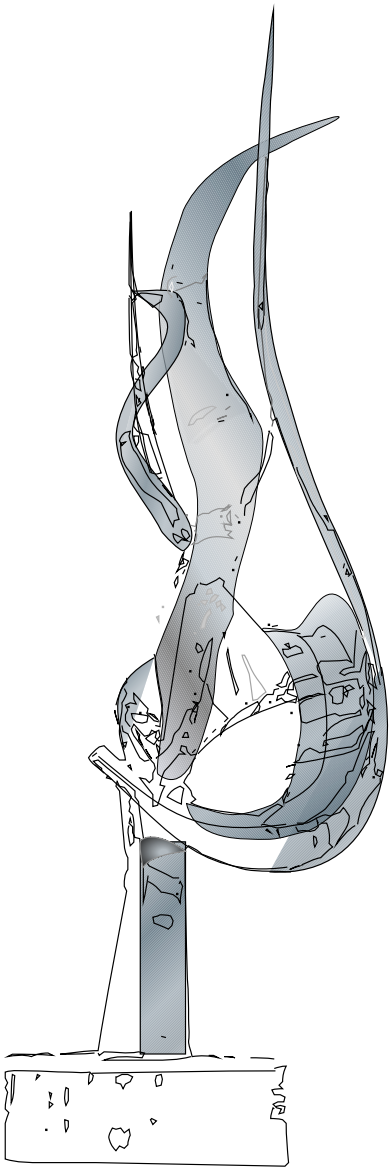
Interdisciplinary Research Collaborative

IRC

Rose-Hulman Institute of Technology



8th Annual IRC Undergraduate Research Symposium

Friday
October 28, 2011

Sponsored by

Edwards Lifesciences

ROSE-HULMAN INSTITUTE OF TECHNOLOGY

Welcome to the

8th Annual IRC Undergraduate Research Symposium

Sponsored by

Friday, October 28, 2011

We are honored to welcome you to the 8th Annual IRC Undergraduate Research Symposium and we sincerely appreciate your participation. The symposium is coordinated by the Interdisciplinary Research Collaborative (IRC), which is supported by funding from Edwards Lifesciences, the Lilly/Guidant Applied Life Sciences Research Center, and Rose-Hulman Institute of Technology. The IRC would like to express its great appreciation for the Symposium sponsorship of Edwards Lifesciences.

The IRC was created to encourage scientific research by undergraduate students and to help them better understand the exciting educational and research opportunities that exist in science and engineering. An appreciation for laboratory research is central to a working understanding of experimental sciences. By participating in research, students add to current knowledge and, furthermore, they enhance their education and broaden their understanding of the scientific method and its application.

Interdisciplinary research is gaining prominence in both academia and industry, as new techniques from one discipline are applied to problems in other disciplines. By acquiring experience in interdisciplinary research, students become more attractive to potential post-graduate programs and employers. The IRC program specifically fosters such interdisciplinary work, and we are pleased to highlight the research of our students, as well as the research of our colleagues in Indiana.

We are delighted to welcome you to this eighth in the annual event series. Our intention in hosting this event is to offer students an opportunity to share their research interests and progress with their colleagues in a nurturing and supportive environment, and to encourage celebration of the undergraduate research experience. We hope you enjoy the dynamic program of speakers.

Mark Brandt
IRC Program Coordinator

Scott McClellan
IRC Program Coordinator

Rose-Hulman Institute of Technology

Symposium Schedule

7:30 AM Registration

Morning Session I (8:00 – 9:30 AM)

Neural Induction of Periodontal Ligament Stem Cells
Yucan Zhao, Herman Cheung, Veronica Fortino and Daniel Pelaez*
GRECC, Miami, Veterans Affairs Medical Center, Department of Biomedical Engineering, University of Miami, Coral Gables, FL, 33146

Carbon Nanotube Fabrication Using Carbon Dioxide
Kellen Stolze, James Folberth, Scott Kirkpatrick, and Elaine Kirkpatrick*
Department of Physics and Optical Engineering, Rose-Hulman Institute of Technology, Terre Haute, IN 47803

The Role of Nitrate Respiration in Chronic Infections of *Ralstonia solanacearum*
Alexandria Williams, Beth Dalsing, Caitilyn Allen*
Department of Plant Pathology, University of Wisconsin-Madison, College of Agriculture and Life Sciences, Madison, WI 53711-2266

Investigation of Polycyclic Aromatic Hydrocarbons in Water-Ecosystems near the Rose-Hulman Campuses
Alex Bledsoe and Justin W. Shearer*
Department of Chemistry & Biochemistry, Rose-Hulman Institute of Technology, Terre Haute, IN 47803

STING Dimerization is Critical for Innate Immune System Surveillance
Hobey Tam, Katia Sotelo-Troha, Dara Burdette, and Russell Vance*
[1]Department of Applied Biology & Biomedical Engineering, Rose-Hulman Institute of Technology, Terre Haute, IN 47803 and [2]Molecular Cellular Department, University of California, Berkeley.

Food Preservation in a Zeer Refrigeration System
Stephanie Biecker, Katelyn Stenger, and Sister Mary Ethel Parrott*
Notre Dame Academy

Morning Session II (9:45 – 11:30 AM)

Characterization and Comparison of Cancer Stem Cells in Human and Dog Glioblastoma Cell Lines
Thomas Clements, Jessamine Osborne, Craig Barcus , Jenna Rickus, and Kari Clase*
Department of Chemistry, Purdue University, West Lafayette IN 47906

Finite-Element Modeling of FRP-Timber Bond Performance
Chris Ohslund[1], Jacob Slifer[2]*, Richard Onyancha[1], and John Aidoo[2]*
[1]Department of Mechanical Engineering and [2]Department of Civil Engineering, Rose-Hulman Institute of Technology, Terre Haute, IN 47803

Direct Interaction with an Assistive Robot for Individuals with Chronic Stroke

Heather Markham, Brandon Kmetz, Bambi Brewer*
Department of Electrical & Computer Engineering, Rose-Hulman Institute of Technology, Terre Haute, IN 47803

Design and construction of a testing device for the evaluation of the effect of loading orientation on the structural properties of the anterior cruciate ligament

Cody Austin and Glen Livesay*
Department of Applied Biology & Biomedical Engineering, Rose-Hulman Institute of Technology, Terre Haute, IN, 47803

Zearalenone's Effects on Breast Cancer

Alexander Krug and Ross Weatherman*
Department of Chemistry & Biochemistry, Rose-Hulman Institute of Technology, Terre Haute, IN 47803

Probing Speciation of Selenium Dioxide and Sodium Selenite as a Function of pH: Understanding Metal-Ion Binding by Selenium Compounds and its Role in Antioxidant Activity.

Robert French[1,2] and Daniel Morris, Jr.[2]*
[1]Department of Chemical Engineering and [2]Department of Chemistry & Biochemistry, Rose-Hulman Institute of Technology, Terre Haute, IN 47803

Investigating the effect of Selenium Dioxide and Sodium Selenite on the formation of 8-OH-dG via Oxidative DNA damage and developing a Liquid Chromatography- Mass Spectrum Method as a means of Analysis.

William Hart and Daniel Morris*
Department of Chemistry & Biochemistry, Rose-Hulman Institute of Technology, Terre Haute, IN 47803

Afternoon Session (2:15 – 5:15 PM)

Investigating the Baking Process

Andrew Harris and David Finn*
Department of Mathematics, Rose-Hulman Institute of Technology, Terre Haute, IN 47803

Synthesis and Characterization of Tamoxifen Conjugates

Paul Himes and Ross Weatherman*
Department of Chemistry & Biochemistry, Rose-Hulman Institute of Technology, Terre Haute, IN 47803

Astaxanthin Production and Extraction in *Synechocystis* sp. PCC 6803

James Jeffryes[1], Stevan Albers[2], and Christie Peebles[2,3]*
[1]Department of Chemical Engineering, Rose-Hulman Institute of Technology, Terre Haute, IN 47803, [2]Cell and Molecular Biology Graduate Program, Colorado State University, and [3]Department of Chemical and Biological Engineering, Colorado State University

Characterization of the Effects of SeO_2 and Na_2SeO_3 on the Interactions Between Metal Ions and DNA by Molecular Absorption Spectroscopy

Steve Marczak and Daniel L. Morris, Jr.*
Department of Chemistry & Biochemistry, Rose-Hulman Institute of Technology Terre Haute, IN 47803

Determination of a Standard Test Method for Assessing FRP-to-Timber Bond Characteristics

Jacob Slifer[1] Chris Ohslund[2]*, Richard Onyancha[2], and John Aidoo[1]*
[1]Department of Civil Engineering and [2]Department of Mechanical Engineering, Rose-Hulman Institute of Technology

Cyclic Voltametric Characterization of Carbon Cryogel Electrodes with Multiple Systems

Zhang Wang[1] and Justin W. Shearer[2]*
[1]Department of Chemical Engineering and [2]Department of Chemistry & Biochemistry, Rose-Hulman Institute of Technology, Terre Haute, IN 47803

Detection of Phytoplasma in *Trillium grandiflorum* using 16s rDNA PCR

Nathan D. Wheeler and J. Peter Coppinger*
Department of Applied Biology & Biomedical Engineering, Rose-Hulman Institute of Technology, Terre Haute, IN 47803

Preparation of Temperature and pH-sensitive micelles through Formation of poly(N-isopropylacrylamide)-b-poly(D,L-lactide-co-glycolide)

Durushka Ahmed and Scott McClellan
Department of Chemical Engineering, Rose-Hulman Institute of Technology, Terre Haute, IN 47803

Comparison of Constant Strain vs. Constant Stress in Rat Tail Tendon

Neil Dorsey, Brianne Widmoyer, Cody Austin, and Glen Livesay*
Department of Applied Biology & Biomedical Engineering, Rose-Hulman Institute of Technology, Terre Haute, IN 47803

The Effects of Catalysts on the Mesoporosity of Carbon Cryogel and the Atom Economy of its Synthesis

Caitlin Anderson, Rebecca DeVasher, and Justin W. Shearer*
Department of Chemistry & Biochemistry, Rose-Hulman Institute of Technology, Terre Haute, IN, 47803

Expression of the Ligand Binding Domain of the Estrogen Receptor-Beta

Melissa Galey and Mark E. Brandt*
Department of Chemistry & Biochemistry, Rose-Hulman Institute of Technology, Terre Haute, IN 47803

All Nanoparticles Spray-Layer-by-Layer Assembly

Yile Gu[1], Will Mulhearn[2], David Lee[3], and Daeyeon Lee[2]*
[1]Department of Chemical Engineering, Rose-Hulman Institute of Technology, Terre Haute, IN 47803, [2]Department of Chemical & Biomolecular Engineering and [3]Department of Mechanical Engineering & Applied Mechanics, University of Pennsylvania, Philadelphia, PA 19104

Poster Session (11:30 AM – 12:30 PM)

Targeting and Trafficking of Quantum Dots

Alex Cochrane[*1], Amanda Riddle[2], and Ian Schneider[2]*

[1]Department of Applied Biology & Biomedical Engineering, Rose-Hulman Institute of Technology, Terre Haute, IN, 47803 and [2]Department of Chemical & Biological Engineering, Iowa State University, Ames, IA 50100

Fabrication of Self-Assembled Polystyrene Nanosphere Monolayers

James Folberth, Kellen Stolze, Scott Kirkpatrick, and Elaine Kirkpatrick*

Department of Physics and Optical Engineering, Rose-Hulman Institute of Technology, Terre Haute, IN 47803

A Simple Algorithm to Relate Surface Roughness to Equivalent Sand Grain Roughness

*Christopher Grant**

Department of Mechanical Engineering, Department of Physics Rose-Hulman Institute of Technology, Terre Haute, IN 47803

Incremental Stress Relaxation

Karah Hickman, Cody Austin, Neil Dorsey, Brianne Widmoyer, and Glen Livesay*

Department of Applied Biology & Biomedical Engineering, Rose-Hulman Institute of Technology, Terre Haute, IN 47803

Analysis of Proximal Tibia Strain Response to Sagittal Alignment in Partial Knee Arthroplasty Tibial Components

Amanda L. Kingman[*1], Derek B. Archer[1], Kelsey R. Hughes[1], Renee D. Rogge[1,2], Scott R. Small[2], and Michael E. Berend[2]*

[1]Rose-Hulman Institute of Technology, Terre Haute, IN 47803, 2Joint Replacement Surgeons of Indiana Foundation, Mooresville, IN

Quantitative study of how the cross-sectional area and cell density of amoebocytes recovers in horseshoe crabs after bleeding 40-50% of the animal's blood volume

Jillian R. Hufgard and William W. Weiner

Department of Applied Biology & Biomedical Engineering, Rose-Hulman Institute of Technology, Terre Haute, IN 47803

The Recovery of Viscoelastic Properties in Creep of Rat Tail Tendon Fibers

Brianne Widmoyer, Neil Dorsey, Cody Austin, and Glen Livesay*

Department of Applied Biology & Biomedical Engineering, Rose-Hulman Institute of Technology, Terre Haute, IN 47803

Separation and detection of nanoparticles using capillary electrophoresis with laser light scattering detection

Kabir Sodhi and Luanne Tilstra*

Department of Chemistry and Biochemistry, Rose-Hulman Institute of Technology, Terre Haute, Indiana 47803

Neural Induction of Periodontal Ligament Stem Cells

Yucan Zhao*, Herman Cheung, Veronica Fortino and Daniel Pelaez
GRECC, Miami, Veterans Affairs Medical Center, Department of Biomedical Engineering, University of Miami, Coral Gables, FL, 33146

To set the groundwork for future therapeutic transplantation of periodontal ligament (PDL) stem cells for the treatment of DPN, finding an optimal protocol for the induction of PDL cells into neural-like cells is the first step. In this project, PDL stem cells were treated with epidermal growth factor (EGF) and basic fibroblast growth factor (bFGF) for ten days. Neurogenic gene expression of the cells was checked using qPCR and immunohistochemistry techniques and it was found that our protocol significantly increases the expression of neurogenic genes.
Morphologically, the EGF-bFGF treated PDL cells have more rigid and compact cell bodies than the control cells, in addition to having longer and thinner processes that extend to nearby cells. Statistically significant increased expression of Nestin and βIII-tubulin (TUBB3) were observed in the EGF-bFGF treated PDL cells. Immunohistochemistry techniques confirm elevated TUBB3 expression of the neuro-induced PDL stem cells. EGF-bFGF treated PDL cells showed significant increase of TrkA expression. Gene expression of IGF-I is elevated but not to statistical significance. In a word, the EGF-bFGF treatment is successful in converting PDL stem cells into neural-like cells.

Carbon Nanotube Fabrication Using Carbon Dioxide

Kellen Stolze*, James Folberth, Scott Kirkpatrick, and Elaine Kirkpatrick
Department of Physics and Optical Engineering, Rose-Hulman Institute of Technology, Terre Haute, IN 47803

Carbon nanotubes, CNTs, have received interest in several fields for their unique properties. The thermal, mechanical, electrical, and structural properties of CNTs have found applications in electronics, optics, nanotechnology, and other areas of material science. There are several ways to fabricate CNTs including chemical-vapor deposition (CVD). Currently most CNTs fabricated by CVD use a hydrocarbon, usually either methane or acetylene. We have developed a procedure which uses CO_2. By using CO_2 the driving energy to formation is smaller which reduces the amount of amorphous carbon produced.

The Role of Nitrate Respiration in Chronic Infections of *Ralstonia solanacearum*

Alexandria Williams*, Beth Dalsing, Caitilyn Allen
Department of Plant Pathology, University of Wisconsin-Madison, College of Agriculture and Life Sciences, Madison, WI 53711-2266

Ralstonia solanacearum is a plant pathogenic bacterium that causes bacterial wilt, a fatal occlusion of xylem vessels. It is known that *R. solanacearum* can respire in vitro on nitrate and it is hypothesized that this metabolism is required for biofilm formation and long-term survival in host xylem vessels. A strain lacking *narG*, a gene predicted to encode an indispensible piece of the nitrate reductase enzyme that confers the ability to use nitrate as a terminal electron acceptor, was created and was unable to form biofilms *in vitro*. Using this Δ*narG* strain, anaerobic growth was monitored in vitro and it was confirmed that *narG* is needed for anaerobic growth with nitrate. Then, plants were inoculated with the Δ*narG* and wild type strains to assess the ability of each to grow in the xylem. It was found that although *narG* is needed for anaerobic growth with nitrate, the gene is not needed for xylem colonization or for virulence.

Investigation of Polycyclic Aromatic Hydrocarbons in Water-Ecosystems near the Rose-Hulman Campuses

Alex Bledsoe* and Justin W. Shearer
Department of Chemistry & Biochemistry, Rose-Hulman Institute of Technology, Terre Haute, IN 47803

Polycyclic aromatic hydrocarbons (PAH's) that are released from combustion reactions from automobiles and commercial trucks are of environmental concern because of the health problems they cause. The waterways near the Rose campuses are of keen interest due to their proximity to a major commercial roadway, Interstate 70. Current research to analyze water samples from near the main campus of Rose-Hulman and Rose Hulman Ventures. Six specified PAH's (Anthracene, Phenanthrene, Pyrene, Benzo(a)pyrene, Fluorene, and Fluoranthrene), that are often found in water ecosystems, were quantified using gas chromatography with mass spectrometric detection. A direct aqueous injection method was developed to permit injection of samples with minimal sample preparation. Solid phase extraction using octadecysilane was employed as a sample clean-up/pre-concentration technique.

This research was funded in part by a Joseph B. and Reba A. Weaver Undergraduate Research Award.

STING Dimerization is Critical for Innate Immune System Surveillance

Hobey Tam[1], Katia Sotelo-Troha[2], Dara Burdette[2], and Russell Vance[2]
[1]Department of Applied Biology & Biomedical Engineering, Rose-Hulman Institute of Technology, Terre Haute, IN 47803, and [2]Molecular Cellular Department, University of California, Berkeley.

Pathogen-sensing by the innate immune system enables the body to react to a broad range of harmful foreign invaders with high specificity. Cells use different sensors ranging from surface membrane bound proteins to proteins in the cytosol to detect a broad range of compounds. We are interested in cytosolic surveillance pathways that sense foreign nucleic acids. Previous experiments in the Vance lab suggest that in addition to DNA, a bacterial nucleic acid, 3´,5´-cyclic di-guanosine monophosphate (c-di-GMP), is sensed in the cytosol leading to the production of IFN. Many pathogenic bacteria produce c-di-GMP but mammalian cells do not, making c-di-GMP an indicator of a foreign invasion. The c-di-GMP response requires the same signaling components as the response to DNA, such as the adaptor STING, the kinase TBK1 and the transcription factor IRF3. Our research suggests STING is the cytosolic sensor for c-di- GMP, but not DNA, although STING is critical for both sensing pathways. We are interested in how the cytosolic surveillance pathway discriminates between DNA and c-di-GMP. The literature suggests STING dimerizes and we are testing the hypothesis that dimerization is a mechanism by which cells discriminate between DNA and c-di-GMP. We are using immunoprecipitation followed by SDS-PAGE to test previously generated STING mutants for their ability to dimerize. We are also using immunofluorescence to determine STING subcellular localization in response to c-di-GMP and DNA stimulation. Our results indicate dimerization is critical for STING function. Understanding STING function will aid in developing better immunotherapies and vaccine adjuvants.

Food Preservation in a Zeer Refrigeration System

Stephanie Biecker, Katelyn Stenger*, and Sister Mary Ethel Parrott
Notre Dame Academy

This research studies the structural integrity of tomato peel and the antioxidant content of tomatoes when refrigerated in a zeer system as compared to tomatoes exposed to warm air outside the system. A zeer system with rain garden soil as the fill layer was constructed. Tomatoes were placed in the center of the refrigeration chamber with probes recording temperature. In four runs, average temperatures were about 3°C lower when under refrigeration. The firmness of the tomato peels was tested by force sensor. Zeer-refrigerated tomatoes required far more force to break than did control tomatoes. The average for 96 measurements was 1.22 N in the refrigerated samples and only 0.56 N in the air. The ascorbic acid content was determined by titration with DCPIP. Zeer-refrigerated tomatoes had more than double the ascorbic acid content of the control. The zeer system shows great promise for food storage in a developing country.

Characterization and Comparison of Cancer Stem Cells in Human and Dog Glioblastoma Cell Lines

Thomas Clements*, Jessamine Osborne, Craig Barcus , Jenna Rickus, and Kari Clase
Department of Chemistry, Purdue University, West Lafayette IN 47906

Glioblastomas (GBMs) are among the most common and most malignant forms of primary brain tumors that occur in humans. Despite advances in numerous treatment methods such as chemotherapy and radiation techniques, upwards of 75% of all individuals diagnosed with this disease die within two years of their initial treatment. A possible explanation of this circumstance is the presence of a "cancer stem-like cell" (also coined cancer stem cell or CSC), which are resistant to the initial treatments, and thus allow the tumor to continue to proliferate throughout the affected individuals lifetime.

GBMs are not only present in humans, but they also exist and are extremely devastating in the canine population. This fact allows scientists the possibility to make a direct comparison of the two systems. A direct comparison could prove to be quite beneficial because cancers in both organisms share many characteristics such as physical appearance, genetics, and even therapeutic response. The perspective gained from these comparisons could prove to be invaluable because the usual rodent model for comparison does not share many of these similarities.

Specifically, this research is oriented around identifying the cancer stem cell (CSC) in glioblastoma (GBM) and reducing its stem-like quality, that is, preventing these cells from proliferating once they have been exposed to various levels of treatment.

Perhaps the best method to identify the CSC is through the detection of one its putative protein markers. These markers include CD 133, Nestin, BMI 1, and OLIG 2. Of these, none of which yield conclusive results, yet CD 133 has been studied thoroughly and will be used in this study. Also, the protein Sox-2 will be used as a potential marker as well because of its use as a neural stem cell marker.

One of the theorized ways to accomplish this is to inhibit the pathways that are up-regulated in GBM. In order to discover which pathways' activity are most increased, proteomic analysis on the GBM cell lines were performed. The two pathways that exhibited the most increased activity is Notch, which has been shown to promote tumor growth when in its up-regulated state and is also related to neural stem cell survival and TGF-B, which has the ability to promote the migration and proliferation of tumor cells in late-stage metastatic cancer. These pathways can be inhibited through the use of DAPT and Rep-Sox respectively. Once the pathways have been inhibited, the cells can be tested for the CD 133 and Sox 2 protein in order to potentially quantify the level of "stemness" that the cells possess. This is accomplished through the use of numerous Western Blots incorporating the antibodies for the markers.

Finite-Element Modeling of FRP-Timber Bond Performance

Chris Ohslund[1]*, Jacob Slifer[2]*, Richard Onyancha[1], and John Aidoo[2]
[1]Department of Mechanical Engineering and [2]Department of Civil Engineering, Rose-Hulman Institute of Technology, Terre Haute, IN 47803

One of the key areas of innovation in civil engineering is the matter of how the performance of common structural materials can be improved. One such method entails coupling a plate of carbon fiber reinforced polymer (CFRP) to an existing beam by means of an epoxy resin; this has the effect of strengthening and stiffening the beam and reducing the effects of damage. The effects of CFRP reinforcement on materials like steel and concrete are widely understood and the practice is common; data on how the addition of CFRP plates affects timber beams, however, is scarcer. This practice offers a great deal of potential, allowing existing wooden structures to be quickly and easily repaired and without requiring extensive renovation of the structure. The inherently variable properties of wood, however, complicate calculation and restrict the use of this process. This paper outlines efforts to model such behavior; it details and analyzes the results of three-point bending tests applied to timber beams to which FRP plates of different widths are bonded, and contrasts the performance of experimental beams with models constructed using general purpose finite-element software. The finite-element modeling method was found to be sufficiently accurate for general use; the load-strain curves constructed from model data differed in slope from those constructed from strain gage data by a maximum of 20%. The finite element model was then used to investigate the effect of notch depth on the response of the beam; strain at the notch base was found to increase with notch size only to a notch depth of 1.66 in, approximately the point at which the ratio of the second moment of area of the beam to the distance between the edge and neutral axis of the beam was maximized, and decreased as the notch was further enlarged.

Direct Interaction with an Assistive Robot for Individuals with Chronic Stroke

Heather Markham, Brandon Kmetz*, Bambi Brewer
Department of Electrical & Computer Engineering, Rose-Hulman Institute of Technology, Terre Haute, IN 47803

Many robotic systems have been developed to provide assistance to individuals with disabilities. Most of these systems require the individual to interact with the robot via a joystick or keypad, though some utilize techniques such as speech recognition or selection of objects with a laser pointer. In this paper, we describe a prototype system using a novel method of interaction with an assistive robot. A touch-sensitive skin enables the user to directly guide a robotic arm to a desired position. When the skin is released, the robot remains fixed in position. The target population for this system is individuals with hemiparesis due to chronic stroke. The system can be used as a substitute for the paretic arm and hand in bimanual tasks such as holding a jar while removing the lid. This paper describes the hardware and software of the prototype system, which includes a robotic arm, the touch-sensitive skin, a hook-style prehensor, and weight compensation and speech recognition software.

Design and construction of a testing device for the evaluation of the effect of loading orientation on the structural properties of the anterior cruciate ligament

Cody Austin* and Glen Livesay
Department of Applied Biology & Biomedical Engineering, Rose-Hulman Institute of Technology, Terre Haute, IN, 47803

The anterior cruciate ligament (ACL) has a poor capacity to heal and is the most commonly injured ligament in the knee. The low healing capabilities of the ACL coupled with the high frequency of ACL injuries leads to roughly 200,000 ACL reconstructions each year. There are many complications with current ACL reconstructions, namely osteoarthritis, poor stability, and improper knee kinematics. Tissue engineered grafts are an alternative to current reconstruction methods. In order for the tissue engineered grafts to be appropriate, their structural properties must be similar to those of the ACL. The structural properties of the ACL are typically only reported for uniaxial orientations; however, it has been shown that the loading orientation has an effect on the structural properties. Since the structural properties of the ACL are dependent on the loading orientation, the tissue engineered graft should have similar structural properties for all load orientations. Currently, the evaluation of the structural properties of the ACL is incomplete because only medial-lateral and anterior-posterior changes in orientation have been evaluated, and for these orientations only stiffness was reported, providing no information on the how the length of the toe region varies. A testing device was developed that allows the orientation of the ACL to be adjusted about the medial-lateral axis in 2.5° increments, about the anterior-posterior axis in 2.5° increments, and about the longitudinal axis in 5° increments, and any combination of the three motions. This testing device will allow the structural properties (both stiffness and length of the toe region) of the ACL to be determined for several different loading orientations which can assist in ensuring that tissue engineered grafts are mechanically appropriate.

Zearalenone's Effects on Breast Cancer

Alexander Krug* and Ross Weatherman
Department of Chemistry & Biochemistry, Rose-Hulman Institute of Technology, Terre Haute, IN 47803

Zearalenone, a mycotoxin from the fungus Fusarium roseum has been identified to exhibit estrogen-like behaviors in a variety of mammalian livestock, cows and pigs. This mycotoxin's effects on humans, has been the subject of investigation by the Environmental Protection Agency. Here we describe and characterize the potency and affects that zearalenone had upon a particular branch of human breast cancer cells. We also demonstrate the affinity that zearalenone has for the different types of estrogen receptor.

This research was funded in part by a grant from the National Institutes of Health.

Probing Speciation of Selenium Dioxide and Sodium Selenite as a Function of pH: Understanding Metal-Ion Binding by Selenium Compounds and its Role in Antioxidant Activity.

Robert French*[1,2] and Daniel Morris, Jr.[2]
[1]Department of Chemical Engineering and [2]Department of Chemistry & Biochemistry, Rose-Hulman Institute of Technology, Terre Haute, IN 47803

Oxidative damage to DNA is associated with cancer, aging, and a host of other diseases and clinical conditions. Metal ions are known to bind to both phosphate groups and individual bases in DNA, and comparing production of the accepted oxidative DNA damage marker 8-hydroxy-2'-deoxyguanosine (8-OH-dG) to the levels of unmodified nucleosides (specifically deoxyguanosine (dG)) provides insight into the degree of site-specific damage. It is accepted that formation of 8-OH-dG results from reactive oxygen species (ROS) generated close to the guanine base while generalized base damage is most likely produced by ROS generated in bulk solution. Selenium compounds are known to exhibit both anti- and pro-oxidant behavior, and recent work suggests that metal ion coordination is a critical part of the mechanism by which inorganic selenium compounds inhibit metal-ion mediated oxidative DNA damage in the presence of hydrogen peroxide. Selenium dioxide (SeO_2) and sodium selenite (Na_2SeO_3) show different antioxidant activity at physiological pH despite predictions that they would exist as the same species ($HSeO_3^-$) at pH 7.0. We present results of a pH-dependent Raman study of SeO_2 and SeO_3^{-2}, as well as a discussion of the experimental protocols that gave the initial results. Our results demonstrate that the two selenium compounds exhibit different antioxidant activity based on metal identity despite the fact that Raman spectroscopy indicates that they are structurally similar at pH 7.0.

Investigating the effect of Selenium Dioxide and Sodium Selenite on the formation of 8-OH-dG via Oxidative DNA damage and developing a Liquid Chromatography- Mass Spectrum Method as a means of Analysis.

William Hart* and Daniel Morris, Jr.
Department of Chemistry & Biochemistry, Rose-Hulman Institute of Technology, Terre Haute, IN 47803

Oxidative DNA damage occurs when essential transition metal ions, including Fe(II), Cu(II) and Cr(III), form reactive oxygen species (ROS) through the Fenton reaction or "Fenton-like" reactions when they come in contact with hydrogen peroxide, a by-product of cell function. The ROS attack DNA at very specific sites causing DNA strand scission and/or chemical modifications that cause problems with replication of DNA in cells [1]. The effect of oxidative DNA damage is linked to many diseases, such as atherosclerosis, Parkinson's disease and Alzheimer's disease, as well as the aging process [2]. Previous research has shown that Selenium Dioxide and Sodium Selenite have an effect on oxidative DNA damage caused by Fe(II) and that these compounds ability to act as anti-oxidants is tied to their ability to complex with the metal ion. For these reasons, we investigated the effect of Selenium Dioxide and Sodium Selenite on their ability to protect Calf thymus DNA from oxidative DNA damage caused by Fe(II), Cu(II), and Cr(III) ions. We investigated the effect these two compounds had by using high performance liquid chromatography (HPLC) to separate our reaction mixtures of digested DNA and used an electro-chemical detector (ECD) and UV-Vis Spectrophotometer as a means of detection. We looked at two factors to compare the effect of these compounds, the amount of deoxyguanosine (dG) and the amount of 8-hydroxy-deoxyguanosine (8-OH-dG) remaining after oxidative DNA damage. We will present this data as a the ratio of 8-Oh-dG to dG remaining, to illustrate how well the selenium compounds effect both site specific damage and generalized damage. In addition to using HPLC-ECD to monitor 8-OH-dG and dG we investigated using liquid chromatography coupled with a mass spectrometer (LC-MS) as a means of detection and identification of the products of oxidative DNA damage with the goal of better understanding the mechanism of how selenium dioxide and sodium selenite affect oxidative DNA damage. This led us to develop and optimize an LC-MS method, and investigate the two ionization process of LC-MS, electrospray ionization (ESI) and atmospheric pressure chemical ionization (APCI). Our research is, to our knowledge, the first time someone has tested the effect of SeO_2 and SeO_3^{2-} on oxidative DNA damage mediated by Cu(II) and Cr(III). Our results can hopeful allow us to gain insight into the differences between the oxidative damage caused by these metal ions and the mechanism by which SeO_2 and SeO_3^{-2} affect oxidative DNA damage.

This research was funded in part by a Joseph B. and Reba A. Weaver Undergraduate Research Award.

Investigating the Baking Process

Andrew Harris* and David Finn
Department of Mathematics, Rose-Hulman Institute of Technology, Terre Haute, IN 47803

A mathematical model for the baking process will be presented. This model is primarily for baking a cake or bread. The model incorporates the temperature distribution, the moisture content, the vapor content, and the deformation induced during the baking process. The resulting model is a system of differential equations. We start the model with an oversimplified situation and add complexity, using basic differential equations, some thermodynamics (evaporation/enthalpy/pressure), numerical methods, and some continuum mechanics. Only basic knowledge of differential equations is needed to understand this presentation.

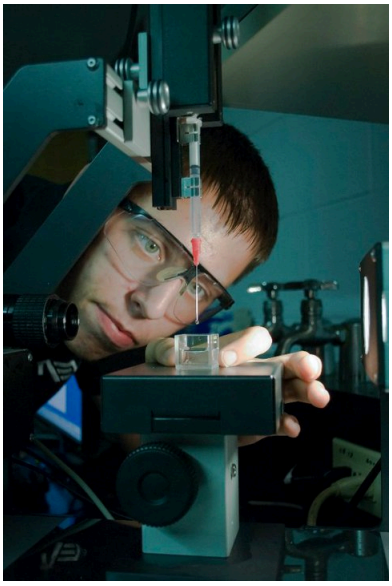
This research was funded in part by Edwards Lifesciences under the auspices of the IRC, and by support from the Departments of Mathematics and Physics & Optical Engineering, Rose-Hulman Institute of Technology.

Synthesis and Characterization of Tamoxifen Conjugates

Paul Himes* and Ross Weatherman
Department of Chemistry & Biochemistry, Rose-Hulman Institute of Technology, Terre Haute, IN 47803

Tamoxifen is a widely used chemo-preventive drug course in dealing with breast cancer. It does this because Tamoxifen is a Selective Estrogen Receptor Modulator (SERM) and by being this, it interferes with the estrogen stimulation growth of breast tumors. More than fifty percent of the tumors will respond, but tamoxifen acts as an estrogen in other tissues which causes problems and breast tumors can develop resistance to tamoxifen. Derivatives can provide a solution to this problem by changing how the drug acts with the receptor. Attaching tamoxifen conjugates to a polymer backbone is shown to increase potency against tamoxifen-resistant breast cancer. It is thought to do this in three ways: the scaffold interacts with the estrogen receptor, the scaffold enables better delivery of the drug, or the scaffold extends the half-life of the drug within the cell. Derivatives of tamoxifen were synthesized using different chemistry techniques and then were tested biologically in MCF7 cells breast cancer cell lines. Starting with 4-hydroxytamoxifen (a more potent form of tamoxifen), different linkers were added that were intended to attach to different macromolecular scaffolds. Three compounds were synthesized and tested in the MCF7 cells. OHT-6C, OHT-6C-MB, and END-5C-OH were tested to see if their potency as an anti-estrogen in the breast was increased or retained. If the potency was retained or increased the drug conjugate was then attached to a polymer backbone and tested again. While the OHT-6C-MB was found to not have significant potency for reasons that are still not clear, both OHT-6C and END-5C-OH showed sub-micromolar potency and promise as ligands for attachment to polymers and dendrimers.

This research was funded in part by Edwards Lifesciences under the auspices of the IRC and by an Eli Lilly Undergraduate Research Grant.



Astaxanthin Production and Extraction in *Synechocystis* sp. PCC 6803

James Jeffryes*[1], Stevan Albers[2], and Christie Peebles[2,3]
[1]Department of Chemical Engineering, Rose-Hulman Institute of Technology, Terre Haute, IN 47803, [2]Cell and Molecular Biology Graduate Program, Colorado State University, and [3]Department of Chemical and Biological Engineering, Colorado State University

Astaxanthin is a natural pigment and antioxidant with significant commercial demand. Its red color makes it a potential reporter molecule for future engineering of the terpenoid pathway. Synechocystis sp. PCC 6803, a model cyanobacterium, could be utilized as a sustainable production platform for astaxanthin and other commercially important compounds. Synechocystis is very amenable to genetic manipulation but lacks the ability to produce astaxanthin due to lack of a di-ketolase. The design of an astaxanthin production construct for Synechocystis will be discussed. The majority of the production construct has been built into E. coli puc19 plasmid as well as a second plasmid providing selective pressure for homologous recombination. In parallel, a method for extraction of carotenoids using ultrasonication was adapted from a protocol for red yeasts which will allow quantification of metabolic products by HPLC.

Characterization of the Effects of SeO_2 and Na_2SeO_3 on the Interactions Between Metal Ions and DNA by Molecular Absorption Spectroscopy

Steve Marczak* and Daniel L. Morris, Jr.
Department of Chemistry & Biochemistry, Rose-Hulman Institute of Technology
Terre Haute, IN 47803

The transition metal ions Fe(II), Cu(II), and Cr(III) undergo reactions with H_2O_2 to produce reactive oxygen species that give rise to damage associated with many disease, clinical conditions, and aging. These metal ions produce oxidative DNA damage in a site-specific manner that is related to whether they bind to DNA through bases and/or the phosphate backbone. Selenium is considered an essential dietary trace element. The inorganic selenium compounds, selenium dioxide (SeO_2) and sodium selenite (SeO_3^{2-}), have been shown to have antioxidant properties by interfering with the binding between metal ions and DNA. The effects of SeO_2 and Na_2SeO_3 on the binding between Cr(III) and Cu(II) and DNA were studied by visible absorption spectroscopy. The spectra indicate that Cr(III) and Cu(II) both form complexes with SeO_2 and with SeO_3^{2-} in solution. Similar spectra are observed when SeO_2 and SeO_3^{2-} are introduced into solution after Cr(III) and Cu(II) are already bound to DNA. We suggest the possibility of formation of a metal ion coordination complex with the selenium compounds. This mechanism could be responsible for the antioxidant properties of SeO_2 and SeO_3^{2-}.

This research was funded in part by Edwards Lifesciences under the auspices of the IRC.

Determination of a Standard Test Method for Assessing FRP-to-Timber Bond Characteristics

Jacob Slifer[1]* Chris Ohslund[2]*, Richard Onyancha[2], and John Aidoo[1]
[1]Department of Civil Engineering and [2]Department of Mechanical Engineering, Rose-Hulman Institute of Technology

The use of advanced composite materials in civil engineering to increase the structural capacity of members is always being investigated. Compared to steel, these materials are superior in the areas of resistance to electrochemical corrosion, strength to weight ratio, fatigue resistance, and their versatility of use. The utilization of fiber reinforced polymer (FRP) composites for rehabilitation purposes is becoming common practice in civil infrastructure. Like concrete repair methods, FRP-to-timber behavior is dominated by the interface bond. Failures are usually adhesive failures at the FRP-to-timber interface; the FRP debonds from the timber substrate causing the member to immediately lose the extra strength the composites once provided. FRP bond to timber requires a clean and sound substrate. The typical application involves the use of a belt sander prior to bonding the FRP laminate. In this study, FRP-to-timber bond is assessed using a modified three-point bending test. This method was adopted from the ASTM Test D 198: Standard Test Methods of Static Tests of Lumber in Structural Sizes. To initiate debonding the timber beam is notched to simulate a defect. This paper addresses the specific issues associated with standardizing such a test specimen including (a) the width of the FRP relative to the timber substrate width; and (b) the effect of providing an initially unbonded region in the vicinity of the notch. Conclusions provide recommended specimen geometry which satisfies the objectives of such a standardized test.

Cyclic Voltametric Characterization of Carbon Cryogel Electrodes with Multiple Systems

Zhang Wang[1]* and Justin W. Shearer[2]
[1]Department of Chemical Engineering and [2]Department of Chemistry & Biochemistry, Rose-Hulman Institute of Technology, Terre Haute, IN 47803

The primary goal of this research is to develop a carbon cryogel electrode which can adsorb heavy metal ions in water by applying a slight electrical bias. Heavy metals are persistent and accumulate in the environment. Carbon cryogels can be made using a sol-gel reaction between resorcinol and formaldehyde using a basic salt, sodium carbonate, as a catalyst. The carbon was obtained after freeze drying and carbonizing the gel at 800 °C in an inert atmosphere. The current research was focused on testing the ability to characterize some electrochemical properties of carbon cryogels. To accomplish these studies, cyclic voltammetry was used to apply voltage to a standard three-electrode system, in which carbon cyrogel electrode serves as the working electrode, a silver/silver chloride electrode serves as the reference electrode, and bare platinum wire serves as the auxiliary electrode. Metallic and organic oxidation-reduction reactions were studied to determine the electrodes ability to distinguish between adsorption-based and outer-shell REDOX processes. The ability of carbon cryogels to serve as metal-removing electrodes was also investigated using a three-electrode system.

This research was funded in part by Edwards Lifesciences under the auspices of the IRC.

Detection of Phytoplasma in *Trillium grandiflorum* using 16s rDNA PCR

Nathan D. Wheeler* and J. Peter Coppinger
Department of Applied Biology & Biomedical Engineering, Rose-Hulman Institute of Technology, Terre Haute, IN 47803

Trillium Grandiflorum plants in Michigan were found to exhibit symptoms typical of a phytoplasmic infection; using PCR and Gel Electrophoresis the infectious agent has been identified to be in the phytoplasm family. *Trillium Grandiflorum*, often referred to as White Trillium, is a perennial, monocot flower indigenous to the North Eastern portion of the United States. Trillium seeds are typically spread by ants, who consume the elaiosome of the seed and disperse the seeds; thus raising the issue of vector transmission of the infection. Trillium samples collected in Michigan displayed signs of phytoplasmic infection; namely floral virulence, or the transformation of floral parts to leafy green structures. Since no published information could be obtained concerning the identification of the infectious agent in White Trillium, the primary focus of the project has been to identify the organism causing the symptoms. Phytoplasmic infections are usually confined to the phloem tissue of the plant so using the blank DNA Extraction kit, DNA was isolated from frozen Trillium samples; an uninfected plant control was also used. Using PCR, phytoplasmic DNA was amplified using universal primers and Gel electrophoresis confirmed the amplification. The sequenced DNA came up with a 97% match with other phytoplasmic species. New *Trillium grandiflorum* samples were obtained from a new site in northern Michigan that exhibit symptoms similar to the original samples suggesting a related phytoplasmic infection. The same process used with the original samples was applied to the new samples. The resulting sequences obtained were entered into a nucleotide BLAST and the sequenced DNA came up with a 98% match with other phytoplasmic species in the database. The matches for both sites were all dicot species, so this species may be a new phytoplasmic agent or a case of transferal from dicot to monocot infection. All flowers exhibiting symptoms produced a band however 2 out of 3 flowers used as negative controls also produced a band. A possible resistance to the phytoplasma may have developed in the population or symptoms had not presented when the flowers were collected. Ongoing research will be focused on affirming that the phytoplasma present in Trillium tissue are the causal agent of the symptoms presented and obtaining the complete 16s rDNA sequence of the bacteria.

This research was funded in part by a Joseph B. and Reba A. Weaver Undergraduate Research Award and in part by the Edwards Lifesciences under the auspices of the IRC.

Preparation of Temperature and pH-sensitive micelles through Formation of poly(N-isopropylacrylamide)-b-poly(D,L-lactide-co-glycolide)

Durushka Ahmed and Scott McClellan
Department of Chemical Engineering, Rose-Hulman Institute of Technology, Terre Haute, IN 47803

pH-sensitive and temperature-sensitive diblock copolymers, poly(N-isopropylacrylamide)-b-poly(D,L-lactide-co-glycolide) (PNIPAAm-b-PLGA) with different PNIPAAm and PLA contents were synthesized in order to be later employed as micelles used to control delivery of doxorubicin. The prepolymer PNIPAAm was synthesized using a hydrolysis mechanism. This prepolymer was employed in a ring-opening polymerization in order to create the gel-like polymer that will be later used to create micelles. The prepolymer and polymer were verified using various analytical methods (HPLC, UV-Vis and NMR chromatography). These techniques indicated the existence of both polymers as well as the accuracy of the assumed measurements.

This research was funded in part by Edwards Lifesciences under the auspices of the IRC.

Comparison of Constant Strain vs. Constant Stress in Rat Tail Tendon

Neil Dorsey*, Brianne Widmoyer, Cody Austin, and Glen Livesay
Department of Applied Biology & Biomedical Engineering, Rose-Hulman Institute of Technology, Terre Haute, IN 47803

Understanding soft tissue mechanics is a crucial step in developing viable recovery solutions in the medical industry. Specifically, additional information on the viscoelastic behavior of collagen-based elements in the body, such as tendon, can greatly improve modern soft tissue repairs, that are often subjected to long-term loading. In the present work, the creep (increase in deformation under constant load) and stress relaxation (decrease in load while held at a constant length) behavior of rat tail tendon (RTT) were examined, as this material is widely utilized as a model for soft tissues in the human body. The objective of the present work is to determine if the viscoelastic response of RTT under creep can be predicted from a stress-relaxation test (or vice versa). All RTT fiber samples were dissected from the same tail, and frozen until needed the morning of testing. After thawing, each end of the RTT was attached to plexiglas tabs using cyanoacrylate to ensure ease of gripping in the testing machines and uniform loading of the sample. While resting, samples were kept hydrated by spraying with physiological buffered saline (PBS); during testing the samples were hydrated using a controlled drip system. Samples (n = 8) were subjected to tensile creep testing for 20 minutes, allowed 20 minutes to recover while unloaded, and then subjected to a 20-minute, tensile stress relaxation (SR) test. The creep tests were conducted using a custom creep testing device as a laser transducer recorded elongation data, all of which was controlled via MATLAB coding. The SR test was conducted using an Instron materials testing machine. Elongation vs. time (creep) and stress vs. time (SR) data were collected via computer. As a first step towards determining whether creep could be predicted from SR (and vice versa), the curves were analyzed to determine if the viscoelastic responses were similar. One measure of the viscoelastic response involved comparing the time taken for each curve (creep and SR) to reach half of their respective maximum stress/strain, which provides some indication of the rate of viscoelastic response. The other measure for comparison utilized the actual, equilibrium maximum stress/strain value reached in each test, which provides some indication of the overall amount of viscoelastic response. Comparisons were made in paired fashion (within fiber), using a two-tailed t-test with significance set at 0.05. The 'half time' comparison yielded a p-value of 0.925, and the 'equilibrium' comparison yielded a p-value of 0.205. Although this is a first pass comparison, the fact that the p-values are both well above the 0.05 level does indicate that the data do show that the viscoelastic responses are significantly different. Calculations regarding the potential Type II error will be required to determine what strength may be placed behind this 'lack of difference'. Future work will involve developing a viscoelastic model based on the data and assessing whether one type of viscoelastic test can be predicted from the other.

The Effects of Catalysts on the Mesoporosity of Carbon Cryogel and the Atom Economy of its Synthesis

Caitlin Anderson*, Rebecca DeVasher, and Justin W. Shearer
Department of Chemistry & Biochemistry, Rose-Hulman Institute of Technology, Terre Haute, IN, 47803

Carbon Cryogels are unique, porous carbon materials that exhibit high surface area, chemical robustness, and thermal stability. Synthesis of the carbon cryogel material varies in the catalyst used and in the materials used for solvent exchange. These changes alter the porosity of the carbon cryogel and the atom economy of the reaction. Carbon cryogels were synthesized through the polycondensation of resorcinol and formaldehyde in the presence of an alkaline or earth-alkaline carbonate catalyst (Na_2CO_3, K_2CO_3, Li_2CO_3, Cs_2CO_3, $CaCO_3$, $BaCO_3$) followed by solvent exchange with acetone and/or tert-butanol, freeze-drying with liquid nitrogen, and then carbonized at 800 ºC in an inert atmosphere. The synthesized carbon cryogel samples were then characterized using Raman Spectroscopy and Scanning Electron Microscopy (SEM). Another focus of this project was to determine the atom economy and environmental (E) factor for the sol-gel reaction.

This research was funded in part by Edwards Lifesciences under the auspices of the IRC.

Expression of the Ligand Binding Domain of the Estrogen Receptor-Beta

Melissa Galey* and Mark E. Brandt
Department of Chemistry & Biochemistry, Rose-Hulman Institute of Technology, Terre Haute, IN 47803

The Estrogen Receptor is a nuclear receptor that primarily interacts with 17b-estradiol, its ligand, in order to regulate various functions including cell proliferation. In the past, it was thought that only one nuclear receptor responded to estradiol as a ligand but in 1995 a second estrogen receptor was discovered. While much is known about the estrogen receptor-alpha (ER-α), little is understood about the estrogen receptor-beta (ER-β). Therefore, we are attempting to determine both the similarities and differences between the two receptors. Our laboratory has previously carried out experiments concerning the ligand binding domain (LBD) of the ER-α and how small organic compounds affect protein dimerization. In order to perform similar tests on the ER-β, it is necessary to obtain purified ER-β protein. We are therefore constructing a plasmid to express the ER-b LBD in *E. coli*. The DNA encoding the LBD of the ER-β was amplified by polymerase chain reaction (PCR). The PCR primers were designed to incorporate restriction sites to simplify construction: the 5' primer (sequence: *5´ ATATCCCGGG**CGAGTGCGGGAGCTGCTGCTGGACG*** 3') includes a *Sma*I site (underlined); the bold sequence corresponds to the N-terminus of the human ER-β LBD. This primer also resulted in the addition of a glycine codon to the beginning of the LBD DNA sequence. The 3' primer (sequence: *5' CGTGAAGCTT**CA**CTTGCACCCGCGAAGCACGTGGGCATTCAGCATCTC 3')* is used to introduce both a stop codon (reverse and complement shown in bold) and a *Hind*III site (underlined) into the DNA. The resulting PCR product was then digested using restriction enzymes *Sma*I and *Hin*dIII. The digested PCR DNA was then ligated to an expression plasmid, which was digested with *Xmn*I and *Hind*III. The resulting construct was then transformed into competent *E. coli* cells. The colonies obtained were screened using PCR for ER-b DNA. Plasmids yielding PCR products were restriction mapped using a digest with *Ssp*I and *Eco*RI. Plasmids found to be positive by both techniques were sent to MC Labs for sequencing. The future direction of the project is to develop purification methods for the ER-β LBD protein. It will then be possible to perform dimerization assays on the ER-β and compare these to the results seen with the ER-α. The ER-β can further be investigated by testing its ability to form a heterodimer with ER-α in solution. The information gathered can later be used to better understand how these ligand-bound receptors interact in the human body with each other and with other molecules.

This research was funded in part by Edwards Lifesciences under the auspices of the IRC.

All Nanoparticles Spray-Layer-by-Layer Assembly

Yile Gu*[1], Will Mulhearn[2], David Lee[3], and Daeyeon Lee[2]
[1]Department of Chemical Engineering, Rose-Hulman Institute of Technology, Terre Haute, IN 47803, [2]Department of Chemical & Biomolecular Engineering and [3]Department of Mechanical Engineering & Applied Mechanics, University of Pennsylvania, Philadelphia, PA 19104

Layer-by-layer assembly is a recent technique which constructs thin films via alternating deposition of two oppositely charged species. All nanoparticle layer-by-layer coatings exhibit attractive and potentially useful antifogging, antireflection, and self-cleaning functionalities. However, the traditional way to accomplish the coatings, dipped layer-by-layer assembly, requires long processing time, which makes the method undesirable for industrial use. In this research, a pre-programmable automatic spray layer-by-layer assembly machine is built and optimized to make layer-by-layer films with comparable thickness and uniformity compared to traditional dipped method, with much decreased time.

Targeting and Trafficking of Quantum Dots

Alex Cochrane*[1], Amanda Riddle[2], and Ian Schneider[2]
[1]Department of Applied Biology & Biomedical Engineering, Rose-Hulman Institute of Technology, Terre Haute, IN, 47803 and [2]Department of Chemical & Biological Engineering, Iowa State University, Ames, IA 50100

Nanoparticles are powerful materials in the rapidly expanding arsenal of biological imaging techniques. One incredibly promising class of nanoparticles is the Quantum Dot (QD). Advantageous and highly controllable abilities of QDs allow us to use multiple types of QDs in one sample, and target both intracellular and cell surface proteins. These applications were looked upon as a possible candidate for locating cancer metastases in our lab. The ability to conjugate any amino acid, peptide, or fatty acid chain boons its ability as an imager, but better qualitative knowledge of the QD is needed. Endocytosis of QDs is predicted to be a major factor in the amount of light given off during analysis, especially when exclusively targeting the plasma membrane. Preferential binding was also called into question in the form of using plectin1 binding peptide on QDs and tested on Panc1 cancer cells, which express plectin1 on the cell surface. Here, I looked at a few unique properties of cell-QD interaction to better understand cell surface association of QDs. I also have used techniques to inhibit clathrin-coated pit and caveolar formation via incubation with the compounds dynasore, chlorpromazine, a mixture of the two, and at 4°C to better understand at which level QDs are binding to the cell surface versus how much is being endocytosed. Through the use of flow cytometry, the conjugation of different tags on the QD was a determining factor in overall endocytosis where lysine tagged dots showed less overall endocytosis and aminopropandiol showed approximately two fold more endocytosis. Increased binding seen in cysteine dots was shown to be attributed to the free sulfhydryl group making disulfide bonds on the cell surface. Lastly, in a step towards use of the QD as an actual biosensor, plectin1 conjugated cell surfaces showed preferential binding to Panc1 cells. The consequence of this is an increased confidence of QDs as a biosensor and provided evidence of QD's high targeting ability.

Fabrication of Self-Assembled Polystyrene Nanosphere Monolayers

James Folberth*, Kellen Stolze, Scott Kirkpatrick, and Elaine Kirkpatrick
Department of Physics and Optical Engineering, Rose-Hulman Institute of Technology, Terre Haute, IN 47803

The fabrication of uniform, spherical polystyrene (PS) nanospheres is commercially feasible. This has led to many nanosphere self-assembly research projects in the scientific community. In the related literature, self-assembly techniques require anywhere from hours to weeks to yield uniform monolayers.

We have developed a procedure for fabricating PS nanosphere monolayers with long range order that only requires about five minutes. This short wait time could be used for larger production runs, and quicker analysis of our samples. We are also collaborating with a research group that is attempting to use PS nanosphere multilayers in the production of high density magnetic data storage devices.

A Simple Algorithm to Relate Surface Roughness to Equivalent Sand Grain Roughness

Christopher Grant*
Department of Mechanical Engineering, Department of Physics Rose-Hulman Institute of Technology, Terre Haute, IN 47803

As one of the most important resources available in the field of fluid mechanics in pipes, the Moody Chart relates Reynolds number, Darcy friction factor, and pipe roughness. Moody created the chart in 1944 by correlating the earlier experimental data of Nikuradse. In Nikuradse's experiments, pipes were roughened by coating their internal surfaces with a monolayer of sand. The pipe wall absolute roughness, ε, was defined as the average diameter of the sand grains. Values of roughness reported in "Pipe Roughness" tables are typically ascertained by comparing pressure drop data to the Moody Chart, an equivalent sand-grain roughness being assigned to each material. It is important to realize that these values are not derived from a direct measure of surface roughness using surface characterization equipment, such as an optical profilometer. Thus, direct measurements of surface roughness may not be appropriate for fluid flow calculations and could lead to significant error, especially when utilizing new materials and/or fabrication techniques.

In this work, we attempted to find an algebraic relationship between physical surface topology measurements and equivalent sand-grain roughness. MATLAB was used to model uniform spheres on a flat surface in two different packing configurations. The spheres' diameter was the analog of the equivalent sand grain roughness value, ε. A profilometer can measure roughness with a number of algorithms including arithmetic roughness (R_a), root mean squared roughness (R_{rms}), and average peak- to-valley roughness (R_{zd}). Using these three algorithms, we simulated a profilometer scan of the MATLAB models. This gave us a relationship between measured roughness and ε.

Although the only material tested was copper due to time constraints, much of the research went towards creating a method of testing that could be used for any material. The copper pipe's internal surface was analyzed with a Zygo NewView 6300 optical profilometer. Using the relationships from the MATLAB models, values for ε were calculated for each of the three algorithms. A pressure drop experiment was also performed to obtain an experimental value for ε, back-calculated using the Haaland equation. This experimental value was compared to the values obtained from the roughness algorithms.

The results from the copper test suggest that an algorithm based on R_{zd}, the average peak-to-valley height, will provide the best conversion algorithm to transfer from a profilometer measurement to a Moody-Chart-compatible absolute roughness. Because these findings are based on a single material, they can only point to a path for future research. The experimental method designed for the copper test can be used for circular pipes of similar size made of virtually any material. With further testing, a more concrete conversion algorithm can be determined.

Incremental Stress Relaxation

Karah Hickman*, Cody Austin, Neil Dorsey, Brianne Widmoyer, and Glen Livesay
Department of Applied Biology & Biomedical Engineering, Rose-Hulman Institute of Technology, Terre Haute, IN 47803

Collagen is one of the most abundant materials in the body, specifically in soft tissues such as tendons and ligaments. Injuries to soft tissues such as those can be life-altering. The methods for repairing the injured soft tissue often involve the use of collagen matrices, which are usually made of materials not normally found in the body. To be able to fully replace the injured soft tissue, the collagen matrices must closely replicate the characteristic non-linear response of collagen.

There are many ways to test if the man-made collagen exhibits a non-linear response. One way is to perform an Incremental Stress Relaxation test on the material. Incremental Stress Relaxation involves pulling on the fiber to a pre-determined length and holding it there for a certain amount of time. The fiber will relax when it is held under that constant length; similar to what happens when silly putty is stretched and held. The load and elongation of the fiber are measured and are then converted to stress and strain data based on the area of the fiber.

Methods

Ten thawed fibers from previously dissected rat tail tendon soaked in PBS to keep moist were chosen to be tested. Fibers were used from the same rat tail to prevent tail-to-tail variation. The fibers were then pulled to failure to find baseline characteristics of individual collagen fibers.

Using this data, it was found a strain rate percentage of 3% was the optimal length to pull the fibers. The next two increment lengths were calculated and loaded into the computer. Ten more thawed fibers from previously dissected rat tail tendon soaked in PBS to keep moist were used in the Incremental Stress Relaxation test. The fibers were pulled at a rate of 300 mm/min and held for ten minutes a total of three times while load and elongation data was taken.

Results

The data was exported and a Cauchy Stress vs. LaGrangian Strain plot was made to best find material properties of the collagen. The slopes at each increment as well as the percent drop at the two increments were calculated. The slope of the top line in the Cauchy Stress vs. LaGrangian Strain plot is the Instantaneous Modulus. The slope of the bottom line in the Cauchy Stress vs. LaGrangian Strain plot is the Equilibrium Modulus.

Discussion

Results are still being compiled for both collagen and man-made collagen. Statistical comparisons will be made between the two materials' Instantaneous Modulus and Equilibrium Modulus to show how man-made collagen compares to natural collagen. This will hopefully give researchers a better understanding of how to more properly make man-made collagen with properties closer to that of collagen found in the body.

Analysis of Proximal Tibia Strain Response to Sagittal Alignment in Partial Knee Arthroplasty Tibial Components

Amanda L. Kingman*[1], Derek B. Archer[1], Kelsey R. Hughes[1], Renee D. Rogge[1,2], Scott R. Small[2], and Michael E. Berend[2]
[1]Rose-Hulman Institute of Technology, Terre Haute, IN 47803, 2Joint Replacement Surgeons of Indiana Foundation, Mooresville, IN

Background:
Partial knee arthroplasty is less invasive than total knee arthroplasty and has shown improved clinical success with modern prostheses designs. Tibial collapse is a clinically observed complication that can require complex revision surgeries. Studies have shown that sagittal alignment has been associated with early mechanical failure of the bone underlying the tibial component. The purpose of this study is to quantify the strain response of the tibia within a range of commonly utilized tibial component orientations.
Method:
Twenty-four composite tibias were divided into four experimental groups, with tibial trays implanted in alignments ranging from 5 degrees of anterior to 10 degrees of posterior slope as measured from the mechanical axis of the tibia. Each bone was fitted with a metal-backed mobile bearing medial tibial prosthesis. Knee motion was simulated by loading the polyethylene meniscal bearing anteriorly, posteriorly, and centrally for each specimen. The tibias were compressively loaded to 1500 N in an electrodynamic materials testing machine, and data was recorded both by the use of strain gauges and digital image correlation.
Results:
Highest strains in the anterior aspect of the tibia were observed in the 5 degrees anterior slope experimental group, while the highest strains recorded in the posterior aspect of the tibia were observed in specimens implanted in 10 degrees posterior slope. Well-balanced tibial loading was observed in tibias implanted with 5 degrees posterior slope; compared to other alignments tested, 5 degrees of posterior slope exhibited 1.32 to 2.46 times more resistance to extreme strain gradients as a result of bearing translation than any of the other slopes tested.
Conclusions:
In partial knee arthroplasty, areas of extremely high strain may lead to tibial collapse, while areas of extremely low strain may lead to stress-shielding and bone resorption. In this study, slope exhibited a significant effect on strain distribution in the tibia. Tibial components implanted in five degrees of posterior slope generated a strain response most evenly distributed among the component alignments tested. By paying close attention to tibial component alignment, clinical complications such as collapse and bone resorption could potentially be reduced.

This research was funded in part by Edwards Lifesciences under the auspices of the IRC.

Quantitative study of how the cross-sectional area and cell density of amoebocytes recovers in horseshoe crabs after bleeding 40-50% of the animal's blood volume

Jillian R. Hufgard and William W. Weiner
Department of Applied Biology & Biomedical Engineering, Rose-Hulman Institute of Technology, Terre Haute, IN 47803

The presence of endotoxins, a potentially life threatening bacterial contamination in medical devices and supplements, is determined by a product know as Limulus Amebocyte Lysate (LAL). LAL is created by collecting hemolymph from the American Horseshoe Crab where the sole cell, the amebocyte, is separated and purified. This lysate is highly sensitive and incredibly important with respect to the health industry, but is known to raise the mortality rates of the crabs up to 10%. These very important animals are seeing population declines because of environmental pressures and fishing industries, aside from the LAL industry, suggesting the need for an alternative to bleeding to keep the $15 million a quart industry alive. This research worked to quantify and characterize cell size, shape, and concentration in amebocytes from horseshoe crab hemolymph following significant bleeds.

In order to systematically quantify the cell size, shape, and concentration of the amebocytes, a protocol was developed to draw their blood and capture images with as little outside contamination as possible. A series of test bleeds was performed which aided in the establishment of a protocol. Through literature reviews, it became obvious that the largest complication results from contamination forcing the amebocyte cells to lysate before they can be counted and measured. We overcame this obstacle by using a computer program, DinoCapture 2.0, and inserting a small camera into the eye piece of a microscope. Using this device, pictures could be taken of the cells before they lysed, which allows for documentation of the cells for later data collection. Using the camera in this fashion allowed for minimal handling of the crabs and time between the bleed and data collection. For the safety and health of the horseshoe crab, the animals were strapped down for easier bleeding, cleaned with a 95% alcohol mixed on the area of soft tissue, and a clean needle and syringe were used for each animal. Originally, a chemical was being used to slow the clotting of the amebocytes, but this was abandoned when it became apparent that the chemical was changing the size of the cells and was not stalling the clotting long enough to be effective. When large bleeds were being performed, the blood was not removed via syringe in order to provide less stress on the animal. The blood was removed by inserting a needle into the soft tissue near the tubular heart and letting the pumping action of the animal's heart drive blood out of the syringe.

The current literature suggests the horseshoe crabs will regain their blood volume within a few days, but the production of new amebocyte cells may take up to six weeks. A long term bleed beginning with 25-35% removal of blood was performed for a span of 8 weeks; the data collected suggested that 35% blood removal was not a significant enough amount to quantify the effects of the bleed on cell size or density. The data also showed that the most important time frame to measure is within the first week following the bleed. Because of this discrepancy with the published data, a series of base line data collections for each of the remaining animals was performed in order to have a better comparison for later collections. After this base line data was collected we began a new series of bleeding with six animals removing 40-50% of their blood. Subsequently, small blood draws were taken 2-3 times daily for future quantification of how cell size and density is affected by this bleed. This bleed series is still being performed, but the data collected over the summer provide both a protocol and a basis for present and future research.

This research was funded in part by a Joseph B. and Reba A. Weaver Undergraduate Research Award and in part by the Edwards Lifesciences under the auspices of the IRC.

The Recovery of Viscoelastic Properties in Creep of Rat Tail Tendon Fibers

Brianne Widmoyer*, Neil Dorsey, Cody Austin, and Glen Livesay
Department of Applied Biology & Biomedical Engineering, Rose-Hulman Institute of Technology, Terre Haute, IN 47803

Biological soft tissues frequently encounter constant loading during normal activities, which is why a creep viscoelastic test can help assess in vivo mechanics. Creep testing involves constant uniaxial loading to a specimen over time and examining strain. The goal of this test was to subject rat tail tendon fiber specimens to low stress, followed by high stress, with a 20 minute rest period in between tests for recovery. The low stress was a constant 20 MPa, and the high stress a constant 35 MPa. A custom creep testing device was built that utilizes gravity to apply constant, uniaxial tension loading to the specimen. Data collection was obtained using a laser distance transducer, which outputted a voltage linearly proportional to the distance the specimen moved into MATLAB code. The samples (n=8) were obtained from the same tail, and adhered to plexiglas tabs with cyanoacrylate. Samples were soaked in a solution of physiologic buffered saline (PBS) until testing time, and during tests, they were hydrated with a custom PBS drip system. Once the data was collected, the overall percentage of creep was compared within and between fibers by using a paired t test, with significance set at 0.05. The comparison, and therefore test, yielded a p-value of 0.070. A power test was performed since the p-value is close to the significance of 0.05, and the power calculated was 0.4526. We conclude that there is not sufficient evidence to disprove that the samples creeped the same amount, regardless of the level of stress applied. This could suggest that percentage of creep stays the same regardless of the applied stress level, or that the tests were conducted at stresses too close in value to produce statistically observable results.

Separation and detection of nanoparticles using capillary electrophoresis with laser light scattering detection

Kabir Sodhi* and Luanne Tilstra
Department of Chemistry and Biochemistry, Rose-Hulman Institute of Technology, Terre Haute, Indiana 47803

Self-assembly of molecules into aggregates has a profound effect on their chemical activity. The goal of our research is to try and understand the mechanism of this self-assembly and aggregation of the insulin peptide. For this we are using a novel combination of capillary electrophoresis with laser light scattering detection. Data is collected at ultra-high frequency (2925 Hz) to assure multiple data points for each particle. The resulting data files are analyzed using software written within the lab.

The objective of our summer research was to quantify this system by injecting submicron polystyrene beads to establish a relationship between 1) the intensity of scattered light and bead size; 2) retention time and bead size; and 3) the number of beads detected and the number of beads injected. Injected polystyrene beads (from PolySciences ®) were 101 nm, 202 nm, 356 nm, 505 nm, and 1025 nm in diameter. Individual beads were detected for all but the smallest size. Detection efficiency varied from 20 to 35 % depending on the size of beads that were injected. The electrophoretic velocity and scattered light intensity were determined for each particle size detected.

2010 IRC Research Participants

Scholars

Durushka Ahmed
Caitlin Anderson
Cody Austin
Alex Bledsoe
Neil Dorsey
Melissa Galey
Andrew Harris
Karah Hickman
Paul Himes
Jillian Hufgard
Amanda Kingman
Alex Krug
Steven Marczak
Kabir Sodhi
Zhang Wang
Nathan Wheeler
Brianne Widmoyer

Faculty Mentors

Mark Brandt, Ph.D.
Peter Coppinger, Ph.D.
David Finn, Ph.D.
Glen Livesay, Ph.D.
Scott McClellan, Ph.D.
Justin Shearer, Ph.D.
Yosi Shibberu, Ph.D.
Scott Small
Luanne Tilstra, Ph.D.
Lee Waite, Ph.D.
Ross Weatherman, Ph.D.
William Weiner, Ph.D.