.21
CHROMOPHORE BINDING TO IN-VITRO ENGINEERED BIO-MIMETIC SURFACES. Albena Ivanisevic, Joseph Kinsella, Nyamjav Dorjderem, Dept of Biomedical Engineering, Purdue University, West Lafayette, IN.

In the retina, a chromophore molecule, 11-cis retinal isomerizes and the protein to which it is bound changes shape. In this work, we describe a proof-of-concept experiment in which we engineer an artificial surface where the physicochemical environment of the retina is mimicked and the key reaction of the visual cycle can occur. We immobilized small peptides on silicon and assessed changes in their surface properties upon adsorption via atomic force microscopy. Our observations suggest that when binding occurs it is accompanied by conformational changes of the surface-anchored peptide.

O7.22
CELLULAR INTERACTIONS AND BIOCOMPATIBILITY OF SELF-ASSEMBLING DIBLOCK POLYPEPTIDE HYDROGELS. Lisa M. Pakstis, Bulent Ozbas, and Darrin Pochan, Univ of Delaware, Dept of Material Sciences and Engineering, Newark, DE; Clifford R. Robinson, Univ of Delaware, Dept of Chemistry and Biochemistry, Newark, DE; Andrew P. Nowak and Timothy J. Deming, Univ of California Santa Barbara, Dept of Materials Science and Chemistry, Santa Barbara, CA.

Self-assembling peptide based hydrogels having a unique nano- and microscopic morphology are being studied for potential use as tissue engineering scaffolds. Low molecular weight amphiphilic polypeptides of hydrophilic lysine (K) or glutamic acid (E) and hydrophobic leucine (L) or valine (V) self assemble into hydrogels in aqueous solution at neutral pH and low fraction of polymer (vol. fraction polypeptide $\geq$0.5 wt%). Various molecular architectures, including di and triblock compositions with different block lengths, secondary structures, and densities of charged species, are being employed to study the effect of polypeptide design on morphology and biofunctionality. The morphology of these hydrogels in both pure water and salt solutions/cell culturing growth media has been characterized using laser scanning confocal microscopy (LSCM), small and ultra-small angle neutron scattering (SANS/USANS), and cryogenic transmission electron microscopy (cryoTEM) imaging. The hydrogel scaffolds consist of interconnected, highly porous membranous network on the nanoscale and a well-defined, highly porous structure on the microscale. Studies using fluorescence recovery after photobleaching (FRAP) highlight the diffusion properties of the hydrogels. Diffusion within the hydrogel matrix is extremely slow whereas diffusion throughout the hydrophilic regions is similar to that of pure water. The structure-bioproperty relationship is assessed through cell culturing biocompatibility studies. Mammalian cell studies indicate that polycationic hydrogels are cytotoxic while polyanionic hydrogels are noncytotoxic.

O7.23
CHARACTERIZATION OF HYDROGELS FORMED VIA SELF-ASSEMBLY OF AMPHIPHILIC β-HAIRPIN MOLECULES. Bulent Ozbas, Lisa Pakstis, Darrin Pochan, Univ. of Delaware, Dept of Materials Science and Engineering, Newark, DE; Karthikan Rajagopal, Juliana Gill, Joel Schneider, Univ of Delaware, Dept of Chemistry and Biochemistry, Newark, DE.

Stimuli-responsive and biocompatible networks formed via self-assembly serve great opportunities in tissue engineering and drug delivery applications. Due to the necessity of versatile biomaterials applications, understanding the relation between molecular design and mechanical-chemical properties is crucial. In this work we present the formation of hydrogels via the intramolecular folding and consequent self-assembly of 20 aminoacid long β-Hairpin peptide molecules. These hairpin molecules are amphiphilic in nature with an alternating sequence of hydrophobic valine and hydrophilic lysine amino acids. These molecules are found to form hydrogels with a unique microstructure and nanostructure at different physical conditions and at low peptide concentrations ($\sim$1 wt%). CD spectroscopy shows that at low pH or salt concentration the molecules are unfolded in a coil conformation and, thus, unassembled. However, gelation is observed at high pH values ($\sim$9) and at high salt concentrations ($\sim$150 mM) where β-sheet secondary structure due to hairpin folding is observed.

The intimate relationship between β-Hairpin molecule turn sequence and the consequent materials properties will be discussed. Laser Scanning Confocal Microscopy data reveals that hydrogel structure is heterogeneous at the microscale with water channels in the order of 10 μm. The self-assembled regions, imaged by cryogenic transmission electron microscopy technique, consist of interconnected fibrillar/tubular networks. Small angle and ultra small angle scattering results are in accordance with the microscopy findings. The viscoelastic properties of the hydrogels, as measured by rheology, are indicative of a shear-thinning (easily processible) network that quickly recovers to the original state after cessation of shear. Hydrogel storage moduli (G) are greater than 1kPa at low concentration of peptide (<3 wt%) indicating significant gel rigidity. Importantly, the gel is also reversible with pH, returning to a viscosity of water with a drop in pH that unfolds, and disassembles, the hairpin molecules.

O7.24
IN VITRO MINERALIZATION AND DEGRADATION OF BIODEGRADABLE POLYURETHANE SCAFFOLDS FOR THE REPAIR AND REGENERATION OF BONE DEFECTS.
Katarzyna Gorna and Sylwester Gogolewski, Polymer Research, AO/ASIF Research Institute, Davos, SWITZERLAND.

Bone defects of critical-size do not heal spontaneously in a lifetime unless the healing process is promoted by filling the defects with osteogenic substances. Primarily, these are cancellous and/or cortical bone grafts, platelet concentrate, bone marrow aspirate or growth factors. To reduce the amount of bone graft used to fill critical-size defects, it is often mixed with various fillers, primarily collagens sponges and fleeces or ceramic bone substitutes. If bone marrow aspirate or platelet concentrate are to be used to fill defects, it would be beneficial to load them into a suitable scaffold thus providing an additional substrate for osteogenic cells and newly formed bone. Optimally, scaffolds for bone regeneration should be three-dimensional, porous to allow for bone ingrowth, biodegradable to be replaced by new bone, radiolucent, and able to complex calcium phosphate salts from body fluids. Biodegradable polyurethanes are among the candidates for such scaffolds. This study addresses in vitro mineralization and degradation of porous 3-D scaffolds for regeneration of skeletal defects produced from new biodegradable aliphatic polyurethanes. The highly crosslinked porous polyurethane scaffolds were obtained in the course of material synthesis using liberated carbon dioxide as a foaming agent. In vitro mineralization was carried out in a newly developed calcifying medium, and in vitro degradation in phosphate buffer at the pH=7.4 and 37°C. The scaffolds underwent controlled calcification and degradation in vitro. The extent of material calcification, the morphology and Ca/P ratio of calcium phosphate crystals grown in the scaffolds and the rate of material degradation could be controlled by adjusting the polymer composition. Incorporation of saccharide units into the polyurethanes promoted scaffold calcification while reducing the rate of degradation; incorporation of polyethylene oxide units enhanced scaffold degradation.

O7.25
DEXTRAN GRAFTED SILICON SUBSTRATES: PREPARATION, CHARACTERIZATION AND BIOMEDICAL APPLICATIONS.
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The design of new biocompatible materials is a topic of great interest both from the practical and theoretical point of view; in particular biodevices used in the cardiovascular system suffer from well-known problems associated with surface-induced gas embolism and thrombosis. Since protein-surface interactions are recognized as critical in determining events leading to thrombus formation, the reduction of protein adsorption by using hydrophobic surface coatings is very often used. Following reported methods for immobilizing polysaccharides on artificial surfaces, we recently synthesized a coating layer of dextran, a relatively simple and well characterized neutral polysaccharide, with the purpose of mimicking the cells' glycocalyx layer, that prevents non-specific cells-protein interactions. The dextrans were activated for covalent immobilization by periodate oxidation, which produced hemiacetal structures that were reacted with the surface amino-groups of a self-assembled monolayer of 3-aminopropyltriethoxysilane (APTES) previously deposited on flat silicon wafer surfaces. Systematic physical chemical characterization, with evaluation of thickness, wettability and roughness, was performed on coatings obtained both from commonly used polydisperse dextrans and monodisperse dextrans in the 1-100 kDalton molecular weight range. The synthetic surfaces were also tested for protein rejection ability, antithrombogenic and gas bubble adhesion properties. The next step will be to combine standard surface analysis techniques, such as ellipsometry, contact angle measurements and AFM, with less traditional vibrational spectroscopy techniques. Micro-Raman and ATR-FTIR spectroscopies will also be utilized to correlate the conformational and molecular aspects of the grafted poly- and monodisperse dextran chains to their attractive biological properties.

O7.26
COMPOSITION AND SURFACE TOPOGRAPHY EFFECTS ON APATITE-FORMING ABILITY OF CERAMIC-POLYMER COMPOSITES.
Susan Rea, Serena Best, William Bonfield, Univ of Cambridge, Dept of Materials Science and Metallurgy, Cambridge, UNITED KINGDOM.

The bioactivity of two ceramic-polymer composites with potential for orthopaedic applications was investigated: HAPEX^{TM} (40 vol% hydroxyapatite in a high-density polyethylene matrix) and AWPEX (40 vol% apatite-wollastonite glass ceramic in a high density polyethylene matrix). These composites, designed to match the structure and mechanical properties of human cortical bone, have had positive in vivo results, and HAPEX^{TM} has had clinical success in middle ear and orbital implants. However, more detailed in vitro information is desirable to improve the range of possible uses for these materials. The effects of controlled surface topography and ceramic filler composition on apatite layer formation in acellular simulated body fluid (SBF) with ion concentration similar to those of human blood plasma were investigated. Samples were prepared as 1 cm x 1 cm x 1 mm tiles with polished, roughened, or parallel-grooved surface finishes, and were incubated in 20 ml of SBF at 36.5°C for 1, 3, 7, or 14 days. The formation of a biologically active apatite layer on the composite surface after immersion was demonstrated by thin-film X-ray diffraction (TF-XRD), environmental scanning electron microscopy (ESEM) imaging and energy dispersive x-ray (EDX) analysis. Variations in sample weight and solution pH over the period of incubation were also recorded. Significant differences were found between the two materials tested, with greater bioactivity in AWPEX than HAPEX^{TM} overall. Results also indicate that within each material the surface topography is highly important, with rougher samples correlated to earlier apatite formation.

O7.27
Abstract Withdrawn.

O7.28
EFFECT OF ENVIRONMENT AND ORGANIC PHASES ON STRESS CORROSION CRACKING IN CALCIUM PHOSPHATE CEMENT.
Victoria C. Jew and Reinhold H. Dauskardt, Stanford Univ, Dept of Materials Science and Engineering, Stanford, CA.

Cementitiously forming calcium phosphate apatites have shown significant potential as bone mineral substitutes for orthopedic and craniofacial applications due to their injectability and to their similarity to the inorganic component of bone. While these materials have promising biocompatible and osteoconductive properties, their application is limited by poor tensile strength properties. Basic mechanical properties of calcium phosphate cements have been well characterized, but only limited information has been reported for the fracture behavior of these materials. We have previously examined both cyclic fatigue crack growth behavior and moisture-assisted stress corrosion cracking in a hydroxyapatite bone cement. Stress corrosion cracking was significantly accelerated by increases in temperature and in moisture, raising implications for the integrity and reliability of such synthetic bone mineral substitutes, particularly in load bearing applications. In this investigation, we examine the effect of a range of acidic and basic pH values on hydroxyapatite cement in order to further elucidate the mechanisms of stress corrosion cracking. In addition, the effect of organic reinforcement phases incorporated into the cement is studied; the introduction of second-phase organics has been shown to increase critical fracture strength of calcium phosphate cements. Models for the kinetics of moisture-assisted subcritical crack extension have been developed based on the observed trends.

O7.29
ELECTROCHEMICAL DEPOSITION OF CALCIUM- DEFICIENT AND SUBSTITUTED (F-, CO3-) APATITES ON TITANIUM ALLOY SUBSTRATE.
Racquel Z. LeGeros, Shujie Lin, Ramin Rohanizadeh, John P. LeGeros, Calcium Phosphate Research Laboratory, Dept of Biomaterials and Biomimetics, New York Univ College of Dentistry, New York, NY.

To provide coating with homogenous composition at low temperature, alternative methods to the commercially used plasma-spray method are being developed. Using electrochemical deposition method, several calcium phosphate compounds have been deposited on titanium (Ti) alloy substrates. These have included: dicalcium phosphate dihydrate (DCPD), calcium-deficient apatite and octacalcium phosphate. The aim of this study was to deposit calcium deficient apatite,

fluoride-subtituted apatite (FAp) and carbonate-substituted apatite (CHA) on Ti alloy substrates using electrochemical deposition method (ECDM). Materials and Methods. Ti alloy (Ti6Al4V) plates (25x25x1.5mm) plates were mechanically polished, ultrasonically cleaned, acid etched, rinsed and dried. Nail varnish was applied on the substrates except for a circular area (1.0 cm dia) at the center. One plate was used as the anode and the other as the cathode, with the separation distance between plates kept constant. ECD was carried out using modulated pulse time electric fields programmed with a custom-made dual microprocessor. Metastable calcium phosphate solutions with or without additional fluoride (F) or carbonate (C) ions was used to obtain the desired apatite coating (calcium deficient apatite, FAp or CHA). Coatings were characterized using x-ray diffraction, scanning electron microscopy and FT-IR. Results. Morphology of the coating depended on the type of apatite obtained. Crystallinity (reflecting crystal size) was highest for FAp and lowest for CHA coating. Resolution of the FT-IR absorption bands was highest for FAp and lowest for CHA. Conclusions. Using ECD and appropriate pH, temperature and electrolyte composition, deposition of the desired apatite coating at low temperature can be obtained. [Supported in part by L. Linkow Professorship Implant Research Fund]

O7.30
CYTOCOMPATIBILITY OF CARBON NANOFIBER/ POLYMER COMPOSITES. Janice L. McKenzie, Michael C. Waid, Purdue Univ, Dept of Biomedical Engineering, West Lafayette, IN; Riyi Shi, Purdue Univ, Dept of Basic Medical Sciences, West Lafayette, IN; Thomas J. Webster, Purdue Univ, Dept of Biomedical Engineering, West Lafayette, IN.

Neural prostheses provide a means for monitoring and applying electrical signals to neural tissue. At an implant site, a cellular process known as gliotic response partitions the implant from surrounding tissue by scar formation. This scar tissue development is largely due to the activity of astrocyte cells, and is thought to interfere with the long-term operation of implanted neural prosthetic devices. The development of biomaterials that enhance nerve cell interactions and deter astrocyte formation of scar tissue provides an attractive alternative to conventional neural prosthetic materials. Recent research confirms that nanoscale biomaterials achieve better cytocompatibility with certain types of cells than their conventionally sized counterparts. Carbon nanofibers possess excellent electrical properties, which may be desirable in the area of neural prostheses. The objective of the present in vitro study was to determine cytocompatibility properties of carbon nanofiber and polymer composites pertinent to neural prosthetic biomaterial formulations. Polycarbonate urethane was used as a supportive matrix for varying weight percents of carbon nanofibers. Cytocompatibility studies were then performed with astrocyte cells to ascertain the ability of the composite to deter astrocytic functions. The adhesion of the astrocytes decreased as the percentage of carbon nanofibers in the composite was increased. These results are promising, and neurite outgrowth on the carbon nanofibers is also being tested. The material characteristics of the carbon nanofiber composites can be engineered for optimal usage as a neural biomaterial.

O7.31
Abstract Withdrawn.

O7.32
MULTIFUNCTIONAL POLYMER SUPPORT FOR THE HYDROLYSIS AND REMOVAL OF METHYL PARATHION. Yongwoo Lee, Ivan Stanish, and Alok Singh, US NRL, CBMSE, Washington, DC.

Cyclodextrin (CD) is known to form inclusion complexes with paranitrophenol (pNP) and other related organic toxic agents including methyl parathion (MPT). Cross-linked CD (PCD) with bifunctional alkyl spacers (i.e., butyl, hexyl, and phenyl ethyl etc.) that have been sieved to 32 - 53 mm, provides an insoluble matrix that is chemically stable in acidic and basic media while preserving accessibility for pNP sorption. At pH 1 - 9, we have observed sorption capacities that range from 14 - 191 mg pNP/g PCD in under an hour. In addition, PCD provides a unique support for the non-covalent immobilization of organophosphorous hydrolase (OPH) which can catalyze the hydrolysis of MPT (i.e, apparent turnover rates near 0.01 s-1) and subsequently sequester its by-products (i.e., pNP and its phosphate derivative) through exploitation of the inclusion phenomenon and secondary interactions. Preparation of the enzyme embedded PCD material, which takes advantage of the layer-by-layer assembly technique, will be discussed in detail. We will also describe the multi- utility of PCD as a single unit biofilter system and characterize its performance in batch and continuous mode as a stable biomaterial for the hydrolysis of MPT and simultaneous adsorption of its by-products.

O7.33
ENHANCED OSTEOBLAST ADHESION ON A NOVEL HYDROXYAPATITE COATING. Michiko Sato, Thomas J. Webster[a], Elliott B. Slamovich, School of Materials Engineering and [a]Department of Biomedical Engineering, Purdue University, West Lafayette, IN.

Titanium coated with hydroxyapatite (HA) has been used as a dental and an orthopedic implant material. Titanium has similar mechanical properties to bone, while HA exhibits good cytocompatibility with surrounding tissue. Plasma spray is widely used to process HA coatings. However, the resulting HA contains a significant number of surface cracks that degrades bonding to juxtaposed bone. This study aimed to develop a different HA coating process, and evaluate its resulting biocompatibility by osteoblat (bone-forming cells) adhesion compared to HA coatings obtained by plasma spray processing. HA was synthesized by dripping calcium nitrate and ammonium phosphate in water. After 24hrs, the resulting precipitate was added to a mixture of titanium alkoxide, chloroform, and poly(di-lactic-glicolic acid)(PLGA). The polymer was added to minimize cracking. Coatings were deposited on glass substrates(1 cm^2)by spin coating, and some of the resulting films were treated hydrothermally in water at 70°C. SEM observations indicated that the coatings were crack-free, and that the hydrothermally treated coatings contained clusters of precipitated particles. EDS and XRD studies suggested that the clusters were mixtures of calcium phosphorus compounds, and titanium oxide compunds. Results in vitro studies demonstrated that coatings with HA gel, titania precursor, and PLGA with hydrothermal treatment enhanced osteoblast adhesion compared to all others formulations tested. Also, the hydrothermally treated coating with the titania precursor and PLGA enhanced the cell adhesion more that the same formulation without hydrothermal treatment($p<0.1$). Further in vitro studies are in progress to determine if this novel coating technique and material would decrease solubility and increase biocompatibility properties necessary for increase osseointegration with bone;criteria critical for orthopedic and dental implant success.

O7.34
HIGH SURFACE AREA NANOFIBER MEMBRANES FOR SELECTIVE AND SPECIFIC BIOAFFINITY CAPTURE OF ANALYTES. Kris J. Senecal, Charlene M. Mello, Philip Pivarnik, Andre G. Senecal, U.S Army Soldier Systems Command, Natick Soldier Center, Natick, MA.

Polymeric materials with specific chemistries have been studied to form a high surface area membrane for selective affinity capture. Through a network of hydrophobic, electrostatic and other interactions, Molecular Recognition Elements (e.g antibodies, aptamers and peptide based receptors) will be attached to the fibrous membrane to impart biochemical specificity. High surface area membranes have the potential to provide a higher density of these MREs on the membrane surface and enhance the efficiency of binding or capturing an analyte of interest. We are using electrospinning technology with polymeric materials to form a porous nonwoven bioactive fibrous membrane. Polymeric fibers resulting from electrospinning are in the nanoscale range and provide a high surface to volume ratio specific for the sensor platform. Antibodies and peptide-based receptors are selectively immobilized onto these nanoporous membranes for bioaffinity capture. We envision these membranes being used for preconcentration/sample preparation devices for biosensors, water filtration and purification, bioseparation media, and sample collection membranes. Initial results involving imaging and quantifying the antibody attachment and activity in association with the electrospinning process will be discussed.

O7.35
CYTOCOMPATIBILITY AND MATERIAL PROPERTIES OF POLY-CARBONATE URETHANE/CARBON NANOFIBER COMPOSITES FOR BONE AND NEURAL APPLICATIONS. Michael C. Waid[†], Janice L. McKenzie, Rachel L. Price, and Thomas J. Webster, [†]Mechanical Engineering, University of Nebraska, Lincoln, NE; Biomedical Engineering, Purdue University, West Lafayette, IN.

Current orthopedic and neural implants lack the cytocompatiblity properties necessary for sufficient integration into the body, so new materials and approaches are necessary. Previous research has shown that carbon nanofibers have excellent cytocompatiblity properties because of their biologically inspired nanostructured dimensions (less than 100 nm). However, carbon nanofibers do not inherently bond strong to each other and, thus, cannot be used alone as an implant material. The present study sought to synthesize a composite material of poly-carbonate urethane (PCU) and carbon nanofibers. Different weight percentages of PCU and carbon nanofibers were created and the cytocompatiblity, mechanical, and electrical properties were characterized for each composition. Also, a composite was created with aligned carbon nanofibers by mechanically stretching the material uniaxially. Results from cells studies demonstrated that

enhanced osteoblast (bone-forming cells) adhesion correlated with increased carbon nanofiber content in PCU. Moreover, adhesion of competitive fibrous forming cells pertinent for bone and neural implant applications (specifically, fibroblasts and astrocytes, respectively) decreased with increasing amounts of carbon nanofibers in PCU. Osteoblast morphology changed with aligned carbon nanofibers such that the cells appeared stretched along the direction of carbon nanofiber alignment. Furthermore, the material properties of the composites were improvements over the individual polymer or pure carbon nanofiber compacts. For example, compared to pure PCU, the tensile modulus of the 75:25 weight percentage PCU:carbon nanofibers composite increased almost 10 times and the electrical resistivity decreased significantly. For these reasons, the present study demonstrates that carbon nanofibers can be used in a PCU composite to create materials that closely match the necessary cytocompatibility, mechanical, and electrical properties for bone and neural implant applications.

O7.36
QD-BIOCONJUGATES ASSEMBLED USING GENETICALLY MODIFIED PROTEINS. S.-Y. Ding, M.E. Himmel, W.S. Adney, M.P. Tucker, National Bioenergy Center, National Renewable Energy Laboratory, Golden, CO; J. Wall, Brookhaven National Laboratory, Brookhaven, NY.; J. Nedeljkovic, O.I. Micic, M. Jones, A.J. Nozik, and G. Rumbles, Center for Basic Science, National Renewable Energy Laboratory, Golden, CO.

We have successfully coupled an engineered cohesin-protein polymer and its monomer protein equivalent to one or more CdSe[ZnS] core-shell colloidal quantum dots. We find these new bioconjugates to be stable, water-soluble, and amenable to separation by aqueous high-pressure size-exclusion chromatography (HPSEC). We have analyzed the fractions eluted from HPSEC using absorption, photoluminescence (PL), multi-angle laser light scattering (MALLS), and scanning transmission electron microscopy (STEM). The QD conjugate preparations based on single cohesin proteins modified genetically to display both N- and C-terminal 6 x histidine tags were found to be dominated by discrete and regular structures surrounded by protein. In addition, single protein coated quantum dots are observed in the later eluting fraction from the same sample. PL studies on the larger structures suggest that the CdSe[ZnS] QDs are strongly coupled electronically, with emission emanating from the largest QDs only. In this presentation, we will discuss the role of the protein in conferring water-solubility, shape, and the mechanism and efficiency of binding to the QD surface. We will also discuss the relative roles of protein directed- and self-assembly of the QDs.

O7.37
OSTEOBLAST-LIKE CELL RESPONSE TO APATITE-WOLLASTONITE/POLYETHYLENE BONE REPLACEMENT COMPOSITES. Susan Rea, Serena Best, William Bonfield, Univ of Cambridge, Dept of Materials Science and Metallurgy, Cambridge, UNITED KINGDOM; Roger Brooks, Addenbrooke's Hospital, Orthopaedic Research Unit, Cambridge, UNITED KINGDOM.

Apatite-wollastonite/high density polyethylene composite (AWPEX) materials, designed to match the structure and mechanical properties of human cortical bone, have been shown to be bioactive in vivo. The effects of surface finish and ceramic filler size and content on osteoblast-like cell attachment, proliferation, and differentiation were examined to understand better and extend AWPEX orthopaedic applications. Glass ceramic content was tested at 30 or 50 vol% and median particle size at 4.5 or 7.7 μm. Samples were prepared as 10 x 10 x 1 mm^3 tiles with polished or roughened surfaces, sterilized by gamma irradiation (2.5 Mrad), and characterized by SEM, surface energy, and surface profilometry. Saos-2 human osteoblast-like cells were cultured on each surface at an initial concentration of 4500 cells/cm^2 in McCoy's media and incubated at 37°C in a humidified atmosphere with 5% CO_2 for 1, 3, or 7 days. At each time point, adenosine triphosphate (ATP) and alkaline phosphatase (ALP) levels were measured to assess cell number and osteoblast differentiation. Significant differences were found at 7 days, confirmed by ANOVA post-hoc testing using Bonferronis correction. For identical A-W volume fraction and particle size, samples with polished surfaces had elevated ATP levels in all except the 50-vol% 4.5-μm composite. Polishing also increased ALP in 50-vol% composites, but had no significant effect in 30-vol% samples. For identical surface finish and particle size, greater A-W content led to elevated ATP levels in 4.5-μm composites, but had no significant effect in 7.7-μm composites. Greater A-W content also elevated ALP in all except the roughened 7.7-μm samples. For identical surface finish and volume fraction, larger particle size raised ATP and ALP levels in all except the 50-vol% roughened samples. Increased exposure of the A-W phase in AWPEX through surface polishing, higher volume fraction, and/or larger particle size, can therefore lead to an improved cell response.

SESSION O8: MOLECULAR RECOGNITION AND TEMPLATES
Chairs: James L. Thomas and Kristi L. Kiick
Friday Morning, April 25, 2003
Franciscan I (Argent)

8:30 AM *O8.1
BIOMOLECULAR CONTROL AND ASSEMBLY OF SEMICONDUCTOR AND MAGNETIC NANOSTRUCTURES. Angela M. Belcher, Seung-Wuk Lee, Brian Reiss, MIT, Dept of Materials Science and Engineering and BioEngineering, Cambridge, MA; Christine Flynn, Chuanbin Mao, Andrew Hayhurst, George Georgiou, Brent Iverson, Univ of Texas at Austin, Dept of Chemistry and Biochemistry, Austin, TX.

Biological systems have a unique ability to control crystal structure, phase, orientation and nanostructural regularity of inorganic materials. We are currently investigating the principles of natural biological molecular recognition in materials and developing new methods to pattern useful non-biological electronic and magnetic materials on new length scales. A peptide combinatorial approach has been employed to identify proteins that select for and specifically bind to inorganic structures such as semiconductor wafers and semiconductor and magnetic nanoparticles. This approach utilizes the inherent self-organizing, highly selective properties of biologically derived molecules. We are currently investigating peptide recognition and interaction with III-V and II-VI semiconductor materials and magnetic materials including Fe_3O_4, Co, FePt and CoPt. We have selected peptides that can specifically bind to and discriminate zinc-blende III-V semiconductor surfaces. These peptides show crystal face specificity and are being used to organize nanoparticle heterostructures. We have also selected peptides that can nucleate and control phase, particle diameter and aspect ratio of semiconductor and magnetic nanoparticles. These peptides are being used to grow nanoparticles and nanowires of specific crystallographic structure and orientation. Using these molecular interactions and specific nanoparticles, we are organizing organic/inorganic hybrid materials into supramolecular architectures.

9:00 AM O8.2
SELECTION OF INORGANIC BINDING PEPTIDES USING PCR-DRIVEN PHAGE PEPTIDE DISPLAY. Rajesh R. Naik and Morley O. Stone, Air Force Research Laboratory, Materials and Manufacturing Directorate, WPAFB, OH; Christopher J. Murray and Joseph C. McAuliffe, Genencor International, Inc., Palo Alto, CA.

Combinatorial phage display is a common technique used to select for ligands that are capable of interacting with specific targets. A phage display library is a library of peptides or proteins that are displayed on the surface of a filamentous bacteriophage. The peptide is expressed as a fusion with a coat protein of the bacteriophage resulting in the display of the fusion protein on the surface of the phage. A number of studies have demonstrated the use of phage peptide display to select for peptides that can recognize inorganic surfaces. By exploiting the molecular recognition properties of the peptides, some of these peptides can serve as templates for inorganic material deposition. Most target-binding peptides are selected using the standard modified pH- or temperature-dependent elution step that allows for the release of ligand(s) that bind strongly to the target surface. However, it has been observed that strongly binding ligands are not easily eluted using standard procedures. Here, we demonstrate the use of a PCR-driven approach to identify peptides that would otherwise be omitted using the standard elution process and also present evidence that specific ligands can be selected in a single step.

9:15 AM *O8.3
BIOMIMETIC SYNTHESIS AND PATTERNING OF SILVER NANOPARTICLES. Rajesh R. Naik, Sarah J. Stringer, Gunjan Agarwal, Sharon E. Jones and Morley O. Stone, Air Force Research Laboratories, Materials and Manufacturing Directorate, Biotechnology Group, Wright-Patterson Air Force Base, Dayton, OH.

An in vitro biosynthetic route for the formation of silver nanoparticles is reported. The process of silver nanoparticles formation is promoted by the presence of a silver-binding peptide that causes the reduction of silver ions to metallic silver. Examination of the silver nanoparticles by transmission electron microscope revealed a variety of crystal morphologies such as hexagons, triangles and spheres. This research is the first demonstration of a peptide-based synthesis of silver nanoparticles. Through the patterned delivery of the silver-binding peptide onto a glass surface using micromolding in capillary (MIMIC) technique, we demonstrate the fabrication of silver arrays as an approach for using the templating function of the biomolecules to organize inorganic materials for bottoms-up fabrication.

9:45 AM O8.4
BIO-IMPRINTED POLYMER SCAFFOLDS FOR SELECTIVE

RECOGNITION OF ECM PEPTIDES. Nicole Bergmann and Nicholas A. Peppas, University of Texas, Department of Biomedical Engineering, Austin, TX.

Fibronectin and a number of other plasma and extracellular matrix (ECM) adhesion proteins contain the tetrapeptide arginine-glycine-apartic acid-serine (RGDS), and this sequence can be summarily recognized and bound by integrins present on cell membranes. Upon integrin binding, cells adhere to the substrate, and this adherence encourages ECM deposition and other cellular remodeling events. By targeting specific chemical functional groups on the peptide using non-covalent molecular imprinting, biomimetic polymeric scaffolds can be designed to mimic protein-ECM binding on both the surface and in the bulk during polymer degradation. Methacrylic acid-ethylene glycol dimethacrylate (MAA-g-EGDMA) copolymer films were prepared by free-radical ultraviolet polymerization in the presence of RGDS to create novel imprinted matrices for possible tissue engineering scaffolds. SEM analysis revealed a highly macroporous structure in peptide-imprinted polymers compared to controls. Optimal crosslinking ratios for peptide imprinting were determined using a small molecular weight fluorescent tag, 4-chloro-7-nitrobenzofurazan, and analyzed using fluorescent microscopy. Higher crosslinking ratios yielded better template recognition and gels exhibited specific recognition in aqueous media to RGDS molecules when in the presence of similar tetrapeptides.

10:30 AM *O8.5
LIVING TEMPLATES FOR HIERARCHICAL ASSEMBLY OF GOLD NANOPARTICLES. Zhi Li, Sung-Wook Chung, Jwa-Min Nam, David S. Ginger, and Chad A. Mirkin, Northwestern University, Dept of Chemistry, Evanston, IL.

Living Templates for Hierarchical Assembly of Gold Nanoparticles Biological systems form sophisticated mesoscopic and macroscopic structures with tremendous control over the placement of nanoscopic building blocks within extended architectures. The promise of borrowing from Natures repertoire to organize inorganic materials on length scales relevant for applications in catalysis, photonics, and electronics has led to the pursuit of a variety of biotemplated and biomimetic strategies for nanoscale materials synthesis and assembly1-6. However, using such methods to independently control the organization of inorganic materials on multiple length scales has remained challenging. Here we demonstrate that living microorganisms can facilitate the assembly of oligonucleotide-functionalized gold nanoparticles into ordered structures with characteristic lengths ranging from less than 1 to over 100 micrometers. We show that the hybridization of the DNA on the nanoparticles with additional DNA-functionalized building blocks can be used to tailor the nanoscopic architecture of the materials while the microorganisms control the microscopic architecture, providing a method to independently direct the assembly of inorganic particles on length scales ranging from a few nanometers to hundreds of micrometers.

11:00 AM O8.6
HYBRIDIZATION-BASED UNQUENCHING OF DNA HAIRPINS ON Au SURFACES: PROTOTYPICAL MOLECULAR BEACON BIOSENSORS. Hui Du, Todd D. Krauss, Benjamin L.Miller, Univ of Rochester, Dept of Chemistry and Dept of Dermatology, Rochester, NY.

Recent intense interest in the use of rapid genetic analysis as a tool for understanding biological processes, in unlocking the underlying molecular causes of disease, and in the development of biosensors, has led to a need for new sensitive and arrayable chip-based analytical tools. Of high importance is the need for techniques that do not require labeling of the target sample, since that increases the time, cost, and potential for error inherent in the analysis. Molecular beacons consist of a DNA hairpin functionalized at one end with a fluorophore, and at the other with a quencher. In the absence of the target DNA sequence, the close proximity of the quencher to the fluorophore inhibits fluorescence. Addition of the target leads to hairpin unfolding and signal generation. In the context of solution-phase biological assays, the molecular beacon concept has proven itself to be both sensitive and reliable. Although a few reports of surface-immobilized molecular beacons exist, to our knowledge the material on which the DNA is immobilized plays a passive role only. Here we will present a "label-free" molecular-beacon biosensor based on unquenching of DNA hairpins on Au surfaces. Two DNA hairpins incorporating portions of the Staphlococcus aureus FemA and mecR methicillin-resistance genes were self-assembled on a gold substrate. Using confocal-epi-fluorescence microscopy we examined the fluorescence of DNA hairpin-functionalized Au films in the presence and absence of complementary targets. Fluorescence was enhanced by at least 20-fold in the presence of the DNA complement. Furthermore, similar measurements performed in the presence of non-specific DNA indicate that DNA hairpins retain their ability to bind complementary sequences selectively. Efforts to implement this design in a microarray format, and to replace dye molecules with highly fluorescent and photostable CdSe quantum dots will be discussed.

11:15 AM O8.7
BIO-INSPIRED TEMPLATE DESIGN TO CONTROL CRYSTAL GROWTH. Simon Champ, Nicholas B. Dinsdale, Denise J. Farrall, Brigid R. Heywood, Susan J. Hill, Mandeep Jandu, Graham P. Mitchell, Kathryn Pitt, Paul A. Tibble, Keele University, School of Chemistry & Physics, UNITED KINGDOM.

Biomimetics is an important branch of materials chemistry, which aims to explore and exploit the mechanisms by which useful and efficient materials are produced in biological systems. One key aspect of the research has been an exploration of the links between supramolecular chemistry and solid state synthesis through the use of a range of ordered organic systems to control and direct the formation of inorganic materials. These studies have shown that when assembled in ordered hierarchical arrays, functionalised organic polymers are capable of directing and controlling crystallisation to produce novel solids. It is proposed that the nano-structural template presented by the organic assembly mimics defined structural motifs within the crystal system; the nucleation of oriented crystals and their subsequent habit modification is effected by the translation of the template imprint at the polymer-crystal interface. As part of a continuing exploration of this process the activity of novel polymer systems has been assayed; for example, metal activated two-component supramolecular arrays, quaternary ammonium functionalised polymers, calixarenes and durable organic gels (three-dimensional polymer networks). In these studies, the ability of these auto-assembling templates to control the growth of barium sulphate and calcium carbonate has been investigated. In each case the impact of specific chemical features within the template (e.g. molecular weight, functionality, mode of assembly) upon the crystal growth process has been highlighted with a view to generating an informed directory of design features which can be programmed into polymer systems to effect the controlled fabrication of novel inorganic materials. This study illustrates the potential of bio-inspired materials chemistry to offer new routes to crystal engineering, and poses the challenge of designing templates "tuned" with respect to their specific chemical and spatial characteristics with a view to producing "tailor-made" crystals of medicinal or commercial value.

11:30 AM O8.8
MAGNETIC IRON OXIDE MINERALIZATION OF PEPTIDE AMPHIPHILE NANOFIBERS. Eli D. Sone and Samuel I. Stupp, Northwestern University, Department of Chemistry, Department of Materials Science and Engineering, and Feinberg School of Medicine, Evanston, IL.

The formation of chains of aligned magnetite (Fe_3O_4) nanocrystals in magnetotactic bacteria allows these bacteria to orient and migrate using the earth's magnetic field. This striking example of biomineralization involves a remarkable level of control over the size, morphology, and orientation of the inorganic product. Synthetically, one approach to controlling inorganic features on the nanoscale is to use supramolecular organic objects as templates for inorganic growth. Peptide-amphiphile (PA) molecules, consisting of an aliphatic hydrophobe and peptidic head group self-assemble into nanofibers which have been shown to act templates that align crystals of hydroxyapatite. We report here on the self-assembly, characterization, and mineralization of a new histidine-containing PA designed to act as a template for the nucleation and growth of magnetite nanocrystals. This molecule can be induced to self-assemble into cylindrical micellar nanofibers ~8 nm in diameter by heating or pH change, leading to the formation of aqueous gels at concentrations as low as 1% by weight. The PA contains histidine moieties at the C-terminus which are exposed at the fiber/solution interface upon self-assembly and can function as effective chelators of Fe^{2+} ions at pH above 6.5. Mineralization of the PA fibers was investigated both in the gel state and also in aqueous suspensions of nanofibers. After introduction of iron precursors, exposure to ammonium hydroxide vapors leads to the formation of magnetic nanocrystals on nanofiber surfaces, with electron diffraction consistent with magnetite. In addition to mineralization studies, details on the self-assembling properties of this new PA will be presented. For instance, films composed of parallel arrays of fibers can be formed by evaporation of dilute (0.03% by weight) solutions of the PA. Such films may ultimately serve as templates for the production of ordered arrays of magnetic crystals.

11:45 AM O8.9
BIOMIMETIC SILICIFICATION TEMPLATED BY SURFACE-GRAFTED POLY(L-LYSINE). Yuli Wang, Ying-Chih Chang, Department of Chemical Engineering and Materials Science, University of California, Irvine, CA.

A surface-grafted poly-L-lysine (PLL) film is used as a template to

induce silica formation in neutral pH and at room temperature. Surface-grafted PLL is obtained by debenzylation of protection groups of surface-grafted poly(N-carbobenzyloxy L-lysine), which is fabricated by the surface-initiated vapor deposition polymerization. To start the biomimetic mineralization process, the grafted PLL film was immersed in a tetraethoxysilane/water solution for a period of time. The progress of the reaction is monitored by in-situ ellipsometry and atomic formic microscopy, and the surface composition was confirmed by Fourier transform infrared spectroscopy. It was found that silica formation occurred spontaneously inside the PLL film, and the growth pattern is fully templated by the surface-grafted PLL films in three dimensions. For instance, for a 170 nm thick PLL film, the silicification process is completed in 20 hours with the silica pattern followed by the dimensions of the template film. The prolonged immersion does not grow silica any further. The growth of silica is anticipated to correlate with the charge density of the grafted PLL film. By utilizing this characteristic, we will demonstrate the formation of high-resolution patterned silica films directed by pre-patterned surface-grafted PLL films with regulated charge distribution.