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#### **RESEARCH SCHOLARS PROGRAM 2009**

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"The program has no set time limits. Research is a lifelong learning experience, and we hope to remain a resource to our students long after 'graduation'."





The Garcia Center for Polymers at Engineered Interfaces is a collaboration of eleven academic, industrial, and government laboratories. The Center was founded in 1996 and is named after the late Queens College professor, Narciso Garcia, a pioneer in the integration of education and research. The Garcia Center is funded by the National Science Foundation as part of its Materials Research Science and Engineering Center (MRSEC) program. The goal of the MRSEC is to combine the instrumentation and expertise of the participating institutions into a coordinated research program on polymer interface science. The principal focus areas include thin films, coatings, nano composites, self assembled structures, biomaterials, and tissue engineering.

These areas address both the fundamental and applied aspects that are relevant to the development of cutting-edge technologies in both engineering and medicine. In the community, the mission of the center is to serve as a valuable resource, providing easy access for technological assistance to educational and industrial institutions. For information on the numerous programs that are available, please see our web site at:

#### http://polymer.matscieng.sunysb.edu

The Research Scholar Program offers the opportunity for high school teachers and students to perform research on the forefronts of polymer science and technology together with the Garcia faculty and staff. Students work as part of focused research teams and are taught to make original contributions of interest to the scientific community. In addition to entering national competitions, the students are encouraged to publish in revered scientific journals and present their results at national conferences.

**Our goal** is to convey to the students the excitement we enjoy daily in research. The program has no set time limits. Research is a lifelong learning experience, and we hope to remain a resource to our students long after "graduation".

Miriam Rafailovich Professor, Garcia MRSEC Jonathan Sokolov Professor, Garcia MRSEC



# GRADS AND POST DOCS



























































# Michael Schiff







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onya Mrasad





# Learning & Speaking

















DENT





### Friday BBQ!!











nterfaces



#### Location: SAC Ballroom A

10:00-10:05

Welcome

#### **Miriam Rafailovich**

10:05-10:15



**Professor John H. Marburger III** Former Science Advisor to the President and Director of The Office of Science and Technology Policy (OSTP) during the George W. Bush Administration (2001-2009)

#### **Session I: Polymer Blends**

10:20—10:30 Chair: Max Plaut, *MIT* Miguel Pimentel, *Universidad Autonoma de Madrid* Anthony Oniwe, *SUNY Stony Brook* 

> Compatibilizing PMMA and EVA David Bender, Academy of Science and Technology

The Syntheis and Characterization of Ecoflex and Ethylene Vinyl Acrylate (EMA) Blends to Maximize Biodegradability and Cost Efficiency *Kyle O'Neil,* South Side High School; *Douglas Hoch,* Solomon Schechter High School

A Novel Procedure in the Creation of a Flame Retardant Polymer in the Nano-Macro- Scale with Focus in Biodegradability Mariah Geritano, Plainview-Old Bethpage John F. Kennedy High School

Nanopatterning with Self-assembled Polymer Masks on Silicon (Si) and Evaporated Gold (Au) Wafers

Evan Chen, Stuyvesant High School

#### Session II: Physiological & Biological Sensors

10:35—10:45	Chair: Suna Lee, SUNY Stony Brook Megan Garber, UC Berkeley
	<b>Optimization of Molecular Combing of Single-stranded DNA</b> Irsal Alsanea, Julia Budassi, Long Beach High School
	Biosensors and Molecular Imprinting: An Engineering Perspective Rohit Basavaraju, Jericho High School; Edan Elias, North Shore Hebrew Academy High School; Lina Lin, Plainview High School
	Engineering a 2D Potentiometric Biosensor for Protein Detection Naveen Murali, Staples High School; Nikhil Mehandru, Roslyn High School
	Digitizing the Moving Face During Dynamic Displays of Emotion Adam Ossip, North Shore Hebrew Academy High School
	Session III: Cells on Hydrogels
10:50—11:00	Chair: Isabelle Afriat, SUNY Stony Brook
	Pluronic F-127 Hydrogels: In-situ Gel Forming Systems for Tissue Engineering Andrea Barrientos, Lawrence High School
	The Use of Hydrogels and Electrospun Fibers as a Scaffold for Tissue Engineering Michelle Leonetti , Long Beach High School; Anirudh Nandan, Los Alamitos High School; Salonee Shah, W. T. Clarke High School
	Session IV: Alternative Energy
11:05—11:15	<ul> <li>Chair: Dr. Joanne Figuereido, Smithtown West High School</li> <li>Improvement of Titanium Dioxide and Dye Interface in Dye Sensitized Solar</li> <li>Cells</li> <li>Joy Jiang, Comsewogue High School; Judith Jacobson, Torah Academy for Girls</li> <li>High School; Aryeh Guterman, Hebrew Academy of Nassau County High School</li> <li>Characterization of Thin Film Morphology and Nanoparticle Confinement in</li> </ul>
	the Active Layer of Ordered Bulk Heterojunction Photovoltaic Cells Mubarrat Nuvid Bhuiyan, Jericho High School; Matthew Miecnikowski, Half Hollow Hills High School East

Understanding and Explaining Mechanisms in the Optimization of Fuel Cell Reactions Using Nanoparticle Catalysts *Alex Spangher, Lucas Spangher, Smithtown High School East* 

#### Session V: Enzymatically Synthesized Polymers

#### 11:20—11:30 Chair: Brandon Hsu, Johns Hopkins University Daniel Peng, Johns Hopkins University

Cutinase-Catalyzed Modification of Poly(ethylene terephthalate) Surfaces by Transesterfication and Transamidation Reactions in Aqueous and Organic Media

Daniel Chun, POB JFK High School; Michael Zhou, Jericho High School

Enzyme Stamping onto Polymer Interfaces Jonathan Nachman, North Shore Hebrew Academy High School; Sandy Guerrero, Longwood High School

#### **Session VI: Flame Retardant Polymers**

11:30—11:45 Chair: John Iraci, Cornell University

Simulating the Effect of Flame Retardant Materials on Heat Diffusion in Polymers Arpon Raksit, Commack High School

Creating Flame Retardant Polymer Blends for Application in Long Distance Cables Debby Leah Greenstein, SAR High School; Katelyn Marie Elizabeth Ireland,

Biodegradable Polymer Composites: Compatibility, Thermal Stability, and Flame Retardancy *Neil Muir, Uniondale High School* 

Effect of Organic Phosphate Oligomer on Roughness in Heat-Resistant Polymers Joong Gon (Kevin) Yim, South Side High School

Langmuir-Blodgett Trough-Assisted Graphene Synthesis Paul Masih Das, Lawrence High School

#### Session VII: Adult Stem Cell Differentiation

 11:50-12:05
 Chair: Jacqueline Belizar, SUNY Stony Brook

 Vladimir Jurokovski, Suffolk County Community College

West Islip Senior High School

The Effect of TiO2 and ZnO Nanoparticles on Dental Pulp Stem Cells Rida Malick, Lawrence High School

The Effect of Electrospun Polybutadiene Fibers on Dental Pulp Stem Cells Julie Chang, Yesha Maniar, Herricks High School

Substrate Induced Osteoblast-Like Differentiation of Stromal Stem Cells Reena Glaser, Matthew Hung, Smithtown West High School

Short and Long Term Stem Cell Lineage Determination: Use of Polymers/Copolymers with Various Mechanical Properties Benjamin Parnes, Michael Schiff, HAFTR High School

An Analysis of the Toxicity of Inert Metal Nanoparticles on Dermal Fibroblasts and Pre-adipocytes Ranjeet Kaur, Half Hollow Hills High School West; Kelsey McKenna, South Side High School

#### Session VIII: Blood Clotting Proteins

12:10—12:20 Chair: Alex Ramek, Harvard University Sara Snow, Yeshiva University

> The Effects of Glucose, Curcumin, and Biomineralization Solution on the Formation of and Development of Fibers from Fibrinogen with Respect to Topography, Modulus and Chemical Makeup.

*Katie Peyser,* North Shore Hebrew Academy High School; *Hannah Jin,* West Windsor – Plainsboro High School South

Characterizing Interactions between Fibrinogen Dimers During Fiber Self-Assembly: A Study of Biomaterials in Contact with Blood *Pooja Rambhia, Jericho High School* 

#### Session IX: Effects of Nanoparticles on Cells

12:25–12:35 Chair: Rafael Holzer, Yeshiva University

**The Effect of Iron Oxide on Cells and their Biomineralization** *Kimberly Lombardi, St. John the Baptist DHS* 

Effect of Zinc Oxide and Iron Oxide Nanoparticles on MC-3T3 cells Anisha Kapoor, Sonya Prasad, The Wheatley School

Investigating the Effects of Varied Concentrations of Titanium Dioxide on the Growth and Biomineralization of Osteoblast Cells Bracha Einzig, Talya Lent, Yeshiva University High School for Girls (Central)

#### The Effects of Titanium Dioxide and Zinc Oxide Nanoparticles on Adipocytes and Dermal Fibroblasts

Kayla Applebaum, Ma'ayanot High School; Alana Warhit, HAFTR High School

#### Session X: Cells Under Stress

#### 12:40—1:00 Chair: Alicia Franco, SUNY Stony Brook

Safety in Numbers?: Evaluating Cell Density's Role in Responses to Oxidative Stress

Sanjay Palat, Smithtown High School East

Evaluating the Effect of Cell Plating Density on UV Exposed Human Dermal Fibroblasts *Tiya Nandi, Jericho High School* 

The Effect of Magnetic Fields on Osteoblast Growth and Biomineralization Using An Iron Oxide-Clay Composite *Aaron Akhavan, Joseph Szpigiel, Rambam Mesivta* 

Whole Cell Patch Clamp Analysis of Human Cervical Carcinoma HeLa Cells Exposed to Gold Nanoparticles

**Tara Jain,** Moravian Academy Upper School; **Anjali Kapur,** Mount Sinai High School

Investigating The Effect of Platinum Nanoparticles in HeLa Cells Jonathan Millings, Emanuel Christian Academy

Phosphoibuprofen Administration and Quantum Dot Imaging Using Nanoparticle Vehicles in Cancer Cells Nevin Daniel, Ward Melville High School; Lucia Wang, Irvington High School

#### **Session XI: Filtration**

1:05—1:15	<b>Chair: Shuang Chen,</b> <i>SUNY Stony Brook</i> The creation of high flux nanofiltration membranes with interfacially polymerized polyamide on cellulose nanofibers <i>Matthew Kim, Commack High School</i>
	The Encapsulation of Alcanivorax borkumensis in a Gelatin Hydrogel for Water Filtration Applications
	<b>Ijeamaka Anyene, Lauren Herrera, Andrew Franco, William Genova,</b> Brentwood High School
1:15-2:00	Luncheon prepared by Wing Wan of West Hempstead Poetry reading: Veronica Collazo
	Musical Performance arranged by Garcia Students, arranged by John Jerome

## **MRSEC MEMORIES**

By Veronica Collazo



During this summer we did a lot, But there are some things that we forgot. So, before we start school again in September, Here are some moments that we should remember: Chemicals dropping on the stairs, And always forgetting to work in pairs. Trying to catch late night REUs, And, in Atlantis, breaking our shoes. Getting Miriam before she's gone, And finding Petri Dishes big enough to hold silicone. Working in labs all night thru, And hearing lectures in 201/102. Hurrying to get the sign-in sheet, And trying to stand the unbearable heat. Constantly avoiding Lourdes' 'Iron Fist', And attempting to stay of Mr. Sachs' 'List.' But there is one thing that we'll never forget The things that we learned here And how much they meant.

# Session 1 Polymer Blends

Session Chairs: Maxwell Plaut, MIT Anthony Oniwe, Stony Brook University, Miguel Pimentel, Universidad Autónoma de Madrid, Spain

Graduate Mentors: Seongchan Park,

Department of Materials Science and Engineering, Stony Brook, University , Stony Brook, NY **John Iraci,** Department of Mechanical Engineering, Cornell University, Ithaca, NY



#### **Compatibilizing PMMA and EVA**

David Bender, Academy of Science and Technology Seoung Chan Pack, Dr. John Jerome, Dr. Miriam Rafailovich Department of Material Science and Engineering, SUNY Stony Brook Maxwell Ethan Plaut, Jaclyn Schein - Massachusetts Institute of Technology John Michael Iraci – Cornell University

The ever expanding field of polymers needs new materials to work with. Making blends out of two different polymers in order to create new polymers with combined or new properties makes polymer research ongoing. This project is about creating a new polymer blend out of polymethyl methacrylate (PMMA) and ethyl vinyl acetate (EVA). A successful blend of these polymers might create a new polymer that has the strength of PMMA and the impact-resistance of EVA. The applications of this new blend are numerous and range from being a clear protective covering on solar cells, to being an impact resistant windshield. A main focus for this new blend is its optically clear properties, which any polymer that's optically clear is more useful. Along with its strength coming from PMMA and impact resistance coming from EVA, being optically clear will help this new blend become useable in a wide variety of applications. The polymers were combined under two different settings, thin film and bulk. Within thin films samples were spun with a blend of both polymers and then put through annealing and/or supercritical carbon dioxide. The bulk samples were brabended and then heat pressed before getting put through supercritical carbon dioxide. The thin film samples were analyzed using the atomic force microscope. The contact angle of these samples, which also helps determine compatibility, turned out to be at an average of 4° for all of the samples. Further analysis will determine how well these two polymers were compatibilized. The bulk samples were put through the dynamic mechanical analysis machine and the instron machine. The DMA measured the modulus while the instron measured the tensile strength. However, none of the bulk samples were optically clear. This occurred due to the phase separation of the polymers, therefore they could not have been compatibilized very well. Even though the polymer blend may not be optically clear, the physical properties themselves may allow this new polymer to become yet another useful invention of polymer science.



Figure 2



AFM close up image 1X1 without any annealing or supercritical CO2

 <sup>&</sup>lt;sup>1</sup> D. L.Tomasko, H. Li, D. Liu, X. Han, M. J. Wingert, L. James Lee, and K. W. Koelling. A Review of CO<sub>2</sub>Applications in the Processing of Polymers Department of Chemical Engineering, The Ohio State University, Columbus, Ohio Ind. Eng. Chem. Res. 42 6431-6456, 2003.
 <sup>1</sup> Fourman, M. Palermo, E., Lubin, S. Si, M. Rafailovich, M H. Sokolov, J C. Increasing the Compatibility of Polymer Blends using Supercritical Fluids (Abstract) APS March Meeting, (2004).

#### <u>The Syntheis and Characterization of Ecoflex and Ethylene Vinyl Acrylate</u> (EMA) Blends to Maximize Biodegradability and Cost Efficiency

Kyle O'Neil South Side High SchoolDouglas Hoch Solomon Schechter High SchoolDr. Miriam Rafailovich, Dr. John Jerome, Seoungchan Pack, Jason WangDepartment of Material Science and Engineering, SUNY Stony BrookMaxwell Plaut, Jaclyn ScheinMassachusetts Institute of TechnologyJohn Michael IraciCornell University

The world consumes approximately 100 million tons of plastic annually<sup>1</sup>, much of which has been estimated to take hundreds of years to degrade. In recent years advancements in the area of biodegradability have been made. One such biodegradable copolymer is BASF's Ecoflex. Biodegradeable and ecotoxilogical tests have shown that when Ecoflex is exposed to micro-organisms in soil (ex. *Photobacterium phosphorum* and *Daphnia Magna*)<sup>2</sup>, over 90% of it biodegrades into CO<sub>2</sub> after only 80 days<sup>3</sup>. However, polymers and copolymers that exhibit such unique properties are relatively expensive to manufacture. This unfortunate fact has led to increased research of polymer and copolymer blends in the hope of synthesizing new polymers that contain strong properties while also remaining cost efficient. With this idea in mind, we decided to combine Ecoflex with EMA, a cheap, non-biodegradable polymer with otherwise similar molecular properties to those of Ecoflex.

Throughout our experimentation three ratios of Ecoflex to EMA were used: 1:1, 3:1 and 1:3. These ratios were also made including 5% of three types of nanocomposites: Cloisite 20A, Cloisite 30B and RDP clay to further the cohesion of the blend. The samples were also exposed to supercritical CO<sub>2</sub> to further compatiblize the two copolymers. The mechanical properties (ultimate tensile strength {UTS}, proportional limit, yield stress and toughness) of each sample were calculated using an Instron. Individual thermal properties such as glass transition temperature, melting point, tandelta and E' were found using Dimensional Mechanical Analysis (DMA) and Differential Scanning Calorimetry (DSC). Ellipsometry was used to compute the sample thicknesses. Finally, with Atomic Force Microscopy (AFM), the samples were visually mapped and the contact angle and height of each trial was obtained.

It was found that the addition of clays caused the toughness of the samples to increase but to also become softer (Figure 1). After the samples were exposed to supercritical  $CO_2$  (ScCO<sub>2</sub>) the thickness of each blend increased by over 100% and some samples foamed (Table 1). The foaming skewed the Instron data because swelling induced the formation of bubbles within the polymer acting as an artificial crack. Therefore, Instron readings could not be taken from the samples that were exposed to ScCO<sub>2</sub>.



<sup>&</sup>lt;sup>1</sup> "Plastics recycling information sheet ." *wasteonline.org.uk.* N.p., Feb. 2006. Web. 10 Aug. 2009 <http://www.wasteonline.org.uk/resources/InformationSheets/Plastics.htm>.

<sup>&</sup>lt;sup>2</sup>Bastioli, Catia. Handbook of biodegradable polymers. Ed. Catia Bastioli. N.p.ISmithers Rapra Publishing, 2005. Print.

<sup>&</sup>lt;sup>3</sup> BASF. "Ecoflex- the entirely biodegradeable plastic." plasticportal.net. BASF, 2002. Web. 10 Aug. 2009.

<sup>&</sup>lt;a href="http://www.plasticsportal.net/wa/plasticsEU/">http://www.plasticsportal.net/wa/plasticsEU/</a> portal/show/common/content/campaigns/Ecoflex\_presentation/english/index.htm>.

#### <u>A Novel Procedure in the Creation of a Flame Retardant Polymer in the Nano- Macro-</u> <u>Scale with Focus in Biodegradability</u>

Mariah Geritano, Plainview-Old Bethpage John F. Kennedy High School, Plainview, NY 11803 Maxwell Plaut, Jaclyn Schien, Massachusetts Institute of Technology, Boston, MA 02210 John Michael Iraci, Cornell University, Ithaca, NY 14850 Dr. Miriam Rafailovich, Dr. John Jerome, Seoung Chan Pack, Department of Materials Science &

Engineering, State University of New York at Stony Brook, Stony Brook, NY 11794

Throughout the engineering community, the need for biodegradeable flame retardant polymers with enhanced tensile and mechanical properties has grown exponentially within the past decade.<sup>1</sup> "In the past 20 years, the production and use of plastics in the world have enormously increased, worsening the problem of waste disposal". <sup>2</sup> These new polymers not only have to be able to withstand immense heat and pressure and biodegrade in a sufficient manner, but they must also be cost efficient.<sup>3</sup> Many polymer blends have found to be immiscible, leading to materials with a poor interfacial adhesion, and ultimately, poor mechanical properties. <sup>4</sup>The novel technique implemented in this procedure allows for a cheap and productive method for creating a flame-retardant, biodegradable material. This experiment aims to create polymer composites using the copolymer Ecoflex and RDP clay in macro- and nano- scales.

In the thin film portion of the experiment, spin curves were produced with varying ratios of Ecoflex and Ecoflex/ RDP solutions. An Ellipsometer was used to measure the thicknesses of the films; to view the surface characteristics of the polymer thin-films, an Atomic Force Microscope was utilized. The samples then underwent the prior procedure after being annealed for 108 hours. In the macro sector of experimentation, 5% and 10% RDP-clay and Ecoflex mixtures were created, and tensile tests along with Dynamic Mechanical Analysis (DMA) were performed in order to juxtapose the thin film properties to the bulk blend properties of the clay mixtures.

Atomic Force Microscopy concluded the presence of phase separation within the copolymer through a direct relationship; as concentration of the thin-film rose, the phase separation found throughout the polymer drastically increased (Scheme 1). The annealed samples (Scheme 2) demonstrated the copolymer at equilibrium, showing that Ecoflex can be flattened, producing a malleable, flat surface which can be implemented in consumer products. In future research, analysis of the Ecoflex/RDP samples will be explored further as well as the phase separation between the polymers found in Ecoflex. Analysis of the biodegradability will also be done through the use of a bacterial testing method.



1. Cireli, Aysun, Nurhan Onar, Faruk Ebeoglugil, Isil Kayatekin, Bengi Kutlu, Osman Culha, and Erdal Ceilk. "Development of Flame Retardancy Properties of New Halogen-Free Phoshphorous Doped SiO2 Thin Films on Fabrics." *InterScience* 105 (2007): 3747-756.

2. Avella, Maurizio, Jan J. De Vlieger, Maria Errico, Sabine Fischer, Paolo Vacca, and Maria Volpe. "Biodegradable Starch/Clay Nanocomposite Films for Food Packaging Applications." *Science Direct* (2004): 467-74.

3. Quede, A., B. Mutel, P. Supiot, C. Jama, O. Dessaux, and R. Delobel. "Characterization of organosilicon films synthesized by N2-PACVD. Application to fire retardant properties of coated polymers." *Science Direct* (2003): 180-85.

4. Ray, Suprakas, and Mosto Bousmina. "Role of Organically Modified Layered Silicate Both as an Active Interfacial Modifier and Nanofiller for Immiscible Polymer Blends." (2007): 1-23

#### Nanopatterning with Self-assembled Polymer Masks on Silicon (Si) and Evaporated Gold (Au) Wafers

Evan Chen, Stuyvesant High School, New York, NY 10282 Raul Romero, Dr. John Jerome, Suffolk County Community College, Selden, NY Anthony Oniwe, Dr. Miriam Rafailovich, Stony Brook University, Stony Brook, NY

The manipulation of surface topographies on the nanoscale can generate surfaces with unique chemical and physical properties. Potential applications include the fabrication of antireflective layers, hard disks, templates for circuits, and surfaces for cell growth and differentiation.<sup>1</sup> Current methods in creating such nanopatterns include the use of conventional photolithography methods developed by the semiconductor industry, reactive ion etching, and block copolymer lithography.

We explore a different approach to creating such nanopatterns using a sputtering technique developed at this lab.<sup>2</sup> It involves the bombardment of surface with argon ions to imprint a pattern on the substrate. The approach is cheaper, simpler, and faster than other processes. It does not require plasma or specialized polymers, and the equipment is readily available for commercial use.

To create our patterns, we rely on the phase separation morphology of our polymer blends of polystyrene (PS), poly methyl methacrylate (PMMA), and ethyl vinyl acetate in different combinations and concentrations. These polymers are immiscible, so they phase separate easily, generating the template for our nanopatterns. Upon sputtering, the template is transferred to the substrate, which in our case was either silicon or gold.

Before patterns were created, we found the sputtering rates of our samples of PS, PMMA, and EVA at concentrations of 30mg/mL and 21mg/mL in toluene. We did this again with our blends of PS-PMMA (figure 1), PS-EVA (figure 2), and PMMA-EVA. Once our rates were established, we used them to predict the length of time required for the argon ions to remove the polymer layer and etch the silicon and gold on our later samples. These patterns were later analyzed under the AFM and the SEM for the contact angles aspect ratios, and fidelity of the various features in the nanopatterns.



Figure 1: AFM image of annealed PS-PMMA sample.



Figure 2: SEM image of a sputtered PS-EVA sample.

References

- 1. M.E. Stewart et al. *Unconventional methods for forming nanopatterns*. Proceedings of the Institution of Mechanical Engineers, Part J, Nanoengineering and Nanosystems p220 (2006) 81
- 2. J. Jerome et al. *Phase Segregation of Thin Film Polymer Blends on Au Nanopatterned Si Substrate*. Macromolecules p37 (2004) 6504-6510

# Session 2

# Physiological and Biological Sensors

Session Chairs: Sunah Lee, Stony Brook University, Megan Garber, University of California at Berkeley Rima Patel, Carnegie Mellon University

Graduate Mentors: Dr. Yantian Wang, Tatsiana Mironava Department of Materials Science and Engineering, Stony Brook, University, Stony Brook, NY



#### Optimization of molecular combing of single-stranded DNA

Irsal Alsanea<sup>1</sup>, Julia Budassi<sup>1</sup>, Sunah Lee<sup>2</sup>, Eli Hoory<sup>2</sup>, Dr. Jonathan Sokolov<sup>2</sup> 1 Long Beach High School 2 Stony Brook University

In this work, we focus on DNA deposition onto Poly-methyl methacrylate (PMMA) coated silicon subtrates. The goal of this work is to engineer optimal conditions and methods to improve resolution of biomolecular detection of DNA on a surface for future applications in genomics, proteomics, genetic suppression of viral and microbial processes, and DNA-protein interactions. The samples are dipped into solutions containing varying DNA concentrations (0.5, 1.25, 5.0, 12.5, & 25.0  $\mu$ g/ml). The  $\lambda$ DNA is diluted in a 6:50 solution of 0.1M NaOH: .02M MES (2-(N-morpholino) ethanesulfonic acid). After soaking in the solutions for times ranging from 30 sec. to 300 sec., the samples are retracted at varying rates (5, 10, 200, 250, 500, 1000, 2500, 5000, 8000  $\mu$ m/sec). DNA molecules can be immobilized either by attaching one end

of the DNA to the surface while stretching the molecule by pulling on the other end with optical or magnetic tweezers, or by a fluid flow concept utilizing a meniscus that would permanently attach the whole molecule onto the surface in a stretched configuration<sup>3</sup>.

PMMA (MW250kg/mol) film of 500-800Å thickness are deposited onto polished Si substrates by spin-casting from toluene solutions. Samples are annealed for 60min at 105°C in a vacuum oven prior to DNA deposition. Fluorescent labeling of DNA is prepared by mixing a 25µg/ml concentration of λDNA (Biolabs<sup>©</sup> E. coliλ cl857 ind1 Sam 7) solution with 7.5µl of 0.1mM YOYO-1 dye (Molecular Probes). Thereafter, the DNA and dye are incubated at 45°C for 90min to promote intercalation of the dye. Single-stranded DNA is produced by heating the solutions to 95°C for 10min, followed by a snap cooling in ice-water. In post-soaked samples, where dye dipping occurs after DNA dipping model.



Fig. 1. Sample 5 of July 30, 2009 exemplifying isolated strands of ssDNA under ideal conditions of 1% DNA at 5000µm/sec with a 30sec soaking time. A significant amount of immobilized isolated strands of DNA are 20µm.

soaked samples, where dye dipping occurs after DNA dipping, most strands recoil or are undetected. Optimal ssDNA length in a 25µg/ml dilution with 242.5bp is 20µm with a 10X magnification of the confocal microscope using a mercury lamp light source. A 5µg/ml λDNA dilution at a dipping rate of 5000µm/sec and a soaking time of 30sec shows a significant number of immobilized isolated single molecules under aforementioned conditions with little or no overlapping DNA (Fig. 1). An immobilized molecule with a stretched length of approximately 20µm would eliminate the overstreching (>20µm) and coiling (<20µm) factors. Longer and overstreched strands are more prevalent at slower Si-chip retraction speed (5 & 10 µm/sec), whereas faster retraction speeds (1000-8000 µm/sec) produce a greater number of isolated DNA molecules that are ~20 µm in length. Increasing DNA concentration and dye soaking time yield a greater abundance of DNA molecules in a given field of view. The soaking time in solution before extraction significantly affects the density of adsorbed DNA, with 30sec soaks showing moderate density (low overlap of molecules as in Fig. 1), while 300sec soaks producing densities too high to enable analysis of individual chains. A simple metrics comparison of the efficiency of combing under different conditions will be used to measure stretched DNA with the formula: Q=(number of DNA molecules)/(area of field of view[µm²] x DNA concentration [pM])<sup>1</sup>.



Fig. 2. Molecular combing with dipping machine.Fluid flow method of immobilizing DNA on a silinized chip with a meniscus acting as the stretching force for DNA combing.

1 Kim, Ji H, and Wei-Xian Shi, and Ronald Larson. "Methods of stretching DNA molecules using flow fields." <u>Langmuir</u> 23 (2007): 755-764. 2 Zammatteo, N, and L. Jeanmart, S. Hamels, et. al. (2000). Comparison between different strategies of covalent attachment of DNA to glass surfaces to build DNA microarrays. Analytical Biochemistry, 280 pp. 143-150. 3 Hansma, H G, and D Laney. "DNA binding to mica correlates with cationic radius: assay by atomic force microscopy." Biophysical Journal 70 (1996): 1933-1939.

#### **Biosensors and Molecular Imprinting: An Engineering Perspective**

Rohit Basavaraju<sup>1</sup>, Edan Elias<sup>2</sup>, Lina Lin<sup>3</sup>, Megan Garber<sup>4</sup>, Rima Patel<sup>5</sup>, Yantian Wang<sup>6</sup>, Miriam Rafaelovich<sup>6</sup> <sup>1</sup>Jericho High School, <sup>2</sup>North Shore Hebrew Academy, <sup>3</sup>Planview High School, <sup>4</sup>UCBerkeley, <sup>5</sup>Carnegie Mellon University, <sup>6</sup>SUNY Stony Brook, <sup>6</sup>SUNY Stony Brook

Biosensor technology is the basis of modern medical diagnostics; specifically through western blotting and micro-arrays as a means of protein analysis. However, disadvantages with both mechanisms are that they rely on expensive antibodies which commercially are not available for all proteins and may lack complete specificity. Molecular imprinting on the other hand, does not rely on antibodies and has been noted to be simple, specific, quick and potentially cheap<sup>1</sup>. Molecular imprinting is divided into two major types: 2D and 3D imprinting. Unfortunately, it is difficult to imprint and remove proteins from the 3D model<sup>2</sup> Therefore we investigated the 2D variety.

Our 2D model is based on a gold plated silicon chip coated with an 11-mercapto-1-undecanol (thiol) monolayer and imprinted with a particular protein. Once the protein is re-introduced, it binds to the imprinted space via electrostatic and hydrophobic/hydrophilic interactions, changing the surface potential (mV) of the chip (see figure1). As this binding process occurs the resulting electric change is measured by a potentiometer.

Our work attempted to make this chip as cost-effective and accessible as possible while keeping it as accurate as possible. For example, we reduced the size of the chip from 5 mm in width to 1mm and found a simple method of storing and reusing the chip. We



Figure 1: A particular protein is imprinted in the thiol monolayer that coats the gold plated silicon chip. When removed, the protein leaves specific spaces which are occupied when the particular protein is reintroduced (diagram from Yantian et al., 2008).

determined the optimal method of storage to be refrigeration; logical, as thiol denatures at room temperature. In theory, if the thiol does not denature i.e. if the chip is continually refrigerated, it may remain viable for years on end. We also found the chip to be reusable if cleaned by deionized water immediately following a test, as the water mechanically displaces the protein.

Our future work and applications for this technology are ambitious, but we believe very plausible. We will attempt to substitute the gold plating with copper; our rationale here is that the thiol binds to the gold via sulfur-metal bonding, which copper should be able to participate in as it and gold are in the same group in the periodic table (they both have similar electron structures). A successful replacement of gold with copper would significantly decrease the price of employing our technology, not only would this increase its accessibility in the US, but also in impoverished ("3<sup>rd</sup> world" countries") communities. This is extremely substantial, as such nations would otherwise be unable to afford other methods of protein analysis or biosensing (e.g. western blotting and molecular arrays).

The potential of 2D molecular imprinting is virtually infinite as theoretically any piece of solid matter can be imprinted, in turn any disorder with a physical identity can be diagnosed (and by definition everything must be hydrophilic or hydrophobic, additionally there may be other specific interactions that can accommodate binding between a particular particle and thiol). We hope to use this technology in the detection of HIV via the sensing of gp41 and/or gp120, prostate cancer through the sensing of PSA and myosin VI in the blood and/or urine<sup>3</sup>, and breast cancer through the sensing of the salivary proteins VEGF, EGF and/or CEA<sup>4</sup>. Note that although several of these proteins do exist naturally in the body, their levels are skewed in the presence of their respective disorder, meaning that their potentiometric readings would have to be compared to a standard curve which models their regular levels and corresponding mV potential.

<sup>1</sup>Belluzo, Ribone and Lagier, 2008, <sup>2</sup> Turner, Jeans, Brain, Allender, Hlady, and Britt, 2009 <sup>3</sup> Dunn, Chenn, Faith, Hicks, Plats, Chen, Ewing, Suavageot, Isaacs, Marzo, Luo, <sup>4</sup> Brooks et al., 2008

#### Engineering a 2D Potentiometric Biosensor for Protein Detection

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There is an increasing need for precise protein detection as a diagnostic tool for identifying diseases,

pathogens, and abnormal protein levels in the body. An ideal biosensor for proteins is one that has both specificity and sensitivity. One common technique for constructing such protein biosensors is molecular imprinting (MI). Existing MI techniques for biosensing application are three dimensional, making use of bulk polymer matrices. This technique is unfavorable because of its long response time and low protein reoccupation of polymer cavities.<sup>1</sup> Alternatively, molecular imprinting on two dimensional surfaces has been proposed through a self-assembled monolayer (SAM) of thiol (11-mercapto-1-undecanol) and gold. Shown in Figure 1, this SAM is comprised of a Si wafer, a gold layer formed through chemical vapor deposition, an additional layer of thiol, and the proteins desired for templating



Figure 1: the SAM with organic thiol molecules and proteins for templating (shown in red).

embedded in the thiol. Proteins in aqueous solution are polyelectrolytes whose charges are dependent on the intrinsic isoelectric point of the protein as well as the ionic composition of the solution.<sup>1</sup> Charged protein adsorption into geometrically complementary cavities of the thiol and gold layer yields noticeable electric potential differences due to gold's conductive properties. We hypothesized that an effective electrochemical biosensor capable of identifying specific protein concentrations, selectively sensing proteins, and *in vitro* protein detection could be engineered through the use of gold-thiol SAMs.

In order to demonstrate the ability of the chip for determining specific protein concentration in an unknown solution, the chip was incubated in a 6% fibrinogen solution. A linear standard curve of fibrinogen concentration vs. voltage difference was produced and found to be y=1.2877x + 3.6674 for fibrinogen



concentration x in µg/mL and voltage difference y in mV. The selectivity of the sensor was also evaluated by cross testing the biosensor with hemoglobin and fibrinogen. Chips were imprinted with hemoglobin and fibrinogen separately and tested in solutions of fibrinogen, hemoglobin, or a combination of the two. Results indicated that for fibrinogen imprinted chips, the combined proteins produced the highest voltage response as the charged hemoglobin protein occupied cavities intended for the uncharged fibrinogen molecules, therefore producing a larger voltage response.

At the same time, previous research has shown that Au nanoparticles are toxic to human dermal fibroblasts and disrupt

the integrity of the cell. Fibronectin is a glycoprotein essential for processes like cell growth and adhesion. Therefore, it was hypothesized that exposure of these cells to Au nanoparticles would reduce fibronectin production. Cell media was extracted from human dermal fibroblasts exposed to 45 nm and 13 nm particles as well as no particles (control). Voltage response curves from the potentiometer (shown in Figure 2) confirmed this hypothesis as the control media had the highest response for fibronectin while 45 nm was lowest due to extensive cell damage from these large particles.

<sup>&</sup>lt;sup>1</sup> Wang, Y., Zhou, Y. "A potentiometric protein sensor built with surface molecular imprinting method." *Biosensors and Bioelectronics*. 2008; 24:162-166

#### Digitizing the Moving Face During Dynamic Displays of Emotion

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Facial motion can be correlated to muscular activity, which has been linked to emotional states. Currently, emotional states are determined by using the Facial Action Coding System (FACS), consisting of 32 Action Units (AU), each representing one or a combination of muscles that contract or relax.<sup>1</sup> However, FACS's accuracy is being questioned. One major limitation is the individual skill of the observer. Therefore, analysis of emotional states and muscular activity can benefit from additional methods. Digital Image Speckle Correlation (DISC) has been introduced recently to quantify the in-plane displacement of soft tissues with sub-pixel precision. The difference of light intensities between the pictures before and after a deformation is analyzed, displaying the direction and amplitude of the tissue motion. So far, DISC has been used to analyze voluntary facial expressions, such as smiling and blinking.<sup>ii</sup> Our goal is to quantify involuntary facial motion following auditory stimuli and to determine if it can be correlated to an emotional state.

First, we confirmed that facial motion following a smile, can be quantified using DISC. Furthermore, we showed that facial asymmetry can be visualized by displaying the areas of maximum motion amplitude during smiling [Fig.1] and can be quantified by calculating the ratio of motion amplitude of each side of the face. Fig. 2 illustrates the degree of asymmetry for identical twins during smiling. Second, we induced involuntary facial motion by introducing auditory stimuli. Subjects kept their heads still while they were listening to clips of relaxing and disturbing music for about 1.5 min. Pictures of their faces were acquired approximately every 10 seconds during the music. We show that DISC can display the direction and amplitude of motion over the entire face during an involuntary facial motion that is not visually detectable on a photo. We can identify the main areas of motion of the face [Fig.4] and therefore correlate the muscles with an emotional state. In this case, the muscles of the jaw (masseter) are contracting. The masseter is associated with anger (according to J.C. Hager). We show that DISC is a powerful tool that will benefit research in human emotional states. Experiments are currently being conducted on a larger number of subjects.



<sup>i</sup> Tracy JP, Robins, RW, Schriber RA. Development of a FACS-verified set of basic and self-conscious emotion expressions. Emotion, 2009 Aug; 9(4):554-9.

<sup>ii</sup> Staloff IA, et al., An in vivo study of the mechanical properties of facial skin and influence of aging using digital image speckle correlation. Skin Res Technol. 2008 May; 14(2):127-34

# **Session 3**

# Cells on Hydrogels

#### Session Chairs: Isabel Afriat, Stony Brook University

#### Graduate Mentors: Divya Bhatnagar, Zhi

**Pan** Department of Materials Science and Engineering, Stony Brook, University, Stony Brook, NY


### Pluronic F-127 Hydrogels: In-situ Gel Forming Systems for Tissue Engineering

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Pluronic F-127 thermoreversible gels are of potential use as an alternative to open surgery, by being injected at the herniated disc sites<sup>1</sup>. For concentrations superior to 18% w/v, the F-127 gels are liquid under 10°C and therefore can be easily injected into the body<sup>2</sup>. They quickly harden and mold into the desired shape once the temperature is over 18°C. The cytotoxicity of F-127 is unclear: a group of researchers indicates a cytotoxic effect at a concentration larger than 10%<sup>3</sup> whereas others demonstrate its suitability at 20% and 30% for bone marrow derived stem cells differentiation<sup>4</sup>. It is critical to determine the cytotoxicity of F-127 gels, relative to concentration and cell type. We examined the behavior of dermal fibroblasts in contact or suspended in F-127 gels by looking at cell survival, cell growth and cell adhesion.

A solution of Pluronic F-127 was prepared at 30% w/v in DMEM, filter-sterilized through a 0.22µm pore size bottle-top filter, enriched with 10% fetal bovine serum and stored at 4°C until use. The assays were performed in a 24-well tissue culture plate and each treatment was performed in triplicate. We plated 2500 neonatal dermal fibroblasts in media (control), underneath the gel and in between two layers of gel in two 24 wells plates, and kept in an incubator at 37°C and 5% CO2, for 24 hours. After 24 hours, the plate is removed from the incubator, full media is added to dilute the gel and the cells are immediately imaged. Then the plate is placed again in the incubator for two hours and the cells are imaged again. In another experiment, the dermal fibroblasts were put in contact with the gel, similarly to the first experiment and were also suspended in the F-127.

The cells that were in contact with the gel for 24 hours are rounded [Fig.1b], indicating their lack of adhesion to the gel. After adding media to dilute the gel and incubating for two hours, the cells recover an elongated shape [Fig.1c], similar to the control [Fig.1a]. Cell count is performed using a hemocytometer showing the reduced growth of the fibroblasts in contact with the gel in comparison to the control [Fig.2]. These results show that the F-127 at 30%w/v is non-cytotoxic for a 24-hour contact with cells. In the second experiment, where the cells were suspended in the gel, trypan blue staining showed a 50% positive staining compared to 5% for the control and 6% under the gel, showing toxicity after 24 hours when the cells are suspended within the gel. The plate incubated for 48 hours was imaged after addition of full media and only cell debris was found [Fig. 1d], showing that the gel is cytotoxic when in contact for 48 hours. The degree of purity of the F-127 is critical to assess its biocompatibility; various grades and concentration of F-127 are being tested. Cell type is also to be taken into account. Current experiment investigates the cytotoxicity of 10, 18 and 25% w/v Pluronic F-127 (BASF NF grade) gels on dermal fibroblasts and endothelial cells. These assays are designed for future use as *in vitro* biological safety release assays for Pluronic F-127 before they are used clinically.



<sup>&</sup>lt;sup>1</sup>Albert, Todd. "Cervical Spine Surgery: An Overview" Spine Universe. 2008. http://www.spineuniverse.com/displayarticle.php/article1545.html

<sup>2</sup> Gutowska, A., Jeong, B., & Jasionowski, M. (2001). Injectable Gels for Tissue Engineering. *The Anatomical Record*. 342-349

<sup>&</sup>lt;sup>3</sup> Khattak S., Bhatia, A., & Roberts S., "Pluronic F-127 as a Cell Encapsulation Material: Utilization of Membrane-Stabilizing Agents" <u>Liebert</u> <u>Online.</u> 2005. http://www.liebertonline.com/doi/abs/10.1089/ten.2005.11974

<sup>&</sup>lt;sup>4</sup> Vashi, A. et. Al. (2008) Adipose Differentiation of Bone Marrow-Derived Mesenchymal Stem Cells Using Pluronic-F127 Hydrogel In Vitro. *Biomaterials*. 573-579

#### The use of hydrogels and electrospun fibers as a scaffold for tissue engineering

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Hydrogel surfaces of different thicknesses were tested to determine the optimal scaffold for cellular proliferation and migration of human dermal fibroblasts (HDF). The discovery of such surfaces can lead to major advancements in the field of tissue engineering and wound healing. Hydrogels, by definition, are viscoelastic surfaces comprised primarily of water [1]. The significance of hydrogels is that they serve as a minimally invasive and biodegradable interface that allows for the facilitation of cell migration to the site of wounds [2]. In order to accomplish this, two novel hydrogels were engineered, one composed of a combination of Hyaluronic Acid (HA) and Laponite clay and the other a combination of gelatin cross-linked with the enzyme microbial transglutaminase (MTG), in varying ratios of 5:1, 25:1, and 250: 1 (gelatin:MTG). Each sample was plated with 5,000 cells on day 0 and cell counts were performed on days 1, 3, 5, and 7 for HA- clay samples and days 2, 4, 6, and 8 for gelatin- MTG samples to establish growth curves. Cells in the HA-clay hydrogels became embedded in the gel making a growth curve impossible to establish, whereas, a normal growth curve was observed for the cells on the gelatin-MTG gel. Also, single cell migration was observed on all gelatin-MTG samples 6, 24, and 48 hours after plating. Imaging technology was then applied to the migration videos to determine the average velocity of cell movement. For all concentrations of gel, the movement of cells after 6h was negligible, however, after 24h, the velocity of cells increased significantly. The mean velocity of the cell population after 24h was recorded at 0.814 µm/min, 0.821 µm/min and 0.593 µm/min for the 5:1, 25:1 and control samples, respectively. Samples that were kept for overnight observance exhibited decreased velocity from the cells that were observed after 24 hours. These results indicate that the gels significantly increased the movement of the cells as compared to the control and that the 25:1 sample was most beneficial to the cells. In addition, it is clear that the optimal time for implantation of gels is after a 24 hour incubation period. This information is essential because it indicates the optimal conditions for cell movement as applied to wound healing. Additionally, the migration of HDFs, en masse, on electrospun fibers was observed. Studying the viability of

electrospun fibers as a scaffold and observing the migration of cells on this surface is particularly crucial to the process of wound healing due to the fact that the extracellular matrix possesses several fibrous components. Cell migration was measured as a function of the fiber angle [3]. In order to do this, bi-layer fibers were electrospun at angles of either 45 or 90°. This served two purposes: to create a 3 dimensional substrate composed of more than one flat layer and to create junctions between the angled layers of fibers that would test a cell's preference towards a certain direction. Silicon wafers were coated with a 30 mg/ml concentration of PMMA (Molecular Weight



**Figure 1.**5X optical microscopy of PMMA electrospun fibers at 45° (fig 1.1) and 90°

120) in toluene. PMMA fibers were spincasted onto one half of each wafer. After annealing, a droplet of cells was plated onto the wafer, and after about 24 hours, the cells were fixed and stained. Confocal images of the now diffused cells were taken, and were examined to determine how the cells migrated along the fibrillar surface.



**Figure 2.** Migration of human dermal fibroblasts cultured on a hydrogel scaffold of gelatin and MTG enzyme (25:1). Images were taken at 10 minute intervals during a 10 hour period. The progression of the cells through images 1.1, 1.2 and 1.3 demonstrates the cells' tendency to follow track established by neighboring cells.

[2]Park, Y. *et al.* "Photopolymerized hyaluronic acid-based hydrogels and interpenetrating networks." <u>Biomaterials</u> 24 (2003): 893-900 [3]Gerardo-Nava, J. *et al.* "Human neural cell interactions with oriented electrospun nanofibers in vitro." Nanomed 4.1 (2009): 11-30

<sup>[1]</sup> Courey, A. et al. "Hydrogels in tissue engineering. " Genzyme Corporation (2006): 1



### Alternative Energy

Session Chairs: Joanne Figueiredo, Smithtown West High School

**Graduate Mentors:** 

**Ping (Rose) Li, Cheng Pan** Department of Materials Science and Engineering, Stony Brook, University, Stony Brook, NY



### Improvement of titanium dioxide and dye interface in dye sensitized solar cells Joy Jiang<sup>1</sup>, Judith Jacobson<sup>2</sup>, Aryeh Guterman<sup>3</sup>, Dr. Joanne Figueiredo<sup>4</sup>, Cheng Pan<sup>5</sup>, Ping (Rose) Li<sup>5</sup>, Dr. Miriam Rafailovich<sup>5</sup>

<sup>1</sup>Comsewogue High School; <sup>2</sup>Torah Academy for Girls High School; <sup>3</sup>Hebrew Academy of Nassau County High School; <sup>4</sup>Smithtown West High School; <sup>5</sup>Stony Brook University

Sunlight is the only truly renewable energy source, and as such it is key in the pressing search for a sustainable, environmentally friendly way to satisfy the immense human demand for power in modern times. Dye-sensitized solar cells (DSSC) are part of a third generation of devices designed to harness the power of sunlight. DSSC are more cost effective than traditional silicon cells and reached a competitive efficiency of 10% in 2004<sup>1</sup>. DSSCs separate the photoelectron provider and charge transfer medium into two different materials. As shown in Figure 1, a monolayer of dye is adsorbed to a suspension of titanium dioxide nanoparticles that are coated on clear conductive glass. The dye acts as the sensitizer, the



material whose electrons are excited by photons to higher energy levels, and the titanium dioxide functions as the semiconductor, which carries the electrons away

from the dye to the conductive glass of the electrode. The electrons flow through an external circuit and load where work is performed, then back to a graphite coated counterelectrode. An electrolyte completes the cell by replenishing the electrons lost by the dye then accepting electrons that have traveled through the external circuit from the counterelectrode<sup>2</sup>.



The goal of this work is to increase the efficiency of these cells by altering the interface between the titanium dioxide and dye. Solar cells dyed with anthocyanin from blackberries were exposed to a solar simulator. It was observed that the thickness of the porous titanium dioxide coating, as shown in Figure 2, affected the degradation of the cell. It was also found that a greater saturation of the titanium dioxide with the dye produced better results, with more consistent voltage and

**Figure 1: SEM image of TiO**<sub>2</sub> current outputs, as shown in coating at 10K magnification the IV curve in Figure 3. Our current research focuses on improving the adsorption of the dye to the titanium dioxide by pre-treating with positively charged lysozyme protein to improve the bond between the dye and titanium dioxide. We may also experiment with different titanium dioxides and dyes in the future.



Figure 3: IV Curve from DSSC

<sup>&</sup>lt;sup>1</sup> Gratzel, Michael. "Conversion of sunlight to electric power by nanocrystalline dye-sensitized solar cells." *Journal of Photochemistry and Photobiology: A Chemistry* 164 (2004) 3-14.

<sup>&</sup>lt;sup>2</sup> Smestad, Greg. "Education and solar conversion: Demonstrating electron transfer." *Solar Energy Materials and Solar Cells* 55(1998) 157-178.

<sup>&</sup>lt;sup>3</sup> From Posttech Pohang University of Science and Technology, http://www.postech.ac.kr/chem/mras/eunju.jpg

### Characterization of Thin Film Morphology and Nanoparticle Confinement in the Active Layer of Ordered Bulk Heterojunction Photovoltaic Cells

Mubarrat Nuvid Bhuiyan, Jericho High School Matthew Miecnikowski, Half Hollow Hills High School East Matthew Alpert, Harvard; Jennifer Segui, John Jerome, Miriam Rafailovich, SUNY Stony Brook

The need for an alternative energy source has become imperative; most promising in availability is solar energy. Typical silicon solar cells possess large areas and a single junction, but are extremely expensive in comparison to polymer-based solar cells. Thus, organic polymer solar cells are the most favorable prototypes because they are relatively harmless to the environment, have nanoscale morphological properties which can be altered in numerous ways, and are much more cost-effective<sup>1</sup>.

Photovoltaic power production occurs in the following steps: 1) light is absorbed by the donor polymer,

commonly regio-regular poly-(3,hexylthiophene) (P3HT), where excitons (e.g. bound electron-hole pairs) are produced 2) excitons then diffuse towards the respective donor-acceptor interface 3) the pair separates at the interface, due to the greater electron affinity of the electron acceptor, generally phenyl-C<sub>61</sub> butyric acid methyl ester (PCBM)<sup>2</sup> and produces a photocurrent.



Figure 1- Representation of columnar structure of ordered BHJ solar cells

In our study we have analyzed the properties of bulk heterojunction (BHJ) solar cells which take advantage of multiple interfaces created by casting a polymer mixture which phase separates into nanoscale regions of each material: P3HT and polystyrene (PS) (Fig. 1). BHJ's also improve exciton dissociation efficiency at interfaces, thereby allowing the active layer to be thicker and permit increased absorption of light. We prepared a variety of blend solutions of PS to P3HT (90:10, 70:30, 50:50) to determine which blend offers the greatest evidence of lateral phase separation into the ideal ordered columnar structure (Fig. 2). To ascertain the depth of P3HT penetration into PS we used a sputtering technique via the ion mill. Due to the quicker etching rate, found by interpretation of ellipsometry results, of P3HT in comparison with PS, P3HT was removed while the PS layer appeared intact. We then compared the etching of P3HT under the ion mill to the chemical etching using UV light exposure and toluene and chlorobenzene submersion. Samples containing PCBM nanoparticles were spincast and observed using atomic force microscopy (AFM) and transmission electron microscopy (TEM) to note any differences in structure and pinpoint nanoparticle confinement.



Figure 2- AFM image of lateral phase separation of 90:10 PS:P3HT blend nanocomposites

Using quantitative data from AFM and sputtering techniques, we calculated the interfacial tension of the polymer-polymer interaction to establish the degree of miscibility. Empirically, we discovered the index of refraction of blend samples of PS and P3HT.

Further research includes observation of all samples treated with scCO<sub>2</sub>, annealed and unannealed containing the different blend ratios, for crystallinity. Thin films for the active layer can be incorporated into a cell, which can be tested using I-V equipment for solar cells. Thus, we hope to maximize the potential of BHJ solar cells by improving both their nanostructure and optoelectronic properties.

<sup>1</sup> Yang, F., Shtein, M., & Forrest, S. R. (2005, January). Controlled growth of a molecular bulk heterojunction photovoltaic cell. *Nature Materials*, *4*, 37-41

<sup>2</sup>Vanlaeke, P., (2005, November). P3HT/PCBM bulk heterojunction solar cells: Relation between morphology and electro-optical characteristics. Solar Energy Materials and Solar Cells, 90, 14, 2150-2158

### Understanding and Explaining Mechanisms in the Optimization of Fuel Cell Reactions Using Nanoparticle Catalysts

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Because of its high efficiency and lack of harmful emissions, the polymer electrolyte membrane (PEM) fuel cell is a potential mechanism for future environmentally-friendly vehicular transport. PEM fuel cells rely on a perfluorosulphonated ionomer membrane to conduct split hydrogen ions from anode to cathode while their electrons are forced through a circuit to generate current. Prior research indicates that gold nanoparticles (AuNP's) significantly increase the efficiency of similar current generating process.

However, there is little, if anything, in the literature pertaining direct manipulation of the PEM membrane and the mechanism that AuNP's use; i.e. how AuNP's speed up the reaction. Therefore, our research was two pronged: we sought to determine the most favorable concentration of AuNP's on the membrane while determining how AuNP's function and we sought to experiment with other metals and mixtures to produce the highest output power possible.



To do this, we synthesized AuNP's through an established two phase method<sup>1</sup>: tetrachloraurate was broken down to the nanoscale and stabilized with dodecanethiol with an experimentally determined optimal gold-thiol ratio of 1:1<sup>2</sup>. We then used AuNP's to coat Nafion-117® membranes at different surface pressures through particle deposition on the Langmiur Blodgett Trough. A low range of surface pressures increased fuel cell efficiency up to 80%. Additionally, we hypothesized that the gold acts as a catalyst not to the hydrogen, but atmospheric and source impurities of carbon monoxide (CO), which acts as an inhibitor to the platinum catalyzed redox reactions of the cell. Thus, we compared blank control membranes to AuNP coated membranes in atmospherically normal levels of CO and reduced levels of CO and found a significant difference in efficiency boosts, proving that the nanoparticles act as CO

reducers. Furthermore, we coated membranes with Titanium Dioxide (TiO<sub>2</sub>) nanoparticles, another established nanocatalyst, and mixtures of Au and TiO<sub>2</sub>. Results indicate that a possible synergism may exist between the two catalysts, opening the way to new research in membrane modification.

We hope to broaden our search to include other metals such as graphite, tungsten, iron and rhodium to search for further ways to increase efficiency. By substituting some of these metals in for the expensive platinum catalysts, we can increase efficiency while lowering costs. Such advances are necessary to make the fuel cell a more competitive alternative on the energy market.



<sup>&</sup>lt;sup>1</sup> Thompson, David T. <u>Using Gold Nanoparticles for Catalysis</u>. Nanotoday, Aug 2007. Vol 2, No 4.

<sup>&</sup>lt;sup>2</sup> Kao, Kenny. <u>Nanoparticle Enhancement of Polymer Electrolyte Membrane Fuel Cell Output.</u> Written for Dr.

Miriam Rafailovitch during a previous SUNY Stony Brook research program.



### Enzymatically Synthesized Polymers

Session Chairs: Daniel Peng, Brandon Hsu, John Hopkins University

**Graduate Mentors: Maddy Manoj,** Department of Chemical Engineering NYU-Polytechnic University, NY, NY **Ping (Rose) Li, Cheng Pan** Department of Materials Science and Engineering, Stony Brook, University, Stony Brook, NY



### Cutinase-catalyzed modification of poly(ethylene terephthalate) surfaces by transesterfication and transamidation reactions in aqueous and organic media Daniel Chun<sup>1</sup>, Michael Zhou<sup>2</sup>, Chong Li<sup>3</sup>, Seongchan Park<sup>4</sup>, Miriam Rafailovich<sup>4</sup> 1. POBJFK High School; 2. Jericho High School; 3. NYU Poly; 4. Stony Brook University

Polyethylene terephthalate (PET), better known as polyester, is the highest volume synthetic fiber in the textile industry. However, hydrophobicity limits its applications such as the dying and surface cleaning (Fischer-Colbrie, 2004). Our goal was to utilize a cutinase-catalyzed reaction to modify PET thin film surfaces through transesterification, transamidation, and hydrolysis in an aqueous and organic media. Cutinase was chosen as an enzyme for surface modification because its natural function is the hydrolysis of cutin, a structural polyester in plants.

PET thin films were spuncast using a 20 mg/ml solution; the solvent consisted of dichloromethane:hexafluoroisopropanol in a 5:1 volume ratio. To modify the PET surface, the thin films were placed in a solution of 1.8 mL acetonitrile (ACN), 0.2 ml deionized water, 2 mg cutinase derived from *Humicola insolens* (HiC), and 1 mg of amine for 3 hours at 70° C. The cutinase breaks the ester bond, which allows either amidation or hydrolysis to occur when the carbon bonds to an amine group (-NH) or hydroxyl group (-OH) respectively. To remove residual cutinase and amine that have absorbed into the PET, the thin films were rinsed in sodium dodecyl sulfate (SDS) and methanol. They were then dried in a vacuum for 1 hour.

In order to determine whether the modification of PET occurred, Fourier Transform Infrared Spectroscopy (FTIR) was employed. Hydrolysis, the bonding of -OH to carbon, is observed as a broad spectrum peak from 3000 to 3500 cm<sup>-1</sup>. Amidation, the bonding of -NH to carbon, is observed as peaks at 1650cm<sup>-1</sup> and 1540cm<sup>-1</sup>. FTIR confirmed that hydrolysis occurred, as expected. However, there appeared to be only slight amidation, as there was only a slight shoulder at 1650 cm<sup>-1</sup>.

To determine whether the amine groups will have an effect on protein adsorption and fiber formation, PET and modified PET surfaces were incubated in 3.7 mg/mL solution of fibrinogen in TBS-EDTA overnight. AFM Images of the PET surfaces incubated in fibrinogen also proved that amidation was limited. In Fig. 1 and 2, fibers formed both on the control and modified surfaces.

In the future, we will use alternative procedures to successfully attain transamidation. We will also calculate the contact angle of water droplets on PET and modified PET surfaces to quantify the change in hydrophilic/hydrophobic nature.



Fig. 1 Fibilitiogen libers of FET Fig. 2 Fibilitiogen libers of Modified FET

Fisher-Colbrie, Gudrun. "New Enzymes with Potential for PET Surface Modification." *Biocatalysis and Biotransformation* (2004).

### **Enzyme Stamping onto Polymer Interfaces**

Jonathan Nachman<sup>1</sup>, Sandy Guerrero<sup>2</sup>, Brandon Hsu<sup>3</sup>, Daniel Peng<sup>3</sup>, Maddy Manoj<sup>4</sup>, Miriam Rafailovich<sup>5</sup>, Yizhi Meng<sup>5</sup>

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For years, commercial polymers have been synthesized through the burning of fossil fuels. However, recently, there have been experiments into creating polymers by using enzymes. Due to the fact that these new polymers were made by enzymes, the enzymes could break the polymer apart as well<sup>1</sup>. If so, these polymers could be the replacements of current commercial plastics. Also of significance is the use of these enzymes to stamp patterns onto polymers. If the deposition of enzymes can be controlled, then these polymers could replace the current lithographic methods, which are used to create circuit boards. This would be cheaper and safer to the environment than all of the solvents used in current methods<sup>2</sup>. Another application of this type of polymer is drug delivery. Drugs could be encapsulated with the enzyme inside the polymer and would thus release the drug over a predetermined period of time in a preset location<sup>3</sup>.

To test this, a unique procedure had to be devised. Samples of the polymer in varying concentrations were

spuncast, on both glass and silicon. A "stamp" was made by pouring PDMS onto an etched piece of silicon (figure 1a), thereby copying the pattern (figure 1b)<sup>3</sup>. The enzyme was applied to the stamp which was, in turn, stamped onto the samples. The samples were incubated, either in buffer solution or dry, at 37° C, and both optical and confocal microscopy images were taken of the samples. In addition, a sample was also spuncast with the enzyme encased in it to see the progression of degradation.





optical microscope)

The results have shown that while the stamp works for the first couple of days, it doesn't stop eating away the polymer. Also, when the enzyme was encased in the polymer it took about a week to dissolve the thin films' attachment to the wafers. This did not allow for future readings In the future, this research will focus on methods to slow the degradation. Also, using other media to apply the enzyme to the polymer is being considered. Dip pen lithography using the atomic force microscope is a primary thought of such methods. This will allow greater precision than a stamp and also allow for more intricate designs than prior methods.

- S. Warwel, C. D. (2000). Polyseters by Lipase-Cataluzed Polycondensation of Unsaturated and Epoxidized Long Chain Dicarboxylic Acid Methyl Esters with Diols. *Journal of Polymer Science*, 1601-1609.
- 2. P. Snyder, M. J. (2007). Biocatalytic Microcontact Printing. *American Chemical Society*, 7459-7461.
- 3. S. Wang, S. G. (2007). Disodium norcantharidate loaded polycaprolactone microspheres Preparation and Evaluation. *Intl. Journal of Pharmaceutics*.

## **Session 6**

## Flame Retardant Polymers

Session Chairs: John Iraci, Department of Mechanical Engineering, Cornell University

**Graduate Mentors: Seongchan Pack** 

Department of Materials Science and Engineering, Stony Brook,

University, Stony Brook, NY **Rebecca Isseroff**, Lawrence High School, Cedarhurst NY

Flame Retardant Addit	tives	
5% Additive (80/20 Pla/Ecoflex)	UL 94V Rating	
Cloisite Na+	V2	
RDP Starch /Cloisite Na <sup>+</sup>	V2	
RDP Starch/ RDP Cloisite Na+	V2	

### Simulating the Effect of Flame Retardant Materials on Heat Diffusion in Polymers

Arpon Raksit, Dilip Gersappe, and Miriam Rafailovich Department of Materials Science and Engineering, Stony Brook University

Polymers find use in various materials and items due to their extensive range of useful properties. However, many commonly used polymers, including polystyrene, polyvinyl chloride, and polyethylene, have low ignition temperatures, presenting the dangers of combustion and thermal degradation of polymers (Kashiwagi, n.d.). Research is being done on the use of flame retardants, materials that inhibit the proliferation of fire or heat, yet not much is known about the physics of heat loss due to these flame retardant materials. Simulating the effect of flame retardants on the spread of heat throughout a polymer may provide a better understanding on how to effectively manipulate and make use of flame retardant materials. Using the lattice Boltzmann method, an algorithm that models the physics of fluid dynamics, a basic simulation of heat diffusion from a heat source to sink was implemented in two dimensions (Chen & Doolen, 1998). The polymer and flame retardant material were incorporated into the system by implementing ignition within the particles of the polymer and by adding heat absorbing filler particles within the polymer matrix. By manipulating the volume fraction of flame retardant particles, their ability to absorb heat, and their efficiency in removing heat from the system, different degrees of polymer combustion were simulated (Figures 1A-C). To quantify the data, the heat in the system was plotted as a function of time and the x-coordinate in the lattice, both illustrating that flame retardant particles inhibit flame movement. From these simulations and graphs, it was concluded that the combustion of polymers and the effect of flame retardant materials on heat diffusion can successfully be simulated, and by altering the efficiency of flame retardant materials, the propagation of heat can be controlled. In the future, the systems will be simulated in three dimensions, providing a more detailed understanding, as well as allowing for complex designs and structures of flame retardant material.



Figure 1A: Complete polymer combustion (10% Filler Particles, 10% Heat Absorption, 0% Heat Removal)



Figure 1B: Low degree of flame quenching (30% Filler Particles, 10% Heat Absorption, 25% Heat Removal)



Figure 1C: High degree of flame quenching (50% Filler Particles, 25% Heat Absorption, 50% Heat Removal)

### Creating Flame Retardant Polymer Blends for Application in Long Distance Cables

Debby Leah Greenstein, SAR High School Katelyn Marie Elizabeth Ireland, West Islip Senior High School Dr. Miriam Rafailovich and Seongchan Pack Department of Material Science and Engineering, SUNY Stony Brook John Michael Iraci, Cornell University

Recently halogens have been banned causing a push to create flame retardants utilizing polymers and organoclays that are environmentally sound. However, because of phase separation and dispersion it is very hard to create flame retardant polymer blends even with the addition of flame retardant fillers.<sup>1</sup>

This experiment explores the efficiency of different additives and compatiblizers in increasing the flame retardancy of Ethyl Methyl Acetate (EMA), blended with either Linear Low Density Poly Ethylene (LLDPE) or Low Density Poly Ethylene (LDPE) in various ratios. Additives to the copolymer blends to increase compatibility include cloisite 25A clay, Aluminum Trihydrate (ATH), and RDP treated clay. In addition, the polymer blends were exposed to supercritical Carbon Dioxide (scCO<sub>2</sub>) for the purpose of increasing compatiblization. We use the Dynamic Mechanical Analysis (DMA) to test the mechanical properties of the materials for the 4:1 EMA/LLDPE copolymer. We also use the Instron to test the tensile strength of the materials. Flammability testing has proven that scCO<sub>2</sub> increased flame retardancy in some sample, whereas cloisite 25A clay and RDP treated clay, when each was combined in 1, 3, 5, and 10 percentages with a 4:1 sample of EMA:LLDPE, did not. Flammability testing also revealed that ATH caused the copolymer blend to be flame retardant as expected since ATH is known to absorb heat cooling the material.<sup>2</sup> Future research will involve blending carbon nanotubes in various quantities with our copolymer blends, as well as exposing the clay nanocomposites to scCO<sub>2</sub> in order to further increase the flame retardancy of the material.



<sup>&</sup>lt;sup>1</sup> Pack, S., Si, M., Koo, J., Sokolov, J.C., Koga, T., & Kashiwagi, T. (2009). Mode-of-action of self-extinguishing polymer blends containing organoclays. *Polymer Degradation and Stability*, *94*, 306-326.

<sup>&</sup>lt;sup>2</sup> F.Laoutid, L. Bonnaud, M. Alexandre, J.-M. Lopez-Cuesta, Ph. Dubois. "New prospects in flame retardant polymer materials: From fundamentals to nanocomposites." <u>Material Science and Engineering R</u> 63 (2009) 100-125.

### Biodegradable Polymer Composites: Compatibility, Thermal Stability, and Flame Retardancy

### Neil Muir, Uniondale High School Seoung chan Pack and Dr. Miriam Rafailovich, Department of Material Science and Engineering, Stony Brook University John Michael Iraci, Cornell University

In recent years, biodegradable polymers have been carefully studied as alternatives to long-lasting polymers in everyday use. This effort has been made in response to rapidly decreasing nonrenewable resources such as petroleum, and the growing burden of plastic waste management in the world today.<sup>1</sup> Biodegradable polymers, often derived from plant sources, degrade safely into the environment, releasing small amounts of CO<sub>2</sub>, CH<sub>4</sub>, water, biomass, and humic matter.<sup>2</sup> However, these recent studies have also revealed unfavorable qualities of biodegradable polymers such as high thermal sensitivity, flammability, and poor polymer compatibility.<sup>1</sup> Furthermore, poor compatibility or polymer phase separation due to large unfavorable enthalpy results in poor mechanical properties of the polymer blend.<sup>3</sup> The improvement of these aforementioned properties, with awareness to the expense of certain biodegradable polymers, (Ecoflex is much more expensive than Poly (lactic acid)), is the goal of this study.

Mechanical, thermal, and flame tests were administered to samples melt-mixed in a C.W. Bradender Plasticorder. A base polymer blend of 80% Poly (lactic acid) (PLA) and 20% Ecoflex was used in each sample to determine the capability of each filler to reduce phase separation between each polymer. Fillers (5% by weight) included Corn starch (Melojel), Cloisite Sodium, and Q-salt Halloysite Nanotubes. Flame retardant additives: Aluminum trihydrate (ATH), a metal hydroxide known to absorb 1050kJ/kg of heat thus cooling the polymer blend<sup>4</sup>; and Resorcinol bis( diphenyl phosphate) (RDP), a phosphorus based flame retardant known to limit the volatilization of fuel and prevent the formation of new free-radicals<sup>4</sup> were also studied in this experiment. RDP was tested after being soaked with separate mixtures of Starch and Cloisite Sodium while ATH was added (5% by weight) in powder form. The results of this study serve to further understand the properties of biodegradable polymers as well as improving the properties of biodegradable flame retardants and composites.

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Flame Retardant Additives			Ī	_	т						
5% Additive (80/20 Pla/Ecoflex) Cloisite Na*	UL 94V Rating V2	1.50E+09 -			1	Ŧ	Т		Ι		
RDP Starch /Cloisite	V2 V2	1.00E+09 -	T				Ţ	Ι		I	
RDP Starch/ RDP Cloisite Na⁺	V2	5.00E+08 -	trol	ch	Clay	Starch		Starch Clay		Starch/ Clay	
Chart 1 depicts flame retardant additives which made the base		0.00E+00	Control	Starch	RDP Clay	RDP	Clay	RDP (	PLA	RDP	
polymer blend flame retardant according to ASTM standards.		Figure 1 depicts th	e aver	age m	odulu	s (MP	a) of s	sample	s (5%	6 by weight	:)

<sup>1</sup> Matkó, Sz., A. Toldy, S. Keszei, P. Anna, Gy Bertalan, and Gy Marosi. "Flame retardancy of biodegradable polymers and biocomposites." <u>Polymer Degradation and Stability</u> 88 (2005): 138-145.

<sup>2</sup>Gross, Richard A.; Kalra, Bhanu. "Biodegradeable Polymers for the Environment" <u>Green Chemistry.</u> (2002) Volume 297: 803-307 <sup>3</sup> Mayu Si, Tohru Araki, Harold Ade, A. L. D. Kilcoyne, Robert Fisher, Jonathan Sokolov, Miriam Rafailovich. "Compatibilizing Bulk Polymer Blends by Using Organoclays." <u>American Chemical Society</u> (2006)

<sup>4</sup>F.Laoutid, L. Bonnaud, M. Alexandre, J.-M. Lopez-Cuesta, Ph. Dubois. "New prospects in flame retardant polymer materials: From fundamentals to nanocomposites." <u>Material Science and Engineering R</u> 63 (2009) 100-125.

### Effect of Organic Phosphate Oligomer on Roughness in Heat-Resistant Polymers

Joong Gon (Kevin) Yim, South Side High School Seong Chan Pack, Stony Brook University, NY Dr. Miriam Rafailovich, Department of Material Science and Engineering, SUNY Stony Brook John Iraci, Cornell University

Flame retardant polymers have been center of research in the field of material science. The problem with some flame-retardant additives is the recyclability and the environmental concerns. To address the environmental issues many researchers are using organophosphorus flame-retardant additives because they are biodegradable<sup>1</sup>. Another problem is when flame-retardant additives are added to the polymers, the additives and the polymers do not mix well, decreasing the flame retardant and mechanical properties of the polymers. Poly Carbonate (PC) was used as the polymer and Resorcinol bis(diphenyl phosphate) (RDP) for flame-retardant additive. RDP is an organic oligomeric phosphate flame-retardant and flow modifier. PC and RDP were spun and dissolved in chloroform in different ratios to get a thin film on silicon wafer. Atomic force microscope (AFM) was used to measure the roughness of the films. Films were then annealed in the vacuum oven for an hour at 120 °C to get better results. The roughness was measured because decreasing roughness means the RDP clay is dispersed well throughout the polymer. The film with 100% PC at the height of 100 µm had mean roughness of 15.038 nm (figure 1). As the weight percentage of RDP increased the roughness started to decrease. Ten percent of RDP was added in PC, the roughness decreased to 5.510 nm (figure 2). As the wt % of the RDP increased, the roughness decreased, showing that RDP disperses throughout the polymer better as the concentration of RDP increases.



<sup>1</sup>D. Bright, S. Dashevsky, P. Moy, B. Williams. *Resorcinol Bi(Diphenyl Phosphate), a Non-Halogen Flame-Retardant Additive* Akzo Nobel Central Research Dobbs Ferry, NY 10522

#### Langmuir-Blodgett Trough-Assisted Graphene Synthesis

Paul Masih Das<sup>1</sup>, Vadim Pozin<sup>2</sup>, Seongchan Pack<sup>2</sup>, Rebecca Isseroff<sup>1</sup>, Miriam Raffailovich<sup>2</sup>

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Graphene is a two-dimensional, one-atom-thick sheet of bonded carbon atoms which arrange themselves into a hexagonal lattice structure (Fig. 1). Discovered in 2004, graphene has become the strongest substance known to man, also exhibiting the highest surface-area-to-volume ratio ever recorded.<sup>1</sup> The cost of producing such a substance is extremely high, about \$1,000 to synthesize a sample the width of a human hair. However, graphene has the potential to become a superior replacement for current electronic transistors because of its ability to support the transportation of electrons at speeds thousands of times greater than any current material. Graphene's electron mobility rating, tensile strength, surface area, and thermal conductivity are all greater than silicon. In fact, most of graphene's properties are the highest ever observed for a solid-state material. Graphene transistors would save time and space, conserve energy, and undoubtedly revolutionize the electronic industry.



Fig. 1: Graphene Nanoribbon www.technologyreview.com/read\_article.aspx?c h=specialsections&sc=emerging08&id=20242

To obtain graphene, graphene oxide was first synthesized through a modified Hummer's



Fig. 2: TEM of Graphene Oxide

method.<sup>2</sup> First, powdered graphite was added to sulfuric acid, sodium nitrate, and potassium permanganate. The graphite was then washed and treated with hydrogen peroxide and hydrochloric acid. The graphene oxide was then reduced with sodium borohydride. Analysis of the graphene and graphene oxide was done through Raman spectroscopy, Fourier Transform Infrared Spectroscopy, Atomic Force Microscopy, Transmission Electron Microscopy, and X-Ray Diffraction. Transmission Electron Microscopy of our graphene oxide revealed that our samples were indeed ideal due to the presence of 5-6 carbon layers (Fig. 2). To further test our graphene sheets, High-Resolution Transmission Electron Microscopy will be done at Brookhaven National Laboratory.

The final stage of our experiment employs the

use of a Langmuir-Blodgett (LB) trough to "sew" our relatively small graphene sheets into one large sheet. By first making a homogeneous solution of graphene (in H<sub>2</sub>O or ethanol), the sheets then be spread on the air-water interface of the LB trough, compressed into one large monolayer, and removed on a silicon wafer (Fig. 3). This interdisciplinary (material science, physics, computer science) technique is unique and revolutionary. It has the potential to change today's silicon-based computer industry into one grounded on graphene.



<sup>&</sup>lt;sup>1</sup> Li et al. Processable Aqueous Dispersions of Graphene Nanosheets. Nature and Nanotechnology **3**:101-105 (2008)

<sup>&</sup>lt;sup>2</sup> Hummers and Offeman. *Preparation of Graphitic Oxide*. National Lead Company **1**:1339 (1957)

## **Session 7**

## Differentiation of Adult Stem Cells

**Session Chairs: Jaqueline Belizar** 

### **Graduate Mentors: Dr. Vladimir**

Jurukovski, Biology Department, Suffolk Community College

### Chung-Chueh Chang, Tatsiana Mironava

Department of Materials Science and Engineering, Stony Brook, University , Stony Brook, NY



### The Effect of TiO<sub>2</sub> and ZnO Nanoparticles on Dental Pulp Stem Cells

Rida Malick, Lawrence High School Dr. Vladmir Jurukovski, Dr. Miriam Rafailovich, Dr. Marcia Simon, Chung-Chueh Chang, Stony Brook University

An increasing demand for efficient tissue and bone regeneration calls for us to think of novel techniques to produce these desired results. Stem cells are optimal for this process because they possess characteristics such as proliferation, plasticity, and differentiation.<sup>1</sup> Dental pulp stem cells (DPSCs) are easily attainable and noncontroversial stem cells that are located in the pulp of the tooth. They can differentiate into a variety of different cells like osteoblasts, neural like cells, and adipocytes. Because they are diverse and ethically acceptable, DPSCs are favorable for tissue and bone regeneration.

In order to see how the DPSCs differentiate in different environments we used TiO<sub>2</sub> and ZnO nanoparticles, small substances about 100 nm or less in size that are present in products like sunscreen, toothpaste, and cosmetics. TiO<sub>2</sub> and ZnO have been previously shown to have adverse effects on DPSCs. Both nanoparticles were tested in non-inducing and inducing media, the only difference in the media being the presence of Dexamethasone, a corticosteroid that encourages differentiation of DPSCs into osteoblasts. Five thousand cells were originally plated on PB thin films and were treated with 0.1mg/ml and 0.5mg/ml of TiO<sub>2</sub>; 0.05mg/ml and 0.1mg/ml of ZnO. It was previously shown that DPSCs grown on PB thin films differentiate towards an osteoblast-like lineage regardless of if they are in non-inducing or inducing media. The cells were incubated for 3 weeks to check for biomineralization.

After 3 weeks confocal microscopy images were taken and later cells were stained for osteocalcin, a protein involved in biomineralization (Fig. 1). The presence of osteocalcin (red color) indicates that the cells were indeed differentiating into osteoblasts. As expected the control samples grown in inducing media show greater osteocalcin expression (shown in red) compared to samples in non-inducing media, which shows that the presence of dexamethasone causes the cells to differentiate. Lack of osteocalcin expression in both non-induced TiO<sub>2</sub> nanoparticle treated samples indicate that damaged cells are not capable of differentiation. The same result was found in the samples grown in inducing media in presence of TiO<sub>2</sub>, however they appeared healthy and were proliferating suggesting that the presence of Dexamethasone prevented excessive cell death. The ZnO nanoparticles caused severe cell damage in the non-induced media and again the presence of dexamethasone 'protected' the cells from the harmful effects of these nanoparticles. In addition, not only are the cells in the inducing media healthy and proliferating like in the case of the TiO<sub>2</sub>, but also are able to differentiate as indicated by the increased amounts of osteocalcin. The presence of both types of nanoparticles in non-inducing media damaged the cells and caused cell death. While TiO<sub>2</sub> is preventing cell differentiation in the presence of dexamethasone the ZnO did not. The major finding of this research is the fact that the presence of dexamethasone protects the cells from the damaging effects of the nanoparticles.



 $Control (+) \qquad TiO_{2} (+) \ 0.1 \ mg/ml \qquad TiO_{2} (+) \ 0.5 \ mg/ml \ ZnO (+) \ 0.05 \ mg/ml \ ZnO (+) \ 0.1 \ mg/ml \ ZnO (+) \ Mg/ml \ ZnO ($ 

**Figure 1.** The effect of  $TiO_2$  and ZnO nanoparticles on the proliferation and differentiation of DPSCs in the absence or presence of dexamethasone. (+) indicates presence of dexamethasone in the media (inducing media).

<sup>&</sup>lt;sup>1</sup> D'Aquino, Riccardo, Gianpaolo Papaccio, *Gregorio Laino, Antonio Graziano. Dental Pulp Stem Cells: A Promising Tool For Bone Regeneration.* 26 February 2008. Humana Press.

#### The Effect of Electrospun Polybutadiene Fibers on Dental Pulp Stem Cells

Julie Chang<sup>1</sup>, Yesha Maniar<sup>1</sup>, Vladimir Jurukovski<sup>2,3</sup>, Marcia Simon<sup>4</sup>, Miriam Rafailovich<sup>3</sup>, Chung-Chueh Chang<sup>3</sup> <sup>1</sup>Herricks High School, New Hyde Park, NY

<sup>2</sup>Suffolk County Community College <sup>3</sup>Dept. of Materials Science & Engineering and <sup>4</sup>Oral Biology and

Pathology, SUNY at Stony Brook, NY

Stem cell research is a growing area of interest, due to the favorable characteristics of stem cells to differentiate into a variety of cell types. Recently, a new source of adult stem cells, dental pulp stem cells (DPSCs), has been successfully isolated and is of interest due to their unique properties. DPSCs differ from other adult stem cells because they can be obtained from both deciduous and adult teeth after necessary extraction. DPSCs, found in the soft living tissue of the tooth, are multipotent and can differentiate into various cell types such as osteoblasts, neuronal cells and

adipocytes. Although DPSCs have lesser differentiation possibilities than those of bone marrow stem cells BMSCs, they have better immunoregulatory characteristics and higher proliferation rates.

We want to quantify the effect of electrospun polybutadiene (PB) fibers with different thicknesses on DPSCs, the differentiation capacity of DPSCs and signs of biomineralization that might occur. Previous research has shown that DPSCs can differentiate into osteoblast-like cells on thin spun cast PB.<sup>1</sup> In order to observe the path of differentiation of DPSCs, a thin film of PB was spun cast onto the Si wafers followed by electrospun PB fibers of different thicknesses, 5µm and 10 µm, thin and thick respectively.

thick respectively. Five thousand DPSCs were cell plated onto the surfaces and grown in standard non-inducing media, and the cells were counted on days 3, 5, and 7, to create a growth curve (Fig. 1). On days 7 and 21, the





atomic force microscope and confocal microscope was used to determine the stiffness of the cells and the cell's orientation along the fibers, respectively, both in non-inducing and inducing media. The scanning electron microscope and the energy dispersive x-ray will be utilized to observe calcium phosphate as sign of biomineralization, indication of osteoblast-like cell differentiation. Finally, the presence of osteocalcin will be examined, as this protein is present in cells that perform biomineralization.



Figure 2. AFM analyses of cell moduli under different conditions





The growth curve (Fig.1) indicated that compared to the control, the DPSCs proliferated slower on both thin and thick films than on thin PB fibers the cells grew faster on the thick PB fibers. This might indicate that the cells grow better when they lay over the fibers. The images from the confocal microscope also confirmed this data (Fig.3). The AFM readings (Fig. 2) showed that the DPSCs were softest on the thick fibers in inducing media, which is unusual since the biomineralizing cells are usually harder. The confocal images showed that the cells grew along and on top of the fibers (Fig.3). Furthermore, the mercury lamp images (Fig. 4) showed possible biomineralization in all samples except the thin fibers in inducing media since the cells were not healthy under this condition. Overall, the cells preferred the thick fibers to the thin fibers and the cells were not healthy on the thin fibers in induced media.



Figure 4. Mercury images of dental pulp stem cells on thick and thin polybutadiene fibers after 7 days

<sup>1</sup> Brahim, J., et al; "Postnatal human dental pulp stem cells (DPSCs) *in vitro* and *in vivo.*" *Proceedings of the National Academy of Science*. 97 (2000): 13625 – 13630.

### Substrate Induced Osteoblast-Like Differentiation of Stromal Stem Cells

Jacqueline Belizar<sup>1</sup>, Reena Glaser<sup>2</sup>, Matthew Hung<sup>2</sup>, Marcia Simon<sup>4</sup>, Jay Gao<sup>4</sup>, Miriam Rafailovich<sup>3</sup> Chungchueh Chang<sup>3</sup> Alice Shih<sup>4</sup>

1. Miller Place High School, Miller Place, NY 2. Smithtown West High School, Smithtown, NY, 3. College of Engineering, Stony Brook University, Stony Brook, NY 4. School of Dental Medicine, Stony Brook University, Stony Brook, NY

Typical procedures for growing bone grafts often lead to complications such as immunorejection, a long operation period, and excess costs (1). This study focused on the potential of Adipose-derived Stem Cells (ASCs) to differentiate along an osteogenic lineage, providing an autologous cell source that involves a non-invasive harvesting procedure. We have demonstrated that these cells can be induced to biomineralize and form calcium phosphate fossilization purely by plating the cells on a polybutadiene (PB) substrate created by spincasting. In contrast to ASCs that were grown on tissue culture plastic, ASCs grown on PB generated calcium phosphate deposits within 7 days in the absence of added dexamethasone. In addition, Confocal microscopy showed that the cells continued to proliferate and retain viability while biomineralizing. The most calcium phosphate crystallization was exhibited on the thin PB surface in a non-induced media as seen on the SEM/EDAX. Concurrently, the control setup-where the cells were plated on tissue culture plastic-did not produce any deposition. Moreover, biomineralization did not occur on the plastic surface in the absence of dexamethasone. In order to determine whether the induction was permanent or transient, we removed the cells exposed to polybutadiene after 14 and 28-day incubation periods, and then cultured the cells for another five days on plastic. These "educated" cells continued to form calcium phosphate mineralization on the plastic substrate, thus proving them to have become stably differentiated. ASCs were then grown on thin and thick PB fibers, where the moduli was unaffected by the thickness of the fibers as seen in figures 1a and 1b. Nonetheless, there was an inverse relationship between the ASC modulus and the dynamic modulus of the PB surface; thus, demonstrating the ability of the cells to sense the mechanics of the substrate on which they were plated. Confocal imaging displayed cell growth along the fibers as calcium phosphate deposits formed. Further data is being collected using RT-PCR and immunohistochemistry for RNA and protein expression, X-ray Scattering microscopy, and continued use of the AFM, Confocal, and SEM/EDAX. Preliminary experiments revealed that runx2 expression increased only in cultures exposed to dexamethasone and that its expression did not correlate with the increased calcium deposition promoted by ASC on PB. This suggests alternative mechanisms to those regulating biomineralization. Due to our positive findings regarding ASCs and their ability to promote calcium phosphate deposition, these easily isolated cells may be considered for development of adjunct therapy for the production of safe autologous bone grafts



Fig. 1a: Moduli of Day 1 PB fibers

Fig. 1b: Moduli of Day 7 PB fibers

1. Bajada S, Mazakova I, Richardson JB, Ashammakhi N. Updates on stem cells and their applications in regenerative medicine. J Tissue Eng Regen Med 2008 Mar 11; 2: 169-83.

### Short and Long Term Stem Cell Lineage Determination: Use of Polymers/Copolymers with Various Mechanical Properties

Benjamin Parnes [1], Michael Schiff [1], V. Jurukovski [2,3], M. Simon [2], M. Rafailovich [2], C.C. Chang [2] [1] HAFTR High School, [2] SUNY Stony Brook, [3] Suffolk County Community College

Currently the recovery period for a broken bone can be weeks or months. Dental Pulp Stem Cells (DPSCs) possess the potential to reduce this recovery period. DPSCs are multi-potent cells that possess the potential to differentiate into neurogenic, osteogenic/odontogenic, adipogenic, myogenic, and chondrogenic-like cells [1]. However, our main concern is differentiation into osteoblasts for bone tissue growth. This ability would enable these cells to be implanted onto scaffolds which would then be placed into the body to help restore broken bone, and speed up recovery time for bone injuries. Currently, one of the main challenges facing this goal is finding the correct polymer surface with the correct thickness to ensure optimal cell proliferation and differentiation.

Past research has indicated that DPSCs grow poorly on Polystyrene (PS), and grow well on Polybutadiene (PB). We propose to study a copolymer, a blend of PS and PB (65% PS: 35% PB) as base material for scaffold building. Our hypothesis was that a copolymer scaffold of PS/PB would be better suited than the PS alone because of the biocompatibility of PB. Although PS does not support cell growth it has mechanical properties that can assemble an effective scaffold. PB on the other hand is a rubber-like substance that cannot be used to construct a sturdy scaffold. Individually one of the polymers might not support cell growth, however blend of two can. Therefore, the best scaffold may be built by blending different polymers of different mechanical and biological properties.

The DPSCs were analyzed using confocal microscopy which showed that the DPSCs did not grow on the PS, and were on the verge of dying (Figure 1a). However, the DPSCs grew well on both PB and the PS/PB copolymer (Figures 1b and 1c). Additionally, Mercury Lamp images showed presence of crystals indicating that biominerilization had occurred in all samples except PS (Figure 2). The results show that scaffold formation can be achieved by using copolymers with different mechanical and biological properties proving our hypothesis to be correct. In addition, we are conducting a project where the DPSCs are induced to differentiate for a short time period on thin PB and are then transferred to thick surfaces that do not induce differentiation. Will the cells "remember" their "education" and continue to differentiate? If the cells continue responding as they did in their primary environment, we could induce differentiation on a substrate and then transfer them into the body, thereby avoiding the adverse effects of chemically inducing cell differentiation.



Figures 1a, b, c. Confocal images of DPSC grown on PS, PB, and PS/PB copolymer films of different thicknesses.

[1] U.S. Department of Health and Human Services. Scientists Discover Unique Source of Postnatal Stem Cells. NIH News. 21 Apr. 2003. Web.

<http://www.nidcr.nih.gov/Research/ ResearchResults/NewsReleases/Arc hivedNewsReleases/NRY2003/PR0 4212003.htm>.

### An Analysis of the Toxicity of Inert Metal Nanoparticles on Dermal Fibroblasts and Pre-adipocytes **Ranjeet Kaur**<sup>1</sup>, **Kelsey McKenna**<sup>2</sup>, **Tatiana Mironova**<sup>3</sup>, **Dr. Miriam Rafailovich**<sup>3</sup> <sup>1</sup>Half Hollow Hills High School West, Dix Hills, NY; <sup>2</sup>South Side High School, Rockville Centre, NY; <sup>3</sup>SUNY Stony Brook, Stony Brook, NY

Especially in recent years, practical applications of nanoparticles have developed and become commonplace. Gold and platinum nanoparticles in particular are used for drug delivery, and cancer detection and treatment.<sup>1</sup> However; these particles can be detrimental to the health of normal cells in addition to target cells.

Previous research has shown the toxicity of gold nanoparticles through their ready uptake by dermal fibroblasts in regular medium. The nanoparticles in this study accumulated in the vacuoles of the cell and resulted in rapid cell death.<sup>2</sup> In another study, nanoparticles scarcely penetrated adipocytes in adipogenic medium. Adipogenic medium is designed to aid in the differentiation of pre-adipocytes into mature adipose cells. In addition to the standard nutrients and anti-biotics necessary for cell growth found in regular media, adipogenic medium contains dexamethasone and insulin. The results of these two experiments left it unclear whether the penetration of the nanoparticles is due to the type of cell (and whether or not it is differentiating) or the type of medium.

We plated dermal fibroblasts and pre-adipocytes in adipogenic and regular medium. We used cells harvested from tissue of the same individual to further eliminate error. 24 hours after plating, we added 13 and 45 nm gold nanoparticles in different concentrations. We used 100, 150, and 200 µl of nanoparticle solution for each size nanoparticle. This resulted in concentrations of .086, .124, and .158 mg/mL for 13 nm nanoparticles, and concentrations of .012, .017, and .022 mg/mL for 45 nm nanoparticles. We counted cells and conducted confocal microscopy at two time points: 3 and 6 days after the addition of the nanoparticles. The confocal microscopy was also further viewed at two separate magnification intervals: 20x and 63x (Figures 1A, and B, 6 Day time point). In the two dermal fibroblasts' controls, we saw confluence in the regular medium, while only 40-50% of the cells grew in the adipogenic medium because dermal fibroblasts do not differentiate in adipogenic medium.

Figure 1A: 0.022mg/mL Au Dermal Fibroblasts in Regular Medium 20x



Figure 1B: 0.022mg/mL Au Dermal Fibroblasts in Adipogenic Medium 20x



We will use the TEM to determine if the nanoparticles were absorbed by the cells. If the nanoparticles are absorbed by the dermal fibroblasts in adipogenic medium, uptake is

most likely based on cell type, and the gold nanoparticles affect skin cells more aggressively than fat cells. If the nanoparticles are not absorbed, the adipogenic medium most likely changes the chemistry of the cell. We would also like to observe if the nanoparticles penetrate the cell membrane or just surround the cell.

Additionally, we exposed separate CF-29 dermal fibroblasts to .077 mg/mL and .141 mg/mL in stock solution of platinum nanoparticles. We found that because the platinum nanoparticles are dramatically smaller (roughly 2 to 4 nm) than the gold nanoparticles, the platinum penetrated faster and more completely into the cells than gold. We counted cells after 26, 48, and 73 hours (Figure 2: Growth Curve). We observed rapid cell death, with the larger concentration of nanoparticles resulting in more extreme damage. (Figures 3A, B, and C: Confocal images, 2 Day time point).





**Figure 3A**: Control CF-29 Dermal Fibroblasts 63x





Figure 3B: 0.077 mg/mL Pt Figure 3C: 0.141 mg/mL PtCF-29 Dermal Fibroblasts63x63x

<sup>1</sup>Borm, Paul JA and Wim H De Jong. "Drug delivery and nanoparticles: Applications and hazards." Int J Nanomedicine Jun 2008 133-149. Web. 7 Aug 2009. <a href="http://www.pubmedcentral.nih.gov/articlerender.fcgi?tool=pmcentrez&artid=2527668">http://www.pubmedcentral.nih.gov/articlerender.fcgi?tool=pmcentrez&artid=2527668</a>>.

<sup>2</sup>Pernodet, Nadine, et al. "Adverse Effects of Citrate/Gold Nanoparticles on Human Dermal Fibroblasts." *Wiley Interscience* 2.6Jun 2006 766-773. Web.14 Jul 2009. <a href="http://www3.interscience.wiley.com/cgi-bin/fulltext/112598020/PDFSTART">http://www3.interscience.wiley.com/cgi-bin/fulltext/112598020/PDFSTART</a>.

## **Session 8**

### The Fibrinogen Molecule

Session Chairs: Alex Ramek, Harvard University, Sara Snow, Stern College for Women

Graduate Mentors: Dr. Yantian Wang, Xaiolan (Nancy) Ba Department of Materials Science and Engineering, Stony Brook, University, Stony Brook, NY



## The Effects of Glucose, Curcumin, and Biomineralization Solution on the Formation of and Development of Fibers from Fibrinogen with respect to Topography, Modulus and Chemical Makeup.

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Fibrinogen is a nonglobular glycoprotein found in the bloodstream involved in the process of clotting. During coagulation, platelets aggregate at the wound site and a signal is sent to release the thrombin into the bloodstream. When thrombin reaches the fibrinogen molecules, it cleaves the peptides at the E domain so that the molecules can self-assemble to form fibrin networks (see Fig. 1). Along with the platelets, these form a fiber network that make up a clot.

Previous research has shown that even without thrombin, hydrophobic surfaces can produce a similar effect on the fibrinogen such that it will behave as though it has been cleaved and self-assemble<sup>1</sup>. Using this information, samples were prepared so the effects of glucose, curcumin, and biomineralization solution on both fibrinogen and fibrin fibers could be studied.

People with diabetes and excess glucose tend to have abnormal clotting. The purpose of this study was to determine the effects of glucose on both the formation and development of fibers from fibrinogen. The effects of glucose on fibrinogen were tested in four concentrations (0 mg/dL, 81 mg/dL, 162 mg/dL, and 270 mg/dL) based on the idea that diabetic glucose level is 120 mg/dL or greater. Samples of polystyrene (PS) were spun on clean, hydrophobic silicon wafers. After annealing for two hours, the samples were taken out of the oven, and a solution of 450  $\mu$ m of TBS EDTA and 50  $\mu$ m of fibrinogen (per sample) was added. The samples were allowed to incubate for about three days. After incubation, thrombin was added to each sample, and the samples were analyzed under the atomic force microscope (AFM).

In studying the effects of incubating fibers in a biomineralization solution, PS thin films were spun on clean, hydrophobic silicon wafers. The samples were annealed in the oven overnight and later incubated in solutions of fibrinogen or fibrinogen and thrombin. The biomineralization solution was then added to all samples except for the control (zero hours in solution). The samples were examined under the AFM at various time points (e.g. zero hours, two days, five days, etc.) to check for fiber formation, height, and modulus data. The expected findings are that over time, the samples will show biomineralization, indicated by an increase in hardness and also calcium and phosphate content under the scanning electron microscope (SEM).

Future research will involve further analysis of the mechanics of the fibers under the AFM with the topography, friction, and modulus options. There are also plans to study the effects of curcumin similar to the study on glucose, but as it pertains to burn victims. Past observations show that people with untreated second degree burns tend to have their burns progress to the third degree. The theory tested here is that the clotting of the local blood vessels cuts off the supply to the surrounding tissue, effectively killing it.



<sup>1</sup>J. Koo et al. Influence of Chemical Properties of Solid Surfaces on Fibrinogen Self-Assembly, unpublished, 2009

#### Characterizing Interactions between Fibrinogen Dimmers During Fiber Self-Assembly: A Study of Biomaterials in Contact with Blood

Pooja Rambhia<sup>1</sup>, Alex Ramek<sup>2</sup>, Adam Fields<sup>3</sup>, Sara Snow<sup>4</sup>, Yantien Wang<sup>6</sup>, Dennis Galanakis<sup>6</sup>, Miriam Rafailovich<sup>6</sup>

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Fiber formation plays a critical role in the process of wound healing, and is also the driving force behind blood clotting. Conversely, inhibition of clotting on blood-contact biomaterials is crucial for preventing deleterious thromboses and improving patient prognoses. In the human body, vascular stents are crucial in the process of correcting artherosclerosis; however, research has shown that current polymer-coated stents induce fiber formation, thus leading to detrimental clotting.<sup>1</sup> Our research was premised on probing surfaces with various chemistry to understand if surface interactions alone could influence the spontaneous formation of a clot. To do so, I exposed polystyrene surfaces with supercritical carbon dioxide to see if protein adsorption was at all affected. I was successful in finding that polystyrene exposed to supercritical carbon dioxide inhibits fiber formation where as normal polystyrene induces fiber formation. This find has potential in the development of optimal stents because polystyrene is a polymer commonly used in the making of such biomaterials. By exposing these coated stents to



Figure 1. Fibrinogen fiber formation on different polymer surfaces

supercritical carbon dioxide, the thrombosis known to occur on stents can be effectively prevented.

This study also served to examine the interactions between epithelial cells and fibrinogen fibers. Normally in the body, clots are formed by the migration of epithelial cells and platelets onto a network of fibrin fibers. In order to see if the fibrinogen fibers being produced via polymer surfaces can produce legitimate clots, the interactions between these cells and fibers is crucial. Therefore, I plan to use confocal microscopy to observe the migration of these cells onto fibrinogen fibers. Additionally, I plan to use atomic force

microscopy to conduct mechanical testing on the fibrinogen fibers.

<sup>1</sup> Nonthrombogenic Plastic Surfaces, R. I. Leininger, C. W. Cooper, R. D. Falb, and G. A. Grode *Science* 152 (1966): 1625-1626.



## Effects of Nanoparticles on Cells

**Session Chairs:** 

Rafael Holzer, Yeshiva University, Ava Rosenbloom, Stephen Poon Stony Brook School of Dental Medicine

Graduate Mentors: Lourdes Collazo, Chien-Hsiu Lin Department of Materials Science and Engineering, Stony Brook, University, Stony Brook, NY



### <u>The Effect of Iron Oxide on Cells and their Biomineralization</u> Kimberly Lombardi, *St. John the Baptist DHS* Lourdes Collazo, Rafael Holzer, Ava Rosenbloom, Stephen Poon, Miriam Rafailovich Department of Materials Science and Engineering, Stony Brook University

Iron oxide nanoparticles are used in a variety of cosmetics, drug capsulations and other products. It is mostly used as a coloring agent in cosmetics such as foundation and eye shadow. A review done by Skin Deep Environmental Working Group's Cosmetic Safety Database on some of Estee lauders products have indicated that some ingredients in their products are linked with cancer, developmental/reproductive toxicity, allergies, etc. The same review stated that iron oxide in one of their products caused enhanced skin absorption, persistence and bioaccumulation<sup>1</sup> The flow of iron oxide in the body begins to infiltrate from the skin to the bones. Therefore this project consist of the study of the effects of iron oxide on bone cells (MC-3T3).

On day zero, 3,000 MC-3T3 cells were plated in 24 well dishes. Nanoparticles were added the next day using alpha-MEM containing 10% Fetal Bovine Serum. The media contained different types of Iron Oxide (red, yellow, blue and black) at different concentrations (0.1 mg/mL, 0.4 mg/mL and 0.8 mg/mL). After counting the cells and observing them through the Confocal Microscope, it was concluded that the cells died and new concentrations were substituted (0.05 mg/mL and 0.1 mg/mL). The osteoblasts did not survive either concentration for the Iron Oxide yellow and Iron Oxide black (Figure A), as observed under confocal, whereas the cells exposed to blue, and red Iron Oxide nanoparticles grew successfully (Figure B).

With different living capacity of cells in different Iron Oxide and their concentrations it is safe to say the less Iron Oxide the better off people are. Also, the more scientists know about these Iron Oxides the better the chances are to find living concentrations for cells. Further experiments are needed to gain this data. These experiments consist of using the transmission electron microscope (TEM) to find the size of the Iron Oxide. This will show if the size of the Iron Oxides affects the rate they enter and kill the cell. Also a living concentration will be found where the cells survive and grow at a normal rate with. Once that concentration is found it can be used in biomineralizing the cells. That will be done to see if the process of biomineralization is affected by the Iron Oxide. After seeing how the Iron Oxide affect biomineralization, the Iron Oxide can be tested on other types of cells, for example cancer cells or epithelial cells. The future hold many answer, that will soon be revealed.



Figure A -Iron Oxide Black on MC Cells Day 3

Figure B- Iron Oxide red 0.1mg/ml day 3 (left), Iron Oxide blue 0.1 mg/ml (middle and right)

Reference :

<sup>1</sup> <u>http://www.cosmeticsdatabase.com/product.php?prod\_id=120511</u>

#### Effect of Zinc Oxide and Iron Oxide Nanoparticles on MC-3T3 cells

Anisha Kapoor, *Wheatley School;* Sonya Prasad, *Wheatley School* Kim Lombardi, *St. Johns Baptist DHS* Lourdes Collazo, Rafael Holzer, Ava Rosenbloom, Stephen Poon, Miriam Rafailovich Department of Materials Science and Engineering, Stony Brook University

Zinc Oxide nanoparticles are abundant in a variety of skin care, cosmetic and sunscreen products. Cosmetics in which zinc oxide nanoparticles are present, such as most mineral makeup, can benefit the products for its non-inflammatory properties, calming effect on acne and rosacea, and its sun protection from absorbing UV rays.<sup>1</sup> An additional nanoparticle was used, Zn66 coated with a hydrophobic polymer. Despite these factors, high concentrations of these nanoparticles can have a possible negative effect on cells. This study focused on the optimal concentration of nanoparticles for cell growth of MC-3T3-E1 osteoblasts cells.

Cells were plated at a density of 3,000 cells/cm<sup>2</sup> and exposed to alpha-MEM containing 10% Fetal Bovine Serum. The media contained Zinc Oxide and Zinc-66 at different concentrations (0.05 mg/mL, 0.1 mg/mL and 0.4 mg/mL). The cells did not survive these concentrations, so new concentrations were substituted, 0.03 mg/mL and 0.6mg/mL for both nanoparticles. The osteoblasts were counted using a hemacytometer and observed under the Confocal Microscope. For Zinc Oxide (at 0.03mg/mL and 0.06mg/mL concentrations), cells grew well on day 1 (Figure B), but began to die by Day 4 (Figure A).

More information is needed on these nanoparticles to find the optimal concentrations to resist toxicity in cells. Further research is needed using the transmission electron microscope (TEM) to find the size of the nanoparticles so we can better determine the size of the nanoparticles and if they can enter the cell. Once that concentration is found, it can be used to determine the effects in biomineralization.

Figure B: Bone cells incubated with ZnO



Figure A: Day 4 Control (left), ZnO 0.03 (middle), ZnO 0.06 (right)



<sup>1</sup> <u>http://www.azom.com/details.asp?ArticleID=2357</u>

### Investigating the Effects of Varied Concentrations of Titanium Dioxide on the Growth and Biomineralization of Osteoblast Cells

**Bracha Einzig, Talya Lent,** Yeshiva University High School for Girls (Central), Holliswood New York Lourdes Collazo, Miriam Rafailovich, Show Lin, Stony Brook University, Stony Brook New York

Titanium Dioxide is a major component used in the coating of many artificial medical implants which are in contact with Osteoblasts. Titanium Dioxide is also found in toothpaste, which comes in contact with teeth. Two types of Titanium Dioxide were used in this experiment- Titanium Dioxide Rutile and Titanium Dioxide Anatase the latter being the more deadly of the two.<sup>1</sup>

The experiment was to determine the impact of Titanium Anatase and Rutile on cell viability and the capacity for biomineralization. Concentrations of 0.1, 0.4, and 0.8 mg/ml of both Rutile and Anatase.

Table 1

	0.01	0.03	0.05	0.075	0.1	0.4	0.6
Rutile	+	ND	+	±	_	-	-
Anatase	+	±	1	ND	-	l	_

In the first experiment we found that  $TiO_2$  Anatase could be used at 0.01 mg/ml and  $TiO_2$  Rutile could be used at 0.05 mg/ml (see Table 1)

After obtaining results from analysis of the cell growth; it was found that cells in Titanium Dioxide Rutile concentrations of 0.01 and 0.05 (Figure2) were viable when compared to the control (Figure1). Titanium Dioxide Anatase did not survive the higher concentration of 0.05 mg/mL (Figure3, Figure4).

Based on the results on table 1, we will use concentrations of 0.05 mg./ml of  $TiO_2$  Rutile and 0.01 mg/ml for  $TiO_2$  Anatase for biomineralization



Figure 1. Osteoblast cell control after 2 days

Figure 2: Osteoblast Cell Rutile .05 after 2 days

Figure 3: Osteoblast Cells Anatase .01 after 2 days

Figure 4: Osteoblast Cells Anatase .05 after 2 days

1. Zhi Pan , Wilson Lee , Lenny Slutsky , Richard A. F. Clark , Nadine Pernodet , Miriam H. Rafailovich-"Adverse Effects of Titanium Dioxide Nanoparticles on Human Dermal Fibroblasts and How to Protect Cells" Wiley Inter Science Volume 5 Issue 4, Pages 511 – 520. 10/14/08

#### The Effects of Titanium Dioxide and Zinc Oxide Nanoparticles on Adipocytes and Dermal Fibroblasts

Kayla Applebaum [1] Alana Warhit [2] Chienhsiu Lin [3] Dr. Marcia Simon [3] Dr. Miriam Rafailovich [3] Ma'ayanot High School [1] HAFTR High School [2] Stony Brook University

Titanium dioxide and Zinc oxide are the main components of sunscreens and cosmetic products. In addition, these chemicals are used in most toothpastes and household items. Although the function of titanium dioxide and zinc oxide in sunscreens is to protect us from sunburns and UV penetration of our skin, they are also photoactive chemicals. This means that UV radiation will catalyze ROS (reactive oxygen species) production, which is known to damage DNA. Previous studies have shown that the two structures of titanium dioxide, rutile and anatase, hinder cell function for human dermal fibroblasts. Human dermal fibroblasts, when exposed to titanium dioxide, have decreased in cell area, cell proliferation, mobility and ability to contract collagen. However, particles that were coated with a dense grafted polymer brush were prevented from penetrating the cell, which in turn decreased the production of ROS.<sup>1</sup> This study has shed light onto the potential that this coating contains in preventing tissue and cell damage. In addition, evidence has shown that titanium dioxide kills keratinocytes and impairs cell function.<sup>2</sup>

The primary focus of our project is the effects of titanium dioxide and zinc oxide nanoparticles on preadipocytes and dermal fibroblasts. Although dermal fibroblasts have been previously studied, we used concentrations of these nanoparticles on the dermal fibroblasts that differed from previous experiments. We tested adipocytes because these are the cells from the hyper-dermis layer, after the keratinocytes and dermal fibroblasts. We cultured the cells in an adipogenic media for 3 weeks. We had 3 Petri-dishes, with 6 wells in each, and 20,000 cells per well.

We ran two I-DNA gel electrophoresises to observe the DNA breakage. When exposed to UVB for 3 hours, rutile and anatase titanium dioxide and zinc oxide nanoparticles break the DNA. However, the zinc coating #66 and the titanium dioxide coating prevent DNA breakage (Figure 1). In fact, the coating proves better than the control with UV exposure. Similarly, when exposed to UVA for a period of 4 and 6 hours, the nanoparticles induced DNA breakage (Figure 2).

Although we have not finished calculations, under a confocal microscope the adipocytes from the control seemingly remain much healthier than the cells exposed to nanoparticles. The coating appears to be an effective protection for the cells. The dermal fibroblasts also appear as we expected. The cells that were exposed to nanoparticles seem to have shrunken and died out more (Figure 3).

Although our results led us to conclude that adipocytes and dermal fibroblasts are adversely affected by these nanoparticles, there was a possibility that these nanoparticles would not penetrate the skin far enough to reach these cells. Therefore, we took a cross section of skin from a human being and coated pieces of skin with these nanoparticles, with some exposed to UV radiation. Under a microscope, we were able to see that these toxic chemicals were able to penetrate deep enough into the dermal fibroblasts and the hyper-dermis layer of skin- the adipocytes.



1 Z. Pan, W. Lee, L. Slutsky, R. Clark, N. Pernodet, and M. Rafailovich, Small 2009, 5, No. 4, 511–520

2 C. Lin, M. Simon, V. Jurukovski, W. Lee, M. Rafailovich, American Physical Society 2009

## Session 10

### Cells Under Stress

**Session Chairs:** 

Alicia Franco Stony Brook University Yehuda Isseroff, Queens College

Graduate Mentors: Tatsiana Mironava, Monica Apostol, Department of Materials Science and Engineering, Stony Brook, University, Stony Brook, NY, Dr. Ting Nie, Department of Medicine, Stony Brook University, Stony Brook, NY Rebecca Isseroff, Lawrence High School, Cedarhurst NY



**Safety in Numbers?: Evaluating Cell Density's role in responses to Oxidative Stress** Sanjay Palat<sup>3</sup> Dr. Richard Clark<sup>1</sup>, Dennis Kim<sup>2</sup>, Michael Sperandeo<sup>1</sup>, Dr. Miriam Rafailovich<sup>1</sup>,

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Oxidative stress has been shown to be damaging on both the cellular and the organismal level, with potential consequences including apoptosis and necrosis, and numerous degenerative diseases, respectively. Although several mechanisms for combating oxidative stress have been identified, including the use of enzymes and non-enzymatic compounds such as glutathione, a factor whose effect on oxidative stress responses that has yet to be explored is that of cell density. Previous investigations by members of the laboratory under Dr. Clark suggest a direct correlation between cell density and its protective effects during periods of oxidative stress. It is hypothesized that the reason for this protective role lies in the fact that a denser sample of cells has a higher total amount of antioxidant compounds such as reduced gluthathione, bestowing a community benefit upon the individual cell.

To test this hypothesis, we plated human dermal fibroblasts (CF31) at varying densities in quadruplicate, and subjected them to incremental doses of hydrogen peroxide, a reactive oxygen species naturally occurring in the human body as a by-product of oxygen metabolism. After twenty hours of incubation, an XTT assay was used to measure cell viability in the samples. Initial results failed to show cell death at expected ranges of hydrogen peroxide. However, it was realized that sodium pyruvate, an alpha-ketoacid and a component of our cell culture media, was degrading the hydrogen peroxide and therefore reducing the effect of hydrogen peroxide as a source of oxidative stress. After repeating the experiment in pyruvate-free media, we observed cell death at expected ranges of hydrogen peroxide. In a second experiment, we also stressed cells in pyruvate-containing media and expanded the range of hydrogen peroxide doses, and observed cell death only at higher doses of hydrogen peroxide, suggesting that the sodium pyruvate had been acting as an antioxidant. In both experiments, it was observed that the denser a sample was, the more oxidative stress in the form of hydrogen peroxide it was able to withstand without undergoing decreased cellular metabolism. This suggests that cell density in a sample yields a community benefit to the individual cells within that sample. These findings call for greater standardization in the design of experiments studying oxidative stress.



References:

- Gurjala, Anandev., Liu, W. Robert., Mogford, Jon E., Procaccini, Piero S. A., Mustoe, Thomas A. "Age-dependent response of primary human dermal fibroblasts to oxidative stress: cell survival, pro-survival kinases, and entrance into cellular senecence" Wound Repair and Regeneration 13 2005; 565-575
- Desagher, Solange., Glowinski, Jacques., Premont, Joel. "Pyruvate Protects Neurons against Hydrogen Peroxide-Induced Toxicity" *The Journal of Neuroscience* 17(23) December 1, 1997; 9060-9067

#### Evaluating the Effect of Cell Plating Density on UV Exposed Human Dermal Fibroblasts

Tiya Nandi-Jericho High School, Jericho, NY, Dr. Miriam Rafailovich-Stony Brook University, Stony Brook, NY, Monica Apostol- Estee Lauder Laboratories, Melville, NY, Tatsiana Mironava-Stony Brook University, Stony Brook, NY

Previously, we created a novel methodology for the non-invasive release of partially confluent cell sheets in order to mitigate the shortcomings of conventional methods of tissue engineering for organ transplantation. We used the photosensitive triblock copolymer polyvinylpyridine-polyvinylphenylketonepolymethylmethacrylate (PVP-PVPK-PMMA) for the assembly of heterogeneous bilayered tissue constructs of cell sheets through layer-by-layer deposition. Although our application proved to be an optimal method for manipulating non-invasively harvested cell sheets, it required the cell sheets to be exposed to UV light. Previously, UV light has always been known to damage cell functions which were seen after the cell sheets were exposed to UV C for a long time. We wanted to find a wavelength of UV light which would be efficient for this method as well as cause no harmful effects on the cell sheets. Concurrently, we proposed to investigate the exact effects of this UV light on the cell sheets by plating high density cell sheets opposed to cell sheets which had a significantly lower plating density. After exposing both samples to UV B for the same duration and distance, the samples were evaluated based on cell counts (Fig. 1) and confocal imagery. The cell sheets with a higher plating density was seen to survive significantly better than the low density sheets which showed the direct impact of plating density. While UV C was seen to kill the cell sheets, the cells were proliferating healthily under UV B. By eliminating the limitations caused by UV C and plating high density cell sheets, this novel methodology can prove to be effective in the field of tissue regeneration and transplantation.





Reference: M. Apostol. "Photosensitive Film-Forming Polymer for Tissue Engineering Applications." Unpublished Results

### The Effect of Magnetic Fields on Osteoblast Growth and Biomineralization Using An Iron Oxide-Clay Composite

Aaron Akhavan, Joseph Szpigiel, Rambam Mesivta Yehuda Isseroff, Queens College Professor Rebecca Isseroff, Rambam Mesivta Dr. Miriam Rafailovich, Dr. Yizhi Meng, Stony Brook University

It has been proven that magnetic fields have a positive effect on Osteoblast growth. However, the presence of external magnets does not penetrate significantly enough to affect cell growth. Therefore, a magnetic nanocomposite has been created to help enhance the magnitude of the magnetic field.<sup>1</sup> The goal of this experiment is to use the magnetic nanocomposite within a biocompatible polymer to effectively enhance the effects of the external magnetic field on Osteoblasts. In order to recreate such a nanocomposite, 3.4 ml of Iron Pentacarbonyl was added to 5g of Cloisite 20A and was then left to oxidize for a period of 48 hours. The composite was sent to Columbia Analytical Services to determine the percentage of iron in the composite. The results showed that 15% of the composite was made up of iron. The sample was also sent for a Mossbauer test to determine the oxidation number of the iron particles, the results of which are still being processed.

In order to test the survivability of the Osteoblasts on the iron oxide-clay nanocomposite, thin films of various concentrations were spuncast onto glass coverslips and silicon wafers. Twenty-five mg/ml solutions of the nanocomposite and Poly(methyl methacrylate) (PMMA), a FDA approved polymer for bone cement, were made in Toluene, with varying concentrations of the nanocomposite (5%, 10%, 15%) to PMMA. The same was done with Cloisite 20A to make sure that it was the presence of the iron oxide particles that were affecting the Osteoblast proliferation. The thin films were viewed on the Atomic Force Microscope (AFM) to see whether there was a uniform film coat.

Cells of the MC 3T3 line (Osteoblasts derived from mouse skulls) were plated onto the thin films. Growth curves are currently being constructed to determine the survivability of the Osteoblasts on the nanocomposite/PMMA thin film. Additional cells are being plated onto thin film surfaces and are going to be induced to biomineralize to test the potential effect of the nanocomposite on the biomineralization of the Osteoblasts.

The composite has also been electrospun with PMMA to produce nanofibers which can be used to create a scaffold for cells to grow on. It is believed that the electrospun fibers will be a better substrate for the cells to grow on. Solutions of PMMA (20% by weight of solution) in choloforom were prepared for electrospining. The nanocomposite was added to the PMMA/chloroform solution with varying percentages (0, 3.5%, and 5% by weight of solution). The resulting fibers were looked at under an optical microscope and pictures were taken under multiple magnifications (Figure 1-3).

The next step in the experiment is to plate cells on the electrospun fibers for growth cuvres and biomineralization tests to determine whether it is a viable substrate for Osteoblast survival. The experiments will be repeated in the presence of a magnetic field to determine the effects of a magnetic field on Osteoblast growth and biomineralization and to see whether the ironoxide-clay nanocomposite will enhance these effects.



Figure 1: Electrospun PMMA fibers under 50x magnification



Figure 2: Electrospun Cloisite 20A + PMMA fibers under 50x magnification



Figure 3: Electrospun nanocomposite + PMMA fibers under 50x magnification

<sup>1</sup> Brenner, Nicole. "Synthesis and Characterization of a Magnetic Nanocomposite that Enhances Bone Cell Growth." ACS Presentation: Aug. 2007. Whole Cell Patch Clamp Analysis of Human Cervical Carcinoma HeLa Cells Exposed to Gold Nanoparticles

**Tara Jain**<sup>1</sup>, **Anjali Kapur**<sup>2</sup>, **Dr. Miriam Rafailovich**<sup>3</sup>, **Tatsiana Mironava**<sup>3</sup>, **Dr. Peter Brink**<sup>3</sup>, **Joshua Kaufman**<sup>3</sup> <sup>1</sup>Moravian Academy Upper School, Bethlehem, PA; <sup>2</sup>Mount Sinai High School, Mount Sinai, NY; <sup>3</sup>SUNY Stony Brook, Stony Brook, NY

Gold Nanoparticles are used in a wide variety of applications, including the facilitation of biological light imaging, heat diffusion, target-specific drug delivery, cancer diagnostics, and biosensing. As the use of gold nanoparticles becomes a common practice in clinical and therapeutic applications, it is important to study the long term effect of these particles on cellular function.<sup>1</sup> In this study, the effect of different sized gold nanoparticles, 13 nm and 45 nm, on HeLa cell current was measured using a patch clamp.

The patch clamp technique involves inserting an electrode into a cell and redirecting the flow of ions through the cell membrane into a glass micropipette. Suction is applied by the micropipette to create high resistance, measured in gigaohms, between the electrode and the cell surface. The average current of the entire cell, measured in picoamperes, is recorded through multiple ion channels simultaneously. A measurement of cell current can be used to determine the efficiency of ion channels in facilitating the passage of ions across the cell membrane. Specific ion channels, including voltage-gated and ligand-gated, are controlled by various proteins and pumps. The ions that flow through these channels, such as K<sup>+</sup>, Na<sup>+</sup>, Ca<sup>2+</sup>, and Cl<sup>-</sup>, play essential roles in cell signaling and communication. Based on levels of current determined by the patch clamp, we can infer whether ion channels have opened or closed.<sup>2</sup> A change in the conductance of these ion channels may indicate a conformational or other type of change in ion channel proteins.

The control HeLa cells were plated without gold nanoparticles, and HeLa cells were also plated and exposed to 13 nm and 45 nm gold nanoparticles for 24 hours. The three conditions of the cells were then patched. Cells were bathed in Tyrode solution to mimic the extracellular fluid, and four consistent, tight patches were performed using the patch clamp and oscilloscope monitor on each condition of the cell. The program Clampex displayed the varying membrane potential of the cell and the corresponding current from the whole cell, and the Clampfit program was used to analyze the data. The HeLa cells were also fixed and stained to be viewed under the confocal microscope on Day 2 to determine the cytotoxity of the gold nanoparticles, as seen in Figure 1.

The exposure to 13 nm gold nanoparticles reduced the outward and inward current of the cells significantly at different membrane potentials, as seen in Figure 2. The control cells were able to produce a current above 1000 pA, while the cells exposed to the 13 nm particles produced almost no current. These results illustrate negative effects of 13 nm gold nanoparticles on the ion channels of HeLa cells. This disrupts cell signaling and communication, essential to cell survival. The 45 nm gold nanoparticles are being patch clamped, and growth curves are being produced for the three conditions of HeLa cells.



Figure 1a Control Group HeLa cells show undisturbed growth



Figures 1b and 1c Cells exposed to 13 nm and 45 nm from left to right



References: 1. N. Sanvicens and M.P. Marco, *Multifunctional Nanoparticles* – properties and prospects for their use in human medicine 2. L.S. Liebovitch and P. Krekora, *The Physical* 



I-V curve comparing HeLa exposed and not exposed to 13 nm gold nanoparticles

### Investigating The Effect of Platinum Nanoparticles in HeLa Cells

Jonathan Millings – Emanuel Christian Academy Tatsania Mironava, Joshua Kaufmam, Dr. Miriam Rafailovich, SUNY Stony Brook

HeLa cells are human epithelial cervical cancer cells. They were the first human cells continuously grown in culture. They will continue to grow and divide indefinitely. Platinum is an inert metal, with the ability to change properties making it useful as a chemotherapeutic drug deliverer. Because cancer cells have more abundant folate receptors than regular cells, combining folic acid with platinum(IV) is used in chemotherapeutic drugs to facilitate increased uptake in cancer cells.

In this investigation, platinum folate nanoparticles were synthesized. A sample of 30,000 cells was exposed to a platinum nanoparticle concentration of  $0.0404\mu$ L.The cells were then incubated for 24 hours. The amount of current emitted by the cell was measured using a patch clamp. Patch clamping recording was accomplished with a glass pipette under a microscope. A very small pipette connected to an electrode was attached to a cell membrane and suction was applied through the pipette. A cell membrane patch was sucked into the glass pipette and formed a high electrical resistance seal. The current that passed through the ion channels in either the membrane patch or the whole cell membrane was then recorded at different voltages. The patch clamp measured the activity across the ion channels of a cell.

We expect an increase in the electrical output of the cells because of the penetration of the platinum nanoparticles. Further studies in this area would include varying the concentrations of platinum folate nanoparticles.



HeLa cels (X20)



HeLa cells with Platinum Nanoparticles (X252)

1. J. Seo, C. Ionescu-Zanetti, J. Diamond, R. Lal, L. P. Lee "Integrated multiple patch-clamp array chip via lateral cell trapping junctions" *Applied Physics* Letters 2004. **84**, pp.11-15

2. Markus Galanski and Bernhard K. Keppler. "Searching for the Magic Bullet: Anticancer Platinum Drugs Which Can Be Accumulated or Activated in the Tumor Tissue": *Anti-Cancer Agents in Medicinal Chemistry* 2007. **7**, pp. 55-73.

3. "What are Ion Channels?" July 16, 2009. Copyright 2004-2007. ICMG Ltd <u>http://</u>www.ionchannels.org

### Phosphoibuprofen Administration and Quantum Dot Imaging Using Nanoparticle Vehicles in Cancer Cells

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Current chemotherapeutic cancer treatments lack specificity, have uncontrolled release within the human body, and are highly toxic to normal cells. In addition, the physical excision of tumors, though specific and effective against fully-developed cancer cells, frequently is unable to remove cancer stem cells, the sources of tumors. For these reasons, researchers have explored the possibilities of encapsulating drugs within nanoparticles to improve the effectiveness of drug delivery.<sup>1</sup>

The purpose of this study is to compare the efficiency of drug release and cytotoxicity of drug delivery vehicles in normal and cancer cells among Poly(ethylene oxide-b-caprolactone) (PEO-b-PCL) micelles,

liposomes, and PAMAM G5 dendrimers. In order to measure the cytotoxicity of the different nanoparticles, MTT and CellTiter-Blue<sup>™</sup> assays of both BXPC3 (pancreatic cancer cells) and NCM460 (normal colon cells) were conducted. Fluorescence and absorbance readings from a spectrophotometer were interpreted to determine respective  $IC_{50}$  values. Furthermore, CdSe/ZnS quantum dots were analyzed for their potential in imaging drug delivery systems as drugs are delivered to cancer stem cells. Through counting and confocal microscopy on

	BXPC3		NCM460		
	24hr (uM)	72hr (uM)	24hr (uM)	45hr (uM)	
Drug	96.2	54.0		99.7	
Old LD + Drug	71.5		103		
New LD + Drug	103.2		165		
PEO-b-PCL	Nontoxic	Nontoxic		Nontoxic	
PEO-b-PCL+					
Drug	221	114		404	
PAMAM	<180uM*	<1.40uM*		<12.5uM*	
PAMAM +					
Drug	<180uM*	<1.40uM *		<12.5uM*	

\*Literature shows PAMAM  $I_{C_{50}}$  value at 24hr to be approx. 160nM<sup>2</sup> Figure 1:  $I_{C_{50}}$  values of different drug delivery vehicles for BXPC3 and NCM460 cell lines



Figure 2: BXPC3; 3 days; PEO-b-PCL + CdSe/ZnS in Medium; [CdSe]=750nM; [PEO-b-PCL]=15uM; AF=green;

BXPC3 and NCM460 cells exposed to quantum dot film and quantum dot solutions, the effect of the quantum dots on cells was determined.

Initial cell toxicity results show that PEO-b-PCL micelles are nontoxic to both cell lines while PAMAM dendrimers are extremely toxic (Figure 1). This result coupled with the significantly lower toxicity of PEO-b-PCL micelles led us to conclude that PEO-b-PCL micelles and liposomes may be viable alternatives for phosphoibuprofen (Drug D) delivery. Furthermore, the results also indicate that Drug D may potentially be more toxic to cancerous cells than normal cells. Our confocal images indicate that quantum dots, specifically CdSe/ZnS, are able to successfully diffuse into cancer cells and be imaged (Figure 2).

Analysis of quantum dot cell toxicity studies as well as confirmation of our initial findings have yet to be completed. Long term future goals of this study include developing an economical method of making PAMAM dendrimers less toxic,

adsorbing multiple quantum dots to PAMAM dendrimers to increase the intensity of emissions, synthesizing biodegradable photoluminescent polymers as a nontoxic substitute for quantum dots, and conjugating drug delivery vehicles to known cancer-targeting molecules such as biotin and folic acid for enhanced specificity.

<sup>2</sup> Lesniak, Wojciech et al. "Synthesis and Characterization of PAMAM Dendrimer-Based Multifunctional Nanodevices for Targeting

<sup>&</sup>lt;sup>1</sup> Brannon-Pappas, Lisa et al. "Nanoparticle and targeted systems for cancer therapy." Science Direct 56 (2004): 1650-1659. Print.

 $<sup>\</sup>alpha_{\nu}\beta_{3}$  Integrins." Bioconjugate Chemistry 18(2007): 1148-1154. Print.



### Filtration

**Session Chairs:** 

Shuan Cheng Stony Brook University

**Graduate Mentors: Ying Liu,** Department of Materials Science and Engineering, Stony Brook, University, Stony Brook,

Rebecca Grella, Brentwood High School, Brentwood, NY



### The creation of high flux nanofiltration membranes with interfacially polymerized polyamide on cellulose nanofibers

Matthew Kim, Xiao Wang, Hongyang Ma, Miriam Rafailovich

The pure water crisis is one of the world's most pressing modern issues. Due to recent shortages in the clean water supply, 1.1 billion people are currently without access to fresh water (World Water Council 2009). Fortunately, pressure-driven membrane filtration processes have arisen as solutions to the growing water crisis. The advances in this type of technology allow it to remove particles as small as monovalent ions (Water Partners International 2009). Membrane filtration is subdivided into the categories of microfiltration, ultrafiltration, nanofiltration, and reverse osmosis, each with decreasing pore size and increased ability to reject smaller particles. Nanofiltration membranes, when used for water treatment tend to have pore sizes of approximately 1 to 5 nanometers and are primarily used to remove salts from water. The membrane tested consisted of four layers: a non-woven polyethylene terephthalate (PET) layer, an electrospun polyacrcylonitrile (PAN) layer, a coated cellulose nanofiber layer, and an interfacially polymerized polyamide layer. The combination of the first two layers (PET, PAN) created a microfiltration membrane, whereas the addition of the cellulose nanofibers created an ultrafiltration membrane. However, the polyamide layer allowed for the rejection of salts, thus creating nanofiltration (Yoon 2008). Polyamide has been previously tested when polymerized on the previously stated microfiltration membrane, but never on the ultrafiltration membrane. The membrane was tested against a commercially bought membrane, with the goal being to achieve a comparable rejection rate with increased flux. Thus far, the membrane rejection after being exposed to pressure has decreased, leading to the conclusion that the polyamide layer, because of its thinness and observed non-uniformity (Fig. 1), is being broken by the water. In order to remedy this problem, the thickness of the polyamide layer will be increased.



Fig. 1: SEM image of nanofiltration membrane at 10000x

### The Encapsulation of *Alcanivorax borkumensis* in a Gelatin Hydrogel for Water Filtration Applications

Ijeamaka Anyene, Lauren Herrera, Andrew Franco, William Genova: Brentwood High School, Brentwood, NY, 11717

Ying Liu, Dr. Miriam Rafailovich, Lourdes Collazo, Shuang Chen, Alicia Franco: Stony Brook University

*Alcanivorax borkumensis* is a species of bacteria that is found in the upper layers of the oceans. It plays a vital role in its marine environment by bio-remediating oil-contaminated waters caused by oil spills [1]. Its multiple oil-degrading enzymes make remediating oil stricken water possible. Because of these characteristics, *A. borkumensis* can be looked upon as a primary tool for bioremediation of oil spills.

The bacteria, *Alcanivorax borkumensis* was encapsulated in a hydrogel composed of porcine skin type A (gelatin), Microbial transglutamine (mTG) and Dulbeco's Phosphate Buffered Saline (DPBS). Then the proliferation of *A. borkumensis* in the hydrogel was tested by cutting a new piece of the hydrogel and staining using Live/Dead Staining at twenty four hour intervals. This stain allowed us to view the bacteria under a Confocal microscope in which the live bacteria appears as green, and dead as red (Figure 1).

Our tentative results show that *Alcanivorax borkumensis* is able to effectively proliferate in the gelatin hydrogel; however their survival wasn't as successful as anticipated. As the total amount of bacteria increased, the amount of dead bacteria increased as well (Figure 2). In the future, the reason why the bacteria's survival in the hydrogel was not as successful as anticipated will be explored. Tests on the efficiency of *Alcanivorax borkumensis* for the remediation of oil will be conducted as well as tests on the modules of the hydrogel.



**Figure 1:** Bacteria encased in hydrogel, submerged in tap water. Sample 3 of Day Four



**Figure 2:** The proliferation of a high concentration of bacteria in the hydrogel.

<sup>1</sup> Schneiker, S. et. Al. "Genome sequence of the ubiquitous hydrocarbon-degrading marine bacterium *Alcanivorax borkumensis*" <u>Nature Biotechnology</u>. (2006) http://www.nature.com/nbt/journal/v24/n8/pdf/nbt1232.pdf



### SUMMER SCHOLAR PROGRAM SCHEDULE OF ACTIVITIES

### EVERY DAY STARTS WITH A GROUP MEETING

#### CHECK SCHEDULE DAILY!

	MONDAY	TUESDAY	WEDNESDAY	THURSDAY	FRIDAY
Week of 6/29	29 10:00 AM General meeting 11:00 AM "Thinking outside the box" Dr. Srinivas Pentyala 12:00 PM CAD Tutorial 1:00 PM Lunch 2:00 PM "Skin Bank" Dr. Marcia Simon 3:00 PM ID cards and campus tour	30 10:00AM General meeting 11:00 AM Dr. Michael Hadjiargyrou 12:00 PM "Patents and I.P." Mrs. Donna Tumminello 1:00 PM Lunch 1:45 PM EH&S Training 3:00 PM "Chemical disposal" Vladimir Zaitsev	1 10:00 AM General meeting 11:00 AM Vladimir Jurokowski 11:45 AM Basic Biology – Lourdes Collazo 12:30 PM Hands on Training Dr. Zaitzev, machine shop tour, lunch 3:15 PM Safety Quiz	2 10:00 AM General meeting 11:00 AM <i>Learning</i> <i>Science Database</i> Ms. Godlind Johnson <i>Excel Tutorial</i> Dr. Vladimir Zaitsev 1:00 PM Pizza Lunch 1:45 PM <i>Journal Club</i>	3 HAPPY 4 <sup>TH</sup> OF DULY!
Week of 7/6	<ul> <li>6</li> <li>10:00 AM General meeting Research minute: John Jerome</li> <li>11:00 AM "Operational Safety" Kim Auletta</li> <li>12:00 PM Distribution of lab boxes, lab notebooks, storage, etc.</li> <li>1:00 PM Lunch</li> <li>1:30 PM "How to decide your future career" Andrea Lipack</li> <li>2:30 PM "Ellypsometer" Jonathan Sokolov</li> </ul>	7 10:00AM General meeting 10:30 AM "Polymers" Dilip Gersappe 11:15 AM Spinning, Journal Club 1:00 PM Lunch 1:30 PM <i>Biosafety</i> <i>Training</i> Bob Haulthausen 2:30 PM Spinning, Journal Club	8 10:00 AM General meeting 10:30 AM Gary Halada 11:15 AM "Solar Cells" Charles Fortman 12:00 PM "DNA Experiments" Jonathan Sokolov 12:45 PM Lunch 1:15 PM Spinning	<ul> <li>9</li> <li>10:00 AM General meeting</li> <li>10:30 AM</li> <li>"Biomineralization- nature's nanomaterials, studied by synchrotron x-rays" Elaine Dimasi</li> <li>11:15 AM "Managing the electric grid using operational research" Jayant R Kalagnanam</li> <li>12:00 PM Lunch</li> <li>12:30 PM Spinning and Journal Club</li> </ul>	<ul> <li>10</li> <li>10:00AM General meeting "Under your skin- what we can see about you" Isabelle Afriat</li> <li>11:00 AM "Enzymes making polymers" Richard Gross</li> <li>12:00 PM Journal Club</li> <li>1:00 PM Pizza Lunch</li> <li>1:30 PM Spinning</li> </ul>
Week of 7/13	<ul> <li>13</li> <li>10:00 AM General meeting Peter Brink</li> <li>11:00 AM Steve Schwarz</li> <li>11:30 AM "Wound Healing" Richard Clark</li> <li>12:30 PM Lunch</li> </ul>	<ul> <li>14</li> <li>10:00AM General meeting</li> <li>10:30 AM "Polymers" Dilip Gersappe</li> <li>11:30 AM "DNA Experiments" Jonathan Sokolov</li> <li>12:30 PM Lunch</li> </ul>	<ul> <li>15</li> <li>10:00 AM General meeting</li> <li>10:30 AM "Diatoms &amp; Coccoliths in the Ocean: From Biomineralization to Global Warming" Cindy Lee</li> <li>11:30 AM Project Distribution</li> <li>12:30 PM Lunch</li> </ul>	16 ATLANTIS MARINE WORLD AQUARIUM TRIP	<ul> <li>17</li> <li>10:00AM General meeting</li> <li>11:00 AM Mr. Allen</li> <li>Sachs - Contests and</li> <li>required paperwork</li> <li>12:00 Pizza Lunch</li> </ul>
Week of	20	21	22	23	24

7/20	10:00 AM General meeting	10:00AM General meeting	10:00 AM General meeting	10:00 AM General meeting	10:00AM General meeting
	10:15 AM Dr. Tom Butcher, Brookhaven National Lab "Energy Research: Bio fuels, Boilers, and Power Generators"	<ul><li>10: 15 AM Dr. Richard</li><li>Fine, Dean SB School of</li><li>Medicine "Renal</li><li>transplant – historical</li><li>perspective"</li><li>WORK!!!!</li></ul>	11:00 AM Mr. Allen Sachs "Research Ethics" WORK!!!	10:30 AM Dr. Miriam Rafailovich – "Statistics" WORK!!!!	11:15 AM Pizza Speakers: Reena, Matt, Jacki Solar blueberry Juice cells: Joan F and co. Mariah - Ecoflex WORK!!!!
Week	27	28	29	30	31
of 7/27	10:00 AM General meeting	10:00AM General meeting	10:00 AM General Meeting	10:00 AM General meeting	10:00AM General meeting
	WORK!!!!	11:00 AM Dr. Pat Bossert – LISEF/ISEF SRC WORK!!!!	WORK!!!!	WORK!!!!	11:00 AM Student Presentations 12:00 Pizza Lunch WORK!!!!
Week	3	4	5	6	7
of 8/3	10:00 AM General meeting WORK!!!!	10:00AM General meeting CANOE TRIP!	10:00 AM General meeting 11:00 AM Student Presentations WORK!!!!	10:00 AM General Meeting 11:00 AM Dr. Hyum Jun Kim "Nanocomposite Research in Korea" WORK!!!!	10:00AM General meeting 11:00 AM Student Presentations BBQ Lunch WORK!!!!
Week	10	11	12	13	14
of 8/10	10:00 AM General meeting 11:00 AM Student Presentations WORK!!!!	10:00AM General meeting 11:00 AM Student Presentations WORK!!!!	10:00 AM General meeting 11:00 AM Student Presentations WORK!!!!	10:00 AM General meeting 11:00 AM Student Presentations WORK!!!!	10:00 AM – 3:00 PM GARCIA SYMPOSIUM SAC Ballroom A



# ACKNOWLEDGEMENTS

Dr. Srinivas Pentyala

Dr. Bob Holthausen

Ms. Godlind Johnson

Dr. Steven Schwartz

Dr. Elaine Dimasi

Dr. Dilip Gersappe

Mrs. Kim Auletta

Dr. Charles Fortman

- Dr. Jayant R. Kalagnanam
- Dr. Marcia Simon
- Dr. Richard Gross
- Dr. Yizhi Meng
- Dr. Peter Brink
  - Dr. Richard Clark
- Mrs. Andrea Lipack
- Dr. Vladimir Zaitsev
- Dr. Michael Hadjiagyrou

Mrs. Donna Tumminello

- Dr. Cindy Lee
- Dr. Hyum Jun Kim
- Dr. Gary Halada
- Dr. Vladimir Jurokovski

- Dr. Pat Bossert
  - Dr. Richard Fine

Dr. Tom Butcher

- Miss Veronica Collazo
- Miss Lauren Collazo
- Mrs. Bracha Cook

KO John Mehad Free. The Batman Debler Greek C whith Jacobson Sonyaparalt PKOUNA كرى den Elics Cara Jain aveen MM Sanjay Pala Special Thanks to our Sponson Michel Ompli Kopur The Morin Foundation feor Intenmann/ Corporation Matthew 4 ks. Odille Garcia lichael Ti*s*chler Lauren M'Herera r. Herb Wei*m* Aoron Allowan Lent Poop Signal Signal Draft Masih hman Park Saloner Sha 'e / Budassi Kayla