Biomaterials Day
October 19, 2018
James B. Hunt Jr. Library,
NC State University

Sponsored by
Welcome to the Society for Biomaterials (SFB) student chapter at North Carolina State University!

The ‘Society for Biomaterials’ is an organization that connects individuals from many disciplines such as technology, health, and government to the field of biomaterials. Materials science, biomedical engineering, medical textiles, microbiology, and polymer science are several of many areas that have utilized biomaterials to improve health and life.

The goal of the Society for Biomaterials student chapters is to identify students interested in biomaterials, to generate student interest and interaction in biomaterials, and to aid the efficacy of students seeking research, education, and professional development opportunities.

The purpose of the Biomaterials Day event is to connect groups from all disciplines to gather and collaborate on advances in biomaterials used in healthcare and medical applications. It is an event designed to provoke inspiration, ideas, and connections under one roof. Welcome to the 2nd annual Biomaterials Day!
# AGENDA

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<td>8:00 am - 8:30 am</td>
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| 8:30 am - 8:45 am | Welcome address  
Dr. Jeffrey Joines, College of Textiles, NC State University |
| **Session 1** | **Development of New Technologies**                                  |
| 8:45 am - 9:35 am | Invited speaker  
Dr. Ke Cheng, Joint Department of Biomedical Engineering, North  
Carolina State University and the University of North Carolina at  
Chapel-Hill  
Biomaterial and biomimetic approaches for cardiac cell therapy |
| 9:35 am - 9:50 am | Ria D. Corder, Chemical and Biomolecular Engineering, NC State  
University  
Quantifying modulus, viscoelasticity, and enzymatic degradation of  
mammalian tissues using rheology |
| 9:50 am - 10:10 am | Coffee break & Poster setup                                           |
| **Session 2** | **Biomaterials for Medical Applications**                             |
| 10:10 am -11:00 am | Invited speaker  
Dr. Edwin R. Cadet, MD, Raleigh Orthopaedics  
Innovations in Rotator Cuff Repair |
| 11:00 am - 11:15 am | Emily P. Mihalko, Joint Department of Biomedical Engineering,  
North Carolina State University and the University of North  
Carolina at Chapel-Hill  
Wound-targeting nanogels deliver tissue plasminogen activator for  
treatment of disseminated intravascular coagulation |
| 11:15 am - 11:30 pm | Ismaeel Muhamed, Joint Department of Biomedical Engineering,  
North Carolina State University and the University of North  
Carolina at Chapel-Hill  
Fibrin nanoparticles coupled with keratinocyte growth factor  
enhance dermal wound closure rate |
| 11:30 pm - 12:30 pm | Lunch & Networking  
Dr. Jason Cramer, the Graduate School, NC State University |
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| 12:30 pm - 1:20 pm | Invited speaker  
Dr. Barbara Nsiah, United Therapeutics Corporation  
Tissue Engineering: From Academia to Industry | Invited speaker  
Dr. Thomas H. LaBean, Materials Science and Engineering, NC State University  
Engineering Molecular Assembly for 3D Electronics & Medicine |
| 1:20 pm - 1:35 pm | Ashish Kapoor, College of Textiles, NC State University  
Fiber based Wearable Sensors for Biomedical Monitoring Applications | Fan Zhang, College of Textiles, NC State University  
A Small Diameter Tissue-Engineered Vascular Graft of Collagen Yarns and Electrospun Collagen Nanofibers |
| 1:35 pm - 1:55 pm | Coffee break | Student poster session |
| 1:55 pm - 2:45 pm | Invited speaker  
Dr. Martin W. King, College of Textiles, NC State University  
Awards & Closing |
| 3:00 pm - 4:00 pm | | |
| 4:00 pm - 4:30 pm | | |

*Session 3: Commercialization of Biomaterials & Medical Products*

*Session 4: Biomaterials in Tissue Engineering and Regenerative Medicine*
INVITED SPEAKERS

Dr. Ke Cheng
Professor, Joint Department of Biomedical Engineering, North Carolina State University and the University of North Carolina at Chapel-Hill
Raleigh, NC

Interests:
● Organ-specific adult stem cells and stem cell derivatives
● Targeted nanomaterials for heart regeneration
● Biomaterials for cardiac tissue engineering
● New mechanisms of cell extravasation

Dr. Edwin R. Cadet, M.D
Orthopaedic Surgeon, Raleigh Orthopaedics
Raleigh, NC

Specialties:
● Elbow
● Hip
● Knee
● Shoulder
Dr. Barbara Nsiah  
Senior Scientist, United Therapeutics Corporation  
Raleigh, NC  

Interests:  
● Research and development  
● Product development  
● Tissue engineering in lungs generation

Dr. Thomas H. LaBean  
Professor, Materials Science and Engineering, North Carolina State University  
Raleigh, NC  

Interests:  
● Self-assembling DNA nanostructures  
● Molecular materials  
● Bioinspired nanoelectronics fabrication  
● Nanomedicine
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Society For Biomaterials
Student Chapter at NC State University
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Quantifying modulus, viscoelasticity, and enzymatic degradation of mammalian tissues using rheology

Ria D. Corder¹, Robert B. Vachieri², Darlene K. Taylor², Jodie M. Fleming³, Friederike L. Jayes⁴, and Saad A. Khan¹

¹Chemical and Biomolecular Engineering, North Carolina State University; ²Chemistry and Biochemistry, North Carolina Central University; ³Biological and Biomedical Sciences, North Carolina Central University; ⁴Obstetrics and Gynecology, Duke University

Biological tissues are complex composite materials whose mechanical properties are often difficult to measure by traditional techniques. Quantification of bulk tissue properties, such as modulus and viscoelasticity, can be used to aid in disease diagnosis and design of novel therapies. The novelty of our work is the application of rheological characterization to mammalian tissues. First, we show data collected from reduction mammoplasty human breast tissues isolated from multiple individuals and demonstrate that rheology can quantify tissue variability. Repeated freeze-thaw studies show the effect of sample history and highlight the need for consistent protocols for handling biological samples.

Next, we demonstrate how rheology can be used to quantify the in-vivo efficacy of a novel injectable treatment for uterine fibroids. Our treatment consists of co-injected collagenase Clostridium histolyticum (CCH) to digest the fibroid collagens¹ and Liquo-gel (LQG), a thermoresponsive polymer gel, to entrap CCH within the fibroid. Fibroids obtained from human women undergoing hysterectomies were injected with our treatment, surgically implanted into mice, and then removed at set day intervals for measurement. We observed that co-injecting LQG+CCH significantly reduced tissue modulus and increased viscoelasticity compared to both buffer controls and free CCH injections. We also show how atomic force microscopy and histological staining of parallel tissue samples can be used alongside rheology to gain a more complete understanding of treatment effects on tissues.

To conclude, we end with two examples of how rheology can characterize the sol-gel transition of biomaterials for drug delivery. The LQG polymer used in our uterine fibroid treatment undergoes reversible gelation upon heating. Using rheology, we identified the gelation temperature and showed that LQG exhibits thermal hysteresis during heating and cooling. Lastly, we present how we measured the gelation time of a crosslinked poly(vinyl alcohol) gel for insulin delivery².

References:
Wound-targeting nanogels deliver tissue plasminogen activator for treatment of disseminated intravascular coagulation

Emily P. Mihalko1,2, Kimberly Nellenbach1,2, Nicholas Mininni1, Ashley C. Brown1,2.
1Joint Department of Biomedical Engineering, North Carolina State University and the University of North Carolina at Chapel-Hill, Raleigh, NC; 2Comparative Medicine Institute, North Carolina State University, Raleigh, NC

Disseminated intravascular coagulation (DIC) is a pathological process that causes systemic coagulopathy through excessive thrombin generation. While over-activation of clotting causes microthrombi throughout the body, tissue injury, and multi-organ failure, DIC also contributes to bleeding as clotting factors are consumed. Different clinical events can lead to DIC including pregnancy, cancers, trauma, and infections. Although treatment focuses on treating the underlying condition, patients may also present with either bleeding or thrombosis creating therapeutic dilemmas in managing. We hypothesize that simultaneously addressing bleeding and microthrombi associated with DIC can improve treatment. We have developed fibrin-specific nanogels (FSNs) loaded with tissue plasminogen activator (tPA) that can be used to treat the multifaceted complications associated with DIC. Poly(N-isopropylacrylamide) nanogels were synthesized in precipitation polymerization reactions, including acrylic acid copolymer as chemoligation sites for EDC/NHS coupling of a fibrin-specific antibody. Particles were purified and then rehydrated in a tPA loading solution followed by subsequent purification. FSNs on their own (i.e. unloaded), target sites of clotting and augment the clotting process by crosslinking fibrin fibers. Simultaneously, loading tPA into nanogels facilitates clot lysis. FSNs exhibit fibrin-binding in vitro at wall shear rates of 1 sec⁻¹. In a lipopolysaccharide-induced rodent model of DIC, animals exhibit darkened organ color, microthrombi, and hemorrhagic areas in the organs, which are diminished in the tPA-FSN treatment group. Elevated fibrin degradation products in DIC-induced animals validate microthrombi formation and fibrin turnover, while platelet concentrations demonstrate recovery to normal levels only in the tPA-FSN treatment group. In addition, fibrin network structure is severely compromised in DIC as clotting factors are consumed in various organs, and is not restored in animals receiving FSNs alone. However, FSNs loaded with tPA restore clot structure to that seen in healthy animals. In conclusion, fibrinolytic loaded fibrin-targeting microgels balance procoagulant and fibrinolytic action in DIC to improve outcomes.

Figure 1: Whole organ pictures and histological sections from organs stained with Martius scarlet blue (MSB) to visualize fibrin (red). Black arrows point to hemorrhagic areas in the organs and red to fibrin deposition in stained tissue sections (A). CryoSEM images from plasma samples with or without LPS induced DIC. DIC-induced animals were treated with either saline, FSNs, or tPA-FSNs 30 minutes prior to blood collection and plasma isolation (B).
Engineering fibrin nanoparticles to enhance the early stages of wound healing

Ismaeel Muhamed\textsuperscript{1,2}, Frances Ligler\textsuperscript{1,2}, Ashley Brown\textsuperscript{1,2}

\textsuperscript{1}Joint department of Biomedical Engineering University of North Carolina Chapel Hill and North Carolina State University; \textsuperscript{2}Comparative Medicine Institute NCSU.

The inability of blood to quickly coagulate and facilitate tissue repair is a major concern, especially in hemophilia, diabetes and Von Willebrand disease (VWD). The CDC reports that delayed blood clotting is responsible for 18.9\% of blood based coagulation deaths in 2010. FDA-approved commercial blood clotting agents use >500 times physiological concentrations of fibrinogen and thrombin to thwart blood loss, but the ensuing wound tissue remodelling events are affected.

Our objective is to synthesize pre-polymerized fibrin nanoparticles that can be used to enhance wound healing by:

- Creating fibrin nanoparticles that assist sealing of wounds.
- Improving cell migration using activated fibrin chemotactic ligands while providing appropriate tissue mechanics and porosity.

Our fibrin nanoparticles are created in a medium throughput two-phase Y-channel microfluidic droplet device (Fig A). The size of the fibrin droplets is tuned by altering the flow velocity and shear ratio. The biocompatibility and functionality of the designed fibrin nanoparticles were studied for their ability to enhance cell migration (at early time points) and wound closure responses (Fig C-E). Furthermore, the generated stable fibrin nanoparticles can be further modified in future studies to deliver growth factors and/or inflammatory mediators to further modulate healing outcomes.

Fig 1. A) The microfluidic 2-phase channel that generates fibrin nanoparticles is shown. B) CryoSEM of fibrin nanoparticles. C-D) The \textit{in vitro} wound healing assay used to functionally validate the particles. E) Results of cell migration analysis.
Fiber based Wearable Sensors for Biomedical Monitoring Applications
Ashish Kapoor, Dr. Tushar K. Ghosh

Soft polymer-based sensors as an integral part of textile structures have attracted considerable scientific and commercial interest recently because of their potential use in healthcare applications. While electronic sensing functionalities can be incorporated into textiles at one or more of the hierarchical levels of molecules, fibers, yarns, or fabrics, arguably a more practical means to introduce the desired electrical characteristics is at the fiber level. In this research, we have investigated sensory capabilities of multicomponent fibers consisting of conducting and insulating materials in the fiber cross-sectional structure in a woven structure for biomedical monitoring applications. The multifunctional characteristics of the sensors and their potential are successfully demonstrated by measuring tactile, tensile, and shear deformations, as well as wetness and biopotential (ECG). The current efforts are towards integration of these fiber based sensors into the socket/liner structure without causing subject discomfort for objective monitoring of the inner prosthetic environment (i.e. pressure, temperature, and humidity) and residual muscle activity (EMG). These multimodal sensors can also be used for gait analysis to study plantar pressure distribution and predict occurrence of diabetic foot.
Cardiovascular disease is the number one cause of death worldwide. Small caliber (<6mm) arterial prostheses made from synthetic materials are known to fail due to thrombosis, mechanical mismatch, and/or activation of a chronic inflammatory foreign material response [1,2]. As the predominant protein component of native tissue, collagen is a promising biomaterial for vascular tissue engineering. However, the major challenge is to design the collagen-based graft with sufficient mechanical performance.

In this study, recently developed electrochemically aligned collagen (ELAC) yarns [3] were circular-knitted into a tubular structure to fabricate a small-caliber vascular graft that mimicked native vessels mechanically and biochemically. Type I collagen was extracted from rat-tail tendons and aligned into collagen yarns, which were knitted into a single-layer tubular structure. A layer of collagen nanofibers was electrospun onto the luminal surface of the prototype graft [4].

Both the single layer and bilayer collagen grafts showed comparable bursting strength and suture retention strength to autologous vessels. There was no significant difference between the single layer knitted collagen sample and the bilayer samples with the knitted outer layer and electrospun luminal layer (p<0.05), indicating that the mechanical properties are contributed primarily by the knitted layer.

The AlamarBlue assay results indicated that the collagen grafts promoted cell attachment to the outer layer, while the inner electrospun layer reduced the pore size and encouraged endothelial cell proliferation.

In previous studies, the most common methods to fabricate collagen grafts have used casting, electrospinning and bioprinting, all of which are limited by inferior mechanical properties [5]. In this study, the combination of collagen yarns and knitting technology produced a small caliber vascular graft with comparable mechanical strength to native blood vessels, and increased the extent of cell proliferation compared to synthetic polymeric grafts.

References:
Hydrogels are three-dimensionally crosslinked, macromolecular and hydrophilic networks insoluble in water. Hydrogels have unique characteristics to absorb and release water solutions in a reversible manner, in response to specific environmental stimuli such as temperature and pH etc. This attribute of hydrogel makes them a versatile candidate for many biomedical applications especially drug delivery and wound healing.

Hydrogel blends (PHG 1-5) were prepared via ultrasonication and blending techniques using micro-crystalline cellulose and sodium carboxymethyl cellulose (Na-CMC) in different concentrations with epichlorohydrin as a cross-linking agent. Cellulose behaves as a strong backbone of the cross-linked network whereas CMC, containing carboxyl (COO⁻) anions, imparts hydrophilic properties to the hydrogel improving overall porosity in the designed network. Hydrogels during the gel formation process, lead to reduced crystallinity and a higher adsorption capacity for heavy metal ions. These hydrogels have been characterized by physical testing (swelling ratios), Fourier transform infrared spectroscopy (FTIR), thermal analysis (TGA), x-ray photoelectron spectroscopy (XPS), field emission scanning electron microscopy (SEM), energy dispersive spectrometer analysis (EDX) as well as in vitro cytotoxicity analysis.

The morphology of hydrogels is an important characteristic to be determined as the water absorbency depends upon their porosity. Figure 1 shows the SEM micrographs and EDX scans for two of the prepared hydrogels. SEM images show that these hydrogels exhibited a three-dimensional well defined microporous network structure with an undulant, channel-like appearance which led to an increase in surface area and capillary action responsible for high absorption. FTIR and EDX confirmed the chemical interaction between Na-CMC and cellulose as well as the elemental ratio between the 5 formulations, respectively. In addition, in vitro cytotoxicity analysis showed a non-toxic performance toward cells viability and functionality.

With increase in the concentration of Na-CMC, the undulating channels became more dominant. The molecular chain repulsion also increased which led to the development of coarser meshes (PHG-5), which favored more water uptake. The developed hydrogels with increasing Na-CMC content facilitated the diffusion of water and improved swelling performance.

The prepared hydrogels can be employed for different biomedical applications such as personal hygiene products, implantable biomedical textiles, wound dressings, artificial muscles, substrates for controlled drug delivery. They also have potential application in the removal and recovery of heavy metal ions.

We are planning to incorporate these hydrogels within spacer fabrics made from inherently antimicrobial polymeric fibers or polymeric fibers imbedded with an antimicrobial agent. The preparation of various wound dressing formulations using these hydrogels and spacer fabrics will require further validation by in vitro and in vivo methods.
Thermoregulatory Fabrics for Personal Comfort
Kony Chatterjee\textsuperscript{a}, PI: Dr. Tushar K. Ghosh\textsuperscript{b}

\textsuperscript{a}Fiber and Polymer Science Program, College of Textiles, North Carolina State University, Raleigh, NC 27606; \textsuperscript{b}Department of Textile Engineering, Chemistry, and Science, College of Textiles, North Carolina State University, Raleigh, NC 27606

As energy costs and global temperatures rise world-over, an important way of saving costs and combating climate change is to eliminate the heating and cooling of buildings, without compromising on the comfort that is imparted by a temperature-controlled living space. Textiles offer an innovative solution in this regard. Textiles have always occupied an important role in human survival, and due to their conformable and flexible nature, they are an obvious choice for integrating various “smart” functionalities without sacrificing their inherent comfort properties. One such functionality that we wish to explore is as personal cooling garments by the integration of thermoelectric (TE) materials into textiles. Thermoelectricity is the property of materials wherein they can convert a temperature difference into electricity (Seebeck effect), or electricity into a temperature gradient (Peltier effect). Using Peltier effect, the objective is to design fiber-based wearable TE fabric that provides a transformative level of comfort via next-to-skin cooling next to the body – thereby eliminating the need to heat or cool an entire living space. Using polymeric TE materials integrated into fabrics in a woven structure, along with heat sink fibers to transport the rejected heat, results in a conformable, unobtrusive, fabric-based cooling system. Such thermoregulatory fabrics can offset the energy demands for building thermo-regulation by up to 15% and reduce the greenhouse gas emissions by 2%, resulting in a truly transcendent textile system, befitting a future where fossil fuels are scarce and renewable energy solutions are important for humanity’s survival.
Platelets are important to stopping bleeding and promoting wound healing following injury. In traumatic injury and chronic wounds, platelets can be depleted and the healing process can be complicated by bacterial infection. Our group has previously developed synthetic platelets (SPs), consisting of highly deformable microgels coupled to fibrin targeting antibodies\(^1\) that augment hemostasis and mimic native platelet clot retraction, which is a feature that we hypothesize to promote healing by promoting durotaxis and enhancing cell infiltration. The purpose of this study is to enhance this design and develop antimicrobial synthetic platelets. To fabricate antimicrobial SPs, we created nanogold composite (NGCs) SPs by covalently and noncovalently incorporating nanogold. Covalent incorporation is achieved via THPC-assisted covalent seeding\(^2\). Noncovalent incorporation is achieved by allowing the microgels to breath in nanogold spheres via suspension in nanogold solution. Deformability was characterized by size and height data collected by AFM and DLS. Composite SP nanometal distribution and stability was characterized by TEM imaging. Clot retraction was characterized by incorporating nGCs into fibrin clots and imaging via CryoSEM. Antimicrobial ability was characterized by a colony forming assay consisting of culturing S. Aureus and E. Coli in the presence of NGCs. A spheroid cell migration assay using human dermal fibroblasts (hDFn) was conducted to characterize durotaxis\(^3\). It was found that NGCs were similar to unloaded microgels in size, height, and clot retraction ability. Antimicrobial activity was higher in covalent NGCs than noncovalent. NGCs were found to induce a difference in cell migration, however, batch-to-batch variability was observed. In conclusion, we have successfully synthesized NGC SPs. Nanogold incorporation does not influence SP clot retraction ability and covalent NGCs have an antimicrobial effect, and NGCs have an influence on cell migration. In the future, nanosilver incorporation will be investigated to better optimize antimicrobial action.

Figure: Section A – Representative TEM images of nanometal composites. Section B – Results of E. Coli for the antimicrobial assay. Section C – Results of cell migration assay.

References:
In Vivo Anchoring Performance and Histological Examination of Barbed Surgical Sutures in a Rat Model

Hui Cong\textsuperscript{1}, Gregory L. Ruff\textsuperscript{2}, Jacqueline H. Cole\textsuperscript{3}, Debra A. Tokarz\textsuperscript{4}, Simon C. Roe\textsuperscript{4}, Martin W. King\textsuperscript{1,5}, Wei Liu\textsuperscript{6}

\textsuperscript{1} College of Textiles, North Carolina State University, Raleigh, NC, \textsuperscript{2} 55 Vilcom Center, Chapel Hill, NC, \textsuperscript{3} Joint Dept. of Biomedical Engineering, North Carolina State University and University of North Carolina at Chapel Hill, \textsuperscript{4} College of Veterinary Medicine, North Carolina State University, Raleigh, NC, USA, \textsuperscript{5} Donghua University, Shanghai, \textsuperscript{6} Shanghai Ninth People’s Hospital, Shanghai Jiaotong University School of Medicine, Shanghai, China

The benefits of barbed surgical sutures such as knots elimination, uniform stress distribution and improved efficiency enable its successful application in various fields for example, dermatology, urology, orthopedics and so on. Polydioxanone (PDO) is a well-known biomaterial for both conventional sutures and barbed sutures. A recently developed biopolymer, poly-4-hydroxybutyrate (P4HB) manufactured by means of bacterial fermentation is a promising material for barbed sutures due to the outstanding tensile properties in a prolonged degradation profile. Anchoring performance generated by barb engagement with the surrounding tissues has been measured by suture/tissue pullout test \textit{in vitro} and wound closure test \textit{in vivo}. The first purpose of the present study is to examine the \textit{in vivo} anchoring performance of PDO and P4HB barbed sutures using suture/tissue pullout test in a rat model. The second purpose is to study the histological interactions between barbs and the surrounding tissues in terms of inflammatory cell infiltration and foreign body reaction in both cross-sectional and longitudinal views.

Size 2-0 PDO and P4HB monofilaments were used to fabricate barbed suture samples using a mechanical barbing machine donated by Quill Medical Inc. Twenty-five female Sprague-Dawley rats were randomly assigned to one of five groups corresponding to the duration of barbed suture implantation and designated postoperative day of assessment: Day 0, 3, 7, 14 and 28. Seven barbed sutures were inserted on the dorsal back of each rat following a series of predetermined three-stitch makers. Customized Elizabethan collars were used to prevent rats from biting and pulling of the implanted sutures. The anchoring performance was measured on a universal tensile tester as the maximum force when individual barbed suture was pulled out from the rat skin tissue in a rate of 20 mm/min. The harvested skin tissues with suture materials were embedded in glycol methacrylate (GMA) using a Technovit® 7100 embedding kit and sectioned in a thickness of 4 \textmu m for histological examination. The relative degree of tissue reaction was scored according to a scoring scheme published by Zaruby et al. \cite{1}.

Results in Figure 1 shows that the anchoring performance of PDO barbed sutures at Day 0 was 5.78 N, which was significantly higher than that of P4HB barbed sutures with 3.28 N. This was attributed to the flimsy barbs fabricated on P4HB monofilaments with low hardness. The significant difference was observed on both Day 3 and Day 7, with the anchoring performance of both suture materials decreased. However, due to the rapid material degradation, the anchoring performance of PDO barbed sutures dropped to 1.38 N at Day 28, which was significantly lower than that of P4HB barbed sutures. Histological results showed that fewer cells were observed to infiltrate into the operative area of P4HB barbed sutures compared with PDO sutures. Tissues inserted with P4HB barbed sutures had a lower degree of necrosis and congestion/edema. The score of foreign body reaction was same for both suture materials.

The anchoring performance of PDO barbed sutures evaluated by suture/tissue pullout test in a rat model decreased gradually with the increase of implant duration days, while that of the P4HB barbed sutures increased from Day 14. For the first time, the tissue reactions with barbs were examined by combination of both cross-sectional and longitudinal views. In a summary, P4HB can be a promising material for barbed suture application, especially for long-term use.

Reference:

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Development of the Components of an Engineered Heart Tissue (EHT) using Extracellular Matrix (ECM)-Derived Hydrogels

Erin C. Dowell¹, Emily A. Wrona¹,², Donald O. Freytes, PhD¹,²
¹ UNC-Chapel Hill/NCSU Joint Department of Biomedical Engineering, Raleigh, NC;
² Comparative Medicine Institute, North Carolina State University, Raleigh, NC

The human heart requires specific structural and functional components to ensure the heart is beating in synchrony and mediating efficient transport of blood. In vitro models for studying the heart function can be advantageous to animal models by allowing large variable screening at a lower cost. Engineered heart tissues (EHTs) are a type of in vitro model in which cardiomyocytes (CMs) are suspended within extracellular matrix (ECM)-derived hydrogels that contract around two posts. To create this model, cellular and hydrogel components were developed separately with the ultimate goal of creating a model that mimics, not only the structural composition of the heart, but also its function. The cellular components of this system include fibroblasts, CMs, and macrophages (MΦs.) MΦs present in the atrioventricular node facilitate the conduction of the heart by connecting with CMs through gap junctions created by a surface protein called Connexin 43 (Cx43.) Since MΦs have not been previously incorporated within EHTs, we need to determine the optimal MΦ subtype that can be used in vitro. Given the availability of monocyte-derived MΦs from human peripheral blood, our goal was to determine which polarization state better resembles the cardiac MΦs found in vivo. To quantify these potential connections, MΦs were polarized in vitro into several functional subtypes and assessed for their expression of Cx43. It was determined that the pro-inflammatory MΦ subtype had the highest expression of Cx43, suggesting that this polarization state was most likely to connect with CMs. To create the hydrogel component, porcine tissue was decellularized by a series of washes until only the ECM remained. The ECM was lyophilized, ground into a powder, and followed by enzymatic digestion, resulting in a self-assembling hydrogel. Future work will continue to validate the efficacy of our ECM-derived hydrogels and MΦ subtype as additions to a superior EHT.
The Development of a Three-Dimensional In Vitro Model of Mast Cell Neoplasia
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Preclinical drug development for canine mast cell tumors (MCTs) is difficult due to the biological and clinical diversity of these diseases. Mutations in the proto-oncogene c-KIT cause activation of the receptor tyrosine kinase KIT, leading to neoplastic mast cell growth in the skin, gastrointestinal tract, and bone marrow. Currently, in vitro models and murine models do not accurately represent the highly variable tumor cell growth that naturally occurs in canine disease, making it difficult to develop new therapeutic strategies. We have developed a three dimensional (3D) in vitro model to study mast cell neoplasia, using extracellular matrix (ECM) hydrogels from decellularized porcine dermis and the canine mast cell line HRMC, derived from a dog with MCTs. We hypothesize that culturing HRMCs within a tissue-specific 3D structure will influence cell behavior, leading to more accurate representation of tumor cell growth in vivo. We confirmed expression of the KIT gene and protein using reverse transcription polymerase chain reaction (RT-PCR) and flow cytometry, respectively. Histochemical staining, alamarBlue, PicoGreen, and LIVE/DEAD viability assays were used to analyze cell survival and proliferation. In comparison to control gels made from collagen, cells in ECM hydrogels indicated increased rates of cellular activity and indices of malignancy. We found good cell viability, defined as roughly 85% live cells, in hydrogels between 24 and 72 hours, indicating that these time points would be optimal for future mast cell phenotype experiments. Our results suggest that this model mimics MCTs in vivo, and is therefore a useful tool for the development of new therapeutics for mast cell neoplasia.
Myocardial Extracellular Matrix Impregnated with Synthetic Cardiac Stem Cells for Cardiac Regeneration

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Stem cell therapy has been a promising strategy for therapeutic heart regeneration. To overcome the limitation of low cell retention/engraftment, the cardiac patch strategy involves seeding a scaffolding material with stem cells before transplantation onto the surface of the heart. These patches have to be freshly made and long-term storage is not feasible. We developed an off-the-shelf therapeutic cardiac patch, composed of the decellularized porcine myocardial extracellular matrix from pig myocardium, and synthetic cardiac stem cells (synCSC) from the secreted factors of human cardiac stem cells. This fully acellular artificial cardiac patch (artCP) maintained its potency after long-term cryopreservation. In a rat model of acute myocardial infarction, transplantation of artCP supports cardiac recovery by reducing scarring, promoting angiomyogenesis, and boosts cardiac function. The safety and therapeutic efficacy of artCP are further confirmed in a porcine MI model. Compare to current cardiac patch strategies, artCP offers clinically feasible, easy-to-store, and cell-free advantages.
Additive manufacturing (3D printing) is emerging as a key manufacturing technique in medical devices. In particular, selective laser melted (SLM) Ti-6Al-4V implants with interconnected porosity have become widespread in orthopedic and other load bearing applications where porous structures encourage bony ingrowth and the stiffness of the implant can be tuned to reduce stress shielding. The SLM technique allows high resolution control over design, including the ability to introduce porosity with spatial variations in pore size, shape, and connectivity. This study investigates the effect of construct design and surface treatment on tensile fatigue behavior of 3D printed Ti-6Al-4V, which represents the worst-case scenario for implants. Samples were designed as solid, solid with an additional surface porous layer, or fully porous, while surface treatments included commercially available rotopolishing and “SILC” cleaning. All groups were evaluated for surface roughness and tested in tension to failure under monotonic and cyclic loading profiles. In solid 3D printed materials, surface treatments were shown to improve surface roughness and fatigue behavior as compared to non-treated surfaces. With sufficient reduction in surface roughness, the fatigue strength of 3D printed solid samples approached 40% to 50% of the ultimate tensile strength of identical 3D printed solid material. Conversely, the surface porous and fully porous materials demonstrated little to no improvement in fatigue behavior with surface treatment. Irrespective of surface treatment and resulting surface roughness, the fatigue strength of 3D printed samples containing bulk or surface porosity was approximately 20% to 25% of the ultimate tensile strength of identical 3D printed porous material. This study highlights the relative effect of surface treatment in solid and porous printed samples and the inherent decrease in fatigue properties of 3D printed porous samples designed for osseointegration.
Platelet-like Particles Augment Fibrin Matrices to Promote Wound Healing Events
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The wound healing process incorporates several functions of native platelets, including binding to fibrin to promote clotting and interacting with fibrin fibers to induce clot retraction in order to increase clot stiffness and stability1,2. Our lab has developed synthetic platelet-like particles (PLPs) capable of recapitulating the fibrin binding and clot retraction capabilities of native platelets. Since changes in matrix stiffness have been shown to affect cell migration behaviors, we hypothesized that PLPs would promote cell migration and thus improve wound healing outcomes3. In these studies, we characterize the forces associated with PLP-mediated clot retraction, concomitant increases in clot mechanics, and the effect of clot retraction on wound repair events in vitro and in vivo, and then investigate the effects of PLPs on clotting in a hemophilic healing model.

PLPs were created by conjugating ultralow crosslinked poly(N-isopropylacrylamide) micro-scale hydrogels (microgels) to fibrin-specific antibodies. To characterize contraction forces we utilized a micropost deflection method; microposts were created from PDMS and seeded with fibrin-only clots or fibrin + PLP clots. Post deflection was analyzed to determine the forces exerted by the PLPs. Clot stiffness and structure were characterized via AFM and CryoSEM. We then examined PLP-mediated cell migration within fibrin clots in vitro and PLP-mediated wound healing in vivo using a full-thickness dermal wound healing mouse model. Post deflection increased in the presence of PLPs. CryoSEM and AFM showed decreased network porosity and increased stiffness in the presence of PLPs. In vitro cell migration and in vivo wound closure were increased in the presence of PLPs. Hemophilia models indicate that clot structure and stiffness is enhanced in the presence of PLPs (Figure 1).

Together, these results demonstrate that PLPs exert forces on a fibrin network, increase clot stiffness and enhance cell migration and wound closure in healthy and hemophilic wound conditions.

Figure 1: PLPs increase density and Young’s modulus and decrease porosity in hemophilic clots relative to controls. *p<0.05; **p<0.01; ***p<0.001; ****p<0.0001
Photodynamic Polymers for Anti-Infective Materials
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Increase of antibiotic resistance in pathogens has directly impacted healthcare industry. With only a few novel discoveries in the field of antibiotics since last two decades, often referred to as the discovery void, drug-resistance in pathogens has increased. Previously, infections that were easily treatable have now become fatal. Infections caused by antibiotic-resistant pathogens can occur anywhere, but, it is observed to take maximum effect in healthcare settings such as hospitals and nursing homes. Adherence and proliferation of microbes on surfaces such as counter tops, drapes, linens, door handles, monitory and sanitation equipment in health-care settings contribute to increase in HAIs. As increase in microbial drug resistance causes conventional methods of treatment to fail, researchers are looking at alternative routes to tackle the infections. Photodynamic therapy (PDT) is such a technique that uses a photosensitizer (PS) and a light source, to treat medical conditions such as acne, wet-age macular degeneration and initial stages of skin cancer. Initially, the PS is applied on a target area of cancer cells. Subsequently, the target area is illuminated by visible light (typically of red color), thus, activating the PS. The activated PS, through interactions with ground state triplet oxygen diffusing through the cells, converts it into singlet state oxygen. Being very reactive, singlet oxygen can oxidize various components in the cancerous cells leading to its death. Instead of ex post facto medical treatment, we intend to incorporate the PS on surfaces that will result in inactivation of microbes by continuous surface disinfection and serve as a preventive measure. In this study, we have incorporated a PS, belonging to the porphyrin class, in an olefin block copolymer (OBC). Melt pressed PS/polymer films were prepared. Five bacterial and three viral strains were tested and all showed at least 99.89% inactivation after 60 min exposure to non-coherent visible light.
Integrating Experiments and Molecular Dynamic Simulations to Understand the Degradation of Bioresorbable Medical Textiles in the Form of Surgical Sutures
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With the advent of new resorbable polymers and their use as medical textiles, it is important to understand their degradation or resorption mechanism inside the body’s living environment, the reaction pathway, and the changes in properties during the resorption process. In this research study, we are evaluating the use of a novel polymer, poly-4-hydroxybutyrate (P4HB), as a surgical suture. P4HB is a thermoplastic, aliphatic polyester produced by bacteria. The sutures from P4HB are manufactured by melt spinning in the form of monofilaments and multifilament yarns. While previous scientific reports explain some of the properties of this novel polymer, there are still gaps in our understanding of its chemical structure and its resorption or degradation mechanism, particularly as a function of pH which plays a significant role in controlling the rate of suture resorption. Because it has a high degree of polymerization, superior tensile strength, and high flexibility compared to polylactic acid (PLA), it is a suitable candidate as a load bearing surgical implant. Like PLA, P4HB also degrades hydrolytically at the aliphatic ester bonds when placed inside the human body. Our research focuses on the effect of pH and temperature on the degradation behavior of P4HB and PLA in different hydrolytic environments using both an experimental study and molecular dynamic simulations. The results from this research study will be valuable in generating fundamental information about the resorption mechanism and how it correlates with the mechanical and tensile performance of P4HB, both as a suture material and in other medical textile applications such as a hernia mesh. By understanding how the chemical environment inside the human body influences the rate, mechanism, and by-products of the P4HB degradation process, we can confirm its biocompatibility, and predict how to tune the spinning and drawing processes to generate the desired resorption profile.
Antimicrobial Finishing of Hernia Mesh with Carboxymethyl Chitosan

Jingmei Wang, Wendy E. Krause, Ashley C. Brown, Martin W. King

The current treatment for ventral, inguinal or umbilical hernia repair is using an implantable mesh to reinforce the abdominal wall without tension. The most commonly used meshes are made from synthetic materials such as polypropylene (PP), polyester (PET) and expanded polytetrafluorethylene (ePTFE). Synthetic meshes are chemically stable, biocompatible, and have adequate mechanical strength that does not deteriorate with time. However, the post-operative or late infection rate for open surgical repair that involves a mesh is 6%~10%. And the infection not only brings suffering to the patient and additional costs to the healthcare system, but also may require the removal of the mesh during a re-operation, which can lead to other complications.

Therefore, the main objective of this study was to identify and evaluate the effectiveness of applying a surface treatment to a synthetic hernia mesh that will prevent localized bacterial infections. In this study carboxymethyl chitosan (CMC) was selected as a potential antimicrobial agent that could either be chemically grafted and cross-linked or coated onto a commercially knitted polyester hernia mesh without changing the mesh’s mechanical, handling and suturing properties. CMC is derived from chitin, which is obtained from shell-fish, and is known for its hemostatic and antimicrobial properties.

By first exposing the polyester fibers to radio frequency plasma under an oxygen-rich atmosphere, the surface was activated by the generation of oxygen containing groups, which were able to react with CMC and its cross-linking agent, citric acid.

Chemical elemental analysis confirmed that the CMC was successfully cross-linked to the PET surface. After a certain level of CMC concentration, the cross-linking efficiency was not advanced further, and the morphology of the polyester fiber surface did not appear to change. In terms of the antimicrobial performance neither the CMC chemically cross-linked treatment nor the application of a physical coating displayed any positive antibacterial performance against E. coli or S. aureus strains of bacteria. And the mechanical properties of both groups, as measured in terms of their bursting strength, were reduced as a result of the chemical grafting and crosslinking, and the physical coating.
Polycarbonate urethanes (PCUs) are gaining in popularity in many biomedical applications due to their low stiffness, biocompatibility, and high strength. As a thermoplastic, PCUs can be 3D printed through a process known as fused deposition modeling (FDM). FDM offers many benefits such as: cheaper, faster, and easier prototyping, custom or complex geometries and architectures, and potentially even final component or device manufacturing. However, such processing will have implications on the material microstructure and ultimately the mechanical properties. Specifically, the strength between the layers (weld strength) and fatigue behavior. Weld strength exposes the weakest point of a printed part while fatigue evaluates how many cycles to failure under certain strains. The purpose of this study was to assess the effects and viability of FDM with PCU in an effort to realize new opportunities in the biomedical field for this promising soft material. Test samples were formed through either injection molding (IM) of pellets or extrusion into filament and then processing via FDM. Mechanical tests included monotonic tension and tensile fatigue. This study demonstrated the effectiveness of FDM as a processing method for PCU based on the performance of FDM samples. The results are both unexpected and significant, as they indicate potential for printing soft devices while maintaining a high level of performance, including in fatigue. This is promising for biomedical applications where custom geometries or complex architectures utilizing a soft, biocompatible material are desired. Potential applications are numerous, ranging from transvaginal meshes to custom, composite osteochondral devices.
Designing an Antimicrobial Knitted Spacer Fabric with Chitosan for Vacuum Assisted Wound Closure

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Open wounds, such as venous ulcers, burn injuries, exposed joints and skin graft donor sites, require wound dressings that protect the wound from blood loss, infection and further trauma, as well as maintain the appropriate level of moisture so as to promote wound healing [1]. Previously, on account of its high flexibility and its ability to cause blood coagulation, we have promoted the use of a 3D spacer fabric knitted from polyester (PET) yarns to serve as a dressing for vacuum assisted wound closure (VAC) therapy. However, the addition of an antimicrobial finish would enhance the prevention and management of wound infections compared to regular polyester materials. Generally, antimicrobial activity at the fiber’s surface can be achieved by applying an appropriate coating or chemical treatment like chitosan. Chitosan, which is highly biocompatible, biodegradable, non-toxic, non-immunogenic and exhibits antibacterial properties, can form a platform to stimulate the synthesis of collagen and promotes fibroblast growth. [2] The primary purpose of this study was to evaluate the mechanical properties and the antibacterial performance of a chitosan–coated polyester knitted 3D spacer fabric that is suitable for vacuum assisted wound closure (VAC) therapy.

Chitosan (Dr. Sam Hudson, NCSU, Raleigh, NC.), with an 95.3% degree of deacetylation and a mean molecular weight of 740 kDa, was used to apply a basic finish to the 100 deniers 34 filament nylon 66 yarns (Unifi Inc. Greensboro, NC). The yarns were used to knit a 3-dimensional spacer fabric on a 4 guides bar Ruis Warp knitting machine using a 24 gauges needle bar. Chitosan was dissolved in 5% acetic acid to prepare a 3% solution by weight. The procedure of surface modification was achieved by treating the nylon fabric with a radio frequency plasma (94.6% helium and 5.4% oxygen) and placed in chitosan solution. The mechanical properties were measured in terms of their tensile strength and stiffness and their moisture retention capacity. The antimicrobial performance was observed by challenging the spacer fabric with E.coli and Staphylococcus aureus bacteria according to ASTM E2149 − 13a.

The spacer fabrics knitted from chitosan-coated polyester yarn have similar mechanical properties to those fabrics without the coating. The structure and appearance of the space fabrics is shown in Figure 1. The views of the face and back, as well as the cross sections indicate the range of different structural morphologies included in this study.

Figure 1. Views of face and back (A and B) and cross sections of spacer fabric(C) by optical microscopy and design software(D).
The objective of this study was to evaluate the positional stability and physical properties of stent-grafts and covered stents that have been used in an aortic arch chimney EVAR for a specific patient who had a thoracoabdominal aneurysm.

To reach this objective, an accelerated mechanical fatigue test of 120 million cycles (equal to 3 life-years) was applied to a Cook Zenith stent-graft and an Atrium covered stent that were deployed inside a customized elastic polyurethane phantom using a chimney EVAR approach, and mounted on an Electroforce® accelerated stent-graft fatigue tester. The fabrication of the polyurethane phantoms was based on the DICOM® images of the patient who was treated with chimney EVAR repair. The fatigued phantom with the endovascular devices inside was removed from the mechanical fatigue system at different time intervals to perform computed tomography (CT) scans and endoscopy views to monitor the devices’ size, shape and positional stability. After 120 million cycles, post-fatigue tests including scanning electron microscopy (SEM), bursting strength and fabric total porosity were performed to determine any changes in the physical properties of the stent-graft fabric during fatiguing.

The results confirmed that the shape and position of the proximal end of the chimney covered stent did not move significantly during fatiguing, while the total length and distal position did experience some displacement. While no apparent surface abrasion was observed by SEM, and no significant changes were measured in bursting strength during fatiguing, the total fabric porosity was observed to change depending on its location.

This chimney EVAR approach for this particular patient maintained acceptable positional and dimensional stability based on this in vitro mechanical fatigue study that mimicked a three-year life period. No significant surface abrasion nor mechanical strength loss was observed for the thoracic stent-graft fabric.
NOTE