

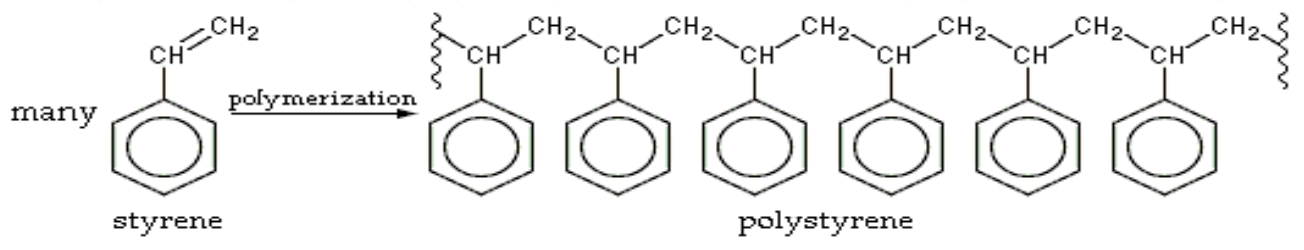


Garcia MRSEC NSF

Polymers at Engineered Interfaces



RESEARCH SCHOLARS PROGRAM 2012





Garcia MRSEC

Polymers at Engineered Interfaces

SUNY Stony Brook • Polytechnic University • CUNY • North Carolina State University

“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’.”



**Jonathan
Sokolov**

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**

Miriam Rafailovich
Professor, Garcia MRSEC

Jonathan Sokolov
Professor, Garcia MRSEC

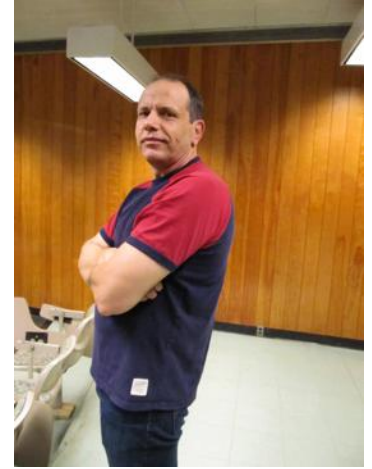
STAFF



DR. JOHN JEROME



DR. CHRISTINE FALABELLA



DR. VLADIMIR JURUKOVSKI

RESEARCH EXPERIENCE FOR TEACHERS



DR. TERRENCE BISSOONDIAL



MRS. ISSEROFF



DR. JOANNE FIGUEIREDO



TOM VAN BELL

FACULTY



DR. MARCIA SIMON



DR. H.Z WANG



CINDY LEE



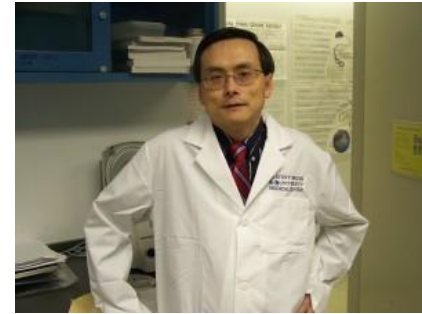
DENNIS GALANAKIS



CHRIS GORDON



PETER BRINK



YUPO MA



DR. YURY YAKUBCHUK

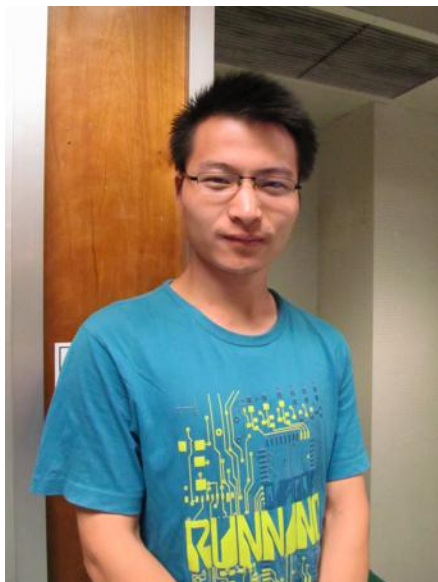


JE BELL AGUILA



STEVEN WALKER

GRADUATE STUDENTS



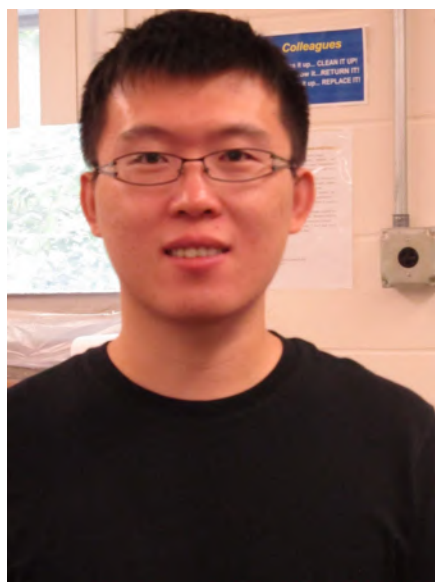
ZHENHUA YING



DIVYA BHATNAGER



SISI QIN



HONGFEI LI



YICHEN GUO



LJUDI ZHANG



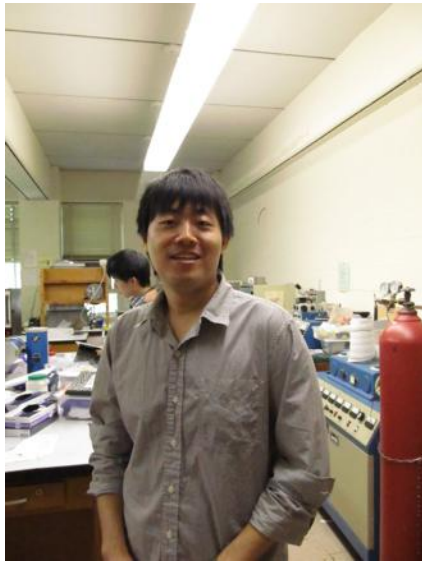
HARRY SHAN HE



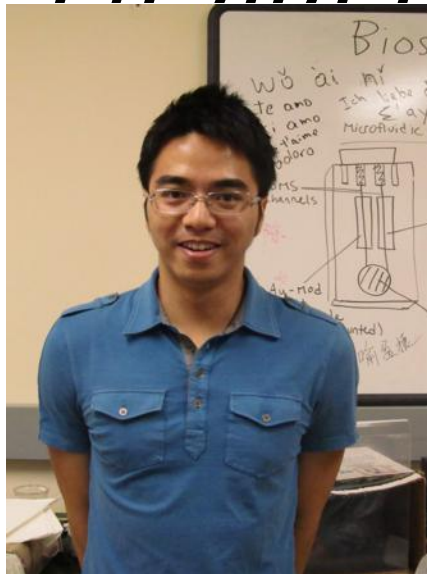
CLEMENT MARMAROT



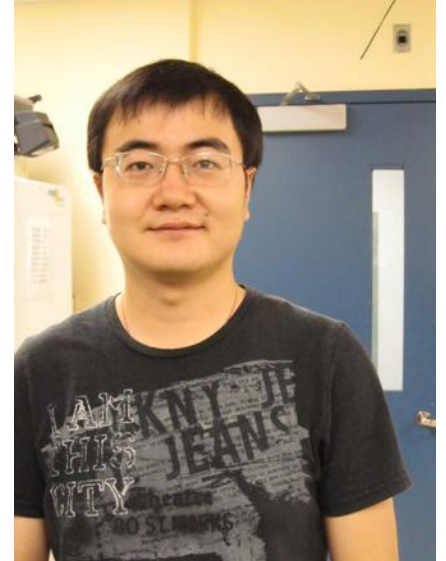
YING LIU



KAI YANG



YINGJIE YU



CHENG PAN



SIMON CHANG



YAN HU

RESEARCH EXPERIENCE FOR UNDERGRADUATES



KAYLA APPLEBAUM



MONIKA BATRA



JULIA BUDASSI

R.E.U Of The Year



DERYA KARATAS



RACHEL DAVIS



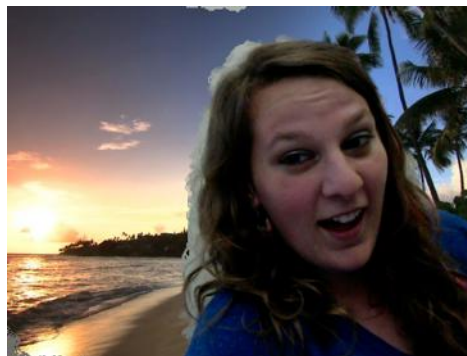
SCOTT DUNAJSKY



HOLLY FLORES



ALANNA FOERTH



MARIAH GERITANO



ILAN GOLD



BENJAMIN GOLDMAN



DANIEL GROSSMAN



JULIA LANDSBERG



DALJA LEIBOWITZ



NIKHIL MEHANDRU



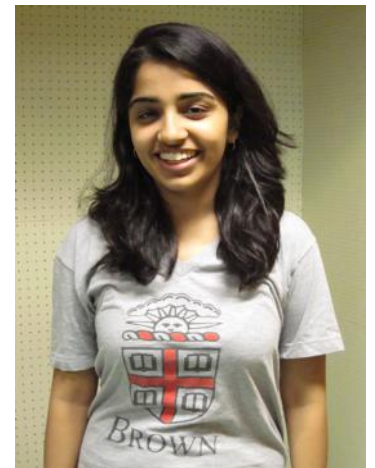
ADAM OSSIP



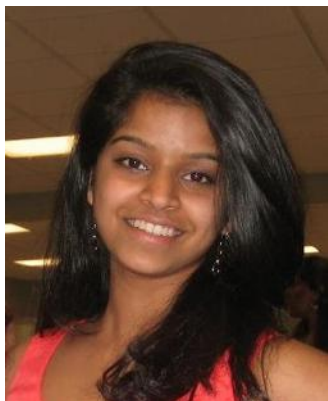
ALINA RANJBARAN



PENINA SAFIER



SANCHITA SINGAL



SNEHA SUBRAMANIAM

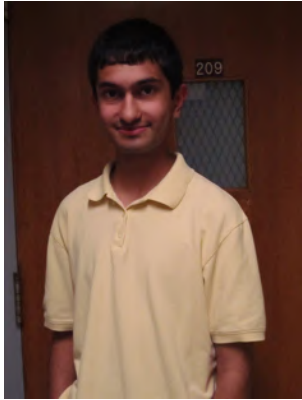


AARON ARHAVAN



JOSE DENIZ

RESEARCH SCHOLARS



AATMAN MAHADIA



ALEXA ASEEL-FINE



ALEXANDER LEE



ALEXANDER NIE



ALEXANDRA TSE



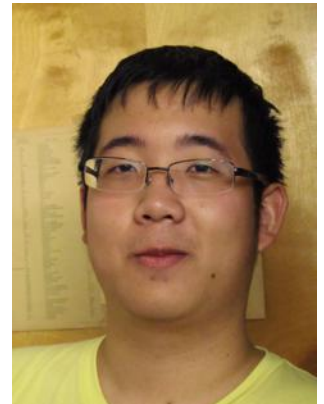
ALLISON LEE



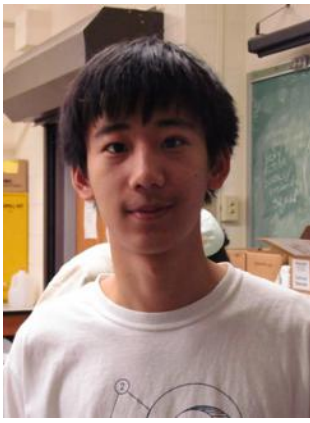
ALYSSA AUERBACH



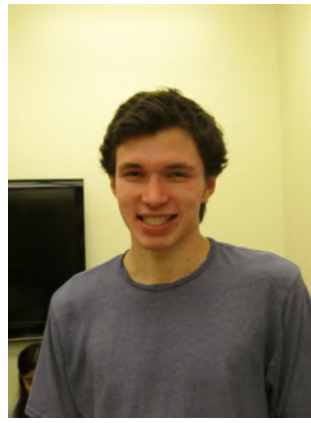
AMY WANG



ANDREW D. CHEN



ANDREW M. CHEN



ANDREW O'NEIL



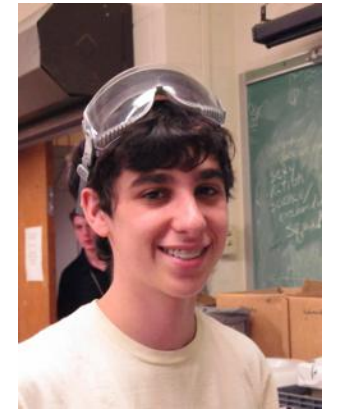
ANERI KINARIWALLA



ARIELLA APPLEBAUM



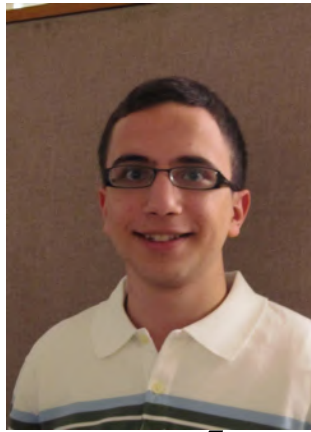
AUSTIN WILD



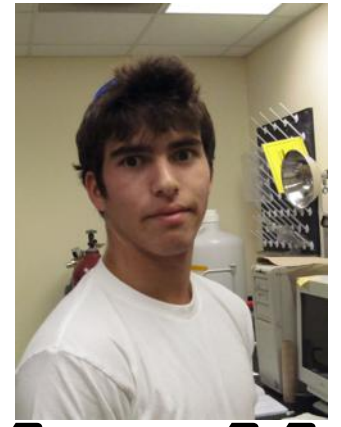
AVERY FEIT



AVIGAEŁ SOSNOWIK



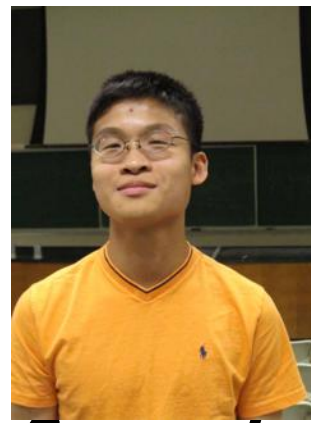
BENJAMIN ARHAVAN



BENJAMIN DUBOW



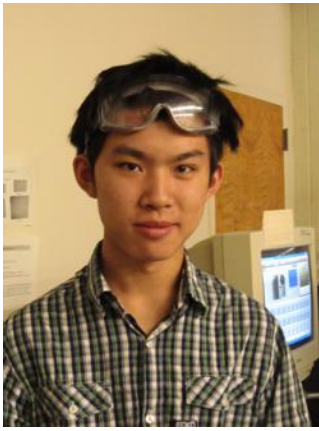
BENJAMIN HOENIG



BENJAMIN LEI



BRADLEY KING



BRIAN WEI



BRIANA FRIEDMAN



CHRISTINE CHANG



DANIELA CZEMERINSKI



DEAN FULGONI



EDA ALGUR



ELIANA APPLEBAUM



EMMA ZAWAKI



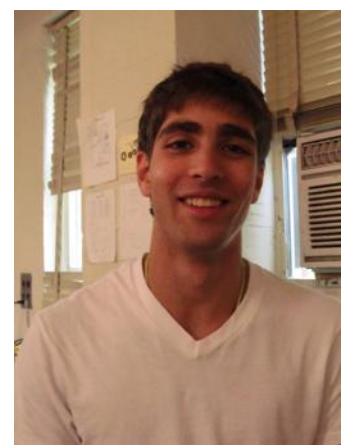
EVAN CHERNACK



GEORGE FEI



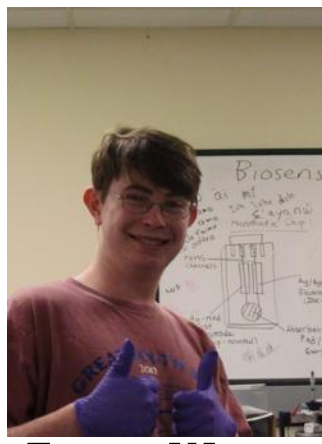
HANNA SILVA



HARIS NAIR



ISSAC ROBSON



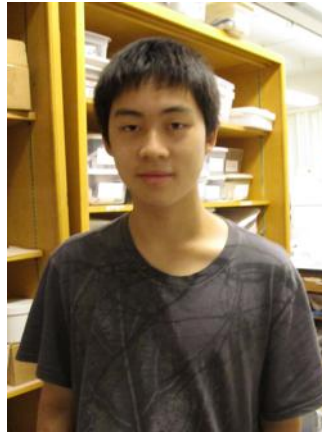
JACOB WAH



JERRY LIU



JOHN MELE



JUSTIN CHIANG



JUSTIN KORITZINSKY



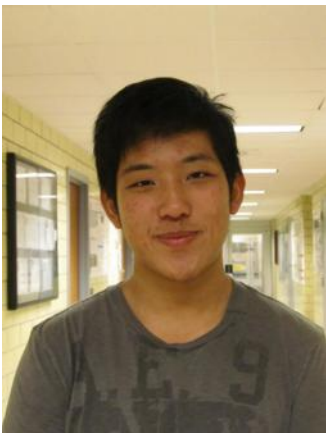
JUSTIN MERKIN



JUSTINE JANG



KAVEH ISSAPOUR



KEVIN CHAN



KEVIN LIU



KIMIA ZIADKHANPOUR



MANASVI VARSHNEY



MATTHEW EMRANI



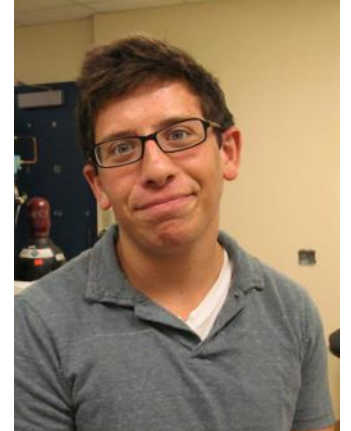
MEGHANA BHAT



MELIH YURSEL



MELISSA CLARK



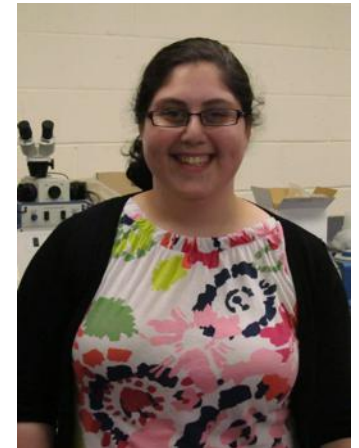
MICHAEL SOSNICK



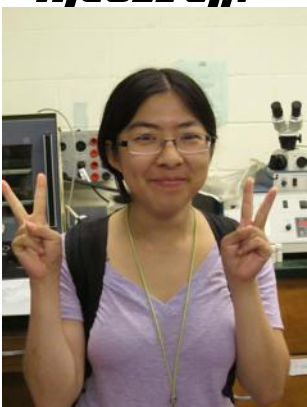
NICOLE LIN



NICOLETTE ALMER



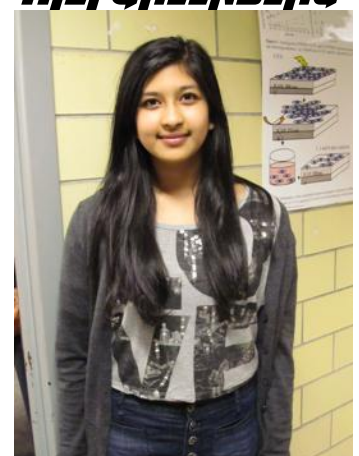
MILJ GREENBERG



PHEOBE WANG



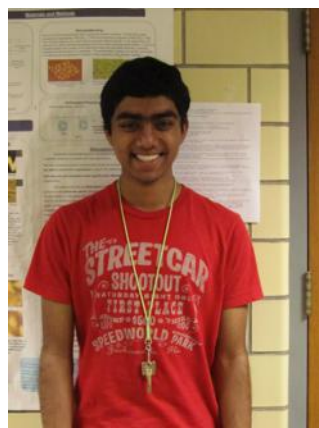
PIERRE MAX ETIENNE



PUJA BANSAL



RACHEL YANG



RAHUL BACHAL



ROBERT ALDANA



ROHIT MEHANDRU



SACHI PATIL



SACHIT SINGAL



SAMANTHA PRASHAD



SHIVRAM CHANDRAMOULI



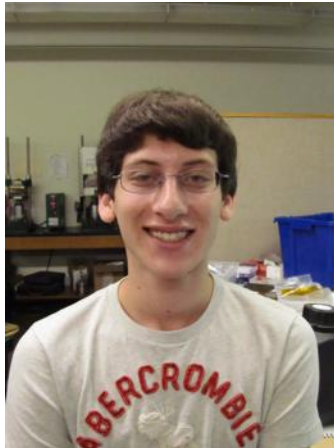
SHOSHANA GUTERMAN



SNEHA CHITTABATHINI



SOHINI UPADHYAY



STEVEN KRIM



TEHILA STONE



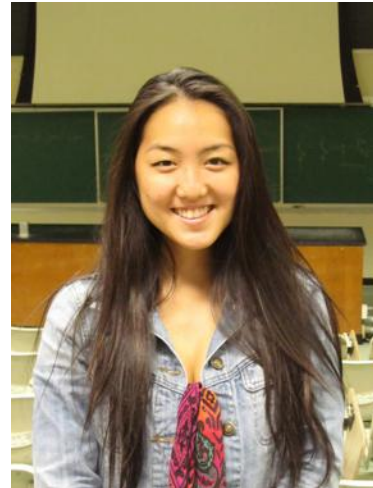
TIMOTHY HART



VICKIE YE

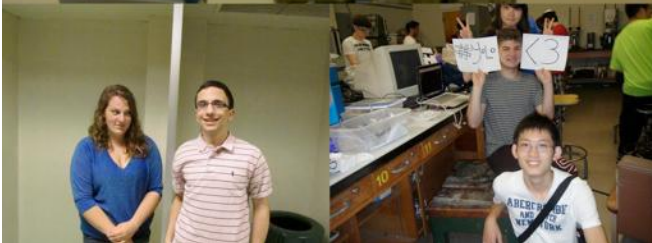


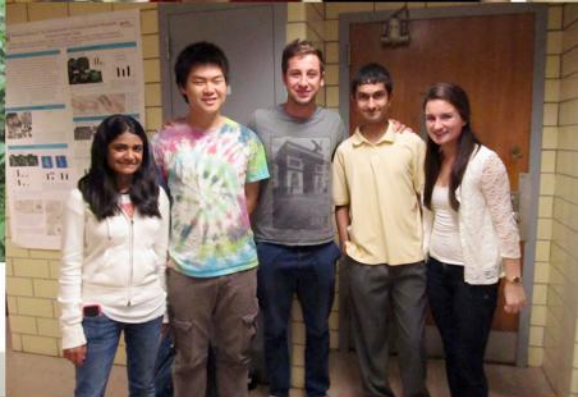
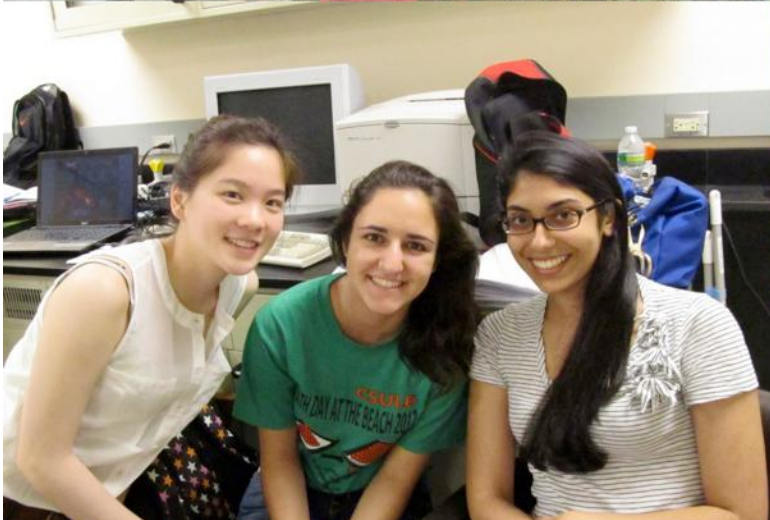
VICTORIA PETROVA



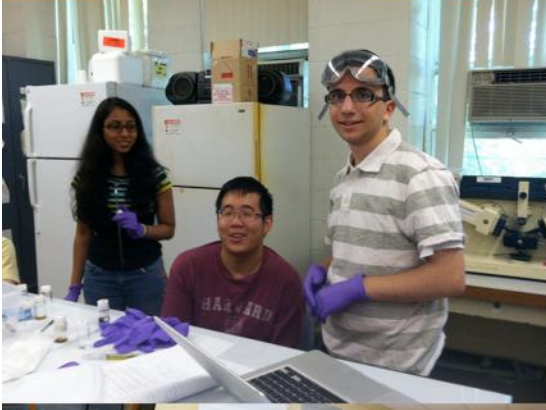
YOU JEONG PARK















SUMMER LECTURE SERIES

June 25, 2012:

"Why Do We Do Research?" - Srinivas Pentyala
"Patents and Intellectual Property" - Donna Tumnello

June 26, 2012:

"Stem Cells and Bioethics" - Brooke Ellison
"LISEF/ISEF Preparation" - Herb Weiss
"Microbiology Studies" - Steve Walker

June 27, 2012:

"Nanocomposites experience at Intel" - Rachel Davis
"Wound Healing" - Marcia Simon
"Rheology and Spin Coating" - Steve Schwarz
"Basic Biology for Engineers" - Vladimir Jurukovski



July 2, 2012:

"Ethical Problems in Medical Care" - Stephen Post
"IPS and Other Types of Stem Cell Research" - Je rell Aguila

July 3, 2012:

"Hydrogen Fuel Cells" - Cheng Pan
"DNA Physics" - Jonathan Sokolov
"Dental Pulp Stem Cell Research" - Vladimir Jurukovski

July 5, 2012:

"How to Keep a Lab Notebook" - Miriam Rafailovich
"Leishmania and TiO₂ Nanoparticles" - Yury Yakubchyk

July 6, 2012:

"TiO₂ and Virology" - Sarah Gross
"Printing with Biodegradable Polymers" - Richard Gross
"Bulk Heterodyne Organic Solar Cells" - Miriam Rafailovich



July 16, 2012:

"Apparent Toxicity of Nanoparticles" - Tatsiana Mironava

July 26, 2012:

Bronx Zoo Trip!

August 3, 2012:

Canoe Trip!

August 7, 2012:

Sopresta Trip





The Garcia Center
Invites you to attend the
Annual Summer Symposium
of the
Research Scholars Program
On

Friday, August 10, 2012

10:00 AM-1:00PM

in the

**Student Activities Center
Ballroom A**

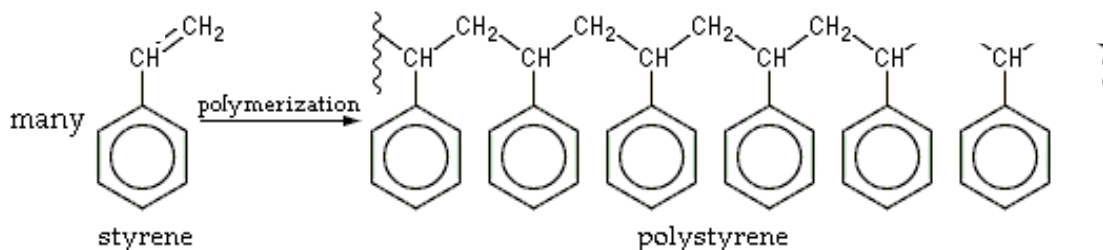
10:00 Coffee, Welcome, Student Musical Arrangements

10:15-12:15 Student Presentations

12:15-1:00 Formal Luncheon arranged by Wing Wan of West Hempstead, NY

Garcia MRSEC

End of *summer* 2012 Symposium



Design by: Sneha Kinariwalla

10: 00 **String Musical Arrangement:** Humoresque, Gymnopedie, No. I, Queen of Sheba, arranged by Matthew Hindson 2004

10: 10 **Welcome** Assemblyman Charles D. Lavine

10:15-10:30 **FLAME RETARDANT NANOCOMPOSITES**

Chairs: Rachel Davis and Dalia Leibowitz, MIT, Cambridge Mass.

A Comparative Analysis of Polystyrene Flame Retardants containing Carbon Nanotubes and Clay Composites;

Robert Aldana, South Side High School, Rockville Centre, NY

Incorporating Flame Retardant Wood Fibers in Polymer Blends

Matthew Emrani and Tehila Stone, The Frisch School, Paramus, NJ

Incorporating Sodium Clay, RDP, And Magnesium Hydroxide Into Nylon 6 To Impart Flame Retardancy

Avery Feit and Justin Merkin, HAFTR High School, Woodmere, N Y

A Study of Biodegradable Polymer Blends and the Effects of Silylation on Their Flame Retardancy and Mechanical Properties

Brad King, Thousand Oaks High School, CA

The Effect of Graphene and Carbon Nanotubes on the Thermal and Electrical Conductivity of Polypropylene

Steven Krim, Lawrence High School, Cedarhurst, NY

Developing Safer Alternatives for Polyvinyl Chloride Using Flame Retardant High-Density Polyethylene Nanocomposites

Brian Wei, Granite Bay High School, Granite Bay, CA

Meghana Bhat, Castilleja School, Palo Alto, CA

10:30-10:40 **MARINE AND ENVIRONMENTAL SCIENCE**

Chair: Dr. Joanne Figueiredo, Smithtown HS, NY

Phytotoxicity of Zinc Oxide: Effects on *Brassica rapa*
Reyna Guzman and Sumaiya Chowdhury, Brentwood HS, N

Influence of Oil on the Aggregation of Diatoms found in the Long Island Sound
Ben Hoenig, South Side High school, Rockville Centre, NY

The effects of Pluronic F98 Prill, a model dispersant, on the formation of marine aggregates
Samuel Lederer, Ashley Crespo, and Kevin Marino, Brentwood High School, NY
Ankita Jain Smithtown West High School, New York

10:40-10:55 **Nanoparticle Cytotoxicity**

**Chairs: Kayla Applebaum Stern College for Women, New York, NY
and Daniel Grossman, Queens College, Flushing, NY**

The Cytotoxic Effects of Titanium Dioxide (TiO₂) & Zinc Oxide (ZnO) Nanoparticles on Human Cervical Adenocarcinoma (HeLa) Cell Membranes **Ariella Applebaum and , Eliana Applebaum**, Ma'ayanot Yeshiva High School, Teaneck, NJ
Shoshana Guterman, Yeshiva University High School for Girls, Holliswood, NY

The cytotoxicity of titanium dioxide nanoparticles and their effect on the infectivity of PRV
Briana Friedman, Nili Greenberg, Yeshiva University High School for Girls, Holliswood, NY
Phoebe Wang, Conestoga High School, Berwyn, PA

Effects of Micelle Coated TiO₂ and ZnO nanoparticles on Targeting Macrophages Infected with *Leishmania tropica* In Vitro
Alexander Lee, Allison Lee Hauppauge High School, Hauppauge, NY

The Effects of Dexamethasone on Dental Pulp Stem Cells Treated with Titanium Dioxide Nanoparticles
Hannah Silva, St. Francis High School, Sacramento, VA
Melissa Clark, Victoria East High School, Victoria, TX

The Effect of Titanium Dioxide Nanoparticles on the Growth and Differentiation of Dental Pulp Stem Cells and Preadipocytes
Nicolette Almer, Kimia Ziadkhanpour, Plainview - Old Bethpage John F. Kennedy High School, Plainview, NY

The Effects of Dexamethasone on the Cytotoxicity of ZnO Nanoparticles in Dental Pulp Stem Cells
Rachel Yang, Commack High School, Commack, NY

10:55-11:05 **Innovative Medical Technologies**

**Chairs: Julia Landsberg Queens College, Flushing NY
Adam Ossip Brandeis University, Waltham, MA**

Using Digital Image Speckle Correlation (DISC) for Analysis of Severe Burn Scarring
Drew O'Neil, Southside High School, NY

Engineering a Multiplexed, Electronic, and Intelligent Drug Delivery Platform for Next-Generation Chemotherapy

Sachit Singal, Herricks High School, New Hyde Park, NY

Rohit Mehandru, Roslyn High School, Roslyn, NY

Engineering A Dynamic Valve System For Hydrocephalus Management

Kaveh Issapour Woodbury, NY

Sohini Upadhyay Port Washington, NY

Analysis of Commercially Available Gutta Percha Materials

Alexa Aseel-Fine Jericho Senior High School, Jericho, NY

11:05-11:20 **Biomolecular and DNA Sensors**

Chairs: Julia Budassi and Jose Deniz Stony Brook University

Controlled Enzymatic Cutting of DNA Using Soft Lithography

Alyssa Auerbach, Yeshiva University High School for Girls, Holliswood, NY

Stretching DNA Molecules On A Flexible Substrate Probed By Polarization-Dependent Fluorescence Microscopy

John Mele, Central Islip Senior High School, Central Islip, NY

Expanding Biosensor Applications Through the Use of Potentiometric Technology

Puja Bansal Half Hollow Hills High School East, NY

Melik Yuksel, Harmony Science Academy Houston TX

Developing Methods of Disease Detection and Wound Healing through Sensing Biomolecules on Surfaces

Daniela Czemerinski, The Wheatley School, NY,

Nicole Lin, El Camino Real Charter High School, CA

Investigating the Sensitivity and Specificity of the Potentiometric Biosensor Mechanism Through Bacteria and Bacterial Spore Cross-Testing

Jacob Wax, Harborfields High School, Greenlawn, NY

11:20-11:30 **Hydrogels**

Chairs: Clement Marmorat, Ecole Polytechnique at Nantes, France and Monika Batra, Stony Brook, NY

Gelatin Hydrogels: The Effect of Physical VS Chemical Hardening on Fibroblast Adhesion and Proliferation

Alex Nie, Livingston High School, NJ

Aatman Makadia, St Anthony's High School, NY

The Effect of Various Concentrations of Glucose and Microbial Transglutaminase on the Mechanical Properties of Cross Linked Gelatin Hydrogels, Biomineralization, and the Growth of Dermal Fibroblasts

Sachi Patil Half Hollow Hills High School East, Melville NY

Emma Zawacki, Smithtown High School East, St James, NY

Using Micropatterned Thin-Films on Silicon Substrates as Carriers for Hydrogel Drug Delivery: A Study of Micropattern Structure on Drug Effusion

Rahul Bachal, Inglemoor High School, 15500 Simonds Rd NE, Kenmore, Washington

Avigael Sosnowik, Stella K. Abraham High School for Girls, 291 Meadowview Avenue, Hewlett Bay Park, New York

11:30-11:50 **Cell Differentiation, Dynamics, and Mechanics**

Chairs: Holly Flores, Stony Brook University, Stony Brook, NY and Alanna Foerth, Messiah College, PA

The Effect of Graphene and Different Concentrations of Iron Oxide on the Proliferation and Differentiation of Dental Pulp Stem Cells

Eda Algur and **Manasvi Varshney**, Smithtown High School West, Smithtown, NY

Analyzing the Role of ROCK/rhoA Kinases in the Differentiation of Dental Pulp Stem Cells

Evan Chernack, South Side High School, Rockville Centre, NY,
Aneri Kinariwalla, Sayville High School, Sayville, NY

Differentiation of dental pulp stem cells on electrospun poly (4-vinylpyridine) and poly (methacrylate)

Justin Koritzinsky Walt Whitman High School (Bethesda, MD)

The Effect of Various Polymers on the Differentiation and Proliferation of Mice Embryonic Stem Cells

You Jeong Park, Half Hollow Hills High School West, Dix Hills, NY;
Kevin Liu, Interlake High School, Bellevue, WA;
Benjamin Lei, Arlington High School, LaGrangeville, NY

The Effects of Polybutadiene, Poly(methyl methacrylate), Sulfonated Polystyrene, and Poly(4-vinylpyridine) on the Proliferation and Differentiation of Hematopoietic Stem Cells

Shivram Chandramouli, Munster High School, Munster, IN

A Study of the Growth and Differentiation of Dental Pulp Stem cells with and without Static Magnetic Fields

Austin Wild, South Side High School, Rockville Centre, NY

The Effect of PMMA Substrates on Keratinocyte Migration

Christine Chang, Palo Alto High School, Palo Alto, CA
Amy Wang, St. Anthony's High School, South Huntington, NY

11:50-12:10 **Hydrogen Fuel Cells**

Chairs: Ilan Gold, University of Md and Aaron Akhavan, Yeshiva University

A Comparative Study on the Structural effects of Noble Metal Nanowires and Nanoparticles as Novel Catalysts for PEM Fuel Cells

Kevin Chan, Stevenson School, Pebble Beach, California
Victoria Petrova, South High School, Torrance, California

Investigating Various Methods of Incorporating Graphene Oxide into PEM Fuel Cell System

Andrew Chen, Dougherty Valley High School, San Ramon, CA
Justin Chiang, Saratoga High School, Saratoga, CA

Using Silver Nanoparticles and Silver/Copper Nanoalloys on the Nafion Membrane Inside of a Hydrogen PEM Fuel Cell to Increase Efficiency

Michael Sosnick, Benjamin DuBow: HAFTR High School, Cedarhurst NY

The Effect of Gold Nanoparticles on a Hydrogen Polymer Electrolyte Membrane Fuel Cell Stack
Timothy Hart, Hauppauge High School, Hauppauge, New York

The Construction of a Microbial Fuel Cell
Featuring *E. coli* Bacteria to Generate Electricity
Haris Nair, Hastings High School Westchester NY;
Samantha Prashad, South Side High School Rockville Centre NY;

Investigating Gold-Palladium Alloy Nanoparticle Enhancement of Proton-Exchange Membrane Fuel Cell Power Output
Vickie Ye, Arnold O. Beckman High School, Irvine, CA

Analysis of Cathodic Waste Gas from a PEM Fuel Cell with Gold Nanoparticle Co-Catalyst
George Fei, George Walton Comprehensive High School, Marietta GA

12:10-12:30 **MATERIALS FOR ENERGY GENERATION**

Chairs: Mariah Geritano Stony Brook University and Sneha Subramaniam, Columbia University

Nanoscale Morphology of Various Polymer Blend Thin Films for Use in Bulk Heterojunction Photovoltaic Cells
Dean Fulgoni, Half Hollow Hills High School West, Dix Hills NY
Pierre Max Etienne, Suffolk Community College, Selden NY

Improving the Nanoscale Morphology of Polymeric Solar Cells Using the LB Trough
Justine Jang, Livingston Senior High School, NJ

Replacement of Aluminum Cathode with Graphene in Organic Polymer Solar Cells via UV/Ozone Exposure and Spin-Coating
Alexandra Tse, Sneha Chittabathini, and Andrew Chen, Lawrence High School, Cedarhurst, NY

Functionalizing Graphene With Nanoparticles
by Blending Nanoparticles Before Reducing Graphene Oxide
Benjamin Akhavan, Rambam Mesivta HS, Lawrence, NY

The Effect of Morphology on Phase Formation, Expansion, and Saturation Time of Silicon Nanowires on Electrodes Using the Lattice Boltzmann Method(LBM)
Jerry Liu, Los Altos High School, Los Altos, CA

Implementing Graphene into a Conductive Polymer Spin Cast
Isaac Robson, Bentonville High School, Bentonville, AR

12:30 Buffet Dinner and Music : Wing Wan of West Hempstead, NY



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Flame Retardant Nanocomposites

Chairs:

Rachel Davis and

Dalia Leibowitz

MIT, Cambridge Mass.

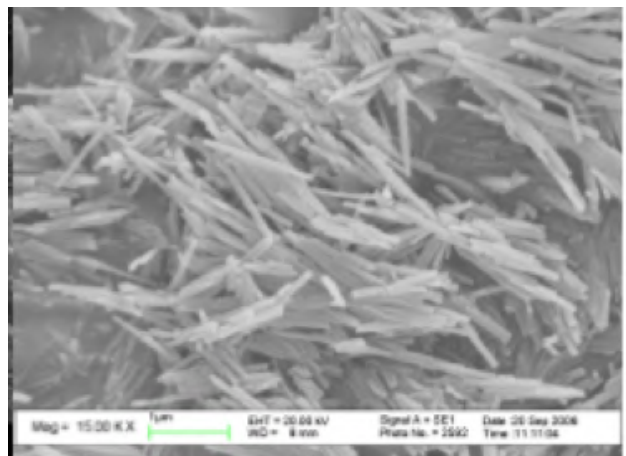
Graduate Students:

Harry Shan He

Kai Yang

Linxi Zhang

Yichen Guo



A Comparative Analysis of Polystyrene Flame Retardants containing Carbon Nanotubes and Clay Composites

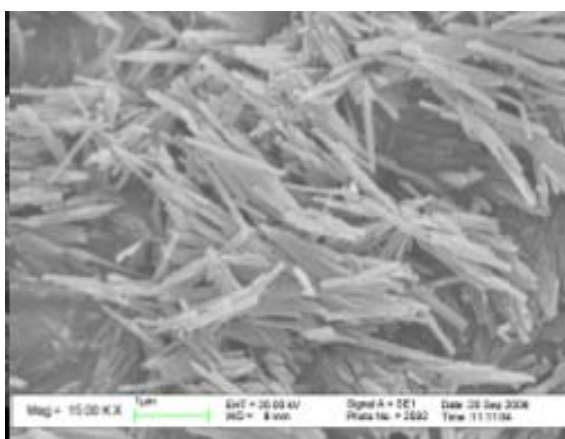
Robert Aldana, South Side High School, Rockville Centre, NY

Rachel Davis, Dalia Leibowitz, Stony Brook University, NY

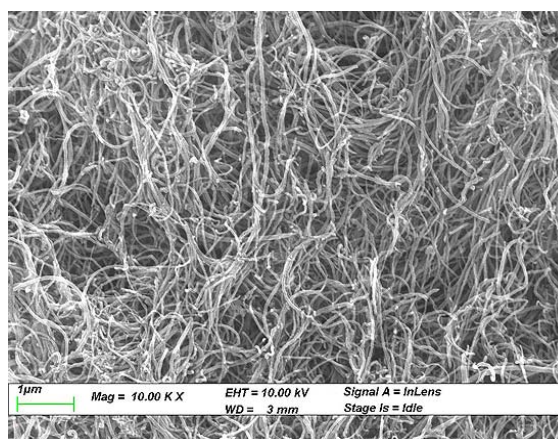
Harry Shan He, Kai Yang, Yichen Guo, Stony Brook University, NY

Miriam Rafailovich, Department of Material Science and Engineering, Stony Brook University, NY

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Every year, residential fires result in approximately 15,000 casualties and over 7 billions dollars lost¹. Often, these fires are exacerbated by structures and accessories composed of flammable plastics. Flame retardant polymers offer a solution to the problem of residential fires without compromising the necessary mechanical properties of plastics. In the past 20 years, Carbon Nanotubes and Halloysites clay nano composites have become increasingly prevalent in the field of material science as flame retardant additives. In 1991, physicist Sumio Iijima discovered Carbon Nanotubes with desirable mechanical, electrical, and thermal properties². Halloysite clay composites were first reported by Berthier as a dioctahedral 1:1 clay mineral of the kaolin group in 1826. In this study, these additives were added to High Impact Polystyrene and were compared because of their similar Nano-tubular structure and large aspect ratios as seen in figures 1a and 1b. These materials were also chosen to compare the efficacy of their different mechanisms for flame retardancy. To increase their flame retardant properties, both the Carbon Nanotubes and the Halloysite composites were soaked in resorcinol bis-diphenyl phosphate (RDP). RDP is an effective additive because of its low volatility and excellent thermal stability. Other additives such as Aluminum Trihydrate (ATH), Brominated Polystyrene (BPS), and Antimony Trioxide (ATO) were used in varying proportions to optimize both flame retardant and mechanical properties. Flame retardancy and mechanical properties were ascertained and studied through several analytical techniques. After reviewing the results from these techniques it was seen that the polymers containing Halloysite are a safer, cheaper, and more practical alternative than those containing Carbon Nanotubes.



1a g g g g g g g



- 1b g
- (1.) "U.S. Fire Administration Fire Estimates." *USFA Statistics: USFA Fire Estimates*. N.p., n.d. Web. 06 Aug. 2012. <<http://www.usfa.fema.gov/statistics/estimates/index.shtm>>.
 - (2.) Moniruzzaman, Mohammad, and Karen I. Winey. "Polymer Nanocomposites Containing Carbon Nanotubes." *Macromolecules* 39.16 (2006): 5194-205.

Incorporating Sodium Clay, RDP, and Magnesium Hydroxide into Nylon 6 to Impart Flame Retardancy

Avery Feit¹, Justing Merkin¹, Rachel Davis², Dalia Leibowitz², Kai Yang³, Harry Shang He³, Yicheng Guo³,
Dr. Miriam Rafailovich⁴, Dr. Sokolov^{4g}

¹HAFTR High School, NY ²Massachusetts Institute of Technology, MA ³Stony Brook University, New York
⁴Stony Brook University Center for Materials Science, NY

The need to develop novel, durable, flame retardant, and efficient polymer coatings of wire is apparent [1]. Although flame retardant coatings suitable for an indoor environment exist, coatings capable of withstanding outdoor elements are not available. Our research endeavored to create such a material to be used as a coating. We determined that Nylon 6, a material with an unusually high melting point compared to other polymers used in the nanocomposite field, contains the mechanical properties necessary to ensure a durable, flame retardant blend. Certain additives, including sodium clay, resorcinol bis(diphenyl phosphate) [RDP], as well as magnesium hydroxide (Mg(OH)₂) were found to further improve and enhance mechanical properties necessary in a flame retardant polymer blend. In order to create polymer blends consisting of the aforementioned additives, a number of machines are necessary to blend and mold the material to allow for the determination of the mechanical properties. Nylon 6, which was observed as a material whose flame extinguishes within 1 second, drips considerably during the UL-94 test. This prompts a 2 result. Had dripping been absent, the result would have been V-0 (an optimal result). Sodium clay, a material known to improve a blend's mechanical properties [2] (including the prevention of dripping), as well as RDP (40% RDP in clay), and magnesium hydroxide, which improve flame retardancy, were blended in different ratios to determine the optimal blend. Currently, none of our blends have yielded successful results in the UL-94 flame test. Despite this, the additives did improve tensile strength of Nylon 6. Each blend exhibited an increased young's modulus (Figure 1) and impact toughness relative to pure Nylon 6. In the future, we will replace sodium clay (with RDP) with Halloysite Clay (with RDP), which may further improve the material's mechanical properties.

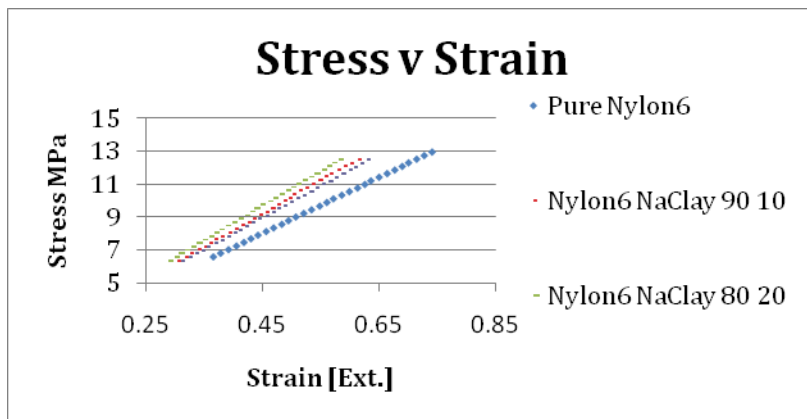


Figure 1: The graph to the left displays the stress vs. strain of pure nylon 6 compared with the nylon blends. The greater the slope of the strength vs strain line the greater the Young's Modulus, which indicates the stiffness of the substance g

- Lewin, Menachem. "Unsolved problems and unanswered questions in flame retardance of polymers" *Polymer Degradation and Stability*. 88. (2005): 13-19.
- Yang, Jiu-Jun, Zhang, Lei, Guo, Pu, and Xue-Ping Wang. "Investigation on mechanical properties of clay-based material prepared from the Yellow River silt." *Applied Mechanics and Materials*. 52.154g (2012): 24-29.

A Study of Biodegradable Polymer Blends and the Effects of Silylation on Their Flame Retardancy and Mechanical Properties

Brad King¹, Kai Yang², Harry Shan He², Yichen Guo², Rachel Davis³, Dalia Leibowitz³, Miriam Rafailovich⁴, Jonathan Sokolov⁴

¹Thousand Oaks High School, CA ²Stony Brook University, NY ³Massachusetts Institute of Technology ⁴Department of Materials Science and Engineering, Stony Brook University, NY

The search for and development of biodegradable and biocompatible plastic products is on. Environmental sustainability is more important than ever as worldly crises such as global warming escalate. The cost, mechanical properties, and flame retardancy of a polymer blend are crucial factors to consider, as are the levels of toxicity and harm to the environment and its organisms. A largely unexplored area of flame retardant engineering is the development of biodegradable thermoplastics. Polylactic acid (PLA) and poly(butylene adipate-co-terephthalate) (PBAT) are two such plastics. PLA is a brittle polymer made from corn starch, while PBAT is tear-resistant and extremely elastic¹.

To create a flame retardant blend with the optimal mechanical properties, PBAT and PLA were blended in a three-to-two ratio to serve as a control sample, and the effect of particular additives was observed. Cellulose, chitin from crab shells, halloysite clay, and silylated cellulose were added to the combination of the two thermoplastics. Resorcinol bis-diphenylphosphate (RDP) was used as a flame retardant, as it is a promising replacement for toxic halogenated flame retardants that are commonly used in plastic manufacturing today. Silylated cellulose was created using triethoxy (3-isocyanatopropyl) silane (ICPTEOS) and tetraethoxy silane (TEOS), forming a repeating chain of silicon and oxygen on the surface of each fiber. This modification resulted in greater thermal stability and increased hydrophobicity, which lead to a higher level of compatibilization with PLA (which is hydrophobic as well).

Under the strict requirements of the American Standards for Testing Materials (ASTM), the blend containing PLA/PBAT/40% RDP Cellulose in both a 40:60:5 ratio and 40:60:10 ratio achieved a V2 rating, as did PLA/PBAT/40% RDP Chitin in a 40:60:5 ratio². Because dripping was excessive in each of these blends, 40% RDP soaked halloysite clay was added to each of them in an attempt to induce charring and reduce the flow of the polymer when exposed to a flame by allowing the clay particles to intercalate along the surface.

As additives were compounded into the polymer blend, the material became stiffer, and thus the Young's Moduli of the samples increased (See Fig. 1). This value is the ratio of stress versus strain, and is indicative of the elasticity of the substance.

Future work includes observing the effects of silylation on the mechanical and flame retardant properties of the control blend, performing a greater variety of tests on the samples, and blending different concentrations of additives, PLA, and PBAT.

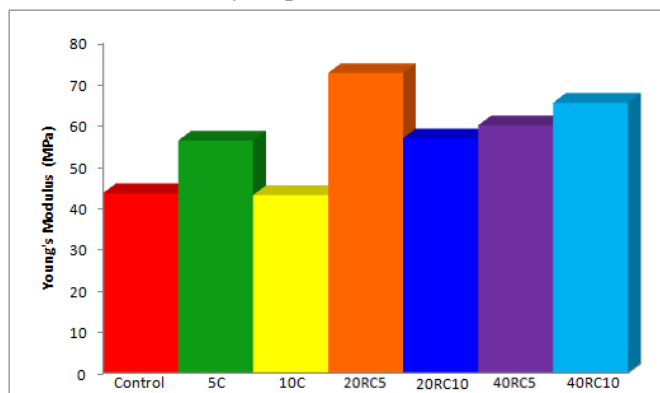


Fig. 1 Elastic moduli for polymer blends (5C = 5% cellulose blended with 40% PLA and 60% PBAT; 20RC5 = 20% RDP cellulose blended with 40% PLA and 60% PBAT; 40RC5 = 40% RDP cellulose blending with 40% PLA and 60% PBAT)

¹Jiang, Long, Michael Wolcott, et al. "Study of Biodegradable Polylactide/Poly(butylene adipate-co- terephthalate) Blends." *Biomacromolecules*. 7.1 (2006): 199-207.

²Dvir, Haim, Moshe Gottlieb, et al. "Optimization of a flame-retarded polypropylene composite." *Composites Science and Technology*. 63. (2003): 1865-1875.

The Effect of Graphene and Carbon Nanotubes on the Thermal and Electrical Conductivity of Polypropylene

Steven Krim, Lawrence High School, Cedarhurst, NY

Rachel Davis, Massachusetts Institute of Technology, Cambridge, MA

Dalia Lebowitz, Massachusetts Institute of Technology, Cambridge, MA

Kai Yang, Stony Brook University, NY

Harry Shan He, Stony Brook University, NY

Lindsay Zhang, Stony Brook University, NY

Yichen Guo, Stony Brook University, NY

Dr. Miriam Rafailovich, Department of Materials Science & Engineering, Stony Brook University, NY

The flame retardancy of polymers has been a growing concern in society in recent years.¹ One polymer that is extremely flammable is polypropylene. Polypropylene is used for automotive components, textiles, and packaging due to its versatile properties. Therefore, it is necessary to increase the flame retardancy of the polymer so that it can meet current standards for flame retardancy.

In this study, graphene was tested as a flame retardant additive to Polypropylene. Polypropylene mixed with differing additives will achieve different ratings according to the UL-94 flame test. When 1% by weight of graphene was blended with polypropylene, the sample was unable to meet the minimum flame retardant requirements of The Underwriters Laboratories Inc, showing that graphene alone, as an additive is unable to make a polymer flame retardant. Next, the graphene was soaked in bis-diphenylphosphate (RDP) and then blended with polypropylene. The RDP soaked graphene blend was unable to pass the flame test. After these tests, Aluminum hydroxide (ATH) and Magnesium Hydroxide (MgOH) were added to the polymer blend due to their flame retardant effect. ATH and MgOH act as fire retardants by releasing water vapor through endothermic decomposition leaving a thermally stable inorganic residue. When used as fillers in polymer composites, they dilute the combustible polymer by decomposition, by becoming water, which cools the condensed phase through endothermic dehydration.² Graphene and carbon nanotubes (CNT) were added to the polypropylene blend due to their thermo and electrical conductive properties.³ When ATH, MgOH, CNT, and graphene were added to polypropylene, a V-0 rating was achieved, but as in (figure 1), the mechanical properties that the young's modulus increased, indicated increased brittleness, and the impact toughness decreased significantly, indicating a decrease in ductility.

Future work includes trying differing concentrations of the additives: ATH, MgOH, Graphene, and CNT to try to preserve the mechanical properties of polypropylene while still having a V-0 rating.

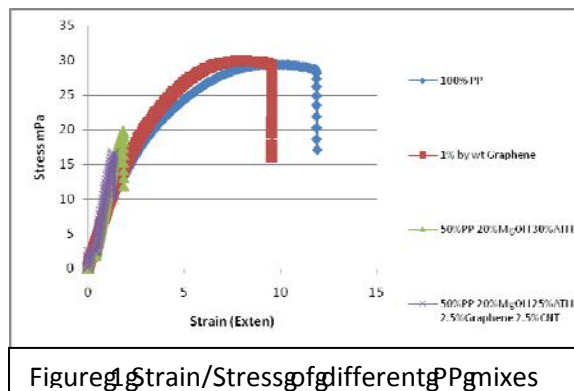


Figure 1. Strain/Stress of different PPMixes

¹ Sheng Zhang, A. Richard Horrocks, A review of flame retardant polypropylene fibres, Progress in Polymer Science, Volume 28, Issue 11, November 2003, Pages 1517-1538, ISSN 0079-6700, 10.1016/j.progpolymsci.2003.09.001.

² L.A. Hollingbery, T.R. Hull, The fire retardant behaviour of huntite and hydromagnesite – A review, Polymer Degradation and Stability, Volume 95, Issue 12, December 2010, Pages 2213-2225, ISSN 0141-3910, 10.1016/j.polyimdeggradstab.2010.08.019.

³ Bernardo Marinho, Marcos Ghislandi, Evgeniy Tkalya, Cor E. Koning, Gijsbertus de With, Electrical conductivity of compacts of graphene, multi-wall carbon nanotubes, carbon black, and graphite powder, Powder Technology, Volume 221, May 2012, Pages 351-358, ISSN 0032-5910, 10.1016/j.powtec.2012.01.024.

Developing Safer Alternatives for Polyvinyl Chloride Using Flame Retardant High-Density Polyethylene Nanocomposites

Brian Wei, Granite Bay High School, Granite Bay, CA, **Meghana Bhat**, Castilleja School, Palo Alto, CA
Rachel Davis and Dalia Leibowitz, Massachusetts Institute of Technology, Cambridge, MA
Harry Shan He, Kai Yang, and Chen Yui, Stony Brook University, NY
Miriam Rafailovich, Department of Materials Science & Engineering, Stony Brook University, NY

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Polyvinyl chloride (PVC) is the third most widespread plastic in the world today, behind only Polypropylene (PP) and High-Density Polyethylene (HDPE).^[1] It is favored for its low cost, versatility, durability, and flame retardant properties.^[2] However, recent studies have raised concerns over the potential health and safety issues regarding the halogenated nature of PVC, the carcinogenic Vinyl Chloride Monomer, and the additives commonly involved in the manufacture and processing of PVC, namely phthalates, aldehydes, dioxins, and heavy metals, as well as the adverse effects that PVC production has on the environment.^[3] Thus, the development of less harmful alternatives to PVC has been of great interest.

HDPE was chosen as an optimal base material for the development of PVC alternative nanocomposites for its relative environmental friendliness, lack of health controversy, and availability.^[4] However, HDPE is highly flammable, which makes it less desirable in many applications. To improve its properties, HDPE was blended into nanocomposites with various concentrations of Ecoflex, Polycarbonate, and resorcinol bis(diphenylphosphate) (RDP) and sodium cloisite clay. These composites were tested for mechanical and chemical properties using the UL-94 flame test, Fourier Transform Infrared Spectrometry (FT-IR) Attenuated Total Reflection (ATR), and Instron tensile test. The goal was a composite that was flame retardant and had comparable mechanical properties to PVC.

None of the composites created achieved a UL-94 rating. However, flame retardancy did improve with the additives to HDPE. A larger portion of the control sample of pure HDPE burned away in constant flaming drops, igniting the cotton. 40% RDP clay did reduce the amount of dripping. At a concentration of 1.0% and 7.0% HDPE 0% PC blend, it stopped the ignition of the cotton in the flame test along with significant reduction of the burning time. After 20% Aluminum trihydride (ATH) was blended into the composite of 0% HDPE and 1.0% Ecoflex with 1.0% of 40% RDP clay in

order to catalyze the reaction that RDP undergoes to increase the sample's flame retardancy, the fire hardly spread from the tip of the composite, and the sample hardly dripped.

Adding Ecoflex, RDP clay, and ATH each did decrease the yield strength and Young's modulus--they could withstand less of a stress and could be deformed by a weaker stress. All the composites snapped cleanly in contrast to the stretchy nature of HDPE. The 0% HDPE 0% PC blend showed similar changes in mechanical properties. On the other hand, an increase in the proportion of PC to create 0% HDPE 7.0% PC blend resulted in higher Young's modulus. Its low impact toughness, in this case, showed the increase in hardness of the composite as compared to that of pure HDPE (Fig. 1). Future research would involve using magnesium

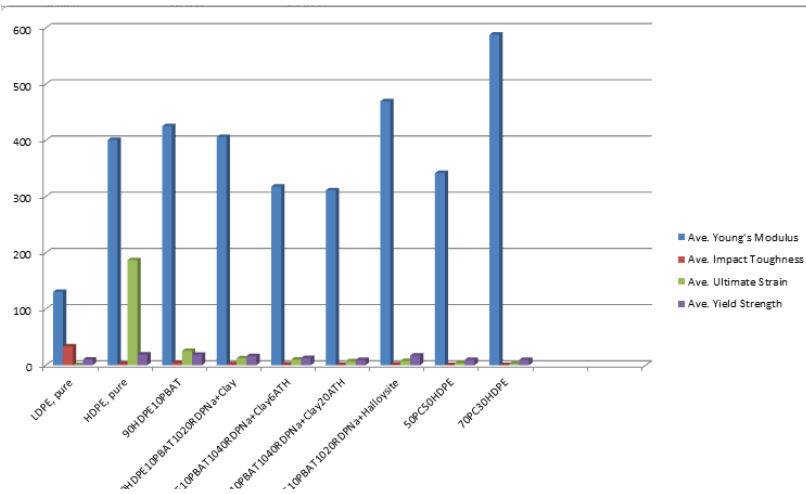


Figure 1: Comparison of Mechanical Properties

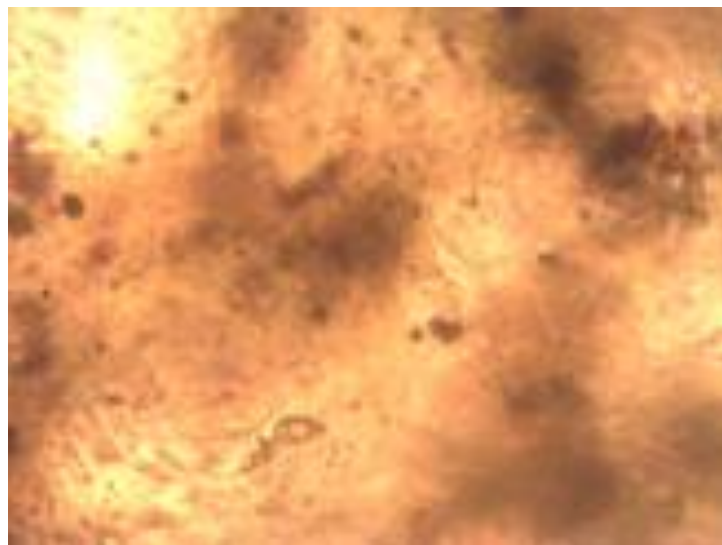
hydroxide powder ($Mg(OH)_2$) as a catalyst instead of ATH, which decomposes at temperatures above 180 degrees Celsius (the HDPE-Ecoflex blends are molded at approximately 204 degrees Celsius). Blending just HDPE with the RDP clay and $Mg(OH)_2$ would also aid in examining whether Ecoflex helps the flame retardancy of the sample and how it affects the mechanical properties of the nanocomposite. TEM analysis was conducted on the samples in order to analyze the effect of the structure of the composites on their properties. Alternative polymers can also be blended with the HDPE to see their effects on flame retardancy and mechanical properties.

1. "Resin Statistical Annual Summary." *American Chemistry Council*. April 2012. Web. 23 July 2012.
 2. Wilkes, Charles F.; Summers, James W.; Daniels, Charles Anthony; Berard, Mark G. (2005). *PVC Handbook*. Hanser Verlag. p. 14, 68.
 3. Altschuler, Kara; Scot Horst; Nadav Malin; Gregg Norris; and Yurika Norioka. *Assessment of the Technical Basis for a PVC-Related Materials Credit for LEED*. Feb. 2007. Task Group Final Report.
 4. "High Density Polyethylene (HDPE)." *Environment Product Declarations of the European Plastics Manufacturers*. Plastics Europe, Nov. 2008. g

Marine And Environmental Science

Chair:

Dr. Joanne Figueiredo,
Smithtown H.S, NY



Phytotoxicity of Zinc Oxide: Effects on *Brassica rapa*

Reyna Guzman, Brentwood HS, NY - Sumaiya Chowdhury, Brentwood HS, NY
Dr. Joanne Figueiredo, Smithtown HS, NY - Dr. Terrence Bissoondial, Hewlett HS, NY

The increased presence of nanoparticles (NP) in a wide range of consumer products raises key concerns about their impacts on the environment and human health. Numerous studies have shown that NPs can be taken up by plants and may bioaccumulate in a variety of tissues (Rico et. al., 2011)¹. Several studies have shown that nanoparticles activate the Reactive Oxygen Species (ROS) pathways inside cells (Langebartels et. al., 2002)². We decided to test the ability of the pigment anthocyanin to protect plant tissue against the negative effects of NPs.

Our work focused on the effect of the nanoparticle Zinc Oxide (ZnO) on *Brassica rapa* growth and development. We studied wild type and a strain of *B. rapa* that overproduces the pigment anthocyanin. Seeds were placed in petri dishes and exposed to 0, 0.1, 0.5 and 1.0 g/L of ZnO and allowed to grow for 7 days in either the light (12 hours light/12 hours dark) or complete darkness. For plants that were grown in the light, we found that for both wild type and high anthocyanin plants, hypocotyl length of control was twice that of those that were exposed to the highest concentration of ZnO (Figure 1.) A similar effect was seen when the plants were grown in the dark with the controls having hypocotyl lengths that were twice that of plants exposed to 1.0 g/L of ZnO. We also compared the amount of anthocyanin present in control plants to those grown in the presence of 1.0 g/L ZnO by extracting the pigment and reading the light absorbance at 520 nm on a spectrophotometer. As shown in Figure 2, Wild type plants that were grown in the light had significantly more anthocyanin than those grown in the dark. When these plants were exposed to 1.0 g/L ZnO, the anthocyanin levels decreased. For the mutant strain, we found that plants grown in the light have more anthocyanin than those grown in the dark. When exposed to 1.0 g/L ZnO anthocyanin levels increased. To demonstrate that this effect was specific to anthocyanin, we also measured plant chlorophyll level with a flourometer and found that ZnO exposure did not alter the production of this pigment.

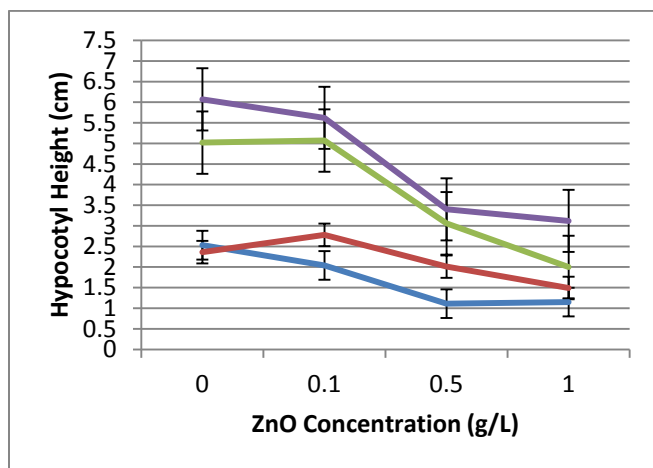


Figure 1: Hypocotyl height in different Zinc Oxide concentrations. Purple: High anthocyanin plants grown in dark; Green: Wild type plants grown in dark; Red: High anthocyanin plants grown in light; Blue: Wild Type plants grown in light.

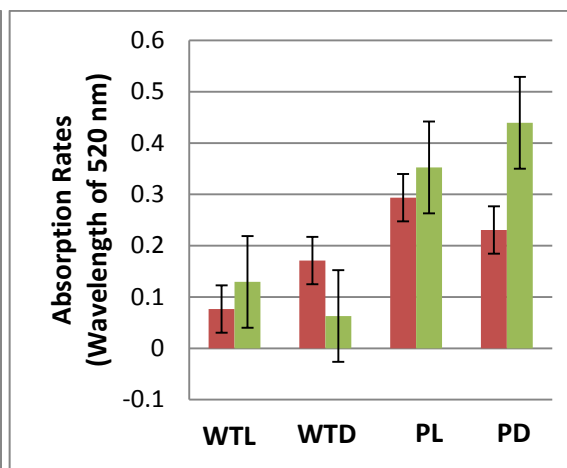


Figure 2: Light Absorption of anthocyanin in plants. Red: Control; Green: 1 g/L; WTD: Wild type grown in dark; WTL: Wild Type grown in Light; PL: High Anthocyanin grown in light; PD: Low Anthocyanin grown in dark.

¹Rico, C. M., Majumdar, S., Duarte-Gardea, M., Peralta-Videa, J. R., & Gardea-Torresdey, J. L. (2011). Interaction of nanoparticles with edible plants and their possible implications in the food chain. *Journal of Agricultural and Food Chemistry*, (59), 3485-3498.

²Langebartels, C., Wohlgenuth, H., Kschieschan, S., Grun, S., & Sandermann, H. (2002). Oxidative burst and cell death in ozone-exposed plants. *Plant Physiology and Biochemistry*, (59), 567-575.

Influence of Oil on the Aggregation of Diatoms found in the Long Island Sound

Ben Hoenig, Southside High School, Rockville Centre, NY

Cindy Lee, Department of Marine Sciences, Stony Brook University, NY

Carolina Cisterna-Novoa, Department of Marine Sciences, Stony Brook University, NY

Joanne Figueiredo, Smithtown High School, NY

Miriam Rafailovich, Material Science and Engineering, Stony Brook University, NY

Diatoms are some of the most important creatures in the world. They are responsible for aggregating into particles, which gradually increase in size through secretion of marine glue and other material, and sink. As these particles sink, they take all the carbon they used for life processes down into the ocean sediment, where it is permanently sequestered from the atmosphere. These marine aggregates are considered the most important element by which carbon is sequestered into the deep ocean. If factors outside elements were to impede diatoms in their aggregation or sinking mechanism, it could have harmful effects on the surrounding environment.¹

In the modern era, the risk of an oil spill into marine ecosystems is very real, as demonstrated by recent events. To observe the effects of oil on the aggregation of marine particles, a study was conducted by culturing diatoms found in the Long Island Sound and placing them in three different roller tanks with varying oil concentrations. The roller tanks had a volume of 1.185 L and encouraged the aggregation of particles, as they would be found in nature.

After aggregates formed and were measured for settling rates, they were collected, dried, and observed using optical microscopy, as well as scanning electron microscopy (SEM). This initial study of aggregates was carried out to verify the diatoms present in the samples were those commonly found in Long Island.² After this initial analysis of the aggregates, two more trials were carried out in order to observe aggregates while they were still wet, and stained with Alcian Blue to study the possible negative effects of oil on the marine glue (EP), which encourages diatom aggregation. It is seen that the particles from the oil tanks are less dense and have lighter coloration than the particles from the control tank. (Figure 1) The aggregates in oil also displayed lower settling rates than those not in oil.



Figure 1. Aggregates of diatoms formed in roller tanks without oil (left) and with *Leptocylindrus danicus* oil (right)

1. Alonso-Gonzalez, Ivan. "Role of Slowly Settling Particles in the Ocean Carbon Cycle." *Geophysical Research Letters* 37. No. 13 (2010)
2. McCarthy, Gina. "Monitoring Phytoplankton Community Composition in Long Island Sound With HPLC Photopigment Profiles"

The effects of Pluronic F98 Prill, a model dispersant, on the formation of marine aggregates

Samuel Lederer¹, Ashley Crespo¹, Kevin Marino¹, Ankita Jain²

Dr. Joanne Figueiredo², Dr. Miriam Rafailovich³

^[1] Brentwood High School, New York ^[2] Smithtown West High School, New York ^[3] MRSEC
Stony Brook University

In the marine ecosystem phytoplankton and diatoms excrete transparent exopolymer particles (TEP) to solidify masses of inorganic and organic conglomerates called marine aggregates¹. Aggregate formation is vital to the marine carbon pump and the structuring of food webs. TEP's role in these processes as a binder is related to the stickiness of its polysaccharide chemical make-up. Oil dispersants have been used in recent oil spills, to decrease the surface tension between crude oil and water. This disperses the oil within the water by creating micelles, thereby lessening the immediate effect of oil toxicity on marine life. The main chemical component of dispersants is a surface-active agent, also known as a surfactant. Our work investigates the effect of the surfactant, Pluronic F98 Prill, on the formation and dynamics of marine aggregates.

To study these processes, we collected sea water from Stony Brook Harbor and grew marine cultures for 5 days. A controlled mesocosm experiment was then conducted in order to compare the aggregates formed under control conditions to those formed in the presence of 0.15% Pluronic. Our results showed that there was a decrease in the number of aggregates formed in control mesocosms (20.6 ± 4.7 aggregates) versus those containing 0.15% Pluronic (14.8 ± 3.4 aggregates.) Furthermore, there was a significant difference ($p=0.0006$) in the rate of sedimentation of aggregates in control seawater (0.652 cm/s) compared to the sedimentation rate of aggregates in 0.15% pluronic (0.398 cm/s.) Microscopic analysis of Alcian blue stained aggregates revealed that there was an increase in free TEP (Figure 1) in the presence of Pluronic when compared to controls. These results suggest that Pluronic may decrease the ability of TEP to form marine aggregates.

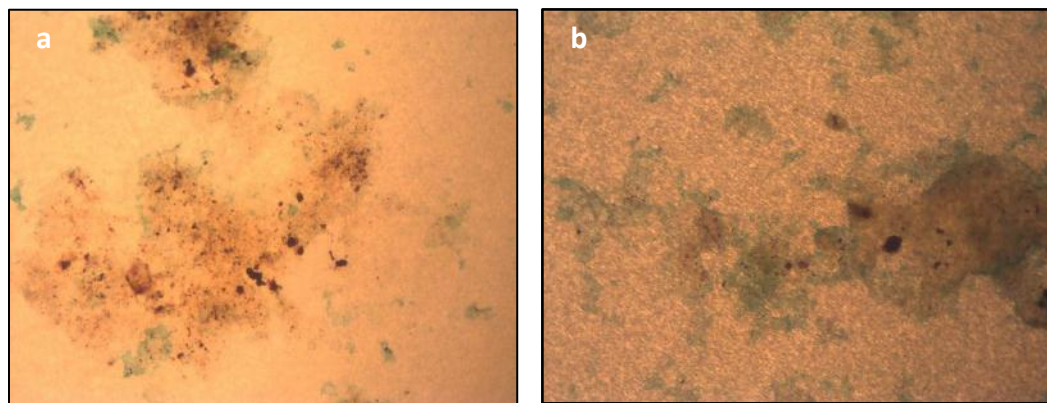


Figure 1: 100× magnification of marine aggregates stained with alcian blue under an optical light microscope. (a) Control, 0% Pluronic F98 Prill. (b) .15% Pluronic F98 Prill.

^[1] Alice L. Alldredge, Mary W. Silver, Characteristics, dynamics and significance of marine snow, Progress In Oceanography, Volume 20, Issue 1, 1988, Pages 41-82

^[2] U. Passow, Transparent exopolymer particles (TEP) in aquatic environments, Progress In Oceanography, Volume 55, Issues 3–4, November–December 2002, Pages 287-333

Nanoparticle Cytotoxicity

Chairs:

Kayla Applebaum, Stern
College for Women, New York

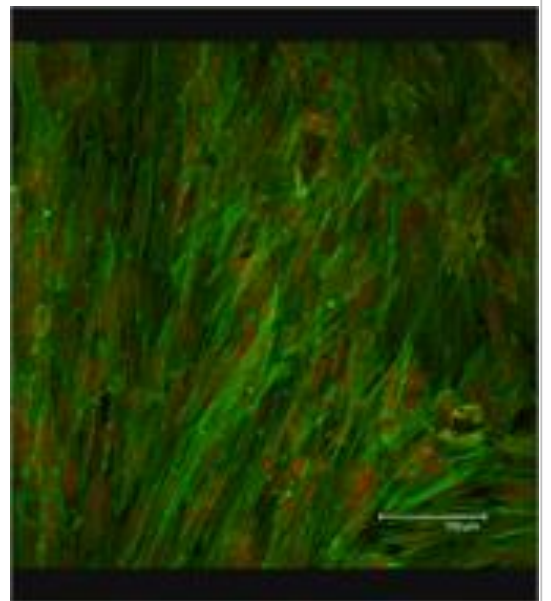
Daniel Grossman, Queens
College, New York

Graduate Students:

Simon Chang

Yan Xu

Ying Liu



The Cytotoxic Effects of Titanium Dioxide (TiO₂) & Zinc Oxide (ZnO) Nanoparticles on Human Cervical Adenocarcinoma (HeLa) Cell Membranes

Ariella Applebaum¹, Eliana Applebaum¹, Shoshana Guterman², Kayla Applebaum³, Daniel Grossman⁴, Chris Gordon⁵, Peter Brink⁵, H.Z. Wang⁵, & Miriam Rafailovich⁵
 Ma'ayanot Yeshiva High School, Teaneck, NJ¹; Yeshiva University High School for Girls, Holliswood, NY²; Yeshiva University, New York, NY³; Queens College CUNY, Flushing, NY⁴; SUNY Stony Brook, Stony Brook, NY⁵

Ultrafine titanium dioxide (TiO₂) and zinc oxide (ZnO) are widely used as active ingredients in cosmetics, sunscreens, lotions, toothpastes and other household items. Despite descriptions of TiO₂ and ZnO as safe commercial ingredients, TiO₂ and ZnO nanoparticles have been shown to generate Reactive Oxygen Species (ROS) known to be cytotoxic and possibly genotoxic leading to cell apoptosis.[1] While the literature to date has described cytotoxic effects of TiO₂ and ZnO on many cell types, few studies have explored the direct effects of these nanoparticles on HeLa cell membrane permeability.[1] This study demonstrates the adverse effect of both rutile and anatase forms of TiO₂ as well as ZnO on human cervical adenocarcinoma (HeLa) cell morphology and proliferation. Additionally, our study shows that HeLa cells exposed to these nanoparticles have increased cell membrane permeability with stable cell membrane resistance. To further elucidate whether TiO₂ and ZnO effect the HeLa cell membrane by close proximity as appose to entering the cell, this study shows that dexamethasone (DXM), a synthetic glucocorticoid that inhibits cell apoptosis, seems to alter the membrane potential of HeLa cells likely preventing TiO₂ and ZnO from entering HeLa cells and results in decreased HeLa cell apoptosis.[2] Initially, HeLa cell controls and HeLa cells incubated with TiO₂ and ZnO were plated for examination of cell morphology by confocal microscopy and cell counting by hemocytometry to determine cell proliferation. Control HeLa cells and HeLa cells incubated with rutile and anatase TiO₂ (0.05 mg/ml & 0.1mg/ml), and ZnO (0.01 mg/ml) were plated. However, these nanoparticle concentrations resulted in high rates of cell death therefore the concentrations of the nanoparticles were lowered to rutile and anatase TiO₂ (0.025 mg/ml & 0.0125 mg/ml), and ZnO (.005mg/ml) respectively. In addition to morphology and cell proliferation, the effect of the above TiO₂ and ZnO concentrations on HeLa cell current was measured using a patch clamp technique.[3] The measurement of cell average current (pA) was used to determine the efficiency of ion channels in facilitating the passage of ions across the membrane. To further study the effects of DXM on HeLa cells membrane potential as compared to non-exposed cells, Controls and TiO₂ and ZnO incubated HeLa cells of the same concentrations listed above were plated and dexamethasone (10⁻⁶M) was added to each set. The patch clamp technique was used in order to observe DXM's effects on the membrane potential of the cells. Additionally, the negative pressure applied in order to patch HeLa cells with and without the added dexamethasone was measured. The patch clamp data was analyzed by the Clamp Fit Program. Under confocal microscopy after 72h of exposure to TiO₂ and ZnO, Control HeLa cells remained healthier than the nanoparticles exposed cells. The HeLa cells exposed to the nanoparticles appeared shrunken and dying. Over a 4d period, average HeLa cell counts of all nanoparticles exposed cells showed a slowing of cell proliferation as compared to controls. ZnO appeared to have the greatest effect on proliferation followed by anatase TiO₂.(fig.1,2) After 24h exposure to above mentioned concentrations of nanoparticles, HeLa cells membrane instantaneous and steady state currents were measured via patch clamp technique.(fig.3) ZnO again seemed to have a greater effect on the current as the voltage was raised. To confirm that the data was based on a strong gigaseal and not due in part to leaking, initial and final resistances were noted and found to be stable. In addition, the patch clamp technique was used on control and nanoparticle exposed HeLa cells with DXM. No significant difference in instantaneous currents of controls with DXM vs nanoparticle exposed HeLa cells with DXM was found. In conclusion, this study shows that both TiO₂ and ZnO decrease HeLa cell proliferation and increase HeLa cell membrane permeability. TiO₂ and ZnO effect on the HeLa cell membrane seems to be blocked by DXM. With a greater understanding of this mechanism, cell specific targeted cytotoxicity of TiO₂ and ZnO may be useful in fighting cancers like cervical adenocarcinoma.

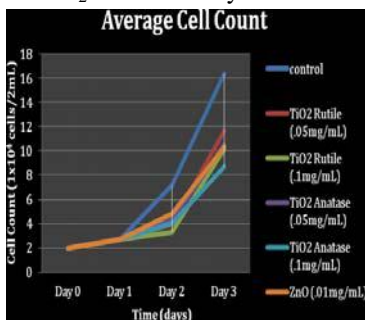


Fig.1: HeLa cell counts with TiO₂ and ZnO and controls over 4 days

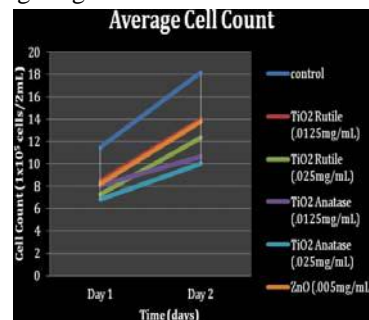


Fig.2: HeLa cell counts with TiO₂ and ZnO and controls over 2 days

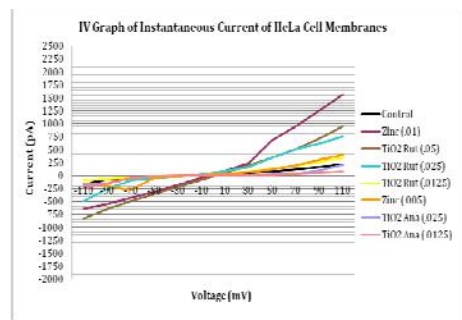


Fig.3 Instantaneous I-V Graph displays the effect of nanoparticles on HeLa cell membrane current.

[1] Zhi, Pan et al. Adverse Effects of Titanium Dioxide Nanoparticles on Human Dermal Fibroblasts and How to Protect Cells. APS (2006) 26: 19-22
 [2] Fratoddi, I. Venditti, I. Cametti, C. Palocci, C. Chronopoulou, L. Marino, M. Acconcia, F. Russo, M.V. Functional polymeric nanoparticles for dexamethasone loading and release. Biointerfaces (2012) 93:59-66.
 [3] Jackson, M.B., Whole-Cell Voltage Clamp Recording. Current Protocols in Neuroscience (2001) 6.6.

The cytotoxicity of titanium dioxide nanoparticles and their effect on the infectivity of PRV

Briana Friedman¹, Nili Greenberg¹, Phoebe Wang²

Mentors: Yan Xu³, Dr. Sarah Gross⁴, Dr. Miriam Rafailovich³

¹*Yeshiva University High School for Girls, Holliswood, NY*

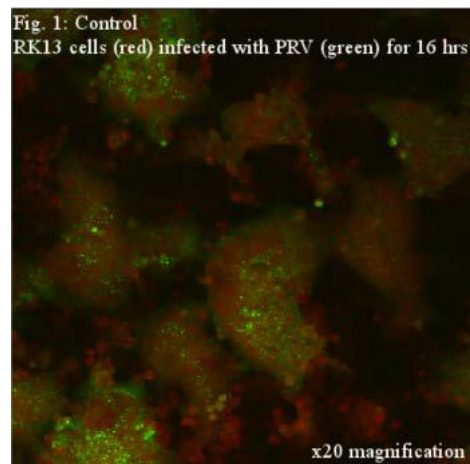
²*Conestoga High School, Berwyn, PA*

³*Department of Materials Science and Engineering, Stony Brook University, Stony Brook, NY*

⁴*Farmingdale State College, Farmingdale, NY*

Titanium dioxide nanoparticles are found in sunscreen, food, cosmetics, paints, and toothpastes because of their white color and UV-absorbing ability. Though small quantities of these nanoparticles are considered non-toxic by federal regulatory agencies¹, the question of the particles' cytotoxicity remains. Titanium dioxide nanoparticles are most commonly found in anatase and rutile forms, which differ in their crystal structure. Anatase has been empirically found to be the more cytotoxic of the two². RK13 cells are from a cell line isolated from the kidney of a rabbit, and are known to be susceptible to pseudorabies virus (PRV), a herpesvirus which infects non-human mammals³ (see Fig. 1). Preliminary research has shown that these nanoparticles cause the cells to be more susceptible to infection. The purpose of this project is to determine the cytotoxicity of anatase and rutile nanoparticles in varying concentrations, and to determine the effect that these nanoparticles have on the infectivity of PRV. RK13 cells were plated in DMEM in 6-well cell culture plates. Twenty-four hours after plating, nanoparticles were added to the wells. There were two groups of plates: one to be only exposed to nanoparticles, and one to be infected with virus as well. For the only- nanoparticle group, the nanoparticles, both anatase and rutile, were added at varying concentrations. The second time this experiment was run, coated rutile nanoparticles were also added. These nanoparticle-only plates were counted at one, two, and three days following adding of nanoparticles. Results suggest that both anatase and rutile nanoparticles are cytotoxic, rutile more so than anatase. Additionally, the nanoparticles seem to cause the cells to be more susceptible to infection.

Future work will include repetition of these experiments to ensure accuracy and AFM microscopy to further examine the effects of the nanoparticles on the cells.



¹Food and Drug Administration, Department of Health and Human Services. (2011). *Title 21--food and drugs, chapter i--food and drug administration, department of health and human services, subchapter a--general, part 73 -- listing of color additives exempt from certification, subpart a--foods, sec. 73.575 titanium dioxide.* (21CFR73.575). Retrieved from website: <http://www.accessdata.fda.gov/scripts/cdrh/cfdocs/cfCFER/CFERSearch.cfm?fr=73.575>

²Pan, Zhi, Wilson Lee, Lenny Slutsky, Richard A. F. Clark, Nadine Pernodet, and Miriam H. Rafailovich. "Adverse Effects of Titanium Dioxide Nanoparticles on Human Dermal Fibroblasts and How to Protect Cells." *Small* 5.4 (2009): 511-20. *Wiley Online Library*. Web. 5 Aug. 2012. <<http://onlinelibrary.wiley.com.libproxy.cc.stonybrook.edu/doi/10.1002/sml.200800798/pdf>>.

³Roisman, B., and Pellett, P. E., (1991) "The family Herpesviridae". In Knipe, DM, Howley, PM (ed) *Fields Virology*, 2nd edn. Raven Press, New York, pp. 2381-2397

Effects of Micelle Coated TiO₂ and ZnO nanoparticles on Targeting Macrophages Infected with *Leishmania tropica* In Vitro

Alexander Lee¹, Allison Lee¹, Mariah Geritano², Yury Yakubchyk²,
Christine Falabella², Wilson Lee³, Ying Liu², Miriam Rafailovich²

¹Hauppauge High School, Hauppauge, NY, ²Stony Brook University, Stony Brook, NY, ³Research and Development, Estee Lauder Inc.

Currently, approximately 12 million people in the world are infected with leishmaniasis¹. *Leishmania tropica*, the cause of leishmaniasis, is widespread through third world countries where treatments are expensive. Furthermore, strains of *Leishmania tropica* are becoming more resistant to current medication making the search for a novel and inexpensive treatment more critical than ever². Fortunately, the rapidly expanding field of nanotechnology provides new hope for combating *Leishmania*; free radical generating titanium dioxide (TiO₂) and zinc oxide (ZnO) have been found to be detrimental to most organisms within close proximity. Since *Leishmania*, TiO₂, and ZnO all sequester within the vacuoles of macrophages, these nanoparticles are potential candidates as an inexpensive treatment to the parasite.

In our experiment, we used J774A.1 macrophage cells and *Leishmania tropica* parasites for infection. To test the effect of UVB radiation on TiO₂ and ZnO on healthy macrophages, various concentrations (0.01 and 0.05 mg/mL) of TiO₂ and ZnO nanoparticles were added to cultures of healthy macrophages. The cultures were then exposed to UVB for 5 minutes and then counted at different time points of 24, 48, and 72 hours. In Figure 1, results showed that with UVB, almost all the macrophages were killed. Without UVB, as the concentration of nanoparticles increase, there was a lower viability rate.

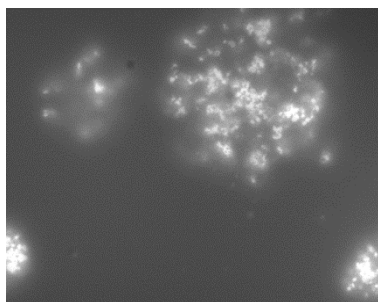


Figure 2: Confocal microscopy image of aggregation of TiO₂ anatase micelles.

It would be desirable to create a drug delivery mechanism to allow a higher number of nanoparticles to accrue into infected macrophages while repel from healthy ones. We hypothesize that by encapsulating the nanoparticles with different charge and surface tension, a particular combination will allow us to drive most of the particles into infected macrophages. We encapsulated TiO₂ and ZnO nanoparticles by sonicating nanoparticles with lectinol phospholipid. Confocal microscopy shown in figure 2 and transmission electron microscopy images were taken to confirm that these nanoparticles were successfully encapsulated. Further experiments are planned to study the effect of micelle coated nanoparticles on healthy and infected macrophages in the future. As the field of nanotechnology expands, potential treatments of untreatable illnesses are revealed. If the results for the encapsulated nanoparticles are successful in targeting infected macrophages, this concept can be applied to aid in curing other diseases based on surface charge and tension.

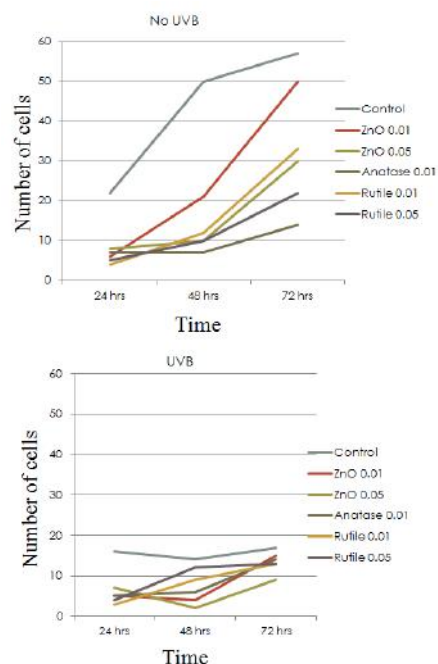


Figure 1: No UVB exposure (top) showed that the macrophages recovered growth rates over time. In contrary, macrophages exposed to UVB (bottom), displayed a detrimental effect to growth rate.

¹ Allahverdiyev, Adil M., Rabia C. Koc, Sezen C. Ates, Malahat Bagirova, Serhat Elcicek, and Olga N. Oztel. "Leishmania Tropica: The Effect of Darkness and Light on Biological Activities in Vitro." Elsevier (2011).

² Allahverdiyev, Adil M., Emrah S. Abamor, Malahat Bagirova, and Miriam Rafailovich. "Antimicrobial Effects of TiO₂ and Ag₂O Nanoparticles against Drug-resistant Bacteria and Leishmania Parasites." Future Microbiology (2011). (in press)

The Effects of Dexamethasone on Dental Pulp Stem Cells Treated with Titanium Dioxide Nanoparticles

Hannah Silva, St. Francis High School, Sacramento, CA

Melissa Clark, Victoria East High School, Victoria, BC

Miriam Rafailovich, Vladimir Jurukovski, and ChungChueh Chang, Dept. of Materials Sciences, Stony Brook University

Marcia Simon, Department of Oral Biology and Pathology, Stony Brook University



Although titanium dioxide nanoparticles are used in a variety of industrial and medical applications, their adverse effects have not been thoroughly researched. Titanium dioxide nanoparticles are known to induce cell death in dental pulp stem cells (DPSCs).¹ The purpose of this experiment is to explore the effects of titanium dioxide nanoparticles on DPSCs when the synthetic glucocorticoid dexamethasone is added to the DPSCs. Dexamethasone has been thought to protect cells from the toxic effects of nanoparticles similar to titanium dioxide.² We hoped to find out which concentration of dexamethasone is most effective against the cytotoxicity of the TiO₂ nanoparticles. For that purpose the proliferation of cells under different treatments was assessed.

Cells were grown on polystyrene well plates and then counted at regular intervals. Cells were observed to proliferate the best in control media and also in the media supplemented with dexamethasone at the concentration of 10⁻⁸ M. The cells treated with TiO₂ (without dexamethasone) experienced the highest degree of cell death, as expected. However, the cells treated with both TiO₂ nanoparticles and dexamethasone at 10⁻⁸ M (D1), 10⁻⁹ M and 0.5x10⁻⁹ M concentration had higher proliferation rates than the cells grown with TiO₂ alone indicating some protective effect of dexamethasone. (Figure 1).

We will continue to analyze the DPSCs using the confocal microscope (pictures taken on days 7 and 9) to determine the effects on cell morphology, the electron scanning microscope and energy dispersive x-ray spectroscopy to determine presence of biomineralization (completed on days 13 and 19), the transmission electron microscope to determine the location and distribution of the nanoparticles within the cells, and RNA extraction followed by RT-PCR to determine the gene expression of osteogenic differentiation markers (completed on days 13 and 19).

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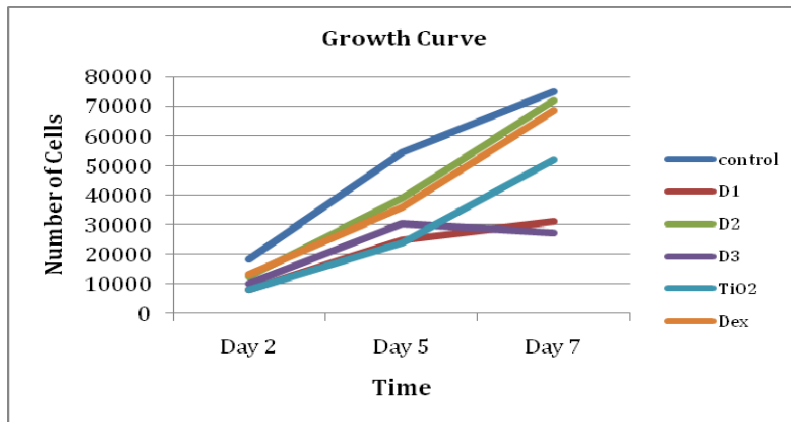


Figure 1: Growth curve of DPSCs growing in different environments. The control contains normal media. D1, D2, and D3 contain cells growing in constant concentration of titanium dioxide (0.1 mg/ml) and varying concentrations of dexamethasone. TiO₂ contains normal media and TiO₂ nanoparticles. Dex contains normal media and dexamethasone at 10⁻⁸ concentration.

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¹ G. Gronthos, B. Ibrahim, W. Li, W. Fisher, N. Cherman, A. Boyde, P. DenBesten, P. Gehron-Robey, and S. Shi. "Stem Cell Properties of Human Dental Pulp Stem Cells." *J Dent Res* (Aug 2002) 81: 31-35.

² B. Alliot-Licht, G. Pluteau, P. Magne, T. Lopez-Cazaux, P. Jubeau, G. Paculsi, and G. Guicheux. "Dexamethasone stimulates differentiation of odontoblast-like cells in human dental pulp cultures." *Cell Issue Res* (2005) 21: 91-100.

The Effect of Titanium Dioxide Nanoparticles on the Growth and Differentiation of Dental Pulp Stem Cells and Preadipocytes

Nicolette Almer, Kimia Ziadkhanpour, Plainview - Old Bethpage John F. Kennedy High School, Plainview, NY
Chung-Chueh Chang, Tatsiana Mironava, Miriam Rafailovich, Marcia Simon, Stony Brook University

Teeth afflicted by apical periodontitis, dental caries, or other oral bacterial infections are in weakened states, and thus are more susceptible to damage. Nanoparticles are particles measuring less than 100 nanometers; at this minute scale, such particles are able to ease their way through the pores in the teeth and reach the pulp chamber, in which dental pulp stem cells reside. Although nanoparticles such as titanium dioxide (TiO₂) and zinc oxide (ZnO) are found in dental materials, such as fillings, most oral contact with nanoparticles occurs when teeth are brushed and treated with toothpaste. Titanium dioxide nanoparticles are often major constituents of toothpastes; however, prior studies have been unable to prove the cytotoxicity of these nanoparticles on stem cells¹.

Dental pulp stem cells (DPSCs) are a type of pluripotent stem cell that have the ability to differentiate into a number of somatic cells, including osteoblasts, odontoblasts, and neurons. The potential of DPSCs and lack of controversy surrounding their retrieval from humans has made them the principal focus of regenerative endodontic studies². Similarly, human preadipocytes (HPAds) are fibroblast-like precursor cells derived from human adipose tissue that have gained increasing attention from the scientific community.

In this experiment, the effect of titanium dioxide nanoparticles on the growth and differentiation of dental pulp stem cells and human preadipocytes was studied. Cells were treated with varying concentrations of rutile and anatase, the two most common crystalline forms of titanium dioxide¹. The cells were plated and counted using standard hemacytometer procedure on days 0, 2, 4, and 7 in order to obtain growth curves. Results indicated that at higher concentrations of nanoparticles (1 mg/mL), DPSCs and preadipocytes tended to die at increased rates than at lower concentrations (0.1 mg/mL). For differentiation, concentrations of anatase and rutile were lowered to 0.05 mg/mL and 0.01 mg/mL to ensure the survival of DPSCs and preadipocytes for the full 28-day duration of the experiment. Confocal microscopy was used to view cells after 7 days of exposure to nanoparticles; it could be seen that the morphology of the cells' actin fibers and nuclei differed according to the concentration of titanium dioxide used and whether the cells were treated with anatase or rutile (Figure 1).

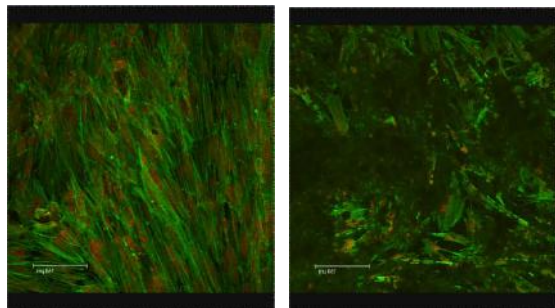


Figure 1. Confocal images of day 7 preadipocytes (left, control; right, after treatment with 0.1 mg/mL rutile).

Our future work will consist of differentiating the DPSCs on thick (2000 Å) and thin (200Å) polybutadiene surfaces. Rt-PCR analysis and SEM imaging on days 21 and 28 of the experiment will indicate the presence of differentiated cells, along with gene expression and extent of biomineralization. In addition, we will be collecting information about the differentiation of human preadipocytes in 6-well plates and will analyze lipid accumulation in these cells over 28 days.

¹Gerloff, Kirsten, Ivana Fenoglio, Emanuele Carella, Julia Kolling, Catrin Albrecht, Agnes W. Boots, Irmgard Forster, and Roel P.F. Schins. "Distinctive Toxicity of TiO₂ Rutile/Anatase Mixed Phase Nanoparticles on Caco-2 Cells." *Chemical Research in Toxicology* 25 (2012): 646-55. Print.

²Güven, Esra Pamukcu, Mehmet Emir Yalvac, Fikrettin Sahin, Munevver M. Yazici, Albert A. Rizvanov, and Gundaz Bayirli. "Effect of Dental Materials Calcium Hydroxide-containing Cement, Mineral Trioxide Aggregate, and Enamel Matrix Derivative on Proliferation and Differentiation of Human Tooth Germ Stem Cells." *Journal of Endodontics* 37.5 (2011): 650-56. Print.

The Effects of Dexamethasone on the Cytotoxicity of ZnO Nanoparticles in Dental Pulp Stem Cells

Rachel Yang, Commack High School, Commack, NY

Ying Liu, Stony Brook University, Stony Brook, NY

Marcia Simon, Oral Biology and Pathology, Stony Brook University, Stony Brook, NY

Miriam Rafailovich, Dept. of Materials Science and Engineering, Stony Brook University, Stony Brook, NY

Nanotechnology is a rapidly expanding field of materials engineering that has many applications in drug delivery, biomedical imaging, and consumer products. Zinc oxide (ZnO) nanoparticles are particularly used in sunscreen and cosmetics due to their ability to absorb UV light. However, there has been concern regarding potential adverse health effects due to the abundance of ZnO in consumer products. ZnO nanoparticles have been shown to be toxic towards proliferating cells, such as cancer stem cells, while leaving differentiated cells relatively unharmed. The glucocorticoid dexamethasone (Dex) induces cell differentiation in human osteoblasts; therefore it may be able to reduce ZnO toxicity. This was used as a basis for investigating the effects of dexamethasone on ZnO cytotoxicity in dental pulp stem cells (DPSCs), multipotent stem cells that have the ability to differentiate into osteoblasts, odontoblasts, and adipocytes.

The day after DPSC plating, the samples with nanoparticles were given a ZnO concentration of 0.05g/mg/mL while the control samples had no nanoparticles. Different concentrations of dexamethasone (0, 1.1x10⁻⁸, 0.5x10⁻⁸, and 1x10⁻⁸ M) were added to cell samples with and without ZnO.

Cell growth curves were used to show the relative growth rates of cells with and without nanoparticles and with varying concentrations of dexamethasone. Cell counts on Day 3 and Day 7 of cell growth show that 0.1g and 0.5g concentrations of dexamethasone do not provide protection from ZnO. On Day 28 the cells will be evaluated for differentiation and bio-mineralization using SEM. In addition, confocal microscopy will be used to determine cell health by viewing morphology, cell density, and actin fiber alignment. **Figure 1. A** shows that the DPSCs without ZnO and with 0.5g Dex have normal actin filaments dyed green with Alexa Fluor 488. **Figure 1. B**, the DPSCs cultured with ZnO and 0.5g Dex had damaged actin filaments, demonstrating that ZnO nanoparticles destroy the cell's actin fibers. Both confocal images were taken on Day 7.

Future studies include investigating the effects of dexamethasone on gold nanoparticle toxicity in DPSCs and ZnO toxicity in adipocytes to determine whether the cell type and nanoparticle material are affected differently by dexamethasone. Furthermore, other future investigations will focus on the effects of polystyrene vs. poly(4-vinylpyridine) (P4VP) surfaces on cell distribution and nanoparticle uptake.

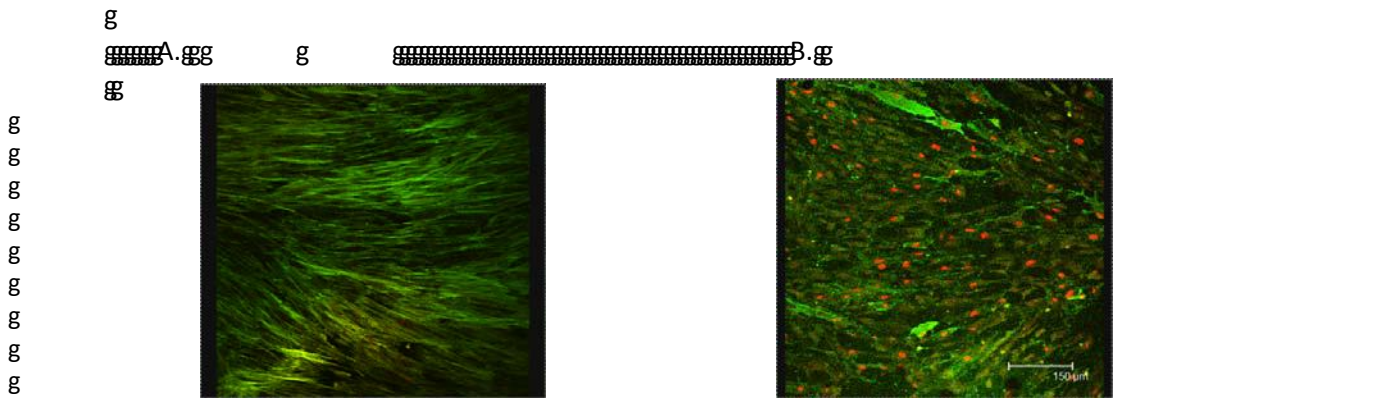


Figure 1. A. The effects of 0.5g Dex without ZnO on DPSCs (Day 7) B. The effects of 0.5g Dex and ZnO on DPSCs (Day 7)

¹ Sharma, V., Anderson, D., & Dhawan, A. (2012). Zinc oxide nanoparticles induce oxidative DNA damage and ROS-triggered mitochondria mediated apoptosis in human liver cells (HepG2). *Apoptosis*, (17), 852-870.

² Taccola, L, et. al. (2011). Zinc oxide nanoparticles as selective killers of proliferating cells. *Int J Nanomedicine*, 6, 1129-1140.

³ Yamanouchi, K., Gotoh, Y., & Nagayama, M. (1997). Dexamethasone enhances differentiation of human osteoblastic cells in vitro. *Journal of Bone and Mineral Metabolism*, 15, 23-29.

⁴ Gronthos, S, et. al. (2000). Postnatal human dental pulp stem cells (DPSCs) in vitro and in vivo. *Proceedings of the National Academy of Sciences*, 97(25), 13626-13630.

Innovative Medical Technologies

Chairs:

Julia Landsberg, Queens
College, New York

Adam Ossip, Brandeis
University, Waltham, MA

Graduate Students:

Clément Marmorat

Kai Yang

Divya Bhatnagar



Using Digital Image Speckle Correlation (DISC) for Analysis of Severe Burn Scarring

Drew O'Neil¹, Adam Ossip², Julia Landsberg³,

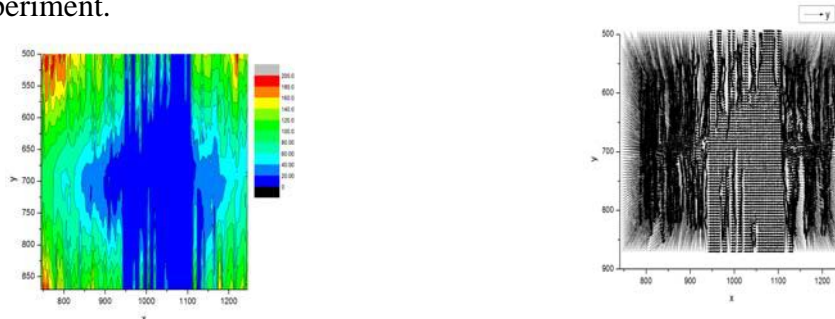
Divya Bhatnagar⁴, Marcia Simon⁴, Miriam Rafailovich⁴

¹ South Side High School, ² Brandeis University, ³ Queens College, ⁴ SUNY Stony Brook

Burns are a widespread injury that can cause permanent damage to the skin and the underlying tissue. Burns are caused by actin and myosin production of myofibroblasts, which results in actin-myosin filaments giving a net force inwards of the wound bed.¹ Burns that experience severe contraction can cause deformation and immobilization. Since the skin is attached to the underlying muscle studying the burn can also gauge the muscle trauma caused by the burn. Developing a means to measure the healing process of a burn can allow us to gauge the effectiveness of the wound healing process.

Digital Image Speckle Correlation (DISC) can track the motion of skin pores when there is an applied force. DISC requires two consecutive images to be taken for the given area. Despite no obvious motion is seen between the two pictures Since DISC only requires photos it can provide a non-invasive, quick and quantitative measurement of scar contracture. Three pigs were used for this experiment with each pig receiving sixteen burns applied to the back of the pig. Sixteen burns were applied to each side of the pig's back. After the burns were implemented, the pigs were tattooed around each of the burns. Three types of ointments were applied to the burn wounds: collagenase (new formulation), Santyl and a pluronic gel. Five burns were treated with each of the ointments, which allowed one control where none of the ointments were applied.

From DISC we can plot the motion of the wound found by contour and vector maps by using Origin Pro 8. Contour maps are generated which show the magnitude of motion using different colors for the burn area. Vector maps are generated show the magnitude and direction of motion for the burn area. These maps allowed us to see how a wound contracted. It was shown that generally all the wounds from day zero to day twenty-eight improved in contraction. Looking more closely at the punch biopsies with DISC we were able to look at the underlying muscle. To further this study it must be to human subjects and this will allow us to understand the scopes and limitations of this experiment.



Eddy, R.J., J.A. Petro, and J.J. Tomasek, Evidence for the nonmuscle nature of the "myofibroblast" of granulation tissue and hypertrophic scar. *Am J Pathol*, 1988. 130(2): p. 252- 60.

Engineering a Multiplexed, Electronic, and Intelligent Drug Delivery Platform for Next-Generation Chemotherapy

Sachit Singal¹, Rohit Mehandru², Miriam Rafailovich³, Jonathan Sokolov³, Nikhil Mehandru⁴, Ilan Gold⁵

¹Herricks High School, New Hyde Park, NY

¹Roslyn High School, Roslyn, NY

³Dept. of Materials Science & Engineering, SUNY at Stony Brook, NY

⁴Harvard University, MA

⁵University of Maryland, MD

According to the World Health Organization (WHO), cancer is “a leading cause of death worldwide” and killed 7.6 million people in 2008.¹ With current chemotherapy techniques such as surgery and radiation therapy, there is a possibility of malignancy reoccurring and of the cancer patient experiencing side effects related to systemic treatment.² As a result, there is a growing need for more precise and inexpensive drug delivery methods that can administer potent medicines to a given region. This study aimed to accomplish this by developing a smart, wireless micropump that can externally inject tumor-targeted chemotherapy drugs. Additionally, this novel drug delivery platform is extremely cost-efficient and entirely disposable.



Figure 1: The aluminum mold used to create our backflow inhibitors.

The pumping unit is able to administer these potent drugs in small quantities over extended periods of time by using a multi-layer mechanism. In creating the pump, we molded two backflow inhibitor valves from high-density polyethylene (HDPE) and kapton, positioned a ceramic piezoelectric actuator, programmed a self-correcting flow sensor dependent on changes in temperature, and attached a drug reservoir component molded from polydimethylsiloxane (PDMS). The code for the wireless microcontroller was written using Code Composer Studio v5 as well as Atmel Studio v4.17 and was externally programmed using each respective field-effect transistors.

After successfully developing the pump, some preliminary testing was performed to assess specific pumping characteristics. We measured the average minimum and maximum force exerted by the pump to eject solutions of variegated viscosities, the average minimum and maximum rate of flow, and the average overall energy assumption of the pumping unit. These tests helped to determine the necessary adjustments needed in order to manipulate the pumping unit for a variety of drug medications.

With the aim to simulate the delivery of chemotherapeutic agents to patients diagnosed with pancreatic cancer, we first sought to mimic the viscosity of drugs such as gemcitabine monotherapy (phase 1), gemcitabine monotherapy (phase 2), gemcitabine in combination with erlotinib, and other cocktail chemotherapeutic drugs by combining deionized water and glycerol in fixed volume ratios. Given the myriad number of viscosity levels for these drugs, we created these water/glycerol solutions along a broad range of viscosities. Preliminary testing was performed by injecting these different solutions (marked by different food coloring) into a closed, transparent cavity. The aforementioned pumping characteristics were then monitored based on the discrepancies in drug viscosity.

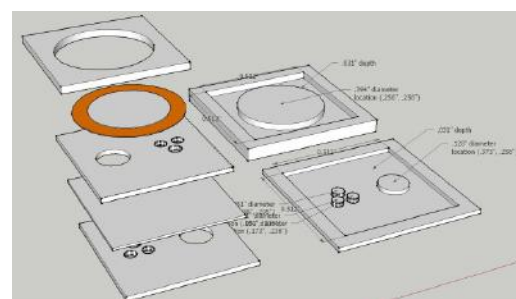


Figure 2: A 3D design of our pumping unit.

In our future work, we hope to reinforce the data collected from our mock-chemotherapeutic drugs by testing with actual gemcitabine. After inoculating two Petri dishes with two distinctive pancreatic cancer cell lines (Panc-1 and BXP3), we hope to perform IC₅₀ tests in each dish to test the precision and effectiveness of our drug delivery platform in a real-time in vitro environment.

¹World Health Organization (WHO) Cancer Fact Sheet No. 297. Geneva, Switzerland: GLOBOCAN Section of Cancer Information, 2008.

²Carelle, N., E. Piotto, A. Bellanger, J. Germanaud, A. Thuillier, and D. Khayat. “Changing Patient Perceptions of the Side Effects of Cancer Chemotherapy.” *Cancer* 95 (2002): 155-63.

³Langer, R. “Drug Delivery and Targeting.” *Nature* 392 (1998): 5-10.

Engineering a dynamic valve system for hydrocephalus management

Kaveh Issapour Woodbury, NY
Sohini Upadhyay Port Washington, NY

Hydrocephalus is a medical condition characterized by abnormal accumulation of cerebrospinal fluid (CSF) in the ventricles, or cavities, of the brain. The accumulation of CSF leads to neurological problems such as convulsions, mental retardation, epileptic seizures, and in some cases death. Hydrocephalus is treated via surgery, usually by creating various types of cerebral shunts. Shunts come in many forms but all of them consist of a pump or drain connected to a long catheter, the end of which is usually placed in the space between the two membranes that separate the organs in the abdominal cavity from the abdominal wall (peritoneal cavity); this

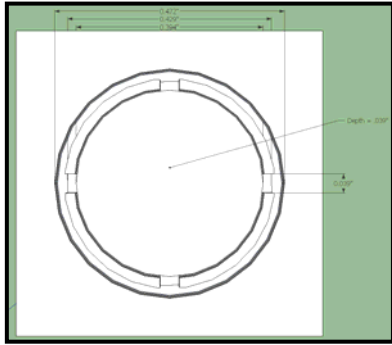


Figure 1: Top View of Valve Molds

bypasses the flow obstructions and drains the excess fluid into other body cavities, from where it is able to be reabsorbed. Excess fluid in existing pumps is drained from the brain gravitationally over a high to low pressure gradient. An unfortunate consequence of this drainage mechanism is a process known as siphoning. Siphoning refers to the formation a vacuum or suction effect in the drainage site due to the uncontrolled acceleration of flow by gravity. This leads to overdrainage, an issue associated with various health risks ranging from migraines to subdural hematomas. This suction effect also acts upon biological debris in the drainage ventricle,

occluding valves and necessitating valve replacement and surgery. Anti-siphon valves on the market offer resistance against accelerated flow, but still rely on gravity for movement. We hypothesized that a valve capable of pumping CSF on its own, independent of gravity, would offer better protection against siphoning. Our current prototype is a Polydimethylsiloxane tube

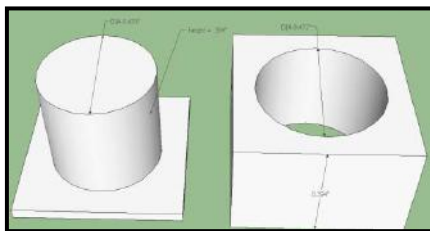


Figure 2: Side Views of Valve Molds

with a piezoelectric actuator. The piezo disc act as a flap, vibrating to move and block flow when stimulated potentiometrically. The piezodisc facilitates pulsatile flow; as the bulk of cerebral fluid dynamics is pulsatile, this prototype is more finely tuned to the brain, dually functioning as a valve for treatment and a valve for furthering the understanding of hydrocephalus' pathology. Currently we are in the process of creating the components of the valve with the use of an aluminum mold, the design of which is pictured below. Once

assembled, we aim to fully characterize the functionality of the valve, tabulating applied voltage against corresponding flow rate. Further aims also include incorporating pressure *sensors* and external modulation of the valve through wifi.

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2. Brian Sweetman, Andreas A. Linninger. (2011) Cerebrospinal Fluid Flow Dynamics in the Central Nervous System. *Annals of Biomedical Engineering* 39:1, 484-496. Online publication date: 1-Jan-2011

Analysis of Commercially Available Gutta Percha Materials

Alexa Aseel-Fine—Jericho Senior High School, Jericho, NY
Dr. George Bruder and Dr. Christopher Joubert—School of Dental Medicine, Stony Brook University
Clément Marmorat—Stony Brook University, Department of Materials Science and Engineering
Kai Yang—Stony Brook University, Department of Materials Science and Engineering
Dr. Miriam Rafailovich—Stony Brook University, Department of Materials Science and Engineering

Root canals are the most common endodontic procedures, with approximately 40 million performed each year. Presently, using gutta percha cones with sealer is one of the most reliable methods for filling the root canal system. If a root canal is not obturated properly, however, the root can become re-infected¹. Gutta percha is a non-cytotoxic rubber material that contains the molecule transpolyisoprene, and can work in both humid and dry conditions^{1,2}. The purpose of this experiment was to determine the proliferating and differentiating effects of various thicknesses of polyisoprene (PI) in a 2D model on dental pulp stem cells (DPSCs) and to analyze the mechanical properties of three different gutta percha products on the market: Lexicon, Protaper, and Guttacore. A 7-day growth curve was performed which showed that the thick had about the same doubling time as the control (1.725 days) while the thin grew at a slower rate (Figure 1). Likewise, AFM to test cell modulus, SEM/EDX to test for biomineralization, confocal microscopy to analyze cell morphology and ST-PCR to analyze gene expression will all be done to further test the effect of PI on DPSCs. Additionally, tensile strength testing was performed on an Instron 5524, but in order for the clamps not to crush the sample, dental filing (compactable composite) was used on the ends of samples and cured with a blue light for 20 seconds on each side to harden it. The results showed that after being worked for two minutes the elastic modulus of Lexicon and Protaper decreased, showing that the material was more pliable in addition to being able to handle more load. Guttacore showed a much smaller elastic modulus than the other samples and could carry far more load than them. Finally, rheology was performed on Lexicon and Protaper. This showed that Protaper was a harder material because it had a higher G' value than Lexicon. At the same time, because Lexicon has a higher breaking stress than Protaper, it is able to handle stress for a longer period of time and is able to handle more stress than Protaper between the two breaking stresses (Figure 2). Future work will include doing rheology on Guttacore and Differential Scanning Calorimetry (DSC) testing. Furthermore, these results will be used to create a new gutta percha material similar to Guttacore that is less crosslinked, easier to remove, and not as brittle.

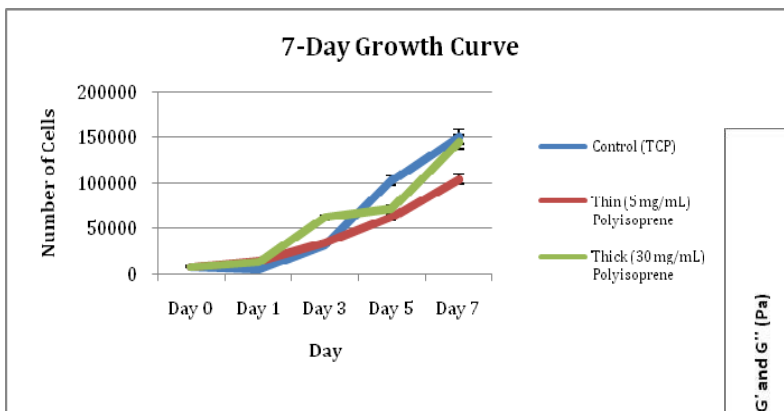


Figure 1

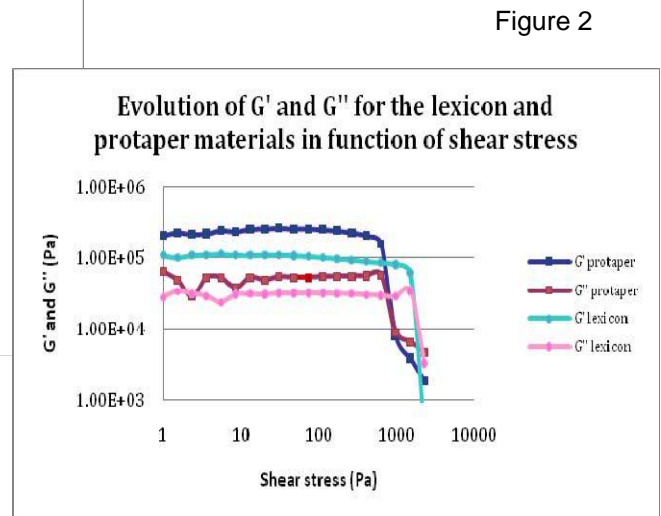
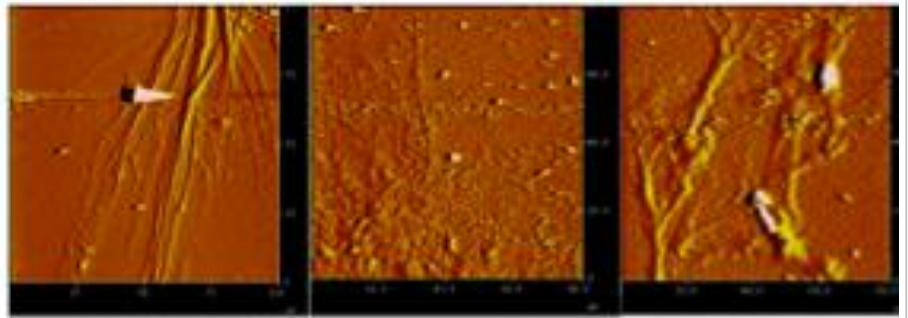


Figure 2

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Biomolecular and DNA Sensors



Chairs:

Julia Budassi, Stony Brook
University, New York

Jose Deniz, Stony Brook
University, New York

Graduate Students:

Ke Zhu

Yingjie Yu

Controlled Enzymatic Cutting of DNA Using Soft Lithography

Alyssa Auerbach¹, Julia Budassi², Ke Zhu², Dr. Jonathan Sokolov²

¹Yeshiva University High School for Girls, Holliswood, NY

²Department of Materials Science & Engineering, Stony Brook University, Stony Brook, NY

Since the discovery of DNA structure, research in molecular genetics has boomed¹, culminating recently in the Human Genome Project, which aimed to completely sequence the entire genome. Current techniques are limited to sequencing DNA molecules of 500 bases or less, requiring long DNA to be cut into many small pieces. The cutting is done either randomly or with restriction enzymes, both requiring complex reconstruction of the master sequence. The ability to cut DNA at regular, known locations would greatly simplify sequencing. Enzymatic soft lithography has been used to create nano-patterns as well by digesting specific substrate layers.² Here, we applied the technique to cutting DNA.

DNA (Lambda or T4) molecules were deposited onto PMMA-coated silicon wafers by withdrawing the substrates from the DNA solutions at 1mm/s in the presence of an electric field (to enhance adsorption). By varying the DNA concentration, either isolated or densely-packed molecules could be obtained, as observed using fluorescence microscopy on YOYO labeled DNA. Soft lithography stamps were made by curing a PDMS solution onto a silicon mold containing a grating pattern. Stamps were coated with the DNA-cutting enzyme DNase 1 and placed in contact with substrates containing adsorbed DNA. Pressure was applied for ten minutes, and then the wafer is once more observed under the confocal microscope. Results were optimized by varying the ratio of sodium hydroxide to MES Buffer suspending the DNA, changing the type of DNA used, and trying many ways of applying DNase to the stamp and peeling the stamp off to optimize the grid pattern. The efficiency of cutting the DNA was optimized by varying the PDMS surface treatment, DNase application methods, and the stamp contact pressure and duration on the substrate. DNA molecules were successfully cut along the pattern set by the stamp. (Figures 1 and 2), enabling high resolution and ordered sectioning of long DNA molecules.

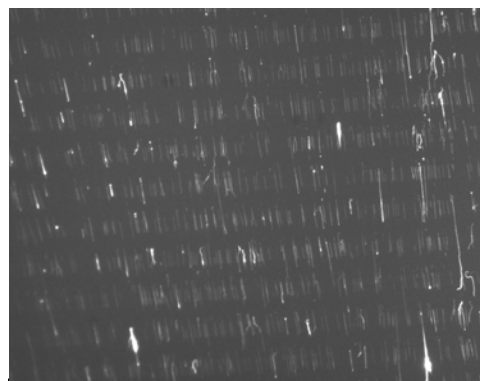


Figure 1. The DNA was able to be cut in single strands.



Figure 2. DNA was systematically cut along the lines of the stamp, even in extremely dense conditions.

¹ Check Hayden, Erika. "Life is Complicated." *Nature*. 464 (2010): 664-7.

² Guyomard-Lack, Aurélie, et al. Site-Selective Surface Modification Using Enzymatic Soft Lithography." *Langmuir*. 27 (2011): 7629-34.

Stretching DNA molecules on a flexible substrate probed by polarization-dependent fluorescence microscopy

John Mele, Central Islip Senior High School, Central Islip, NY

Ke Zhu, Julia Budassi, and Dr. Jonathon Sokolov, Department of Materials Science and Engineering, Stony Brook University, Stony Brook, NY

DNA molecules adsorbed and stretched onto surfaces can be used to analyze DNA structure by imaging fluorescence of labeled hybridization probes or enzymes. A recently proposed method for sequencing by electron microscopy requires either adsorbed single-stranded DNAs or twisted double-stranded DNA. In this experiment, studies were undergone on the adsorption of isolated DNA molecules to a flexible PDMS substrate, which permits continuous stretching, until breakage, as shown in Figure 1, of the DNA molecules. λ and λ DNAs (48.5 and 165.6 kilobase pairs, respectively) were adsorbed onto PDMS out of solution by withdrawing a submerged substrate at a rate of 1 mm/s, producing linear molecules deposited on the surface. The DNAs were labeled with the dye YOYO-1 and imaged using a 40x oil immersion lens. A rotatable polarizer allowed us to vary the incident light polarization and fluorescence emission intensity and was measured as a function of polarization angle and degree of stretching of the DNA. Emission spectra for unstretched and stretched molecules are shown in Figure 2.

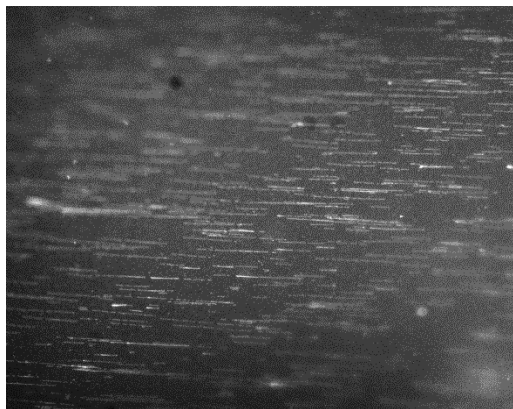


Fig. 1: DNA displays breakage at 35 cm stretch.

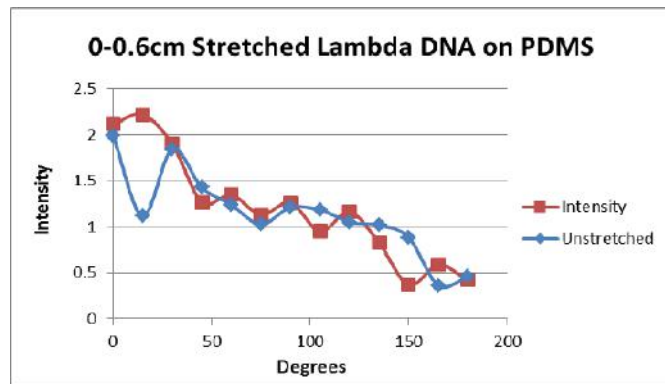


Fig. 2: Polarization data of stretched and unstretched DNA molecules on PDMS substrate.

Future work includes identifying the stretching and breakage properties of T-4g DNA on the PDMS substrate, along with measurements of pixel intensity of varying degrees of polarization under the confocal microscope. Applications of this work include the ability to view unstretched double-stranded DNA molecules for the purpose of genomic sequencing of the human DNA molecular structure. This was begun with estimations of the necessary length to nearly break particles for varying kilobase pairs of DNA, including combined and recombinant DNA¹.

¹Shiao, J., De, M., Ziegler, A. "ATP for the DNAigation Step in Base Excision Repairs Generated from Poly(ADP-ribose)" the journal of Biological Chemistry.

Expanding Biosensor Applications Through the Use of Potentiometric Technology

Puja Bansal¹, Melik Yuksel^{2g}

Alina Ranjbaran³, Nikhil Mehandru³, Yingjie Yu⁴, Dr. Miriam Rafailovich⁴

¹Half Hollow Hills High School East, NY, ²Harmony Science Academy Houston, ³Harvard University,

⁴Department of Materials Science & Engineering, Stony Brook University

As disease prevalence rises globally, research has become increasingly centered around developing inexpensive and efficient detection methods. A biosensor is a biomedical device utilizing self-assembled monolayers and potentiometric technology. One goal of this experiment is to characterize molecular imprinting on biosensors to detect the presence of virus analytes. Additionally, as technology and efficiency of health care improve, it is becoming possible to bypass lengthy traditional testing by conducting patient tests using small electronic devices that utilize the same methods used in the laboratory. Thus, this study also sought to replicate the already-established testing principles used in large-scale biosensors by miniaturizing the testing apparatus into an integrated, handheld device capable of using microfluidic technology to detect virus analytes with the same accuracy observed in laboratory-based potentiometric biosensors.

In an effort to expand the applications of the biosensors, chips were created for Adenovirus Ad12 and Poliovirus. Adenovirus Ad12 is an oncogenic virus that has been linked to gastrointestinal cancer.¹ Poliovirus is an RNA virus that has been mostly eradicated in the world today. Both of these viruses have complex structures and successful imprinting is indicative of future success with a larger variety of viruses.² Current methods of detection for viral infection are ELISA (Enzyme-linked immunosorbent assay), cell cultures, and nucleic acid hybridization.³ However, these tests are often time consuming and unspecific. Moreover, the laboratory setup is often large and generally inaccessible in third world regions.

Gold-coated silicon wafers are incubated with Dulbecco's Phosphate-Buffered Saline, alkanethiol (11-mercapto-1-undecanol), dimethyl sulfoxide, and the virus template molecules. Because of the hydrophobic interactions, a self-assembled monolayer (SAM) binds to the surface of the wafer.⁴ Deionized water is used to cleanse the surface after incubation to leave cavities specific per molecule shape. Once the molecule is reintroduced, the change in surface potential between the working and reference electrode causes a significant voltage change that is registered by a potentiometer.⁵ Meanwhile, a microprocessor and electronic components including an 18-bit voltmeter were used to create a device to automatically interpret voltage change in novel types of biosensor utilizing microfluidic technology. These chips contain two electrodes (Au/Si and Ag/AgCl) set in PDMS channels and utilize the MI and SAM methods to measure analyte presence. Furthermore, the device also collects data from usage and uploads it to a database for later analysis on measurement effectiveness.⁵

The Adenovirus Ad12 chips were cross-tested with molecules of similar sizes to ensure for selectivity. *Figure 1* shows the results of these tests. We also found the optimum imprinting concentration of Ad12 (2 μ l Adv/mL) as well as the optimal thiol concentration (0.4mg thiol/1ml DMSO). Due to time constraints, no data on the effectiveness of the Poliovirus has yet been produced. Future work will include testing the microfluidic chip with the same concentrations of Poliovirus and Adenovirus and comparing the results to common biosensors for accuracy.

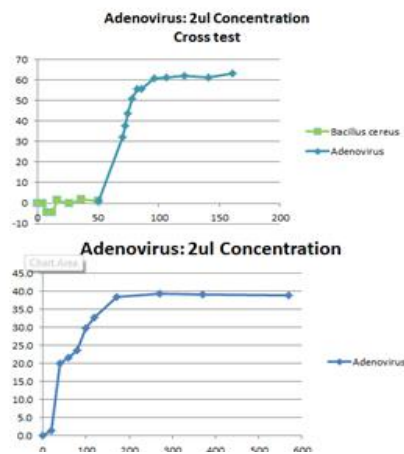


Figure 1: Adenovirus Ad12 Tests, Volume Added vs. Voltage Change in millivolts (mV)

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¹ Proffitt, Joanne. "Adenovirus 12-mediated down-regulation of the major histocompatibility complex (MHC) class I promoter: identification of a negative regulatory element responsive to Ad12 E1A." *Oxford Journals, Life Sciences*. 22.22 (1994): 4779-4788. Web. 8 Aug. 2012.

² "Progress Toward Interruption of Wild Poliovirus Transmission." *CDC*. 58.12 (2008): 308-312. Print.

³ Greer S, Alexander GJ. Viral serology and detection. *Baillieres Clin Gastroenterol*. 1995 Dec;9(4):689-721

⁴ Wang, Yantian, et al. "Potentiometric Sensors Based on Surface Molecular Imprinting: Detection of Cancer Biomarkers and Viruses." *Sensors and Actuators B: Chemical* 146 (2010): 381-87. Print.

⁵ Wang, Yantian, et al. "A Potentiometric Protein Sensor Built with Surface Molecular Imprinting Method." *Biosensors and Bioelectronics* 24.1 (2008): 162-66. Print.

Developing Methods of Disease Detection and Wound Healing through Sensing Biomolecules on Surfaces#

¹Daniela Czemerinski, ²Nicole Lin

³Alina Ranjbaran, ³Nikhil Mehandru, ⁴Yingjie Yu, ⁴Liudi Zhang, ⁵José Deniz, ⁶Dr. Miriam Rafailovich,

¹The Wheatley School, NY, ²El Camino Real Charter High School, CA, ³Harvard University, Cambridge MA,

⁴Department of Materials Science & Engineering, Stony Brook University

In the past few decades, biosensors have been greatly developed and researched in order to make medical diagnosis more efficient, less costly, and more reliable.¹ A biosensor's sensitivity, specificity, and stability must be assured before any technology can be introduced into the market. One such biosensor, the potentiometric biosensor, works by transducing a biological reaction into an electrical signal. When a gold-plated silicon wafer is immersed into a mixed solution of alkanethiol (11-Mercapto-1-undecanol) and template molecules, a process known as molecular imprinting (MI) occurs. A thiol self-assembled monolayer (SAM) binds to the surface of the wafer via sulfur-metal bonds, and template molecules are adsorbed through hydrophobic interactions and electrostatic forces.² After incubation, the template molecules can be washed off the surface of the wafer using deionized water (DI), leaving behind a thiol monolayer with cavities corresponding to the template molecules. When template molecules are reintroduced, an electrochemical signal caused by the change in surface potential of the chip can be measured using a potentiometer.³

In an effort to expand the applications of the potentiometric biosensor, we developed a biosensor to detect fibrinogen (Fb), a glycoprotein that is converted by thrombin to fibrin during blood coagulation. Abnormal Fb levels and structures are associated with cardiovascular disease, liver damage, thrombosis, and bleeding disorders.⁴ Current methods of detecting Fb-related diseases, such as global screening tests and immunological assays, lack specificity and are cumbersome.⁵

The final potentiometric measurements of our tests demonstrated that a selective biosensor can be constructed to detect Fb, even in the presence of other protein molecules that are slightly different in structure, such as damaged fibrinogen lacking α C regions (dFb), albumin (ABS), and hemoglobin (Hb). *Figure 1* shows the results of these tests. We also found the optimum imprinting concentration of Fb (0.02mg Fb/mL), the optimal thiol concentration (0.4mg thiol/1ml DMSO), and the optimal imprinting time (90 minutes).

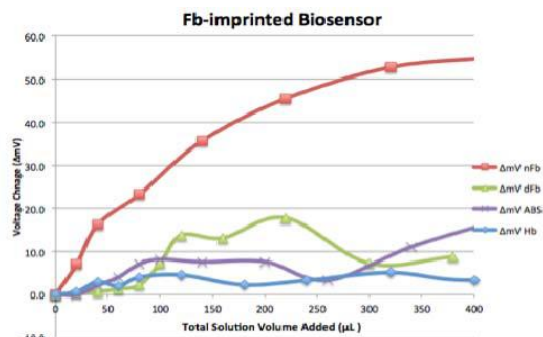


Figure 1: Selective Fb potentiometric detection, cross-tested with damaged Fb (dFb), albumin (ABS), hemoglobin (Hb).

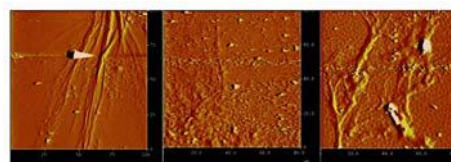


Figure 2: Examining fiber growth through AFM scans (contact mode): PS (left), PMMA (Mid), PMMA/PS blend (Right).

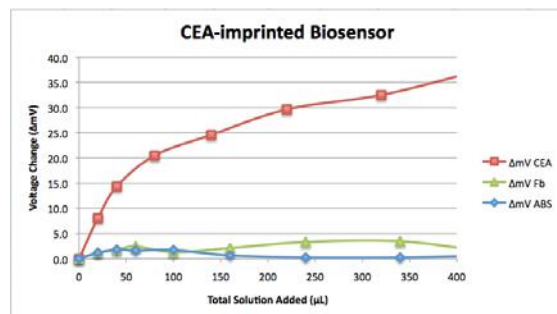


Figure 3: Selective CEA potentiometric detection, cross-tested with Fb, albumin (ABS).

specificity of this biosensor, cross tests were performed with other proteins, as shown in *Figure 3*. In the future, we hope to expand the applications of our biosensor by detecting CEA in actual patient serum.

In another study, Fb was placed onto silicon wafers with different polymer surfaces in order to investigate hydrophobic surface-induced Fb aggregation. A hydrophobic surface exposes the α C domains and other cryptic functional sites of Fb molecules, allowing lateral aggregation to occur and causing the production of fibers.⁶ Using the atomic force microscope (AFM), we examined the fibrin fibers that formed without the presence of thrombin, as shown in *Figure 2*. According to the results, the greatest fiber growth occurred on the PS surface. Further tests will be conducted to analyze whether damaged Fb lacking the intact α C region will be able to develop fibers and compare how sterilized and unsterilized conditions affect fiber growth.

To further examine the applications of biomarker detection, we constructed a biosensor selective to carcinoembryonic antigen (CEA). Increased levels of CEA in the blood of normal adults have been shown to have a correlation with pancreatic, lung, and colon cancer, leading to the use of CEA measurements as tumor markers. In order to test the

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¹ Reyes De Corcuera, Jose I. and Ralph P. Cavalieri. "Biosensors." *Encyclopedia of Agricultural, Food, and Biological Engineering* 10.1081 (2003): 119-123. Marcel Dekker, Inc. Washington State University. Web. 03 Aug. 2012. #

² Wang, Yantian, et al. "Potentiometric Sensors Based on Surface Molecular Imprinting: Detection of Cancer Biomarkers and Viruses." *Sensors and Actuators B: Chemical* 146 (2010): 381-87. #

³ Wang, Yantian, et al. "A Potentiometric Protein Sensor Built with Surface Molecular Imprinting Method." *Biosensors and Bioelectronics* 24.1 (2008): 162-66. #

⁴ Koo, Jaseung, et al. "Evaluation of Fb Self-assembly: Role of its α C Region." *J Thromb Haemost* 8(12): 2727-2735 (2011). #

⁵ Bérubé, Caroline MD. "Disorders of Fb." *UpToDate*. 26 Jun. 2012. #

⁶ Collet, Jean-Philippe, et al. "The α C Domains of Fb Affect the Structure of the Fibrin Clot, Its Physical Properties, and Its Susceptibility to Fibrinolysis." *Journal of the American Society of Hematology* 106.12 (2005): 3824-3830. #

Investigating the Sensitivity and Specificity of the Potentiometric Biosensor Mechanism Through Bacteria and Bacterial Spore Cross-Testing

Jacob Wax, Harborfields High School, Greenlawn, NY

Alina Ranjbaran, Harvard University

Yingjie Yu, Department of Materials Science & Engineering, Stony Brook University

Miriam Rafailovich, Department of Materials Science & Engineering, Stony Brook University

Bacterial infection, biological warfare and other threats that stem from infectious bacteria make the ability to detect the presence of certain strains of bacteria a low-cost and high-efficiency pressing and important goal. Using a biosensor—in this case, a potentiometric biosensor—could be an excellent method for low-cost and efficient early detection of bacterial infection. The goal of this experiment was to determine if this potentiometric biosensor could detect the difference between different strains of bacteria and bacterial spores; ultimately, the sensitivity and specificity of this biosensor mechanism was to be examined.

The potentiometric biosensors enable to detect the presence of a biomarker in a serum or solution through molecular imprinting (MI) on a gold-plated silicon wafer. Self-assembled monolayers (SAM) have proven to be an efficient way to detect small molecules, such as hemoglobin.¹ However, imprinting with organisms poses much more of a challenge; unlike proteins, organisms are unique, and differences in outer structure could cause differences in the cavities created by imprinting. To imprint with organisms, imprinting must be used; this requires a “rough” silicon wafer.

This experiment focused on testing the biosensor mechanism to see if it could detect the differences between *Bacillus cereus* spores and *Bacillus subtilis* spores. A rough gold-plated silicon wafer was introduced into solutions of dimethyl sulfoxide (DMSO), thiol (11-mercapto-1-undecanol), Dulbecco’s phosphate buffered saline (DPBS), and the *Bacillus cereus* spores concentration of 6.82×10^8 spores/mL. The wafer incubated for one to two hours in order to create a self-assembled monolayer; gas thiol molecules bind to the gold on the silicon wafer, the spores become imprinted in the SAM. The wafer was washed off after the incubation period with de-ionized water in order to wash off the biomarkers—the *Bacillus cereus* spores—and leave behind cavities that are complementary to the spores.

In order to test the specificity of the biosensor, the imprinted wafer was then connected to the two-electrode potentiometer mechanism and submerged in a well containing 2 mL of DPBS. *Bacillus cereus* spores of the same concentration as those used for imprinting were then added to the well, resulting in a jump in potential; this was a result of the spores falling into the complementary cavities. The same test was run again, but instead of *Bacillus cereus* spores being reintroduced into the system, *Bacillus subtilis* spores were added to see if the biosensor was sensitive enough to tell the difference between the two. No significant jump in potential was observed, indicating that the biosensor had successfully been specifically imprinted for *Bacillus cereus* (Fig. 1).

Future work includes using live bacteria to cross-test, as well as other types of bacterial spores such as *Bacillus thuringiensis*.

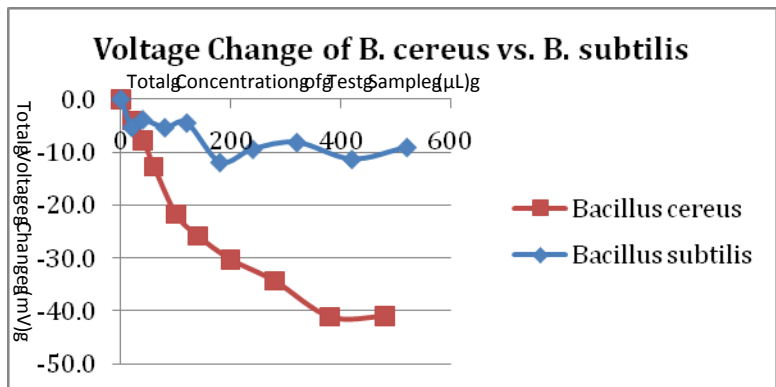


Figure 1: *Bacillus cereus* vs. *Bacillus subtilis* cross-test

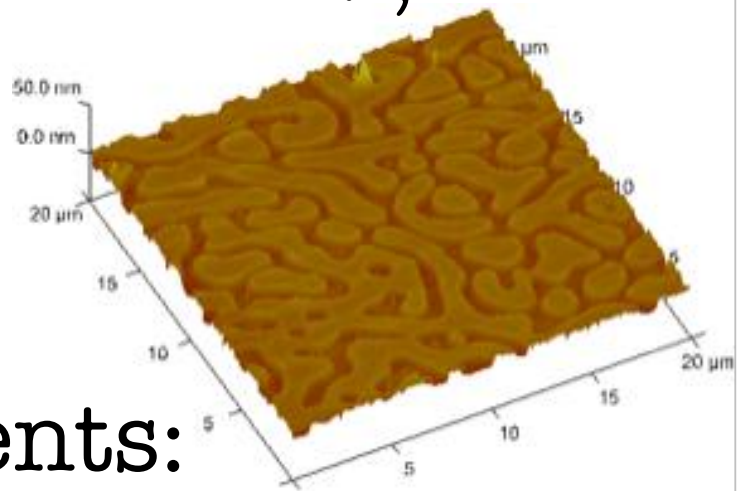
¹Wang, Y., Zhou, Y., Sokolov, G., Rigas, B., Levan, K., and Rafailovich, M. A Potentiometric Protein Sensor Built with Surface Molecular Imprinting Method. "Biosensors & Bioelectronics" 4 (2008): 62-66.

Hydrogels

Chairs:

Monika Batra, Stony Brook
University, New York

Clement Marmorat, Ecole
Polytechnique at Nantes,
France



Graduate Students:
Clément Marmorat
Yan Xu
Divya Bhatnagar

Gelatin hydrogels: The effects of physical versus chemical hardening on fibroblast adhesion and proliferation

Alex Nie¹ and Aatman Makadia²

Sneha Subramaniam³, Clément Marmorat^{4,5}, Yan Xu⁵, Divya Bhatnagar⁵,
Dr. Miriam Rafailovich⁵, and Dr. Marcia Simon⁵

¹Livingston High School, NJ, ²St. Anthony's High School, ³Columbia University,
⁴Ecole Polytechnique at Nantes, ⁵SUNY Stony Brook University

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Hydrogels have shown much promise in the field of regenerative medicine [1]. In particular, this study sought to develop gelatin hydrogels as scaffolds for cell delivery. Because hydrogel hardness influences fibroblast growth [2], the study examined fibroblast behavior under different hydrogel hardnesses. The gels were hardened physically by glucose and chemically by microbial transglutaminase (mTG), an enzyme which cross-links collagen fibers [3]

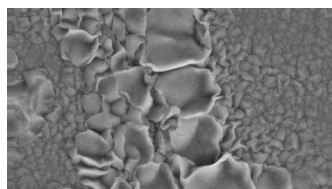


Figure 1: SEM of calcium phosphate deposits on 2mg/ml glucose hydrogel. As seen, plating occurred over the entire surface.

A frequency sweep rheology was used to analyze the hardness of gels. Overall it was shown that that longer incubation periods and temperatures around 55°C corresponded to higher hardness, evincing the importance of mTG cross-linking to hardness. In addition, hardness varied with glucose concentration, with maximum hardness at 2mg/ml glucose.

In previous studies, it was shown that mTG crosslinked hydrogels undergo biomineralization of calcium phosphate, even in the absence of cells [4]. To understand this effect in diabetics, a scanning electron microscopy was performed on the gels immersed in a solution of calcium and phosphate ions. As shown in Figure 1, deposits appeared on the surface, which Electron Dispersive X-ray Spectroscopy (EDX), confirmed were calcium phosphate. Even deposition was seen at 2mg/ml glucose while uneven, clumped deposition was seen at 5mg/ml.

In a study of cell adhesion, confocal microscopy was used to examine fibroblasts plated chemically hardened. As seen in Figure 2, fibroblasts plated on harder gels tended to have parallel actin filaments, revealing better adhesion than those plated on softer gels.

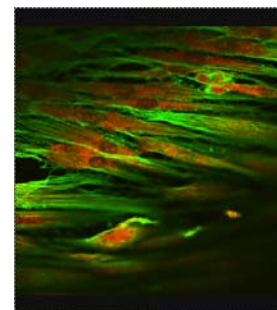


Figure 2: Confocal image of fibroblasts on hard gelatin. Parallel actin (green) indicates good cell adhesion

To analyze the effect of gel hardness on cell proliferation, a cell count was performed on fibroblasts plated on hydrogels for 24 hours with various glucose concentrations. Images of the cells were taken using an optical microscope and analyzed using ImageJ, a computer software for cell counting. For all gels, the 2mg/ml glucose gel showed the highest cell counts.

Although optimal hardness conditions have been found for cell adhesion and proliferation, research remains to be done to better understand cell behavior on gels. Future experiments include fibroblast migration on the hydrogels as well as similar studies for endothelial cells to study blood vessel formation, another process critical to tissue regeneration. Overall, the results of this study show promise in modeling wound healing in diabetics and developing better gels for cell delivery to wound sites.

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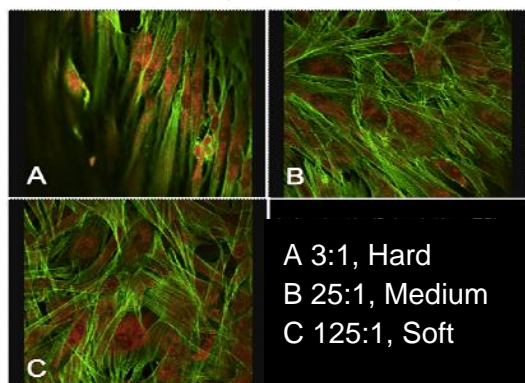
The Effect of Various Concentrations of Glucose and Microbial Transglutaminase on the Mechanical Properties and Biomineralization of Cross Linked Gelatin Hydrogels, and Hydrogel Support of Dermal Fibroblast Growth

Sachi Patil Half Hollow Hills HS East, Melville NY, Emma Zawacki, Smithtown HS East, St James NY, Sneha Subramaniam, Columbia University, Clement Marmorat, Stony Brook University, Ecole Polytechnique; Dr. Marcia Simon, Oral Biology and Pathology, Stony Brook University, Yan Xu, Divya Bhatnagar, and Dr. Miriam Rafailovich, Materials Science and Engineering, Stony Brook University

Fibroblasts, which play a crucial role in wound healing, comprise the extracellular matrix in animals and are widely studied in the tissue engineering field. Aside from the cells themselves, scaffolds are imperative to cell growth and differentiation. For the development of specific tissues, scaffolds must contain a physical and chemical nature that induces growth and differentiation of cells¹. In this study, gelatin cross-linked with microbial transglutaminase enzyme (mTG) was used as the scaffold for cell delivery. Gelatin is an ideal scaffold because of its affordability and biocompatibility². Further, gelatin can easily be controlled in the laboratory due to the reversible nature of the creation of physical bonds between gelatin molecules. In this experiment, glucose was also added as a component of the hydrogel in order to investigate its effect on the physical properties of the substrate. Thus, the goal of our project is to study the substrate mechanics of mTG cross-linked gelatin in terms of fibroblasts.

A multitude of techniques were used in order to determine the substrate mechanics of the cross-linked hydrogel at the various levels of glucose as well as enzyme concentration. Using the Bohlin Rheometer, elastic modulus (G') was noted as a function of the various incubation times of the hydrogel in order to find the ideal incubation period with and without glucose. The data from the rheometer clearly indicated that the most time would be saved if samples were kept incubated for 0-12 hours—once the gels were incubated for 15 hours there was no change in the elastic modulus. The effects of temperature were also tested using similar methods; gels were prepared under various temperatures between 37-57°C and no differences in hardness were observed. Finally, it was discovered that the amount of mTG and glucose added to the gels directly impacted the hardness; for example, the higher the ratio of mTG to gelatin the harder the gel.

With the various effects of time, temperature, mTG, and glucose on the hardness of hydrogels, cells were cultured on the gels to see the effects of the different hardness. Using confocal microscopy, fibroblasts were plated on gels of different mTG to gelatin ratios were observed. It was found that the fibroblasts plated on the hard ratio (3:1, gelatin: mTG) reacted better—their actin was more stretched and elongated (Fig 1). Currently, the effects of hardness on cell migration are also being studied. Dermal fibroblasts are being added to gels hardened by different ratios of mTG and their behavior on the different matrices will be observed.



As previously stated, glucose also has an effect on the hardness of hydrogels. A Leo 1550 Scanning Electron Microscope (SEM) was used to observe the effects of various glucose amounts, 0 mg to 5mg, on the effect of calcium phosphate mineralization in the absence of cells. Images showed that gels with the medium amounts of glucose, 2 mg, had more homogenous biomineralization—deposits were not scattered in random locations on the gel.

Further experiments will include a study on cell migration and differential scanning calorimetry (DSC) to determine the degree of cross-linking. Additionally, these gels

hold promise for the use of wound healing and cell delivery.

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Using Micropatterned Thin-Films on Silicon Substrates as Carriers for Hydrogel Drug Delivery: A Study of Micropattern Structure on Drug Effusion

Rahul Bachal¹, Avigael Sosnowik², Monika Batra³, J. Jerome³, M. H. Rafailovich³, J. Sokolov³

¹Inglemoor High School, 15500 Simonds Rd NE, Kenmore, Washington 98028, United States

²Stella K. Abraham High School for Girls, 291 Meadowview Avenue, Hewlett Bay Park, New York 11557, United States

³Chemical and Molecular Engineering Program, Department of Material Science and Engineering, Stony Brook University, Stony Brook, New York 11794-2275, United States

Drug delivery is one of the most widely researched topics in the medical field today. Due to limitations in controlled drug release with current systems¹, new alternatives for drug delivery systems are being researched extensively. This experiment focuses on a novel drug delivery system based on spuncast polymer blends, namely polystyrene with poly(methyl methacrylate), polystyrene with polycaprolactone, and poly(methyl methacrylate) with polycaprolactone. Polymers are usually immiscible and phase-separate when spuncast together, forming natural micropatterns. A hydrogel containing a drug can then be spread over these micropatterns, filling the depressions. The drug, salicylic acid in this case, will effuse out of the hydrogel at rates dependent on the structure of the micropatterns.

Ratios of 1:3, 1:1, and 3:1 of polystyrene to poly(methyl methacrylate), respectively, were blended and spuncast. Additionally, polystyrene and poly(methyl methacrylate) were each spuncast with polycaprolactone in concentrations of 2%, 4%, 6%, and 8% by mass. Polycaprolactone was selected due to its confirmed degradability on contact with *Candida-Antarctica* Lipase B enzyme as well as its crystalline structure. In accordance with previous studies², micropatterns were found in the polystyrene and poly(methyl methacrylate) blends (Figure 1), and micropatterns were found in PCL blends with PCL concentrations of 2% and 4% (Figure 2). In addition, samples of each ratio and blend were annealed, and micropatterns were more defined for these samples. The process of annealing removed toluene from the samples and induced further phase separation.

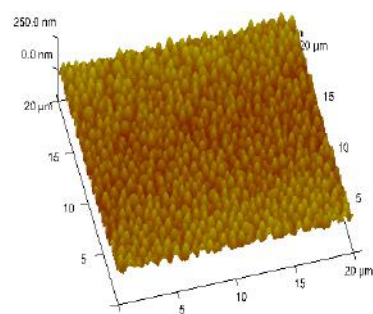


Figure 2: 2% PCL PS Annealed 3D AFM Image

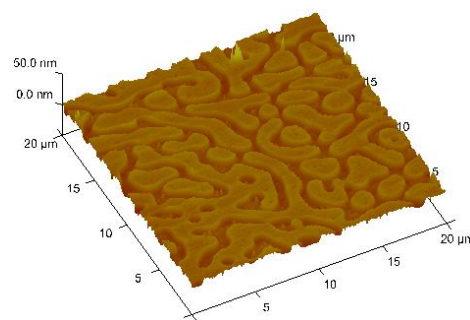


Figure 1: 25% PS 75% PMMA 3D AFM Image

Next, solutions of 0.05%, 0.5%, 1%, and 2% salicylic acid dissolved in butylene glycol were dissolved in a stock solution of 25% pluronic f127 hydrogel dissolved in deionized water. Thin-films were dipped in the hydrogel solutions and left to heat to room temperature, at which the pluronic f127 hydrogel phase transformed from liquid to gel. A thin layer of water was spread over the hydrogel and after various time intervals, was pipetted off and analyzed with Ultraviolet-Visible Spectroscopy to determine the amount of salicylic acid present in the water. Results remain to be collected, but it is expected that the rate of drug effusion will be directly proportional to the increased size and ubiquity of depressions. Future steps also include inkjet printing on PCL thin-films, using CalB dissolved in DPBS as ink, to create artificial patterns that will be tested identically to the blend-induced micropatterns for hydrogel drug effusion.

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Cell Differentiation, Dynamics and Mechanics

Chairs:

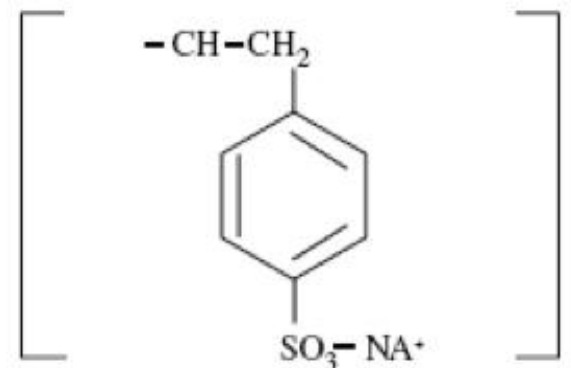
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University, New York

Alanna Foerth, Messiah
College, PA

Graduate Students:

Simon Chang

SiSi Qin



The Effect of Graphene and Different Concentrations of Iron Oxide on the Proliferation and Differentiation of Dental Pulp Stem Cells

Eda Algur and Manasvi Varshney, Smithtown High School West, Smithtown, NY

Dr. Vladimir Jurukovski, Suffolk County Community College, Brentwood, NY

Chung Chueh Chang, Dr. Miriam Rafailovich, Dept. of Materials Sciences, Stony Brook University, Stony Brook, NY

Known for their pluripotency, dental pulp stem cells (DPSCs) are post-natal stem cells affiliated with odontogenic and osteogenic differentiation. They can also differentiate into adipocytes and neural-like cells. The nanoparticles being used to treat the DPSCs in this experiment are graphene and iron oxide. Graphene is a biocompatible, two-dimensional, one-atom thick substance made up of carbon. Studies have shown graphene to cause osteogenic differentiation in stem cells.¹ Superparamagnetic iron oxide nanoparticles (SPIONs), are the nanoparticles of choice in magnetic resonance imaging. SPIONs have been proven to act as peroxidases in cells by decreasing intracellular hydrogen peroxide levels.^{2,3} Although there is some data regarding the use of graphene and iron oxide in cellular applications, this field is still expanding; thus, the biological properties and cytotoxicity of graphene and iron oxide have not been researched extensively. In addition, we will pair these nanoparticles with dexamethasone as it is known to induce osteogenesis and will thus be used as a control for osteogenic differentiation.

The purpose of this experiment is to test the effects of graphene and iron oxide on both the proliferation and differentiation of DPSCs and how these effects may change when the nanoparticles are combined with dexamethasone (Dex). The experiment consists of six parts: assess proliferation using the growth curve, assess biomineralization and differentiation using RT-PCR to determine the type of differentiation (osteogenic or odontogenic) and ESM-EDAX, TEM analysis to determine if nanoparticle uptake occurred, and confocal microscopy imaging to study cell morphology. Five treatments were designed for the cells: a control treatment with regular cell media, a treatment with 0.1 mg/mL of graphene, and three treatments with 0.05 mg/mL, 0.1 mg/mL, and 0.5 mg/mL of iron oxide suspended in regular media. For assessment of proliferation cells were counted on days 2, 5, and 7 after the start of treatments, each in absence or presence of Dex. In absence of Dex (Fig. 1), the cells treated with graphene showed the most proliferation, exceeding the control group. At 0.05 and 0.1 mg/ml iron oxide concentrations the cells showed proliferation similar to that of the control, while the concentration of 0.5 mg/ml iron oxide significantly decreased cell proliferation. In presence of Dex (Fig. 2), all samples treated with nanoparticles showed decreased proliferation compared to both control groups (with or without Dex).

We intend to analyze the staining, EDAX, and RT-PCR samples on days 14 and 21, and the confocal microscopy and TEM samples on day 7.

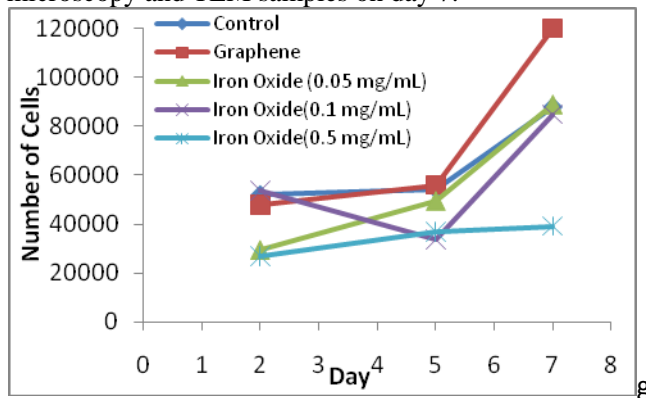


Figure 1: Growth Curve of samples treated with the nanoparticles only

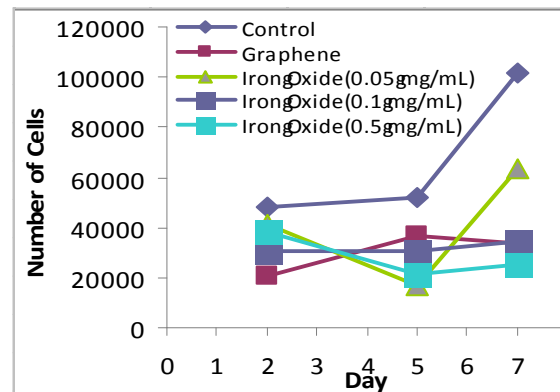


Figure 2: Growth Curve of samples treated with the nanoparticles and dexamethasone

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² Dong-Ming Huang, Jong-Kai Hsiao, Ying-Chun Chen, Li-Ying Chien, Ming Yao, Yin-Kai Chen, Bor-Sheng Ko, Szu-Chun Hsu, Lin-Ai Tai, Hui-Ying Cheng, Shih-Wei Wang, Chung-Shi Yang, Yao-Chang Chen. (2009) The promotion of human mesenchymal stem cell proliferation by superparamagnetic iron oxide nanoparticles. Biomaterials 30, 3645-3651

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Analyzing the Role of ROCK/rhoA Kinases in the Differentiation of Dental Pulp Stem Cells

Evan Chernack, South Side High School, Rockville Centre, NY, **Aneri Kinariwalla**, Sayville High School, Sayville, NY

Holly Flores, Stony Brook University, Stony Brook, NY

Aneel Bherwani, Dept. of Oral Biology and Pathology, Stony Brook University, Stony Brook, NY

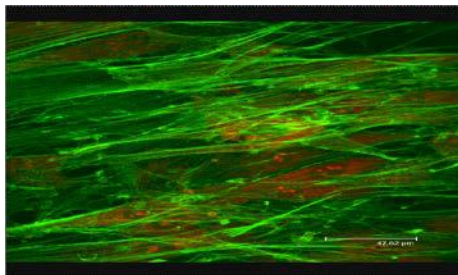
Dr. ChungChueh Chang, **Dr. Miriam Rafailovich**, Dept. of Material Sciences, Stony Brook University, Stony Brook, NY

The dental pulp of adult human teeth is a high-yielding source of dental pulp stem cells (DPSCs). Displaying multipotency, DPSCs are predominantly well suited to differentiation into osteoblasts¹. As a result, DPSCs hold immense promise as a method for osteoregenerative therapies.

Several factors are involved in the process of DPSC differentiation, such as chemical signals, environmental conditions in the extracellular matrix, and various protein kinases¹. Kinases serve as crucial components of the signaling pathways responsible for differentiation by transferring phosphates. Among these kinases are those in the ROCK pathways. These proteins can be inhibited by the chemical compound Y-27632 dihydrochloride (Fig.1), a specific inhibitor of Rho-Associated kinases². The cells were treated with this y-factor in order to analyze the function of the abovementioned kinases in the differentiation of DPSCs.

Polybutadiene, a biocompatible polymer, was spun cast onto silicon wafers to create films of varying thickness. DPSCs were plated onto the wafers using standard procedure, and half of the samples were treated with the Y-27632. Several tests were run at different time-points in order to collect data on the rate of DPSC differentiation. Images were obtained through confocal microscopy at days 4,6, and 9. The data indicated that the cells treated with Y-27632 formed actin fibers less organized than the control. Such results suggest that ROCK/rhoA kinases are involved in DPSC-ECM interaction. This appeared to show that the absence of ROCK/rhoA kinases results in a disruption of actin fiber organization.

Future research will include the use of atomic force microscopy (AFM) to analyze topography of the extracellular matrix (ECM). Additionally, tests with scanning electron microscopy and RT-PCR will be performed so as to visualize the cells and analyze the gene expression of differentiation-related markers,



(Figure 1.) Day 9 confocal image of DPSCs treated with both dexamethasone and Y-27632 dihydrochloride

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Differentiation of dental pulp stem cells on electrospun poly(4-vinylpyridine) and poly(methyl methacrylate)

Justin Koritzinsky Valt Whitman High School Bethesda, MD

Aneel Bherwani & Marcia Simon School of Dental Medicine, Stony Brook University Stony Brook, NY

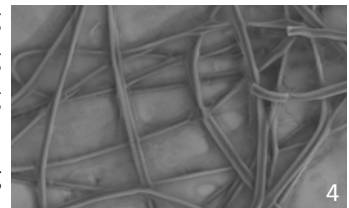
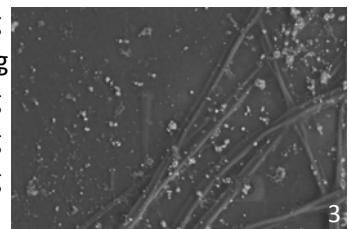
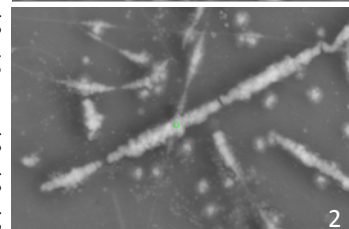
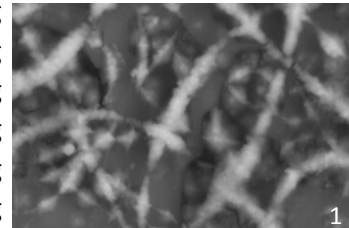
Miriam Rafailovich Department of Materials Science, Stony Brook University Stony Brook, NY

Dental pulp stem cells are pluripotent stem cells with the potential to differentiate into adipocytes, neurons, odontoblasts and osteoblastsⁱ. Environmental factors such as the substrate the cells are cultured on and the nutrients they are provided induce DPSCs' differentiation. DPSCs' ability to undergo osteogenesis suggests an application in tooth repair and other bone regenerative therapiesⁱⁱ. There is not, however, a biocompatible method for controlling the resulting biomineralization. DPSCs were cultured on P4VP and PMMA substrates to test their viability for use in bone generation.

Both P4VP and PMMA thin films, microfibers and nanofibers were spun-cast onto silicon wafers. DPSCs were then plated onto the silicon wafers, and half of them were induced with dexamethazone, a steroid that is known to induce differentiation into osteoblasts. The cells were cultured for 5 days and samples were taken on days 1, 7, 14, 21 and 25. Images of a few of the days 28 samples were taken with scanning electron microscopy. Figs. 1 and 2 show biomineralization throughout both P4VP and PMMA induced samples. While the non-induced PMMA samples

such as Fig. 4 showed no biomineralization, the non-induced P4VP samples such as Fig. 2 showed significant biomineralization along both microfibers and nanofibers. The biomineralization is the result of the DPSCs differentiating into osteoblasts, which produce bone tissue.

The biomineralization of non-induced P4VP fibers is not noteworthy as the number of side effects make dexamethazone incompatible with the human body. These results are similarly significant because the biomineralization occurred only along the fibers, which means that it can be controlled. Further studies involving the RNA extracted from the samples performing real-time polymerase chain reaction will be used to confirm that the differentiation of DPSCs into osteoblasts caused the biomineralization along the non-induced P4VP microfibers and nanofibers. Additional experiments may include the introduction of P4VP microfibers and nanofibers into dental implants in both mice and dogs.



From top: fig. 1 shows biomineralization throughout induced P4VP nanofibers, fig. 2 shows biomineralization along non-induced P4VP nanofibers, fig. 3 shows biomineralization throughout induced PMMA microfibers, fig. 4 shows no biomineralization throughout non-induced PMMA microfibers)

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The Effect of Various Polymers on the Differentiation and Proliferation of Mice Embryonic Stem Cells

You Jeong Park, Half Hollow Hills High School West, Dix Hills, NY; Kevin Liu, Interlake High School, Bellevue, WA; Benjamin Lei, Arlington High School, LaGrangeville, NY

Miriam Rafailovich, Ph. D., Department of Material Sciences and Engineering, Stony Brook University
 Yupo Ma, M.D., Ph. D., Health Sciences Center, Stony Brook University
 Marcia Simon, M.A., Ph. D., School of Dental Medicine, Stony Brook University
 Jerell Aguila, Ph. D., Health Sciences Center, Stony Brook University
 Wenbin Liao, Ph. D., Health Sciences Center, Stony Brook University
 Sisi Qin, Department of Material Sciences and Engineering, Stony Brook University
 Chungchueh Chang, Department of Material Sciences and Engineering, Stony Brook University
 Derya Karatas, Stony Brook University
 Alanna Foerth, Messiah College

Human embryonic stem cells (hESCs) are currently being used in many fields of medicine to test drugs, perform cell-based therapies, and provide a better understanding of human development. With their pluripotency, or ability to differentiate into the three primary germ layers, these cells can differentiate into all cell types found in the body. This makes them favorable to multipotent somatic stem cells, which can only differentiate into a limited spectrum of cell bodies. However, research in this field has been limited due to ethical arguments as these experiments require destroying human embryos.

In our experiment, we worked with mice embryonic stem cells (mESCs), ethical safe models often used to further understand the development of hESCs, to investigate certain conditions in which these cells could proliferate without differentiating. Undifferentiated cells have the ability to replicate an infinite number of times; however, once they differentiate, these cells are limited with a set number of divisions they can go through. Because traditional culture conditions depend on xenogeneic medium to maintain the cells' stemness, utilizing known polymers in an environment without any additives would bypass a significant obstacle for research and regenerative medicine applications. In addition, eliminating the need for gelatin and feeder cells, which usually accompany the stem cells, will provide a more cost-efficient method in the culturing process.

In our study, we observed the effects of the polymers poly(methyl methacrylate) (PMMA), poly(4-vinylpyridine) (P4VP), poly(styrene-co-4-styrene) (SPS), and two different thicknesses of polybutadiene (PB) (200 Å and 2000 Å) on the differentiation and proliferation of mESCs. These polymers were dissolved in solutions of toluene and N,N-dimethylformamide, and spun-casted on micro-glass coverslips and silicon wafers cut with a Miller's index of [1-0-0]. These polymers were plated in a 24-well plate under three different groups for experimentation: gelatin with feeder cells and mESCs, feeder cells and mESC without gelatin, and mESCs without gelatin or feeder cells.

From our confocal images, we were able to compare the mESCs' morphology within each group. In the control group (cells plated on feeder cells with gelatin), we were able to confirm that the cells remained undifferentiated because of their round colonies with distinct edges. After comparing the control group to the cells without any additives, SPS seems to be the best polymer to retain stemness in the cells. Although the cells without additives on both PBs were able to grow, many of the colonies were composed of cells that were already differentiated (signified by the indistinguishable edges of the colonies). On P4VP, both the feeder cells and the mESCs had trouble adhering to the surface of the polymer. Similarly, the mESCs were unable to adhere efficiently to the PMMA polymer surface. The addition

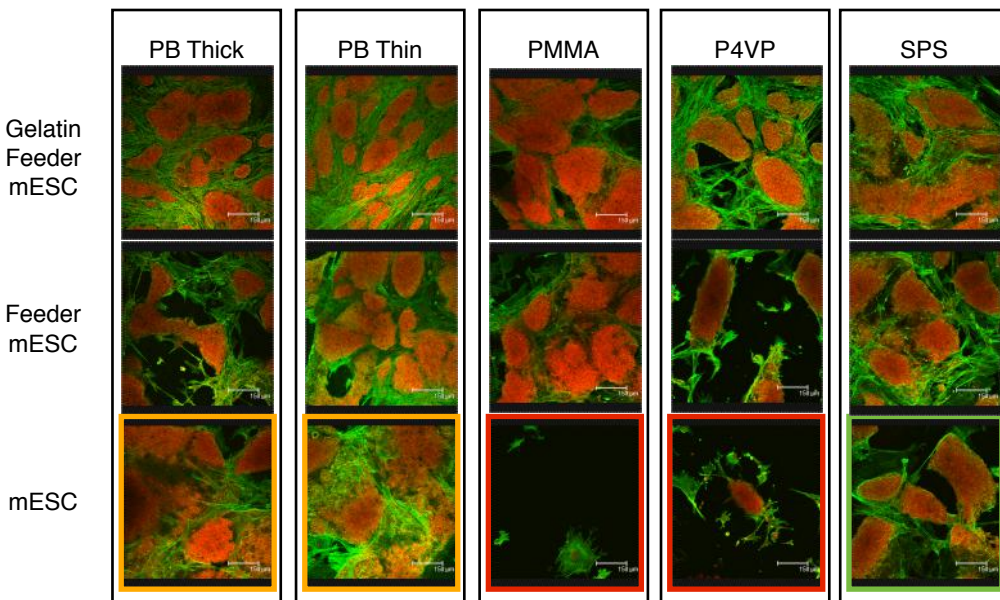


Figure 1. Confocal images of polymers and mESCs. Nucleuses of the mESCs are dyed red and the actin fibers dyed in green.

of gelatin to the polymer surfaces, although it effected the growth of feeder cells, seems to have little effect on preventing the mESCs from differentiating.

In the future, in order to obtain a more accurate measure of the polymers' ability to retain stemness in mESC growth, we will be passaging and observing the mESCs every two days. After two passages, differentiation of the mESCs will become more evident. The most efficient polymer will be the one that retains the most undifferentiated mESCs. In addition, we will carry out this experiment on induced pluripotent stem cells (iPSCs).

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The Effects of Polybutadiene, Poly(methyl methacrylate), Sulfonated Polystyrene, and Poly(4-vinylpyridine) on the Proliferation and Differentiation of Hematopoietic Stem Cells

Shivram Chandramouli, *Munster High School, Munster, IN,*

Alanna Foerth, *Messiah College, Mechanicsburg, PA*

Derya Karatas, *Stony Brook University, Stony Brook, NY*

Jerrell Aguila, Wenbin Liao, Yupu Ma, *Stony Brook University Medical Center, Stony Brook, NY*

Marcia Simon, *Stony Brook University School of Dental Medicine, Stony Brook, NY*

ChungChueh Chang, Sisi Qin, Miriam Rafailovich, *Stony Brook University Department of Materials Science, Stony Brook, NY*

Hematopoietic stem cells (HSCs) have become the most studied of all stem cells as a result of their widespread clinical use. HSCs, obtained from bone marrow, are a rare source of the specialized cells present in the human hematopoietic system (blood). These specialized cells include erythrocytes, megakaryocytes, B-lymphocytes, and T-lymphocytes. The human body replenishes about one hundred billion of these specialized blood cells each day, depending directly on HSCs for this action.¹ HSCs are used in clinics as part of stem cell therapies to treat blood-related diseases. Researchers have not yet been able to find a method for ex-vivo expansion of HSCs, which would greatly aid the availability of HSCs.²

In this experiment, the polymers polybutadiene (PB), poly(methylmethacrylate) (PMMA), sulfonated polystyrene (SPS), and poly(4-vinylpyridine) (P4VP) were tested to see if they could act as surfaces that promote the proliferation of undifferentiated human HSCs. In doing so, we were able to analyze the unique effects each polymer had on the growth of these cells.

First, each of the polymers was spun-cast onto appropriate wafers. The samples were annealed overnight. The next day, approximately 500,000 donated hematopoietic stem cells were divided into 15 wells, and assuming some error during the transfer process, we can say that each well began with about 25,000 cells. Stem Span media was added to each well in addition to 3 cytokines. The cells were counted on days 4 and 6 by means of hemocytometers. Furthermore, the cells were marked with CD34 and CD38 on day 6, and flow cytometry was performed on them.

From the cell counting data, we were able to deduce that the polymers indeed affected the growth rates of the HSCs. Though the cells were able to survive on all the polymers, at the end of day 6, PMMA yielded the best results (Fig. 1). Furthermore, from the flow cytometry data, we noted that the cells grown on each polymer had a different mix of differentiated and undifferentiated cells, with PB Thin yielding the best results.

Future experiments will focus upon testing the effects of these polymers on other stem cells. Results from such experiments will be juxtaposed with those of this experiment to observe how each polymer performs in comparison to others.

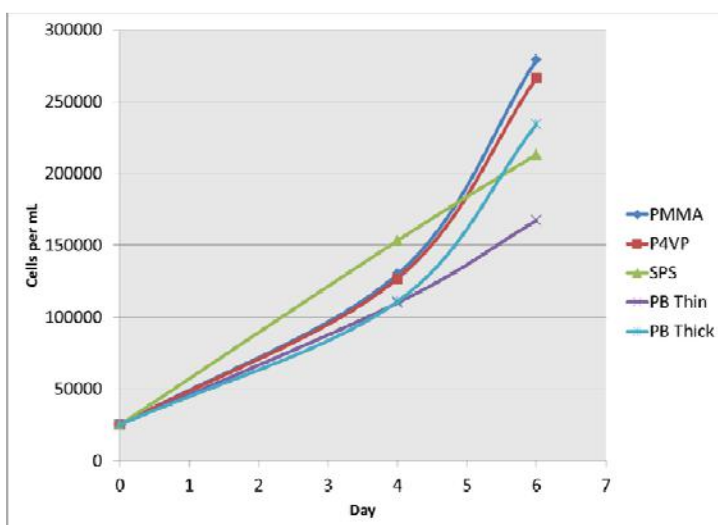


Figure 1. All the polymers allowed the hematopoietic stem cells to survive, but at the end of day 6, PMMA yielded the best results.

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A Study of the Growth and Differentiation of Dental Pulp Stem cells with and without Static Magnetic Fields

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Austing Wild, *South Side High School, Rockville Centre, NY*

Sanchita Singal, *Brown University, Providence, RI*

Vladimir Gurukovski, *Suffolk Community College, Seldon, NY*

Chung Chueh Chang and Miriam Rafailovich, *Stony Brook University, Stony Brook, NY*

Dental Pulp Stem Cells [DPSCs] can differentiate into osteoblastic or odontoblastic lineages and show great promise in regenerative medicine. Research has shown that the extracellular matrix [ECM] is critical in regulating cell function, and manipulating the ECM can affect cell differentiation. A sulfonated polystyrene [SPS] surface (Figure 1) stimulates ECM fibrillogenesis making it ideal for the analysis of the actual fibers of the ECM before the cells become confluent. A magnetic field can be used to test how magnets affect the ECM and cells with or without dexamethasone; a chemical steroid used to induce osteoblastic differentiation.

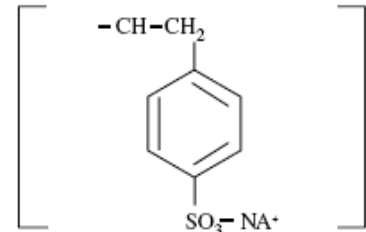


Figure 1 Sulfonated Polystyrene

The purpose of this experiment was to continue the research conducted last year on the differentiation of dental pulp stem cells on SPS in the presence or absence of static magnetic fields. Magnetic apparatuses were created to hold one 24-well plate in between two sets of magnets. Also, a thin film of the biocompatible polymer sulfonated polystyrene was spun cast onto the wafers in order to induce spontaneous fibrillogenesis that would allow the ECM to be analyzed.

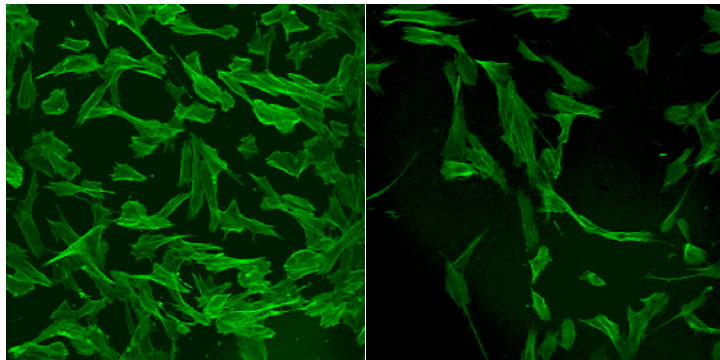


Figure 2, Confocal Day 3 Images. The image on the right are cells grow in a magnetic field and on the ones on the left are cells only grow on SPS without magnets

All sets of samples were plated on the SPS and all of those samples were separated into four parts; samples with both magnetic fields and dexamethasone, samples with only one or the other, and a control, which consists of cells plated only on SPS. Confocal microscopy images were taken on days 3, 5, and 7 and it can be seen that there is more growth with the cells that are plated in a magnetic field than they are without. (Figure 2)

These tests will include scanning and modulus tests using Atomic Force Microscopy to analyze the width and height of the fibers of the ECM, as well as

the stress over strain of the cells, immunohistochemistry to analyze the presence of osteocalcin [OCN] within the samples, Reverse Transcription Polymerase Chain Reaction [RT-PCR] to analyze gene expression, and Scanning Electron Microscopy/EDAX [SEM] in order to image and analyze the deposits of the bio-mineralization of the cells onto the silicon wafer.

g g

1) Gronthos, S., M. Mankani, B. Ibrahim, P. Gehron-Robey, and S. Shi. "Postnatal Human Dental Pulp Stem Cells (DPSCs) in Vitro and in Vivo." *PNAS* 97.25 (2000): 13625-13629. Web. 25 June 2012. <<http://www.pnas.org/content/97/25/13625.full.pdfhtml>> 2) El-Amin, S. F., H. G. Khan, B. P. P. Mitchell, R. T. S. G. Laurencin. "Extracellular Matrix Production by Human Osteoblasts Cultured on Biodegradable Polymers Applicable for Tissue Engineering." *Biomaterials* 4.7 (2003): 213-221.

The Effect of PMMA Substrates on Keratinocyte Migration

Christine Chang, Palo Alto High School, Palo Alto, CA

Amy Wang, St. Anthony's High School, South Huntington, NY

Sisi Qin, Miriam Rafailovich, Dept. of Material Sciences, Stony Brook University,
Stony Brook, NY

Wound healing is a complex, natural response to tissue injury. While many different cells are involved in the process, two of the main cell types are dermal fibroblasts of the dermal layer of skin and human keratinocytes of the epidermal layer of skin. Research has indicated that fibroblasts recognize and adhere to electrospun fiber scaffolds and move in a "crawling" manner.¹ Keratinocyte migration and response to different polymer substrates is unknown. The purpose of this experiment is to analyze the effects of different PMMA substrates on keratinocyte migration, more specifically on substrates of PMMA thin film, 8 μm microfiber, and 700 nm nanofiber.

First, a solution of PMMA was spuncast onto glass slip covers, then 1D parallel fibers were electrospun onto the spuncast glass slip covers, and then were annealed. Keratinocytes were cultured and then plated onto aforementioned glass slip covers. Pictures were taken every fifteen minutes for an hour of each sample for four days. The images were processed using Matlab to measure cell migration speed and confocal microscope images were also utilized to identify focal adhesion points and cell morphology.

Preliminary results indicate that keratinocyte migration differs significantly from that of fibroblast migration. Fibroblasts migrate by "crawling" along fibers, while keratinocytes move by rolling. Results also show that the migration speed of keratinocytes is faster than that of fibroblasts; additionally, while fibroblasts tend to adhere to the fiber scaffolds, keratinocytes recognize the nanofiber scaffold but ignore the microfiber scaffold.

This information can be used in further wound healing experiments. In further research, keratinocytes and fibroblasts can be co-cultured for additional examination. If the respective tendencies of keratinocytes and fibroblasts do continue to prove valid, the two may then be potentially separated for wound healing purposes.

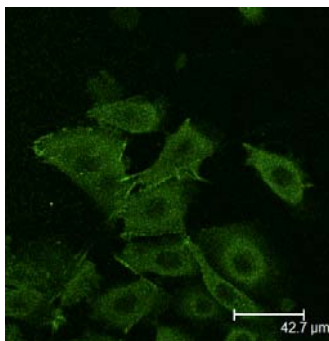


Figure 1: Confocal image of keratinocytes en mass, on nanometer fibers

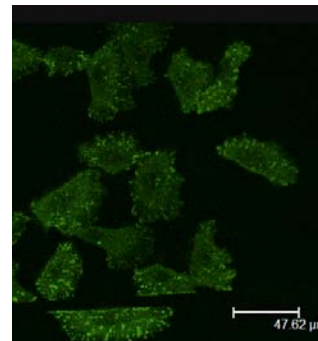


Figure 2: Confocal image of keratinocytes en mass, on micrometer fibers

¹Krawczyk, Walter S. "A Pattern Of Epidermal Cell Migration During Wound Healing." *The Journal of Cell Biology* 49.2 (1971): 247-63.

Hydrogen Fuel Cells

Chairs:

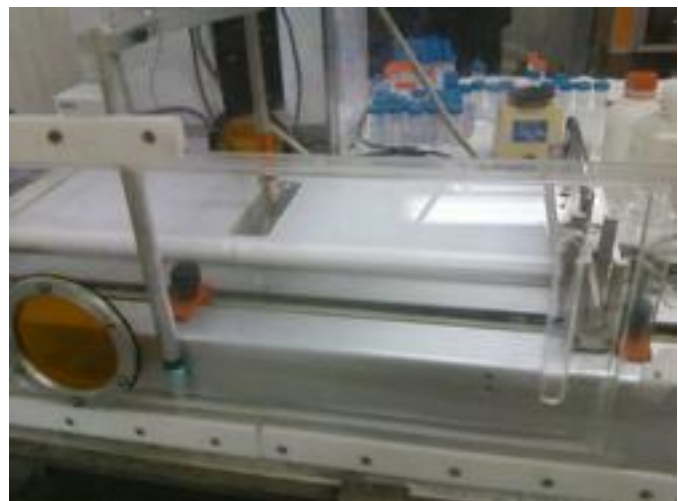
Aaron Akhavan, Yeshiva
University, New York

Ilan Gold, University of
Maryland

Graduate Students:

Cheng Pan

Hongfei Li



A Comparative Study on the Structural effects of Noble Metal Nanowires and Nanoparticles as Novel Catalysts for PEM Fuel Cells

Kevin Chan, Stevenson School, Pebble Beach, California
 Victoria Petrova, South High School, Torrance, California

Cheng Pan, Department of Materials Science and Engineering, Stony Brook University
 Dr. Miriam Rafailovich, Department of Materials Science and Engineering, Stony Brook University

Hydrogen fuel cells are currently being considered as a main alternative to traditional fuels such as petroleum. Fuel cells are already being tested and used in cars for everyday use as well as public transportation in buses and taxis.¹ Obstacles that prevent hydrogen from becoming the main source of energy include the low efficiency of a single fuel cell and the expensive platinum on the electrodes. Recent research has focused on adding various metals as co-catalysts to improve the efficiency.

The goal of this project is to see how varying the morphology of different catalysts affects the power output of a hydrogen fuel cell; in particular, to compare the performance of nanoparticles versus nanowires. The hypothesis is that nanowires would make better catalysts since they have a higher surface area to volume ratio. Furthermore, the structural changes of metal nanowires will increase the availability of contact sites to catalyze the oxygen reduction reaction.² Gold and palladium nanoparticles and nanowires were synthesized, then deposited them onto Nafion® membranes with the Langmuir-Blodgett trough and tested in a single stack hydrogen fuel cell.

The gold and palladium nanoparticles were synthesized by the two-phased Brust method, which produced hydrophobic nanoparticles with Thiols attached. The palladium nanowires were synthesized through a self-assembly method using a stabilizer and reducing agent.² Gold nanowires were synthesized by performing a reaction with a stabilizer and a reducer at high temperature.³

To coat the Nafion® membrane with nanoparticles or nanowires, each was dissolved in a solvent, which was then dispersed onto the surface of the water within the Langmuir-Blodgett Trough. *Figure 2* Isotherms were used to locate an ideal target pressure to confirm that a monolayer was deposited onto the Nafion® membrane. The nanoparticles or nanowires were then tested in an h-tec fuel cell kit with platinum coated electrodes and hydrogen gas within the cell. The voltage and current were recorded as the resistance was varied.

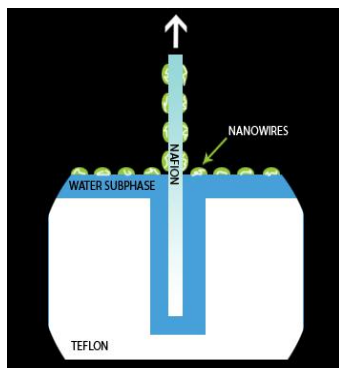


Figure 2: The process of coating a monolayer of nanowires on a Nafion membrane using the Langmuir-Blodgett Trough.

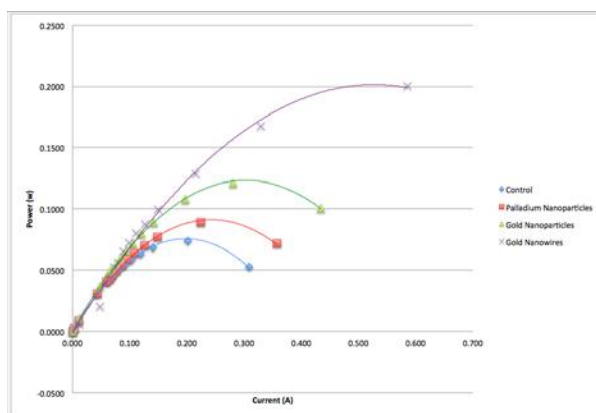


Figure 1: Power vs. Current graph comparing the power outputs of fuel cell test of Nafion® membranes with various coatings.#

Data was retrieved from a control fuel cell test that consisted of a single stack fuel cell operating with a clean Nafion® membrane. In

addition, control tests were performed with Nafion® membranes coated with gold and palladium nanoparticles. Nafion® membranes coated with metal nanowires were also tested in a single stack hydrogen fuel cell under the same conditions of the control. The experimental data obtained was compared to control data to determine the overall change in efficiency of nanowires versus nanoparticles within a PEM fuel cell. *Figure 1* Preliminary tests show that nanowires are promising in increasing the efficiency of the hydrogen fuel cell.

TEM will be performed on the synthesized metal nanowires to determine average diameter, average length, and degree of aggregation. Cyclic voltammetry tests will also need to be performed on the nanowires to determine the electrochemical and catalytic properties of the gold and palladium that were synthesized.

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¹ Thomas, S., & Zalbowitz, M. (2006). *Fuel Cells: Green Power*. Los Alamos National Laboratory.#

² Jiajun Wang, Yougui Chen, Hao Liu, Ruying Li, Xueliang Sun “Synthesis of Pd nanowire networks by a simple template-free and surfactant-free method and their application in formic acid electrooxidation” Elsevier. Vol. 12, 2010, page 219#

³ Yueming Tan,a Jingmin Fan,a Guangxu Chen,a Nanfeng Zheng and Qingji Xie “Au/Pt and Au/Pt3Ni nanowires as self-supported electrocatalysts with high activity and durability for oxygen reduction reaction” The Royal Society of Chemistry, 2011 page S1#

Investigating Various Methods of Incorporating Graphene Oxide into PEM Fuel Cell System

Andrew Chen, Dougherty Valley High School, San Ramon, CA

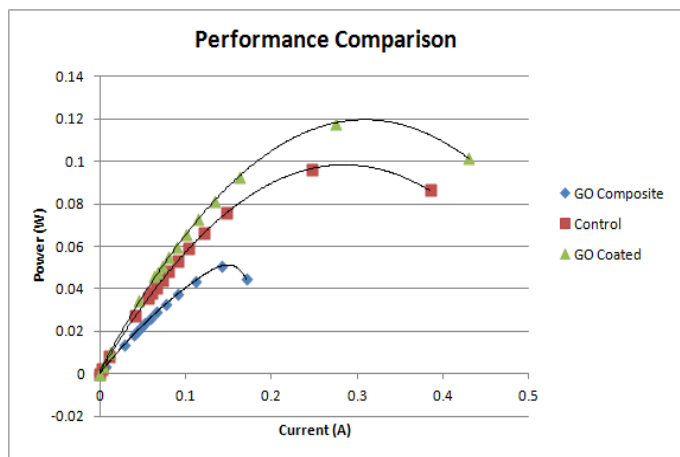
Justin Chiang, Saratoga High School, Saratoga, CA

Holly Flores, Cheng Pan & Miriam Rafailovich, Stony Brook University, Stony Brook, NY

World oil demand is projected to grow 50 percent by 2025. At the same time, due to the continued exhaustion of the world's conventional oil reserves, crude oil production is expected to enter terminal decline by the end of the decade.¹ Some reports even suggest that global production of petroleum may already have peaked as early as 2006. For economic reasons, the widening of the supply-demand gap is certainly a cause for worry. Needless to say, tackling a problem of such global magnitude will necessarily require rapid and aggressive penetration of alternative energy technologies.

As such, the polymer electrolyte membrane (PEM) fuel cell, a technology which runs on hydrogen gas, is widely recognized as a leading candidate for future power generating devices, especially in the automotive and in portable electronic applications.² Not only does the PEM fuel cell run on hydrogen gas, the most abundant element in the universe, it also can achieve efficiencies up to 60%, while the typical gasoline engine is less than 20% efficient in producing power.³ Moreover, the PEM fuel cell is able to operate quietly, at low temperatures, and with few moving parts.

Despite its many advantages, the PEM fuel cell suffers from a high cost to power output ratio. One of the major contributors to this high cost is Nafion[®], the most commonly used membrane in contemporary fuel cell applications. While Nafion[®] is still considered one of the best materials, we were interested in better understanding the mechanisms of proton transport in order to enhance fuel cell performance.⁶ Since graphene-based materials are currently under intense research due to their exceptional conductive properties, we decided to test and compare the effects of incorporating graphene oxide onto and into the Nafion[®] membrane on power output.



Graphene Oxide was produced using a modified Hummer's Method in which graphite powder is oxidized by strong acids and oxidants such as concentrated sulfuric acid and potassium permanganate. The LB Trough was used to coat the membrane with a uniform monolayer of graphene oxide while a recasting process using liquid nafion was used to create a Graphene Oxide Nafion nanocomposite membrane.

Fuel cell performance measurements show that a uniform GO monolayer at the surface of the Nafion significantly improves power output while the composite membrane decreases performance. Future research includes testing whether other graphene-derivative materials can similar impact fuel cell performance and further characterization of membranes.

Using Silver Nanoparticles and Silver/Copper Nanoalloys on the Nafion Membrane Inside of a Hydrogen PEM Fuel Cell to Increase Efficiency

Michael Sosnick, Benjamin DuBow: HAFTR High School, Cedarhurst NY Cheng Pan, Hongfei Li, Miriam Rafailovich: Department of Material Sciences, Stony Brook University, NY

Now more than ever the necessity of finding a clean, renewable energy source is clear. One new method of using a PEM fuel cell with hydrogen and oxygen is very promising.

A hydrogen PEM fuel cell utilizes the electrons that split from hydrogen when hydrogen gas oxidizes in the presence of a platinum catalyst. The resulting hydrogen nuclei (protons) travel through a proton exchange membrane in the middle of the cell while the electrons are forced to travel through an external circuit, creating an electrical current. The hydrogen ions then combine with oxygen at the cathode to form water, which escapes from the cell as a gas¹.

However, PEM fuel cells have a major problem because they are still more expensive to use than petroleum or coal to fuel transportation, commercial, and residential services. For our project we have experimented with a metal nanoparticle, silver, and one of its nanoalloys, silver/copper, as a way to help increase the efficiency of the fuel cell by assisting the platinum in catalyzing the reactions.

The way we included the nanoparticles into the fuel cell was by coating the Nafion PEM membrane with our particles using an LB trough. We then tested a clean Nafion membrane and our coated membranes and calculated the different amounts of energy produced. After the tests we concluded that the PEM fuel cell with the Nafion membrane coated with Ag/Cu nanoalloys at a pressure of 5 mN/m worked better than at other pressures and almost doubled the power output of the control as shown in Figure 1.

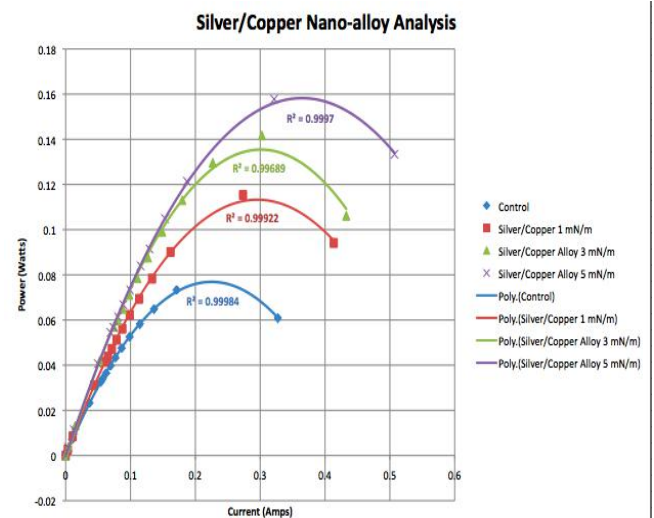


Figure 1: Graph Analysis of Ag/Cu coated

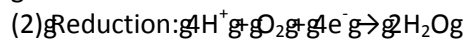
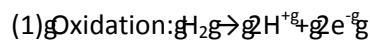
¹ http://www.fueleconomy.gov/feg/fcv_PEM.shtml

The Effect of Gold Nanoparticles on a Hydrogen Polymer Electrolyte Membrane Fuel Cell Stack

Timothy Hart¹, Cheng Pan², Hongfei Li², Mariam Rafailovich²

¹Hauppauge High School, Hauppauge, New York; ²Department of Materials Science & Engineering, Stony Brook University, Stony Brook, New York

As depletion of resources of the world's supply of fossil fuels has opened up the eyes of many nations to alternative energy, it is imperative to avoid an impending energy crisis, and it is necessary to investigate the use of alternative sources of energy. One interesting source of such energy is the Polymer Electrolyte Membrane Fuel Cell, or PEMFC. The PEMFC is a device that uses oxidation (1)-reduction (2) reaction between hydrogen and oxygen gas to produce a useful electrical current. PEMFCs may be connected in series to increase the power output and what is called a fuel cell stack. Fuel cell stacks are the future for clean, energy-efficient vehicles. However, at the present moment, fuel cell stacks must be investigated further in order to incorporate such technology in a real-world scenario.



In this experiment, five Nafion[®] 117 membranes were coated with a monolayer of gold nanoparticles using the Langmuir-Blodgett trough (Figure 1). Coating the electrolyte membrane with a monolayer of gold nanoparticles has already been proven to increase the efficiency of a single fuel cell.¹ However, in this experiment, investigation of the gold nanoparticles in a fuel cell stack will encompass many more variables and would be a realistic situation.

In the future, we will be constructing Membrane Electrode Assemblies (MEAs) to be used in the fuel cell stack. Using the constructed MEAs, we will test the fuel cell stack under various stresses to test its viability in a real-world scenario. We will also be investigating how the gold nanoparticle enhanced Polymer Electrolyte Membrane Fuel Cell Stack compares to a control PEM Fuel Cell stack in terms of durability and efficiency. In addition, we would like to do an analysis of the catalyst layer using TEM imaging.



Figure 1 (left): The Langmuir-Blodgett trough, or LB trough.

¹Cheng Pan, Kenny Kao, Siqi Qing and Mariam Rafailovich, *Gold Nanoparticle Enhancement for Polymer Electrolyte Membrane (PEM) Fuel Cell*

The Construction of a Microbial Fuel Cell Featuring *E. coli* Bacteria to Generate Electricity

Haris Nair, Hastings High School Westchester NY; **Samantha Prashad**, South Side High School Rockville Centre NY; **Dr. Miriam Rafailovich**, Stony Brook University, Department of Engineering NY; **Cheng Pan**, Stony Brook University NY; **Dr. Stephen Walker**, Stony Brook University, Department of Oral Biology and Pathology NY

In the modern world there is a need for alternative sources of energy that are affordable, versatile and environmentally friendly. The most common sources of energy are oil, natural gas and coal. These types of energy sources are non-renewable and give off harmful chemicals into the environment. Since there is a finite amount of these natural resources it makes them very expensive and sometimes difficult to obtain, and microbial fuel cells offer a novel means to help address some of these issues.

²Microbial fuel cells (MFCs) are devices that are able to produce energy through the oxidation of organic

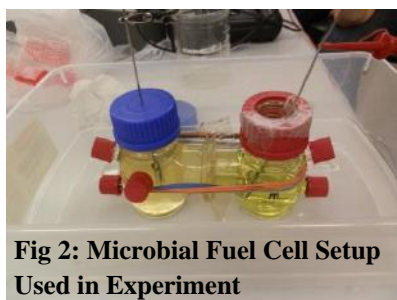


Fig 2: Microbial Fuel Cell Setup Used in Experiment

compounds by microbes. Bacteria go through anaerobic respiration, which is a natural process involving an electron transport chain. The process involved moves the electrons, allowing a current to be produced and electricity to be formed (Fig. 1). An advantage of this process is that it can utilize a variety of carbon sources, making both renewable (wastewater can be used to fuel this process) and versatile. Microbial fuel cells have various components that can be slightly altered in order to improve its efficiency. The microbial fuel cell developed in this study (Fig. 2) implemented *E. coli* as the main bacterial catalyst and a glucose solution as the organic fuel. The other

solutions used were potassium ferricyanide (50mM) and a mediator (methylene blue) to help facilitate the movement of electrons from the anode to the cathode. The electrodes used in this MFC were graphite and they were connected to stainless steel wires. The voltages and currents generated in the cell were recorded and analyzed using two multi-meters connected to the Meterview1.0 software. This program allowed us to see the change in both voltage and current over an extended period of time. The control test was done using just potassium ferricyanide in the cathode chamber and in the anode chamber there were simply the *E. coli* bacteria, LB growth medium and a 20% glucose solution. We used a plain Nafion117 PEM as well.

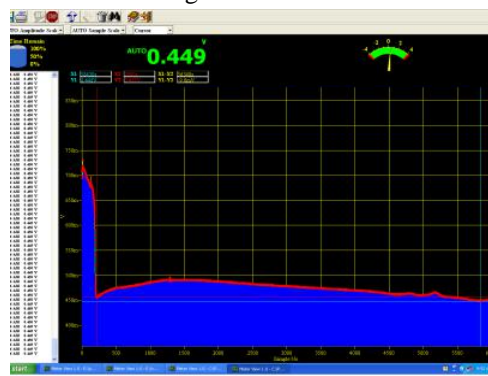


Figure 3 a (left) and b (right): Current ranged from ~4.2 to ~ 4.5 mA and voltage ranged from ~700 to ~ 450 mV

we will continue work by experimenting with variations to our setup, such as nanoparticle blends or the addition of graphene. Using alternative sources of energy is one of the most essential areas in research right now and microbial fuel cells, due to their affordability, renewability, and environmental sustainability, have great potential in this field.

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2. M.Rahimnejad, N.Mokhtarian, G.D. Najafpour, W. Ramli Wan Daud and A.A. Ghoreyshi, *Low Voltage Power Generation in a Biofuel Cell Using Anaerobic Cultures*, World Applied Sciences Journal 6 (11): 1585-1588, (2009)

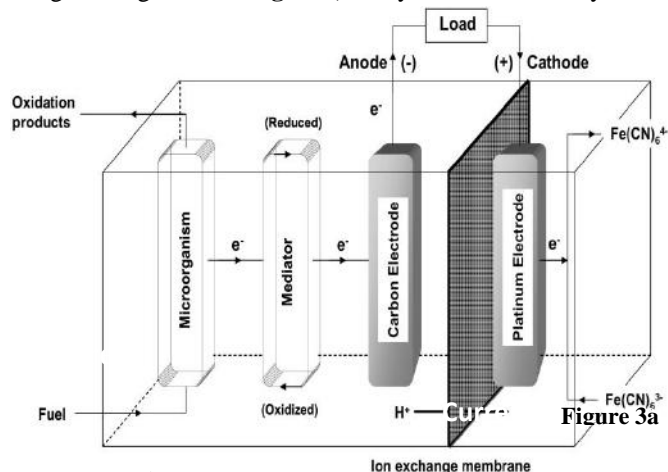


Fig. 1: General schematic setup of a catalyzed microbial fuel cell

Bacteria go through anaerobic respiration, which is a natural process involving an electron transport chain. The process involved moves the electrons, allowing a current to be produced and electricity to be formed (Fig. 1). An advantage of this process is that it can utilize a variety of carbon sources, making both renewable (wastewater can be used to fuel this process) and versatile. Microbial fuel cells have various components that can be slightly altered in order to improve its efficiency. The microbial fuel cell developed in this study (Fig. 2) implemented *E. coli* as the main bacterial catalyst and a glucose solution as the organic fuel. The other

The current ranged from 4.2 to ~ 4.5 mA and the voltage ranged from 700 to ~ 450 mV (fig 3). In order to see how the population density of bacteria were changing we developed a method for extracting bacteria anaerobically and took three samples at different times in order to see how the bacteria were developing. In the future,

Investigating Gold-Palladium Alloy Nanoparticle Enhancement of Proton-Exchange Membrane Fuel Cell Power Output

Vickie Ye, Arnold O. Beckman High School, Irvine, CA

Cheng Pan, Dept. of Materials Science and Engineering, Stony Brook University, NY

Dr. Miriam Rafailovich, Dept. of Materials Science and Engineering, Stony Brook University, NY

Proton-exchange membrane (PEM) fuel cells have been widely acknowledged as the most promising zero-emission power sources for transportation, stationary, and mobile devices because of their low operating temperatures and high power densities. Currently, however, fuel cells are impractical because of the high cost of the essential platinum catalyst, which contributes to over 40% of the cost of a fuel cell unit. In order to make PEM fuel cells cost-efficient, power output must be increased to reduce the cost per watt of operation. To do this, the operational unit of the PEM fuel cell, the membrane-electrode assembly, must be enhanced.

¹It has been shown that thiol-coated gold and palladium nanoparticles synthesized by the two-phase Brust can form Langmuir films of oblong, platelet-shaped nanoparticles¹. Coating the Nafion membrane of the PEM fuel cell with these Langmuir monolayers has been shown to dramatically increase the power output of the fuel cell unit. The addition of gold-palladium alloy nanoparticles monolayers gives an even greater enhancement, due to ligand effects on the surface reactivity of the nanoparticles caused by alloying. Here we demonstrate and investigate the enhanced activity of PEM fuel cells with Langmuir films of Au-Pd nanoparticles.

Thiol-coated alloy nanoparticles were synthesized via the two-phase Brust method. The thiols prevent the nanoparticles from aggregating and determine interparticle distance in Langmuir films². The nanoparticles were then deposited onto Nafion membranes using the Langmuir-Blodgett trough, which controls the surface pressure of particles suspended on the surface of the water. The surface pressure at which the nanoparticles were deposited onto the membrane was varied over five samples. A target pressure of 4 mN/m resulted in the highest power output (Figure 1). At this surface pressure, the nanoparticles suspended on the surface of the trough form a condensed monolayer of platelet particles, which is ideal for electrocatalysis. At lower surface pressures, the nanoparticles are not close enough to cause to form a monolayer and at higher surface pressures, the monolayer collapses into a less catalytically active multilayer. TEM imaging and X-ray reflectivity measurements will be taken to further analyze the surface morphology and thickness of the Langmuir films. HRTEM imaging will also be done to analyze the crystal lattices of the alloys, which influence surface reactivity of the particles.

The dramatic increase in power output suggests that the nanoparticles are catalyzing some sort of reaction at the electrodes. To determine whether the nanoparticles are inducing a reaction at either electrode, Nafion membranes with only one side coated with nanoparticles were prepared and tested. There was a much more dramatic increase in power output in the sample with nanoparticles at the cathode of the PEM fuel cell, confirming suspicions that a reaction is being induced at the cathode. Mixed gases experiments and gas chromatography will be performed to further understand the nature of the catalysis of the nanoparticles.

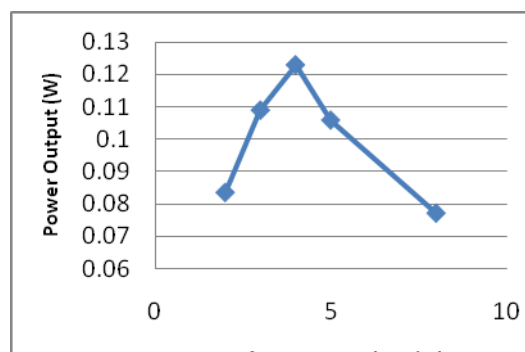


Figure 1 Effect of surface pressure on power output

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Analysis of Cathodic Waste Gas from a PEM Fuel Cell with Gold Nanoparticle Co-Catalyst

George Fei, George Walton Comprehensive High School, Marietta GA
Cheng Pan, Hongfei Li, and Dr. Miriam Rafailovich, Department of Materials Science & Engineering, Stony Brook University, Stony Brook, NY
Alex Pereira, Farmingdale State College, Farmingdale NY

The proton-exchange membrane (PEM) hydrogen fuel cells has been looked at as a strong potential candidate as an alternative energy source, especially for transportation. Attractive characteristics include minimum waste production, reasonable power output, and efficient energy conversion¹. However, the platinum electrodes used to catalyze the reaction are expensive and are susceptible to poisoning, especially since commercial hydrogen gas includes gases like carbon monoxide¹.

Recent studies have shown that adding gold nanoparticles to the Nafion membrane in a PEM cell improves the efficiency of a cell¹. However, it is unclear whether the increased power output is a result of gold's conductivity or some catalytic mechanism by the gold. If the mechanism reduces carbon dioxide to carbon monoxide, the waste gas could potentially poison the platinum catalyst.

Gas that flowed out from the cathode while the fuel cell ran was analyzed using a gas chromatograph. The gas samples were analyzed for carbon monoxide with gas-specific columns. The curves obtained from the resulting gas chromatography indicate the absence of carbon monoxide in the waste gas. However, it is possible that the unused oxygen gas is overwhelming the presence of the carbon monoxide. The chromatographer showed no presence of carbon monoxide (See Figure 1) even when the sensitivity of the chromatograph and the volume of the gas were increased.

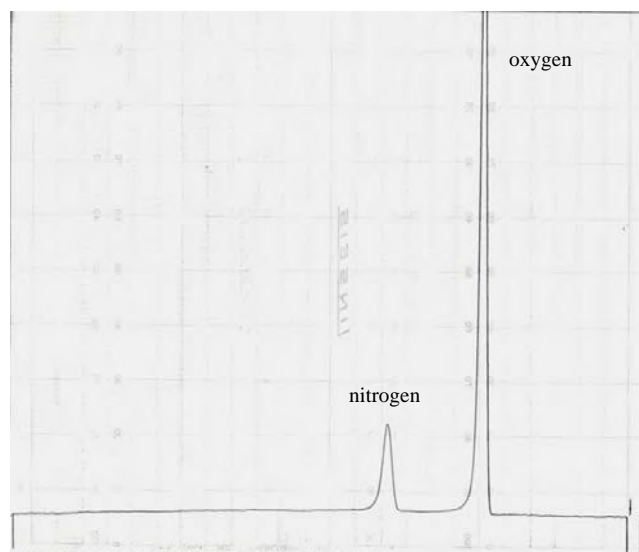


Figure 1:

The figure above shows the composition of the waste gas with the chromatograph at increased sensitivity. Reading from the right to left, the first large peak represents oxygen. This is the left over oxygen that did not react in the fuel cell and exited the system as waste. The peak was so high that the printer could not properly record the peak, indicating a strong presence of oxygen, as expected. The second peak is from nitrogen, which diffused into the gas syringe as gas was transported from the sample into the chromatographer.

Reference:

¹Mazumder, V., Lee, Y., & Sun, S. (2010). Recent Development of Active Nanoparticle Catalysts. *Advanced Functional Materials*, 1224–1231.

Materials For Energy Generation

Chairs:

Mariah Geritano, Stony Brook
University, New York

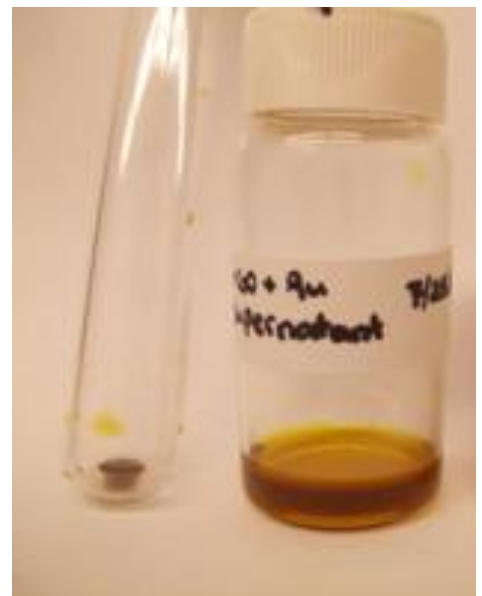
Sneha Subramaniam,
Columbia University, New York

Graduate Students:

Zhenhua Yang

Hongfei Li

Cheng Pan



Nanoscale Morphology of Various Polymer Blend Thin Films for Use in Bulk Heterojunction Photovoltaic Cells

Dean Fulgoni, Half Hollow Hills HS West, Dix Hills NY, Pierre Max Etienne, Suffolk Community College, Selden NY, Scott Dunaisky, Northwestern University, Evanston IL,

Zhenhua Yang and Dr. Miriam Rafailovich, Materials Science and Engineering, Stony Brook University, Stony Brook NY

Polymer based Bulk Heterojunction (BHJ) solar cells are known for their flexibility, low cost, and abundant availability¹. Previous study shows that by introducing polystyrene (PS), a third, non-conductive polymer, to the active layer, a self-assembled columnar structure forms which allows for higher power conversion efficiency, (Fig. 1). Thin films

consisting of one of three non-conductive polymers, Poly (methyl methacrylate) (PMMA), Poly (2-vinyl pyridine) (P2VP), and styrene-acrylonitrile (SAN), each blended in optimal ratios with poly (3-hexylthiophene-2,5-diyl) (P3HT) and [6,6]-phenyl C61 butyric acid methyl ester (PCBM), are spin casted onto silicon substrates and annealed to allow for phase segregation. The optimal polymer blend is determined before the addition of PCBM by examining images obtained by atomic force microscopy (AFM), and transmission electron microscopy (TEM) cross-section analysis is later used to observe columnar structure.

Neutron reflectometry and contact angle reduction are later used to indicate the presence of PCBM at the P3HT-polymer interface. Solar cells will be fabricated at Brookhaven National Laboratory and tested to find a correlation between structure, polymer blend, and power conversion efficiency.

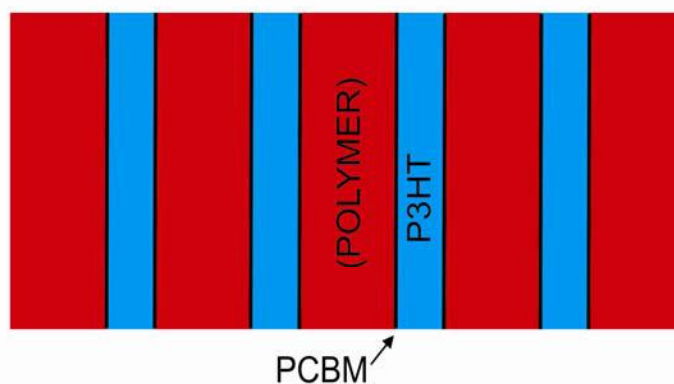


Figure 1 (Picture) – A third polymer added to the active layer allows for the formation of columns of P3HT after time is allowed for annealing. PCBM naturally goes to the interface between P3HT and the third polymer to reduce interfacial energy.

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- 1) g. Li, Y. Liang, C. Ray, S. Tsai, Y. Wu, J. Xia, Z. Xu, L. Yu, “For the Bright Future – Bulk Heterojunction Polymer Solar Cells with Power Conversion Efficiency of 7.4%”, Adv. Mater. **2010**, 22, E135-E138 g

Improving the Nanoscale Morphology of Polymeric Solar Cells Using the LB Trough

Justine Jang¹, Hongfei Li², Cheng Pan², Miriam Rafailovich², Zhenhua Yang²
Livingston Senior High School¹, Stony Brook University²

Although the efficiency for organic solar cells is lower relative to solar cells with semi-conductor technology, organic polymers have much higher potential because of lower material and production costs and the flexibility associated with designing and arranging organic polymers¹. In particular, P3HT (poly(3-hexylthiophene)) and PCBM ([6,6]-phenyl-C61-butyric acid methyl ester) have been shown to be the most promising electron donor/electron pair for the construction of the layer². In our experiment,

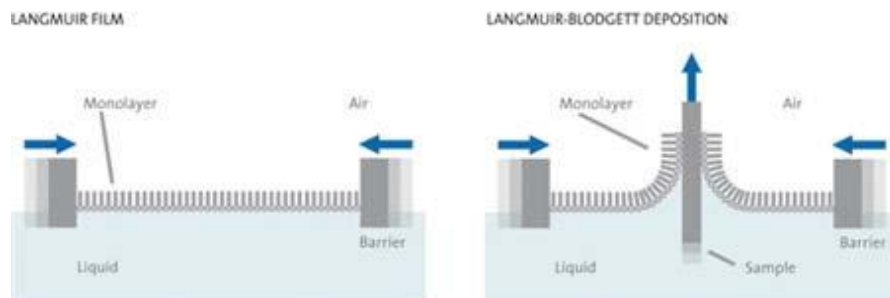
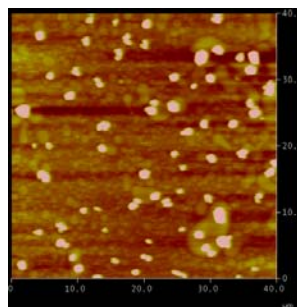


Figure 1. Diagram showing the deposition of nanoparticles onto the surface of a substrate using the LB trough. (www.ksvnima.com/langmuir-blodgett-langmuir-schaefer-technique)

PEDOT:PSS was spin-casted onto silicon wafers. PCBM and P3HT were then coated onto the wafer using the lifting method with the LB trough, as shown by Figure 1. The wafers were then examined under the Atomic Force Microscope, which recorded the topography of the layers coated onto the wafers (Figure 2). At first, for the PCBM/P3HT mixture, there

were many circular globules ranging in size from 0.3 nanometers to 5 nanometers in length. After annealing overnight in a vacuum oven at 140°C, the P3HT and PCBM appeared to have separated into two different layers. The PCBM formed small, uniform and widely dispersed globules on top of a layer of P3HT. The layers appear to be more uniform and better structured, as seen in Figure 2, which supports the hypothesis that using an LB trough would improve the structural uniformity of the solar cell. In the future, different annealing times can be used to investigate adjusting the degree of phase separation. In addition, the active layer may be coated onto an ITO wafer, and tested by exposing it to photons and recording the power output.

Figure 2. AFM image of annealed P3HT/PCBM on top of PEDOT and Silicon. There is noticeable phase separation and uniformity of the PCBM aggregates.



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Replacement of Aluminum Cathode with Graphene in Organic Polymer Solar Cells via UV/Ozone Exposure and Spin-Coating

Alexandra Se, Sneha Chittabathini, and Andrew Chen, Lawrence High School, Cedarhurst, NY 11516
 Mentors: Dr. Miriam Rafailovich, Rebecca Seroff, Cheng Pan, and Benjamin Goldmann
 Stony Brook University, Stony Brook, NY 11794

Organic polymer solar cells are a type of photovoltaic cell that uses conductive organic polymers or organic molecules for light absorption and charge transport.¹ The main disadvantages associated with organic photovoltaic cells are low efficiency, low stability and low strength compared to inorganic photovoltaic cells. Some of these disadvantages are caused by the aluminum cathode, which is evaporated under an ultra-high vacuum, making large industrial production difficult, and is easily oxidized. Graphene's potential to substitute the aluminum because it has a similar work function, 4.5 eV compared to aluminum's 4.3 eV, it does not oxidize easily, and is more conductive.

Graphite was first oxidized using a modified Hummer's method, synthesizing graphene oxide (GO).² The GO was easily suspended in a variety of solvents, such as 5:75 Ethanol/Water or N,N-dimethylformamide. The resulting suspension was then sonicated for 15 minutes to exfoliate the graphene oxide sheets and centrifuged for approximately 5-10 minutes at 600 RPM. The reducing agent, sodium borohydride (NaBH₄), was freshly made in H₂O and added dropwise into the stirring GO solution to a final concentration of 15g millimolar and allowed to stir overnight at room temperature. FTIR spectroscopy, TGA, and TEM (fig. 1) were performed in order to confirm the quality of the graphene and graphene oxide synthesized.

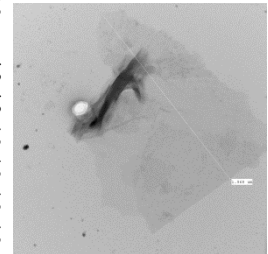
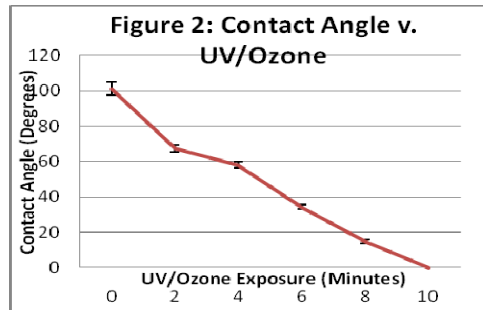


Figure 1: TEM images showing single-layered graphene sheets

A typical bulk heterojunction solar cell has an indium tin oxide (ITO) anode, a hole transporting layer composed of Poly(3,4-ethylenedioxythiophene) poly(styrenesulfonate) (PEDOT:PSS), followed by an active layer usually made up of [6,6]-phenyl C-61 butyric acid methyl ester (PCBM), poly(3-hexylthiophene-2,5-diyl) (P3HT), and finally the aluminum cathode. In our attempt to replace the aluminum cathode, it was necessary to address the interfacial tensions between the graphene suspended in 5:75 EtOH/H₂O and the active layer polymers, P3HT:PCBM, which are typically dissolved in hydrophobic solvent.



Surface modification using UV/Ozone treatment is one of the easiest, fastest, and low-cost ways to increase the surface hydrophilicity. P3HT:PCBM films were spin-coated at 2500 RPM for 30 seconds, creating thin films with an average thickness of 248.5 ± 13.263 Å. The thin films were then exposed to UV/Ozone for 0, 2, 4, 6, 8, and 10 minutes. The results of the contact angle measurements taken after the modification show a positive correlation between the length of exposure and the hydrophilicity of the surface. After exposing the active layer thin film to UV/ozone, a graphene suspension was spin-coated at 2500 RPM. Atomic Force Microscope (AFM) images (fig. 3) were taken after spin-coating and show that the graphene had homogeneously adhered. Solar simulation still needs to be performed in order to determine if graphene is a viable replacement for aluminum.

Surface modification using UV/Ozone treatment is one of the easiest, fastest, and low-cost ways to increase the surface hydrophilicity. P3HT:PCBM films were spin-coated at 2500 RPM for 30 seconds, creating thin films with an average thickness of 248.5 ± 13.263 Å. The thin films were then exposed to UV/Ozone for 0, 2, 4, 6, 8, and 10 minutes.

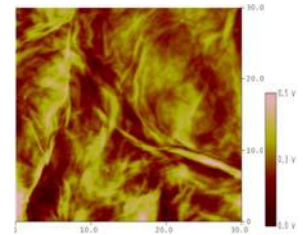


Figure 3: The graphene is represented by the wrinkles on the AFM image. The graphene is well-distributed on the active layer.

¹Hoppe, H.; Sariciftci, N.S. "Organic solar cells: An overview". *J. Mater. Res.*, vol. 19, no. 7, p. 1204 (2004)
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Functionalizing Graphene With Nanoparticles by Blending Nanoparticles Before Reducing Graphene Oxide

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Benjamin Akhavan, Rambam Mesivta HS, Lawrence, NY

Benjamin Goldman, CUNY, Queens, NY

Rebecca Isseroff, Lawrence HS Chemistry Department, Cedarhurst, NY

Dr. Miriam Rafailovich, Department of Materials Science & Engineering, Stony Brook University, Stony Brook, NY

Graphene is a one atom thick sheet of carbon atoms arranged in a hexagonal lattice structure which allows for its superior mechanical, electrical, and thermal properties¹. Functionalized graphene and doped graphene have different properties than regular graphene. By attaching different nanoparticles to graphene, we hope to alter the properties of graphene to produce specialized graphene with superior ability to regular graphene.

The procedure involves mixing graphene oxide (GO) with nanoparticles, allowing them to attach to the graphene, possibly with the help of the functional groups already attached, and then reduce the solution. Four salts have been mixed and reduced with graphene oxide; $\text{HAuCl}_4 \bullet 3\text{H}_2\text{O}$, KAuCl_4 , K_2PtCl_4 , and K_2PdCl_4 . First we mixed $\text{HAuCl}_4 \bullet 3\text{H}_2\text{O}$ at a relatively high concentration in GO in H_2O and reduced the solution, leaving the solution a gold color, as seen in Figure 1. When repeated with all the metals in GO in H_2O , at relatively lower concentrations, all of the solutions precipitated out, leaving a clear supernatant after reduction. The same occurred when the metals were placed in GO in 25/75 Ethanol/ H_2O solution.

Some of the metals, when examined with FTIR were reduced when NaBH_4 was added, but some were not. Some of the metals may be reduced before allowing the GO to reduce.

We hope to attach iron nanoparticles to graphene, using iron pentacarbonyl, in order to enhance its magnetic properties and attach gold nanoparticles to enhance the graphene's catalytic abilities. Previous research has shown that diffusing iron pentacarbonyl into clay has allowed the iron to latch onto the clay and give it magnetic properties².

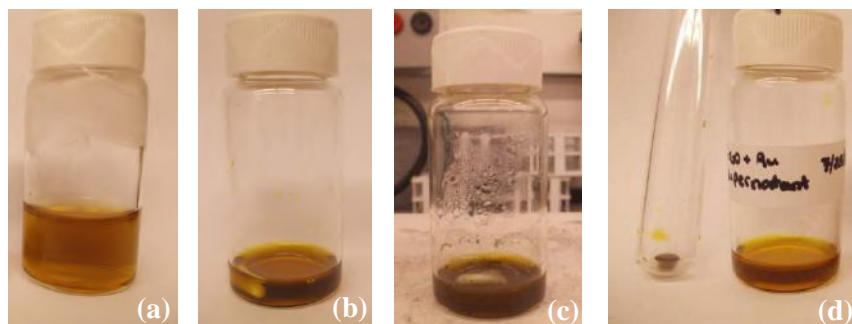


Figure 1 (a) Graphene oxide in H_2O (b) $\text{HAuCl}_4 \bullet 3\text{H}_2\text{O}$ in GO solution (c) Reduced HAuCl_4 and GO with NaBH_4 (d) The precipitate (left) and supernatant (right) of the reduced HAuCl_4/GO solution separated for FTIR

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¹ Huang, Xiao, et al., "Graphene-Based Materials: Synthesis, Characterization, Properties, and Applications", *Small*, Volume 7 Issue 12 (2011) Page 1876

² Rafailovich, Miriam, et al., "The effect of organoclay and adsorbed FeO_3 nanoparticles on cells cultured on Ethylene/Inyl Acetate substrates and fibers", *Biomaterials* Volume 30 (2009) Page 18g

The Effect of Morphology on Phase Formation, Expansion, and Saturation Time of Silicon Nanowires on Electrodes Using the Lattice Boltzmann Method(LBM)

Jerry Liu¹, Ning Sun², Dilip Gersappe³, Miriam Rafailovich⁴

¹Los Altos High School, Los Altos, CA

^{2,3,4}Department of Materials Science and Engineering, Stony Brook University, NY

Lithium ion batteries encounter great utility in the field of portable electronics. Graphite has traditionally been used as the negative electrode material, but silicon has garnered much interest as a more beneficial replacement, with a theoretical capacity of 4200 mAhg^{-1} , or more than ten times that of graphite¹. Yet silicon undergoes large volume expansion(as high as 400%) during charging cycles, causing fracture and tremendous capacity loss. To counteract this property, researchers have developed silicon nanowires (NW's), grown directly on a current collector, which effectively reduces stress and strain². A model simulating the phases of the lithiated silicon and the anisotropic swelling may offer explanations for the results of other researchers. Furthermore, the morphology of the nanowire has yet to be investigated, and measuring a time of ion saturation in the nanowire, after which harmful plating occurs, has also been lacking in research.

The Lattice Boltzmann Methods(LBM) are an innovative series of computational methods designed to simulate fluids. Each block of fluid can be represented as a node on a lattice with multiple directions. At each time step, a part of each block streams into the next node, while the rest undergoes collision; this is modeled by the Bhatnagar-Gross-Krock(BGK) operator³. LBM is used in the project as the core foundation for the model, providing a fluid-based approach.

The model was constructed in C++, using Palabos version 1.1, which is a code library encapsulating the LBM. The earliest model focused purely on diffusion, without incorporating any electrochemical reactions. Diffusion across the boundary from the bulk electrolyte to the nanowire was governed by a first order reaction. **Figure 1** shows an inverse relation between the thickness of the nanowires (while maintaining a constant total nanowire surface area) and the time of first saturation.

A more advanced model was then developed to analyze silicon nanowire dynamics more specifically. For the purpose of isolating a variable, a single nanowire was constructed on the left electrode; the aspect ratio of the nanowire(while maintaining a constant area) would be the independent variable. Along with the time of saturation, the phase formation and expansion of the nanowire would be analyzed as well. Lithium ions were reduced according to the Nernst equation. Part of the lithium atoms that formed would gradually lithiate the silicon according to a second order reaction, and the other part would diffuse through the lithiated silicon. At each node, the lithiation reaction was cut off once $\text{Li}_{3.75}\text{Si}$

was reached, which is the form of fully lithiated silicon. Volume expansion was handled by an incremental process, dependent on the extent of the lithiation of the silicon.

Extensive simulations will be run using the advanced model.

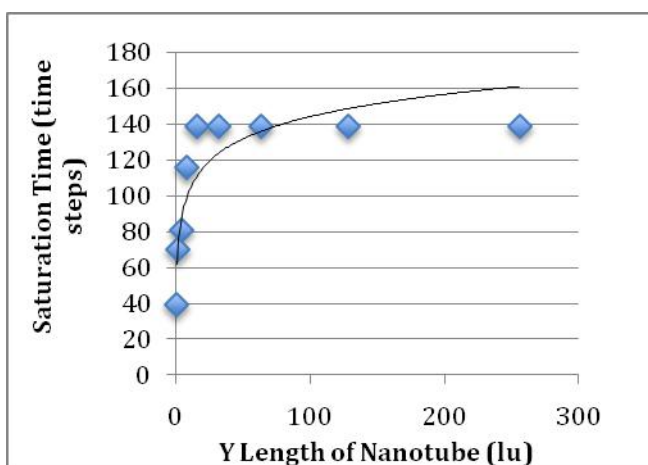


Figure 1. Indicates the saturation time of the nanowire, depending on the thickness.

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Implementing Graphene into a Conductive Polymer Spin Cast

Isaac Robson, Bentonville High School, Bentonville, AR
Zhenhua Yang and Miriam Rafailovich, Stony Brook University, Stony Brook, New York

Graphene is a highly conductive material, consisting of solely hexagonally-arranged carbon atoms in a two dimensional plane. Utilizing graphene to its full potential is very difficult however, because it reverts into graphite. Graphite is an allotrope of carbon with almost the opposite properties of graphene, and the 're-stacking' of graphene into graphite is highly undesirable¹; one potential method to preserve the properties of graphene would be to blend small amounts of graphene into polymers which share conductive characteristics. Through blending, the electrical properties of graphene could be transferred into a polymer blend. Alternatively, graphene could serve to enhance the efficiency of these polymers in organic solar cells².

Graphene was spin casted onto silicon wafers with mixtures of poly-2-vinylpyridine (P2VP) and poly(3-hexylthiophene) (P3HT) which were analyzed after 1 hour of annealing at 150° C. These mixtures were prepared by suspending grade 2 graphene from cheaptubes.com (commercial graphene) in a 50:50 chlorobenzene:methyl-2-pyrrolidinone mixture. The graphene was diluted to 0.5 mg/mL, and then mixtures were created with 5 mg/mL P2VP, 5 mg/mL P3HT, and a 10 mg/mL P2VP:P3HT blend (1:1 by weight). Large graphene chunks were noticeable throughout the surface of the films, but TEM imaging would be necessary to determine how graphene interacted at the interfaces of the polymers. In the combined mixture, little phase separation occurred, because of this, a much longer annealing time would be needed for visible separation.

A second experiment was conducted to determine how the location of the graphene on the surface changed with annealing, along with P2VP/P3HT interaction. 30:10:5, 72:10:5 and 144:10:5 samples of P2VP:P3HT:Graphene were prepared. These samples were analyzed with atomic force microscopy, and re-analyzed after overnight annealing at 170° C. Significant changes in the domain presence were noted, however more data is necessary to distinctly determine significant differences in the allocation of graphene platelets.

Additionally, a height vs. concentration graph of a P3HT spin cast was plotted, to aid future work with P3HT. Figure 1 displays the results of ellipsometer readings. As expected, the determined film thickness increases linearly with concentration. The determined

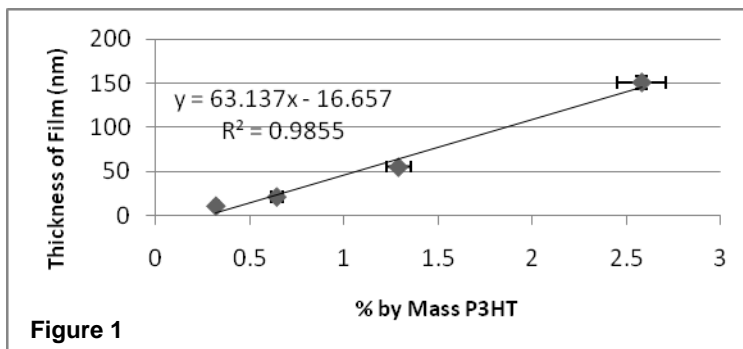


Figure 1

equation for the experimental results was $\text{Thickness}_{\text{film}} = 63.1 \times (\text{Percent}_{\text{mass}}) - 16.66 \text{ nm}$.

The primary data collected was in the ability to suspend graphene. In many previous experiments, groups had difficulty suspending commercial graphene in any solvent, and failed to suspend any type of graphene in chlorobenzene. In this experiment however, 0.5 mg/mL (high) concentrations of commercial graphene were produced in a mixed solvent of chlorobenzene and NMP (1-methyl-2-pyrrolidinone). This suggests that commercial graphene might suspend well in NMP, and more experiments should be conducted to determine the compatibility of graphene with NMP, as well as other mixed solvents. Difficulties occurred when trying to dissolve P3HT with graphene, and more chlorobenzene should be used in subsequent trials.

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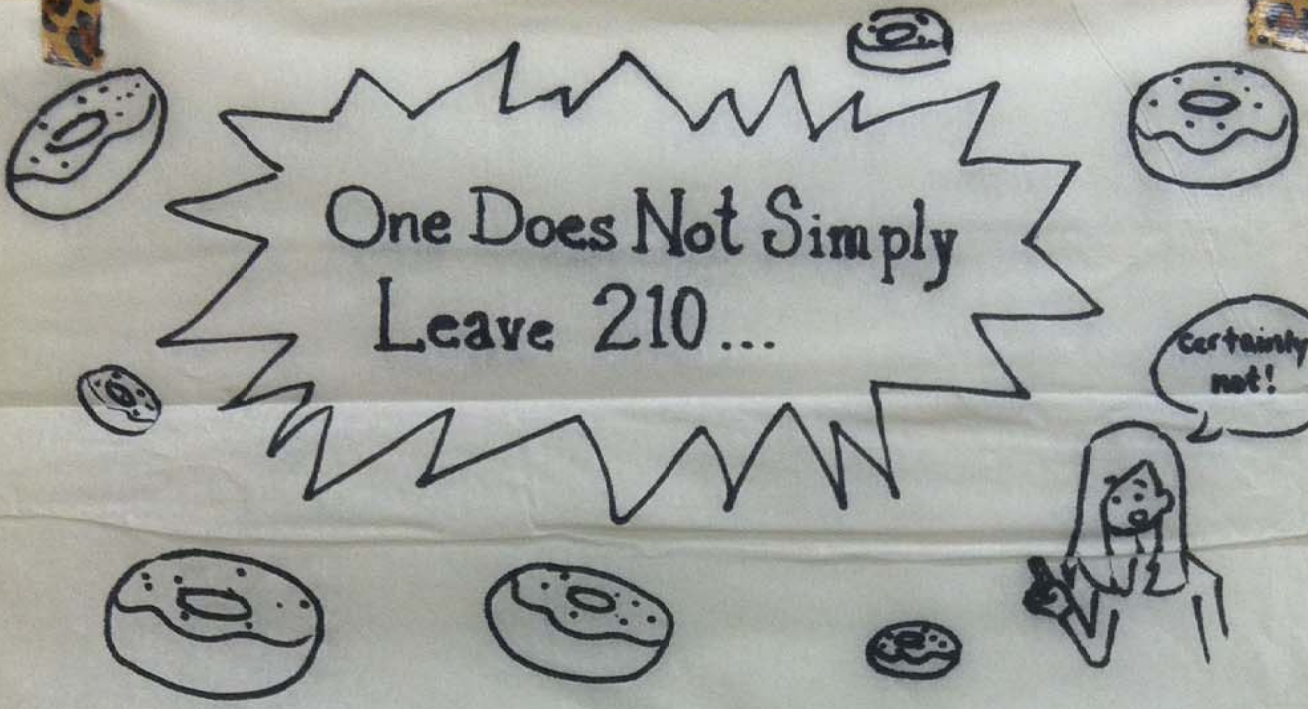


Entenmann's

A collection of handwritten signatures and names, including: Niki Greenberg, melo, Rachel yang, Kimia Liadkhanpon, Rachel Davies, Rachel Yang, Mykhailo, Jacob Wax, Raviel Bachal, Michael Blum, Nicolette Almer, Adam Fuzurs, Holly Flores, Puya Bansal, Alex Na, Monica Butra, Justin Jang, Daniel Shostman, Alex Lee, Allison Lee, Sachit Singal, Jay Sin, Ben Hoent, Sean Ferguson, Sreha Chittabathini, Sanjita Singal, Andrew Chen, Vicky Li, Alina Panikaran, Ariella Applebaum, Hannah Lin, George Fei, Womeh Ueepar, Justin Markin, Haris Nair, Rob Aldana, Benjamin Akhavan, and others. There are also some drawings, including a stylized face and a signature that looks like 'ALTA LEDBOWITZ'.

One Does Not Simply
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