SYMPOSIUM LL
Orthopaedic/Dental Biomaterials

November 27 – 29, 2000

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*Invited paper
SESSION L11: ORTHOPEDIC/DENTAL/TISSUE ENGINEERING
Chair: C. Mauli Agrawal
Monday Morning, November 27, 2000
Berkeley (Sheraton)

8:30 AM L11.1
STRUCTURAL TEMPLATES AND BIODEGRADABLE FOR CARTILAGE TISSUE ENGINEERING
Gordana Vunjak-Novakovic and Lian E. Freed, Massachusetts Institute of Technology, Division of Health Sciences and Technology, Cambridge, MA.

Functional substitutes of native cartilage can be created in vitro using chondrogenic cells, biodegradable polymeric scaffolds and bioceramics. Scaffolds provide a structural template for tissue formation, while bioceramics provide a controlled environment that embodies the chemical and physical regulatory signals for the development of a functional tissue. The structure (composition, morphology) and function (biochemical activity, integrative potential, mechanical function) of engineered constructs can be modulated by the conditions and duration of in vitro culture. The paper reviews the main requirements for scaffold and bioceramic design and recent progress in tissue engineering studies of engineered cartilage.

9:00 AM L11.2
FORMATION OF THREE DIMENSIONAL CELL-POLYMER CONSTRUCTS IN BIODEGRADABLE FOR BONE TISSUE ENGINEERING
Vassilios Skalitis, Gregory Bancroft, Antonios Mikos, Rice University, Dept of Bioengineering, Houston, TX.

The aim of this study is to investigate the ability of bone marrow stromal cells seeded in three dimensional PLGA biodegradable scaffolds to be cultured in a spinner flask or a rotating vessel. The studied bioceramics generate good mixing and thus better nutrient transport to the seeded cells. Poly(D,L lactico-glycolic acid) (PLGA) copolymer scaffolds have been selected for this study because they are osteoconductive and biocompatible, degrading into products that can be either metabolized or excreted. Bone marrow stromal cells were harvested from femurs and tibias of six week old male Sprague-Dawley rats and placed in T-75 culture flasks using DMEM with 10% fetal bovine serum from selected lots. On the third day non-adherent cells were removed and on the seventh day the cells were seeded on porous 75-25 PLGA cylindrical forms prepared by the method of solvent casting and particulate leaching and cultured in the spinner flask, the rotary vessel and under static conditions in the presence of dexamethasone, beta-glycerophosphate and ascorbic acid-2-phosphate. On the 14th, 21st and 28th day, forms were removed and the cell number, radio-labelled thymidine incorporation, alkaline phosphatase activity and osteocalcin, were measured. The forms were also stained for mineralization. The cell number demonstrated a higher number of cells on day 21 in the spinner flask culture. The static culture had the lowest cell number but on the 28th day the difference in cell number in all three cultures was statistically insignificant. Histological sections of the forms on the 28th day showed higher cell densities at the exterior of the form providing strong evidence for the existence of nutrient concentration gradients at the interior of the scaffolds. Although the good mixing provided in the spinner flask allowed the cells to proliferate faster, high proliferation was limited to the cells located at the exterior of the forms.

9:15 AM L11.3
DEVELOPMENT OF A NOVEL PHOTOCROSSLINKABLE HYALURONAN MATRIX FOR CARTILAGE REPAIR
Kimberly A. Smeds, Duke Univ, Dept of Chemistry, Durham, NC; Jean Y. Wang, Anthony E. Baer, Lori A. Setton, Duke Univ, Dept of Biomedical Engineering, Durham, NC; Mark W. Grinstaff, Duke Univ, Dept of Chemistry and Ophthalmology, Durham, NC.

Repair of degenerated articular cartilage with exogenous scaffolds and cellular supplementation is an area of major interest in musculoskeletal research. In this study, we describe a novel scaffold for tissue-engineered cartilage repair which may be crosslinked in situ. This feature is extremely attractive for the ability to fill irregular defects and provide for optimal tissue integration. We have embedded chondrocyte cells in a photocrosslinkable hyaluronan based matrix. The photocrosslinkable methacyrylated hyaluronan, HA-MA, was prepared by reacting a 2% w/v solution of polyacrylamide with a 20 fold excess of methacrylic anhydride for 24 hours at 5°C. The photocrosslinkable polyacrylamide was next precipitated and washed with ethanol to remove remaining methacrylic acid and methacrylic anhydride. 1H-NMR spectroscopy confirmed that 14% of the free hydroxyls on hyaluronan were modified. Upon exposure of the HA-MA to an argon ion laser (514 nm) in the presence of a photoinitiating system, V. L-phenyl proline monomer, and triethyl amine (a stable hydroxyl) is formed. Next, primary chondrocytes were isolated from porcine knee joints and were mixed with a 2% solution of the HA-MA biopolymer at a concentration of 10^6 cells/mL. The suspension was divided into four equal portions and poured into cylindrical molds, photocrosslinked with an argon ion laser, and cultured out to two weeks. In control studies, the photoinitiating system was found not to be cytotoxic to chondrocytes at the concentrations required for the photoinitiating photocrosslinking. Viability studies showed that by 6 hours 91% of the cells were still visible and by 13 days 77% were visible. Additional studies will focus on quantitative measures of DNA content, cell proliferation, SAGG content, and collagen accumulation.

10:00 AM L11.4
CHITOSAN/TRICALCIUM PHOSPHATE SCAFFOLDS FOR BONE TISSUE ENGINEERING
Yong Zhang, Ming Zhang, University of Washington, Dept of Materials and Engineering, Seattle, WA.

This research involves synthesis and characterization of a biodegradable 3-dimensional polymer/calcium phosphate hybrid matrix as scaffolds for bone tissue regeneration. The scaffolds are comprised of chitosan and tricalcium phosphate (β-TCP) and have combined optimum mechanical and biological properties of the two materials. Chitosan is a biocompatible and biodegradable polymer, and its rich hydroxyl groups for surface modification. β-TCP with chemical composition close to the inorganic components of bone is used as a powder filler to reinforce the scaffolds and increase bioactivity of the scaffolds. The scaffolds are fabricated using a thermally induced phase separation technique. The principal advantages of this technique are its low cost, low shrinkage levels, low sintering temperatures, and its ability to produce a variety of microstructures of various shapes and sizes. The scaffolds are fabricated using the polymer/calcium phosphate ratio and material processing conditions. The osteoblast-like MG63 cells are seeded on the scaffolds to study the attachment and proliferation of the cells. The hydroxyl groups on the scaffolds are used to graft polyethylene glycol (PEG) linked arginine-glycine-aspartate (RGD) peptides for promoting bone cell attachment and new tissue formation. The PEG chains are expected to suppress the nonspecific protein adsorption for the prevention of scar tissue formation.

10:15 AM L11.5
CONTROLLING DIFFUSION OF SOLUTES THROUGH IONICALLY CROSSLINKED ALGINATE HYDROGELS DESIGN FOR TISSUE ENGINEERING
Catherine K. Kuo, Peter X. Ma, Univ of Michigan, Dept of Biologic and Materials Sciences, Macromolecular Science and Engineering Center, Dept of Biomedical Engineering, Ann Arbor, MI.

Tissue engineering aims at creating new tissues as alternatives to organ transplants. Our approach is to incorporate biodegradable polymer scaffolds designed to temporarily support tissue biodegradation. An important requirement of scaffolds is biodegradability. Homogeneity ensures structural integrity, uniform distribution of cells, and uniform porosity throughout the scaffold. Controlling pore sizes is necessary to regulate exchange of nutrients and waste products for cells. Pores too large can provide entryways to immune cells that can harm allogenic cells and developing tissue. We have fabricated three-dimensionally defined, homogenious, ionically crosslinked alginate gels with a controlled slow-solution system involving CaCl2 and D-glucuronic-d-mannuronic. We varied the structural parameters and alginate types of these gels to control the diffusion of calcium, vitamin B12 and FITC-dextran (molecular weights of 180, 1355 and 9500, respectively) through the gel. Experiments were performed with gel discs placed between side-by-side donor and receptor chambers in a humidified incubator at 37°C. Gels were taken periodically and measured on a UV-Vis spectrophotometer. Generally, diffusion coefficient (D) increased with decreasing solute size. Varying structural parameters of the gel did not have a significant effect on diffusivity of vitamin B12. In contrast, gels made with a Ca2+ to crosslinker ratio of 0.399, gels, FITC-dextran increased from 2.98 x 10^-7 ± 0.52 x 10^-7 to 4.66 ± 10^-7 ± 0.48 x 10^-7 cm^2/sec as alginate concentration decreased from 3.18% to 1.50%, respectively. D of FITC-dextran also increased from 3.17 x 10^-7 ± 0.30 x 10^-7 to 4.66 ± 10^-7 ± 0.48 x 10^-7 cm^2/sec as crosslinking density decreased for 1.50% alginate gels from a Ca2+ to carbonyl molar ratio of 0.72 to 0.26, respectively. D was highest for alginate gels with the highest phenolic content. Controlling diffusion allows gels with specific properties to be fabricated for tissue engineering scaffolding and other biomedical applications.

10:30 AM L11.6
DEVELOPMENT OF BIODEGRADABLE POLYMER SCAFFOLDS FOR TISSUE ENGINEERING USING CG EXTRUSION
TECHNIQUES. Newell R. Washburn, Alahgir Kamar, Eric J. Amis, National Institute of Standards and Technology, Gaithersburg, MD; Kimberly Potter, National Institutes of Health, Bethesda, MD.

The development of biodegradable polymer scaffolds using co-extrusion techniques is presented. Poly(ε-caprolactone) (PCL) and poly(l-lactic acid) (PLLA) were blended in a twin-screw extruder at 100°C. After subsequent annealing, the PEO was dissolved with water resulting in either open or closed porous material with characteristic length scales in excess of 100 μm, depending on composition and processing conditions. Additionally, the PCL scaffolds have compressive moduli on the order of 1 MPa at low loads. Scaffolds were seeded with osteoblasts isolated from embryonic chick calvaria. Non-invasive analysis of the mineralization process was performed using proton nuclear magnetic resonance microscopy and complementary information was obtained by histologic analyses.

10:45 AM L117
TEMPORARY ENCAPSULATION OF RAT MARROW OSTEOBLASTS IN GELATIN MICROSPHERES FOR BONE TISSUE ENGINEERING. R.G. Page, A.W. Yasko, M.J. Yaszemski, and A.G. Mikos. Inst. of Biosciences and Bioengineering, Rice University, Houston, TX. Dept. of Orthopedics, M.D. Anderson Cancer Center, Houston, TX. Mayo Clinic, Rochester, MN.

We have developed a procedure which may have utility in applications involving cell/polymer constructs. One such application is the delivery of cells as part of an injectable material, such as a poly(propylene fumarate) based composite, for the purpose of bone regeneration. The temporary encapsulation of the cells from environmental effects for a short time period, such as composite curing. After approximately an hour, the gelatin would dissolve in the body fluid surrounding the hardened composite, leaving behind a porous scaffold that would attach to the walls of the pores, allowing them to recover nutrients via diffusion through these pores. Following sufficient bone formation, the composite would degrade and be removed from the body. The purpose of this study was to develop: 1) to determine if the osteoblasts could be encapsulated in gelatin microspheres, and 2) to determine if, after gelatin dissolution, the encapsulated cells retain their phenotypic expression. Briefly, primary rat marrow osteoblasts were suspended in a 10% gelatin solution at body temperature medium at 37°C. This solution was added dropwise to stirred mineral oil at 10°C. The hydrophobic effect and temperature change caused microspheres to form. The microspheres were then rinsed, filtered, placed into tissue culture wells containing medium, and incubated. Preliminary results indicate that the cells were successfully encapsulated, and that mineralization and osteocalcin expression do take place at time points (i.e. 28 days). Additional studies are being conducted to help determine evolvement of viability, proliferation, and phenotypic expression over time. These qualities will be elucidated by analysis of DNA content, 3H-thymidine incorporation, alkaline phosphatase activity, osteocalcin levels, and mineralization of the encapsulated cells with respect to unencapsulated controls. We view temporary cell encapsulation as a promising technique with potential for use in a variety of orthopedic applications.

11:00 AM L118
DESIGNING BIOMATERIALS FOR USE IN BONE. Kenneth James, Howard Levene, Joseph Kohn, Department of Chemistry, Rutgers University; Patricia Hoff, J. Russell Parsons, Department of Orthopedics, UMD-New Jersey Medical School, Newark, NJ.

In a series of homologous, tyrosine-based polycarbonates, small changes in the chemical structure of the polymer pendant chain were found to affect the bone response in a long-term (120 day) implantation study. Identically sized pins, prepared from poly[DiT carbonate], poly[DiT carbonate], poly[DiT carbonate], and poly[DiT carbonate] were implanted in the calvarial bone of a transgenic strain of the New Zealand White rabbits. The tissue response at the bone-implant interface was characterized in terms of the presence of a fibrous capsule (direct bone apposition) or presence of a fibrous capsule (reflected to the encapsulation response). The relative frequency of direct bone apposition versus encapsulation was recorded for each polymer throughout the entire period of the study and compared with that of similar pins of polylactic acid. While all four polymers were tissue compatible, there was a correlation between the chemical structure of the pendant chain and the type of bone response observed, with poly[DiT carbonate] having the highest tendency to elicit direct bone apposition. Based on in vitro degradation data and the ability of model polymers with carbamate groups at their surface to chelate calcium ions, it is proposed that the ability of poly[DiT carbonate] to elicit “bone apposition” is caused by the facile hydrolysis of the pendant ethylene group which creates calcium ion chelation sites on the polymer surface. The incorporation of calcium chelation sites into the chemical structure of an implant material appears to be a key requirement if direct bone apposition/bone bonding is desired. This study demonstrates that very subtle changes in the chemical composition of an implant material can have significant effects on the long-term tissue response in a clinically relevant model.

11:15 AM L119
A COLLAGEN/HYALURONATE BIYLAYER MATRIX FOR TISSUE REPAIR. Lin-Shu Liu, Andrew Y. Thompson, Robert C. Spira, Orquest, Inc. Mountain View, CA.

The composition and structure of the extracellular matrix plays a key role in determining the cellular response that results in the formation of new or reparative tissue. In defects or wounds that involve more than one type of tissue, it would be ideal to have a matrix grafting that can simultaneously support distinct tissue repair requirements. For example, in most joint injuries, matrices with the potential to stimulate both bone and cartilage repair would have a significant clinical impact. We have developed a novel bilayer matrix composed of collagen (COL) and hyaluronate (HA) layers covalently cross-linked by divinyl sulfone. Fetal rat calvarial cells seeded on the matrix showed a distinct cellular morphology in each layer after 4 weeks in vitro culture. Cells in the HA layer had a rounded, aggregated, and chondrocyte-like morphology, while those grown in the COL layer were flattened and spread. Biochemical analysis demonstrated that cells in the COL layer expressed high levels of alkaline phosphatase activity (ALP) and low levels of sulfated glycosaminoglycans (GAGs) compared to those in the HA layer. This phenomenon was enhanced by the addition of recombinant human growth and differentiation factor (rhGDF-5). In response to rhGDF-5, cells in the HA layer formed extensive regions of cartilaginous nodules while only small patches of nodules were observed in the COL layer. The rhGDF-5 also increased the level of ALP activity in the COL layer compared to that found in the COL layer in the absence of growth factor in the HA layer. Intramembranous implants of the bilayer matrix with rhGDF-5 retrieved at 28 days revealed the presence of bone and cartilage tissue in the COL and HA layers, respectively. These results demonstrate that the differentiation of cells within distinct regions of the bilayer matrix can be influenced by specific compositional changes.

11:30 AM L120
RECONSTITUTING THE STRUCTURE AND INFORMATION CONTENT OF ACTUAL TISSUE SCAFFOLDS. Eugene Bell, Tissue Engineering, Inc.

ABSTRACT NOT AVAILABLE.

SESSION L1.2 DENTAL/MAXILLOFACIAL BIOMATERIALS

Chair: John Brunski

Monday Afternoon, November 27, 2000
Berkeley (Sheraton)

1:30 PM L12.1
OVERVIEW OF DEVICE RETRIEVAL AND ANALYSIS. John Lemons, University of Alabama at Birmingham, Department of Dental Biomaterials, Birmingham, AL.

Surgical implant devices constructed from synthetic origin biomaterials (metals, polymers, ceramics and composites of these) continue to be utilized in large numbers for musculoskeletal and surgical reconstructions. A number of research laboratories conduct investigations to analyze characteristics of devices explanted at revision or removal surgery. The device is initially classified in terms of: the role of the clinical history (from MEI or DMD), and any observed surface or bulk characteristics that indicate device alterations associated with in vivo function. If unexpected alterations are noted, more detailed in vitro studies are conducted after confirming adequate sample, source confidentiality and available resources. Approximately 5000 explanted devices have been evaluated in our laboratory since 1970 and current results are several hundred per year with the primary sources being orthopedic and dental. Study outcomes will be reviewed in general, followed by selected examples of anticipated outcomes (Tier I), where device changes are found that have not influenced clinical outcome (Tier II) and unanticipated outcomes where in vitro studies have been conducted (Tier III).

2:00 PM L12.2
DESIGN AND CHARACTERIZATION OF NEW Ti-Ag AND Ti-Ag Bi-Alloys FOR CRANO-MAXILLOFACIAL PROSTHESSES MADE BY THREE-DIMENSIONAL PRINTING. Sung-Bam Hong, Harvard School of Dental Medicine, Boston, MA; Noam Eliaz, Gary G. Leik, Ronald M. Lerman, Massachusetts Institute of Technology, H.H. Ullrig Corrosion Laboratory, Cambridge.
New Ti-5Ag and Ti-5Ag-35Sn (wt %) alloys were designed, synthesized by three-dimensional printing (3DPM), and characterized by means of microscopic observations, microhardness and electrochemical tests, and surface analysis. Silver nitrate was found to be an appropriate inorganic reactive binder for atomized titanium powders. The best mechanical properties of the printed material through sintering and liquid-Sn infiltration were determined for the Ti-5Ag and Ti-5Ag-35Sn alloys, respectively. While the Ti-5Ag alloy exhibited superior corrosion and mechanical behavior to the Ti-5Ag-35Sn latter showed better dimensional stability. The applicability of 3DPM for fabricating complex ceramic-micr-no-facial (and other) prostheses is discussed in detail.

2:15 PM LL2.5
STRUCTURE-NANO MECHANICAL PROPERTY STUDY IN ENAMEL USING MOLECULAR APPROACHES VIA BIOIMMUTICS. Hassan Fang and Mehmet Serkan Y., Materials Science and Engineering, University of Washington, WA; Michael L. Paine, Wen Luo and Malcolm L. Sned, The Center for Craniofacial Molecular Biology, School of Dentistry, The University of Southern California, Los Angeles, CA; Shire N. White, School of Dentistry, University of California, Los Angeles, CA.

Enamel is the unique composite material forming the outer covering of vertebrate teeth. It is the hardest tissue in the vertebrate body, containing long-range-ordered hydroxyapatite (HAP). Enamel functions under immense loads, in a wet, bacterial/indenter environment generally without catastrophic failure. Unlike most other bi-crystalline materials, which are monoclinic in origin, enamel is a mixed-crystal domain called amelodentine. We have focused our investigations on the formation of enamel using cell and molecular approaches and by coupling findings from these techniques to biophysical investigations at the mesoscopic. We have employed 3DPM to the role of amelogenin protein by creating “gain of function” and “loss of function” transgenic animal models. For example, the principle protein of forming enamel, we have identified two domains. We engineered amelogenin containing deletions to eliminate both of these domains. We investigated enamel formation in transgenic mice. Morphological, structural and crystallographic variations were correlated to non-mechanical properties of mouse teeth by electron and atomic force microscopy (nano-indentation) techniques. We found that there are significant changes in the mineralization behavior of the teeth generated using these domain deletions that correspond to decreased in mechanical performance. For example, non-bonded domains by as much as 20% while elastic modulus up to 15% in comparison with nontransgenic teeth. The formation of enamel is an intricate biomineralization process requiring proper formation and assembly of amelogenin. Failing to do so results in altered processes leading to incomplete biomineralization of the hard tissue with inferior mechanical properties. Supported by grants from the NIH, The National Institute of Dental and Craniofacial Research.

3:15 PM LL2.5
APATITE GROWTH ON BIOACTIVE GLASSES IN ARTIFICIAL SALIVA. Sarah E. Einfeldt, Robert F. Cook, Lorraine F. Francis, University of Minnesota, Dept of Chemical Engineering and Materials Science, Minneapolis, MN.

Bioactive materials have been widely researched for orthopaedics and dental implants because of their ability to bond to bone. When bioactive materials are placed against bone in vivo, they form a layer of bone similar to their own growth. The best known bioactive glass is a porous apatite layer on soaking in simulated body fluid correlates with these in vivo results. Like bones, teeth are primarily composed of hydroxyapatite. Our results show dentin from extracted human teeth adheres to bioactive glasses when soaked in saliva at 37°C. To understand this result further, bioactive glasses in the Na2O-CaO-P2O5-SiO2 and MgO-CaO-P2O5-SiO2 systems were soaked in artificial saliva and the growth of crystal-like apatite on the bioactive glasses was monitored. Preliminary results showed growth over soaking times varying from 1 to 42 days was assessed as well as the effects of three different glass surface finishes. Samples were analyzed using x-ray diffraction, ice beam scattering, infrared spectroscopy and electron and optical microscopy. X-ray data reveals apatite peaks on the Na2O-containing glass as early as 10 days exposure with continuous evolution of more crystalline material through 42 days; in contrast, apatite peaks are not apparent on the MgO-containing glass until 42 days. The most interesting observation is that apatite development was greatest on the surfaces of intermediate roughness, implying competition between nucleation and growth on the glass surface. In addition to the difference in the rate of apatite development, the morphology of the apatite was different for the two glasses: micro-crystalline and plate-like for the Na2O and MgO-containing glasses, respectively.

3:30 PM LL2.5

In the present work we present the theoretical and experimental results from the synthesis of hydroxapatite by using natural precursors. Mellite-Eugeduuro Barrosolii sp. Nov. was used in raw material in the synthesis of the precursors: Calcium Oxide (CaO), Calcium Hydroxide (COH) and Calcium (CaCO3). The reaction among the precursors was carried out under hydrothermal conditions. Two distinctive pressures were selected: 1.4 MPa and 6.5 MPa. The reaction time was varied from 2, 4, 6, 8 10 to 20 hours. In order to compare the products of the stoichiometric reaction and the effect of adding an excess of CaO, both sets of products were characterised by XRD, LV-SEM, EDS, FTIR and TEM. One observed several crystalline habits such as: needle-like, plate-like and needle-like and there is a yield dependence on the reaction conditions. The gradual formation of hydroxypatite was studied by comparison of the experimental results with the theoretical calculations obtained by molecular simulation.

4:00 PM LL2.5
ATOMIC-SCALE INTERFACIAL STRUCTURE OF HYDROXYAPATITE HIGH-RESOLUTION TRANSMISSION ELECTRON MICROSCOPY. Kimiyasu Sato, Yoshihiro Kogure, Toshiyuki Ikoma, Yura Kumagai and Junzo Tsukada, CHEST, Japan Science and Technology, Saitama, JAPAN.

Hydroxyapatite (HAp) is a main mineral component of bone and tooth, and is widely used in orthopaedic, dental and industrial fields. To achieve the design of novel bioactive materials by combining HAp with organic substances, a thorough understanding of the surface structure of HAp is fundamentally important. HAp crystals are often faceted with six equilateral facets, expressed as {100} planes for a hexagonal symmetry. We tried to elucidate the atomic configuration of the {100} faces in stoichiometric HAp crystals by high-resolution transmission electron microscopy (HRTEM). We prepared carbonate-free and almost stoichiometric HAp sintered body. HAp studied has been proved to have monoclinic symmetry, because ordered hydroxyl ions lower the symmetry of the crystal structure. As crystallographic difference between hexagonal and monoclinic symmetries is, however, quite insignificant, the crystal structure is formally described by the hexagonal symmetry in the present paper. The sintered HAp was polished mechanically and thinned by argon ion etching. HRTEM observations were performed on a JEM-2010 (Hitachi) at 200 kV. To determine the atomic configuration, HRTEM
images were compared with calculated images obtained by multislice image simulations. Grain boundaries with many planes were observed from the [100] direction. HAP grains had interfaces between hydroxyl columns, where existed PO₄ tetrahedra and Ca(2) ions. When specimens were subjected to electron beam irradiation for a long duration, HAP crystals partially divided. According to TEM observations, even from the [001] direction, the divided regions took roughly hexagonal shapes surrounded by many planes. The interfacial structures between crystalline and amorphous regions were identical to that observed at the grain boundaries. As both grain boundaries and crystalline/amorphous interfaces were terminated with the [100] faces of the same atomic configuration, such structure was conjectured to be essential for interatomic bonds at the HAP crystal interfaces, and gives a fundamental design concept for organic/HAP composites.

4:00 PM LL2.8
THE NANOSTRING MECHANICAL PROPERTIES OF HUMAN MOLAR DENTAL ENAMEL. Adam B. Mian, Suhm Kim, Janet Ouy, Ken Livi, Mark Tesfai, Timothy P. Wells, The Johns Hopkins University, Dept of Materials Science and Engineering, Baltimore, MD.

The mechanical behavior of dental enamel has been the subject of many investigations. These initially assumed the tooth enamel was a more or less homogeneous material with uniform mechanical properties. Now it is generally recognized that the mechanical response of the enamel depends upon location, prism orientation and chemical composition. This study used nonindentation testing to map out the properties of dental enamel over the axial cross-section of an upper 2nd molar. The local variations in mechanical characteristics are correlated with changes in microstructure and chemical content across the cross-sectional enamel sample. SEM is used to examine the microstructure and micromorphology techniques are used to examine changes in chemical composition. The range of hardness (H) and elastic modulus (E) observed over an individual tooth have been found to be far greater than previously reported. At the enamel surface H > 6 GPa and E > 15 GPa, while at the amelodental junction H < 3 GPa and E < 7 GPa. These variations correspond closely to variations in the prism orientation. At the enamel surface the prisms' axial positions are generally perpendicular to the occlusal surface, while at the amelodental junction they are oblique in orientation with many lying parallel to the occlusal surface. The chemical composition of the enamel was also found to vary in a similar manner to the mechanical properties with the constituents of hydroxyapatite (PO₄, CaO) and chloride highest at the surface. There was no discernable trend in fluorescence content. By the amelodental junction the amount of Na₂O and MgO were found to increase. Intriguingly, on the buccal side of the tooth, at the occlusal surface, the enamel was harder and more elastic than on the lingual side. The interior enamel, however, was softer and more compliant on the buccal side than the lingual side.

4:15 PM LL2.9
MAXILLOFACIAL IMPLANTS OF POLARIZED HYDROXY-APATITE PLASMA-SPRAY-COATING TITANIUM. Hideki Sugawa, Tokueki Kobayashi, Masuo Usulhaim, Suzuki Nakamura, Morinosuke Ogushi, Kikuiro Shimizu, Tokyo Medical and Dental Univ, Inst of Biomaterial and Bioengineering, Tokyo, JAPAN.

We have proved that sintered hydroxyapatite (HAP) ceramics are electrically polarized and that the polarized HAP provides characteristic interactions with bone tissues both on the positive and the negatively charged surfaces. In this research, electrical polarizability of plasma-sprayed HAP coatings on titanium was demonstrated by thermally stimulated current (TSC) measurement. Moreover, the biological reactions of the polarized HAP coatings were investigated using bones of beagle dogs. The HAP layer with a thickness of 30 µm was coated on titanium by plasma-spray technique. The HAP coated titanium was electrically polarized in a dc field of 1 kV/cm "̈" at 300°C. The samples were implanted into the defects formed on bones by dental burrs. The bones were extracted at 1, 2 and 4 weeks after the administration and fixed with 20% neutral buffered formalin solution. The bone tissue sections were observed by light microscopy and scanning electron microscopy. The electrical storage of the polarized HAP coatings was clearly detected by TSC measurement. The new bone formation in contact with the Nsurfacle was observed at 1 week. The bone formation was previously correlated to the vicinity of the negatively charged surface (Nsurfacle) was significantly promoted compared to that of the non-polarized HAP coating surface. Histological results suggest that the electrical polarization treatment is effective for osteoconductivity improvement of the HAP coating layers and confidently predicted that the polarization technique is applicable to maxillofacial implant systems of HAP plasma-spray coated titanium.

4:30 PM LL2.10
ENGINEERING MONOLAYER DENTAL CERAMIC CROWN STRUCTURES. Yan Deng, Hoon Kim, Young-Woo Rhee and Brian R. Lawn. National Institute of Standards and Technology, Materials Science and Engineering Laboratory, Gaithersburg, MD.

Simple relations are derived for the onset of competing fracture modes in ceramic coatings on compliant substrates from Hertzian-like contacts. These structures are intended to simulate the basic features of ceramic crown systems in oral function. Special attention is given to a deleterious mode of radial cracking that initiates at the lower coating surface beneath the contact, believed responsible for the bulk of all-ceramic crown failures. Critical load relations for the onset of cracking are expressed in terms of well-documented material parameters and as geometrical parameters: coating thickness and sphere radius. Data from selected ceramic coating materials on polycarbonate substrates are used to demonstrate the validity of the relations. The formulation provides a basis for designing ceramic crowns with optimum damage resistance.

4:45 PM LL2.11
CONTACT DAMAGE MODES IN MULTILAYERED STRUCTURES FOR DENTAL APPLICATIONS. Pedro Miranda, Fernando Guibert, Universidad de Extremadura, Dept Electromecánica, Badajoz, SPAIN; Antonio pajares, Francisco L. Cumbres, Universidad de Extremadura, Dept Fisica, Badajoz, SPAIN; Brian R. Lawn, Materials Science and Engineering Laboratory, National Institute of Standards and Technology, Gaithersburg, MD.

We have characterized contact damage modes induced in model brittle polymer coatings on soft substrates that emulate the essential trilayer structure and material properties of dental crowns. To simulate oral conditions we have conducted indentation tests using hard spheres as indenters, over clinically relevant loads. Experiments have been performed on model transparent trilayer structures, to enable in situ observation during the contact. Individual layer surfaces are preferentially abraded to introduce uniform flaw states and so allow each crack type to be studied separately and controllably. Fracture occurs by one cracking at the top surface or by radial cracking at the bottom surface of the brittle coating layers. Critical loads for crack initiation are measured as a function of layer thickness and elastic modulus mismatch. Finite element analysis (FEA) has been used to calculate contact stresses and shear stress fields in the layer structure using ABAQUS codes, with material parameters input (modulus, yield stress, work hardening, strength) obtained from independent indentation tests on monolithic materials. Fracture criterion are imposed to calculate critical loads for each crack mode for comparison with experimental values. The results yield functional fracture mechanics relations for the critical loads, providing the basis for design of trilayer systems with optimal damage thresholds.

SESSION L13: ORTHOPEDIC BIOMATERIALS
Chair: Orhan K. Muratoglu
Tuesday Morning, November 28, 2000
Berkeley (Sheraton)

8:30 AM LL3.1
Henry Rubash, Massachusetts General Hospital, Boston, MA.

ABSTRACT NOT AVAILABLE

9:00 AM LL3.2
STRUCTURE AND PROPERTIES OF A NEW NANOCOMPO SITE POLY(METHYL METHACRYLATE) BONE CEMENT. Anuj Bellare, Andreas Gonoll, Wolfgang Fitz, Richard D. Scott, Thomas S. Thornhill, Dept of Orthopedic Surgery, Brigham and Women's Hospital, Harvard Medical School, Boston, MA; David A. Baker, Lisa A. Prieto, Dept of Mechanical Engineering, University of California at Berkeley, Berkeley, CA.

Orthopedic bone cement is widely used for fixation of total joint replacement prostheses. These cements usually comprise a partially polymerized poly(methyl methacrylate) or a poly(methyl methacrylate-co-styrene) random copolymer powder and a methyl methacrylate liquid component. The powder component also contains a reaction initiator to initiate setting when the two components are mixed. In addition, micrometer size filler particles of either barium sulfate or zirconium oxide are present in the powder to radiopaque the cement, thereby enabling the orthopedic surgeon to monitor fractures in the implanted bone cement using radiographic diagnostics. Incomplete dispersion of the radiopaque particles during mixing of the two components is known to result in the formation of particle agglomerates of 50–200 micrometer diameter. These large defects can reduce the fracture toughness of PMMA cements, leading to early
fracture of the cement and loosening of the implant, ultimately necessitating early revision surgery to replace the implant. In this study, the microstructure and radiographic properties were replaced by commercially available nanosized (100 nm diameter) barium sulfate radiopaque particles. The resulting noncomposite PMMA bone cements were thereafter characterized using low voltage high resolution scanning electron microscopy, and by light and thermal x-ray scattering at the UNICAT beamline of the Advanced Photon Source, Argonne National Laboratory. The noncomposite and microcomposite PMMA bone cements were thereafter subjected to uniaxial tensile and fatigue testing. The outcome of such work-of-fracture and number of cycles to failure, respectively, of the noncomposite cement. These results, the morphological observations and the structure-property relationships governing fracture toughness of PMMA cements will be presented.

9:15 AM LL13.3
CONTRIBUTIVE EARLY STAGE MINERALIZATION ADJACENT TO PROSTHETIC IMPLANTS IN CANINE AND HUMAN
MODEL BONE EXPLANTS, Alexandra Porter, Cambridge University, Dept of Materials Science & Metallurgy, Cambridge, UNITED KINGDOM; Melanie J. Courty and Gordon W. Blunn, Royal Orthopedic Hospital, Biomedical Engineering Centre, Stanmore, Middlesex, UNITED KINGDOM; Myron Spector, Brigham & Women's Hospital and Harvard Medical School, Department of Orthopedic Research, Boston, MA; Linn W. Holb, MIT, Dept of Materials Science & Engineering, Cambridge, MA.

Experiments of femoral bone containing hydroxyapatite (HA) coated implants from animal and human models have been studied to compare the course of early-stage mineralization of new bone forming adjacent to the coated implants. The canine implants were from distal femur sites implanted with 6-mm HA-coated Ti-6Al-4V rods for periods between 2 hours and 14 days. The human implants come from an inventory of 90 hip replacements involving HA-coated femoral stems which were retrieved at autopsy; the shortest residence time was 14 days. Explants were fixed in formalin and embedded in PMMA, mechanically cross-sectioned, then embedded in epoxy and microtomed for transmission electron microscopy (TEM) in the vicinity of the bone/implant interface. Diffraction contrast and high-resolution TEM were used to study bioceramic mineral modification and apatite mineralization of the extracellular matrix, while heavy metal stains were used to locate fibroblast proteins in the interface region. The canine model studies revealed that HA platelet precipitation occurs as early as 2 hours without apparent foci and cluster nucleation of HA ribbons within 3 days, collagen invades by 10 days and directs the subsequent mineralization of HA platelets which appear to nucleate in the hole zones of the collagen molecular fibril assemblies. Results from the ongoing human explant studies will be compared.

9:30 AM LL13.4
BIOLOGICAL REACTION OF POLARIZED HYDROXYPATITE PROSTHESIS IN BONE MARROW. Tomoko Saki1,2,3, Tatsuki Kobayashi1, Mieko Ueshima1, Sacoshi Nakamura1, Sadashi Morita1, Ken-ichi Shinomiya1, Kenhiro Yamashita1, Tokyo Medical and Dental University, Institute of Biometrics and Bioengineering, Dept of Orthopedics, Tokyo, JAPAN.

We have already reported that hydroxyapatite (HAP) is electrically polarizable and that the higher rate of bioactivity is attained on the polarized HAP surface compared with the non-polarized HAP surface by histological examination in vivo. HAP disks with a size of 10 mm diameter x 1 mm thickness were prepared. These specimens were cramped with Pt plates, heated up to 300°C, then subjected to the polarization treatment in a dc field of 1 kV/cm for 2 hours. As a control, HAP disks without the polarization treatment were used. The femora of six Japanese white rabbits were fenestrated and the specimens were implanted. The rabbits were sacrificed at 1, 2 and 4 weeks after the invasion and the samples containing the surrounding bones. The specimens of femora were fixed and examined by optical microscopy and scanning electron microscopy. The positively charged surface (P-surface) demonstrated the obvious new bone formation at 1 week after the operation. The negatively charged surface (N-surface) showed different osteointegration characteristics from the positively charged surface of the HA. The negatively charged HAP was characterized by a new bone formation as 2 weeks after the operation, though the thickness and direct bone attachment was extensively recognized on the P-surface. These facts showed that the surfaces of the electrically polarized HAP have different osteointegration characteristics from the non-polarized HAP. The earlier new bone formation appeared on the both surfaces of the polarized HAP. These findings suggest that the electrical polarization can accelerate the new bone formation on HAP surfaces and will be of great benefit to the bone regeneration in orthopedic surgery.

10:15 AM LL13.5
STRENGTH AND TOUGHNESS OF AN APATITE CEMENT. Victoria C. Jing, Reinaldo H. Daakkass, Stanford University, Dept of Materials Science and Engineering, Stanford, CA.

Apatic materials resembling bone mineral have received considerable attention due to their bioocompatibility and osteoinductive properties. Particularly when formed cementingly at physiological temperature, they present significant potential for orthopedic applications such as rapid bone repair, fracture fixation, and augmentation of load bearing implants. To date, the tendency of fracture of such apatites have been extremely low, limiting their use in load-bearing applications. To better characterize the mechanical properties of a hydroxyapatite bone cement, this study investigates its fracture toughness and flexure strength, as well as the mechanisms of failure. Strategies to enhance the mechanical properties of this bone cement are also examined, utilizing the addition of organic phases such as collagen. Second phase constituents are found to increase the fracture strength and reliability. Micromechanical models have been developed to account for these strengthening and toughening effects. Implications for the integrity and reliability behavior of such synthetic bone mineral substitutes in load bearing applications are considered.

10:30 AM LL13.6
INFLUENCE OF HIGH SPEED MIXING ON AGGLOMERATION OF NANOMETER BARIUM SULFATE IN BONE CEMENT. Wolfgang Fitz, Andreas Gomoll, Anuj Bhide, Richard D. Scott, Thomas S. Thorndill Orthopedic Research Laboratory, Brigham and Women's Hospital, Harvard Medical School, Boston, MA.

Bone cements revolutionized implant fixation of the low friction arthroplasty introduced by Sir Charnley in the mid 1950s. The bone cement attaches the implant to the bone and transfers forces to the bone while minimizing material fatigue. To date, the mechanical properties of the bone cement have been the leading cause of mechanical loosening. A comparison of radiopaque bone cement with conventional radiographic bone cements with different percentages of barium sulfate resulted in a two-fold reduction of impact strength of different bone cements with different amounts of radiopacifiers added. It is our opinion that, the presence of radiopaque fillers weakens the interface matrix. Either a needle-like matrix platelet agglomeration of the radiopaque or the presence itself may be the cause for the inferior mechanical properties. Our hypothesis is that a uniform dispersion of barium sulfate within the interface matrix will not weaken the mechanical performance compared to radiopaque cement. Furthermore using the principals of the emerging nanotechnology we hope to strengthen the fracture toughness with the use of nanometer sized barium sulfate particles compared to radiopaque bone cement and even surpass its mechanical properties without changing the composition, setting time and handling characteristics of this new nanocomposite acrylic bone cement. We claim that agglomeration of radiopacifiers can be prevented with high speed mixing of small uncoated barium sulfate particles with an average particle size of 10 nm, without significantly changing viscosity and setting time. Futureprognostic analysis will be used to demonstrate the beneficial effect of high speed mixing of nanometer sized radiopacifier compared to commercial formulations and radiopaque cement. In this study, an experimental high speed mixing of nanocomposite cement is compared to vacuum mixed Simplex P CMW (radio-opaque and radiolucent) and high-speed mixed Simplex P JEOL 6200FV low voltage scanning electron microscope (LVHRSEM) examined the surface fracture of all cements. Agglomeration of barium sulfate fillers were not seen in the experimental nanomaterial cement as compared to the commercial formulations. However we observed a uniform dispersion of the 100 nm large barium sulfate particles. High speed mixing of nanometer sized radiopacifier prevails a uniform dispersion with no agglomeration and no significant changes in viscosity and setting time. Further mechanical testing will provide the benefit of this new nanocomposite bone cement.

10:45 AM LL13.7
CONDITIONS LEADING TO INCREASED INTERFACIAL POROSITY IN CEMENTED FEMORAL STEM ALUMINUM. Stephen Spiegelberg, Cambridge Polymer Group, Inc., Somerville, MA.

This paper examines the dependence of the rheological properties of bone cement on the femoral stem-bone cement interfacial porosity in total hip and knee replacements. A study by James et al. (1993) showed that interfacial porosity from in vivo and in vitro components did not vary with surface finish, stem material, or centrifugation. They concluded that the rheological, or flow, behavior of the bone cement could explain the presence of these porosities. We present a study that examines how bone cement rheometry and stem insertion studies that fully characterizes the cure behavior of bone cement, and provides a quantitative analysis of the advantages and disadvantages of each method that will lead to femoral stem cement interfacial porosity. It was proposed that
the level of porosity at the cement-femoral stem interface is a function of the advancing contact angle at the bone cement wetting line, which in turn is a function of the stem insertion rate and the relaxation time of the bone cement. This later relationship is known by the dimensionless parameter the Deborah number.

11:30 AM *LI3.10 EVALUATION OF ARTIFICIAL KNEE JOINTS. Peter Walker, Royal National Orthopedic Hospital Trust.

ABSTRACT NOT AVAILABLE

SESSION LI4: MUSCULOSKELETAL BIORATIONAL MATERIALS CONSIDERATIONS. Mike Yasuiwaki, Mayo Clinic.

ABSTRACT NOT AVAILABLE

2:00 PM LI4.2 STEREOLITHOGRAPHIC PROCESSING OF CERAMIC ORGANIC COMPOSITES FOR ORTHOPEDICS. Jim H Lee, Robert K. Pearlstone and Hye A. Ahn, Princeton University, Dept. of Chemical Engineering and Princeton Materials Institute, Princeton, NJ.

Ceramic stereolithography (CSL) is used to fabricate complex-shaped ceramic/organic composites by laser photocuring a concentrated ceramic dispersion in photocurable solutions layer-by-layer. The main processing parameters in CSL such as layer thickness, resolution, hatch spacing, and cure depend on the knowledge of light propagation in concentrated multiple scattering dispersions. By incorporating biocompatible polymers as the matrix phase, bioceramics with controlled microstructure may be constructed.

In studies dealing with the processing of these reinforced biocomposites, we investigated the depth of curing for model resin systems as a function of photoinitiator concentration. Alumina [Al₂O₃] was used as the reinforcing phase in a matrix of 2,2-bis(4′-hydroxy-3-methylcylohexyl)propionate (Bis-GMA), a commonly used monomer in dental fillings. Free radical polymerization was initiated with 2-benzyl-2,4,4-trimethylpentanoyl-1-(4-morpholinophenyl)-4-butanone (DBMP).

An optimal photoinitiator concentration that maximized the gel cure depth was observed. Two regimes were shown to exist in which the swell ratio was minimized or maximized. The study showed that photoinitiator plays a significant role in controlling the quality and performance of the formed gel network, with special regard to thickness of cured layers. This holds strong implications not only for stereolithographic production of microscale biocomposites, but for dental restorations in which coatings are photocured, as well.

2:15 PM LI4.3 PROPERTIES AND PROPERTIES OF NANOSTRUCTURED HYDROXYAPATITE-BASED BIOMATERIALS FOR ORTHOPEDIC APPLICATIONS. Edward Ahn, Jackie Ying, Massachusetts Institute of Technology, Dept. of Chemical Engineering, Cambridge, MA; Myron Spector, Harvard Medical School, Dept of Orthopedic Surgery, Boston, MA.

Hydroxyapatite (HAP) has generated great interest in the search for advanced orthopedic and dental implant materials as it elicits a favorable biological response. However, it is sensitive to nonstoichiometry and impurities in its synthesis and processing. Thus, conventionally processed HAP materials often lack phase purity and homogeneity, resulting in poor mechanical properties.

Nanostructure processing is employed for microstructural control to achieve the desired mechanical characteristics for HAP. It also further enhances surface reactivity to promote tissue integration. Through nanostructure processing, superior chemical homogeneity, microstructural uniformity, and ultrafine grain sizes (125 nm) have been achieved so that flaw sizes can be significantly reduced in fully dense HAP monoliths. As a result, nanostuctured HAP monoliths provide for superior compressive (900 MPa) and bending (200 MPa) strengths and fracture toughness (1.3 MPa m¹/2).

Microstructure processing is also utilized to further improve mechanical properties by dispersing zirconia nanoparticles in the HAP matrix. The effects of yttria-stabilized zirconia dispersions and unmodified zirconia dispersions on the mechanical properties have been investigated. The incorporation of highly dispersed unmodified zirconia (3 wt%) significantly increased the fracture toughness (2.9 MPa m¹/2) and bending strength (280 MPa) of the HAP-based systems. Our high strength, nanostuctured HAP monoliths also provide a model two-dimensional
 Micropatterned Surfaces Modified with Select Peptides Promote Exclusive Interactions with Osteoblasts. M.E. Hansen, S.C. Deslandes, T.T. Andersen, R.B. Bizios, Department of Biomedical Engineering, Rensselaer Polytechnic Institute, Troy, NY.

Osteointegration is an integral part of the clinical success for orthopedic/dental implants and is dependent on the interaction of osteoblasts with material surfaces. For anchorage-dependent cells, such as osteoblasts, a crucial event in this process is adhesion, a prerequisite for subsequent cell functions. Micropatterning of bioactive, adhesive peptides represents a novel, design approach to enhance osteoblast/biomedical interactions at the tissue/implant interface by promoting and spatially-directing cell attachment to implant surfaces. In the present study, standard microcontact printing techniques were used to pattern circles [diameters 10, 50, 100, and 200 micrometers] with (remarkably lipophilic) diethyl hexamethylenetriamine (DETA) or hydrophilic compound, surrounded by octadecyltrichlorosilane (OTS), a hydrophobic compound, on borosilicate glass, a model substrate. The DETA regions were further modified with (deposited by immersion in either the adhesive peptides Arginino-Glycine-Aspartic Acid-Serine (RGDS) and Lysino-Arginino-Serino-Arginino (KSR) or the non-adhesive peptides Arginino-Acidic-Acidic-Serine (RDGS) and Lysino-Serino-Serino-Arginino (KSR)). After four hours under standard cell culture conditions, adhesion of either osteoblasts or fibroblasts in Dulbecco’s Modified Eagle Medium in the absence of serum on surfaces modified with the non-adhesive peptides RDGS and KSR was observed. In contrast, both osteoblasts and fibroblasts adhered and formed clusters onto the DETA circles modified with the adhesive peptide RGDS; whereas only osteoblasts adhered and formed clusters onto DETA circles modified with KSR, a peptide that selectively promotes osteoblast adhesion (Lee et al., 1998). These results provide evidence that patterning of select peptides can directly adhesion of specific cell lines to predetermined regions on material surfaces. Establishing such region-selective compatibility specifically for osteoblasts but not for fibroblasts] on the surface of the next generation of orthopedic/dental biomaterials will promote proactive interactions at the bone/implant interface leading to improved in vivo osteointegration of implants. References: Lee et al., Tissue Engineering, 1:135-145, 1995; Lee et al., J. Biomed. Mater. Res., 30:371-377, 1996.

Mechanical Behaviour of Poly-Hydroxyalkanoates (PHA) and Poly-Hydroxybutyrate As Bone Implant Biomaterials. Rubén Sanchez, Márco Moriera, Norma Galego, Polymer Section, Advanced Materials Laboratory, North Fluminense State University, UENF, Campos de Goiás, RJ, BRAZIL.

Various synthetic materials have been usually utilized as reinforcement in fracture fixation to the human bone. The poly-(beta-hydroxyalkanoates) (PHA) could replace traditional polyolefins based on their biocompatibility, adequate fracture toughness and fatigue strength. The composites were developed using previously characterized polyhydroxyalkanoates (PHA) with 30, 40 and 50% of poly(lactic acid) composition, and poly-hydroxybutyrate (HA) with 0.1 mm particle diameter. A polymer solution was previously prepared and mixed with HA. The mixture was homogenized and pressed by hot uniaxial pressing at 130 °C above polymer melting point. The mechanical properties of the composites were studied by compression test, and the fracture area was explored by Scanning Electron Microscopy (SEM). HA was used due to its chemical and structural similarity with the human bone. On the other hand biodegradation of PHA is a secondary component, hence being biodegradable and introduce an appropriate mechanical property, tensile strength values close to bone (357 MPa) and cortical bone (41 MPa). The rate of chemical hydrolysis of PHAs is variable, and may be hindered by biomedical implants where the bioactivity of HA and the biodegradability of PHAs can be combined to avoid particle migration from the implant area before growth of a new tissue and a favorable answer to induce a bone formation taking advantages of the slow polymer degradation.

3:30 P.M. LL4.6

A NOVEL COMPOSITE BIOMATERIAL FOR APPLICATIONS INVOLVING ELECTRICAL STIMULATION OF BONE. Peter Sung, Polytechnic Institute, Department of Biomedical Engineering, Troy, NY; Puckel A. Attia, Rensselaer Polytechnic Institute, Department of Materials Science and Engineering, Troy, NY; Klaus Ullmann, Rensselaer Polytechnic Institute, Department of Biomedical Engineering, Troy, NY; and David Esakk, Stanford University, Albuquerque, NM.

Literature reports have shown that bone repair (for example, in animal models) occurs at an accelerated rate under electrical stimulation delivered to the injury site via implantable, metal electrodes. Novel biomaterials (for example, current-conducting composites) provide new unexplored alternatives for biomedical applications that require delivery of electrical stimulation for the purpose of bone fracture repair. Materials with current-conducting properties could eventually lead to alternative therapeutic methodologies; in conjunction with cultured cells, they also provide models to experiment and, thus, elucidate the cellular/molecular-level mechanisms responsible for osteogenesis under electrical stimulation. In the present in vitro study, the effects of electric current stimulation on select functions of rat calvarial osteoblasts (the bone-forming cells) on novel current-conducting polymeric acid/carbon nanotube [80:20 w/w] composites were investigated. Compared to control, that is, osteoblasts cultured on polymeric acid/carbon nanotube composites under standard cell culture conditions, but no electrical stimulation), exposure of osteoblasts (occurring in the absence of 10 to 10 Hz for 6 hours daily for 2 consecutive days resulted in a 40% increase in cell proliferation. Moreover, exposure of osteoblasts to electrical stimulation of 10 µA at 10 Hz for 6 hours daily for 21 consecutive days resulted in a 300% increase in the concentration of extracellular matrix calcium. Osteoblasts exposed to 10 µA at 10 Hz for 6 hours resulted in upregulation of Collagen Type I mRNA expression; during the same time interval, there was no Collagen Type I mRNA expression by osteoblasts cultured on the acid/carbon nanotube composites under similar conditions but no electrical stimulation. These results provide evidence that electrical stimulation promotes osteoblast function for the chemical composition and structure of the organic and inorganic phases of bone.

3:45 P.M. LL4.7

FABRICATION AND CHARACTERISATION OF CALCIUM PHOSPHATE AND LIPOSOMES COMPOSITES AS AN IMPLANT COATING. Tariq Posangotam, Ian T. Chernouksa, Department of Materials, University of Oxford, Oxford, UNITED KINGDOM.

Calcium phosphate ceramics, especially hydroxyapatite have been effectively utilized in dental and orthopedic applications due to their excellent osteoconduction and biocompatibility. It is also known that liposomes are widely used as targetable drug delivery devices. The purpose of this paper is to show that we synthetically combine calcium phosphate and liposomes. This composite can be deposited electrochemically as a coating. Thus, we combine the osteogenic potential of hydroxyapatite with the potential for targeting drugs and active ingredients to bone. This will lead to improved treatments for such conditions as osteoporosis, arthritis and tumours. Agents to prevent infection and enhance bone formation may also be added. Liposome vesicles were prepared by sonication of phospholipid-dendron and then introduced into an aqueous solution of calcium and phosphate ions supersaturated with respect to hydroxyapatite. Precipitation occurred on the outer surface of the liposome vesicles. These composite precipitates were deposited onto a cathode using an electrodeposition method at physiological temperature (37°C). Scanning electron microscopy (SEM), powder x-ray diffraction (PXRD) and Fourier transform infrared spectroscopy (FTIR) were used to analyse the morphology, structure and chemical composition of the composite coatings. The results from PXRD and FTIR show a mixture of amorphous and poorly crystalline hydroxyapatite. Enhanced crystallinity developed on ageing in the conditioning medium. This was very likely due to electrodeposition-impregnant interaction, subsequently enhanced the precipitated HA deposited solely at the outer surface of the liposome. SEM micrographs demonstrated a thin uniform coating at the nanoscale level. These results suggest that liposomes can be coated externally with calcium phosphate. The fraction of crystallinity can also be altered. It is expected that these liposome composite assemblies will have widespread use in aiding the treatments of bone disorders.
4:00 PM LL4.8
TRANSPORT BEHAVIOUR, MECHANICAL PROPERTIES AND BIOMCOMPATIBILITY OF A BINARY NiTi AND A TERNARY NiTiCu ALLOY. Mohammed Es-Souni, University of Applied Sciences, Materials Testing and Joining, Kiel, GERMANY; Martin Es-Souni, Hedge-Fischer-Brendle, Clinic of Orthodontics, Christian-Albrechts University, Kiel, GERMANY.

In the present paper, the transformation behaviour, mechanical properties and biocompatibility of two commercial NiTi and NiTiCu shape memory alloys were investigated. The experimental results were obtained from calorimetric results in the temperature range from -80 to +80 C and the bending properties in the temperature range from 0 to 60 C were reported. Furthermore, the results of biocompatibility experiments using the MTT (3-[4,5-dimethylthiazol-2-yl]-5-diphenyltetrazolium bromide)-test on epithelial cell cultures from human gingiva in the presence of both alloys are described. The morphology of the cells after allografting with the alloys is also investigated using SEM. It is shown that the mechanical properties are strongly temperature dependent. Biocompatibility is shown to depend on the alloy composition with the binary alloy being characterized by a higher biocompatibility.

4:15 PM LL4.9
NANOCERAMICS AS THE FUTURE ORTHOPAEDIC/DENTAL BIOMATERIAL. Thomas J. Webster, Richard W. Siegel*, Rena Bizios, Departments of *Materials Science and Engineering and Biomedical Engineering, Remsen Polytechnic Institute, Troy, NY, USA.

The objective of the present in vitro study was to investigate, for the first time, cytocompatibility properties of nanophase (that is, novel material formulations with grain sizes less than 100 nm) ceramics pertinent to clinical/orthopedic/dental applications. Nanophase was conventional (that is, grain sizes greater than 100 nm) alumina, titania, and hydroxyapatite compacts were prepared as previously described (Webster et al., 1999). Neutronic rat calvaria osteoblasts (bone-forming cells) were seeded (at a cell density of 40,000 cells/cm2) onto the ceramics of interest to the present study and cultured under standard cell culture conditions (that is, a 37°C humidified, 5% CO2/95% air environment) in Dulbecco Modified Eagle Medium (DMEM) (supplemented with 10% fetal bovine serum (FBS), 50 μg/mL ascorbic acid, and 10 mM β-glycerophosphate) for 21 and 28 days. At that time, calcium content in the extracellular matrix was quantified using a commercially available kit (Sigma) and following manufacturer's instructions. Primary osteoblast-like cells (bone-resorbing cells) isolated from femurs of Wistar rats were seeded (2x103/cm2) onto the ceramics of interest to the present study and cultured under standard cell culture conditions in DMEM (supplemented with 10% FBS, 10-8 M-algin, 25 μg/mL vitamin D3, and 1% antibiotic/antimycotic solution) for 10 and 13 days. At that time, resorption pits on the ceramic surfaces were visualized and counted using electrical microscopy (Olympus IX70) with image analysis software (Image Pro). Compared to conventional ceramics, deposition of calcium-containing mineral by osteoblasts and formation of resorption pits by osteoblast-like cells were significantly (p < 0.01) greater on nanoporous ceramic surfaces all after time periods tested in the present study. By demonstrating that bioceramics can be designed and fabricated through control of grain size to possess improved cytocompatibility properties for select osteoblast and osteoclast functions, we believe that these results demonstrate for the first time, that nanophase ceramics have great potential to become the next generation, choice orthopedic/dental biomaterial to enhance bonding to juxtaposed bone and, thus, increase implant efficacy.

Webster et al., Biomater. 20 1221-1227, 1999.

4:30 PM LL4.10
SYNTHESIS AND CHARACTERIZATION OF CORALLINE HYDROXYAPATITE MICROSPHERES FOR ORTHOSTEIC AND BONE FILLING MATERIALS. M. Sivasubramanian, K. Pradhanmou R, Biomaterials Laboratory, Central Leather Research Institute, Adyar, Chennai, INDIA.

Many efforts have been made towards the development of new bone substitute materials. Among these, hydroxyapatite/polymer composites have attracted much attention since such composites have some unique characteristics over their conventional constituent components. Poly(methyl methacrylate) (PMMA) is an adhesive in bone orthopaedic applications as well known. In the present paper, attempts were made to prepare functional microspheres of PMMA having epoxy and hydroxy functionality using glycidyl methacrylate and carboxytactic agent, mercaptopentaoil PMMA functional microspheres were prepared by using dispersion polymerization technique. These microspheres were subsequently grafted with coralline hydroxyapatite using coupling grafting technique via dicoumaroyl. These grafted ceramic-polymeric composites materials were characterized by various techniques such as IR, FT-IR, TGA, DSC and optical microscope. These microspheres and grafted materials were analyzed in term of particle size distribution and particle size analyzer and found to be 260 microns and 310 microns respectively. The optical micrographs clearly indicated that the coralline hydroxyapatite was grafted onto the functional microspheres. The percentage grafting of PMMA in the grafted materials was determined and it was 120%. Gentamicin was incorporated in these grafted materials and in its in-vitro release was carried out in phosphate buffer. It is aimed to use these grafted microspheres in orthopedics particularly in the repair and regeneration of bone and dental.

SESSION LL5 POSTER SESSION
ORTHOPAEDIC/DENTAL BIOMATERIALS
Tuesday Evening, November 30, 2000
8:00 PM
Exhibition Hall D (Hynes)

LL5.1
POLYURETHANE BIOMATERIALS WITH ANTINFECTION PROPERTIES. Erkeos O. Burybekev, Reem M. Iskand, Bahit A. Zhubanov, Institute for Chemical Sciences, Almaty, KAZAKHSTAN.

Bioceramic-related infection are frequently observed with prosthetic implants and these may result in post-surgical failure of the device. In present study the strategy for the prevention of foreign body infection by incorporation of antimicrobial agents in polyurethane biomaterials is described. A procedure has been developed for making antibiotic-loaded polyurethane films. Rifampicine, oxicillin and ciprofloxacin were used as antimicrobial agents. Antibiotics were incorporated in segmented polyurethanes based on various polyetherol, diolomycetes and bumetamed. The antibiotics release kinetics and biological efficacy have been investigated. It was found that the release rate depends on the concentration of incorporated drugs and show two significant periods consisting of burst effect" and typical Fickian diffusion-controlled release. The high antibacterial effect of antibiotic-loaded biomaterials against five different strains of bacteria was shown. The preliminary results in rabbits show the successful application of obtained polyurethane implants for the prevention of foreign body infection. The continuous release of effective doses of antibiotics over a long period in the interface where the biomaterial is contacted with the body can prevent the bacterial colonisation of the implants.

LL5.2
NOVEL POLYMERIC/CERAMIC NANOCOMPOSITES FOR ORTHOPAEDIC/DENTAL APPLICATIONS: MECHANICAL AND CYTOMCOMPATIBILITY PROPERTIES. Anastasios M. Myrillas, Richard W. Siegel*, Robert H. Doremus, Rena Bizios, Remsen Polytechnic Institute, Departments of Biomedical Engineering and *MSOE, Troy, NY.

To date, homogeneous, large (over 200 nm)-grain size, traditional materials that either satisfy mechanical (e.g., metals) or cytocompatibility (e.g., ceramics) requirements have been used almost exclusively for orthopedic/dental applications. The mechanical properties of these materials, however, these materials have not produced ideal bone implants. In an attempt to more closely match the microstructure and mechanical properties of bone (a composite 30% organic/protein, 65% inorganic/organic nanocomposite material), novel polymer/ceramic nanocomposite formulations were designed, fabricated, and characterized (i.e., mechanical and cytocompatibility properties). Formulations of polylactic acid (PLA) composites with varying (30, 40, 50, and 60) weight percent of nanophase alumina, hydroyxapatite, and titania loadings were prepared. The mechanical properties, including bending modulus and fracture toughness, of all materials tested in the present study were determined. The nanocomposites exhibited mechanical properties that were very different than those of the constituent component materials. For example, the polylactic acid/nanophase alumina composites exhibited bending modulus one and three orders of magnitude lower than those of the homogeneous polymer and ceramic constituent components, respectively; most importantly, the bending modulus of these novel composites were in the range of values (1-20 GPa) of human cortical bone. The cytocompatibility of the nanocomposites was determined by comparing osteoblast and fibroblast adhesion and proliferation; formation of mineral deposits in the extracellular matrix by osteoblasts was also examined. Osteoblast adhesion on the alumina nanocomposite formulations increased as a function of nanophase alumina weight percent; the results on the 50% nanophase alumina/PLA composites surfaces were similar to those obtained on the homogenous alumina surfaces, on which osteoblast adhesion was a maximum. The results of the present study provide evidence that these novel polymer/ceramic nanocomposite formulations closely match the
mechanical properties of native bone, an advantage over traditional orthopedic implant materials; most importantly, because of their enhanced biocompatibility, these nanocomposites could promote osteointegration, which is a crucial requirement for the clinical success of orthopedic implants.

**LL5.3 PHYSICAL AND MECHANICAL PROPERTIES OF A PHOTOCLINKED HYALURONAN FOR BIOMEDICAL APPLICATIONS** Kimberly A. Sneed and Mark W. Grinstaff, Duke University, Department of Chemistry, Durham, NC

Novel biomaterials that can be polymerized in situ via an optical trigger are of current interest to a variety of disciplines including dentistry, angioplasty, and orthopedics. Hydrogel type biomaterials are one class of biomaterials currently under investigation for these applications. We have synthesized a novel hydrogel based on the natural polymer hyaluronan (HA); HA, an alternating co-polymer of \( \beta \)-D-guluronic and \( \beta \)-D-glucuronic acids, is known to be biocompatible, non-antigenic and generally non-toxic reactive. By modifying HA with methacrylate moieties (HA-MA), the polymer can be photo-crosslinked in the presence of a radical initiating system of cation V and triethylmethane to form a solid, transparent, soft, flexible hydrogel. This rapid photo-induced hydrogel formation occurs on glass and Teflon surfaces as well as dry and moist tissues. The surface properties of this novel hydrogel were characterized by SEM and AFM and revealed a uniform and smooth surface. Studying wells showed that the HA-MA hydrogel can absorb over ten times its weight in water. The mechanical properties of this photo-crosslinked polyacrylate were evaluated using compression and creep compliance experiments. The HA-MA hydrogel exhibits solid-like (elastic) behavior with a low phase angle and a \( G' \) value of 1.6 kPa. The physical and mechanical properties of this novel hyaluronan hydrogel s, together with its ability to be rapidly photo-crosslinked suggest that this polymer is useful for inert cell encapsulation and in situ ophthalmic wound repair.

**LL5.4 EFFECTS OF DEMINERALISED BONE MATRIX IMPLANTS FOR FRACTURE HEALING IN CANINES** B.V. Suresh Kumar, Department of Surgery and Radiology, College of Veterinary Science, Tirupati, INDIA

The aim of this study is to evaluate Demineralised bone matrix (DBM) with tricalcium phosphate (TCP) and hydroxyapatite (HA) implants for fracture healing in dogs. The same were compared to untreated and cancellous bone graft treated animals. Under general anaesthesia tranverse fractures segmental defects were created in all animals and stabilized with intramedullary pinning. The defects were filled with materials in three groups and left unfilled in control groups. Postoperatively various parameters like clinical, biochemical, radiological, and histological changes were recorded and compared. Early functional limb usage was noticed in DBM TCP implanted group. Biochemical parameters did not show any significant changes post operatively in the study. Radiological studies showed visible segmental defect even at the end of the 12th week in untreated animals. The union of graft to host bone was observed at 4th week in cancellous bone group whereas union was visible at 6th week in DBM implanted groups. Periosteal reaction was a prominent feature in untreated and cancellous bone grafted groups. Angiographic changes revealed slightly hypervascularity at the fracture site in DBM implanted groups which came to normal by 9 weeks. However these findings are not significant in other groups. Osteodestruktographs showed medullary cavity continuity and no leakage of contrast medium at 12 weeks in all groups. Histological studies revealed fibrovascular reaction in early stages of post graft group whereas osteoclastic spikes were noticed in treated groups. 12th week radiographs showed excellent formation of cancellous tissue and periosteum with complete harvesting system. Based on the above observations, it is concluded that DBM implants are proved to be the best substitutes compared to autogenous cancellous bone grafts.

**LL5.5 PHOTOCLINKED POLY(PROPYLENE FUMARATE) SCAFFOLDS FOR ORTHOPEDIC APPLICATIONS** John P. Fishier, Therese A. Holland, Antonios G. Mikos, Rice University, Department of Bioengineering, Houston, TX

A tissue engineering scaffold based upon the biodegradable polymer poly(propylene fumarate) [PPF] has been produced using a UV crosslinking/polymerization leaching strategy. PPF [Mn = 14000 to 27000] was crosslinked using 365 nm ultraviolet light and the photoinitiator bis[2,4,6-(trimethylbenzoyl) phenylphosphine oxide] [BAPO, 5 mg BAPO/\( g \) PPF] by forming covalent crosslinks across fumarate units of PPF chains. With the inclusion of a NaCl porogen (50, 70, 90 and 90 wt%) and its removal by water leaching after UV crosslinking, a porous PPF scaffold was formed. Porosity was characterized by weight loss, mercury porosimetry, and scanning electron microscopy. Results showed that porosity can be controlled by porogen content and that scaffolds of at least 70 wt% porogen possess an interconnected pore structure. Fourier transform infrared spectroscopy was used to evaluate the crosslinked PPF, showing that the exterior surface of the porous samples undergoes greater crosslinking than the interior of the sample. Scaffold compressive mechanical strength was found to increase with increasing PPF molecular weight and decrease with increasing porogen content. The strongest samples which possessed an interconnected pore structure (PPF Mn = 2000 and 70 wt% porogen) were found to have an elastic modulus of 4.8 ± 1.52 MPa and a compressive strength at 1% yield of 1.84 ± 0.79 MPa. This work has shown that the UV crosslinking/polymerization technique is a simple method to form porous solid state photoinitiated materials and therefore allows for the material's further evaluation for tissue engineering applications.

**LL5.6 SYNTHESIS AND CHARACTERIZATION OF HYDROXYapatITE PARTICULATES** Chen-Wei Chen, Kuhlsh Byrappa, Charles Oakes, Wojciech Suchanek, Richard Riman, Rutgers

Three-dimensional porous scaffolds have been utilized as conduits for guided tissue regeneration. Existing methods for the manufacturing of porous scaffolds require pre-fabrication before implantation and are not suitable for injectable systems. A tri-block copolymer of poly(propylene fumarate) and poly(ethylene glycol)-monomethylether combined with poly(ethylene glycol)-diacrylate was crosslinked utilizing a water soluble persulfate-initiated system. The composition of the injection system was varied to assess its effect on the sol fraction and swelling characteristics for thin hydrogel films. The sol fraction was measured to be less than 5% while the equilibrium water content was greater than 40%. Porous from the injection of these hydrogels were further fabricated by carbon dioxide formation from the dissolution of sodium bicarbonate in water in the presence of acetic acid; the sodium bicarbonate was added to the injection formulation. The effect of the sodium bicarbonate and acetic acid reaction on the porosity of the resulting hydrogels was also determined for porous scaffolds of porosity up to 90%. Porcine morphology was analyzed using scanning electron microscopy for dried hydrogels and cryosectioning combined with stereology for hydrated hydrogels. Interconnected porous scaffolds with a controllable porosity were synthesized which hold promise for dental applications.

**SESSION LL6: NOVEL ORTHOPAEDIC/DENTAL BIOMATERIALS**

**Wednesday Morning, November 29, 2000**

**Berkeley (Sheraton)**

8:30 AM **LL6.1 POSTER SESSION - CERAMIC CROWNS - A DURABILITY/FATIGUE PROBLEM** Van Thompson, University of Medicine and Dentistry of New Jersey, NJ

Clinical results with all-ceramics dental crowns on molar teeth indicate that high stress states result in significant failure rates for monolithic materials. Recent investigations have shown that initial strength of a ceramic material has little to do with its clinical performance. Instead, tolerance to damage, introduced a number of different ways, is a more important characteristic. Damage from repeated Hertzian contact (blunt, cyclic contact similar to occlusal contact) accumulates, leading to catastrophic failure. Both near field (cone cracking) and far field (radial cracking) can occur. Routine sandblasting can create damage equivalent to a million loading cycles for some ceramics. Esthetic monolithic materials are susceptible to significant reduction in strength/failure after damage equivalent to 4-5 years of clinical use. Layered ceramic structures, soft esthetic porcelains on stiff and strong substrates, offer an alternative with improved damage tolerance. Unfortunately, recent investigations suggest that fatigue life remains a significant problem as these ceramic systems must perform in this critical section (1.2-2 mm) on a low elastic modulus dentin substrate (15-17 GPa) while cemented with an even lower modulus resin layer (2.8 GPa) of significant thickness (215-50 microns). The wet oral environment with pH transients and millions of complex load cycles presents a challenge for designing future esthetic crown systems. This presentation will compare Hertzian contact laboratory fatigue studies of monolithic and layered ceramics results with long-term clinical performance data on molar crowns.

9:00 AM **LL6.2 DESIGN, SYNTHESIS, AND CHARACTERIZATION OF HYDROXYapatITE PARTICULATES** Chen-Wei Chen, Kuhlsh Byrappa, Charles Oakes, Wojciech Suchanek, Richard Riman, Rutgers
Biological hydroxyapatite \( [\text{HAP}, \text{Ca}_{10}(\text{PO}_4)_{6}(\text{OH})_2] \) is a natural mineral that forms a wide range of calcium and inorganic ion substitutions. Use of synthetic HAP-based materials depend upon the ability to design HAP particulates with well-characterized chemical and physical characteristics. A rational approach to this problem is to control the chemical equilibria in a function of processing variables, generate equilibrium diagrams to map processing variable space for the phases of interest, design hydrothermal experiments to test and validate the predicted diagrams, and use processing variable space to explore opportunities for control of reaction and crystallization kinetics. This paper will detail our progress in making such design HAP particulates. Predictive phase diagrams were constructed with well-characterized HAP calcia for temperatures of 250°C to 350°C. Yield diagrams were calculated using an accurate model of the HAP. Thermodynamic calculations were conducted using temperature-dependent functions for the relevant solution phase and solid-solid equilibria and solute species activity coefficients. Model accuracy was then evaluated through experiments at 50, 100, and 200°C and a pH range between 2.2 and 8.9. The thermodynamic calculations and experimental results are in good agreement. Stoichiometric, crystalline HAP has also been prepared by heterogeneous reaction of \( \text{Ca(OH)}_2 \) powder and aqueous \( (\text{NH}_4)_2\text{PO}_4 \) at room temperature using mechanochemical-hydrothermal methods. Using this approach we have accomplished complete and uniform substitution of hydroxyl groups in HAP with a high level of reproducibility. HAP products prepared via the above methodologies have been characterized for physical properties and structure. Thermal behavior and chemical analysis. BET, SEM, TEM, and X-ray diffraction and X-ray spectroscopy were used to confirm the phase and microstructural features. HAP surface area varies from 45 to 150 m²/g depending upon synthesis conditions with average particle sizes varying from 240 nm to 400 nm.

9:15 AM LL6.3 EFFECTS OF DOPING SUBSTITUTIONS IN HYDROXYAPATITITE ON OSTEOSTRAL ADHESION: Thomas J. Webster, Caleb A. Ennis, Matthew A. Draper, and Biomedical Engineering and Materials Science and Engineering, Rensselaer Polytechnic Institute, Troy, NY.

The present in vitro study investigated the effect of dopants, specifically, Magnesium, Calcium, Zinc, and Yttrium, present in hydroxyapatite (HA) on osteoblast attachment. For this purpose, HA substrates were prepared by adding calcium nitrate (used either undoped or containing 2 mol% of either Magnesium, Calcium, Zinc, or Yttrium) to ammonium phosphate-doped water solutions. The resultant substrates were sterilized for 24 h, centrifuged, filtered, dried, and sintered at 100°C for 60 minutes. Neutral red calcein osteoblasts were seeded at 2,500 cells/cm² in Dulbecco’s Modified Eagle Medium (DMEM, Gibco) supplemented with 10% fetal bovine serum (FBS) and were allowed to adhere under standard cell culture conditions (i.e., in a 37°C, humidified 5% CO₂/95% air environment). For the cell survival study, cells were washed, stained, and counted in situ using fluorescence microscopy. Proteins adsorbed onto the substrates of interest to the present study were quantified following immersion of each sample into 0.5 mL of 5 µg/mL of bovine serum albumin supplemented with 10% fetal bovine serum (containing undetermined amounts of proteins such as albumin, laminin, denatured collagen, fibronectin, vitronectin, etc.) or in a solution of 5 µg/µL of bovine serum albumin, laminin, denatured collagen, fibronectin and vitronectin (all chemicals from Sigma) per mL of phosphate-buffered saline under standard cell culture conditions for 4 hours. Adherent proteins were then desorbed; protein concentration in each eluent was determined using a commercially available kit (BCA Assay, Pierce). HA-attachment was significantly \( (p < 0.01) \) greater on HA doped with Yttrium than on any other HA formulation tested in the present study. Moreover, compared to HA doped with either Zinc or Calcium, significantly \( (p < 0.01) \) greater adhesion of fetal bovine serum, denatured collagen, fibronectin, and vitronectin was observed on HA doped with Yttrium. For the first time, these results demonstrated that modification of HA with Yttrium enhances adsorption of select proteins (specifically, denatured collagen and vitronectin) that, subsequently, promote osteoblasts attachment.

10:00 AM LL6.4 A BIOACTIVE GLASS FIBER REINFORCED COMPOSITE: Anthony B. Brennan, Univ of Florida, M$S$E and Biomedical Eng, Gainesville, FL; Rodrigo Ortejo, Fed Univ of Minas Gerais, Dept of Met and Mater Eng, Belo Horizonte-MG, Brazil; A.E. Clark, Univ of Florida, Florida, Gainesville, FL; Larry L. Hench, Imperial College, London, UNITED KINGDOM.

Bioceramic fibers were produced using a sol-gel method. The rheological properties of different sol compositions prepared from a mixture of ThO₂, H₂PO₄, and calcium nitrate, in a water-ethanol solution are reported. The sols were extruded through a spinneret to produce continuous 10-micron fibers. Discontinuous fibers and fibers mats were prepared by air-spraying the sol-gel into hydroxyapatite particles. In bioceramic fibers by three different thermal treatments at either 600, 700, or 900°C for three hours. SEM, BET, EDX and FTIR were used to characterize the morphology and structure of the fibers. The BET measured specific surface area of each fibers was 25 m²/g compared to a value of 200 m²/g for a typical sol-gel derived particle of similar composition. Both the continuous and discontinuous fibers exhibited in vitro bioactivity in a similar body fluid. For the first time, the bioactivity of the glass is demonstrated to be dependent upon composition. A hybrid composite prepared with novel surface active polymer is described. The preliminary viscoelastic properties and in vitro bioactivity of the composite from continuous fibers containing the discontinuous bioactive glass fibers.

10:15 AM LL6.5 SYNTHETIC BIODEGRADABLE POLYMER NETWORKS MODULATING MARROW STROMAL OSTEOBLAST FUNCTION: Heungsoo Shin, Antonios G. Mikos, Dept of Bioengineering, Houston, TX.

We are interested in the synthesis of new orthopedic biomaterials for the treatment of bone defects by guided tissue regeneration. We have previously shown that composite scaffolds based on poly (propylene fumarate) (PPF) exhibit desirable mechanical properties for trabecular bone replacement. We have further demonstrated that PPF supports marrow stromal osteoblast adhesion and proliferation. In this study, we have synthesized new unstarnted polymers based on PPF with covalently bonded peptide sequences, which act as a functional group on the polymer. The polymer is a linear oligomer of poly(ethylene glycol) and fumaric acid (OPF) with the peptide covalently attached to the PEG end (hydroxyl) group. We have investigated the effects of the peptide content in the polymer network for a model GAG peptide as well as the PEG spacer length on the function of attached marrow stromal osteoblasts. Contact angle analysis confirmed that the hydrophilic domain of OPF and immobilized peptide were homogeneously distributed throughout the polymer network, maintaining the hydrophilic PPF. The functionalized polymer network may provide a new means of modulating cellular function and therefore would be useful as a scaffold for bone tissue engineering.

10:30 AM LL6.6 CRYSTAL MORPHOLOGY CONTROL BY MELT PHASE SEPARATION IN BIODEGRADABLE POLYMER BLENDS: Yvonne A. Akgun, Rensselaer Polytechnic Institute, Department of Chemistry, Troy, NY; J. Caroline Meredith, Georgia Institute of Technology, School of Chemical Engineering, Atlanta, GA; Eric J. Ams, Polymers Division, National Institute of Standards and Technology, Gaithersburg, MD.

The effect of lower critical solution temperature (LCST) phase separation on the crystal growth mechanism and fine structure of PLLA (poly(lactic acid))/PGA (poly(glycolic acid)) blends is studied by simultaneous small-angle x-ray scattering (SAXS) and wide-angle x-ray scattering (WAXS). We have followed the structural changes (2 nm to 100 nm) during crystallization at 45°C of critical- and off-critical blends from two different phase and two-phase melts. Phase separation is induced by controlled temperature jumps into the LCST (two-phase) region, which is above the melting temperature (60°C) of PCL. The large-scale morphology (1-100 µm) resulting from phase separation is observed with optical microscopy. The SAXS profiles arise from two scattering species: (1) amorphous domains outside the lamellar stacks that give rise to diffuse scattering and (2) lamellae in lamellar stacks that give rise to discrete scattering. Wide-angle and small-angle x-ray scattering data are used to study the two scattering contributions by fitting the observed scattering to a model that accounts for a significant amorphous phase between lamellar stacks. Some morphological parameters are independent of blending and melt phase separation, the fraction of crystals within lamellar stacks (60%) and the size of the amorphous regions surrounding stacks (29 nm) are the same for pure PCL and for blends, and are independent of blend composition and the extent of melt phase separation. The long period (17 nm) obtained for PCL is also independent of blend composition and melt phase separation, indicating that PDLA is excluded from the lamellar stack. Other morphological parameters are determined by blend composition and the extent of melt phase separation. For example, the width of the stacking (48 nm) obtained for PCL is reduced to 45 nm in the blends. In the off-critical blends, the fraction of crystals that develop outside stacks increases from 0% to 6% as melt phase separation increases. In the critical blend, however, crystals are restricted to lamellar stacks. Our results show that the melt polymer can determine the extent
to which new crystals are formed in the amorphous regions
surrounding lamellar stacks, the ultimate crystallinity obtained, and the
primary crystallization rate. The effective use of melt phase
separation for controlling the crystal morphology is discussed.

10:45 AM LI6.7
NON-WOVEN SHEETS OF BIOACTIVE FIBERS PRODUCED BY
A SOL-GEI PROCESS Rosana Domingues, Universidade Federal de
Minas Gerais, Dept of Chemistry, BUIAZ; Arthur Clark, University
of Florida, College of Dentistry, FL; Anthony Brennan, Dept of
MSE; University of Florida, FL.

A high velocity spray process was used to prepare SiO₂·CaO·P₂O₅ fibers
from sol-gel method. Both, dispersed fibers and non-woven
sheets of bioactive fibers were produced. Temperature and pH of the
solutions were evaluated during all synthesis and used to evaluate the
progress of sol-gel reaction. Two compositions having different silicon
contents were produced. Sol viscosities were studied and the
optimized range for produced non-woven sheets was determined.

The formation of hydroxy carbonate apatite, HCA, on the surface of fibers
was used to evaluate the kinetics of the biocactivity in a simulated
body fluid. Diffuse Reflectance Infrared Fourier Transform
Spectroscopy analysis confirmed the presence of HCA formed after 3
hours immersion in the test fluid for both compositions tested.

Samples higher in silicon exhibited faster kinetics. Scanning Electron
Microscopy revealed a homogeneous layer of HCA on surface of fibers.
Fiber morphology was maintained almost unchanged after 30 days of
immersion. Potential applications are as scaffolds for both mineralized
and non-mineralized structural tissues.

11:00 AM LI6.8
SYNTHESIS OF NOVEL POLYSACCHARIDE MCMS. Matthew T.
Sheehy, Elizabeth B. Walsh and Mark W. Grinstaff, Duke Univ.
Dept of Chemistry, Durham, NC.

Polysaccharides are of widespread use in the biotechnology, food and
pharmaceutical industries. We have synthesized two novel analogs of
alginate acid, poly(5,6-dihydroxyxycarbonyl carboxylic acid) and
poly(5,6-dihydroxyxycarbonylcarboxylic acid). Ring opening
metathesis polymerization (ROMP) of a cyclic olefin in the presence of
Grubbs catalyst yielded these new polymers. Further modifications
then afforded hydrophobic polymers possessing a repeating cyclic ring
structure, two secondary alcohols, and a carboxylic acid. These
polymers were characterized by IH NMR, IR, and GPC. The synthetic
procedure described involves altering the functional groups of the
polymer such that its chemical, physical, and mechanical properties
can be optimized for a specific tissue engineering application.

11:15 AM LI6.9
SURFACE TREATMENT OF BIOMEDICAL POLYMERS FOR
ENHANCED ADHESION Valerie Barron, Dept of Physics, Trinity
College Dublin, IRELAND; Martin Buggy, Dept of Materials Science
and Technology, University of Limerick, IRELAND.

As part of a project to develop a bioimimetic artificial hip joint, it was
necessary to bond carbon fibre reinforced polyetheretherketone to
thermoset polyurethane. In order to produce a strong and durable
adhesive joint, surface treatment was necessary prior to bonding.
Surface treatments may involve one or more of the following: removal
of a weak boundary layer, increase in surface roughness, change in
cellular nature of the surface, or modification of the physical
structure of the surface. Methods of surface treatment for
thermoset polymers involve altering surface tension and surface
chemistry using techniques such as plasma treatment, corona
discharge, oxidising flame treatments and laser treatments. Prior to
bonding, techniques such as surface tension and wettability were
examined to optimise the method of surface treatment. Surface
chemistry was examined using Fourier transform infrared spectroscopy
(FTIR) and x-ray photoelectron spectroscopy (XPS). In addition
surface topography was examined using a number of techniques
including atomic force microscopy (AFM), scanning electron
microscopy (SEM). Mechanical testing by peel testing according to
British standards was carried out to establish the durability of these
adhesive joints in a biomedical environment. The human body is one
of the most aggressive environments that an adhesive bond has to
endure. The durability of these APC2/polyurethane joints is
investigated in various aqueous environments including Ringers
solution and distilled water. While the bond was stable in air for over
1 year, storage in aqueous media leads to rapid deterioration of bond
strength. This phenomenon was observed both in distilled water and
in Ringers solution. Measured diffusion rates for both environments
were very similar and it is suggested that the observed faster rate of
loss of bond strength in Ringers solution is due to the interaction of
buffered components from the Ringers or stabilisation of low
molecular weight material by the Ringers.

11:30 AM LI6.10
THE SYNTHESIS OF HYDROXYAPATITE / POLYSACCHARIDE
NANOCOMPOSITES THROUGH A SELF-ORGANIZATION
MECHANISM. Toshiyuki Ikoma1,2 and Junzo Tanaka1,2.
1National Institute for Research in Inorganic Materials, Ibaraki, JAPAN.
2CREST, Japan Science and Technology, Saitama, JAPAN.

The purpose of this study is to prepare novel inorganic/organic
composites using a self-organization process. Both
polymerocomposites, hydroxyapatite (HAP)/hyaluronic acid (HyA) and
HAP/chondroitin sulfite (ChS) composites, were prepared by a
coprecipitation method using a calcium hydroxide suspension
involving polysaccharides and a dithiol phosphate solution. HyA and
ChS are the ubiquitous glycosaminoglycan found in almost all tissues,
which are the linear polysaccharides consisting of glucuronic acid and
N-acetylgalactosamine (HyA) or N-acetylglucosamine (ChS). Therefore,
it is expected that HyA and ChS are bioactive and can interact with HAP
crystal surfaces via carboxy groups and/or sulfate groups. According to TEM observations, the HAP/HyA composites
consisted of island-like assembled particles of 300nm in length and
30nm in width, while the HAP/ChS composites those of 150nm in
length and 50nm in width. When electron diffraction was separately
took for respective particles, 002 and 004 diffraction spots ascribed to
HAP were observed along the longitudinal axes of the particles;
therefore, the c-axes of HAP nanocrystals (30nm in size) were
regularly aligned along polysaccharide chains in the respective
assembled particles. This result means that the HAP nanocrystals can
be selforganized through the nucleation and growth of HAP crystals
on the carboxyl groups in ChS/HyA. The self-organization mechanism
was elucidated by a molecular orbital method using a cluster of HAP
surface and carboxyl group, in which calcium ions were assumed to be
coordinated by seven oxygen ions on the HAP (110) surface. As a
result, the configuration of a CO₂ plane of the carboxyl group was
attainable when it was perpendicular to the c-axis of HAP, and the
chemical bond formed between Ca and CO₂ was considerably
covalent with the bond order of 0.15. It was considered that such
coordination bond play an essential role for the self-organization
mechanism.