Distribution of salts and proteins, and structural changes in spider glue revealed by confocal Raman microscopy

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Modern orb weaving spiders use micron-sized glue droplets on their viscid silk to retain prey in webs. A combination of low molecular weight salts and proteins makes the glue viscoelastic and humidity responsive in a way not easily achieved by synthetic adhesives. We recently reported that different spider species tune glue viscosity to maximize adhesion at their foraging humidity\textsuperscript{1}. In this study we used optical and confocal Raman microscopy to determine the spatial chemical structure of glue droplet. Optically, the glue droplet shows a heterogeneous structure, but we found that salts and proteins are present ubiquitously throughout the droplet\textsuperscript{2}. The distribution of adhesive proteins in the peripheral region explains the superior prey capture performance of orb webs as it enables the entire surface area of the glue droplet to act as a site for prey capture. Recent results on the primary structural changes in glue with an increase humidity and upon stretching will be discussed. Understanding the function of individual glue components and the role of the droplet’s macro-structure can help in designing better synthetic adhesives for humid environments.
References


Thiol-Mediated Miniemulsion Polymerizations: A New Route to Antimicrobial Nanoparticles

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Engineered polymer nanoparticles – with sizes ranging from 20-500 nm – are playing an increasingly important role in the advancement of emerging technologies for industrial, agricultural, pharmaceutical, and biological sectors. The prospects of advancing these and other technologies have provided great impetus for the development of rapid, low-cost methodologies for the synthesis of functional polymer nanoparticles. Emulsion-based processes – such as miniemulsion polymerizations – provide well-studied synthetic routes to polymer nanomaterials. The first part of this presentation will highlight our recent efforts employing thiol-mediated photopolymerizations in miniemulsion as a simple, rapid, and one-pot synthetic approach to polythioether nanoparticles with tunable particle size, clickable functionality, and composite compositions. The latter part of this presentation will focus on our recent results employing this versatile synthetic platform for the encapsulation of plant-based essential oil (EO) extracts (i.e. monoterpenoid phenols such as carvacrol/thymol) within polythioether nanoparticles as model, sustained-release antimicrobial nanomaterials. The thymol/carvacrol-loaded nanoparticles show effective antimicrobial activity (>99.9% kill efficiency at 24h) against gram-positive (*B. subtilis* and *S. aureus*) and gram-negative (*E. coli* and *B. cenocepacia*) bacteria. The simplicity, modularity, and efficacy of this essential oil encapsulation platform may combat bacteria with intrinsic resistance to conventional antibiotics, and is potentially adaptable for delivery of EOs as active packaging materials and topical antiseptics. The antimicrobial activity of against inherently resistant *B. cenocepacia* (an opportunistic pathogen with significant clinical importance in persons with cystic fibrosis) may provide a route to innovative pulmonary therapeutics by appropriately engineering the nanoparticle properties.
Title: Phosphorylated Poly(ester-urea) based Biomimetic Degradable Bone Adhesives

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Abstract:

Bone and tissue adhesives are an integral part of surgeries not only for wound healing but also for hemostasis and drug delivery \[1\]. Currently a number of tissue adhesives are commercially available in market. However, they lack mechanical strength, degrade into carcinogenic, toxic byproducts or are difficult and expensive to apply. There is still an urgent need to develop alternative tissue adhesives that are degradable, form bioresorbable byproducts with high adhesion strengths. Aquatic animals like mussels, barnacles, sandcastle worms and caddisflies are known to exhibit strong underwater adhesion virtually on any surface. Numerous studies have shown amino acids or their modified forms like DOPA (dihydroxyphenylalanine) and phosphorylated serines as major components imparting adhesion to natural glues.\[2\] Taking inspiration from these natural adhesives, we aim at developing their synthetic mimics from poly(ester urea)s (PEU) which are semicrystalline polymers with tunable degradation rates. PEUs are synthesized by solution polymerization of amino acid based esters and are demonstrated to degrade into bioresorbable byproducts. Recently, we have studied adhesion properties of catechol functionalized PEUs, a mimic of mussel adhesive with adhesion strengths comparable to that of commercial fibrin glue on porcine skin.\[3\] To expand more in the area of tissue adhesives, we have now synthesized phosphoserine based PEUs as synthetic mimics of caddisfly silk. Phosphoserine-Valine copolymer with 2% and 5% phosphate functionality are synthesized and characterized by \(^1\)H, \(^13\)C and \(^31\)P NMR, ATR-IR, DSC, TGA, SEC and contact angle. The adhesion properties are studied by lap shear adhesion tests both before and after crosslinking with Ca\(^{2+}\) ions and compared with commercially available PMMA bone cement on both aluminum substrates and bovine bone.

References:

Qinyuan Chai, Yongshun Huang and Neil Ayres

Covalently crosslinked polyurethane/urea networks were successfully synthesized by reacting 2,4-toluene diisocyanate with a glucose functionalized diamine, poly(ethylene glycol) (PEG) and 1,1,1-tris(hydroxymethyl)ethane (triol) comonomers through step-growth polymerization. The polymer showed repeatable shape memory behavior with a tunable switching temperature by changing the mole ratio between glucose-diamine and PEG. The surface of a film of the polymer can be sulfated, therefore mimicking heparin, achieving increased biocompatibility without sacrificing the shape memory properties. This prepared material can be processed into highly compressive interconnected foams by solvent casting/particulate leaching technology for potential aneurysm treatment. This strategy has been extended to use isophorone diisocyanate (IPDI) and a lactose functionalized diamine. Due to the relatively low reactivity of IPDI compared with TDI, dibutyltin dilaurate (DBTDL) was needed to catalyze the crosslinking reaction after preparing a NCO-premix first, which was obtained by mixing the lactose diamine with excess IPDI under room temperature. The pore structure/porosity of the foams can be controlled by the salt size, salt fusion level and macromonomer concentration. The mechanical properties of the foam depend on the pore morphology. A decrease in the compressive modulus is seen with increased porosity. The prepared polymer foam can be compressed into a small
volume and expand back to ~70% of its original size.
Crosslinked cationic polyester films for prevention of *P. Aeruginosa* colonization and biofilm formation

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Hospital related infections are a major concern in the healthcare industry with 1.7 million affected patients worldwide at any time. At surgical sites one of the main causes of implant failure is the formation of biofilms which result from the attachment of bacterial colonies to surfaces. One of the current practices to inhibit bacteria colonization on the surfaces is using antimicrobial coatings. Controlling physico-chemical properties of the biomedical surface by coating with quaternary ammonium, pyridinium groups or encapsulation with silver, are current routes to inhibit bacteria colonization and biofilm formation. However the lack of stability of such coatings and in some cases their toxicity necessitates the development of more stable and non-toxic coatings.

The current work details our efforts to further develop stable initiator free-crosslinkable cationic polyester coatings designed from coumarin and N-substituted diols as monomers to provide stable antimicrobial coatings. This initiator free-crosslinkable cationic polyester coating were tested against *Pseudomonas aeruginosa* (PAO1) in static mode. Results from biofilm assay showed that the cationic surface not only inhibits the biofilm formation, but it also resulted in killing of the attached bacteria. Additionally, these polyester coatings exhibit high thermal stability, low cytotoxicity and minimal hemolysis activity. Comparison with polyesters having neutral or anionic functional groups demonstrated the need for role of cationic charges in antimicrobial activity.
Additive Manufacturing of Tissue Scaffolds Using Novel Biodegradable Photopolymers

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Additive Manufacturing, or 3D Printing, is becoming a popular tool for fabricating tissue scaffolds and other biological constructs for use in Tissue Engineering and Regenerative Medicine. In this work, we present the use of Microstereolithography (μSLA) to fabricate tissue scaffolds from novel biodegradable photopolymers. By selectively patterning UV light, μSLA can achieve extremely high resolution and create scaffolds with feature sizes of just a few tens of microns. We have developed a custom μSLA machine capable of printing a wide variety of photopolymers that can achieve feature sizes below 50 μm.

One of the primary barriers to the greater use of μSLA for the fabrication of tissue scaffolds is the lack of materials with low cytotoxicity, good cell adhesion, and degradability. A novel biodegradable polyester, poly(tri(ethylene glycol) adipate) dimethacrylate (PTEGA-DMA), was synthesized and evaluated for its printability. The curing parameters for printing were developed and three-dimensional parts were made. Optical and electron microscopy were used to determine the achievable feature sizes and accuracy of the polymer in the μSLA system.

Cell studies were conducted to evaluate adhesion and viability on PTEGA-DMA. MC3T3-E1 mouse preosteoblasts were seeded on cast films of the polymer and observed after 1, 4, and 7 days. Viability was determined using an MTS assay.
Tuning Insulin-sensitizing Activity of Polyoxovanadate Derivates by Kinetically Control Their Self-assemblies

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Abstract:
Insulin resistance plays a crucial role in the pathophysiology of type 2 diabetes mellitus and vascular disease. Vanadium is an essential element in most living beings and is accumulated in several oxygen-dependent organisms to participate in biological processes, owing to the similarity in size and charge of phosphorus(V) and vanadium(V). Therefore, vanadate has been demonstrated an effective inhibitor toward the activity of many enzymes, especially those related to phosphate reactions such as Protein tyrosine phosphatase 1B (PTP1B). The insulin-sensitizing activity of a series of polyoxovanadate derivates (POVs) covalently modified with hydrophobic ligands is investigated. PTP1B inhibition test revealed that the high inhibition rates benefit from hydrophobicity of the ligands. However, the most two effective POVs in inhibiting PTP1B showed reduced insulin receptor phosphorylation resistance and glucose uptake promoting ability. Solution behavior study provided essential information for POVs cellular uptake which dominanted their insulin-sensitizing activities. Self-assembly of POVs into different architectures was kinetically controlled. Two morphologies, spherical micelles and needle-shape crystals were determined by laser light scattering and observed by transmission electron microscopy in aqueous solution. Spherical micelles were stable and demonstrated much higher internalization by adipocytes, which may provide insights into new ways to treat insulin resistance.
TUNABLE POLY(ESTER UREA)S FOR TENDON-BONE REPAIR IN ROTATOR CUFF APPLICATIONS

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Injuries that result in damage to the bone-tendon interface represent a significant problem in orthopedic surgery. While there are several options at the disposal of surgeons, most of the current treatment strategies are fraught with complications. Despite the extensive investigation of synthetic materials, autograft, and allograft materials, there is no ideal, translationally-relevant, regenerative solution to enhance healing at the tendon-bone interface. Particularly lacking are degradable polymeric materials that have tunable growth factor release rates. Our lab has developed fully resorbable, amino acid-based poly(ester urea)s (PEU) that meets all of these requirements. Preliminary studies have shown that these PEUs have controllable degradation rates\(^3\), and tunable functionalities\(^4\). The Becker Lab has also shown that in vivo, PEUs have non-toxic hydrolysis byproducts which do not induce unfavorable inflammatory responses\(^4\). In this work PEUs will be utilized to develop 3D-materials for adsorption of autologous growth factors or use at the interface of tendon and bone to enhance the healing process. If successful, we feel that these materials will provide multiple new avenues for investigating solutions to unmet medical needs in orthopaedic and regenerative medicine applications beyond what we are proposing to investigate.

References

Fabrication of chitosan/gelatin/keratin composite as a buccal mucoadhesive patch to treat Desquamative gingivitis

Zahra Davoudi, Mohammad Rabiee, Behzad Houshmand, Niloofar Eslahi

Abstract

Mucoadhesive films bring up the advantage of more efficiency and available drug in injured region. In this work mucoadhesive films for delivery of Hydrocortisone sodium succinate were prepared using different ratios of chitosan, gelatin and keratin. In the first step, chitosan and gelatin proportions were optimized after evaluating the mechanical properties, swelling, water uptake, stability and biodegradation of films and a 3:1 w/w chitosan/gelatin ratio was found as the optimized proportion. Then keratin was added at different percentages to the optimum composite of chitosan and gelatin together with the drug. The results of surface pH showed that none of the samples were harmful to the buccal cavity. FT-IR analysis confirmed the influence of keratin-on the structure of composite. The presence of higher amount of keratin (1.5% W/V) in the composite films resulted in high mechanical, mucoadhesive properties and stability, low water uptake and biodegradation in phosphate buffer (ph=7.4) containing $10^4$ U/ml Lysozyme. Release profile of films illustrated that Keratin is a rate controller in release of the drug.

Keywords: Mucoadhesion, Chitosan, Gelatin, Keratin, Hydrocortisone sodium succinate, Composite, Drug delivery
Synthesis of Functionalized Self-immolative Polymers with Biological Activity
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Conventional polymer chemistry has been dedicated to assemble small molecules together to construct long chain macromolecules. In contrast, “self-immolative” polymers just do the opposite. They are long chain macromolecules, which are capable of disintegrating into their small components when exposed to a specific stimulus. The degradation of traditional bioresorbable polymers is initiated by nonspecific triggering events and cleaved at random sites throughout the polymer backbone, thus leading to an uncontrolled degradation profile. On the contrary, self-immolative polymers are robust under normal environmental conditions, yet undergo continuous and complete depolymerization within minutes in response to the removal of stimuli-responsive end-caps. A variety of self-immolative polymer backbones and architectures have been developed for applications such as, tissue engineering, drug delivery, responsive coatings, microfluidics and shape-memory applications. Here, we demonstrate a novel concept using a new type of thermosetting plastics which can possess dynamic 3D printing and subsequent erasing of polymeric objects. A strategy to form rigid 3D thermosetting materials is the photo-chemically induced cross-linking of stimuli-responsive polymers with spatial control. Whereas conventional 3D-printed objects cannot be further processed, erasing of specific parts of a self-immolative 3D printed object is acheived by triggered depolymerization of main chains. This chemical technology is currently being explored as a platform for advanced functional biomaterials including new tissue engineering platforms and antimicrobial or antifouling surfaces.
Myocardial infarction (MI) affects millions of people in the western countries. Cardiac fibrosis naturally occurs after MI and progresses with time. The increase of cardiac fibrosis leads to gradual decrease of cardiac function. Myofibroblasts, differentiated from cardiac fibroblasts mainly through TGFβ signaling pathway, are responsible for cardiac fibrosis. Therefore, to inhibit cardiac fibrosis, it is essential to prevent TGFβ pathway-induced myofibroblast formation. However, ideal therapeutic approaches to achieve this goal remain to be established. We hypothesized that controlled release of anti-fibrotic agent basic fibroblast growth factor (bFGF) will efficiently inhibit myofibroblast formation. bFGF has been shown to prevent myofibroblast activation in various tissues. It counteracts the profibrotic activity of TGFβ. In the present work we developed an injectable and thermosensitive hydrogel, and tested if it could deliver bFGF to inhibit cardiac fibroblasts from differentiating into myofibroblasts. The hydrogel was based on N-isopropylacrylamide, acrylate-polylactide, and 2-hydroxyethyl methacrylate, and synthesized through free radical polymerization. Hydrogel sol-gel transition temperature was around 26°C measured using DSC. 20% hydrogel solution (w/v) could be injected through a 26-gauge needle at 4°C. bFGF was encapsulated in the 4% hydrogel solution. Release study was conducted at 37°C using PBS as release medium for 28 days. The released bFGF medium at each time point remained bioactive that could promote the cell proliferation. The effect of released bFGF on cardiac fibroblast differentiation into myofibroblast was evaluated in two and three dimensional model (2D and 3D) in the presence of TGFβ. In 2D, cardiac fibroblasts (RCF) was cultured on bFGF encapsulated hydrogel; in 3D, bFGF was injected into 3D collagen gel seeded with rat cardiac fibroblasts RCF. At both protein and gene level, this bFGF release system significantly attenuated myofibroblast differentiation characterized by αSMA and CTGF. The results demonstrated that this system has potential to control cardiac fibrosis after MI.
**H₂S-Releasing Nanoparticles with Morphologically-Dependent Release Kinetics Exhibit Anti-Cancer Activity**

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Gasotransmitters such as hydrogen sulfide (H₂S) are biologically produced signaling gases with physiologically relevant activities. For example, H₂S suppresses oxidative stress, modulates inflammation, and protects endothelial tissue via vasodilation. Of particular interest is the mounting evidence that H₂S can exert both pro- and anti- apoptotic activity on cultured cells. For example, Lee et. al. reported a time- and concentration-dependent activity of H₂S on human cancer cells, with low concentrations of H₂S applied over a period of days resulting in inhibited cancer cell proliferation as well as apoptosis.³ To achieve a desired outcome *in vivo*, H₂S must be delivered locally and at the correct concentration. Therefore, the vehicle of delivery must be rationally designed to release H₂S at the desired rate. A number of H₂S-releasing compounds have been reported including S-aryltiooximes, which release H₂S controllably (sensitive to thiooxime structure) in response to thiol functionality.³ Thiooximes have been successfully conjugated to methacrylate polymers with pendant aldehyde functionality in a post-polymerization modification approach.⁴ This philosophy was extended to the preparation of H₂S-releasing nanoparticles by preparing thiooxime-functionalized block copolymers that self-assemble in aqueous solution to form spherical micelles. In this case, the kinetics of H₂S-release can be tuned by manipulating various parameters of the polymer aggregates (i.e., polymer thermal properties, particle size, aggregate morphology, etc.). This release reaction occurs on the timescale of days, in contrast to most H₂S-relasing small molecule compounds that exhibit full release in a few hours. These H₂S releasing micelles exhibit anti-cancer activity on human colon cancer cells *in vitro* but are well-tolerated by healthy cells.

**Figure 1.** H₂S release from thiooxime micelles exhibits anti-cancer activity on human cancer cells (HCT116) but is well-tolerated by healthy cells (NIH3T3).

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**References**

Increased Monomer Content of Acrylate-Based Shape Memory Polymers (SMPs) Increases Endothelial Cell Attachment on SMP Surfaces

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Increased understanding of interactions between polymer surfaces and cells has paved the way toward expanding the application of polymers to medical devices that require healthy cell-surface interactions for optimal in vivo performance. Over the last decade, we have adapted bulk characteristics of acrylate-based shape memory polymers (SMPs) and have deployed these in a number of minimally invasive device applications. While thermomechanical and bulk properties of these materials have been investigated extensively, surface properties, which govern cell attachment, have been explored less. Surface properties can be manipulated, via surface modification, to tailor these materials to achieve a specific purpose, such as endothelialization of the surface to minimize thrombotic response. By encouraging endothelial cell attachment on the surface of SMP devices, we should be able to facilitate the expansion of these materials for use in cardiovascular stents and other blood-contacting devices.

In this work, we investigate the effect of changing the chemistry of the SMP on endothelial cell attachment. Nine different formulations were fabricated by changing the weight percent ratio of the monomer, tert-Butyl acrylate (tBA), with respect to the crosslinker, poly (ethylene glycol) dimethacrylate (PEGDMA). The surface characteristics quantified were wettability, obtained from contact angle measurements, and surface roughness, quantified using the roughness coefficient ($R_a$) from atomic force microscopy (AFM).

We found that increasing the acrylate monomer content, which results in rougher, more hydrophobic surfaces, leads to increased endothelial cell attachment on SMP surfaces. Successful SMP formulations had high cell viability, visualized using Live/Dead assay as well as increased cell metabolism, quantified by increased reduction of non-fluorescent resazurin into fluorescent resofurin, both of which were visualized using fluorescent techniques. Increased understanding of surface issues for these acrylate-based SMPs may help these materials become viable options for use in cardiovascular devices.
Effect of Humidity on the Adhesion of Black Widow Spider’s Gumfoot Silk

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Adhesion in nature is an important aspect with respect to humidity given that many biomaterials perform well in presence of water while commercial glues show a loss of adhesion. Capture silk in spider webs is a potential investigative system for fabricating bio-inspired humidity responsive materials since its performance depends on humidity for the prey adhesion on the web structure. In the case of orb web spiders, the capture silk is known as ‘viscid silk’ and is a combination of glycoproteins and a cocktail of hygroscopic, low molecular weight organic and inorganic salts that aid in water uptake that make the silk tacky in presence of humidity. The salts also directly interact with and stabilize the glycoproteins. Relatively less is known about the composition, adhesion and humidity response for ‘gumfoot silk’ produced by cobweb spiders. In the present study, we first explore the composition of gumfoot silk produced by Black Widow by a host of spectroscopic and staining methods. We use thread pull-off measurements to understand the role of material properties in the adhesion in different humidity conditions. The adhesion results showed an enhancement in adhesion at relative humidity~30% with no further change in adhesion with increase in humidity. To complement adhesion studies, we explored the molecular effects of humidity on silk components using Solid State NMR. Results showed that the salts became mobile with increase in humidity and also there was a synergistic role of salts and glycoproteins in preserving the adhesion across the range of humidity conditions. The findings can serve as a guide for designing synthetic materials including adhesives that work in presence of humidity.

A Low Modulus Multi-functional Polyester Platform for Room Temperature 3D Printing

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3D printing has enabled bench-top fabrication of customized bioengineered constructs with intricate architectures. However, residual solvents, monomers, and initiators typically involved in the fabrication of 3D printed structures create long term translational and regulatory challenges. There is a need for new synthetic soft biodegradable materials that can be well characterized and 3D printed at room temperature. We present the synthesis and 3D printing of a solvent, initiator, and monomer-free biodegradable low modulus polyester platform. Soybean oil derived aliphatic side chains inhibit packing and impart viscous behavior to the polyester. Coumarin pendant side groups crosslink the patterned polymer melts into elastomeric solids through exposure to UV irradiation at 365 nm. Furthermore, we examine the rheological behavior of polyesters within this platform and demonstrate the ability to perform post printing ligand conjugation through click attachment of FITC to surface amines of the 3D printed scaffolds.
Promote encapsulated mesenchymal stem cells survival and differentiation with anti-inflammatory peptides functionalized hydrogels

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Stem cell transplantation has been considered as a promising approach to regenerate ischemic limbs. However, the therapeutic efficacy remains low. One of the reasons is immune rejection after transplantation where the pro-inflammatory cytokines such as IL-1β and TNF-α attack the transplanted cells leading to cell death. In this work, we hypothesized that a cell delivery system that prevents IL-1β and TNF-α from attacking the encapsulated stem cells will enhance cell survival in the ischemic limb. The system was based on a biodegradable and thermosensitive poly(N-isopropylacrylamide)-based hydrogel conjugated with anti-IL-1β and anti-TNF-α peptides. The hydrogel was synthesized by copolymerization of N-isopropylacrylamide, 2-hydroxyethyl methacrylate, dimethyl-γ-butyrolactone acrylate and N-acryloxsuccinimide (NAS) followed by biotinylation. The hydrogel had a LCST of 26.1°C. Peptide conjugation significantly improved cell survival. No cell death was found after peptide conjugation when cultured in the medium containing IL-1β and TNF-α. In contrast, more than 80% of encapsulated mesenchymal stem cells (MSCs) died without peptide conjugation. After transplantation of the developed system into mouse ischemic limbs for 2 weeks, more MSCs were survived with the protection of peptides. The survived MSCs were able to proliferate and differentiate for muscle and blood vessel regeneration. These results demonstrated that the developed anti-inflammatory stem cell delivery system has potential for ischemic limb regeneration.
Improving the Biopharmaceutical Properties of Oligonucleotides by Brush-Polymer-Assisted Compaction

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Abstract: Nucleic acids have long been envisioned as biopharmaceutical agents in many forms of therapies. However, difficult biopharmaceutical characteristics of nucleic acids, such as poor enzymatic stability, rapid clearance by reticuloendothelial organs, immunostimulation, and coagulopathies, limit their application as therapeutics. Many of these side effects are initiated via sequence-specific or non-sequence-specific interactions with proteins. We envisage that a strategy capable of inhibiting protein access but retaining nucleic acid hybridization should bypass many if not all of the side effects associated with oligonucleotide therapeutics. Herein, we developed a novel form of PEG brush polymer-DNA conjugate that provides the DNA with nanoscale steric selectivity: hybridization kinetics with complementary DNA remains nearly unaffected, but interactions with proteins are significantly retarded. We demonstrate that these unimolecular nanoparticles are capable of entering cells and suppressing gene expression without the need of a cationic polymer co-carrier. The PEG brushes also improves the in vivo bio-distribution of oligonucleotide. Therefore, our strategy is a radically new approach to addressing several long lasting challenges in oligonucleotide therapeutics, and the general approach has the potential to be applied to essentially all forms of oligonucleotides, i.e. antisense DNA, siRNA, microRNA, aptamers, ribozymes, etc., to improve their biopharmaceutical characteristics.
Extraction of Cellulose Nanostructures from the Pistachio Shell

By Josh Marett.
Advised by Dr. Johan Foster.

The goal of this project is to determine if the shell of the pistachio nut is a viable material for the extraction of cellulose nanostructures and to determine if there are any special structures in this system. The method of extraction was acid hydrolysis using both sulfuric acid and hydrochloric acid. Characterization is being carried out via FESEM and X-ray diffraction methods. We have confirmed nanostructures which appear to be nano-fibulas cellulose (NFC) based on SEM micrographs as well as the strong response of the [110] and [200] planes as well as a response from the [004] planes, which appear congruent with NFC [1]. Additionally, differences between the two acids have been noted, including degree of crystallinity in the final product. There is still work to determine the best extraction method to remove the excess cellulose and compare the time and temperature of extraction with the degree of crystallinity and the dimensions of the nanostructures. We eventually hope to create a saleable method to extract the nano-structures from this common waste product for industrial use.

- [1] Influence of drying method on the material properties of nanocellulose I: thermostability and crystallinity by Yucheng Peng et al.
Polymeric nanofiber/anti-bacterial formulations using a novel co-extrusion approach

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The present work demonstrates the first successful fabrication of poly(ethylene-co-vinyl acetate) (EVA) fibers using a novel melt co-extrusion process developed at CWRU (1), along with the incorporation of tetracycline hydrochloride (TCH) during co-extrusion and assessment of in-vitro antibacterial activity. This approach can process significantly greater quantities (~3lbs/hr) of EVA-TCH fibers compared with similar electrospun systems (2). The process involves co-extrusion of EVA-TCH with PEO (5% drug) followed by removal of the PEO with a water jet. The fibers derived from the co-extrusion process are typically rectangular. Nominal average fiber dimensions were thicknesses of 3.14±1.23µm, and widths of 1.80±0.65µm (Figure 1). TGA experiments demonstrated that EVA-TCH blend was stable at the thermal processing temperature of 180°C. The in-vitro antibacterial activity of TCH in electrospun and co-extruded nanofibers was determined against Methicillin-Resistant Staphylococcus Aureus and Methicillin-Resistant Staphylococcus Epidermidis (MRSA and MRSE) (Figure 2).

The results demonstrate EVA fiber production by a novel co-extrusion process, along with the ability to successfully incorporate and release TCH. Clinical translation of this and related polymer/drug systems is in progress. Appealing advantages of the continuous co-extrusion process compared with, electrospinning include much higher throughput and better process control.

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OGP-functionalized phenylalanine-based poly(ester ureas) for enhanced mechanical properties and osteoinductive ability for bone regeneration applications

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While there are several clinical repair options at the surgeon’s disposal for treating severe bone injuries and fractures, many of the current treatment strategies result in additional complications for the patient. As a widely expanding field in polymer science, a regenerative medicine approach can be used to overcome these limitations. Phenylalanine-based poly(ester ureas) (poly(PHE)) are high modulus, resorbable polymers with many potential uses in the surgical repair of bone defects. In vitro and in vivo studies have shown that poly(PHE)s have non-toxic hydrolysis byproducts, tunable degradation times, and significant synthetic flexibility.\(^1,2\) OGP is a naturally occurring growth factor which aids in proliferation, differentiation, and mineralization of osteoblasts.\(^3,4\) In vitro data of OGP-tethered poly(PHE) porous scaffolds showed enhanced signaling for osteogenic differentiation of human mesenchymal stem cells compared to control scaffolds lacking tethered OGP.\(^5\) These results led to the synthesis of the second generation poly(PHE)s crosslinked with OGP peptide for increased stiffness and osteoinductive ability for bone repair. Poly(PHE) copolymers were synthesized with pendant allyl functionality for efficient radical-induced crosslinking using cysteine-functionalized-OGP peptides. The synthesis of OGP-peptide crosslinkers was conducted via Fmoc-protected solid phase peptide synthesis. Poly(PHE) copolymers and crosslinked networks were synthesized and characterized, followed by preliminary biological testing to show enhanced osteoinductive ability of OGP crosslinked into a polymer network.

References
Electroactive Polymeric Nanofibers As Active Component in Biosensors.

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Electroactive nanofibers decorated with functional groups that specifically interact with biomarkers have tremendous potential to be utilized as single molecule detectors. Previous investigations in our group used functionalized and processable electronic conductive polythiophenes to study polymer-biomarker interactions. Another approach is to use normally insulating polymer and mix it with single wall carbon nanotubes (SWCNT) to obtain a functional conductive composite. The advantages of using these composite nanostructures include good electrical sensitivity and biocompatibility because of their sizes related to biomolecules. Fabrication of the electroactive composite into nanofibers have shown to be effective not only as carriers of therapeutic agents but also as the active component is biosensor. Nanofibers can be conveniently prepared by electrospinning. Variables affecting the quality of nanofibers include polymer molecular weight, polymer type, and solution viscosity. The molecular weight, concentration, and microphase separation in the nanofibers contributes to the overall physical characteristics of the nanofibers. In this study, we used a thermoplastic elastomeric triblock copolymer as an additive to prepare nanofibers with significantly improved physical properties.

The triblock copolymer poly(styrene)-b-poly(dimethylsiloxane)-b-poly(styrene) (PS-b-PDMS-b-PS) was synthesized by living anionic polymerization. Each block was 10K in molecular weight. A solution with PS-b-PDMS-b-PS / PS at a w/w ratio of 1:5 in DMF along with SWCNT at 1% wt was electrospun onto a silicon wafer at a flow rate of 20µl/min and a potential of 10kV. The fibers were characterized by SEM. I-V plots of the fibers were determined using a four-point probe. A solution with PS 900,000 Mw/SWCNT (100:1) was also spun for comparison with the fiber prepared using the triblock copolymers.
Title: Polyester Brushes as Dynamic Surfaces

Presenter: Cameron L. Stevens
Co-author: Christopher B. Gorman

Borne of nature’s ability to selectively tailor molecular assemblies and interfaces to provide specific, precise molecular interactions, dynamic surfaces, stimuli-responsive surfaces, or smart surfaces have become an increasingly large area of study. This emerging class of materials has the ability to change surface properties in response to a stimuli (e.g., force, pH, temperature, solvent, light), and may find utility in anti-biofouling surfaces. Polyesters brushes, which are known to degrade in a controlled manner, are an example of a dynamic surface. Our initial studies of polyester brushes showed they were far too hydrophobic to resist protein adsorption. Efforts to synthesize new polyesters with more hydrophilic character will be presented, in addition to a new route for creating polyester brushes.
Effects of Lattice Imperfections on the Electrical and Transport Properties of Ultra-Thin MoS$_2$

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Over the past decade, two-dimensional materials such as graphene have shown unique electronic and mechanical properties, making them attractive for applications in micro- and nanoscale electronics. Transition metal dichalcogenides (TMDs), including molybdenum disulfide (MoS$_2$), have recently been considered as alternatives to graphene in transistor applications due to their unique transformation from indirect to direct bandgap semiconductors when reduced to monolayer or few-layer systems. Here, density functional theory (DFT), in combination with a non-equilibrium Green's function (NEGF) approach as implemented in the Atomistix ToolKit (ATK) software package, is used to create a two-probe device model for nanoscale transport characterization of TMDs. The conductivity of few-layer TMD devices can be examined by varying the number of grain boundaries, defects, or dopants within the central scattering region while adjusting the drain-source bias. Information gathered from the device setup includes the density of states (DOS), transmission spectra, IV curves, optical spectrum, transport pathways, eigenstates, and electron density. Through this nanoscale view of transport in TMDs, we hope to guide further experimental investigations and explore the properties and effects of modifications that would give rise the feasibility of TMD devices.
In vivo and in vitro testing of thermoplastic degradable elastomers that mimic natural rubber for soft tissue regeneration applications

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Most biological tissues are highly elastic in nature, a property that has been difficult to mimic in synthetic polymeric biomaterials. Many elastomer biomaterials attempt to replicate the mechanical properties of natural rubber by using hard and soft segments to achieve similar results, but these do not typically contain the cis-1,4 structural component of natural rubber that imparts such high elasticity to the material. Poly(isoprene) is the only synthetic polymer that contains stereoregular cis alkene functionalities, but it is difficult to control the amount of cis content that is incorporated and is not biologically or hydrolytically degradable. Recent work by the Dove group has discovered a series of elastomers with controllable ratios of cis:trans double bonds in the backbone allowing for tunable elasticity. Furthermore, copolymerizing with succinate-functionalized monomers renders this elastomer degradable. This degradability can also be tuned by the monomer feed ratio in order to control the degradation rate. A series of these elastomers containing varied cis:trans ratios and also varied succinate content have been characterized and tested for their potential to be used in soft tissue regeneration applications. Of primary importance, these materials have been tested in vitro and in vivo for their degradation, cell response, and to observe any inflammatory response. Thermal characterization has shown that these materials can be processed at moderately high temperatures. This exciting discovery has led to developments of processing techniques for three-dimensional scaffolds that may be used for regeneration of a variety of soft tissues.
Multimodal Polymeric Nanoparticles for $^{19}$F MRI and Optical Imaging.
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Magnetic Resonance Imaging (MRI) is a popular and promising diagnostic imaging technique, as it offers spatial and anatomical resolution of tissues while providing insight into pathological features through the use of contrast agents. Recent work has focused on using fluorinated MR tracers to provide researchers with a robust handle for imaging tissue. Fluorine is an MR active nuclei, is nearly as sensitive as hydrogen, and no MR active fluorine is present in the body. Fluorine MRI compliments proton MRI by imaging only the contrast agent. Hence, overlaid images provide insight into anatomical features while visualizing diseased tissue via the fluorine tracer. With this in mind, we synthesized a random copolymer comprised of trifluoroethyl methacrylate and polyethylene glycol methylether methacrylate via atom transfer radical polymerization. In water, the copolymer self-assembled into micelles of approx. 350nm. The copolymer offers a single strong peak in the $^{19}$F NMR leading to favorable properties in the $^{19}$F MRI. In-vitro studies using murine macrophages verified viability and uptake of the copolymer into the cells. Using the click-reaction, the copolymer was decorated with an imaging agent allowing for optical detection of the material. In-vivo results from the fluorescently-labeled copolymer indicated localization of the copolymer in a solid tumor with little to no accumulation of the copolymer in other organs. The copolymer design lends itself well to applications in imaging and targeted therapeutics.
Title: Reversible Calcium Ion Contraction and ATP-Induced Re-Expansion of Poly(acrylic acid) Gels

Author: Yu Jen Wang, Case Western Reserve University

Abstract:

The principal biochemical role of adenosine triphosphate (ATP) involves the driving of metabolic processes linked to its large, negative free energy of hydrolysis. However, less well known is ATP's ability to act as a chelator for divalent metal ions [1]. We recently have become very interested in poly(acrylic acid) (PAA) gels as model systems for biomimicking physiological systems such as muscle and nerve [2], especially the ability of PAA to switch from a compact to a swollen gel as the result of divalent/monovalent ion exchange. Swelling of calcium ion-containing gels can be accomplished using common chelators such as EDTA or citrate, but we have found that such gels efficiently re-swell using ATP as the chelator. This is of great interest because two biochemically-significant species, Ca$^{2+}$ and ATP, can be used to switch PAA gels between compact and swollen states in water. A comparison of the relative abilities of EDTA, citrate and ATP to swell calcium ion- contracted gels will be presented.

References:

Dynamic Covalent Assembly of Peptoid-based Ladder Oligomers and Its Registry Mechanism
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Dynamic covalent chemistry, in conjunction with template-directed assembly, enables the fabrication of extended nanostructures that are both precise and tough. Here, we demonstrate the dynamic covalent assembly of peptoid-based molecular ladders with up to 16 rungs via scandium (III)-catalyzed imine metathesis. Owing to their monomer diversity and synthetic accessibility, sequence-specific oligopeptoids bearing dynamic covalent pendant groups were employed as precursors for molecular ladder fabrication. The assembled structures were characterized using matrix-assisted laser desorption/ionization (MALDI) mass spectrometry and gel permeation chromatography (GPC), confirming successful molecular ladder fabrication. To determine the mechanism of hybridization registry, a kinetics study was performed to examine the formation of molecular ladders with different lengths by MALDI mass spectrometry. In addition, distance measurements using Förster resonance energy transfer (FRET) were employed to confirm the registry mechanism.
Synthetic biodegradable polyesters comprise a family of polymers with many potential clinical applications due to their vast diversity and versatility. In this study, a method was developed to functionalize citrate-based biodegradable polyesters in order to utilize them as platforms for drug delivery. The biomolecule incorporated to polymeric systems, nitric oxide (NO), is an endogenous free radical involved in many physiological processes. Integration of NO into polymeric materials is advantageous due to its remarkable broad-spectrum antimicrobial action, as well as its ability to increase cell proliferation and modulate inflammation in wounds. Furthermore, studies have shown that NO can mitigate the foreign body response that often negatively affects the outcome of implantable devices. In addition, the polycondensation reaction included citric acid as a monomeric unit due to its well-studied hemocompatibility and inherently low toxicity. Considering the wide range of potential applications for these materials, their NO-release properties and degradation profiles were studied under physiological conditions. The average total NO loading of poly(citric-co-maleic acid-co-1,8-octanediol)-cysteamine was determined to be 0.45 ± 0.07 mmol g⁻¹, while the NO loading for poly(citric-co-maleic acid-co-1,8-octanediol)-ethyl cysteinate was 0.16 ± 0.04 mmol g⁻¹. Preliminary cell viability assays and morphological studies with human dermal fibroblasts suggested that the citrate-based polyesters may be suitable for future biomedical applications.
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A TISSUE-ENGINEERED MODEL OF A TENDON-TO-BONE ENTHESIS

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ABSTRACT

The tendon-to-bone enthesis has a four-zone gradation of connective tissue that includes tendon followed by uncalcified cartilage, calcified cartilage, and bone. The loss of the tendon-to-bone attachment is a common and difficult problem in orthopedic injuries of the hand, elbow, shoulder, knee, heel and foot. These injuries may be treated surgically by repair, reconstruction or grafting methods, processes that may be beneficial to the patient but may also lead to complications such as adhesion formation, tenolysis, persistent tears and sub-optimal function\(^{1-2}\).

This study develops a tissue-engineering approach to generate a functional tendon-to-bone insertion site (an ‘enthesis’), utilizing a phalanx model well characterized previously\(^{3-5}\). This complete tissue-engineering process included fabrication of tendon-to-bone constructs, culture of constructs for 1-2 weeks, implantation of constructs in nude mice serving as bioreactors in vivo, construct harvest from mice, and retrieved construct immunohistological and biomechanical assessments. Preliminary data from a recently harvested 20-week construct demonstrated that the fabricated cell-seeded enthesis (II-IV, Fig. A) was viable and possessed stiffness three times greater than its non-seeded aspect (I, Fig. A). Subsequent studies will compare morphology, immunohistochemical markers, and additional biomechanical properties of fabricated entheses to the normal tendon-to-bone attachment of cadaveric or non-cadaveric adult surgical specimens of the human phalanx. Methods developed here could be applied in a broad translational sense for treatment and repair of isolated tendon injuries such as tears of the flexor tendon, Achilles tendon, and rotator cuff.

Figure A. A schematic of a fabricated enthesis model. I: poly(caprolactone-co-L-lactic acid) (PCL/PLLA), II: fresh (< 24 hr old) cadaveric human periostium wrapped about human allograft bone, III: poly(glycolic acid) seeded with human chondrocytes, and IV: PCL/PLLA seeded with human tenocytes. Figure B. Image of an enthesis construct after implantation in an athymic mouse for 20 weeks.
References


