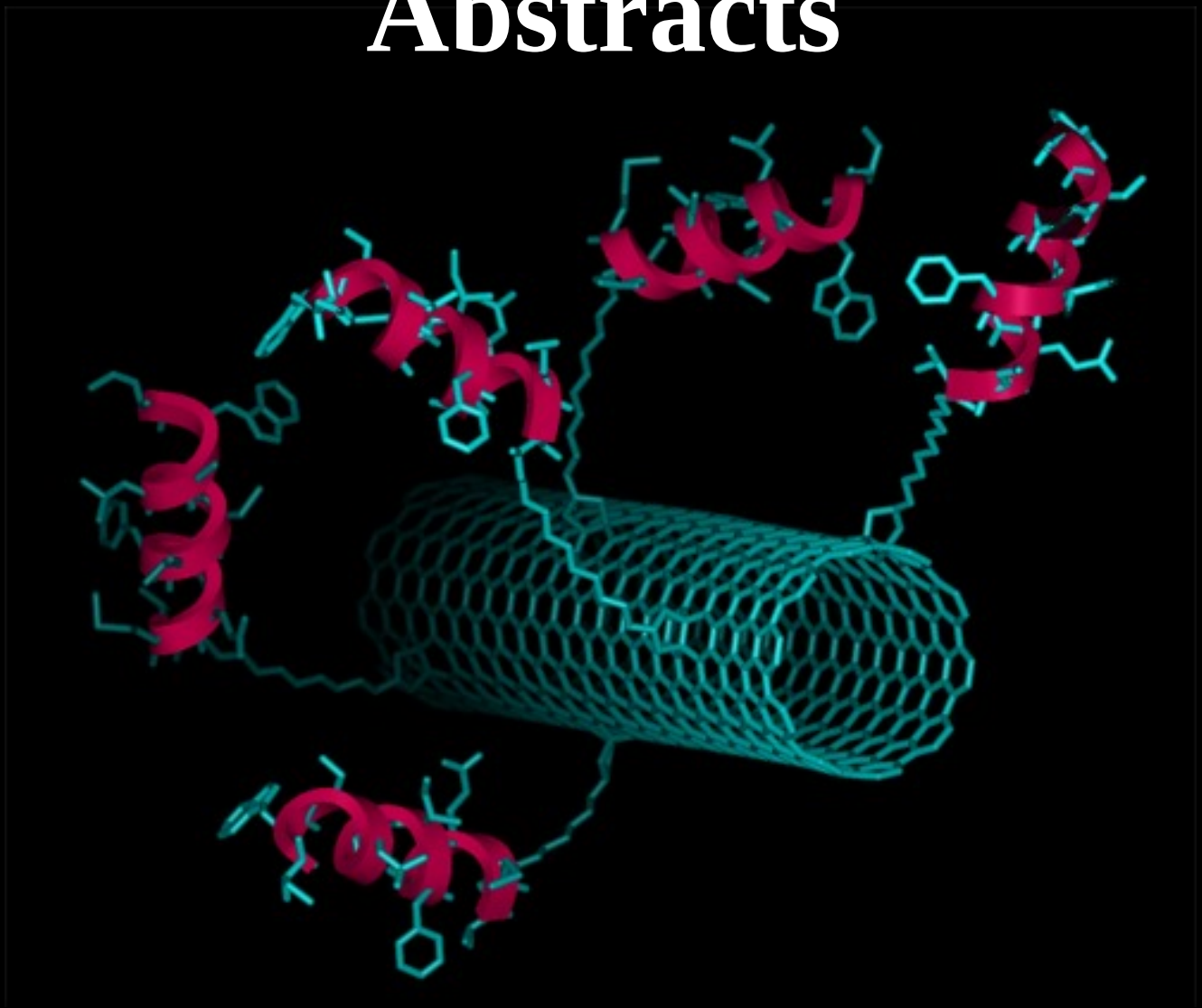
NanoBio Summit 2016 - Poster Abstracts



October 13-14, 2016

Auburn University - Student Center

Auburn, Alabama

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Poster Abstracts: Undergraduate Students

UP-01. Nano-encapsulated proprietary antimicrobial peptides and their antimicrobial activity

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Purpose: A current major concern is the resistance of bacterial microbes to available antibiotics. Nanoparticles in recent years have been increasingly used as a sufficient alternative to antibiotics. Even so, antimicrobial peptides due to their antibacterial activity against multi-drug resistant bacteria have also gained much attention. However, their stability is a major concern. In the present study, we hypothesize that the encapsulation of antimicrobial peptide TP557 in Polylactic-co-glycolic acid (PLGA) will provide stabilized degradation of PLGA, releasing peptide TP557 at a controlled rate and may exhibit better antimicrobial activity.

Methods: First we determined the antibacterial effect of antimicrobial peptide 557 (TP557) from Therapeutic Peptides Inc.® against *Staphylococcus aureus* and *Escherichia coli* using minimum inhibitory concentration (MIC) assay, and growth curve analysis of *Staphylococcus aureus* and *Escherichia coli* exposed to peptide TP557. The encapsulation efficiency of the peptide incorporation into PLGA will be determined and the antibacterial effect of the encapsulated antimicrobial peptide will be evaluated using molecular, microbiological and microscopic methods.

Results: For both bacteria, the data showed that the MIC of the peptide against *Staphylococcus aureus* and *Escherichia coli* was 7.81\ µg/ml. The sequential monitoring of the growth of the bacteria exposed to TP557 showed that the bacterial growth was inhibited in a time and concentration dependent manner.

Conclusions: This information will be beneficial for future research of encapsulation of the peptide. PLGA is advantageous as it is biodegradable, biocompatible, and provides sustained release.

UP-02. Multispectral Opto-Acoustic Tomography and Segmental 360-Degree Bioluminescent Imaging Both Show Potential for the Dynamic Study of Bacterial Infections

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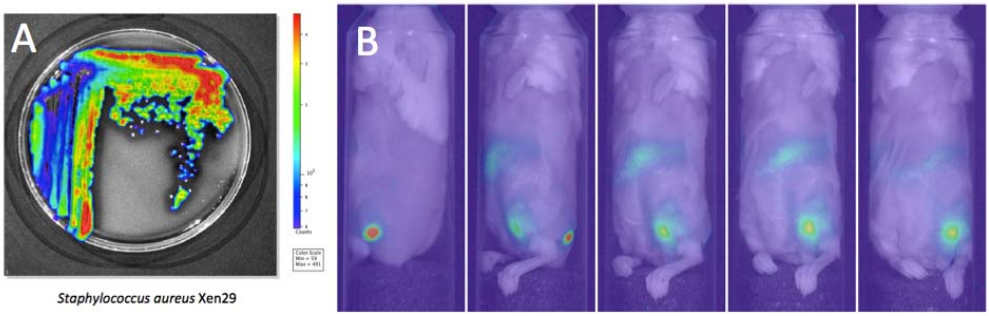
Purpose: Bio-luminescence imaging (BLI) of light-producing pathogens (**Panel A**) has made it possible to track infections non-invasively in mice. However, there exist inherent biases in tracking these bacterial infections as they spread in the mouse. One of the primary issues is the consistent positioning of animal in the imager in relation to the camera or detector. There are also cases where the signal from a deep tissue or bone infection has difficultly being seen, due to the light absorption and scattering properties of blood, fat, and muscle. Here, we use two techniques to overcome these issues and monitor *in vivo* propagation of a pathogen in mouse models over time.

Methods: First, we used our newly developed semi-automated full 360-degree imager called the Mouse Imaging Spinner (MiSpinner), which allows for the actuated rotation of the animal during the imaging process to reduce any positioning biases. Secondly, we used multispectral opto-acoustic tomography (MSOT) to image bacteria via their photo-acoustic signal generated after systemic administration of the near-infrared bacterial surface probe called XenoLight RediJect.

Results: In the images collected by the MiSpinner prototype housed in the IVIS Lumina XRMS system, we were able to appreciate liver signal in 1 of 5 mice on day 5 following bolus injection of *Staphylococcus aureus* Xen29 (1e5 CFUs) (**Panel B**). Using the MSOT imager and XenoLight RediJect agent, we could detect 2 of the 5 animals imaged had splenic and liver abscesses present on day 5 following Xen29 injection.

Conclusion: Taken together, these results suggest new avenues available for the *in vivo* study Staphylococcal infections.



UP-03. Superabsorbent Hydrogels Based on Interpenetrated, Biocompatible Networks

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Purpose: Hydrogels are crosslinked polymers that can reversibly absorb and release large amounts of water. They fall into the category of superabsorbent polymers (SAP's) and have numerous applications. These include 3D printing artificial human tissue, controlled drug release, and slow release fertilizer. There are limitations to creating these innovative technologies though. These hurdles consist of creating a hydrogel that is both mechanically strong and biocompatible.

Methods: To create a mechanically strong hydrogel, interpenetrated polymer networks (IPNs) were synthesized using alginate and acrylate networks. Nanofibrillated cellulose was also added to these IPNs as nano filler, increasing the biocompatibility of the resulting hydrogel. During this research, simultaneous and sequential polymerization of the polymers were studied.

Results: The swelling ratio for all the hydrogels was quite large as expected. Simultaneous polymerization was determined to result in a better swelling ratio than that of sequential polymerization. Swelling was also observed to decrease with increasing cellulose concentration. The mechanical testing of the cellulose/alginate Ca2+ network confirms that adding cellulose as a filler increases the mechanical strength of the polymer up until the point at which it inhibits network formation. From rheology experiments, it was observed that increasing cellulose concentration beyond 70% decreased the storage modulus in all networks except the EPH crosslinked networks. The cellulose/alginate/acrylate networks were determined to have a higher storage modulus than the non-interpenetrated networks as well.

Conclusion: The cellulose/alginate network interpenetrated with an acrylate network was prepared. Cellulose was also confirmed to increase the elastic modulus of the resulting hydrogel up to a point. Beyond this point it was found that cellulose decreased the elastic modulus and shear storage modulus of the hydrogel.

UP-04. Sulfathiazole as a Prophylaxis in Drug Delivery Polyurethane

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Purpose: Urinary catheters are vulnerable to infections, such as *Escherichia coli* and *Candida* spp, necessitating a material that can prevent the infections without compromising the functionality of a catheter.

Methods: The antimicrobial Sulfathiazole was dissolved in dimethylformamide and tetrahydrofuran and then films were cast by the solvent casting process. The drug released from the film into water was analyzed by the Shimadzu UV-2450 spectrometer at 270 nm and compared against calibration curves.

Results: The data shows a two-phase release mechanism. The release mechanism for the first phase is Korsemeyer-Peppas kinetic model while the second phase correlates to the first-order kinetic model.

Conclusion: Our results indicate that sulfathiazole in thermoplastic polyurethane is a potentially viable method to prevent catheter-based infections.

UP-05. Novel Antimicrobial Peptide Mitigates Inflammation in Human Lung Cells Infected with Pseudomonas aeruginosa

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Pseudomonas aeruginosa is an opportunist Gram-negative pathogen that causes infection in immuno-compromised individuals, such as cystic fibrosis (CF) patients. We have shown that antimicrobial proprietary peptides (AMPs) have the ability to inhibit the in vitro growth of both mucoid and non-mucoid strains of P. aeruginosa. Numerous studies now indicate that AMPs can modulate immune responses by inducing cytokines/chemokines production and inhibiting pro-inflammatory cytokine production. In this study, we investigated the anti-inflammatory properties of a proprietary antimicrobial peptide, TP359, in human A549 lung cells infected with P. aeruginosa. We hypothesized that the peptide, TP359, will reduce the inflammation caused by P. aerugonisa infection. Cytokine ELISA was used to determine the anti-inflammatory effect of TP-

359 by measuring pro-inflammatory cytokines Interleukin-6 (IL-6), Interleukin-8 (IL-8), and Tumor Necrosis Factor (TNF). The A549 cells were exposed to TP359 at 4 different concentrations (12.5, 25, 50, and 100 ug/mL). Three types of exposure were utilized: A549 lung cells were exposed to P.aerugonisa 1 hour before TP359 treatment, A549 lung cells were exposed to TP359 1 hour before exposure to P. aerugonisa, and simultaneous exposure of P. aerugonisa and TP359 to A549 lung cells. Our results show that TP359 reduced the levels of pro-inflammatory cytokines in a dose- and time-dependent manner at all treatment conditions. These results indicate that TP359 can modulate P.aerugonisa -induced inflammatory responses in lung cells. Due to its confirmed anti-microbial and suggested anti-inflammatory properties, TP359 is an attractive agent for the development of therapeutics to treat Pseudomonas infections.

UP-06. Bioactive Multiscale 3-D Tissue Scaffolds

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Purpose: Injuries to the Anterior Cruciate Ligament (ACL) tears are one of the most detrimental injuries for athletes. The increasing prevalence of this injury along with the shortage of tissue from the patient's body and the potential of rejection of tissue from donors underscores the need for synthetic tissue sources. Tissue engineering provides a means by which we can potentially develop scaffolds that mimic the structure and function of the ACL.

Methods: We have combined 3-D printing, 3DP and electrospinning of nanofibers to prepare scaffolds that combine the best attributes of both. Polylactic acid (PLA) was used to fabricate both types of structure. PLA nanofibers containing hydroxyapatite (HA) were also prepared to enhance the osteoconductivity. Three-dimensionality and structural integrity is imparted by 3-D printing, while nanoscale surface topology (which is important to cell attachment) will be contributed by the electrospinning, ESP. Scanning electron microscopy (SEM) was used to characterize the morphology.

Results: Scanning electron microscopy (SEM) showed a hybrid morphology, with electrospun nanofibers deposited on a 3-D printed scaffold. Cell studies were utilized to link cell affinity to the different compositions of electrospun scaffolds with human mesenchymal stem cells (hMSCs). Confocal and immunofluorescence microscopy were employed to view cells on the scaffolds using florescent dyes. Attachment and growth of the cells is enhanced by the presence of the HA.

Conclusions: We fabricated a hybrid 3-dimensional tissue scaffold that combines multiple size scales and chemical composition in a manner that mimics the extracellular matrix and promotes cell adhesion and growth. Further characterization is underway and will be presented.

UP-07. Effective Uptake of Poly (lactic acid)-b-Poly (ethylene glycol) Nanoparticles by Mouse Dendritic Cells for Chlamydia Vaccine Development

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Purpose:*Chlamydia* is a sexually transmitted disease, which can cause asymptomatic infections and multiple complications to the human body if not treated. Some of these complications include infertility, pelvic inflammatory disease and epididymitis. We need a vaccine to boost the immune system by generating memory against infection and to provide protection against the asymptomatic nature of disease.

Methods: In this study, we nano-encapsulated peptide from the outer membrane protein of Chlamydia in poly (lactic acid)-b-poly (ethylene glycol) biodegradable nanoparticle (PLA-PEG-NPs) to boost immune responses that can provide protection against its infection. Dendritic cells (DCs) are specialized cells that present antigens to T cells for development of specific adaptive immune responses. We did experiments to determine if DCs uptake PLA-PEG-NPs and activate IL-6 cytokine production. Uptake studies were done using JAW II a mouse dendritic cell line. **Results**: Immunofluorescence microscopy images confirmed localization of PLA-PEG-NPs in DCs and production of IL-6 by ELISA.

Conclusion: Collectively, these data suggests that our nano-encapsulated vaccine drives maturation of DCs and efficient antigen uptake nonspecifically for elicitation of enhanced adaptive immune responses.

UP-08. Development of Procedures for the Differentiation of Human Epithelial Type 2 Cells, Prostate Cancer and Breast Cancer Cells Using Laser Induced Breakdown Spectroscopy

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[1]CNBR Program, [2]Alabama State University

Purpose: Laser induced breakdown spectroscopy (LIBS) is a technique that uses laser pulses to create atomic emission representative of the sample. In the present study, LIBS was explored as a viable technique in discriminating between diseased and healthy human cells by comparing the elemental intensities of each cell type.

Methods: We used three types of cells in this experiment: healthy human epithelial type 2, prostate cancer, and breast cancer cells. Various cell counts and volumes were used in to improve the quality of our LIBS signal. We also manipulated the energy of the laser and used two different spectrometers (ANDOR and Avantes) in an effort to find certain elements suggested in literature, such as Cu II at 518.3 nm or Ca at 610.27 nm.

Results: Our results indicated that the cells should be suspended in deionized water rather than their respective growth mediums since each of the mediums contained trace minerals that greatly affected our results. The data indicated different intensities at different wavelengths for each cell type, such as the human epithelial cells having a significantly higher relative intensity than the prostate cancer cells at 610.27 nm, which is the emission line for calcium. The differences in intensity could be the result of significantly different cell counts for each cell type even though our preparation methods were the same. As an example, our last two experiments placed great effort on keeping cell counts and volume nearly the same. Despite our efforts, the breast cancer cells still managed to produce remarkably different results in each experiment.

Conclusion: Further investigation of the study would require an improved method of sample preparation and possibly a more rigorous data analysis scheme which explores the use of elemental ratio.

UP-09. Antibacterial effect of antimicrobial peptides and functionalized carbon nanotubes at different pH conditions using Kirby Bauer diffusion assay

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Purpose: Bacterial resistance against current antibiotics is important issue. Antibiotic resistant bacteria are difficult to treat and hence novel approaches are needed. Over the past few years, the nanoparticles and antibacterial peptides (APs) have been effective against multi-drug resistant bacteria. Of relevance, APs functionalized silver coated carbon nanotubes (fAgCNTs) have been very effective as antibacterial. The functionalization of AgCNTs with these peptides has additive effect but the antibacterial activity at different pH has never been tested.

Methods: In the present study, we determined the antibacterial activity by Kirby Bauer disc diffusion assay of three peptides from Therapeutic Inc., designated as TP557, TP556 and TP226 and fAgCNTs with these peptides. The antibacterial effect was investigated by performing disc diffusion assay of different concentrations of peptides or fAgCNTs against gram-negative bacteria *Salmonella* Typhimurium and gram-positive bacteria *Staphylococcus aureus* grown on Luria-Bertani (LB) agar plates at different pH conditions such as pH=7.4 and pH=9. The KB assay was performed by using four concentrations such as 20µg, 10 µg, 5µg, and 2.5 µg of each peptide and each peptide-functionalized AgCNTs against bacteria grown on LB agars of pH 7.4 and 9.

Results: After overnight, zones of inhibition indicated inhibition of bacterial growth. The zones of inhibition were measured subsequently. At both the pH conditions *Staphylococcus aureus* growth was inhibited at all four concentrations of either peptides or functionalized AgCNTs. On average the zones of inhibition observed at ph7.4 and 9 were as follows: 8 for 20 µg, 7 for 10 µg, 8 for 5 µg, and 6 for 2.5 µg. Similarly, for *Salmonella* Typhimurium the zones of inhibition at pH=7.4 were, 7 for 20 µg, 6 for 10 µg, 8 for 5 µg and 5 for 2.5 µg. On the other hand at pH 9 the zones of inhibition were, 8 for 20 µg, 8 for 10 µg, 7 for 5 µg and none for 2.5 µg.

Conclusion: Our results clearly indicated the antibacterial effects of the peptides and functionalized AgCNTs against *Salmonella* Typhimurium and *Staphylococcus aureus* grown at pH 7.4 and 9.

UP-10. Comparison of LIBS Emission at Three Different Wavelengths for Nanoparticle Enhanced LIBS

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Department of Physical Sciences, Alabama State University

Purpose: Laser Induced Breakdown Spectroscopy (LIBS) is an economic alternative to most other methods of elemental analysis. This technique has many advantages because it is quick, easy, relatively simple and requires little to no sample preparation. However, a major disadvantage of LIBS as compared to other techniques is that its sensitivity has very apparent limitations. For this reason, there have been many techniques developed to increase LIBS sensitivity such as double pulse LIBS or resonance-enhanced LIBS. However, these techniques are rather complicated and can be tedious to set up. As a result, there has been a recent increase in LIBS research that incorporates depositing nanotechnology on samples to increase LIBS sensitivity.

Methods: In this project, 2 µL drops of an aqueous silver nanoparticle solution at concentrations: 0.003 µg/µL, 0.0015 µg/µL and 0.000375 µg/µL were deposited onto gold film. Once dried, the gold film was pulsed with 15 mJ of energy using an Nd:YAG laser at wavelengths: 266 nm, 532 nm and 1064 nm. Five replicates were performed for each concentration, as well as the pure gold control, at each wavelength.

Results: The data was analyzed for a gold line at 267.6 nm (Au I), and results suggest that the silver nanoparticle solution reduced the intensity of the gold signal. At each wavelength, maximum reduction in gold emission intensity was observed when the concentration of silver nanoparticles was the highest.

Conclusion: Data shows that silver nanoparticles did not enhance the signal of the gold film. However, future studies may look at the significance of the amount of energy delivered.

UP-11. Characterization of Poly Lactic Acid/Polyhyroxybutyrate-valerate (PLA/PHBV) hybrid nanostructured biocomposite

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Purpose: The objective of this research is to prepare a hybrid biopolymer blend using PLA and PHBV with enhanced mechanical and thermal properties.

Methods: Bio-based PLA and PHBV blends were prepared using the melt-mixing procedure. Tensile, FTIR, DSC, TGA, optical microscopy (OM), and scanning electron microscopy (SEM) tests were performed to investigate mechanical properties, bonding interaction, glass transition temperature, melting and crystalline enthalpy, thermal decomposition, and morphological analysis. Different concentrations (1, 2, and 3 wt%) of nanoclay was added to the system to observe the bonding interaction.

Results: It was observed that the crystallinity increases with increasing amount of nanoclay. The result showed that the tensile strength of PLA thin film and PHBV film was found to be 31.1 MPa and 14.41 MPa, respectively. Hence, PLA has better mechanical properties than PHBV. On the other hand, the thermal properties of PHBV thin film were found to be better than that of PLA. To optimize both mechanical and thermal properties, a hybrid biopolymer blend of PLA and PHBV, using various combinations of PLA/PHBV including 25/75, 50/50 and 75/25 wt%, was prepared. Among them, PLA-PHBV (75/25 wt%) with 2 wt% nanoclay resulted in the best outcome. The tensile strength of this prepared polymer blend was 29.34 MPa. Thermal analysis demonstrated two melting temperatures: 238.37 ºC and 308.31 ºC, respectively. Two glass transition temperatures were found from thermal tests which are the indication of the solution immiscibility. It was also observed that the adding of nanoclay enhances tensile properties as well as thermal stability up to 2 wt%. Optical and SEM micrographs revealed that the 2 wt% NC was dispersed uniformly throughout the resin blend.

Conclusion: A combination of two individual resins with differing advantageous properties resulted in a resin with both properties: the bio-based PLA and PHBV blend effectively created a composite with improved mechanical and thermal properties. Specifically, PLA-PHBV (75/25 wt%) with 2 wt% nanoclay performed best mechanically and thermally.

UP-12. Fabrication of Polymeric Tissue Scaffolds by 3-D Printing

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Purpose: A number of approaches for generating porous, 3-dimensional biodegradable polymeric scaffolds have been reported, including solvent and particulate leaching, phase-separation, freeze drying, and self-assembly. While all of these approaches have advantages and disadvantages, some of the key shortcomings include the lack of precise control of pore geometry, size, distribution and interconnectivity. These shortcomings can be addressed by fabricating the scaffolds using 3D printing.

Methods: In this study, three diverse scaffold architectures were 3-D printed and used for mechanical testing. We systematically controlled various structural design parameters (eg., control pore geometry, size, distribution, and interconnectivity) in order to determine if we are able to directly control properties (modulus, porosity, strength, strain, etc.).Samples were fabricated using a fused deposition type three dimensional printer. The dynamic mechanical properties of the resulting structures was characterized and compared to their architecture and crystallinity.

Results: The orthogonal sample exhibited the most resistance to torsional deformation, resulting in the higher modulus. The unidirectional sample exhibited the lowest modulus and the multidirectional sample exhibited a modulus that was intermediate to the other two. The multidirectional sample was the most porous due to it's geometry resulting from the 45˚ stack sequence, followed by the orthogonal sample and lastly the unidirectional sample.

Conclusions: The capability to fabricate biodegradable scaffolds with controlled strength, modulus, and porosity has many potential applications in tissue engineering. We have demonstrated the effect of scaffold geometry on mechanical properties. We are currently designing more realistic structures and investigating their mechanical behavior and cell compatibility

UP-13. Novel Green Synthesis of Silver Nanoparticles Using Beringia Ligulata Plant extract

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Purpose: This study was conducted to develop an environmentally safe, and cost effective novel method for synthesis of silver nanoparticles using aqueous extract of the plant "Bergenia Ligulata". The inherent anti-microbial activity of Bergenia Ligulata may add or synergize the anti-microbial efficacy of silver nanoparticles and can be useful in many biomedical applications.

Method: A rapid, simple approach was applied for synthesis of silver nanoparticles using Bergenia Ligulata plant extract. The plant extract acts both as reducing agent as well as stabilizing agent. In brief, an aqueous solution of silver nitrate (0.1 mL, 0.1 M) mixed with freshly prepared aqueous plant extract (9 mL) and volume was made up to 10 mL with distilled water. The mixture was incubated at 30^{o}C in dark and continually stirred for 48 hrs. After 48 hours, the solution was removed and stored for characterization and testing.

Results: UV-Visible spectroscopic studies showed characteristic Surface Plasmon resonance (at 500 nm) for silver nanoparticles. TEM studies revealed that silver nanoparticles were spherical in shape and in the size range of 10-100 nm. Elemental analysis using Energy dispersive X-ray (EDX) spectroscopy confirmed the presence of silver in nanoparticles.

Conclusion: In this study, we have successfully developed one-step biosynthesis approach for silver nanoparticles using aqueous extract of Bergenia Ligulata, for the first time.

UP-14. Nanobiomaterials and their applications in tissue/skin regeneration

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Synthetic or natural biomaterials are used for generating artificial organs and prostheses or to replace tissues. Some of biomaterials have been engineered to work with biological systems and used as therapeutics and other medical purposes. Some biomaterials can be synthesized while others are found naturally. There are three different categories of biomaterials which include bioinert, bioactive and bioresorble. Bioinert materials can be used in the human body with minimal adverse effects with its surrounding tissues such as use of titanium in medical devices. Bioactive materials once introduced in the human body interact with the surrounding bones and soft tissues. This occurs through a time dependent kinetic modification of the surface of the material and is often triggered by being implanted within the living bone and tissue. Bioresorbable materials start to dissolve and are slowly replaced by advancing tissues in the body. Common examples of bioresorbable materials are tricalcium phosphate and polylactic–polyglycolic acid copolymers.

Future applications of these types of biomaterials include skin regeneration for various skin types and wound healing. To further enhance the applications of these biomaterials, various nanomaterials such as silver and carbon nanotubes can be incorporated with antimicrobial properties. The skin/tissue regeneration with nanobiomaterials will have its applications in wound healing as well as other types of tissue replacement for treatment of diseased tissues in patients.

UP-15. Liposomal Encapsulation of Genipin and Toxicity Analysis

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Purpose: Liposomes are spherical vesicles comprised of a phospholipid bilayer and an aqueous core. Over two decades, liposomes have been engaged in drug delivery. More than a dozen liposomal formulations have entered the market. Liposomes due to their structural attributes can be used for targeting. Genipin is a natural product extract derived from the gardenia fruit. Past research confirms the potential of genipin against breast cancer.

Methods: Liposomes were prepared using conventional thin film hydration method. Briefly, the lipid components *1,2-disteroyl-sn-phosphatidylcholine* (DSPC): Cholesterol (5:2 w/w) were dissolved in an organic solvent mixture of chloroform and methanol with the addition of lipophilic drug, followed by solvent evaporation to obtain a thin dry film. The film was then hydrated with phosphate saline buffer (PBS), resulting in the formation of multi-lamellar vesticles (MLV). This MLV suspension was then sonicated using probe sonication and SUV were obtained. The sonicated liposomal suspension was then centrifuged to remove titanium debris and the final suspension was then tested for size and zeta potential analysis. Drug encapsulated liposomes were prepared having genipin was dissolved in buffer. Initial analysis of genipin has confirmed that it is soluble in water as well as ethanol. Therefore, possibility was concluded that the genipin will get entrapped into the aqueous core as well as the lipid bilayer. Entrapment was quantified using UV/Visible spectrophotometry.

Results: It was observed that 28.2% (n=2, ±0.2) was encapsulated into the liposomes. The efficacy of genipin and genipin encapsulated liposomes was tested against MDB-MA 231 cell line and the results show that the genipin encapsulated into the liposomes is more prominent compared to effect of genipin alone.

Conclusion: The genipin encapsulated liposome were prepared successfully and encapsulated genipin is more efficient for cell death compared to effect of genipin alone

UP-16. Functional Role of SOCS on IFN-Y signaling in *Chlamydia trachomatis*

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Background: *Chlamydia trachomatis* (CT) is a Gram-negative, obligate intracellular pathogen and the most reported bacterial sexually transmitted pathogen globally. A hallmark of CT infection is its capacity to mount a massive inflammatory response such as pelvic inflammatory disease (PID) in the female genital tract. Suppressor of cytokine signaling (SOCS) proteins are negative regulators for various cytokines, and SOCS1 is known to negatively regulate IFN-γ signaling. We have observed that CT induced the up-regulation of SOCS1 in mouse J774 macrophages, which suggest that SOCS1 may act as a direct modulator of IFN-γ cytokine signaling in macrophages to control its own inflammation.

Purpose: The objective of this research project is to investigate the functional role of induced SOCS1 by CT on IFN-γ signaling in mouse J774 macrophages. **Hypothesis:** Our hypothesis is that SOCS1 protein, induced by stimulation of macrophages with CT, inhibits the expression of IFN-γ and therefore prevents macrophage activation.

Methods: Mouse macrophages were exposed to various concentrations of IFN-γ (12.5 to 50 ng/mL) and LPS (1 µg/mL) and TaqMan qRT-PCR was performed to assess expressions of SOCS1 mRNA gene transcripts, with SOCS3 serving as a control. We also performed western blot to assess SOCS protein expression. **Results:** Our results showed that all concentrations of IFN-γ induced marked upregulation of SOCS1 in macrophages as compared to SOCS3 over a 24 hr time-period. Western blotting further confirmed the raised level of the corresponding SOCS1 protein.

Conclusions: We have shown that CT has the capacity to regulate its own inflammation by inducing SOCS proteins. The ability of CT to also induce SOCS3 suggest that it may regulate other cytokines, like IL-6, in addition to IFN-γ. These results will uncover a potential mechanism of how CT controls its induced inflammation early after infection of the host. Studies are ongoing to determine the capacity of CT-induced SOCS1 to negatively regulate the expression of STAT1 (signal transducer and activator of transcription 1) in mouse macrophages.

UP-17. Processing of soy protein-based nanostructured biocomposite

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Purpose: A completely biodegradable composite was fabricated from an herbal polymer, soy protein concentrate (SPC) resin. Soy protein was modified by adding 30 wt% of glycerol and 5 wt% of poly vinyl alcohol (PVA) to enhance its mechanical as well as thermal properties. 3%, 5%, 10%, and 20% nanoclay (NC) were infused into the system to evaluate how they affected the strength of the matrix.

Methods: Tensile tests, x-ray diffraction (XRD), thermogravimetric analysis (TGA), differential scanning calorimetry (DSC), fourier transform infrared spectroscopy (FTIR) tests, optical microscopy (OM), and scanning electron microscopy (SEM) evaluations were performed to evaluate the mechanical properties, crystallinity, thermal properties, bonding interaction, and morphological evaluation of each composite respectively.

Results: Tensile tests showed that the addition of nanoclay improved the mechanical properties of the modified resin. Soy protein is hydrophilic due to the presence of amino acids that contain various polar groups such as amine, carboxyl, and hydroxyl. As a result, polar nanoclay particles that are exfoliated can be evenly dispersed in the SPC resin. From experimental results, it is clear that adding of nanoclay with SPC resin significantly increased the sti□ness of the SPC resin. A combination of 5% clay, 30% glycerol, and 5% PVA with the modified SPC resulted in the maximum stress of 18 MPa and Young modulus of 958 MPa. The modified SPC showed a reduced failure strain as well. X-ray diffraction curves showed an improvement of crystallinity of the prepared resin with increasing amount of nanoclay. Interaction among soy, glycerol, PVA, and nanoclay was clearly demonstrated from the FTIR analysis. Optical microscopy (OM) and scanning electron microscopy (SEM) micrographs revealed rougher surface in the nanoclay infused SPC samples compared to that of the neat one. SEM evaluation revealed rougher fracture surface in the NC infused samples.

Conclusions: The Soy Protein-based nanostructured biodegradable composite has adequate properties to replace its non-biodegradable counterparts in indoor applications such as furniture or automobile interiors.

UP-18. 3-D Articular Cartilage Scaffolds Based on PVA/PLA Composites

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[1]Montgomery Catholic High School, Montgomery; [2]Department of Biology, Alabama State University; [3]Biomedical Engineering Program; Alabama State University

Purpose: Articular cartilage is a smooth, transparent tissue that covers the ends of bones in joints such as the knee and elbow. Defects and loss of cartilage can be caused by traumatic injuries and degenerative joint diseases. Unfortunately because of its lack of vasculature, cartilage has a limited capacity to undergo self-repair. While some treatments are available to repair defective cartilage, they do not present a long term solution. Tissue engineering presents an approach to synthetically develop replacements for articular cartilage. The aim of this research is to fabricate 3-dimensional scaffolds that mimic the complex architecture and biochemical nature of articular cartilage. **Methods:** We have fabricated scaffolds based on electrospun nanofibers of polylactic acid (PLA) infused with a hydrogel system based on polyvinyl alcohol (PVA) and sodium alginate (SA). The microstructure, chemical composition, mechanical properties and cell attachment and proliferation have been studied.

Results: Microscopy of the scaffolds reveals a two-phase, porous structure. A gradient in the structure and properties is also evident. The chemical composition of the gradient was characterized using infrared spectroscopy. The mechanical properties of the two components have been characterized separately using dynamic mechanical analysis. Characterization of the hybrid system is underway.

Conclusions: We have fabricated a hybrid, three dimensional scaffold that mimics the structure and of articular cartilage**.** The samples exhibit and two phase system with interconnected porosity in each phase. The sample also exhibits a well-defined, diffuse interphase region. The impact of the interphase structure on the mechanical properties will be presented.

UP-19. Development of Cell-membrane Coated Nanoparticle Platform for Targeted Drug Delivery

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Introduction: Cell membrane coated PLGA nanoparticles loaded with anti-cancerous drug are the recent development of

personalized medicine. This approach significantly lowers the cytotoxicity compared to free drug treatment by its specific targeting taking advantage of inherent homotypic binding and excellent immunocompatibility.

Methods: we have designed a naturally targeting nanoparticles using cancer cell membranes loaded with a hydrophobic/hydrophilic drug in an effort to more efficiently deliver chemotherapeutics to the site of action. First, we verified purification procedure for the membranes isolated from cancer cell using western blotting. We also estimated protein and DNA after each step. With the collected membrane, cancer cell membrane vesicles were then prepared by physical extrusion through a 400 nm and 200 nm porous polycarbonate membrane. Concurrently, we prepared the core of the nanoparticles which is composed of poly (D,L-lactic-co-glycolic acid) (PLGA) biodegradable polymer. The PLGA nanoparticles were loaded with the hydrophobic (curcumin)/Hydrophilic (doxorubicin) drug through a well-studied nanoprecipitation and solvent evaporation process. Cell membrane vesicles and nanoparticles were measured for their physiochemical characteristics.

Results: Results reveal an average hydrodynamic diameter for vesicles under 300 nm (using 0.4 um filter) and 147.6 nm (using 0.2 um filter). The drug loaded polymeric nanoparticle were approximately 60 nm for curcumin and 165.98 nm for doxorubicin in size. The zeta potential below -35 mV and – 17.7 mV are expected in alignment with those seen with PLGA nanoparticles and cell membranes. We did not see any size difference after lyophilization of drug loaded nanoparticles. The drug loading was found to be 2.4 % for curcumin and 2.1 % for doxorubicin. Purified cancer cell membrane will be coated onto the nanoparticles' surface which will allow for high-affinity binding to the homotypic site of cancer. Functional assays and cell culture studies are being planned for future studies to better understand the efficacy of such cancer cell coated nanoparticles.

Conclusion: This novel concept by combining natural targeting of cancer cells using a currently approved drug, to create a novel class of targeted drug delivery with strong potentials to improve on therapeutic efficacy for treatment of a variety of diseases.

UP-20. Eco-friendly Methods of Synthesis of Silver Nanoparticles and their Anti-microbial Activities

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We have adopted "go green' strategy for the synthesis of silver nanoparticles by using plant extracts such as banana and tea. Bana peels and tea leaves are rich in polyphenols that can serve as substrate as well as reducing agent for the silver particles. Our results showed that tea extract had limited ability as a reducing agent or as stabilizing agent and required the addition of starch or glucose as a stabilizer and to reduce metal precursor, silver nitrate into silver nanoparticles. Ultraviolet-Visible (UV-Vis) was used to determine whether or not silver nanoparticles were present. The surface morphology of the nanoparticles was determined by the Scanning Electron Microscope (SEM), Zetasizer and zeta potential was used to determine the size and surface charge of the particles, and Laser Induced Breakdown Spectroscopy (LIBS) was used to determine whether or not the nanoparticles were silver. Also, the susceptibility and antimicrobial activity of the silver nanoparticles against bacterial strains of *Salmonella enterica* serovar Typhimurium was studied using Kerby Bauer Disc Diffusion assay and MIC method, and found to be highly effective.

UP-21. Polymeric Tissue Scaffold Design using 3-D Printing

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The field of 3-D printing has experienced rapid growth over the last few years, with a number of potential applications for tissue engineering. However, to assist in realizing this potential, there is a need for a better understanding of the structure-property relationships of the materials, particularly the polymers that are being fabricated and studied as tissue scaffolds. Therefore, the objective of this work is to develop a fundamental understanding of the link between the chemical structure and composition, physical properties and cytocompatibility. We have used 3-D printing to prepare tissue scaffolds that vary in composition and structure. Preliminary correlation of the structure (i.e. chemical makeup; crystallinity and scaffold architecture) with the physical properties (melting behavior; mechanical properties; porosity) shows that the pore size and pore geometry are key variables for a viable scaffold. Optimization of the pore geometry, porosity and correlation of physical properties with cytotoxicity is underway.

UP-22. Mechanical characterization of nanophased jute fiber reinforced PLA-PHBV biocomposite

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Purpose: The objective is to develop nanophased jute fiber reinforced PLA-PHBV biocomposites as leading alternatives to synthetic composites in the areas of biomedical, automobile, construction, and consumer goods. The incorporation of nanoclay in the polymer matrix is expected to improve physical, mechanical, viscoelastic, thermal, fire resistance as well as electrical properties of the resultant composite.

Methods: Untreated/treated jute fiber reinforced composites are manufactured using PLA and PHBV (PLA-PHBV) blend as a matrix modified without/with 0-3 wt% Closite NA+ nanoclay. The purpose of using polymer blend is to reduce the brittleness of PHBVand taking the advantage of higher mechanical strength of PHBV. PLA-PHBV blend was dissolved into chloroform at a ratio of 1:10 at room temperature and stirred by magnetic stirrer for 24 hours to prepare a homogeneous solution. In case of nanophased composites, nanoclay was dispersed into the PLA-PHBV blend using ultrasonication technique followed by magnetic stirring as well as vacuum mixing. This nanophased blend was then poured into a mold to prepare a thin film and dried at room temperature to evaporate chloroform. Dried films were then placed in the hot press and force was applied at 180 °C for 2 minutes to prepare flat thin films of the composite. Jute/PLA-PHBV blend composites were produced by stacking films and fibers like a sandwich using the compression molding process applying 1.5 ton of pressure at 180 °C for 20 minutes. Mechanical characterization and fracture morphology evaluation were performed using flexure test and optical microscopy, respectively.

Results: The experiment proved to be a success in forming sufficient biocomposites. The nanophased treated jute fiber reinforced composites demonstrated enhanced mechanical properties compared to those of the untreated one.

Conclusion: Mechanical properties and fracture morphological study demonstrated that these biocomposites can be easily used as an interior structural material to replace synthetic fiber reinforced composites.

UP-23. Computational Study of Electrostatically Tunable Band Offsets in MoS_2 Multilayer Heterostructures

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Purpose: In the past years, the isolation of semiconducting transition-metal-dichalcogenide molybdenum disulfide (MoS_2) has opened promising opportunities for flexible optoelectronic applications. However, achieving low-contact resistance in these layered materials is an important limitation in their application in electronic devices as it is limited by Fermi level pinning. More recently, the metallic 1T-phase of MoS_2 (metastable) has been produced experimentally and has been proposed as a solution to the contact issue in two-dimensional materials.

Methods: We perform first principles calculations within the density functional theory to study the electronic properties MoS_2 multilayers. In particular we characterize the band offsets changes in MoS_2 multilayers of different thicknesses (number of layers) and composition (phases of MoS_2) under external electric fields in different geometry configurations.

Results: We find that the introduction of 1T-MoS_2 layers alleviates the Fermi level pinning observed in these materials. In field-effect transistor geometries, configuration where the 1T-phase resides at the top of the stack offers the largest response to external fields. A comparison of the results using the generalized gradient approximation and the HSE hybrid functional (which determines the band alignments more accurately) shows that both approaches yield similar band offsets variations despite their different band alignments.

Conclusion: Using multilayer hetorostructures combining the different phases of MoS_2 enhances the band alignment susceptibility to external electric fields by alleviating the Fermi level pinning at the metal/semiconductor interface. The modulation of the band offsets based in transition dichalcogenide multilayers may prove useful for the realization of flexible two-dimensional electronics.

UP-24. *Salmonella* Typhimurium and *Staphylococcus aureus* Gene Transcriptional Changes in Response to Functionalized Carbon Nanotubes and pH Variations

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Purpose: Bacterial resistance to modern antibiotics is a serious concern and new strategies are needed to inhibit the infection from spreading. Metallic nanoparticles such as silver coated carbon nanotubes (AgCNTs) have demonstrated good

antibacterial activity. Antimicrobial peptides (APs) have also shown efficacy against drug-resistant pathogens. However, the APs are unstable at different pH conditions, and may need to be combined with nanoparticles to be effective. In this study, we investigated the antibacterial effect of AgCNTs functionalized with a proprietary peptide TP226 (Therapeutic Peptides, Inc., fAgCNTs) under two different pH conditions (acidic pH=5 and alkaline pH=9) against a gram- negative bacterium, Salmonella Typhimurium and a gram-positive bacterium, Staphylococcus aureus.

Methods: Studies included minimum inhibitory concentration assays and gene transcription assays carried out by qRT-PCR. The expression of ATP-binding cassette transporter genes (dpp, artP and livJ) in Salmonella Typhimurium and cell wall biosynthesis gene (ftsZ) in Staphylococcus aureus were investigated. The MIC assay showed that fAgCNTs were highly effective (MIC values lower than 0.9 μg/ml) as opposed to the plain AgCNTs (MIC value 62.5 μg/ml). Transcriptional analysis of the genes associated with transporter systems in Salmonella Typhimurium and cell wall biosynthesis in Staphylococcus aureus revealed that these genes were either up-regulated or down-regulated several folds (~3-4 folds) in fAgCNTs treated bacteria at alkaline pH compared to neutral pH.

Results: The expression of dppA was up regulated and expression of artP and livJ was down regulated at pH=9 in Salmonella Typhimurium exposed to fAgCNTs. The down-regulation of artP and livJ and up-regulation of dppA at pH=9 validates the better antibacterial activity of the fAgCNTs at pH=9. Similarly, the up regulation of cell division mediating gene ftsZ at pH=9 in Staphylococcus aureus indicated a strong stress response.

Conclusions: Our results show that fAgCNTs are much more effective at alkaline pH compared to neutral pH. Currently, we are investigating the effects of fAgCNTs on the expression of several genes associated with cell division and amino acid biosynthesis in both the pathogens.

Poster Abstracts: Graduate Students

GP-01. Acyclovir Nanogel Formulation for Human Skin Cadaver Permeation Enhancement

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Purpose: Topical application of acyclovir has a potential purpose to target Herpes Labialis infection due its efficacy as anti-viral agent. Topical application of acyclovir demonstrates a low efficiency compared to the other routes owing to low skin permeation of acyclovir. There is a need to enhance the permeation of acyclovir to reach deep layers of skin and treat the infection. We proposed formulating acyclovir nanoparticles based gel to enhance the drug permeation across skin layers.

Methods: Acyclovir nanoparticles were prepared via ball milling method. The physicochemical evaluations were tested such as PH, viscosity, uniformity, drug content and particle size. We have made modification on the formula utilizing different chemical penetration enhancers with various % and compositions (Ethanol, PG, Oleic acid, Transcutol p, n-methyl-2-pyrrolidone (pharmasolve). In vitro release and permeation kinetics using regenerated cellulose membrane and dermatome human cadaver skin, respectively were studied in comparison with the commercial product (Zovirax), the acyclovir concentrations in the samples were analyzed by a validated HPLC-UV method.

Results: Ball milling produced acyclovir particle size at 269.5 nm with PI, 0.321. These nanoparticles in the gel formulations showed a pH 6, viscosity 7177 cP (5 rpm), and drug content 24.9 mg/gm (99.6%). This formulation showed a skin permeation of 0.22 $\mu g/cm^2$/hr. The modified formula with various chemical penetration enhancers showed a permeation enhancement 0.50 $\mu g/cm^2$/hr. (5% ethanol) and 2.68 $\mu g/cm^2$/hr (10% ethanol), which were 15 and 81 fold higher as compared with the flux produced by Zovirax (0.033 $\mu g/cm^2$/hr).

Conclusion: Developing Acyclovir nanoparticles in conjunction with ethanol as a penetration enhancer is showing pronounced effect on enhancing acyclovir permeation upon topical application.

GP-02. Evaluation of Diacetyl Boldine Loaded Microemulsion Formulations for Topical Drug Delivery: Preparation, Characterization, In Vitro Release and Cytotoxicity Studies

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Purpose: The object of this study was to prepare novel microemulsion formulations of Diacetyl Boldine (DAB) for topical delivery and to evaluate in vitro cytotoxicity of microemulsion formulations against melanoma cell line (B16 BL6).

Methods: A simple, reproducible, accurate and sensitive method was developed for quantitative analysis of DAB in microemulsions using high performance liquid chromatography (HPLC) with UV detection. Pseudo-ternary phase diagrams was plotted to identify the formulation region and optimal microemulsions were characterized for their particle size, viscosity, pH and in vitro release properties. Permeation studies were performed using excised human skin using Franz diffusion cells. The cytotoxicity of the reported formulations on B16BL6 melanoma cell lines were evaluated by MTT assay.

Results: HPLC method for DAB was established by optimizing isocratic flow parameters of the mobile phase. Based on the pseudo-ternary phase diagram, four microemulsion formulations were selected, and the PH of the selected formulations ranged from 5.26 to 6.5. Optimized formulations showed globule size of <50 nm, and polydispersity index of 0.31. The ex vivo skin permeation study demonstrated that the microemulsions exhibited a potent skin enhancement effect to penetrate skin layers up to 8-13 fold higher compared with the control (DAB-MCT oil). Furthermore, there is a significant increase in cancer cell death for all DAB microemulsions compared to the control. The optimized formulations showed 160-180% higher cytotoxicity toward B16BL6 cell lines compared to control. The half-maximal inhibitory concentrations (IC50) of F12, F20, F26, and MCT formulations against B16BL6 cells were calculated to be 1 µg/mL, 10 µg/mL, 10 µg/mL, and 50 µg/mL respectively. By comparison, the IC50 of F12 was 50 fold lower relative to that of the DAB-MCT formulation.

Conclusion: Results of present study suggest that microemulsion could be a promising formulation for topical administration of DAB.

GP-03. Evaluation of the *in vitro* and *in vivo* optoacoustic properties of PEGylated, Cy7 conjugated-iron oxide nanoparticles using multispectral optoacoustic tomography (MSOT)

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Purpose: Addition of a hydrophilic, electrostatically neutral polymer such as polyethylene glycol (PEG) to the surface of nanoparticles provides a "stealth" property through steric hindrance to opsonization and macrophage uptake; thus minimizing plasma clearance in the body. By prolonging circulation, nanoparticles have a greater opportunity to passively enter the compromised, leaky tumor vasculature through the enhanced permeability and retention effect. The purpose of this study is to examine the effect of the size of PEG, which alters the surface properties of nanoparticles, on the pharmacokinetics and biodistribution profiles of PEGylated superparamagnetic iron oxide nanoparticles (SPIONs). By conjugating a near-infrared fluorophore (Cy7), we are able to image and quantify nanoparticle accumulation in various tissues using multispectral optoacoustic tomography (MSOT).

Methods: The *in vitro* and *in vivo* effect of PEGylation was tested both *in vitro* and *in vivo***.** We performed *in vitro* experiments to mimic the first two steps (protein adsorption and macrophage uptake) of the reticuloendothelial system (RES) clearance process. Next, we conducted MSOT phantom studies on our PEGylated, Cy7 conjugated-SPIONs to measure the optoacoustic properties of our probe and determine imaging sensitivity and reliability. Finally, MSOT was used to visualize and quantify the probe's pharmacokinetics and biodistribution *in vivo* following intravenous injection.

Results: PEG-SPIONs demonstrated enhanced colloidal stability, significantly lower protein binding, lower toxicity and reduced macrophage uptake *in vitro* than the unmodified SPIONs. We then created scattering agar MSOT phantom gels (1.5% agar, 1% intralipid) and tested the photoacoustic response of Cy7 conjugated-SPIONs, and were able to detect very small concentrations (0.0005 mg Fe/mL) of SPIONs. The MSOT signal correlated strongly with SPION concentration. Finally, we were able to use MSOT able to quantify SPION concentration in the ischiatic vein, liver, spleen and kidneys following intravenous injection. The size of PEG affected nanoparticle plasma PK half-life, with smaller size PEG (2K) extending half-life.

Conclusion: We used *in vitro* and *in vivo* methods to validate the enhanced circulation properties of PEGylated SPIONs over unmodified SPIONs. The size of PEG also had an effect on *in vitro* toxicity, macrophage uptake, and *in vivo* plasma PK.

GP-04. Evaluation of the Antibacterial activity of Plain, PVP and Polymer coated Silver Nanoparticles

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Purpose: Novel antibacterials are the need of the hour due to accelerating rates of bacterial resistance to current antibiotics. Silver nanoparticles are effective antibacterial agents, but may prove toxic at therapeutic concentrations. Silver-Polyvinyl Pyrrolidone (Ag-PVP), , and other polymer-functionalized Ag nanoparticles may reduce the toxicity of the silver nanoparticles while inhibiting bacterial growth. In the present study, we tested the antimicrobial activity of plain, PVP and polymer coated nanoparticles against Gram-negative *Escherichia coli* and Gram-positive *Staphylococcus epidermidis*,both of which can quickly develop resistance to common antibiotics such as methicillin. *E. coli* is the most frequent Gram-negative pathogen associated with high mortality of patients with ventilator associated pneumonia (VAP) in intensive care units and affects immuno-compromised individuals. *S. epidermidis* is a Gram-positive pathogen that causes human disease such as skin and soft tissue infections (SSTI) and invasive disease that lead to bacteremia, sepsis, endocarditis, or pneumonia.

Methods: The antimicrobial activity of plain Ag-NP, Ag-PVP, and functionalized Ag nanoparticles against *E. coli* and *S.epidermidis* was investigated using the minimum inhibition concentration (MIC) assay, minimum bactericidal concentration (MBC) assay and bacterial growth curve analysis in a time and concentration dependent manner. We are also testing the effects of the nanoparticles against various genes of *Escherichia col*i and *Staphylococcus epidermidis* using quantitative polymerase real-time chain reaction (qRT-PCR) to determine the possible mechanism of action. The genes include those related to virulence (*aufA, yeD*), amino acid metabolism (*argC, metL, and gadB*) and the TCA Cycle (*frdB* and *aceF*).**Results**: Results available so far showed that the MIC for AgPVP and the polymer coated Ag nanoparticles against *E. coli* and *S. epidermidis* ranged from 0.3125 µg/mL to 0.1565 µg/mL with an MBC of 0.6215 µg/mL. However, *S. epidermidis* treated with plain AgNP was inhibited at concentrations between 1.25 µg/mL and 0.6215 µg/mL, with an MBC > 1.25 µg/mL. The bacterial growth curve further revealed that *E. coli* and *S.epidermidis* treated with AgPVP showed inhibition at a concentration of 0.1565 µg/mL and 0.3125 µg/mL at 8 and 16 hour time points, respectively. The data demonstrates that relative to the positive control the bacteria treated with their respective nanoparticles can reduce the growth of both *E. coli* and *S. epidermidis*

Conclusion: All the nanoparticles tested were able to inhibit both *S.Epidermidis* and *E.Coli*. However, the polymer coated nanoparticles were effective at lower concentrations. The qrt-PCR assay will reveal the effects of the nanoparticles on the genes of both bacteria and studies on mammalian cells will determine which nanoparticles are least toxic.

GP-05. Synthesis and Characterization of BioNovolac-Epoxy Interpenetrating Polymer Networks

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Purpose: The emphasis on the research of bio-based polymer networks is due to the ever increasing demand for renewable resources. Biomass pyrolysis oil (bio-oil) can be focused as an alternative to the limited petroleum resources. Multi-hydroxyl functional pyrolysis oil can be utilized for specific polymerization to yield tailor-made polymer networks. Additionally, other bio-based polymers can be chemically grafted to yield thermoset networks for better thermo-mechanical performance.

Methods: Bio oil was partially used for synthesis of phenol-formaldehyde novolac type polymer. The molar excess of bio-oil/phenol was reacted with formaldehyde in acidic medium to form BioNovolac. Fourier Transform Infrared spectroscopy (FTIR) was used to monitor the progress of synthesis of BioNovolac. After purifying BioNovolac, it was physically blended with epoxy resin synthesized from α-resorcylic acid, a model compound chemically similar to gallic acid found in different natural sources. The epoxy resin was cross-linked in the physical presence of BioNovolac to form interpenetrating type networks. The polymer networks were analyzed using different characterization methods including differential scanning Calorimetry (DSC), dynamic mechanical analysis (DMA).

Results: FTIR spectra of BioNovolac at successive intervals confirmed the consumption of phenol. The technique also helped to observe the polymerization of epoxide groups of α-resorcylic acid based epoxy resin. DSC and DMA results showed the thermal transitions of the interpenetrating polymer networks, and the glass transition temperatures were measured for all the systems. Additionally, with the help of DMA, initial modulus and the modulus in the rubbery plateau of bio-based IPNs, were proved to be comparable to the conventional polymer networks.

Conclusion: It can be concluded that the bio-oil has chemical functionality which can certainly be utilized for polymerization reactions. The bio-oil derived polymers are also compatible with other polymers synthesized from non-bio-oil sources. The comparison with conventional polymer network revealed that the bio oil can be successfully used without compromising the thermo-mechanical performance of the polymers.

GP-06. Influence of Biogenic nano Silica-amorphous Carbon hybrid on Thermo-Mechanical Properties of Biodegradable Polymer (Ecoflex)

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Purpose: Biodegradable polymers lead the packaging industry including food packaging, foam packaging, compostable bags for domestic applications etc., though these polymers have some inferior properties such as poor thermal stability, excessive brittleness, and poor melt strength and processability issues which restrict their wide range of applications. Nano phased silica a high temperature material can be used as a filler to improve the thermo-mechanical properties of biodegradable materials, which is also a natural sourced such as rice husk. Agricultural waste rice husk is a good source of silica, mostly used as energy source which creates CO_2 emission to the environment or dumped as a waste. Therefore, the prime aim of this work is to improve the structural properties of biodegradable polymer (Ecoflex) through nanocomposites approach by making micron thick Ecoflex/SCNPs (silica-carbon nanoparticles) thin films using 3D printing process for future targeted sensing application.

Methods: In this study, 3D printed flexible Ecoflex thin films integrated with biogenic silica-carbon nanoparticles (SCNPs), were investigated to determine the influence of the silica/carbon material on the thermal and mechanical properties of biopolymer. The high pressure reactor was used to synthesize silica/carbon nanoparticles and they were further characterized by X-ray Diffraction, Raman Spectroscopy, FTIR and TEM analysis. The composites were also characterized by DSC, TGA, X-ray Diffraction, Raman spectroscopy, TEM, SEM and Tensile analysis.

Results: X-ray diffraction and Raman analysis revealed the formation of crystalline cristobalite nano silica with amorphous carbon hybrid material. BET surface area measurement showed the highest surface area (706.23 m^2/g) of the nanoparticles. The TEM analysis of the composites identified the nanoparticles in the biopolymer while SEM proved the microstructure of the composites. TGA and Tensile test revealed significant enhancement in thermal stability, maximum strain and strain to failure properties due to the integration of 0.5 and 1.0 wt. % nano silica/carbon particles (SCNPs).

Conclusion: Agricultural waste rice husk is being used to synthesize valuable engineering materials silica/carbon nanoparticles and used as a filler material to enhance the thermo-mechanical properties of biopolymer**.** The nanocomposites approach shows a significant increase in thermal properties of polymer which minimizes the drawbacks of biodegradable polymers and thereby broadens their application areas.

GP-07. Novel Method For The Synthesis of New Polyaniline Derivatives And Their Potential Applications

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Purpose: Polyaniline exhibits many unique properties including electrical conductivity, radar absorption, antimicrobial activity and anticorrosive capabilities. Most research done on producing polyaniline derivatives has focused on improving processibility, often at the expense of its other properties. Development of new polyaniline derivatives in combination with its unique properties will expand the scope of potential applications.

Methods: Polyaniline is functionalized through a reductive addition mechanism. The common polyaniline emeraldine base is treated with aqueous oxidant to produce the fully oxidized pernigraniline base, providing an active substrate for the reaction. The pernigraniline base is then reacted with a 0.1 M solution of amine at room temperature. Addition of the amine nucleophile leads to reduction of the quinoid ring structure, lowering the overall oxidation state back to emeraldine. The product is collected by filtration and rinsed with ethanol to remove any residual amine.

Results: UV-Visible and FTIR spectroscopy confirm the reduction of the polymer backbone as well as the addition of the various amines. Through this method previously unreported derivatives including amines, alkenes, nitriles, aryl, carboxylic acids and alkyl groups have been created. These derivatives were also able to undergo reversible doping to the conductive state. Diamine functionalized polyanilines were used to cure epoxy resins, acting as a combination curing agent and functional filler. Several of the alkyl derivatives were utilized for corrosion protection of steel and provided a 200 mV anodic shift compared to the parent polyaniline.

Conclusion: The reductive addition of amines to the pernigraniline form provides a simple yet effective method for the derivatization of polyaniline. These derivatives create new potential uses for polyaniline as well as the opportunity for improvement in current fields of application.

GP-08. High-throughput screening methodologies for targeted melanoma drug discovery

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Purpose: Gain-of-function mutations in the ErbB4 receptor tyrosine kinase have been found in a significant fraction of melanoma cell lines that are dependent on ErbB4 for proliferation. However, there is a scarcity of therapeutics for treating these ErbB4-dependent tumors.

Methods: Consequently, we have developed high-throughput screening assays to identify small molecule ErbB4 antagonists that may hold promise as targeted melanoma therapeutics. Our approach is based on the observation that the Q43L mutant of the ErbB4 agonist Neuregulin 2beta (NRG2b) functions as a partial agonist at ErbB4. NRG2b/Q43L stimulates ErbB4 tyrosine phosphorylation, fails to stimulate ErbB4 coupling to cell proliferation, and competitively antagonizes agonist stimulation of ErbB4 coupling to cell proliferation. Therefore, we have developed three high-throughput assays to identify ErbB4 partial agonists that function as antagonists. The primary screen identifies molecules that stimulate ErbB4 tyrosine phosphorylation. The secondary screen identifies molecules that stimulate or fail to stimulate ErbB4-dependent proliferation. The tertiary screen identifies molecules that antagonize agonist stimulation of ErbB4-dependent proliferation.

Results: Our automated phospho-ErbB4 sandwich ELISA identifies molecules that stimulate ErbB4 tyrosine phosphorylation with high sensitivity and fidelity ($Z' > 0.5$). Our MTT assays using a cell line that displays ErbB4-dependent proliferation identify molecules that stimulate or fail to stimulate ErbB4-dependent proliferation ($Z' > 0.5$) and identify molecules that antagonize agonist stimulation of ErbB4-dependent proliferation.

Conclusions: The validated ELISA has identified 20 small molecule compounds that stimulate ErbB4 tyrosine phosphorylation. Efforts to determine whether these hits function as ErbB4 full agonists or partial agonists/antagonists are currently underway. ErbB4 partial agonists that function as ErbB4 antagonists may hold promise as targeted therapeutics for ErbB4-dependent melanomas.

GP-09. Quantitative *in situ* speciation of multicomponent transport through Nafion 117™ membranes

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Purpose: Polymeric membranes are an important technological platform for a broad range of applications. The main quantitative analysis used to characterize membrane properties, specifically solute permeability and membrane selectivity, have relied on single component transport experiments as multicomponent experiments have typically required arduously taking aliquots for analysis by *ex situ* chromatographic or spectroscopic techniques. Herein, we demonstrate the utility of *in situ* Attenuated-Total-Reflectance Fourier Transform Infrared (ATR FTIR) spectroscopy for determining the transport properties of multiple components permeating through polymeric membranes.

Methods: First, aqueous concentration standards of methanol and acetone were prepared and used to determine effective extinction coefficients at a series of wavenumbers via ATR FTIR spectroscopy. Multicomponent aqueous solutions were then prepared at varying concentrations and their ATR FTIR spectra were collected. Finally, permeability experiments were performed using a custom-built diffusion cell apparatus consisting of two jacketed cells, for temperature and evaporative control, separated by a fully-hydrated (>12hrs) Nafion 117™ membrane. One cell contains pure deionized water and the ATR FTIR probe and the other a one molar aqueous solution of the desired permeate(s). Spectra are recorded every 10 minutes for 48 hours.

Results: Effective extinction coefficients of methanol and acetone were determined from the aqueous concentration standards using Beer's Law. Utilizing the additive nature of light absorption, expressions are derived from Beer's Law and used to quantitatively determine solute concentrations in multicomponent static solutions from ATR FTIR spectra with high fidelity. The permeability of methanol and acetone, and the corresponding membrane selectivity, through Nafion 117™ membranes were determined from single component diffusion cell permeability experiments utilizing these concentration expressions under the assumption of a solution-diffusion transport mechanism and the method of Yasuda. Permeabilities and the corresponding membrane selectivity were also determined from analogous multicomponent permeability experiments.

Conclusions: We find ATR FTIR to be a quantitative and simple technique for the determination of solute concentrations in both single and multicomponent static solutions. Furthermore, ATR FTIR spectroscopy provides a new and facile route to characterize multicomponent transport without the need for time-consuming aliquots when adopted as an *in situ* technique for characterizing transport in diffusion cell permeability experiments.

GP-10. Interleukin-10 Encapsulated within Poly (lactic acid)-b-Poly (ethylene glycol) Nanoparticles is Functional by Down-regulating Cytokines and Inducing Suppressor of Cytokine Signaling 1 and 3 in Mouse Macrophages Exposed to *Chlamydia trachomatis*

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Purpose: Inflammation which is induced by the presence of cytokines and chemokines is an integral part of *Chlamydia trachomatis* (CT) infection. Nevertheless, it can be regulated using effective alternative therapeutics, including anti-inflammatory molecules. We recently reported that the anti-inflammatory cytokine, IL-10 can effectively regulate CT inflammation by inhibiting its associated-induced inflammatory mediators in mouse J774 macrophages. A major problem with IL-10 however, is its short biological half-life thus requiring frequent applications at high dosages for biomedical applications. Our goal in this study was to encapsulate IL-10 within the biodegradable polymer, PLA-PEG (Poly (lactic acid)-b-Poly (ethylene glycol) nanoparticles in an attempt to prolong its half-life.

Methods: IL-10 was encapsulated in PLA-PEG by the double emulsion method, followed by physiochemical characterizations and functional studies. The anti-inflammatory effect of encapsulated IL-10 was then tested using various concentrations (1-1000 ng/mL) over a 24-72 hour time-point in mouse J774 macrophages exposed to the recombinant major outer membrane protein of CT. We then used specific ELISAs (Enzyme-linked immunosorbent assays) to measure the production of pro inflammatory cytokines IL-6 and IL-12 in our cell-free supernatants and qRT-QCR to quantify the mRNA gene transcripts of SOCS 1 and SOCS 3.

Results: Data from Ultra Violet (UV) visible and Fourier Transform-Infrared Spectroscopy (FT-IR) revealed the successful encapsulation of IL-10 within PLA-PEG. Encapsulated IL-10 had an average size of ~ 100 to 200 nm, with an encapsulation efficiency > 90 %. Temperature stability of encapsulated IL-10 was up to 89°C as shown by differential scanning calorimetry analysis. Cytokine specific ELISAs showed that encapsulated IL-10 reduced the levels of both IL-6 and IL-12 in macrophages in a time- and concentration-dependent fashion, correlating with its stability and slow release capacity. By Taqman qRT-PCR, we further demonstrated that encapsulated IL-10 induced higher levels of SOCS3 as compared to SOCS1, independent of the encapsulated IL-10 concentration, suggesting their probable role in the encapsulated IL-10 molecular mechanism of inhibition.

Conclusion: Our data shows successful encapsulation of IL-10 and that PLA PEG can prolong the half-life of IL-10. More importantly, encapsulated IL-10 is functional by down-regulating cytokines and inducing SOCS1 and 3 in macrophages exposed to CT at relatively low dosages.

GP-11. Ultrastructure evaluation of boar spermatozoa following interaction with iron oxide nanoparticles

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Purpose: Semen ejaculates contain a mixed population of viable and non-viable spermatozoa. The presence and magnitude of a non-viable sperm cell population can negatively impact male fertility. Currently, various commercial techniques allow detection of non-viable spermatozoa (i.e., apoptotic membrane damages), but their selective removal from semen doses before artificial insemination remains unsuccessful. In previous studies, we used specifically designed magnetic nanoparticles to target and easily discard non-viable spermatozoa from semen doses. The goal of this study was to conduct ultrastructure imaging of labeled spermatozoa to confirm their interaction with nanoparticles.

Methods: Freshly prepared insemination doses (n=3) of boar semen were purchased from a local stud and were subsequently mixed with or without (Control) iron-oxide nanoparticles designed to target moribund or non-viable spermatozoa. Two successive incubations for subsequent removals of acrosome reacted and apoptotic spermatozoa through an electromagnetic field were performed. While nanoparticle-bound (moribund) spermatozoa were trapped by magnetism, unbound (viable or nanopurified) spermatozoa were collected by simple elution. From each semen dose motility characteristics were assessed by a computer assisted-sperm analyzer (CASA). Control, nanopurified, and moribund spermatozoa prepared from a single semen dose were subjected to transmission electron microscopy (Jeol TEM) imaging.

Results: Motility characteristics of spermatozoa were not significantly different between both control and nanopurified groups (T-test, $P>0.05$). The ultrastructure of spermatozoa was not affected by the presence of nanoparticles. In the moribund sperm group, TEM imaging revealed nanoparticles binding to the sperm plasma membrane with some displaying accumulations in the cytosol and nucleus. In contrast, only few sperm cells exhibited traces of nanoparticles in the nanopurified group, while the majority of cells did not.

Conclusion: The application of a magnetic nanopurification procedure revealed successful for massive targeting of moribund spermatozoa, without impairment of sperm motility. Nonetheless, the minor detection of nanoparticles in the nanopurified group indicates additional improvement is necessary to reach a total elimination of added nanoparticles from semen insemination doses.

GP-12. Effect of Peptide Nano-tag and Nanoparticles on MPER epitope of HIV exposed on Qβ exterior Surface

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HIV infection has been of great concern to public health with the recent estimate of 1.2 million people infected in the United States. There is a critical need for a safe and effective vaccine that can protect the uninfected. Studies have shown that the membrane proximal external region (MPER) represents one of the most neutralizing targets for HIV and mediates viral entry. We anticipate that the MPER will serve as an antigen to stimulate antibodies once presented on the platform of the Qβ phage. There is no stable commercial monoclonal antibody against MPER. We have fused MPER and a nano-tag for evaluation and detection. We designed the PCR template using oligos that represent a specific region of the 50 amino acid MPER. We performed the cloning by fusing the MPER motif with and without nano-tag in frame at the end of the A1 of our Qβ genome. The recombinant plasmid was used to transform *E. coli* HB101, and the binding of the phage exposing MPER motif on antibody was achieved using ELISA. Further analysis will be done using western blot, dot blot and EM to reveal the fusion of the MPER-A1 of the surface of Qβ. Through tedious EM and Cryo-EM imaging using Ni-NTA-Nanogold, we have determined the position of each MPER peptide on the surface of the phage. Also, analysis of antibodies from HIV patients with QβMPER using ELISA was successful and did not show a significant difference between both forms of phage. Future studies include animal immunization with QβMPER, and. We hope that the MPER-Qβ will serve as a vaccine candidate and as a standard for vaccine efficacy.

GP-13. Multi-Factor Role of Secretory Phospholipase A_2 and PLA_2 Receptor Expression in Altering Liposome Nanoparticle Uptake/Disposition and Tumorgenesis within a Prostate Cancer Model

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Purpose: The over-expression of Secretory Phospholipase A_2 ($sPLA_2$) in prostate and other cancers can be potentially exploited by phospholipid-based nanomedicines (liposome nanoparticles) to target and identify various grades of cancerous tissue and to initiate liposome degradation for effective release of chemotherapeutic drugs. The presence of a regulatory membrane receptor for $sPLA_2$ (PLA2R1) may also modulate enzymatic interaction with liposomes and their endosomal uptake. Understanding the impact of these proteins on liposome disposition and tumor microenvironment provides insight for improving nanomedicine delivery and therapeutic outcomes.

Methods: Flow cytometry and fluorescence microscopy were applied to evaluate the *in-vitro* uptake of $sPLA_2$ Responsive Liposomes (SPRL) and clinically-used Sterically Stabilized Liposomes (SSL). After modification with DiR, a near-infrared fluorescent lipid probe, formulations were dosed in metastasis-derived human prostate adenocarcinoma cells (PC-3) and a PLA2R1 knock-down variant (PC-3-PLA2R-KD) while varying supplementation of $sPLA_2$ enzyme isoforms (IIA, X, V). *In-vivo* disposition and efficacy of doxorubicin-loaded SSL and SPRL formulations in mice with subcutaneous prostate tumors were also investigated. IVIS Lumina XR-II and iThera enVision MSOT-256 devices provided non-invasive imaging for tracking DiR-liposome-nanoparticle biodistribution and monitoring tumor vascularization within mice following intravenous treatment.

Results: Within 48hrs of formulation dosing, *in-vitro* PC-3-PLA2R-KD cells demonstrated higher fluorescent intensity vs. control PC-3 cells. Liposomal uptake was also enhanced at early time points in cells supplemented with IIA enzyme isoform, which shows augmented expression in PLA2R-KD. More importantly, fluorescent signal indicated SPRL uptake is significantly greater than SSL ($p<0.05$) beyond 24hrs. During *in-vivo* studies, overall growth was limited in PLA2R-KD tumors, for which imaging reveals notable vascularization and enhanced deposition of both liposome formulations. Overall, SPRL targeted more efficiently across tumor types, and chemotherapeutic-loaded liposomes demonstrated efficacy towards inhibiting tumor growth.

Conclusion: Greater *in-vitro* uptake of SPRL nanoparticles and increased accumulation of SPRL to prostate tumor regions *in-vivo* demonstrate an advantage of the formulation for drug delivery. Elevated enzyme expression and activity caused by enzyme dysregulation in PLA2R-KD may be enhancing access of nanoparticles to the tumor site through vascular pathways and accelerating liposomal degradation and drug release. Ultimately, PLA2R1 provides a viable therapeutic target and is an important modulator of liposome nanoparticle drug delivery.

GP-14. Construction of Solid State Nanoreactor for the Synthesis and Characterization of Large Scale Metallic Nanoparticles

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Purpose: The current state of nanoparticles fabrication via polymer or solid state means is limited by the instability of the formed nanoparticles against self-aggregation and the ability to produce monodispersed nanoparticles on the macro-scale.

Methods: Hence, motivation for this project is to construct a solid state nanoreactor that uses multifunctional reduction methods (chemical reduction and photoreduction) to produce technologically relevant monometallic and multimetallic nanoparticles with controllable shapes and sizes and with specific designed cores and shell compositions at the macroscale "gram scale". In particular, this design will take advantage of polymeric means (Dendrimers and high temperature polymers such as PEI) to control the growth modes of CuNi, CoPt, and CuNiCo particles during formation.

Results: The resulting morphologies and structures of the formed nanoparticles were characterized by UV-vis, TEM, and XRD. Tests were conducted to establish the influence of its effect on biological systems.

Conclusion: Furthermore, TEM characterization of the formed nanoparticles were an average of 5 nanometers. The nanoparticles were proven to be non-toxic based on the results of the biological studies.

GP-15. Electrospun Mn-doped Titanium dioxide and Bismuth ferrite nanofibrous composite network: Effective photo-degradation in visible light

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Purpose: Titanium dioxide (TiO_2) is a semi-conducting material utilized for water treatment and environmental remediation via photo-catalysis. Photo-degradation efficiency of inorganic contaminants at or near the surface is dependent on material structure and irradiation. Furthermore, TiO_2 has optimal photo-catalytic behavior in the UV region (<5% of sunlight) due to a relatively large band gap of 3.2 eV.

Methods: Within the following study, fabrication of four electrospun samples including pure TiO_2, Mn-doped TiO_2, bismuth ferrite: TiO_2, and Mn-doped TiO_2: bismuth ferrite nanofibrous networks followed by pyrolysis. Pyrolysis was optimized to attain the ideal photo-catalytic behavioral phase of titanium dioxide, known as anatase. Subsequently, investigation of fiber morphology/ porosity distribution (SEM), structural characterization of existing phases (XRD), and photo-degradation efficiency of methylene blue under the influence of visible light (UV-vis spectrophotometry) was performed.

Results: Pure TiO_2 nanofibrous networks had a smooth surface texture morphology while the surface roughness increased in correlation with the introduction of metallic species. The samples consisting of bismuth ferrite had some agglomerations dispersed throughout fibrous network, while the Mn nanoparticles were dispersed on the surface and within the thickness of the fibers. The average fiber diameter distributions of TiO_2 network was 12.2 nm, while the Mn-doped TiO_2, bismuth ferrite: TiO_2 and bismuth ferrite: Mn- TiO_2 were 1.82, 2.4 and 2.9 microns respectively. XRD pattern shows that as metallic content increases, there is a yield in TiO_2 phase transformation from rutile to anatase following the heat treatment.

Conclusion: The optimal composition to improve photo-catalytic activity of TiO_2 in visible light requires introduction of Mn and bismuth ferrite to minimize the band gap. A balance between dopant/metallic component content without completely inhibiting the anatase phase of TiO_2 is necessary. The ideal form of the material should be within an entangled fibrous form due to the maximization of specific surface area and optical properties for effective photo-catalysis.

GP-16. Smart pH Responsive Nanocomposites Particles Integrated Contact Lens for Ophthalmic Drug Delivery

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Purpose: A number of ocular diseases, such as glaucoma, are typically treated with medicated eye drops, which provide sub-optimal therapy due to low bioavailability and poor patient compliance. Drug eluting contact lenses have been explored because of their potential to extend residence time in the post-lens tear film, which ultimately leads to a higher drug flux through the cornea and a lowering of the toxicity side effects.

Methods: In these studies, we present a novel device, engineered using a biocompatible, pH responsive nanocomposite material (silica/alginate NCs) integrated into a contact lens, for triggered drug release into the tear film. These pH responsive nanocomposite particles are synthesized using a water in oil microemulsion method and characterized by DLS, TEM, and TGA for their size, shape and alginate weight ratio.

Results: The size of nanocomposite particles range in diameters from 70 to 120 nm. TGA result confirms the encapsulation of the alginate polymer. These silica/alginate NCs show pH dependent release and incorporation of these particles into contact lens results in less than 5% loss in optical clarity.

Conclusion: Silica/alginate nanocomposite nanoparticles have been developed by sol-gel technique and characterized for size and composition. These spherical shape nanoparticles have pH responsive release properties to trigger release the loaded drug at pH 7.4. Contact lens which integrate these nanoparticles can be used to controlled ophthalmic drug delivery. Moreover, the integrated nanoparticles have little effects on the optical and mechanical properties of the contact lens.

GP-17. Examining the Potential of Phages to Avoid Endosomal Entrapment in MDA MB 231 Cells

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Breast cancer remains a disease of major concern, as it is one of the most common forms of cancer. Since untargeted treatments such as chemotherapy have harsh side effects, research has turned toward developing more targeted therapeutics. Phage display is a technique that screens a library of modified bacteriophages to identify those with an affinity for a particular subset of cells. The DNA of selected phages can then be sequenced and synthesized into a targeting peptide. One of the major obstacles preventing the efficacy of targeted therapeutics, particularly of small molecule drugs, is the tendency of the molecules to become trapped inside cellular endosomes and later degraded in cellular lysosomes. Through phage display technology, we hypothesized that a phage could not only target MDA MB 231 cells, a metastatic breast cancer cell line, but could also be "trained" to avoid endosomal entrapment. To select phages capable of endosomal escape, we treated MDA MB 231 cells with NH_4Cl to disrupt endosomes within the cells, and then used those cells for screening through four rounds of phage display selection and amplification. After selection, we will use the amplified phages to perform a binding assay to obtain a narrow our selection to those with the highest binding and endosomal escape potential. We will obtain the DNA sequences of these phages and synthesize peptides to later be used as the targeting component of a small drug therapeutic for metastatic breast cancer. The results obtained from this study will allow us to determine the potential of phages and their peptide derivatives to avoid endosomal entrapment and deliver therapeutics to the cellular cytoplasm.

GP-18. Role of zinc metal nanoparticles in olfactory sensory neuron signal transduction

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Purpose: Zinc metal nanoparticles enhance olfactory responses to odorant stimulation *in vitro*. These particles can increase responses by about 3-fold when presented to the epithelium in an odorant mixture, but they do not stimulate an odor response when administered alone. At these concentrations, effects are dose-dependent and reversible. They have demonstrated spontaneous elimination from the system after they are administered and induce their enhancing effect, therefore these particles could provide a specific, sensitive, and effective method of olfactory response control. These effects are specific to zinc metal nanoparticles, as copper, gold, and silver metal nanoparticles do not present the results observed for Zn. Gold and silver nanoparticles created a transient enhancement, while copper nanoparticles were ineffective. The purpose of our work is to investigate whether this odorant response enhancement by zinc nanoparticles observed *in vitro* is translated throughout the olfactory sensory circuitry to be perceived cognitively by the brain.

Methods: We utilize electroolfactogram (EOG) or whole cell patch-clamp electrophysiological recordings of isolated rodent olfactory epithelium responses to odorants in order to study the role of zinc metal nanoparticles *in vitro.* We then use noninvasive *in vivo* functional magnetic resonance imaging (fMRI) of awake dogs to investigate whether there is enhanced odor response of olfactory receptor neurons by zinc nanoparticle delivery leading to increased activity in olfaction-related and higher order brain regions.

Results: We determine that zinc nanoparticles function at the olfactory receptor level and are engaged in the initial events of olfaction. A kinetic model of olfactory receptor/odorant/metal interactions based upon experimental results described the stoichiometry of metal nanoparticles and receptors, as well as their mechanism of action. The kinetic olfactory model estimated that one metal nanoparticle binds two receptor molecules to form a dimer. Our canine fMRI results indicate that the addition of zinc nanoparticles results in a significant increase of brain excitation in response to odorants consistent with the increase in excitation observed in response to higher vs. lower concentration odorants at the epithelial level. The results from *in vivo* functional magnetic resonance imaging are consistent with those demonstrated *in vitro* using electrophysiology.

Conclusion: Zinc nanoparticle enhancement was seen in awake dogs, both young and mature cell cultures, and dissected olfactory epithelium. These results along with the fact that endogenous zinc nanoparticles are found in live animal blood reveals the significance of this enhancement for initial events in olfaction

GP-19. Novel Method for Synthesizing Janus Nanoparticles to Develop "Smarter" Nanostructures

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Purpose: Developing a method to produce Janus nanoparticles that will self-assemble into multiple complex structures. The hemispheres of Janus nanoparticles have different properties which can drive the self-assembly of monolayers, liposomes, and micelles. Potential uses for Janus nanoparticles included antimicrobial and/or water resistant coatings, drug delivery vehicles, and emulsion stabilizers to name a few

Methods: Janus nanoparticles will be produced by modifying the hemispheres of different nanoparticles (iron oxide or silica) with hydrophobic lipids, hydrophilic PEG derivatives, fluorescent dyes, and other smaller nanoparticles (iron oxide or gold). Dynamic Light Scattering (DLS) was used to determine the size of the Janus nanoparticles after formation as well as the size of the self-assembled structures. Fourier-transform Infrared Spectroscopy (FTIR) was used to verify the existence of the compounds on the nanoparticles surface. Transmission Electron Microscopy (TEM) and Dark-field/fluorescent Optical microscopy were used to visualize the hemispheres of the Janus nanoparticles. Fluorescent assays were used to quantify the hemispherical coverage ratio of the Janus nanoparticles for samples modified with fluorescent compounds.

Results: We have seen evidence from TEM and dark-field imaging of nanoparticles with side-specific modifications. DLS analysis has shown that we can control the self-assembly of Janus nanoparticles by changing the suspension solvent, and FTIR verified the presence of key compounds on the nanoparticles' surfaces. Optical microscope images and fluorescent assays have verified the ability to attach fluorescent molecules to one hemisphere of the nanoparticles and TEM has verified side-specific modifications on the nanoparticle surface.

Conclusion: We are able to produce different Janus nanoparticle configurations with side-specific modifications using multiple compounds. Multiple methods have been used to analyze and visualize these nanoparticles. The next step is to examine the structures that can be developed using these Janus nanoparticles and develop a nanoparticle configuration with real world application.

GP-20. Fabrication and characterization of cellulose nanocrystal derived lab-on-chip biosensors for early cancer detection

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Purpose: Cellulose nanocrystals (CNC) have garnered attention in recent years for their renewability, outstanding mechanical properties, tailorable surface chemistry, and relatively inexpensive price point. Biomedical applications for micro-electrochemical systems (MEMS) is a growing sector within the MEMS industry. Lab-on-a-chip microdevices have the potential to advance point of care (POC) medical applications and improve the quality of life for developing nations with poor healthcare infrastructure. Our work ultimately aims to test the feasibility of fabricating CNC derived MEMS for cancer detection by increasing the hydrolytic stability of CNC film, improving the electrical conductivity of the device, and evaluating protein immobilization on the device surface.

Method: A thin film of sulfonated CNC dispersion is sheared onto a patterned 4" single crystalline silicon wafer and cured at 100°C. The wafer is then treated with water plasma and submerged in an octyltrichlorosilane(OTS)/hexane solution for 1 hour at 32°C to deposit an OTS monolayer resulting in increased hydrophobicity of the CNC film. CNC derived micro-cantilever structures were fabricated using photolithographic process that utilizes a positive photoresist to imprint the mask design onto the substrate. Time-lapsed water contact angle measurements on the CNC film were made to determine the hydrolytic stability using a Ramé-hart Model 200 Standard Contact Angle Goniometer. Cross-polarized optical images were captured using a Nikon Eclipse 80i optical microscope to determine the uniformity of CNC alignment.

Results: Average water contact angle on OTS coated CNC film measured 107° (at t=0 minutes) and fell to 56.4° (at t=30 minutes). Overnight humidity exposure of CNC film resulted in a uniform film with no prominent defects in the monolayer structure present when viewed using optical microscopy. Furthermore, cross-polarized images indicate a great degree of birefringence indicating anisotropic alignment and ultimately increased mechanical strength in the film.

Conclusion: CNC films with improved hydrolytic stability were produced. Further work on process optimization is needed to validate hydrolytic stability post-release of MEMS devices. Next steps include evaluating CNC protein functionalization and cancer specific protein immobilization on the surface of micro-cantilever beams.

GP-21. Study on Thermal, Mechanical, Durability and Fire Retardant property of PHBV/HNT nanocomposite

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Purpose: PHBV is a biodegradable polymer with low thermal stability and poor mechanical property. But it is thermoplastic which is easy to process as well as biocompatible. Our goal is to improve the thermal stability, mechanical property, water resistance and fire retardancy of PHBV by incorporating a natural nanoclay as halloysite nanotube (HNT). At the same time maximum amount of HNT were investigated to obtain the optimum properties.

Methods: In this study we have used solution casting method to prepare the thin film. Chloroform was used as a solvent. In this whole process, ultrasonication of HNT into chloroform was followed by the magnetic stirring of PHBV to get the PHBV/HNT solution. The final thin films were characterized by thermogravimetric analysis (TGA), differential scanning calorimetry (DSC), fourier transform infrared spectroscopy (FTIR), universal tensile testing machine as well as scanning electron microscopy (SEM). Water absorption test was done into distilled water solution and fire test was done according to ASTM D635-10.

Results: The FTIR graph showed the decrease of peak intensity at 1030 cm^{-1}, 1113 cm^{-1} and 912 cm^{-1} which are representing the perpendicular Si-O stretching, apical Si-O vibration as well as the stretching vibration of Al-O/Al-OH bond. This is the proof of the HNT presence in PHBV matrix. TGA and DSC graph showed $7^{O}C$ and $10^{O}C$ improvement of thermal degradation as well as melting temperature for 10%HNT(optimum) loading compared to the 0% HNT loading respectively. Water absorption test was done for 315 hours which followed the Fickian distribution. It showed the improved water resistance of PHBV matrix with increased HNT loading. SEM image of fracture surface of resulting nanocomposite resembled good bonding between PHBV and HNT for 3 wt% which was investigated by the crack length. Fire test showed the improved barrier property for higher HNT loading.

Conclusion: The incorporation of HNT into PHBV matrix assists to enhance the thermal, mechanical, durability as well as fire retardant property. Good bonding between polymer and nanoclay ensures the improved property. Compatibility of nanoclay and polymer is a key factor to enhance the properties which can be done by nanoclay modification.

GP-22. Conversion of Egg and Seashell Waste Into Nanoparticles for Tissue Engineering

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Purpose: 76 billion eggs are consumed each year in the United States alone, resulting in 1.2 Million Kg of eggshell waste each year. Calcium based materials, such as Calcium Silicate, have the potential to lead to an array of scientific discoveries across numerous scientific disciplines do to their biocompatibility and natural porosity. The purpose of this study is to synthesize Calcium Silicate nanoparticles from egg and seashell waste that can potentially be used for the reminieraliztion of the dentin, bone regeneration, or scaffold design.

Methods: In these studies, we used eggshells and three different sources of seashells as natural sources of Calcium Carbonate, we then ball milled each source of Calcium Carbonate in a 1:1 molar ratio with amorphous Silicon Dioxide. After each sample was ball milled for approximately 100 minutes, we then sintered each sample to 1000 C and burned the samples at 1000 C for 1 hour before cooling down at room temperature. Once each sample was burned, they were then crushed in a ceramic mortar and then characterized with X-Ray Diffraction (XRD), Scanning Electron Microscope (SEM), and Energy Dispersive X-Ray Spectroscopy (EDS).

Results: XRD results showed that eggshells and the three different seashells sources that we used can be used to produce Calcium Silicate nanoparticles. SEM results indicate that we synthesized particles in the nanometer range along with EDS results showing that composition of our samples matched that of Calcium Silicate as well.

Conclusion: In conclusion, eggshell and seashells can be converted from waste into a valuable nanomaterial, Calcium Silicate. This method doesn't require the use of toxic and acidic chemicals and can be produced with industrially scalable methods such as ball-milling and sintering. Future studies will use these nanoparticles and incorporate them into either dental nanocomposites or scaffolds for tissue engineering.

GP-23. *In vitro* wound healing model using PLGA

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Wound healing is a natural process but there still remains major public health issues because burns/deep tissue damages either fail to, or slowly regenerate. Autografts are preferred for skin regeneration but at times are insufficient for severe cases. Skin graft substitutes are now providing significant improvement over traditional allografts and keratinocytes play an important role by proliferating and differentiating to restore the barrier and allowing re-epithelialization. We have encapsulated biomaterials within poly (lactic-co-glycolic acid) (PLGA) nanoparticle and shown it's non-toxic to eukaryotic cells *in vitro* and *in vivo*. Additionally we have shown that naringenin, a naturally occurring polyphenolic compound is non-toxic to cells and has anti-inflammatory properties. Here, we propose to develop a 3D wound healing model using natural polymers. Our 3D model includes a pre-established COCA cell line derived from the epidermis of mice, PLGA as the

extracellular matrix to support structure stability and flexibility, and collagen for binding between the polymer and keratinocytes. PLGA is biocompatible and biodegradable and lactate (a component in PLGA) promotes wound healing and vascularization. We are also incorporating reduced Glutathione (GSH), naringenin and N-acetyl cysteine (NAC) all known for cellular health to enhance the healing process. Our future studies include development of an in *vitro* wound model and application of the 3D skin substitute to reduce healing by continuous growth and differentiation. The release of biomaterials from PLGA will provide continuous benefits to the cells for regeneration. Our developed 3D polymeric skin model will be efficacious in accelerating wound healing and deep tissue damages.

GP-24. The Effect of Functionalized Cellulose on the Thermal and Mechanical Properties of ABS and HIPS-Reinforced Blends

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Purpose: An increase in plastic production has contributed to plastic waste being one of the largest municipal solid waste categories in industrial countries. Consequently, plastics leach brominated flame retardants (BFRs) into the atmosphere upon landfill disposal. Therefore, the need for more biodegradable plastic materials has arisen. As a result, labs across the globe have devoted many hours and research dollars to the development of techniques to reduce the impact of such hazardous waste on the environment by using reinforcement fillers, such as cellulose, to increase plastic biodegradability upon disposal. However, the extraction of cellulose from different biomasses is a rigorous process and, often, modification of its structure is needed to obtain the desired chemical and physical structural properties. Therefore, the objective of this research is to functionalize CreaTech cellulose to increase its hydrophobicity and uniform distribution within acrylonitrile-butadiene-styrene (ABS) and high impact polystyrene (HIPS)-matrix composites.

Methods: In this research, CreaTech cellulose was oxidized using the Albright-Goldman and Jones methodologies to increase its hydrophobicity and uniform distribution within a thermoplastic-polymer matrix. The polymorphic structure and crystallinity percentage of the cellulose was determined using XRD analysis, while FTIR analysis ensured the introduction of carbonyl-functional groups on the cellulosic structure. After chemical functionalization, the cellulose was incorporated into ABS-HIPS blends, where the thermal and mechanical properties were observed with DMA, DSC, and TGA analyses.

Results: TGA determined that a 50:50 ABS-HIPS ratio with 30 wt.% cellulose content was the most thermally stable. However, upon modification via the Jones oxidation methodology, an increase in the thermal stability was observed. Furthermore, DMA analysis proved an increased stiffness at higher cellulose loading contents, but a significant regression in the storage moduli with the incorporation of functionalized cellulose.

Conclusion: Therefore, modification of the cellulose improved the thermal properties, but showed a significant decrease in the mechanical properties. Future work includes determining the best functional groups to introduce onto the cellulosic structure that will increase the interaction between the cellulose and polymeric matrix.

GP-25. Synthesis of highly porous carbon from waste packaging material

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Purpose: Packaging waste is a huge environmental problem which needs immediate attention. Recent survey indicates that about one-third of all municipal trash is packaging waste. This waste can be recycled, or reused as a raw material for synthesis of value added products. In this project we studied the conversion of packaging waste materials into a highly porous activated carbon for water purification and polymer filler applications.

Methods: Highly porous carbon was synthesized from waste packaging material (foam) mainly made of water soluble starch and other biodegradable ingredients. We have carbonized this starch using thermal pyrolysis process in the presence and absence of the potassium hydroxide (KOH) chemical activation. This process was carried out in a batch reactor at high pressure and temperature conditions. The initial thermal degradation temperature was investigated through thermogravimetric analysis (TGA). Resulting biochar was characterized by XRD and Raman spectroscopy. The surface morphology was examined using field emission scan electronic microscopy (FE-SEM). *The surface area measurements were carried out using* Brunauer–Emmett–Teller (BET) experiments.

Results: XRD results show that the as prepared carbon is crystalline with slight amorphous background. The Raman spectroscopy experiments were also indicated the crystalline carbon. The BET results indicated that the carbon is highly porous with high surface area.

Conclusion: this packaging waste material is promising source for synthesis of porous carbon that can be utilized in different application such as water purification, polymer filler and electronic applications. The further studies in this direction are in progress.

GP-26. PolyAcrylic Acid Coated Iron Oxide Nanoparticles cause Reproductive and Developmental Damage in *Drosophila*

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Purpose: For over a decade, nanotechnology has been rapidly developing to include many broad-reaching medical and industrial applications. As different nanoparticles are engineered using different coatings, not much is known about the toxicity of these coated particles when they are ingested in the body and their interaction with tissues. Therefore, it is extremely important to study how each coating affects cells. Our work focuses primarily on Poly Acrylic Acid (PAA) coated Iron oxide nanoparticles (IONPs) due to their clinical relevance.

Methods: To study toxicity, we fed the 2nd instar larvae varying concentrations (0 µg/ml – 100 µg/ml) of PAA IONPS orally for 24 hours to carry out assays. At first, we carried out microscopy to test for cellular uptake of these particles. We then carried out larval mortality, pupation and eclosion by feeding the larvae for 24 hours and then scoring for their survival, percentage of pupation and eclosion. Longevity studies were carried out to test for any long term effects. Magnetometer analysis was used to test for the presence of Iron residues. We also carried out fertility and fecundity assay to see if there are any effects on their reproductive ability. We then carried out immunohistochemistry to look for any reproductive tissue damage. Also, we carried out hemolymph extraction and hemocyte counts to see if there is any immune activation.

Results: We found that low concentrations of PAA IONPs cause larval mortality, reduction in fecundity and reproductive tissue damage to ovaries. In addition, we also found that there is immune activation, in which Hemocyte numbers were elevated at IONPs concentrations above 10 µg/ml.

Conclusion: Overall, it is apparent that interactions of IONPs with tissues in whole organisms are IONPs concentration dependent. Surprisingly, very low concentrations appear to be more deleterious than higher concentrations. Understanding the mechanism by which these nanoparticles affect the tissues or the route for toxicity is critical for the evaluation of the therapeutic and diagnostic potential of these nanoparticles.

GP-27. Simultaneous Electrocatalytic Reduction of CO_2 and Photoelectrochemical Water Splitting: Routes Toward Efficient and Selective Electrocatalysts

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Photoelectrochemical water splitting and reduction of CO_2 into useful products are very challenging topics and have emerged as very attractive areas of research. In this work, photocatalytic anode material is combined with cathode catalysts made of a series of selective Cu nanoparticles and a Re(I) pyridyl N-heterocylic carbine complexes to demonstrate a low cost approach to reduce CO_2 selectively and water oxidation under sunlight. The catalytic performance, stability and selectivity was analyzed using cyclic voltammetry (CV), gas chromatography (GC) equipped with a mass spectrometry (MS). A rotating ring-disk electrode study was also done to provide the kinetics of the catalyzed reaction. This study will allow insight into improving the performance and selectivity of advanced electrocatalysts for CO_2 reduction and solar water splitting with optimal light absorption and conversion as well as efficient surface charge transfer kinetic rates.

GP-28. Nanoporous Graphene for Energy Storage Applications

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Purpose: Graphene is a unique material with a very high specific surface area, excellent conductivity and high tensile strength. Because of this, it can be considered as a futuristic material for energy storage especially in supercapacitors and possibly in hydrogen storage applications. The best method to produce graphene today is a solution based method through oxidation of graphite to graphene oxide (GO) by Hummer's Method. Reduced graphene oxide (RGO) with modified morphologies has shown potential application in energy storage. By creating nanopores within the RGO sheets, the specific surface area of electrode and diffusion of active species can be improved which further enhances energy density.

Methods: We present a simple solution based process to make nanoporous graphene from graphene oxide (GO) via microwave (MW) radiations. GO aqueous dispersion prepared by modified Hummer's method can be converted to RGO via thermal, chemical or photochemical processes. Further, RGO in presence of hydrogen peroxide is irradiated with microwaves. The MW generated OH• radicals oxidize the carbon atoms around active defective sites in RGO, leaving behind carbon vacancy, which extends to form the nanopores. This nanoporous conductive material is further investigated for electrochemical and diffusional properties.

Results: We were able to make nanoporous graphene with increased surface area which was confirmed by material characterization techniques like TEM imaging, UV-Visible spectroscopy, XPS and Methylene Blue surface area determination. Further, work is in progress to show electrochemical and diffusional properties of the material.

Conclusion: Graphene based materials are safe, non-toxic, and show unique properties to be used in energy storage applications. The nanoporous graphene have shown promising results and they have significant advantages over conventional materials.

GP-29. The effects of Carbon Nanotubes on plant growth and select Soil Microorganisms

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Purpose: As the nanomaterials such as carbon nanotubes are in high demand in today society, their used in industry is increasing. Nanomaterials such as multi-walled carbon nanotubes (MWCNTs) are increasingly being used in various industrial products. This increasing use will result in the release of the nanomaterials to the environment since there are no strict rules regulating their production, usage and disposal. Their potential effect therefore needs to be determined. In this study we investigated the effect of MWCNTs on *Phaseolus vulgaris* and soil microbes: *Mesorhizobium* sp., and *Nitrosomonas stercoris*

Methods: In this study, *Phaseolus vulgaris (*bush beans) were grown under hydroponic conditions. Two weeks after germination, the plants were exposed to different concentrations of dispersed multi-walled carbon nanotubes [0µg (control), 50µg/mL, 500µg/mL and 1000µg/mL]. The effects of the treatments were observed. Cultures of *Mesorhizobium sp*., and *Nitrosomonas stercoris* were also exposed to the same concentrations of dispersed multi-walled carbon nanotubes. The effects of the treatment were observed by incubating the samples in a BioScreen reader for 24 hours.

Results: Our preliminary results show that that at 50µg/mL, bean plants were able to tolerate the presence of the multi-walled carbon nanotubes whereas at 500µg/mL /mL of MWCNTs resulted in very poor growth and development and even plant death. There was growth inhibition of the microbes used in the study.

Conclusion: Our results suggest that concentrations of MWCNTs at 500µg/mL and above adversely affect plant growth as well as lowered the microbial biomass. This may serve as guideline in regulating the release of MWCNTs into the environment.

GP-30. Examining *in vitro* Cytotoxicity of Solid Nanoparticles

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Purpose: Solid nanoparticles have proven useful in overcoming many of the barriers to drug delivery. Additional development has seen the attachment of a myriad of surface coatings such as polyethylene glycol (PEG). This commonly used PEG layer masks particles from innate host defense mechanisms within the body and increases circulation half-life of the therapeutic upon injection. Although this approach is widely accepted, the underlying mechanism is not well understood.

Methods: Solid silica nanoparticles have been coated with 2k, 5k, and 20k Dalton PEG via EDC/NHS chemistry. Two nanoparticle sizes, 60 and 120 nm, were chosen to examine size dependent effects. Rhodamine b isothiocyanate was used for visualization of particle uptake with fluorescence microscopy and flow cytometry. Characterization of the functionalized materials was conducted with Fourier transform infrared spectroscopy, dynamic light scattering, and laser doppler electrophoresis. Chinese Hamster Ovary (CHO) cells were used for *in vitro* analysis and growth inhibition was determined using an enzymatic (MTT) assay.

Results: In vitro distribution of the silica nanoparticles was found to directly correlate with their surface coating. Pegylated particles were found to remain interspersed among the CHO cells, whereas amine coated particles were found to produce punctate regions of fluorescence on and around the cells. A similar trend was correlated with flow cytometry. It was found that all PEG coatings decreased particle uptake by the CHO cells in comparison to those with an amine coating. The

aminated particles had the greatest effect on cell growth. The 60 nm particles yielded greater half maximal inhibitory concentrations values over the 120 nm particles. Growth inhibition directly correlated with particle concentration regardless of surface character.

Conclusion: This study found that size and surface coatings have a dramatic influence on the cell viability and uptake in vitro. In addition, these studies show there may be an interplay between both of these variable sets. This study has provided a basis of comparison that could be extended to various nanoparticle systems. Understanding the impact of widely used solid nanoparticle compositions, sizes, coatings and their combinations on mammalian systems is an important step in developing robust drug delivery vehicles.

GP-31. Optimization and Control Studies on the Synthesis and Characterization of Epoxidized Soybean Oil (ESO)

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Purpose: The greenhouse gas emissions from petroleum based polymers makes unattractive options as there have been adverse influences in climate changes and weather conditions and often non-biodegradable when disposed. For these reasons, bio-based polymers are used to replace petroleum derived polymers that offer advantages of being renewable and biodegradable that improves sustainability, and economic and environmental concerns.

Methods: The current study focuses on synthesizing a high bio content resin via an epoxidation process using soybean oil. The epoxidation process uses hydrogen peroxide as an oxygen donor to form the epoxide groups in the presence of formic acid as the oxygen carrier. Studies were performed to determine an optimized system by analyzing the effect of temperature (at 50 and 60 ºC), reaction time (at 2, 4, and 6 hours), and variation of hydrogen peroxide content (at 1, 1.5, and 2 molar ratio). Initial characterization uses ASTM D1652-11 to determine the number of epoxide groups, Fourier transform infrared (FTIR) spectroscopy and Gas chromatography (GC) to analyze the change in functional groups and the formation of epoxy groups and provide a quantitative analysis of the number of double bonds, and Rheology to analysis the viscosity of each sample for use in various fabrication processes.

Results: Initial results revealed low OOC values at 2 and 4 hours due to the formation of side reactions that includes the presence of water and epoxy ring opening. FTIR spectroscopy revealed epoxide groups at 824 cm^{-1} only at 6 hours which revealed conversion of double bonds to epoxide groups. The total area percent in GC decreased with increasing time and increasing molar ratio of hydrogen peroxide, indicating the reduction of double bonds. Rheology results further verified results from the previous methods and indicated possible use of the optimized system in resin transfer molding (RTM) and filament winding.

Conclusion: The optimum system was achieved at conditions of 50 ºC for 6 hours using 2 mole ratio of hydrogen peroxide, achieving a 98% relative conversion to epoxide groups. The studies performed in this research resulted in stability and control for further use of the optimized system in fiber reinforced composites.

GP-32. Spray Deposition of Cu_2ZnSnS_4 (CZTS) for Cost-Effective and Non-Toxic Photoelectrodes

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The most direct and benign technology to capture abundant solar energy is the photovoltaic cell as long as the cell itself isn't made of harmful elements. In that regard, Copper, Zinc, Tin and Sulfide based Cu_2ZnSnS_4(CZTS) solar cell holds a great attention as the elements are earth abundant and environment friendly. However, CZTS solar cell lacks in power conversion efficiency to meet the grid parity. The research presented here focuses on improving the efficiency of CZTS photovoltaics through the incorporation of a mesoscopic hole acceptor, nickel oxide.

GP-33. Fabrication of Nanocrystalline Two-Stage Diamond Anvils for High Pressure Research Applications

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Single crystal diamond (SCD) anvils have been used for decades for the generation of high pressures and the study of materials. Due to the inherent yield point of single crystal diamond, these anvils are greatly limited as to what maximum pressure they can produce before experiencing failure. To increase the yield point of diamond anvils, a two-stage design has been implemented in which a second stage nanocrystalline diamond structure is grown in a selective region of the culet of the SCD primary anvil. Due to the higher yield point of nanocrystalline diamond, larger pressures can be generated without anvil

failure in a two-stage configuration because of the higher fracture resistance of the nanocrystalline second stage. The second stage is grown utilizing nanocrystalline growth chemistry in a chemical vapor deposition (CVD) system, and growth is localized by creating a tungsten mask via DC-sputter deposition and maskless lithography. Two stage anvils have been employed in high pressure experiments on osmium and tungsten, achieving a high-pressure region of 264 GPa without anvil fracture. This is a factor of four greater than the maximum pressure that the equivalent single crystal anvil absent of a second stage is capable of generating.

GP-34. Role of different sulfonic acid dopants on electrodeposition of polypyrrole and their effect on corrosion protection of carbon steel

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Purpose: The suitable system of dopants was studied to found out the best system for polypyrrole coating on carbon steel to suppress the corrosion and also helps us to obviate the use of toxic heavy metal primer in commercially available paints. The Elizabeth Lipke <eal0003@auburn.edu>dopants used were p-toluene sulfonic acid (p-TSA), sulfuric acid (SA), (±) camphor sulfonic acid (CSA) and sodium dodecyl sulfate (SDS) and sodium dodecylbenzene sulfonate (SDBS).

Methods: In this study we used electrochemical method (constant potential) to polymerize pyrrole and form a uniform coating over the steel surface to minimize the processing issues, control the film thickness. The linear weep voltammetry technique was used to test the coated samples for polarization and passivation behavior of steel surface in acidic dopants.

Results: It was found out that p-TSA and SDBS dopants are best for polypyrrole coating because of presence of extra benzene ring that helps in forming lamellar (sheet like) barrier to the aggressive chloride ion to enter the coating system. However, other dopants are also protecting the steel with significant shift in corrosion potential in positive direction.

Conclusion: The polypyrrole was synthesized electrochemically on carbon steel with four different dopants sub categorized in short chain (p-TSA, CSA and SA) and long chain dopant (SDS and SDBS). The potentiodynamic scan was performed on the bare steel sample to study the passivation behavior of steel in short chain dopant and found out lowest passivation potential have better coating characteristics and better anodic protection against corrosion. In this study, p-TSA and SDBS found to be best dopant for polypyrrole coating for carbon steel. This is due to presence of benzene ring that form π orbital overlapping resulting in π- π stacking which creates lamellar like structure which prevent the ingress of chloride ions in the coating system to the maximum. The outcomes of this work can be further used in combination of commercially available organic paint topcoat to have longer life and higher corrosion potential. Our system can also be used as replacement of toxic metal primer, which is hazardous to many living organism including human being.

GP-35. Utilizing LC-MS/MS to evaluate in vitro and in vivo intracellular uptake and degradation of $sPLA_2$ responsive liposome in prostate cancer

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Purpose: Secretory phospholipase A_2 ($sPLA_2$) are increased in various cancers. The lipid specificity and reactivity of sPLA2 and its ability to interact with PLA2 receptors (PLA2R) are potential targets for development of liposome drug delivery system. However, many phospholipids used to prepare liposomal formulations can be found endogenously and have biological isomers that complicate quantitative analysis. Thus, one challenge associated with the development and optimization of liposome nanoparticles is the difficulty to extract them from biological milieu and to distinguish liposome formulations and their metabolites from endogenous phospholipids.

Methods: In these studies, we incorporated deuterated lipids in our $sPLA_2$ responsive liposomes (SPRL) and developed an acidified Bligh-Dyer extraction method in combination with liquid chromatography tandem mass spectrometry (LC-MS/MS) to evaluate their intracellular uptake and degradation in prostate cancer. The ability to distinguish and quantify d70-DSPC from tumor samples was determined using a mouse xenograft model of human prostate adenocarcinoma (PC-3) cells implanted subcutaneously in athymic NCr (nu/nu) mice. The method was also used to examine uptake of different formulations using PC-3 “wild” and PLA2R knock-down (KD) cells.

Results: The LC-MS/MS chromatograms showed no interfering peaks from endogenous phospholipids with d70-DSPC and d35-LysoPC and had lower limits of quantification of 2 pg (S/N > 10) on column. Analysis of LC-MS/MS results showed an accumulation of SPRL in tumor based on the quantification of d70-DSPC and its metabolite d35-LysoPC. The uptake of liposome was coordinated to drug (doxorubicin) disposition. The d70-DSPC labeled SPRL formulation was also incubated with “wild” and PLA2R knock-down PC-3 cells for 48 hr. PLA2R knock-down resulted in a decrease in the uptake of SPRL

based on LC-MS/MS quantification. The observed decrease further supports the role of PLA2R in the intracellular uptake of SPRL.

Conclusion: This method strengthens the ability to evaluate and optimize lipid-based drug carriers such as liposomes. Such tools are critical to gaining mechanistic insights into the distribution and intracellular fate of nanomedicines.

GP-36. Evaluation of antibacterial activity of single walled carbon nanotubes

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Purpose: There is significant controversy in the literature over whether single walled carbon nanotubes (SWNT) have antibacterial activity and the mode of action of said antibacterial activity. There are conflicting results in experiments involving cytotoxic effects due to factors such as SWNT purity and functionalization, cell culture media, and cell type. The mode of action of the antibacterial activity is difficult to determine due to factors that affect nanoscale SWNT-bacterial interactions. Hypothesized modes of action in the literature include metabolic inhibition, oxidative stress and physical damage to the cell membrane. It is important to understand SWNT antibacterial properties to provide a foundation for possible future SWNT applications.

Methods: Antibacterial assays were performed using commercially produced SWNT-OH and SWNT that was acid functionalized, and tested against cultures of gram-negative Escherichia coli and gram-positive methicillin-resistant Staphylococcus aureus (MRSA) strain Xen 29. Each experiment was conducted in triplicate and was initiated by inoculating dispersed SWNTs into the respective log-phase bacterial culture within a tryptic soy broth (TSB) medium. The SWNT-bacterial suspension was incubated for 1 hour while slowly shaking at 15 rpm, then sub-cultured into TSB at a 10^{-1} dilution. The bacterial cultures were then incubated with shaking at 200 rpm and bacterial growth was monitored over time. In the case of E. coli, the optical density at 600 nm was recorded, whereas the MRSA strain Xen29 emits bioluminescence and this was recorded using a microtitre plate reader. Uninoculated TSB was the negative control and TSB containing an antibiotic that inhibited the respective bacterium was the positive control.

Results: The commercially produced SWNT was observed to have very poor dispersion while the oxidized SWNT produced manually was well-dispersed. Interestingly, the commercial SWNT-OH showed strong activity against both E. coli and MRSA, while the manually produced oxidized SWNT only showed strong activity against MRSA.

Conclusions: This experiment indicated that SWNTs prepared in different ways was observed to have antibacterial activity. However, it is important to conduct further experiments to determine the effect of different SWNT dispersion methods on SWNT antibacterial activity, and the mode of action of this activity in different bacterial species.

GP-37. Perfusion Studies of Ophthalmic In-Situ Nepafenac Gel on Porcine Eyes

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Purpose: Nepafenac is a nonsteroidal anti-inflammatory drug (NSAID) with low water solubility, currently only available in suspension form (Nevanac). Suspensions irritate eyes leading to limited residence time and low drug bioavailability. We successfully solubilized nepafenac using hydroxypropyl-beta-cyclodextrin (HPBCD) to improve its ocular bioavailability. We then incorporated the complex into an ion-activated gel formulation using sodium alginate, Protanal PH 1033, to reduce repeat administration and increase residence time.

Methods: Nepafenac was solubilized using HPBCD and a boric acid buffer. Gellation capacity studies comparing varying concentrations of HPMC to Protanal PH 1033, were done by dropping the formulation into simulated tear fluid (STF) and noting the time and duration of gelation. We determined the rheological properties of each formulation before and after the addition of STF. Diffusion studies across dialysis membrane and trans-corneal permeation studies across porcine corneas were performed using Franz diffusion cells on the nepafenac formulations with varying concentrations of Protanal using the commercial product as a reference. Perfusion studies across porcine eye balls were performed for the gel versus the suspension formulations.

Results: Gellation studies revealed that Protanal PH 1033 concentrations of 0.1% (F15), 0.3% (F16), and 0.5% (F17) all formed gels upon contact with the STF; however, F15 and F16 dissolved immediately while F17 remained in a gel form for more than 8 hours. Rheological studies revealed that the viscosity of all of the formulations doubled when exposed to the STF at 35°C, with F17 producing the highest viscosity. All of the formulations displayed pseudo-plastic flow as exhibited by the shear thinning profile. The permeation rate of all three in-situ formulations was approximately 14 times higher than the commercial product and had significantly higher amounts of nepafenac retained in the cornea when compared to the

commercial formulation. Perfusion studies revealed that the corneal concentration was not significantly different from the HPBCD solution; however, the amount of drug on the sclera was higher when compared to the suspension and solution, thus more drug is retained on the cornea.

Conclusion: An in-situ solution formulation, using 0.3% Protanal PH 1033, was created for the sustained release of nepafenac on the cornea.

GP-38. Macroscopic Control of Planar Helix in Cellulose Nanocrystal Suspensions for Improved Optical Properties in Films

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Purpose: The objective of this research is to control the chiral nematic helix in cellulose nanocrystal (CNC) suspensions during drying to obtain planar domains in the dried films. In planar orientation, the helix becomes perpendicular to the film surface giving rise to improved and uniform selective reflection of light on a macroscopic level. Circularly polarized light of same handedness with chiral nematic layers of CNC is reflected while light of the opposite handedness is transmitted. This phenomenon of selective reflection of visible light make CNC films a promising material for selective reflectors, band pass filters, optical sensors, security papers, and decorative coatings/display.

Methods: A novel method of producing CNC films from liquid crystalline dispersions was developed. The optical properties of the resulting films were measured using circularly polarized reflected light on a CRAIC Microspectrophotometer. Additional insights into the film microstructure and resulting optical properties were gained through cross-polarized optical microscopy and scanning electron microscopy.

Results: Circularly polarized light selective reflection spectra and scanning electron microscopy revealed excellent macroscopic planar alignment throughout the film. Compared to previous methods of film fabrication, domain sizes were increased from tens of micrometers to hundreds of micrometers. Domain shapes and sizes were found to be dependent on the initial dispersion concentration and shear flow. Also, planar defects called spatially varying pitch and/or nematic layer defect which cause phase jumps were also observed for the first time in planar CNC films.

Conclusion: Surface anchoring on dispersion surface facilitated the pinning of nematic layers of helical orders while slowed down capillary flow and orbital shear interplayed to give excellent control to planar helix giving intriguing self-assembly induced photonic films.

GP-39. *Chlamydia trachomatis* recombinant MOMP encapsulated within PLGA 85/15 nanoparticles triggers Th1 type cytokine response and enhanced co-stimulatory molecules in mouse dendritic cells

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Chlamydia trachomatis (CT), the leading cause of bacterial sexually transmitted diseases, is of public health significance because it causes considerable morbidity and socioeconomic burden worldwide. The quest for vaccine development against this pathogen has escalated to reduce its occurrence and global economic burden. Recently nanovaccines have emerged as attractive therapeutics due to flexibility in their formulation, biocompatibility and adjuvant properties. We have developed a nanovaccine against CT employing its MOMP (major outer membrane protein) and encapsulating it in PLGA [poly (D, L-lactic-co-glycolic acid) 85/15 nanoparticles. We hypothesized that encapsulated MOMP will be effective in activating dendritic cells (DCs) for induction of Th1 immune responses, which are needed for protection against CT. Exposure of mouse DCs to encapsulated MOMP at concentrations of 0.01 to 1 μg/mL revealed marked IL-6 and IL-12p40 secretions, with the latter being 20-fold more abundant. Encapsulated MOMP induced lower IL-10 and TNF levels with an elevated IL-12p40/IL-10 ratio, suggesting a predominant Th1 response. Time-kinetics studies (4 to 96 hr) showed increasing IL-12p40 concomitant with decreasing IL-10 concentrations. Analyses of CD11c+ DCs by FACS indicated heightened expression of the co-stimulatory molecules, CD80, CD86 and CD40 in response to encapsulated MOMP stimulation. TaqMan qPCR for mRNA transcripts validated the upregulation of TLR-2, CD80, CD86 and particularly CD40, which are important for adaptive immune responses. Our data suggest that encapsulated MOMP is a potential nanovaccine candidate by triggering enhanced co-stimulatory molecules and Th1 responses, which are required to protect against CT.

GP-40. Synthesis of biodegradable nanocomposites for vaccine delivery

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Purpose: Vaccines have saved more lives and prevented more human and animal suffering than any other single medical intervention. Many vaccination schedules require multiple visits to healthcare providers to receive a booster shot, which limits sustained protection due to patient non-compliance. Our goal is to overcome this limitation using a nanoparticle-based formulation that releases vaccines at predetermined intervals after a single injection - mimicking the effect of reimmunization (boosting).

Methods: Preliminary studies were carried out using GFP-expressing adenovirus. Adenovirus and adenovirus encapsulated in PLGA-chitosan nanocomposites were incubated at various dilutions with 293 Human Embryonic Kidney (HEK) cells in a 24-well plate, in order to quantify their activity. Fluorescence microscopy and ImageJ software was used to quantify the adenoviral activity in our samples. A spectrophotometric assay was developed to quantify the viral activity in the samples. The nanocomposites were characterized using Dynamic Light Scattering (DLS), Fourier Transform Infrared Spectroscopy (FTIR) and Scanning Electron Microscope (SEM).

Results: Viral activity in samples were successfully quantified using an assay based on bicinchoninic acid (BCA). DLS results indicate that nanocomposite particles had a size distribution ranging from 500 nm to 1500 nm and surface charge (zeta potential) was found to be +30mV, indicating the presence of chitosan on the surface. Further analysis by SEM provided evidence that nanocomposites can be synthesized with various morphologies (e.g. spherical, porous and hollow). Fluorescence microscopic analysis exhibited that the activity of adenovirus was preserved upon encapsulation.

Conclusion: We have successfully synthesized a biodegradable, hydrophilic, cationic nanocomposite with control of size over a wide range. The control of surface morphologies as well as the size of these composites will enable modulation of the release profile of the vaccine

GP-41. Physical characterization of zinc nanoparticles involved in olfaction enhancement

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Purpose: Previous studies have demonstrated that zinc metal nanoparticles, at low concentrations, can enhance electroolfactogram (EOG) or whole cell patch-clamp responses to odorants by about 3-fold (1). Zinc nanoparticles alone create no odor effects, rather increase the odor response only when mixed with an odorant. This study examined the physical characteristics of the zinc nanoparticle based on size, structure and oxidation status to assist in clarifying the mechanisms involved in the initial events of olfaction.

Methods: The zinc nanoparticles were prepared by the underwater electrical discharge method. The total concentration of metal in suspension was measured by atomic absorption spectra and the particle size and number determined by the atomic force microscopy. The crystallinity of small nanoparticles was analyzed by the Transmission Electron Microscopy (TEM). The degree of nanoparticle oxidation was obtained from the X-ray photon spectroscopy. Zinc nanoparticles were size-selected for small (1 nm to 2 nm diameters) and a subgroup oxidized by the percolation method. Oxidized zinc nanoparticles were also supplied by National Institute of Standards and Technology (NIST) at 15 nm and 70 nm diameters (2).

Results: Small (1.2±0.3 nm) zinc nanoparticles significantly enhanced electrical responses of olfactory neurons to odorants. Large and small oxidized zinc nanoparticles were not able to modulate responses to odorants. Zinc nanoparticles under TEM showed a crystalline structure. The calculated estimate of shell and core atoms was 12 atoms comprising the core and the total 59 atoms with the surface atoms of the 1.2 nm zinc nanoparticle account for 80% of all atoms, while the surface of the 15 nm zinc oxide nanoparticle contains only 10% of all atoms.

Conclusion: The enhancement seen with zinc nanoparticles is limited to small non-oxidized zinc nanoparticles capable of evoking considerable responses in olfactory sensory neurons only in the presence of odorant. The crystalline structure and high number of surface atoms found in combination with the atomic shell numbers of these small 1.2 nm zinc nanoparticles suggests properties of added stability and a high chemical reactivity.

GP-42. Using DNA aptamer-conjugated nanoparticles as cancer targeting and drug delivery vehicles

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Purpose: Lung cancer is the most diagnosed cancer with 1.8 million cases annually and accounts for ~20% of all cancer deaths worldwide. Small cell lung carcinoma (SCLC), one of the two lung cancer subtypes, is characterized by rapid growth and poor prognosis. Untreated SCLC is fatal within 2-4 months following diagnosis. Over the past 40 years there has been no significant improvement to the survival rate. Our objective is to synthesize, characterize, optimize, and investigate the control of drug release and cellular uptake of our drug-loaded nanocarrier towards selected SCLC cell lines.

Methods: 5' Modified anchor DNA was covalently bound to gold nanoparticles (AuNps) using the protocol designed by Mirkin et al. The anchor DNA sequence chosen has been shown to have a high binding efficiency to AuNps from previous work in our lab. A DNA aptamer molecule selected for targeting SCLC was engineered to contain drug binding sites using known doxorubicin DNA binding sequences. This cell-specific drug carrying aptamer will base pair with the anchor DNA bound to the AuNp. Our therapeutic carrier is designed for binding of intercalating agents only in the location of the double-stranded drug-binding sequences.

Results: Our lab has developed a highly tunable DNA-functionalized AuNp, which can bind approximately 800 molecules of daunomycin, while controlling the amount of anchors bound per AuNp. Engineering of the nanocarrier allows for regulating the interactions between the drug and its DNA binding sites, releasing in a controlled manner, as well as interactions between the cell-specific aptamer and its cellular target. *In vitro* viability studies have indicated that our nanocarrier is more efficient at killing cancer cells than free daunomycin at the same concentration.

Conclusion: The proposed research involves the first developed avidity-driven high payload programmable nanocarrier. The release rate and quantity of drug can be controlled by manipulating the sequence and length of the drug-binding region. Preliminary data indicate that our carrier will be able to deliver more drugs per particle than any other AuNp platform. Ongoing work includes an extensive analysis and testing efficacy of drug delivery platform in vitro and in vivo.

GP-43. $Ni(OH)_2$ as a hole mediator for visible light-induced urea photo-oxidation

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Purpose: Hydrogen is a potential substitute of fossil fuels due to its environmentally friendly and sustainable properties. Solar-driven method to produce hydrogen is a promising way as solar energy is the green and renewable energy source that excesses the human needs. Urea [$CO(NH_2)_2$], a major component of human urine, has been proposed as a source of hydrogen.

Methods: we used CdS-sensitized TiO_2 assembled with catalyst $Ni(OH)_2$ on fluorine-doped tin oxide (FTO) coated glass ($FTO/TiO_2/CdS/Ni(OH)_2$) as the photoanode for water reduction and urea oxidation under visible light irradiation. A photoelectrochemical reactor was employed to demonstrate that $Ni(OH)_2$ can serve as hole mediator to drive urea oxidation. The steady-state response of the semiconductor-catalyst electrode in the photoelectrochemical system was investigated, and the photoelectrode transient charge dynamics were studied by employing transient absorption spectroscopy.

Results: The spectroelectrochemistry reveals the change of $Ni(OH)_2$ to NiOOH in basic urea solution. The cyclic voltammetry shows that the $Ni(OH)_2$ is the photocatalyst of the urea oxidation. The hole transfer rate from CdS to $Ni(OH)_2$ and from $Ni(OH)_2$ to urea is 7.31×10^{11} s^{-1} and 4.15×10^{9} s^{-1}, respectively.

Conclusion: The $Ni(OH)_2$ can be a hole mediator for visible light-induced urea photo-oxidation. The obtained carrier dynamics can provide useful information on the competitive processes such as charge transfer and/or recombination within the photoanode, as well as on rational selection of suitable materials and structure design for photoelectrochemical system.

Poster Abstracts: Postdoc, Research Assistant, Faculty & Industry

OP-01. Branched amphiphilic cationic oligo-peptides for delivery of HPV-16 DNA vaccines

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Introduction: Peptide vesicles are emerging as an alternative gene delivery approach over lipid-based methods, featuring resistance to oxidation and thermal stability. We recently reported on a new class of branched amphiphilic peptides that self-assemble into extremely stable nano-spheres. The Branched Amphiphilic Peptides Capsules (BAPCs) display a uniform size of 20-30 nm and are resistant to detergents, proteases and chaotropes. In the presence of DNA, they can act as cationic nucleation centers around which DNA wraps. The BAPCs-DNA nanoparticles are capable of delivering plasmid DNA of different size into cells in culture, yielding high transfection rates and minimal cytotoxicity. Furthermore, BAPCs were tested for in vivo delivery of a DNA vaccine previously designed to activate immune responses and capable of controlling tumors induced by type 16 human papilloma virus (HPV-16).

Results and Discussion: BAPCs mixed with DNA form compact clusters with sizes ranging on average from 50 to 250 nm. Comparable to how histones compact DNA to form nucleosomes, BAPCs interact with plasmid DNA acting as a cationic nucleation centers with the negatively charged DNA coating the outer surface. HeLa cells transfected with the BAPCs-DNA complexes showed transfection rates approaching 55 % (higher than cells treated with Lipofectin). Furthermore, we tested the ability of the DNA-coated BAPCs to deliver DNA in vivo. For that purpose, we tested a DNA vaccine that encodes the HPV-16 E7 oncoprotein (pgDE7). Mice immunized with pgDE7-coated BAPCs at N:P of 1.3 managed to constrain tumor growth up to one month after transplantation of the TC-1cells. In addition, the survival time was enhanced by two-fold in comparison to that observed in the non-complexed DNA group without detectable acute toxicity.

Conclusion: Here we report the ability of nano-sized DNA-BAPCs to safely deliver plasmid DNA both in vitro and in vivo. In vitro, DNA-BAPCs nanoparticles transfected cells in culture with higher efficiency than that observed with a popular lipid-based commercial product and with less cytotoxicity. In vivo, they induce immune modulatory effects leading to enhancement of the anti-tumor effects of a DNA vaccine in a murine model.

OP-02. Functionalization of silver coated single walled carbon nanotubes with antimicrobial peptide: a novel covalent approach

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Purpose: Increasing antibiotic resistance is a global issue and the use of silver coated single walled carbon nanotubes (SWCNTs-Ag) and antimicrobial peptides (APs) is becoming popular due to their antimicrobial properties. However, toxicity to human cells and stability in a solution are some major issues with APs and SWCNTs-Ag, respectively. Our hypothesis is to functionalize APs covalently to SWCNTs-Ag and investigate their synergistic effect.

Methods: We first carboxylated SWCNTs-Ag using Tri sodium citrate (TSC) at 37 °C and then covalently functionalized them with an effective antimicrobial peptide from Therapeutic Inc., TP359 (FSWCNTs-Ag). Non covalent functionalization was also performed by simple mixing of TP359 and SWCNTs-Ag (SWCNTs-Ag-M). The characterization of the FSWCNTs-Ag (covalent) and SWCNTs-Ag-M (non-covalent), were performed by Fourier transform infrared spectroscopy (FT-IR), Ultraviolet visualization (UV-VIS), atomic force microscopy (AFM) and transmission electron microscopy (TEM). Also, the antibacterial activity of both and TP359 were investigated against two gram positive (*Staphylococcus aureus* and *Streptococcus pyogenes*) and two gram negative (*Salmonella enterica* serovar Typhimurium and *Escherichia coli*) pathogens and the cellular toxicity of TP359 and FSWCNTs-Ag was compared with plain SWCNTs-Ag using murine macrophages and lung carcinoma cells.

Results: FT-IR, TEM and AFM analysis revealed the successful functionalization of the peptide to the SWCNTs-Ag. More importantly, the present study results further showed that the minimum inhibitory concentration (MIC) of FSWCNTs-Ag were much lower (~7.8-3.9 µg/ml with IC50: ~4-5 µg/ml) compared to SWCNTs-Ag-M and plain SWCNTs-Ag (both 62.6 µg/ml, IC50: ~31-35 µg/ml). Also, FSWCNTs-Ag were non-toxic to the eukaryotic cells at their MIC concentrations (5-2.5 µg/ml) compared to SWCNTs-Ag (62.5 µg/ml).

Conclusion: In conclusion, our data showed that covalent functionalization of SWCNTs-Ag and TP359 exhibited an additive antibacterial activity. This study described a novel approach to prepare SWCNT-Ag bio-conjugates without loss of

antimicrobial activity and reduced toxicity, and this strategy will aid in the development of novel and biologically important nanomaterials.

OP-03. Identification of Novel, Prostate Cancer Targeting Peptides Using Phage Display Screening

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Purpose: The efficacy of chemotherapeutic drugs, as well as their safety profile, could be potentially enhanced dramatically through cell-specific targeting. Identification of ligands which bind to cell-specific surface proteins could enable diagnostic and therapeutic tools and also permit the investigation of receptor function. Phage display technology, with its high throughput capacity, has proven to be a useful tool for identifying targeting ligands, especially in the targeting of cancer cells.

Methods: We describe the use of a f8/9 landscape phage display library to identify a number of novel targeting peptides against metastatic prostate cancer cells (PC-3M). F8/9 phage display library underwent three rounds of selection against PC-3M. Selected phage clones were sequenced and extensively characterized for their binding affinity and selectivity.

Results: A total of 144 individual phage sequences were identified as binding to the PC3 cells. Assessment of phage selectivity and specificity for PC3M cell binding further reduced the number of candidate phage particles. A nuclear binding assay and confocal immunofluorescence microscopy further indicated that several phage particles achieve intracellular localization.

Conclusion: We identified new PC3M prostate cancer binding peptides that are expressed on the pVIII major coat protein of phage. A potential application of our findings is the use of these cell targeting peptides for targeted delivery of diagnostic or therapeutic agents to highly metastatic, PC3M prostate cancer cells. Ongoing *in vitro* and *in vivo* studies will confirm their utility for drug delivery.

OP-04. Mathematical model of actin cytoskeleton in kidney podocyte cells applied to glomerular disease.

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Purpose: Podocytes kidney cells morphology is essential for their function: the maintenance of the size selective filter in the glomeruli. Fully differentiated podocytes interdigitate their foot processes with neighboring cells, establishing a signaling hub for its actin cytoskeleton. Such cytoskeleton is composed of cortical actin and actin bundles, essential to sustain the local mechanical stress. Disruption of the actin cytoskeleton is linked to glomerular disease, such as proteinuria.

Methods: The analysis of a recent mathematical model for the actin cytoskeleton of the podocyte foot processes (http://biorxiv.org/content/early/2016/07/26/065839) is extended to better understand the interplay between regulators of actin (polymerization, bundling and turnover) and cytoskeleton stability. Recent literature review on podocyte actin regulators was performed to relate parameters of the mathematical model and podocyte proteins.

Results: The mathematical model predicts that global competition for freely diffusing monomeric actin may cause local cytoskeleton catastrophe and loss foot processes; prolonged imbalance among regulators may lead to progressive damage, while short lived imbalance would result in localized damage; hyperactive bundling competes with the machinery for cortical F-actin polymerization, and can be compensated by increased bundle turnover. Similarly, high bundle turnover (due to increased mechanical stress, for example) can be compensated by enhancing F-actin cross-linking; the cell morphology is resilient to mild spatial asymmetries in the F-actin polymerizing machinery (due to cell-cell interaction for example), however, significant disparities will result in localized enhanced bundles at the cost of the cytoskeleton of neighboring foot processes.

Conclusion: Quantitative methods and analytical and numerical models can be used to generate new hypotheses and potentially accelerate research on kidney disease.

OP-05. Forensic Drug Detection using Nanotechnology

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The purpose of this work is to develop novel methods in forensic drug analysis using nanotechnology and seek potential applications in forensic community. Although nanotechnology has made great contributions in medicine and energy science,

the applications of nanotechnology in forensic science are limited. The hypothesis of this work is that the infrared spectra of samples containing drugs and nanoparticles should provide the structural information of the drugs on nanoparticles. Here I will share some of the work on forensic drug detection in fingerprint samples using nanotechnology in my laboratory during the past three years. Our results showed that combination of nanotechnology and spectroscopy is a promising and informative tool for the detection of illegal substances cocaine, marijuana and benzodiazepines. Four types of fingerprint powders and four types of nanoparticles were used for preparing fingerprint samples. We found that the cocaine showed almost no IR signal in the presence of the fingerprint powders; in contrast, the IR signals of cocaine were observed in the presence of gold nanoparticle. It indicated that one is able to detect the forensic drugs in fingerprint samples using nanoparticles. Additionally, we found that the gold nanoparticles modified with peptide showed higher IR signals of cocaine than the native gold nanoparticles. We examined two procedures for detection of cocaine in the simulated fingerprint samples and found that both methods are effective. The chemical interactions between the cocaine and gold nanoparticle were investigated and summarized in a possible model. We further extended our effort to other forensic drugs, cocaethylene, norcocaine, ecgonine methyl ester, cannobinol, and flunitrazepam. In conclusion, the nanoparticles or functionalized nanoparticles is able to enhance the IR signals of forensic drugs and likely be used in forensic drug analysis of fingerprint samples. In the future, we intend to synthesis functionalized nanoparticles and investigate the potential improvement in detection of forensic drugs using these nanoparticles. The experimental data may provide useful and specific information in assessing whether the fingerprint suspect individual is an illicit drug dealer or even a drug abuser.

OP-06. Detecting Tumor Response to a Vascular Disrupting Agent Using Multispectral Optoacoustic Tomography

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Purpose: Vascular disrupting agents (VDAs) are a promising class of drugs that selectively disrupt the tumor vasculature, and induce tumor necrosis by preventing the delivery of critical nutrients. CA4P is a VDA that has undergone extensive clinical trials and has been evaluated in pre-clinical studies using multiple modalities. Its effect on tumor vasculature and oxygenation has been assessed by MRI and bioluminescence imaging, but these methods are time consuming and expensive. In this study we used Multispectral Optoacoustic Tomography (MSOT) to image the fraction (%) of hemoglobin (HbO_2), the fraction of deoxyhemoglobin (Hb), and tumor perfusion (DCE MSOT) in mouse model of breast cancer before and after treatment with CA4P.

Methods: Five nude mice were implanted subcutaneously with MDA-MB-231 breast cancer cells, and imaged by MSOT at baseline and at 4 and 24 hours after I.P. administration of CA4P (120 mg/Kg). Dynamic OA Imaging after injection of Indocyanine green (ICG,~80 nmol/kg) was performed by interleaving the acquisition of data after excitation at 800 nm (maximum for ICG) and 900 nm for 15 minutes. The MSP data was used to estimate the relative concentrations of Hb and HbO2. DCE MSOT data was used to estimate the area under the curve after of the OA signal (AUC). The Kruskal Wallis test was used to test equal distributions between time points for Hb, HbO2, and AUC.

Results: All tumors showed lower HbO_2 fractions and higher AUC that other tissues. Upon administration of CA4P, all tumors showed a 50 % decrease in HbO_2 fraction ($p \leq 0.016$), and a similar decrease in their AUC of DCE MSOT ($p \leq 0.032$). After 24 hours, the HbO_2, Hb, and AUC returned to their baseline levels ($p \geq 0.74$). The total hemoglobin was not significantly different among the three time points. These results are consistent with a decrease in perfusion after VDA administration.

Conclusion: MSOT can be used to detect response to VDAs after 4 hours therapy was started, using two complementary biomarkers of response, the % fraction of HbO_2 and the AUC of the DCE MSOT curve. These two biomarkers can be used to track two different aspects of the treatment.

OP-07. Evaluation of in vitro release of β-galactosidase from pH responsive nano-polymersomes as treatment for neurodegenerative disease

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Purpose: Currently, no treatment is available for patients with GM1 gangliosidosis, a neuropathic lysosomal storage disease in which β-galactosidase is not produced. Current methods of treatment for lysosomal storage disorders include infusions of free enzyme. However, this treatment cannot cross the blood-brain barrier and ameliorate severe central nervous system

symptoms. Encapsulating enzyme into targeted polymeric vesicles, called polymersomes, facilitates transport into the brain through IV injections, effectively extending therapy to the brain for the first time.

Methods: In these studies, polymersomes were created through polyethylene glycol-b-poly(lactic acid) (PEGPLA) self-assembly via solvent injection. Fluorescently tagged β-galactosidase (βgal) was loaded into lyophilized polymersomes, with loading measured via dialysis and spectroscopy prior to in vitro release in buffers created to mimic physicologic and lysosomal conditions. Apolipoprotein-E and CF350 attachment was facilitated through the introduction of a homobifunctional PEG strand during injection. A feline fibroblast line, GM1SV3, was used in in vitro studies to confirm particle uptake and treatment efficacy.

Results: PEGPLA polymersomes loaded βgal at 0.08 ± 0.03 mg βgal/mg polymersome with activity maintained at 452 ± 82 a.u. Fickian release of βgal was observed in pH 4.8, showing burst release expected in the lysosome. Limited release occurred in pH 7.4, demonstrating protection of βgal in the blood. CF350 Amine was attached to polymersome surface, confirmed by a diameter increase, microscopy, and 75 ± 28% of polymersomes labeled in flow cytometry. Attachment of ApoE was confirmed with both a diameter and protein content increase. GM1SV3 cells present low density lipoprotein receptors, indicating that they are a good model for Apolipoprotein-E mediated delivery. Apolipoprotein-E increases the uptake of polymersomes into GM1SV3 fibroblasts when compared to control polymersomes.

Conclusion: This work highlights our ability to consistently create polymersomes capable of simultaneously targeting and delivering βgal to the lysosome of neural cells using tunable chemistry. This combination of enzyme replacement therapy and nanotechnology demonstrates the capability of our carrier to transport enzymes to the brain while maintaining their activity and thus, will create a paradigm shift in the treatment of CNS disease, providing cures for currently untreatable and fatal diseases like GM1 gangliosidosis.

OP-08. The role of frataxin in anthracycline mediated cardiac hypertrophy

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Purpose: Doxorubicin (DOX) is a widely used anthracycline which has been used as a front line cancer chemotherapy regimen for decades. However the clinical application of DOX is limited due to its cumulative dose dependent cardiotoxicity which eventually leads to irreversible pathological hypertrophy transitioning towards heart failure. In recent years, research has attributed the cause of DOX cardiotoxicity to dysregulation in mitochondrial energy metabolism and the resulting damage due to overproduction of iron mediated intracellular reactive oxygen species (ROS). Thus the central focus of our study is to better understand at the molecular level, how deficits in mitochondrial energetics lead to cardiac hypertrophy. The present study investigates the role of frataxin (FXN), a mitochondrial iron-sulfur cluster biogenesis protein, and its role in development of DOX mediated cardiac hypertrophy.

Methods: We have treated 10 week old athymic mice with chronic DOX treatment (5mg/kg for 5 wks followed by 2 wks no treatment) to measure the development of cardiac hypertrophy as measured by MRI. In addition heart lysate and over expressing FXN and FXN knock down cells will be investigated for iron accumulation, mitochondrial respiration and mitochondrial energetics.

Results: We observed in DOX mice hearts a significant reduction in FXN expression leading to alterations in mitochondrial iron importer TfR1 and exporter ABCB8 expression, thus resulting in mitochondrial labile iron accumulation. Our results were confirmed in our FXN knock down (FXN-KD) cardiomyoblasts using FRET assay and a Ferrozine colorimetric assay. DOX treated and FXN KD cardiomyoblasts, via catalysis of fenton chemical reaction displayed increased mitochondrial ROS levels. However, over expression of FXN promoted mitochondrial iron homeostasis offering cardioprotection against ROS production and conferred significant protection against DOX mediated mitochondrial energy dysregulation as observed by improvements in aconitase, NADH dehydrogenase, basal oxygen consumption and ATP levels.

Conclusions: The current findings led us to postulate that FXN is primarily involved in DOX mediated cardiac hypertrophy and FXN over expression is protective against mitochondrial energy dysregulation. Our future work will determine the cardioprotective nature of FXN-OE mice against hypertrophy induced by DOX and pressure overload induced via Transverse Aortic Constriction (TAC).

OP-09. Fluorescence imaging of liposome nanoparticle interaction with boar spermatozoa

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Purpose: Production of transgenic livestock animals is of great interest for basic research, veterinary medicine, and both agriculture and biomedical applications. However, the current strategies to produce such animals are still inefficient. Amongst them, the sperm-mediated gene transfer (SMGT) has been described as one easy and rapid method that can be improved, and the use of liposome nanoparticles appears as a viable approach to increase transfection efficiency. As a first step to produce transgenic piglets, we investigated the toxicity of liposomes on boar spermatozoa.

Methods: Commercial doses of freshly harvested boar semen were centrifuged and sperm pellets were resuspended in PBS to $2x10^8$ spermatozoa/ml. Sperm aliquots were mixed with various concentrations of fluorescent liposomes (0, 91.4, 182.8, 457, and 914 M phospholipid basis) and incubated for 30 minutes at 37℃ under mild rotation. Afterwards, sample aliquots were subjected to sperm motility analysis (Computer-Assisted Sperm Analyzer –CASA-) and remaining cells were washed three times with pre-warmed PBS to remove the liposome excess. Fluorescence intensities of both supernatants (PBS) and sperm pellets were analyzed (In Vivo Imaging System –IVIS-). Sperm cells were subsequently fixed with 4% paraformaldehyde, smeared and mounted onto microscope slides, and observed under an epifluorescence microscope (EVOS FL-Auto). Experiments were repeated 3 times and data were analyzed with ANOVA-1. The threshold of significance was fixed at $P<0.05$.

Results: The proportions of motile, progressive, and rapid (≥30 μm/s) spermatozoa, as well as the velocity (average path – VAP- and curvilinear – VCL-) parameters were not affected by the liposome incubation. However, the straight-line velocity (VSL) and sperm linearity and straightness were dose-dependently increased ($P<0.05$). Fluorescence intensities of labeled samples using IVIS increased in a dose-dependent manner ($P<0.05$), with less or no detectable signals in supernatants derived of the third wash. Epifluorescence imaging confirmed the sperm-liposome interactions, with labeling generally found throughout the sperm.

Conclusion: Liposomes successfully interacted with boar spermatozoa in a dose-dependent manner, without altering the sperm motility. There was no observed toxicity effect, and the liposome nanoparticles applied in this study showed high potential in sperm transfection to produce transgenic animals.

OP-10. Development of graded materials and interfaces for periodontal tissue engineering

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Purpose: Periodontitis is a chronic infectious disease that causes destruction of tooth-attachment tissues and bone. Regeneration of functional periodontal tissues is feasible with the use of an ideal scaffold/membrane favoring the repopulation of regenerative cells residing in the defect or by scaffold-based cell-therapies. Both approaches need an ideal scaffold/membrane that prevent the growth epithelial tissues into the defect and favor the growth of periodontal ligament cells or stem cells into complex periodontal tissue.

Methods: The development of a new composite gradient-scaffold based on a physiological biomatrix (Biogel), biodegradable polymer and nanohydroxyapatite (nHA) (bone mineral). Toward this end, an innovative Biogel/nHA gradient in a continuous z-axis (throughout the thickness) is proposed to form as asymmetric scaffolds. Here, the bone-interfacing surface is rich in bone-mimetic physiological matrix milieu with high-density Biogel and nHA, while the ligament/connective tissue interfacing surface of the asymmetric scaffold is rich in polymer with low-density Biogel.

Results: Simultaneous electrospinning/spraying of the functionally graded membranes resulted in a nanocomposite tri-layer system averaging 452 (±17) μm in thickness. SEM images have shown the nanostructured morphology with randomly oriented fibers having nHA/gelatin particles embedded in the matrix. SEM image analyses indicated an average fiber diameter of 720 (±10) nm, 676 (± 20) nm, and 740 (± 20) nm for PDO+Maxon, PDO+Maxon+gelatin/HA, and PDO+Maxon with increased gelatin/HA layers respectively. Average particle sizes were measured to be 790 (± 25) nm and 820 (± 20) nm for the lower and higher HA/gel content layers, respectively. The mechanical properties and cell-response are favorable for barrier membrane application.

Conclusion: The "ideal" periodontal membrane must be clinically manageable, biocompatible, biodegradable, and sufficiently strong to avoid membrane collapse. We have fabricated new functionally-graded bioresorbable fibrous composite membranes with nHA gradient for potential use as a barrier membrane interface tissue engineering by promoting the bone growth and preventing the bacterial colonization.